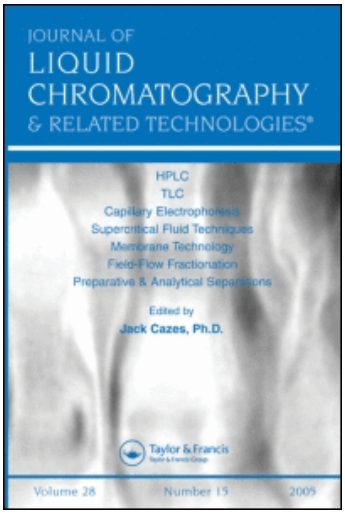

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INERTNESS OF BONDED SILICA GEL PACKINGS

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ABSTRACT

Stability and inertness of bonded silica gel packings were examined from the retention behavior of acidic and basic compounds, and also metal sensitive compounds. Bonded silica gels made from pure silica gel were stable in acidic and basic solutions, and did not interfere with chromatography of chelate reagents in reversed-phase liquid chromatography.

INTRODUCTION

A variety of bonded silica gels are marketed, however a chromatogram obtained with one type of packing often could not be reproduced with another similar packing. The difference was not simply a difference in retention time; occasionally the elution order was changed or some compounds completely disappeared. The reasons may be related to the difference in surface area, pore size, pore structure, pore volume, particle size, particle shape or size distribution. In addition to these physical parameters, the chromatographic performance is undoubtedly affected by the chemical composition of silica gel, such as the existence of untreated silanol groups and/or trace metals, especially polar

basic compounds [1,2]. In effect, the removal of impurities by boiling with hydrochloric acid can result in improved properties of the silica gel bonded phase for chromatography of cinnamic acid and hop bitter acids [3].

There are many reports regarding the synthesis method of bonded silica gel packings which limits the work of residual silanol groups and trace metals, where by silica gels are first washed in acid and carefully silanized to obtain maximum surface coverage packings. One of such ODS silica gels is suitable for liquid chromatography of peptides [4].

In a previous report, octadecyl- and octyl-bonded silica gels were synthesized from very pure silica gel and their stability in acidic and basic solutions was discussed [5]. The new bonded silica gels were stable for over 500 hours immersed in 0.1 v% aq. trifluoroacetic acid solution (pH 2) and 0.01 M sodium phosphate buffer with 50 v% methanol solution (pH 9). In this report, several ODS silica gels were synthesized from various silica gels. An octyl- and a phenyl-bonded silica gels were synthesized from pure silica gel and their chemical stability in both acidic and basic solutions was determined. Additionally, their inertness for residual silanol and trace metals is discussed based on results obtained with marketed octadecyl-bonded silica gels.

EXPERIMENTAL

The details of the liquid chromatograph equipment have been previously described [5]. The HPLC pump, the UV/VIS detector, the column oven, the degassing unit and the Rheodyne injector, models 576, 502U, 553, 545, and 7125, respectively were obtained from Gasukuro Kogyo Inc., Tokyo. Chemicals and HPLC solvents were obtained from Kishida Kagaku, Tokyo, and the deionized water was

further purified through a Pure-line from Yamato Kagaku, Tokyo. The chromatographic condition is given in each figure legend.

Five silica gels were prepared, one original non acid-washed silica gel (NAW), a once acid-washed silica gel (AW1), a twice acid-washed silica gel (AW2), a different type of 10 μm silica gel (PREP-SIL) and a pure silica gel (Inertsil SIL). The trace metals were washed out after boiling in 6N hydrochloric acid. The metal content, measured by atomic adsorption spectroscopy and ICP, is given in Table I. There was a significant difference in the metal content in all silica gels except between AW1 and AW2.

The carbon content of bonded packings was measured by an elemental analysis method. The end-capping (EC) process was completed by silanization with trimethylsilyl reagent until the inert parameter I* became constant [5]. The carbon content and inertness are listed in Table II, where NEC means non end-capped.

The packings were packed in 15 cm x 4.6 mm id. stainless steel columns using a high pressure slurry-packing method [6]. The chromatographic behavior of these packings was compared to that of the following two ODS silica gel columns: STR ODS-H (15 cm x 4.0 mm id.) from SHIMADZU TECHNO-RESEARCH Inc., (Kyoto), CAPCELL PAK C18 (SG) (15 cm x 4.6 mm id.) from SHISEIDO Co. Ltd., (Tokyo), and two house-packed columns of Nucleosil 100-C18, 5 μm from Machinery-Nagel Co. and YMC-GEL ODS-5 AM Type from YAMAMURA CHEMICAL LAB. Co. Ltd., (Kyoto). The column size of latter packings was 15 cm x 4.6 mm id..

RESULTS and DISCUSSION

After the acid-wash process, the surface of silica gel may become acidic. The effect of the acid-wash process can be seen from the chromatographic behavior of acids. The effect of the

Table I Metal content of silica gels

silica gel	PS* um	ps# A	SF$ m /g	Metal ppm Na	Mg	Al	Ca	Ti	Cr	Mn	Fe	Ni	Zn
NAW	5	122	333	190	250	150	730	160	nd	nd	22	nd	19
AW1	5	126	328	56	97	54	310	99	nd	nd	10	nd	4
AW2	5	132	320	40	92	41	350	95	nd	nd	11	nd	9
PREP-SIL	10	100	350	6	nd	63	nd	70	nd	nd	13	nd	6
Inertsil SIL	5	138	364	5	1	5	10	nd	nd	nd	1.5	nd	1

NAW: non acid-washed silica gel, AW1: once acid-washed NAW, AW2: twice acid-washed NAW, PREP-SIL and Inertsil SIL are marketed silica gels. nd: not detectable, *: particle size, #: pore size, $: surface area.

Table II Characterization of octadecyl-bonded silica gels

ODS-silica gel	C w%	I*	RRT1	RRT2	RN	PS	RH1	RH2
A NAW NEC	19.2	nc	2.30	1.22	1.13	>8	nc	nc
B NAW EC	19.2	0.45	1.00	0.65	0.52	3.40	0.11	nc
C AW1 NEC	20.2	nc	0.56	0.60	1.14	3.90	0.13	nc
D AW1 EC	20.2	0.44	0.49	0.54	0.93	1.74	0.63	0.13
E AW2 NEC	19.6	nc	0.51	0.55	1.04	3.43	0.13	nc
F AW2 EC	19.6	0.48	0.45	0.49	0.91	1.73	0.73	0.05
G Inertsil ODS-2	18.5	0.35	0.41	0.44	1.13	1.35	1.68	0.91
H Inertsil C8	10.5	0.30	0.38	0.40	1.10	1.25	1.51	1.22
I Inertsil Ph	9.5	0.40	0.44	0.49	1.07	1.80	-	0.84
J Inertsil PREP-ODS	20.0	0.40	0.47	0.51	1.11	1.35	2.12	0.80
K commercial ODS	-	-	0.43	0.46	0.34	2.06	2.06	0.69
L commercial ODS	14#	-	0.50	0.55	0.94	2.22	0.94	nc
M commercial ODS	17$	-	0.47	0.52	0.51	1.89	0.76	0.01
N commercial ODS	20$	-	0.62	0.65	1.10	3.21	1.12	0.11

C w%: carbon content, I*: polarity index (ref. 5), RRT1: ratio of retention time of pyridine/phenol, RRT2: ratio of retention time of aniline/phenol, RN: ratio of plate number of ethylbenzoic acid/phenol, PS: peak synmetry of diphenylhydramine, RH1: peak height ratio of acetylacetone/naphthalene, RH2: peak height ratio of 8-quinolinol/toluene, NEC: non end-capped, EC: end-capped, AW1 and AW2: the same as those in Table I, nc: could not calculated, # from ref. 13, $ from ref. 14

end-capping process was not clear on the chromatograms of benzoic acids as seen in Fig. 1. The peak shape of p-ethylbenzoic acid was better on the non acid-washed and end-capped ODS silica gels. A further comparison of chromatography of p-ethylbenzoic acid was done by comparing the plate number of p-ethylbenzoic acid and phenol. The results are summarized in Table II as RN value. RN value is the ratio of the plate number of p-ethylbenzoic acid to that of phenol. Higher values indicate better packings for chromatography of acids. Even acid-washed and end-capped packings gave poor RN values. Bonded silica gels made from highly pure silica gels gave better RN values. Only one commercial ODS column had an RN value of greater than 1.0 and the other three gave an RN of less than 1.0.

The existence of residual silanol groups particularly influences the chromatography of nitrogen-containing compounds [2,7-9]. Here this influence was also seen (Fig. 2). Aniline was eluted after p-nitroaniline on non-acid-washed and end-capped ODS silica gel. However, end-capping was not the answer to reduce the tailing of aniline, where the peak height of aniline was lower than that of p-nitroaniline on acid-washed and non-end-capped ODS silica gels. Further investigation was based on the comparison of the ratio of the retention time of aniline to that of phenol; the value is given in Table II as RRT2. The end-capping process together with the use of purer silica gels gave a better chromatogram of aniline, and this was also true for the chromatography of pyridine. This is given in Table II as RRT1 value, which is the ratio of retention time of pyridine to phenol. The smaller RRT1 and RRT2 values indicate better packings for polar nitrogen-containing compounds. Four commercial packings were appeared to be acid-washed but the efficacy of the end-capping process was uncertain from this test.

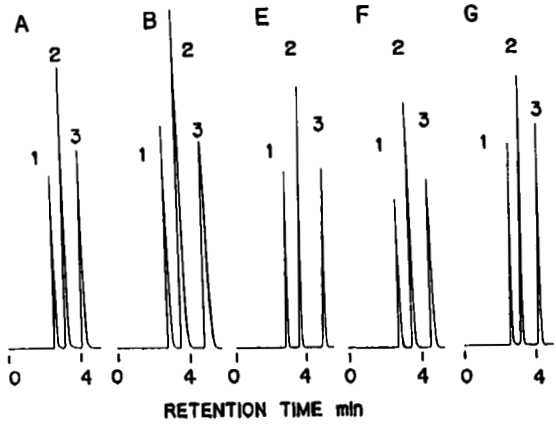

Fig. 1 Chromatograms of benzoic acids
Column: 15 cm x 4.6 mm id, eluent: 0.1 v% phosphoric acid in 50 v% aq. acetonitrile, flow rate: 1 mL/min, column temperature: 40 °C. Peaks: 1) benzoic acid (0.42 v%), 2) p-toluic acid (0.1 v%) and 3) p-ethylbenzoic acid (0.1 v%), Packings: A) NAW NEC, B) NAW EC, E) AW2 NEC, F) AW2 EC and G) Inertsil ODS-2 from Table II.

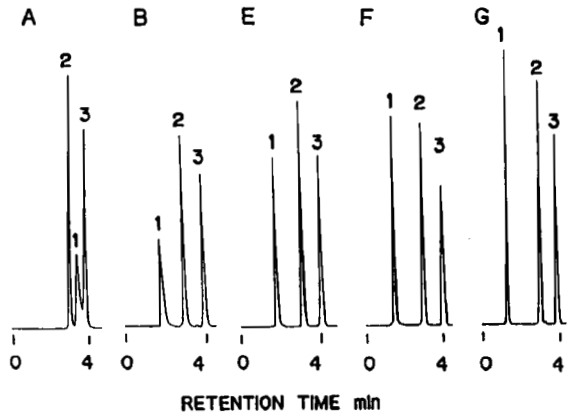

Fig. 2 Chromatograms of anilines
Column size, eluent, flow rate and column temperature are the same as those in Fig. 1. Peaks: 1) aniline (0.6 v%), 2) p-nitroaniline (0.1 v%) and 3) 2,4-dinitroaniline (0.13 v%). Packings A, B, E, F and G are the same as those in Fig. 1.

Diphenylhydramine was reported to be a metal-sensitive compound [10]. However such a large molecule did not give a good plate number as seen in Fig. 3, thus a their peak symmetry (PS) of 10% peak height was used for the comparison. The values of PS are given in Table II. Acid-wash and the end-capping process improved the peak symmetry. Three commercial packings appeared to be acid-washed and end-capped; one was not sufficiently acid-washed.

Acetylacetone is also a metal-sensitive compound [11] and makes chelate with several metal ions. As seen in Fig. 4, the chromatographic behavior was similar to that of aniline. The peak height was, however, lower than that of naphthalene on acid-washed and end-capped ODS silica gel compared to the result obtained on Inertsil ODS-2. The peak height ratio of acetylacetone to naphthalene is given as RH1 in Table II. Two commercial ODS packings gave an RH1 value of greater than 1.0.

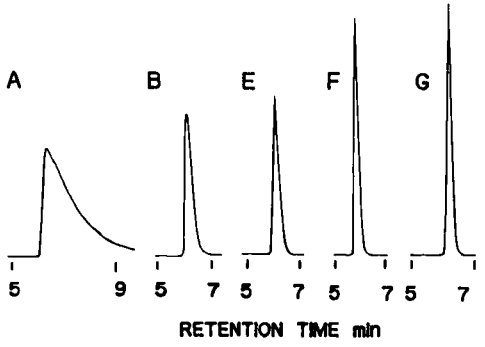

Fig. 3 Chromatograms of diphenylhydramine
Column size, flow rate, column temperature and packings are the same as those in Fig. 1. Eluent: 0.01M sodium laurylsulfonate in 60 v% aq. acetonitrile with phosphoric acid (pH 2.5). Peaks: diphenylhydramine (5 mg/mL).

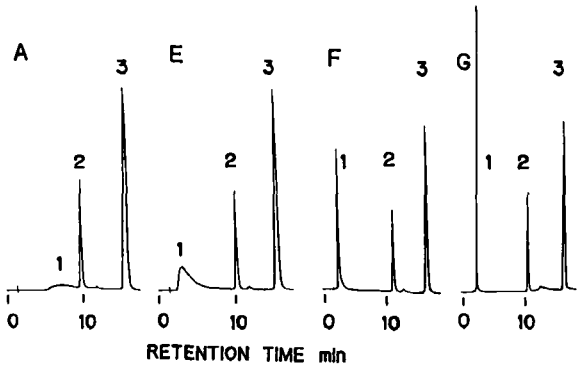

Fig. 4 Chromatograms of acetylacetone
Column size, flow rate and column temperature are the same as those in Fig. 1. Eluent: 0.5 w% sodium acetate in 60 v% aq. methanol. Peaks: 1) acetylacetone (20 μL/100 mL), 2) 5-nitronaphthalene (5 mg/100 mL) and 3) naphthalene (30 mg/100 mL). Packings: A) NAW NEC, E) AW2 NEC, F) AW2 EC and G) Inertsil ODS-2 from Table II.

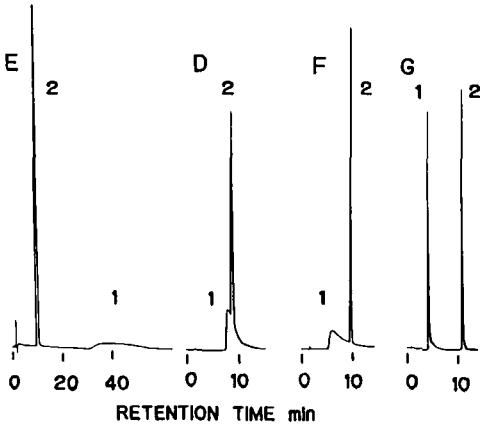

Fig. 5 Chromatograms of 8-Quinolinol
Column size, flow rate and column temperature are the same as those in Fig. 1. Eluent: 50 v% aq. acetonitrile. Peaks: 1) 8-quinolinol (1 mg/mL) and 2) toluene (20 μL/mL). Packings: E) AW2 NEC. D) AW1 EC, F) AW2 EC and G) Inertsil ODS-2.

8-Quinolinol is an active chelate reagent. It was impossible to measure the retention time [12] and difficult to obtain a reasonable peak shape for the analysis of bonded packings as seen in Fig. 5. The measurement of peak symmetry and the plate number of 8-quinolinol was difficult, therefore the peak height ratio of 8-quinolinol to toluene was measured and the values are given in Table II as RH2. If the silica gel contained a certain amount of metals, the silanization process was not beneficial to facilitate a well-bonded silica gel for chromatography of 8-quinolinol. This test seemed to be the most suitable test to measure the presence of trace metals in silica gels.

CONCLUSION

The acid-wash process can improve column efficiency. If the end-capping process is complete the peak symmetry of nitrogen-containing compounds becomes better. However, use of very pure silica gels can solve part of the problems of bonded silica gel packings, the chemical stability and chromatography of metal-sensitive compounds.

REFERENCES

1 G. Schomburg, A. Deege, J. Kohler and U. Bien-Vogelsang, J. Chromatogr., 282, 27 (1983).

2 J.S. Kiel, S.L. Morgan and R.K. Abramson, J. Chromatogr., 320, 313 (1985).

3 M. Verzele and C. Dewaele, J. Chromatogr., 217, 399 (1981).

4 K. Larsson, W. Hermann, P. Moller and D. Sanchez, J. Chromatogr., 450, 71 (1988).

5 T. Hanai, M. Ohhira and T. Tamura, Magazine of Liquid and Gas Chromatography, 16, 922 (1988).

6 Y. Yamaguchi and J. Kumanotani, J. Chromatogr., 210,512 (1981).

7 N.H.C. Cooke and K. Olsen, Am. Lab., 11, 45 (1979).

8 C.H. Lochmulee and D.B. Marshal, Anal. Chim. Acta, 142, 63 (1982).

9 S. Hara, T. Nakajima and M. Hirobe, eds., Biomedical Chromatography, (Nankodo, Tokyo, 1981) Vol. 2 (in Japanese).

10 Y. Ohtsu, O. Shirota, Y. Shiojima, K. Komatsu, H. Fukui, K. Nakamura and O. Nakata, in Abstracts of the 31st Liquid Chromatography Symposium, Kyoto, (1988), p. 33.

11 M. Verzele and D. Dewaele, Chromatographia 18, 84 (1984).

12 T. Hanai and J. Hubert, J. Liq. Chromatogr., 8, 2463 (1985).

13 1987 Catalog of Chemco, Osaka (1987), p. 38.

14 S. Nakagawa and K. Makino, eds., HPLC for Life Science (Hirokawa, Tokyo, 1988), p. 107 (in Japanese).