Supplementary Information for

Fluorescent Recognition of L- and D-Tryptophan in Water by Micelle Probes

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Supporting Information

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1.General experimental methods

All amino acids and other chemicals were purchased from Sigma Aldrich Chemical Co. or Alfa Aesar, and used without further purification. NMR spectra were recorded on Varian-600 MHz spectrometer or Bruker-800 MHz spectrometer. Chemical shifts for ^{1}H NMR were reported in parts per million relative to a singlet at 7.26 ppm for deuterated chloroform and 2.50 ppm for deuterated DMSO. Chemical shifts for ^{13}C NMR were reported in parts per million relative to a triplet at 77.16 ppm for deuterated chloroform. Steady-state fluorescence emission spectra were recorded on Horiba FluoroMax-4 spectrofluorometer. Low-temperature fluorescence emission spectra were recorded on Horiba FluoroMax-3 spectrofluorometer. Nanoparticle sizes and polydispersities were analyzed via dynamic light scattering (DLS, Wyatt, DynaPro). Cryo-TEM images were recorded on a Tecnai F20 TEM system. For cryoTEM, nanoparticles were concentrated by centrifugation at 8000 rpm for 6 min in a cellulose filter tube (Amicon, Ultra-15, 30000 Da MW cutoff). High-resolution mass spectra were obtained from the University of Illinois at Urbana-Champaign (UIUC) Mass Spectrometry Facility. Deionized water was used for all the experiments.

Nano precipitation to prepare the micelle encapsulated probe. 1.6 µmol of the (*S*)-organic probes (1.0 mg/mL in DMF) were added to 80.0 mg block copolymer mPEG-PLLA [mPEG-PDLA for the (*R*)-probes]. More DMF was added to dissolve the block copolymer and the volume was calibrated to 8 mL. The solution was sonicated for 10 min and added dropwise to a vortex of 72 mL water at a rate of 1 mL/min. The vortex was allowed to last 30 more min after the addition of DMF solution. The final mixture was dialyzed in DI water to form micelle solutions containing 20 µM corresponding organic probes.

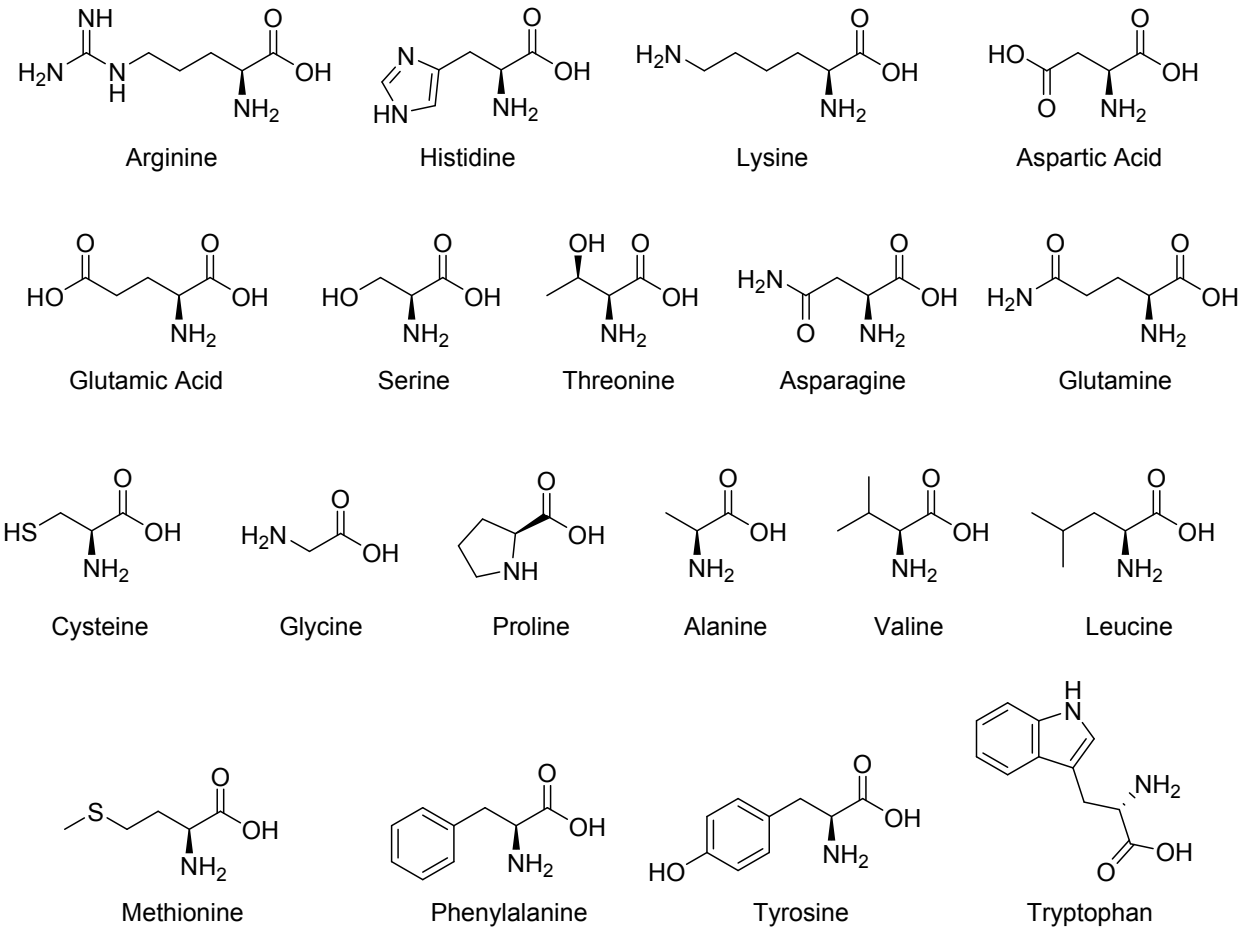
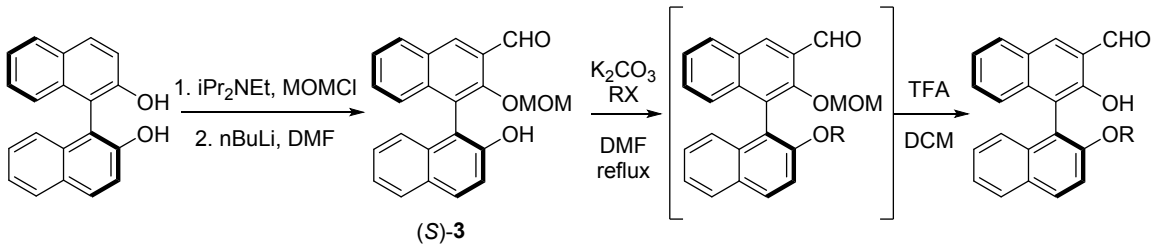


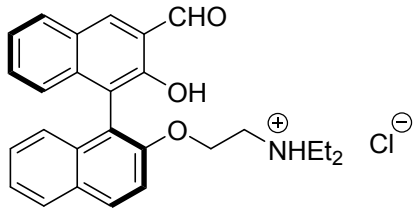
Figure S1. Structure of the Chiral Substrates

2. Synthesis and characterization data



General Procedures:

After pumping off air and refilling with nitrogen gas three times, the mixture of 200 mg of (*S*)-**3**, 4 equiv of potassium carbonate, and 2.5 equiv of alkyl halide (RX) was heated to 100 °C for 8 h in dimethylformamide. The reaction mixture was neutralized with 2M HCl, extracted with ethyl acetate, and dried with sodium sulfate. The concentrated mixture was dissolved in dichloromethane to which was added 1 mL trifluoroacetic acid at 0 °C. Then, the mixture was allowed to warm up to room temperature. After 40 min, the reaction mixture was neutralized with sodium bicarbonate, extracted with dichloromethane, and dried with sodium sulfate. The concentrated mixture was purified by column chromatography to afford the products.

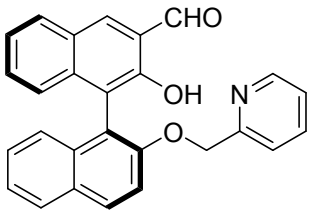


(*S*)-**2a** HCl

(*S*)-2'-(2-(diethylamino)ethoxy)-2-hydroxy-[1,1'-binaphthalene]-3-carbaldehyde hydrochloride (*S*)-**2a**·HCl :

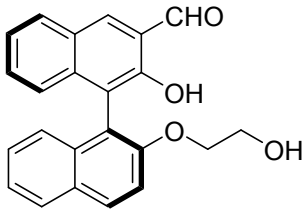
2-Bromo-N,N-diethylethylamine hydrobromide was used as RX. After the reaction mixture was treated with trifluoroacetic acid, the mixture was neutralized with sodium bicarbonate, acidified with 2M HCl, and dried with sodium sulfate. The concentrated mixture was purified by column chromatography (DCM/MeOH) to afford the products. Yield 57%, yellow oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 12.04 (s, 1H), 10.44 (s, 1H), 10.21 (s, 1H), 8.33 (s, 1H), 8.04 (d, J=9.2Hz, 1H), 8.00 (dd, J_1=3.2Hz, J_2=6.3Hz, 1H), 7.91 (d, J=8.2Hz, 1H), 7.41 (m, 4H), 7.28 (t, J=7.6Hz, 1H), 7.1 (d, J= 8.8Hz, 2H), 4.58 (m, 1H), 4.47 (m, 1H), 3.16 (m, 2H), 2.72 (m, 2H), 2.58 (m, 2H), 1.07 (t, J=7.3Hz, 3H), 0.71 (t, J=7.2Hz, 3H). ^{13}C{^{1}H} NMR (201 MHz, $CDCl_3$) δ 196.84, 153.49, 152.92, 137.92, 137.89, 133.57, 130.99, 130.87, 129.97, 129.93, 128.50, 127.65, 127.32, 125.23, 124.91, 124.87, 124.67, 122.14, 118.57, 117.86, 113.96, 77.32, 77.16, 77.00, 63.76, 49.84, 47.27, 46.60, 8.97, 8.10. Mass calculated (M+H$^+$) 414.2069, found 414.2062.

$[\alpha]_D^{20}$ = -62.233 (c= 1 mg/mL $CHCl_3$)



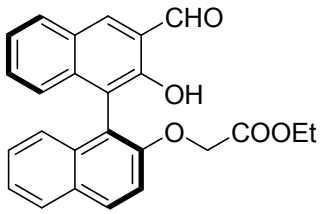
(*S*)-**2b**

(*S*)-2-hydroxy-2'-(pyridin-2-ylmethoxy)-[1,1'-binaphthalene]-3-carbaldehyde (*S*)-**2b**:
2-(Bromomethyl)pyridine hydrobromide was used as RX, product yield 76%, yellow solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ 10.48 (s, 1H), 10.22 (s, 1H), 8.47 (d, J=4.1Hz, 1H), 8.34 (s, 1H), 8.00 (m, 1H), 7.95 (d, J=9.1Hz, 1H), 7.87 (d, J=8.2Hz, 1H), 7.42 (d, J=9.0Hz, 2H), 7.38 (m, 2H), 7.35 (t, J=7.6Hz, 1H), 7.27 (t, 8.2Hz, 2H), 7.20 (two d, J_1=10Hz, J_2=9.0Hz, 2H), 7.10 (m, 1H), 6.92 (d, J=7.6Hz, 1H), 5.30 (d, J=14.0Hz, 1H), 5.24 (d, J=14.0Hz, 1H). ^{13}C{^{1}H} NMR (201 MHz, $CDCl_3$) δ 196.89, 156.67, 153.61, 153.46, 146.53, 139.05, 138.01, 133.76, 130.63, 130.54, 129.92, 129.76, 128.42, 127.70, 127.05, 125.52, 125.00, 124.56, 124.32, 123.15, 122.28, 122.06, 118.61, 118.19, 114.89, 69.95.
Mass calculated (M+H$^+$) 406.1443, found 406.1441.
$[\alpha]_D^{20}$ = -104.13 (c= 1 mg/mL $CHCl_3$)



(*S*)-**2c**

(*S*)-2-hydroxy-2'-(2-hydroxyethoxy)-[1,1'-binaphthalene]-3-carbaldehyde (*S*)-**2c**:
2-Chloroethanol was used as RX, product yield 40%, yellow solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ 10.58 (s, 1H), 10.20 (s, 1H), 8.33 (s, 1H), 8.00 (m, 2H), 7.89 (d, J=8.5Hz, 1H), 7.45 (d, J=9.0Hz, 1H), 7.40 (m, 2H), 7.37 (t, J=7.4Hz, 1H), 7.27 (t, J=7.4Hz, 1H), 7.17 (m, 2H), 4.24 (m, 1H), 4.11 (m, 1H), 3.64 (m, 1H), 3.57 (m, 1H). ^{13}C{^{1}H} NMR (201 MHz, $CDCl_3$) δ 188.87, 155.13, 152.10, 131.88, 131.55, 131.52, 131.35, 130.36, 129.79, 129.42, 129.31, 128.44, 126.65, 125.80, 125.57, 125.54, 124.93, 121.71, 121.30, 118.53, 113.00, 29.85, 0.12.
Mass calculated (M+H$^+$) 358.1283, found 359.1281.
(*S*)-**2c** $[\alpha]_D^{20}$ = -62.133 (c= 1 mg/mL $CHCl_3$)
(*R*)-**2c** $[\alpha]_D^{20}$ = 62.833 (c= 1 mg/mL $CHCl_3$)



(*S*)-**2d**

Ethyl (*S*)-2-((3'-formyl-2'-hydroxy-[1,1'-binaphthalen]-2-yl)oxy)acetate (*S*)-**2d**:

Ethyl bromoacetate was used as RX, product yield 92%, yellow solid. ^{1}H NMR (600MHz, $CDCl_3$) δ 10.38 (s, 1H), 10.21 (s, 1H), 8.32 (s, 1H), 7.98 (m, 2H), 7.89 (d, J=8.3Hz, 1H), 7.37 (m, 4H), 7.27 (m, 1H), 7.23 (m, 1H), 7,17 (d, J=8.6Hz, 1H), 4.62 (d, J=17Hz, 1H), 4.55 (d, J=17Hz, 1H), 4.11 (q, J=7.1Hz, 2H), 1.16 (t, J=7.1Hz, 3H). ^{13}C{^{1}H} NMR (150 MHz, $CDCl_3$) δ 196.78, 169.34, 154.04, 153.59, 137.96, 133.77, 130.46, 130.36, 130.08, 129.85, 128.35, 127.74, 126.99, 125.70, 125.17, 124.48, 124.45, 122.30, 118.91, 118.29, 115.48, 67.31, 61.25, 14.19.

Mass calculated ($M+H^+$) 401.1389, found 401.1395.

$[\alpha]_D^{20}$ = -91.533 (c= 1 mg/mL $CHCl_3$)

3. Fluorescence spectra

Fluorescent measurement at room temperature:

To a suspension of a micelle-encapsulated probe (1 mL, 2.0 x 10^{-5} M) and a carbonate buffer solution (1 mL, CBS, 25 mM of sodium carbonate with 25 mM of sodium bicarbonate, pH = 10.1) were added amino acids (10 µL, 20 mM in CBS) and $Zn(OAc)_2$ (10 µL, 4 mM in water). After standing at rt for 3 h, the mixtures were used for fluorescence measurement. Slit = 3/3 nm, int time = 0.1 s. Fluorescence emission spectra were recorded on Horiba FluoroMax-4 spectrofluorometer.

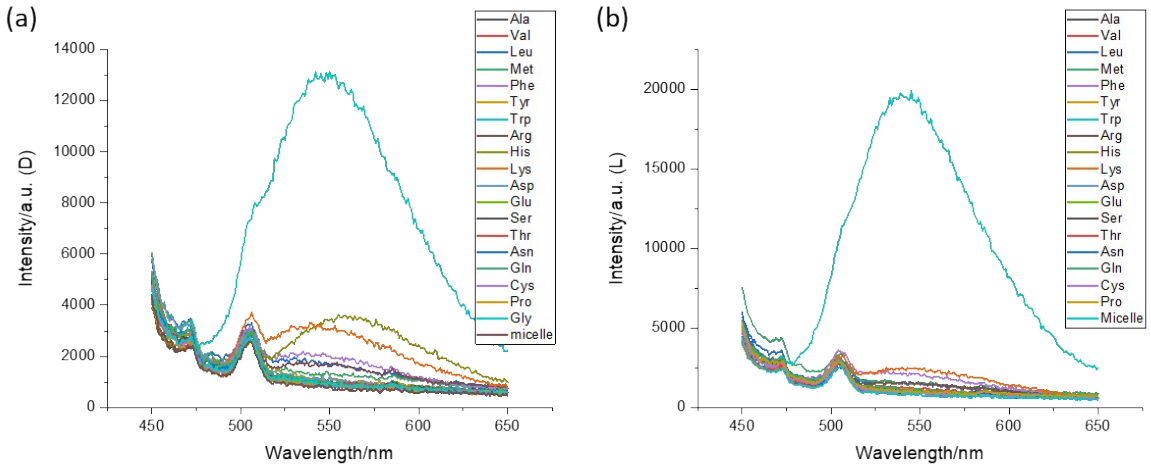


Figure S2. Fluorescence spectra of (*S*)-**2a**@PEG-PLLA with various (a) D-amino acids; (b) L-amino acids. Corresponding organic sensor concentration = 10 µM, with 2 equiv zinc acetate, 10 equiv amino acids. Excited at 430 nm, slit = 3/3 nm, integration time = 0.1 s.

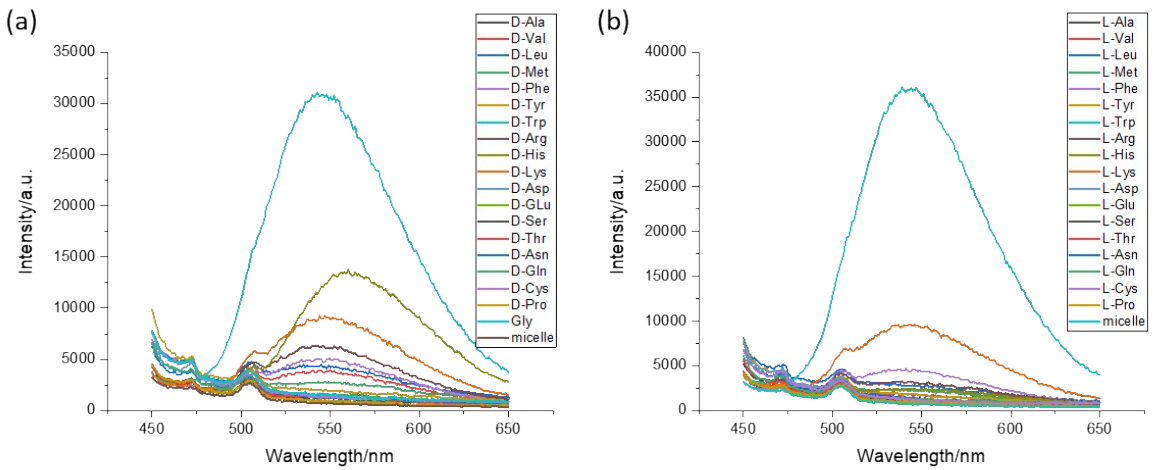


Figure S3. Fluorescence spectra of (*S*)-**2b**@PEG-PLLA with various (a) D-amino acids; (b) L-amino acids. Corresponding organic sensor concentration = 10 μM, with 2 equiv zinc acetate, 10 equiv amino acids. Excited at 430 nm, slit = 3/3 nm, integration time = 0.1 s.

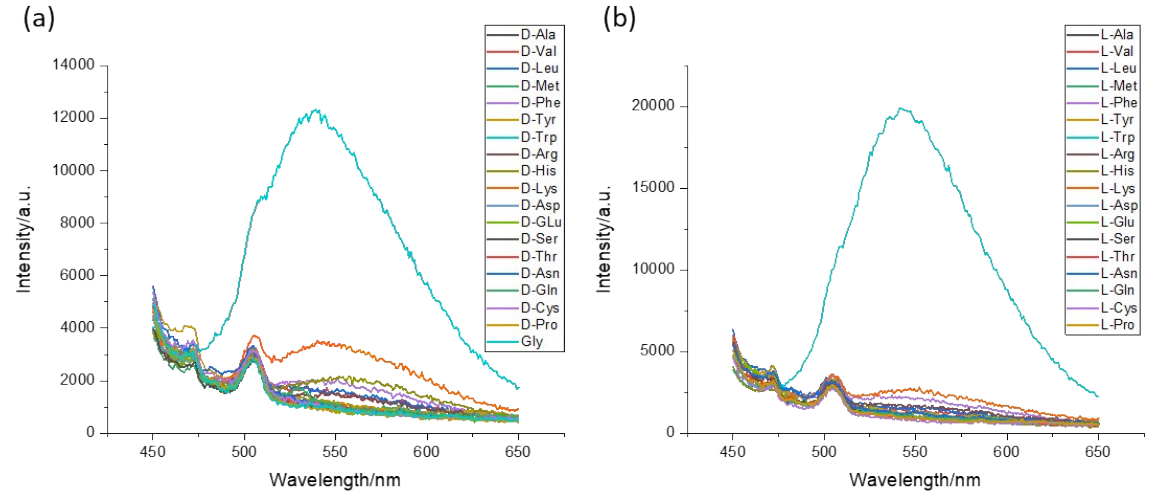


Figure S4. Fluorescence spectra of (*S*)-**2c**@PEG-PLLA with various (a) D-amino acids; (b) L-amino acids. Corresponding organic sensor concentration = 10 μM, with 2 equiv zinc acetate, 10 equiv amino acids. Excited at 430 nm, slit = 3/3 nm, integration time = 0.1 s.

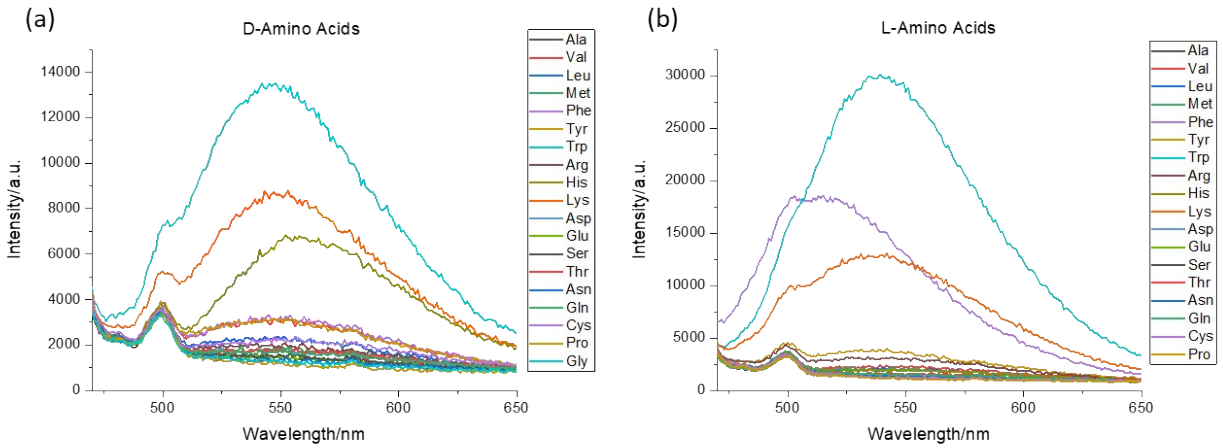


Figure S5. Fluorescence spectra of (*S*)-**2d**@PEG-PLLA with various (a) D-amino acids; (b) L-amino acids. Corresponding organic sensor concentration = 10 μM, with 2 equiv zinc acetate, 10 equiv amino acids. Excited at 430 nm, slit = 3/3 nm, integration time = 0.1 s.

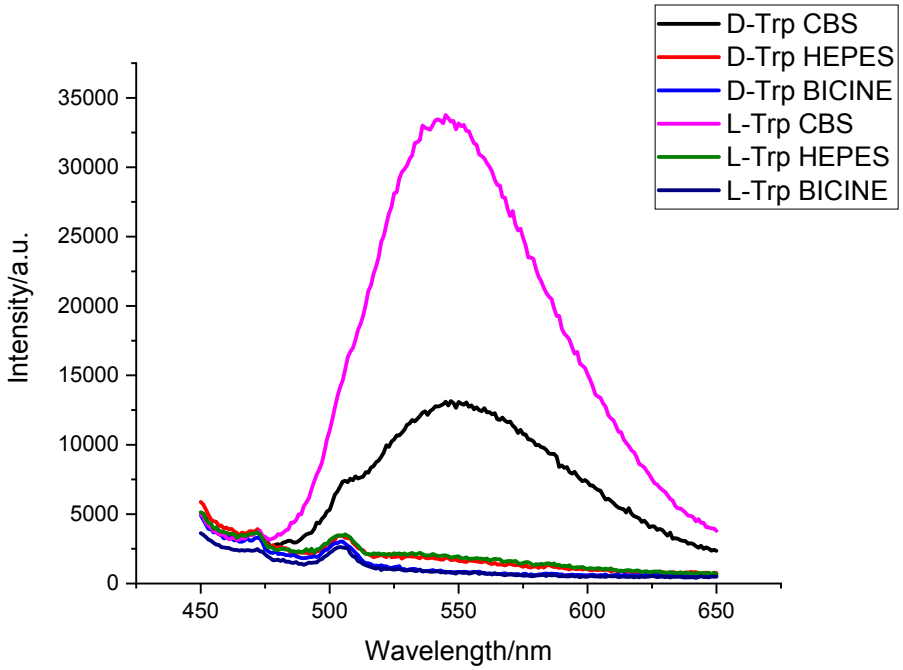


Figure S6. Fluorescence spectra of (*S*)-**2c**@PEG-PLLA in various buffers. Corresponding organic sensor concentration = 10 μM, with 2 equiv zinc acetate, 10 equiv amino acids. Excited at 430 nm, slit = 3/3 nm, integration time = 0.1 s.

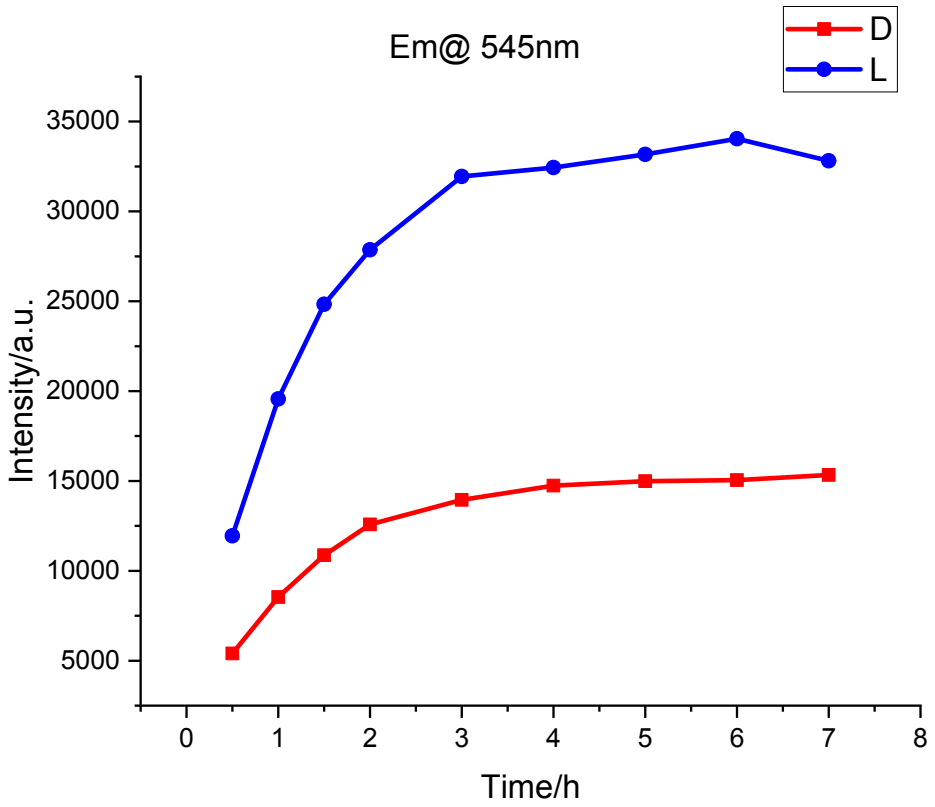


Figure S7. Fluorescence response of (*S*)-**2c**@PEG-PLLA toward D- and L-Trp at rt versus reaction time. The intensities stabilized at 3 - 5 h.

Low-temperature fluorescence study:

To a suspension of 1 mL of 20 μM Probe and 1 mL CBS buffer, were added with 10 μL of 20 mM amino acid (in water) and 10 μL of 4 mM $Zn(OAc)_2$ (in water). After standing at rt for 3 h, the mixtures were chilled in an ice bath, and tested for fluorescence with FluoroMax3 at 5 °C with a continuous nitrogen flow. Slit = 3/3 nm, int time = 0.3 s.

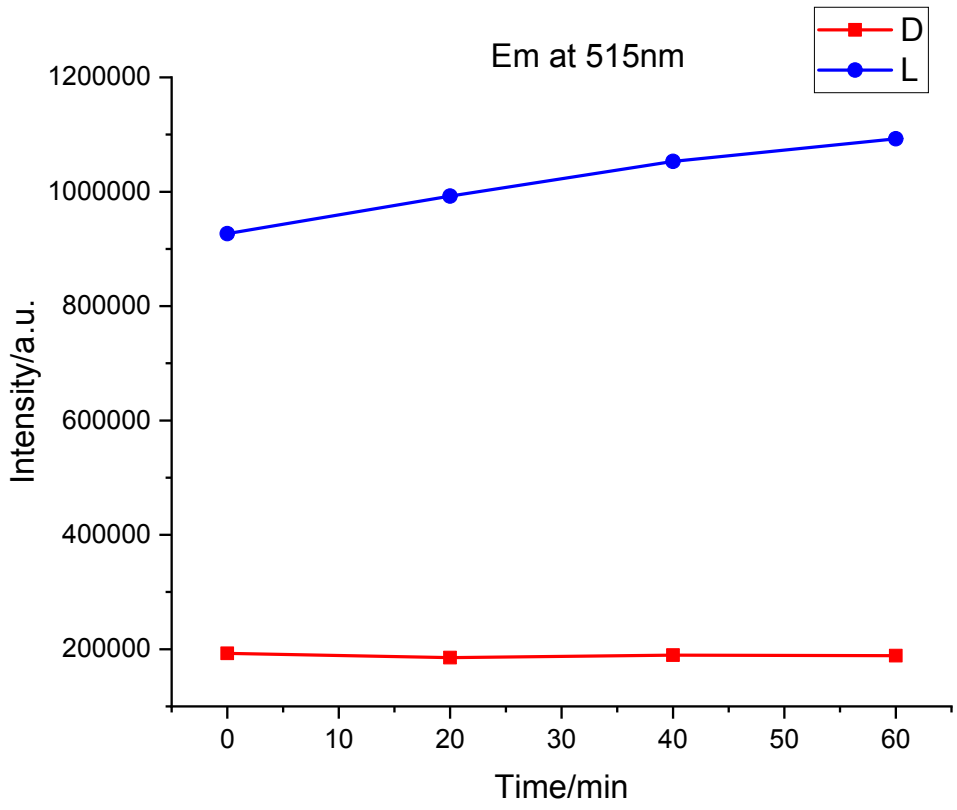


Figure S8. Fluorescence intensity of (*S*)-**2d**@PEG-PLLA in the presence of phenylalanine and Zn(II) versus time. Conditions: (*S*)-**2d (**10 μM), Zn(II) (2 equiv), phenylalanine (10 equiv), in carbonate buffer. Cooled with an ice water bath after 3 h reaction at rt, and measured at 5 °C versus time. The intensity changed 17% within an hour. λ_{ex} = 430 nm, slit = 3/3 nm, int time = 0.1 s.

4. NMR studies for the reaction of (*S*)-2c with tryptophan

General conditions: 2 mM of (*S*)-**2c** in d_6-MeOH, 5 equiv Trp-TBA salt in d_6-MeOH.

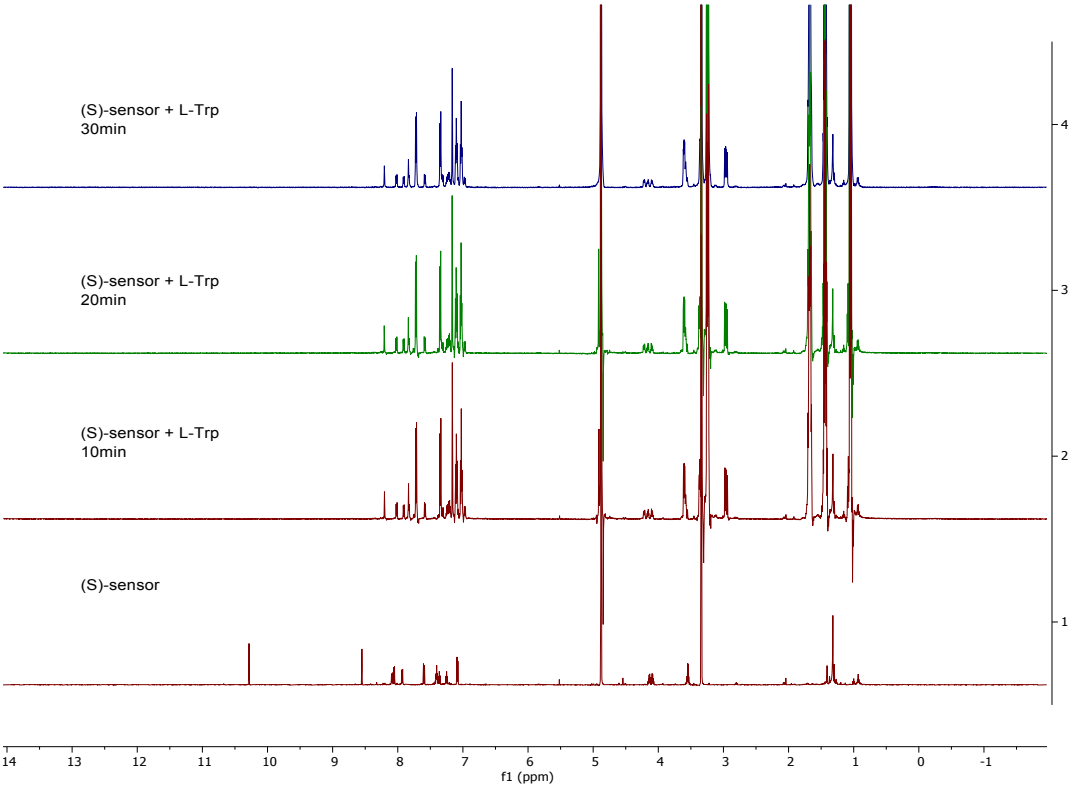


Figure S9. Kinetic studies of the reaction between (*S*)-**2c** and L-Trp.

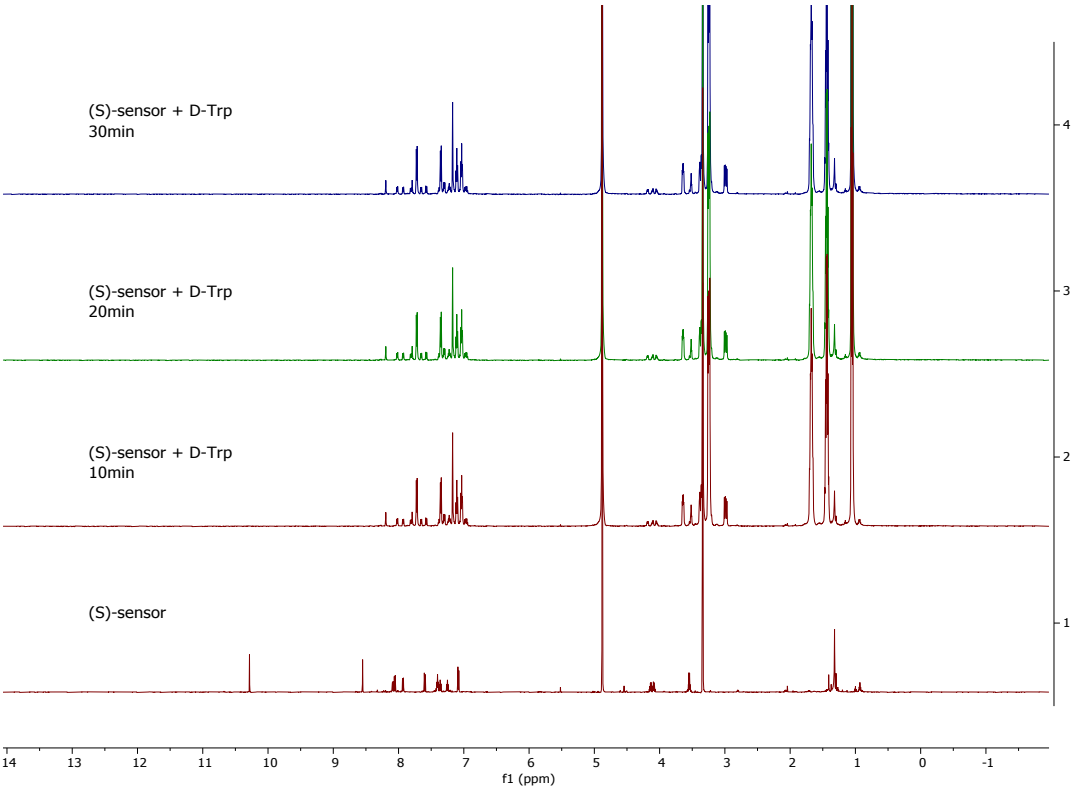


Figure S10. Kinetic studies of the reaction between (*S*)-**2c** and D-Trp.

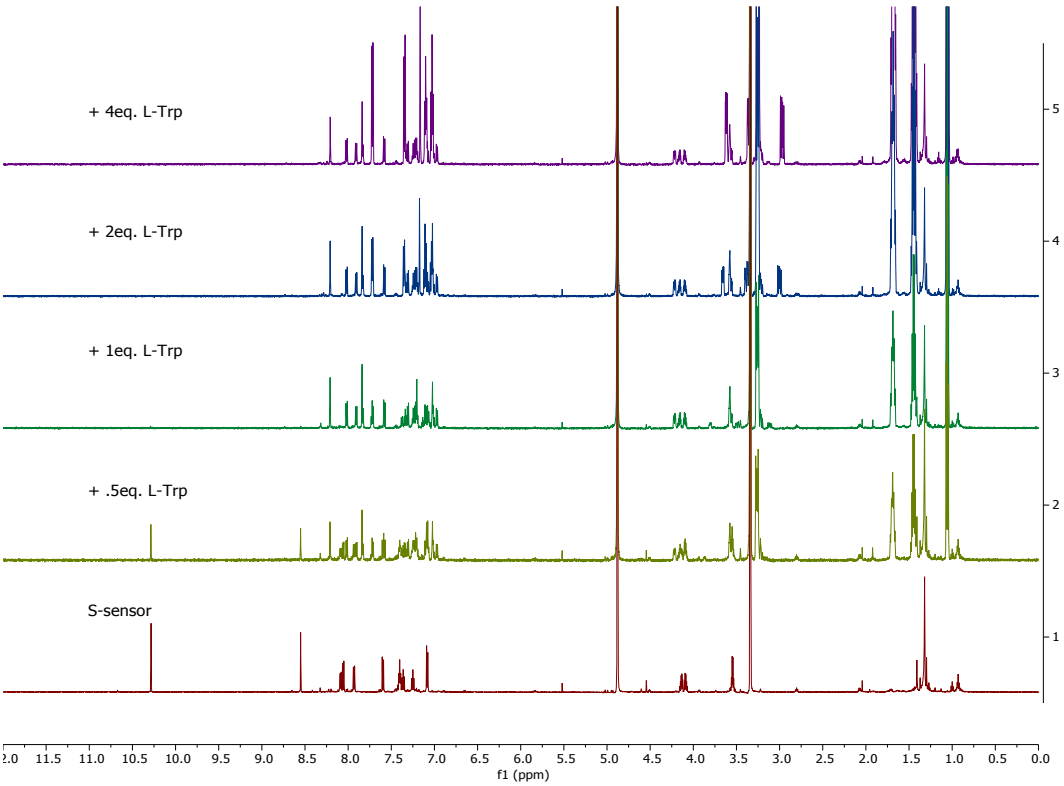


Figure S11. Stoichiometric studies of the reaction between (*S*)-**2c** and L-Trp.

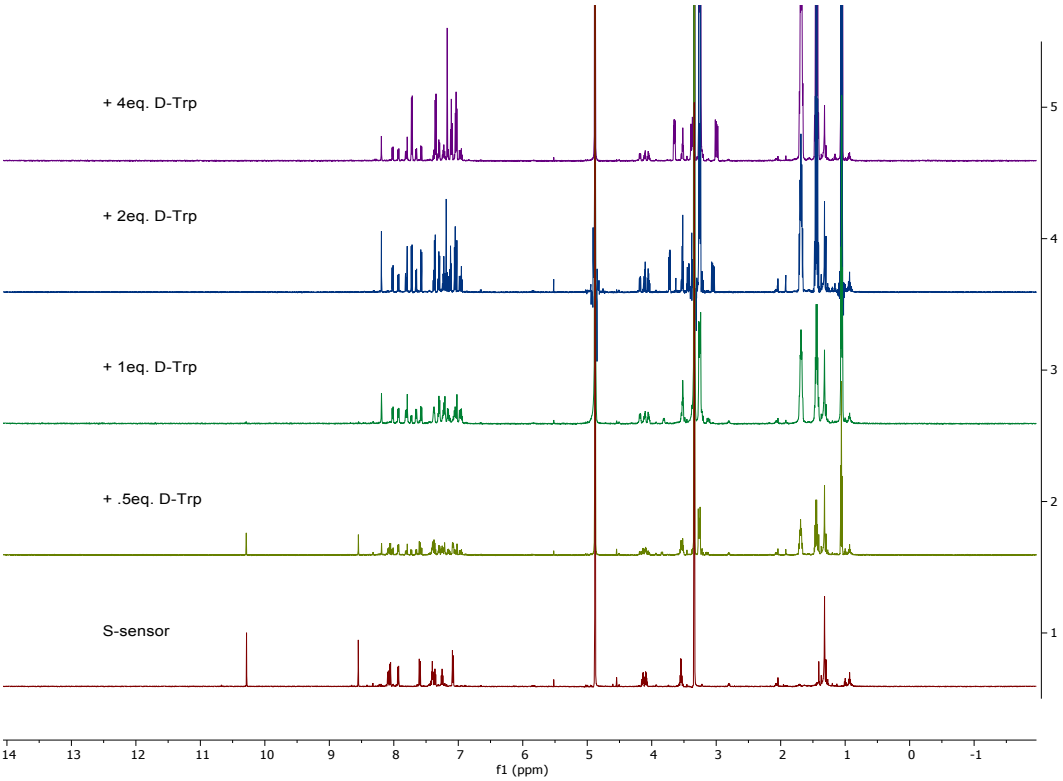


Figure S12. Stoichiometric studies of the reaction between (*S*)-**2c** and D-Trp.

5. UV-vis spectra of (*S*)-2c with tryptophan

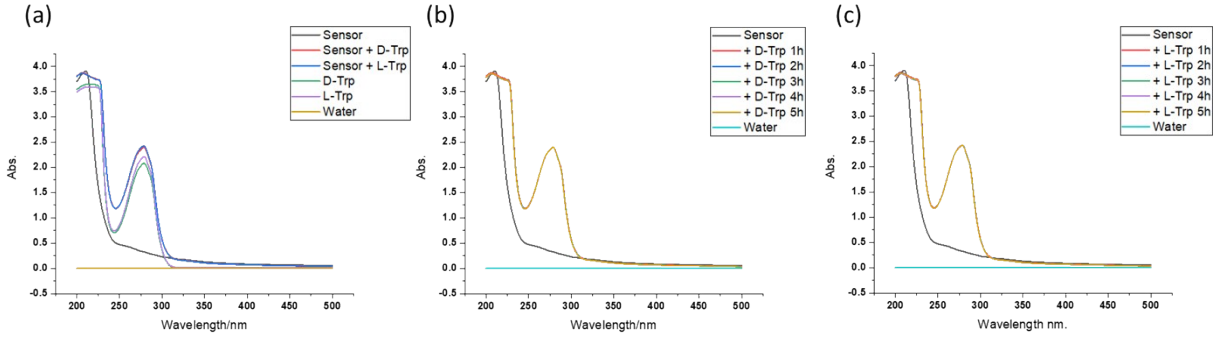


Figure S13. (a) Overview; (b) Time course with D-Trp; (c) Time course with L-Trp. Corresponding concentration = 10µM, 2eq. zinc acetate, 10eq. trp.

6. DLS studies of (*S*)-2c@PEG-PLLA with tryptophan

Procedures: To a suspension of 1 mL of 20 μM Probe and 1 mL CBS buffer, were added with 40 μL of 20 mM amino acid (in water) and 10 μL of 4 mM $Zn(OAc)_2$ (in water). After standing at rt for 3 h, the mixtures were analyzed via dynamic light scattering (DLS, Wyatt, DynaPro).

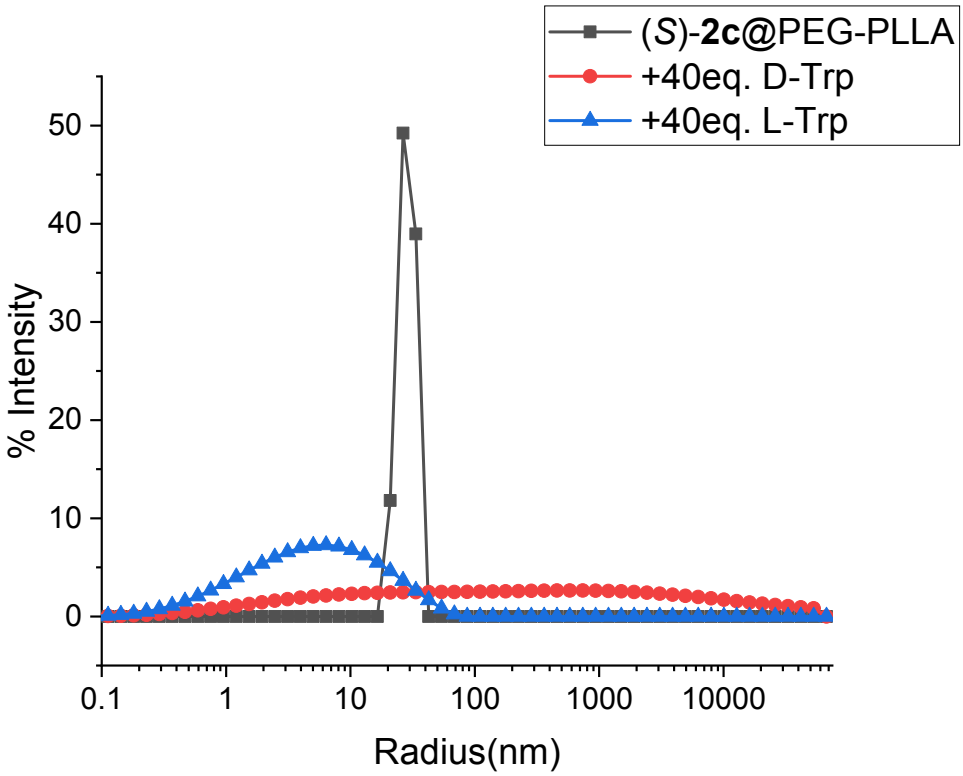
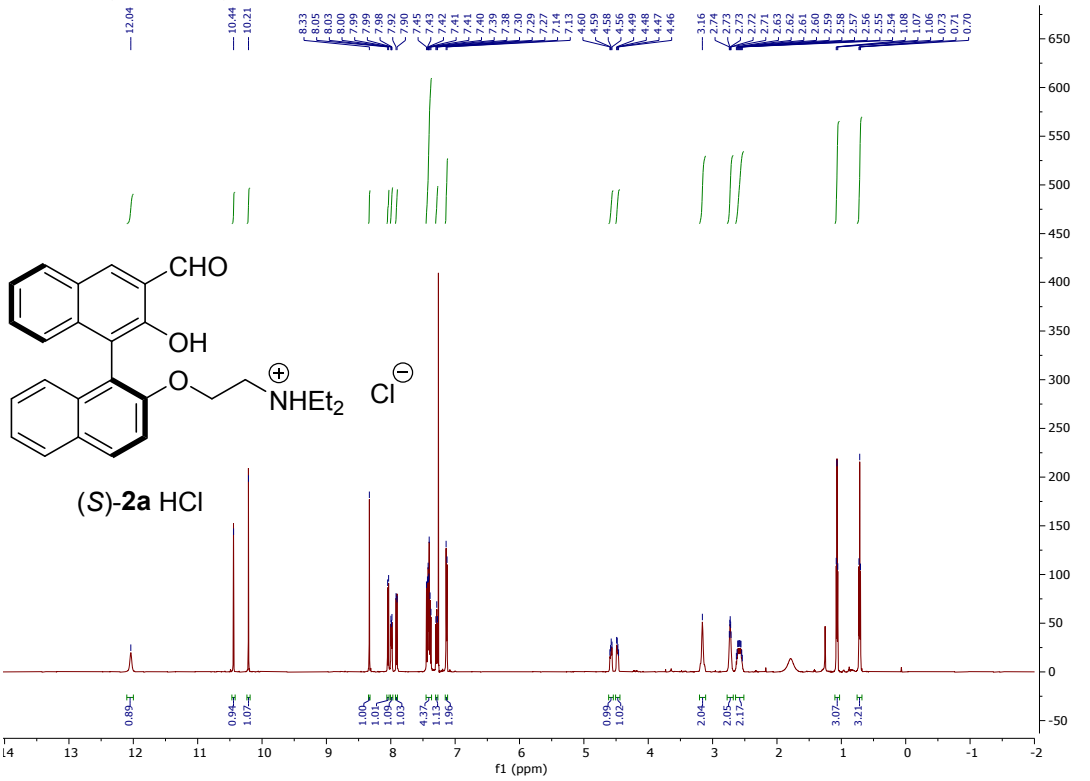


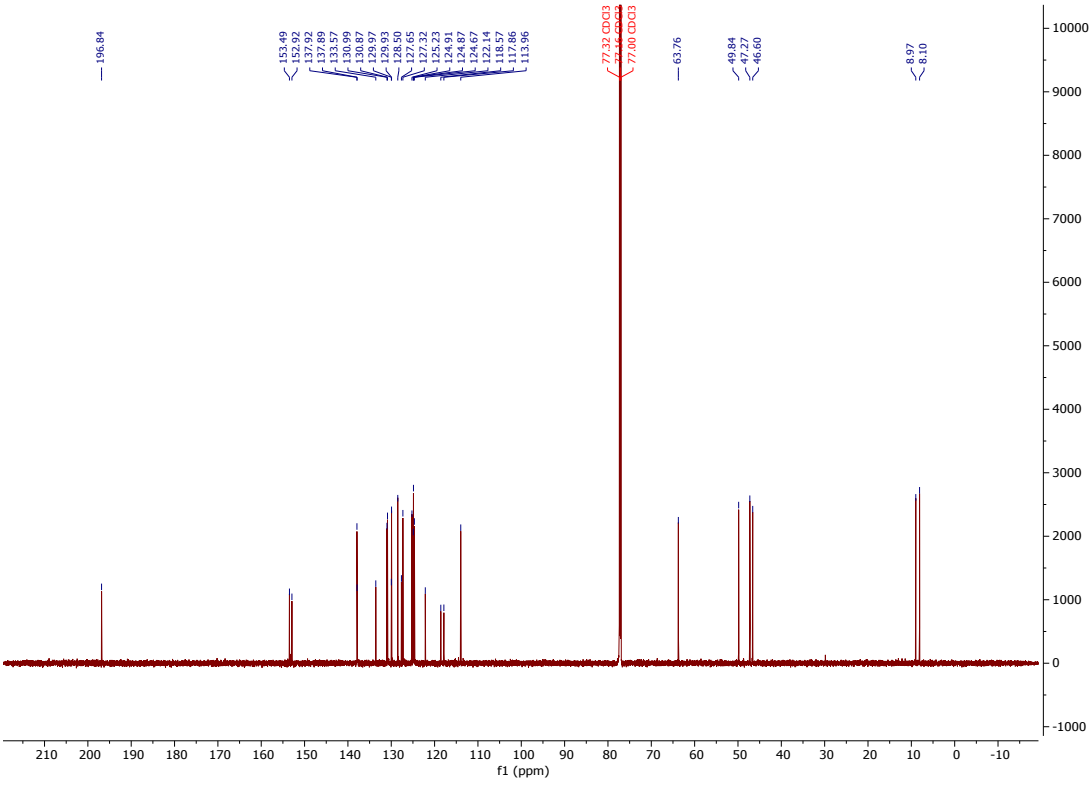
Figure S14. DLS studies.

7. NMR spectra

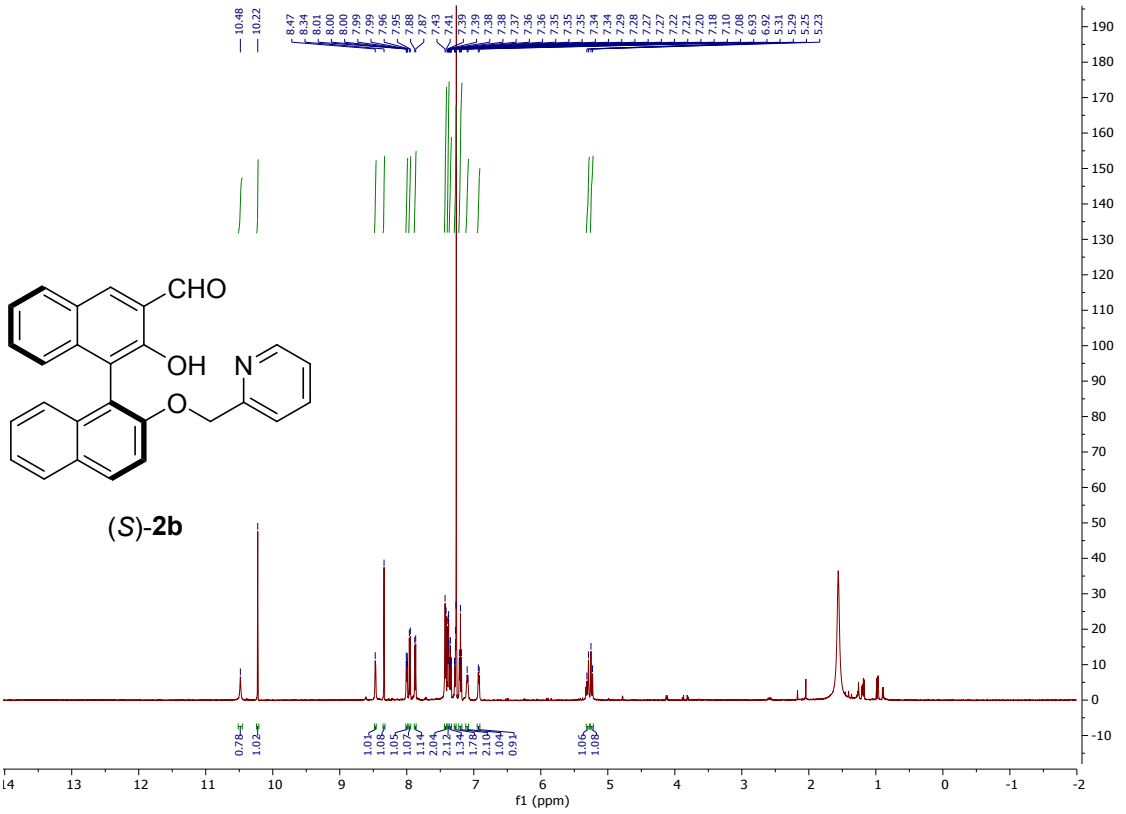
^{1}H-NMR (600 MHz), $CDCl_3$



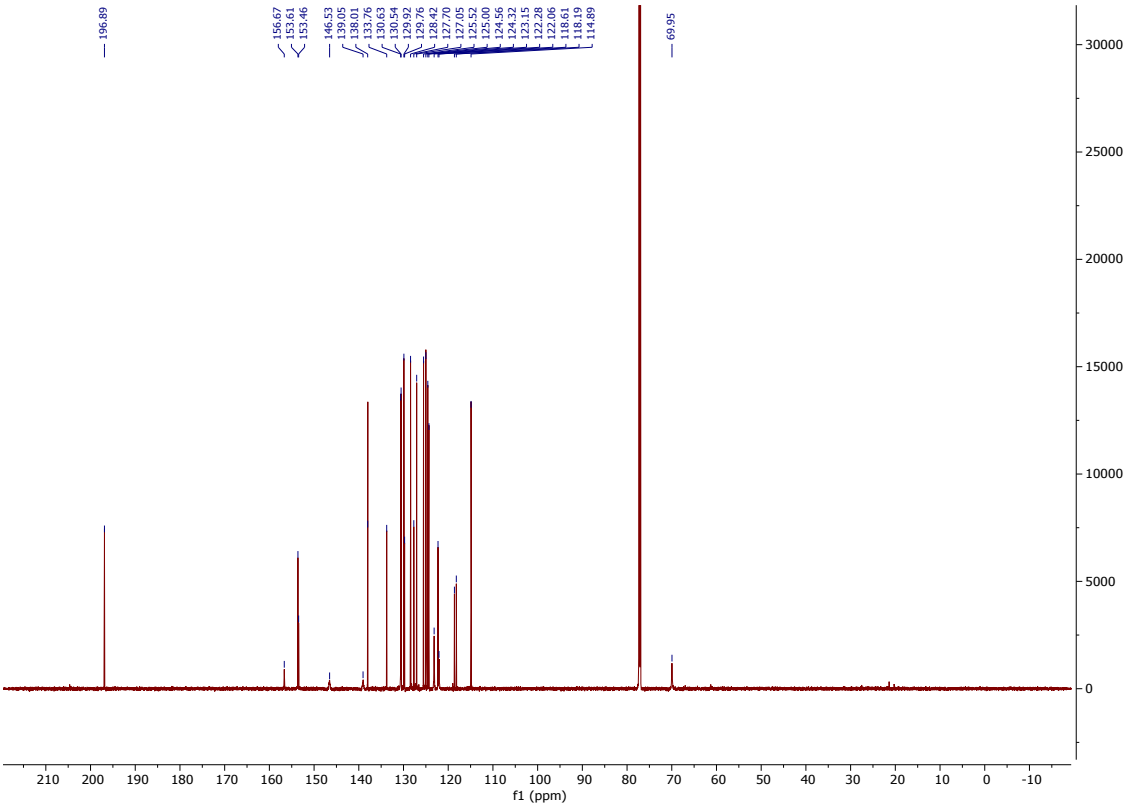
^{13}C-NMR (201MHz), $CDCl_3$



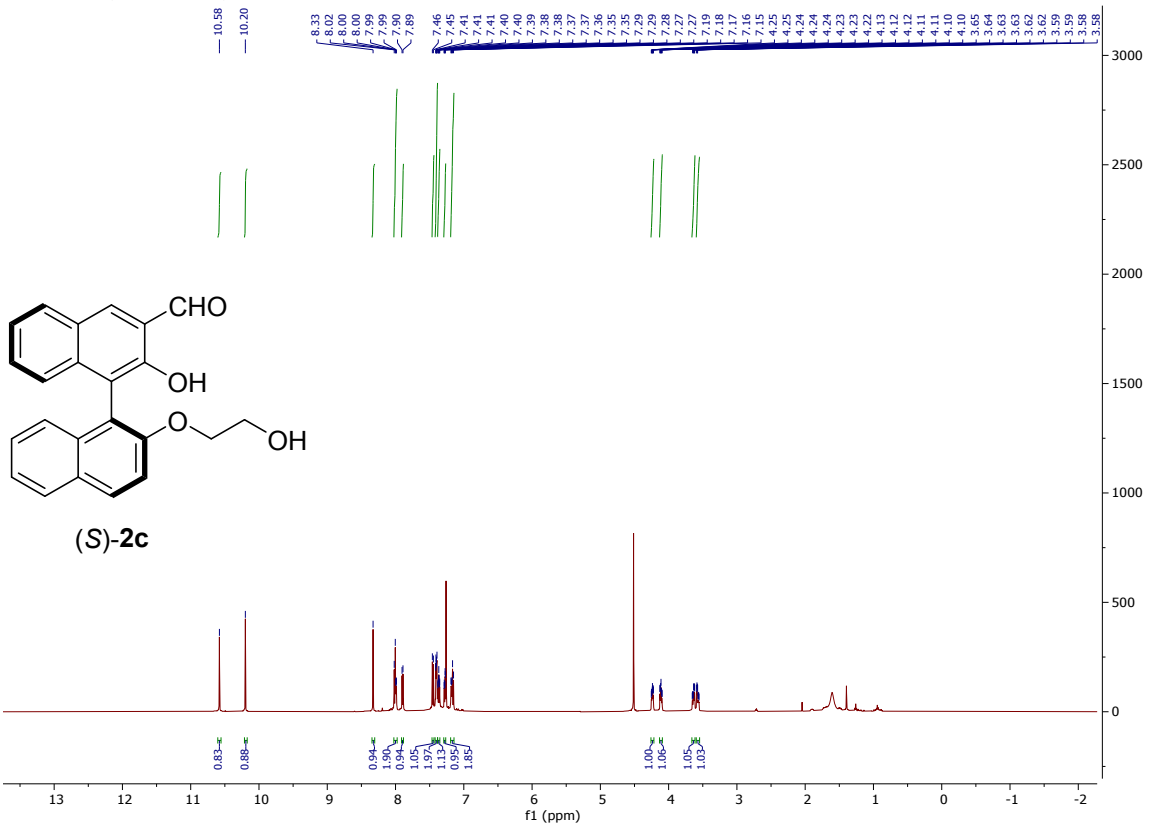
^{1}H-NMR (600 MHz), $CDCl_3$



^{13}C-NMR (201 MHz), $CDCl_3$

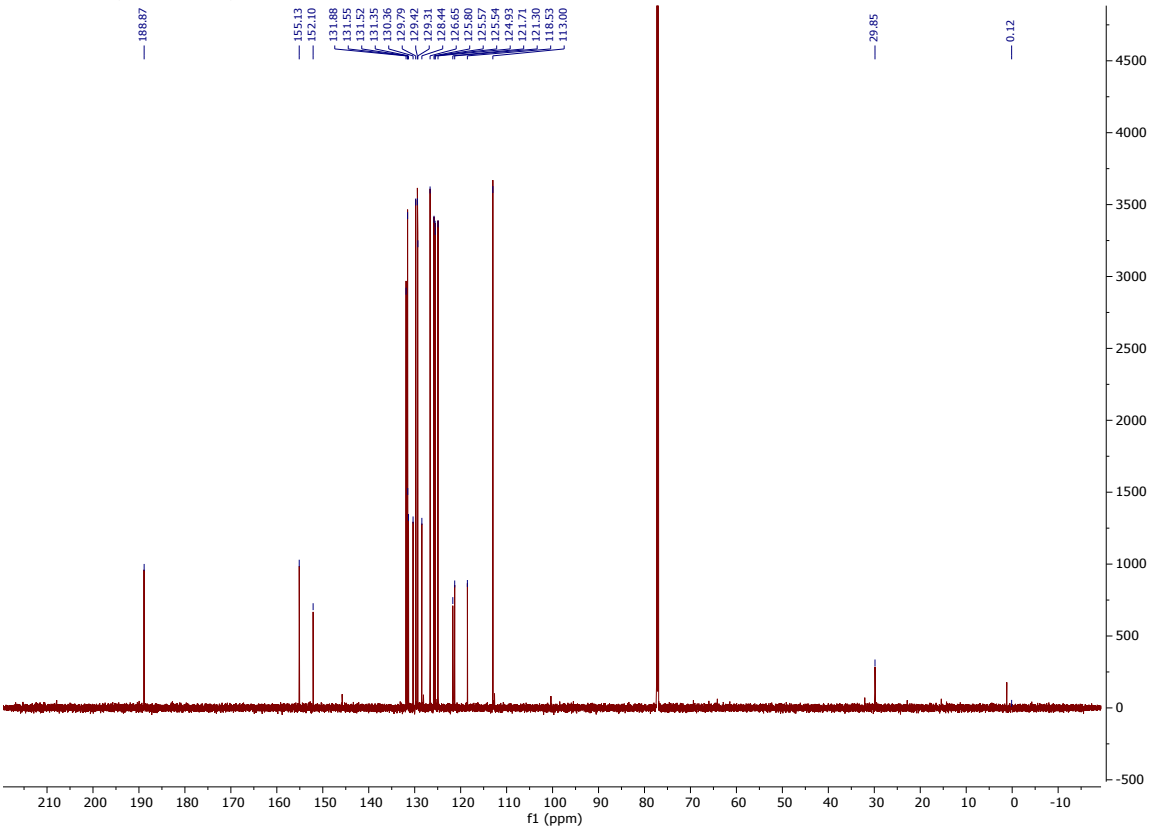


^{1}H-NMR (600 MHz), $CDCl_3$

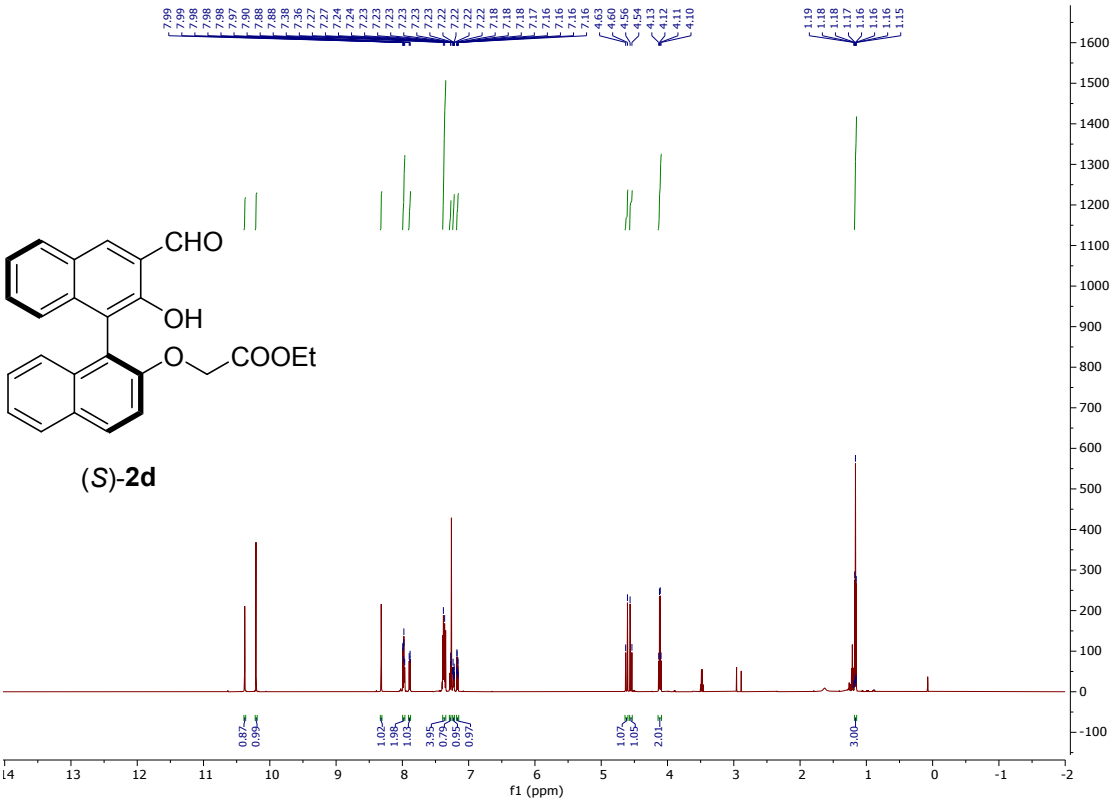


(*S*)-**2c**

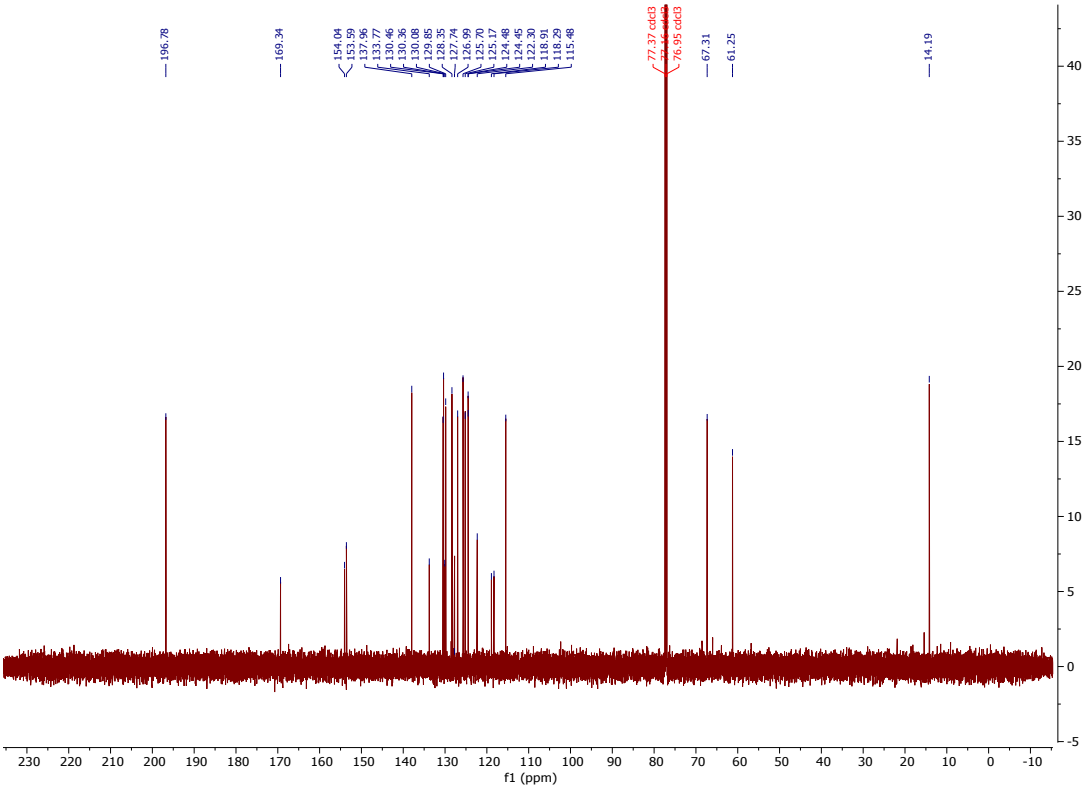
^{13}C-NMR (201 MHz), $CDCl_3$



^{1}H-NMR (600 MHz), $CDCl_3$



^{13}C-NMR (201 MHz), $CDCl_3$



8.Mass Spectra

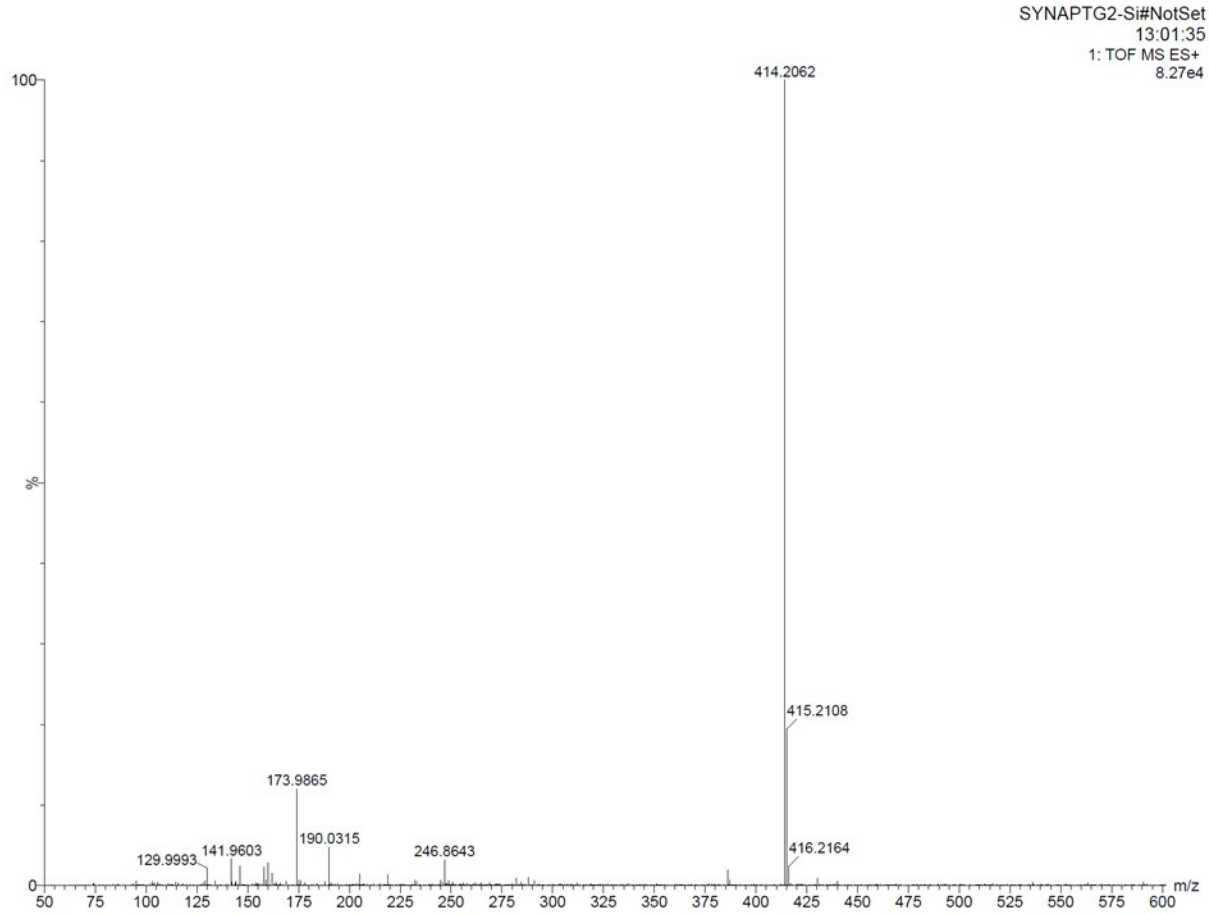


Figure S15. Mass spectrum of (*S*)-**2a**.

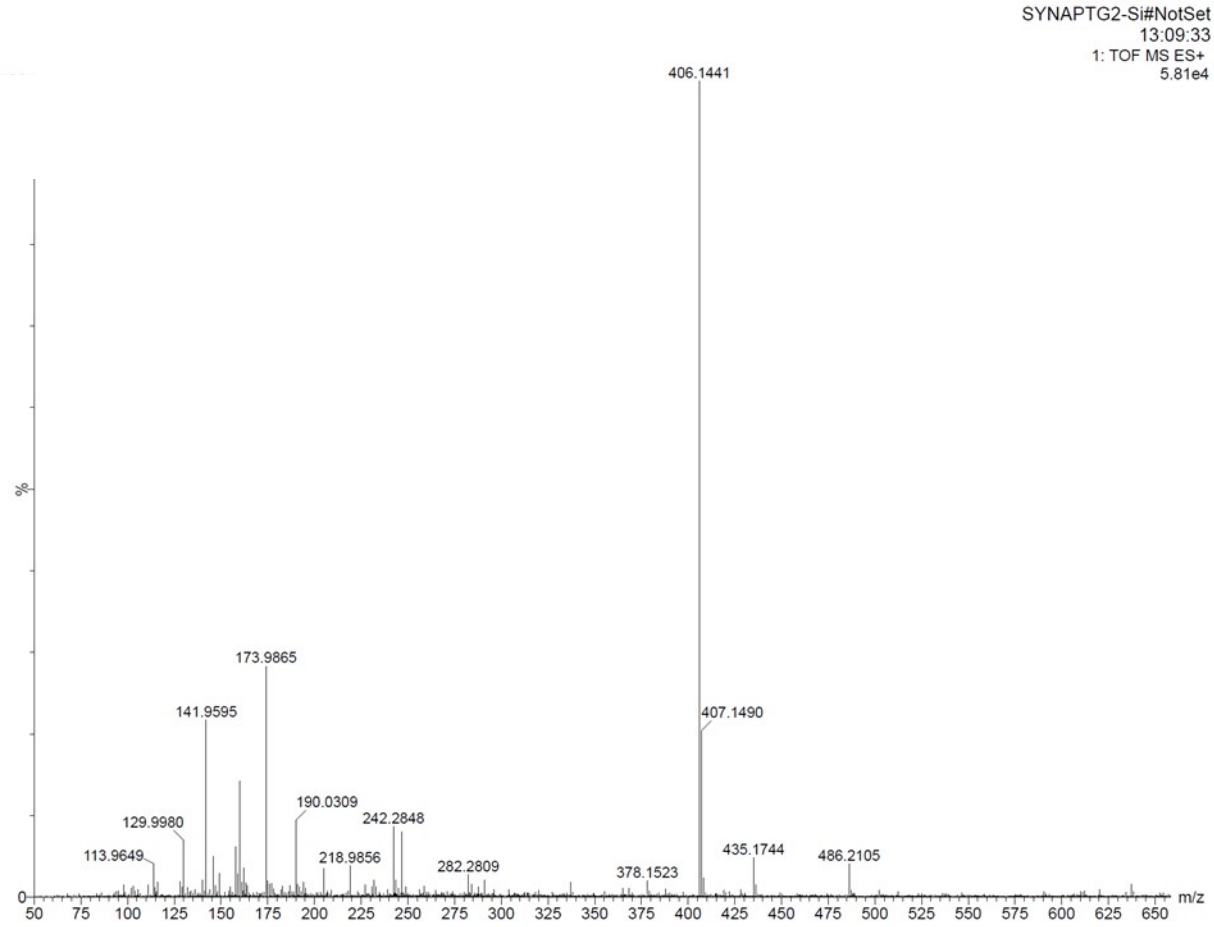


Figure S16. Mass spectrum of (*S*)-**2b**.

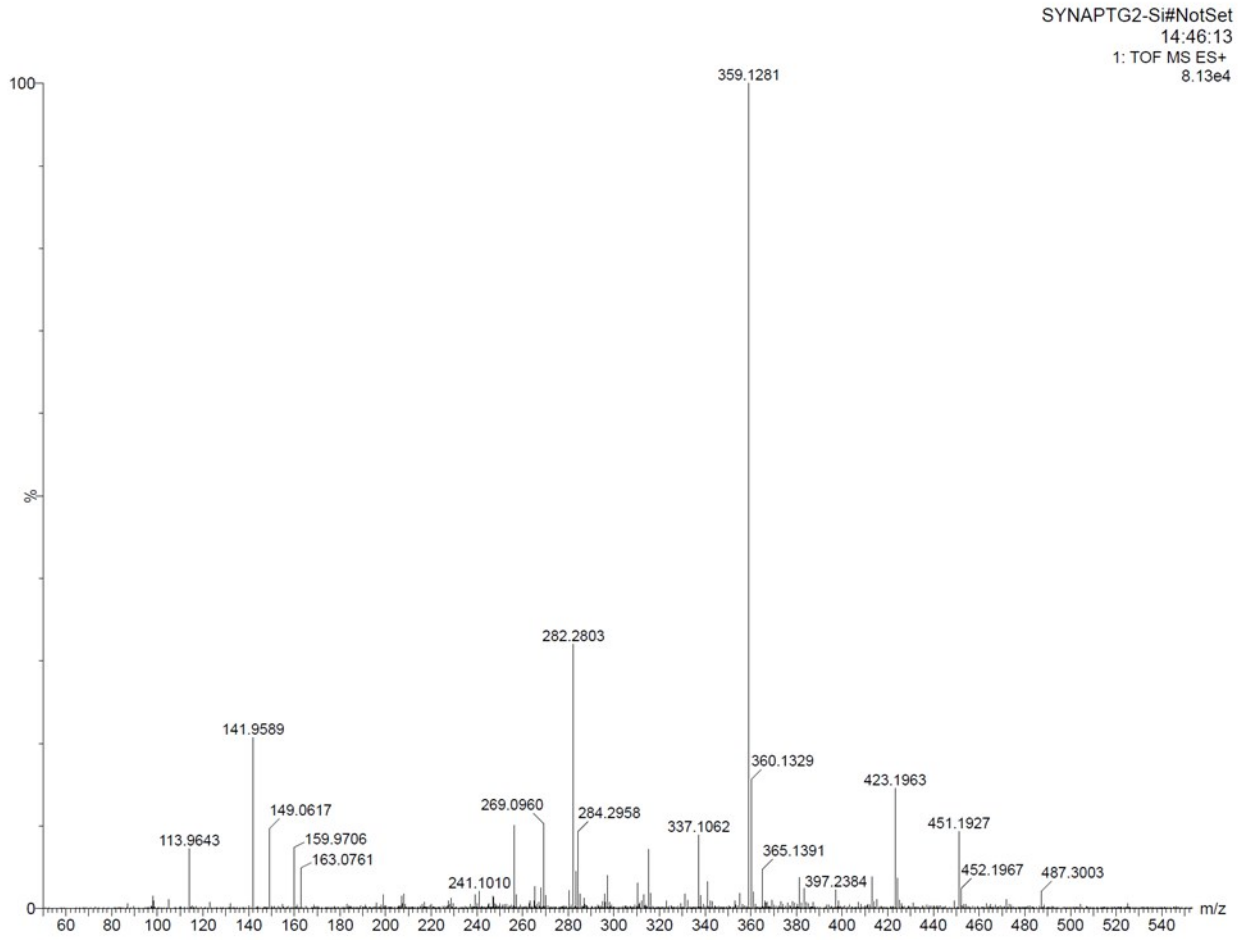


Figure S17. Mass spectrum of (*S*)-**2c**.

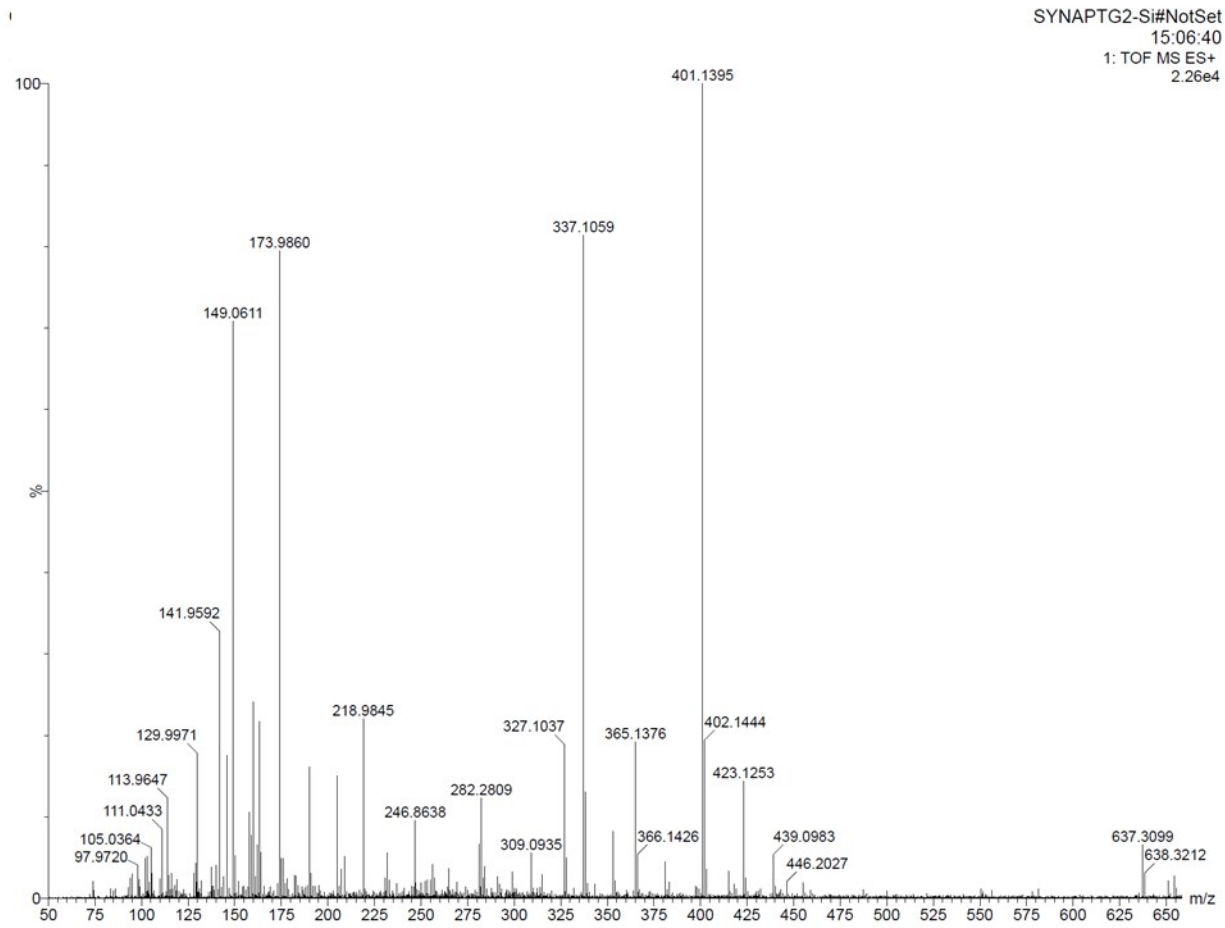


Figure S18. Mass spectrum of (*S*)-**2d**.

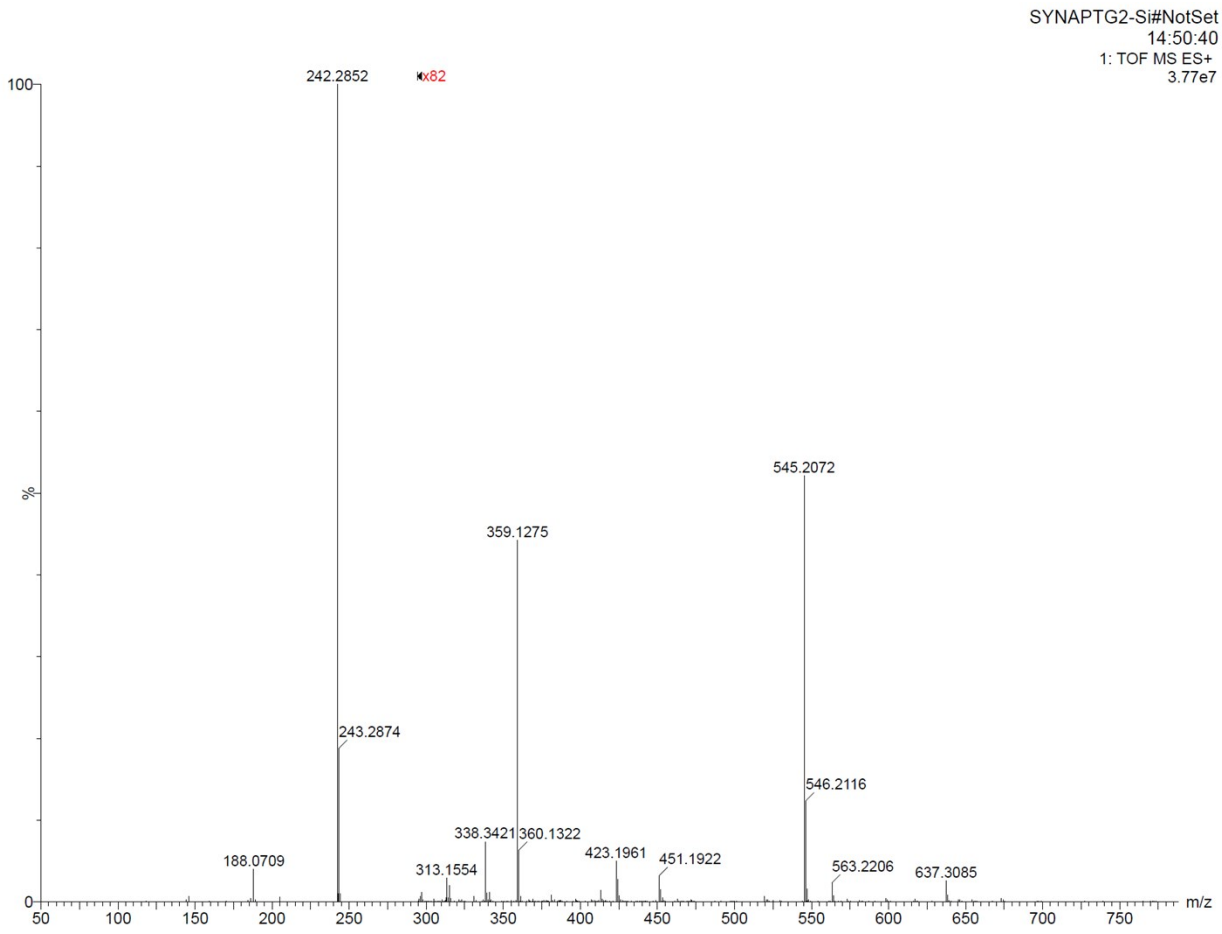


Figure S19. Mass spectrum of the mixture of (*S*)-**2c** with 4 equiv of L-Trp (TBA salt form).

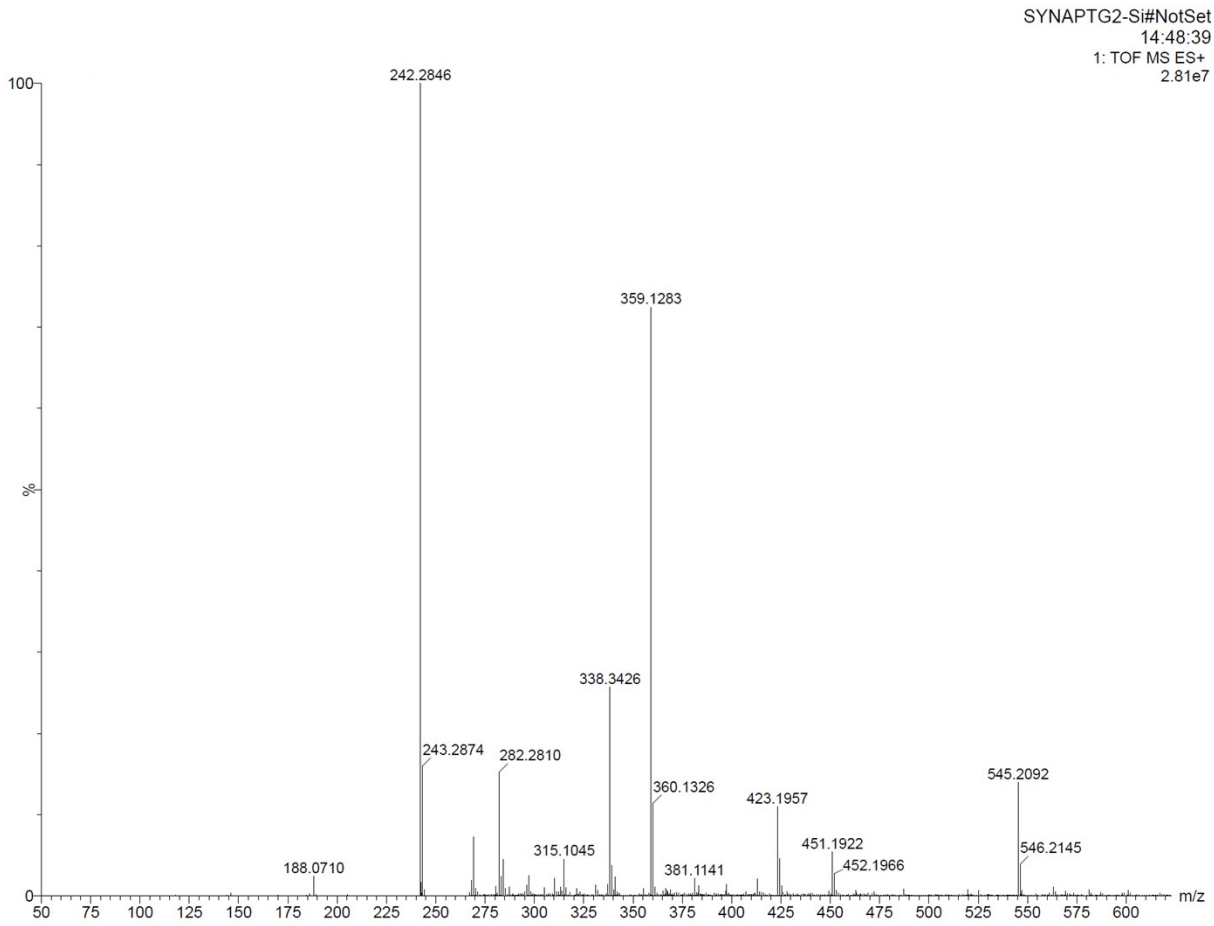


Figure S20. Mass spectrum of the mixture of (*S*)-**2c** with 4 equiv of D-Trp (TBA salt form).