Electronic Supplementary Information for

Silicon Oxynitride Microspheres as Stationary Phase for High Performance Liquid Chromatography This is the title

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Experimental

Reagents. All reagents were used without further purification and the following chemicals and reagents were used: spherical silica from Fuji Silysia Chemical (Kasugai, Japan); NH_3 (99.999%) from gas supplier center of dalian institute of chemical physics; nonanoyl chloride from Alfa Aesar (Ward Hill, MA, USA); acetonitrile (CH_3CN, HPLC grade) from fisher Chemicals (Fair Lawn, NJ, USA); NH_3/H_2O (10%) from Fluka (Switzerland); HAc and NH_4Ac from Acros (Cambridge, USA). All the water used in experiments was purified with a Milli-Q system (MA, USA). All solvents employed in the derivatization of sph-SiON microspheres, and test solutes for chromatographic evaluation were analytical quality.

Sph-SiON Preparation. In a typical synthesis, spherical amorphous silica (3.0 g) was placed in a quartz boat and inserted into tubular quartz furnace. Prior to nitridation treatment, the quartz tube was purged by N_2 for 2 h to remove the air. Afterwards, the temperature of the furnace was increased at a ramp rate of 5 $^oC\ min^{-1}$ and maintained at 1050 ^oC for 12 h under the NH_3 atmosphere with a flow rate of 250 mL min^{-1}. The furnace was then cooled down to room temperature and purged again with N_2 for 1 h. The process was repeated for 10 times to ensure homogenous nitridation.

Sph-SiON Surface Modification. Nonanoyl chloride was used to functionalize sph-SiON spheres. sph-SiON (4.0 g) was dried in vacuum 100 ºC for 6 h and dispersed in dried toluene (100 mL) under N_2. Subsequently, dried pyridine (400 μL) and nonanoyl chloride (6.4 mL) in 20 mL of dried toluene were added, separately. The reaction mixture was refluxed for 12 h under N_2 and then filtered. The obtained nonanoyl bonded sph-SiON spheres were washed with toluene, methanol, methanol

/H_2O, and methanol, respectively. The resultant modified spheres were dried at room temperature for 6 h and then in an oven at 80 ^oC overnight.

Apparatus. Powder X-ray diffraction (XRD) patterns are recorded on an X'Pert Pro (PANAnalytical) diffractometer with Cu Ka radiation at 40 kV and 40 mA. Scanning electron microscopy (SEM) images were recorded using a JEOL JSM-820 scanning electron microscope. N_2 adsorption/desorption experiments were undertaken isothermally at 77 K on an automatic ASAP 2010 Micromeritics apparatus. The samples were degassed at 300 ^oC and 10^{-3} Pa for 2 hours prior to the adsorption measurements. Specific surface areas were calculated using Brunauer-Emmet-Teller method based on adsorption data in the partial pressure (P/P_0) range 0.05-0.2 and total pore volume was determined from the amount of the nitrogen adsorbed at P/P_0=0.99. Elemental analysis of the nitrided samples was measured using a Vario EL III elemental analysis system (Elementar, Germany). All solid-state NMR spectra were recorded on a Varian Infinityplus-400 specctrometer using a 5 mm MAS probed. $^1H \rightarrow ^{29}Si$ CP/MAS NMR spectra were recorded at 79.4 MHz and a spinning rate of 6 kHz with a contact time of 6 ms, a recycle delay of 2 s. Chemical shift were referenced to 4,4-dimethyl-4-silapentane sulfonate sodium (DSS) at 0 ppm. The spectra were accumulated for 640 scans with a $\pi/4$ pulse width of 1.7 μs and a 30 s recycle delay. $^1H \rightarrow ^{13}C$ CP/MAS NMR spectra were recorded at 100.5 MHz and a spinning rate of 8 kHz with a contact time of 8 ms, a recycle delay of 2 s. The chemical shifts were referenced to admantane with the upfield methine peak at 29.5 ppm. Infrared spectra were recorded using a Bruker Equinox55 FTIR spectrometer on self-supporting sample wafers in a Pyrex vacuum IR cell. Prior to the collection of spectra the samples were heated in the cell for 1 h at 490 ^oC under vacum, after which the spectra were recorded at room temperature. The ζ-potential measurements were carried out using a Malvern Zetasizer Nano-ZS90 instrument (Malvern, UK). Stock sample solutions were prepared by suspending 50 mg of material in water (30 mL). The final samples for ζ-potential measurement were prepared by mixing stock sample (1 mL) with ammonium acetate buffer (100 mM, 2 mL), and then diluted with water to 10 mL. After a suspension solution was made, it was thoroughly mixed and

immediately thereafter transferred to the measurement cell.

Chromatographic Evaluation. The unmodified or modified sph-SiON, or SiO_2 microspheres were packed into the stainless-steel tubes (length of 150 mm or 50 mm, inner diameter of 4.6 mm) by slurry packing method under a maximum pressure of 40 MPa using methanol as propulsion solvent. Experiments were performed on a Waters HPLC system, which consisted of a Waters 2695 HPLC pump, a Waters 2489 diode array detection (DAD), and a Waters 2424 evaporative light scattering detection (ELSD) system (Waters, Milford, MA, USA). The Empower workstation software was used for data recording. All high pH chromatographic experiments were performed on the HPLC system (Agilent 1100 Series, USA) consisted of a quaternary pump, an auto-sampler, a degasser, an automatic thermostatic column compartment, and a diode array UV detector. Chromatographic parameters, e.g. plate number and column pressure, were determined for each column using a standard test protocol. All chromatographic experiments were performed at flow rate of 1.0 mL min^{-1} and column temperature of 30 ^{o}C. The ELSD detection for oligosaccharide was set as: gas pressure 30 psi, tube temperature 75 ^{o}C, and gain 10. The UV detection for other analytes was 254 nm. All the compositions of mobile phase were mixed with volume ratios.

The sph-SiON phase packed in 50 mm × 4.6 mm i.d. column was subjected to an aggressive aging test using mobile phase of 20 mM NH_4Ac adjusted by 10% aqueous NH_3 at pH 9.0 at 30 ^{o}C. After each 60 min time period, the column was washed with H_2O for 10 min, equilibrated with 90:10 ACN/ H_2O for 6 min, and 1μL of solution composed of toluene and cytosine was automatically injected and separated using the same mobile phase. To detect any change, the chromatographic plate number and column pressure were monitored as functions of the volume of the pH 9.0 mobile phase that had passed through the columns. For comparison, a commercial silica phase was packed into similar 50 mm × 4.6 mm i.d HPLC columns and submitted to the same aging test.

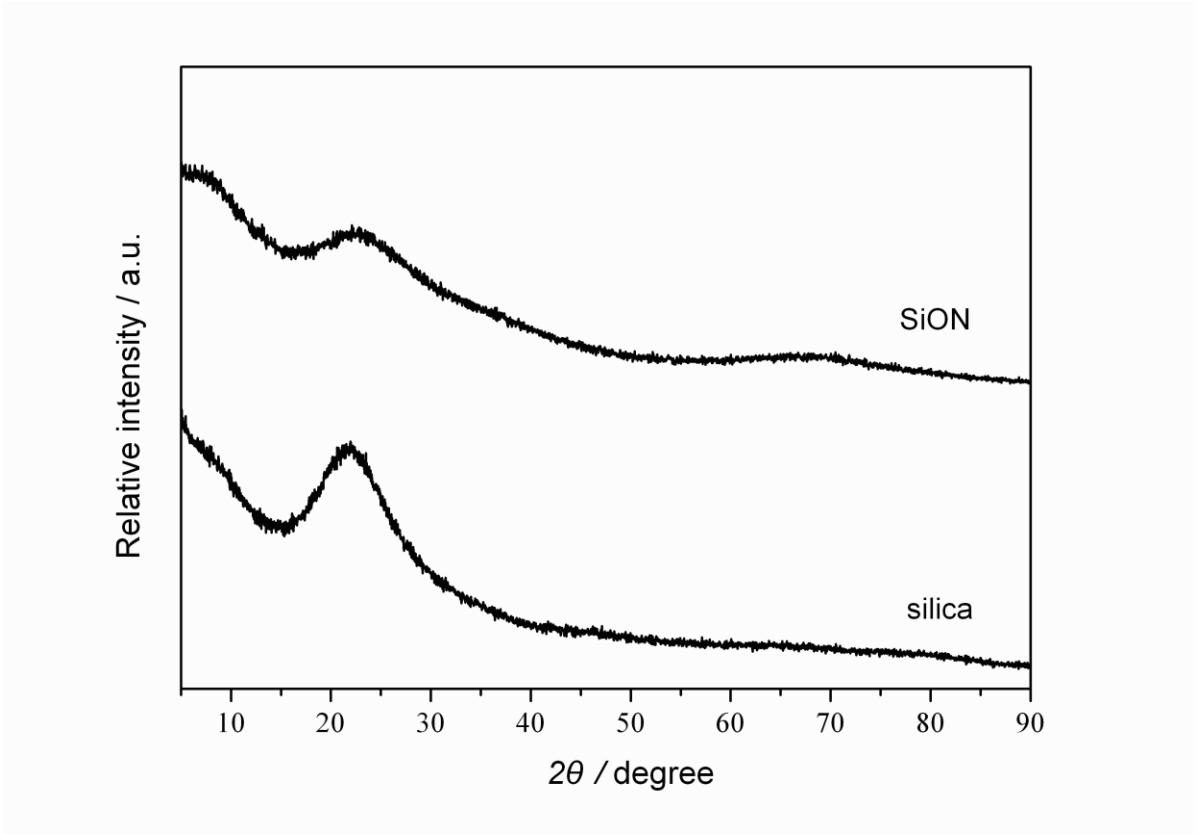

Fig. S1 XRD patterns of parent silica and sph-SiON.