Journal of Chromatography A, 1217 (2010) 6085–6091

Contents lists available at ScienceDirect

Journal of Chromatography A

journal homepage: www.elsevier.com/locate/chroma

Cyano bonded silica monolith—Development of an *in situ* modification method for analytical scale columns

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ARTICLE INFO

Article history:
Received 14 April 2010
Received in revised form 15 July 2010
Accepted 17 July 2010
Available online 22 July 2010

Keywords:
Silica monolithic columns
Cyano reversed phase
Carbon load axial homogeneity

ABSTRACT

This study investigates the synthesis and chromatographic behaviour of an analytical size cyanopropyl "cyano" bonded silica monolith. Surface modification was undertaken by treating a neat silica monolith with chloro(3-cyanopropyl)dimethyl silane in dry heptane over a two day period. The resulting monolith showed stability over the duration of the testing program that involved flushing the column with more than 2000 column volumes of mobile phase. Efficiency measurements before and after sylation verified that the integrity of the silica monolith itself was not affected by the modification process, the highest number of theoretical plates (N/m) using anisole was 81,650. A brief selectivity test was then undertaken to assess methylene selectivity and phenyl selectivity. Elemental analysis was used to determine the homogeneity of the carbon load throughout the monolithic bed, and was compared to two commercial C18 and one 'self' modified C18 silica monoliths. The development of the *in situ* modification is also discussed.

1. Introduction

Monolithic columns have established their importance in high performance liquid chromatography (HPLC). Their structure allows for high speed and high efficiency separations as they can be operated at high flow rates with low back pressures. However, in the current market the variety of ceramic based monoliths available compared to that of particle packed columns is very limited, with only three moieties available—silica, C8 and C18. If the impact of monolithic columns is to reach the same level as that of particle packed columns, a broader range of stationary phase chemistries is required. In particular the development of *in situ* modification methods for silica monoliths would be worthwhile as it would allow chromatographers the opportunity to prepare columns with tailor-made surfaces, more in tune with their own separation problem.

The majority of the research published to date that involve modifications of monoliths has been focused on both silica and polymer based capillary monoliths, for applications in capillary electrochromatography (CEC). There has not yet been a strong drive for the modification of the silica rods encased in PEEK tubing that are suitable for analytical scale chromatography, i.e. ~4.6 mm i.d. A recent review by Núnez et al. [1] detailed methods for the chemical modification of silica monolithic columns, albeit most were for capillaries, aimed at microscale chromatography.

There are only few studies involving modified analytical scale silica monoliths in the current literature [2–9]. Sutton and Nesterenko prepared an aminopropyl-silica monolith [3], which was then used in normal phase HPLC for the separation of alkyl benzenes, and the sub-fractionation of a complex petroleum fraction. The column efficiency of the modified monolith measured in the number of theoretical plates (N) was less than aminopropyl-silica particulate columns. Aminopropyl-silica monoliths were also prepared by Bayer et al. [2] and Lubda et al. [5], who further modified these monoliths with a cyclodextrin (CD) derivative and used them for enantiomeric separations. The column efficiency for 2-nitroanisole was 73,584 N/m for the column prepared by Bayer et al. [2] after amino-propyl derivatisation, but there was no measure of the column efficiency following CD derivatisation. The column prepared by Lubda et al. [5] showed that the highest N value reached was 36,770 N/m using methadone. A similar method used by Lubda et al. [5] was also used to prepare a *tert*-butyl-carbamoylquinine

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doi:10.1016/j.chroma.2010.07.052

modified monolith (*t*-BuCQN) [6] with high *N*/m values. Yin et al. [4] prepared a *p*-*tert*-butylcaliz[8]arene modified monolith that was used for the separation of dihydropyridines and fullerenes. The column efficiency using naphthalene was not as high as the values reported by Bayer et al. or Lubda et al. for their columns, with only 22,400 *N*/m. Sugrue et al. [7] modified a silica monolith with iminodiacetic acid (IDA) for the separation of alkaline earth, alkali and selected transition metal ions. The efficiency for the column prepared with this *in situ* method using Ba (II) was 18,050 *N*/m. Yang et al. [8] studied the efficiency of columns as a result of end-capping procedures using *N*-(trimethylsilyl)imidazole (TMSI). They showed that commercial C18 columns, as well as self modified columns, could be improved using their technique. No information was given, however, with respect to the carbon load of either the C18 ligands or the end-capping agent. Calleri et al. [9] prepared a penicillin G acylase (PGA)-based monolithic silica support as a chiral stationary phase. Column efficiencies measured at half height were higher than for comparative particle packed columns, but peaks were more asymmetrical on the monolithic column. The surface coverage was 2.9 μmol/m^2.

In this study, we illustrate the cyano modification of analytical scale silica rod encased monoliths using a procedure similar to that of Bayer et al. [2] and Lubda et al. [5,6]. We employed these procedures (with some modifications) as they yielded the highest column efficiencies for analytical scale columns reported in the literature. The procedure employed here involves a sylation reaction using chloro(3-cyanopropyl)dimethyl silane in dry heptane. In the current literature there has not yet been a cyano modified silica monolith aimed for analytical scale chromatography, although Allen and El Rassi [10] have previously reported the preparation of a cyano bonded silica monolith prepared in micro-scale dimensions aimed for capillary electrochromatography (CEC). They prepared two types of cyano monoliths; a cyano monolith with an average efficiency of 213,000 *N*/m and another with a hydroxyl group acting as a spacer between the cyano ligand and the silica backbone (CN–OH–monolith), which had an average efficiency of 199,000 *N*/m. The separation performance of these two cyano phases was compared and they found that the CN–OH–monolith separated polar compounds more efficiently using normal phase capillary electrochromatography than the cyano monolith.

It is the purpose of this study to successfully bind this functionality to an analytical sized silica monolith and study the chromatographic behaviour, and in particular evaluate the ligand density along the axial direction of the bed.

2. Experimental

2.1. Chemicals

HPLC grade methanol, isopropanol, and heptane were obtained from Merck Pty. Ltd. (Kilsyth, Victoria, Australia). Heptane was dried by reflux over sodium. HPLC grade tetrahydrofuran was obtained from LabScan Analytical Sciences distributed by LOMB Scientific (AUST) Pty. Ltd. (Taren Point, NSW, Australia). Test solutes and chloro(3-cyanopropyl)dimethyl silane were obtained from the Aldrich Chemical Company, Inc. (Sigma–Aldrich Chemical Company Inc., Castle Hill, NSW, Australia). Chloro-trimethyl silane was obtained from Gelest (USA). An Onyx neat silica monolith (100 × 4.6 mm) was purchased from Phenomenex Pty. Ltd. (Lane Cove, NSW, Australia).

2.2. Equipment

Chromatographic tests 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, DGU-14A online degasser, SPD-M10A*VP* diode array detector (set at 254 nm), and Shimadzu Class-*VP* version 6.14 software on a Pentium III 700 MHz processor. In addition to the on-line degasser, mobile phases were periodically sparged with helium. The temperature of the column was thermostated at 30 °C using a HPLC column heater (Thermasphere TS-130) from Phenomenex.

The analysis for total carbon was performed at the Central Science Laboratory, University of Tasmania, using a Thermo Finnigan EA 1112 Series flash elemental analyser.

2.3. In situ modification method

Prior to surface modification, dried heptane (50 mL) was pumped through the monolith. A 1% v/v solution of chloro(3-cyanopropyl)dimethyl silane in dried heptane was used as the cyano ligand bonding silane solution and a 1% v/v solution of chlorotrimethyl silane was used as the end-capping silane solution. The cyano silane solution was pumped through the monolith at 30 min intervals (at flowrates of up to 4 mL/min) using each time five column volumes of the silane solution until 100 mL had passed through the monolith. This step was repeated twice in the forward direction and twice in the reverse direction. Once completed, the end-capping silane solution was passed through the monolith (forward direction only) using the same procedure as for the cyano silane solution. These solutions were pumped through the monolith using a Waters 501 HPLC pump thermostated at 80 °C using a HPLC column heater (Thermasphere TS-130) from Phenomenex.

After completion of the sylation, the monolith was washed at room temperature with 100% heptane (50 mL), isopropanol (30 mL) and methanol (30 mL) using flowrates of up to 4 mL/min.

2.4. Column efficiency

Efficiency tests were performed before modification (normal phase on the bare silica bed), after CN ligand modification, and after end-capping (reversed phase). Plate counts were determined using the second peak moment (variance) method, i.e., $N = t_r^2/\sigma^2$, where N is the number of theoretical plates, t_r is the retention time, σ is the standard deviation. Performance was measured at 20 different flow rates between 0.3 and 3 mL/min.

2.4.1. Normal phase (NP)

Column efficiency was measured using toluene (9 mmol/L) and acetone (68 mmol/L) dissolved in the mobile phase. The mobile phase was a binary mixture of heptane (50% saturated in water)/methanol (99.1/0.9) at ambient temperature.

2.4.2. Reversed phase (RP)

Column efficiency was measured using acetone (136 mmol/L) and anisole (87 mmol/L) dissolved in methanol. The mobile phase was a binary mixture of methanol/water (20/80) at 30 ± 0.2 °C.

2.5. Linear chromatographic separations

Alkyl benzene test solutes were dissolved in methanol/water (80/20), and made up to concentrations between 7 and 14 mmol/L. The mobile phase was prepared accurately by mass, correcting also for the solvent density at the laboratory temperature at the time of preparation. Polycyclic aromatic hydrocarbons (PAH) test solutes were dissolved in THF and then diluted further in methanol/water (80/20), and made up to concentrations between 0.01 and 3 mmol/L. Chromatographic behaviour was assessed in at least four mobile phases (methanol/water compositions), at a constant temperature (30 ± 0.2 °C), and at a flow rate of 3 mL/min. Experiments were randomised, and duplicates were performed for

each injection. Void volumes were measured using the minor disturbance method [10].

2.6. Longevity

The stability of each column, i.e., silica bed quality and surface coating was tested using a standard test mix of substituted aromatics (acetone, *p*-cresol, benzene, anisole, toluene and phenetole) dissolved in methanol/water (40/60) and made up to concentrations between 3 and 52 mmol/L run initially and at every 500 column volumes of solvent passage (methanol/water). The log of the retention factors (log *k*) were calculated for each test solute and compared throughout the entire duration of the experimental work to ensure the integrity of the stationary phase. These longevity tests sought only to ensure that the stationary phase remained stable throughout the duration of the selectivity tests.

2.7. Surface coverage

The homogeneity of the *in situ* method was determined through the measurement of percentage of carbon (%C) through elemental analysis. Each column was cut into sections ~1 cm in length (8–10 sections per column) and the carbon load was measured for each section. Characterisation by this method is destructive and therefore all chromatographic studies were completed prior to the surface coverage measurement. The surface coverage of four columns (two commercial C18 columns, one C18 modified monolith and the cyano modified monolith) was calculated according to the Berendsen equation (Eq. (1)) [12,13].

$$x = \frac{10^6 \times \%C}{1200n_c - \%C(M_1 - n_c)} \times \frac{1}{S_{BET}} \quad (1)$$

Here, x is the ligand density in μmol/m^2, n_c is the number of carbon atoms of the ligand, M_1 is the formula weight of the ligand, and S_{BET} is the specific surface area of the unmodified support. It is difficult to measure the ligand density for the %C associated only to the bonded ligand as characterisation involves the destruction of the sample. Therefore the %C measured represents the total carbon of the ligand bonded and the end-capping agent. For the cyano column the surface coverage for the ligand was calculated using the same equation replacing the %C with %N (% loss of nitrogen) and n_n (number of nitrogens in ligand) instead of n_c. The surface coverage for the end-capping ligand for the cyano column was calculated by subtracting the ligand surface coverage (calculated from the %N) from the surface coverage associated to the ligand and end-capping surface coverage (calculated from the %C).

3. Results and discussion

The modification conditions employed in this work were chosen according to processes that we have previously used in the modification of particles, the difference being that the current technique was adapted to an *in situ* process, which was necessary because the monolithic bed is itself prepared within the column tubing. The development of this *in situ* method was kept as simple, cost and time efficient as possible to produce a chemically bonded analytical sized monolith [2,5,6,14].

3.1. Column efficiency

The performance of the native silica monolith prior to modification displayed typical performance expected of this type of column (Fig. 1a and b, Si labelled curve). The monolith efficiency was tested after modification with the cyano functionality and also after end-capping. Fig. 1a illustrates the column efficiency for an unretained marker using toluene (NP) and acetone (RP) and Fig. 1b for a retained marker using acetone (NP with a *k* value of 1.25) and anisole (RP *k* values of 1.27 for the cyano phase and 1.40 for the cyano end-capped phase). Both efficiency plots show that after each modification step, the monolithic performance remained similar to that of the native silica monolith (Fig. 1a and b, curves labelled CN and CN end-capped). The highest *N*/m value was 81,650 using anisole on the end-capped cyano bonded monolith.

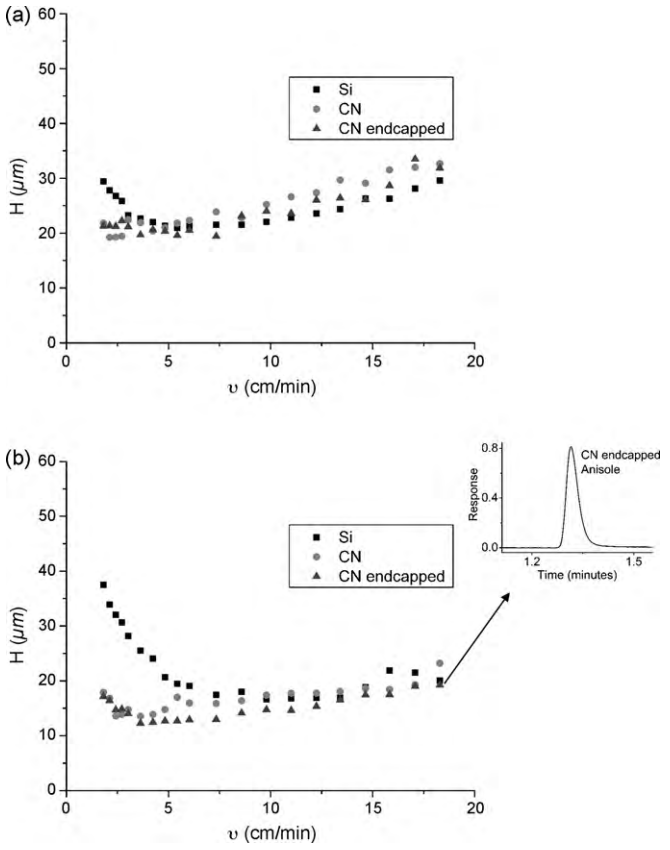

Fig. 1. HETP curve using an unretained marker (a) and retained marker (b). Inset illustrates anisole peak shape at 3.0 mL/min.

3.2. Longevity

Column and stationary phase stability were tested using a standard test mix of acetone, *p*-cresol, benzene, anisole, toluene and phenetole. Retention of these compounds was assessed approximately every 500 column volumes. No stationary phase degradation was observed, within the period of study, which exceeded 2000 column volumes. The log of the retention factor (log *k*) values for each test solute were calculated from the retention times and were compared for the entire duration of chromatographic work performed on each column. The largest difference of log *k* for this study was 0.08 (unretained marker) for acetone and 0.03 for the other test solutes. Fig. 2 details the variation of log *k* on the cyano-silica monolith with an illustration of the anisole peak shape at the various test points.

3.3. Methylene selectivity

The linear solvent strength theory (LSST) is used to determine and compare the retention of different solutes in a reversed phase chromatographic system [16]. The theoretical relationship between the retention factor (*k*) and the mobile phase composition

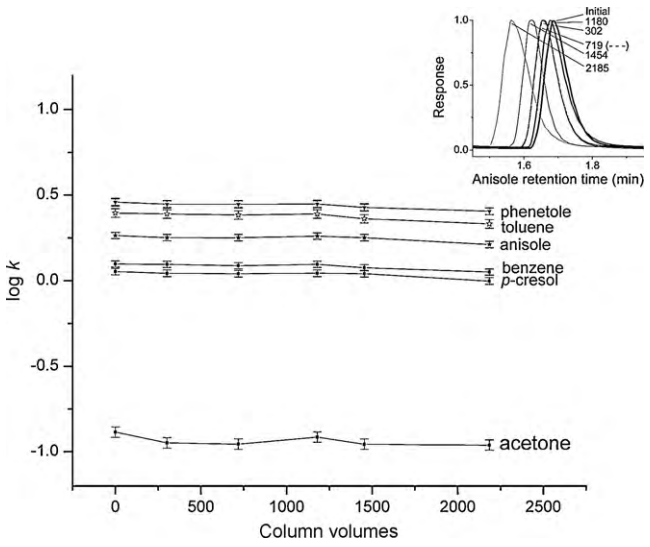

Fig. 2. Longevity: plot of log k vs column volumes through-put. Inset illustrates the peak shape during this period.

(ϕ) is described by Eq. (2).

$$\text{Log}\, k = \log\, k_w - S\phi \quad (2)$$

where k_w is the retention factor in pure water and S is the slope of a log k v ϕ plot and is analogous to the hydrophobic contact area [16]. The magnitude of S is useful for the optimisation of separations.

The chromatographic retention behaviour of the alkyl benzenes resulted in linear plots of log k vs ϕ as shown in Fig. 3a. All correlation coefficients were above 0.91. The S values derived from the log k vs ϕ plots for the alkyl benzenes are shown in Table 1. A plot of S vs the substituent alkyl chain length (n) is shown in Fig. 4a with an R^2 value of 0.9996. This type of plot gives an indication of the global methylene selectivity (across all solvent compositions) and provides more meaningful information than a simple plot of log k verses n at a single composition. The methylene selectivity results fit the LSST with S increasing as the alkyl chain increased with excellent linearity.

3.4. Phenyl selectivity

Plots of log k vs ϕ for the linear PAHs on the cyano bonded monolith are linear as shown in Fig. 3b with correlation coefficients above 0.98. The retention of anthracene and 2,3-benzanthracene was almost coincident across the entire solvent range tested, which is interesting since there was not a concordant increase in retention consistent with the addition of an addition phenyl ring. This is discussed in more detail following.

S values derived from these plots are given in Table 1, which also includes values from a previous study involving different particle packed stationary phases [17]. The magnitude of the S value for each linear PAH (with the exception of benzene) was greater on the cyano monolith than on the other columns tested, which included a variety of phenyl-type stationary phases. This is a surprising result, which suggests a larger hydrophobic contact area between the stationary phase and the test solutes given that the propyl-cyano stationary phase produces a relatively shallow layer in comparison to the C18 and phenyl-type stationary phases [17], which reinforces the significance of the solute–stationary phase interactions that are experienced by the PAHs on this type of surface, and the relative change in the strength of these interactions as a function of the mobile phase composition.

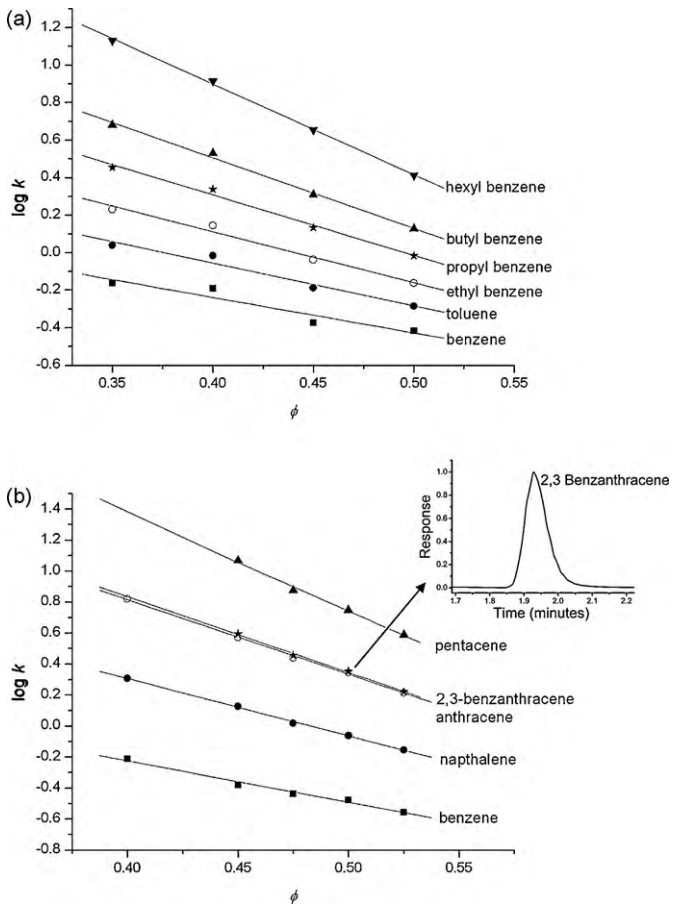

Fig. 3. Log k vs Φ at 30 °C for the alkyl benzenes ($R^2 > 0.91$) (a) and linear PAHs ($R^2 > 0.98$) (b). Inset illustrates the peak shape for 2,3-benzanthracene at a mobile phase composition of 50/50 methanol/water at 3.0 mL/min.

Fig. 4b illustrates a plot of S vs n (where n represents the number of aromatic rings) for the linear PAHs. This plot represents the phenyl selectivity of the phase across all mobile phase compositions (since S is a measure of the slope of the log k v ϕ plot). The relationship between S and n was discontinuous between the three ring and four ring PAH members in the series (anthracene and 2,3-benzanthracene). This trend was also observed in the work of Kayillo et al. [17–19] and Stevenson et al. [14] on a variety of phenyl-type and C18 phases, and was a consistent trend at temperatures of 30 °C [14] and 40 °C [17–19]. The importance of this discontinuity in S vs n is that there is not a monotonic increase in resolution as a

Table 1
S values for alkyl benzenes and linear PAHs [17].

Alkyl benzenes	CN	Linear PAHs	CN	C18	C18Aqua	Propyl-phenyl	Syngery polar-RP	Cosmosil 5PBB
Benzene	1.90	Benzene	2.67	2.89	2.75	2.51	2.71	2.61
Toluene	2.29	Napthalene	3.70	3.51	3.3	3.06	3.37	3.19
Ethylbenzene	2.72	Anthracene	4.81	4.23	3.96	3.92	4.13	3.71
Propylbenzene	3.23	2,3-Benzanthracene	4.90	3.8	3.49	4.09	4.04	3.85
Butylbenzene	3.76	Pentacene	6.28	4.42	4.19	5.14	4.91	4.25
Hexylbenzene	4.84							

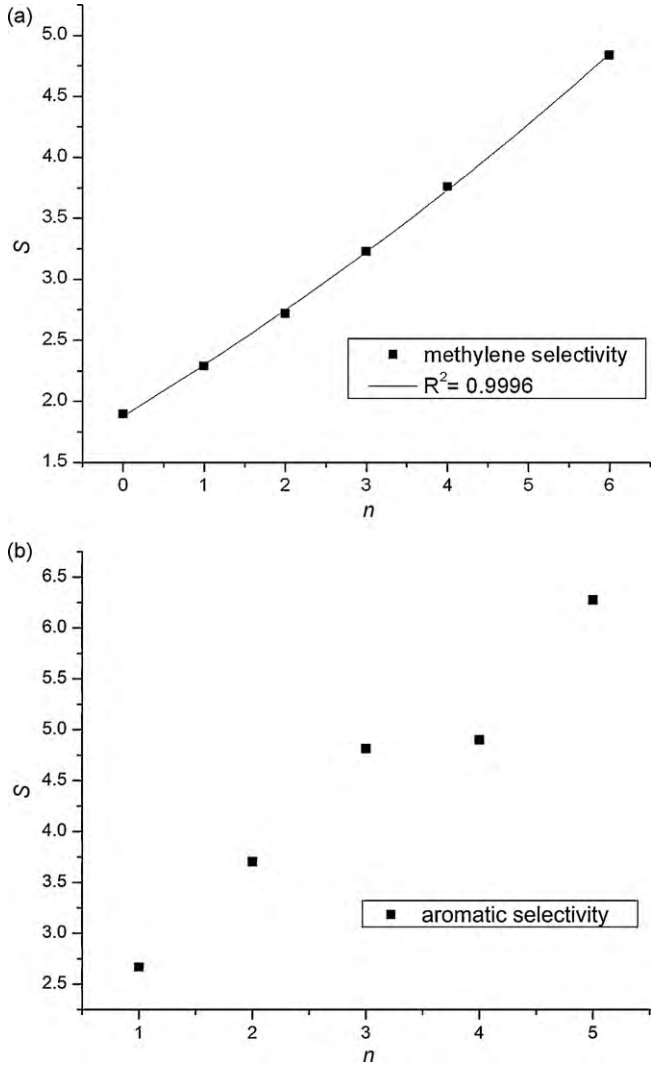

Fig. 4. Plots of *S* vs *n* for alkyl benzenes (a) and linear PAHs (b).

function of solvent strength between all members of the homologue series and at some point in time, the elution orders may change, and the components anthracene and 2,3-benzanthracene exhibit co-elution with limited phenyl selectivity, further complicating separation optimisation processes. This translates into a more complex separation problem for the separation of analytes with no specific relationship. The cause of this discontinuity is still under investigation, but it is interesting that such an effect occurs irrespective of the type of stationary phase ligand. Our initial studies on phenyl phases, for which we observed this same effect, indicate that the extent of this discontinuity is related to the

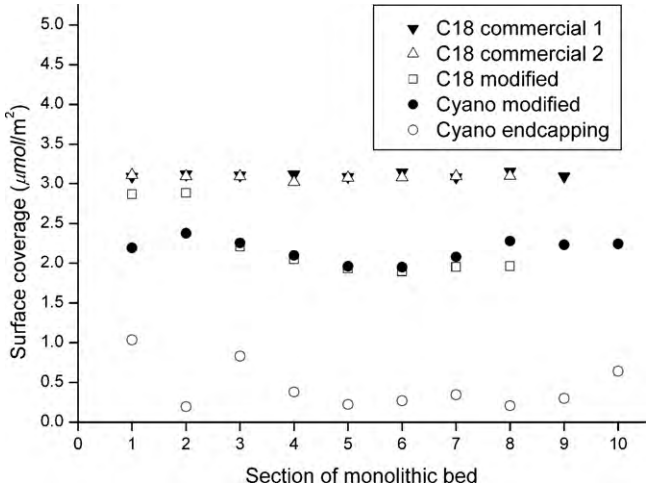

Fig. 5. Plot of surface coverage (μmol/m²) as a function of axial location of the monolithic bed (1 = inlet).

carbon loading of the phase [14]. These results indicate that there exists essentially a critical molecular size in relation to the ligand environment such that retention and the resulting selectivity is not consistent with the increase in the molecular size. We have discussed this in greater detail in reference [17]. We should point out, however, that the retention was not inconsistent with chromatographic predictions in relation to retention, that is, within certain limits (dependent on solvent composition, the type of column and the stationary phase ligand density [17]) retention increased monotonically for higher order members of the homologue series.

3.5. Surface coverage results

Initial trials into the *in situ* modification of silica rod monoliths were undertaken using C18 phases. These studies involved pumping a total of 20 mL of the C18 silylation solution (1% v/v solution of chlorodimethyloctadecyl silane in dried toluene) through the monolith, followed by a period of non-flow for 30 min. This procedure was repeated for a total of 5 h (eluting 2 mL of the C18 sylation solution every 30 min). Following the C18 modification of the silica rod monolith the column was dissected and the carbon load was determined at intervals of approximately 1 cm along the column axis. The surface coverage results are detailed in Table 2, and plotted in Fig. 5. On this C18 column, ligand density within a 3 cm region from the inlet was ~33% higher than in the remaining sections of the bed, after which the carbon load was essentially homogeneous.

Commercial C18 monoliths were subsequently dissected in order to assess whether these columns were axially homogenous with respect to the carbon load. Two columns were tested, and the ligand densities of these columns are also detailed in Table 2 and plotted in Fig. 5. The magnitude of the carbon load for both commer-

Table 2
Surface coverage results (μmol/m²).

Section	C18 commercial 1	C18 commercial 2	C18 modified	Cyano modified	Cyano end-capping
1	3.09	3.11	2.87	2.19	1.03
2	3.12	3.09	2.89	2.38	0.20
3	3.11	3.09	2.21	2.26	0.83
4	3.12	3.02	2.05	2.10	0.38
5	3.09	3.08	1.93	1.96	0.22
6	3.14	3.08	1.90	1.95	0.27
7	3.08	3.10	1.95	2.08	0.34
8	3.15	3.10	1.96	2.28	0.21
9	3.09			2.23	0.30
10				2.24	0.64

cial C18 monoliths was constant and their values uniform across the entire axial length of the column. It was interesting to note also, that the carbon load of the laboratory prepared C18 monolith had a similar carbon load to that of the commercial C18 monolith at the column inlet, prior to the decrease that was observed after the inlet 3 cm region. Following these results we deduced that the unidirectional flow through the monolith may have been the cause of the non-uniform carbon load of the C18 phase, despite the period of time (30 min) in which the silane reagent remained stagnant in the rod column. Hence, future modifications of the rod monoliths were undertaken using bidirectional flow through the column, with periods of 30 min of zero flow, and this was the procedure used here for the cyano modified monolith.

The surface coverage results for the cyano modified rod monolith are also detailed in Table 2 and plotted in Fig. 5. We were able to determine the surface coverage of the cyano ligand and the end-capping agent separately using the %N and %C elemental analysis results. We found that the end-capping agent was bound heterogeneously distributed across the column in the axial direction, and that the cyano ligand was distributed in a parabolic distribution, with the inlet and outlet sections having the highest ligand densities. The minima of this distribution corresponded to the column mid point. Unlike the commercial C18 columns these rod columns were not homogeneous, despite the multiple forward and reversed flows through the bed and the stagnant periods of reaction. However, the degree of heterogeneity was less than for the C18 modified monolith prepared using only the unidirectional flow through the bed.

Unfortunately, evaluating the homogeneity of the ligand density of the monoliths is destructive. This largely explains why, to date, there are only two papers [5,6] that describe the heterogeneity of monolithic phases. In the study by Lubda et al. [5], a two step procedure was used to prepare a cyclodextrin phase. The first step involved the preparation of an aminopropyl modified phase, which was then subsequently modified with the cyclodextrin phase. The authors reported that the modification process for the first step in the preparation of the cyclodextrin phase, i.e. the aminopropyl phase, resulted in a homogeneous surface coverage, although close inspection of the results reveal the same degree of heterogeneity that we report here, except that, in their work, three regions only were tested for carbon load. No information regarding the homogeneity of the final step in the preparation of the cyclodextrin phase was presented.

The ramifications of the non-uniformity of the carbon load can be serious, because the carbon load can influence the retention mechanisms (adsorption/partitioning) [20], and how the ligands are structurally organised onto the silica surface [20–22]. Furthermore S vs n plots (which influences the optimisation as discussed in previous Sections 3.3 and 3.4) are also influenced by carbon load [14].

Gritti and Guiochon [20] showed that changing the ligand density of C18 stationary phases impacted the retention process in accord with the degree of coverage. This occurred as the topology of the stationary phase surface changed; ligand densities with less than 2 μmol/m^2 resulted in large voids between chains. The chains also tended to cluster, leaving large spaces providing the analyte, and solvent, access to the end-capped surface. Higher ligand densities (greater than 3 μmol/m^2) resulted in close-packed monolayers with few inter-ligand cavities. They found that as the surface coverage increased the average distance between C18 chains decreased from 15.1 to 5.5 Å for surface coverages of 0.42–3.15 μmol/m^2, respectively.

Lork and Unger [22] proposed that for alkyl phases, once the critical ligand density has been reached, stationary phase ligands are closely packed, the chains have restricted freedom and the ability for solute penetration into the stationary phase is strongly reduced. They found that the critical ligand density was not fixed, but had a range of 2.3–3.2 μmol/m^2 depending on the ligand chain length and the size of the solute molecule.

Critical ligand density, and therefore, stationary phase topology have an impact on solute retention processes. For example, as the C18 ligand bonding density increases above 2 μmol/m^2 the space between adjacent C18 ligands decreases, and the mechanism of retention shifts towards adsorption, rather than partitioning [20]. Retention, as a function of ligand density, thus decreases as the ligand density increases, as once a critical ligand density is achieved, only the outer surface of the bonded-phase is accessible for solute interaction, indicating that stationary phase ligands require a degree of flexibility in order to interact effectively with the analytes [21,22]. As the surface coverage increases the exclusion of larger molecules becomes more apparent, even though smaller molecules are still able to penetrate into the bonded layer. At some point, however, a surface coverage would be achieved, where, even for small molecules retention is almost exclusively dominated by adsorption and the analyte then can only explore part of the length of the alkyl chain [20,21]. Subsequently, variations in ligand density across the column length may lead to significant changes in retention mechanisms, which may be solute dependent.

4. Conclusion

A cyano functional silica based monolithic column was successfully prepared using a simple silylation procedure in less than two days. Efficiency tests showed that the integrity of the monolith itself was not comprised during the modification procedure. The highest number of theoretical plates (N/m) using anisole was 81,650. Longevity tests undertaken on a range of substituted aromatics showed that the column performance was stable for over 2000 column volumes. The methylene selectivity of the cyano phase showed the plots of S vs n (n = member of the series) to be linear, whereas the aromatic/phenyl selectivity for a homologous series of linear PAHs showed discontinuity in the S vs n plot at the four member ring (2,3-benzanthracene). This had been observed on other types of phases (C18 and Phenyl-type) particle packed columns. Conceivably this type of modification process, due to its simplicity, will allow chromatographers the option to prepare almost any type of monolith-modified stationary phase and to overcome the limitations associated with selectivity in the ceramic based monolith commercial market. However, future work is needed in order to improve the homogeneity of the surface coverage throughout different sections of the monolithic bed.

Acknowledgments

AS acknowledges the receipt of a UWS Post Graduate Research Award. This research was supported under Australian Research Council's Discovery Projects funding scheme (project number DP0987318). We also acknowledge Dr. Thomas Rodemann of the Central Science Laboratory, UTAS for assistance with elemental analysis.

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