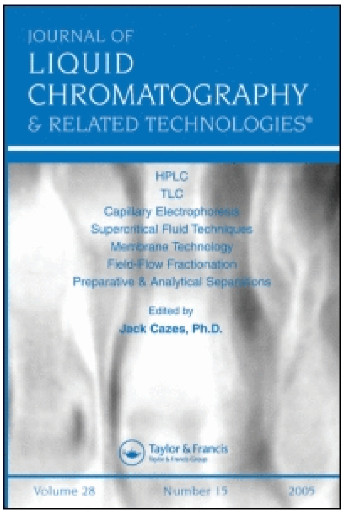

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RETENTION BEHAVIOR OF BASICA SOLUTES ON ZIRCONIA-SILICA COMPOSITE STATIONARY PHASE SUPPORTS IN NORMAL PHASE LIQUID CHROMATOGRAPHY

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ABSTRACT

Impregnation of sodium iodide into the matrix of zirconia-silica composite stationary phases during calcination was shown to improve the band profiles of basic compounds analysed using these surfaces. These compounds (pyridine and aniline) displayed severe band distortion associated with interactions between Lewis acid sites or residual hydroxyl groups on columns that were not subjected to the sodium-iodide impregnation. On the sodium iodide impregnated stationary phase, pyridine and aniline eluted with band shapes similar to neutral solutes, such as benzene. Stationary phase materials of this nature may prove to be useful for the separation of basic solutes that are

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unstable in aqueous environments, and hence, demand normal phase separations in which residual hydroxyl groups are often a significant component of the stationary phase surface.

INTRODUCTION

The surface chemistry of zirconia is complex and as a chromatographic surface, zirconia has been discussed in detail (1,2). In short, the zirconia surface is highly heterogeneous, containing Bronsted base sites arising from surface hydroxyl groups. But, more important to the surface chemistry of zirconia in liquid chromatography are the Lewis acid sites, which are formed as a consequence of unsatisfied co-ordination valency of the zirconium atom. This gives rise to a positively charged zirconium atom (1). The neighboring oxygen atoms then become Lewis bases (1).

In liquid-solid chromatography the Lewis acid sites may be occupied by water molecules or hydroxide ions depending on the pH (1), however, these sites also become competitive binding sites for other Lewis bases. It is through this competitive binding, that in many respects the surface interaction can be controlled. For instance, the hard Lewis base–fluoride, may be added as a mobile phase modifier, which then strongly binds to the Lewis acid sites. Other Lewis bases, as a consequence of the fluoride binding, experience less interaction with the Lewis acid sites on the zirconia surface. A detailed Lewis base eluotropic series was developed by Carr and coworkers (3). In this study, fluoride and phosphate were found to have the strongest interaction with the zirconia surface. However, the interaction between the Lewis base and the Lewis acid site is pH dependent and, consequently, a new eluotropic series would be required for each mobile phase and pH. For instance, the concentration of the fluoride ion adsorbed to the zirconia surface decreased dramatically in alkaline solvents (3). Nevertheless, these studies serve as a very useful guide in the evaluation of Lewis base adsorption strengths.

Silica too, has a heterogeneous surface, albeit the nature of the silica surface is less problematic in chromatographic separations than its zirconia counterpart. Basic substances such as pyridine do, however, typically display severe peak skewing due to interactions with the hydroxyl groups. Consequently, silica based columns that are used for separations of basic compounds like pyridine, must have a surface in which the solute can not interact with the residual hydroxyl groups. Reversed phase C18 columns prepared in such a manner are rigourously end capped to improve the chromatographic performance. Regardless, successful chromatographic performance of such compounds on silica supports requires that the surface of the support be modified to remove the unwanted secondary retention effects.

Recently, composite stationary phase supports have been prepared consisting of silica-zirconia, silica-alumina, silica-titania, and silica-magnesia (4). Kaneko and coworkers illustrated that these supports, when used in a normal phase mode for the separation of benzene, dimethyl phthlate, and pyridine displayed retention behaviour that was different from supports prepared from the individual components. Only the silica-magnesia composite stationary phase allowed for the elution of pyridine without severe peak tailing. All the other supports that were tested either eluted pyridine as a broad band, or in the case of native silica and the silica-zirconia composite, the pyridine was not observed to elute at all.

In the present study, the retention behaviour of a variety of compounds, including basic substances such as pyridine and aniline are evaluated on silica, zirconia, and zirconia-silica composite stationary phases. A unique aspect of this work is the modification of the zirconia-silica surface with a Lewis base during the calcination of the chromatographic support.

EXPERIMENTAL

Chemicals

HPLC grade hexanes and *iso*propanol were obtained from Mallinckrodt, Australia. Milli-Q water was obtained in-house and filtered through a 0.2 μm filter. All mobile phases were sparged continuously with helium. The stationary phase materials used for the preparation of columns in this study were synthesised according to methods previously described (5,6). Nucleosil silica (3 μm) was obtained from Alltech Associates, Australia.

Equipment

All chromatographic experiments were performed on a Shimadzu LC system (Shimadzu Scientific Instruments, Rydalmere, NSW, Australia) incorporating a LC-10AT*VP* pumping system, SIL-10AD*VP* auto injector, SPD-10A*VP* UV detector set at 254 nm, SCL-10A*VP* system controller, and Shimadzu Class-*VP* version 5.03 software on a Pentium II 266 PC. Data acquisition was achieved using a Lawson Labs model 203 serially interfaced 20-bit data acquisition system with a custom (±1 volt gain range operated at 10 Hz (Lawson Labs Inc, Malvern, PA, USA). Columns were packed using a Haskel air driven fluid pump (Haskel International, Burbank, CA, USA).

Preparation of Stationary Phase Supports

The synthesis of zirconia and zirconia silica mixed oxide stationary phase supports has been described in detail in previous communications (5,6). Specifically, aspects that are relevant to the current study involve the preparation of mixed oxide zirconia and silica composites and the process of surface modification.

In the present work, two composite materials were prepared. The first composite was that of a zirconia-silica composite prepared as previously described (5). The second composite material was also a zirconia-silica mixed oxide support. However, the calcination of this support was carried out under conditions that would allow a change in the crystal structure from the amorphous state to the tetragonal phase, at a temperature that corresponded to the melting of sodium iodide (7), namely 655°C. This specifically involved the hydrolysis of zirconyl chloride octahydrate (80 g) in water (60 mL) and the formation of zirconia microspheres with particle sizes of approximately 2 µm. Sodium metasilicate pentahydrate (52.7 g) was hydrolysed in water (80 mL), which was then added to the zirconia microspheres in a total volume of 500 mL. The reaction was allowed to proceed for 48 hours, allowing the water to evaporate until a thick paste was obtained. The resulting composite was washed and calcined for two hours, either in the absence of sodium iodide (at 810°C) to yield the unmodified zirconia-silica composite, or in the presence of sodium iodide (at 655°C) to yield the surface modified support.

Preparation of the surface modified zirconia-silica composite involved stirring equal parts of the amorphous zirconia-silica composite with sodium iodide in a water slurry overnight. The total volume of the slurry was such that the concentration of the zirconia-silica composite was approximately 5% w/v. The water was allowed to evaporate and the composite/salt mixture was dried overnight at 110°C. This composite/salt mixture was then calcined for two hours at 655°C. The calcined materials were then washed with copious quantities of water to remove the salt.

Preparation of Chromatographic Columns

Chromatographic columns were prepared in 50.0 × 2.1 mm stainless steel tubes fitted with 2 µm stainless steel frits. All columns were packed in a downward slurry mode. In all cases, the packing and slurry solvent was methanol. The displacement solvent was also methanol for all columns except that of the silica column in which the displacement solvent was dichloromethane. Slurrys were prepared by weighing approximately 0.5–0.7 g of the stationary phase material into 5 mL of methanol. The slurry was stirred for 30 minutes, then ultrasonicated for a period of 10 minutes, followed by a further 10 minutes of stirring. The columns were then packed using a Haskel air driven fluid pump at a pressure of 7000 p.s.i. for a period of twenty minutes.

RESULTS AND DISCUSSION

All relevant information regarding phase transitions during the calcination process for each of the zirconia and zirconia-silica composite stationary phase supports has been previously published (5–7). The silica stationary phase was used as received from the manufacturer. All materials had particle sizes in the order of 3 μm. Detailed information regarding the pore structure of these materials may be found in previous publications (5–7).

The primary reason why salt was added to stationary phase materials during the calcination process was for the control in the pore size and pore size distribution. In many respects, salts were shown to be beneficial for such purposes (6,8). However, in the preparation of the zirconia-silica composite, in which sodium iodide was used as the pore size controlling salt, the stationary phase material had a permanent brown-yellow discoloration. This discoloration remained even after washing with the solvents hexane, dichloromethane, methanol, water, and 5 M sodium hydroxide. Consequently, the benefits of incorporating a salt such as sodium iodide into the sample matrix during calcination become immediately obvious. That is, because the stationary phase was discolored, the surface must to some extent be modified. This modification may involve the incorporation of the Lewis base I^- into the support matrix. As a result, this type of surface modification may provide a mechanism through which unwanted interaction with Lewis acid sites may be controlled, irrespective of the mobile phase.

The scanning electron micrograph in Figure 1 illustrates the particles of the sodium iodide impregnated zirconia-silica composite stationary phase. Elemental analysis of these particles revealed that iodide remained within the sample matrix. Also present was sodium, silicon, and zirconium, confirming the formation of the silica-zirconia composite. The ratio of silicon to zirconium remained approximately constant over the cross section of the broken particle shown in Figure 1 compared to the whole sphere, indicating that the preparation of this material may yield an almost homogeneous composite. The broken particle shown in Figure 1 was not representative of the particles present in the sample, moreover, its presence was an interesting aspect, which was worthy of reporting and now makes us wonder how many of these particles are, in fact, hollow as this image suggests. During three weeks submersion in 5 M sodium hydroxide, the yellow color of the composite slowly changed, and when analysed using the electron microscope, it was apparent that the microspheres were fused together. Probably,

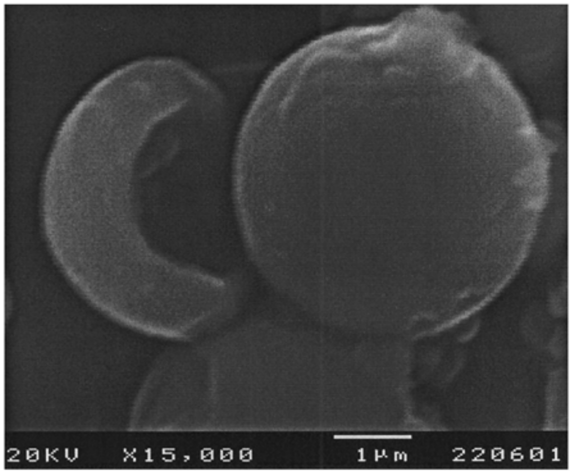

Figure 1. Scanning electron micrograph of zirconia-silica particles. Weight % composition: Fractured sphere, Si (21.1%), Zr (60.5%), I (3.2%), Na (10.9%), Al (4.3%) (Note: mounting cells were made from aluminium and hence the presence of the Al in the sample analysis). Whole sphere, Si (22.4%), Zr (68.8%), I (1.4%), Na (7.3%), Al (0.0%).

the silica content of the composite was soluble in the basic solvent following extended exposure, although the composite was stable following the short term exposure associated with washing the support. We have not performed detailed chemical stability studies in alkaline solvent environments.

The chromatographic performance of these four stationary phase supports was evaluated in a normal phase mode using benzene, anisole, aniline, and pyridine as test compounds. The mobile phase was 99% hexane and 1% isopropanol. Elution of benzene on all four columns resulted in a nearly symmetrical band, as shown in Figure 2. For ease of comparison, which becomes more important for the basic compounds, the peak height has been normalised. As can be seen in Figure 2, the peak shape for benzene on each of the columns is almost identical on all of the four columns. The difference in the elution volume reflects the differences in the void volume more so than the degree of retention, as for the most part, benzene was not retained on each of these columns. In the following discussions, differences in peak shapes may be attributed to the interactions between solutes and the stationary phase surface, rather than differences in the quality of the packed bed associated with eddy diffusion, or diffusion from within the porous network.

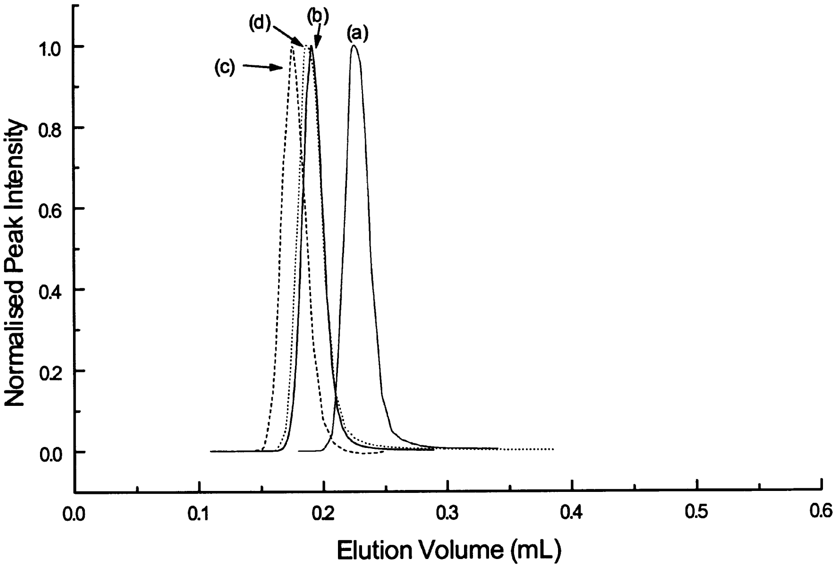

Figure 2. Elution profiles of benzene on each of the four columns. Column (a) silica; (b) zirconia; (c) zirconia-silica composite; and (d) iodide impregnated zirconia-silica composite.

Table 1 lists the capacity factors and asymmetry factors for each of the compounds tested on all four columns. Very little difference in the degree of retention is observed for the compounds, benzene, and anisole on any of the columns tested. Each of these compounds was unretained in 99% hexane However, significant differences in retention were observed for the basic compounds aniline and pyridine. In the first instance, pyridine was observed to elute with a capacity factor of 9.50 on the silica column and 0.99 on the zirconia column, while on the unmodified zirconia-silica composite column the capacity factor of pyridine was 0.86. The degree of retention is more similar to that of the zirconia column, despite the fact that the process of synthesis involved coating zirconia microspheres with silica and then allowing homogenisation to occur through the process of calcination (5). The peak shape of pyridine on each of these columns was severely tailed with asymmetry factors greater than 1.8 and up to 2.4 on the silica stationary phase. Kaneko and coworkers did not observe the elution of pyridine at all from their silica column or their silica-zirconia column (4). The difference between our zirconia-silica column and their composite column probably reflects the differences in the procedure for the preparation of the support. With such large differences, however, we envisage that the

Table 1. Retention Data for the Solutes Benzene, Anisole, Aniline, and Pyridine Eluting from the Zirconia and Silica Stationary Phase Materials

Column	Test Solute	Capacity Factor	Asymmetry Factor
Zirconia	Benzene	0.00	1.165
	Anisole	0.03	1.173
	Aniline	2.16	0.810
	Pyridine	0.99	1.840
Silica	Benzene	0.00	1.265
	Anisole	0.07	1.195
	Aniline	3.06	1.035
	Pyridine	9.50	2.450
Zirconia-Silica	Benzene	0.00	1.190
	Anisole	0.06	1.250
	Aniline	0.57	2.060
	Pyridine	0.86	1.795
NaI-Zirconia-Silica	Benzene	0.00	1.235
	Anisole	0.02	1.060
	Aniline	0.45	1.260
	Pyridine	0.24	1.330

preparation of composites with highly reproducible surface chemistry may be a formidable task, at least for those of us who work on small scale stationary phase preparations.

The peak shape of the pyridine band eluting from the sodium iodide impregnated zirconia-silica column was uniform with an asymmetry factor equal to 1.33,which is not too dissimilar from that of the unretained solute, benzene. On this column, the capacity factor of the pyridine was 0.24. The improvement in peak shape as a result of the surface modification is most clearly shown in Figure 3, which compares the benzene and pyridine bands on the zirconia-silica and sodium iodide zirconia-silica composite stationary phases. Once again, for clarity, the peak height of each band has been normalised. Clearly, sodium iodide impregnation significantly reduced the extent of peak tailing. In addition to the improved peak shape gained by the impregnation of sodium iodide, the retention of pyridine was reduced.

The other compound of interest in this separation system was aniline. Aniline demonstrated significant peak tailing on the zirconia-silica composite stationary phase (asymmetry factor = 2.06), but was observed to elute from the sodium iodide impregnated zirconia-silica column with an asymmetry factor of 1.33. This is clearly illustrated in Figure 4. Aniline was most strongly retained on the silica column, but the elution order between the pyridine and the aniline was

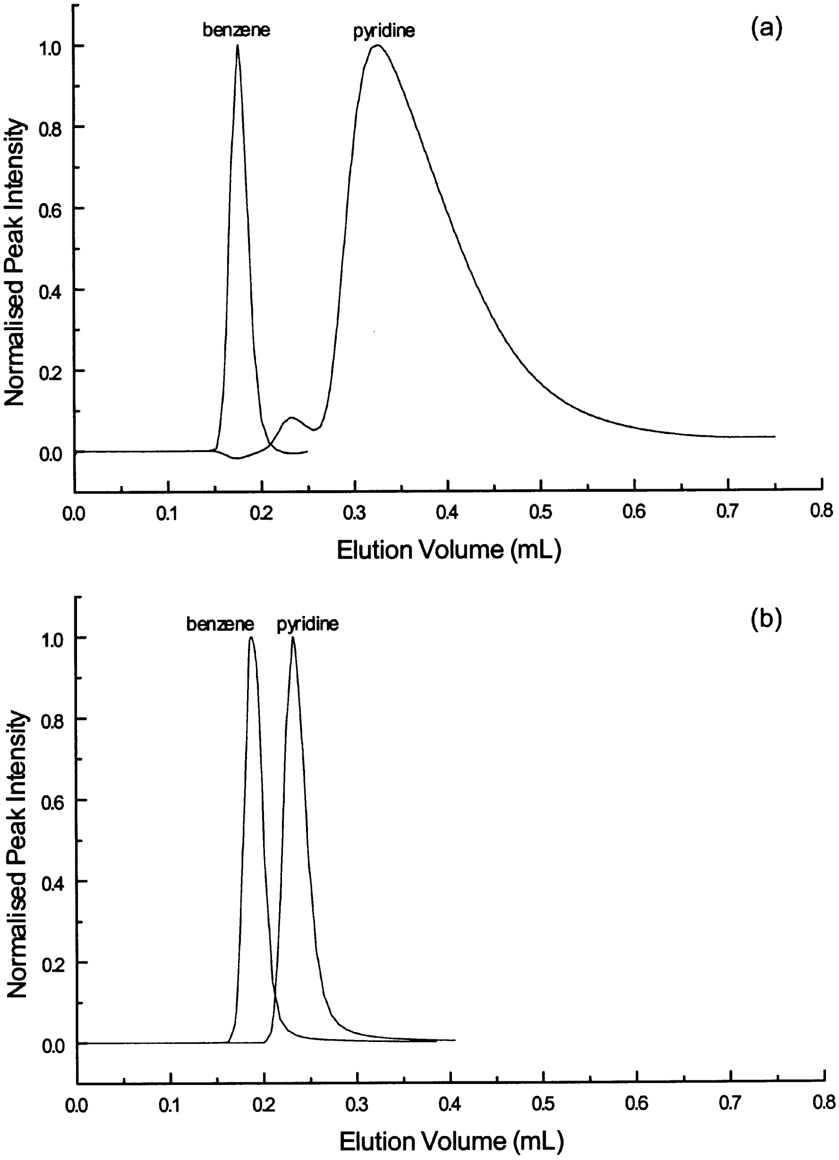

Figure 3. Comparison in peaks shapes between pyridine and benzene on each of the composite stationary phase surfaces. (a) Zirconia-silica composite. (b) Iodide impregnated zirconia-silica composite.

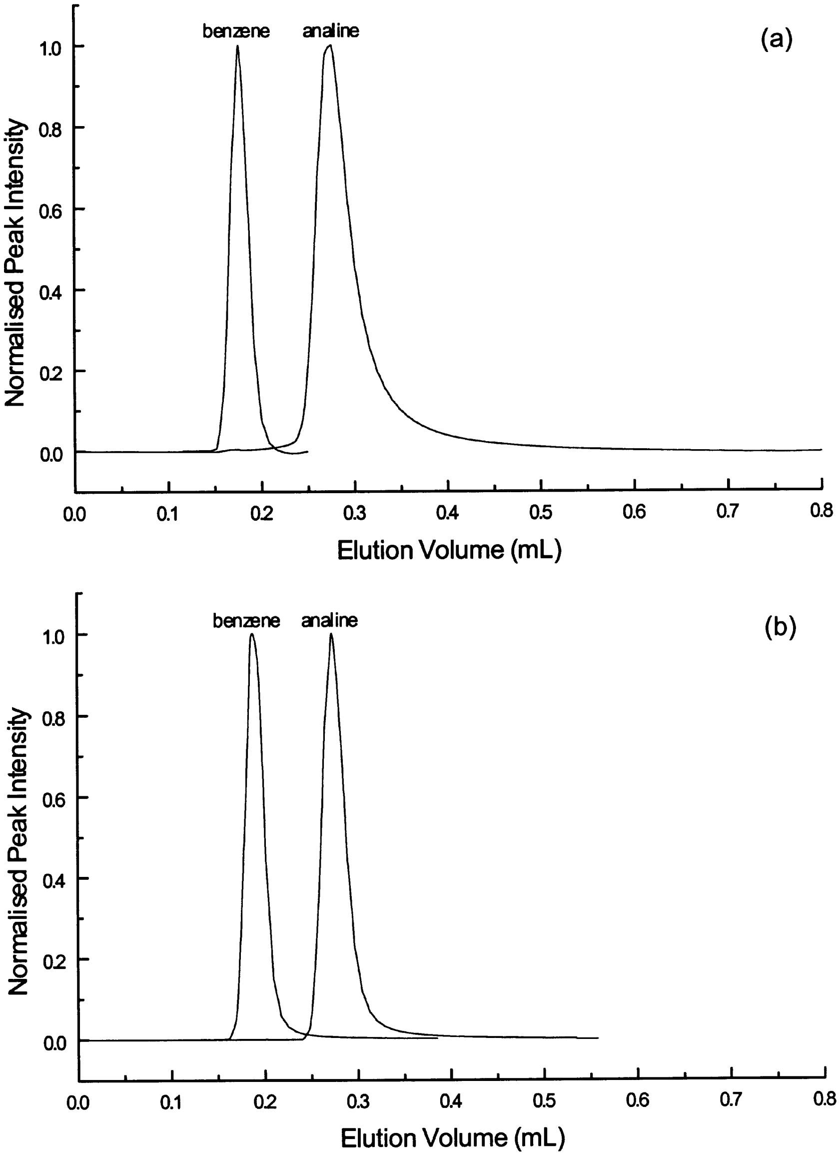

Figure 4. Comparison in peaks shapes between aniline and benzene on each of the composite stationary phase surfaces. (a) Zirconia-silica composite. (b) Iodide impregnated zirconia-silica composite.

reversed on the zirconia and sodium iodide impregnated zirconia-silica composite columns, compared to the silica and the zirconia-silica composite columns.

In general, all the solutes tested in this study had less retention on the sodium iodide impregnated zirconia-silica column than any of the other three columns tested. Furthermore, the peak shapes of the basic compounds pyridine and aniline were more symmetrical. One interesting aspect of the four different stationary phases, was that retention on the composite stationary phase supports was entirely different from the supports made from the individual components, as was also observed by Kaneko and co-workers (4).

Finally, the protection of the solute from secondary adsorption interactions on these heterogeneous surfaces was shown to be possible following calcination of the support in a media such as sodium iodide. These types of columns may prove to be particularly useful for the separation of compounds that display similar secondary adsorption phenomena to that of pyridine, yet are unstable in aqueous environments. While we demonstrated the behaviour of solutes in a normal phase mode, we are yet to test the ligand exchange properties of these columns in aqueous media.

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REFERENCES

1. Nawrocki, J.; Rigney, M.P.; McCormick, A.; Carr, P.W. J. Chromatogr. A. **1993**, *657*, 229.
2. Nawrocki, J.; Dunlap, C.J.; Carr, P.W.; Blackwell, J.A. Biotechnol. Prog **1994**, *10*, 561.
3. Blackwell, J.A.; Carr, P.W. Anal. Chem. **1992**, *64*, 863.
4. Kaneko, S.; Mitsuzawa, T.; Ohmori, S.; Nakamura, M.; Nobuhara, K.; Masatani, M. J. Chromatogr. A. **1994**, *669*, 1.
5. Shalliker, R.A.; Rintoul, L.; Douglas, G.K.; Russell, S.C. J. Materials Sci. **1997**, *32* (11), 2949.
6. Shalliker, R.A.; Douglas, G.K.; Comino, P.R.; Kavanagh, P.E. Powder Technology **1997**, *91* (1), 17.
7. Shalliker, R.A.; Douglas, G.K.; Rintoul, L.; Comino, P.R.; Kavanagh, P.E. J. Liq. Chromatogr. and Relat. Technol. **1997**, *20* (10), 1471.

8. Shalliker, R.A.; Douglas, G.K. J. Liq. Chromatogr. and Relat. Technol. **1997**, *20* (11), 1651.

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