Electronic Supplementary Information

Preparation of novel chiral stationary phases based on a chiral trianglsalen macrocycle by thiol-ene click chemistry for enantioseparation in high-performance liquid chromatography

Jia-Lei Wu,[a] Hua-Can Zhang,[a] Li-Qin Yu,[a] Sheng-Ming Xie,[a] Jun-Hui Zhang,[a] Bang-Jin Wang,*[a] and Li-Ming Yuan*[a]*

[a]Yunnan Key Laboratory of Modern Separation Analysis and Substance Transformation, College of Chemistry and Chemical Engineering, Yunnan Normal University, Kunming 650500, P.R. China.

*Corresponding authors: Jun-Hui Zhang, Bang-Jin Wang, and Li-Ming Yuan

Table of Contents

Fig. S5. Chromatograms for comparison of the separation of racemates on CSP-A-packed column A, CSP-B-packed column B, Chiralpak AD-H, and Chiralcel OD-H columns.

Fig. S6. Separation chromatograms of (A) 1-phenylethanol on column A and (B) ethyl mandelate on column B tested again after hundreds of injections.

Fig. S7 Resolutions of (A) 1-phenylethanol on different batches of prepared column A, and (B) ethyl mandelate on different batches of prepared column B (column temperature: 25 ℃; mobile phase: n-HEX/IPA = 90/10 (v/v); flow rate: 0.1 mL/min).

Tables

Table S1. Data of elemental analysis of SiO_2-SH, CSPs, and the surface-bound amount of CSPs.

Table S2. Values of ΔH, ΔS, ΔG, and correlation coefficient (R^2) for the separations of racemic 1-phenylethanol on column A and ethyl mandelate on column B.

References

Additional Experimental Details

1. The source details of each racemate

Racemates including methyl mandelate, 4-chlorobenzhydrol, 4-methylbenzhydrol, 1,1'-bi-2-naphthol, 3-hydroxy-2-butanone, ethyl mandelate, 1-phenylethanol, 1-indanol, ketoprofen, benzoin, mandelic acid, 2-phenylcyclohexanone, 1-(4-fluorophenyl)ethanol, 1-phenyl-1-propanol, 2-phenyl-1-propanol, 1-(4-fluorophenyl)ethanamine, styrene oxide, amlodipine, 3-(benzyloxy)propane-1,2-diol, mexiletine hydrochloride, and hesperidin were purchased from Adamas-beta (Shanghai, China). 4,4'-Dimethylbenzoin and 2,2,2-trifluoro-1-(9-anthryl)ethanol were purchased from TCI (Kyoto, Japan). 2-Methoxy-2-phenylethanol was purchased from Aladdin (Shanghai, China). 4,4'-Difluorobenzoin and 1-phenyl-1-pentanol were purchased from Macklin (Shanghai, China).

2. Preparation of CTSM-based CSPs via thiol-ene click reaction

2.1. Preparation of thiol-functionalized silica

According to the literature method,[1] thiol-functionalized silica (SiO_2-SH) was synthesized. At a nitrogen (N_2) atmosphere, the spherical silica gel (5.0 g) was weighed and dispersed into 10% HCl solution (50 mL). The system was refluxed overnight at 100 ℃ for 12 h. After the system was cooled to room temperature, it was filtered to collect solids. The solids were washed with deionized water until the washing solution was neutral, and the resulting solids were vacuum dried at 180 ℃ for 10 h to obtain activated silica (SiO_2).

At a N_2 atmosphere, activated silica (4.0 g), anhydrous toluene (50 mL), (3-mercaptopropyl)trimethoxysilane (3.0 mL), and anhydrous pyridine (2.0 mL) were sequentially added to a 100 mL round-bottom flask. The reaction was slowly warmed to 100 ℃ and refluxed for 2 days. After cooled to room temperature, the mixture was filtered, then the collected solids were sequentially washed twice with toluene, methanol, and acetone, and finally the resulting solids were dried at 100 ℃ for 12 h to obtain SiO_2-SH.

2.2. Preparation of the CTSM-based CSP-A

The synthetic route of CSP-A was shown in Fig. 2A. Under a N_2 atmosphere, the

synthesized CTSM (0.50 g, 0.52 mmol) was dissolved in anhydrous chloroform (60 mL), followed by the addition of K_2CO_3 (0.40 g, 2.89 mmol). The mixture was heated to 60 °C, and 1,4-dibromobutane (0.54 g) was added to the system. The reaction was maintained at 60 °C under reflux for 48 h. After cooling to room temperature, the reaction mixture was filtered to remove the solution. The resulting solid was collected and dried under vacuum at 80 °C for 6 h to obtain compound 1.

Under a N_2 atmosphere, compound 1 (0.50 g) was dissolved in anhydrous chloroform (60 mL), and the solution was heated to 60 °C. 1-Allylimidazole (0.26 g) was then added to the reaction system. The mixture was stirred at 60 °C for 48 h. Upon completion, the solvent was removed under reduced pressure. The resulting solid was collected and dried under vacuum for 8 h to yield compound 2.

Under a N_2 atmosphere, a mixture of compound 2 (0.50 g), SiO_2-SH (1.50 g), and AIBN (70 mg) was refluxed in MeOH (60 mL) at 60 °C for 72 h. After completion, the solvent was removed. The solid was sequentially washed with methanol and acetone, followed by drying overnight at 80 °C to yield the CTSM-based CSP-A.

2.3. Preparation of the CTSM-based CSP-B

The synthetic route of CSP-B was shown in Fig. 2B. Under a N_2 atmosphere, NaH (0.36 g, 15 mmol) was added to anhydrous chloroform (50 mL) and cooled to 0 ℃. Subsequently, a solution of CTSM (0.5 g, 0.52 mmol) in chloroform (10 mL) was added to the mixture. The suspension was maintained at 0 ℃ with stirring for 30 min, followed by dropwise addition of 5-bromo-1-pentene (0.8 mL). The reaction system was gradually heated to 60 ℃ and refluxed for 72 h. The reaction was quenched with water (10 mL), and the mixture was filtered. The organic phase was separated, while the aqueous phase was back-extracted with chloroform (3 × 10 mL). All organic fractions were pooled, dried over anhydrous $MgSO_4$, and rotary evaporation of solvent to obtain compound 3.

Under a N_2 atmosphere, a mixture of compound 3 (0.50 g), SiO_2-SH (1.50 g), and AIBN (70 mg) was refluxed in methanol (60 mL) at 60 °C for 72 h. After completion, the solvent was removed. The solid was sequentially washed with methanol and acetone, followed by drying overnight at 80 °C to yield the CTSM-based CSP-B.

3. Calculation of the surface-bound amount of these CSPs

The surface-bound amount of CSPs was calculated according to the following equation 1.[2,3]

$$\frac{\mu mol}{m^2} = \frac{C\% \times 10^6}{S \times 12.001 \times N_C \times \left(100 - \frac{C\%}{N_C \times 12.001} \times M_r\right)} \quad (1)$$

C% represents the increment of carbon content compared to the SiO_2-SH, *S* represents the surface area of silica gel (300 m^2 g^{-1}), N_C represents the carbon atom numbers of CTSM after modification with either 1,4-dibromobutane in combination with 1-allylimidazole or 5-bromo-1-pentene, M_r represents the molecular weight of CTSM after modification with either 1,4-dibromobutane in combination with 1-allylimidazole or 5-bromo-1-pentene.

4. Calculation of the thermodynamic parameters

The enthalpy change (ΔH), entropy change (ΔS), and Gibbs free energy change (ΔG) were calculated according to the following equations (Eqs 2 and 3).[4]

$$\ln k' = -\frac{\Delta H}{RT} + \frac{\Delta S}{R} + \ln \Phi \quad (2)$$

$$\Delta G = \Delta H - T\Delta S \quad (3)$$

where k' is the retention factor, *R* is the gas constant, *T* is the absolute temperature, and Φ is the phase ratio. k' was calculated according to the following Eq. 4.

$$k' = \frac{t_R - t_0}{t_0} \quad (4)$$

where t_R is the retention time of the enantiomers and t_0 is the dead time.

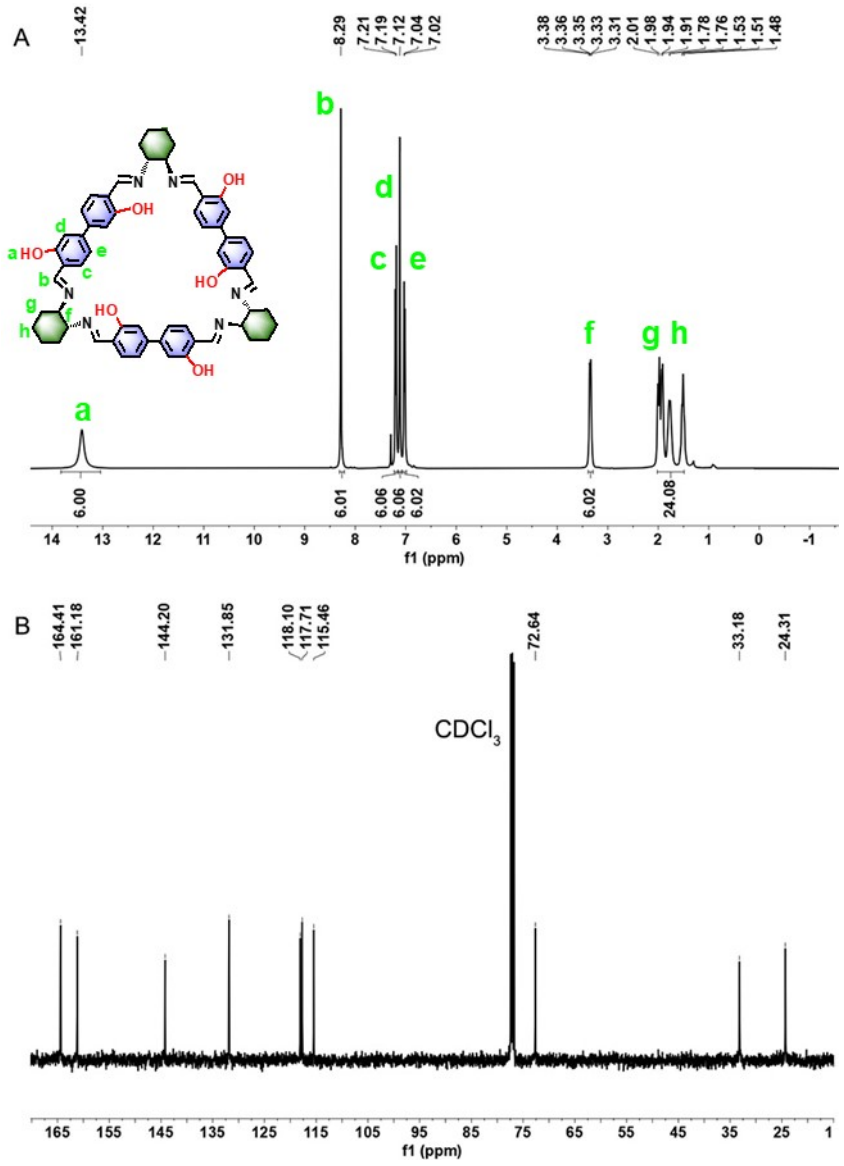


Fig. S1. NMR spectra of the synthesized CTSM. (A) ^{1}H NMR spectrum; (B) ^{13}C NMR spectrum.

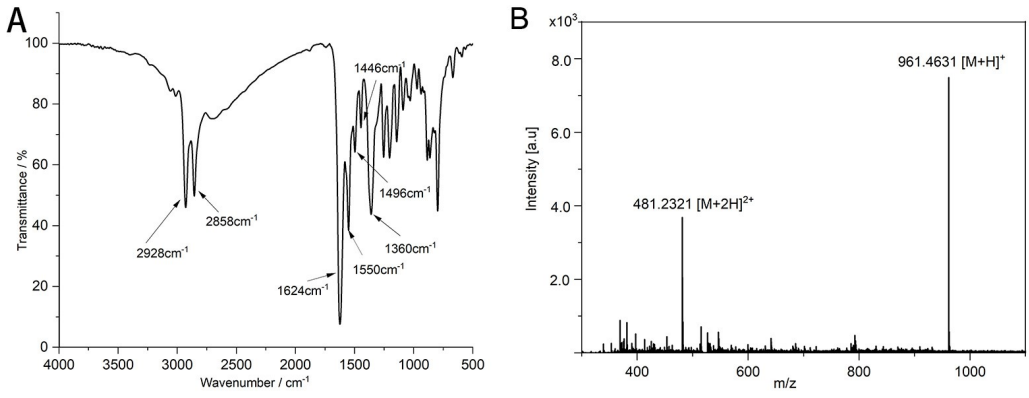


Fig. S2 (A) FT-IR spectrum of the synthesized CTSM; (B) ESI-MS spectrum of the synthesized CTSM.

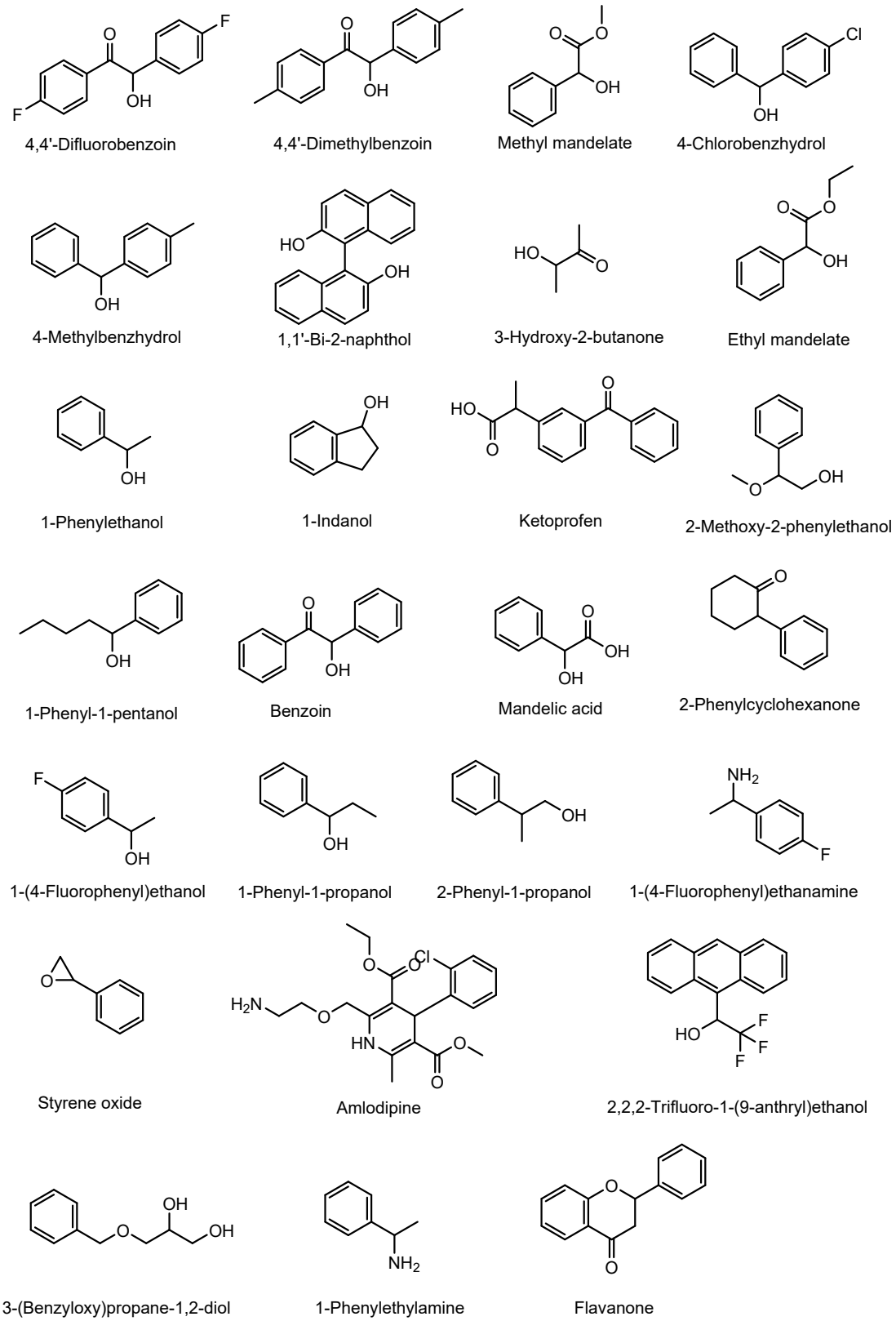


Fig. S3 Molecular structures of the racemates.

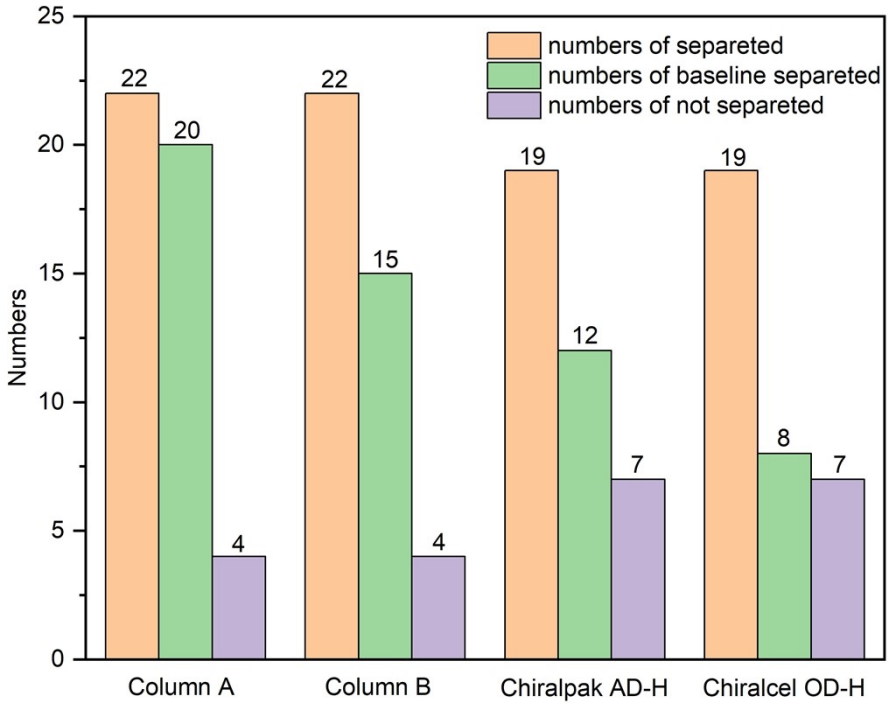
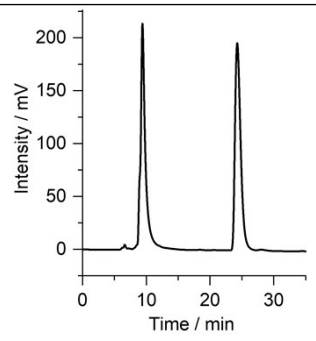
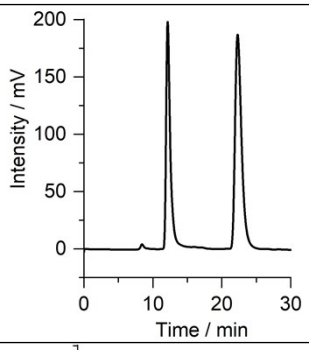
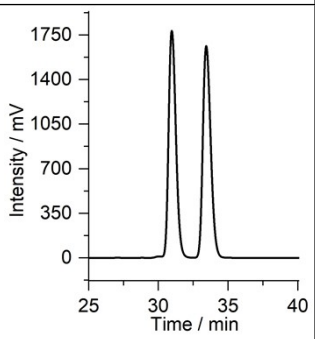
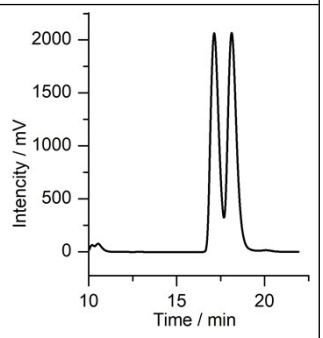
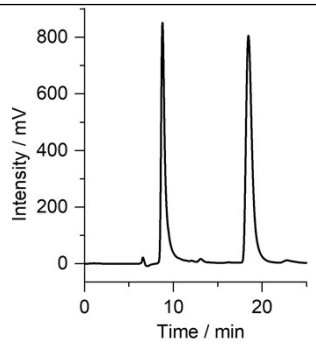
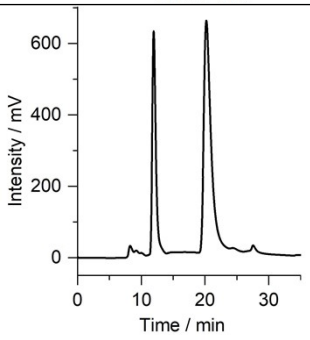
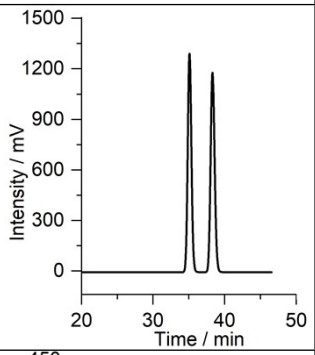
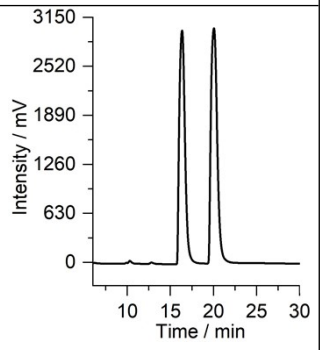
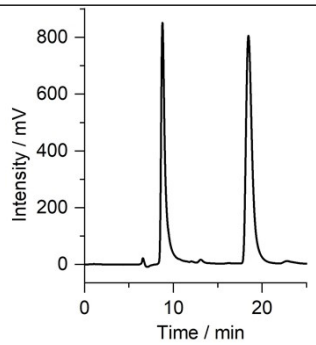
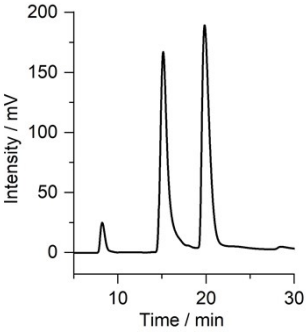
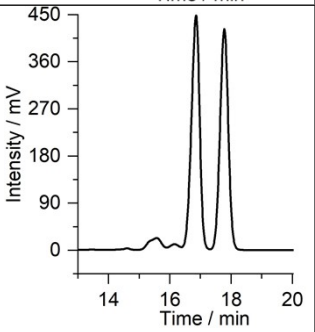
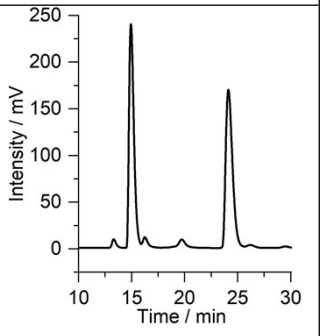
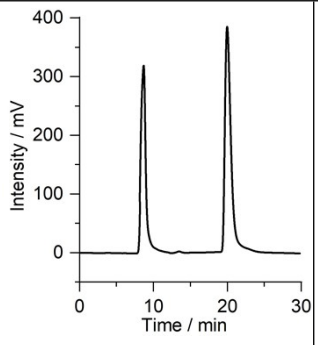
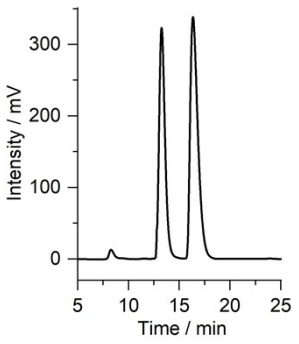
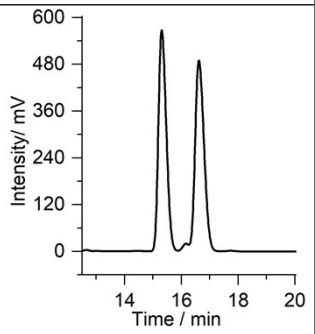
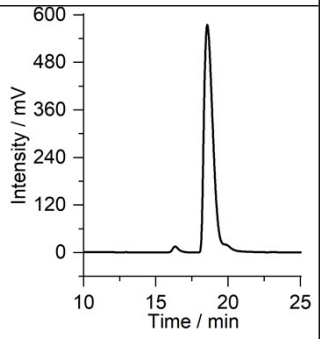
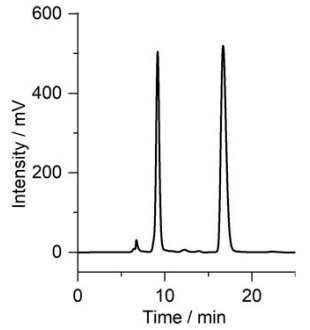
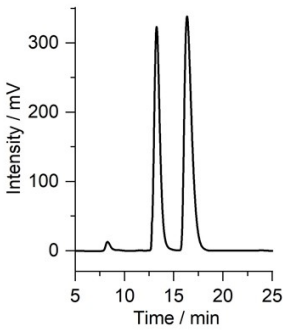
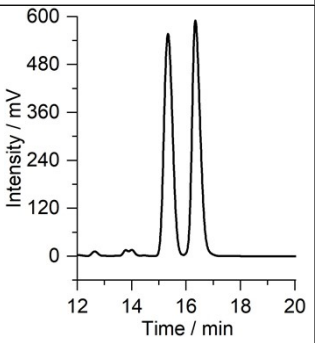
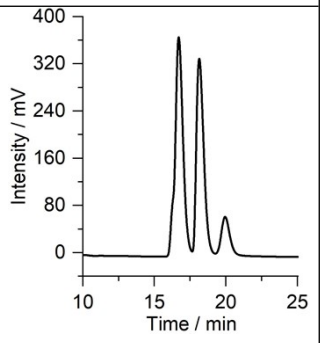
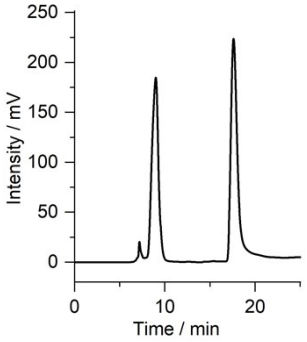
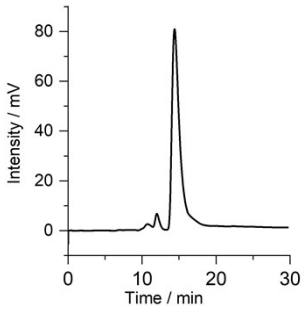
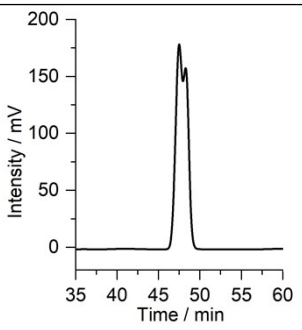
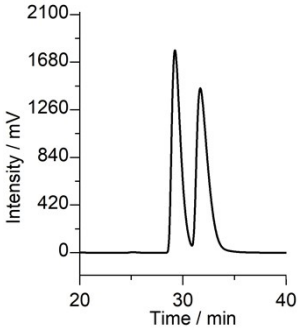
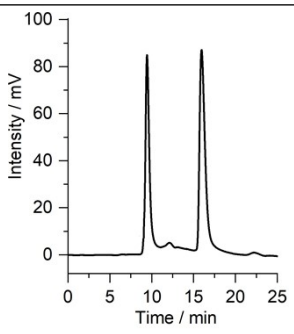
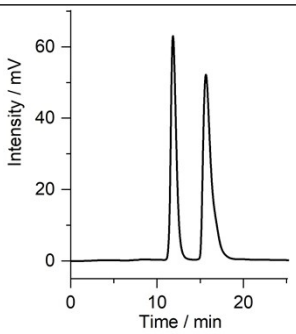
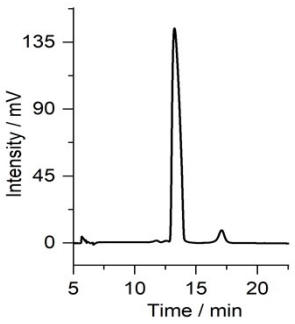
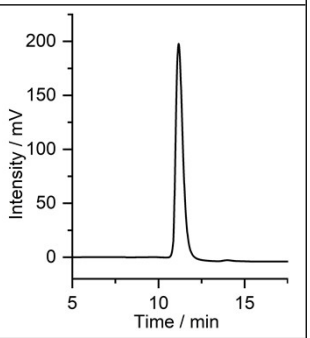
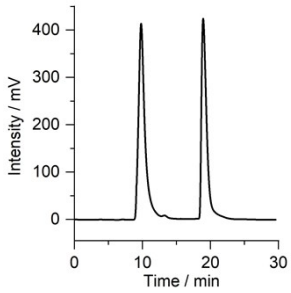
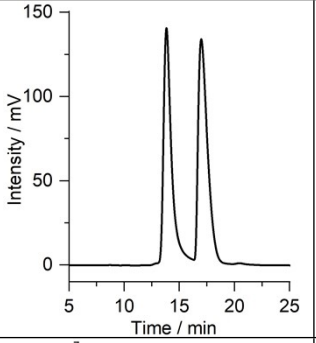
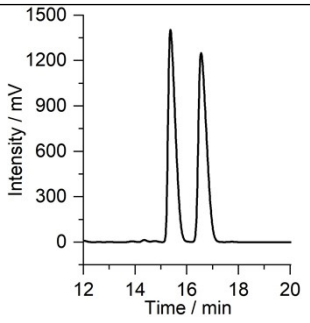
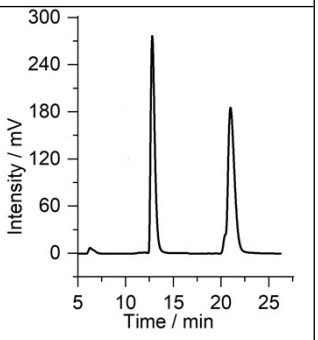
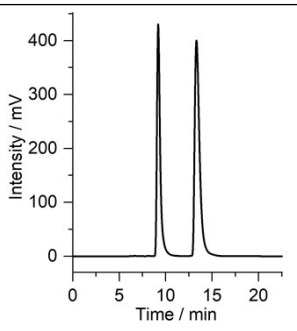
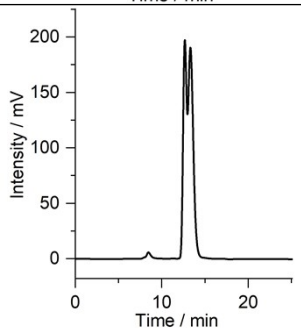
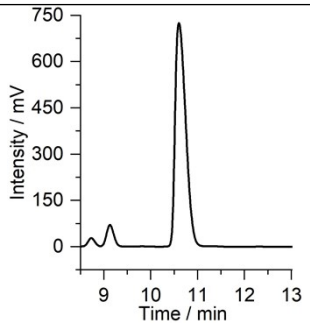
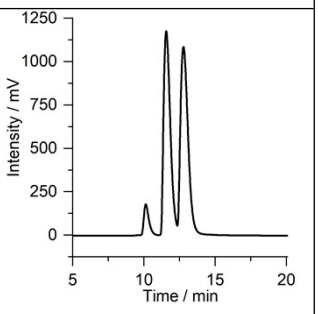
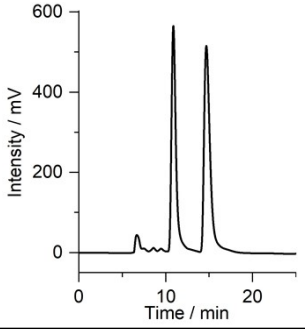
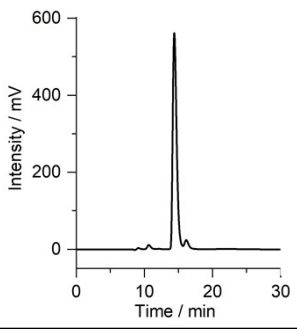
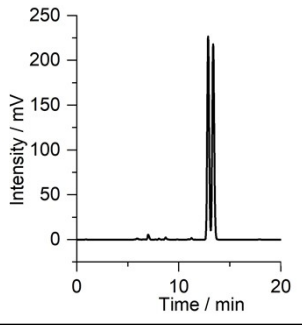
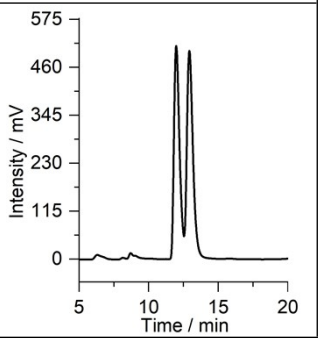
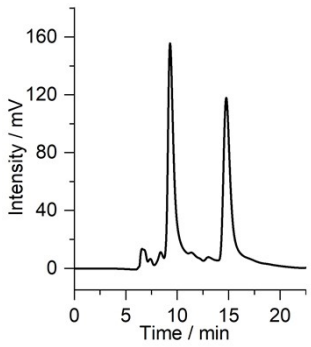
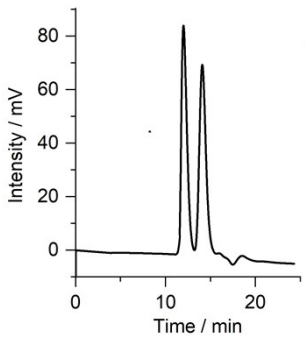
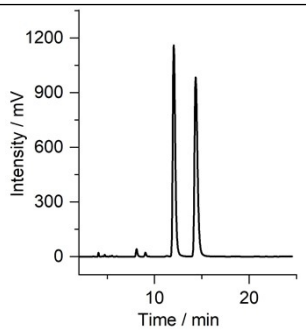
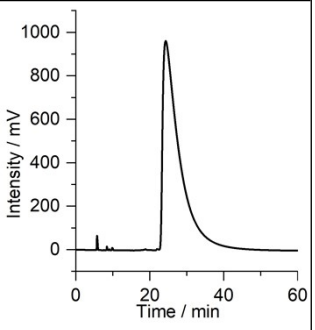
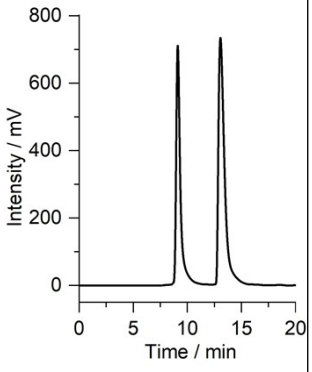
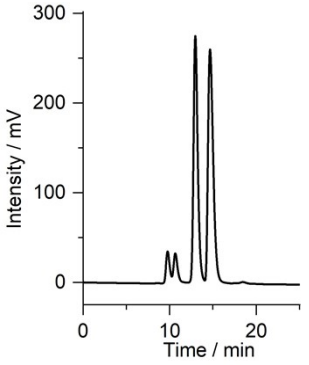
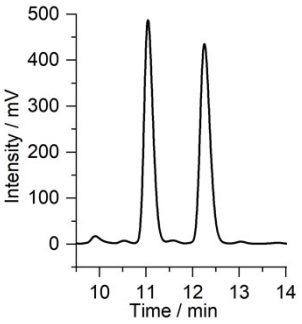
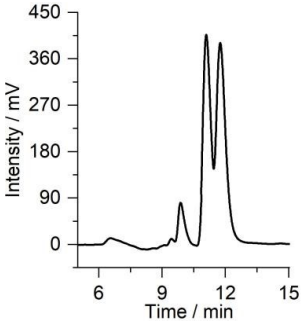
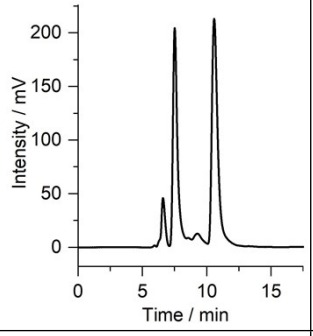
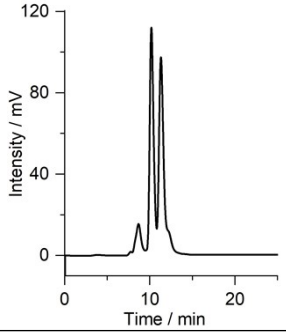
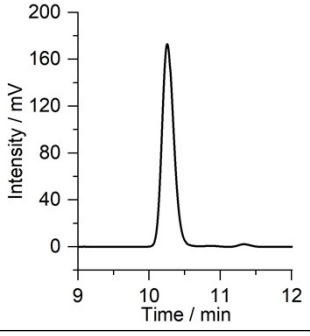
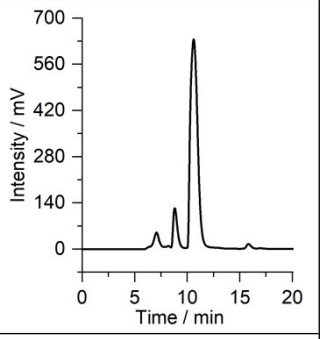
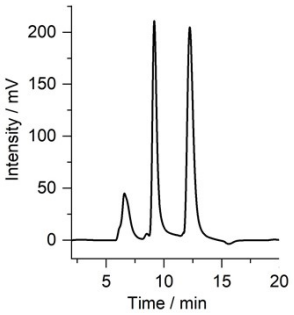
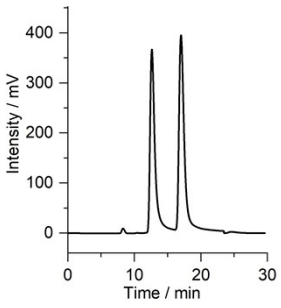
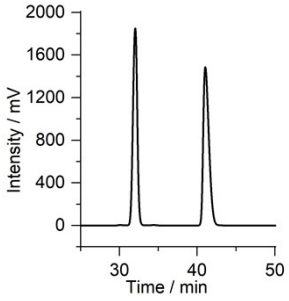
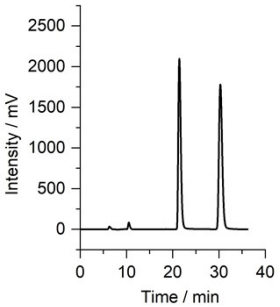
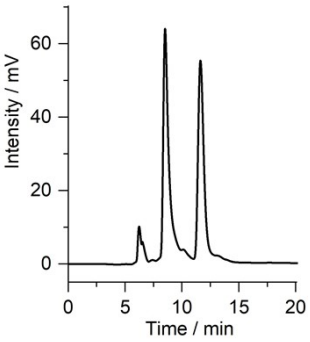
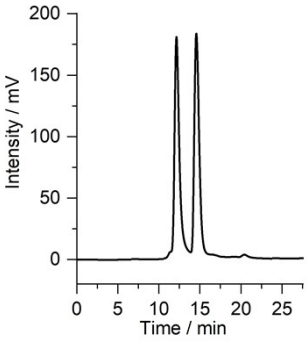
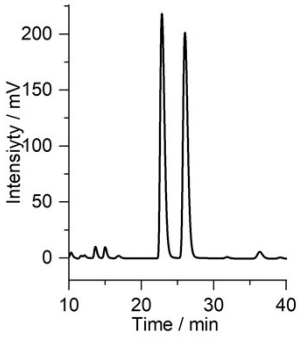
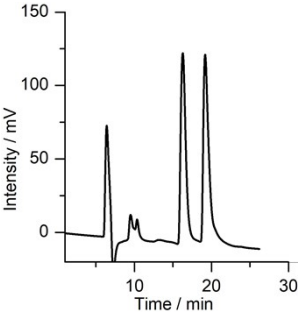
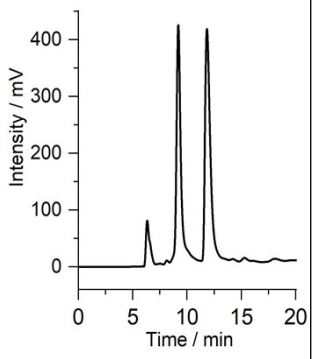
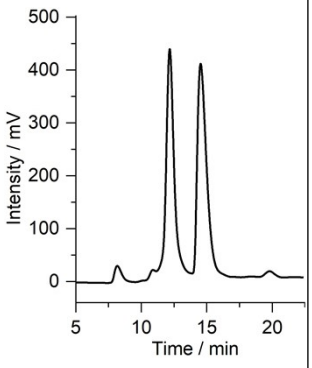
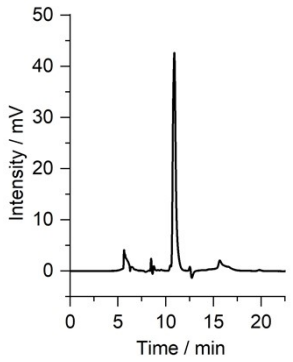
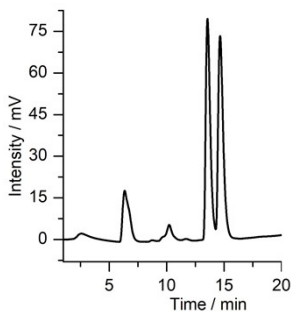
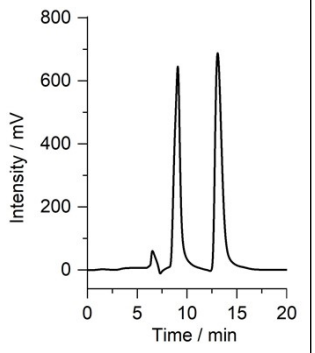
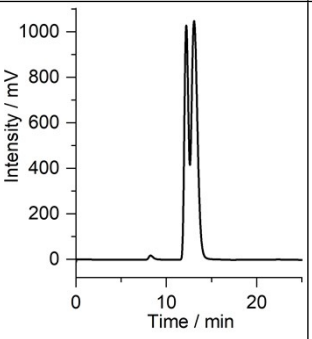
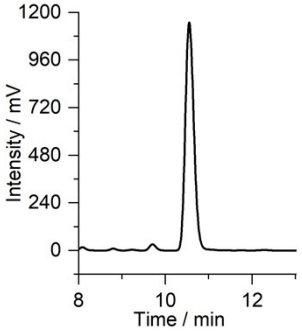
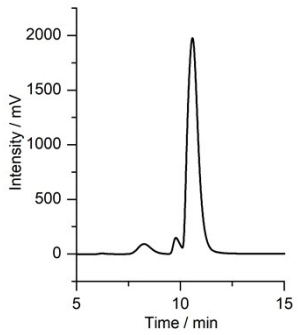
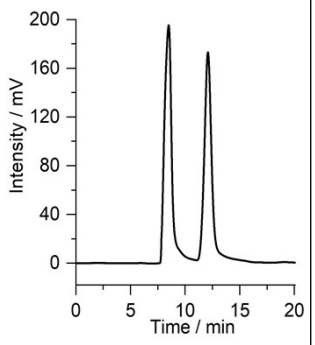
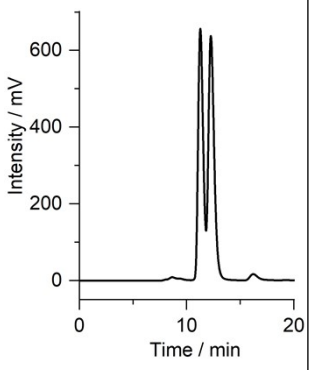
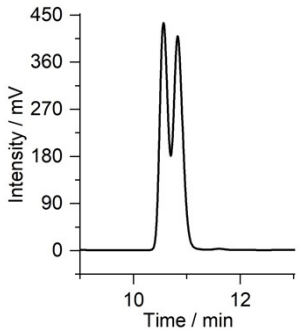
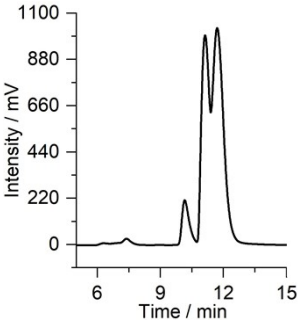
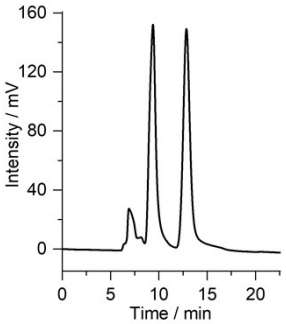
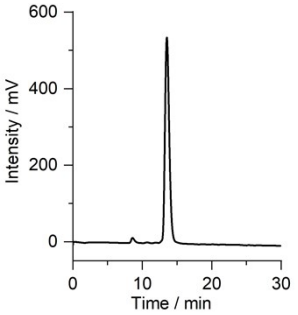
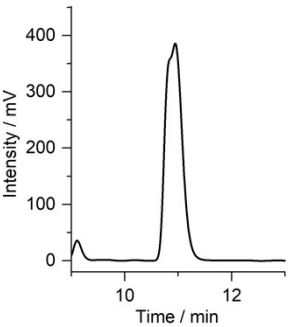
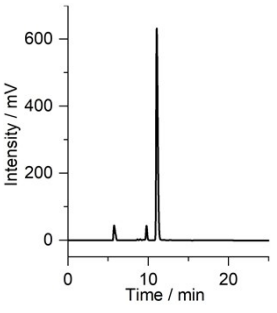
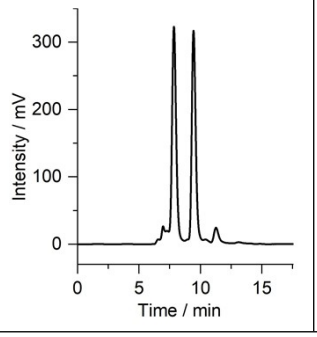
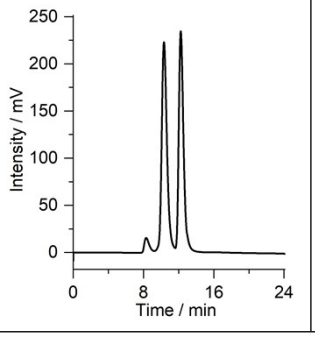
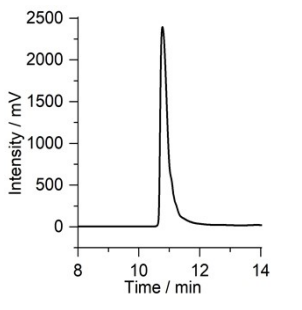
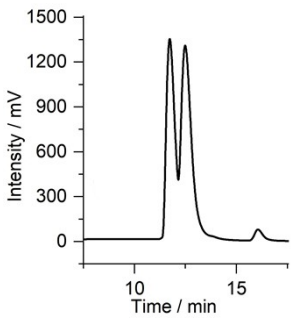


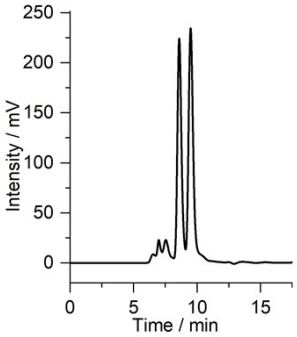
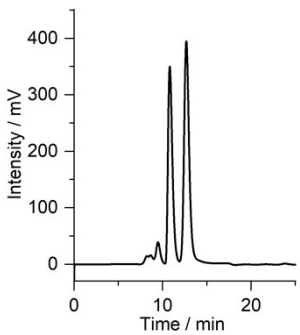
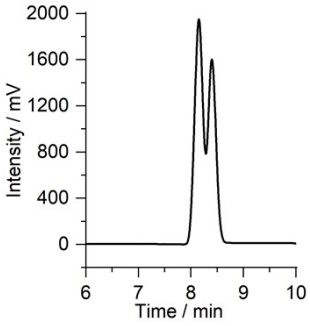
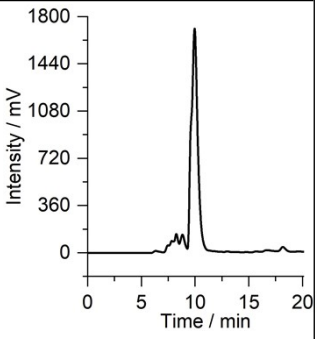
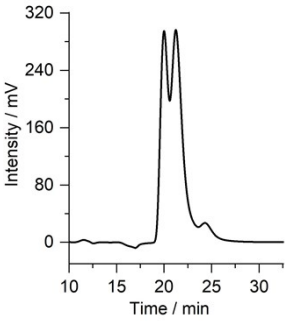
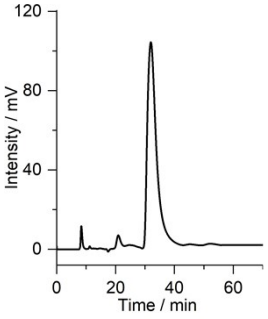
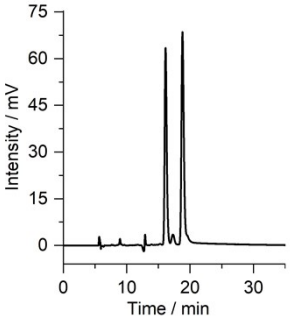
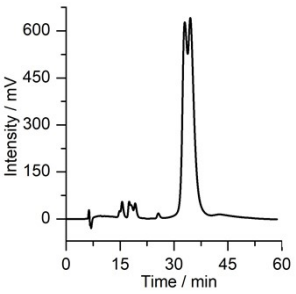
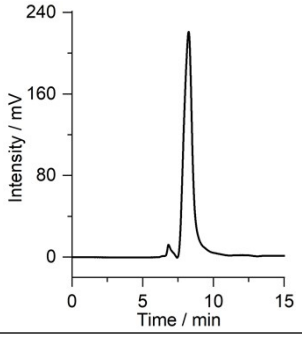
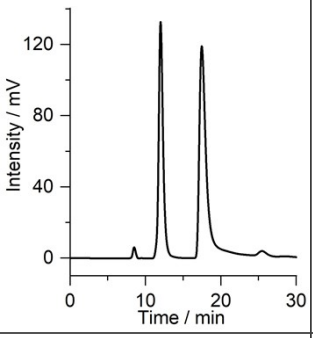
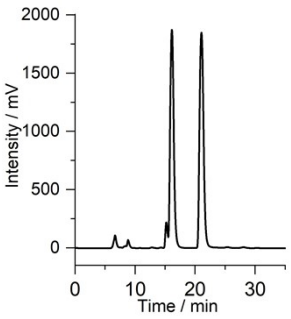
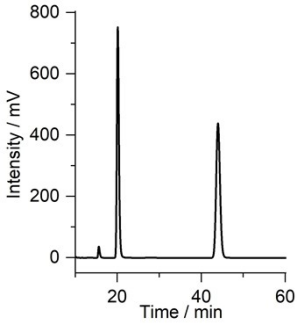
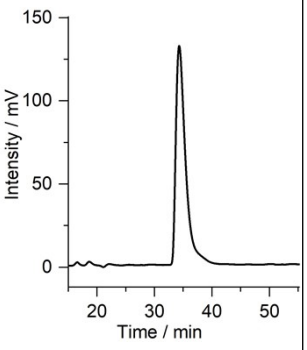
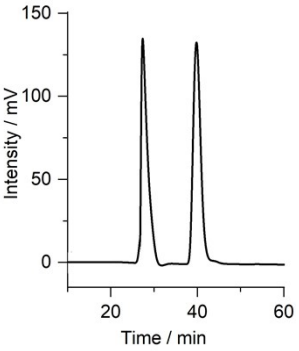
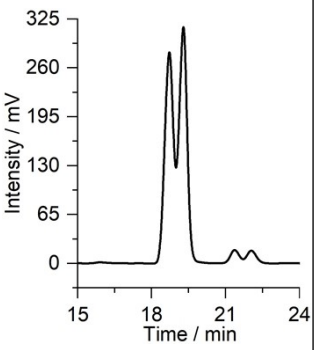
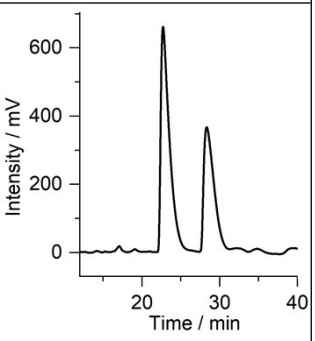
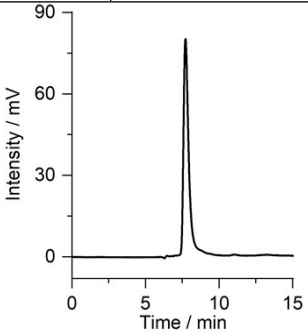
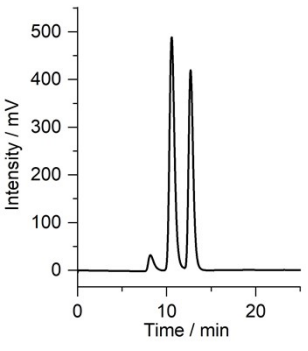
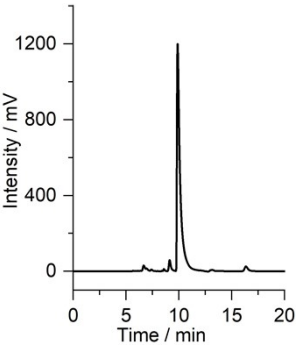
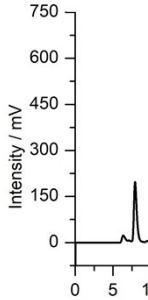
Fig. S4 Statistical data of the tested 26 racemates on column A, column B, Chiralpak AD-H, and Chiralcel OD-H.

Racemates	Column A	Column B	Chiralpak AD-H	Chiralcel OD-H
4,4'-Difluorobenzoin				
4,4'-Dimethylbenzoin				
Methyl mandelate				
4-Chlorobenzhydrol				
4-Methylbenzhydrol				

Racemates	Column A	Column B	Chiralpak AD-H	Chiralcel OD-H
1,1'-Bi-2-naphthol				
3-Hydroxy-2-butanone				
Ethyl mandelate				
1-Phenylethanol				
1-Indanol				

Racemates	Column A	Column B	Chiralpak AD-H	Chiralcel OD-H
Ketoprofen				
2-Methoxy-2-phenylethanol				
1-Phenyl-1-pentanol				
Benzoin				
Mandelic acid				

Racemates	Column A	Column B	Chiralpak AD-H	Chiralcel OD-H
2-Phenylcyclohexanone				
1-(4-Fluorophenyl) ethanol				
1-Phenyl-1-propanol				
2-Phenyl-1-propanol				
1-(4-Fluorophenyl) ethanamine				

Racemates	Column A	Column B	Chiralpak AD-H	Chiralcel OD-H
Styrene oxide				
Amlodipine				
2,2,2-Trifluoro-1-(9-anthryl)ethanol				
3-(Benzyloxy)propane-1,2-diol				
1-Phenylethylamine				

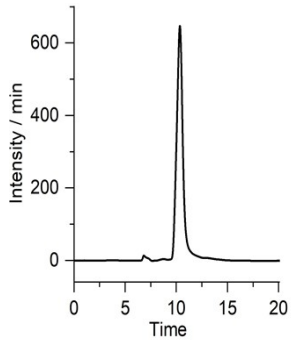
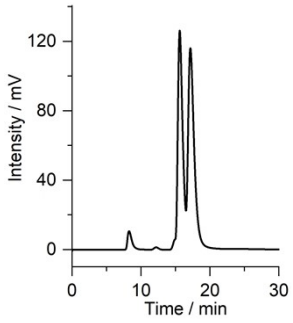
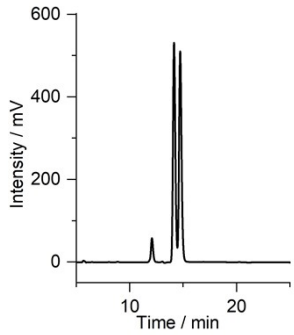
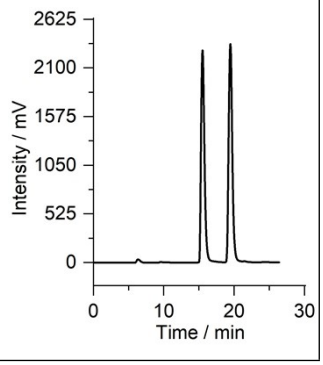
Racemates	Column A	Column B	Chiralpak AD-H	Chiralcel OD-H
Flavanone				

Fig. S5 Chromatograms for comparison of the separation of racemates on CSP-A-packed column A, CSP-B-packed column B, Chiralpak AD-H, and Chiralcel OD-H columns.

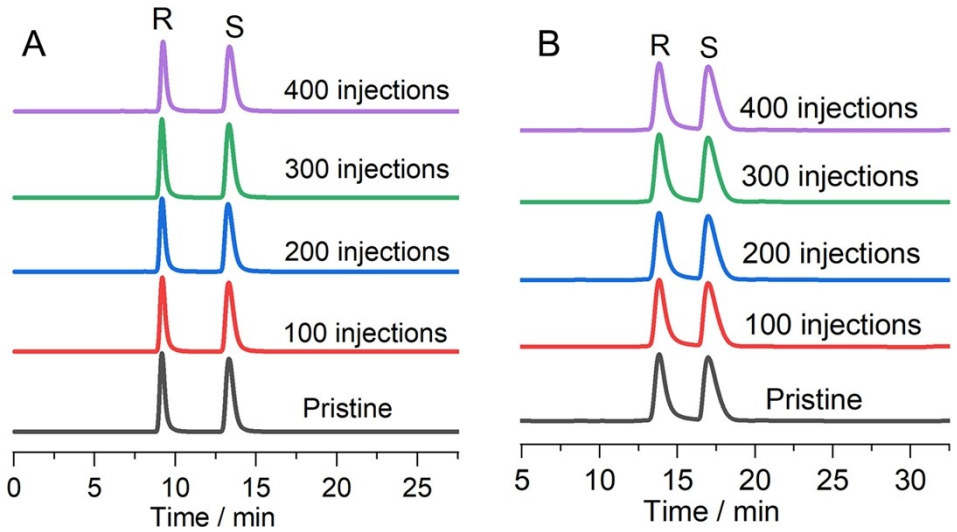


Fig. S6 Separation chromatograms of (A) 1-phenylethanol on column A and (B) ethyl mandelate on column B tested again after hundreds of injections (column temperature: 25 °C; mobile phase: *n*-HEX/IPA = 90/10 (v/v); flow rate: 0.1 mL/min).

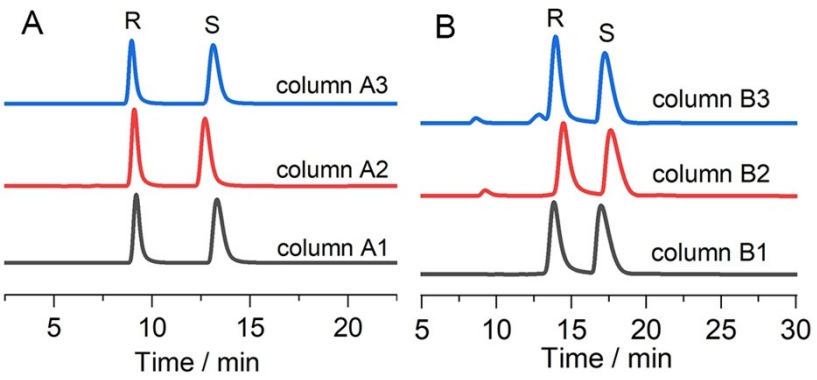


Fig. S7 Resolutions of (A) 1-phenylethanol on different batches of prepared column A, and (B) ethyl mandelate on different batches of prepared column B (column temperature: 25 ℃; mobile phase: n-HEX/IPA = 90/10 (v/v); flow rate: 0.1 mL/min).

Table S1. Data of elemental analysis of SiO_2-SH, CSPs, and the surface-bound amount of CSPs.

Samples	Elemental content			CSP bonding amounts
	C %	N %	H %	(μmol/m^2)
SiO_2-SH	4.30	< 0.05	1.09	-
CSP-A	8.56	1.46	1.75	0.11
CSP-B	8.48	0.31	1.66	0.14

Table S2. Values of ΔH, ΔS, ΔG, and correlation coefficient (R^2) for the separations of racemic 1-phenylethanol on column A and ethyl mandelate on column B.

Analytes	ΔH (kJ·mol^{-1})	ΔS (J·mol^{-1}·K^{-1})	ΔG (kJ·mol^{-1})	R^2
R-1-phenylethanol	-9.84±0.27	-31.46±0.90	-0.45±0.27	0.998
S-1-phenylethanol	-5.86±0.18	-11.37±0.59	-2.47±0.18	0.998
R-ethyl mandelate	-6.97±0.65	-16.57±2.15	-2.03±0.65	0.997
S-ethyl mandelate	-8.72±0.75	-21.33±2.18	-2.36±0.75	0.996

References

1 X. B. Yao, T. T. Y. Tan and Y. Wang, *J. Chromatogr. A*, 2014, **1326**, 80-88.

2 B. A. Siles, H. Brian Halsall and J. G. Dorsey, *J. Chromatogr. A*, 1995, **704**, 289-305.

3 X. B. Yao, H. Zheng, Y. Zhang, X. F. Ma, Y. Xiao and Y. Wang, *Anal. Chem.*, 2016, **88**, 4955-4964.

4 C. X. Yang and X. P. Yan, *Anal. Chem.*, 2011, **83**, 7144-7150.