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Chemically Modified Polymeric Resins for High Performance Liqui Chromatography, Solid-phase Extraction and Organic Separation b LC and GC

by

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Chemically modified polymeric resins for high performance liquid chromatography, solid-phase extraction and organic separation by LC and GC

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Polystyrene divinylbenzene (PS/DVB) was chemically modified by the introduction of several hydrophilic functional groups, $-COCH_3$, $-CH_2OH$, $-CH_2CN$, $-COCH_2CH_2COOH$, and one hydrophobic group, $-C(CH_3)_3$. Those functional groups changed the chromatographic behavior of various test compounds on PS/DVB resin. On the hydrophilically modified resins, the capacity factors (k′) of polar compounds (such as phenol, p-cresol) are increased, while the k′ of non-polar compounds (such as toluene, cumene) are decreased. The opposite is true for hydrophobically modified resin. The change of k′ of some compounds resulted in the change of separation factor α ($=k_2'/k_1'$). The modified resins offer an additional selectivity parameter for liquid chromatographic separation.

The chemically modified polymeric resins (PS/DVB-$COCH_3$ and PS/DVB-CH_2OH) are easily wetted by water. They are excellent for solid-phase

extraction (SPE), especially for polar compounds such as phenols. The recoveries of phenols and multihydroxy phenols are overwhelmingly higher than those obtained using commercial SiC_{18}. An HPLC system with a mini-column (4.0 x 4.0 mm) was used to study SPE of various compounds on different stationary phases. The elution efficiency of various solvents was also studied.

Sulfonated PS/DVB was used for separation of neutral, basic and weakly basic organic compounds in this research. The basic and weakly basic compounds were separated by an on-line mini-column. The neutral and basic compounds were separated by an off-line mini-column. The sulfonated PS/DVB was also used for separation of amine isomers, showing better separations than a commercial SiC_{18} column.

A special, 5 μm non-porous, spherical PS/DVB resin was sulfonated and then packed into the split liner of a capillary gas chromatography to be an amine abstractor. When a sample mixture of basic amines and neutral compounds was injected into the liner, all the basic amines were abstracted quantitatively while all the neutral compounds passed through. The quantitative determination of passed neutral compounds was excellent. A 200-fold excess concentration of a basic compound in the presence of a neutral compound with the same retention time could be abstracted.

This special non-porous PS/DVB resin was converted synthetically to a polymeric-mercuric resin, which was used as a selective abstractor of mercaptan compounds in the split liner of capillay GC. Non-mercaptan (both sulfur and non-sulfur) compounds can pass the liner and be quantitatively determined well. The injection temperature was also studied.

Chemically modified polymeric resins for high performance liquid chromatography, solid-phase extraction and organic separation by LC and GC

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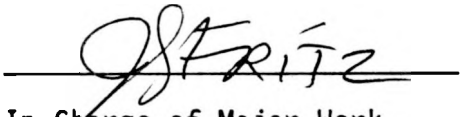

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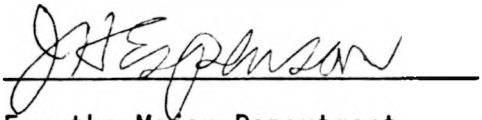

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GENERAL INTRODUCTION

Polymeric Resins

Polymeric resins are being used more frequently as packings or stationary phases in modern (high performance) liquid chromatograhy as scientists seek solutions to separation problems. Although silica columns currently dominate the liquid chromatography industry, a part of columns sold today contain polymeric packing materials. The percentage will increase as new polymers are developed and synthesis technique are improved.

Although silica-based columns are very efficient, they do have several drawbacks. The most serious of these is the instability of silica in alkaline solution or in highly acidic solution. Silica packings also possess residual silanol groups that can cause peak broadening or tailing by interacting with polar compounds such as amines and alcohols. After a gradient elution, silica-based columns may require a rather extended treatment to re-establish equilibrium for the next run.

In contrast, modern polymeric resins are unaffected by the pH of mobile phases. Many polymers are stable from pH 0 to 14. Consequently, for applications in which column stability is important, and in most

applications involving ions, polymeric columns are the appropriate choice.

A large number of polymeric resins have been synthesized for chromatography, which are reviewed in depth [1]. They include polystyrene divinylbenzene (polystyrene as monomer and divinylbenzene as crosslinker), polyacrylamide (acrylamide as monomer and N,N′-methylenebisacrylamide as crosslinker) [2,3], polyvinylacetate (vinylacetate as monomer and divinyl adipate as crosslinker) [4], polymethyl methacrylate (methylmethacrylate as monomer and glycol dimethacrylate as crosslinker) [5,6], polyethylene glycol (methacrylate as monomer and bisethylene glycol methacrylate as crosslinker) [7], etc.

Two types of polymeric structures are used in modern liquid chromatography: microporous and macroporous. "Microporous" polymers (sometimes called "gels") are cross-linked copolymers in which the porosity is determined by the amount of crosslinking. A lower crosslinking gives larger porosity, but a softer resultant product. Low crosslinked polymers (i.e., containing less than ca. 8% of the crosslinking agent) are usually not sufficiently rigid to be used in modern LC equipment. "Macroporous" polymers, as the term is used here, are copolymers with a high degree of crosslinking that are synthesized in the presence of a "porogen". This is a compound that is soluble in the

monomer but insoluble in the polymer. Polymers synthesized in the presence of a porogen can be made to contain large voids or pores. Usually, polymers formed in this manner are also rigid.

The most important and widely used polymeric resin is polystyrene divinylbenzene (PS/DVB) resin. The synthesis of crosslinked copolymers of styrene and divinylbenzene has been studied intensively and is well documented [8, 9]. The starting monomer is styrene and divinylbenzene (DVB) is used as crosslinker. The amount of DVB can reach about 55% (w/w). At 55% DVB, the copolymer shows practically no swelling and possesses a permanent porosity [9]. The network structure of PS/DVB is illustrated in figure 1. Commercial products differ in bead size and pore size. There are even non-porous products on the market, designed for the rapid separation of peptides and proteins by reversed phase HPLC [10]. Since PS/DVB resins are stable in the pH range of 0-14, they find increasing applications in the separation of low molecular weight compounds, peptides, and proteins by means of reversed phase chromatography [11, 12] and as parent materials for the synthesis of derivatized packings in interactive chromatography of biopolymers [13, 14].

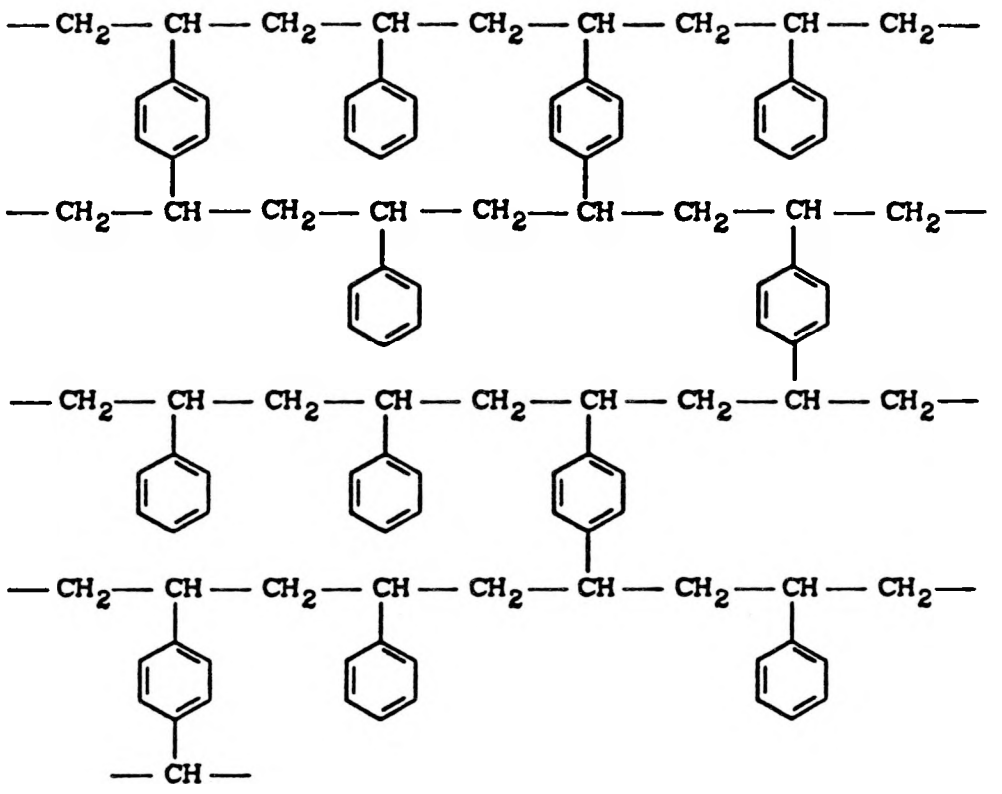

Figure 1. Structure of polystyrene divinylbenzene

Polystyrene Divinylbenzene resins in HPLC

PS/DVB itself is a lipophilic stationary phase and widely used for reversed-phase HPLC. A large number of publications have appeared on its chromatographic behavior and application.

F. Nevejans and M. Verzele studied the pore structure and its influence on their behavior of polystyrene phase [15]. They found that microporisity played an important role in the chromatography performance of this type of phase. For polystyrene compatible solutes, the partitioning (adsorption) process involves not only the surface of polystyrene packing material, but also the whole bulk mass. In other words, the solutes penetrate into the polystyrene matrix. This is promoted by microporosity, Microporosity in polystyrene phase is not constant but can change with the nature of the eluent and sample.

Richard I. Greyson and Andrea M. Patch compared PS-DVB and SiC_{18} for HPLC of gibberellins [16]. They studied three performance characteristics of these two reversed-phase materials as they affect gibberellin chromatography: capacity factors (k′), selectivity (α) and efficiency (N). They found that polystyrene gave significantly higher capacity factor, higher selectivity but low efficiency.

S. Coppe, et al. characterized some PS/DVB sorbents for reversed-phase chromatography [17]. Chromsorb 101 (surface area 11 m^2/g, 0.3 ~

0.4 μm pore diameter), Porapak Q (surface area 557 m^2/g, 0.075 μm pore diameter), and PRP-1 (surface area 415 m^2/g, 0.075 μm pore diameter) were compared. The efficiency of first two are reported satisfactory, but PRP-1 gave best results. The swelling of polymers in acetonitrile was shown to be higher than in methanol.

H. M. Smith and D. R. Garside studied the effect of organic modifiers on the separation and efficiencies of homologous on porous polymeric columns [18]. The retentions of the homologous and test compounds using different eluents were compared. Considerable improvements were found in the peaks shapes and column efficiency when changing the organic components of eluent from methanol and acetonitrile to tetrahydrofuran.

A typical PS/DVB column is PRP-1 (Hamilton Co.). It has been used for a variety of separations and determinations of various analytes, such as determination of multifunctional carboxylic acids by fixed-site ion exchange [19], determination of nitrilotriacetic acid, ethylenediaminetetraacetic acids and related aminopolycarboxylic acids [20], analysis of plant phenolics by HPLC [21], HPLC assay for dilevalol in human plasma and urine [22], determination of ergot alkaloids in wheat [23], separation of quaternary ammonium compounds [24], separation of amino acids, peptides and derivatives [25], determination of

acetaminophen in human plasma by ion-pair reversed-phase HPLC [26], quantitative analysis of doxycycline and related substances by HPLC [27], determination of erytyromyein in human plasma [28], separation and indirect ultraviolet detection of phosphorylated sugars [29], analysis of industrial waste water with pre-column technology [30], preconcentration and analysis of selected pollutants in industrial effluent [31], purification and analysis of DNA oligomers [32], determination of 5-fluorouracil in plasma and whole blood by ion-pair HPLC [33], determination of inhibition of monoamine oxidase activity in platelet rich plasma of depressed patients treated with phenelzine [34], and LC enrichment, separation and determination of chlorophenol and phenoxyacetic acids [35], etc.

Although PS/DVB resin is widely used in HPLC, there are still some problems associated with this resin. Since the surface of this resin is highly hydrophobic, the retention times of some non-polar compounds may be too long. Polar compounds with similar molecular weights and small difference in polarity might not be discriminated well and, therefore, poorly separated. Protein compounds can be denatured by this highly hydrophobic surface.

In section I of this dissertation, PS/DVB resins were chemically modified by the introduction of various functional groups, including

several polar functional groups and one non-polar functional group. The chromatographic behavior of various sample compounds were compared on the derivatized and un-derivatized PS/DVB resins. The results showed very interesting phenomena which occurred on those resins. Some sample separations showed the advantages of the derivatized resin over the un-derivatized resin.

Polymeric Resins and Solid-Phase Extraction

Solid-phase extraction (SPE) is gaining wide acceptance as a tool for sample clean up, isolation and preconcentration [36, 37, 38]. The principle and type of sorbents of SPE have been studied in detail [38, 39]. Although, SPE can be used in different phases, such as sorption from the gaseous phase and desorption into gaseous or liquid phase, sorption from liquid phase and desorption into gaseous or liquid phase, the most commonly used one is sorption in liquid phase and desorption into liquid phase. Also, SPE can be used for both organic and inorganic compounds. My research in this dissertation focuses on SPE of trace amounts of organic compounds.

The stationary phases used in solid-phase extraction of organic compounds actually are the same as those used in reversed-phase liquid chromatography, such as silica based C_{18}, C_8, cyclohexyl and polymer

based PS/DVB [40, 41]. Of those, SiC_{18} and PS/DVB (XAD) resins are the most widely used.

Labib Ghaoui compared the silica based bonded phase sorbents for isolation of polynuclear aromatic hydrocarbons from water [42]. He found out that those sorbents sometimes are not efficient.

B. Tippins studied the methods of solid phase extraction for sample preparation of a variety of endogenous compounds [43]. The sorbents and techniques were discussed.

S. H. Hoke, E. E. Bruggemenn, L. J. Boxter, and T. Trybus determined phenoxy acid herbicides using solid phase extraction instead of liquid-liquid extraction, then followed by HPLC method [44].

A. K. Barnham, G. V. Calder, J. S. Fritz, et al used macroreticular XAD resin to extract trace organic contaminants from potable water and determined ppb ~ ppm levels of organic compounds [45].

Pieter B. Rossum and Ronald G. Webb measured the recovery efficiency of XAD-2,4,7,8 resin and their mixtures using distilled water samples which contained thirteen organic pollutants. They found that an equal-weight mixture of XAD-4 and XAD-8 was the most efficient [46].

Thomas G. Greco and Robert L. Grob extracted specific nitrogen-containing organic compounds from soil samples, concentrated by solid phase extraction, and separated and quantified by capillary GC [47].

Hassan Salari compared AL-LE and six other solid absorbents as effective tools for extraction of phospholipids from plasma. He found out that Si gel and SiC_{18} were the poorest and that XAD-2 and XAD-4 were the best [48].

M. W. F. Nielen, U. A. Th. Brinkman and R. W. Frei used small precolumns packed with SiC_{18}, PRP-1 and cation exchange materials for on-line group separation and trace enrichment of industrial waste water samples. The concentrated solutes were eluted and separated by LC [49].

I. Ogawa and J. S. Fritz used a small column containing zeolite ZSM-5 for concentration of low concentration of aldehyde and ketones in aqueous samples. Then the compounds were eluted and converted to 2,4-dinitrophenylhydrazones and separated by LC [50].

Andrzey Przyjany evaluated XAD-2,4,7 and Chromsorb 102, 105, 106 for preconcentration of organosulfur compounds from water. Chromsorb and XAD-4 were found to be the best [51].

A. Tateda and J. S. Fritz used XAD-4 or Spherocarb in a mini-column to effectively absorb most organic contaminates. The absorbed organic compounds were eluted by an organic solvent and the organic solutes were separated by GC [52].

C. Chriswell, C. Chang and J. S. Fritz determined phenols in natural water and treated drinking water by sorption on macroporous anion exchange resin, elution with acetone and then quantified by GC [53].

G. A. Junk, J. J. Richard, et al. isolated organic impurities in water by macroporous resin, then eluted by diethylether, concentrated by evaporation, finally separated and determined by GC [54].

Although polymeric resins are successfully used for solid-phase extraction of many organic compounds, it is really difficult for solid-phase extraction of polar compounds, such as phenols and multi-hydroxy phenols. This problem is even greater when using silica based resin, such as SiC_{18} or SiC_8. The extraction efficiencies are lower than 40% for phenols.

In section II of this dissertation, some chemically modified polymeric resins were synthesized by the introduction of hydrophilic functional groups. The chemically modified resins are easily wetted by water and therefore have a good contact with organic compounds in aqueous solution. They greatly improved the solid-phase extraction efficiency, especially for polar compounds such as phenols. The high efficiency of solid-phase extraction of chemically modified resins was improved by a systematic study of capacity factors (k′) of several organic compounds on different stationary phases using an HPLC system. The efficiencies of

various solvents in elution of organic compounds absorbed on the stationary phase were also studied.

Special applications of chemically modified polymeric resins in LC and GC

Although LC and GC are the most powerful separation methods known, scientists are constantly looking for ways to achieve greater selectivity. A very common and useful method is to separate sample components into various groups by solvent extraction or by the use of special stationary phases. Some types of on-line preliminary group separations are fast and convenient. These are performed with a short, packed column (called a mini-column), placed just before or after the LC and GC columns to remove various sample constituents.

The packing materials in the mini-column can be varied according to necessity. Various inorganic materials have been used quite often. Chemically modified polymeric resins are more and more commonly used as packing materials. Chemical modification of polymeric sorbents allows alteration of the chemical characteristics of the polymer surface in order to obtain sorbents with greater selectivity or special usage.

K. I. Sakodynskii, G. P. Terekhova and L. I. Panina studied the GC properties of sorbents with nitro and amino functional groups bonded directly to the benzene ring in a polymeric matrix. The selectivity of

the sorbents was determined and practical recommendation for the application of sorbents in GC are made [55].

K. I. Sakodynskii, L. I. Panina and S. B. Makarova studied GC properties of some chemically modified polymeric sorbents, which were based on macroporous styrene, 2-methyl-5-vinylpyridine and divinylbenzene copolymers. The specificity and selectivity of the sorbents were estimated [56].

R. I. Hirsch, H. C. Stober et al. used macroreticular cation exchange resins in gas-solid chromatography. They found that these resins were highly adsorptive packings for gas-solid chromatography and showed good selectivity among hydrocarbons. The silver form of the resin retained aromatic and olefinic compounds strongly and separate geometric isomers of the olifins [57].

R. I. Hirsch and S. G. Phillips used lightly sulfonated porous polymer in gas-solid chromatography, which were efficiently selective packings for gas-solid chromatography [58].

C. D. Chriswell, L. D. Kissinger, and J. S. Fritz used Chromsorb F AW/DMCS coated with copper (II) chloride as amine abstractor, which was packed into a pre-column before GC separation column. The pyridine solvent was removed and the samples were well separated [59].

J. Frycka and J. Pospisil used orthophosphoric acids as a substracting agent to remove bases in an chromatographic pre-column quantitatively [60].

R. R. allen used a short column of FFAP to selectively remove aldehyde from complex mixtures during GC [61].

J. Mcconagh, W. G. Maggoner, E. G. Hamilton, B. Hindenach and R. P. McDonagh used organomercuric agarose to purify plasma factor XIII, fraction 4 with affinity chromatography [62].

Z. Slovak, M. Smrz, B. Docekal and S. Slovakova prepared the sorbent Spheron Thiol, which is a hydrophilic glycolmethacrylate gel with side chains containing thiol groups, and used it to absorb Hg, Sb, Bi, As, Ag, Cu and Pt in solutions up to 1-3 mole concentration of sulfuric or hydrochloric acid. [63].

M. W. F. Nielen, R. Bleeker, R. W. Frei and U. A. Th. Brinkman used a pre-column packed with a mercury (II)-8-hydroxyquinoline phase for the selective trace enrichment and cleanup of 2-mercaptobenzimidazole [64].

Many workers have been using different methods for various special applications by these pre- or post- mini-column techniques in chromatography. Along with the development of packing materials and more rigid requirements of analysis, there are even more opportunities for

using these special techniques to solve difficult problems in chromatography.

In sections III, IV, and V of this dissertation, some special applications of chemically modified PS/DVB resins are described. In section III, sulfonated PS-DVB was used for separations of neutral and basic compounds as well as basic and weaker basic compounds in LC. The neutral and basic compounds were separated by an off-line mini-column packed with sulfonated PS/DVB resin at first and then determined by HPLC. The basic and weakly basic compounds were separated by an on-line mini-column packed with sulfonated PS/DVB resin. Also, some amine isomers were separated by a sulfonated PS/DVB column and the results are better than those obtained with a commercial SiC_{18} column. In section IV, a special non-porous PS/DVB resin was sulfonated and packed into the split liner of a gas chromatograph. This resin was used as amine abstractor to selectively abstract amine compounds in gas phase and allow neutral compounds to pass through. Up to 200-fold excess of amine present in the sample can be cleaned up. In section V, the same non-porous PS/DVB resin was synthesized into a polymeric-mercuric resin and used as mercaptan abstractor. This polymeric-mercuric resin exclusively abstracted mercaptan compounds in capillary gas chromatograph while allowed non-

mercaptan compounds (bother sulfur and non-sulfur) to pass through. Also, the injection temperature was studied for those abstractors.

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SECTION I. CHEMICALLY MODIFIED POLYMERIC RESINS FOR HIGH PERFORMANCE LIQUID CHROMATOGRAPHY

INTRODUCTION

Reverse-phase high performance liquid chromatography (HPLC) is generally performed by silica packing with C_{18}- or C_8- bonded phased silica [1]. Although such columns are very efficient, they do have several drawbacks. The most serious of these is the instability of silica in alkaline solutions or in highly acidic solutions. Silica is readily degraded by water and eluent with pH value greater than 8. Therefore, many separations occurring optimally at elevated pH levels cannot be addressed with silica columns. Silica resins also have residual silanol groups that may cause peak broadening or tailing by interactions with polar compounds such as amines and alcohols. After a gradient elution, silica-based columns require a rather extended treatment to re-establish equilibrium for the next run [2]. Silica columns have also displayed irreversible adsorption of solutes which may cause shortening of column lifetime.

In recent years there has been a growing interest in polymeric materials for use in reversed-phase HPLC. In particular, resins based on polystyrene-divinylbenzene (PS/DVB) are stable with eluents from pH 1-14 and give excellent separations [3-6]. Polystyrene packings for chromatography are organic porous spheres obtained by copolymerization of

styrene and divinylbenzene. Such chemicals are chemically stable and insoluble in solvents. Some difficulties have been associated with polymeric resins, such as swelling in the presence of organic solvents. This can be troublesome when solvent gradients are used [7]. However, rapid improvements are being made in stability and performance so that polymeric resins are now considered to be attractive for HPLC. The improvements in the techniques used to synthesize polystyrene beads have led to their use as reversed-phase HPLC packing materials much more successfully than before.

In order to take advantage of the desirable properties of polymeric resins, a considerable variety of polymeric resins have been prepared and used for HPLC [8-13]. There has been a remarkable tendency to bond C_{18}-hydrocarbon groups to the surface so that the retention behavior of the polymeric resins will closely approximate that of the popular C_{18} silica materials [11,13,14]. In fact, silica resins with a C_{18}- or C_8- bonded phase dominate the field of reversed-phase liquid chromatography. Using these resin columns, necessary changes in selectivity are brought about by varying the solvents that make up the mobile phase. Several investigators have used silica columns with more polar bonded phases to provide an additional parameter for varying selectivity in liquid chromatography [15-17]. A number of people have investigated the

synthesis and modifications of different kinds of polymeric resins. Yang and Verzele described a hydrophilic modification of polystyrene resin and its application [8,18]. Tanaka, Sato, Miyazak, and Yamada described the synthesis of stearyl methacrylate copolymer and its application [9]. Yasukawa, Yamura, Uchida, Yangihara and Noguchi described the introduction of octadecyl group into vinyl alcohol copolymer [10]. Tennikova, Horak, Svec et al described how to make hydrophilic resin by hydrolysis of copolymer glycidyl methacrylate-ethylene dimethacrylate (GMA-EDMA) [19]. Dawkins, Lloyd and Warner described the synthesis of polyacrylamide-based octadecyl (PLRP-C_{18}) packing and made a comparison with polystyrene based polymer [5]. Interaction Chemicals Company (Mountain View, CA) provides a ACT-1 column which actually is polystyrene-based C_{18} column and made some comparison with PRP-1 packing material and octadecyl silica packing material [20].

All these studies showed that more and more people are developing polymeric packing materials for reversed-phase liquid chromatography because of its advantage. Polystyrene divinylbenzene resin is one of the favorite polymeric resins to be modified. Since it has reactable benzene rings, many functional groups can be introduced by the Friedel-Crafts reaction or other reactions. The introduced groups change the property

of the original polystyrene resin and therefore change the chromatographic behavior.

In the present work cross-linked polystyrene resins have been modified by introduction of any of several hydrophilic functional groups, and in one case by introduction of a more hydrophobic group. The modified resins are easily prepared by the Friedel-Crafts reaction with the benzene ring of the polymer. It is shown that the derivatized resins can be used for practical liquid chromatographic separations and that the type of functional group incorporated in the resin has a major effect on the retention behavior of various analytes.

EXPERIMENTAL

Apparatus

A Gilson 302 HPLC system equipped with a microprocessor controller (Gilson Medical Inc., Middleton, WI), a 7125 Rheodyne injector (Rheodyne, Berkeley, CA) equipped with a 20 microliters loop, a Spectroflow 783 Kratos variable-wavelength UV-Vis detector (Kratos Analytical Instruments, Ramsey, NJ), a Fisher Redcordall series 5000 recorder (Fisher Scientific/Instrument Lab, Itasca, IL), a Hitachi D-2000 intergrater (EM Science, Cherry Hill, NJ), were used for high performance liquid chromatography. A Shandon HPLC packing pump (Shandon Southern Products Limited, Sewichley, PA) was used for column packing. An IBM FT-IR instrument (USA Bruker Instruments Inc., San Jose, CA) was used for structure determination.

Reagents and chemicals

Two kinds of polystyrene divinylbenzene (PS/DVB) resins were used in this experiment, non-spherical XAD-4 resin (Rohm and Haas Co., Philadelphia, PA) of 45-58 μm particle size, 50 Å pore size and 784 m^2/g surface area and 10 μm spherical resin (Sarasep Inc., Sanata Clara, CA) of 80 Å pore size and 415 m^2/g surface area. The XAD-4 resin has the

most surface area and, therefore, is easier for derivatization. The XAD-4 resin was ground and then sieved with a Model L3P sonic sifter (Allen-Bradley Co., Milwaukee, WI). The particles of size 45-58 μm (325 ~ 250 mesh) were chosen for further derivatization. The ground resin was washed with water and acetonitrile, Soxhlet-extracted overnight with methanol, ether and acetonitrile, and then dried. The 10 μm spherical resin was cleaned by the same way.

The reagents used for derivatization were all reagent grade. The solvents used for Friedel-Crafts reaction, such as tetrachloroethane, nitrobenzene, carbon disulfide, were dried by molecular sieve. The catalysts aluminum chloride, which is used for Friedel-Crafts reaction, and zinc chloride, which is used for introduction of hydroxymethyl group were dried. The reagents used in Friedal-Crafts reaction such as tertiary-butyl chloride, succinic anhydride, acetyl chloride and chloroacetonitriles, were also dried by molecular sieve.

The solutes used in high performance liquid chromatography were reagent grade. The eluents (acetonitrile and methanol) were HPLC grade. The water used was deionized by Barnstead Nanopure II (SYBRON Barnstead, Boston, MA).

Synthesis procedures and characterization

Five different functional groups, either hydrophilic or hydrophobic, were introduced into the benznene ring of PS/DVB resins by the following procedures:

(1) P-ϕ-CH_2OH (P-Polystyrene divinylbenzene resin; ϕ-benzene ring on the resin)

Add 1.2g of paraformaldehyde, 16 ml of acetic acid and 4 ml of acetic anhydried to 5.2 g of PS/DVB resin. Stir for a few minutes, then add 6.0 g of anhydrous zinc chloride and keep at 60°C overnight. Filter the resin, rinse with methanol, then heat with a solution of methanol-conc.HCl (90:10) for 1 hour. Wash the final product with methanol and dry.

The capacity of -CH_2OH group was determined as 1.3 mmol/g. The determination procedure was as follows. An exact amount of XAD-4-CH_2OH resin (about 0.1 g) was weighed. An exact volume of acetic anhydride (about 0.44 ml) and 5.00 ml pyridine was added. The mixture was stored overnight. The reaction

$$(CH_3CO)_2O + \text{XAD-4-}CH_2OH = \text{XAD-4-}CH_2OCOCH_3 + CH_3COOH$$

occurred. Then an excess of water was added to hydrolyzed the unreacted acetic anhydride.

$$(CH_3CO)_2O + H_2O = 2\ CH_3COOH$$

The resin was isolated and washed with water. The wash solutions and the isolated solution were collected together and titrated by a standard basic solution of known concentration. The volume of basic solution required was recorded. After that, an equal volume of acetic anhydride as previous and 5.00 ml pyridine were mixed with the same volume of water as previous to hydrolyze the acetic anhydride. After several hours, this solution was titrated with the same basic solution. The volume difference of basic solution used between this time and last time was calculated. The moles calculated according to this difference of basic solution equaled the moles of $-CH_2OH$ group. The determinations were performed three times and the capacity is 1.3 ± 0.1 mmol/g.

(2) P-ϕ-$COCH_2CH_2COOH$

Mix 2.5 g of XAD-4 resin with 3.6 g of succinic anhydride, 30 ml of tetrachloroethane and 15 ml of nitrobenzene. Stir for a few minutes, then add 10.7 g of anhydrous aluminum chloride and keep at 45°C for 24

hours. Pour the product into ice water. Isolate the resin, wash with acetone, methanol and water, then dry.

The capacity of -COOH group was determined to be 1.1 mmol/g by back titration. The determination procedure was as follows. An exact amount of resin was weighed and an exact volume of NaOH solution of known concentration was added. After 1 hour, the resin was isolated and washed several times. All the wash solutions and the isolated solution were collected together and titrated with standard acidic solution. The determination were repeated three times. The capacity of -COOH was determined as 1.1 ± 0.03 mmol/g.

(3) P-ϕ-$C(CH_3)_3$

Mix 4.6 g of resin, 60 ml of nitromethane, 8.0 g of tert.-butylchloride and 1.2 g of $AlCl_3$. Keep at 60°C for 24 hours. Pour into ice water, wash with acetone, methanol and water then dry. The capacity of the tert.-butyl group is difficult to determine because of lack of suitable methods, though the chromatographic behavior is greatly different from that of the underivatized resin.

(4) P-ϕ-$COCH_3$

To 5.1 g of resin add 30 ml of carbon disulfide, 9.5 g of anhydrous aluminum chloride, add 5.5 g of acetyl chloride, added dropwise. Keep at

50°C for 24 hours. Pour the product into ice water. Isolate the resin, wash with acetone, methanol and water, then dry.

The cabonyl group (-CO-) was determined by FT-IR. Comparing the FT-IR spectrum of parent XAD-4 resin and derivatized XAD-4 resin with $-COCH_3$ group, the vibration at 1690 cm^{-1} present in the derivatized resin proved the existence of carbonyl group.

The concentration of $-COCH_3$ on the resin was 1.2 mmol/g, determined by oxygen analysis.

(5) P-ϕ-CH_2CN

To 5.4 g of resin add 30 ml of tetrachloroethane and 20 ml of chloroacetonitrile. Keep at 85°C for 24 hours. Pour the product into ice water, then wash and dry as above.

The concentration of $-CH_2CN$ on the resin was 0.9 mmol/g, determined by nitrogen analysis.

(6) The reaction procedures were the same for the 10 μm spherical PS/DVB resin as that of the XAD-4 resins.

Packing materials and packing procedure

The parent XAD-4 resin, parent 10 μM spherical PS/DVB resin and their derivatives were used as packing materials for packing columns. A certain amount (about 2 g) of packing materials (Non-spherical XAD-4

resin or spherical 10 μM PS/DVB resin) was suspended in 30 ml of acetonitrile/water (50/50). The suspension was sonicated for 10 minutes and then packed into a stainless-steel column by the Shandon HPLC packing pump. The packing pressure was set at 3000 psi. The packing eluent was acetonitrile/water (50/50). Two kinds of stainless-steel column were used for packing, 250 x 2.1 mm and 100 x 4.6 mm.

RESULTS AND DISCUSSION

XAD-4 resin derivatives

Ground up and sieved XAD-4 resin was chosen for the first experiments because it is highly cross-linked and has a very high surface area (784 M^2/g). This allows easy preparation of the various derivatives. XAD-4 resins were prepared containing each of the following functional groups attached to the benzene ring: $-C(CH_3)_3$, $-CH_2OH$, $-COCH_3$, $-COCH_2CH_2CO_2H$, and $-CH_2CN$.

Hydrophilic derivatives (such as XAD-4-CH_2OH, XAD-4-$COCH_3$, etc.) are easily wet with water. The objective of this derivatization is to give hydrophobic XAD-4 resin good contact with aqueous phase and therefore good contact with the solutes in aqueous samples. The un-derivatized, the hydrophobic XAD-4 resin must be wetted with a solvent such as methanol or acetone to attain good contact with water. These solvent can be washed off and the resin becomes hydrophobic again. The derivatized resins don't show this problem since the functional groups are chemically bonded to the resin.

Hydrophobic derivatives (such as XAD-4-$C(CH_3)_3$) has the opposite effect, which makes the parent material more hydrophobic and therefore more difficult to wet. The objective of this derivatization is to make a

comparison with hydrophilically derivatized XAD-4 resin. This property was shown in Table I by the k′ values of non-polar organic compounds (such as cumene, o-dichlorobenzene etc), which are larger on the derivatized resin than that on the parent XAD-4 resin.

Liquid chromatographic behavior of XAD-4 resin and its derivatives

The parent XAD-4 resin and its derivatives were packed into stainless-steel columns of 250mm length and 2.1 mm inside diameter (250 x 2.1 mm). The eluent composed of 50% acetonitrile (volume) and 50% water was used to determine the retention times and capacity factor (k′) values of several organic compounds from non-polar to polar with these packed columns. The detection wavelength (λ) used was 270 nm. Since the chemically introduced hydrophilic and hydrophobic functional groups modified the parent resin, the liquid chromatographic behavior of these derivatized resins were different from that of the parent resin. The retention times or k′ values of the solutes were different for the different derivatized resins under the exact same chromatographic conditions. The results were shown in Table I. The k′ values in Table I are defined as

$$k' = \frac{t_r - t_0}{t_0}$$

Table I. Capacity factors (k′) of test compounds on XAD-4 resin and its derivatives. R is the ratio of k′ on the derivatized resin to k′ on the XAD-4 resin. Chromatographic conditions: 250 x 2.1 mm column; Acetonitrile-water (50-50) eluent, adjusted to pH 1.7 with HCl; 1 ml/min flow rate

Compounds	XAD-4	Derivatives of XAD-4 resin									
		$-C(CH_3)_3$		$-CH_2OH$		$-COCH_3$		$-COCH_2-CH_2COOH$		$-CH_2CN$	
	k′	k′	R	k′	R	k′	R	k′	R	k′	R
Cumene	19.3	23.2	1.20	18.4	0.80	14.2	0.74	17.7	0.92	15.6	0.81
o-Dichlorobenzene	18.7	22.0	1.18	17.0	0.91	16.9	0.91	18.7	1.00	17.1	0.91
Toluene	9.83	11.5	1.17	8.83	0.90	8.50	0.86	8.83	0.90	8.78	0.89
Anisol	6.53	7.33	1.12	5.81	0.89	6.22	0.95	5.83	0.89	5.67	0.87
Diethylphthalate	6.25	7.30	1.19	4.83	0.77	4.58	0.73	5.17	0.83	3.89	0.78
Methylbenzoate	5.25	5.83	1.11	4.84	0.92	4.61	0.88	4.15	0.90	4.78	0.91
2,4-Dinitro-fluorobenzene	4.92	4.41	0.89	4.50	0.92	4.28	0.87	4.83	0.98	4.08	0.83
Acetophenone	3.17	3.25	1.03	3.17	1.00	3.00	0.95	3.00	0.95	3.00	0.95
p-Cresol	2.17	1.92	0.88	2.25	1.04	2.69	1.24	2.08	0.96	2.11	0.97
Phenol	1.50	1.42	0.94	1.67	1.11	1.94	1.30	1.67	1.11	1.61	1.07

in which t_r is the retention time of solute and t_0 was the dead time. The t_0 is determined by Na_2SO_4 as a solute at λ = 220 nm. Na_2SO_4 is an ionic solute and is not retained on the resin. An easy comparison of K′ for the parent and daughter resins can be made by the use of an R value, which is the ratio of k′ of the daughter to k′ of the parent. Since all the comparisons in our experiments were made under the same chromatographic conditions, the results of different derivatization were showed.

Compared to the parent XAD-4 resin, the k′ values of non-polar solutes (o-dichlorobenzene, toluene, etc) decreased on the polar derivatized resins (such as XAD-4 resins with $-CH_2OH$, $-COCH_3$, $-CH_2CN$ groups), while they increased on non-polar tertiary-butyl resin derivative.

Compared to the parent XAD-4 resin, the k′ values of polar solutes (p-cresol, phenol, etc) increased on polar resin derivatives, while they decreased on the non-polar tertiary-butyl resin derivative.

Solutes intermediate between these extremes had somewhat shorter or longer retention times on the derivative resins, depending on the polarity of the solutes and polarity of derivatized resins.

Capacity factors for some of the analytes were measured as a function of the percentage of acetonitrile in the eluent. A change in

acetonitrile percentage of the mobile phase caused a change of retention time of solutes. A better separation factor at low percentage of acetonitrile in the eluent was seen. The plots of log k′ vs percentage of acetonitrile for toluene, methylbenzoate, p-cresol, and phenol on parent XAD-4 resin and XAD-4-CH_2OH resin clearly showed the effects of derivatization of resins (see Figures 1, 2). The plots were not quite linear. They curved upward at low acetonitrile percentage. Comparing the plots on XAD-4 resin (Figure 1) and that on XAD-4-CH_2OH resin (Figure 2), the curves of two relative non-polar solutes, toluene and methylbenzoate, moved down a little while the two relative polar solutes, p-cresol and phenol, moved up a little on the more polar XAD-4-CH_2OH resin. The same plots of log k′ vs. acetonitrile percentage were prepared by SiC_{18} column under the same conditions and similar results were seen. The plots were not quite linear and curved upward at low acetonitrile percentages (see Figure 3). The slopes of those solutes were similar to that on the XAD-4 resins.

Methanol-water is a weaker eluent than acetonitrile-water for eluting organic solutes. Approximately 75% methanol was required to obtain the similar k′ values to 50% acetonitrile in our experiments. An eluent of 75% methanol and 25% water was used to determine the retention time and k′ values for these same solutes. The results in

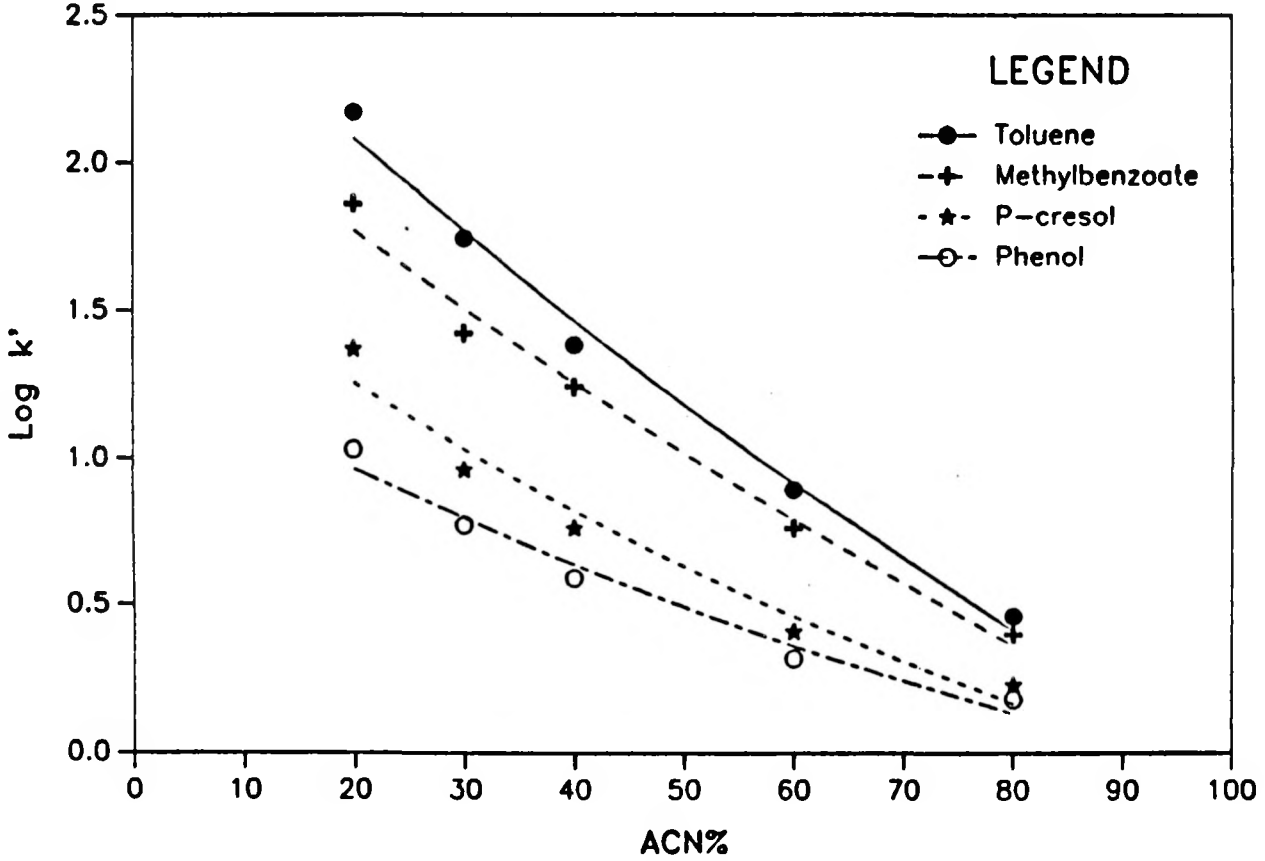

Figure 1. Log k' on un-derivatized XAD-4 resin column as a function of the percentage of acetonitrile (ACN) in the eluent.

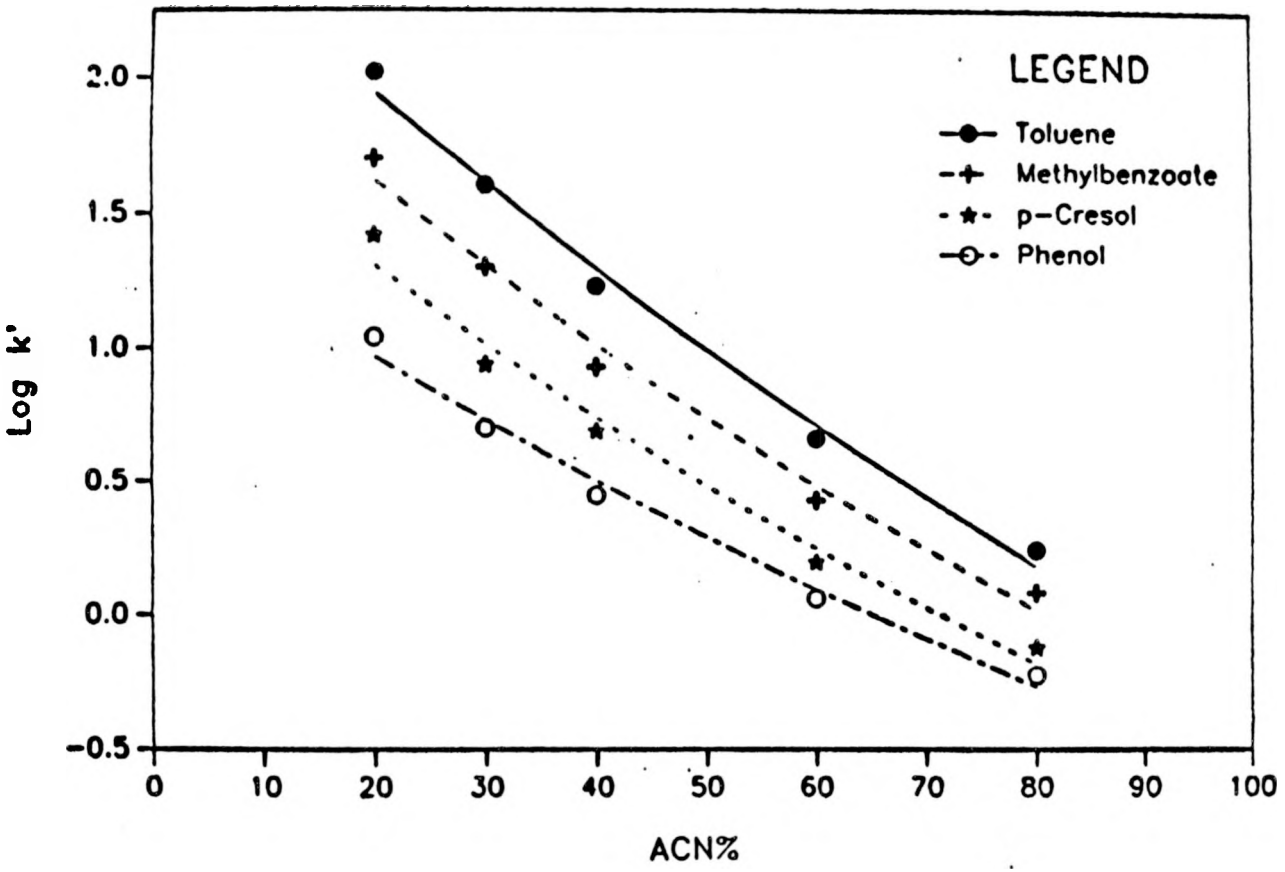

Figure 2. Log k' on XAD-4-H_2OH resin column as a function of the percentage of acetonitrile in the eluent

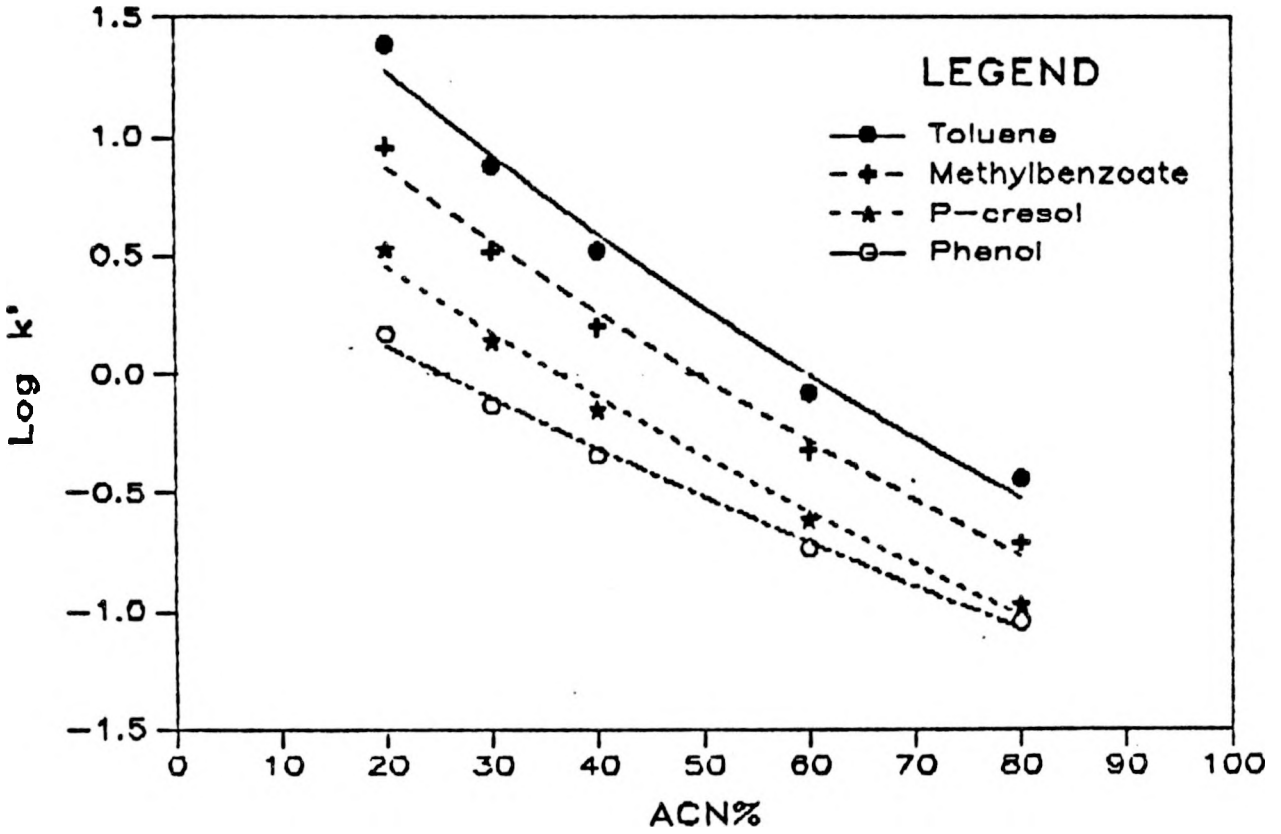

Figure 3. Log k′ on SiC_{18} column as a function of the percentage of acetonitrile in the eluent

Table II, in which three packing materials were used, showed some different elution orders obtained going from XAD-4 to XAD-4-CH_2OH resin. The general tendency of k′ values were similar to that with acetonitrile-water eluent.

The determinations of k′ values with SiC_{18} column (Vydac column, 250 x 4.6 mm, 5 μM spherical particle, Anspec Company, Inc., Ann Arbor, MI) and SiCN column (Spheresorb CN column, 250 x 4.6 mm, 5 μM spherical particle, Anspec Company, Inc., Ann Arbor, MI) were also made with eluent of 50% acetonitrile and 50% water (see Table III). These results were compared with those of XAD-4 resins. Some interesting phenomena were noticed. XAD-4 columns have much larger retention than the SiC_{18} column. For example, k′ for cumene is 19.3 on XAD-4 column and 3.51 on SiC_{18} column. The volume of SiC_{18} column is 4.80 times of that of the XAD-4 column. Therefore, the ratio of k′ for cumene on a column of similar volume is

$$\frac{k'(\text{on XAD-4})}{k'(\text{on } SiC_{18})} = \frac{19.3 \times 4.80}{3.51} = 26.4$$

if we compare XAD-4-$C(CH_3)_3$ resin, the ratio for cumene is still larger,

$$\frac{k'(\text{on XAD-4-}C(CH_3)_3)}{k'(\text{on } SiC_{18})} = \frac{23.2 \times 4.80}{3.51} = 31.7$$

Table II. Capacity factors (k') of test compounds on derivatized and underivatized resins using methanol-water elutent (75-25). Other conditions were the same as Table I

Compounds	XAD-4	Derivatives of XAD-4 resin			
		$-CH_2OH$		$-C(CH_3)_3$	
	k'	k'	R	k'	R
Cumene	22.31	13.84	0.62	28.28	1.27
o-Dichlorobenzene	21.75	15.57	0.72	21.75	1.00
Toluene	15.12	11.20	0.74	15.93	1.05
Anisol	12.76	9.83	0.77	13.41	1.05
Diethylphthalate	10.91	6.93	0.64	12.33	1.13
Methylbenzoate	10.39	7.88	0.76	11.69	1.12
2,4-Dinitro-fluorobenzene	7.56	7.80	1.03	7.50	0.99
Acetophenone	5.76	4.41	0.77	6.19	1.07
p-Cresol	2.19	2.38	1.09	2.09	0.95
Phenol	0.93	1.41	1.14	1.21	0.98

Table III. Comparison of capacity factors (k′) on C_{18} silica, and cyano silica columns. The size of the columns is 250 x 4.6 mm. The eluent was acetonitrile-water (50-50)

Compounds	C_{18} Silica	Cyano Silica	
	k′	k′	R
Cumene	3.51	1.33	0.38
o-Dichlorobenzene	2.40	1.17	0.49
Toluene	1.57	0.97	0.62
Anisol	0.92	0.80	0.87
Diethylphthalate	1.13	0.89	0.79
Methylbenzoate	0.82	0.75	0.91
2,4-Dinitro-fluorobenzene	0.69	0.90	1.30
Acetophenone	0.53	0.64	1.21
p-Cresol	0.40	0.56	1.40
Phenol	0.29	0.49	2.45

The main reason for this difference may be the large surface area of the XAD-4 (784 M^2/g). Also the XAD-4 resin is ground up and sized, so that the particles are not spherical.

k′ values going from SiC_{18} to SiCN were greatly reduced. For example, the ratio for cumene is 0.38, for toluene is 0.62. The ratio of range of k′ values (cumene:phenol) is 12.1 on SiC_{18} and only 2.71 on SiCN. For these reasons, cyanosilica is seldom used for reverse-phase liquid chromatography.

k′ values going from XAD-4 to XAD-4-CH_2CN show much less difference. For example, the ratio for cumene is 0.81, for toluene is 0.89. The ratio of range of k′ values (cumene:phenol) is 12.87 on XAD-4 resin, 16.34 on XAD-4-$C(CH_3)_3$ resin, 11.02 on XAD-4-CH_2OH resin, 7.32 on XAD-4-$COCH_3$ resin, and 9.69 on XAD-4-CH_2CN. Generally speaking, the larger the polarity of derivatized resin, the smaller the ratio of range of k′ values. This is similar to the tendency of Si based column (SiC_{18} and SiCN).

Derivatives of 10 μM spherical PS/DVB resins for liquid chromatography

The XAD-4 resin and its derivatives have clearly showed the effects of derivatized functional groups on the resins. However, the fairly large and somewhat irregular particle size limit the separation ability.

Usually, spherical particle shape and small particle size improves the efficiency of the column. For better separation and practical application, a spherical PS/DVB resin of 10 μM size and 425 M^2/g surface area was derivatized and packed into columns. The derivatizations and packing procedures were the same as those for XAD-4 resins. The parent 10 μM spherical PS/DVB resin and its derivatives with the non-polar group ($-C(CH_3)_3$) and the polar group ($-COCH_3$) were packed into columns. The same solutes were tested to see the effects of the functional groups on spherical and smaller size resins with two mobile phases, 50% acetonitrile-50% water and 75% methanol-25% water. The results were shown in Table IV and Table V.

The k′ values were similar to that of the XAD-4 resin and its derivatives, but not identical. This is due to the differences in surface area, particle size and the amounts of resins in the columns. The relative k′ values (R) were also similar. The peaks were narrower and more symmetric on the 10 μM spherical resin.

The general trends of k′ values of various solutes was similar to that observed with XAD-4 resin and its derivatives. The non-polar solutes had smaller k′ values on the 10 μM spherical resin with polar functional group ($-COCH_3$) but larger on the resin with non-polar functional group ($-C(CH_3)_3$). The opposite was true for the polar

Table IV. Capacity factors (k') of test compounds on derivatized and un-derivatized spherical, polymeric resin columns using acetonitrile-water (50-50) eluent. Other conditions were the same as Table I

Compounds	10 μm PS/DVB	Derivatives of 10 μm PS/DVB resin			
		$-COCH_3$		$-C(CH_3)_3$	
	k'	k'	R	k'	R
Cumene	22.98	11.25	0.49	28.71	1.25
o-Dichlorobenzene	19.18	12.38	0.65	24.29	1.27
Toluene	10.83	6.61	0.61	12.84	1.19
Anisol	6.93	4.50	0.65	7.73	1.11
Diethylphthalate	6.33	2.97	0.47	6.72	1.06
Methylbenzoate	5.48	3.57	0.65	6.12	1.12
2,4-Dinitro-fluorobenzene	4.56	4.08	0.89	4.29	0.94
Aetophenone	3.06	2.05	0.67	3.25	1.06
p-Cresol	1.70	1.96	1.15	1.65	0.87
3-Nitrophenol	1.59	2.07	1.30	1.50	0.94
4-Nitrophenol	1.43	1.97	1.37	1.27	0.89
Phenol	1.23	1.41	1.15	1.11	0.90

Table V. Capacity factors (k') of test compounds on derivatized and un-derivatized resin columns using methanol-water (75-25) eluent. Other conditions were the same as Table I

Compounds	10 μM PS/DVB	Derivatives of 10 μM PS/DVB resin			
		$-COCH_3$		$-C(CH_3)_3$	
	k'	k'	R	k'	R
Cumene	35.08	15.06	0.43	40.22	1.15
o-Dichlorobenzene	25.96	17.01	0.65	28.89	1.12
Toluene	15.02	11.96	0.79	16.51	1.10
Anisol	14.34	10.70	0.75	15.89	1.11
Diethylphthalate	14.28	5.68	0.39	16.20	1.13
Methylbenzoate	12.78	7.92	0.62	13.92	1.09
2,4-Dinitro-fluorobenzene	10.10	8.16	0.81	10.04	0.99
Acetophenone	6.57	4.54	0.69	7.09	1.08
p-Cresol	2.21	2.91	1.32	1.95	0.88
Phenol	1.27	2.11	1.66	1.14	0.89

solutes. When 75% methanol-25% water was used as eluent, some difference in elution order were observed (see Table V).

Several test compounds were separated on each of the columns (PS/DVB, PS/DVB-$COCH_3$, PS/DVB-$C(CH_3)_3$) using a very fast chart speed so that column efficiency and peak shape could be observed. The theoretical plate numbers of the polymeric resin columns were rather low (~ 10,000 plates/m), but this would be expected because the average resin diameter was around 10 μm rather than 5 μm as used in silica-based columns.

Peak asymmetry factors for the four test compounds are measured and calculated, which are given in Table VI for the three polymeric resins and for a commercial 5 μm silica C_{18} column. The tertiary-butyl column shows the most tailing. The parent polymeric resin column shows essentially no tailing for three of the four test compounds. The acetyl column also compares favorably with the silica column.

In Table VII the capacity factors (k′) for 20 aromatic compounds are compared to the k′ of benzene on each of the three polymeric columns. In this way the effect of a single aromatic substituent can be measured. Several of the test compounds have more than one substituent, but the effects are not additive nor would they be expected to be additive [15].

The k′ values relative to benzene in Table VII are listed in decreasing order. Compared to the un-derivatized resin the relative k′

Table VI. Asymmetry factors of four test compounds on different resin coluns

Resin	Peak assymetry factor			
	Toluene	Anisol	Acetophenone	4-Nitrophenol
PS-C(CH_3)$_3$	1.97	1.43	1.57	1.86
PS	0.96	0.95	0.96	1.33
PS-$COCH_3$	1.46	1.48	1.44	1.50
silica C18	1.38	1.50	1.53	1.77

Table VII see next page.

Table VIII. Relative k' values ($k'_X/k'_{benezene}$) for halogenated benzenes (C_6H_5X) on three resin columns

X	Relative k'			k'(t-Bu) / k'(Unsub.)	k'($COCH_3$) / k'(Unsub.)
	t-Bu	Unsubst.	-$COCH_3$		
I	3.22	2.71	2.87	1.19	1.06
Br	2.74	2.36	2.68	1.16	1.14
Cl	2.09	1.82	2.11	1.15	1.16
F	0.95	0.91	1.07	1.04	1.18

Table VII. Capacity factors (k′) and capacity factors relative to k′ (benzene) derivatives on three polymeric resin columns. R is the ratio of k′ on the derivatized resin compared to the un-derivatized resin. The eluent was 50-50 acetonitrile-water, acidified with HCl

Compounds	PS/DVB		PS/DVB-$COCH_3$			PS/DVB-$C(CH_3)_3$		
	k′	k'_x/k'_b	k′	k'_x/k_b	R	k′	k'_x/k'_b	R
Benzene	6.59		4.82			7.21		
Biphenyl	41.28	6.27	28.46	5.90	0.69	50.63	7.02	1.23
Cumene	22.98	3.49	11.25	2.33	0.49	28.71	3.98	1.25
o-Dichlorobenzene	19.18	2.91	12.38	2.57	0.65	24.29	3.37	1.27
Iodobenzene	17.89	2.71	13.83	2.87	0.77	23.19	3.22	1.30
Bromobenzene	15.53	2.36	12.93	2.68	0.83	19.78	2.74	1.27
Chlorobenzene	11.98	1.82	10.18	2.11	0.85	15.05	2.09	1.26
Toluene	10.83	1.64	6.61	1.37	0.61	12.84	1.78	1.19
Anisol	6.93	1.05	4.50	0.93	0.65	7.73	1.07	1.12
Diethylphthalate	6.33	0.96	2.97	0.62	0.47	6.72	0.94	1.06
Fluorobenzene	5.99	0.91	5.17	1.07	0.86	6.85	0.95	1.14
Methylbenzoate	5.48	0.79	3.57	0.74	0.65	6.12	0.85	1.12
Nitrobenzene	5.24	0.80	4.70	0.98	0.90	5.99	0.83	1.14
2,4-DiNitrofluorobenzene	4.56	0.69	4.08	0.85	0.89	4.29	0.60	0.94
Acetophenone	3.06	0.46	2.05	0.43	0.67	3.25	0.45	1.06
p-cresol	1.70	0.26	1.96	0.41	1.15	1.65	0.23	0.97
3-Nitrophenol	1.59	0.24	2.07	0.43	1.30	1.50	0.21	0.94
4-Nitrophenol	1.43	0.22	1.97	0.41	1.38	1.27	0.18	0.89
phenol	1.23	0.19	1.42	0.29	1.15	1.11	0.15	0.90
Benzoic Acid	0.78	0.12	2.55	0.53	3.27	0.75	0.10	0.96
Benzyl Alcohol	0.76	0.12	0.93	0.19	1.22	0.71	0.10	0.93

values on the tertiary-butyl resin are higher for the more hydrophobic test compounds and lower for the polar compounds. However, the order of the relative k′ values is the same for tertiary-butyl resin and un-derivatized resin, with the minor exception of diethylphthalate and fluorobenzene.

The relative k′ values of non-polar test compounds are lower on the resin derivatized with a polar acetyl group than on the un-derivatized resin. Exactly the opposite is true for the polar test compounds. More significantly, there are numerous changes in the elution order of the various test compounds on the acetyl resin.

It is interesting to compare the relative k′ values for the halogenated benzenes on the three resin columns (Table VIII). the relative k′ values for the four simple halobenzenes are higher on both the butyl and acetyl columns than on the un-derivatized resin column. However, the ratio of relative k′(butyl) to relative k′(un-substituted) increases in the order F, Cl, Br, I. The ratio of relative k′ (acetyl) to relative k′(un-substituted) decreases in the same order.

Following the treatment of Sadek and Carr [15], log k′ was plotted against the number of carbon atoms in the side chain of alkyl benzenes and of alkyl phenols. Excellent linear plots were obtained for both classes of compounds (Figure 4 and Table IX). The slopes show the effect

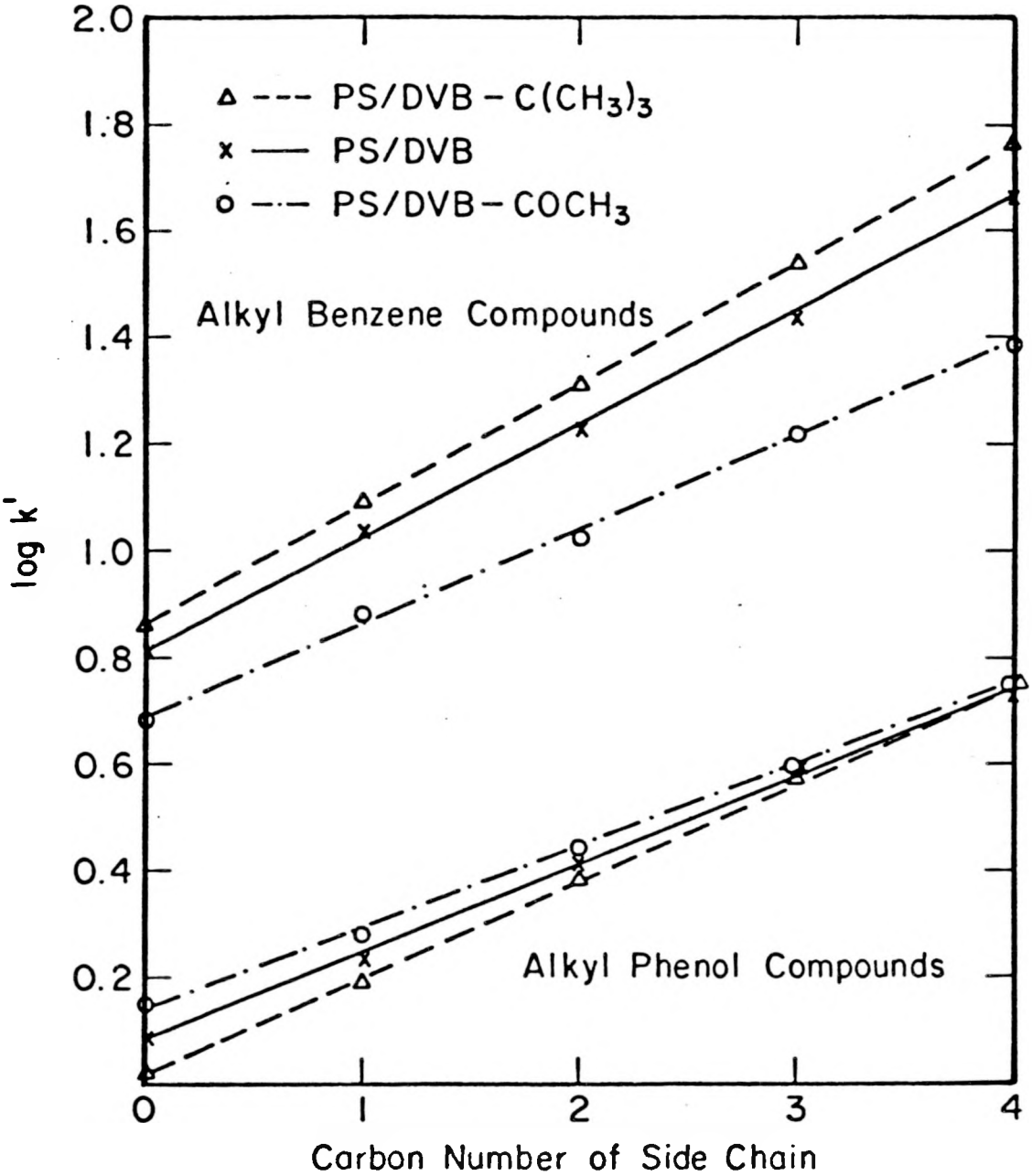

Figure 4. Log k′ of homologous compounds on different resin vs. their carbon number of side chain

Table IX. Linear regression data for plots of log k′ against the number of carbon atoms in the R group for C_6H_5R and for HOC_6H_4R

For C_6H_5R, where R=0, 1, 2, 3, 4

Resin	Slope	y-intercept	corr.(v)
PS-$C(CH_3)_3$	0.23	0.86	0.9997
PS	0.21	0.81	0.9995
PS-$COCH_3$	0.18	0.69	0.9995

For HOC_6H_5R, where R=0, 1, 2, 3, 4.

Resin	Slope	y-intercept	corr.(v)
PS-$C(CH_3)_3$	0.185	0.016	0.9996
PS	0.165	0.081	0.9995
PS-$COCH_3$	0.15	0.140	0.9992

of a single -CH_2 group on the increased retention of a test compound. The higher slopes on the butyl resin indicate a greater affinity for a -CH_2 group than on the un-derivatized phenyl resin. The lower slopes on the more polar acetyl resin indicated a lower affinity for a -CH_2 group. The slopes of each of the three resins were higher for the alkyl benzenes than for the alkyl phenol.

The intercepts for the alkyl benzenes (Table IX) are in the order of decreasing polarity of the resins acetyl < un-derivatized < tertial-butyl. The intercepts of the more polar alkyl phenols are in exactly the opposite order: tertiary-butyl < un-derivatized < acetyl.

Bonded-phase silica with cyano, amino, and other polar groups have, of course, been available for a number of years. Columns containing these packings are used for normal-phase chromatography but seldom for reversed-phase liquid chromatography. A comparison of the retention times of various analytes on cyano silica and cyano polymeric columns using a typical reverse-phase eluent (acetonitrile-water) showed significantly longer retention times for all of the analytes on the polymeric cyano columns (Table X). In Table X, the capacity factors of these test compounds on a column packed with cyano polymeric resins are compared with values obtained under identical conditions using un-derivatized resin. Polar test compounds are retained a little longer and

Table X. Comparison of capacity factors (k') on cyano silica to C_{18} and of cyanomethyl polymeric to polymeric resin. The eluent was acetonitrile-water (50-50)

Compound	Silica columns			Polystyrene columns		
	$k'C_{18}$	$k'(CN)$	$\frac{k'(CN)}{k'(C18)}$	$k'(PS)$	$k'(CN)$	$\frac{k'(CN)}{k'(PS)}$
Cumene	3.51	1.33	0.38	23.0	20.6	0.90
o-Dichlorobenzene	2.40	1.17	0.49	19.2	16.4	0.85
Toluene	1.57	0.97	0.62	10.8	10.5	0.97
Anisol	0.92	0.80	0.87	6.93	6.32	0.91
Diethylphthalate	1.13	0.89	0.79	6.33	5.08	0.80
Methylbenzoate	0.82	0.75	0.91	5.48	4.79	0.87
2,4-Dinitrofluoro-benzene	0.69	0.90	1.30	4.56	4.41	0.97
Acetophenone	0.53	0.64	1.21	3.06	3.08	1.01
p-Cresol	0.40	0.56	1.40	1.70	1.74	1.02
Phenol	0.29	0.49	2.45	1.23	1.35	1.10

non-polar compounds are less retained on the cyanomethyl column, although the differences are generally less than those between un-derivatized and acetyl resin column.

It is interesting to compare the effects of introducing a cyanomethyl group into a polystyrene resin with changing from a C_{18}- to cyanoalkyl bonded-phase silica resin column. The results in Table X show a much lower retention for cumene and o-dichlorobenzene on cyano silica than on C_{18} silica. This is probably the reason that cyano silica columns have generally been less used for reversed-phase LC than for normal-phase chromatography.

It is logical to expect a large change in capacity factors with cyanoalkyl silica column compared to an alkyl silica column because of the drastic difference in polarity of the bonded groups. The difference in polarity between a phenyl group and a cyanomethyl phenyl group in polystyrene resins is less drastic. Accordingly, smaller differences in capacity factors of test compounds would be expected.

Separations of several mixtures by 10 μM spherical PS/DVB resins

Separations of several mixtures were compared on a parent 10 μM spherical resin and an acetyl-derivatized one. The column size was 100 mm in length with a 4.6 mm inside diameter for practical applications.

Although columns packed with 10 μm resins are no longer state-of-the-art, the separations are still good enough to be of practical value and to show clearly the effects of derivatization.

Examination of the data in Table 3 showed better separation factors for several pairs of the analytes on the derivatized column than on the parent polymeric resin column. The separation factors α calculated ($\alpha = k'_1/k'_2$, where k'_1 and k'_2 are the capacity factors of separated pair analytes) for three pairs of analytes (anisol and diethylphthalate, 3-nitrophenol and phenol, 4-nitrophenol and phenol) were 1.09, 1.29 and 1.16 on the parent polymeric resin, compared with 1.52, 1.47 and 1.40 on the acetyl-derivatized resin. Figure 5 and figure 6 showed the instances in which significantly better separations were obtained on the column containing the acetyl-derivatized resin under the same experimental conditions.

In some cases, the elution order of test compounds on the derivatized resin differs from the parent polymeric resin. Figure 7.A and B showed examples where the elution orders of methylbenzoate and diethylphthalate, o-dichlorobenzene and cumene were reversed on those two columns. Also the separations are better.

For non-polar analytes, the retention times greatly decreased on the acetyl-derivatized polymeric resin compared to the parent resin.

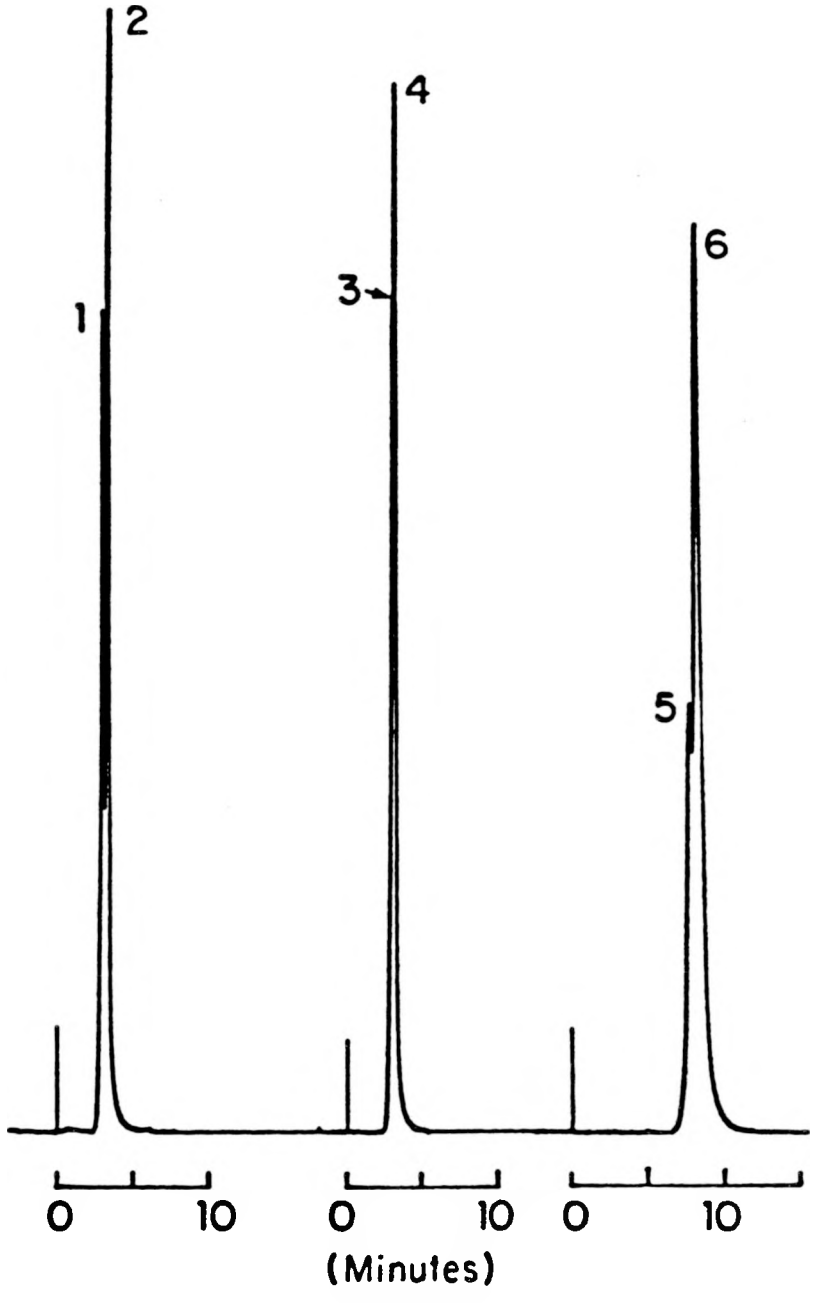

Figure 5. Chromatographic separations on PS/DVB resin column (100 x 4.6 mm), using 60% acetonitrile (pH=1.7) eluent. 1=phenol, 2=3-nitrophenol, 3=phenol, 4=4-nitrophenol, 5=diethylphthalate, 6=anisol

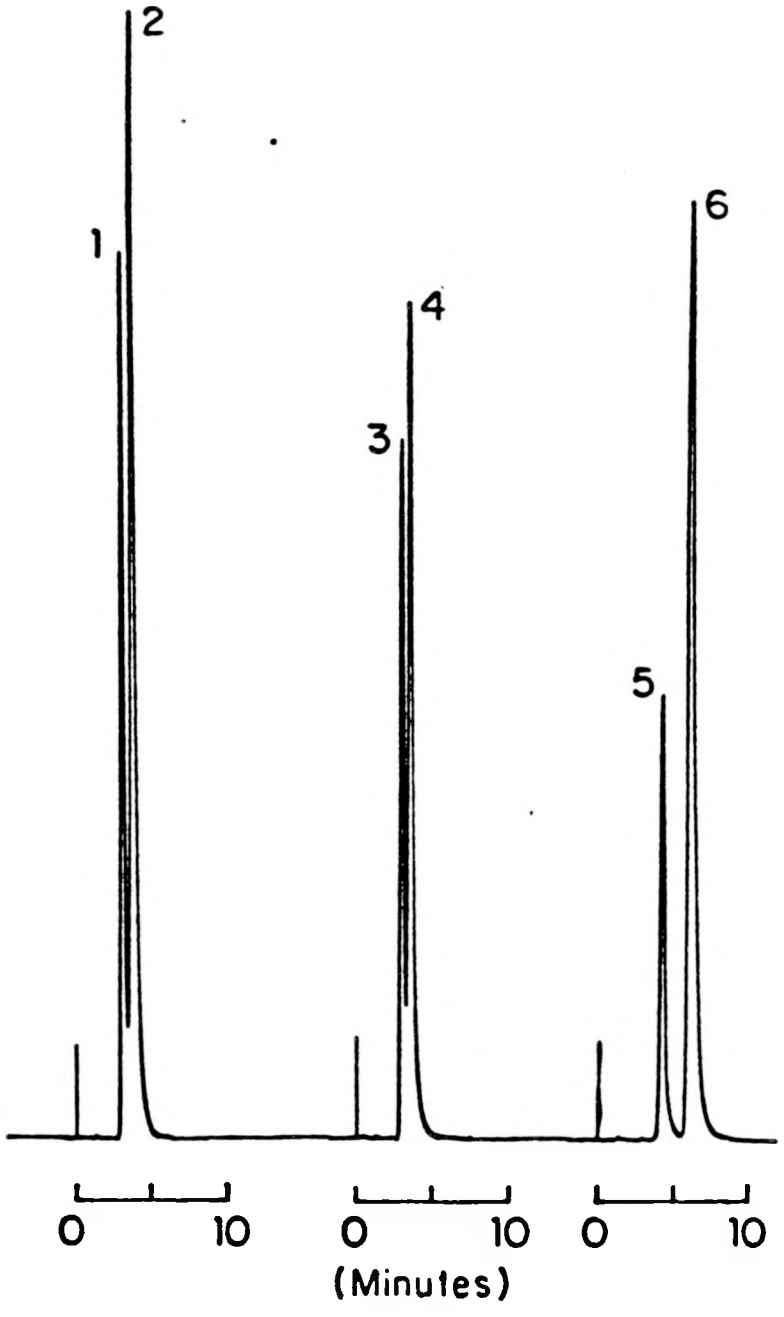

Figure 6. Chromatographic separations on an acetyl derivatized PS/DVB column. Conditions are the same as Fig.5

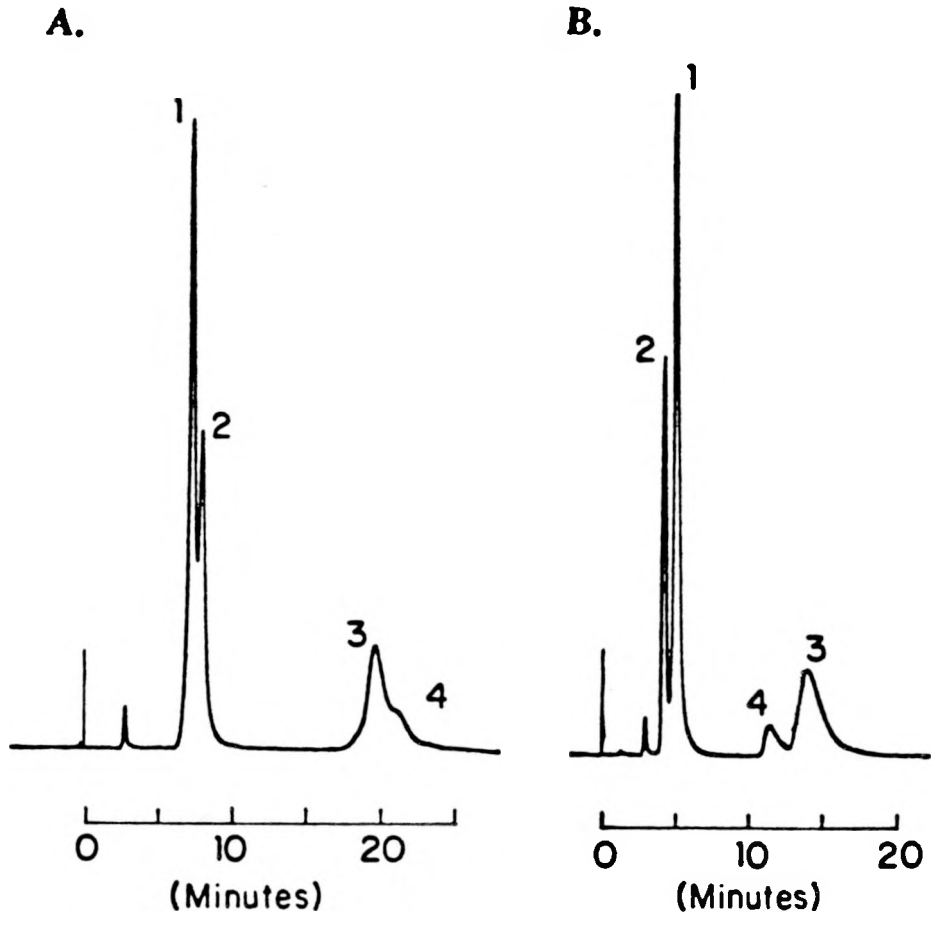

Figure 7. A. Chromatographic separations on PS/DVB column. 1=methylbanzoat, 2=dieyhlphthalate, 3=o-dichlorobenzene, 4=cumene. Conditions are the same as Fig.5. B. Chromatographic separations on acetyl PS/DVB column

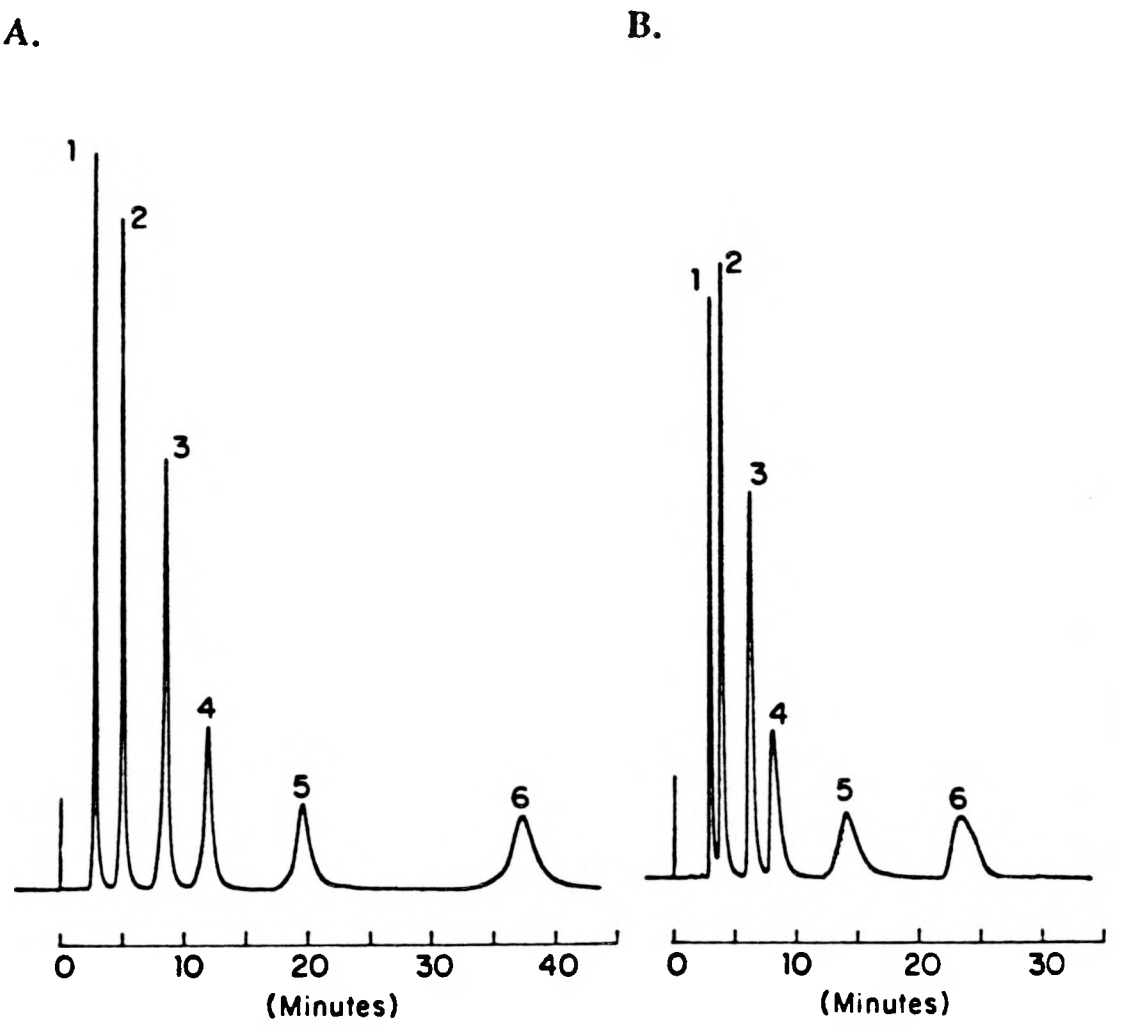

Figure 8. A. Chromatographic separations on PS/DVB coloum. 1=phenol, 2=acetophenone, 3=anisol, 4=toluene, 5=o-dichlorobenzene, 6=biphenyl. Conditions are the same as Fig.5. B. Chromatographic separation on aceyl PS/DVB column

Figure 8.A and B showed the difference in retention times for six solutes (phenol, acetophenone, anisol, toluene, o-dichlorobenzene and biphenyl) on these two columns under the same chromatographic conditions. The retention time on acetyl-derivatized polymeric resin was much smaller.

CONCLUSION

Even with the limited number of test compounds studied, it is apparent that functional groups introduced into PS/DVB resins have an appreciable effect on the retention times and k′ values obtained in reverse-phase liquid chromatography. The retention times of all the compounds are distinctly different on the derivatized columns, and in several cases the relative retention times (R) of many of the test compounds are significantly different on the derivatized columns. Therefore, derivatized polymeric columns offer an additional selectivity parameter (e.g. interaction of analyte with the resin) over reverse-phase chromatography with C_{18} or C_8 silica bonded resins in which the selectivity effects are determined primarily by interaction between the analytes and the solvents in the mobile phase.

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SECTION II. CHEMICALLY MODIFIED RESINS FOR SOLID PHASE EXTRACTION

INTRODUCTION

Solid-phase extraction (SPE) is now widely used for preconcentration and cleanup of analytical samples, purification of various chemicals, and for large-scale applications such as removal of toxic or valuable substances from a variety of predominately aqueous solution. Typical applications include methods for determination of trace amounts of pesticides (1,2), analysis of trace organic contaminants in water (3,4), analysis of industrial wastewater (5), determination of azaarenes in water (6), evaluation of solid phase extraction effect of porous polymers (8), isolation of organic compounds in ground water (7), sampling of priority pollutants in wastewaters (9), collection and concentration of environmental samples (10), and pretreatment of urine samples (11, 12).

For analytical purposes, SPE is usually performed using a small column or cartridge containing an appropriate resin. Membranes loaded with appropriate resins have also been used for SPE. Following uptake of extractable solutes from a predominantly aqueous sample, it is common practice to elute the adsorbed materials from the resin by a small amount of an organic solvent.

Chemically-bonded silica, usually with a C_{18} or C_8 organic group, is by far the most commonly used material for SPE. Minor use has been made

of porous polystyrene or other polymeric resins in SPE. However, these materials have several shortcomings for use in SPE.

1) They are hydrophobic and therefore do not make good surface contact with predominately aqueous solutions.

2) Pretreatment of the SPE materials with an activating solvent (such as methanol, acetone, or acetonitrile) must be used to obtain better surface contact with the aqueous solution being extracted. However, the activating solvent can be gradually leached out of the resin, thereby causing the extraction to become ineffective. This is particularly true if the SPE column inadvertently goes dry, causing air to be sucked into the column.

3) Many types of organic compounds are incompletely extracted from predominately aqueous solutions. This is especially true with chemically bonded silica resins.

Some other resins were tried to solve the problem. An anion exchange resin was used to abstract phenols and the results were good, though the procedure was a little tedious (13). A methyacrylate polymeric resin with an amino group attached was tried and the recoveries were improved (14). The reason for low recovering of phenolic or other polar compounds by SiC_{18} or polystyrene-divinylbenzene is probably the hydrophobicity of those sorbents, which makes the polar compounds

difficult to contact the resin surfaces. When the sample solution passes through the SPE column, only part of the polar organic compounds are absorbed while the rest just passes through.

We tried to solve this problem by modifying the resin to increase its hydrophilicity non-ionically while still keeping its extraction ability. The polystyrene-divinylbenzene resin is the most suitable choice for us since it is easy to modify the benzene ring in its structure. Also it is more chemically stable than SiC_{18} under strong acidic or basic conditions.

In the present work, new chemically and non-ionically modified polystyrene-divinylbenzene resins were prepared, which are hydrophilic and easily wetted by water while still keeping its extraction ability. Various organic compounds were tested by solid-phase extraction using these modified resins. The recovery results were compared with those using SiC_{18} and un-derivatized polystyrene-divinylbenzene resin. The new resins showed superior recoveries for test compounds in solid-phase extraction.

EXPERIMENTAL

Preparation of modified resins

Several chemically modified resins were prepared from porous, crosslinked polystyrene materials. The Amberchrome 161 resin (Rohm and Haas Co., Philadelphia. PA) is spherical with average particle size of approximately 50 μm and a surface area of about 720 m^2/g. The Sarasep resin (Sarasep, Inc., Santa Clara, CA) has an average particle size of approximately 10 μm and a surface area of about 415 m^2/g. The resins were cleaned by Soxhlet-extraction with methanol, ether and acetonitrile over night, dried and underwent following synthetic procedures.

Amberchrome 161-$COCH_3$ derivative: To 5.1 g of Amberchrome 161 resin add 30 ml of carbon disulfide, 9.5 g of anhydrous aluminum chloride, and 5.5 g of acetyl chloride dropwise. Keep at 50° for 24 hours. Pour the product into ice water, isolate the resin, wash with acetone, methanol and water, then dry. The presence of the carbonyl group was proved by a strong band at 1690 cm^{-1} on the spectrum obtained by FT-IR. The concentration of -$COCH_3$ was determined as 1.2 mmol/g by elemental oxygen analysis.

Amberchrome 161-CH_2OH derivative: Add 1.2 g of paraformaldehyde, 16 ml of acetic acid, and 4 ml of acetic anhydride to 5.2 g of Amberchrome

161 resin. Stir for a few minutes, then add 6.0 g of anhydrous zinc chloride and keep at 60°C overnight. Filter the resin, rinse with methanol, then heat with a solution of 90% methanol-10% concentrated HCL for 1 hour. Wash the final product with methanol and dry. The concentration of $-CH_2OH$ on the resin was 1.3 mmol/g. This was determined by a standard acetylation procedure using acetic anhydride in pyridine as the reagent.

Apparatus and components

Solid-phase extractions were performed on small columns (SPE columns) packed with 100 mg of resins. Several different resins were tested and compared in this experiment. The commercial SiC_{18} SPE column with resin of 40μm particle size was obtained from Alltech Associates, Inc. (Deerfield, IL). The inside diameter and the length of the SPE column are 6 and 55 mm. 100mg of derivatized and un-derivatized Amberchrom 161 resin, and derivatized and un-derivatized Sarasep resinsn were packed into empty columns, which were obtained from P.J.Cobert Associates, Inc. (St. Louis, MO), of the same size of SiC_{18} SPE column. The length of silica resin in the column is about 10 mm. The length of Amberchrom resins is about 12 mm. The length of Sarasep resins is about 8 mm because of the smaller particle size. The SPE column was connected

to a home-made reservoir by an adaptor (P. J. Cobert Associates, Inc.) for solid-phase extraction. The flow rate of the sample solution from the reservoir through the SPE column was controlled by a compressed air pressure on the top of the reservoir. The apparatus used is shown in Figure 1.

The organic compounds eluted from SPE column by ethyl acetate were collected and then analyzed by a HP 5880A gas chromatography instrument with a flame ionization detector, a HP 5880A series level 4 integrator, and a HP7673A automatic sampler (Hewlett-Packard Co., Avondale, PA). The gas chromatography columns used were J. & W. fused silica capillary megabore DB-5 and minibore DB-1 (Alltech Associates, Inc.).

A Bruker FT-IR 98 instrument (USA Bruker Instruments Inc., San Jose, CA) was used for structure determination.

Reagents and chemicals

The reagents and solvents used for the derivatization reactions were reagent grade and were dried by molecular sieves. The chemicals used were all reagent grade or analytical grade. Laboratory distilled water was further deionized by Barnstead Nanopure II system (Sybron Barnstead, Boston, MA).

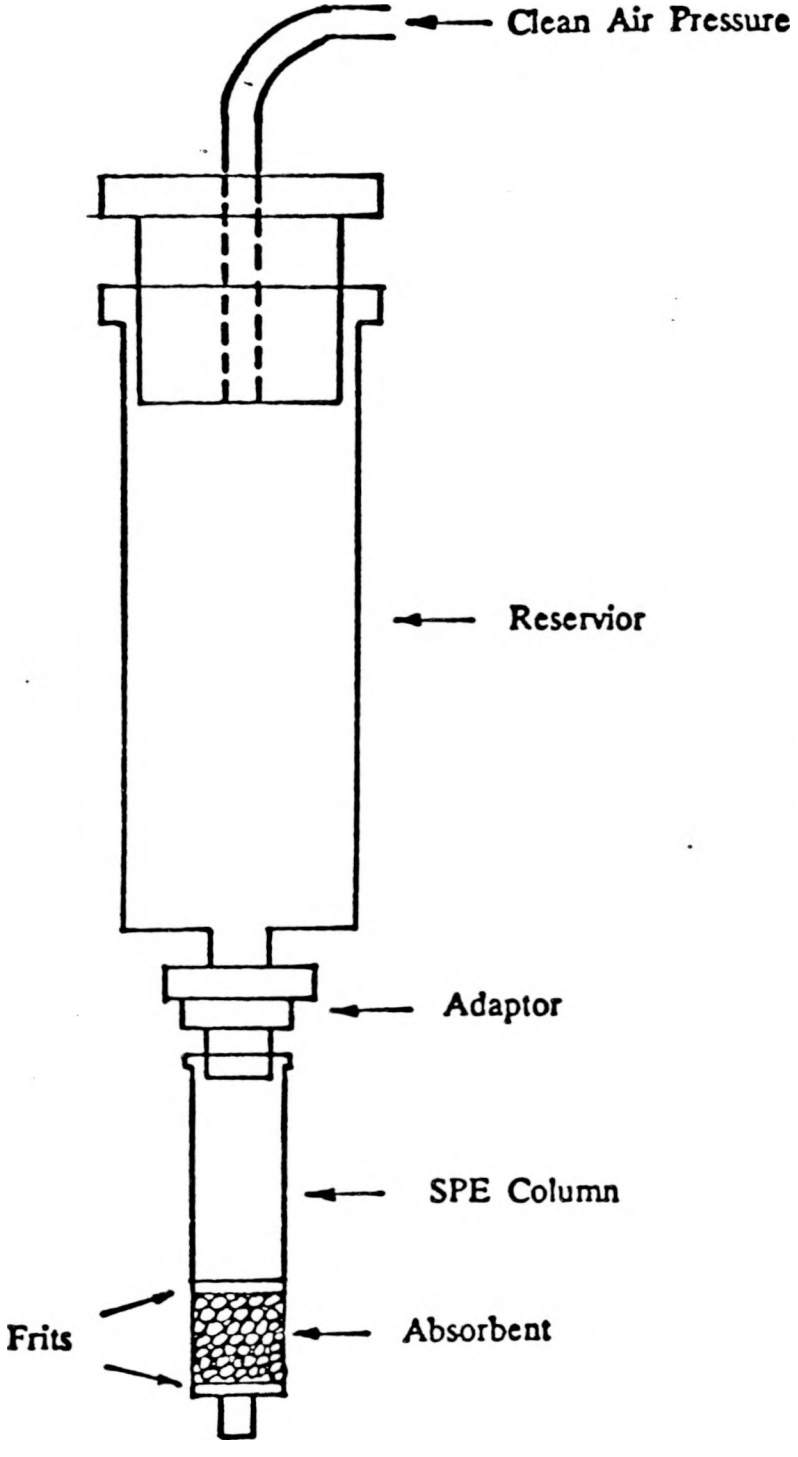


Figure 1. Solid phase extraction apparatus

Loading methods

SPE columns were cleaned with methanol, ethyl acetate and acetonitrile, and then dried. Two loading methods were compared in this experiment: wet column loading and dry column loading. In wet column loading, which can also be called preconditioned loading, the SPE column was wetted with methanol and then the sample solution was forced through the SPE column. In dry column loading, the SPE column was totally dried before the sample solution was forced through the SPE column. The results of dry and wet column loading are quite different and will be discussed later in this section.

Sorption procedures

In the solid phase extraction experiments, the sample organic compounds to be tested were added into 20 ml of pure water. The concentration of sample compounds were in the range of 1 to 5 ppm. The sample solution was then added into a reservoir and the air pressure on the top of the reservoir was adjusted to make the flow rate of the sample solution to be 5ml/min when passing through the SPE column. After the sample solution was passed through the SPE column, the column was washed with 1 ml of pure water. Until the last drop of water was passed

through, air was blown through the column for a few seconds to ensure that all the water was removed.

Elution procedure

The SPE column was then taken off from the reservoir and 1 ml of ethyl acetate was added. It was connected to the reservoir again and the air pressure was adjusted to make the eluate to pass through the column at a flow rate of 1 ml/min. The retained organic compounds were eluted and the eluate was collected in a small vessel until the last drop of ethyl acetate passed through the column. An internal standard of exact amount was added into the vial, which was capped immediately, mixed well and ready for gas chromatographic determination.

Gas chromatography separation and quantitation

The vessel was directly used in the automatic injection sampler on the gas chromatograph. The injection volume was 1 μl. The carrier gas was helium, flow rate was 15 ml/min, and the split ratio was 1:40. Temperature programming was used to optimize the separations. An exact aliquot of the loading standard samples dissolved in 1 ml of ethyl acetate with the same amount of internal standard was also made, mixed well in another vessel and injected immediately into gas chromatograph

under the exact same conditions. The relative peak areas were used for quantitation and the percentage recoveries of the organic compounds were calculated from relative peak areas directly. Recoveries were calculated as average values and represented more than two different analysis.

RESULTS AND DISCUSSION

Selection of resins

In section I various hydrophilic functional groups were chemically attached to porous, crosslinked polystyrene resins. These included $-CH_2OH$, $-COCH_3$, and $-CH_2CN$ groups that were attached directly to the benzene rings of the polymer. The ability of these resins to retain various organic compounds was surveyed by packing a column with each resin and determining the retention times of the test compounds using 50% acetonitrile - 50% water as the eluent. The retention times of non-polar test compounds were somewhat shorter on the columns with hydrophilic substituents. While the retention times of more polar test compounds such as p-cresol and phenol were significantly longer on the columns with hydrophilic substituents. These results follow rules that are well established for silica resins. However, comparison of retention times for test compounds on a cyano silica resin column and a cyanomethyl organic resin column shows much greater retention on the modified organic column.

It is well known that the logarithm of adjusted retention time, or of capacity factor (k′), is inversely proportional to the volume percentage of organic solvent in an eluent containing an organic solvent

plus water. Extrapolation of the data in Figure 1 of section I to zero percent acetonitrile suggests that organic resins containing a hydrophilic group would strongly retain all of the test compounds from predominantly aqueous solution, as in SPE. Of the resins studied, the acetyl resin showed the best ability to retain phenol. The resin with a hydroxy-methyl group also retained phenols more strongly than the un-derivatized resin.

Various organic compounds, which include aromatic, phenolic, multi-hydroxy phenolic, aliphatic compounds, were tested under dry and wet loading conditions with SiC_{18}, un-derivatized and derivatized Amberchrom 161 resins. The results were shown from Table I through Table VI.

Dry column loading (non-preconditioning loading) and wet column loading (preconditioning loading)

SiC_{18} resin is one of the most widely used sorbents in solid phase extraction by analysts for preconcentration, sample preparation and purification. One basic and very important stage of solid phase extraction method is the preconditioning of the SPE column. One purpose of preconditioning is to make the sorbent resin compatible with the loading solvent, which is water in most cases, and analytes of interest, which are trace organic compounds. Since the octedecyl group is very hydrophobic, SiC_{18} resin had much lower recovery efficiency without

preconditioning. Tables II, IV and VI showed the recoveries under dry column loading condition which were much lower than that under wet column loading condition in Tables I, III and V. It was also shown that there was a big difference on recovery efficiency by un-derivatized Amberchrome 161 between dry and wet column loading conditions. Although SiC_{18} and polystyrene-divinylbenzene are good absorbents for extraction, their hydrophobic properties prevent the organic compounds from achieving good contact with resin surface, especially for polar compounds. However, the derivatized Amberchrom 161-$COCH_3$ resin showed much higher recovery efficiency and a much smaller difference between dry and wet column loading condition. For some compounds (see table I), the recoveries of dry column loading were as good as that of wet column loading. This is because that the chemically bonded -$COCH_3$ group is polar, making the resin compatible to water and organic compounds without preconditioning. A preconditioning solvent on SiC_{18} or un-derivatized polystyrene-divinylbenzene resin could evaporate or be washed off, while the -$COCH_3$ group is permanently bonded to the resin. This is an important advantage of the acetyl derivatized PS/DVB resin, allowing it to be used in cases where preconditioning is impossible or inconvenient.

Table I. The recoveries (%) of phenolic and aromatic compounds by different sorbents under wet SPE column loading condition

Compound	SiC_{18}	Amberchrome	Amberchrome $-CH_2OH$	Amberchrome $-COCH_3$
Phenol	6.3	90.7	94.0	99.7
p-Cresol	16.2	91.1	98.1	100.9
p-Ethylphenol	65.9	96.0	98.5	101.2
2-Nitrophenol	44.7	92.9	94.9	96.0
3-Nitrophenol	<5	81.0	84.9	92.5
4-Nitrophenol	<5	87.0	85.7	86.6
2,4-Dimethylphenol	70.8	94.7	97.3	100.2
4-t-Butylphenol	82.9	88.2	95.5	99.5
Anisol	77.9	90.6	94.1	98.1
Aniline	9.1	94.0	96.1	99.5
Benzylalcohol	10.2	91.5	98.2	99.2
Nitrobenzene	53.6	92.4	96.3	99.9
2,4-Dinitroflorobenzene	43.9	83.0	96.0	98.4
o-Hydroxyacetophenone	88.1	84.9	94.7	96.0
Isopentylbenzoate	83.8	71.8	89.2	95.2
Diethylphthalate	90.2	87.2	95.5	100.1
Average	47.1	88.6	94.3	97.6

Table II. The recoveries (%) of multi-hydroxy phenolic compounds under wet SPE column loading condition

Compound	SiC_{18}	Amberchrome	Amberchrome $-CH_2OH$	Amberchrome $-COCH_3$
Catechol	0	71.9	88.6	75.1
Resorcinol	0	61.3	88.2	97.4
o-Methylresorcinal	0	82.9	97.2	98.6
Hydroquinone	0	25.7	71.8	86.9
Methylhydroquinone	0	77.7	97.7	99.0
Phloroglucinol	0	0	23.8	55.5

Table III. The recoveries (%) of phenolic and aromatic compounds by different sorbents under wet SPE column loading condition

Compound	SiC_{18}	Amberchrome	Amberchrome -CH2OH	Amberchrome -COCH3
Pentanone	20.4	91.0	94.1	97.2
Octanone	90.8	88.5	96.8	98.3
Hexylacetate	85.4	70.0	87.8	92.1
Mesityl oxide	58.1	76.2	97.2	97.5
Ethyl crotonate	76.3	73.7	96.5	97.0
Hexenyl acetate	71.5	58.5	86.1	85.0
3-Picoline	41.4	91.6	96.3	97.0
3-Ethylpyridine	75.7	95.3	97.3	96.5
Average	65.0	80.6	94.0	95.1

Table IV. The recoveries (%) of phenolic and aromatic compounds by different sorbents under dry SPE column loading condition

Compound	SiC_{18}	Amberchrome	Amberchrome $-CH_2OH$	Amberchrome $-COCH_3$
Phenol	<3	3.1	75.2	92.8
p-Cresol	4.4	12.4	87.9	93.7
p-Ethylphenol	15.3	37.1	96.8	98.8
2-Nitrophenol	17.2	47.0	95.8	95.6
3-Nitrophenol	<5	<5	74.9	72.5
4-Nitrophenol	<5	<5	77.3	84.8
2,4-Dimethylphenol	20.7	41.6	96.0	98.1
4-t-Butylphenol	49.0	50.0	89.6	95.0
Anisol	58.4	55.9	94.8	96.2
Aniline	<5	25.8	89.5	96.1
Benzylalcohol	<5	17.4	85.1	98.5
Nitrobenzene	27.4	50.5	96.0	97.2
2,4-Dinitroflourobenzene	3.7	23.2	92.0	94.3
o-Hydroxyacetophenone	66.7	54.3	94.0	94.2
Isopentylbenzoate	60.2	73.0	83.8	84.7
Diethylphthalate	70.2	58.0	84.2	89.5
Average	26.0	34.9	88.3	92.6

Table V. The recoveries (%) of multi-hydroxy phenolic compounds under dry SPE column loading condition

Compound	SiC_{18}	Amberchrome	Amberchrome $-CH_2OH$	Amberchrome $-COCH_3$
Catechol	0	0	8.6	29.9
Resorcinol	0	0	0	94.5
o-Methylresorcinal	0	0	15.6	95.6
Hydroquinone	0	0	0	80.9
Methylhydroquinone	0	0	4.9	93.8
Phloroglucinol	0	0	0	42.0

Table VI. The recoveries (%) of phenolic and aromatic compounds by different sorbents under dry SPE column loading condition

Compound	SiC_{18}	Amberchrome	Amberchrome -CH2OH	Amberchrome -COCH3
Pentanon	7.6	60.5	91.5	92.6
Octanone	70.1	84.5	91.4	94.6
Hexylacetate	78.1	64.8	75.8	83.0
Mesityl oxide	37.6	23.8	97.0	97.2
Ethyl crotonate	54.7	28.2	98.0	97.1
Hexenyl acetate	65.4	26.9	77.7	78.6
3-Picoline	31.0	48.2	88.4	91.1
3-Ethylpyridine	59.8	70.2	92.6	95.5
Average	50.5	50.9	89.1	91.2

Recovery study

The solid-phase extraction recoveries of various compounds of four different resins were shown in Tables I through VI under both dry and wet column loading conditions. Among the four resins, the derivatized Amberchrome 161-$COCH_3$ showed the highest efficiency under both dry and wet column loading conditions.

For the aromatic and phenolic compounds tested in this experiment (see Tables I and II), the average recoveries by Amberchrome 161-$COCH_3$ were 97.6% under wet column loading condition and 92.6% under dry loading condition. The difference of the averages under wet and dry column loading condition was 5.0%. The average recoveries by Amberchrome 161-CH_2OH were 94.3% under wet column loading condition and 88.3% under dry loading condition. The difference of the averages under wet and dry column loading condition was 6.0%. The average recoveries by un-derivatized Amberchrome were 88.6% under wet column loading condition and 34.3% under dry column loading condition. The difference of these averages was 54.3%. The recoveries by SiC_{18} were the lowest. They were 46.5% under wet column loading condition and 24.5% under dry column loading condition. The difference of these averages was 21.9%.

Several things might have caused these results. First, the polar functional groups -$COCH_3$ and -CH_2OH make the resin more easily wetted

with water and, therefore, in more contact with the compounds dissolved in it. Second, the structure of polystyrene-divinylbenzene is more compatible to aromatic compounds because of the π electron system of the benzene ring. Third, hydrogen bonding between the functional group and some compounds might help the extraction. The results also showed that preconditioning is critical for solid phase extraction of aromatic and phenolic compounds by both SiC_{18} and un-derivatized Amberchrome 161 absorbent, while it has a smaller effect on Amberchrome 161-$COCH_3$ and -CH_2OH sorbents due to the presence of these polar functional groups.

The solid-phase extraction of multi-hydroxy phenolic compounds showed an even greater effect on the derivatized Amberchrome 161-$COCH_3$ resin compared with the other resins, as seen in tables III and IV. The SiC_{18} sorbent showed almost zero recoveries for those compounds under both wet and dry column loading conditions. The un-derivatized Amberchrome 161 got almost zero recoveries under dry column loading conditions and 63.5% average recovery (except phloroglucinol) under wet column loading conditions. The phloroglucinol was not extracted under both loading conditions. The derivatized Amberchrome 161-$COCH_3$ showed 78.9% average recovery under dry column loading condition and 91.4% average recovery under wet column loading condition, with the exception of phloroglucinol, which were 42.0% and 55.5% recovered under dry and wet

column loading conditions. The recoveries of phloroglucinol were lower than other phenolic compounds with two hydroxy group. This might be because that phloroglucinol was partially ionized greatly and therefore not easily extracted.

Some aliphatic and pyridine compounds were chosen for solid phase extraction tests. The recoveries with derivatized Amberchrome 161-$COCH_3$ were still the best under both dry and wet column loading conditions, which were 91.2% and 95.1% respectively. The recoveries of un-derivatized Amberchrome 161 and SiC_{18} were similar under dry column loading conditions, which were 50.5% and 50.9%. Under wet column loading condition, the recoveries of un-derivatized Amberchrome 161 were somewhat higher than that of SiC_{18}, which were 80.6% and 65.0% respectively. The aliphatic compounds are more compatible to SiC_{18} resin than aromatic compounds. However, the important role of -$COCH_3$ functional group was still obviously seen for those aliphatic and pyridine compounds, especially pyridine compounds which have a structure similar to aromatic compounds.

The effect of derivatization was further tested by comparing resins from a different source. A highly crosslinked, spherical polystyrene resin was obtained from Sarasep, Inc. This resin has an average particle

size of 10 μm and a surface area of 415 m^2/g, which is appreciably lower than the area of the Amberchrom 161 resins.

The results in Table VII show excellent recoveries for the four phenolic compounds on the acetyl resin with significantly lower recoveries for two of the phenols on the un-derivatized Sarasep resin. Incomplete recoveries were obtained for the four multihydroxy phenols on the acetyl resin, but the recoveries on the un-derivatized resin ranged from zero to 8 per cent.

Several organic compounds (0.7 to 7 ppm in water) were selected to compare recoveries of the acetyl derivatives of the Amberchrome 161 and Sarasep resins. The recoveries of these two resins were quite similar (Table VIII). Again the recoveries with the silica C_{18} were much lower.

Chromatographic study of solid phase extraction on various stationary phase

The study of solid-phase extraction is actually similar to the study of liquid chromatographic behavior of solutes in mobile phases and stationary phases. Of course, there are some major differences between solid phase extraction and liquid chromatography. In liquid chromatography, the solutes are expected to be moved along with the column by the mobile phase and separated. In solid-phase extraction,

Table VII. Recoveries (%) of phenols and multi-hydroxy phenols using 10 μm PS/DVB resins under wet SPE column loading condition

Compound	PS/DVB	PS/DVB-$COCH_3$
Phenol	23.8	97.6
p-Cresol	91.4	100.0
2,4-Dinitro fluorobenzene	100.4	100.3
3-Nitrophenol	81.5	101.7
Catechol	0.7	39.7
Hydroquinone	0	13.8
2-Methyl resorcinol	8.0	79.9
Methyl hydroquinone	4.4	58.9

Table VIII. Comparison of different acetyl resins for SPE. Amberchrome 161 approximately 720 M^2/g surface area and 50 μm average particle size. Sarasep approximately 415 M^2/g surface area and 10 μm average particle size

Compound	SiC_{18}	Amberchrome 161	Sarasep
Benzene	41.2	85.7	84.9
Toluene	75.2	84.9	87.9
Indene	72.9	84.0	85.0
Naphthalene	67.9	78.9	79.4
Anthracene	58.5	66.6	70.5
Phenol	6.9	97.8	97.3
p-Cresol	33.3	99.8	98.9
Dibutyl phthalate	65.8	84.3	88.3
Average	52.7	85.3	86.5

however, the solutes are ideally expected to be retained on the stationary phase without moving when the solution (aqueous mobile phase) passes through in the first step (loading step), and then totally eluted by another mobile phase (e.g. organic solvent) in a small volume in the second step (elution step). Actually even in the first step, some or even most solutes are still moving on the stationary phase. The moving speed depends on the affinity of this solute to the stationary and mobile phase.

A mini-column packed with a small amount of resin is usually used for solid phase extraction. Whether the solute is retained on the column or leached out primarily depends on the moving speed (which could be anything from 0 to several mm/min) of the solute on the stationary phase and the length of the stationary phase. It also depends on the flow rate of the mobile phase and other factors. If the moving speed of a solute on the stationary is very slow, a short length of stationary phase is enough to retain the solute. If the moving speed is fast, a longer one is needed to retain the solute before the total mobile phase (solution) is passed through. But the length of mini-column cannot be too long or one of two elution problems is caused: incomplete elution or too much solvent needed. The usual commercial mini-column contains about 100 mg of resin, which is about 1 cm in length and 0.5 cm inside diameter.

It would be very helpful to understand solid-phase extraction if the solutes could be monitored as the solution passes through the stationary phase. For this purpose, an HPLC system with a very small column was used, which was 4.0 x 4.0 mm (see Figure 2). The detector was UV-VIS and wave length was set at 270 nm. Water was used as a loading mobile phase. A mixture of water and acetonitrile with different percentage were used as eluting mobile phases. Several different resins (3-10 μm PS/DVB, 3-10 μm PS/DVB-$COCH_3$, 10 μm PS/DVB-$COCH_3$, and 12 μm SiC_{18}) were chosen as stationary phases and tested with five compounds. Since phenolic compounds are the most difficult ones to be extracted, homologous phenolic compounds were chosen: phenol, p-cresol and p-ethylphenol. Also a non-polar compound, toluene, and an intermediate compound, acetophenone, were chosen.

These solutes were first injected into the LC system with pure water as mobile phase (loading mobile phase), the pump was then stopped two seconds after the injection. The mobile phase was then changed to a mixture of water plus some percentage of acetonitrile (eluting mobile phase). The organics were then eluted by this eluent.

The k′ of these compounds on each stationary phase under mobile phases with different percentage of acetonitrile were determined. The results are shown on Tables IX-XII. The log k′ of each compound was

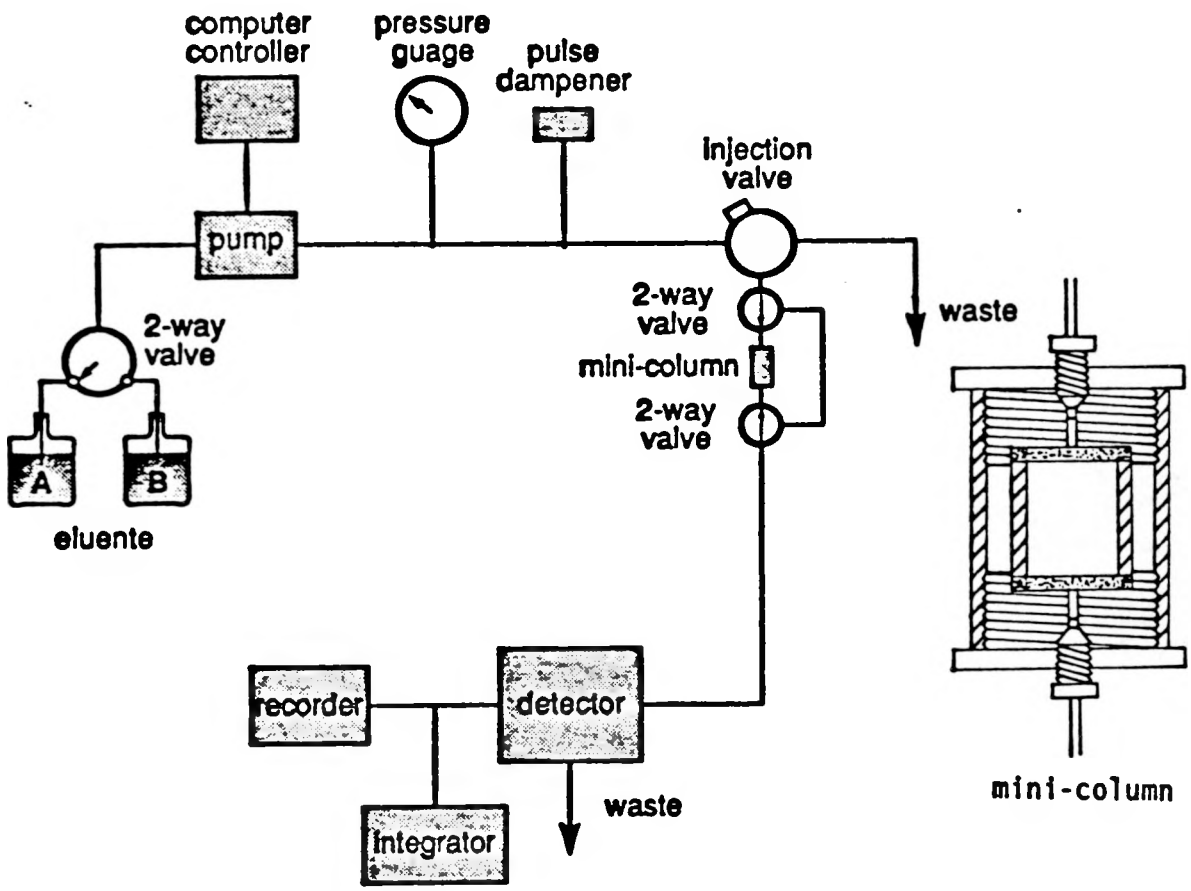


Figure 2. HPLC separation system

Table IX. k′ of various compounds on a PS/DVB resin (3-10μm) column (4mm L x 4mm ID)

ACN%	0%	2.5%	5.0%	10.0%	20.0%	30.0%
Phenol	119.5	66.9	45.3	26.3	14.3	10.1
p-Cresol	799.0	288.4	181.1	83.2	25.8	14.3
p-Ethyl-phenol	2472.7	1030.6	501.6	195.0	53.2	22.2
Aceto-phenone	*	1114.8	396.4	154.9	41.1	19.5
Toluene	*	*	1214.8	588.8	173.8	61.6

* the compound is not eluted.

Table X. K′ of various compounds on a PS/DVB-$COCH_3$ resin (3-10μm) column (4mm L x 4mm ID)

ACN%	0%	2.5%	5.0%	10.0%	20.0%	30.0%
Phenol	515.8	136.4	69.0	36.3	17.8	12.2
p-Cresol	*	589.0	229.1	83.2	28.2	14.1
p-Ethyl-phenol	*	1641.1	597.4	204.2	47.4	18.5
Aceto-phenone	*	877.9	269.2	125.9	42.2	17.4
Toluene	*	1388.5	751.6	338.8	115.8	60.6

* The compound is not eluted.

Table XI. k′ of various compounds on a SiC_{18} resin (12μm) column (4mm L x 4mm ID)

ACN%	0%	2.5%	5.0%	7.5%	10.0%	20.0%	30.0%
Phenol	23.7	17.4	14.8	11.7	10.6	6.9	5.8
p-Cresol	73.2	52.2	39.5	30.6	23.2	11.6	7.4
p-Ethyl-phenol	227.4	145.8	104.3	80.1	56.9	21.1	9.5
Aceto-phenone	150.1	70.1	46.9	33.2	25.7	13.2	10.2
Toluene	254.8	200.1	182.2	154.3	124.8	59.0	25.3

Table XII. k' of various compounds on a PS/DVB-$COCH_3$ resin column (4mm L x 4mm ID)

ACN%	0%	2.5%	5.0%	7.5%	10.0%	20.0%	30.0%
Phenol	2156.9	310.6	146.4	118.5	77.9	30.1	1.3
p-Cresol	*	1451.6	432.7	218.8	180.1	43.7	1.3
p-Ethyl-phenol	*	4114.8	1399.0	776.2	518.4	116.9	1.6
Aceto-phenone	*	1988.5	595.8	331.1	252.7	63.7	27.4
Toluene	*	2867.4	1504.3	1109.5	652.7	209.7	80.6

* The compound is not eluted.

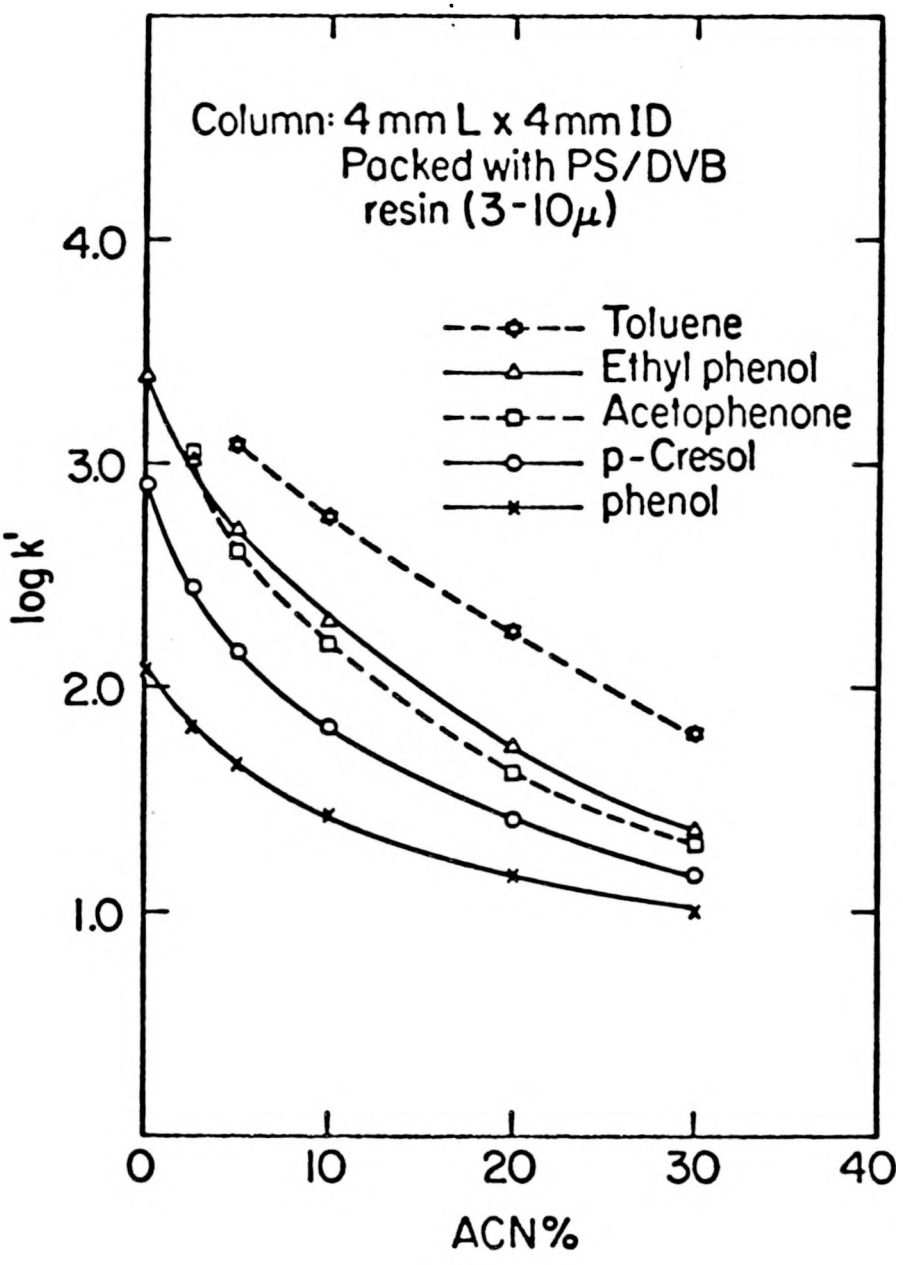


Figure 3. Log k' on PS/DVB (3-10 μm) vs. percentage of acetonitrile (see Table IX)

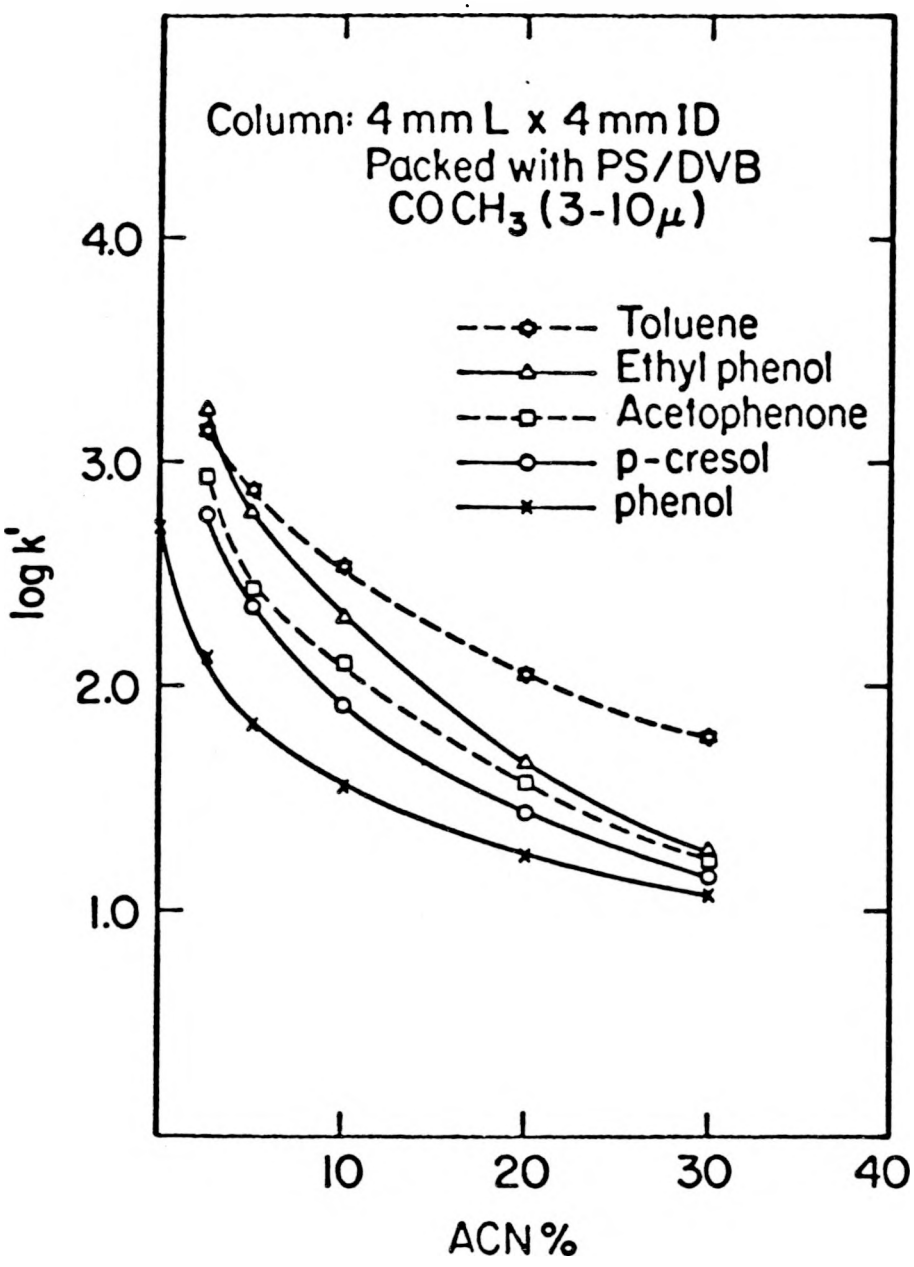


Figure 4. Log k′ on PS/DVB-$COCH_3$ (3-10 μm) vs. percentage of acetonitrile (see Table X)

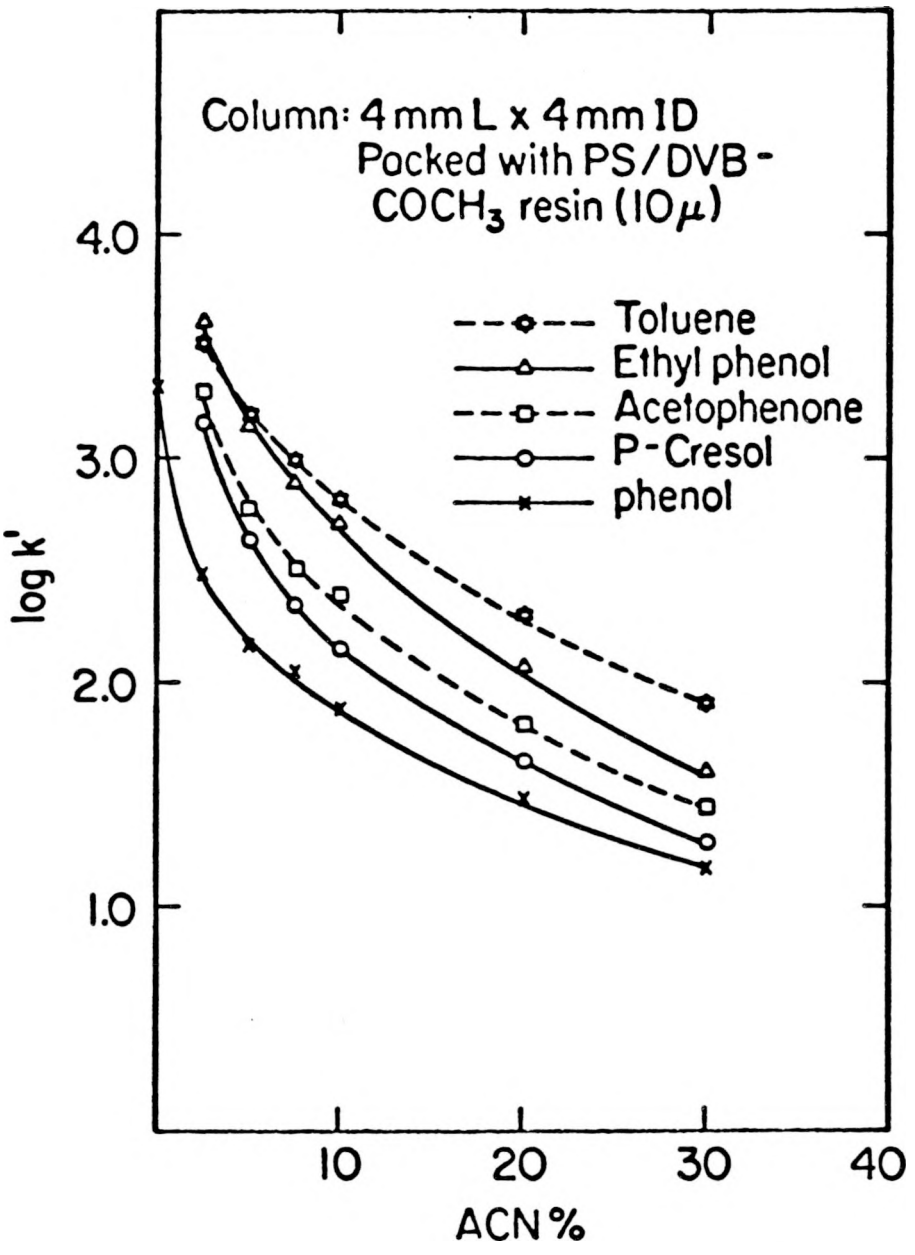


Figure 5. Log k' on SiC_{18} (12 μm) vs. percentage of acetonitrile (see Table XI)

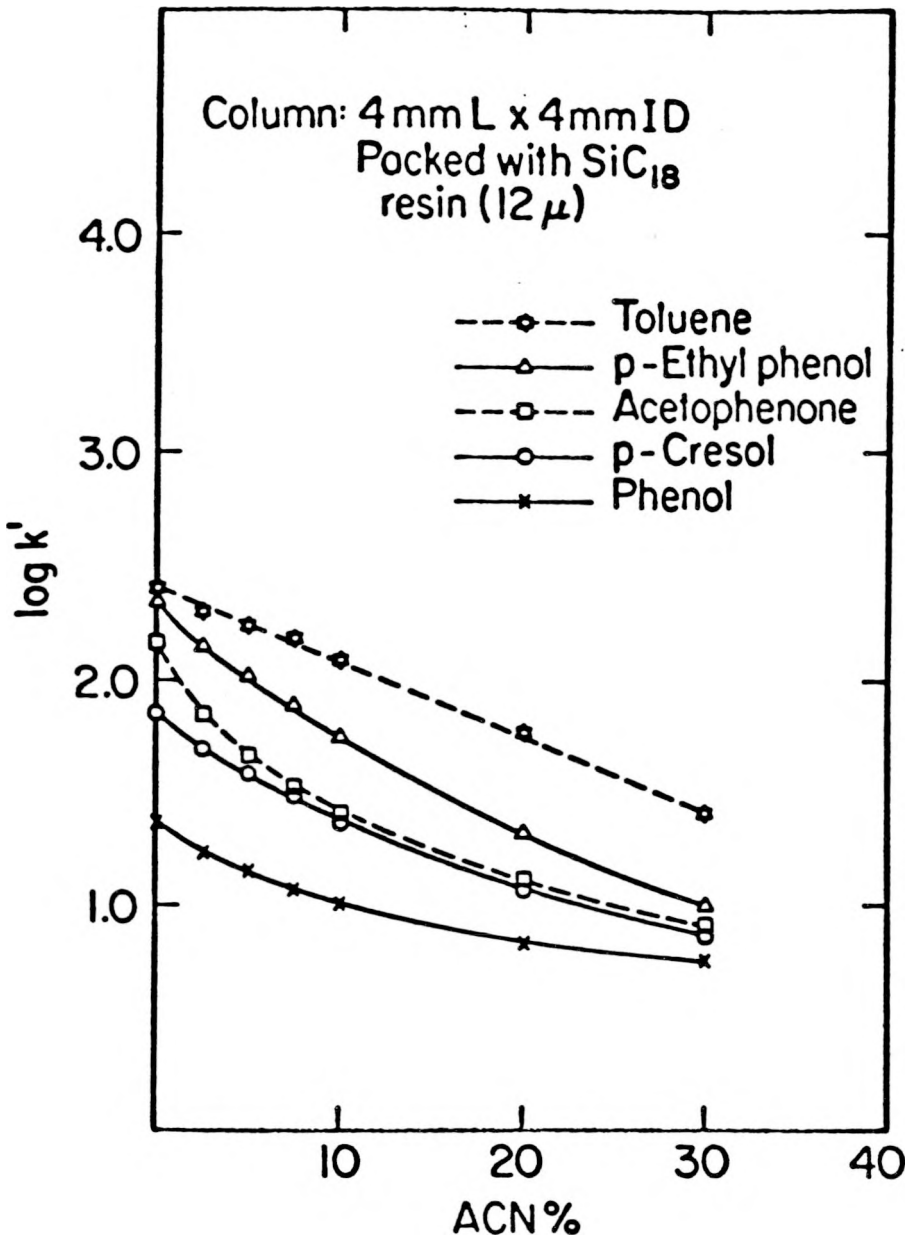


Figure 6. Log k′ on PS/DVB-$COCH_3$ (10 μm, larger surface area) vs. percentage of acetonitrile (see Table XII)

plotted against percentage of acetonitrile on each stationary phase (see Figures 3-6).

Comparing the retentions of various compounds on 3-10 μm PS/DVB and PS/DVB-$COCH_3$ resins, the acetyl group plays a key role in the retention of solutes (see Tables IX and X). For the polar compounds phenol, p-cresol and p-ethylphenol, k's were higher on PS/DVB-$COCH_3$ than on PS/DVB, especially at low percentages of acetonitrile. At 0% of acetonitrile (pure water), p-cresol and p-ethylphenol were not eluted from mini-column of PS/DVB-$COCH_3$ resin at all, however, they were eluted from PS/DVB resin. The k' of phenol on PS/DVB-$COCH_3$ was much higher than that on PS/DVB. This explained why the PS/DVB-$COCH_3$ achieved such a high extraction recovery for polar compounds. For the non-polar compound toluene and less polar compound acetophenone, k's were higher on PS/DVB than that on PS/DVB-$COCH_3$. However, at 0% of acetonitrile (pure water), none of them was eluted from PS/DVB-$COCH_3$. So extraction of these solutes from aqueous solution is not a problem with either PS/DVB or PS/DVB-$COCH_3$ resins. On the other hand, since the k' is smaller on PS/DVB-$COCH_3$ resin in organic eluent, it is easier to elute these organic compounds from PS/DVB-$COCH_3$ than on un-derivatized PS/DVB resin.

Comparing extraction of these solutes on SiC_{18} (12 μm) resin (see Table XI), retention of polar, less polar and non-polar compounds were

much worse. For polar phenolic compounds, none of them were retained on the resin at 0% acetonitrile, even non-polar toluene passed through the mini-column. This explains why the extraction efficiency on SiC_{18} was so low. This can also be seen by those k′ values under different percentages of acetonitrile in the eluent, which were much lower than those on PS/DVB and PS/DVB-$COCH_3$ resins.

Surface area of the resin may affect the extraction efficiency. Comparing the extraction by 10 μm and 3-10 μm PS/DVB-$COCH_3$ (see Tables X and XII), the extraction efficiency was higher on 10 μm than that on 3-10 μm. The main difference of these two resins is the surface area. The surface area of these resins, which were determined by a special method [17], were 497 m^2/g on 10 μm resin and 466 m^2/g on 3-10 μm resin.

From the chromatographic studies using HPLC system with a mini-column, it is clearly shown that the retention, therefore the extraction, of organic compounds on PS/DVB-$COCH_3$ is better than that on un-derivatized PS/DVB, and is overwhelmingly superior to that on SiC_{18}, especially for polar compounds. Also, the higher surface area is helpful for extraction.

Solvent effect in elution of extracted solutes

The procedure of solid phase extraction includes two steps, extraction and elution. It is extremely important to chose a suitable solvent to quantitatively elute the extracted solutes. Taking PS/DVB-$COCH_3$ as the stationary phase, various solvents, from polar to less polar, were tested: methanol, acetonitrile, tetrahydrofuran, tertiary-butylmethyl ether, ethyl acetate and methylene dichloride. Seven compounds including benzene, phenol, indene, p-cresol, naphthalene, anthracene and dibutylphthalate at concentrations of about 70 ppb were used as testing solutes.

The solutes were loaded at the mini-column first. 0.25 ml solvent was used to elute these solutes into a vessel to be used for gas chromatographic injection. Another 0.75 ml of same solvent and an internal standard (p-ethylphenol) was added to the vessel. The vessel was capped, shaken well and ready for injection onto the gas chromatograph. This procedure was repeated three times. Then 1 ml of ethyl acetate was used to elute anything left on the mini-column and collected in the vessel, in which internal standard was added, capped and ready for injection onto the gas chromatograph.

All the collected portions of each solvent were injected into gas chromatograph. The eluted amount in each portion was calculated as

percentage of the total solutes loaded on the mini-column. Tables XIII from (1) to (8) showed the results of those seven compounds with six solvents. Figures 7 from (1) to (8) showed the pattern of elution of seven compounds (benzene, toluene, indene, p-cresol, phenol, naphthalene, dibubylphthalate) with some solvents corresponding to tables.

Of all the solvents, three seem to be the best choices: tetrahydrofuran, ethyl acetate and methylene chloride. Generally speaking, methylene chloride is the best for non-polar compounds (benzene, indene, naphthalene, anthracene and dibutylphthalate) but the worst for polar compounds (phenol and p-cresol). Ethyl acetate is the best for polar compounds (phenol and p-cresol) and good for some non-polar compounds (benzene and indene), but not as good for other non-polar compounds (naphthalene, anthracene and dibutylphthalate). Tetrahydrofuran is good enough for both polar compounds (phenols and p-cresol) and non-polar compounds (benzene, indene, naphthalene, anthracene and dibutylphthalate). One advantage of using tetrahydrofuran is that it is miscible with water. There is not two layers (water and organic solvent) in the vessel when the solutes are eluted from the loading mini-column, therefore there is no worry about the distribution of the solutes in the two phases which might cause error in the gas chromatographic determination.

Table XIII. Elution efficiency of various solvent on Amberchrome 161-$COCH_3$ resin for various compounds. 0.25 ml solvent is used in each portion (1st, 2nd, 3rd & 4th). The numbers are the eluted percentage of the total amount of compounds extracted on the resin.

(1) Methanol

compound	1st	2nd	3rd	4th	2nd 1 ml
Benzene	0	49.6	42.6	4.8	3.7
Toluene	0	31.0	33.1	18.6	17.5
Phenol	10.1	67.1	19.2	0	0
Indene	2.6	3.0	2.3	48.7	42.6
p-Cresol	15.6	12.1	20.7	51.1	0
Naphthalene	0.3	43.8	20.3	15.9	4.0
Anthracene	0	9.2	18.0	36.1	37.1
Dibutyl phthalate	3.6	24.9	34.8	16.6	16.0

(2) Acetonitrile

Compound	1st	2nd	3rd	4th	2nd 1 ml
Benzene	33.8	56.4	9.7	0	0
Toluene	25.1	49.0	13.1	7.5	5.3
Phenol	35.7	64.0	0	0	0
Indene	19.3	44.6	23.9	9.2	2.9
p-Cresol	32.5	61.4	6.1	0	0
Naphthalene	14.8	38.7	33.7	8.5	4.2
Anthracene	0	7.8	21.1	20.8	50.3
Dibutyl phthalate	29.8	54.0	13.0	2.6	0.7

(3) Acetone

Compound	1st	2nd	3rd	4th	2nd 1 ml
Benzene	15.7	74.2	10.1	0	0
Toluene	10.6	67.6	11.9	0	9.8
Phenol	9.8	91.2	0	0	0
Indene	6.7	73.7	19.3	0	0
p-Cresol	14.4	80.8	5.2	0	0
Naphthalene	4.5	66.0	22.3	4.2	3.0
Anthracene	0	59.3	37.3	3.4	0
Dibutyl phthalate	5.8	82.6	9.7	1.9	0

(4) Tetrahydrogenfuran

Compound	1st	2nd	3rd	4th	2nd 1 ml
Benzene	50.2	49.8	0	0	0
Toluene	32.8	48.0	7.1	0.9	11.1
Phenol	45.7	52.0	2.3	0	0
Indene	30.0	58.0	11.2	1.2	0
p-Cresol	41.1	54.3	4.5	0	0
Naphthalene	27.7	57.2	13.0	1.3	0.7
Anthracene	23.0	63.8	13.2	0	0
Dibutyl phthalate	33.6	57.6	7.3	0.8	0.8

(5) Ethyl acetate

Compound	1st	2nd	3rd	4th	2nd 1 ml
Benzene	39.1	54.4	6.4	0	0
Toluene	27.2	37.8	14.1	10.8	10.2
Phenol	38.3	61.7	0	0	0
Indene	26.4	59.5	14.1	0	0
p-Cresol	36.9	60.0	3.1	0	0
Naphthalene	18.9	52.5	19.2	5.0	4.5
Anthracene	7.0	48.2	27.8	9.2	7.9
Dibutyl phthalate	0.7	74.4	15.7	6.8	2.3

(6) Methylene chloride

Compound	1st	2nd	3rd	4th	2nd 1 ml
Benzene	75.9	24.1	0	0	0
Toluene	66.8	22.6	-	-	10.6
Phenol	1.5	49.1	41.2	8.1	0
Indene	68.8	31.2	0	0	0
p-Cresol	3.5	64.9	28.1	3.5	0
Naphthalene	63.4	36.6	0	0	0
Anthracene	57.8	40.8	1.4	0	0
Dibutyl phthalate	70.8	27.6	0.8	0.7	0.2

(7) Methylene chloride 50% + Ethyl acetate 50%

Compound	1st	2nd	3rd	4th	2nd 1 ml
Benzene	76.9	23.1	0	0	0
Toluene	52.1	25.1	8.5	7.9	6.4
Phenol	49.2	50.8	0	0	0
Indene	53.7	44.0	2.3	0	0
p-Cresol	50.0	47.7	1.7	0.6	0
Naphthalene	45.9	53.0	1.1	0	0
Anthracene	29.8	63.5	6.3	0.4	0
Dibutyl phthalate	65.0	31.9	1.4	1.3	0.3

(8) Ethyl acetate on 10 μm PS/DVB resin

Compound	1st	2nd	3rd	4th	2nd 1 ml
Benzene	100.0	0	0	0	0
Toluene	64.9	9.0	9.4	9.0	8.6
Phenol	100.0	0	0	0	0
Indene	95.1	4.2	0.7	0	0
p-Cresol	99.0	1.0	0	0	0
Naphthalene	89.3	8.4	2.3	0	0
Anthracene	73.7	25.2	1.3	0	0
Dibutyl phthalate	89.5	7.6	1.6	0.7	0.4

(1). Compound: Benzene

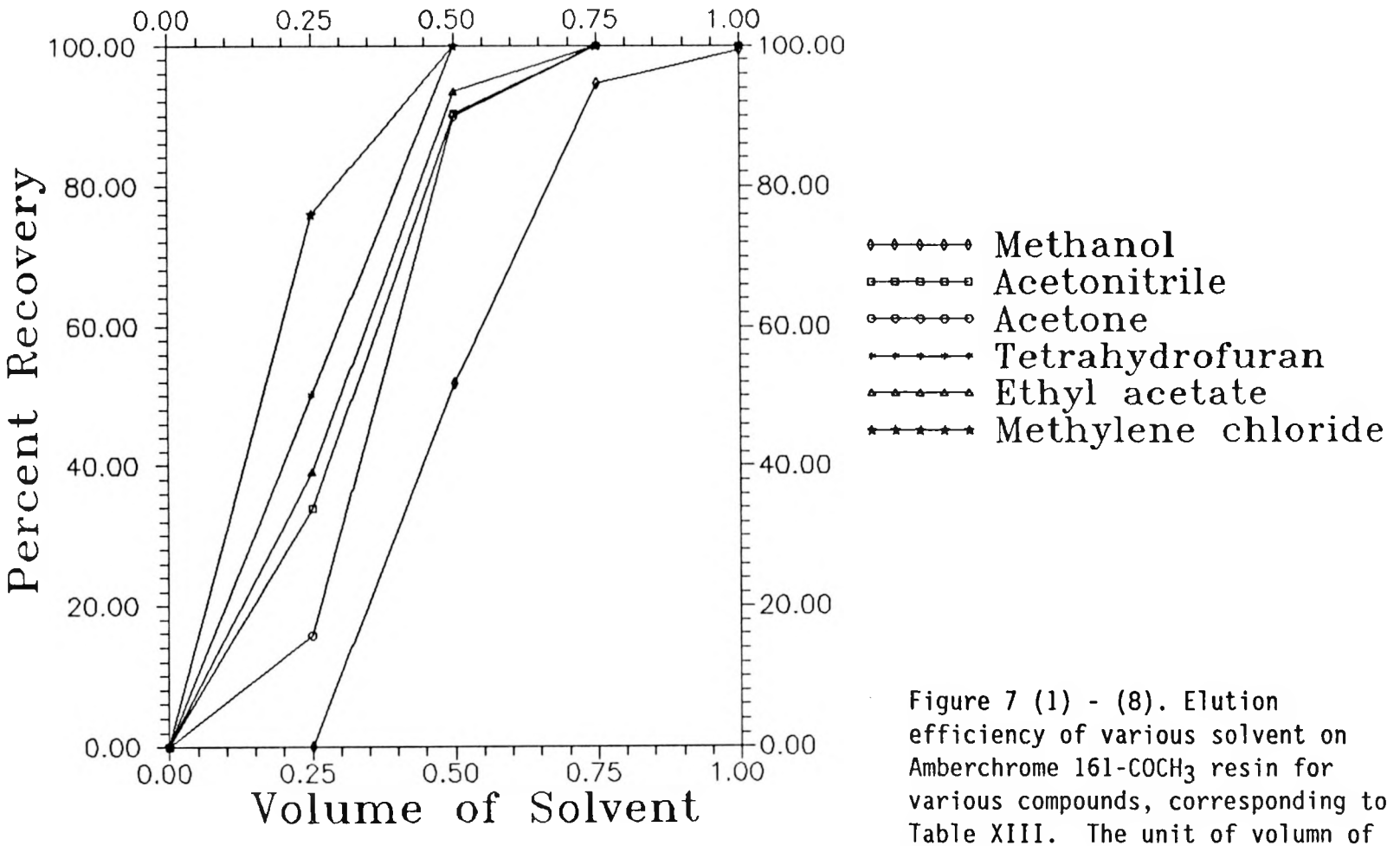
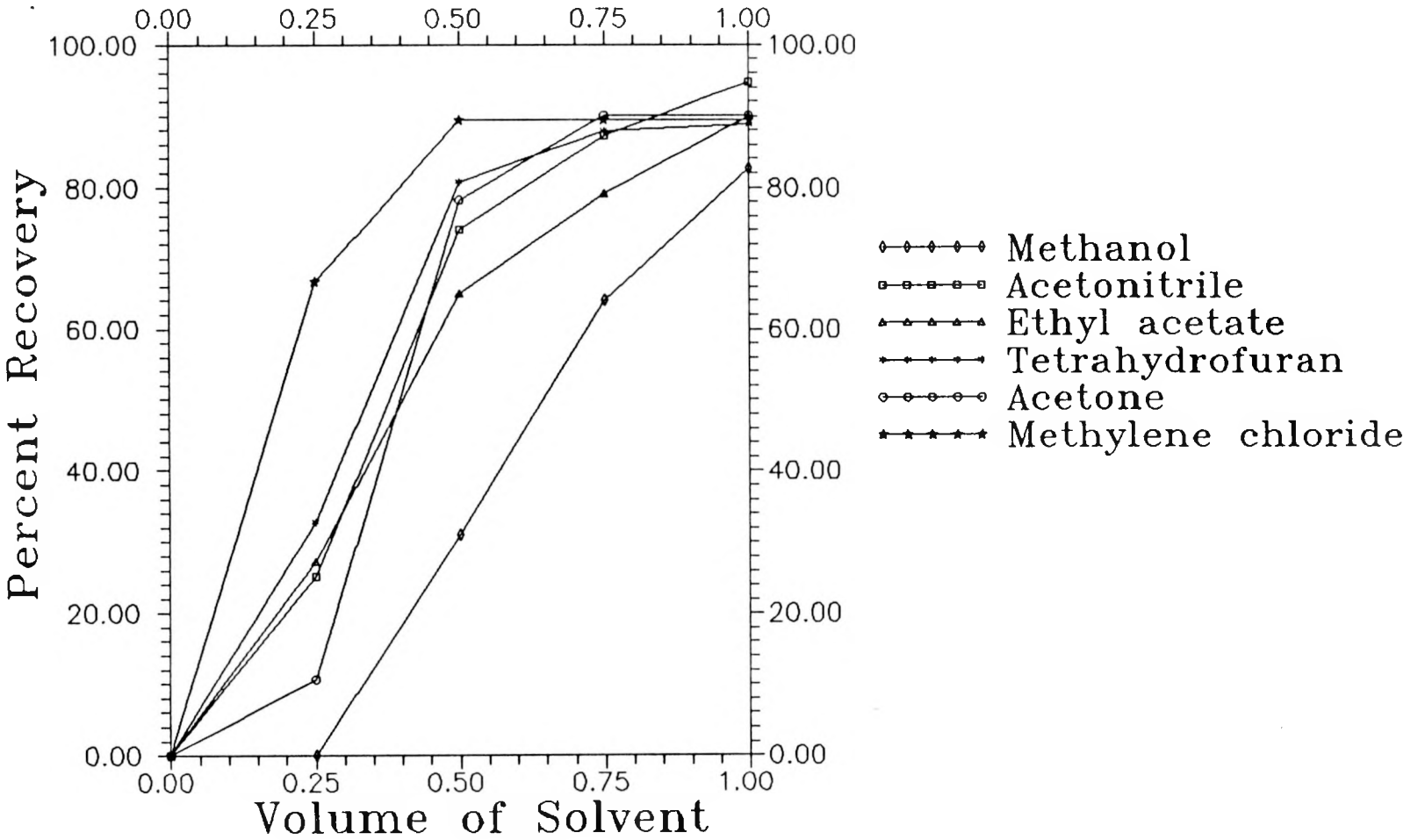
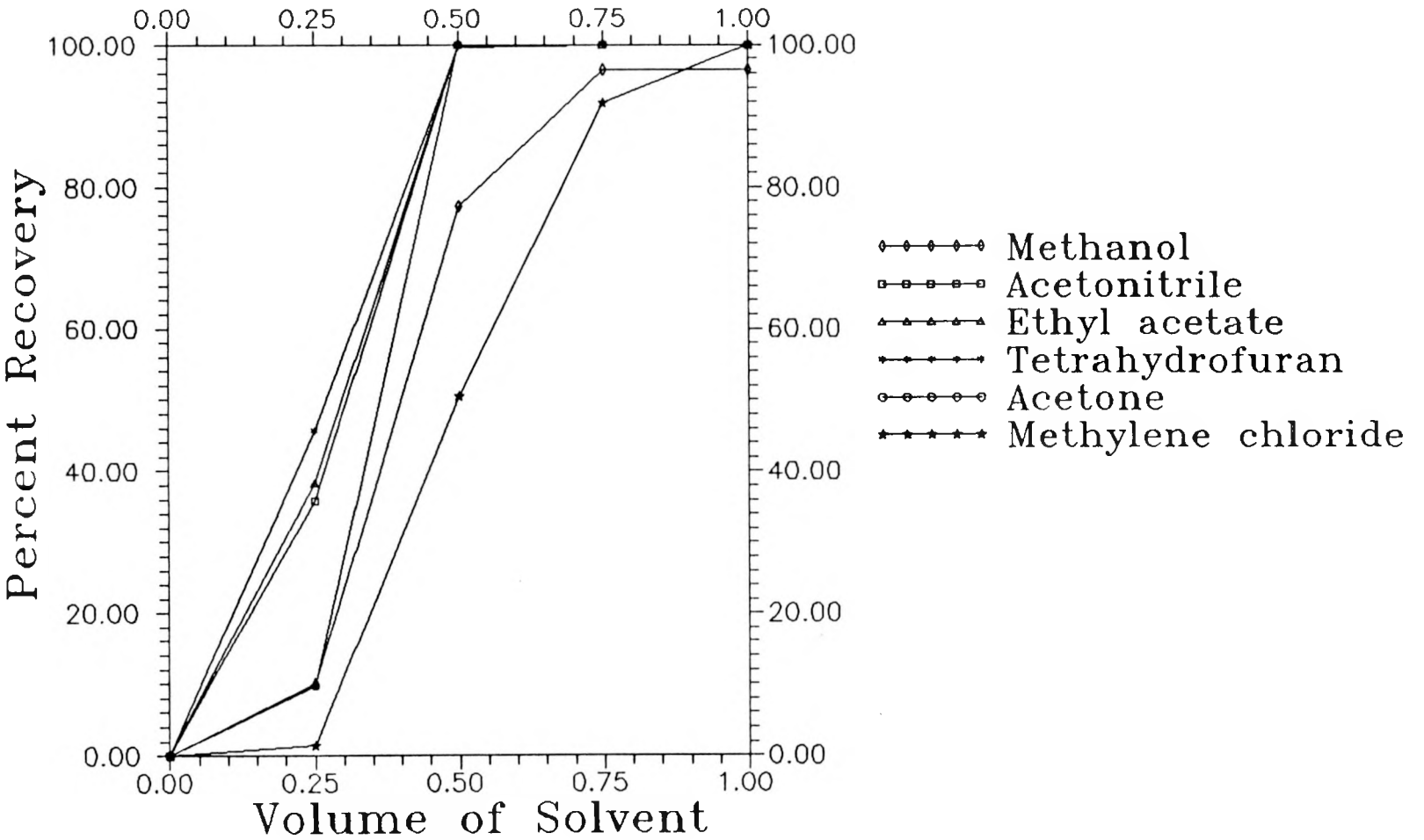


Figure 7 (1) - (8). Elution efficiency of various solvent on Amberchrome 161-$COCH_3$ resin for various compounds, corresponding to Table XIII. The unit of volumn of solvent is ml.

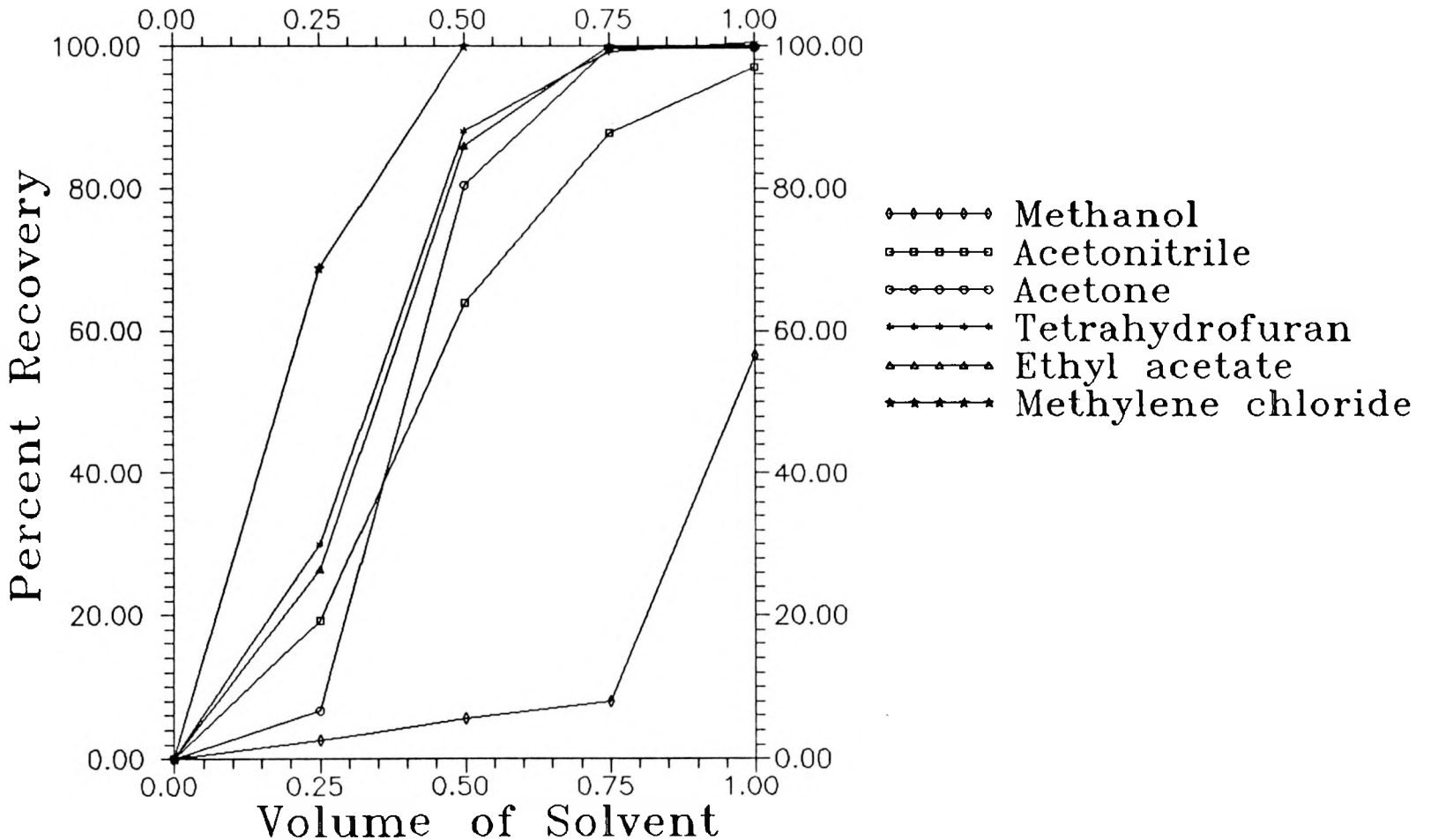
(2). Compound: Toluene



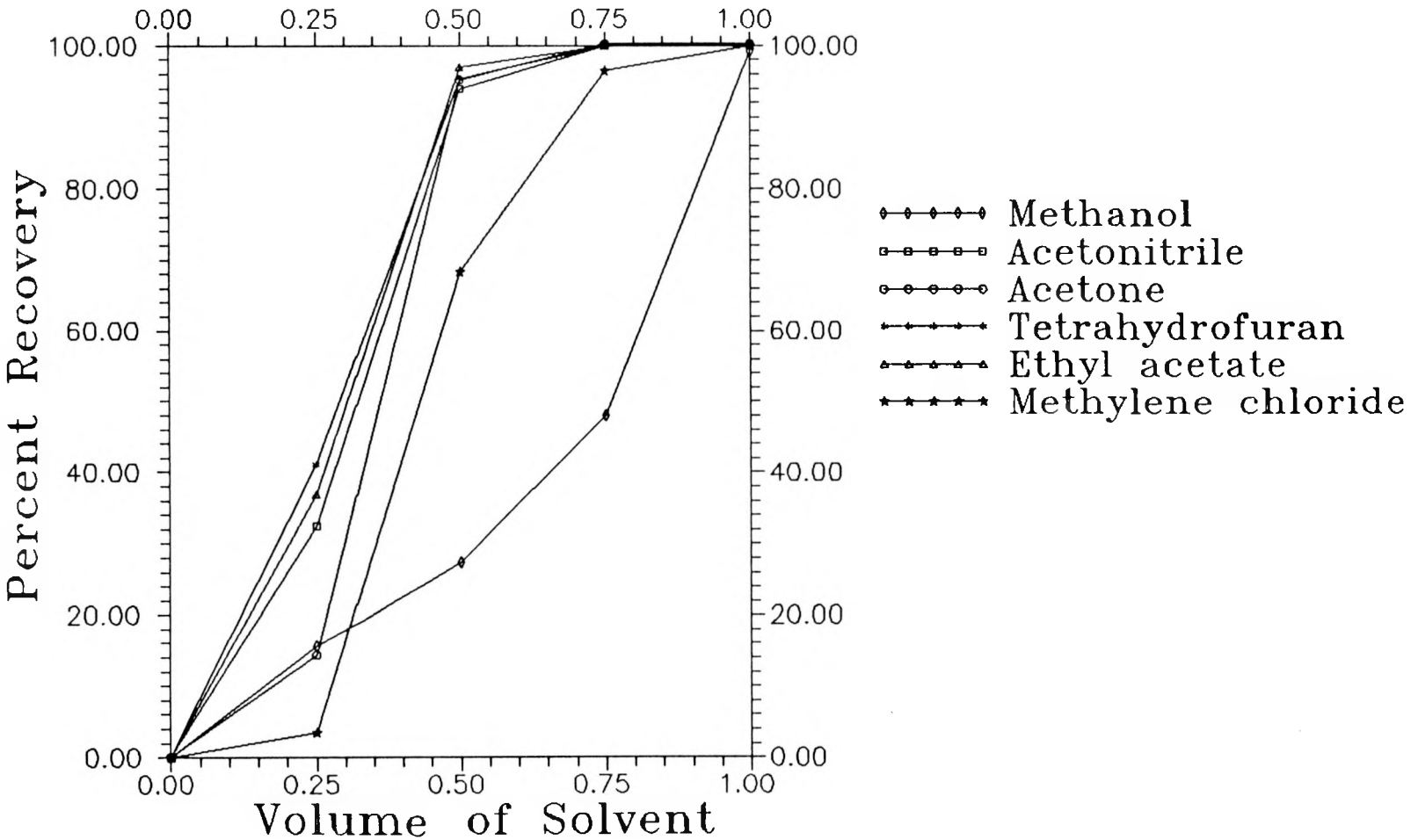
(3). Compound: Phenol



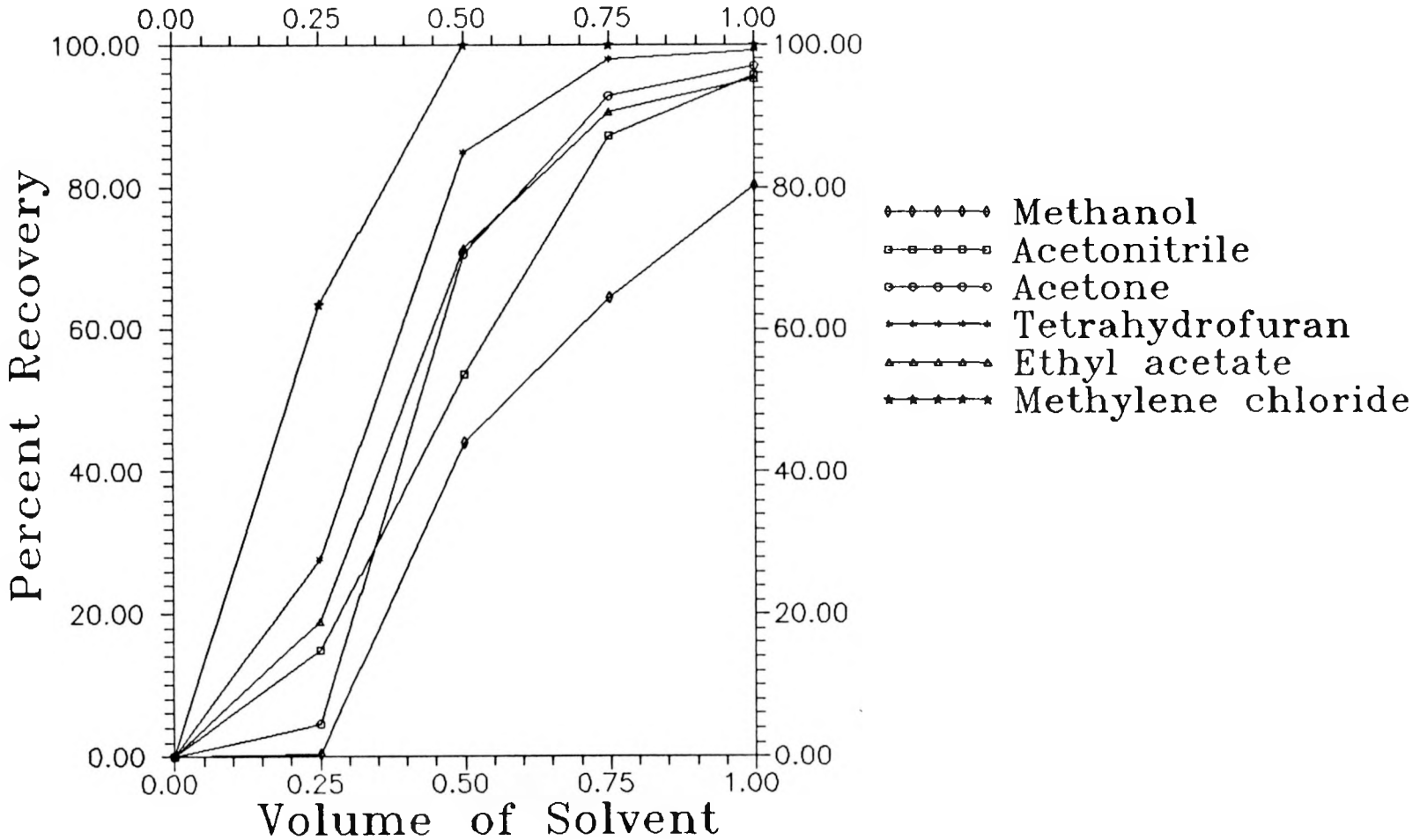
(4). Compound: Indene



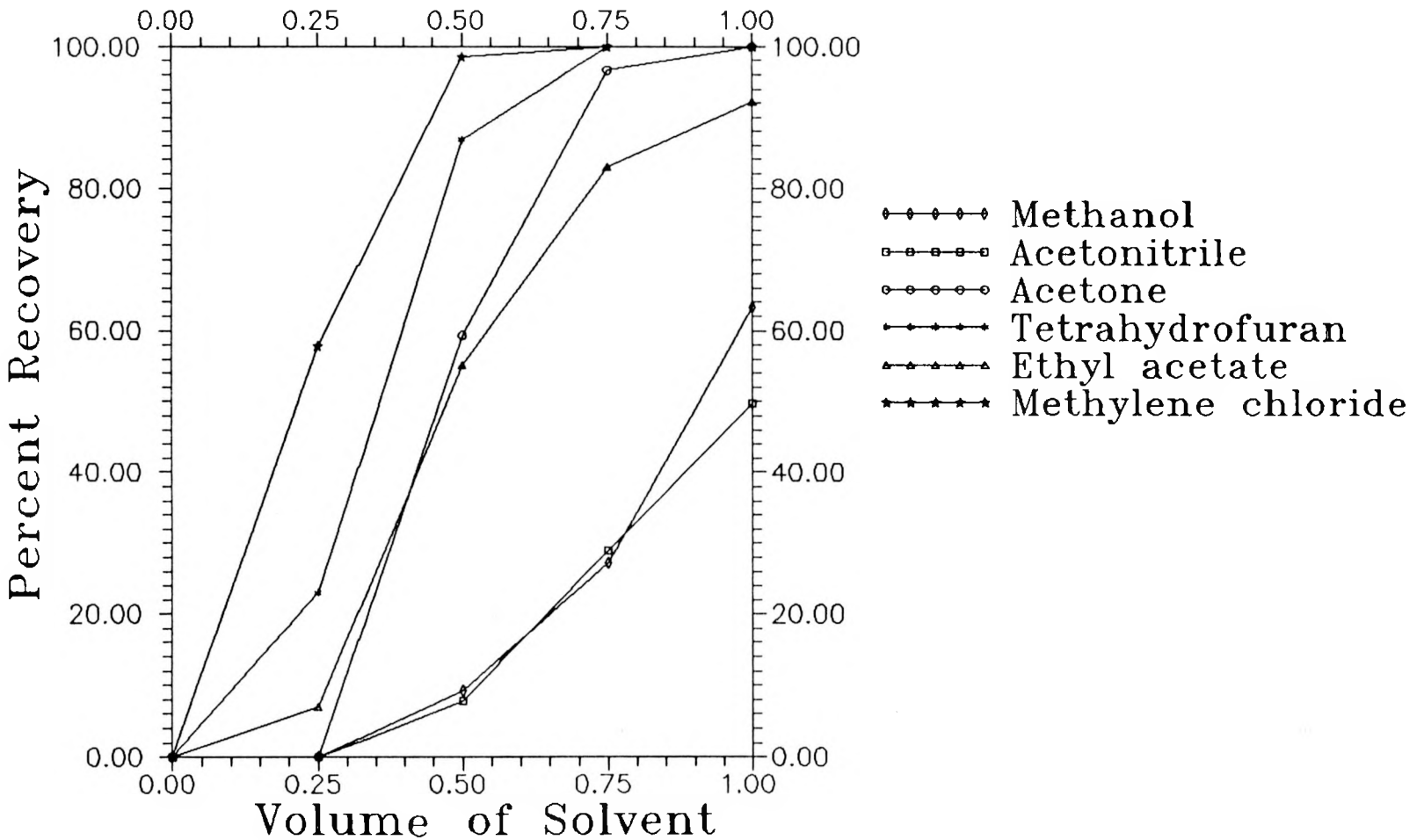
(5). Compound: p-Cresol



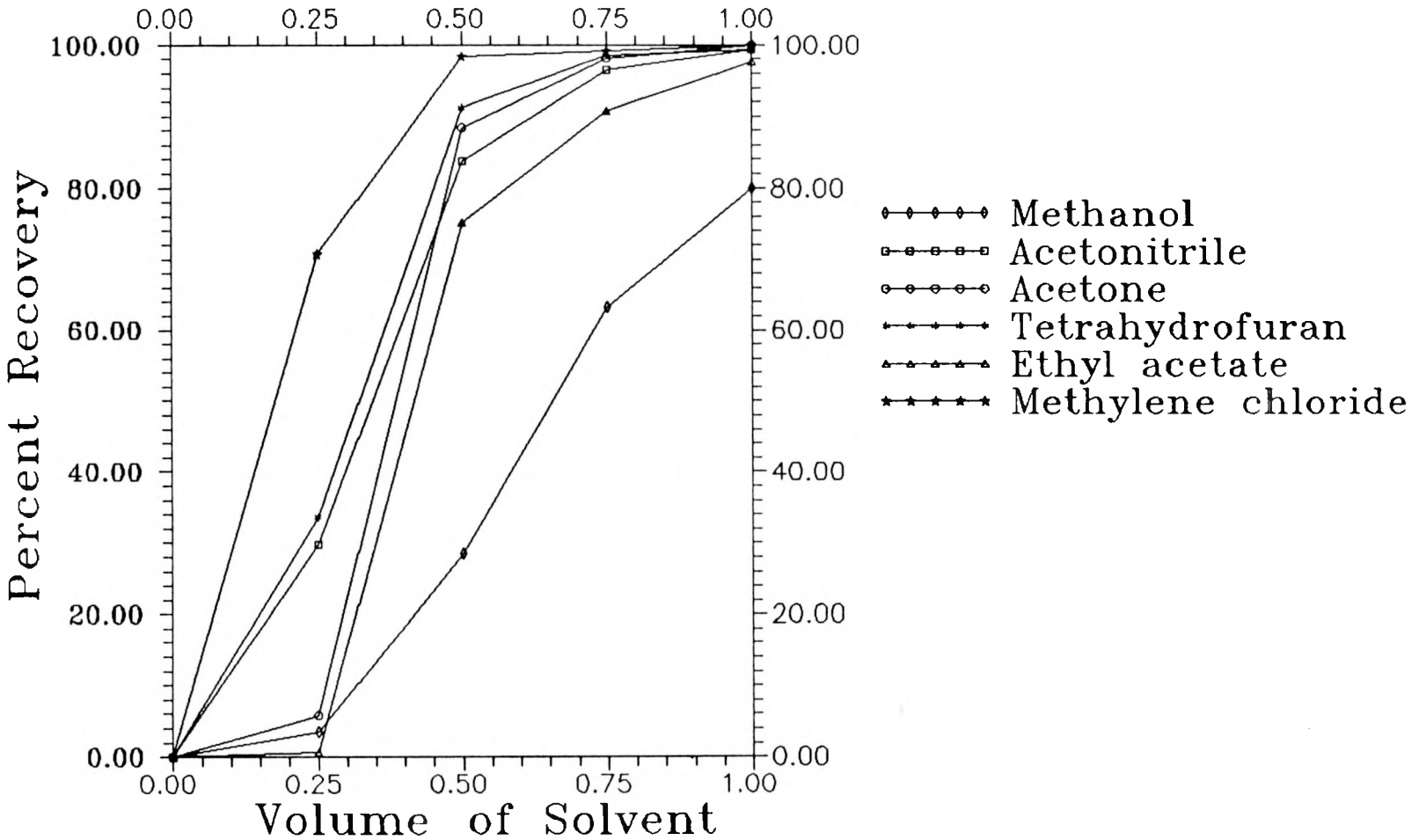
(6). Compound: Naphthalene



(7). Compound: Anthracene



(8). Compound: Dibutylphthalate



CONCLUSION

Incorporation of hydrophilic groups into a porous polymeric resin enables excellent surface contact to be made with an aqueous sample without resorting to any pretreatment of the resin with an organic solvent. Aromatic- and other relatively non-polar organic compounds are taken up from aqueous solutions almost as strongly by the derivatized resins as by the un-derivatized resins. More polar compounds (particlarly phenols) are retained more strongly by the resins with a hydrophilic group, especially when the resin has an acetyl group. The uptake of phenols may involve some interaction such as hydrogen bonding between the carbonyl group and the phenolic hydroxyl group.

The amount of surface area of a porous resin seems to have a major effect on its efficiency for SPE. The better recovery of test compounds by polymeric resins compared to bonded-phase silica resins could be due in part to the significantly higher surface area of the polymeric resins.

Chromatographic studies of retention of several compounds on the different stationary phase using an HPLC system with a mini-column proved the high extraction efficiency of hydrophilic derivatized resin, which is much better than un-derivatized resin and overwhelmingly superior to SiC_{18} resin.

Also, it should be noted that silica-based resins partially dissolve in basic aqueous solutions. Polymeric organic resins, including those with hydrophilic substituents, are stable enough to be used over a wide pH range.

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SECTION III. SULFONATED POLYSTYRENE DIVINYLBENZENE RESINS FOR HIGH PERFORMANCE LIQUID CHROMATOGRAPHY

INTRODUCTION

Sulfonated PS/DVB resin has long been used as a cation exchanger since Small first introduced the concept of ion chromatography in 1975 [1]. Sulfonated PS/DVB is the most widely used cation exchanger in ion chromatography for separation of cations [2-5]. In addition to a reversible, stoichiometric ion exchange, sulfonated PS/DVB resins have the ability to absorb organic and non-ionic solutes from a solution. The number of sulfonic functional groups on the resin will influence the sorption but are not directly involved in the stoichiometry of the sorption. Because of the chemical stability of PS/DVB resin and other advantages, sulfonated PS/DVB resins are also frequently used for various applications, such as analysis of amino acid [6, 7], determination of organic acids [8, 9], determination of carbohydrate [10, 11], etc.

Organic and non-ionic solutes are absorbed by ion exchangers because of several kinds of interactions that may occur. Interactions between the solutes and polymeric matrix of the ion exchanger is one of the main reason. Such interactions could be dipole-dipole interactions, π-electron and other interactions.

In present work, we studied the relationship between the capacity factor and capacities of sulfonated PS/DVB of various organic compounds.

We used sulfonated PS/DVB resins to separate neutral and basic compounds by an off-line mini-column as well as basic and weaker basic compounds by an on-line mini-column in an HPLC system. Also, some aromatic amine isomers were separated by sulfonated PS/DVB resin.

EXPERIMENTAL

Preparation of sulfonated PS/DVB resin

The starting material for the preparation of sulfonated PS/DVB resin was 10 μm porous, spherical (Serasep Inc., Santa Clara, CA). The resin was cleaned with acetonitrile, ethyl acetate, hexane thoroughly and dried. About 2 grams of resin and 20 ml of concentrated sulfuric acid were placed in a round bottom flask. Different reaction temperature and time were used to get different capacity of the resin: 0.34 Meq./g at 5^{0}C, 20 seconds; 0.63 Meq./g at 25^{0}C, 70 seconds; 0.93 Meq./g at 25^{0}C, 5 minutes; 1.64 Meq./g at 75^{0}C, 15 minutes. The resins were thoroughly washed with deionized water and the capacity was determined by titration. The resins of different capacities were packed into columns by the packing equipment.

Apparatus and chemicals

A Gilson 302 HPLC system quipped with a microprocessor controller (Gilson Medical Inc., Middleton, WI), A 7125 Rheodyne injector (Rheodyne, Berkeley, CA) equipped with a 20 microliter loop, a Spectrofolw 783 Kratos variable wavelength UV-Vis detector (Kratos Analytical Instruments, Ramsey, NJ), A Fisher Recordall series 5000 recorder (Fisher

Scientific/Instruments Lab, Itasca, IL), a Hitachi D-2000 intergrater (EM Science, Cherry Hill, NJ), were used for high performance liquid chromatography. A Shandon HPLC packing pump (Shandon Southern Products Limited, Sewichley, PA) was used for column packing. Two home-packed separation columns were used: original PS/DVB resin and sulfonated PS/DVB resin columns which were 100 x 4.6 mm. A home-made mini-column (6.4 x 2.1 mm) designed by Rodney R. Walters [12], was used for group separation and placed between the injection valve and the separation column. This mini-column can be used for group separation in the system or by-passed by two two-way valves. The whole HPLC separation system was shown in Figure 1. A SiC_{18} commercial column (Vydac column, 5μm particle size, 250 x 4.6 mm, Alltech Associates, Inc., Deerfield, IL) was used for comparason of isomer separation.

The reagent grade compounds were used for the HPLC experiments and reactions. HPLC grade acetonitrile was used as eluent and laboratory distilled water was further deionized by a Barnstead Nanopure II system (Sybron Barnstead, Boston, MA).

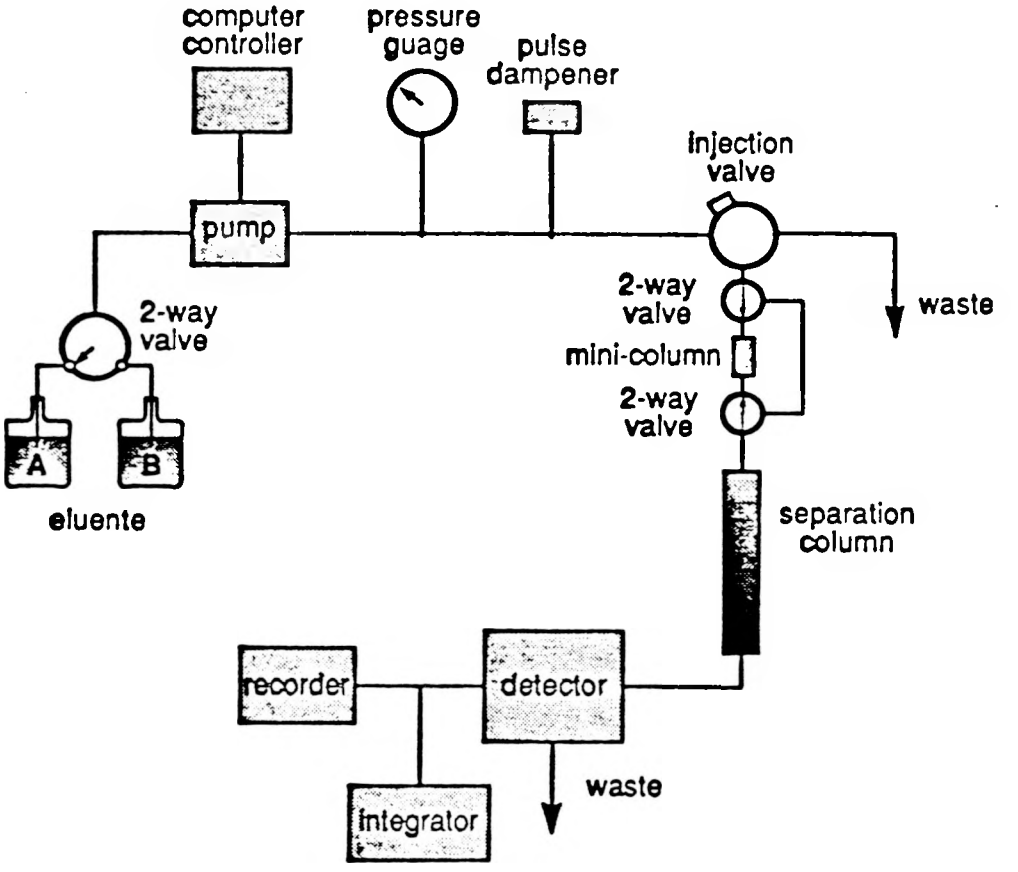


Figure 1. HPLC separation system

RESULTS AND DISCUSSION

Effect of the capacity of resin on t_r of organic compounds

PS/DVB resin is hydrophobic and is usually used in reversed-phase liquid chromatography quite often with a hydrophilic mobile phase. The introduction of sulfonic groups make it hydrophilic, and the t_r of various organic compounds decreases.

The sulfonated PS/DVB resins with different capacities (0.34, 0.63, 0.93 and 1.64 meq./g) were packed into 100 x 4.6 mm column. Various neutral organic compounds, which included o-dichlorobenzene, toluene, methylbenzoate, acetophenone and phenol, were tested on those columns with an eluent of 50% acetonitrile + 50% water and flow rate of 1 ml/min. The k′ values of those compounds were calculated and the log k′ were plotted against the capacities of the resin. The plot showed liner relationship between the log k′ and the capacity (see Figure 2) and the slope of various compounds were almost the same. By calculation of the slopes of those compounds, it is shown that the more non-polar the compounds (e.g.o-dichlorobenzene) have a more linear slope, and the more polar the compounds (e.g.phenol) have a less linear slope (see table I). Since the retention of organic compounds on the resin of different capacity was predictable, it can deduced that it

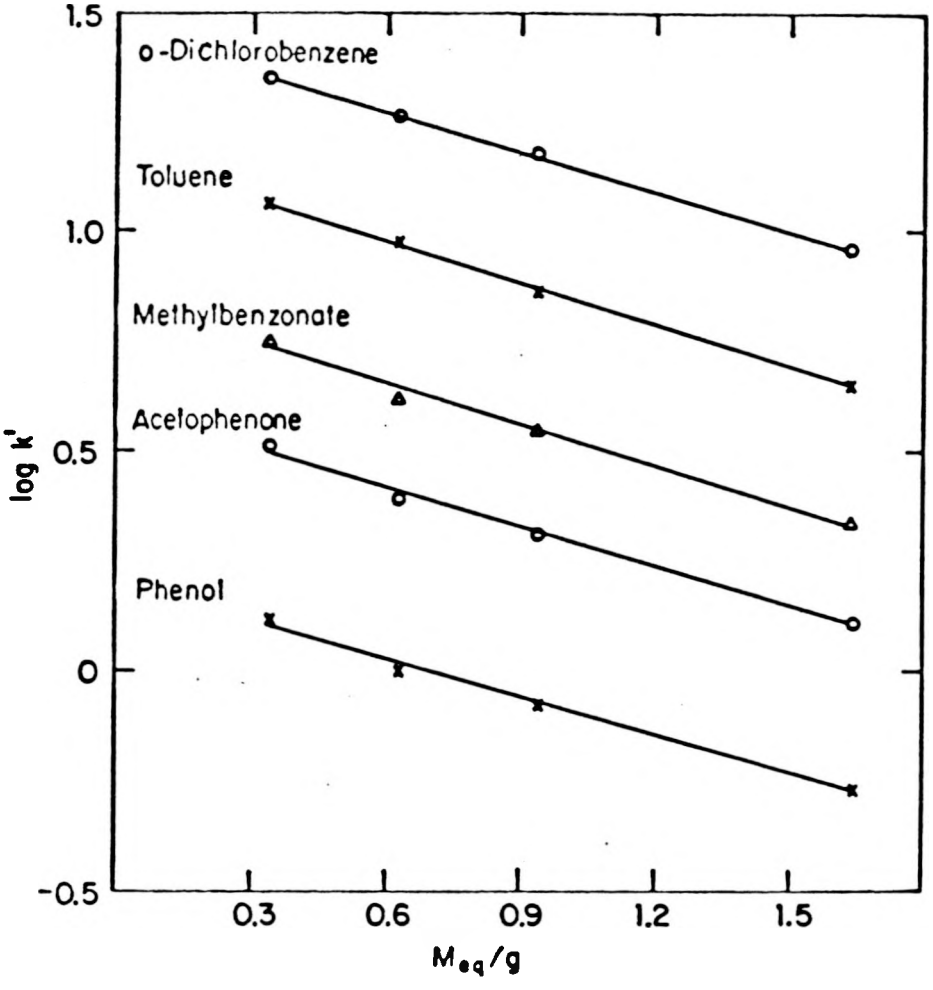


Figure 2. Log k'of various compounds as a function of capacity of sulfunated PS/DVB resins. Column conditions: 100 x 4.6 mm, packede with 10 μm particle. Eluent: 50% ACN + 50% H_2O

Table I. The liner regression coefficient of slopes of various compounds on plot of log k′ against capacity of the PS/DVB resin

Compound	Coefficient
o-Dichlorobenzene	0.999
Toluene	0.999
Methylbenzoate	0.996
Acetophenone	0.995
Phenol	0.993

would be easier to elute retained organic compounds on resins of higher capacity. This conclusion was useful later in group separation of neutral and basic compounds using sulfonated PS/DVB resin.

Separation of basic and weaker basic compounds by an on-line mini-column

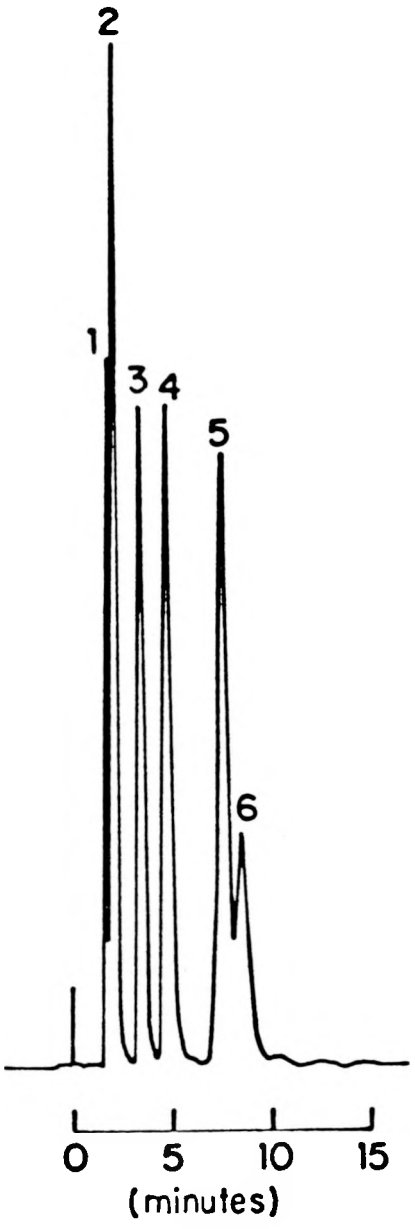
The basicities of aromatic amines could be quite different, depending on the nitrogen atoms or other heterogeneous atoms they contain. For example, the pK_b of pyridine and quinoline, which contain only one nitrogen in the molecular structure, are 8.8 and 9.1 [2] in aqueous solution respectively. The pK_b of pyrazine and quinoxaline, which contain two nitrogen atoms in the molecular structure, are 13.4 and 13.4 [3]. An interesting question arises: Can these two groups of amines, basic and weaker basic amines, be separated according to their difference between pK_b values?

Separation of these two groups was tried using an HPLC system. A home-made mini-column was packed with sulfonated PS/DVB resin of capacity 0.93 meq/g. The mini-column was connected in the HPLC system 1 cm before the separation column, which was packed with porous, spherical 10 μm PS/DVB resin, and after the injector. Only the separation column of PS/DVB resin can be used here because the eluent would contain 0.1M NH_4OH

(pH is about 13) later which is too basic for the Si based column to withstand.

A mixture of six amines was made with the concentration of each compound around 50 ppm. Three amines, pyridine, quinoline and 2-benzylpyridine, which contained only one nitrogen atom in the molecular structure, were relatively strong basic compounds. The pK_b of these three amines in aqueous solution were 8.8, 9.1 and 8.8 respectively. The pK_b of quinoline (9.1) is a little higher than the other two. This was most likely due to the benzene ring participating in the conjugated system, and therefore the lone pair of electrons, which cause the basicity, on the nitrogen atom to be more distributed than in the other two amines. The other three amines, pyrazine, quinoxaline and benzothiazole, contain either two nitrogen atoms or one nitrogen atom and one sulfur atom, and are relatively weak basic compounds. The pK_bs of pyrazine and quinoxaline are both 13.4 [13]. The pK_b of benzothiazole was not found, however, it can be predicted that the pK_b is similar to the other two amines due to the existence of a sulfur atom in the structure.

This mixture was injected into the HPLC system by-passing the mini-column with 50% acetonitrile + 50% water as eluent. All six compounds passed through the separation column and some of them overlapped one



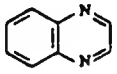
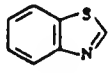
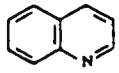
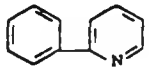
Relatively weak basic compounds:		pK_b
1=Pyrazine		13.4
3=Quinoxaline		13.4
5=Benzothiazole		
Relatively strong basic compounds:		
2=Pyridine		8.8
4=Quinoline		9.1
6=2-Benzylpyridine		8.8

Figure 3. Chromatographic separation of six compounds by the HPLC system without using the mini-column, Separation column: 100 x 4.6 mm, packed with 10 μm PS/DVB resin. Eluent: 50% ACN + 50% H_2O

another (see Figure 3). However, if the mini-column was used in the system when injecting the same sample, only three weaker amines passed through the mini-column and were separated on the separation column (see Figure 4.A). The three relatively strong amines were retained on the mini-column. Then the eluent was changed to another one which was composed of 50% acetonitrile + 50% water + 0.1M NH_4OH, the three retained amines were eluted from the mini-column and then separated on the separation column. Although these three amines were separated very well on the separation column, the change of the eluent caused a change of the base line at the beginning time when the earliest compound (pyridine) was eluted, therefore affecting the quantitative determination of this first compound. This problem can be solved by running a blank eluent and doing a base line correction with a computer. However, we solved this problem by another method. The separation column was disconnected from the HPLC system and the retained amines were eluted by the second eluent. The eluted amines were quantitatively collected into a volumetric flask while monitoring the UV-VIS detector. The collected solution was then injected into the HPLC system without use of the mini-column. Three relatively strong amines were separated well without baseline disturbance (see Figure 4.B).

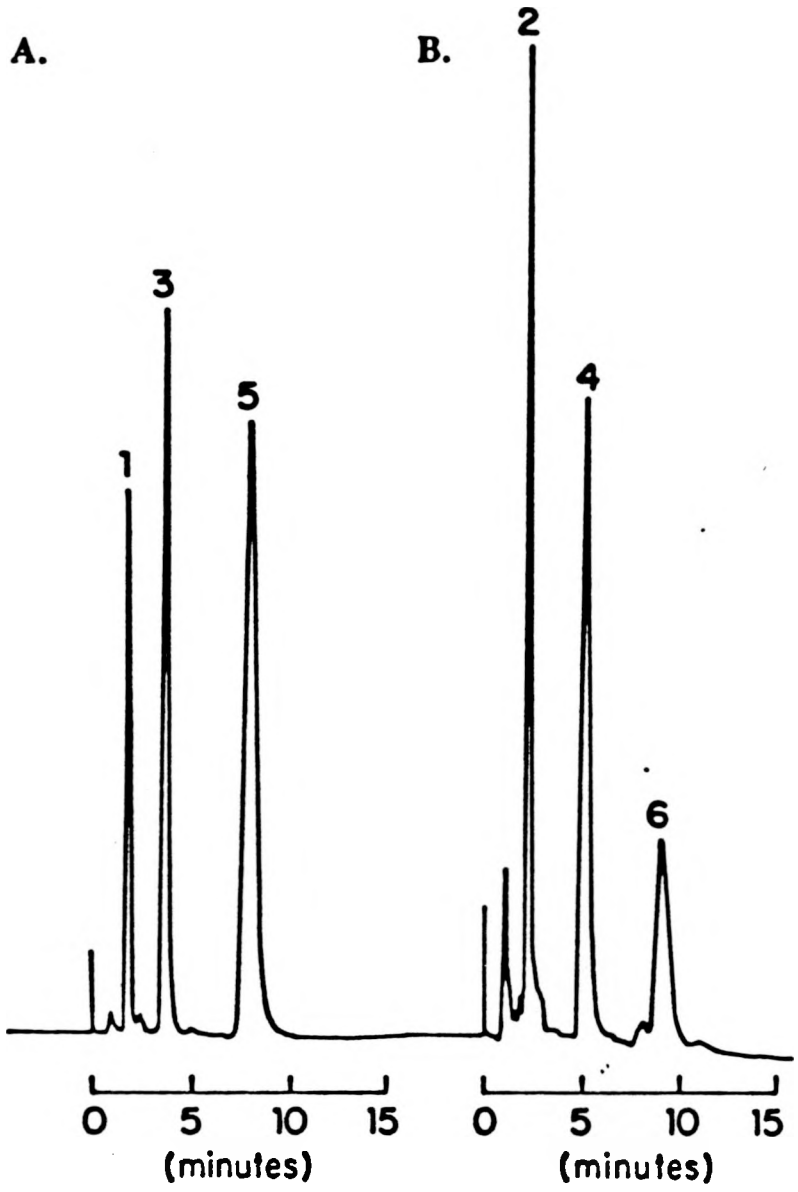


Figure 4. A. Chromatographic separation of six compounds using the mini-column. All other conditions are same as fig.3. B. Chromatogram of three retained amines eluted from the mini-column by 0.1M NH_4OH + 50% ACN and then separated

Several other basic and weaker basic compounds were tested. The same results were obtained, that is, relatively strong amines which had only one nitrogen atom, were retained on the mini-column, while the relatively weaker basic amines, which had two nitrogen atoms, passed through the mini-column. Examples such as 2,6-dimethylquinoline and 5,6-benzoquinoline were retained on the mini-column, while uracil and pyridizine passed through the mini-column.

Quantitative determination of basic and weaker basic amines

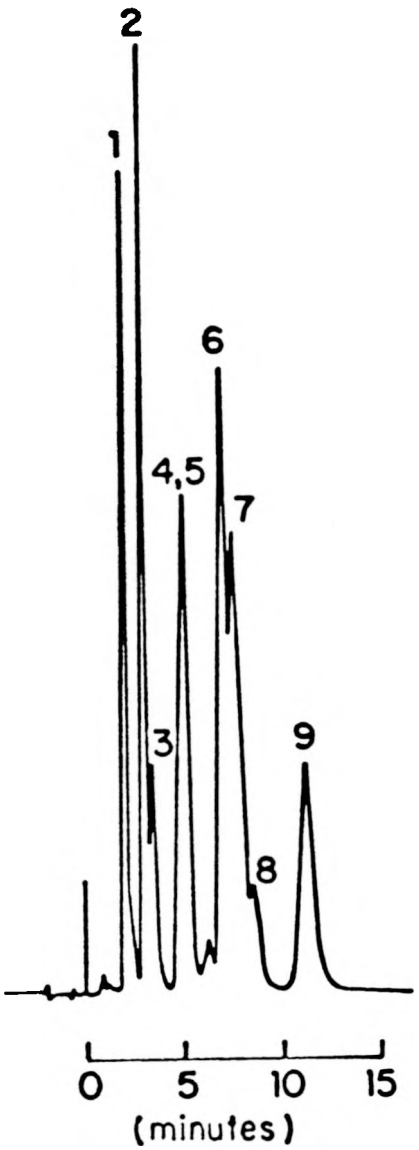
The relatively weaker amines, which passed through the mini-column and were separated by the HPLC column, can be determined directly by normal liquid chromatographic methods without problems. The relatively stronger amines, which were retained on the mini-column and then eluted and collected, were determined by normal liquid chromatographic method also. The results of quantitative determination were very good. The recoveries of these three relatively stronger amines were 98.7% (pyridine), 99.4% (quinoline) and 97.6% (2-benzylpyridine), compared with the original concentration.

Separation of neutral compounds and basic amines by an off-line mini-column

Since the sulfonic group on sulfonated resin is able to react with basic amines to form an ion-pair, a mini-column, which is similar to that used in solid phase extraction, was packed with about 1 g of sulfonated PS/DVB of 0.93 Meq/g and used for separation of neutral and basic compounds.

A mixture solution of five neutral compounds, 2,4-dichlorophenol, phenol, ethylphenol, benzonitrile, 2,4-dinitrofluorobenzene, and four basic amines, which were pyridine, quinoxaline, quinoline and benzothiazole, was prepared. The exact concentration of each compound was approximately 100 ppm. The chromatogram of this solution was shown in the Figure 5. Since some neutral compounds and basic amines had similar retention times, they overlapped one another.

The mini-column was wetted with acetonitrile before loading the sample. 10 ml of mixture solution was passed through the mini-column at flow rate of 4 ml/min. Then two 1-ml portions of acetonitrile was used as eluent to elute the neutral compounds. Since the basic amines were paired ionically with the sulfonic group, they were not eluted by acetonitrile. The eluted solution was injected into the HPLC



Basic compounds:

1=Pyridine

3=Quinoxaline

5=quinoline

7=Benzothiazole

Neutral compounds:

2=2,4-Dichlorophenol

4=Phenol

6=Ethylphenol

8=Benzonitrile

9=2,4-Dinitrofluorobenzene

Figure 5. Chromatographic separation of four basic and five neutral compounds by the HPLC system without using the mini-column. Column conditions and eluent are the same as fig.3

system (the mobile phase was 50% ACN + 50% H_2O) and the chromatogram is shown in Figure 6.A. All five neutral compounds were separated.

The retained basic amines were eluted by two 1-ml 0.1M NH_4OH + 100% ACN. The eluted solution was collected and injected into the HPLC system directly. All four basic amines were well separated. The chromatogram is shown in Figure 6.B.

Off-line quantitative determination

This procedure actually can be used both for group separation and preconcentration. Both the neutral and basic compounds can be absorbed on the resin similar to a solid-phase extraction. However, with the help of sulfonic group on the resin, the neutral compounds can be eluted first, then the basic amines can be eluted from the resin by eluent with 0.1M NH_4OH.

The recoveries of both neutral and basic compounds were very good. The eluted solutions were collected in a 5 ml volumetric flask, which was filled with suitable eluent to the mark line and mixed well. This solution was then injected into the HPLC system and the recovery of the compounds determined quantitatively by a calibration curve. The recoveries of all these compounds are listed in Table II, the percentage

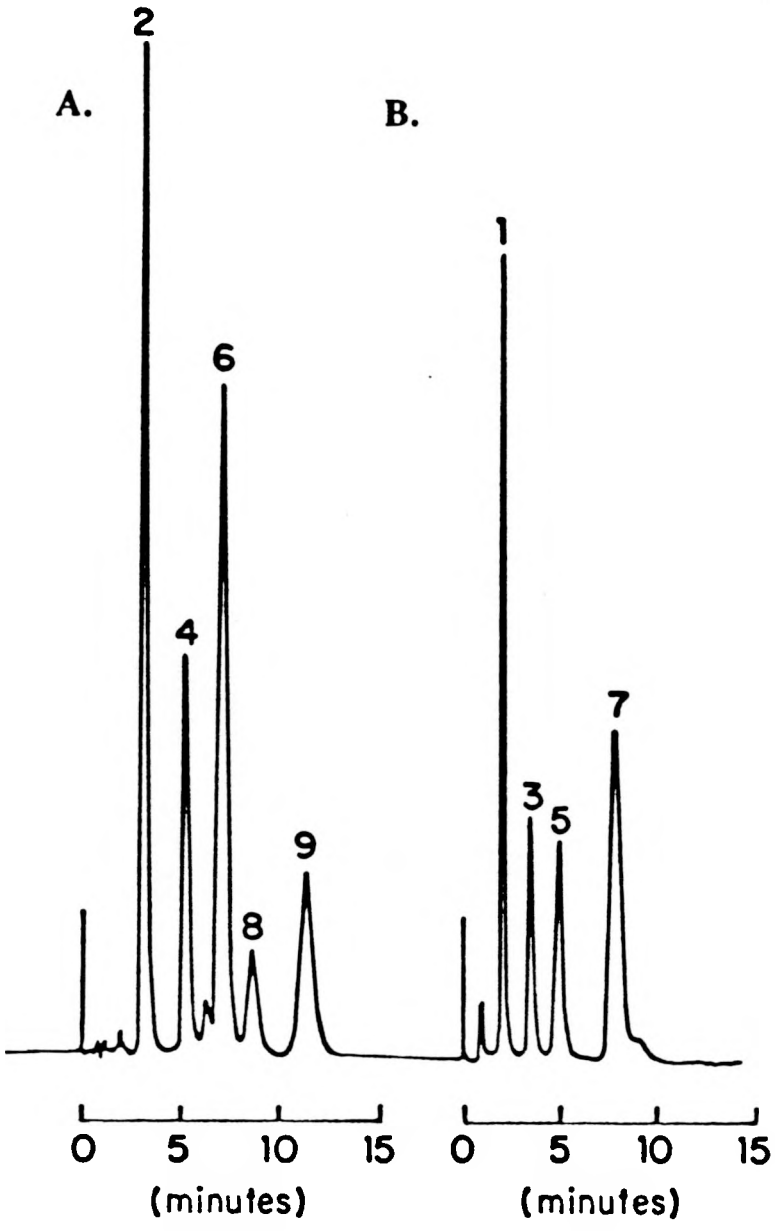


Figure 6. A. Chromatographic separation of the mixture compounds after passing through the off-line mini-column. B. Chromatogram of four basic amines eluted by 0.1M NH_4OH + 50% ACN from the off-line mini-column and then separated

Table II Recovery percentage of the compounds in off-line quantitative determination

Compound	Recovery (%)
Dichlorophenol	98.1
Phenol	98.0
Ethylphenol	99.4
Benzonitrile	99.5
2,4-Dinitro-fluorobenzene	96.3
Pyridine	98.6
Quinoxaline	95.7
Quinoline	98.4
Benzothiazole	101.2

was compared with the original sample solution, and an average of 98.3% was obtained.

Separation of basic isomeric compounds

Sulfonated PS/DVB resin can also be used for the separation of amine isomers. Although the basicity of aromatic amine isomer might be slightly different because of the different substituted position of nitrogen atoms on the ring, this small difference may not be discriminated well by the reversed-phase column. However, this small difference might be discriminated by sulfonated PS/DVB resin. One example is the separation of two basic isomers, pyridazine (pk_b = 11.76) and pyrazine (pk_b = 13.35). They were almost baseline separated on sulfonated PS/DVB resin (see Figure 7.A); the eluent used was composed of 10% acetonitrile + 90% water + 0.1M NH_4OH. The same sample was not separated well on a commercial SiC_{18} column (see Figure 7.B); the eluent used was optimized as 10% acetonitrile + 90% water. The sulfonated PS/DVB resin was 10 μm particle size and the column size was 100 x 4.6 mm, while the commercial SiC18 resin was 5 μm particle size and the column size was 250 x 4.6 mm. This also illustrated one of the advantages of the sulfonated PS/DVB resin, that is, it can withstand a

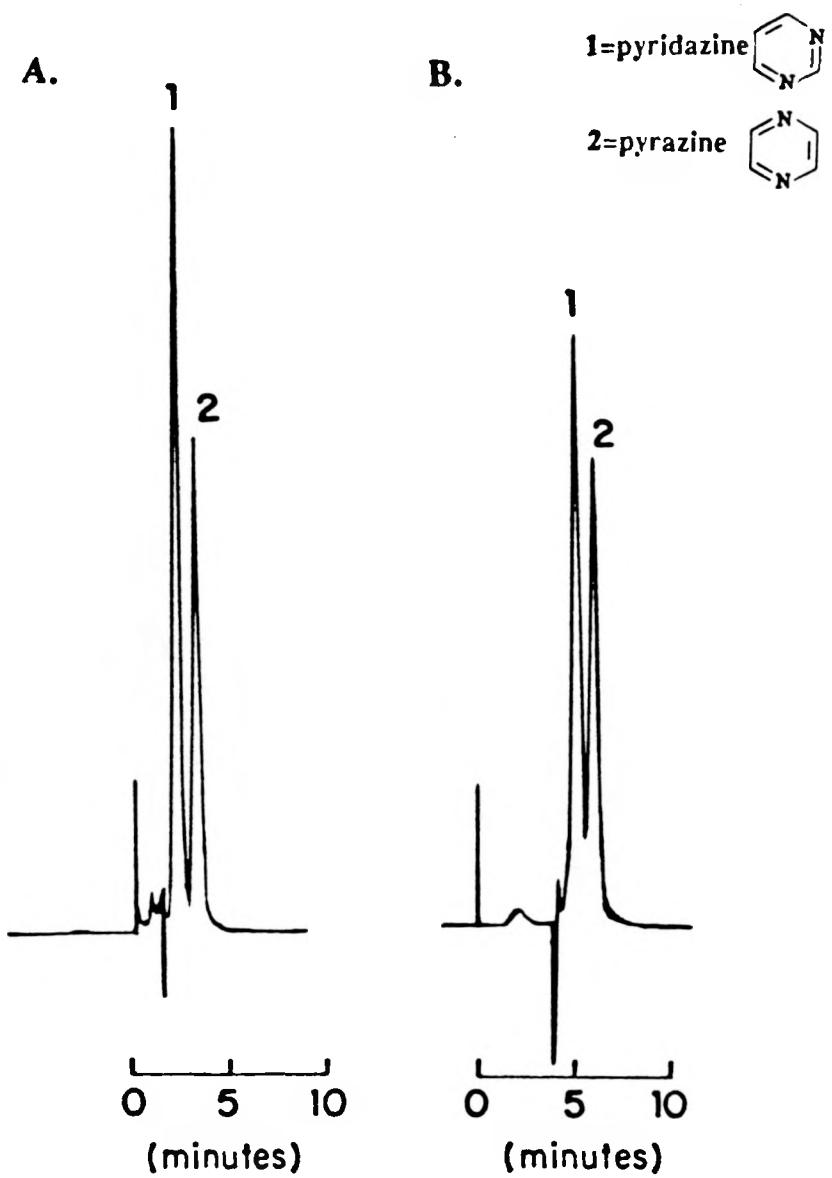


Figure 7. Separation of two isomers on: A. Sulfonated PS/DVB, 10 μm. Column: 100 x 4.6 mm. Eluent: 10% ACN + 0.1 M NH_4OH. B. Commercial SiC_{18}, 5 μm. Column: 250 x 4.6 mm. Eluent: 10% ACN

high pH value if the sample is in basic condition which the Si based column can not be used.

CONCLUSION

Sulfonated PS/DVB resin can be used for group separations of neutral and basic compounds. Also it can be used for separation of basic and weaker basic amines. The group separated compounds can be further separated by an HPLC system. The quantitative determinations were excellent for both groups. This method can also be used for sample preconcentration with almost quantitative recoveries and would be very useful in practical applications. The amine isomers can be separated by this sulfonated PS/DVB column and often the result was better than that on the commercial SiC18 column.

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SECTION IV. ABSTRACTION OF BASIC COMPOUNDS BY SULFONATED POLYSTYRENE DIVINYLBENZENE RESIN IN CAPILLARY GAS CHROMATOGRAPHY

INTRODUCTION

Sample clean up can be a critical step before gas chromatographic separation and quantitative determination for some samples. For example, gas chromatographic analysis of biological samples requires extensive sample clean up to remove interfering components and simplify the matrix. Various methods of removing impurities, which exist in the sample and interfere the analysis, are used in the practical analysis. The most widely used methods are liquid-liquid extraction and solid phase extraction [1, 2, 3]. By using these methods, compounds to be determined or impurities to be removed can be extracted for the further analysis of the sample.

Although gas chromatograhy is one of the most powerful separation methods known, scientists are constantly looking for ways to achieve greater selectivity. One popular approach is to use a selective detector, such as nitrogen or phosphorous detector. Another method is to separate sample components into various groups by solvent extraction or by the use of ion-exchange columns [4, 5]. Some type of an on-line preliminary group separation would be faster and more convenient. Several workers have incorporated a short, packed column placed just before or just after GC column, to remove various sample constituents.

These include copper salts as an amine abstractor [6], liquid FFAP to remove aldehyde [7], phosphoric acid for removing nitrogen amines [8] and epoxides [9], boric acid for abstracting alcohols [10], and sulfuric acid, mercuric perchlorate or molecular sieves for removing olefins and paraffins [11]. This latter arrangement has always been used in conjunction with packed GC column.

Since the rapid development of polymeric resins and highly improved techniques of synthesis, a special type of PS/DVB is now available, which is 5 μm spherical, non-porous PS/DVB resin. In the present research, a small amount of this sulfonated polymeric resin is placed in the injection split liner. This completely removes basic organic compounds while allowing neutral sample components to pass into the GC column. A capillary gas chromatographic column gives excellent separation of the neutral sample compounds. So far as we can determine, this represents the first time a vapor-phase reaction in the injection split liner has been used to quantitatively exclude a major class of organic compounds from separation by capillary GC.

EXPERIMENTAL

Preparation of sulfonated polystyrene divinylbenzene resins

Several different kinds of polystyrene divinylbenzene (PS/DVB) resins, which included porous, non-spherical XAD-4 (Rohm and Haas Co., Philadelphia, PA), porous spherical resin (Sarasep Inc, Santa Clara, CA) and non-porous spherical resin (Rohm and Haas Co.), were used for sulfonation. Resin were cleaned by acetonitrile, ethyl acetate and hexane thoroughly several times and then dried. About 3g resin was put in the round bottom flask, 30 ml of concentrated sulfuric acid was placed into the flask. The reaction temperature was kept at 50^{o}C with stirring. The reaction time varied for each resin. The porous, non-spherical XAD-4 and porous spherical resins took about 20 minutes to get about 1 Meq./g capacity. The non-porous spherical resin took about 24 hours to make surface area sulfonated as much as possible and get about 0.1 Meq./g capacity. After sulfonation, the resins were washed thoroughly with deionized water, dried and ready for use. The capacity was determined by titration.

Apparatus and chemical

A HP 5880A gas chromatography instrument with a flame ionization

detector, a HP 5880 series level 4 integrator, and a HP 7673 automatic sampler (Hewlett-Packard Co., Avonadale, PA), were used for separation of organic compounds and integration. The gas chromatography column used for separation was Supelco fused silica capillary columns (30 m, 0.32 mm ID, 0.25 μm film thickness, Supelco, Inc., Bellefonte, PA). Split mode was used in separation and abstraction.

The chemicals used were all reagent grade or analytical grade, used in purchased form. The solvent used to dissolve the chemicals was acetone. Laboratory distilled water was further deionized by Barnstead Nanopure II system (Sybron Barnstead, Boston, MA).

Abstraction of basic amines from a mixture of basic and neutral compounds

A split injection was used in this method. The split liner in the gas chromatograph instrument was used for pre-column abstractor holder. The sulfonated resins were placed in the split liner to form an even column of different length from 2 mm to 5 mm, which was supported and covered with glass wool. The samples which contained amines and neutral compounds of about 0.3 ppm concentration were injected into the liner by automatic sampler and vaporized quickly. The basic compounds were retained on the sulfonated resin in the liner, while the neutral compounds passed through and were then separated in the GC column. If

silicone resin was placed in the split liner instead of sulfonated PS/DVB resin, both basic amines and neutral compounds would pass through. Metal ions, silver and copper, replaced the hydrogen form and were tried, the results of which will be discussed later.

Gas chromatography

Split injection was used. Samples were made up in acetonitrile and a 1 μm volume was injected at a split ratio of 1:80. The samples contained approximately 0.3 ppm of each individual compound. For base abstraction experiments sulfonated resins were placed in the split liner. The injection port temperature was usually 200°C. For samples containing a large number of compounds, the following temperature program was used: 65°C for 2 minutes, then 15°C/min up to 245°C with a 5 minute hold. For samples containing base:neutral in a 200:1 ratio, the following program was used: 50°C for 2 minutes, then 2°C/min up to 65°C, with a 5 minute hold.

RESULTS AND DISCUSSION

Type of resins used for abstraction

The principal of the proposed abstraction of organic bases is as follows. Injection of a sample containing bases (B) and neutral compounds (N) converts both sample types to the gaseous state. Bases are expected to react with the solid resin to form cations that are retained by the resin.

$$\text{Resin-}SO_3^-H^+ + NR_3 \longrightarrow \text{Resin-}SO_3^-HNR_3^+$$

Gaseous neutral compounds will not react and should pass onto the GC column for separation.

Several different types of sulfonated polystyrene divinylbenzene resins, which were porous non-spherical XAD-4, porous spherical, and non-porous spherical resins, were tried. Only the non-porous spherical sulfonated PS/DVB resin was successful in abstraction of basic amines. When the mixture of basic and neutral compound was injected into the liner packed with sulfonated <u>porous</u> PS/DVB, both the basic and neutral compounds were retained on the liner since the sulfonated porous PS/DVB resin had surface area of both hydrophilic parts (sulfonated parts) and

hydrophobic parts (unsulfonated parts in pores which were impossible to be totally sulfonated). However, when the same mixture of basic and neutral compounds was injected into the liner packed with sulfonated non-porous PS/DVB resin, only the basic compounds were retained, while the neutral compounds passed. This is probably because that the surface area of this resin is much less than that on the porous PS/DVB resin and could be totally sulfonated by long sulfonation (24 hours). Therefore it has only sulfonated surface area which is unable to retain neutral compounds, but able to retain basic compounds.

Three typical PS/DVB resins were tried as listed in Table I. Though the capacity on the non-porous sulfonated PS/DVB (about 0.1 meq./g), is lower than that on the porous sulfonated PS/DVB (about 1 meq./g), the surface area unit capacity (meq./m^2) was much higher. XAD-4 resin has surface area 720 m^2/g and capacity 1 meq./g, so the surface area unit capacity is 1.4×10^{-3} meq./m^2 (equivalent to 0.84 sulfonic functional group per nm^2). Sarasep resin has surface area 415 m^2/g and capacity 1 meq./g, so the surface area unit capacity is 2.4×10^{-3} meq./g (equivalent to 0.14 sulfonic functional group per nm^2). While the non-porous spherical resin (5 μm) has calculated surface area about 1.1 m^2/g and capacity 0.1 meq./g, the surface area unit capacity

Table I. Characteristics of sulfonated PS/DVB resins

Resins (Sulfonated PS/DVB)	Surface Area	Capacity	Capacity/Unit Area
Porous, non-spherical, 150 μm (XAD-4, Rohm & Haas Co.)	720 m^2/g	1 mequi./g	0.0014 mequi./m^2 (0.084 gp/nm^2)
Porous, spherical, 10 μm (Sarasep Inc.)	415 m^2/g	1 mequi./g	0.0024 mequi./m^2 (0.14 gp/nm^2)
non-porous, spherical, 5 μm (Rohm & Haas Co.)	1.1 m^2/g	0.1 mequi./g	0.091 mequi./m^2 (5.5 gp/nm^2)

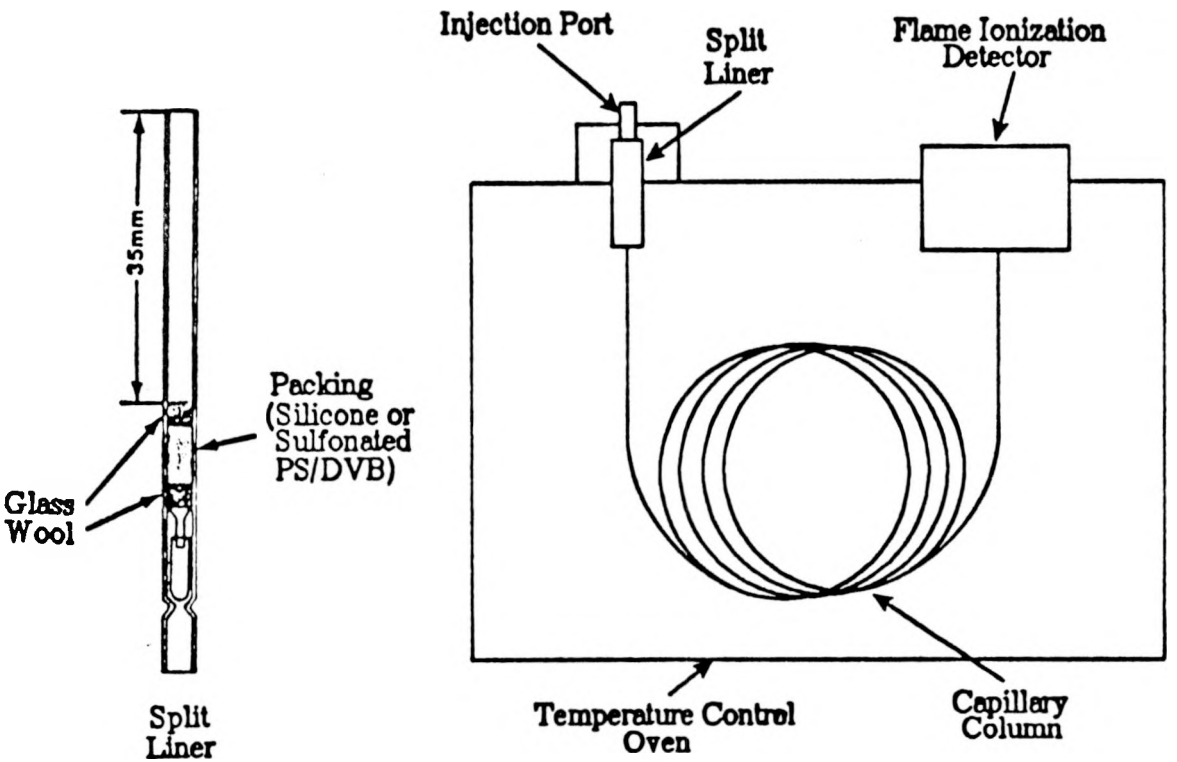

Figure 1. Apparatus for abstraction of amines in GC

is about 9.2×10^{-2} meq./m^2 (equivalent to 5.5 sulfonic functional group per nm^2), which is almost 66 times of that on XAD-4 resin and 38 times of that on Sarasep resin.

Injection temperature

Considering the possible effect of high temperatures on sulfonated PS/DVB resin in the liner, low injection temperatures, such as 100°C, were tried at first. Since the evaporation was not quick enough and the chromatographic separation was not satisfied. Several different injection temperatures were then tried. The chromatographic separation was not satisfied until the temperature got to 200°C. After a long period of use (several weeks) at 200°C, no decrease in the abstraction of basic amines by resins was observed. So the temperature of 200°C was chosen as the injection temperature.

The amount of sulfonated resin in the liner

The inside diameter of the liner is about 5 mm (see Figure 1). Different amounts of sulfonated PS/DVB resins, about 100 mg, 200 mg and 300 mg which formed columns in the liner of about 2 mm, 4 mm and 6 mm, were tried. No effects on the chromatographic separation were observed. Since the capacity of the resin (even with the least amount resin) was

much larger than the amount of the amines to be abstracted (more than 500:1), there were no problems of overloading. In the liner packed with 200 mg resin, 50 samples were injected, and no over loading was observed. Since it takes only a few minutes to change the liner, this method is very practical for abstraction of amines in routine separation.

Abstraction of basic amines

Thirteen basic compounds, which included cyclohexylamine, 3-picoline, 3-ethylpyridine, sym-collidin, 2-aminopyridine, quinoxaline, 2,6-dimethylquinoxaline, tributylamine, benzothiazole, quinoline benzylamine, diethylenetriamine and benzylpyridine, and ten neutral compounds, which included ethylcrotonate, propylbenzene, phenol, octanol, octyl acetate, 4-propylphenol, 3-phenyl propanol, nonanol, 2,4-dibromophenol and 3-nitrophenol, were used to make a mixture solution in acetone. Also, another solution of neutral compounds of exact same concentration was made. The mixture solutions of basic and neutral compounds was injected into the gas chromatograph with liners of two different packing materials, sulfonated polystyrene divinylbenzene and silicone (3% silicone OV-1, Alltech Associates, Arlington Heights, IL) while all the other chromatographic conditions were the same (capillary

column, carrier gas flow rate, temperature programming, etc.). The results were shown in Figures 2 and 3.

When the silicone liner was used, all the compounds in the mixture solutions, both basic and neutral, passed through. Some of these basic and neutral compounds have close retention times and overlapped one another (see Figure 2 and table II)., Examples are compounds b (propylbenzene) and m (3-ethylpyridine), as well as h (nonanol) and u (diethylenetriamine).

When the sulfonated PS/DVB liner was used, only the neutral compounds passed while all the basic compounds were retained on the liner. So when the mixture sample of thirteen basic amines and ten neutral compounds was injected into system, all the amines were cleared away and those previously overlapped neutral compounds could be well determined (see Figure 3).

Figure 4 is the chromatogram of only ten neutral compounds with sulfonated PS/DVB liner. Comparing Figures 3 and 4, it is seen that the chromatogram of ten neutral compounds and the chromatogram of the mixture of ten neutral compounds and thirteen basic amines are almost the same. Benzylnitrile, which has a nitrogen atom and is not an amine, and also passes.

Table II. Retention time of tested compounds

Neutral compounds (t_r)	Basic compounds (t_r)
a=Ethyl crotonated (4.03)	k=Cyclohexylamine (4.30)
b=Propylbenzene (5.68)	l=Picoline (4.30)
c=Phenol (5.85)	m=Ethylpyridine (5.66)
d=Octanol (7.06)	n=Sym-collidin (6.09)
e=Octyl acetate (8.03)	o=2-Aminopyridine (6.22)
f=p-Propylphenol (8.27)	p=Quinoxaline (6.42)
g=3-Phenylpropanol (9.04)	q=2,6-Dimethylquinoxaline (6.51)
h=Nonanol (9.24)	r=Tributylamine (8.57)
i=2,4-Dibromophenol (10.41)	S=Benzothiazole (8.92)
j=m-Nitrophenol (11.39)	t=Quinoline (9.11)
	u=Benzylamine (9.25)
	v=Diethylenetriamine (10.96)
	w=Benzylpyridine (11.62)

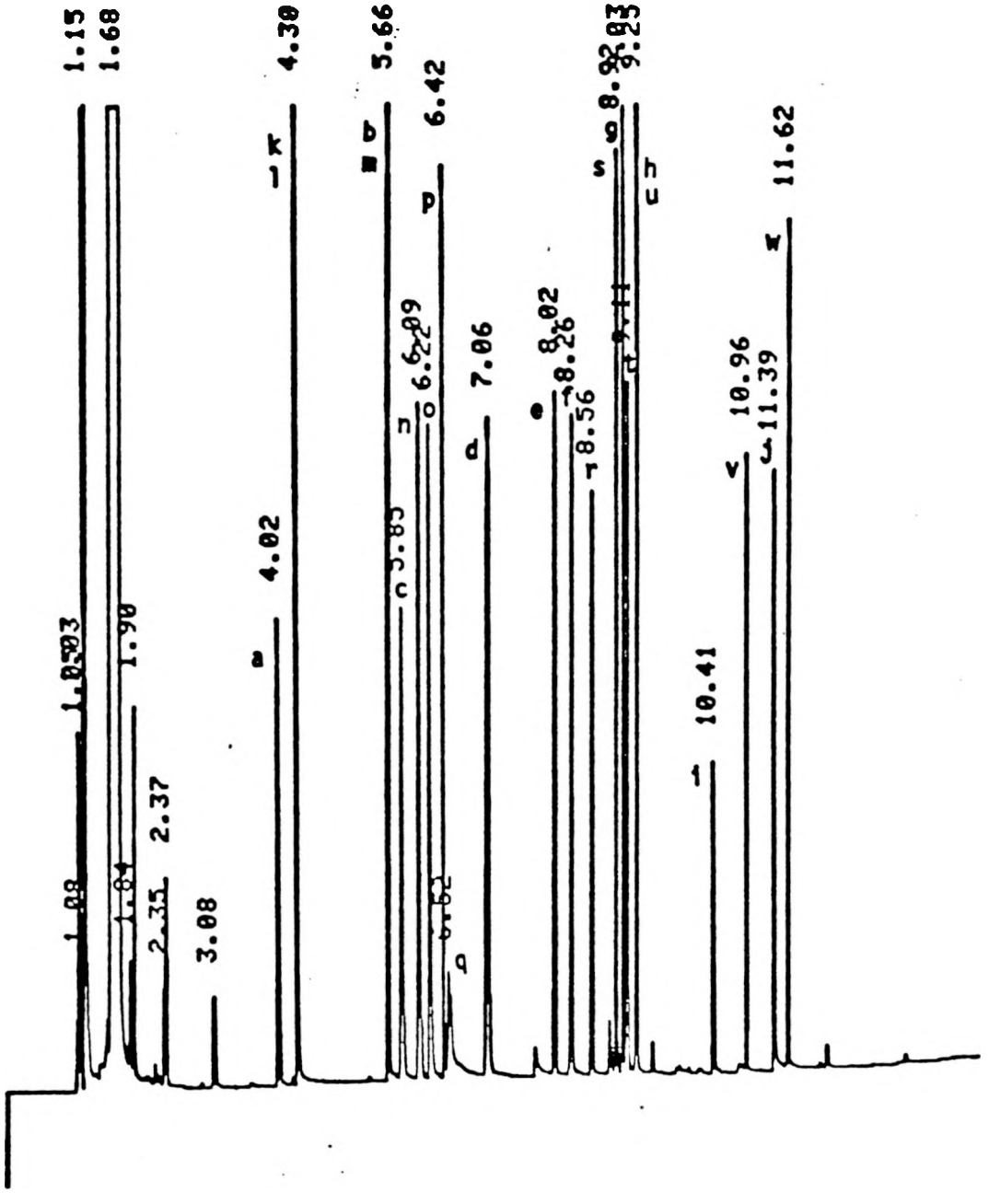

Figure 2. Chromatogram of thirteen basic amines and ten neutral compounds using the liner with silicone

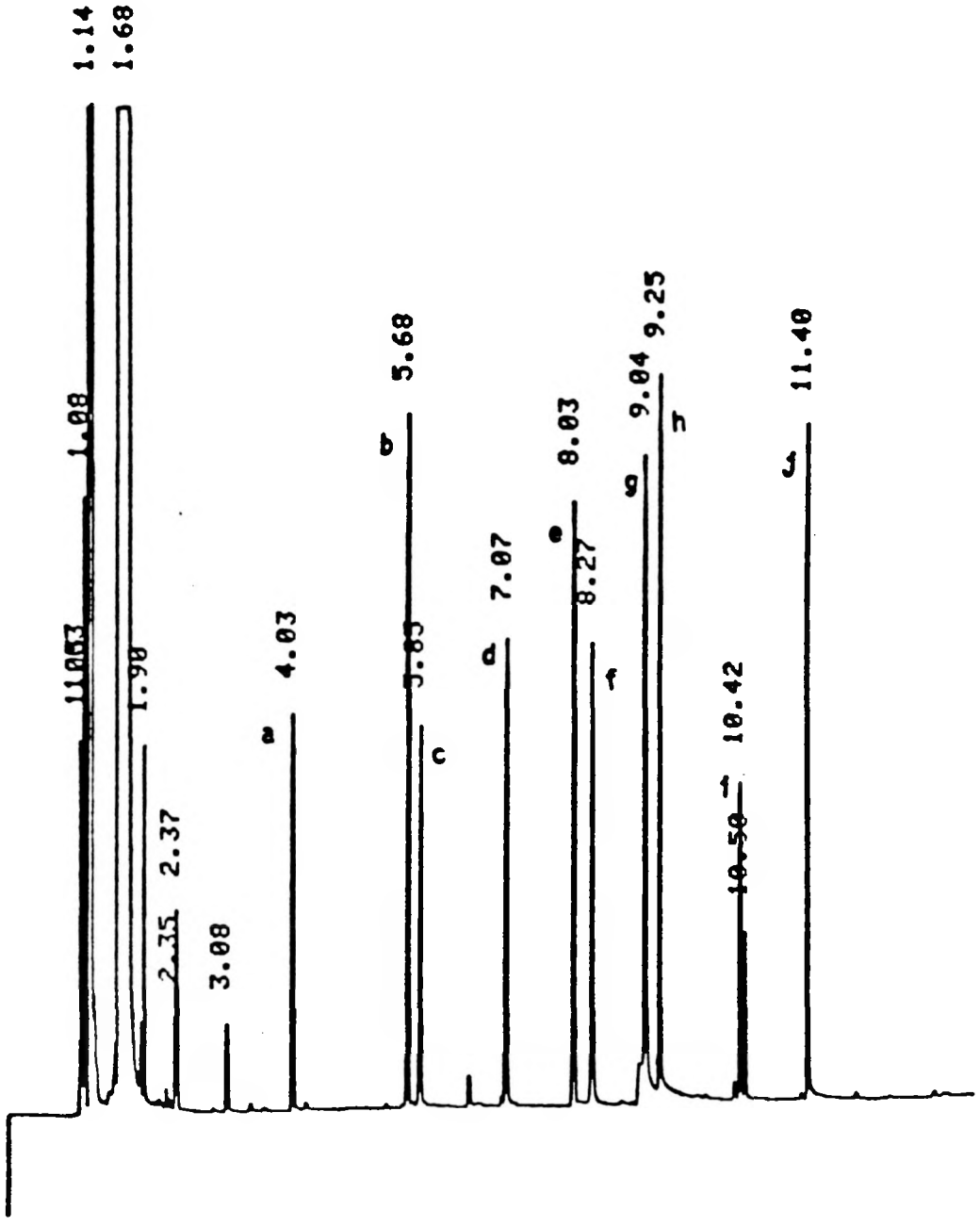

Figure 3. Chromatogram of the same compounds in fig.3 using the liner with sulfonated non-porous PS/DVB resin. Only ten neutral compounds passed through

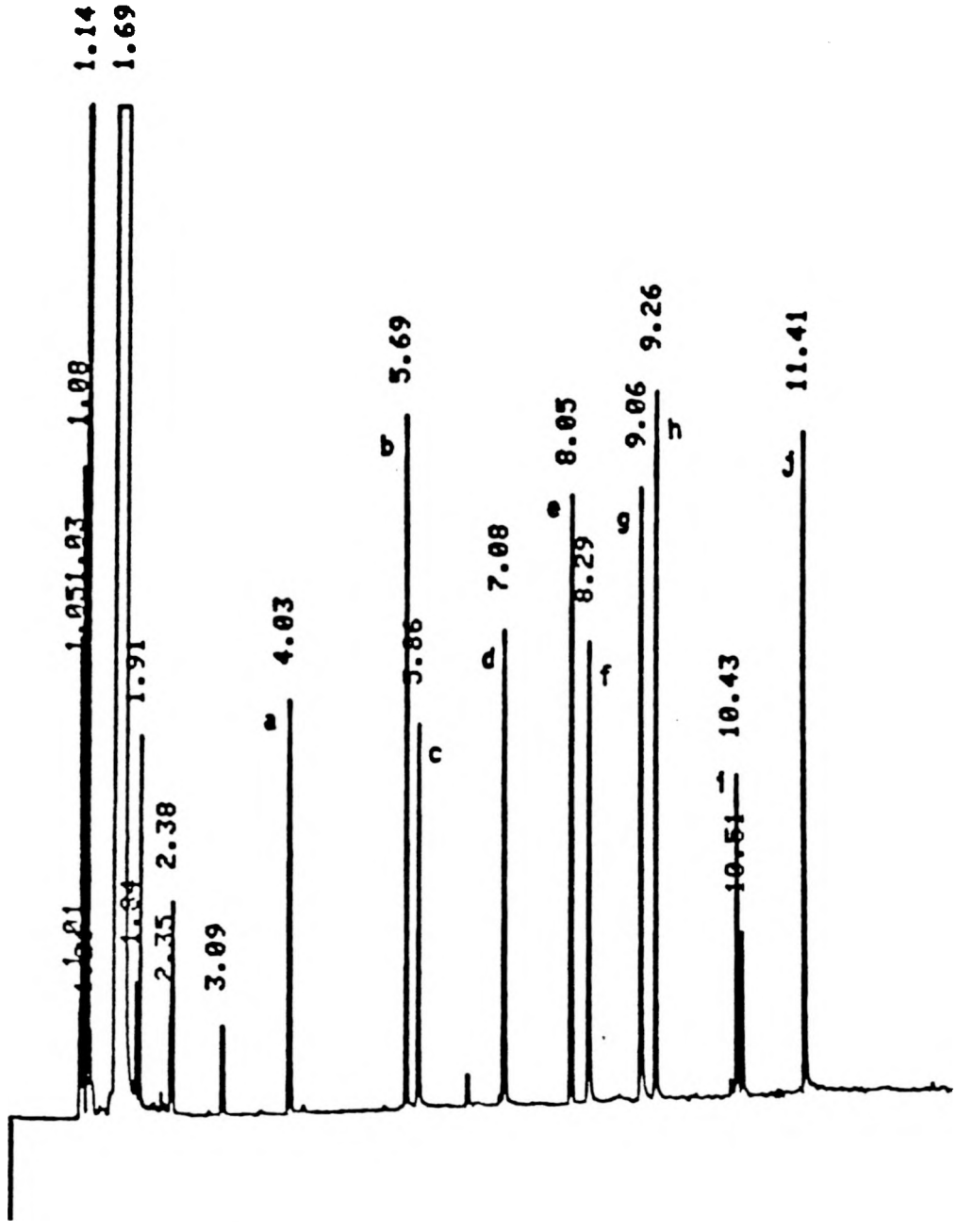

Figure 4. Chromatogram of ten neutral compounds using the liner with sulfonated non-porous PS/DVB resin

Different cation forms of this sulfonated PS/DVB resin were tried. Ag^{+} and Cu^{2+} were replaced H^{+} form and the cationic form resins were placed in the liner. The thirteen amines were injected into the gas chromatography instrument. For both cation forms, nine amines were absorbed on the resin (cyclohexylamine, 3-picoline, 3-ethylpyridine, sym-collidin, 2-aminopyridine, quinoxaline, 2,6-dimethylquinoxaline, tributylamine, benzylamine), while three amines passed through the liner (benzothiazole, quinoline, diethylenetriamine). One amine, benzylpyridine, was partially absorbed.

It is interesting to compare the retention of amines by sulfonated PS/DVB resin in liquid chromatography and gas chromatography [15]. In gas chromatography, when sulfonated PS/DVB resin was packed into the split liner in the injection port, both relatively strong amines (quinoline and 2-benzylpyridine) and relatively weaker amines (quinoxaline and benzothiazole) were absorbed on the liner. However, in liquid chromatography, only the relatively stronger amines are retained on the mini-column. One reason is that the liquid solvent is a stronger mobile phase carrier than gas.

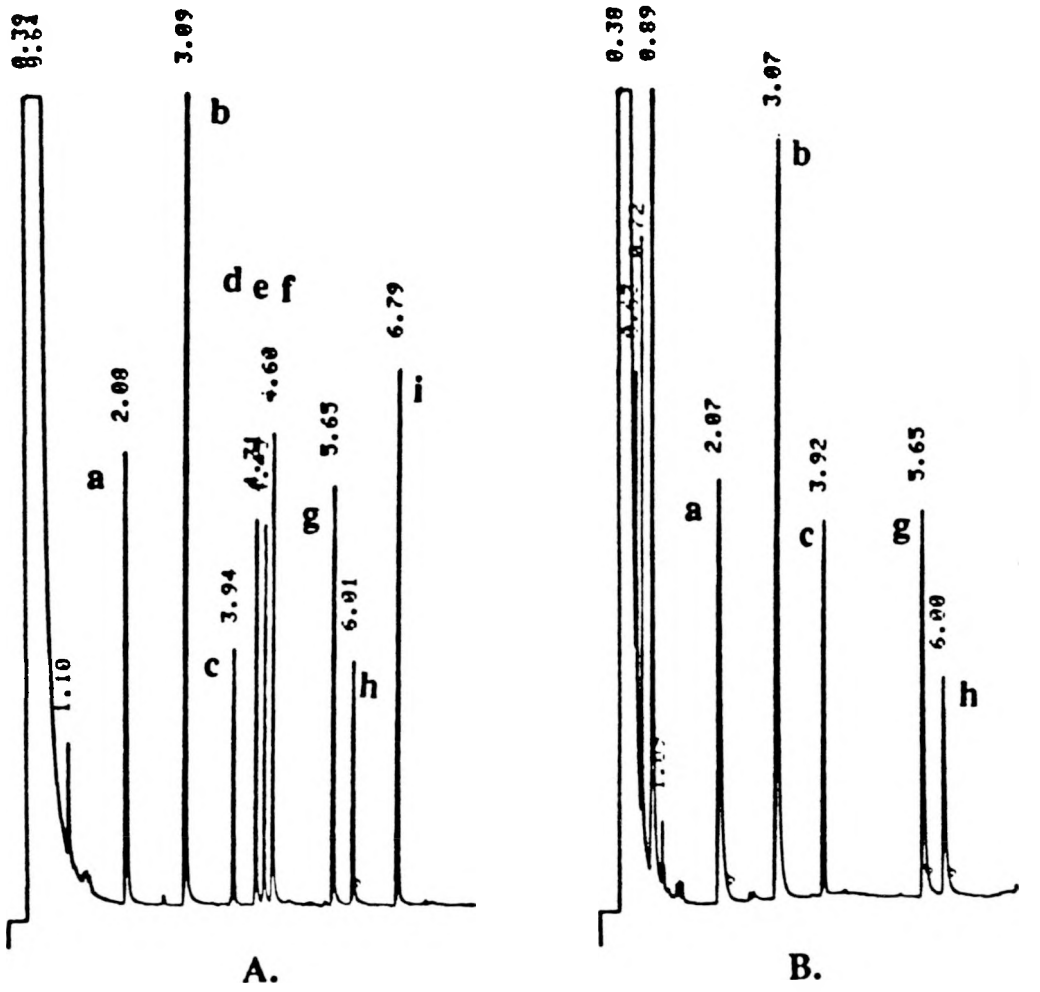

Figure 5. Quantitative determination. A. Using the liner with silicone. B. Using the liner with non-porous sulfonated PS/DVB. Basic compounds: d=quinoxaline, e=benzothiazole, f=quinoline, i=2-benzylpyridine. Neutral compounds: a=benzylnitrile, b=p-cresol, c=octyl acetate, g=2,4-dibromophenol, h=2,4-dinitrofluorobenzene.

Table III. Linear regression data for plots of relative area of various compounds against the concentration of these compounds, using a silicone liner

Basic Compounds	Corr.(v)	Slope	y-Int.
Quinoxaline	0.99994	29.9	0.072
Benzothiazole	0.99993	30.0	0.022
Quinoline	0.99992	30.0	0.083
2-Benzylpyridine	0.99993	30.0	0.051
Neutral compounds			
Benzylnitrile	0.99999	30.7	0.080
Octyl acetate	0.99998	30.0	0.082
2,4-Dibromophenol	0.99996	29.7	0.168
2,4-Dinitro-fluorobenzene	0.99998	29.5	0.285

Table IV. Linear regression data for plots of relative area of neutral compounds against the concentration of these compounds, using a sulfonated PS/DVB resin liner

Neutral compounds	Corr.(v)	slope	y-Int.
Benzylnitrile	0.99991	51.5	-0.093
Octyl acetate	0.99997	50.6	-0.017
2,4-Dibromophenol	0.99995	49.6	0.198
2,4-Dinitro-fluorobenzene	0.99992	48.0	0.539

Quantitative determination using sulfonated PS/DVB resin liner

A mixture of four basic compounds (quinoxaline, benzothiazole, quinoline and 2-benzylpyridine) and four neutral compounds (benzylnitrile, octyl acetate, 2,4-dibromophenol and 2,4-dinitrofluorobenzene) was made with an internal standard compound p-cresol (see Figure 5). A series of different concentrations of these compounds were prepared. The solutions were injected into the gas chromatograph with both silicone and sulfonated PS/DVB liners. With the silicone liner, all the compounds passed through. The relative area of each compound was plotted against its concentration, the linear correlation coefficients were calculated (see table III), and found all to be 0.9999_{+}. While with the sulfonated PS/DVB liner, only the neutral compounds passed and the linear correlation coefficients were obtained, also being 0.9999_{+} (see Table IV). The slopes of same compound but with different liner were different. This was because that the different resins and amounts in the liner affected the gas flow and split ratio. The abstraction of amines by sulfonated PS/DVB resin in the liner would not effect the quantitative determination of neutral compounds in mixture of basic and neutral compounds.

Determination of neutral compound in the presence of large amount of basic compounds

Sample clean up is a very important step for "dirty" samples before the separation and determination. It can be a tedious and difficult work, even impossible at sometime. Part of the compound to be determined can be lost during the cleanup step, causing inaccuracy in the determination. This is especially true when trace amounts of compounds are to be determined. Abstraction of amines from a mixture of basic and neutral compounds by sulfonated PS/DVB resin could be very useful for practical quantitative determination, making sample cleanup unnecessary. Since the capacity of the sulfonated PS/DVB resin was relatively large in comparason with the amount of samples injected, a large amount of amines in the mixture can be tolerated and extracted. Two pairs of basic and neutral compounds, 3-picoline and ethylcrotonate, and 3-ethylpyridine and propylbenzene, were chosen to test this concept. Figure 6.A and B shows that a large excess of 3-picoline has a considerable effect on the peak height of ethyl crotonate with ordinary silicone resin liner, even though the retention times are somewhat different. However, with the sulfonated resin liner the ethyl crotonate peak height is almost the same in the presence and absence of a 200-fold excess of the base (Figure 6.C and D).

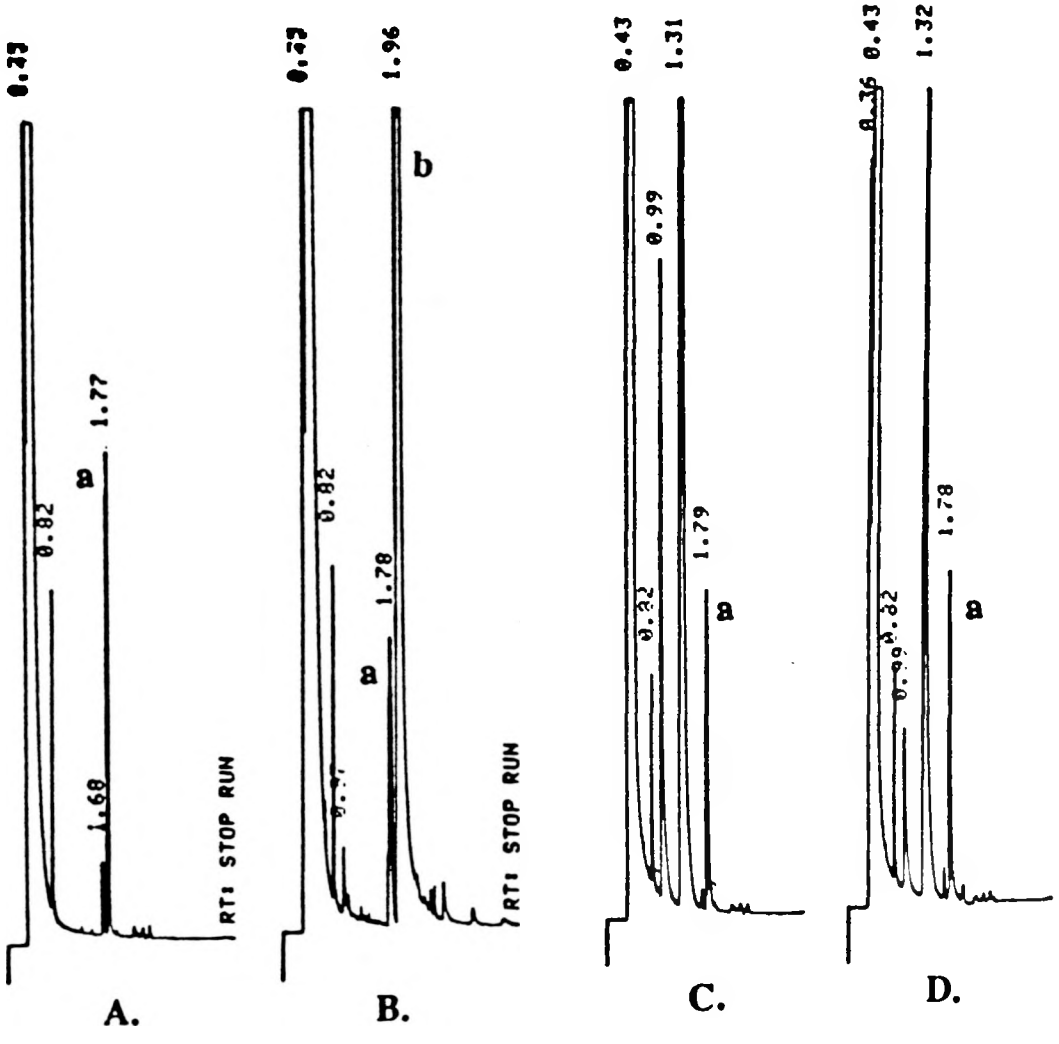

Figure 6. A and B using liner with silicone. A. Only neutral compound a=ethyl crotonate (t_r=1.77-1.79). B. Ethyl crotonate with 200 time concentration of basic compound b=3-picoline (t_r=1.96). C and D using liner with sulfonated PS/DVB resin. C. Same as A. D. Same as B.

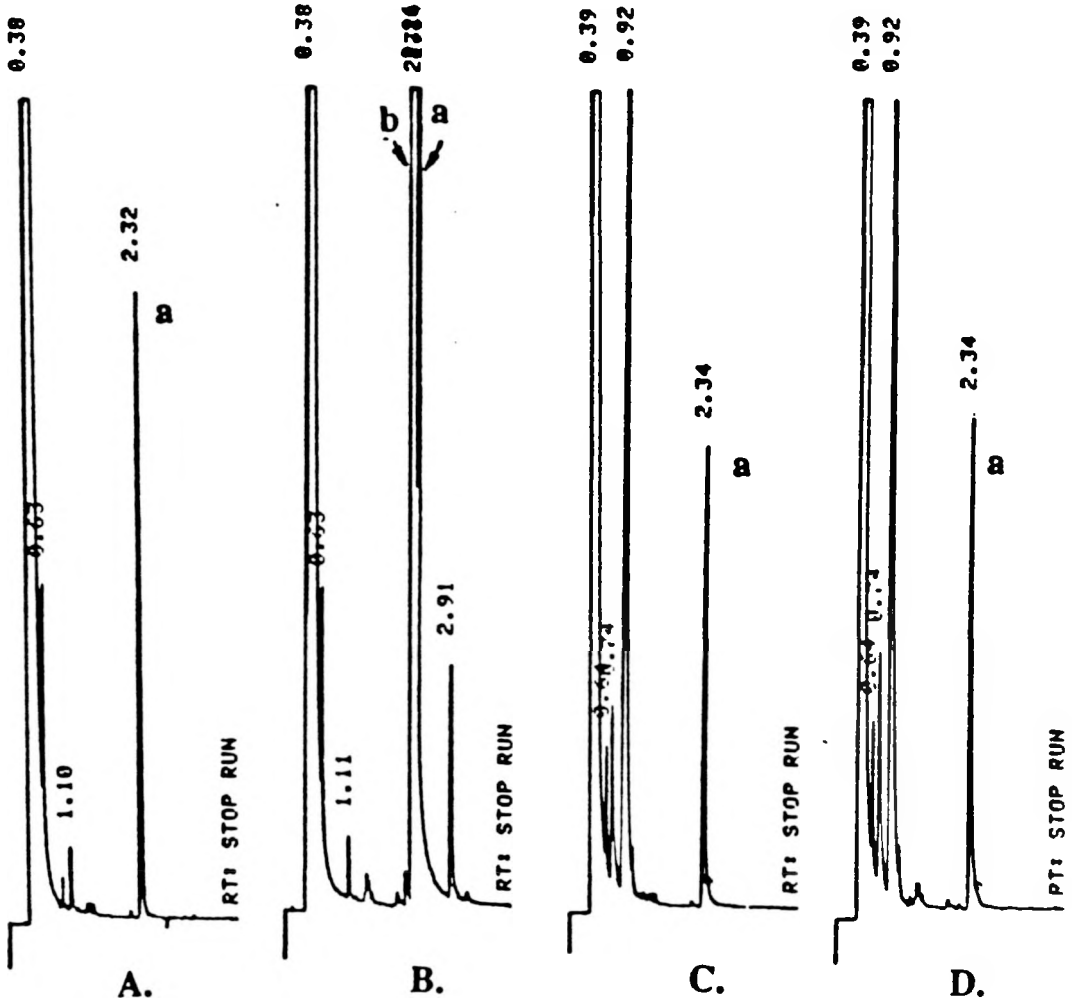

Figure 7. A and B using the liner with silicone. A. Only neutral compound a=propylbenzene (t_r=2.32-2.34). B. Propylbenzene with 200 time concentration of basic compound b=3-ethylpyridine (t_r=2.28). C and D using the liner with sulfonated PS/DVB resin. C. Same as A. D. Same as B.

A neutral compound and base with very similiar retention times was considered next. Figure 7.B shows very poor resolution of mixture of 3-ethylpyridine and n-propylbenzene in a 200:1 molar ratio. However, with the sulfonated resin liner the n-propylbenzene peak height is almost the same when injected alone or with 200 times as much 3-ethylpyridine (Figure 7.C and D).

While a mole ratio of base:neutral of 200 always gave a peak only for the neutral compound, a mole ratio of base:neutral of 500:1 resulted in only partial retention of the basic compound. Increasing the depth of resin in the liner did not greatly improve retention of the base. The resin is not being overloaded by injection of a large amount of a basic compound, because subsequent injections of smaller samples of a base resulted in complete abstraction of the base chromatographic peak. The limitation in abstracting ability seems to be kinetic in that the vapor surge through the split liner is too rapid to retain extremely large amounts of basic compounds.

CONCLUSION

Non-porous sulfonated PS/DVB resin is an excellent abstractor for amines in gas chromatography. The neutral compounds could be well separated in the presence of many basic amines which have similar retention times to the neutral compounds. This special type of sulfonated PS/DVB resin does not affect the quantitative determination of neutral compounds. Also the presence of a 200-fold excess of amines in the sample solution was tolerated and cleaned up by the liner packed with this resin. It would be useful in practical separations of amines and neutral compounds and in quantitative determination of neutral compounds in the "dirty" samples without sample clean up.

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SECTION V. SELECTIVE ABSTRACTION OF MERCAPTAN COMPOUNDS BY CHEMICALLY MODIFIED POLYMERIC-MERCURIC RESIN IN CAPILLARY GAS CHROMATOGRAPHY

INTRODUCTION

Selective removal of one group of compounds is at times a helpful way to solve difficuties in liquid and gas chromatography. A pre- or post- minicolumn is usually used to remove a special group of compounds. In gas chromatography, various inorganic salts or acids, either directly used or coated on some stationary phase, have been used as abstractors. Examples are copper salts as an amine abstractors [1], phosphoric acid for removal of amines [2] and epoxides [3], boric acid for abstracting alcohols [4], and sulfuric acid and mercuric perchlorate for removing olefins and paraffins [5]. As discribed in section IV, a special sulfonated resin was used as an amine abstractor in capillary gas chromatography.

In liquid chromatography, immobilized Hg (II) on 8-hydroxyquinoline (oxine) has been used for selective trace enrichment and cleanup of 2-mercaptobenzimidazole [6], and Pt (IV) coated on 2-amino-1-cyclopentene-1-dithiocarboxylic acid (ACDA) has been used for retaining anilines [7].

In the present research, a chemically modified polymeric-mercuric resin was synthesized and a small amount of this resin was placed in the injection split liner to act as a mercaptan abstractor. This resin selectively and completely removed the mercaptans while allowing non-

mercaptans, which included both surfur and non-sulfur compounds, to pass through. Non-mercaptan compounds were well separated and determined quantitatively. We believe it was the first time that a chemically modified polymeric-mercuric resin was used as a gas phase abstractor to selectively remove mercaptan compounds.

EXPERIMENTAL

Preparation of polymeric-mercuric resins

Several different kinds of polystyrene divinylbenzene (PS/DVB) resins, which included porous spherical Amberchrome 161 (about 50 μm and 720 m^2/g surface area, Rohm and Haas Co., Philadelphia, PA), porous spherical resin (about 10 μm and 415 m^2/g surface area, Sarasep Inc, Santa Clara, CA) and non-porous spherical resin (about 5 μm, Rohm and Haas Co.), were used for the preparation of a polymeric-mercuric resin. The resins were cleaned by acetonitrile, ethyl acetate and hexane thoroughly, and dried. The reaction included two steps. First, about 3 g resin and 4.8 g mercuric acetate were placed in a round bottom flask. 40 ml of acetic acid was added into the flask. The reaction temperature was kept at 130°C with stirring and tap water as condensation for two hours in an oil bath. The first step was finished. The chemical reaction was as follows:

$$\text{PS/DVB-}\Phi + \text{Hg(OAC)}_2 \xrightarrow[130^\circ\text{C}]{\text{HAc}} \text{PS/DVB-}\Phi\text{-HgOAC} + \text{HAc}$$

Φ — Benzene ring on PS/DVB resin.

After the first step of the reaction, the resin was filtered,

washed with methanol and deionized water and dried. The cleaned resin that has undergone the first reaction was placed in a round-bottom flask. 40 ml of a mixture of methanol (80%) and pyridine (20%) was placed in the flask. 40 ml of sodium chloride solution (10%) was added to the flask dropwise. The reaction temperature was kept at 80°C for 24 hours and the resin was kept stirring. Then the resin was filtered, washed with methanol, water, acetone and dried. The resin is ready for use. The chemical reaction of the second step is as follows:

$$\text{PS/DVB-}\Phi\text{-HgOAc} + \text{NaCl} \xrightarrow[80^{\circ}\text{C}]{\text{Methanol, Pyridine}} \text{PS/DVB-}\Phi\text{-HgCl} + \text{NaOAc}$$

The capacity of this chemically modified polymeric-mercuric resin was determined as 1.49 mmole/g by mercuric elemental analysis (29.81% by mass).

Apparatus and chemical

A HP 5880A gas chromatograph with a flame ionization detector, a HP5880 series level 4 integrator, and a HP 7673 automatic sampler (Hewlett-Packard Co., Avonadale, PA), were used for separation of organic compounds and integration. The column used for separation was a Supeleco fused silica capillary column (30 m, 0.32 mm ID, 0.25 μm film

thickness, Supelco, Inc., Bellefonte, PA). Split mode was used in separation and abstraction.

The chemicals used were reagent grade or analytical grade, used in purchased form. The solvent used to dissolve the chemicals was acetonitrile. Laboratory distilled water was further deionized by a Barnstead Nanopure II system (Sybron Barnstead, Boston, MA).

Capillary gas chromatography

A split injection was used in this method. The split ratio was 1:80. The split liner was used as the pre-column abstractor holder (see Figure 1 in section IV). The chemically modified polymeric-mercuric resin was placed in the split liner to form a even column of different lengths of about 2 mm to 5 mm , which was supported and covered with glass wool. The injection temperature was 200°C. Various injection temperatures were tested and the results will be discussed later. A temperature program was used for separation, which was 65°C for 2 minutes, then 10°C/min up to 250°C with a 5 minute hold.

The sample solution contained both mercaptan and non-mercaptan compounds, which includes sulfur (about 3 ppm each) and non-sulfur compounds (about 0.1 ppm each). 1 μl of sample solution was injected into the liner. The mercaptan compounds were retained on the chemically

modified polymeric-mercuric resin while the non-mercaptan compounds (both sulfur and non-sulfur compounds) passed through the liner to be separated by the capillary column.

RESULTS AND DISCUSSION

Mercuric resin

It is well known that the mercaptan group can form very strong complexes with mercury. Injection of sample solution which contains mercaptan and non-mercaptan compounds into the split liner of gas chromatography converts both types to the gaseous state. If the liner is packed with non-porous polymeric-mercuric resin, the mercaptan compounds react with the mercuric functional group on the resin to form a strong complex compounds and therefore were retained on the resin. However, the non-mercaptan and other non-sulfur compounds can't form such complex compounds and passed through the liner.

Although several different kinds of PS/DVB resin were used for synthesis of polymeric-mercuric resins as mentioned in the experimental part, only the non-porous polymeric-mercuric resin selectively abstracted mercaptan compounds. The porous polymeric-mercuric resin retained non-mercaptan compounds too strongly, either totally absorbing or severely tailing the non-mercaptan compounds. This is due to the large surface area of the porous resins. However, the surface area of non-porous resin is only about 1.1 m^2/g. Since the capacity is 1.49 mmol/g, all the surface area is covered by the mercuric functional

groups, which can retain only mercaptan compounds by forming complexes. The capacity of mercuric functional group on the non-porous PS/DVB resin was determined as 1.49 mmol/g by mercuric elemental analysis. The non-porous PS/DVB resin used in this research is 5% crosslinked, which means the resin is composed of 5% divinylbenzene and 95% styrene. The capacity of benzene ring can be calculated as follows.

The mass percentage of benzene ring in styrene is 77/104 = 74.0%, which is equivalent to 9.62 mmol/g. The mass percentage of benzene ring in divinylbenzene is 76/130 = 58.5%, which is equivalent to 7.69 mmol/g.

If η is the crosslink percentage in the PS/DVB resin, the capacity of benzene ring (mmol/g) in 1 gram of crosslinked PS/DVB resin can be expressed as:

$$\text{mmol benzene ring/g resin} = (1-\eta) \times 9.62 + \eta \times 7.69$$

$$= 9.62 - 1.93\eta$$

Since the polymeric-mercuric resin contains 29.81% of mercury and 5.28% of chlorine, which are equivalent to 1.49 mmol/g, the capacity of benzene ring in this resin is

$$1\ g \times (1 - 29.81\% - 5.28\%) \times (9.62 - 1.93 \times 5\%)\ mmol/g$$

$$= 6.18\ mmol/g$$

So the reaction percentage of mercuric functional group on the benzene ring of the resin is

1.49/6.18 = 24.1%

That is, approximately one out of every four benzene rings had a mercuric functional group introduced. This might help to understand the ability of non-porous polymeric-mercuric resin to retain mercaptan compounds while not absorbing non-mercaptan compounds.

Abstraction of mercaptan compounds

Eight sulfur compounds, which included six mercaptan compounds (1-butanethiol, 2-mercaptoethanol, ethyl-2-mercaptoacetate, thiophenol, thiolactic acid, 3-mercapto-1,2-propanediol, and benzylmercaptan) and two non-mercaptan compounds (butyl sulfide and benzylmethylsulfide), were ramdomly picked and a solution was prepared in ethyl acetate. This is called sulfur compounds solution, and the concentration of each compound is about 0.3 ppm. Eight non-sulfur compounds (toluene, phenol, indene, p-cresol, p-ethylphenol, naphthalene, anthracene and dibutyphthalate) were ramdomly picked and a solution was prepared in ethyl acetate. This is called non-sulfur compounds solution, and the concentration of each compound is about 0.1 ppm. A third solution of exact the concentration of the sulfur and non-sulfur compounds was made in ethyl acetate, which is called mixture solution.

Two injection liners were prepared, one packed with "conventional" packing material (3% silicone OV-1, Alltech Associates, Aflington Heights, IL), the other packed with the polymeric-mercuric resin. The retention times of these compounds with the silicone liner under the gas chromatographic conditions mentioned before are listed in Table I.

When the non-sulfur compound solution was injected into the gas chromatograph with both liners (silicone and polymeric-mercuric resins), all the non-sulfur compounds passed through the liners and separated well.

When the sulfur compounds solution was injected into the gas chromatograph with silicone resin in the liner, all compounds passed through (see Figure 1). However, when the sulfur compounds solution was injected into the gas chromatography with polymeric-mercuric resin in the liner, only two compounds out of seven, butylsulfide and benzylmethylsulfide, passed through, while all other seven compounds, which are mercaptan compounds (2-mercaptoethanol, ethyl-2-mercaptoacetate, thiophenol, thiolactic acid, 3-mercapto-1,2-propanedilo and benzylmercaptan), were retained on the mercuric resin in the liner (see Figure 2).

Table I. Retention time (t_r) of sulfur and non-sulfur compounds

Sulfur Compounds	t_r
2-Mercaptoethanol	1.32
Ethyl-2-mercaptoacetate	2.40
Thiophenol	3.24
Thiolactic acid	3.24
3-mercapto-1,2-propanediol	4.05
Benzylmercaptan	4.99
Butylsulfide	5.11
Benzylmethylsulfide	6.33
Non-sulfur Compounds	
Toluene	1.40
Phenol	3.50
Indene	4.46
p-Cresol	4.94
p-Ethylphenol	6.38
Naphthalene	6.62
Anthracene	14.56
Dibutylphthalate	16.36

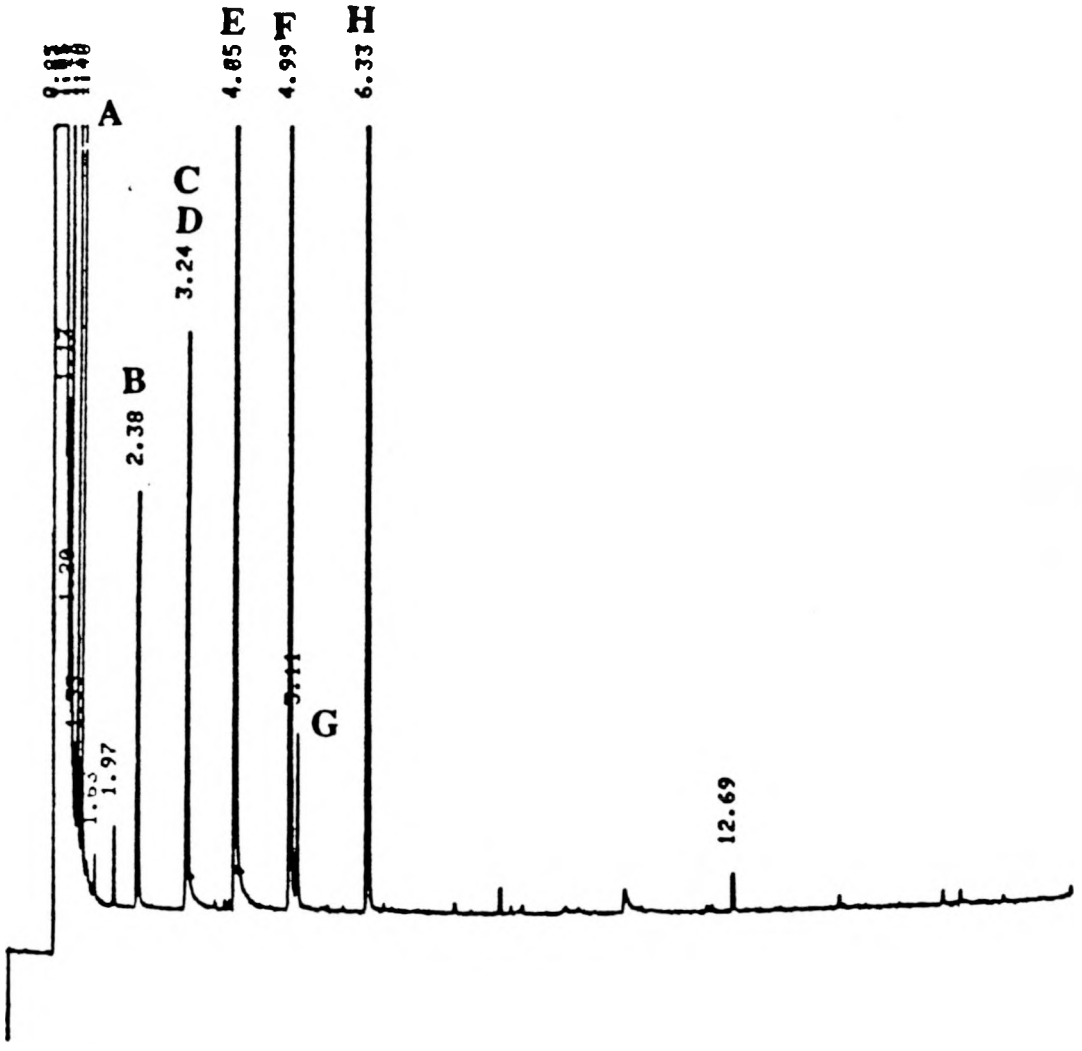

Figure 1. Chromatogram of sulfur compounds (both mercaptan and non-mercaptan compounds) with silicone resin liner

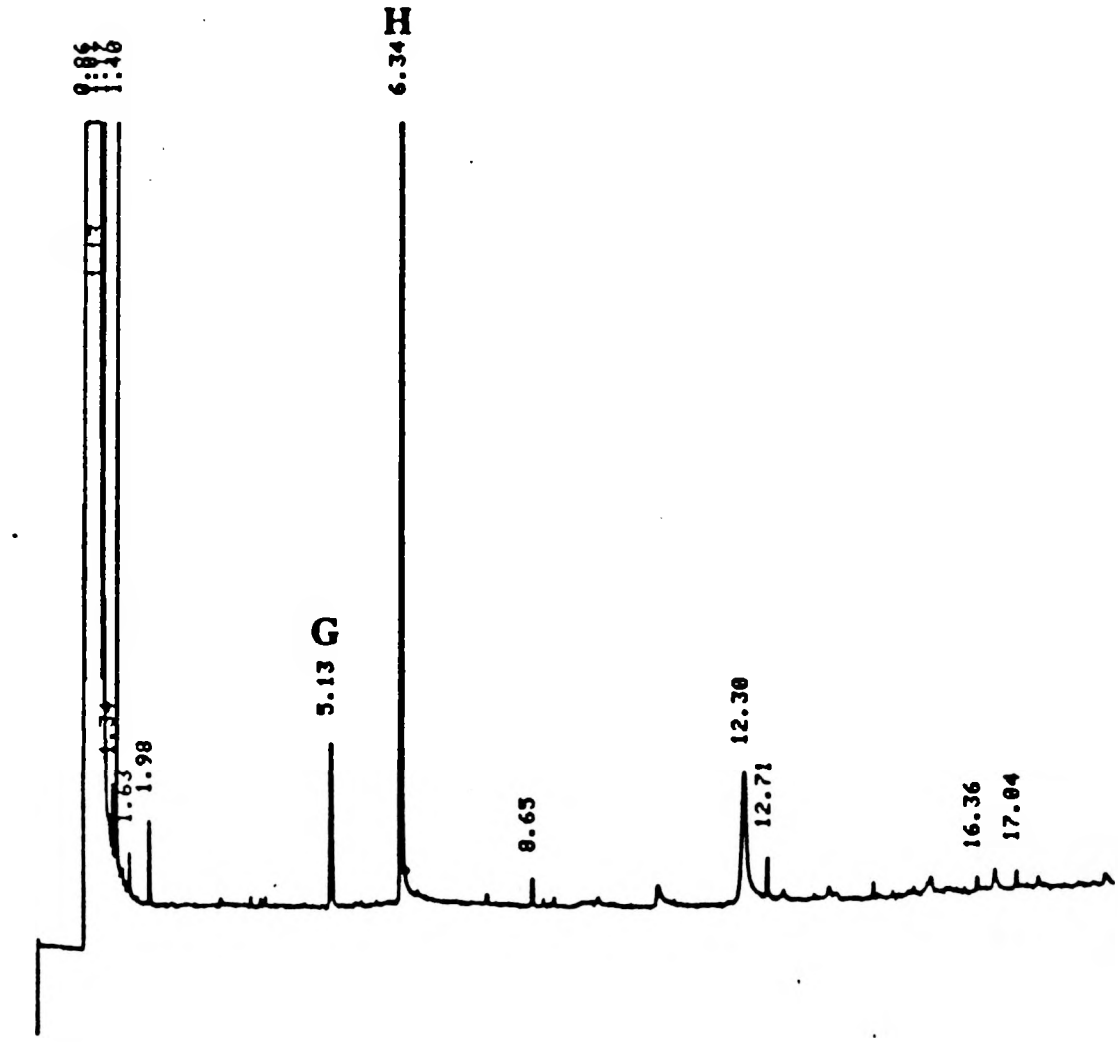

Figure 2. Chromatogram of sulfur compounds (both mercaptan and non-mercaptan compounds)with mercuric resin liner

The mixture solution (including both sulfur and non-sulfur compounds) was injected into the gas chromatograph with silicone resin in the liner, all the compounds passed through (see Figure 3). When the mixture solution was injected into gas chromatograph with polymeric-mercuric resin in the liner, all the non-mercaptan compounds passed through the liner while all the mercaptan compounds were retained on the mercuric resin (see Figure 4).

When the mercuric resin liner was used in both sulfur and mixture compounds solutions, the qualities of these chromatograms were as good as those obtained with the silicone resin liner.

Abstraction selectivity of polymeric-mercuric resin

It was observed that non-sulfur compounds and sulfide compounds, (butylsulfide (C_4H_9-S-C_4H_9) and benzenylmethylsulfide ($C_6H_5CH_2$-S-CH_3)), were not abstracted by the mercuric resin. Various other sulfur, non-mercaptan compounds were tested. Sulfur powder which was dissolved in methylene chloride, dimethylsulfoxide (CH_3-SO-CH_3), methylsulfone (CH_3-SO_2-CH_3), ethyldisulfide (C_2H_5-S-S-C_2H_5), and dithiooxamide (H_2N-CS-CS-NH_2) were injected into the gas chromatograph with both silicone resin liner and mercuric resin liner. The chromatograms of each compound with different resin liners were compared. It was found that none of these

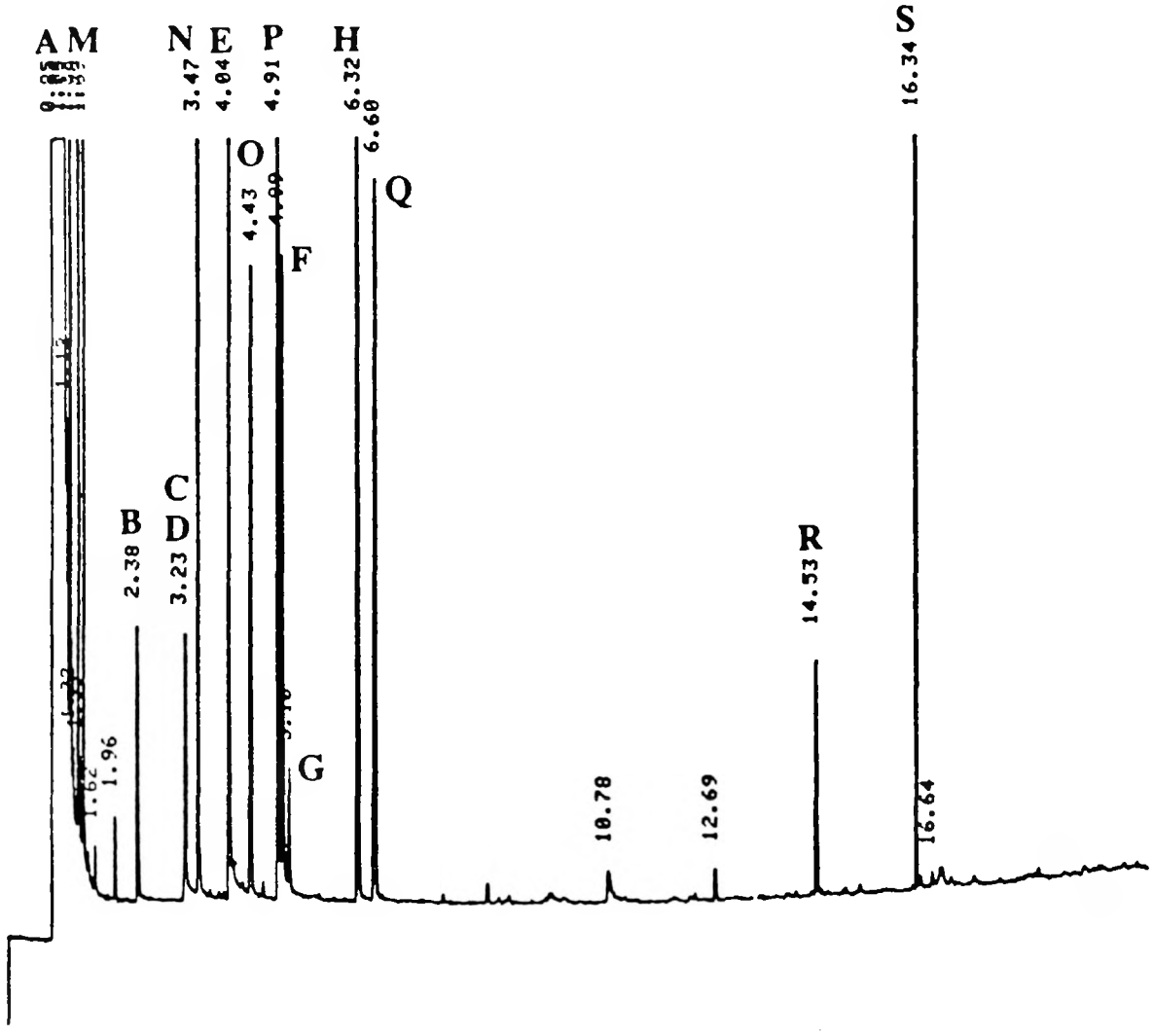

Figure 3. Chromatogram of mixture compounds (both sulfur and non-sulfur compounds) with silicone resin liner

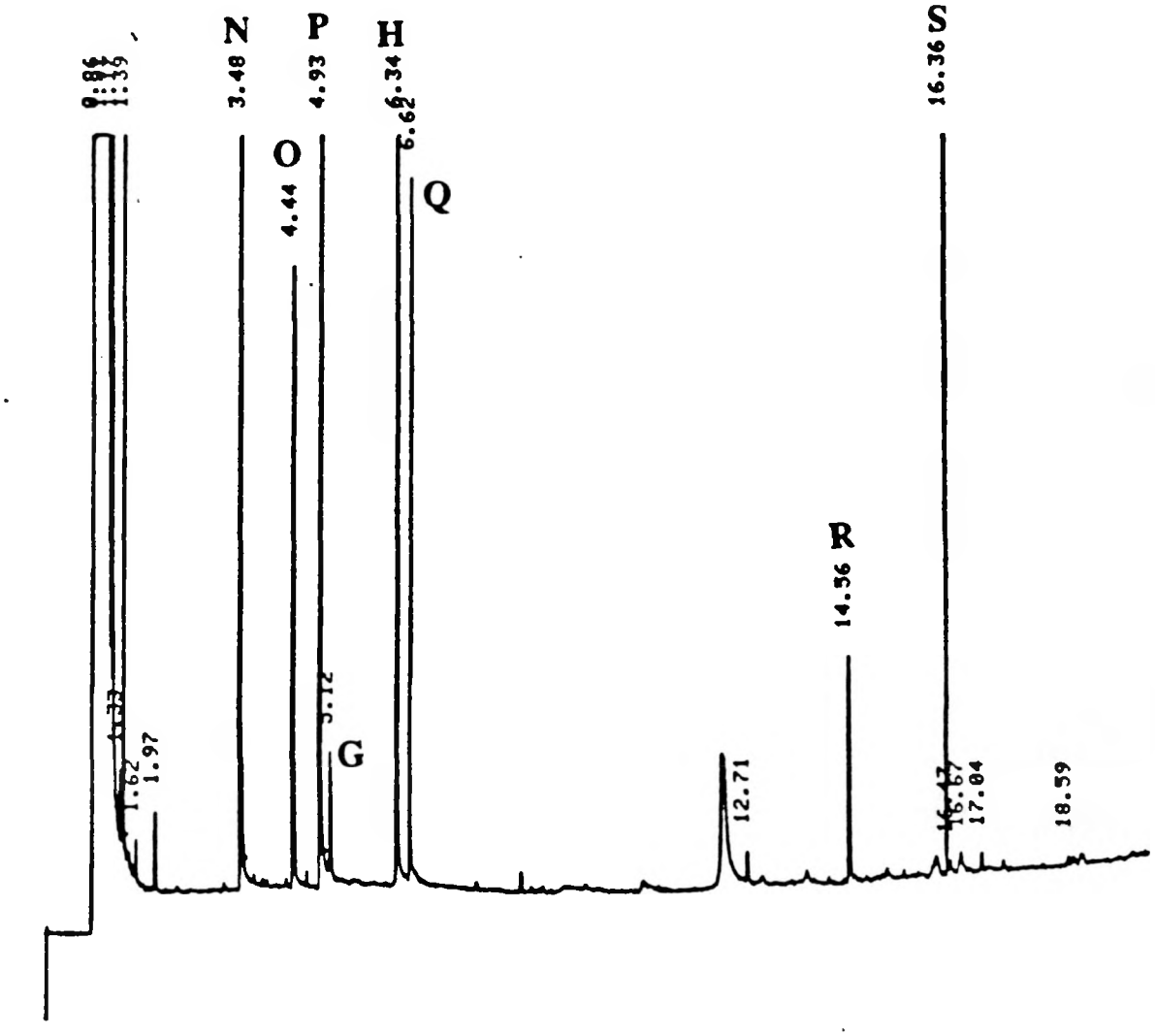

Figure 4. Chromatogram of mixture compounds (both sulfur and non-sulfur compounds) with mercuric resin liner

sulfur non-mercaptan compounds were retained by polymeric-mercuric resin in the liner.

Various mercaptan compounds were tested, which included aromatic, aliphatic, ester, alcohol, etc, and they were all retained by the mercuric resin. Various sulfur but non-mercaptan were tested, including sulfide, disulfide, sulfoxide, sulfone, and sulfur powder. None of these was retained on this resin. Also various non-sulfur compounds were tested and were not retained on this resin. Thus it can be concluded that only mercaptan compounds were abstracted by this polymeric-mercuric resin.

Injection temperature

Injection temperature is one of the critical conditions to be considered. If the injection temperature is too low, sample evaporation is slow and an unsatisfactory chromatographic separation is obtained. On the other hand, if the injection temperature is too high, the PS/DVB resin could be damaged or even destroyed. The chromatogram is unacceptable until the injection temperature reaches around 200°C. Starting from 200°C, various injection temperatures were tested, which included 200°C, 210°C, 220°C, 230°C, 240°C, and 250°C. As the injection temperature increased, more and more unknown inpurity peaks come out and

the baseline became increasingly noisy, especially at retention time after 18 to 20 minutes. When the injection temperature reached 250°C, huge amounts of unknown peaks appeared (see Figure 5). These are propably the compounds or polymers released from PS/DVB at high temperature. Even at 250°C, however, the mercuric functional group on the resin still worked well and all the mercaptan compounds were abstracted on the resin. The injection temperature was reduced to 200°C again, the amounts of unknown and impurity peaks reduced and the baseline improved. Some impurity peaks still remained on the chromatogram from the resin after being subjected to temperatures higher than 200°C (see Figure 6). However, the mercuric functional group still worked well after experiencing high injection temperatures. It is concluded that the high injection temperature (>200°C) did not affect the abstracting function of mercuric group on the resin, but would affect the structure of PS/DVB. The higher temperature caused the release of some impurities and/or polymers, or even destroyed part of the polymeric structure of the resin. So the 200°C injection temperature was chosen for this experiment.

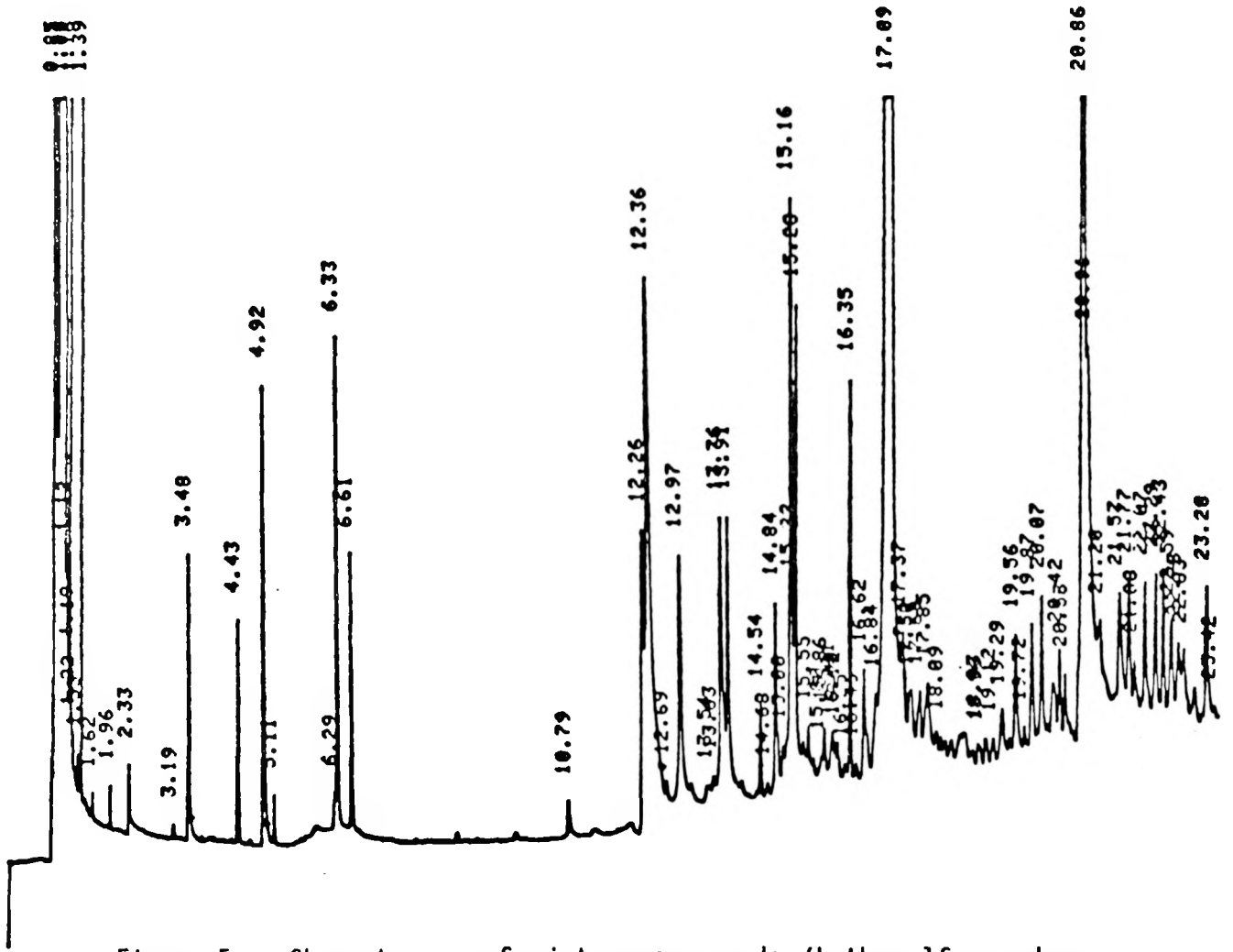

Figure 5. Chromatogram of mixture compounds (both sulfur and non-sulfur compounds) with mercuric resin liner at 250°C injection temperature

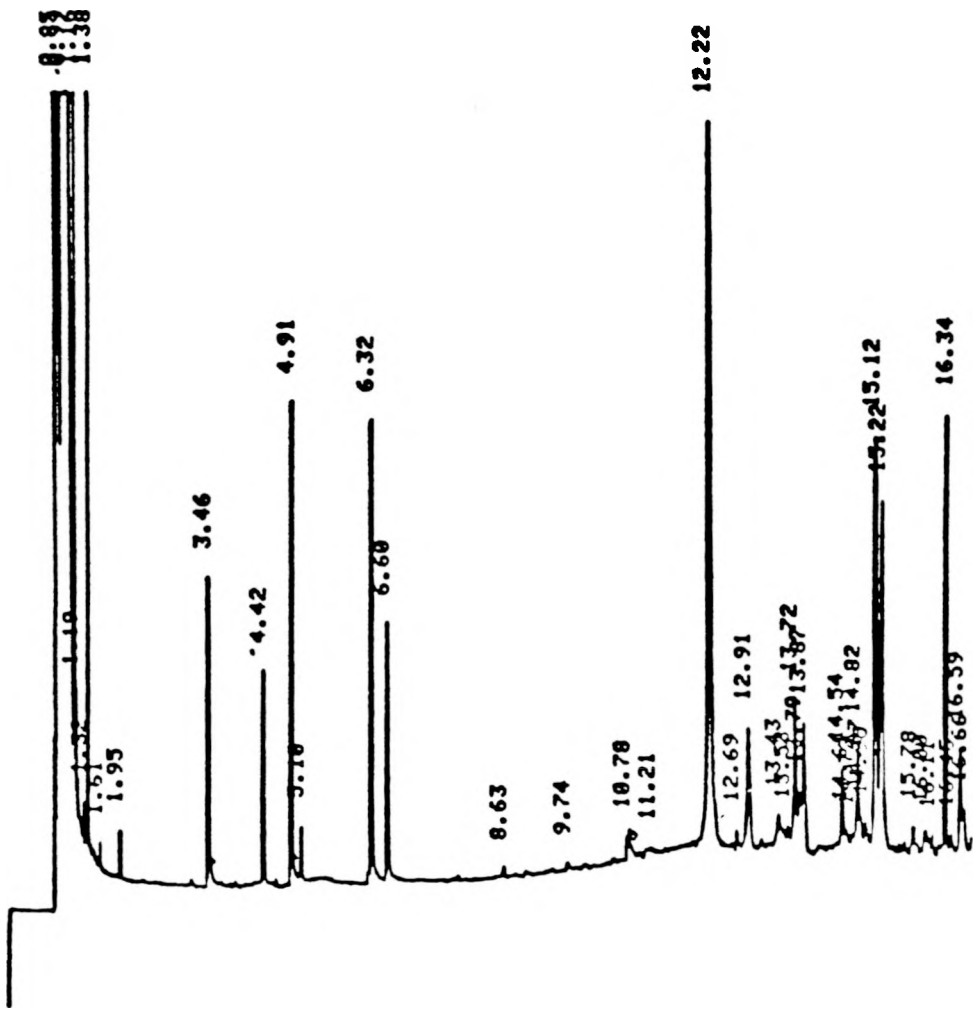

Figure 6. Chromatogram of mixture compounds (both sulfur and non-sulfur compounds) with mercuric resin liner at injection temperature back down to 200°C from 250°C.

Quantitative determination using polymeric-mercuric resin packed split liner

A series of mixture solutions (sulfur and non-sulfur compounds) were made. The concentration of sulfur compounds were fixed at about 2 ~ 10 ppm. The concentrations of non-sulfur compounds ranged by a factor of 0.25 to 50 of test concentrations for each compound, being about 25 ~ 500 ppb. Since benzylmethylsulfide could pass both silicone liner and mercuric liner, it was chosen as an internal standard. This was interesting as we can compare the linear regression parameters (correlation coefficients, slopes and intercepts) in both liners (silicone and mercuric liners) with the same internal stardard. The solutions were injected into the gas chromatograph with both liners respectively. The relative area of each compound at different concentrations was calculated and plotted against concentrations.

All the non-sulfur compounds gave linear plots (correlation coefficients all 0.999_+) with both silicone and mercuric resin liners (see table II). With the silicone resin liner, both sulfur and non-sulfur compounds passed through. Excellent linear plots of relative peak area vs. concentration were obtained (correlation coefficients 0.999_+). The average slope was 0.101 and the intercept -0.0256. With the mercuric resin liner, only the non-mercaptan compounds passed

Table II. Linear Correlation Coefficients of Non-Sulfur Compounds on Silicone and Mercuric Resin Liner

	Silicone Liner			Mercuric Resin Liner		
Compounds	Coef.	Int.	Slope	Coef.	Int.	Slope
Phenol	0.9993	-0.0526	0.104	0.9994	-0.0787	0.108
Indene	0.9997	-0.0546	0.102	0.9995	-0.128	0.109
p-Cresol	0.9995	-0.0335	0.103	0.9995	-0.0462	0.105
Naphthalene	0.9996	-0.0063	0.100	0.9995	-0.0231	0.101
Anthracene	0.9998	-0.0340	0.102	0.9994	-0.0432	0.100
Diethyl-phthalate	0.9997	0.0274	0.095	0.9999	-0.0117	0.102
Average:	0.9996	-0.0256	0.101	0.9995	-0.0552	0.104

through. Still excellent linear plots of relative peak area vs. concentration were obtained (correlation coefficients 0.999_{+}) for non-sulfur compounds. The average slope was 0.104, which was very close to that obtained with the silicone resin liner (0.101), and the intercept was -0.0552, which was also close to that with silicone resin liner (-0.0256). The quantitive determination of non-mercaptan compounds were not affected by the abstraction of the mercaptan compounds in the sample solution by the mercuric resin in the liner.

CONCLUSION

Chemically modified polymeric-mercuric resin is an excellent and apparently specific abstractor for mercaptan compounds, either aromatic or aliphatic, in capillary gas chromatography. Sulfur compounds other than mercaptans and non-sulfur compounds are not absorbed by this mercuric resin. The chromatographic separation of non-mercaptan compounds was good with the split liner packed with mercuric resin and with mercaptan compounds present in the sample solution. The quantitative determination of non-mercaptan compounds is excellent. An injection temperature as high 250°C does not affect the mercuric functional group, but does affect the structure of PS/DVB. So a 200°C injection temperature is suggested. This method could be very useful in sample cleanup, for example, to clean up tert.-butyl mercaptan present in perfumes in the perfume industry.

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GENERAL CONCLUSION

Polystyrene divinylbenzene resins were chemically modified by introduction of various functional groups, which included polar, non-polar, ionic and metallic groups. These chemically modified polymeric resins were used successfully for high performance liquid chromatography, solid phase extraction and some special applications in liquid and gas chromatography. The introduced functional groups offer an additional selectivity parameter for liquid chromatographic separation. The polar derivatized polymeric resins dramatically increased the recoveries of solid phase extraction, especially for polar compounds. The sulfonated polystyrene resins were used for separation of neutral and basic compounds as well as basic and weaker basic compounds. The sulfonated non-porous resin was used as amine abstractor and the polymeric-mercuric resin was used as mercaptan abstractor in capillary gas chromatograph. The researches in this dissertation has shown the very promising applications of polystyrene divinylbenzene resin in chromatographic field.

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