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Electronic Supplementary Information (ESI)

Carbonic Anhydrase Mimics with Rationally Designed Active Sites for Fine-Tuned Catalytic Activity and Selectivity in Ester Hydrolysis

Foroogh Bahrami and Yan Zhao*

Department of Chemistry, Iowa State University, Ames, Iowa 50011-3111, USA

zhaoy@iastate.edu

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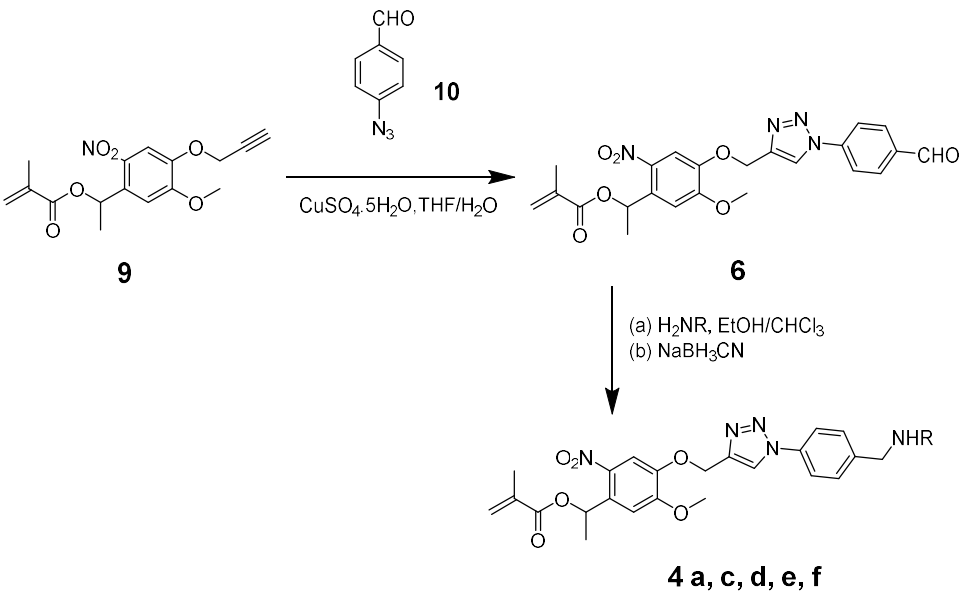
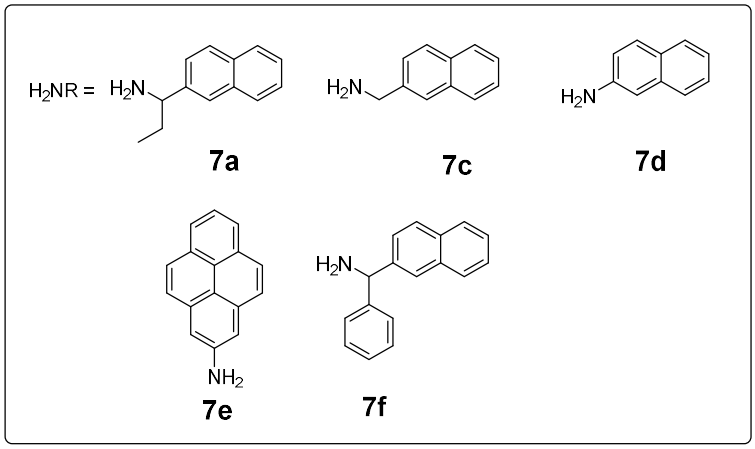
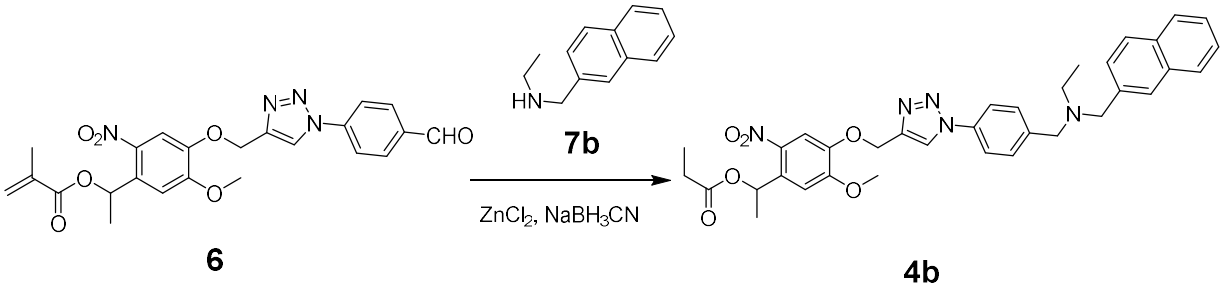
General experimental methods

All reagents and solvents were of ACS-certified grade or higher and used as received from commercial suppliers. Millipore water (18.2 MU; Millipore Co., USA) was used to prepare buffers and nanoparticles. All new compounds were characterized by ^{1}H NMR, ^{13}C NMR and HRMS. NMR spectra were recorded on a Bruker DRX-400, a Bruker AV III 600, or a Varian VXR-400 spectrometer. High resolution mass spectra (HRMS) were recorded on Agilent QTOF 6540 mass spectrometer with a QTOF detector. Dynamic light scattering (DLS) was recorded at 25 °C on a Malvern Zetasizer Nano ZS instrument. TEM was analyzed on a 200kV JEOL 2100 scanning/transmission electron microscope (STEM). UV-vis spectra were recorded on a Cary 100 Bio UV-visible spectrophotometer. GC-MS analysis was performed on Agilent 7250 GC Q-TOF MSMS with FID detector.

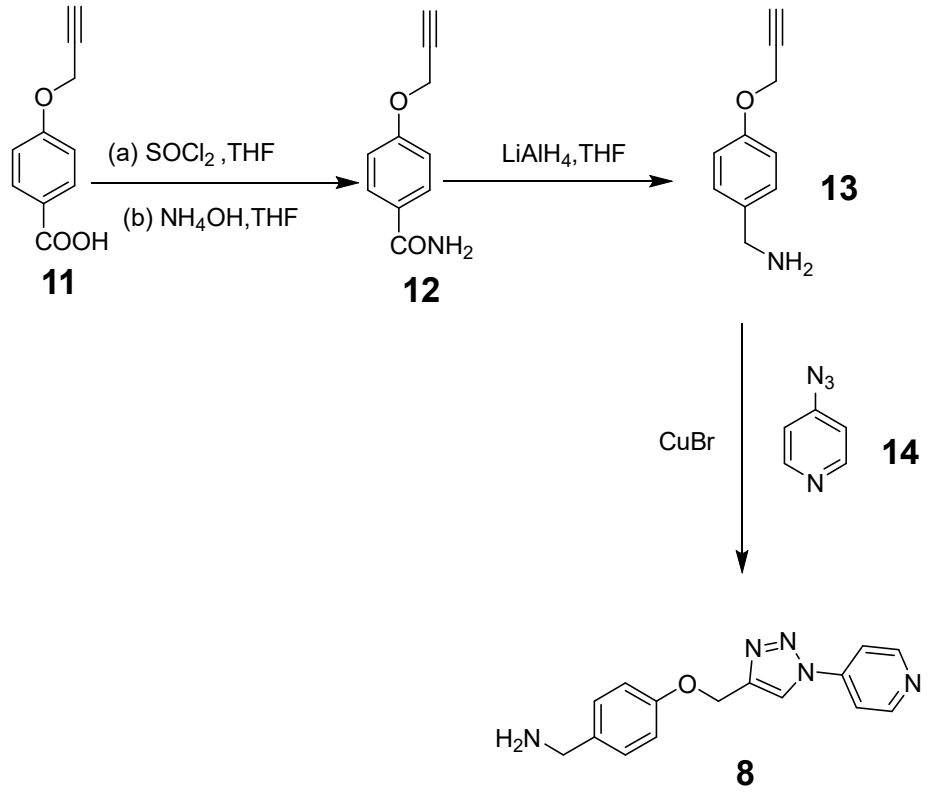




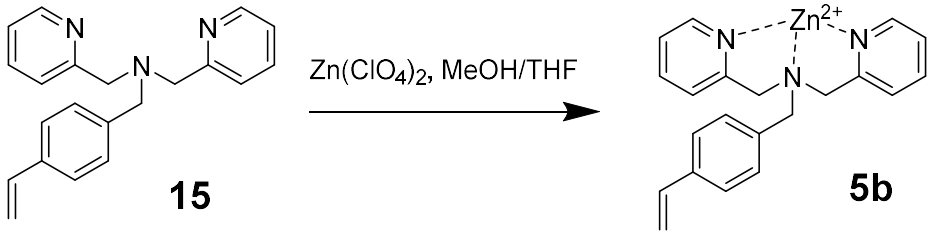
Scheme S1. Synthesis of compounds **4a**, **4c**, **4d**, **4e**, and **4f**.



Scheme S2. Synthesis of compound **4b**.



Scheme S3. Synthesis of compound **8**.



Scheme S4. Synthesis of compound **5b**.

Syntheses

Syntheses of compounds **1**,[1] **2**,[2] **3**,[3] **5a**,[4] **7a**, **7f**,[5] **7b**,[6] **9**,[7] **10**, [8] **11**,[9] **14**,[10] **15**,[11] followed previously reported procedures.

Compound 6. Compound **9** (319 mg, 1.0 mmol) in THF (1 mL) was added dropwise to a solution of compound **10** (136.33 mg, 0.93 mmol), sodium ascorbate (297mg, 1.5 mmol), and $CuSO_4.5H_2O$ (250 mg,1 mmol) in a 2:1 THF/H_2O mixture (5 mL). After being stirred at 40 °C for 12 h in dark, the solvents were removed by rotary evaporation. The residue was dissolved in ethyl acetate, organic solution was washed with water (3 × 10 mL). The combined organic phase dried over Na_2SO_4 and concentrated by rotary evaporation. The residue was purified by column chromatography over silica gel using 44:1 dichloromethane/MeOH as the eluent to give a light-yellow powder (312 mg, 72%). ^{1}H NMR (400 MHz, 298 K, $CDCl_3$) δ 10.03 (s, 1H), 8.30 (s, 1H), 8.02 (d, *J* = 8 Hz, 2H), 7.94 (d, *J* = 8 Hz, 2H), 7.79 (s, 1H), 7.05 (s, 1H), 6.48 (q, *J* = 12 Hz, 1H), 6.16 (s, 1H),5.60 (s, 1H), 5.36 (s, 2H), 3.91 (s, 3H), 1.92 (s, 3H), 1.63 (d, *J* = 8 Hz, 3H). ^{13}C NMR (100 MHz, 298 K, $CDCl_3$) δ 190.6, 166.1, 154.1, 146.3, 140.8, 139.6, 136.2,136.1, 134.4, 131.4, 125.8, 120.6, 110.1, 110.0, 108.3, 68.7, 69.9, 56.3, 22.0, 18.3, HRMS (ESI^+/QTOF). Calcd for $C_{17}H_{16}ClN_3O_6$ m/z: $[M+H]^+$ 467.1567; Found m/z 467.1451.

General procedure for the synthesis of compounds 4a, **4c**, **4d**, **4e**, and **4f.** To a solution of compound **6** (0.25 mmol) in a 4:1 EtOH/$CHCl_3$ mixture (5 mL) was added the appropriate amine (**7a**, **7c**, **7d**, **7e**, or **7f**) (0.30 mmol). The reaction mixture was heated to reflux overnight under N_2. After it was cooled to room temperature, $NaBH_3CN$ (0.75 mmol) was added. The reaction mixture was stirred for another 48 h at room temperature. The solvents were removed by rotary evaporation and the residue was dissolved in $CHCl_3$. The organic solution was washed with water (10 mL), dried over $NaSO_4$, and concentrated by rotary evaporation. The residue was purified by column chromatography over silica gel using 60:40 hexane/ethyl acetate as the eluent to give product.

Compound 4a. The product was prepared according to the general procedure above by using 0.25 mmol of compound **6** and 0.3 mmol of compound **7a** to afford a yellow solid (98 mg, 62%). ^{1}H-NMR (400 MHz, 298 K, $CDCl_3$): δ 8.12 (s, 1H), 7.92-7.86 (m, 3H), 7.84 (s, 1H), 7.79 (d, *J* = 8.7 Hz, 2H), 7.68 (d, *J* = 8.7 Hz, 2H), 7.64 -7.48 (m, 4H), 7.07 (s, 1H), 6.55 (q, *J* = 6.4 Hz, 1H), 6.20 (d, *J* = 1.6 Hz, 1H), 5.64 (d, *J* = 1.6 Hz, 1H), 5.40 (d, *J* = 2.9 Hz, 2H), 4.49 (s, 1H), 4.15 (p, *J* = 7.0 Hz, 2H), 3.96 (s, 1H), 3.95 (s, 3H), 1.97 (s, 3H), 1.92-1.86 (m, 2H), 1.72-1.66 (m, 3H), 0.92 (t, *J* = 7.4 Hz, 3H). ^{13}C-NMR (100 MHz, DMSO-d_6): 166.1, 153.9, 146.7, 143.3, 141.1, 140.0, 137.6, 136.1, 135.6, 133.3, 132.7, 132.6, 130.1, 128.2,127.9, 127.9, 127.4, 127.2, 126.8, 126.4, 126.0, 123.7, 120.6, 109.7, 109.3, 68.3, 62.4, 57.6, 56.8, 56.6, 47.0, 21.7, 18.3, 12.0. HRMS (ESI^+/QTOF) Calculated for $C_{36}H_{37}N_5O_6$ m/z: $[M+H]^+$ 636.2822; Found 636.2820.

Compound 4c. The product was prepared according to the general procedure above by using 0.25 mmol of compound **6** and 0.3 mmol of compound **7c** to afford a yellow solid (66 mg, 44%). ^{1}H-NMR (600 MHz, 298 K, $CDCl_3$): δ 8.11 (s, 1H), 7.84-7.81 (m, 4H), 7.78 (s, 1H), 7.69 (d, *J* = 8.5 Hz, 2H), 7.52 (d, *J* = 8.4 Hz, 2H), 7.51 -7.43 (m, 3H), 7.08 (s, 1H), 6.56 (q, *J* = 6.4 Hz, 1H), 6.20 (d, *J* = 1.6 Hz, 1H), 5.66-5.62 (m, 1H), 5.35 (d, *J* = 2.2 Hz, 2H), 4.0 (s, 2H), 3.94 (s, 3H), 3.92 (s, 2H), 3.72 (s, 1H), 1.98(s, 3H), 1.68 (d, *J* = 6.4 Hz, 3H),). ^{13}C-NMR (100 MHz, $CDCl_3$): 166.1, 154.1, 146.4, 143.3, 140.5, 139.6, 136.5, 136.2, 135.7, 134.1, 133.3, 132.7, 129.6, 129.6, 128.2, 127.7, 127.6, 126.8, 126.5, 126.1, 125.9, 125.7, 121.6, 120.6, 114.4, 114.3, 114.2, 109.9, 108.2,

68.7, 62.9, 56.2, 53.5, 52.8, 51.9, 22.0, 18.3. HRMS (ESI$^+$/QTOF) Calculated for $C_{34}H_{33}N_5O_6$ m/z: $[M+H]^+$ 608.2509; Found 608.2509.

Compound 4d. The product was prepared according to the general procedure above by using 0.25 mmol of compound **6** and 0.3 mmol of compound **7d** to afford a yellow solid (78 mg, 53%). ^{1}H-NMR (600 MHz, 298 K, $CDCl_3$): δ 8.09 (s, 1H), 7.90 (d, *J* = 12 Hz, 1H), 7.85 (s, 1H), 7.83 (d, *J* = 6.4 Hz, 1H), 7.70 (d, *J* = 6.4 Hz, 2H), 7.59 (d, *J* = 6 Hz, 2H), 7.47-7.51(m, 2H), 7.28-7.23 (m, 2H), 7.08 (s, 1H), 6.57-6.56 (s, 1H), 6.56 (m,1H), 6.21 (s, 1H), 5.65 (s, 1H), 5.37 (d, *J* = 2.8 Hz, 2H), 4.93 (s, 1H), 4.61 (s, 2H), 3.95 (s, 3H), 1.99 (s, 3H), 1.70 (d, *J* = 6.4 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$): 166.0, 154.0, 146.4, 143.4, 142.6, 140.4, 139.6, 136.2, 135.9, 134.3, 134.2, 128.7, 128.6, 126.4, 125.9, 123.4, 121.5, 120.9, 119.8, 118.0, 110.0, 108.2, 105.0, 68.7, 63.0, 56.2, 47.7, 29.7, 22.0, 18.3. HRMS (ESI$^+$/QTOF) Calculated for $C_{33}H_{31}N_5O_6$ m/z: $[M+H]^+$ 594.2353; Found 594.2227.

Compound 4e. The product was prepared according to the general procedure above by using 0.25 mmol of compound **6** and 0.3 mmol of compound **7e** to afford a yellow solid (78 mg, 47%). ^{1}H-NMR (600 MHz, 298 K, $CDCl_3$): δ 8.11 (s, 1H), 7.87 (s, 1H), 7.72 (d, *J* = 8.4 Hz, 2H), 7.53 (d, *J* = 8.2 Hz, 2H), 7.33 (dd, *J* = 8.4, 7.3 Hz, 2H), 7.29 (s, 1H), 7.16-7.08 (m, 3H), 7.01 (d, *J* = 8.0 Hz, 2H), 6.57 (q, *J* = 6.4 Hz, 1H), 6.48-6.41 (m, 2H), 6.36 (s, 1H), 6.21 (s, 1H), 5.65 (s, 1H), 5.42 (d, *J* = 2.0 Hz, 2H), 4.42 (s, 2H), 3.96 (s, 3H), 1.99 (s, 3H), 1.70 (d, *J* = 6.4 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$): 166.0, 154.0, 146.3, 143.4, 139.6, 136.2, 136.1, 134.2, 127.5, 126.1, 125.7, 121.3, 120.8, 119.3, 110.0, 108.2, 68.7, 62.9, 56.2, 22.0, 18.3. HRMS (ESI$^+$/QTOF) Calculated for $C_{39}H_{34}N_5O_6$ m/z: $[M+H]^+$ 668.2376; Found 668.2376.

Compound 4f. The product was prepared according to the general procedure above by using 0.25 mmol of compound **6** and 0.3 mmol of compound **7f** to afford a yellow solid (116 mg, 68%). ^{1}H-NMR (600 MHz, 298 K, $CDCl_3$): δ 8.12 (s, 1H), 7.94 (s, 1H), 7.88-7.85 (m, 2H), 7.82 (d, *J* = 8.5 Hz, 2H), 7.72 (d, *J* = 8.5 Hz, 2H), 7.57 -7.46 (m, 7H), 7.36 (t, *J* = 7.7 Hz, 2H), 7.30-7.24 (m, 1H), 7.11 (s, 1H), 6.58 (q, *J* = 6.4 Hz, 1H), 6.21 (s, 1H), 5.42 (s, 2H), 5.06 (s, 1H), 3.97 (s, 3H), 3.90 (s, 2H), 2.00 (s, 3H), 1.71 (d, *J* = 6.5 Hz, 3H),). ^{13}C-NMR (100 MHz, $CDCl_3$): 166.0, 154.1, 146.4, 146.4, 143.3, 136.3, 134.1, 133.3, 132.7, 129.5, 128.5, 128.3, 127.8, 127.5, 127.3, 126.0, 125.7, 125.6, 125.5, 121.5, 120.6, 109.9, 108.3, 68.5, 62.9, 56.2, 21.8, 18.1. HRMS (ESI$^+$/QTOF) Calculated for $C_{40}H_{37}N_5O_6$ m/z: $[M+H]^+$ 684.2822; Found 684.2812.

Compound 4b. The compound was synthesized according to a modified procedure from the literature.[12] Compound **7b** (77.5 mg, 0.42 mmol) was added to a solution of compound **6** (233 mg, 0.50 mmol), $NaBH_3CN$ (32 mg, 0.50 mmol), and $ZnCl_2$ (29 mg, 0.21 mmol) in MeOH (5 mL). After the reaction mixture was stirred for 24h at room temperature, the solid was removed by filtration. The filtrate was concentrated under reduced pressure. The residue was purified by column chromatography over silica gel using 3:2 hexane/ethyl acetate as the eluent to give a white powder (60 mg, 38%). ^{1}H-NMR (600 MHz, 298 K, $CDCl_3$): δ 8.09 (s, 1H), 7.87 (s, 1H), 7.84 (d, *J* = 8.0 Hz, 2H), 7.80 (s, 1H), 7.69 (d, *J* = 8.0 Hz, 2H), 7.64 -7.55 (m, 3H), 7.55 -7.43 (m, 3H), 7.08 (s, 1H), 6.57 (q, *J* = 6.4 Hz, 1H), 6.21 (s, 1H), 5.65 (s, 1H), 5.41 (d, *J* = 6.4 Hz, 2H), 3.96 (s, 3H), 3.78 (s, 2H), 3.69 (s, 2H), 2.61-2.60 (m, 2H), 1.99 (s, 3H), 1.70 (d, *J* = 6.5 Hz, 3H), 1.15 (m, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$): 166.0, 154.0, 146.4, 143.3, 139.6, 136.2, 134.2, 133.2, 132.7, 129.9, 127.9, 127.6, 127.1, 125.9, 125.7, 125.5, 121.4, 120.5, 110.0, 108.2, 68.7,63.0, 58.0, 56.2, 47.3, 22.0, 18.3, 11.92. HRMS (ESI$^+$/QTOF) Calculated for $C_{36}H_{37}N_5O_6$ m/z: $[M+H]^+$ 636.2822; Found 636.3168.

Compound 12. The compound was synthesized according to a modified procedure from the literature.[13] Thionyl chloride (0.89 mL, 12.3mmol) was added to a solution of compound **11** (1.40 g, 8 mmol) in dry THF (20 mL). The reaction mixture was heated to reflux until TLC showed the consumption of the acid. The volatiles were removed by rotary evaporation. The residue was dissolved in chloroform (5 mL) and cooled with an ice bath. The cooled solution was added slowly to a stirred solution of NH_4OH (36%, 6 ml) at 0 °C. After 15 min, water (30 mL) was added. The mixture was extracted with chloroform (3 × 50 mL). The combined organic phase was dried over Na_2SO_4 and concentrated by rotary evaporation. The residue was purified by column chromatography over silica gel using 30:1 dichloromethane/MeOH as the eluent to afford **6** as a white powder (1.01 g, 72 %) ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.86 (d, J = 9.0 Hz, 2H), 7.22 (s, 1H), 7.04 (d, J = 8.9 Hz, 2H), 4.88 (d, J = 2.3 Hz, 2H), 3.61 (t, J = 2.3 Hz, 1H)^{13}C-NMR (100 MHz, DMSO-d_6): 167.7, 159.8, 131.7, 129.7, 127.6, 115.0, 114.7, 79.3, 78.9, 55.9HRMS (ESI$^+$/QTOF) Calculated for $C_{10}H_{10}NO_2$ m/z: $[M+H]^+$ 176.0717; Found 176.0517.

Compound 13. The compound was synthesized according to a modified procedure from the literature.[14] $LiAlH_4$ (0.38 g, 10 mmol) was added in small portions over 25 min to a solution of compound **7** (1.57 g, 9 mmol) in anhydrous THF (20 mL) at 0 °C. The reaction mixture was heated to reflux for 4 h and cooled to room temperature. Silica gel (3 g) was added, followed by 1 M NaOH solution (5 mL). After stirred for 20 min, the mixture was filtered through a pad of sodium sulfate and the filter cake was washed with diethyl ether (30 mL). The combined filtrate was concentrated by rotary evaporation to afford a white powder (986 mg, 68 %). ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.27 (d, J = 8.7 Hz, 2H), 6.93 (d, J = 8.7 Hz, 2H), 4.77 (d, J = 2.5 Hz, 2H), 3.66 (s, 2H), 3.55 (t, J = 2.4 Hz, 1H), 2.34 (s, 2H).

Compound 8. Compound **13** (500.0 mg, 3 mmol) in DMF (2 mL) was added dropwise to a solution of compound **14** (360 mg, 3.0 mmol), CuBr (430.35 mg, 3.0 mmol), N,N,N′,N″,N″-Pentamethyldiethylenetriamine (PMDETA, 0.63 ml, 3.0 mmol) in DMF (5 mL). After stirred at room temperature for 24 h, the reaction mixture was diluted with water. The mixture was extracted with ethyl acetate (3 × 50 mL). The combined organic phase was washed with water (5 × 10 mL), brine (2 × 10 mL), dried over Na_2SO_4, and concentrated by rotary evaporation. The residue was purified by column chromatography over silica gel using 25:4 dichloromethane/MeOH as the eluent to give a light-yellow powder (472.3 mg, 56%).^{1} H NMR (400 MHz, CD_3OD) δ 8.84 (s, 1H), 8.73 (d, J = 4.9 Hz, 2H), 7.99 (d, J = 4.9 Hz, 2H), 7.28 (d, J = 8.6 Hz, 2H), 7.02 (d, J = 8.6 Hz, 2H), 5.25 (s, 2H), 3.75 (s, 2H). ^{13}C-NMR (100 MHz, CD_3OD): δ 157.4, 150.9, 145.4, 143.6, 133.9, 128.6, 121.8, 121.8, 114.5, 113.9, 60.9, 44.3. HRMS (ESI$^+$/QTOF) Calculated for $C_{15}H_{15}N_5O$ m/z: $[M+H]^+$ 282.1355; Found 282.1134.

Compound 5b. The compound was synthesized according to a modified procedure from the literature.[4] $Zn(ClO_4)_2 \cdot 6H_2O$ (3.40 mg, 0.009 mmol) was dissolved in a solution of compound **15** (12.6 mg, 0.04 mmol) in MeOH (2 mL) and THF (2 mL). After the mixture was stirred at room temperature for 4 h, the solvents were removed under reduced pressure to give a white powder (15.0 mg, 98%). ^{1} H NMR (400 MHz, DMSO-d_6) δ 8.65 (d, J = 5.0 Hz, 2H), 8.10 (t, J = 7.7 Hz, 2H), 7.75 – 7.57 (m, 6H), 7.39 (d, J = 8.1 Hz, 2H), 6.83 (dd, J = 17.6, 10.9 Hz, 2H), 5.94 (d, J = 17.6 Hz, 1H), 5.36 (d, J = 10.9 Hz, 1H), 4.29 (s, 2H), 3.85 – 3.64 (m, 4H).

Typical Synthesis of MINPs

To a micellar solution of **1** (9.3 mg, 0.02 mmol) in H_2O (2.0 mL), divinylbenzene (DVB, 1.4 μL, 0.01 mmol), the appropriate template–FM complex (**4a–f·5**) in DMSO (10 μL of 0.04 M solution, 0.0004 mmol), and AIBN in DMSO (10 μL of 8.2 mg/mL, 0.0005 mmol) were added. The mixture was subjected to ultrasonication for 10 min. Cross-linker **2** (4.1 mg, 0.024 mmol), $CuCl_2$ in H_2O (10 μL of 6.7 mg/mL, 0.0005 mmol), and sodium ascorbate in H_2O (10 μL of 99 mg/mL, 0.005 mmol) were added and the reaction mixture was stirred slowly at room temperature for 12 h. Compound **3** (10.6 mg, 0.04 mmol), $CuCl_2$ in H_2O (10 μL of a 6.7 mg/mL solution, 0.0005 mmol), and sodium ascorbate in H_2O (10 μL of a 99 mg/mL solution, 0.005 mmol) were added and the solution was stirred for another 6 h at room temperature. The reaction vial was sealed with a rubber stopper, and the reaction mixture was purged with nitrogen for 15 min before it was stirred at 75 °C for 16 h. The resulting MINP(**4a-f·5a**) solution was purged with nitrogen for 15 min before it was irradiated in a Rayonet reactor for 12 h to cleave the template at the *o*-nitrobenzyl ester site. The final solution was cooled to room temperature and poured into acetone (8.0 mL). The precipitate formed was washed three times with 1:4 water/acetone mixture (5.0 mL), seven times with methanol (5.0 mL), two times with acetone (5ml), and air-dried to give **MINP**(**4a-f·5a**)-COOH as an off-white powder. Typical yields were >75%.

Installation of the base center followed protocols reported previously.[7, 15] EDCI in dry DMF (10 μL of a 61 mg/mL solution, 0.004 mmol) was added to a stirred solution of **MINP**(**4a-f·5a**) (20.0 mg, 0.0004 mmol) in dry DMF (0.5 mL) at 0 °C under nitrogen. After 2 h, compound **8** (10 μL, 0.004 mmol) in dry DMF was added, and the mixture was stirred for 24 h at room temperature. The mixture was concentrated in vacuo and poured into 5 mL of acetone. The precipitate formed was collected by centrifugation and rinsed five times with 3 mL of acetone to afford **MINP**(**4a-f·5a**+**8**) as an off-white powder. Typical yields were >75%.

Procedures for Ester Hydrolysis

Hydrolysis of non-activated esters

Stock solutions (20 mM) of nonactivated esters in methanol were prepared. Stock solutions of the appropriate catalytic MINPs (100 μM) in 25 mM HEPES buffer were prepared. A typical procedure is as follows: An aliquot of 75 μL of the catalytic MINP stock solution and 15 μL of a nonactivated ester were combined with 410 μL of the same HEPES buffer in a vial. The concentration of the ester in the reaction mixture was 600 μM and the concentration of the catalyst 15 μM. The sealed vial was kept at 40.0 °C for 4 h, after which the reaction mixture was cooled to room temperature and extracted with diethyl ether (500 μL). The hydrolysis was monitored by the disappearance of the substrate (ester) by GC-MS analysis using an internal standard (*p*-xylene, 600 μM).

Kinetic Measurements for the hydrolysis of p-nitrophenyl acetate

A stock solution (10 mM) of *p*-nitrophenyl acetate (PNPA) in methanol was prepared. The stock solution was stored in a refrigerator and used within 3 d. Stock solutions of the appropriate catalytic

MINPs (100 μM) in 25 mM HEPES buffer (pH 7.0) were prepared. For the kinetic experiment, a typical procedure is as follows: An aliquot of 300 μL of the catalytic MINPP stock solution was combined with 1690 μL of the same HEPES buffer in a cuvette. The cuvette was placed in a UV-vis spectrometer and equilibrated to 40.0 °C. After 5 min, an aliquot of 10 μL of the PNPH stock solution was added. The concentration of the substrate (PNPH) in the reaction mixture was 50 μM and the concentration of the NP catalyst was 15 μM in all cases. The hydrolysis was monitored by the absorbance of *p*-nitrophenoxide at 400 nm. The amount of the product formed was calculated from a calibration curve obtained from *p*-nitrophenol at pH 7.0.

Solvent isotope effect

Table S1. Pseudo-first-order rate constants and solvent kinetic isotope effects of the hydrolysis of PNPA catalyzed by MINP(**4a**·**5a**+**8**) at 40 °C.[a]

entry	pH or PD	system	k ($\times 10^{-4}$ s^{-1})	k_{H2O}/k_{D2O}
1	7	H_2O	21.8 ± 2.8	
				1.23
2	7	D_2O	17.1 ± 1.4	

[a] Reaction rates were measured by monitoring the formation of *p*-nitrophenolate at 400 nm. [catalyst] = 15 μM. [PNPA] = 50 μM. The pD values were determined by adding 0.4 to the pH meter reading.

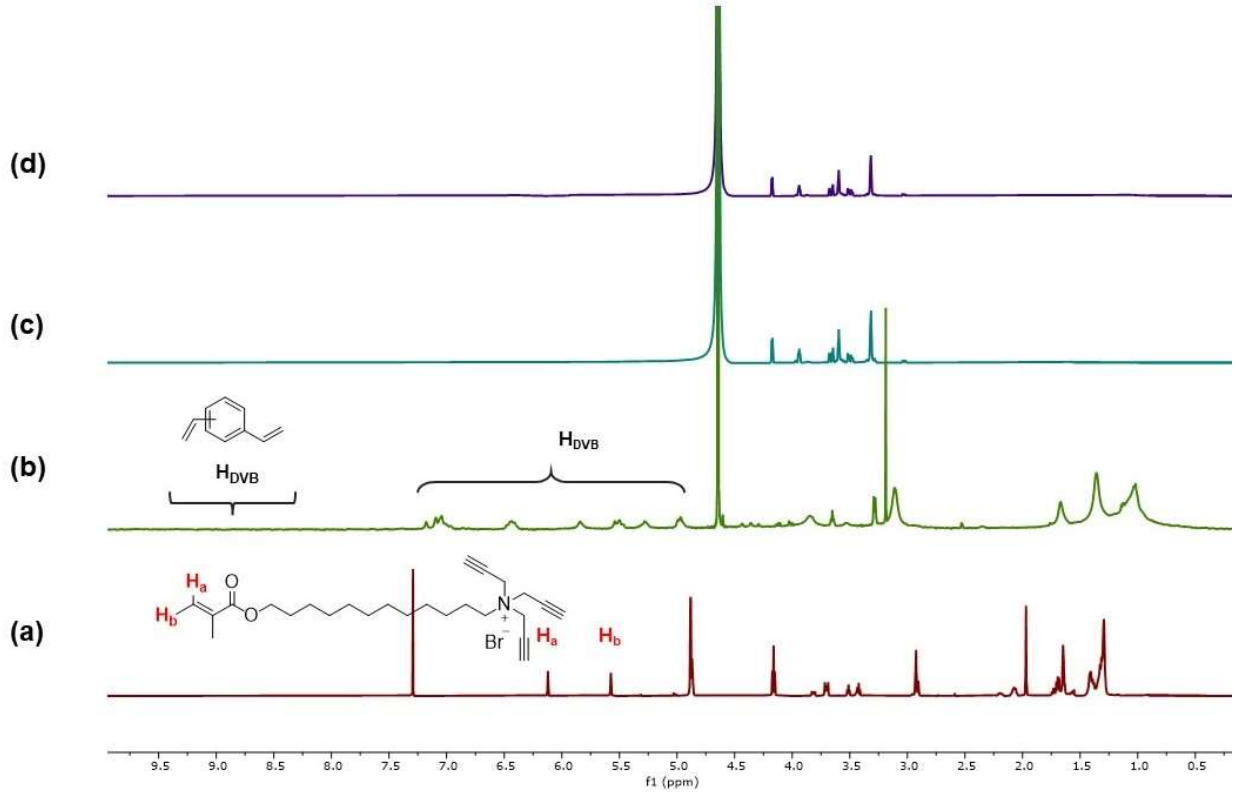


Figure S1. Stacked ^{1}H NMR spectra of (a) surfactant **1** in $CDCl_3$, (b) Typical alkynyl surface cross-linked micelle in D_2O, (c) Typical surface-functionalized micelle in D_2O, (d) Typical MINP after washing and re-dissolving in D_2O.

Dynamic Light Scattering

Particle size of MINP was determined on a Malvern Zetasizer Nano ZS using the Zetasizer software according to the Stokes-Einstein equation (1). The volume of a spherical nanoparticle (V_{D_h}) was calculated from equation (2). Assuming a density of 1.37 g/cm^3 (the density of protein), the molecular weight of the particle can be calculated using equation (3).[16] A nanoparticle with hydrodynamic diameter of 4.87 nm has a calculated molecular weight of 50 kDa.

$$D_h = \frac{k_B T}{6\pi\eta D_t} \quad (1)$$

in which D_h is the hydrodynamic diameter, D_t the translational diffusion coefficient measured by dynamic light scattering, T the temperature, k_B the Boltzmann's constant, and η is dynamic viscosity of water (0.890 cP at 298 K).

$$V_{D_h} = \frac{4\pi}{3}\left(\frac{D_h}{2}\right)^3 \quad (2)$$

$$\text{Mw in dalton} = \left(\frac{D_h}{0.132}\right)^3 \quad (3)$$

in which D_h is the hydrodynamic diameter in nm.

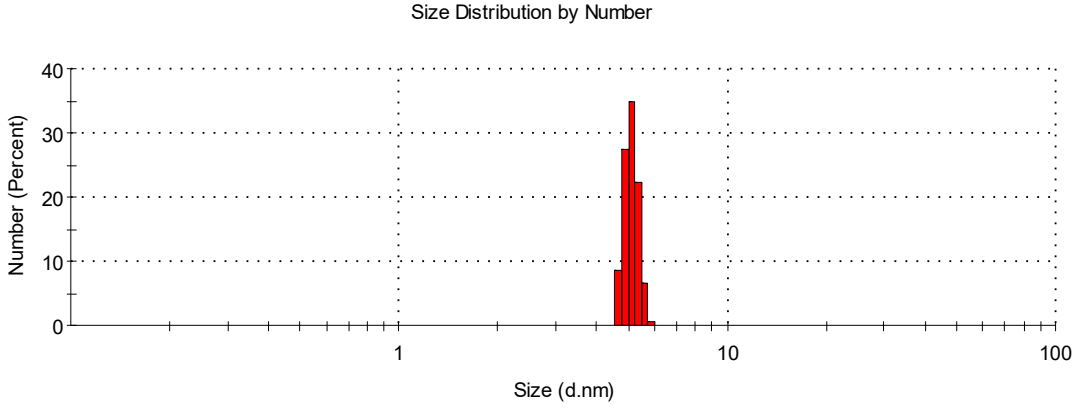


Figure S2. Distribution of the hydrodynamic diameters of the nanoparticles in water as determined by DLS for typical alkynyl-SCM (surface-cross-linked micelle). D = 5.14 ± 0.19 nm.

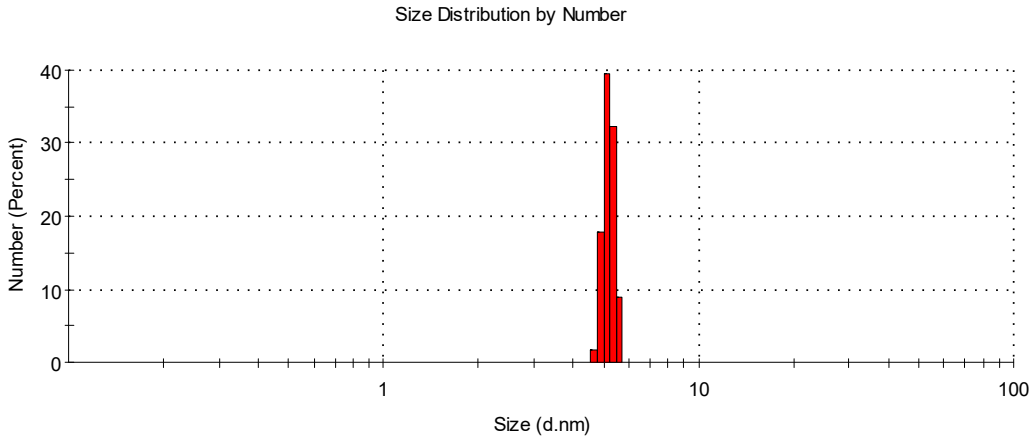


Figure S3. Distribution of the hydrodynamic diameters of the nanoparticles in water as determined by DLS for typical surface-functionalized SCM (surface-cross-linked micelle). D = 5.20 ± 0.13 nm.

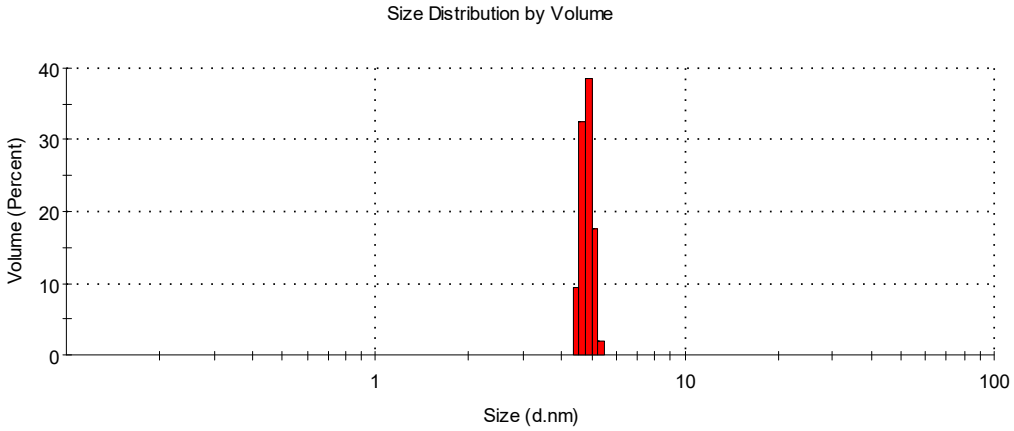


Figure S4. Distribution of the hydrodynamic diameters of the nanoparticles in water as determined by DLS for typical MINP. D = 4.83 ± 0.13 nm.

Transmission electron microscopy

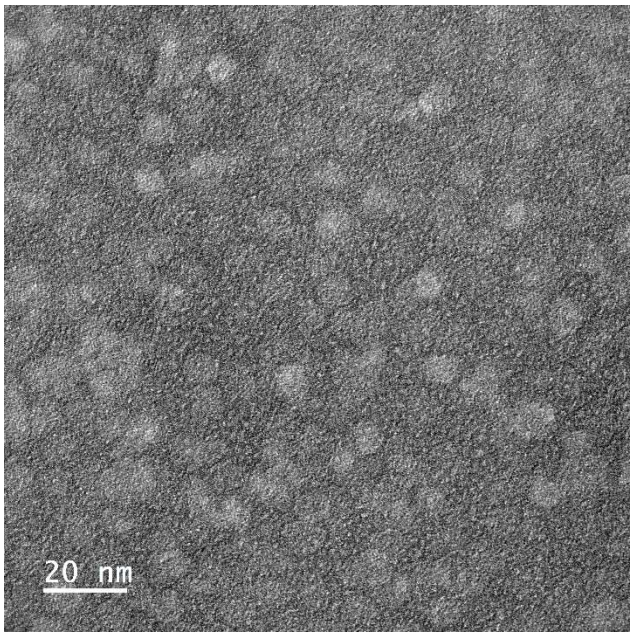
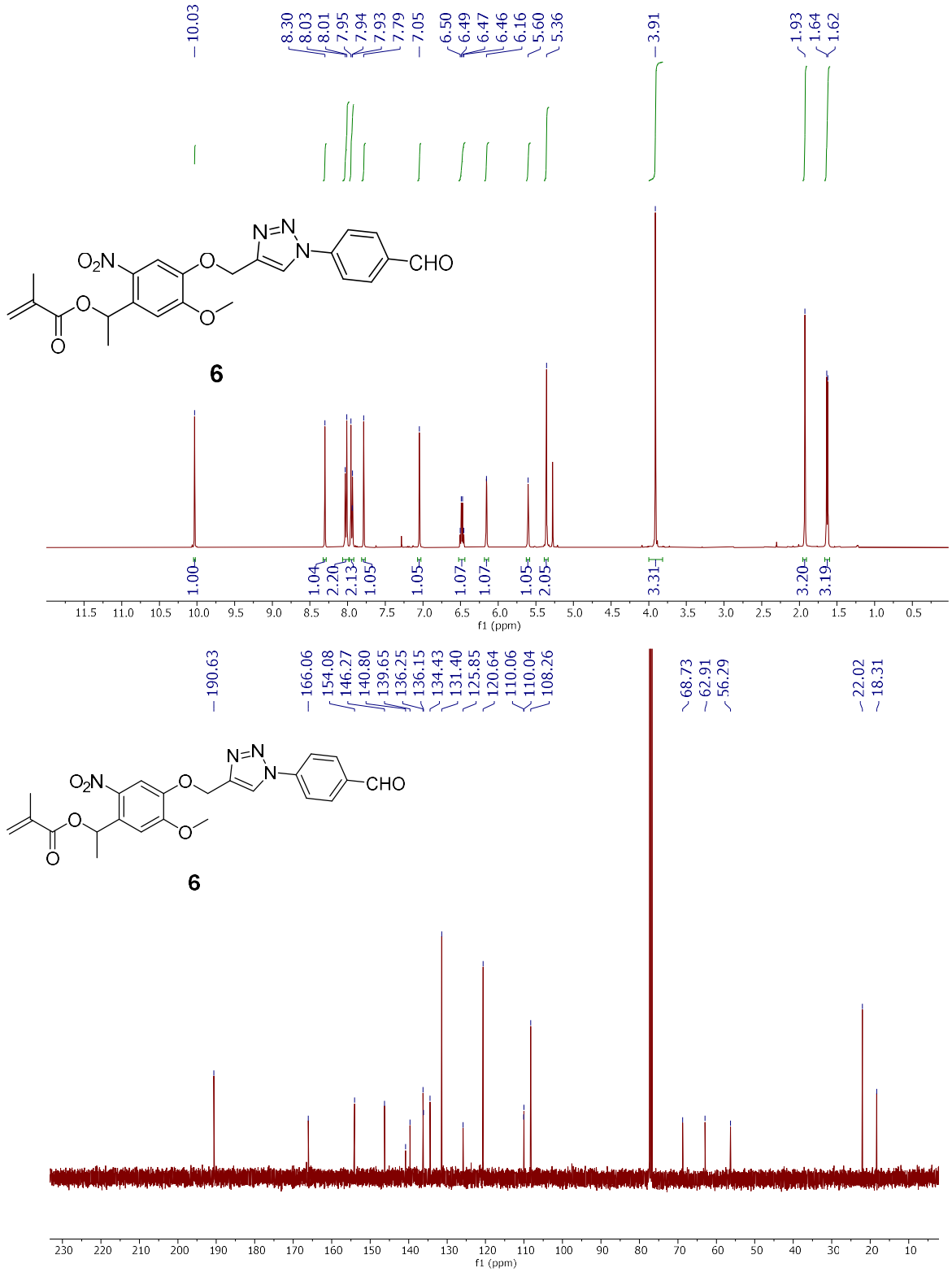
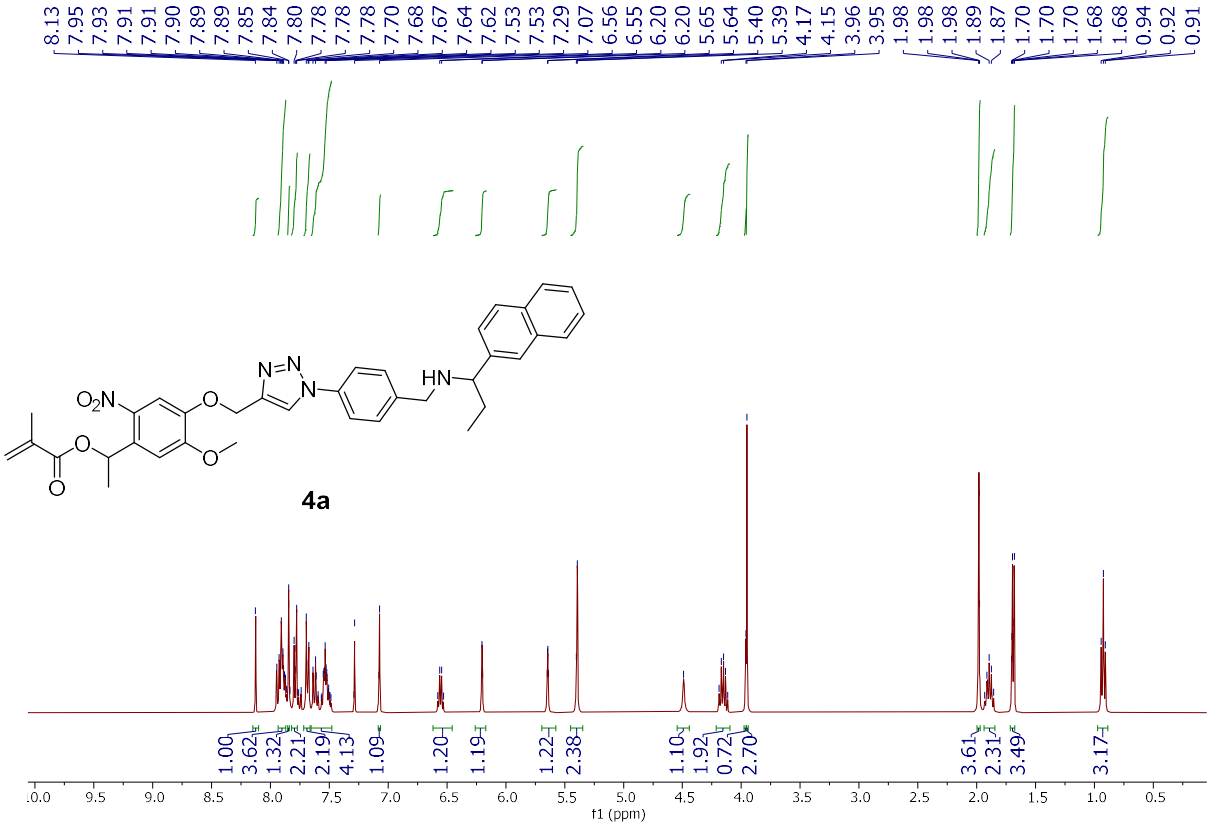


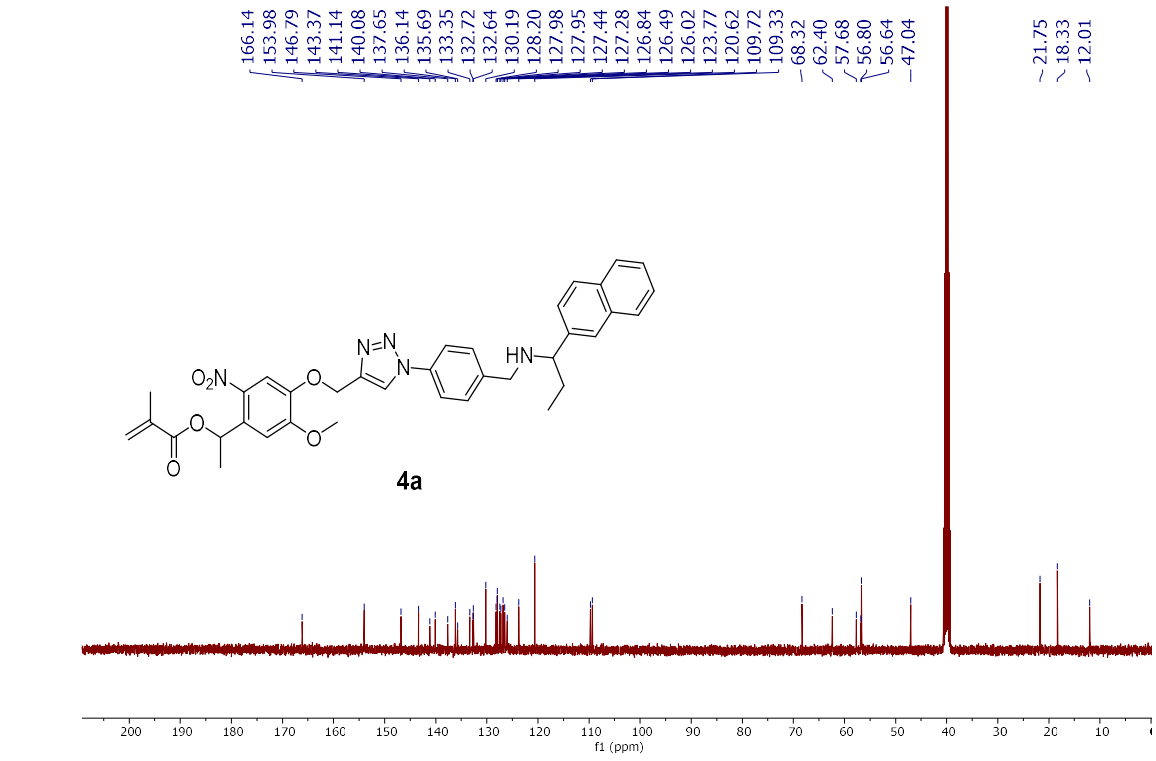
Figure S5. TEM image of typical MINPs (scale bar = 50 nm).

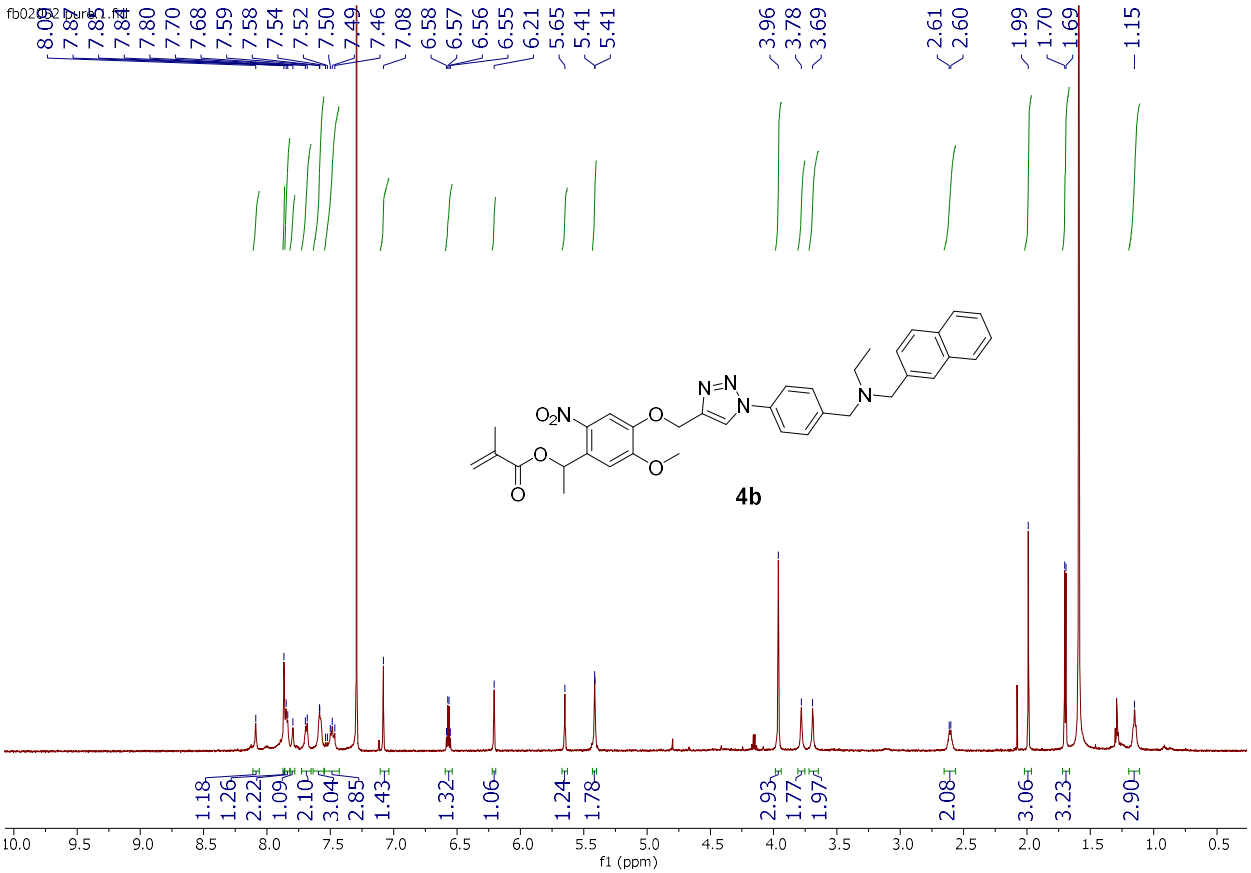
For the TEM imaging, 0.1 mg of MINP was dissolved in 1 mL of Millipore water and the solution was ultra-sonicated for 10 min. A micro syringe was used to load one small drop (~1 μL) of the above solution onto a TEM copper grid covered with carbon film. The sample was left to form a thin layer, and then one small drop (~1 μL) of 2% uranyl acetate solution was loaded on the grid for the negative staining. The sample was left to dry and analyzed on a 200kV JEOL 2100 scanning/transmission electron microscope (STEM).

NMR spectra of key compounds




8.13 7.95 7.93 7.91 7.91 7.90 7.89 7.89 7.85 7.84 7.80 7.78 7.78 7.78 7.70 7.68 7.67 7.64 7.62 7.53 7.53 7.29 7.07 6.56 6.55 6.20 6.20 5.65 5.64 5.40 5.39 4.17 4.15 3.96 3.95 1.98 1.98 1.98 1.89 1.87 1.70 1.70 1.70 1.68 1.68 0.94 0.92 0.91
4a
1.00 3.62 1.32 2.21 2.19 4.13 1.09 1.20 1.19 1.22 2.38 1.10 1.92 0.72 2.70 3.61 2.31 3.49 3.17
f1 (ppm)


166.14 153.98 146.79 143.37 141.14 140.08 137.65 136.14 135.69 133.35 132.72 132.64 130.19 128.20 127.98 127.95 127.44 127.28 126.84 126.49 126.02 123.77 120.62 109.72 109.33 68.32 62.40 57.68 56.80 56.64 47.04 21.75 18.33 12.01
4a
f1 (ppm)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7.84
7.80
7.70
7.68
7.59
7.58
7.54
7.52
7.50
7.49
7.46
7.08
6.58
6.57
6.56
6.55
6.21
5.65
5.41
5.41
3.96
3.78
3.69
2.61
2.60
1.99
1.70
1.69
1.15
4b
1.18
1.26
2.22
1.09
2.10
3.04
2.85
1.43
1.32
1.06
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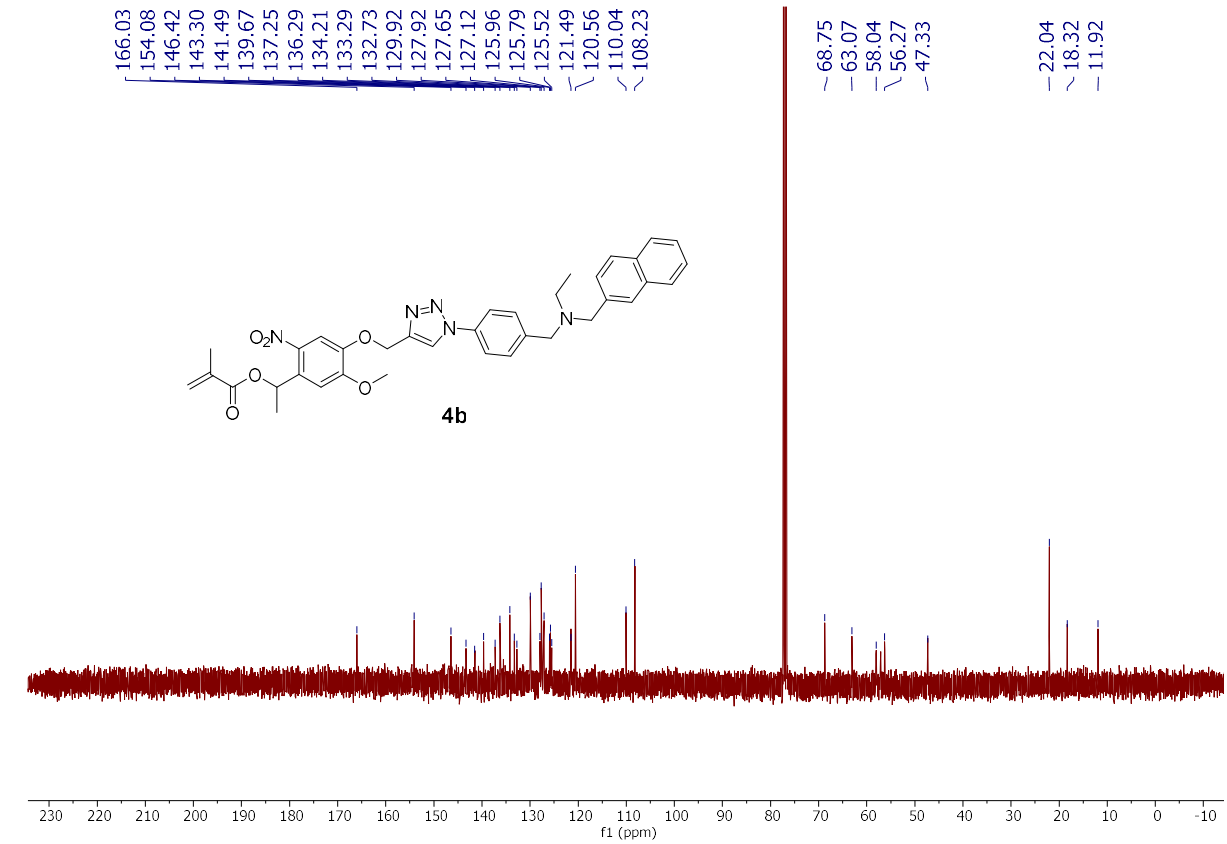
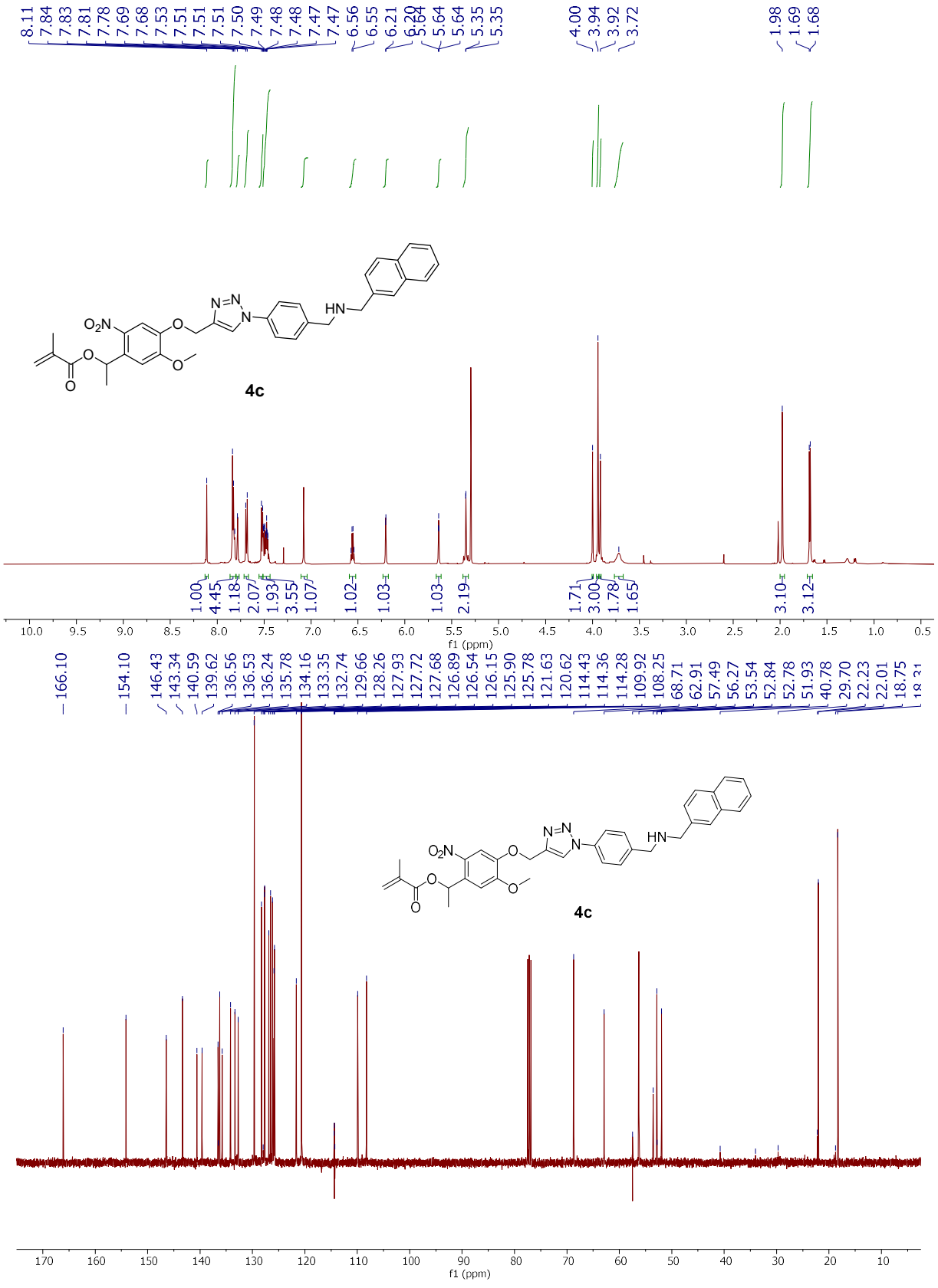
1.78
2.93
1.77
1.97
2.08
3.06
3.23
2.90
f1 (ppm)


166.03
154.08
146.42
143.30
141.49
139.67
137.25
136.29
134.21
133.29
132.73
129.92
127.92
127.65
127.12
125.96
125.79
125.52
121.49
120.56
110.04
108.23
68.75
63.07
58.04
56.27
47.33
22.04
18.32
11.92
4b
f1 (ppm)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7.83
7.81
7.78
7.69
7.68
7.53
7.51
7.51
7.51
7.50
7.49
7.48
7.48
7.47
7.47
6.56
6.55
6.21
6.20
5.64
5.64
5.64
5.35
5.35
4.00
3.94
3.92
3.72
1.98
1.69
1.68
4c
1.00
4.45
1.18
2.07
1.93
3.55
1.07
1.02
1.03
1.03
2.19
1.71
3.00
1.78
1.65
3.10
3.12
f1 (ppm)
166.10
154.10
146.43
143.34
140.59
139.62
136.56
136.53
136.24
135.78
134.16
133.35
132.74
129.66
128.26
127.93
127.72
127.68
126.89
126.54
126.15
125.90
125.78
121.63
120.62
114.43
114.36
114.28
109.92
108.25
68.71
62.91
57.49
56.27
53.54
52.84
52.78
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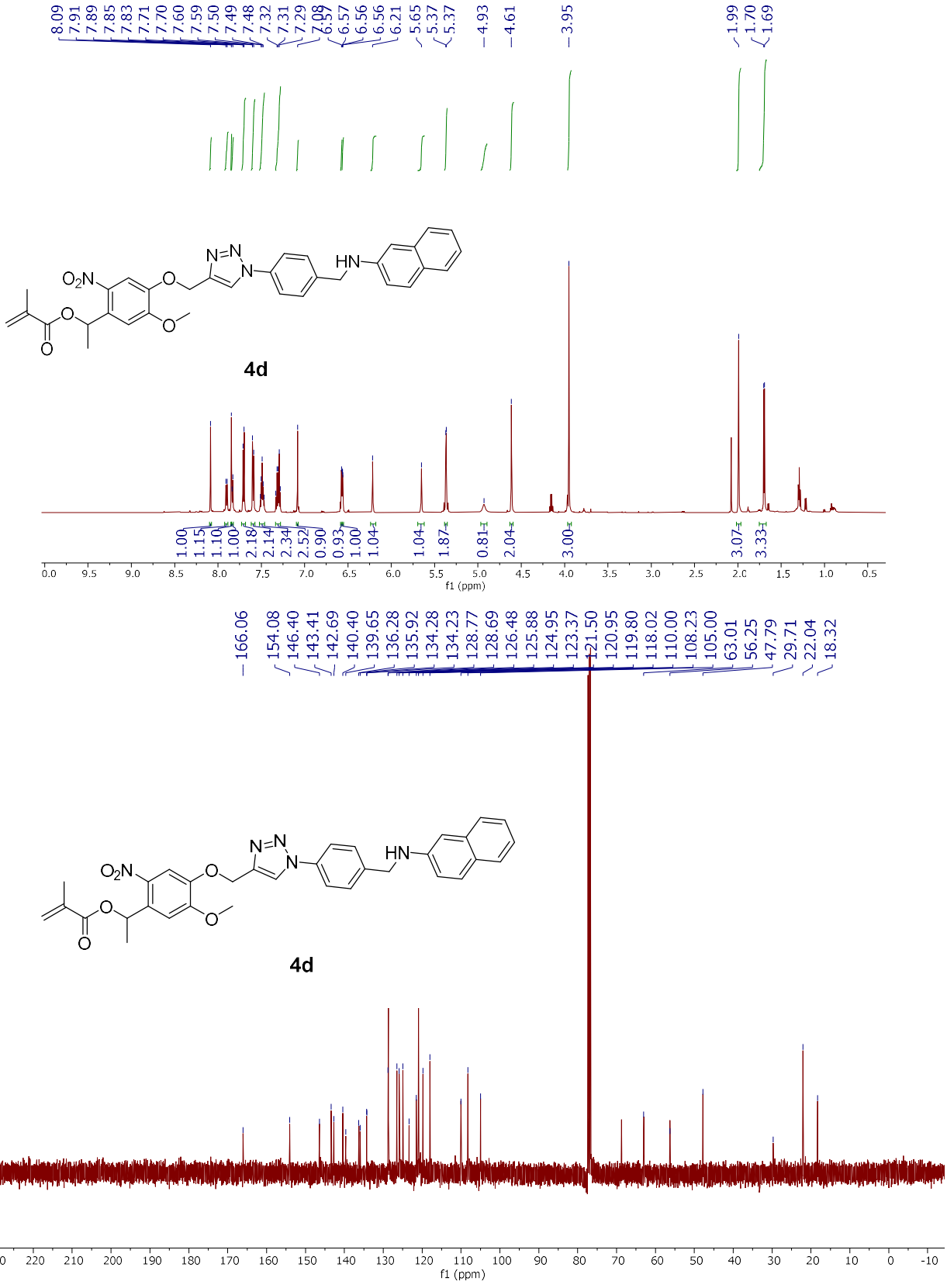
40.78
29.70
22.23
22.01
18.75
4c
f1 (ppm)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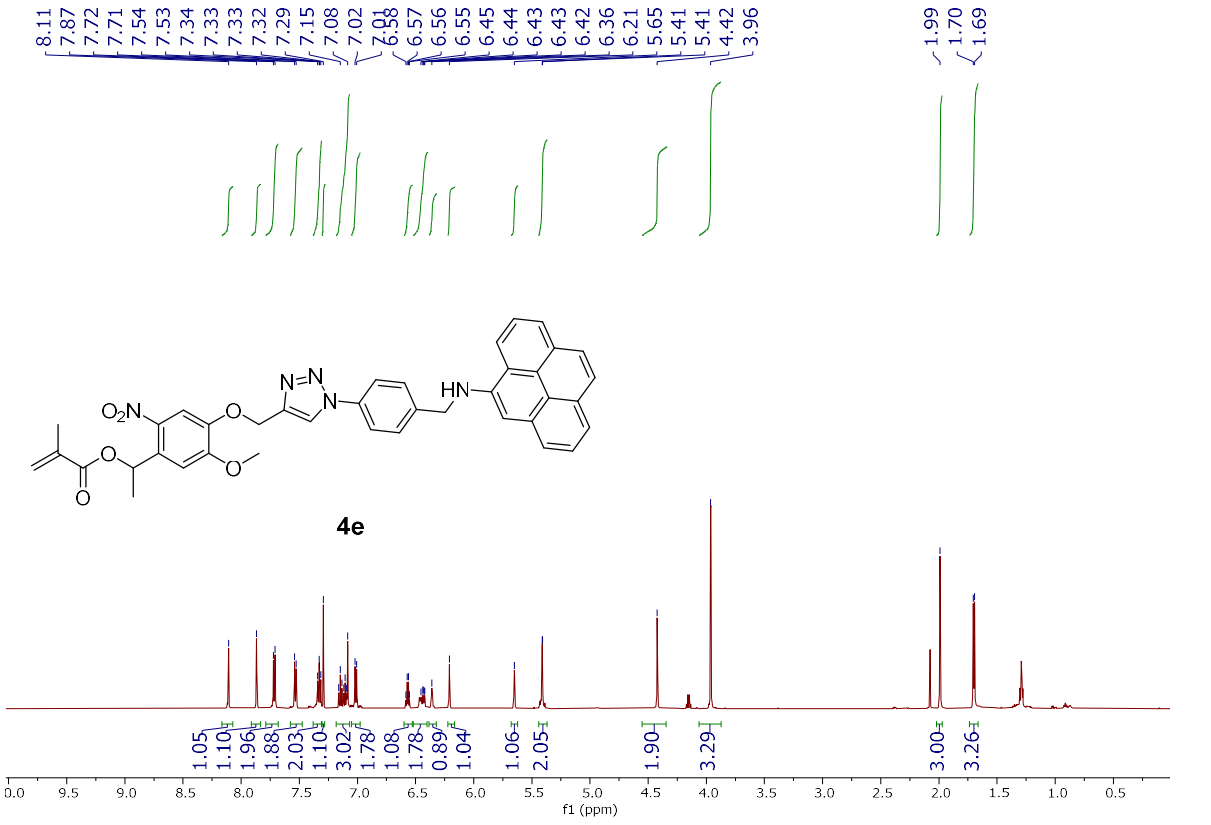
7.91
7.89
7.85
7.83
7.71
7.70
7.60
7.59
7.50
7.49
7.48
7.32
7.31
7.29
7.08
6.57
6.56
6.56
6.21
5.65
5.37
5.37
4.93
4.61
3.95
1.99
1.70
1.69
4d
1.00
1.15
1.10
1.00
2.18
2.14
2.34
2.52
0.90
0.93
1.00
1.04
1.04
1.87
0.81
2.04
3.00
3.07
3.33
f1 (ppm)
166.06
154.08
146.40
143.41
142.69
140.40
139.65
136.28
135.92
134.28
134.23
128.77
128.69
126.48
125.88
124.95
123.37
121.50
120.95
119.80
118.02
110.00
108.23
105.00
63.01
56.25
47.79
29.71
22.04
18.32
4d
f1 (ppm)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7.87
7.72
7.71
7.54
7.53
7.34
7.33
7.33
7.32
7.29
7.15
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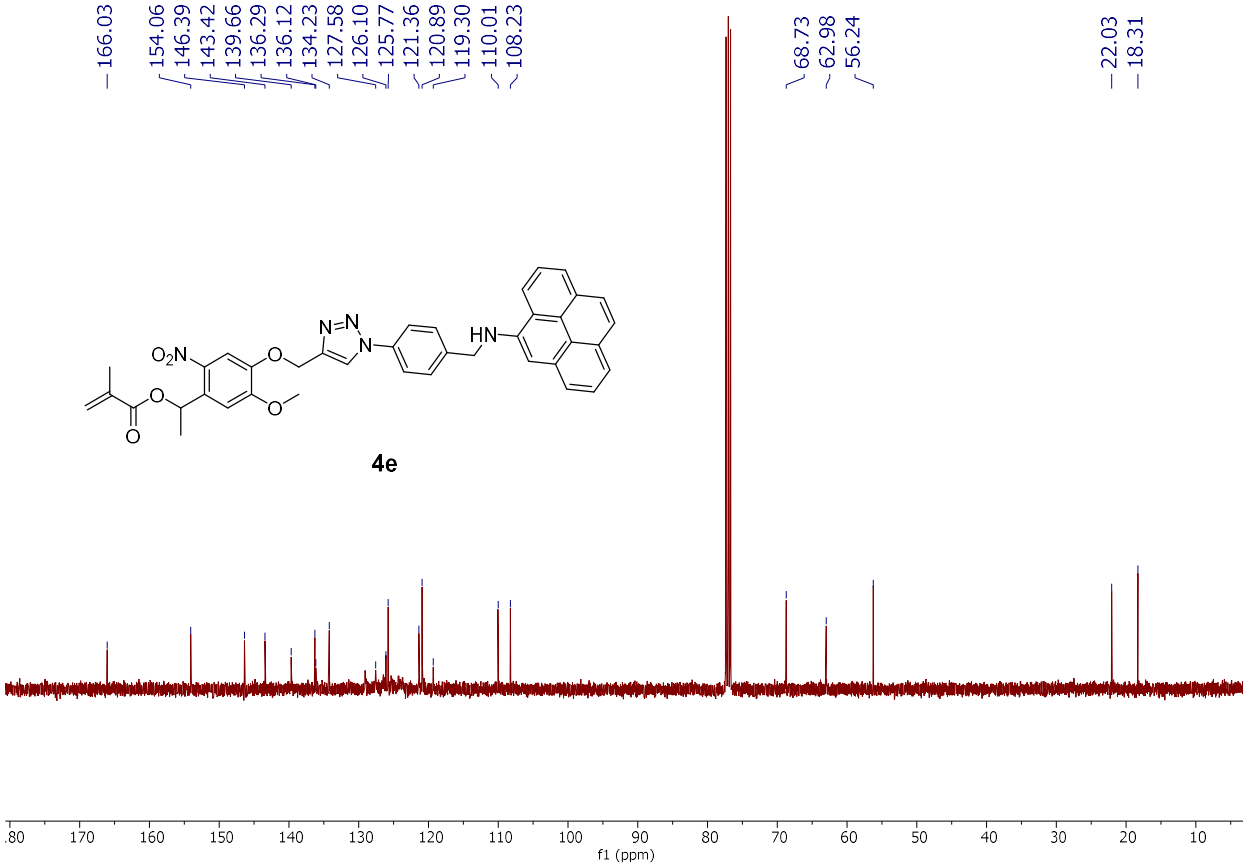
7.02
7.01
6.58
6.57
6.56
6.55
6.45
6.44
6.43
6.43
6.42
6.36
6.21
5.65
5.41
5.41
4.42
3.96
1.99
1.70
1.69
O2N
N=N
HN
O
O
O
O
4e
1.05
1.10
1.96
1.88
2.03
1.10
3.02
1.78
1.08
1.78
0.89
1.04
1.06
2.05
1.90
3.29
3.00
3.26
f1 (ppm)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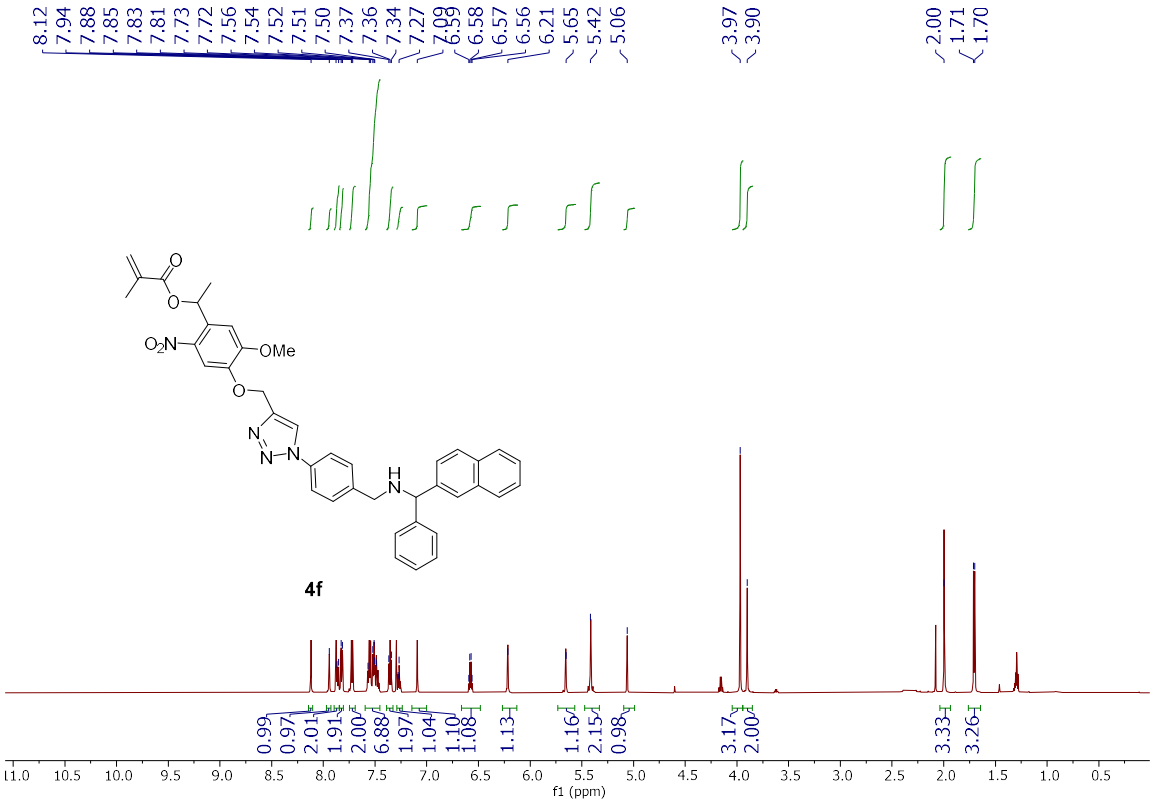
143.42
139.66
136.29
136.12
134.23
127.58
126.10
125.77
121.36
120.89
119.30
110.01
108.23
68.73
62.98
56.24
22.03
18.31
O2N
N=N
HN
O
O
O
O
4e
f1 (ppm)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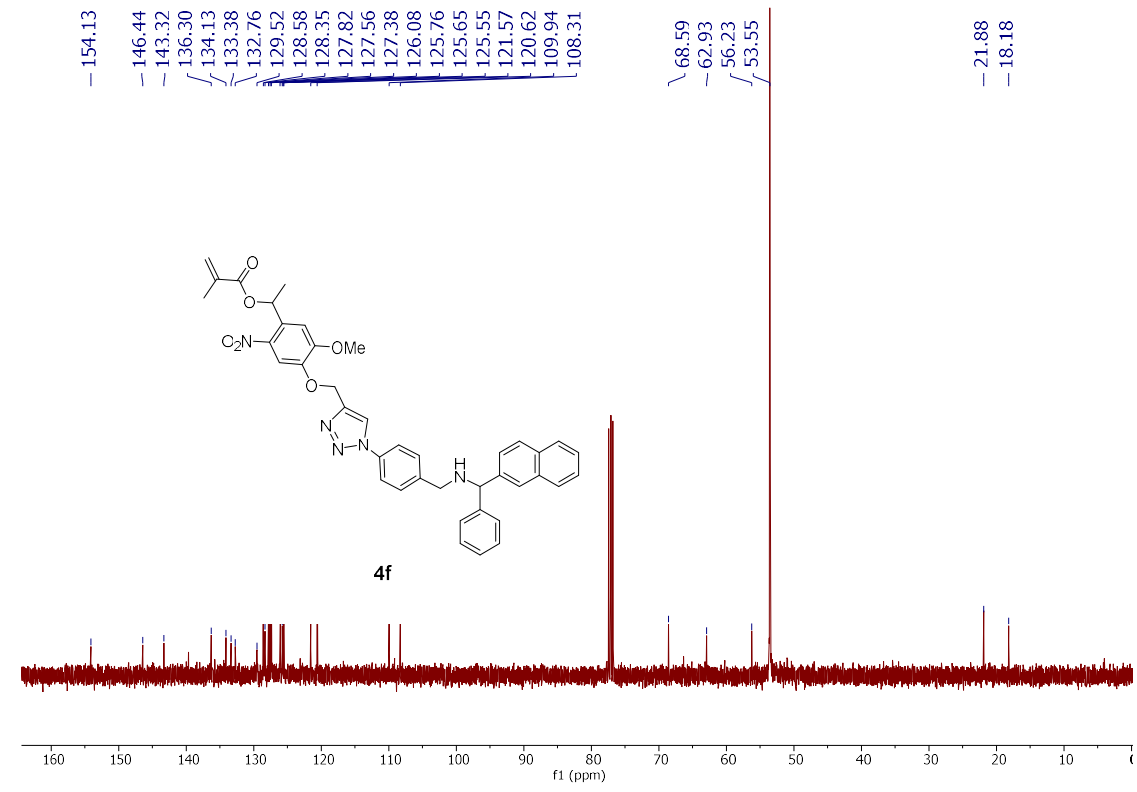
7.37
7.36
7.34
7.27
7.09
6.59
6.58
6.57
6.56
6.21
5.65
5.42
5.06
3.97
3.90
2.00
1.71
1.70
OMe
4f
0.99
0.97
2.01
1.91
2.00
6.88
1.97
1.04
1.10
1.08
1.13
1.16
2.15
0.98
3.17
2.00
3.33
3.26
f1 (ppm)


154.13
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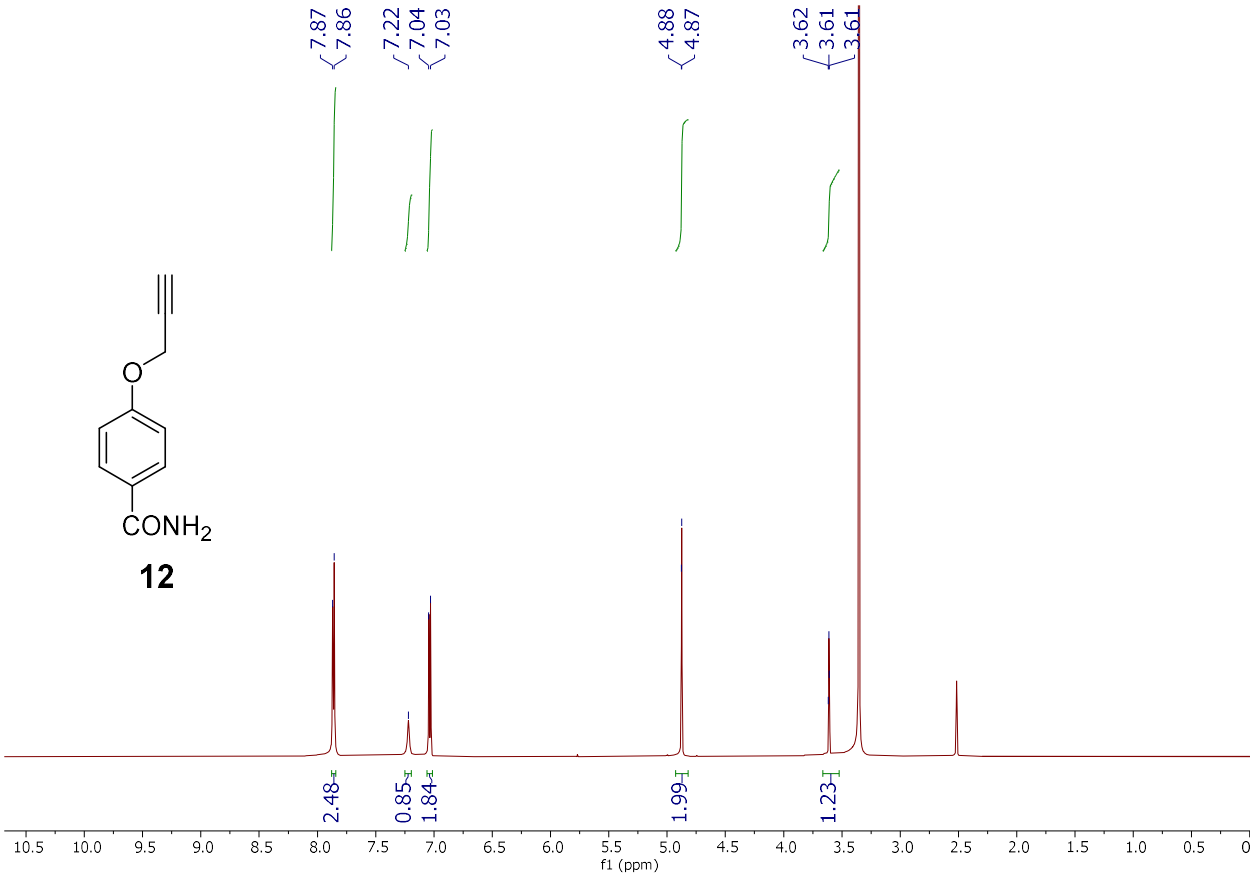
133.38
132.76
129.52
128.58
128.35
127.82
127.56
127.38
126.08
125.76
125.65
125.55
121.57
120.62
109.94
108.31
68.59
62.93
56.23
53.55
21.88
18.18
OMe
4f
f1 (ppm)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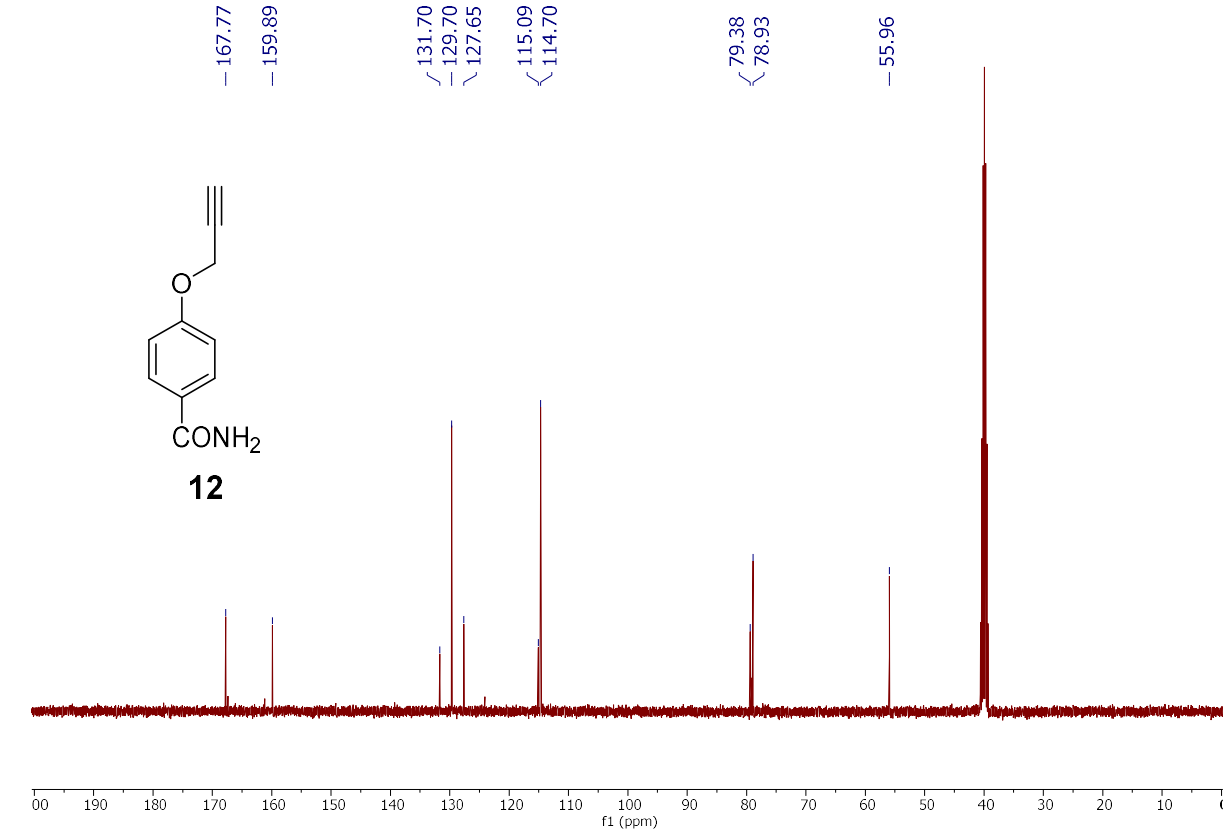
7.86
7.22
7.04
7.03
4.88
4.87
3.62
3.61
3.61
O
CONH2
12
2.48
0.85
1.84
1.99
1.23
f1 (ppm)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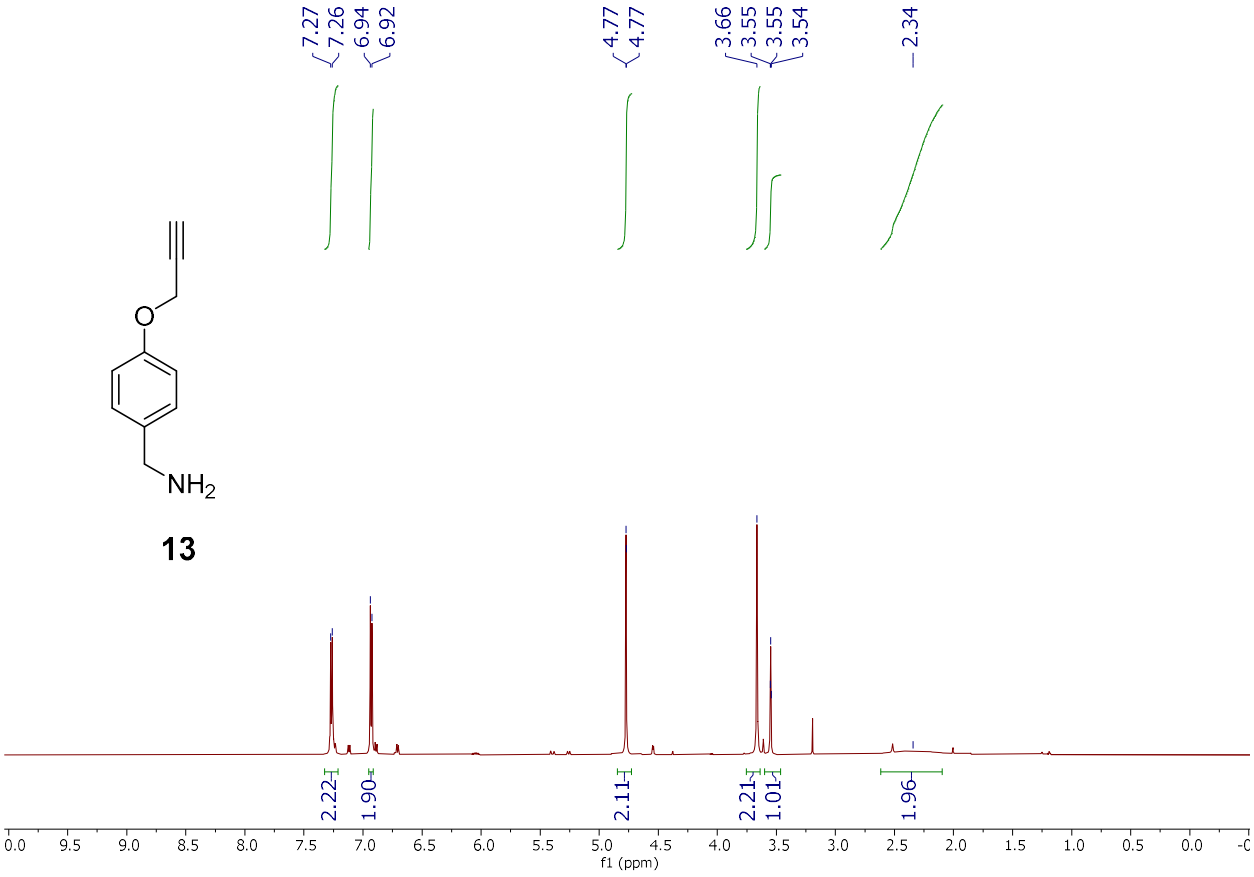
127.65
115.09
114.70
79.38
78.93
55.96
O
CONH2
12
f1 (ppm)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4.77
3.66
3.55
3.55
3.54
2.34
O
NH2
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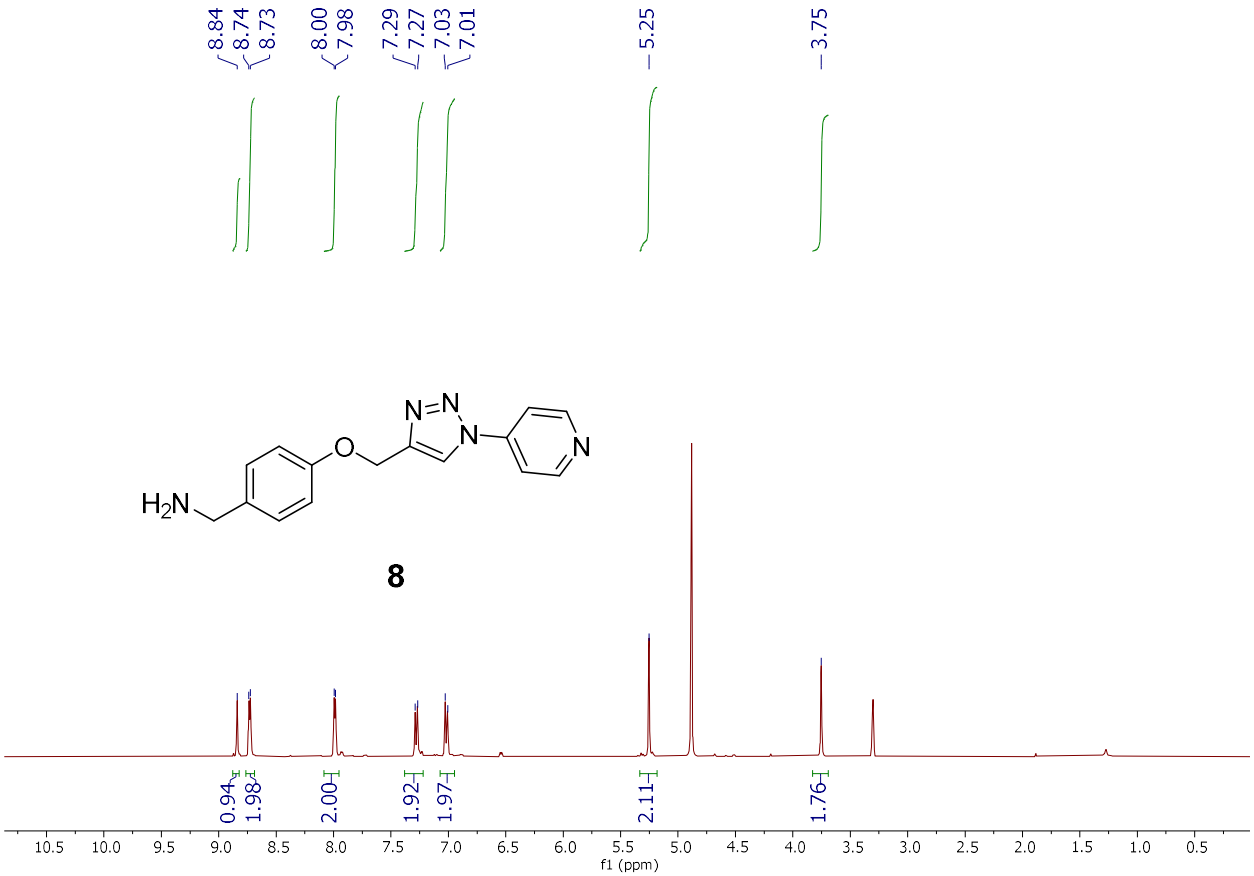
2.22
1.90
2.11
2.21
1.01
1.96
f1 (ppm)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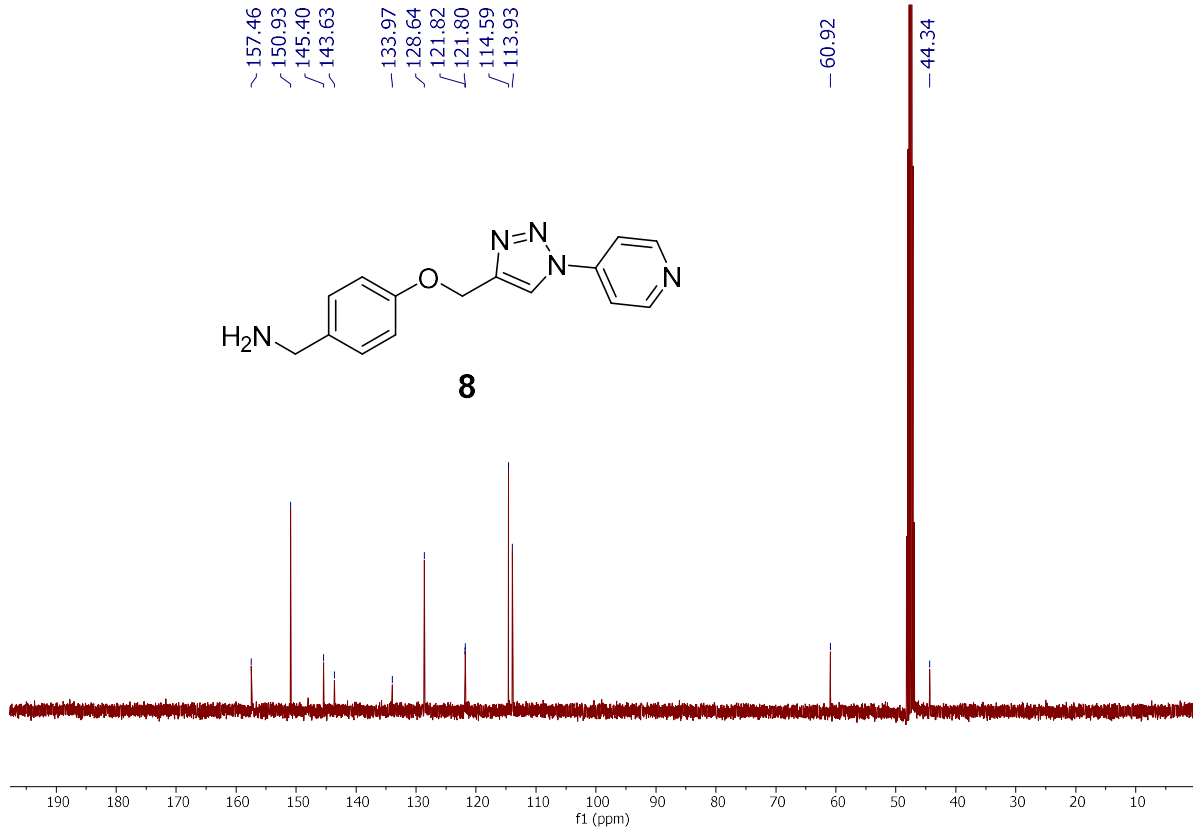
8.73
8.00
7.98
7.29
7.27
7.03
7.01
5.25
3.75
N=N
N
N
O
H2N
8
0.94
1.98
2.00
1.92
1.97
2.11
1.76
f1 (ppm)


157.46
150.93
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133.97
128.64
121.82
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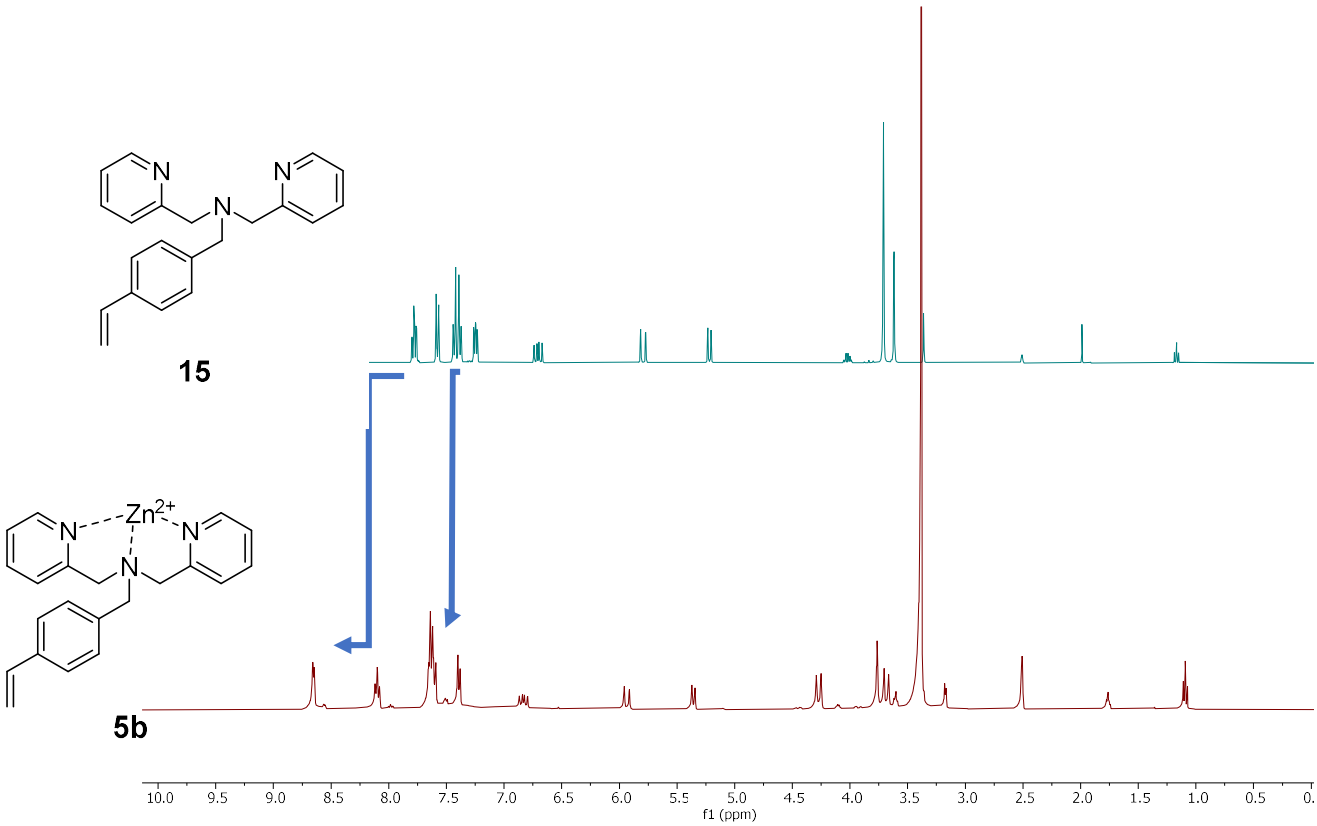
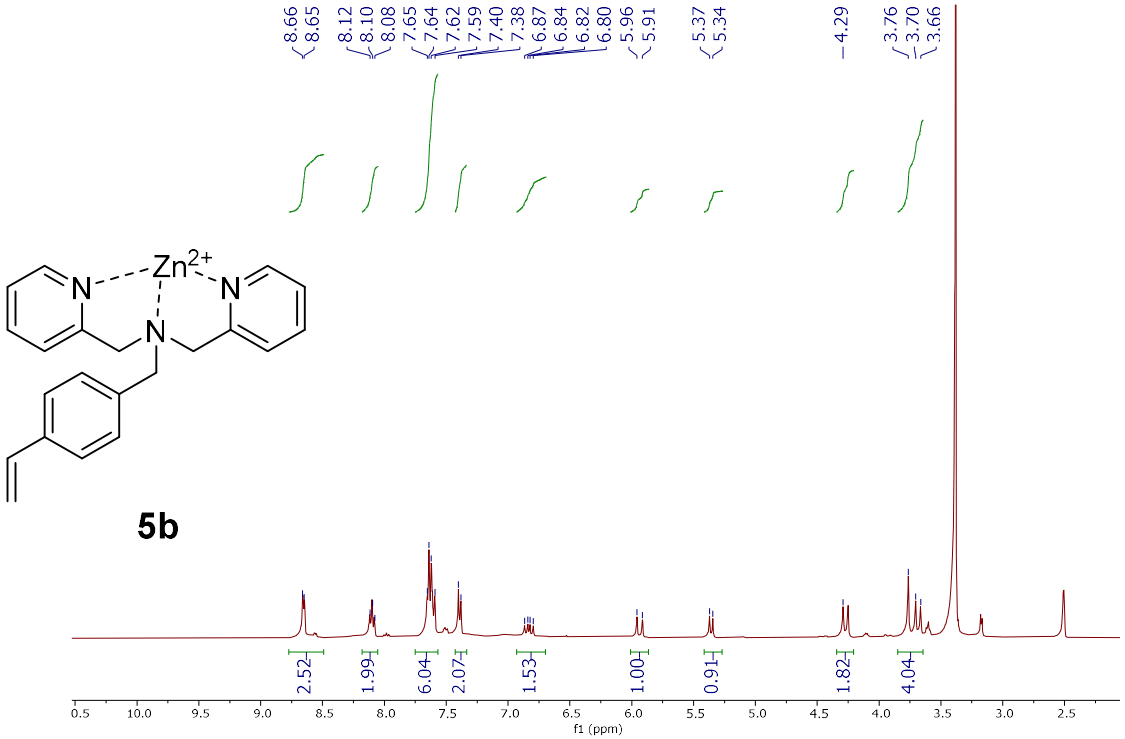
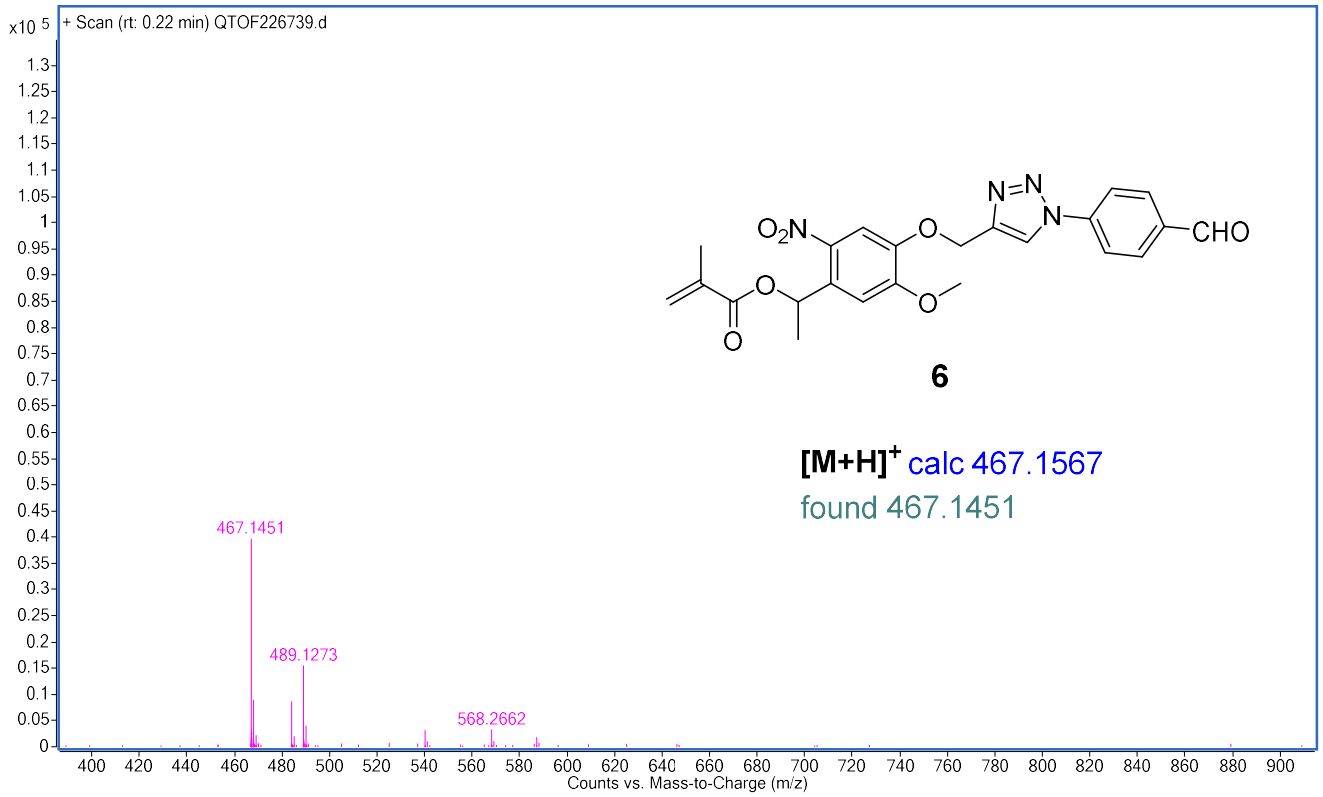
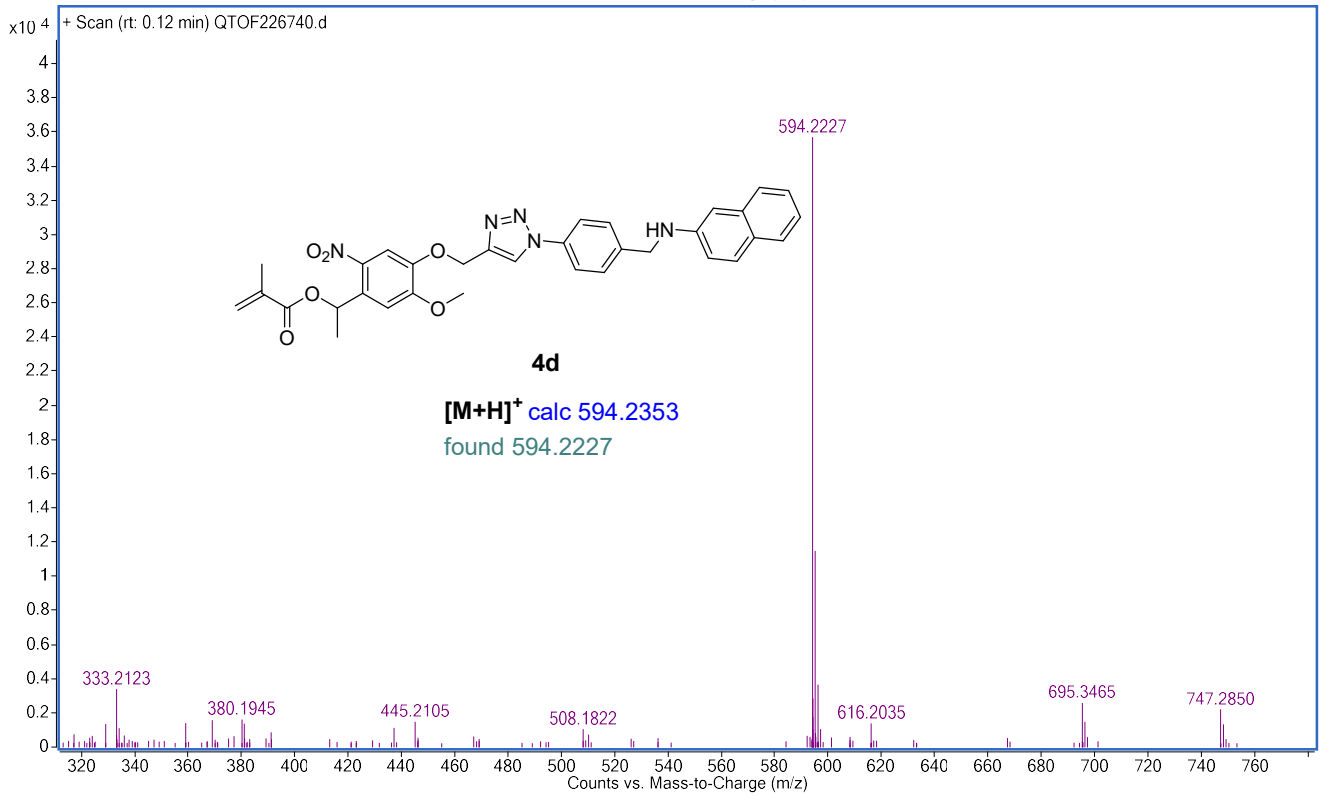
114.59
113.93
60.92
44.34
N=N
N
N
O
H2N
8
f1 (ppm)

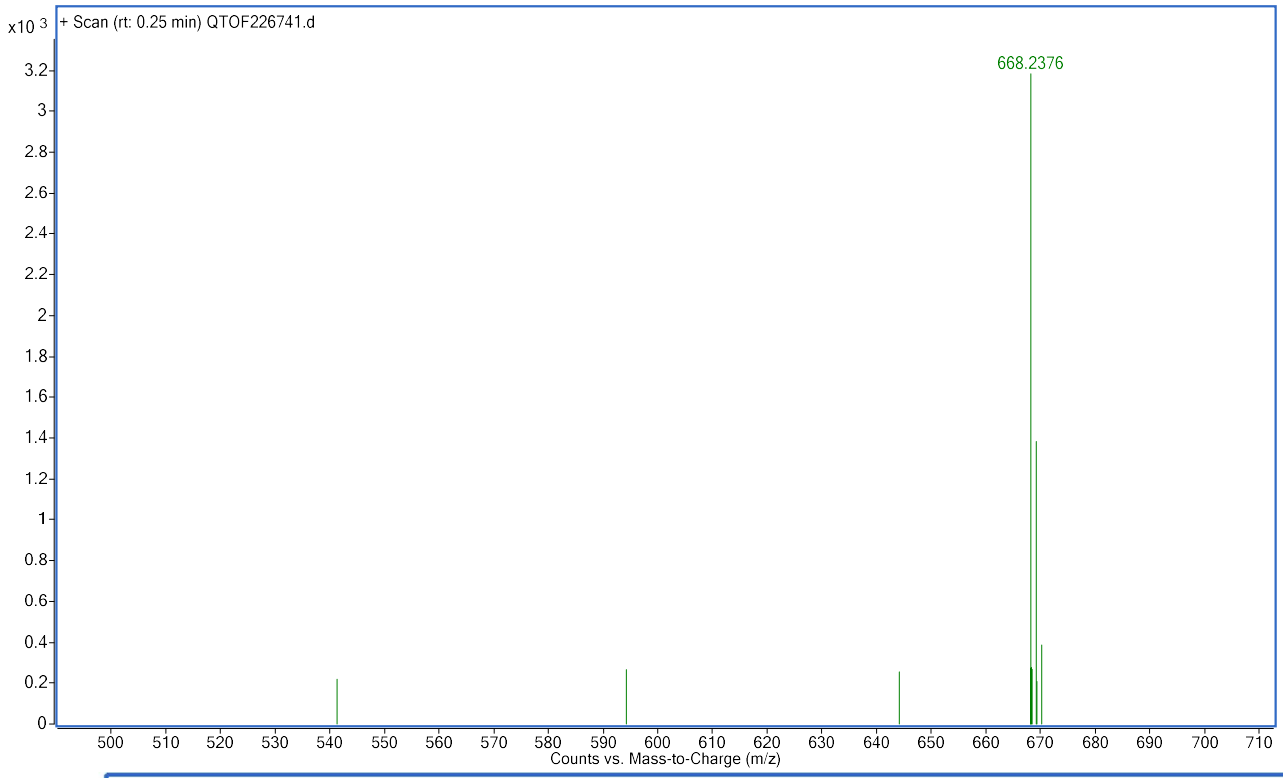


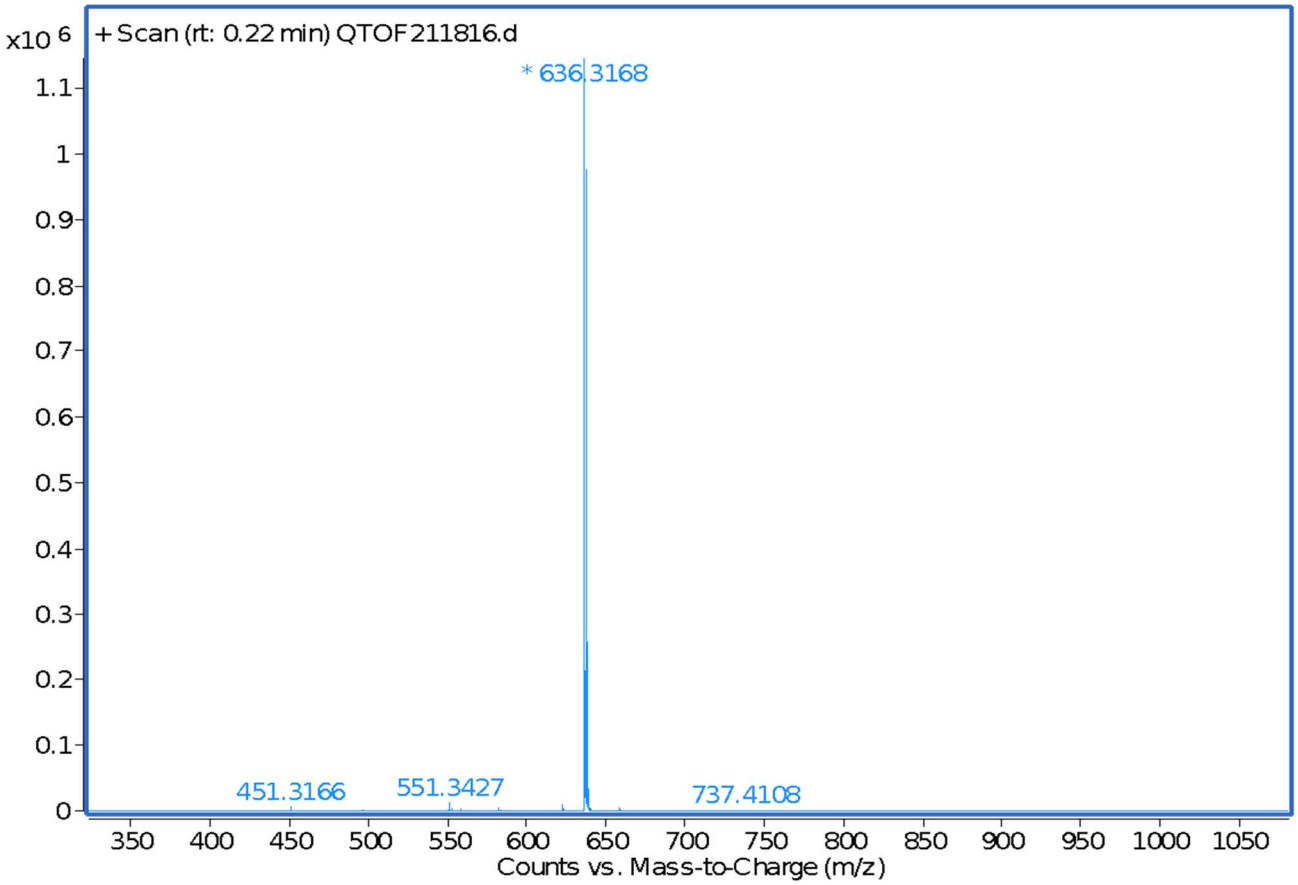


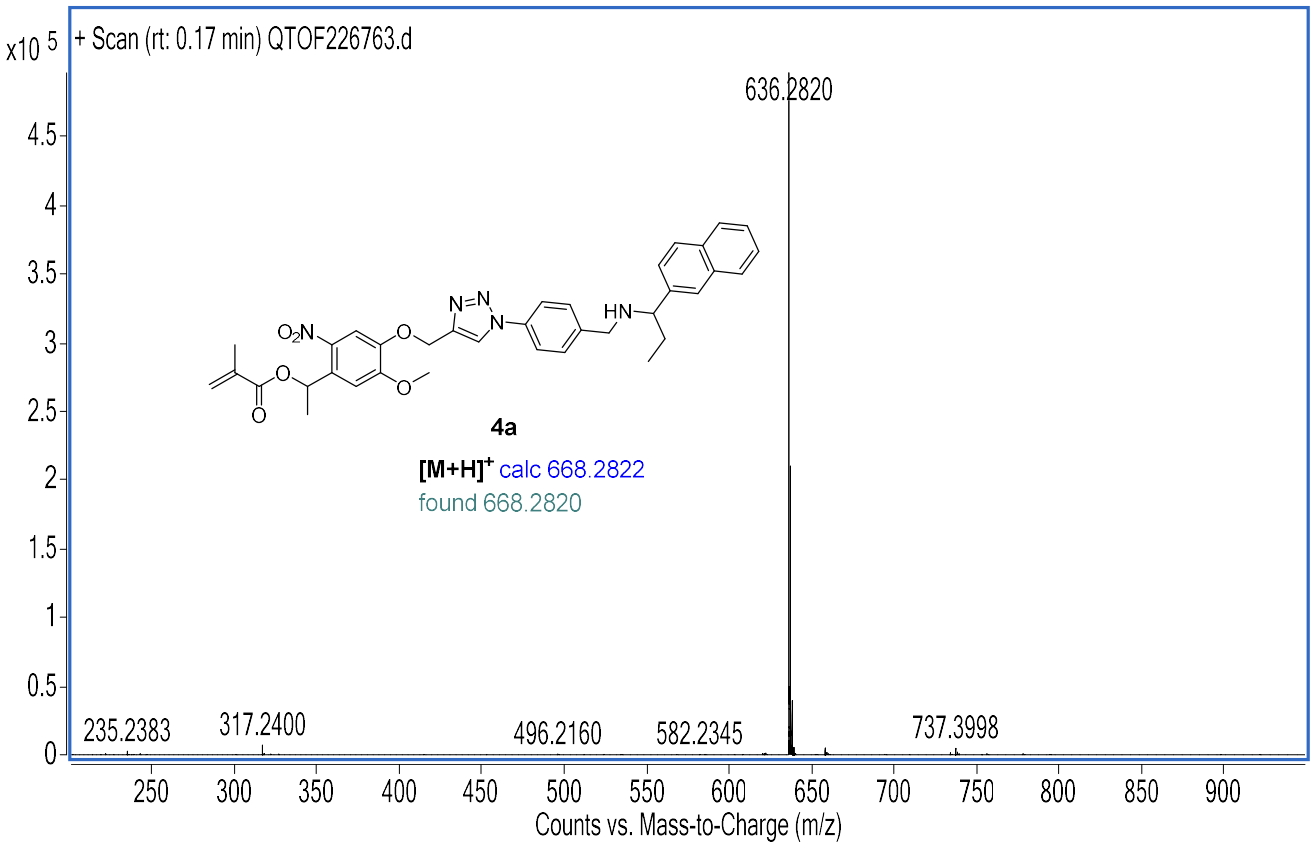
ESI-MS Spectra for New Compounds

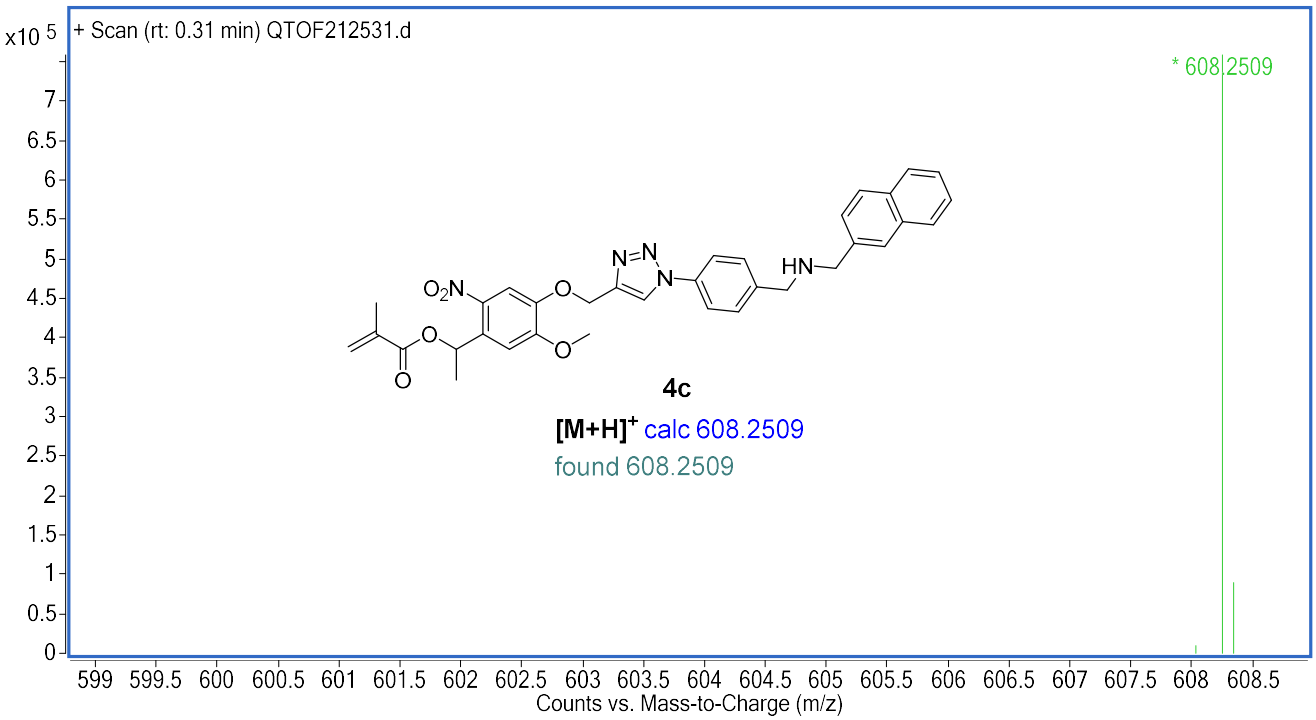

x10 5
+ Scan (rt: 0.22 min) QTOF226739.d
467.1451
489.1273
568.2662
Counts vs. Mass-to-Charge (m/z)
6
[M+H]+ calc 467.1567
found 467.1451

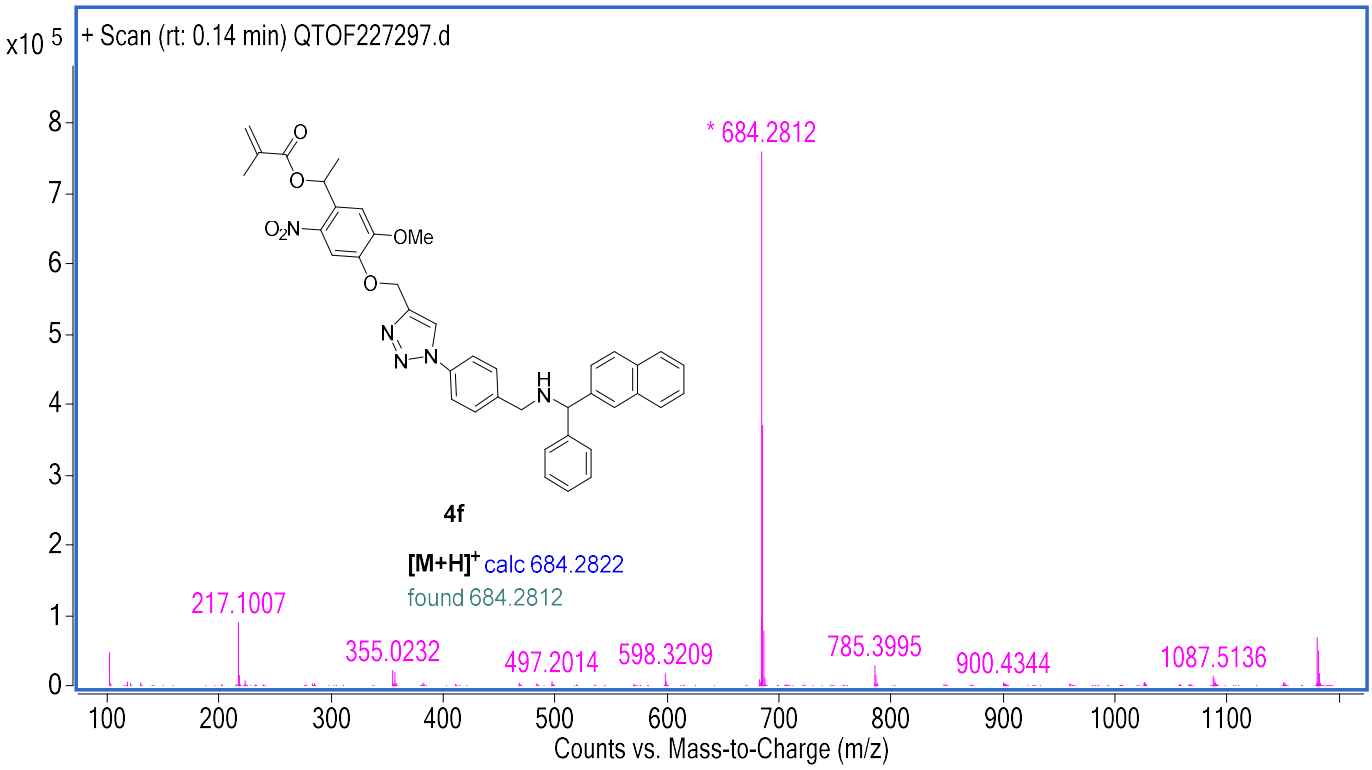

x10 4
+ Scan (rt: 0.12 min) QTOF226740.d
594.2227
333.2123
380.1945
445.2105
508.1822
616.2035
695.3465
747.2850
Counts vs. Mass-to-Charge (m/z)
4d
[M+H]+ calc 594.2353
found 594.2227

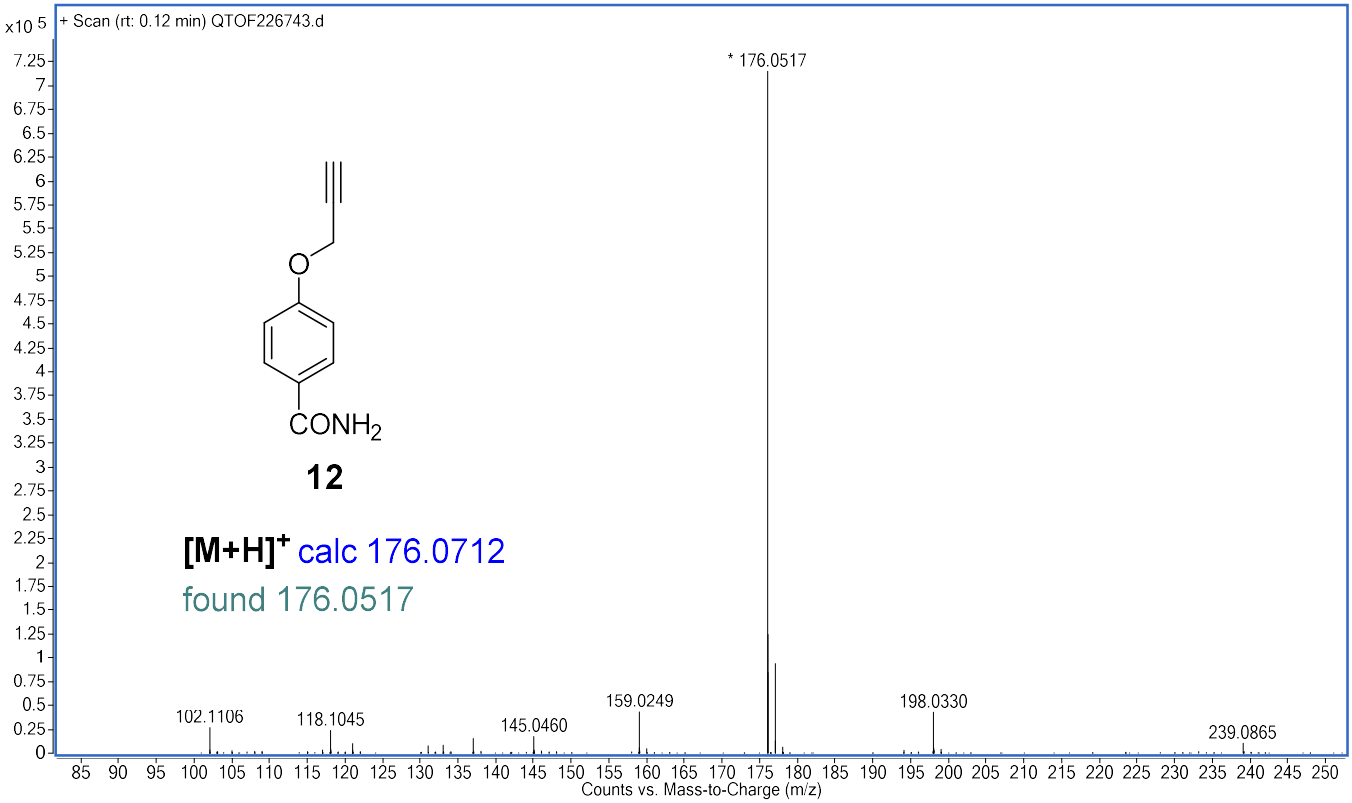

x10 3
+ Scan (rt: 0.25 min) QTOF226741.d
668.2376
Counts vs. Mass-to-Charge (m/z)

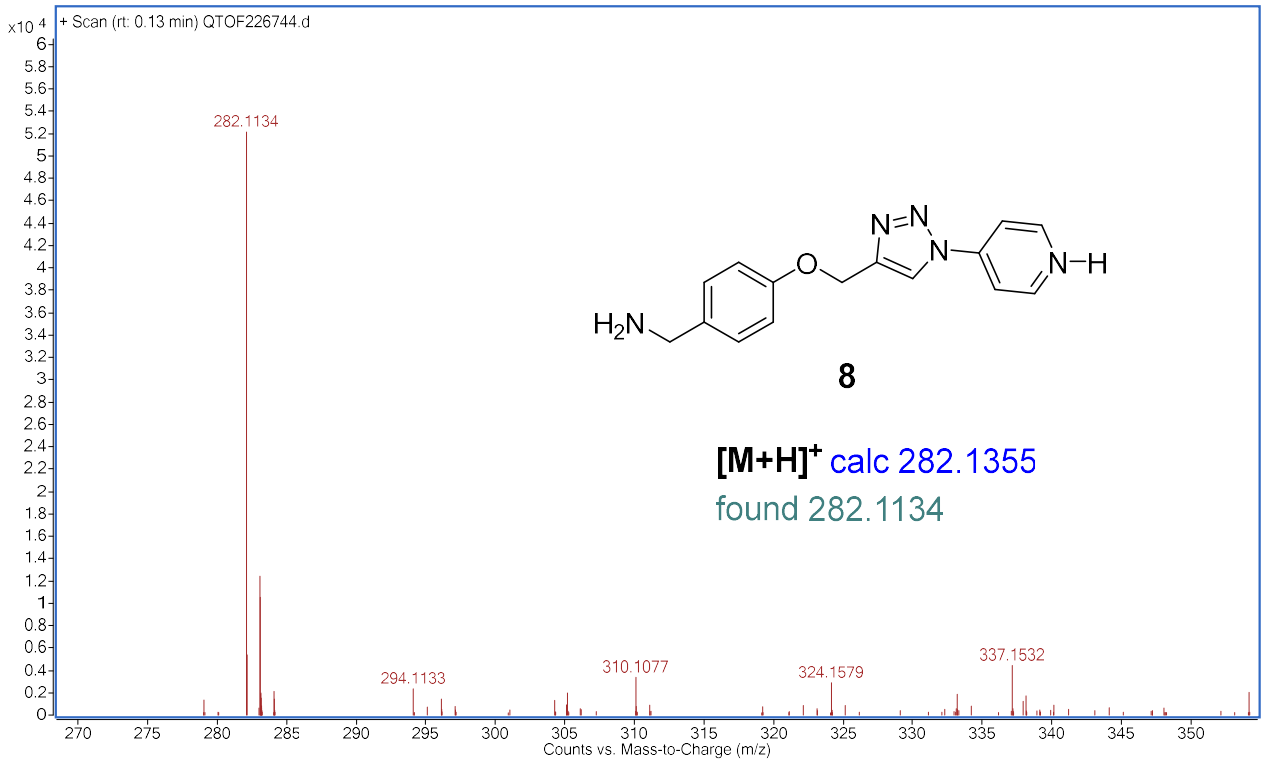

x10 6
+ Scan (rt: 0.22 min) QTOF211816.d
* 636.3168
451.3166
551.3427
737.4108
Counts vs. Mass-to-Charge (m/z)


+ Scan (rt: 0.17 min) QTOF226763.d
x10 5
636.2820
4a
[M+H]+ calc 668.2822
found 668.2820
235.2383
317.2400
496.2160
582.2345
737.3998
Counts vs. Mass-to-Charge (m/z)


+ Scan (rt: 0.31 min) QTOF212531.d
x10 5
* 608.2509
4c
[M+H]+ calc 608.2509
found 608.2509
Counts vs. Mass-to-Charge (m/z)


x10 5
+ Scan (rt: 0.14 min) QTOF227297.d
* 684.2812
4f
[M+H]+ calc 684.2822
found 684.2812
217.1007
355.0232
497.2014
598.3209
785.3995
900.4344
1087.5136
Counts vs. Mass-to-Charge (m/z)


x10 5
+ Scan (rt: 0.12 min) QTOF226743.d
* 176.0517
12
[M+H]+ calc 176.0712
found 176.0517
102.1106
118.1045
145.0460
159.0249
198.0330
239.0865
Counts vs. Mass-to-Charge (m/z)


+ Scan (rt: 0.13 min) QTOF226744.d
x10 4
282.1134
294.1133
310.1077
324.1579
337.1532
Counts vs. Mass-to-Charge (m/z)
8
[M+H]+ calc 282.1355
found 282.1134

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