Journal of Chromatography, 136 (1977) 221–231
© Elsevier Scientific Publishing Company, Amsterdam — Printed in The Netherlands

CHROM. 9883

HIGH-SPEED LIQUID CHROMATOGRAPHY ON CADMIUM-MODIFIED SILICA GEL

CORAZON R. VOGT*, TIMOTHY R. RYAN** and JANET S. BAXTER
Environmental Trace Substances Research Center, University of Missouri, Columbia, Mo. 65201 (U.S.A.)

(Received December 15th, 1976)

SUMMARY

A novel procedure for the preparation of low-surface-area porous silica adsorbents, modified by cadmium salts, is described. The salt is introduced concurrently with a reduction of the surface area of an inexpensive silica gel by hydrothermal treatment. The resulting materials contain mg/g amounts of the metal. Liquid chromatography on the cadmium-modified adsorbent is shown. Charge-transfer chromatography is favored when the salt is introduced at high hydrothermal temperature which yields ultra low surface area and dense cadmium population.

INTRODUCTION

Silica gels are the most widely used adsorbents for column liquid chromatography. This preference prevails because these materials have large surface areas, diverse surface activities, easily controllable porosity and finally because they are available in narrow-size-range spherical particles. The preparations and characteristics of silicas for highly efficient liquid chromatography have been described by Kirkland[1] and Unger *et al.*[2]. While water[3,4] and polar organic compounds[1] are generally used as modifiers on active silica surface, Bebris *et al.*[5] used layers of carbon and weakly polar polymers.

Charge-transfer chromatography of organic compounds on surfaces containing impregnated metal salts has been reviewed[6,7]. The reviews include its use in gas–liquid chromatography (GLC), gas–solid chromatography (GSC) and liquid chromatography (LC).

Silica surfaces are made more selective to olefins by coating the adsorbent with silver ions. In particular, this technique has been used for column, paper, and thin-layer chromatography (TLC)[8]. Impregnation of TLC silica adsorbent with silver oxide[9], cadmium sulfate[10], cadmium acetate[11], and zinc salts[12] yields efficient isomeric separations of aromatic amines.

---

* To whom correspondence should be addressed.

** Present address: Department of Chemistry, Iowa State University, Ames, Iowa 50010, U.S.A.

Chromatography on complex forming materials has been extended to high-speed liquid chromatography. Mikeš *et al.*[13] obtained good separations of various olefins with Corasil II coated with either rhodium(II) acetate or silver nitrate–ethylene glycol. Heath *et al.*[14] used silver nitrate coated silica adsorbent to obtain preparative separations of unsaturated acetates, aldehydes and hydrocarbons by high-pressure LC. Columns packed with metal ion-impregnated silica gel usually suffer fast deterioration due to the loss of the salt with the mobile phase. Improved stability and chromatographic reproducibility of nitrogen-containing compounds on silica-based adsorbents impregnated with silver nitrate has been reported by Vivilecchia, Thiebaud and Frei[15]; with cadmium iodide by Kunzru and Frei[16]. Aigner, Spitzy and Frei[17] reported the bonding of silver to silica gel. In each case, the metal ions were introduced, initially or totally, to the silica surface by way of NaOH treatment. Chromatography on the resulting supports demonstrated largely a donor–acceptor mechanism with some contributions from adsorption.

Treatment of silica gels with water at high temperatures results in a reduction of surface area and better chromatographic properties[18]. Aue *et al.*[19] and Kapila *et al.*[20] converted technical silica gels by a similar "hydrothermal treatment" in liquid water to well-performing supports for GLC. In our own systematic study of the latter process, some evidence was obtained indicating that the activity of the silica gel changed with the hydrothermal temperature, presumably caused by the re-deposition of the dissolved silica on to the particles in the cooling solution. In this work, we report the hydrothermal treatment of silica gel in the presence of cadmium chloride and the performance of the resulting materials in modern LC.

EXPERIMENTAL

A borosilicate glass insert was used with the 500-ml cylinder of the Model 4652 Parr bomb (Parr, Moline, Ill., U.S.A.). The unit included the electric heater and the automatic temperature controller.

Preparation of the supports

A saturated solution of cadmium chloride was poured into the glass insert. Silica gel Davison 62, ground to 325–400 mesh, was added to form a slurry. The insert was placed in the bomb assembly and the heat turned on. It generally took 1–1.5 h to reach the desired temperature (250° or 350°). After 16 h of hydrothermal exposure, the bomb was taken out of the heating assembly and allowed to cool. In approximately 2½ h the temperature decreased to less than 100° and the bomb was opened. The treated silica gel was then transferred to a sintered glass funnel and washed with at least 500 ml of distilled water, followed by washings with methanol and hexane. The packing was dried at 45° under vacuum and re-sieved to 325–400 mesh.

At a hydrothermal temperature (HT) of 350°, the silica gel particles became considerably finer, and re-sieving yielded only a small fraction of 325–400 mesh. Therefore, in succeeding hydrothermal treatment at this temperature, silica gels of larger mesh size (270–325) were used to improve the yield of the 325–400 fraction.

The sieved portion was slurry-packed using a balanced-density mixture of tetrabromoethane and carbon tetrachloride. A Haskel air-driven pump was used at about 5000 p.s.i. on the chloroform packing solvent. All columns were 11 cm × 4.5 mm I.D., precision bore, stainless steel and fitted with 5-μm stainless-steel snubbers at the inlet and outlet.

Portions of 325–400 fractions were digested with perchloric acid and cadmium was determined by atomic absorption (AA).

Chromatography

A Model 202 (Waters Assoc.) liquid chromatograph with UV (254 nm) detector was used. The columns were tested with model isomeric compounds typically used with this type of columns: mixture 1 consisted of *o*-, *m*- and *p*-chloroaniline, containing 0.67 μg/μl of the *ortho* and *meta*, and 0.33 μg/μl of the *para* isomer. Mixture 2 consisted of 2-, 3- and 4-picolines, containing 0.17, 0.33 and 0.50 μg/μl, respectively. Both mixtures were in hexane.

Two binary solvent mixtures were used as mobile phase: acetonitrile–hexane and methanol–hexane. Hexane was also used to demonstrate that the blank supports can effect separations.

Capacity factor

The k' values of the columns were determined by using 1-μl injections of a series of 2-picoline solutions in hexane. At the given composition of the eluting binary solvent, the retention time of benzene was 19–22 sec at a flow-rate of 4 ml/min.

RESULTS AND DISCUSSION

Table I lists the specific surface area and the amount of cadmium present in the adsorbents produced in this study. Hydrothermally treated silica gel in the absence of cadmium was analyzed for the trace metal by flameless AA, others by flame AA. The sensitivity of the former was approximately 100 times better.

Using the more conventional chromatographic term, cadmium is present in 0.3–0.6% load. Incidentally, in a batch equilibration procedure of loading copper(II) complexes on silica gel, Datar and Ramanathan[21] reported a decrease uptake of the copper complexes with increasing surface area, which is opposite to those shown in Table I.

Figs. 1A and 2A show the LC separations obtained with the acetonitrile–hexane system on columns modified by cadmium. Also shown are the chromatographies on the blank columns (≡ hydrothermally treated in the absence of cadmium) obtained under identical solvent composition (Figs. 1B and 2B) and when hexane is vsed alone (Figs. 1C and 2C). The strong influence of cadmium is apparent. While metal-loaded columns give excellent separations of the chloroaniline isomers with acetonitrile–hexane, the same solvent system is inept on the blank columns. Use of pure hexane on cadmium with HT = 250° and 350° (HT-250 and HT-350) results in excessively retained *o*-chloroaniline while the *meta* and the *para* isomers are practically irreversibly adsorbed. On the other hand, the weak adsorption sites produced by hydrothermal treatment without the benefit of cadmium can effect minor separations (especially at HT-250) if hexane is used. Fig. 3 shows the separation of the same mixture when acetonitrile is replaced by methanol. Methanol is a strong solvent. However, it is less likely to compete in complex formation with cadmium than acetonitrile. Therefore, one would assume better sepa-

TABLE I

HYDROTHERMALLY MODIFIED SILICA GEL

Hydrothermal temp. (°C)	*$CdCl_2$ modifier*	*Surface area (m^2/g)**	*Amount of Cd (ppm)*
250	No	14.84	1.6
250	Yes	87.33	3000
300	No		0.32
300	Yes		6440
350	No	1.64	0.23
350	Yes	21.85	6340

* Multipoint BET analysis using krypton (Micromeritics, Norcross, Ga., U.S.A.).

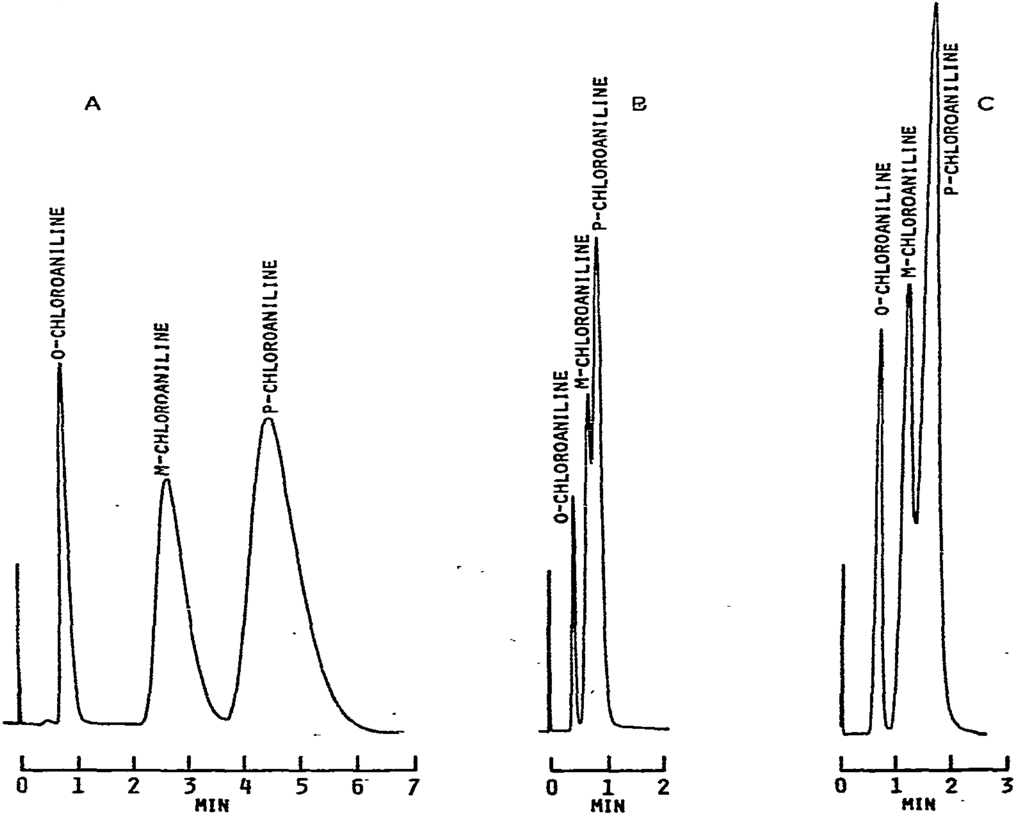

Fig. 1. Separation of chloroaniline isomers on hydrothermally (250°) treated silica gel in the presence and absence of cadmium chloride. (A) HT-250; 0.5% acetonitrile in hexane; attenuation, × 0.05. (B) Blank HT-250; 0.5% acetonitrile in hexane; attenuation, × 0.2. (C) Blank HT-250; hexane; attenuation, × 0.05. Column, 11 cm × 4.5 mm I.D.; eluent (as shown) at a flow-rate of 4 ml/min; detector, UV (254 nm). Injected amount, 2 μg each of *o*- and *m*-, and 1 μg of *p*-chloroaniline.

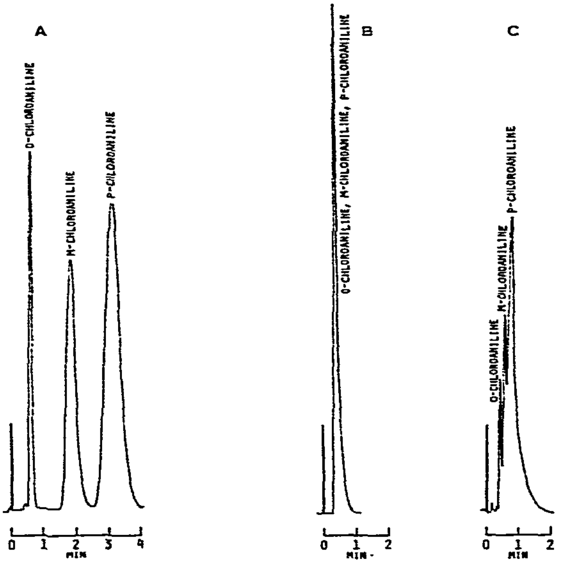

Fig. 2. Conditions are the same as in Fig. 1 except the hydrothermal temperature (350°). (C, attenuation × 0.1.)

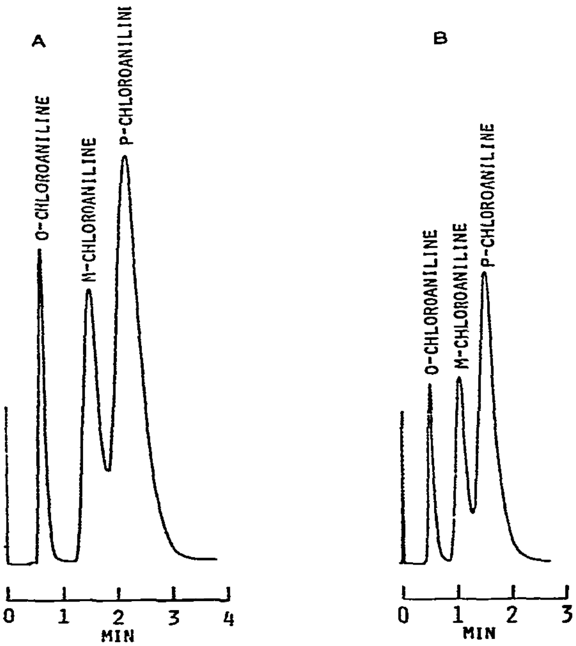

Fig. 3. Separation on cadmium-loaded silica gel. Eluent, 0.1% methanol in hexane (4 ml/min); column, 11 cm × 4.5 mm I.D.; detector, UV (254 nm). (A) HT-250; attenuation, × 0.05. (B) HT-350; attenuation, × 0.2.

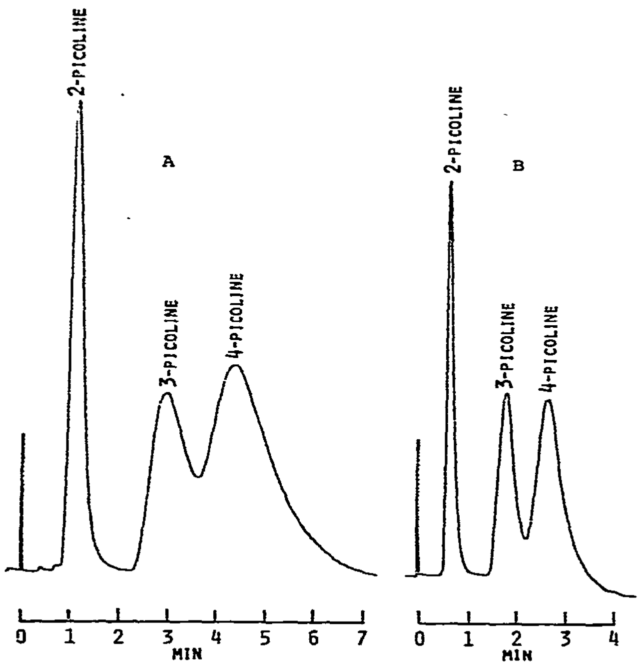

Fig. 4. Separation of picoline isomers on cadmium-loaded silica gel at (A) HT-250, 0.3% methanol in hexane (4 ml/min); attenuation, × 0.02; (B) HT-350, 0.25% methanol in hexane (4 ml/min); attenuation, × 0.05. Injected amounts of 0.5, 1.0 and 1.5 μg of 2-, 3-, and 4-picoline, respectively.

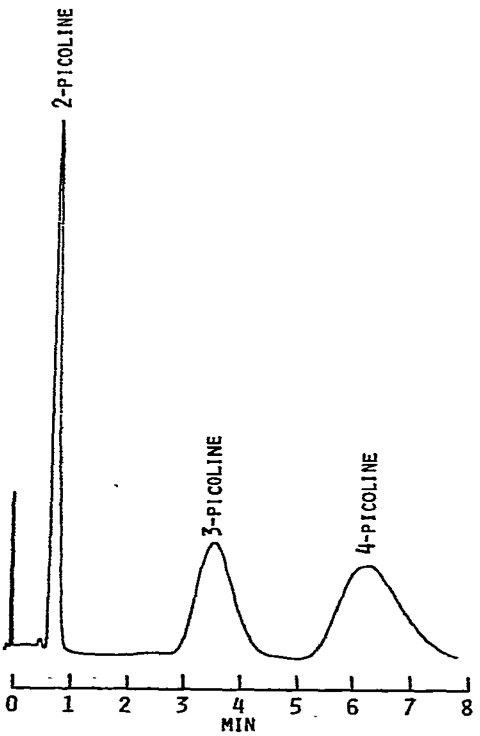

Fig. 5. Same conditions as in Fig. 4B except eluent (2% acetonitrile in hexane).

ration with acetonitrile if a donor–acceptor mechanism predominates. On the chromatograms shown, the cadmium HT-250 (0.03 mg Cd per m^2) is more retentive but less efficient than cadmium HT-350 (0.29 mg Cd per m^2). This seems to suggest the significant contribution of surface adsorption on two different sites in the former. Initial testing on cadmium HT-300 showed close chromatographic resemblance to cadmium HT-350; unfortunately however, the packed bed was disturbed before further evaluations could be made.

Chromatography of the more basic compounds, the isomeric mixture of picolines, is shown on Fig. 4. In this separation, the methanol–hexane composition is approximately twice that for the chloroanilines. Again, the important role played by a competing solvent acetonitrile in the separation is realized by comparing Figs. 4B and 5. Separation is obviously improved in the latter case but efficiency is decreased. Chromatographic efficiency shown on Fig. 5 can be, and was considerably, improved after introducing trace amounts of *n*-propylamine to the column. The separation of α- and β-naphthylamines is shown on Fig. 6A.

The sorption activity appears to have been decreased sufficiently to enable the sorbent to perform satisfactorily without the polar compound. The change of one

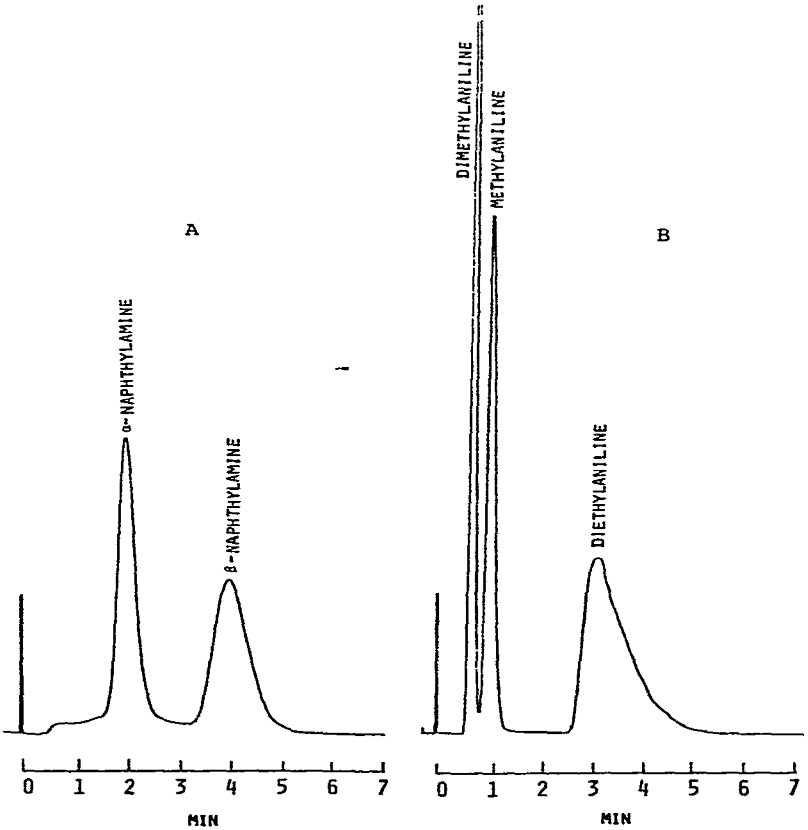

Fig. 6. Separations on 11 cm × 4.5 mm I.D. column packed with cadmium HT-350. (A) 0.5% acetonitrile–hexane; (B) 0.1% acetonitrile–hexane. Flow-rate, 4 ml/min.

mobile phase to another, or return to the initial condition in a gradient elution, requires only a short time for equilibration except for one solvent containing more than trace amount of *n*-propylamine (0.1%) which, however, was still useful.

The chemical form of the cadmium finally introduced into the silica gel by hydrothermal treatment is unknown at this time. On chromatography, the cadmium can act as surface moderator, electron-pair acceptor, or both. In the first case, the cadmium may deactivate the sorption sites effecting to what may be equivalent to a more uniform surface; it may alter the accessible surface area, and lastly, it may effect changes in pore sizes and distributions. In the second case, the cadmium would form a donor–acceptor complex with the nitrogen atom of the solutes to effect separations. If this is the case, the cadmium must be able to function as electron-pair acceptor. It appears that both factors are contributing to the chromatographic properties of cadmium-modified silica gel. It would be desirable to test solutes unlikely to form complexes with cadmium. In this regard however, the UV detector is limiting.

The contribution of the donor–acceptor complexing to the retention is influenced by the accessibility of the electron pair and the $\mathrm{p}K_a$ of the solutes. Both these effects are shown in Table II by the retention times on cadmium HT-350 of a number of amines. The toluidines followed the elution order *para* $>$ *meta* $>$ *ortho*. However, the elution compared to chloroanilines is: *p*-toluidine $>$ *p*-chloroaniline $\approx$ *m*-toluidine $>$ *o*-toluidine $\approx$ *m*-chloroaniline $>$ *o*-chloroaniline. It is interesting to note that while N,N-dimethylaniline elutes before N-methylaniline, N,N-diethylaniline elutes after N-ethylaniline (Fig. 6B). In a similar separation, the use of 0.05% acetonitrile–hexane (instead of 0.1%) gave 60% resolution of methylaniline from ethylaniline while diethylaniline is excessively retained with a broad tailing peak. Pure acetonitrile

TABLE II

RETENTIONS ON CADMIUM HT-350

Compound	*$\mathrm{p}K_a$ (ref. 22)*	*t_R (sec)*	*Mobile phase (4 ml/min)*
o-Chloroaniline	2.62	38	0.05% Acetonitrile–hexane
Dimethylaniline	5.07	72	
Methylaniline	4.85	102	
Ethylaniline	5.11	138	
Diethylaniline	6.56	52	0.2% Acetonitrile–hexane
m-Chloroaniline	3.32	100	
o-Toluidine	4.38	105	
Aniline	4.62	166	
m-Toluidine	4.67	166	
p-Chloroaniline	3.81	176	
p-Toluidine	5.07	262	
Pyridine	5.14	238	2% Acetonitrile–hexane
m-Phenylenediamine	4.88	119	100% Acetonitrile
2-Aminopyridine	6.7	55	10% Methanol–acetonitrile
p-Phenylenediamine	6.08	55	10% Methanol–acetonitrile
Benzylamine	9.34	109	0.001% *n*-Propylamine–methanol
o-Phenylenediamine	4.47	143	

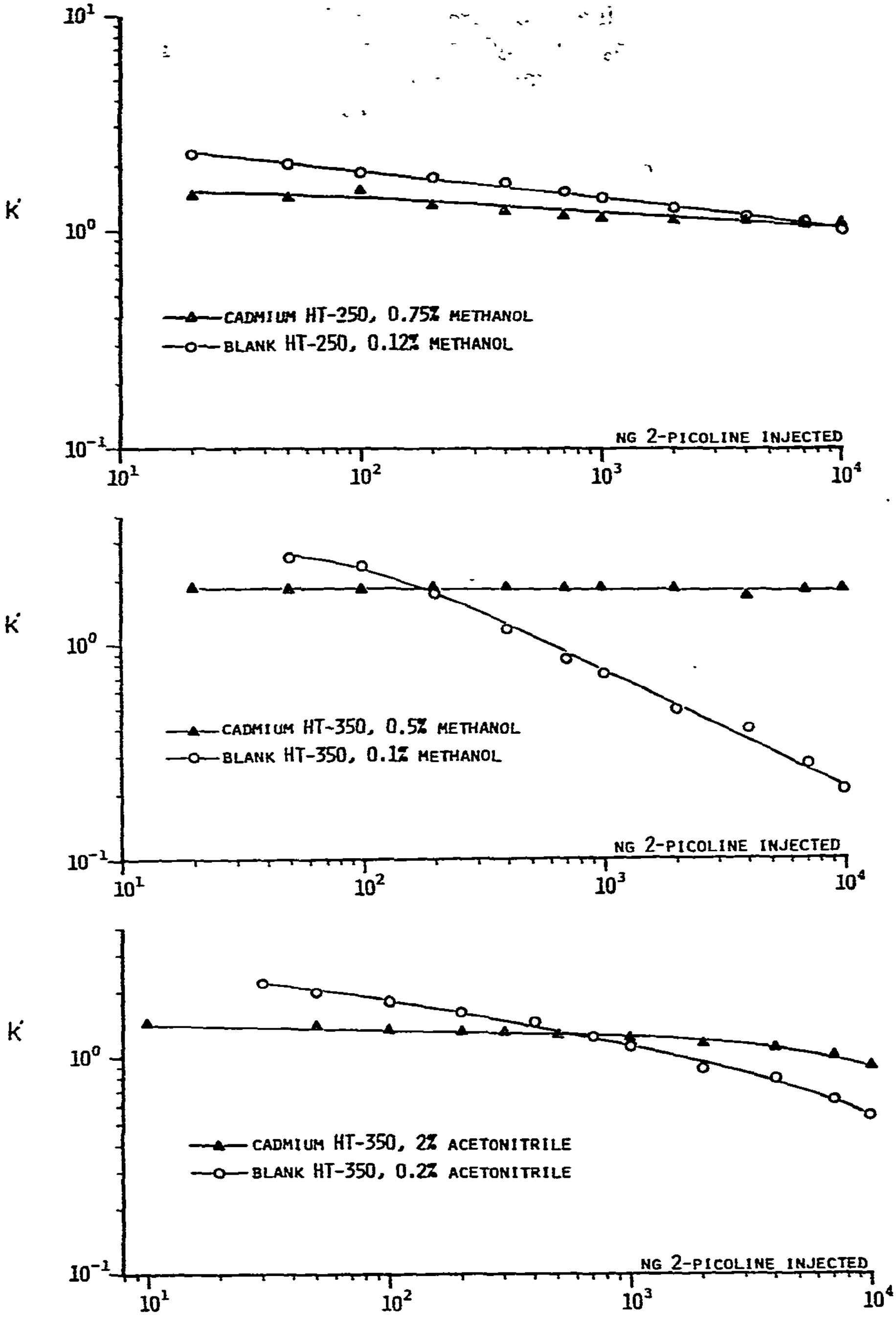

Fig. 7. Sorption isotherms expressed by the capacity factor (k') *versus* amount of 2-picoline injected to 11 cm × 4.5 mm I.D. column packed with silica gel modified with cadmium. Binary solvent as indicated in hexane; flow-rate, 4 ml/min.

will elute *m*-phenylenediamine while the *para* and *ortho* isomers will not. 10% methanol–acetonitrile and 0.001% *n*-propylamine–methanol are required to elute *p*-phenylenediamine and *o*-phenylenediame, respectively.

The column packed with cadmium HT-350 was heavily used with organic solvents for three months without loss of efficiency. At the end of the 3-month testing, the column was subjected to 3 h of continuous flushing with phosphate buffer (pH'5). Re-testing of the column with the chloroaniline mixtures gave tailing peaks not observed before.

In the present study, the capacity factor, *k'*, is expressed as the ratio of the corrected retention time of 2-picoline to the retention time of benzene. Sorption isotherms (expressed as *k' versus* amount injected) of the cadmium-modified silica gel and their corresponding blanks are compared on Fig. 7. The adsorbent loaded with cadmium-HT-250 yields a non-linear isotherm while the HT-350 gives linearity up to at least 10 μg. Nonetheless, the wide linearity range observed with cadmium HT-350 using methanol–hexane is diminished when acetonitrile–hexane is used. It is well known, that strong adsorbing solvents give wide linear ranges. The amount

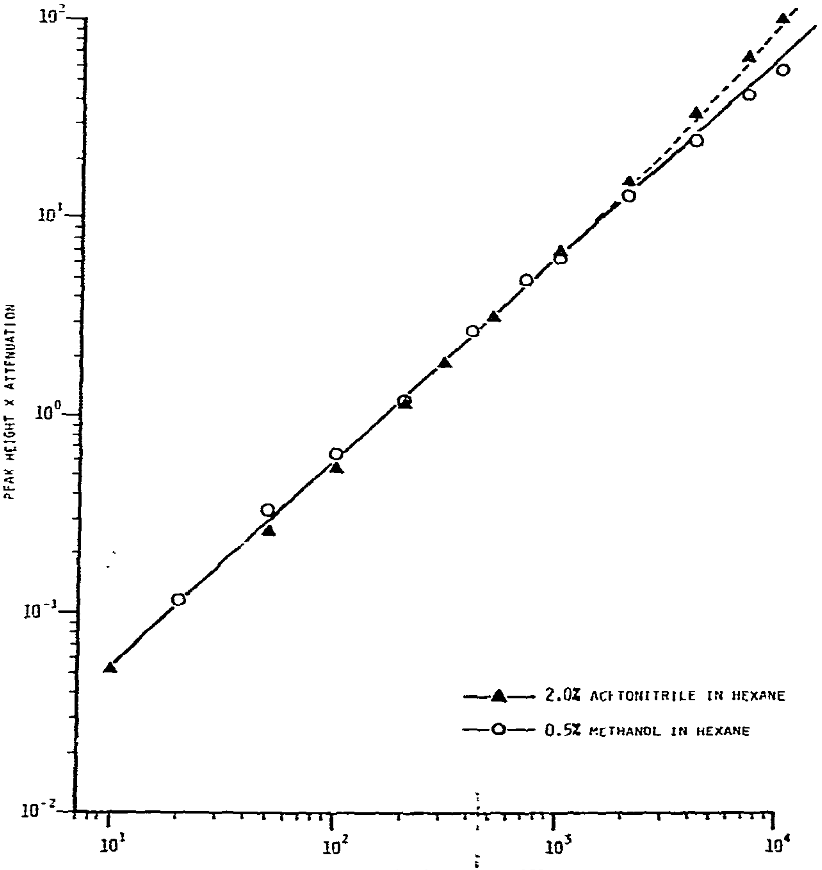

Fig. 8. Response linearity of silica gel loaded with cadmium at 350° hydrothermal temperature.

of 2-picoline injected is plotted *versus* the detector response (peak height × attenuation) on Fig. 8. While solvent strength increases linear range capacity, the separation usually degrades. Therefore, sorbent deactivation is generally used to improve capacity. The metal deactivation described in this work is concurrent with the alteration (decrease) of surface area and porosity in the presence of a saturated solution of the salt. The consequence of an unsaturated cadmium chloride solution on the amount deposited at different HT temperatures was not investigated. The effect of cooling rate on the size and shape of the particles has likewise not been investigated. Yet, it is easy to imagine that a variety of cadmium loadings with different surface areas can be made by this procedure.

CONCLUSIONS

Low-surface-area LC adsorbents can be derived from low-priced silica gel by hydrothermal treatment. The LC activity and selectivity is modified by the presence of a cadmium salt introduced during the treatment, and can be manipulated, to a degree, to yield separations of model amines.

ACKNOWLEDGEMENTS

We are grateful to Mr. E. Hinderberger and Dr. F. Lichte for the atomic absorption analysis. Thanks are due to Dr. W. A. Aue for helpful suggestions. This study was supported by the National Science Foundation, Grant No. MPS74 18084

REFERENCES

1 J. J. Kirkland, *J. Chromatogr.*, 83 (1973) 149.
2 K. Unger, J. Schick-Kalb and K-F. Krebs, *J. Chromatogr.*, 83 (1973) 5.
3 L. R. Snyder, *Principles of Adsorption Chromatography*, Marcel Dekker, New York, 1968, Ch. 4.
4 H. Engelhardt, J. Asshauer, U. Neue and N. Weigand, *Anal. Chem.*, 46 (1974) 336.
5 N. K. Bebris, R. G. Vorobieva, A. V. Kiselev, Y. S. Nikitin, L. V. Tarasova, I. I. Frolov and Y. I. Yashin, *J. Chromatogr.*, 117 (1976) 257.
6 O. K. Guha and J. Janák, *J. Chromatogr.*, 68 (1972) 325.
7 R. J. Laub and R. L. Pecsok, *J. Chromatogr.*, 113 (1975) 47.
8 A. S. Gupta and S. Dev, *J. Chromatogr.*, 12 (1963) 189 and refs. cited therein.
9 S. Tabak and M. R. M. Verzola, *J. Chromatogr.*, 51 (1970) 334.
10 K. Yasuda, *J. Chromatogr.*, 60 (1971) 144.
11 K. Yasuda, *J. Chromatogr.*, 72 (1972) 413.
12 K. Yasuda, *J. Chromatogr.*, 74 (1972) 142.
13 F. Mikeš, V. Schurig and E. Gil-Av, *J. Chromatogr.*, 83 (1973) 91.
14 R. R. Heath, J. H. Tumlinson, R. E. Doolittle and A. T. Proveaux, *J. Chromatogr. Sci.*, 13 (1975) 380.
15 R. Vivilecchia, M. Thiebaud and R. W. Frei, *J. Chromatogr. Sci.*, 10 (1972) 411.
16 D. Kunzru and R. W. Frei, *J. Chromatogr. Sci.*, 12 (1974) 191.
17 R. Aigner, H. Spitzy and R. W. Frei, *Anal. Chem.*, 48 (1976) 2.
18 A. V. Kiselev and Y. I. Yashin, *Gas Adsorption Chromatography*, Plenum Press, New York, 1969.
19 W. A. Aue, S. Kapila and K. O. Gerhardt, *J. Chromatogr.*, 78 (1973) 228.
20 S. Kapila, W. A. Aue and J. M. Augl, *J. Chromatogr.*, 87 (1973) 35.
21 A. G. Datar and P. S. Ramanathan, *J. Chromatogr.*, 114 (1975) 29.
22 H. A. Sober, *Handbook of Biochemistry*, CRC Press, Cleveland, Ohio, 1970, pp. J-203–220.