New Strategy for Drastic Enhancement of Selectivity via Chemical Modification of Counter Anions in Ionic Liquid Polymer Phase

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Experimental details

Chemicals

1-Vinylimidazole (99%) was purchased from TCI (Tokyo, Japan). 1-Bromooctadecane was obtained from Wako (Osaka, Japan). 3-Mercaptopropyltrimethoxysilane (MPS) was purchased from Azmax (Chiba, Japan). Azobisisobutyronitrile (AIBN) was obtained form Nacalai tesque, Inc. (Kyoto, Japan) and purified by recrystallization from methanol before use. Methyl orange (MO) was obtained from Kanto Chemical Co., Inc. (Kyoto, Japan). Porous silica particles (diameter 5 μm, pore size 300 Å, specific surface area 99 m^2 g^{-1}) were kindly supported from Fuji Silysia Chemical Ltd. (Aichi, Japan). All PAHs and alkylbenzenes were commercially available and used without any purification. Standard Reference Material (SRM) 869b, Column Selectivity Test Mixture for Liquid Chromatography, was obtained from the Standard Reference Materials Program (NIST, Gaithersburg, MD). Prednisolone, estriol, testosterone, estrone, and 17β-estradiol were obtained from Sigma (St Louis, MO, USA). 17α-Estradiol was purchased from Dr Ehrenstorfer (Augsburg, Germany).

Synthesis

Sil-MPS. The activated silica (3.0 g) was suspended in 15 ml of dry toluene and then an excess of 3-mercaptopropyltrimethoxysilane (1.5 g) was added in a 50 ml round-bottomed flask with three necks. The suspension was mechanically stirred and refluxed for 48 h. After refluxing, the reaction was stopped and the modified silica was cooled to room temperature, and washed with large volumes of toluene, methanol, deionized water, methanol and ether successively. Then Sil-MPS was dried under vacuum. Elemental analysis result of Sil-MPS: C, 1.22%; H, 1.00%; 3.5 μmol m^{-2}.

[C_{18}VyIm]Br. 1-Vinylimidazole (4.71 g, 0.05 mol) was solved in 50 mL acetonitrile; a little excess 1-bromooctadecane (18.33 g) was added. The mixture was stirred at 60 °C for three days. The obtained [C_{18}VyIm]Br was precipitated in diethyl ether and dried under vacuum, giving a white solid powder: yield 96.5%; [C_{18}VyIm]Br ($C_{23}H_{43}BrN_2$): Calcd. C 64.62, H 10.14, N 6.55; Found: C 62.86, H 9.75, N 6.41. ^{1}H NMR (CD_3Cl, 400 MHz, ppm): δ 11.38 (1H, s), 7.48-7.54 (2H, m), 7.26-7.32 (1H, d), 5.89-5.94 (1H, m), 5.43-5.46 (1H, d), 4.39-4.43 (2H, t), 1.92-2.00 (2H, m), 1.35-1.37 (2H, d), 1.25 (28H, s), 0.86-0.90 (3H, t).

Sil-PImC$_{18}$-Br. [C_{18}VyIm]Br was grafted onto Sil-MPS through a surface radical chain-transfer polymerization as shown in Fig. 1. Sil-MPS (3.0 g) was added to a 100 mL three-neck round-bottomed container. [C_{18}VyIm]Br (3.0 g) dissolved in 12 mL of chloroform and 0.03 g of AIBN was added into the container. The mixture was stirred at 60 °C for 24 h. The precipitates were filtered and washed with chloroform, methanol, and diethyl ether, and dried under vacuum. Elemental analysis result of Sil-PImC$_{18}$-Br: C, 9.87%; H, 1.93%; N, 0.88%; 3.74 μmol m^{-2}. Sil-PImC$_{18}$-Br-1 was prepared with a similar condition and packed as a referenced column. The elemental analysis result of Sil-PImC$_{18}$-Br-1: C, 12.2%; H, 2.5%; N, 1.13%; 5.04 μmol m^{-2}.

Sil-PImC$_{18}$-MO. Sil-PImC$_{18}$-Br (3.0 g) was dispersed in the mixed solvent (methanol/ chloform/ water = 1: 1: 6) and then stirred with a little excess methyl orange dye at room temperature. After 6 h, the precipitate was washed with large mixed solvent stated above, DMF and diethyl ether. The obtained Sil-PImC$_{18}$-MO was packed into a column and characterized after being dried under vacuum. The elemental analysis result of Sil-PImC$_{18}$-MO: C, 13.52%; H, 1.98%; N, 2.04%; 3.57 μmol m^{-2}.

Characterization

Elemental analyses were carried out on a Yanaco CHN Corder MT-6 apparatus (YANACO Co., Ltd., Kyoto, Japan). Diffuse reflectance infrared Fourier transformation (DRIFT) spectra were obtained on a FT/IR–4100 (JASCO Co., Ltd, Tokyo, Japan) in the range of 4000–400 cm^{-1}. Thermogravimetric analysis was performed on a Seiko Exstar 6000 TG/DTA 6200 thermal analyzer (Seiko Instruments Inc., Chiba, Japan) in static air from 35 to 800 °C with a heating rate of 10 °C min^{-1}. ^{1}H NMR spectra for [C$_{18}$VyIm]Br were recorded by a JEOLJNM-LA400 instrument (JEOL, Tokyo, Japan). Differential scanning calorimetric measurements (DSC) were carried out from 10 to 90 °C at a heating rate of 2 °C min^{-1} using a Seiko EXTRA 6000 with a DSC 6200 instrument and an empty pan as reference.

Chromatographic conditions

The Sil-PImC$_{18}$-Br-1 and Sil-PImC$_{18}$-MO stationary phases were packed into stainless-steel columns (150 × 4.6 mm i.d.) separately. Two C$_{30}$ columns (3 μm or 5 μm silica supports) (150 × 4.6 mm I.D.) were bought from Nomura Chemical Co., Ltd. (Seto, Japan). HPLC-graded methanol and Millipore water were used as components of the mobile phase. All samples were directly dissolved in methanol. The chromatographic system (JASCO, Tokyo, Japan) consisted of a LC-NetII/ADC communication device, a DG-2080-53 3 Line degasser, a PU-2080 Plus Intelligent HPLC pump, a UV-2075 Plus Intelligent UV/vis detector, and a Rheodyne injector with a 20 μl sample loop. All chromatographic data were obtained by a JASCO ChromNAV Chromatography Data System. The column temperature was controlled at 10 °C using a column jacket with a circulator having a heating and cooling system. The flow-rate was 1.0 mL min^{-1}, the detection wavelength was UV 254 nm, and the injection volume was 5 μL. The retention time of D_2O was used as the void volume (t_0) marker (The absorption for D_2O was measured at 400 nm, which is actually considered as the injection shock). The retention factor (k) of an analyte was calculated according to the equation: $k = (t_R - t_0)/t_0$, where t_R is the retention time of the analyte. The separation factor (α) is the ratio of the retention factors for the two solutes being analyzed: $\alpha = k_2/ k_1$. The water/1-octanol partition coefficient (log P), usually used to represent molecular hydrophobicity, was determined from the retention factor with the ODS column stated above as $\log P = 3.759 + 4.207 \log k$ ($r = 0.99997$), according to the procedure described in our previous work [1]. The data for C_{18} (mon), C_{18} (poly), Sil-ODA$_n$ and Sil-DSG were directly obtained from the reference [2].

References to the ESI

1. H. Ihara, T. Sagawa, Y. Goto and S. Nagaoka, *Polymer*, 1999, **40**, 2555.

2. M. M. Rahman, M. Takafuji, H. R. Ansarian and H. Ihara, *Anal. Chem.*, 2005, **77**, 6671.

Table S1 Elemental analysis.

Sample	%C	H%	N%	Br%	C/N	Coverage (μmol m^{-2})
Sil-MPS	1.22	1.00	-	-	-	3.50 (C)
Sil-PImC$_{18}$-Br	9.87	1.93	0.88	2.51	9.83	3.74 (N)
Sil-PImC$_{18}$-Br-1	12.20	2.50	1.13	3.23	9.72	5.04 (N)
Sil-PImC$_{18}$-MO	13.52	1.98	2.04	-	6.03	3.57 (N)

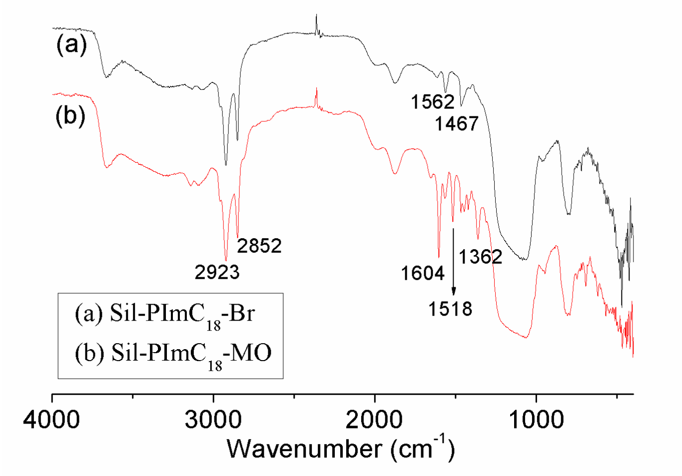


Fig. S1 DRIFT spectra of Sil-PImC$_{18}$-Br (a) and Sil-PImC$_{18}$-MO (b).

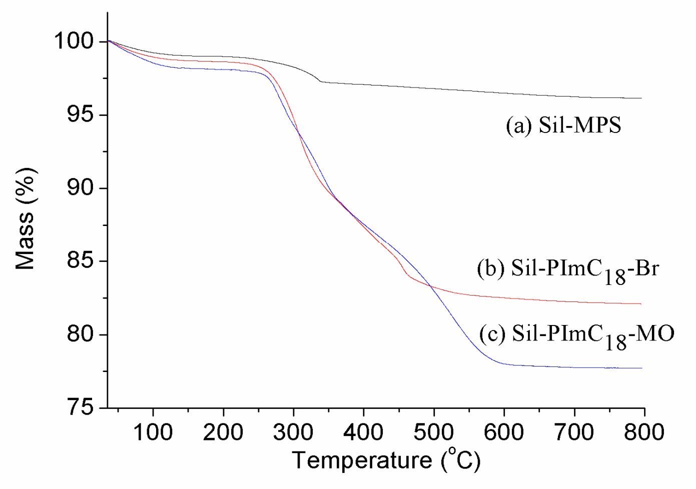


Fig. S2 Thermogravimetric curves of Sil-MPS (a), Sil-PImC$_{18}$-Br (b) and Sil-PImC$_{18}$-MO (c).

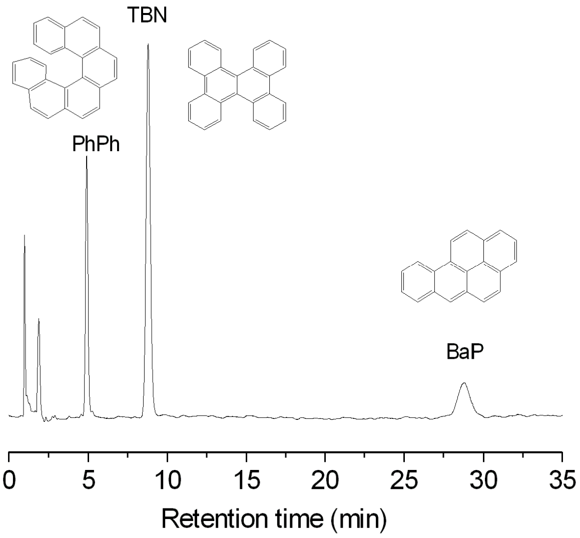
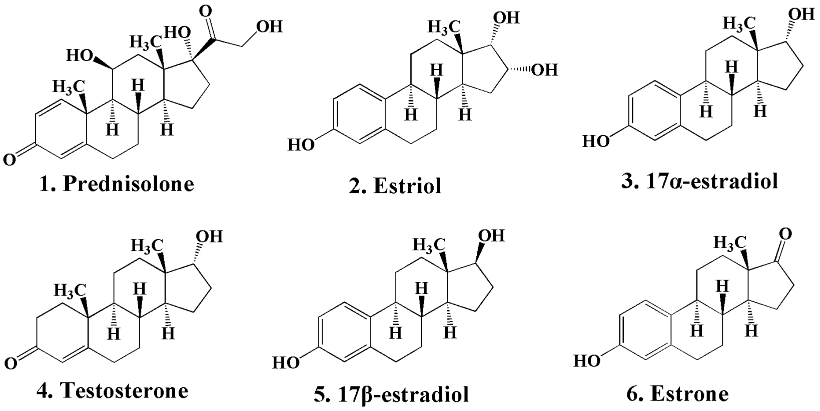


Fig. S3 Separation of SRM 869b with Sil-PImC$_{18}$-MO. Eluent: methanol; Wavelength: 254 nm.

Flow rate: 1 ml min^{-1}. Column temperature: 10 °C.



1. Prednisolone
2. Estriol
3. 17α-estradiol
4. Testosterone
5. 17β-estradiol
6. Estrone

Fig. S4 Structures of 6 steriods separated as shown in Fig. 4.

Table S2 Alkyl chain densities of different phases.

Columns	Surface area of silica (m^2 g^{-1})	% C	Alkyl chain densities (μmol m^{-2})
C18 (mon) [a]	450	13.8	1.72
C18 (poly) [a]	300	17.5	3.40
Sil-ODA$_{25}$ [a]	300	15.7	2.61
Sil-DSG [a]	339	11.7	2.13
C30 (3 μm) [b]	-	-	-
C30 (5 μm) [c]	297	17.8	1.66
Sil-PImC$_{18}$-Br-1	99	12.2	5.04
Sil-PImC$_{18}$-MO	99	13.5	3.57

[a] Taken from ref. 10a, [b] Not obtained, [c] Obtained from Nomura Chemical Co., Ltd.