Supporting Information for

Synthesis of a STnThr analogue structurally based on a TnThr antigen mimetic

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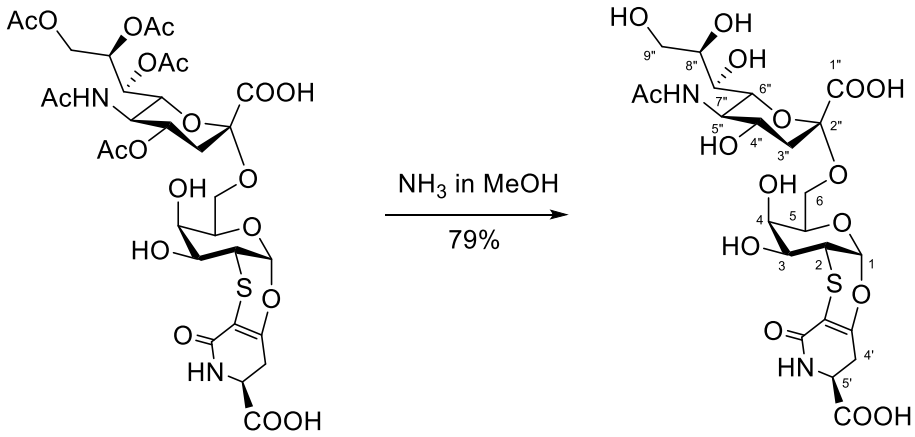
General method

Analytical grade solvents and available reagents were purchased from commercial sources and used without further purification unless noted. For anhydrous reactions, solvent stored over 3 Å molecular sieves were used. Silica gel flash column chromatography purifications were performed using Geduran® Si 60 (0.040-0.063 mm). TLC analyses were performed on glass Merck silica gel 60 F_{254} (0.25 mm, E. Merck) plates. NMR spectra were recorded on a 500 MHz Bruker AVANCE II at 298 K. All chemical shifts are reported in parts per million (δ) referenced to residual nondeuterated solvent. Assignments of resonances, including those for mixtures of compounds, were made on the basis of 2D NMR experiments. Multiplicity abbreviation: b = broad, s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet were used. ESI-MS spectra were carried out on a linear ion-trap double quadrupole mass spectrometer using electrospray ionization (ESI) technique (LTQ-XL - Thermo Fisher). HRMS were performed on a LTQ-IT-Orbitrap in either positive or negative modes. Optical rotation measurements were performed on a JASCO DIP-370 polarimeter. Melting point were recorded on a BUCHI 510. Elemental analyses were performed with an Elemental Analyzer 2400 Series II from Perkin-Elmer.

Experimental details and data

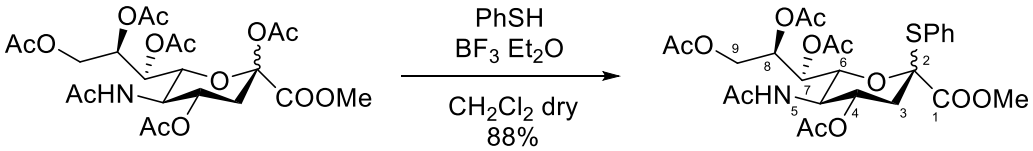
Synthesis of compound 2



12 (43 mg, 54.24 µmol) was solubilized in NH_3 in MeOH 4M (1 mL) and the reaction was stirred at room temperature. After 4 days, the solvent was removed and the product was purified by several washing with Et_2O to give pure **2** (27 mg, 79 % yield).

ESI-MS *m/z* (%): 623.42 (100) $[M]^-$, 311.25 (73) $[M]^{-2}$, $[\alpha]^{25}_D$ = +109.5 (c 0.001, H_2O), ^1H NMR (500 MHz, D_2O) δ: 5.72 (d, 1H, $J_{1,2}$= 2.7 Hz, H-1), 4.23-4.18 (m, 1H, H-5), 4.07-4.03 (m, 1H, H-5'), 3.99-3.95 (m, 1H, H-4), 3.92-3.86 (m, 1H, H-6_a), 3.86-3.71 (m, 4H, H-3, H-5'', H-8'', H-9''$_a$), 3.68-3.48 (m, 5H, H-4'', H-6_b, H-6'', H-7'', H-9"S_b), 3.41 (dd, 1H, $J_{2,1}$= 2.7 Hz, $J_{2,3}$= 11.2 Hz, H-2), 2.89 (dd, 1H, $J_{4'a,4'b}$= 16.9 Hz, $J_{4'a,5'}$= 6.8 Hz, H-4'$_a$), 2.72-2.58 (m, 2H, H-4'$_b$, H-3''$_{eq}$), 1.95 (s, 3H, CH_3), 1.67-1.59 (m, 1H, H-3''$_{ax}$), ^{13}C NMR (125 MHz, D_2O) δ: 177.3 (C_q), 174.9 (C_q), 173.4 (C_q), 167.5 (C_q), 158.3 (C_q), 100.4 (C_q), 96.0 (C-1), 93.7 (C_q), 72.6 (C-6''), 71.9 (C-5), 71.7 (C-8"), 68.4 (C-4), 68.2 (C-7", C-4"), 65.0 (C-3), 63.7 (C-6), 63.0 (C-9"), 52.8 (C-5'), 51.8 (C-5''), 40.1 (C-3"), 38.0 (C-2), 31.1 (C-4'), 22.0 (CH_3), HRMS (ESI): $[M]^-$ calcd for $C_{23}H_{31}N_2O_{16}S^-$, 623.1400; found, 623.1382; Elemental anal. calcd for $C_{23}H_{32}N_2O_{16}S$: C, 44.23; H, 5.16; N, 4.49; Found: C, 44.17; H, 5.19; N, 4.43.

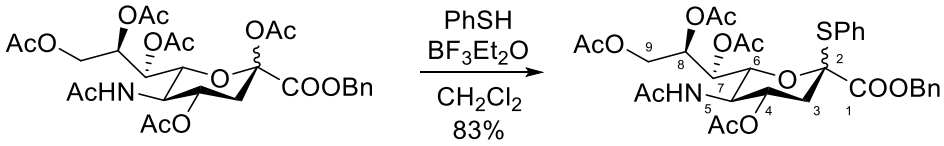
Synthesis of compound 3a



To a solution of **7a** (3.25 g, 6.09 mmol) in CH_2Cl_2 dry (50 mL), cooled to 0 °C, PhSH (730 mg, 6.63 mmol) and BF_3Et_2O (1.01 g, 7.09 mmol) were added and the solution was stirred at room temperature overnight. After complete conversion, the mixture was diluted with CH_2Cl_2 and washed with $NaHCO_3$ s.s. (x5). The organic layer was dried over Na_2SO_4 and concentrated under vacuum. The crude was purified by flash chromatography (DCM/MeOH 9:1) to give **3a** as yellow solid (3.10 g, 88% yield; β anomer 87 % - α anomer 13 %).

Characterization of β anomer: ESI-MS *m/z* (%): 606.25 (100) $[M+Na]^+$, ^1H NMR (β anomer) (500 MHz, $CDCl_3$) δ: 7.46-7.19 (m, 5H, SPh), 5.56 (d, 1H, $J_{NH,5}$= 10.3 Hz NH), 5.46 (dd, 1H, $J_{7,6}$= 2.5 Hz, $J_{7,8}$= 2.3 Hz, H-7), 5.41-5.34 (m, 1H, H-4), 4.94 (ddd, 1H, $J_{8,9b}$= 8.6 Hz, $J_{8,9a}$= 2.3 Hz, $J_{8,7}$= 2.3 Hz, H-8), 4.62 (dd, 1H, $J_{6,5}$= 10.5 Hz, $J_{6,7}$= 2.5 Hz, H-6), 4.48 (dd, 1H, $J_{9a,9b}$= 12.3 Hz, $J_{9a,8}$= 2.3 Hz, H-9a), 4.12 (ddd, 1H, $J_{5,6}$= 10.5 Hz, $J_{5,4}$= 10.4 Hz, $J_{5,NH}$= 10.3 Hz, H-5), 3.99 (dd, 1H, $J_{9b,9a}$= 12.3 Hz, $J_{9b,8}$= 8.6 Hz, H-9b), 3.58 (s, 3H, OCH_3), 2.66 (dd, 1H, $J_{3eq,3ax}$= 13.8 Hz, $J_{3eq,4}$= 4.8 Hz, H-3_{eq}), 2.16-2.11 (m, 1H, H-3_{ax}), 2.09 (s, 3H, CH_3), 2.07 (s, 3H, CH_3), 2.03 (s, 3H, CH_3), 1.95 (s, 3H, CH_3), 1.89 (s, 3H, CH_3), ^{13}C NMR (β anomer) (125 MHz, $CDCl_3$) δ: 171.3 (C_q), 171.1 (C_q), 170.4 (C_q), 170.4 (C_q), 170.3 (C_q), 168.3 (C1), 136.3 (CH, SPh) 129.9 (CH, SPh), 129.2 (CH, SPh), 129.0 (C_q, SPh), 89.1 (C-2), 73.3 (C-8, C-6), 69.2 (C-4), 69.0 (C-7), 62.8 (C-9), 52.7 (OCH_3), 49.5 (C-5), 37.6 (C-3), 23.3 (CH_3), 21.2 (CH_3), 21.0 (CH_3), 20.9 (CH_3), 20.8 (CH_3)

Synthesis of compound 3b

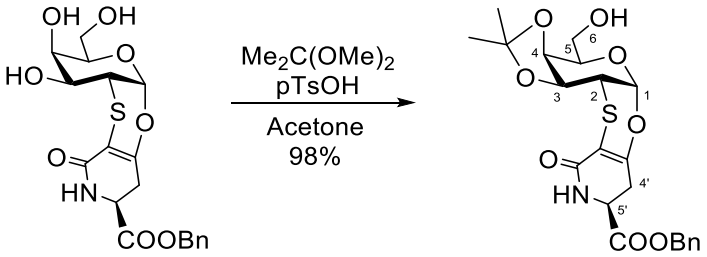


To a solution of **7b** (2.20 g, 3.61 mmol) in CH_2Cl_2 dry (25 mL), cooled to 0 °C, PhSH (432 mg, 3.92 mmol) and BF_3Et_2O (580 mg – 4.09 mmol) were added and the solution was stirred at room temperature, overnight. After complete conversion, the mixture was diluted with CH_2Cl_2 and washed with $NaHCO_3$ s.s (x5). The organic layer was dried over Na_2SO_4 and concentrated under vacuum. The crude was purified by flash

chromatography (EtOAc/Hexane 8:2) to give **3b** as white solid (1.97 g, 83% yield, β anomer 86 % - α anomer 14 %).

Characterization of β anomer: ESI-MS *m/z* (%): 682.17 (100) $[M+Na]^+$, ^{1}H NMR (β anomer) (500 MHz, $CDCl_3$) δ: 7.38-7.16 (m, 10H, SPh, Bn), 6.10 (d, 1H, $J_{NH,5}$= 10.3 Hz NH), 5.47-5.46 (m, 1H, H-7), 5.44-5.38 (m, 1H, H-4), 5.08-5.05 (m, 2H, CH_2Bn), 4.90-4.87 (m, 1H, H-8), 4.62 (dd, 1H, $J_{6,5}$= 10.5 Hz, $J_{6,7}$= 2.7 Hz, H-6), 4.38 (dd, 1H, $J_{9a,9b}$= 12.3 Hz, $J_{9a,8}$= 2.4 Hz, H-9a), 4.19-4.09 (m, 2H, H-5, H-9b), 2.65 (dd, 1H, $J_{3eq,3ax}$= 13.8 Hz, $J_{3eq,4}$= 4.8 Hz, H-3_{eq}), 2.17-2.06 (m, 4H, H-3_{ax}, CH_3), 2.03 (s, 3H, CH_3), 2.03 (s, 3H, CH_3), 1.94 (s, 3H, CH_3), 1.88 (s, 3H, CH_3), ^{13}C NMR (β anomer) (125 MHz, $CDCl_3$) δ: 171.2, 171.0, 170.4, 170.2, 170.2, 167.4, 135.8, 134.9, 129.6, 129.1, 128.7, 128.7, 128.5, 128.5, 88.5 (C-2), 73.1 (C-8, C-6), 69.2 (C-4), 68.8 (C-7), 67.6 (CH_2Bn), 62.2 (C-9), 49.1 (C-5), 37.5 (C-3), 23.1 (CH_3), 21.0 (CH_3), 20.9 (CH_3), 20.8 (CH_3), 20.7 (CH_3)

Synthesis of compound 4



5 (600 mg, 1.42 mmol) was suspended in acetone (15 mL), $Me_2C(OMe)_2$ (10.3 g, 99 mmol) and pTsOH (pH 3) were added and the solution was stirred at room temperature for 5 h. After complete conversion, the mixture was quenched with Et_3N (pH 8), diluted with CH_2Cl_2 and washed with HCl 1M (x4). The organic layer was dried over Na_2SO_4 and concentrated under vacuum. The crude was purified by flash chromatography (EtOAc/MeOH 9:1) to give pure **4** as white solid (628 mg, 98% yield).

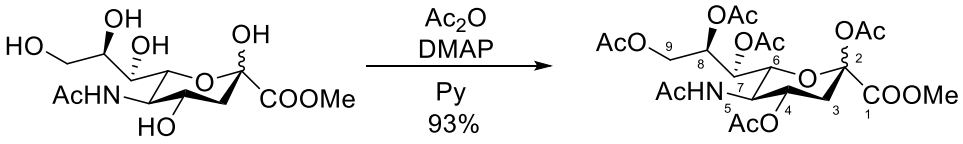
ESI-MS *m/z* (%): 486.17 (100) $[M+Na]^+$, 502.17 (90) $[M+K]^+$, $[\alpha]^{26}_D$ = +118.5 (c 0.001, CH_3OD), ^{1}H NMR (500 MHz, $DMSOd_6$) δ: 7.80 (d, 1H, $J_{NH,5'}$ 3.2 Hz, NH), 7.42-7.31 (m, 5H, Bn), 5.48 (d, 1H, $J_{1,2}$= 2.9 Hz, H-1), 5.22-5.16 (m, 2H, CH_2Bn), 4.38-4.32 (m, 1H, H-5'), 4.24-4.19 (m, 1H, H-5), 4.18-4.15 (m, 1H, H-4), 4.07 (dd, 1H, $J_{3,2}$= 8.7 Hz, $J_{3,4}$= 5.0 Hz, H-3), 3.66-3.55 (m, 2H, H-6_a, H-6_b), 3.17 (dd, 1H, $J_{2,3}$= 8.7 Hz, $J_{2,1}$= 2.9 Hz, H-2), 3.06 (dd, 1H, $J_{4'a,4'b}$= 16.7 Hz, $J_{4'a,5'}$= 7.1 Hz, H-4'$_a$), 2.61 (dd, 1H, $J_{4'b,4'a}$= 16.7 Hz, $J_{4'b,5'}$= 5.0 Hz, H-4'$_b$), 1.45 (s, 3H, CH_3), 1.27 (s, 3H, CH_3),^{13}C NMR (125 MHz, $DMSOd_6$) δ: 171.7 (C_q), 154.7 (C_q), 155.5 (C_q), 136.2 (C_q), 128.9 (CH, Bn), 128.6 (CH, Bn), 128.2 (CH, Bn), 109.1 (C_q), 96.6 (C_q), 95.2 (CH, C-1), 72.5 (CH, C-3), 72.4 (CH, C-4), 69.9 (CH, C-5), 66.9 (CH_2, CH_2Ph), 60.9 (CH, C-6), 51.0 (CH, C-5'), 39.11 (CH, C-2), 30.78 (CH_2, C-4'), 28.51 (CH_3), 26.61 (CH_3); HRMS (ESI): $[M+Na]^+$ calcd for $C_{22}H_{25}NO_8SNa^+$, 486.1193; found, 489.1174; Elemental anal. calcd for $C_{22}H_{25}NO_8S$: C, 57.01; H, 5.44; N, 3.02; Found: C, 56.98; H, 5.47; N, 3.05.

Synthesis of compound 6a

N-acetylneuraminic acid (5.05 g, 16.31 mmol) was suspended in MeOH (170 mL), Dowex 50WX8 (1.5 g) was added and the suspension was stirred at 40 °C. After complete dissolution, Dowex 50WX8 was filtered off and the organic layer was concentrated, under reduce pressure, to give **6a** as white solid (5.06 g, > 95% yield; β anomer 93 % - α anomer 7 %).

Characterization of β anomer: ESI-MS *m/z* (%): 346.17 (100) $[M+Na]^+$, 668.92 (65) $[M+Na]^{2+}$, 386.17 (55) $[M+K]^+$, 708.83 (35) $[M+K]^{2+}$, ^{1}H NMR (β anomer) (500 MHz, CD_3OD) δ: 4.08-3.99 (m, 1H, H-4), 3.99 (dd, 1H, $J_{6,5}$= 10.1 Hz, $J_{6,7}$= 1.4 Hz, H-6), 3,84-3,76 (m, 5H, H-5, OCH_3, H-9_a), 3.72-3.68 (m, 1H, H-8), 3.62 (dd, 1H, $J_{9b,9a}$= 11.2 Hz, $J_{9b,8}$= 5.7 Hz, H-9_b), 3.48 (dd, 1H, $J_{7,8}$= 9.1 Hz, $J_{7,6}$= 1.4 Hz, H-7), 2.22 (dd, 1H, $J_{3eq,3ax}$= 12.9 Hz, $J_{3eq,4}$= 5.0 Hz, H-3_{eq}), 2.00 (s, 3H, NHAc), 1.89 (dd, 1H, $J_{3ax,3eq}$= 12.9 Hz, $J_{3ax,4}$= 11.6 Hz, H-3_{ax}), ^{13}C NMR (β anomer) (125 MHz, CD_3OD) δ: 175.1 (C_q - NHAc), 171.8 (C-1), 96.7 (C-2), 72.1 (C-6), 71.7 (C-8), 70.2 (C-4), 67.9 (C-7), 64.9 (C-9), 54.3 (OCH_3), 53.1 (C-5), 40.7 (C-3), 22.6 (NHAc).

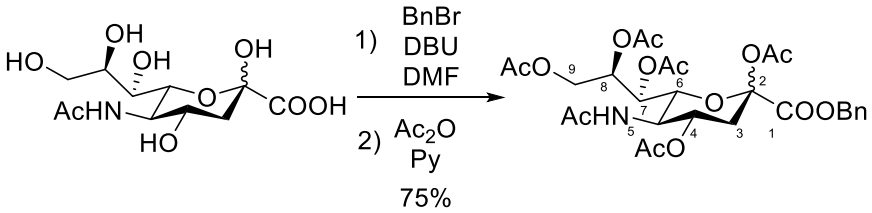
Synthesis of compound 7a



To a solution of **6a** (5.05 g, 15.62 mmol) in pyridine (80 mL), Ac_2O (10.3 g, 99 mmol) and DMAP (100 mg) were added and the solution was stirred at room temperature overnight. After complete conversion, the mixture was diluted with CH_2Cl_2 and washed with HCl 1M (x5). The organic layer was dried over Na_2SO_4 and concentrated under vacuum. The crude was purified by flash chromatography (DCM/MeOH 9:1) to give **7a** as white solid (7.73 g, 93% yield, β anomer > 90 %).

Characterization of β anomer: ESI-MS *m/z* (%): 556.17 (100) $[M+Na]^+$, 572.17 (35) $[M+K]^+$, ^{1}H NMR (β anomer) (500 MHz, $CDCl_3$) δ: 5.40-5.35 (m, 2H, NH, H-7), 5.27-5.20 (m, 1H, H-4), 5.07-5.04 (m, 1H, H-8), 4.48 (dd, 1H, $J_{9a,9b}$= 12.4 Hz, $J_{9a,8}$= 2.6 Hz, H-9_a), 4.14-4.07 (m, 3H, H-5, H-6, H-9_b), 3.80 (s, 3H, OCH_3), 2.53 (dd, 1H, $J_{3eq,3ax}$= 13.6 Hz, $J_{3eq,4}$= 5.0 Hz, H-3_{eq}), 2.34 (s, 3H, CH_3), 2.13 (s, 3H, CH_3), 2.11-2.07 (m, 1H, H-3_{ax}), 2.09 (s, 3H, CH_3), 2.02 (s, 3H, CH_3), 2.02 (s, 3H, CH_3), 1.88 (s, 3H, CH_3), ^{13}C NMR (β anomer) (125 MHz, $CDCl_3$) δ: 171.1 (C_q), 171.1 (C_q), 170.7 (C_q), 170.4 ($2C_q$), 170.3 (C_q), 168.3 (C_q), 166.4 (C_q), 97.6 (C-2), 72.97 (C-6), 71.5 (C-8), 68.4 (C-4), 67.9 (C-7), 62.2 (C-9), 53.3 (OCH_3), 49.4 (C-5), 36.0 (C-3), 23.3 (CH_3), 21.1 (CH_3), 21.0 (CH_3), 20.9 ($2CH_3$), 20.8 (CH_3).

Synthesis of compound 7b

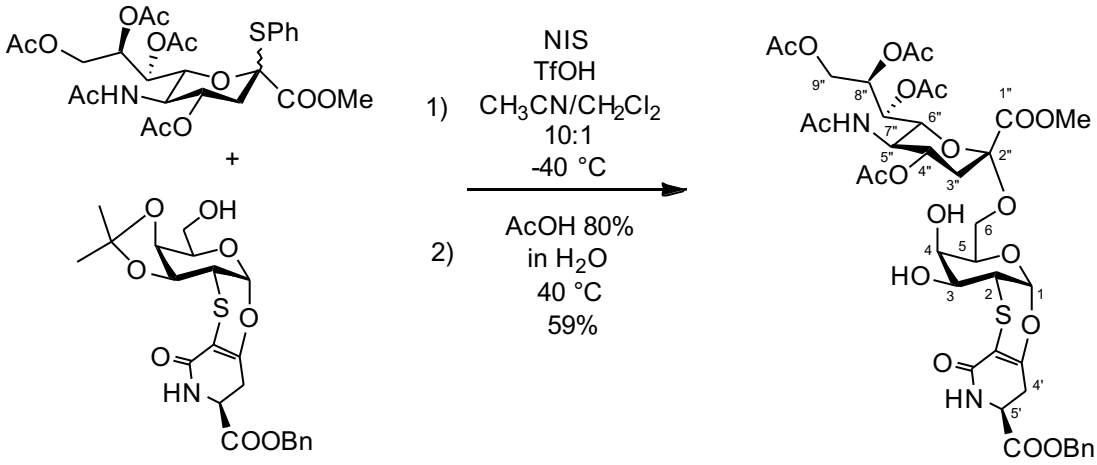


To a suspension of N-acetylneuraminic acid (5.00 g, 16.16 mmol) in DMF (25 mL) cooled to - 0 °C, DBU (3.91 g, 25.72 mmol) and BnBr (4.40 g, 25.72 mmol) were added and the mixture was stirred at room temperature overnight. After complere conversion, the solvent was removed *in vacuo.* Crude **6b** was dissolved in pyridine/Ac_2O 2:1 (100 mL) and a catalitical amount of DMAP was added. After overnight, the reaction was diluted with CH_2Cl_2 and washed with HCl 1M (x5). The organic layer was dried over Na_2SO_4 and concentrated under vacuum. The crude was purified by flash chromatography (DCM/MeOH 9:1) to give **7b** (7.39 g, 75% yield; β anomer > 90 %).

Characterization of β anomer: ESI-MS *m/z* (%): 632.17 (100) $[M+Na]^+$, ^{1}H NMR (β anomer) (500 MHz, $CDCl_3$) δ: 7.41-7.33 (m, 5H, Bn), 5.42-5.33 (m, 2H, NH, H-7), 5.31-5.16 (m, 3H, H-4, CH_2Bn), 5.13-5.08 (m, 1H, H-8), 4.46 (dd, 1H, $J_{9a,9b}$= 12.6 Hz, $J_{9a,8}$= 2.6 Hz, H-9_a), 4.21-4.07 (m, 3H, H-5, H-6, H-9_b), 2.57 (dd, 1H, $J_{3eq,3ax}$= 13.6

Hz, $J_{3eq,4}$= 5.2 Hz, H-3_{eq}), 2.14 (s, 3H, CH_3), 2.13-2.07 (m, 4H, H-3_{ax}, CH_3), 2.05-2.03 (s, 9H, CH_3, CH_3, CH_3), 1.91 (s, 3H, CH_3), ^{13}C NMR (β anomer) (125 MHz, $CDCl_3$) δ: 171.0 (C_q), 170.5 (C_q), 170.3 (C_q), 170.2 (2C_q), 168.3 (C_q), 165.5 (C_q), 134.9 (C_q), 128.6 (CH-Bn), 128.5 (CH-Bn), 128.3 (CH-Bn) 97.7 (C-2), 72.9 (C-6), 71.2 (C-8), 68.3 (C-4), 68.0 (CH_2-Bn), 67.8 (C-7), 62.0 (C-9), 49.4 (C-5), 35.8 (C-3), 23.2 (CH_3), 20.9 (CH_3), 20.8 (3CH_3), 20.7 (CH_3).

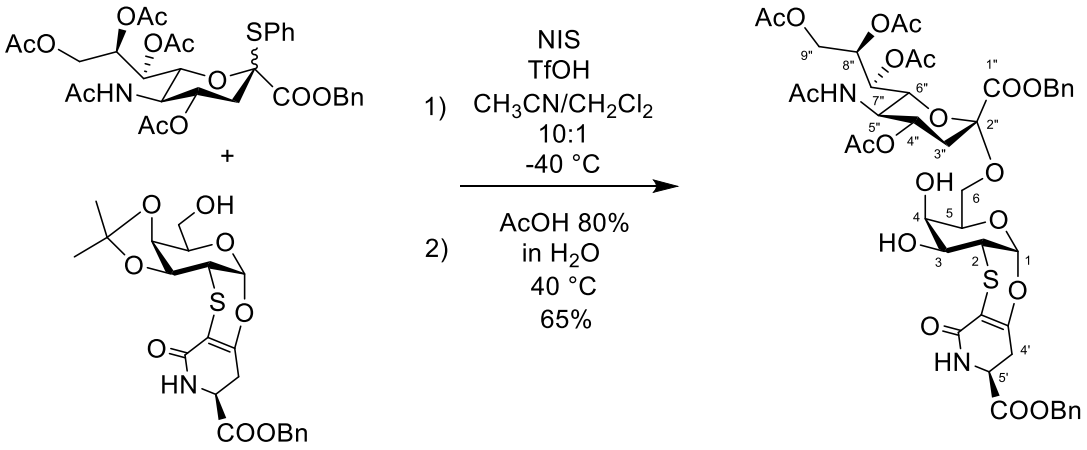
Synthesis of compound 8a



To a suspension of **4** (500 mg, 1.08 mmol) and **3a** (1.50 g, 2.57 mmol) in a mixture of CH_3CN/CH_2Cl_2 dry 10:1 (10 mL) cooled to - 40 °C, NIS (1.04 g, 4.62 mmol) and TfOH (203 mg, 1.35 mmol) were added and the solution was stirred at - 40 °C under an N_2 atmosphere. After 2 h, the reaction was quenched with Et_3N (pH 8), diluted with CH_2Cl_2 and washed with $Na_2S_2O_3$ 1 M (x3). The organic layer was dried over Na_2SO_4 and concentrated under vacuum. The crude was solubilized in AcOH 80% in H_2O (20 mL) and the reaction was stirred overnight at 40 °C. The solvent was removed *in vacuo* and the product was purified by flash chromatography (EtOAc/MeOH 98:2) affording **8a** (571 mg, 59 % yield calculated over 2 steps; α anomer 90 % - β anomer 10 %).

Characterization of α anomer: ESI-MS *m/z* (%): 919.42 (100) $[M+Na]^+$, 935.42 (90) $[M+K]^+$, ^{1}H NMR (α anomer) (500 MHz, $CDCl_3$) δ: 7.37-7.27 (m, 5H, Bn), 6.71 (bs, 1H, NH), 5.71 (d, 1H, $J_{NH,5''}$= 9.8 Hz, NH), 5.57 (d, 1H, $J_{1,2}$= 2.7 Hz, H-1), 5.37-5.33 (m, 1H, H-8''), 5.31-5.28 (m, 1H, H-7''), 5.21-5.14 (m, 2H, CH_2Bn), 4.91-4.81 (m, 1H, H-4''), 4.38- 4.27 (m, 2H, H-5', H-9''$_a$), 4.19-3.98 (m, 5H, H-4, H-5, H-5'', H-6'', H-9''$_b$), 3.95-3.86 (m, 1H, H-6_a), 3.78 (s, 1H, OCH_3), 3.72-3.63 (m, 2H, H-3, H-6_b), 3.48 (dd, 1H, 1H, $J_{2,1}$= 2.7 Hz, $J_{2,3}$= 10.7 Hz, H-2), 2.95-2.87 (m, 1H, H-4"$_a$), 2.80-2.71 (m, 1H, H-4'$_b$), 2.57 (dd, 1H, $J_{3''eq,3''ax}$= 12.8 Hz, $J_{3''eq,4''}$= 4.6 Hz, H-3''$_{eq}$), 2.10 (s, 3H, CH_3), 2.09 (s, 3H, CH_3), 1.98-1.90 (m, 1H, H-3"$_{ax}$), 2.00 (s, 3H, CH_3), 1.99 (s, 3H, CH_3), 1.85 (s, 3H, CH_3), ^{13}C NMR (α anomer) (125 MHz, $CDCl_3$) δ: 171.0, 170.6, 170.4, 170.2, 169.8, 168.2, 165.8, 155.9, 134.9, 128.8, 128.4, 98.8, 96.5 (C-1), 96.3, 72.8, 71.3, 69.2 (C-8''), 69.1 (C-4''), 67.9, 67.8 (CH_2Bn), 67.6 (C-7''), 65.7 (C-3), 63.2 (C-6), 62.7 (C-9''), 53.1 (OCH_3), 51.4 (C-5'), 49.2, 39.1 (C-2), 37.4 (C-3''), 30.7 (C-4'), 23.2 (CH_3), 21.1 (CH_3), 20.9 (CH_3), 20.9 (CH_3), 20.8 (CH_3).

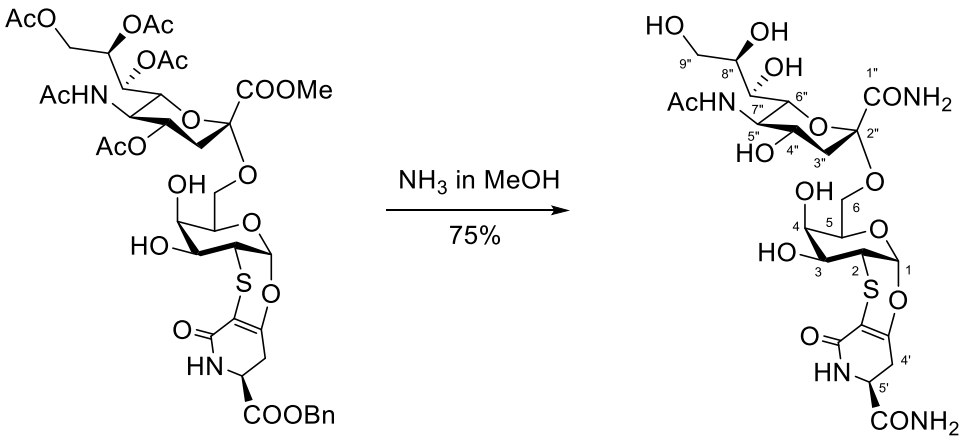
Synthesis of compound 8b



To a suspension of **4** (134 mg, 0.29 mmol) and **3b** (460 mg, 0.69 mmol) in a mixture of CH_3CN/CH_2Cl_2 dry 10:1 (3 mL) cooled to - 40 °C, NIS (260 mg, 0.29 mmol) and TfOH (86 mg, 0.57 mmol) were added and the solution was stirred at - 40 °C under an N_2 atmosphere. After 2 h, the reaction was quenched with Et_3N (pH 8), diluted with CH_2Cl_2 and washed with $Na_2S_2O_3$ 1 M (x3). The organic layer was dried over Na_2SO_4 and concentrated under vacuum. The crude was solubilized in AcOH 80% in H_2O (10 mL) and the reaction was stirred overnight at 40 °C. The solvent was removed *in vacuo* and the product was purified by flash chromatography (EtOAc/MeOH 98:2) to give **8b** (183 mg, 65 % yield calculated over 2 steps >95%; pure by ^1H NMR)

ESI-MS *m/z* (%): 995.17 (100) $[M+Na]^+$, 1011.17 (35) $[M+K]^+$, $[\alpha]^{24}_D$ = +112.3 (c 0.001, $CHCl_3$), ^1H NMR (α anomer) (500 MHz, $CDCl_3$) δ: 7.42-7.34 (m, 5H, Bn), 6.21 (bs, 1H, NH), 5.61 (d, 1H, $J_{NH,5''}$= 9.8 Hz, NH), 5.61 (d, 1H, $J_{1,2}$= 2.7 Hz, H-1), 5.40-5.28 (m, 3H, NH, H-7'', H-8''), 5.27-5.19 (m, 4H, CH_2Bn, CH_2Bn), 4.93-4.84 (m, 1H, H-4''), 4.40 (dd, 1H, $J_{9''a,9''b}$= 12.6 Hz, $J_{9a,8}$= 2.6 Hz, H-9''$_a$), 4.36-4.30 (m, 1H, H-5'), 4.18-3.93 (m, 6H, H-4, H-5, H-5'', H-6$_a$, H-6'', H-9''$_b$), 3.73-3.63 (m, 2H, H-3, H-6$_b$), 3.53 (dd, 1H, 1H, $J_{2,1}$= 2.7 Hz, $J_{2,3}$= 10.7 Hz, H-2), 3.44 (bs, 1H, OH), 3.15 (bs, 1H, OH), 2.95-2.86 (m, 1H, H-4''$_a$), 2.85-2.75 (m, 1H, H-4'$_b$), 2.66 (dd, 1H, $J_{3''eq,3''ax}$= 12.8 Hz, $J_{3''eq,4''}$= 4.6 Hz, H-3''$_{eq}$), 2.15 (s, 3H, CH_3), 2.14 (s, 3H, CH_3), 2.05-2.03 (m, 7H, H-3''$_{ax}$,2CH_3), 1.89 (s, 3H, CH_3), ^{13}C NMR (α anomer) (125 MHz, $CDCl_3$) δ:171.1, 170.8, 170.4, 170.3, 170.2, 169.5, 167.5, 165.1, 155.5, 134.8, 134.7, 128.8-128.4 (5C), 98.8, 96.6 (C-1), 96.2, 72.9, 71.3, 69.1 (C-8''), 68.9 (C-4''), 68.0, 67.9 (2C), 67.5 (C-7''), 65.7 (C-3), 63.5 (C-6), 62.7 (C-9''), 51.4 (C-5'), 49.2, 39.2 (C-2), 37.3 (C-3''), 30.7 (C-4'), 23.2 (CH_3), 21.1 (CH_3), 20.8-20.7 (3CH_3).

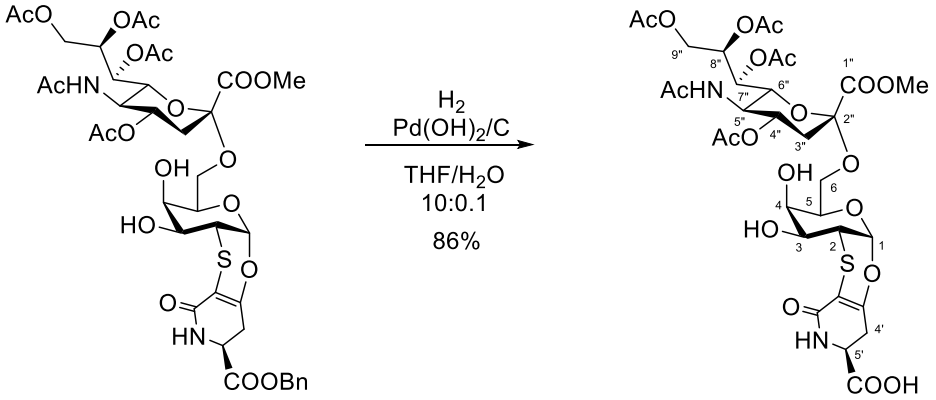
Synthesis of compound 10



8a (50 mg, 55.75 µmol) was solubilized in NH_3 in MeOH 4M (1 mL) and the reaction was stirred at room temperature. After 4 days, the solvent was removed and the product was purified by several washing with Et_2O to give pure **10** (26 mg, 75 % yield).

ESI-MS *m/z* (%): 621.33 (100) $[M]^-$, $[\alpha]^{25}_D$ = +111.2 (c 0.001, H_2O), ^{1}H NMR (500 MHz, D_2O) δ: 5.85 (d, 1H, $J_{1,2}$= 2.7 Hz, H-1), 4.37-4.34 (m, 1H, H-5'), 4.33-4.28 (m, 1H, H-5), 4.10-4.07 (m, 1H, H-4), 4.03-3.94 (m, 2H, H-5'', H-6$_a$), 3.93-3.83 (m, 3H, H-6'', H-8'', H-9"$_a$), 3.82-3.75 (m, 3H, H-3, H-4'', H-6$_b$), 3.73-3.66 (m, 2H, H-7'', H-9"$_b$), 3.53 (dd, 1H, $J_{2,1}$= 2.7 Hz, $J_{2,3}$= 11.2 Hz, H-2), 3.14 (dd, 1H, $J_{4'a,4'b}$= 17.1 Hz, $J_{4'a,5'}$= 7.5 Hz, H-4'$_a$), 2.84 (dd, 1H, $J_{4'b,4'a}$= 17.1 Hz, $J_{4'b,5'}$= 5.7 Hz, H-4'$_b$), 2.77 (dd, 1H, $J_{3''eq,3''ax}$= 13.1 Hz, $J_{3''eq,4''}$= 4.7 Hz, H-3''$_{eq}$), 2.08 (s, 3H, CH_3), 1.90-1.82 (m, 1H, H-3''$_{ax}$), ^{13}C NMR (125 MHz, D_2O) δ: 175.7 (C_q), 175.1 (C_q), 171.7 (C_q), 167.4 (C_q), 157.6 (C_q), 99.5 (C_q), 96.3 (C-1), 94.2 (C_q), 73.5 (C-6''), 71.9 (C-5), 71.0 (C-8''), 68.4 (C-4), 67.7 (C-7''), 67.1 (C-4''), 65.2 (C-3), 63.2 (C-6), 63.0 (C-9''), 51.7 (C-5''), 51.2 (C-5'), 38.3 (C-3''), 37.9 (C-2), 30.4 (C-4'), 22.1 (CH_3); HRMS (ESI): $[M]^-$ calcd for $C_{23}H_{33}N_4O_{14}S^-$, 621.1719; found, 621.1702; Elemental anal. calcd for $C_{23}H_{34}N_4O_{14}S$: C, 44.37; H, 5.50; N, 9.00; Found: C, 44.41; H, 5.46; N, 9.05.

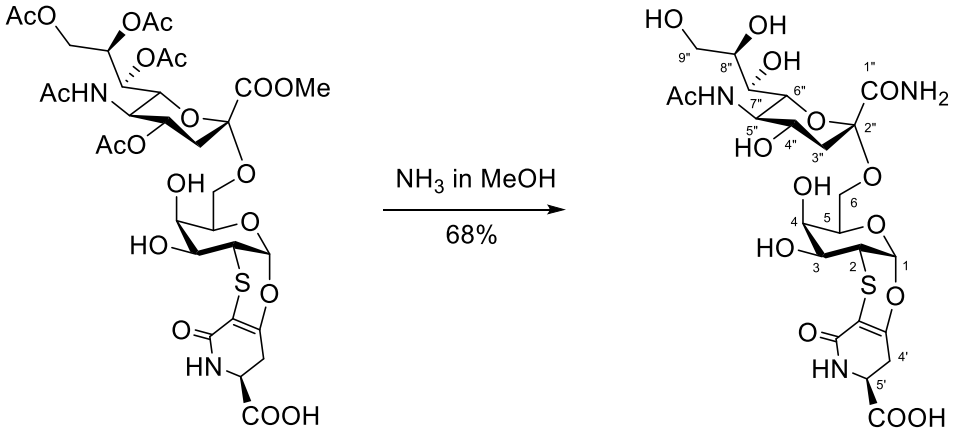
Synthesis of compound 11



8a (100 mg, 0.11 mmol) was solubilized in THF/H_2O 10:0.1 (2 mL) $Pd(OH)_2$/C (20 wt.%, 10 mg) was added and the reaction was stirred at room temperature, under H_2 atmosphere. After complete conversion, the suspension was filtered through a pad of Celite$^{(R)}$, the solvents were removed and the crude was purified by flash chromatography (CH_2Cl_2/MeOH 8:2) to give pure **11** (77 mg, 86 % yield).

ESI-MS *m/z* (%): 805.67 (100) $[M]^-$, $[\alpha]^{26}_D$ = +107.6 (c 0.002, CH_3OH), ^{1}H NMR (500 MHz, CD_3OD) δ: 5.69-5.62 (m, 1H, H-1), 5.45-5.37 (m, 1H, H-8''), 5.37-5.36 (m, 1H, H-7''), 4.84-4.79 (m, 1H, H-4''), 4.38- 4.31 (m, 1H, H-9''$_a$), 4.19-4.06 (m, 4H, H-5, H-5', H-6'', H-9''$_b$), 4.02-3.90 (m, 3H, H-4, H-5", H-6$_a$), 3.86 (s, 1H, OCH_3), 3.78-3.70 (m, 1H, H-3), 3.69-3.60 (m, 1H, H-6$_b$), 3.50-3.43 (m, 1H, H-2), 2.91-2.81 (m, 1H, H-4"$_a$), 2.79-2.70 (m, 1H, H-4'$_b$), 2.70-2.62 (m, 1H, H-3"$_{eq}$), 2.14 (s, 3H, CH_3), 2.12 (s, 3H, CH_3), 2.02(s, 3H, CH_3), 2.00 (s, 3H, CH_3), 1.90-1.80 (m, 4H, H-3"$_{ax}$, CH_3)

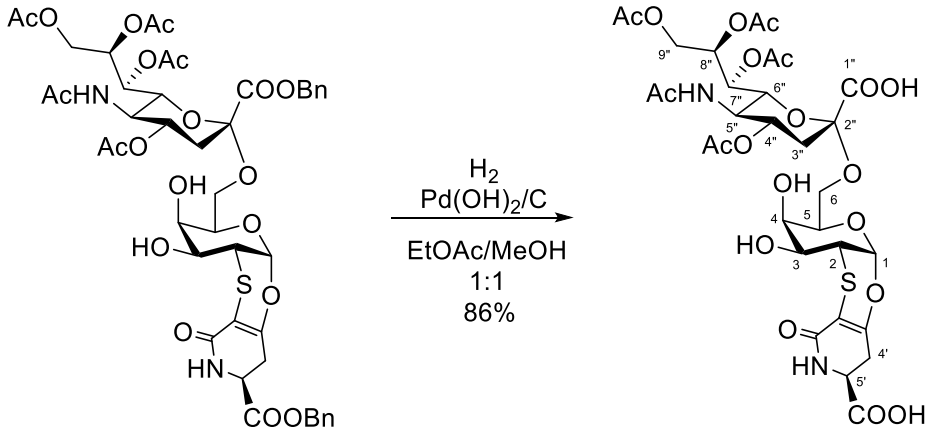
Synthesis of compound 12



11 (31 mg, 38.42 µmol) was solubilized in NH_3 in MeOH 4M (1 mL) and the reaction was stirred at room temperature. After 4 days, the solvent was removed and the product was purified by several washing with Et_2O to give pure **12** (16 mg, 68 % yield).

ESI-MS *m/z* (%): 622.33 (100) $[M]^-$, $[\alpha]^{25}_D$ = +109.1 (c 0.001, H_2O), ^{1}H NMR (500 MHz, D_2O) δ: 5.81 (d, 1H, $J_{1,2}$= 2.7 Hz, H-1), 4.34-4.29 (m, 1H, H-5), 4.17-4.12 (m, 1H, H-5'), 4.08-4.05 (m, 1H, H-4), 4.01-3.73 (m, 8H, H-3, H-4'', H-5'', H-6_a, H-6_b, H-6'', H-8'', H-9''$_a$), 3.72-3.62 (m, 2H, H-7'', H-9''$_b$), 3.51 (dd, 1H, $J_{2,1}$= 2.7 Hz, $J_{2,3}$= 11.2 Hz, H-2), 2.96 (dd, 1H, $J_{4'a,4'b}$= 16.9 Hz, $J_{4'a,5'}$= 6.8 Hz, H-4'$_a$), 2.81-2.71 (m, 2H, H-4'$_b$,H-3''$_{eq}$), 2.05 (s, 3H, CH_3), 1.88-1.80 (m, 1H, H-3''$_{ax}$), ^{13}C NMR (125 MHz, D_2O) δ: 177.3 (C_q), 175.0 (C_q), 171.7 (C_q), 167.5 (C_q), 158.2 (C_q), 99.5 (C_q), 96.1 (C-1), 93.9 (C_q), 73.4 (C-6''), 71.8 (C-5), 71.0 (C-8''), 68.4 (C-4), 67.7 (C-7''), 67.1 (C-4''), 65.1 (C-3), 63.2 (C-6), 63.0 (C-9''), 52.9 (C-5''), 51.6 (C-5'), 38.2 (C-3''), 38.1 (C-2), 31.2 (C-4'), 22.0 (CH_3); HRMS (ESI): $[M]^-$ calcd for $C_{23}H_{32}N_3O_{15}S^-$ 622.1560; found, 622.1583; Elemental anal. calcd for $C_{23}H_{33}N_3O_{15}S$: C, 44.30; H, 5.33; N, 6.74; Found: C, 44.24; H, 5.36; N, 6.82.

Synthesis of compound 13



8b (90 mg, 92.50 µmol) was solubilized in EtOAc/MeOH 1:1 (1 mL) $Pd(OH)_2$/C (20 wt.%, 9 mg) was added and the reaction was stirred under H_2 atmosphere at room temperature. After complete conversion, the suspension was filtered through a pad of Celite $^{(R)}$, the solvents were removed to give **13** (60 mg, 86 % yield).

ESI-MS *m/z* (%): 791.75 (100) $[M]^-$, $[\alpha]^{25}_D$ = +110.6 (c 0.001, CH_3OH), ^{1}H NMR (500 MHz, CD_3OD) δ: 5.67 (d, 1H, $J_{1,2}$= 2.7 Hz, H-1), 5.40-5.37 (m, 1H, H-8''), 5.37-5.33 (m, 1H, H-7''), 5.04-4.96 (m, 1H, H-4''), 4.59-4.53 (m, 1H, H-6''), 4.40 (dd, 1H, $J_{9''a,9''b}$= 12.4 Hz, $J_{9a,8}$= 2.7 Hz, H-9''$_a$), 4.23-4.14 (m, 3H, H-5, H-5', H-9''$_b$), 4.06-4.03 (m,

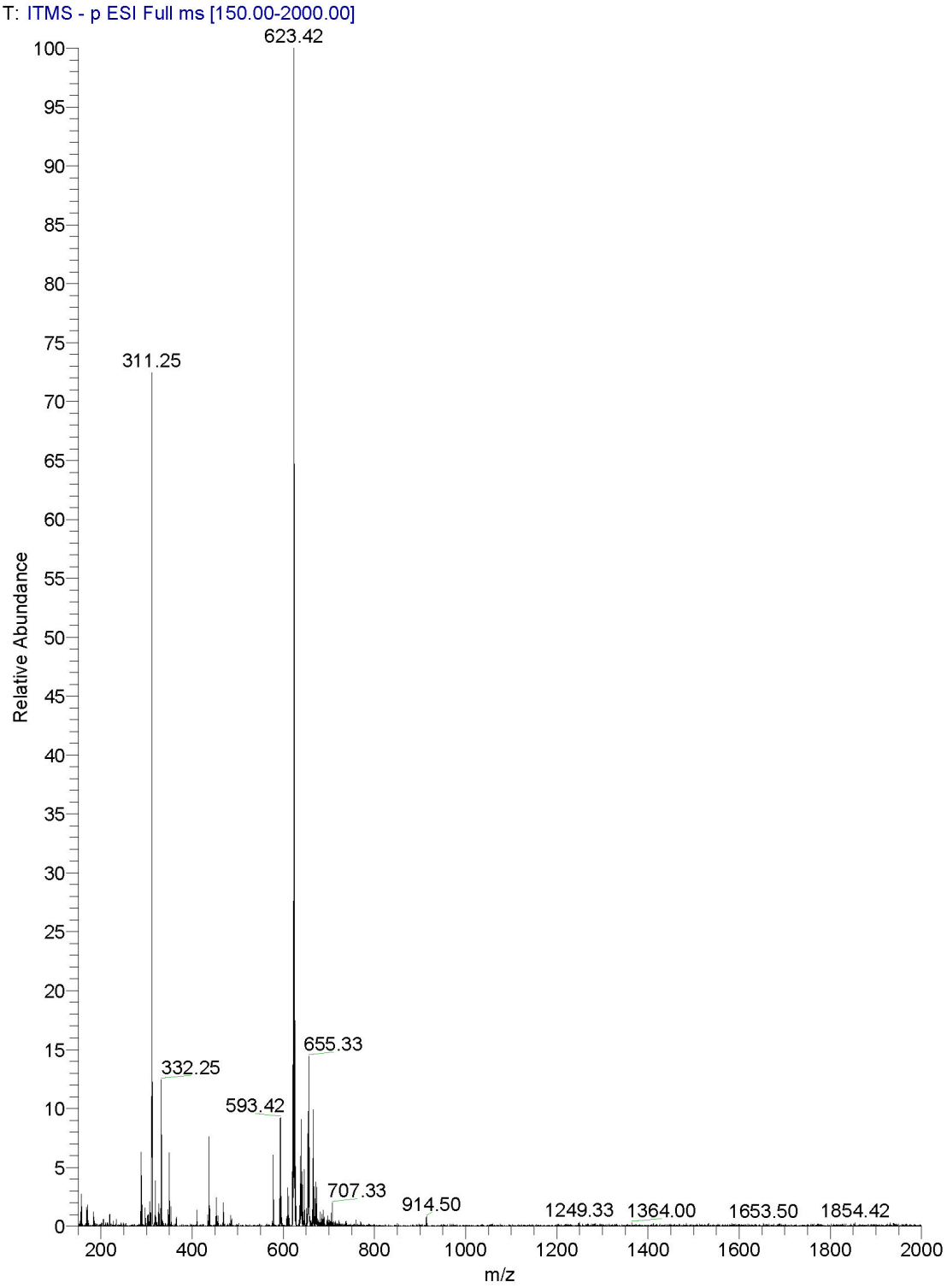
1H, H-4), 4.00-3.89 (m, 2H, H-5", H-6_a), 3.76-3.66 (m, 2H, H-3, H-6_b), 3.48 (dd, 1H, 1H, $J_{2,1}$= 2.9 Hz, $J_{2,3}$= 11.1 Hz, H-2), 3.00-2.92 (m, 1H, H-4"$_a$), 2.81-2.73 (m, 1H, H-4'$_b$), 2.64 (dd, 1H, $J_{3''eq,3''ax}$= 12.4 Hz, $J_{3''eq,4''}$= 4.6 Hz, H-3''$_{eq}$), 2.12 (s, 3H, CH_3), 2.11 (s, 3H, CH_3), 2.03(s, 3H, CH_3), 1.98 (s, 3H, CH_3), 1.85 (s, 3H, CH_3), 1.79-1.69 (m, 1H, H-3"$_{ax}$)

H1N1/ Compound 2 complex Modeling

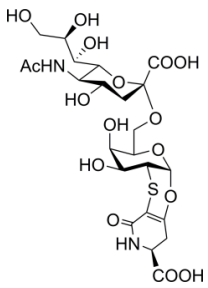
H1N1 neuraminidase structure bound to zanamivir (PDB 3B7E)[1] was used as a starting template for complex generation. Topology and parameters files for compound **2** were obtained by Antechamber program using AM1-BCC charges[2]. The complex model was then immersed in a periodic water box (TIP3) and neutralized by adding Na+ ions. This model was initially equilibrated by several cycles of minimizations (10000 steps, steepest descent) and Molecular Dynamics (50 ps, 200K). Then the protein backbone was constrained and the ligand was docked into H1N1 active site by Soft-Restrained Molecular Dynamics, using Zanamivir coordinates in 3B7E structure as template [3]. Finally, complex was equilibrated and energy minimized using minimization/dynamics cycles without restrained. All computational studies were performed using Amber ff14SB forcefield [4] with NAMD software[5].

NMR and ESI-MS spectra

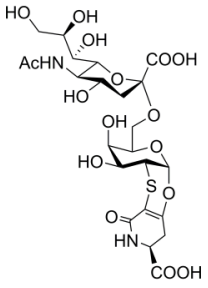
NMR and ESI-MS spectra of compound 2



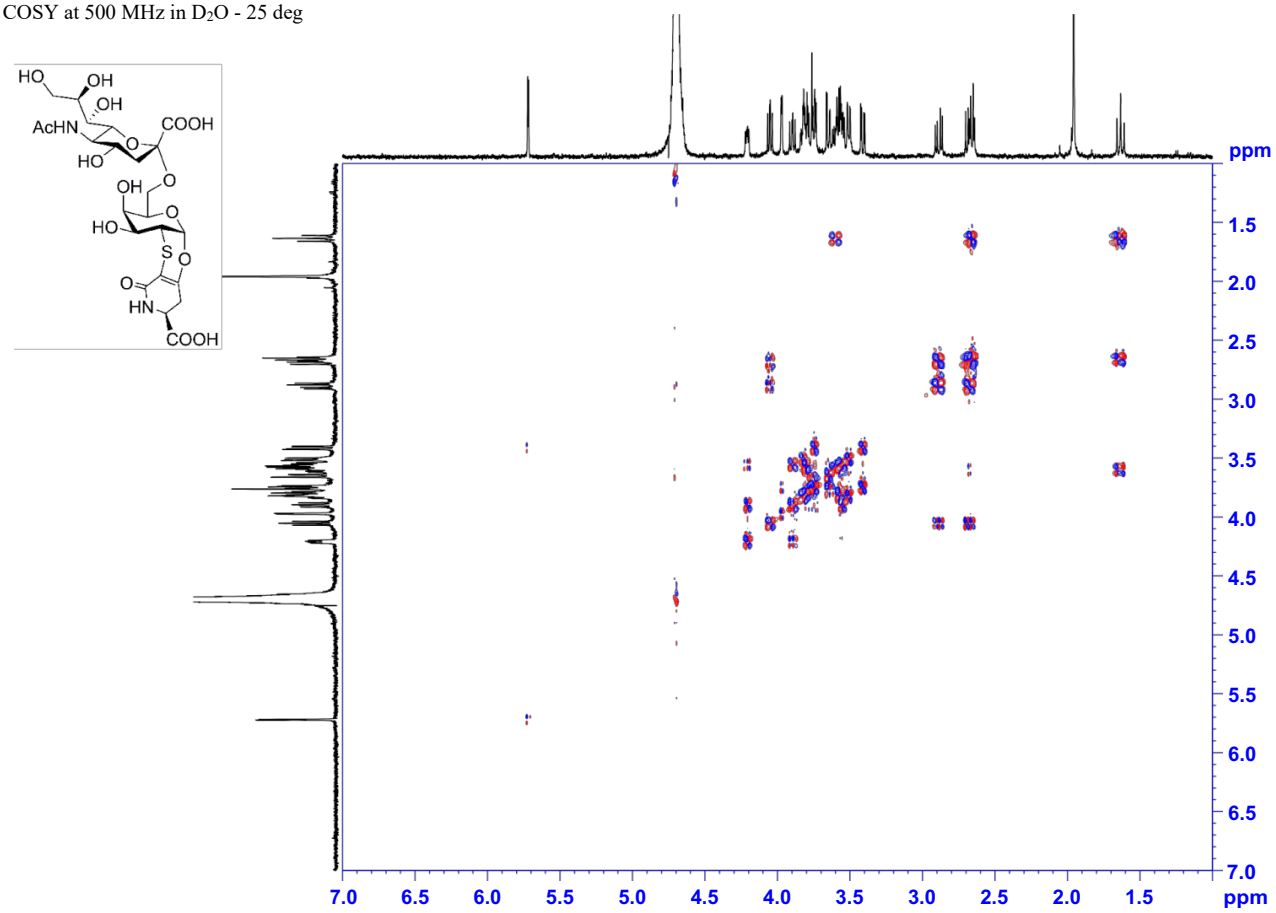
^{1}H at 500 MHz in D_2O - 25 deg



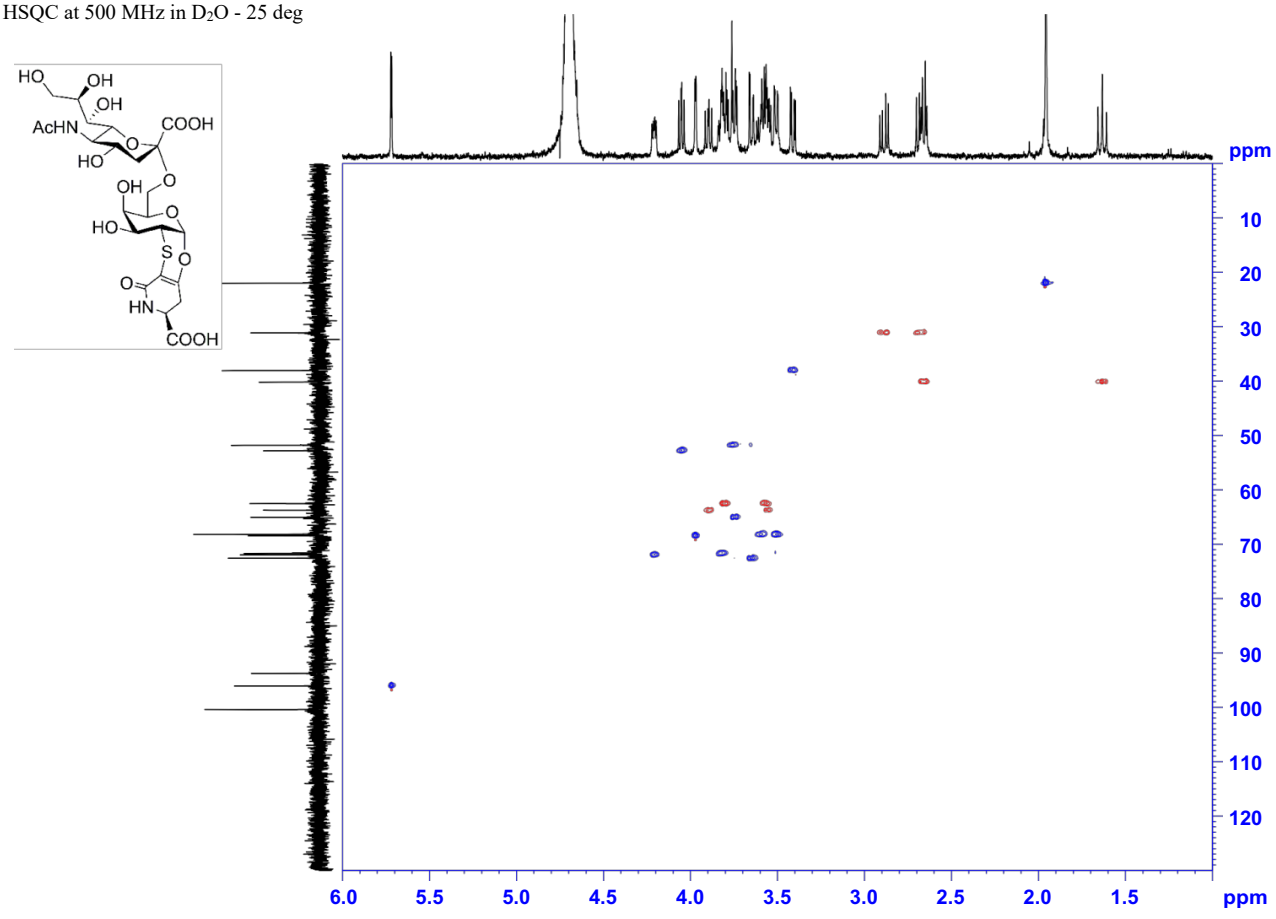
^{13}C at 125 MHz in D_2O - 25 deg



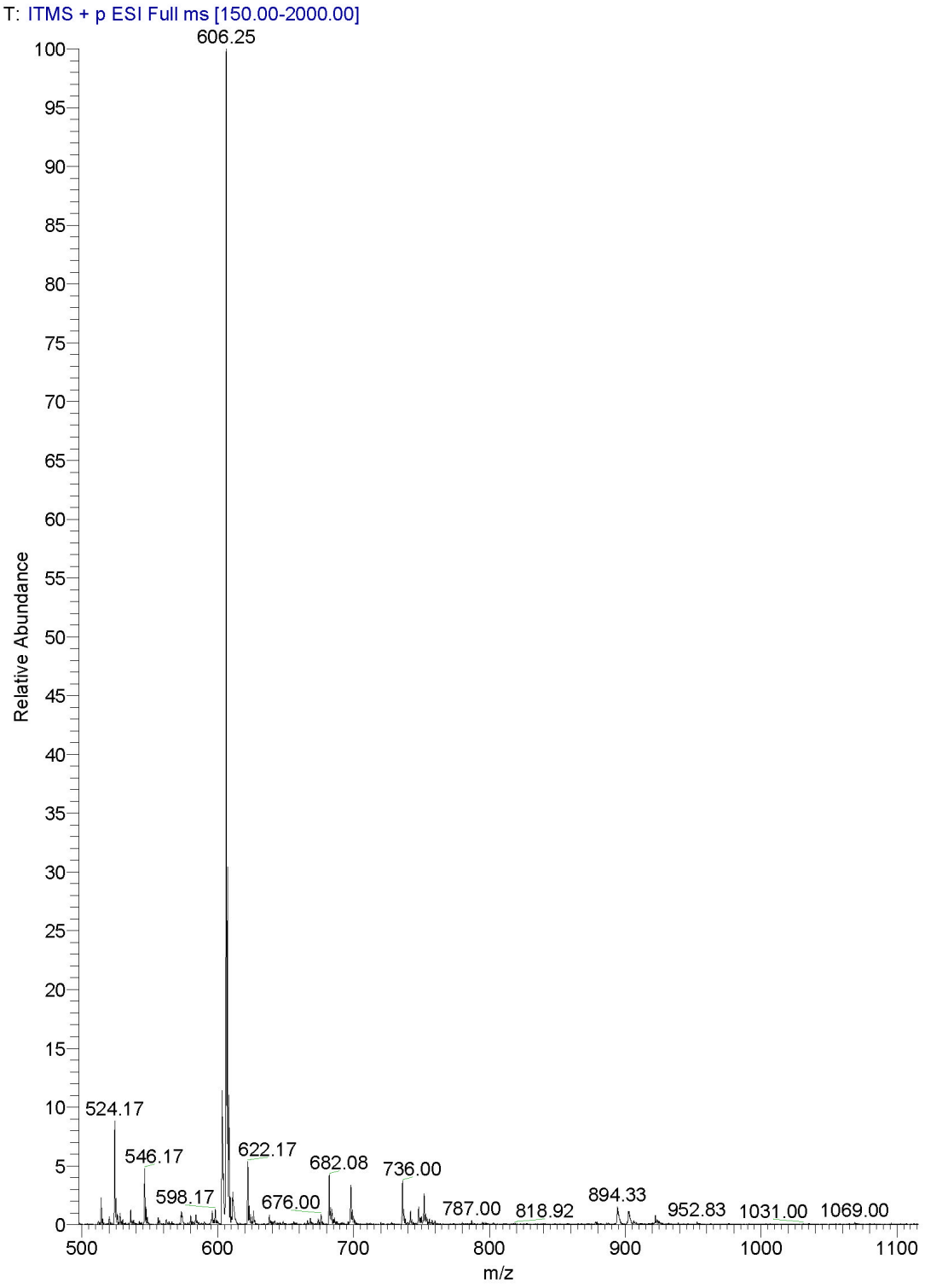
COSY at 500 MHz in D_2O - 25 deg



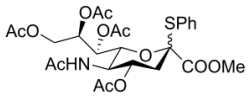
HSQC at 500 MHz in D_2O - 25 deg



NMR and ESI-MS spectra of compound 3a

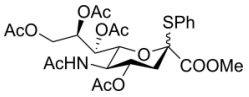


^{1}H at 500 MHz in $CDCl_3$ - 25 deg



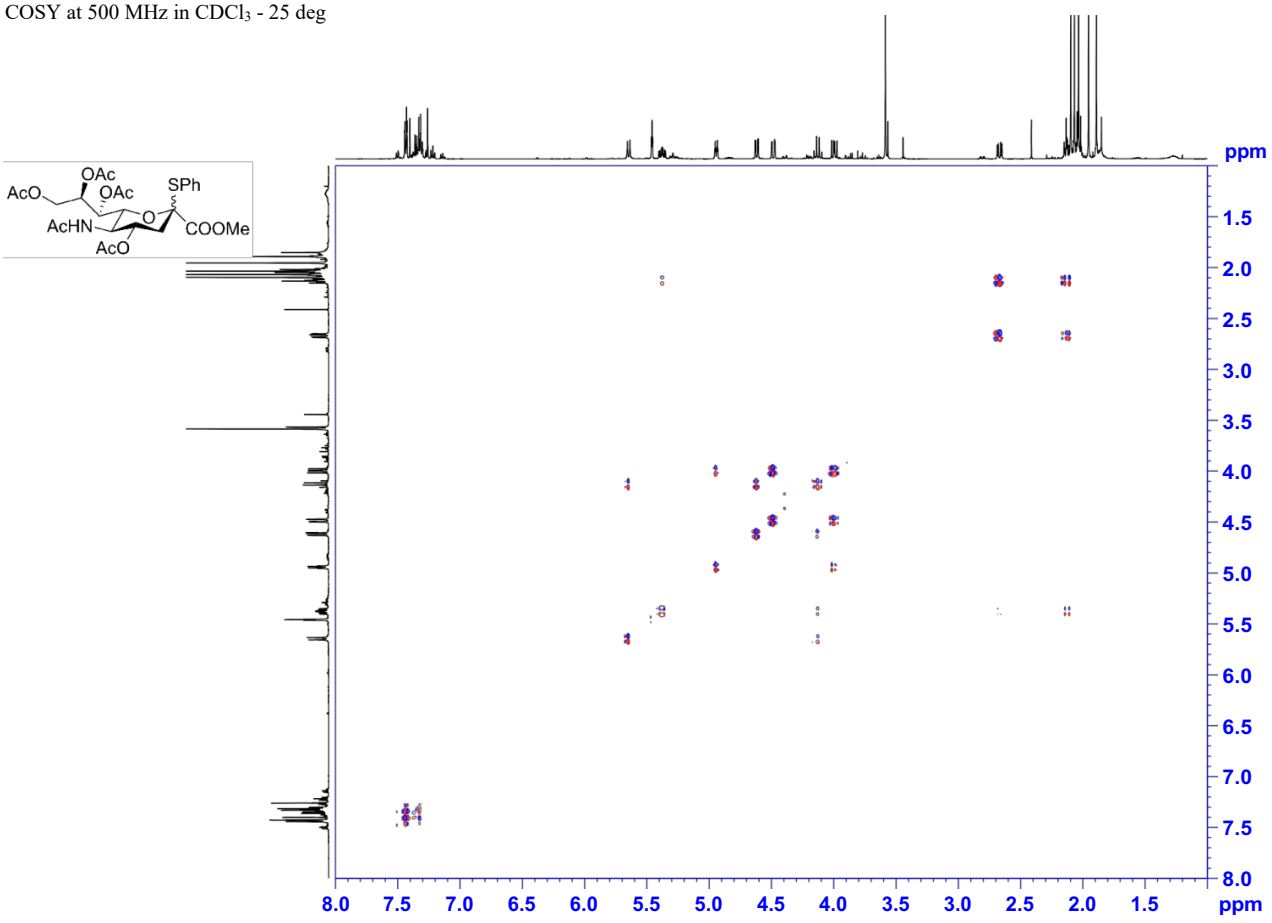
alpha anomer 13%
beta anomer 87%

^{13}C at 125 MHz in $CDCl_3$ - 25 deg

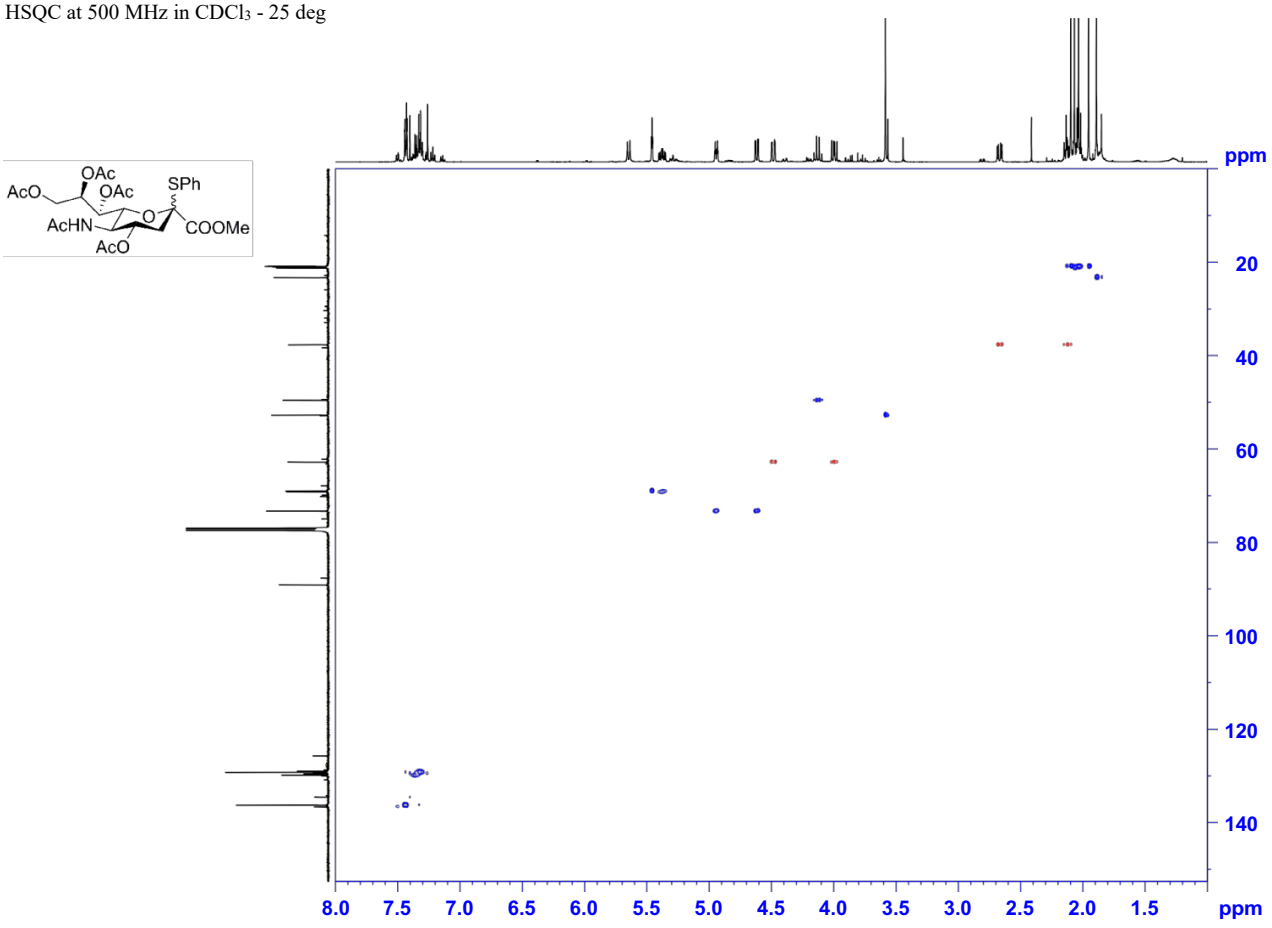


alpha anomer 13%
beta anomer 87%

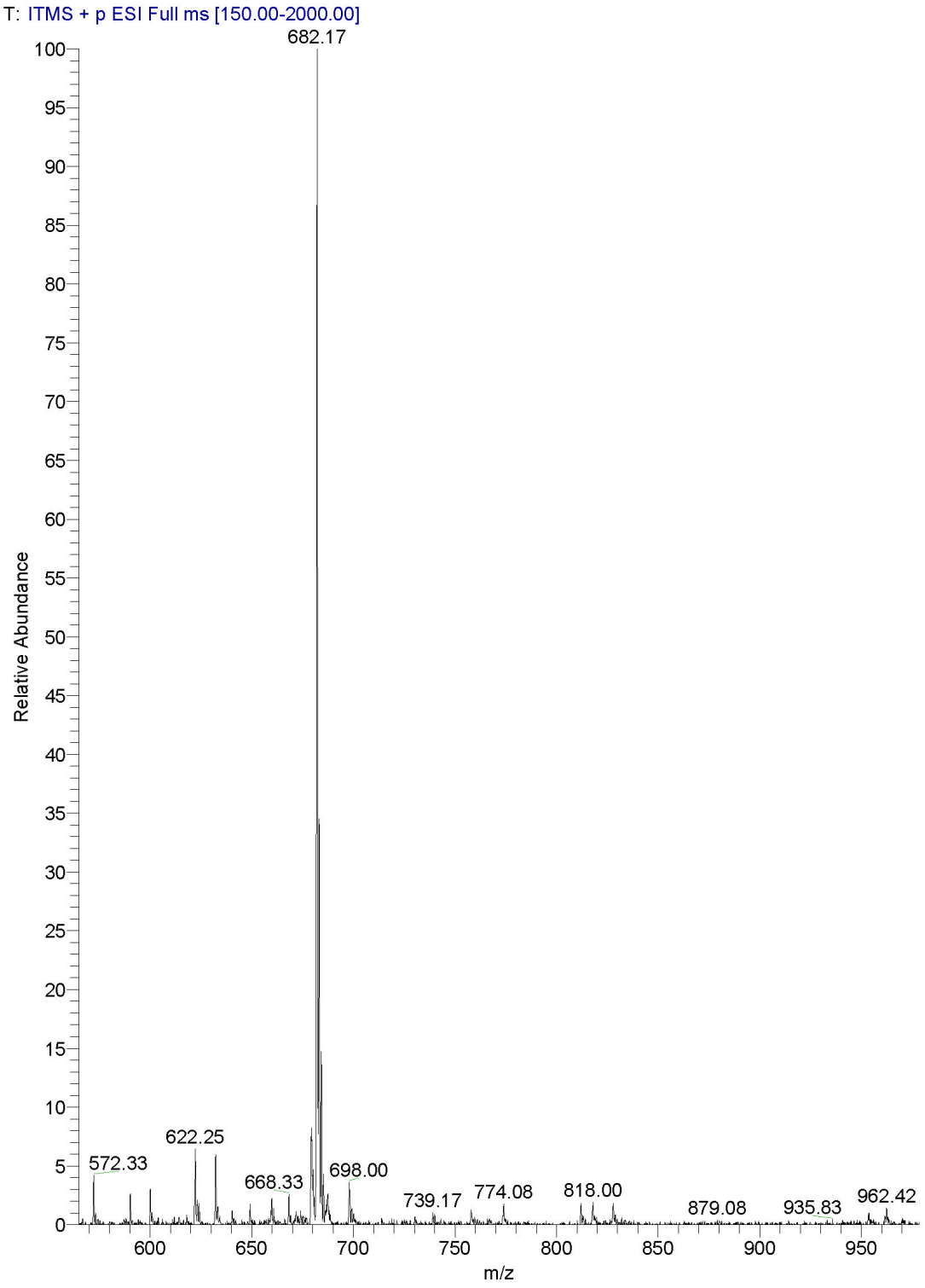
COSY at 500 MHz in $CDCl_3$ - 25 deg



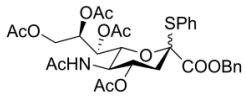
HSQC at 500 MHz in $CDCl_3$ - 25 deg



NMR and ESI-MS spectra of compound 3b

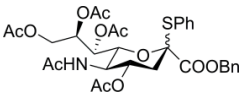


^{1}H at 500 MHz in $CDCl_3$ - 25 deg



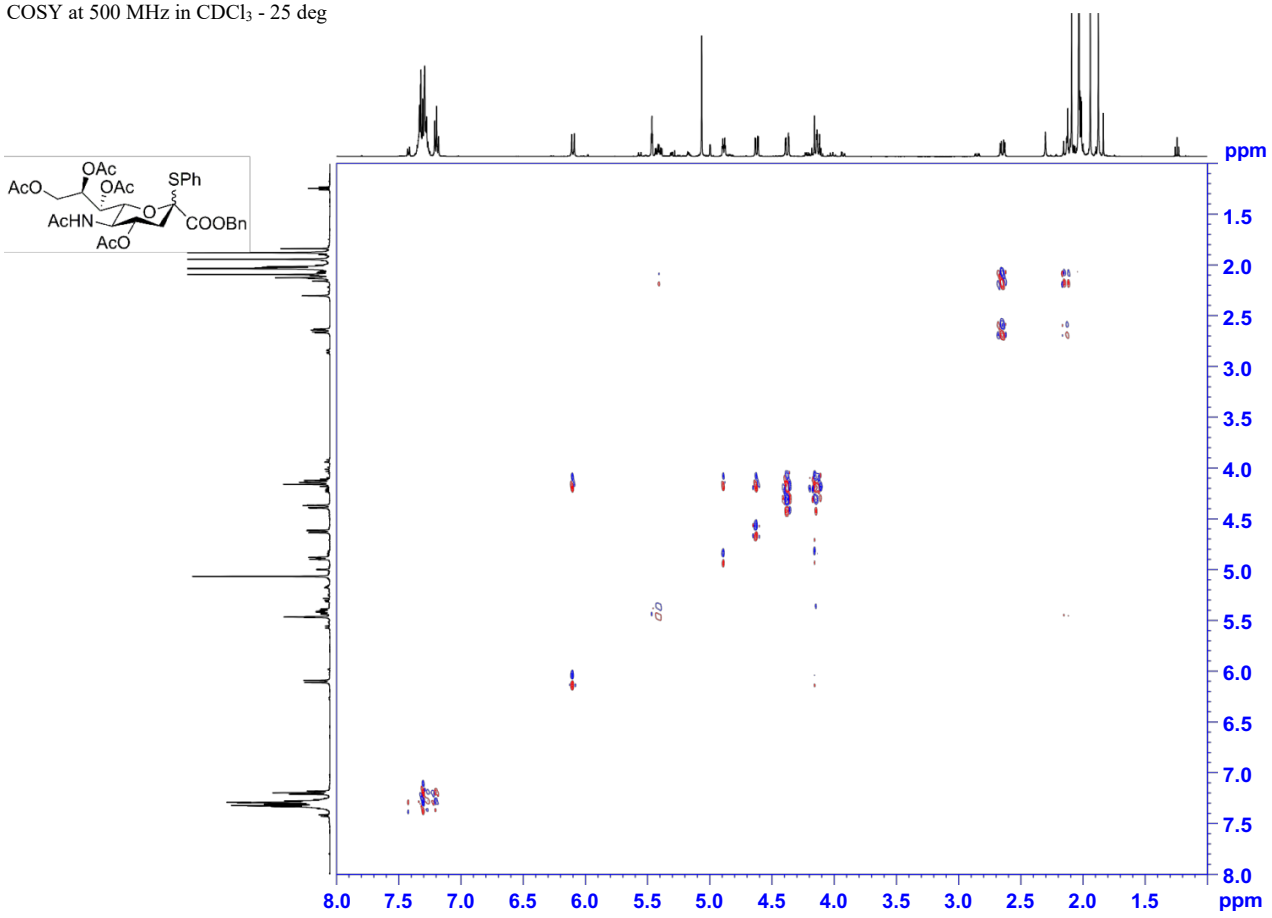
alpha anomer 14%
beta anomer 86%

^{13}C at 125 MHz in $CDCl_3$ - 25 deg

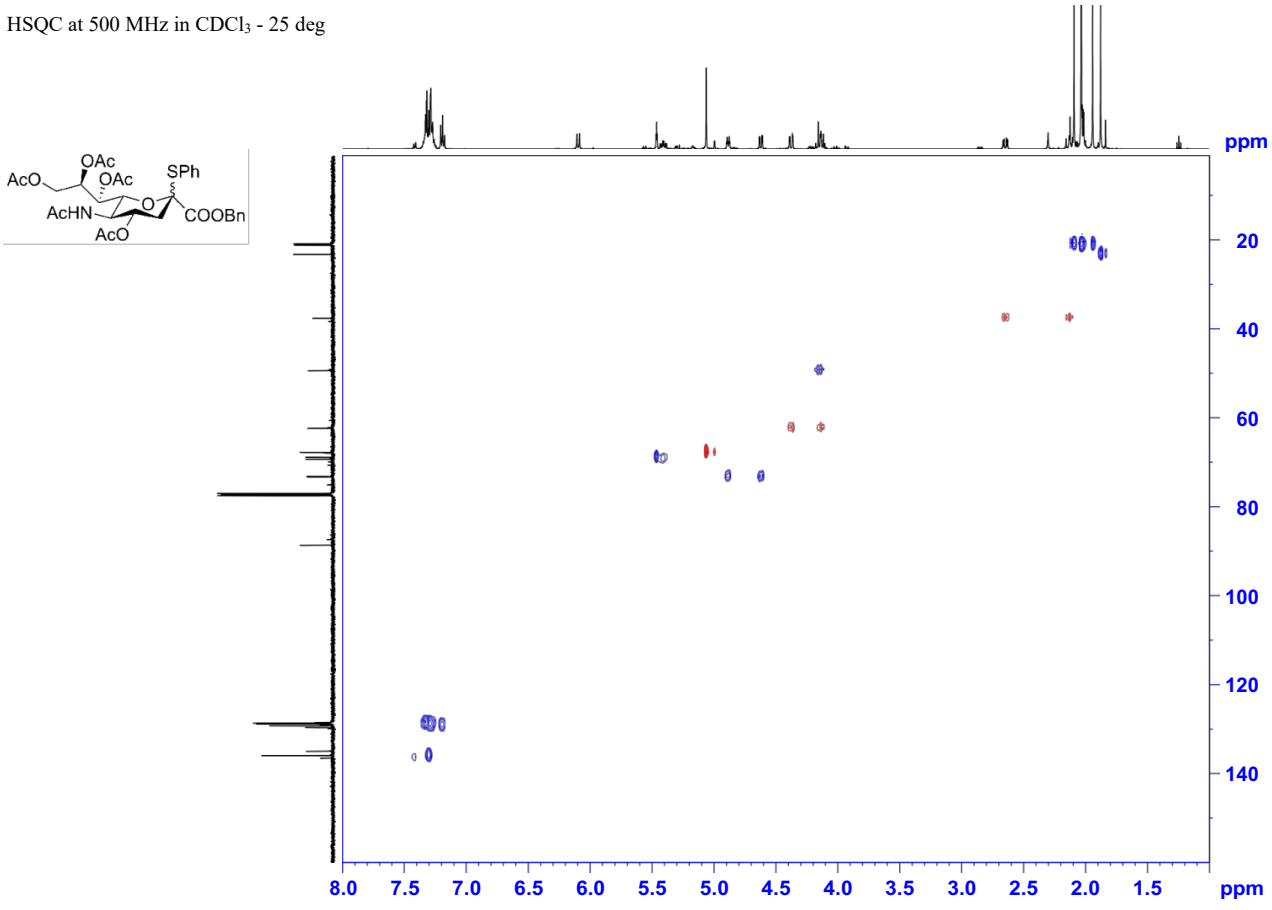


alpha anomer 14%
beta anomer 86%

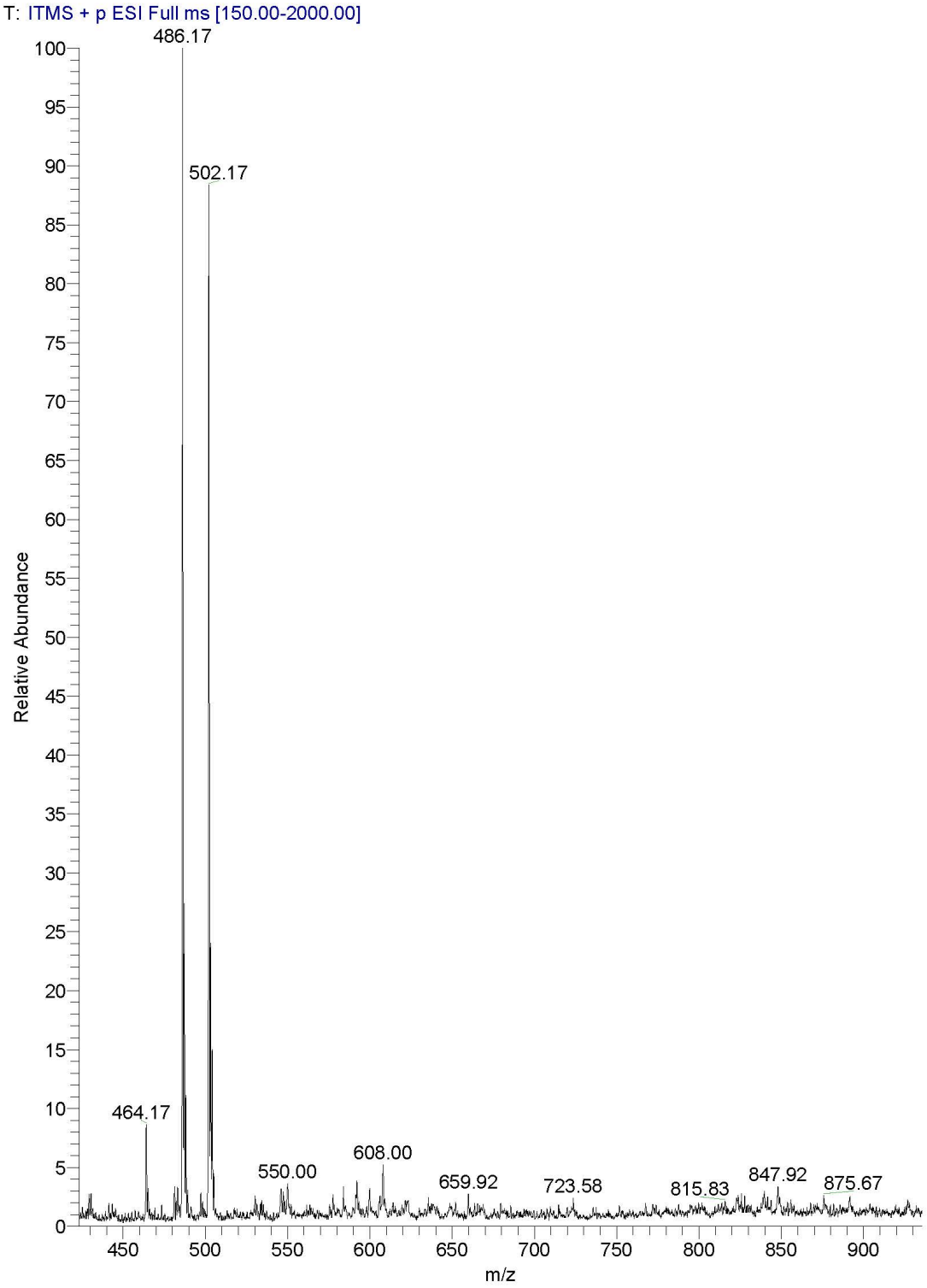
COSY at 500 MHz in $CDCl_3$ - 25 deg



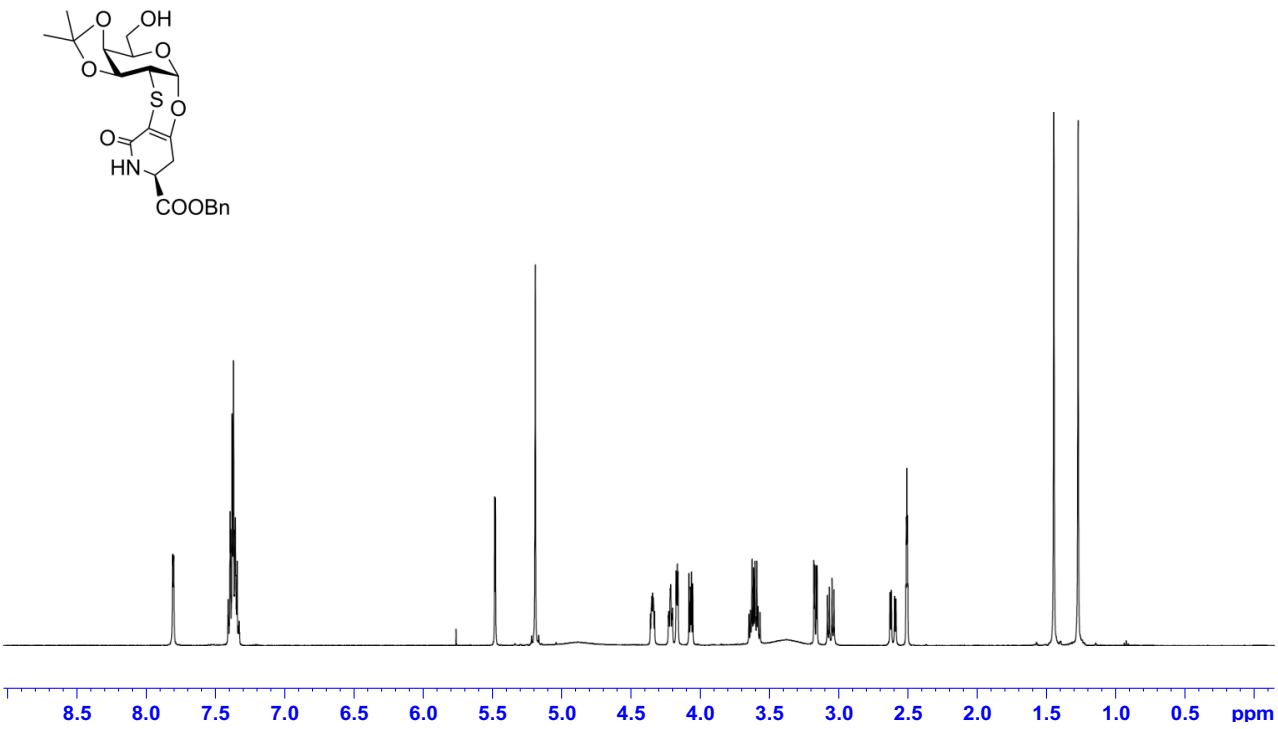
HSQC at 500 MHz in $CDCl_3$ - 25 deg



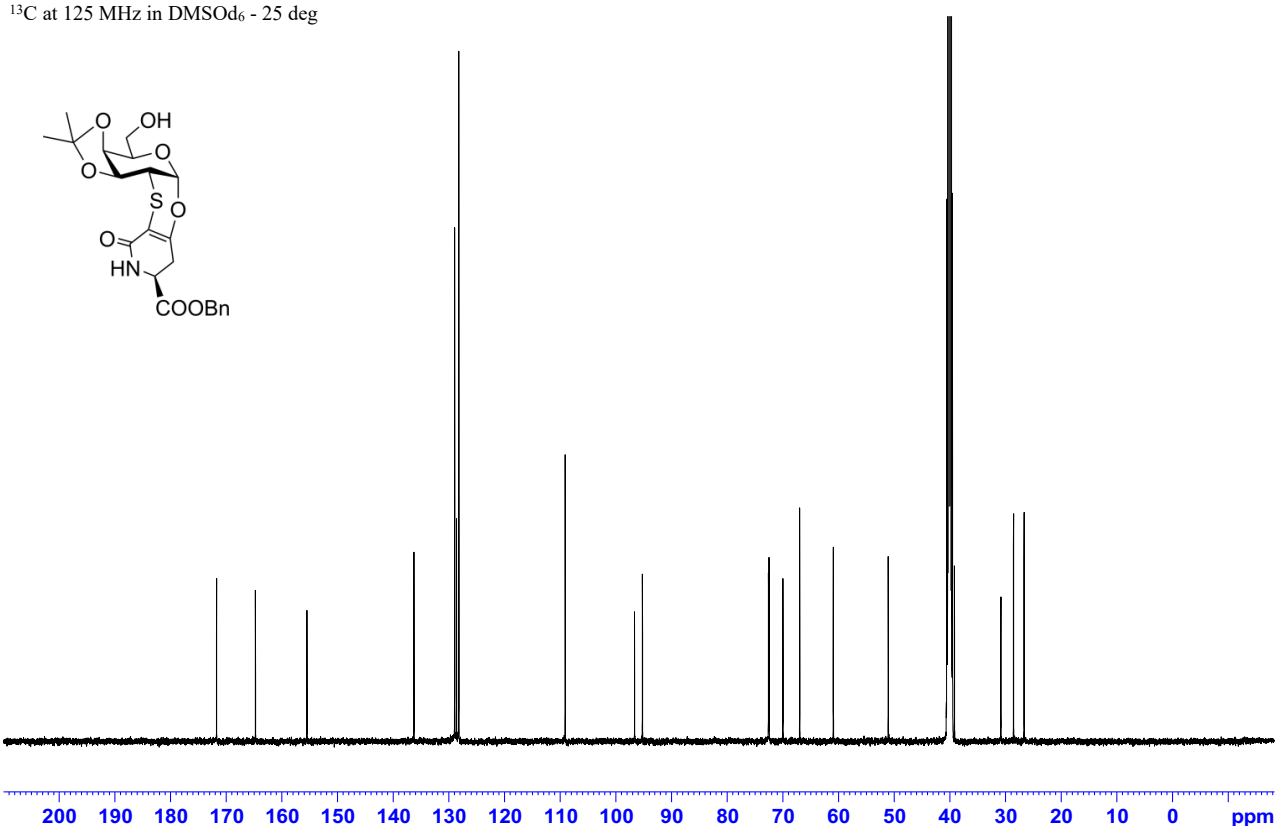
NMR and ESI-MS spectra of compound 4



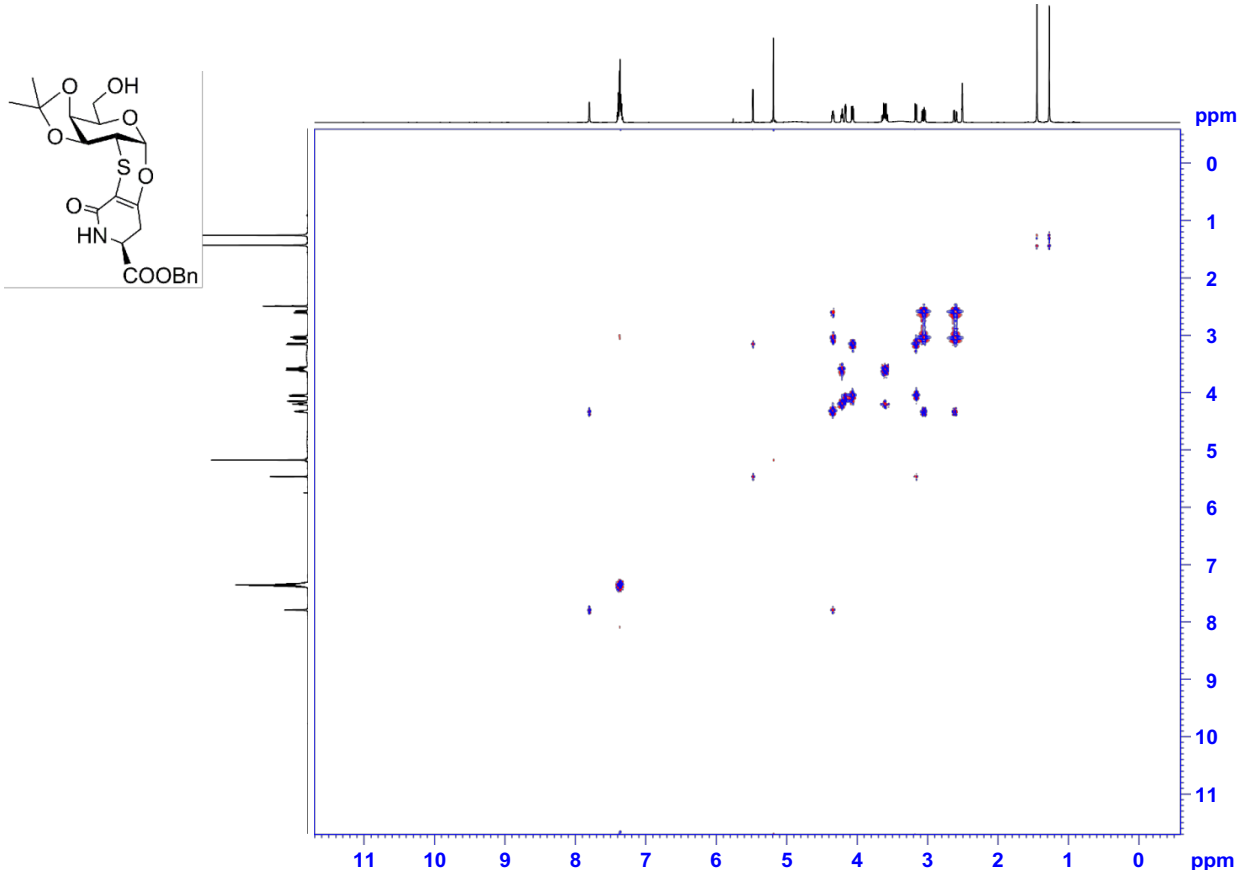
^{1}H at 500 MHz in DMSOd_6 - 25 deg



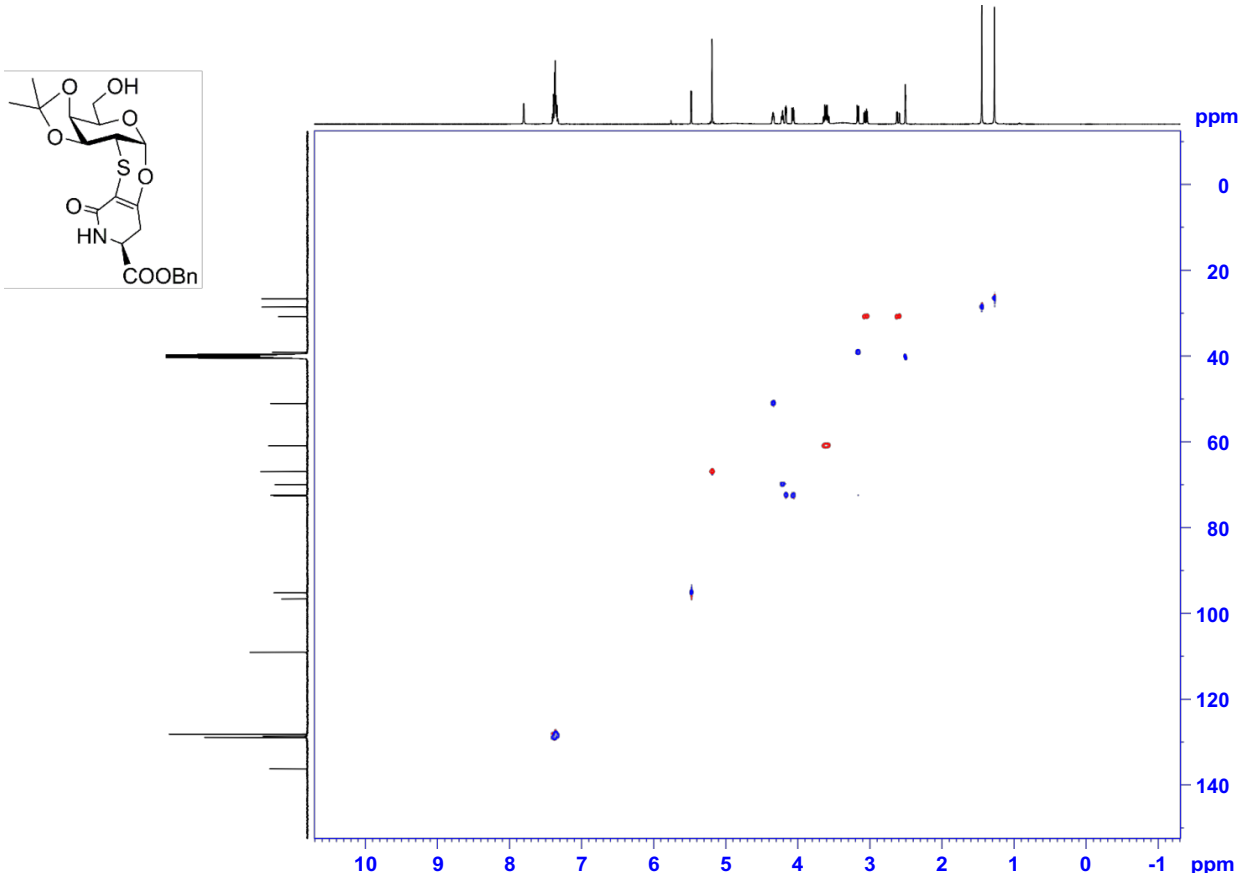
^{13}C at 125 MHz in DMSOd_6 - 25 deg



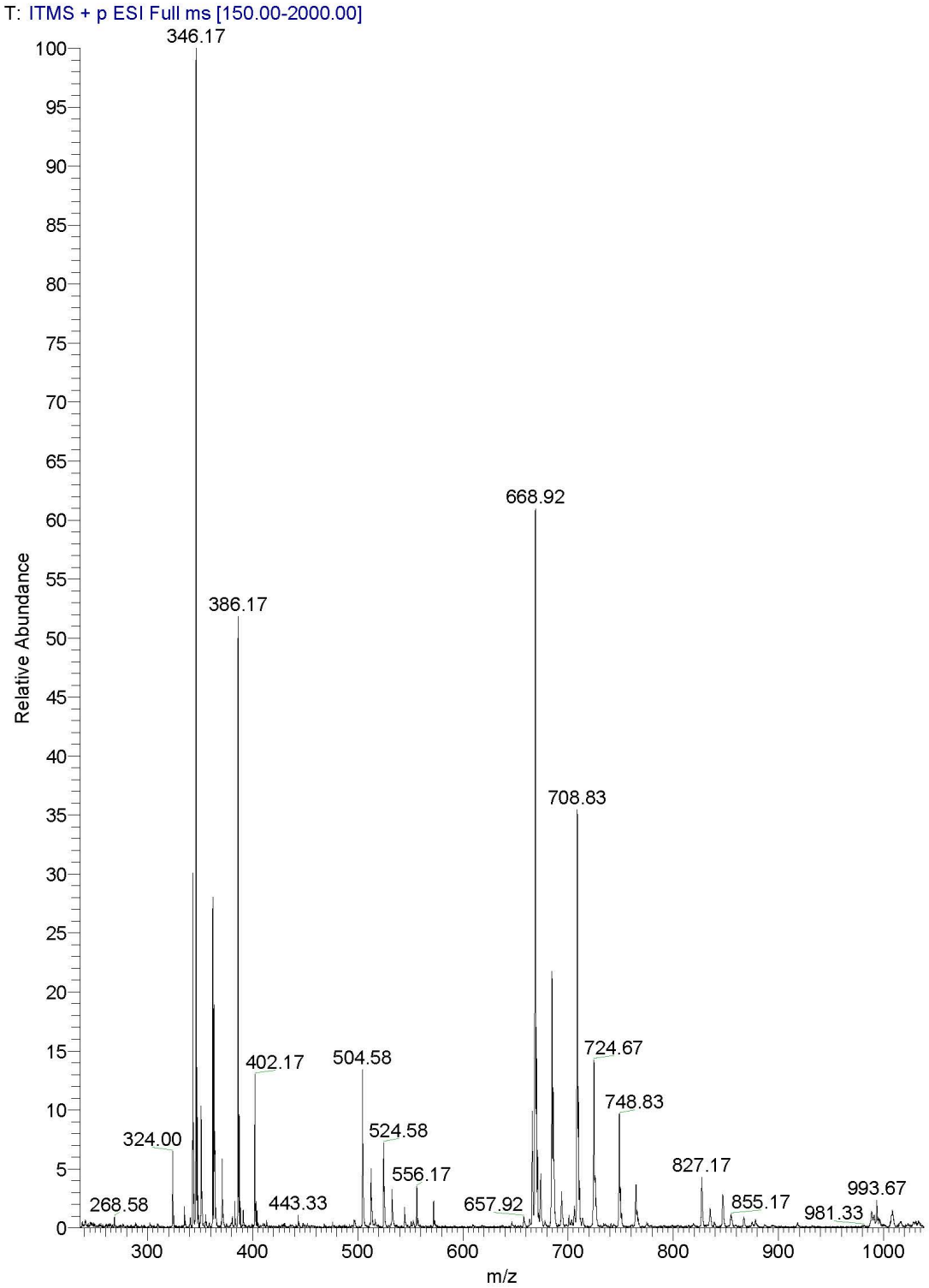
COSY at 500 MHz in $DMSOd_6$ - 25 deg



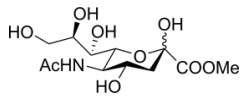
HSQC at 500 MHz in $DMSOd_6$ - 25 deg



NMR and ESI-MS spectra of compound 6a

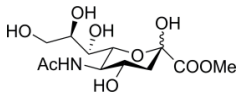


^{1}H at 500 MHz in CD_3OD - 25 deg



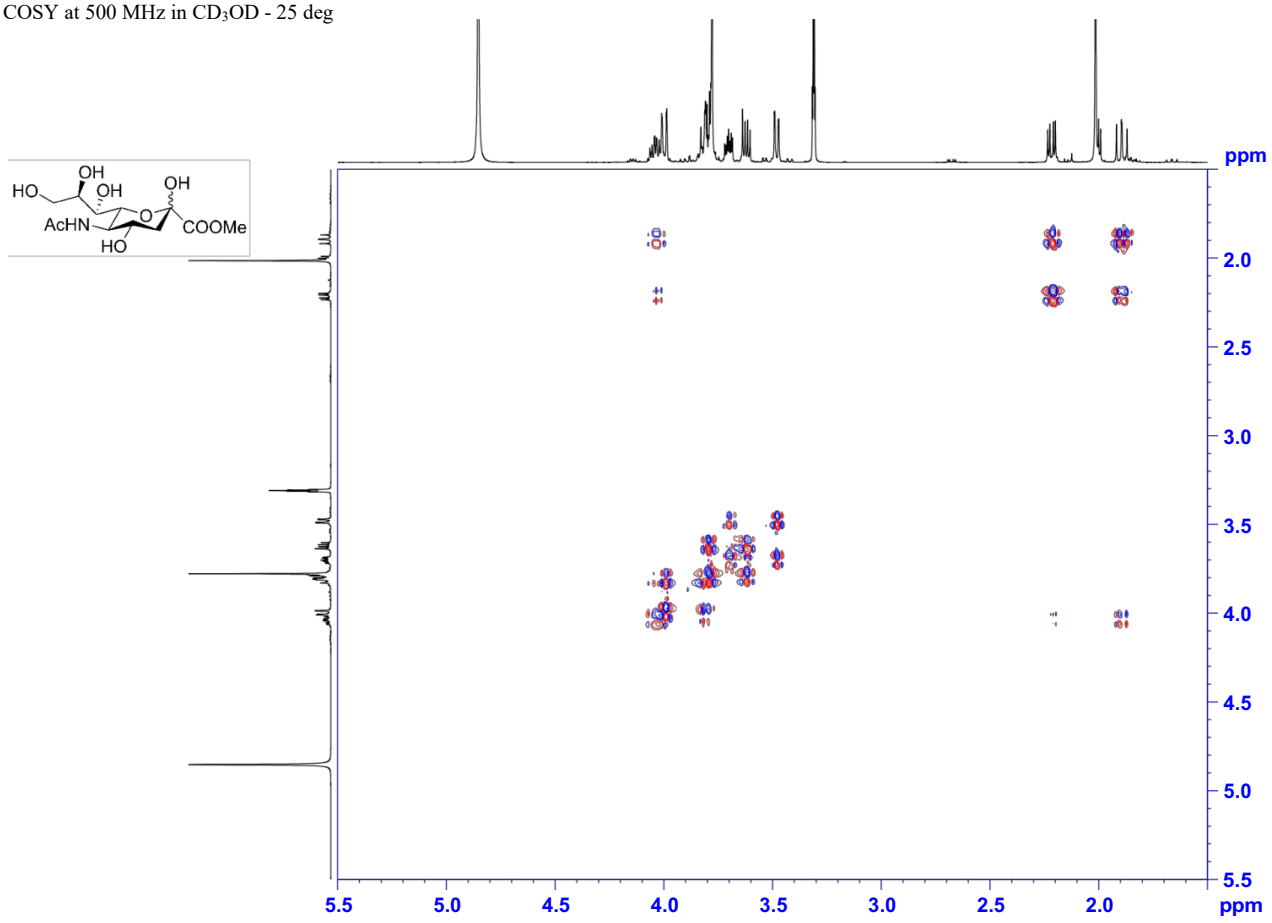
alpha anomer 7%
beta anomer 93%

^{13}C at 125 MHz in CD_3OD - 25 deg

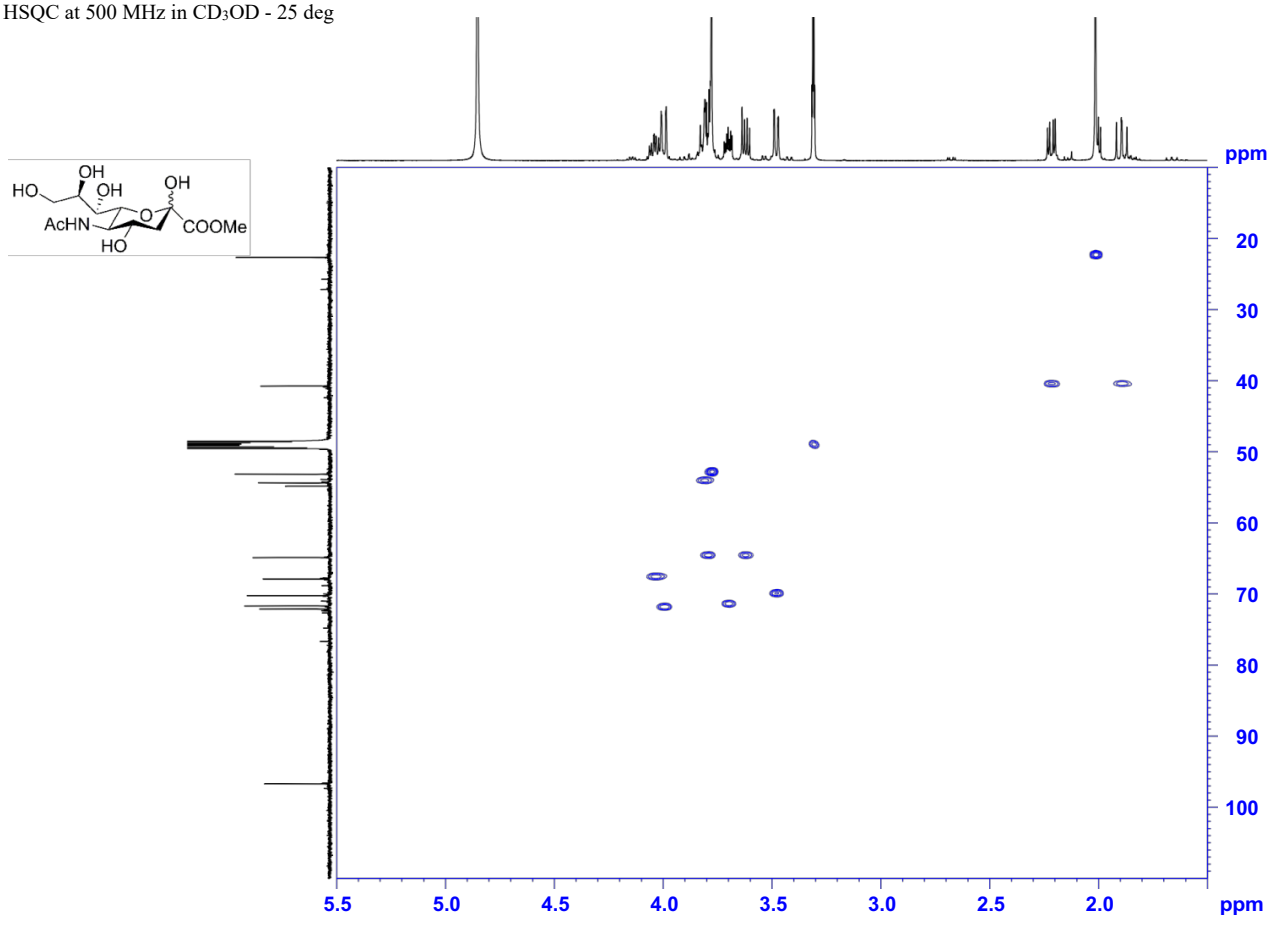


alpha anomer 7%
beta anomer 93%

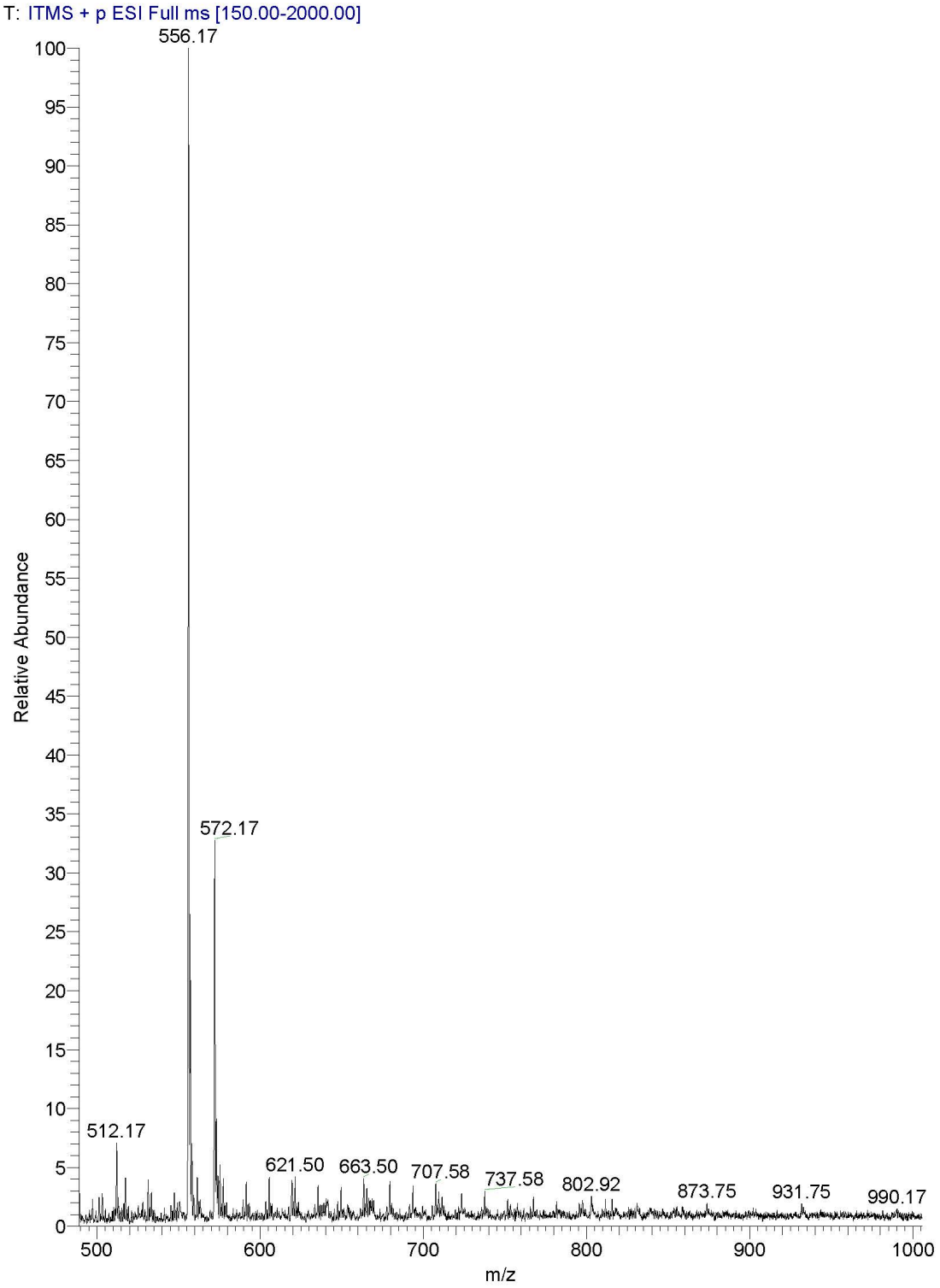
COSY at 500 MHz in CD_3OD - 25 deg



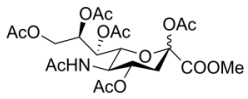
HSQC at 500 MHz in CD_3OD - 25 deg



NMR and ESI-MS spectra of compound 7a

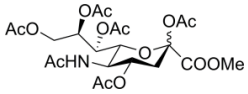


^{1}H at 500 MHz in $CDCl_3$ - 25 deg



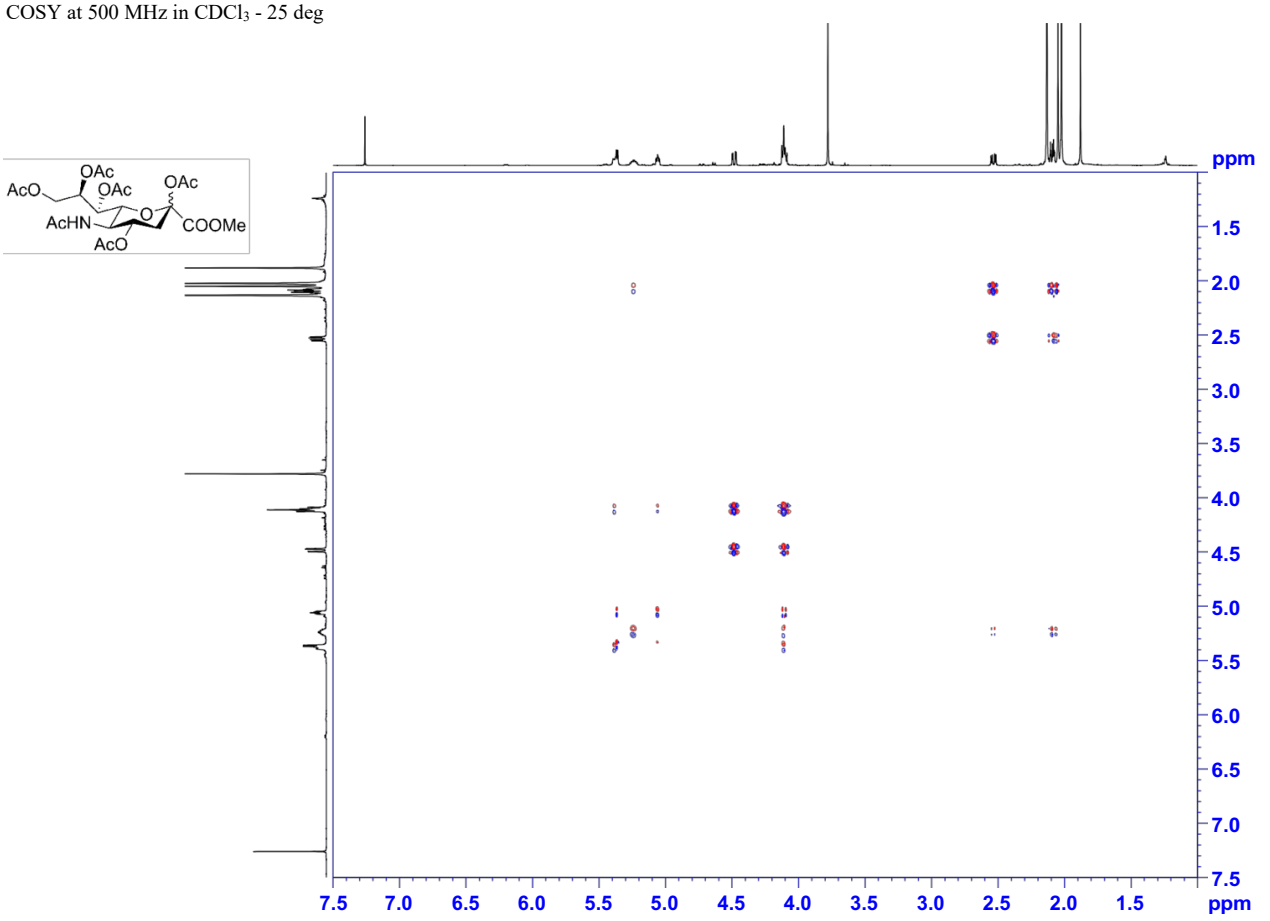
beta anomer > 90%

^{13}C at 125 MHz in $CDCl_3$ - 25 deg

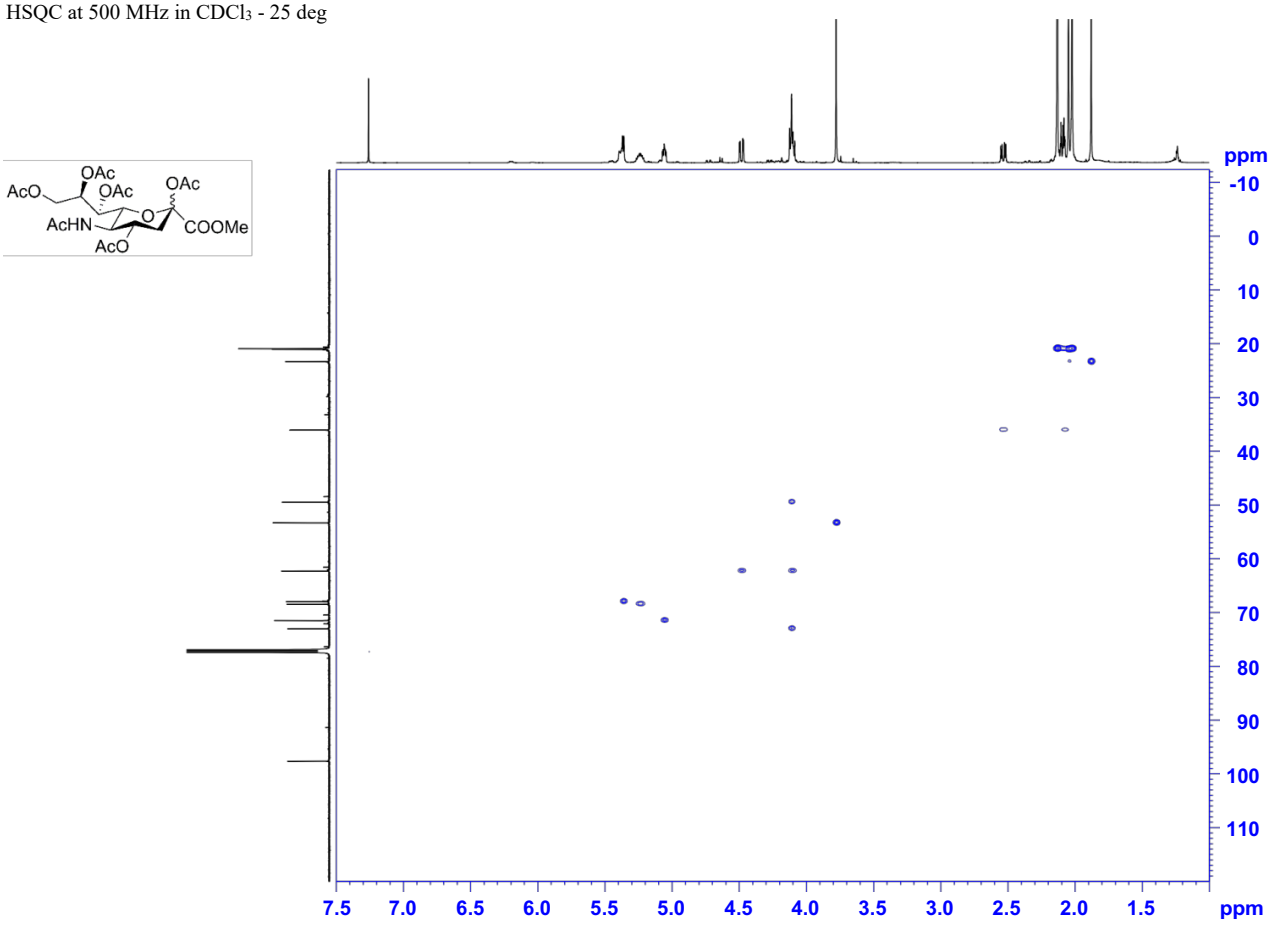


beta anomer > 90%

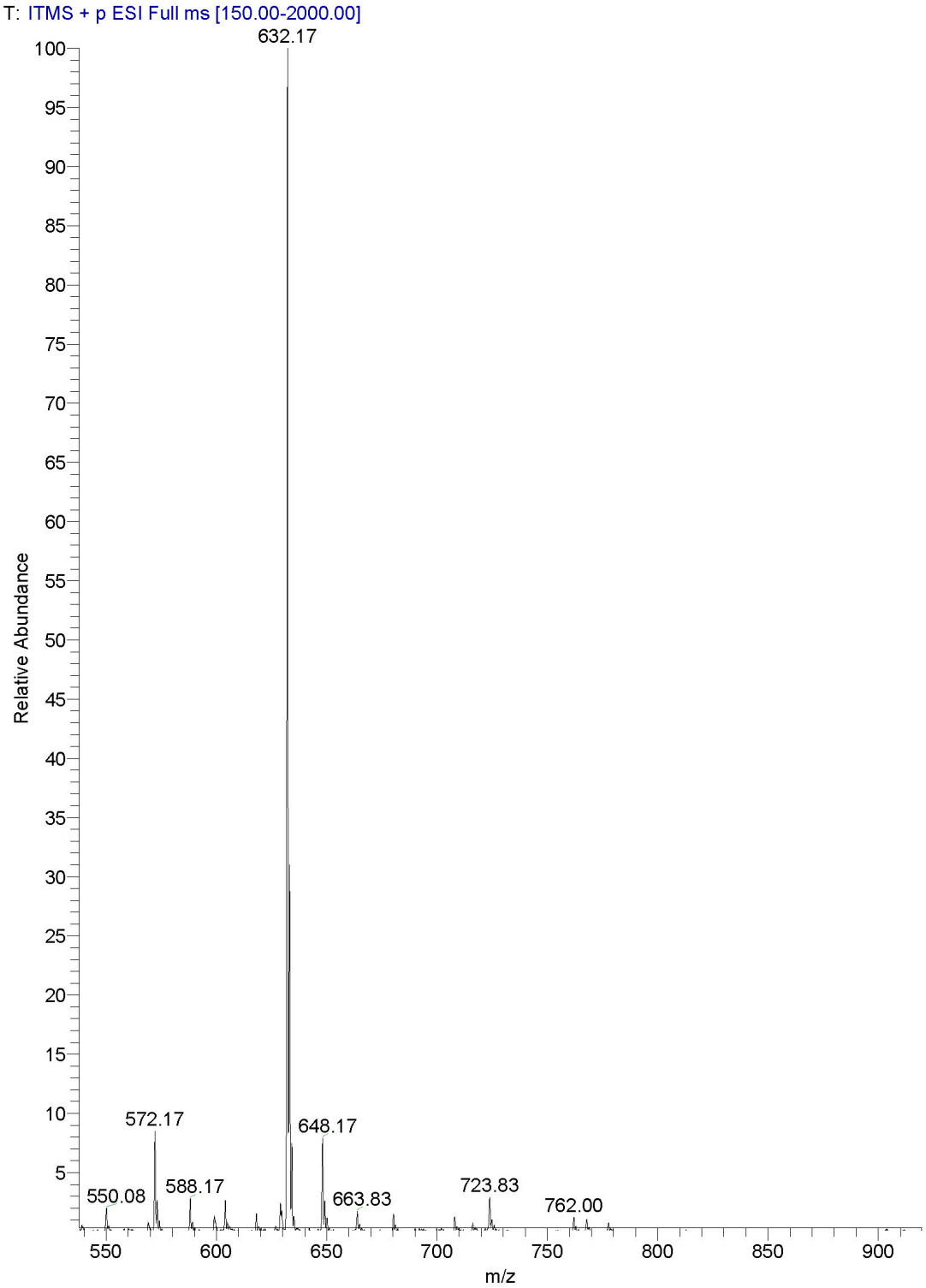
COSY at 500 MHz in $CDCl_3$ - 25 deg



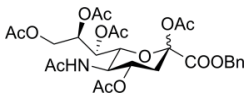
HSQC at 500 MHz in $CDCl_3$ - 25 deg



NMR and ESI-MS spectra of compound 7b

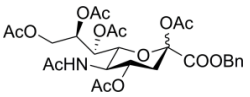


^{1}H at 500 MHz in $CDCl_3$ - 25 deg



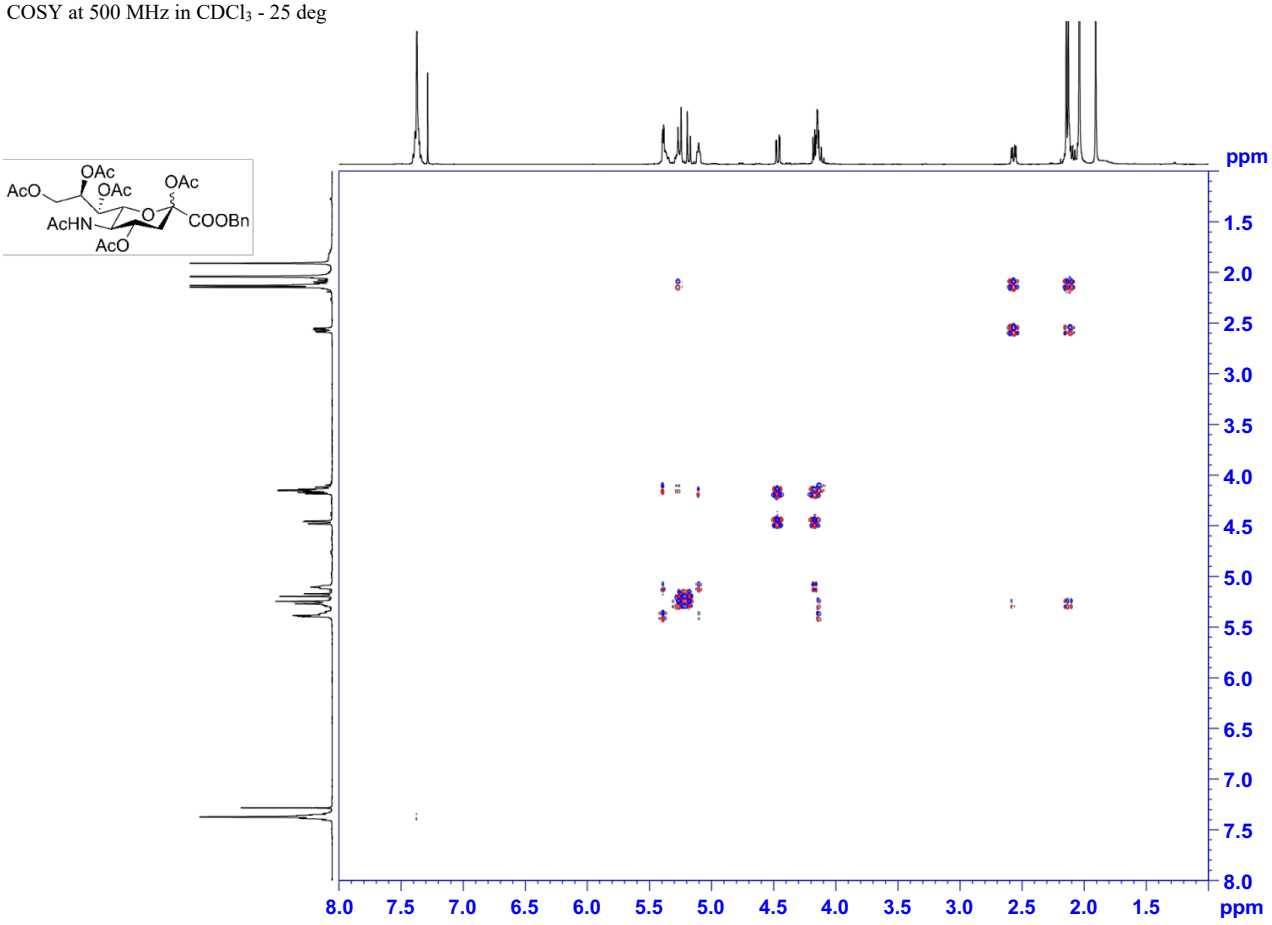
beta anomer > 90%

^{13}C at 125 MHz in $CDCl_3$ - 25 deg

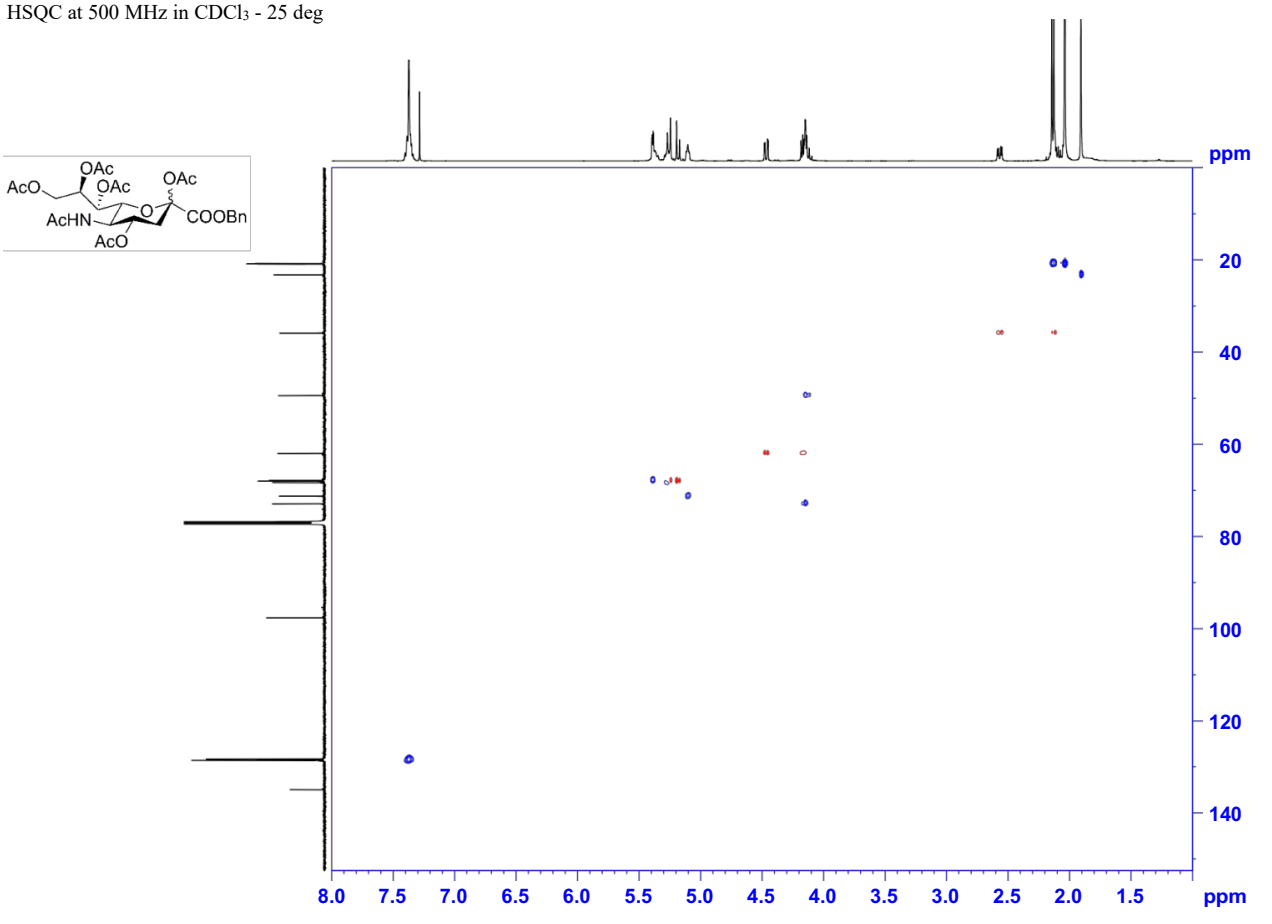


beta anomer > 90%

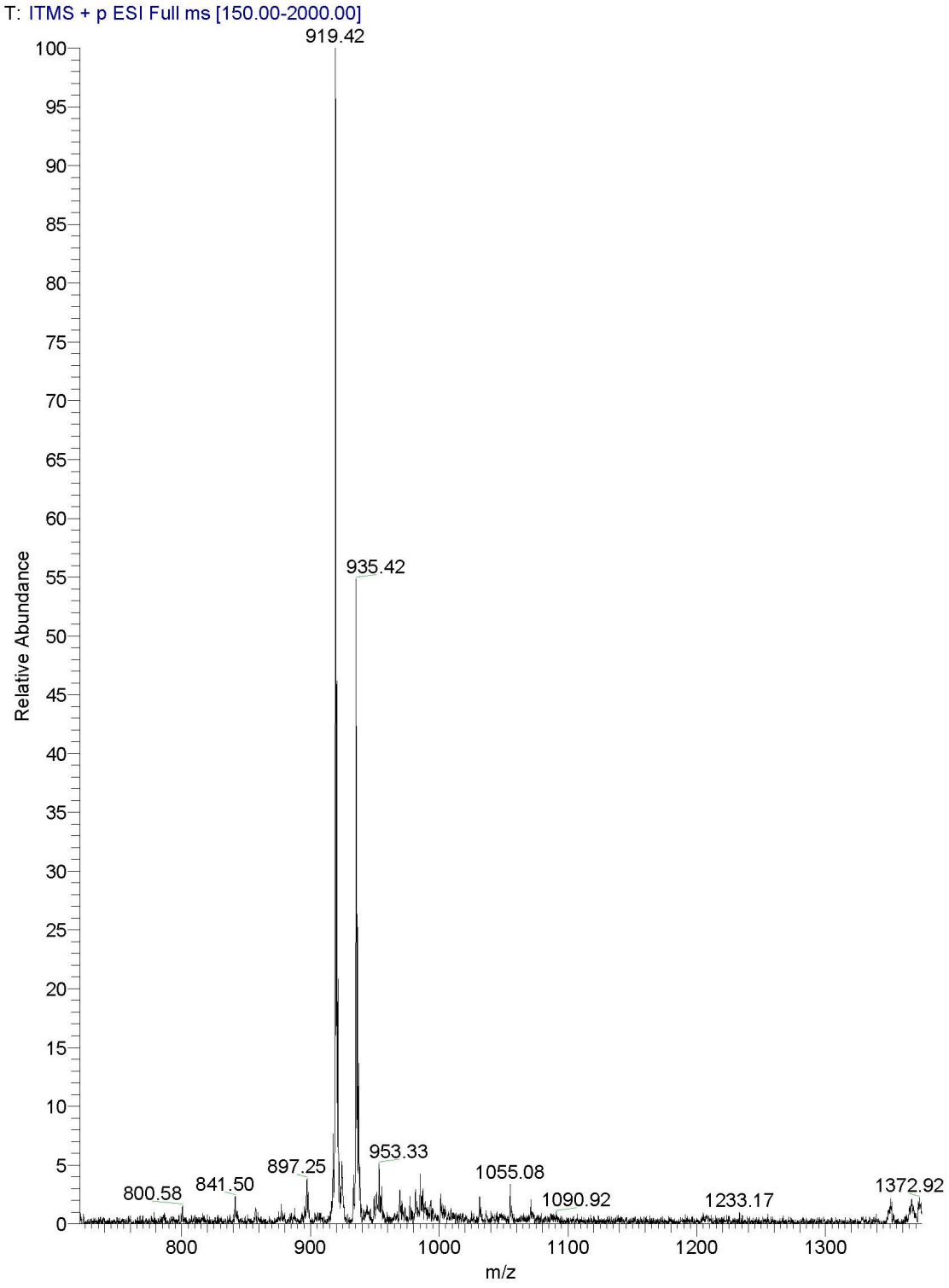
COSY at 500 MHz in $CDCl_3$ - 25 deg



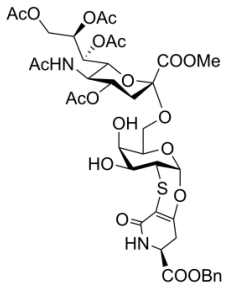
HSQC at 500 MHz in $CDCl_3$ - 25 deg



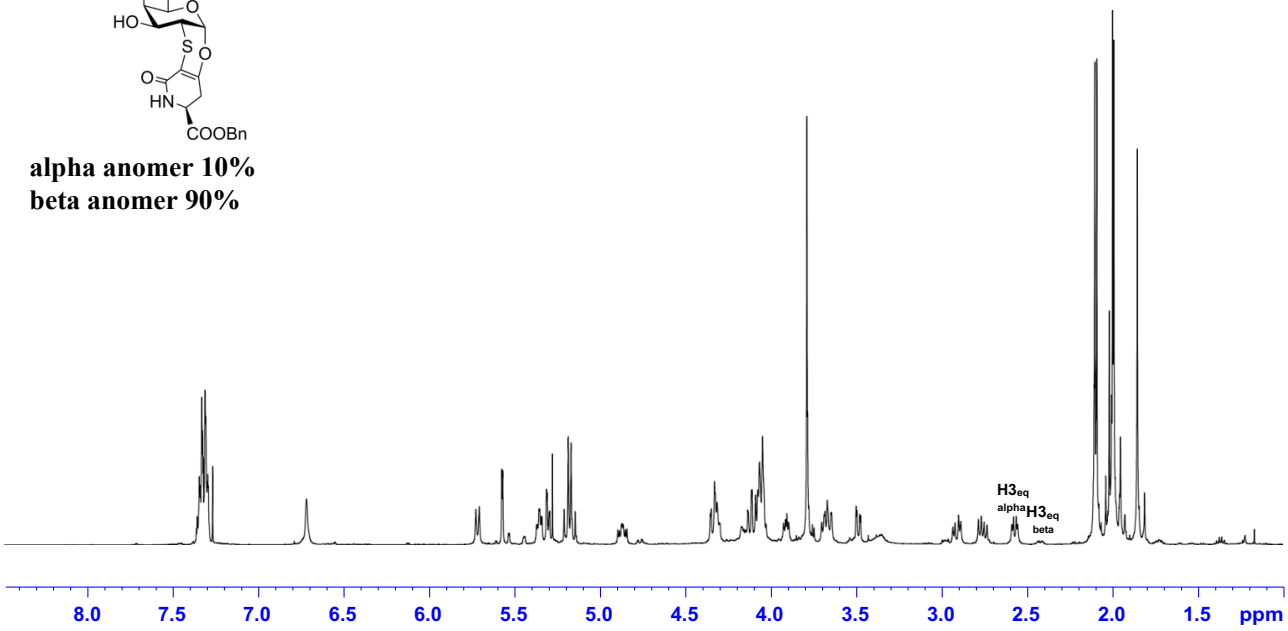
NMR and ESI-MS spectra of compound 8a



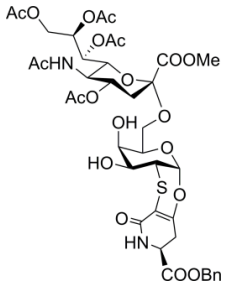
^{1}H at 500 MHz in $CDCl_3$ - 25 deg



alpha anomer 10%
beta anomer 90%

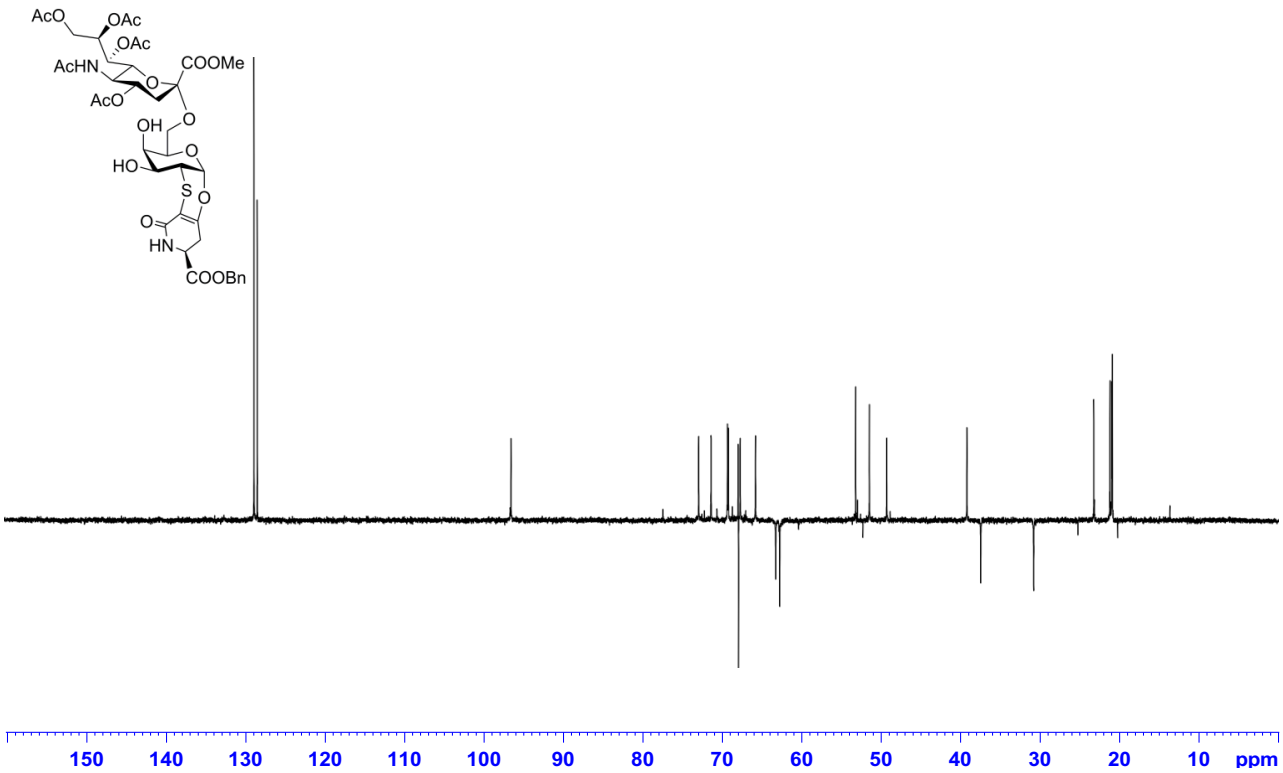


^{13}C at 125 MHz in $CDCl_3$ - 25 deg

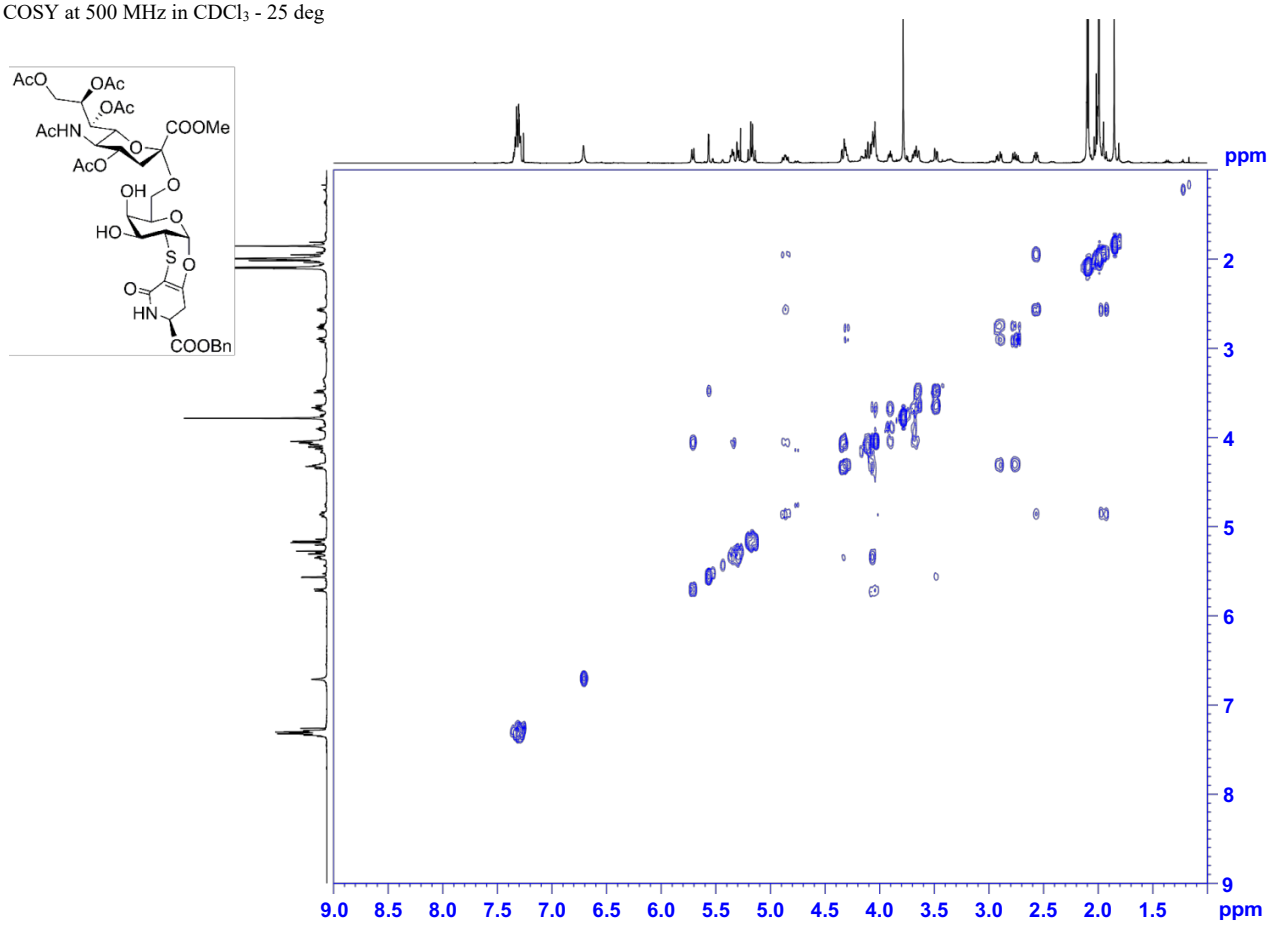


alpha anomer 10%
beta anomer 90%

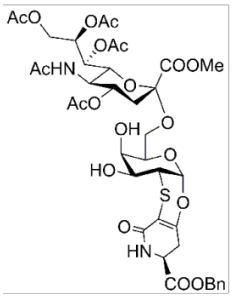
DEPT at 125 MHz in $CDCl_3$ - 25 deg

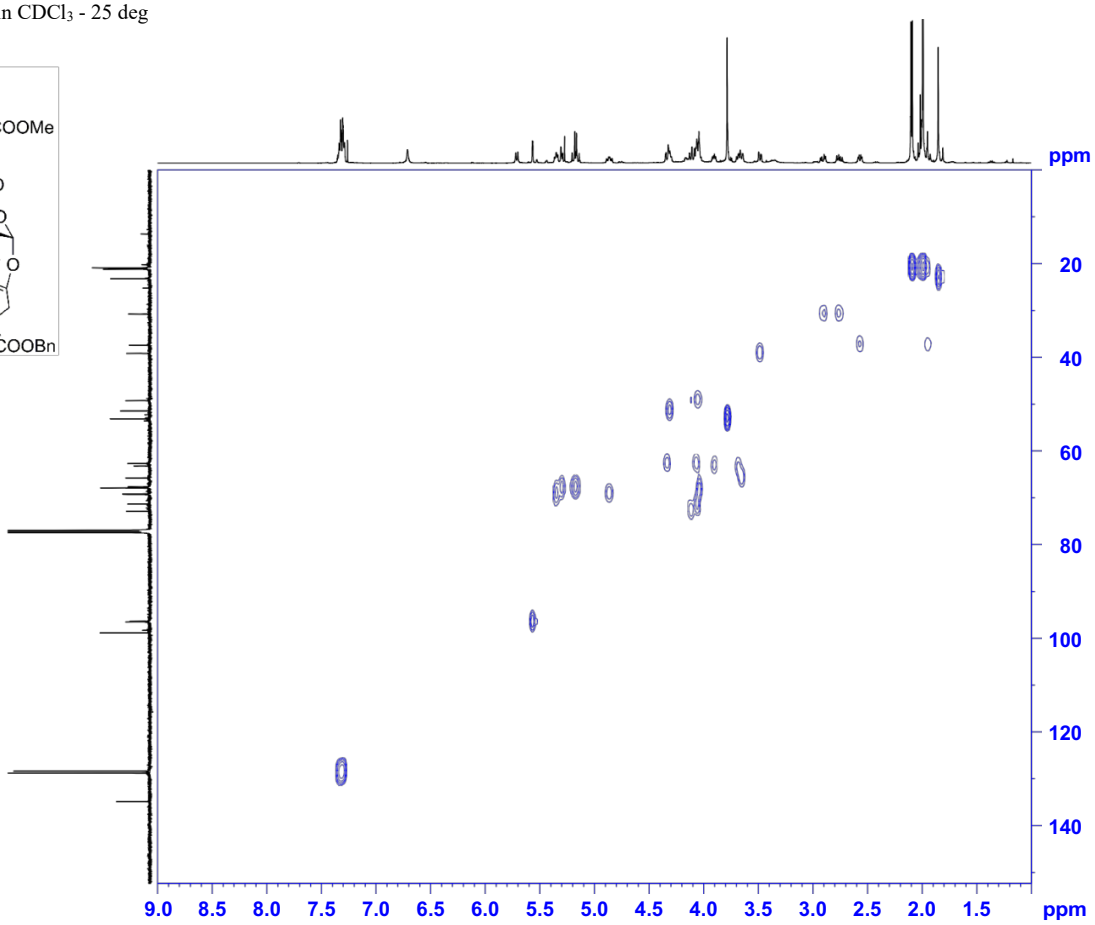


COSY at 500 MHz in $CDCl_3$ - 25 deg

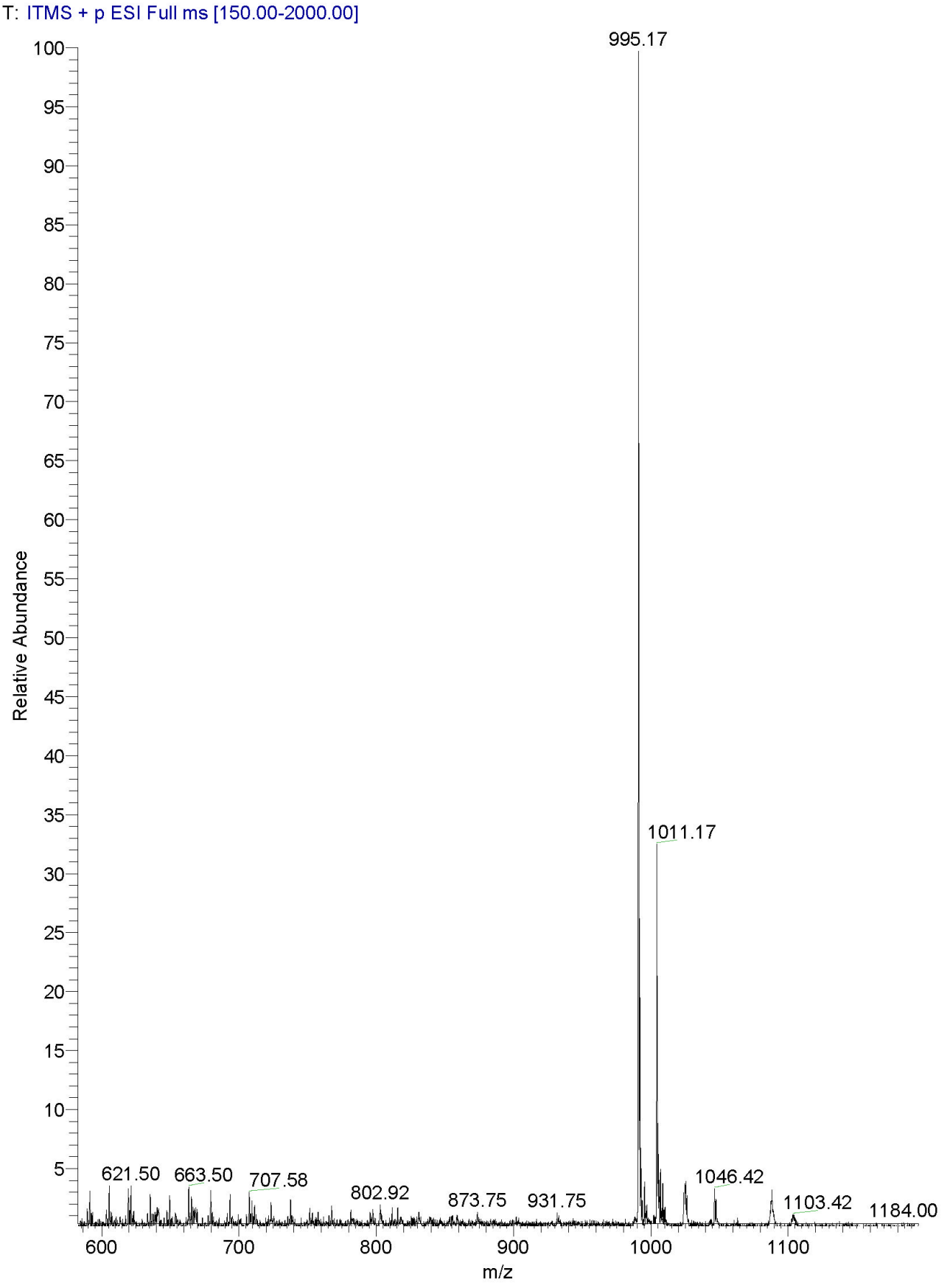


HSQC at 500 MHz in $CDCl_3$ - 25 deg

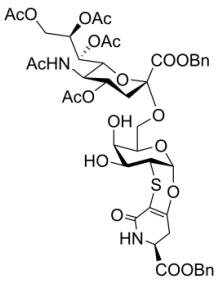




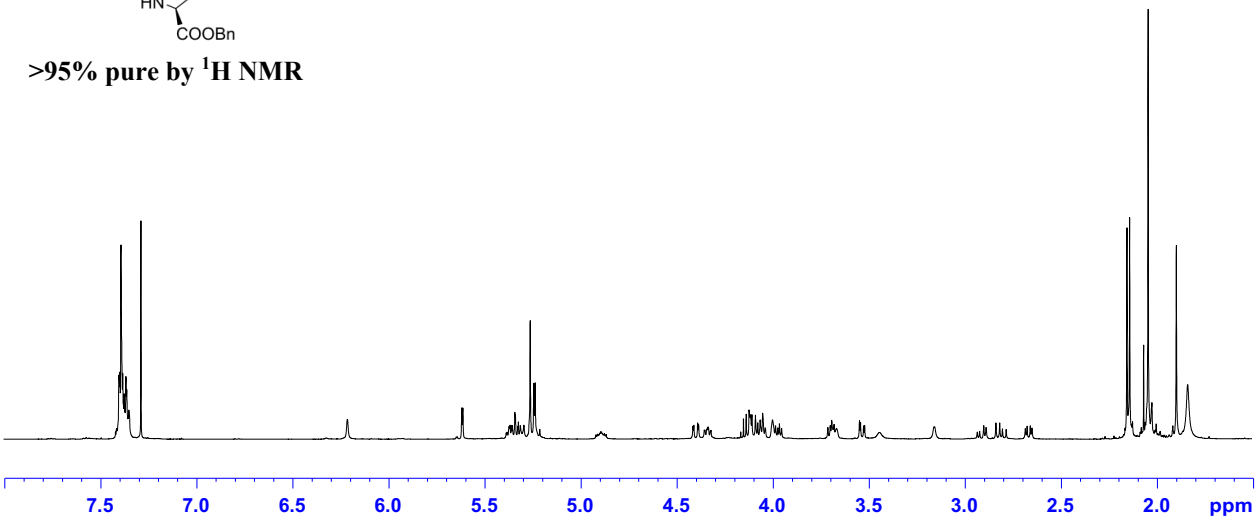
NMR and ESI-MS spectra of compound 8b



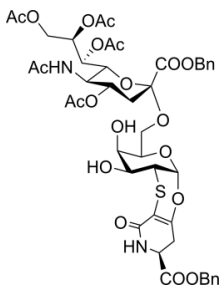
^{1}H at 500 MHz in $CDCl_3$ - 25 deg



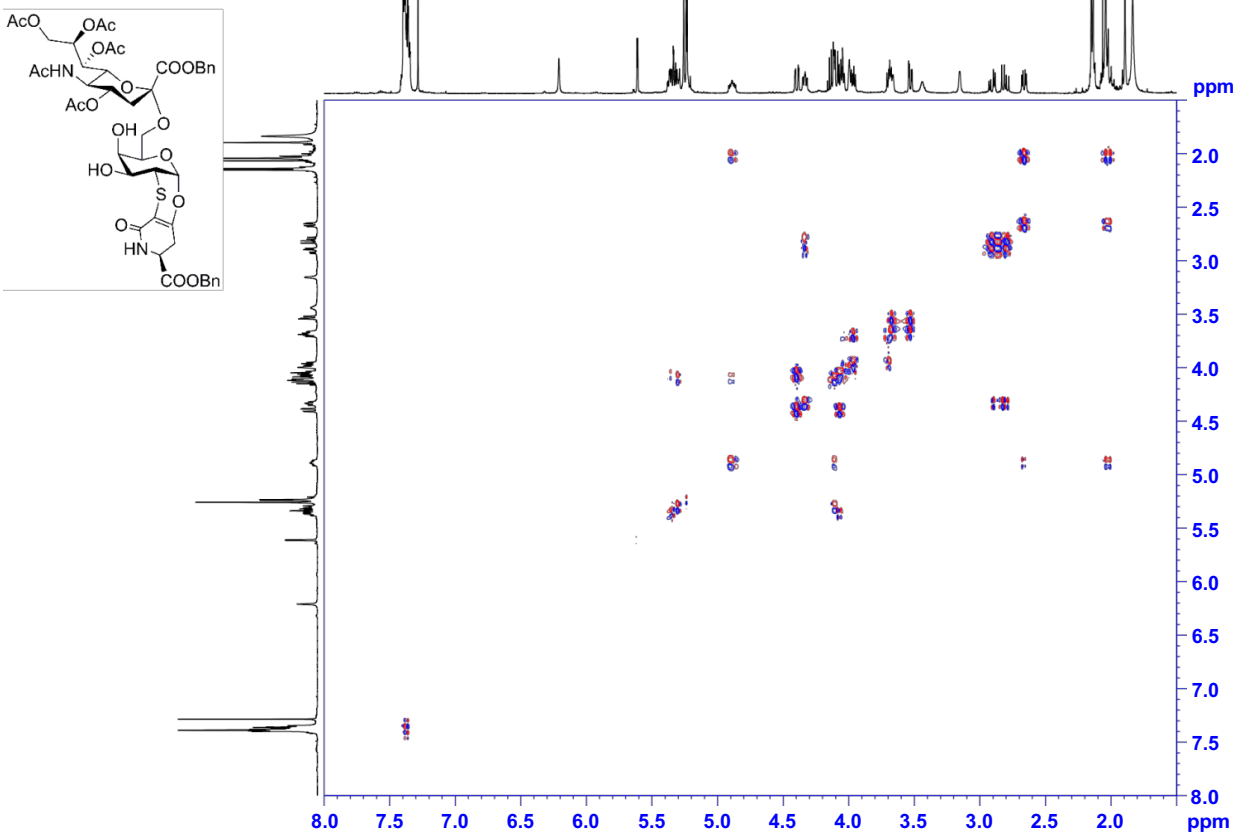
>95% pure by ^{1}H NMR



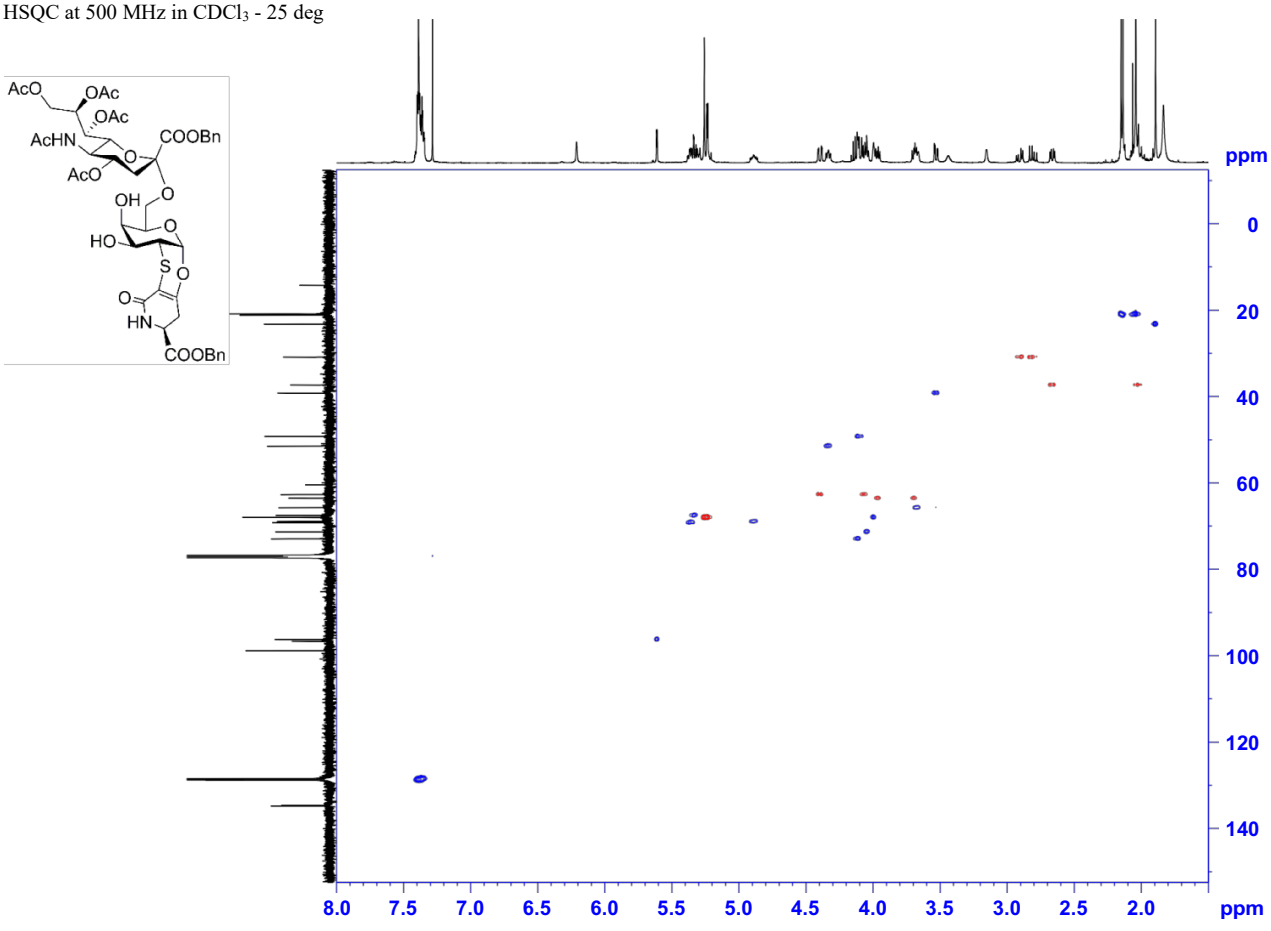
^{13}C at 125 MHz in $CDCl_3$ - 25 deg



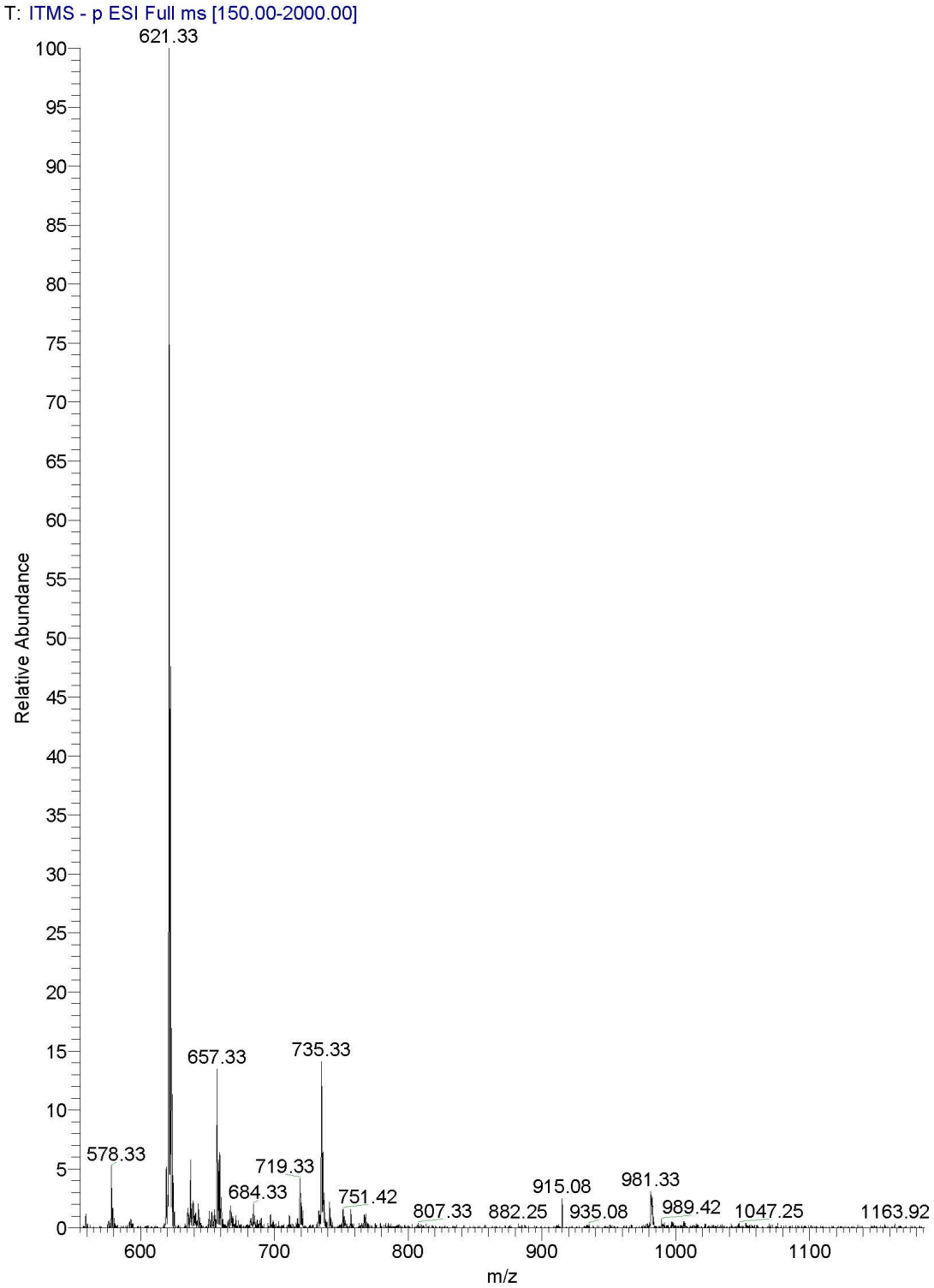
COSY at 500 MHz in $CDCl_3$ - 25 deg



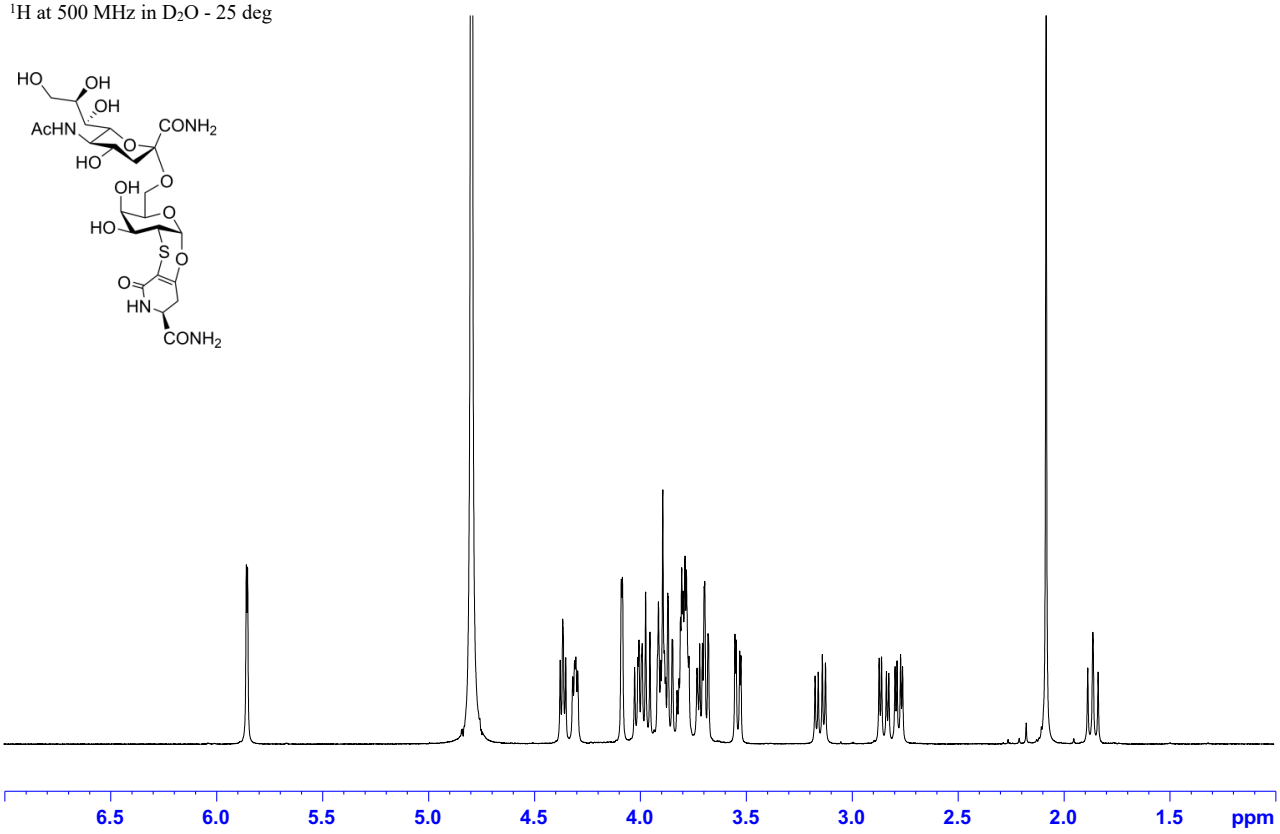
HSQC at 500 MHz in $CDCl_3$ - 25 deg



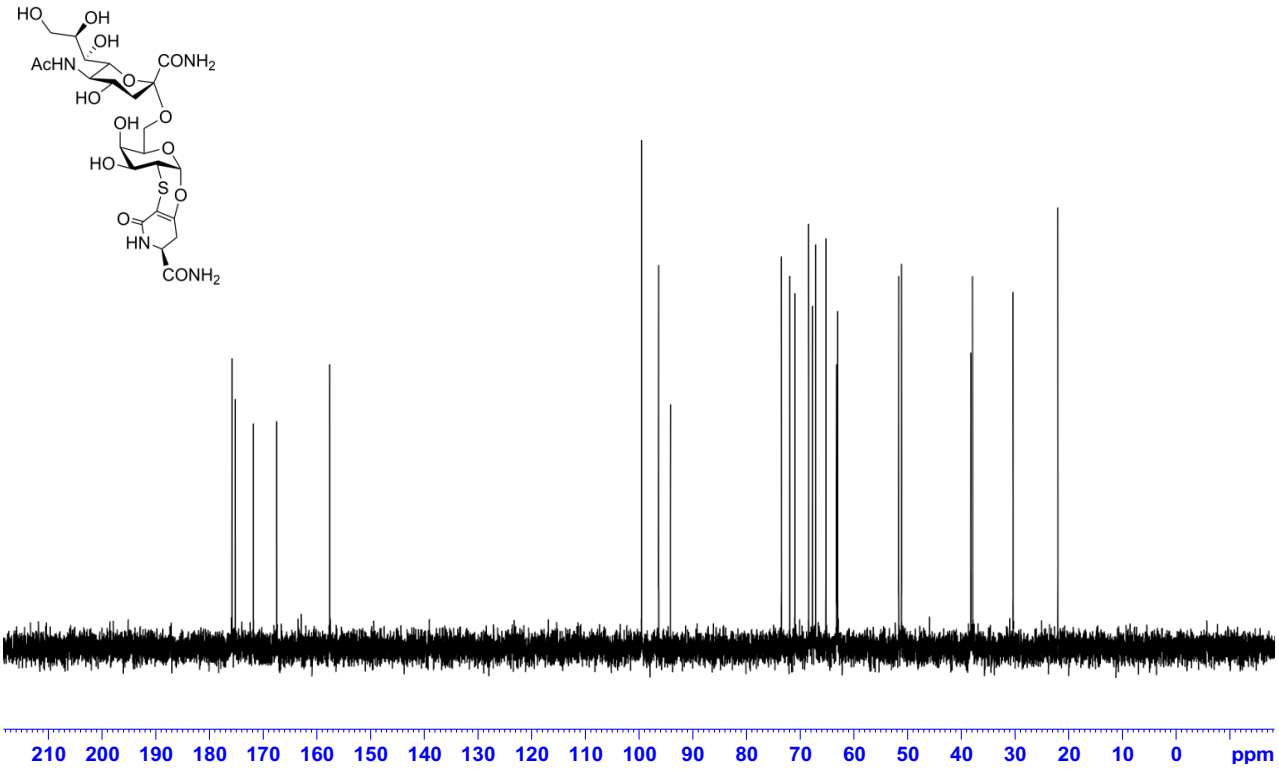
NMR and ESI-MS spectra of compound 10



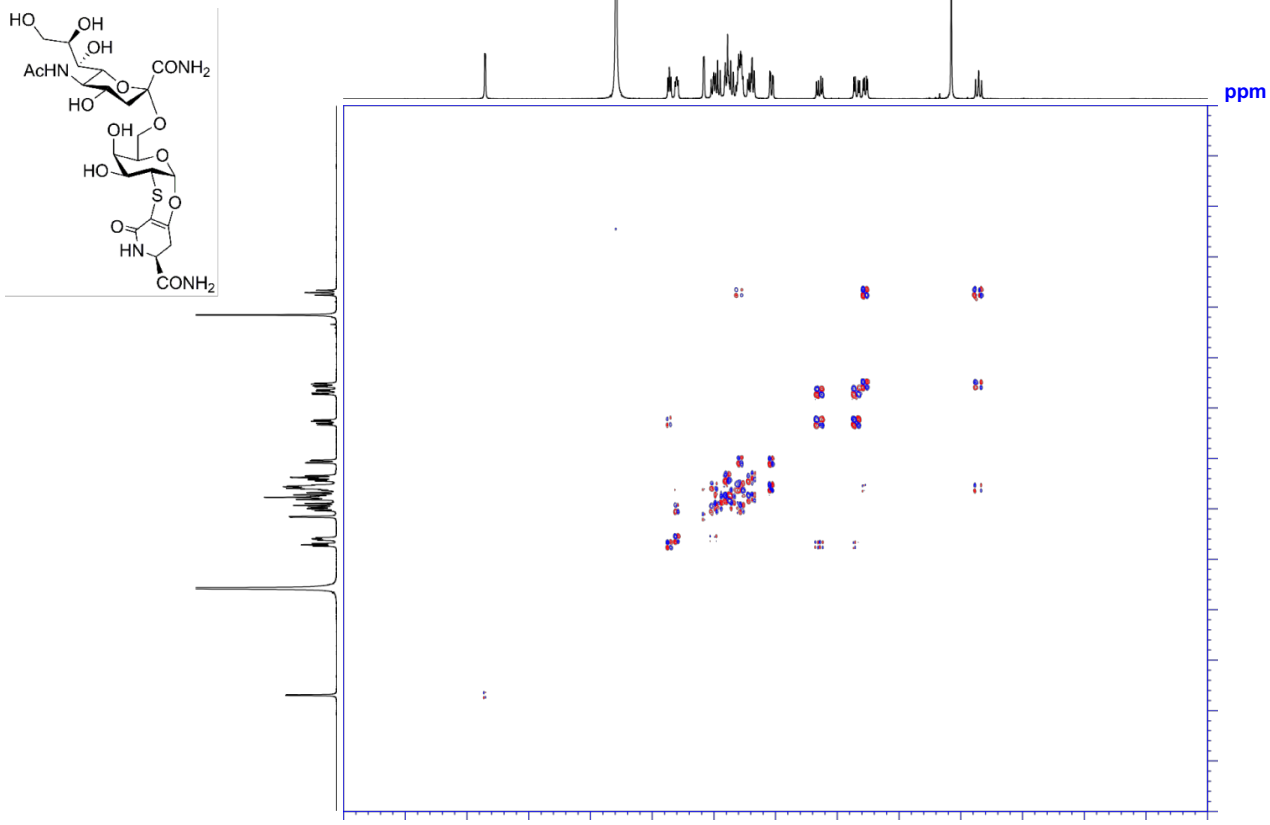
^{1}H at 500 MHz in D_2O - 25 deg



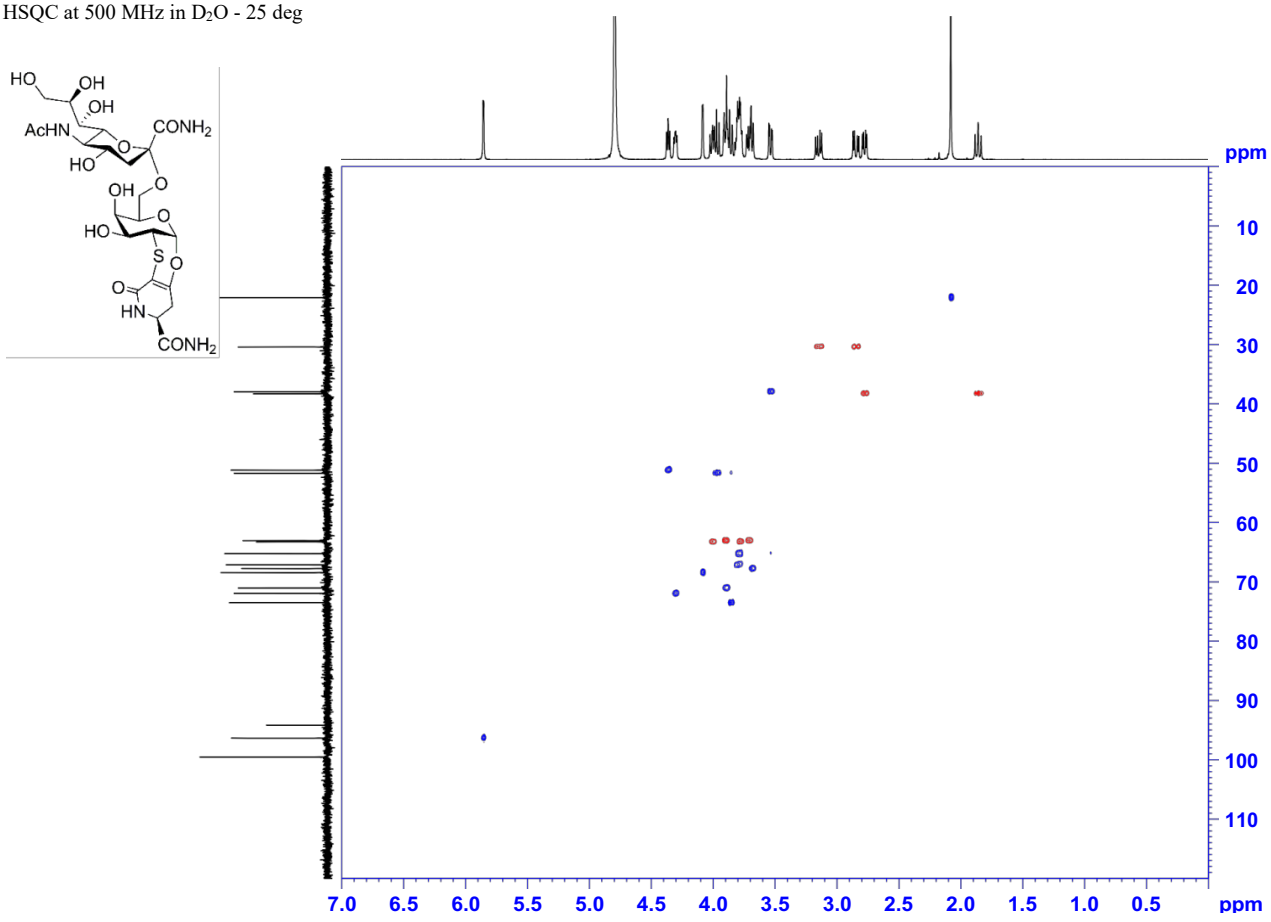
^{13}C at 125 MHz in D_2O - 25 deg



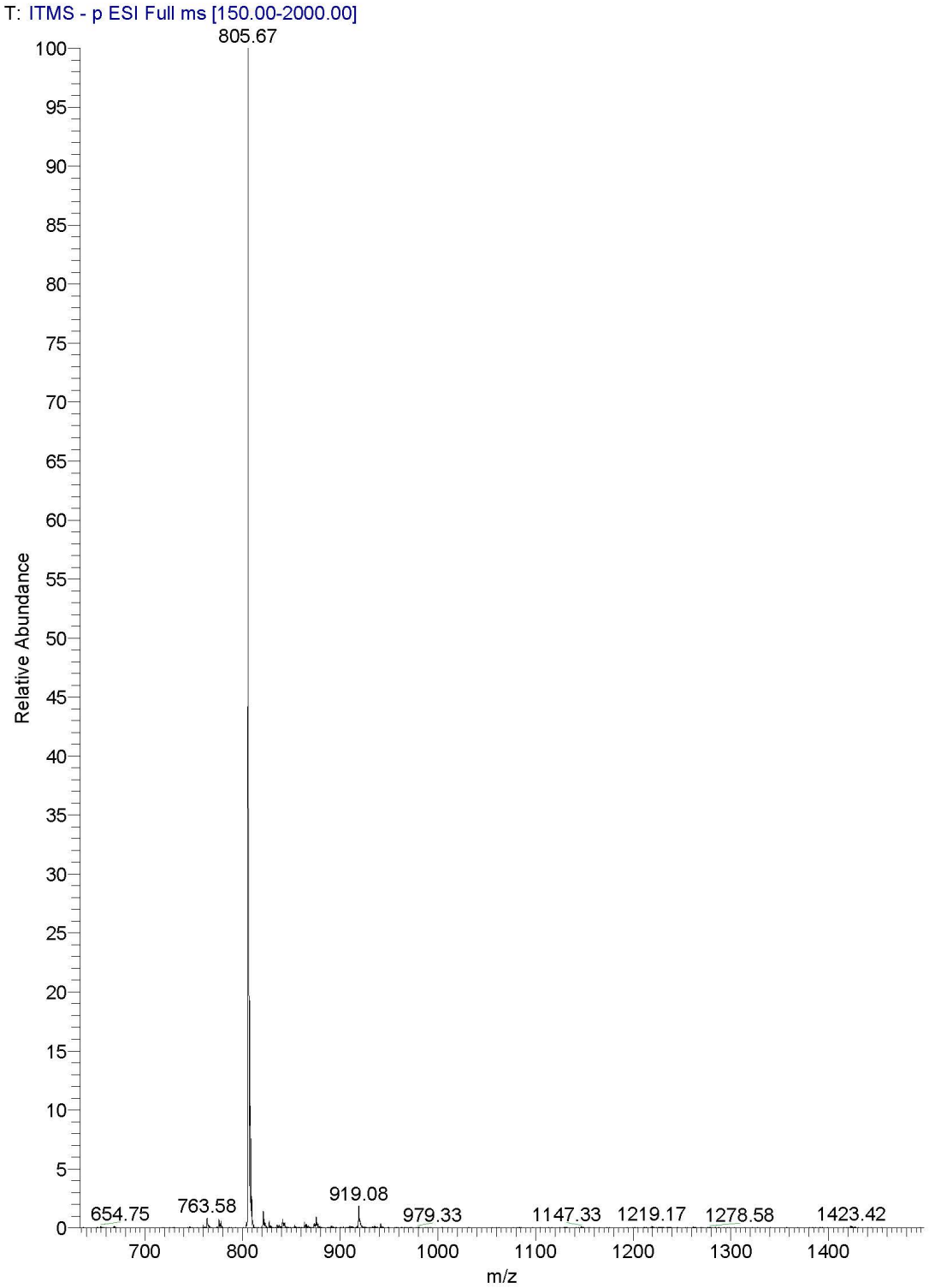
COSY at 500 MHz in D_2O - 25 deg



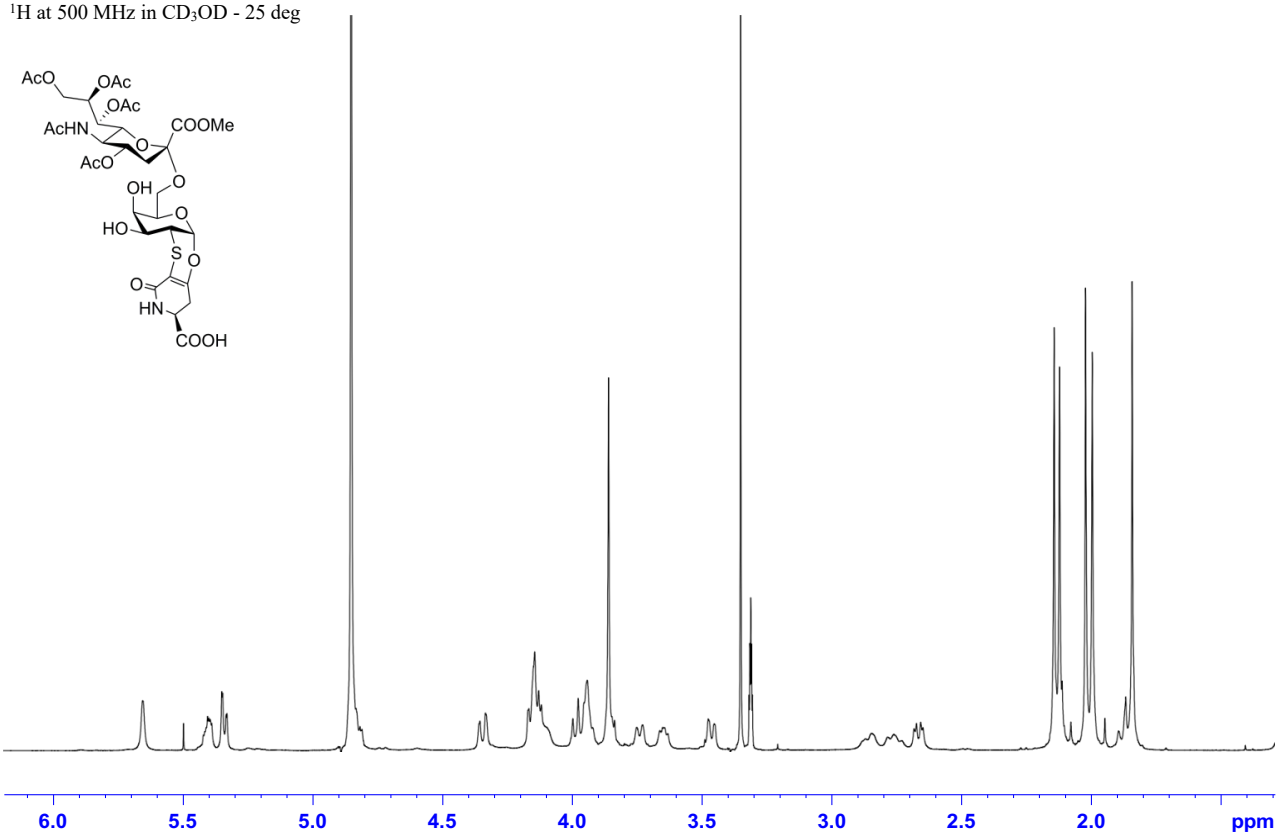
HSQC at 500 MHz in D_2O - 25 deg



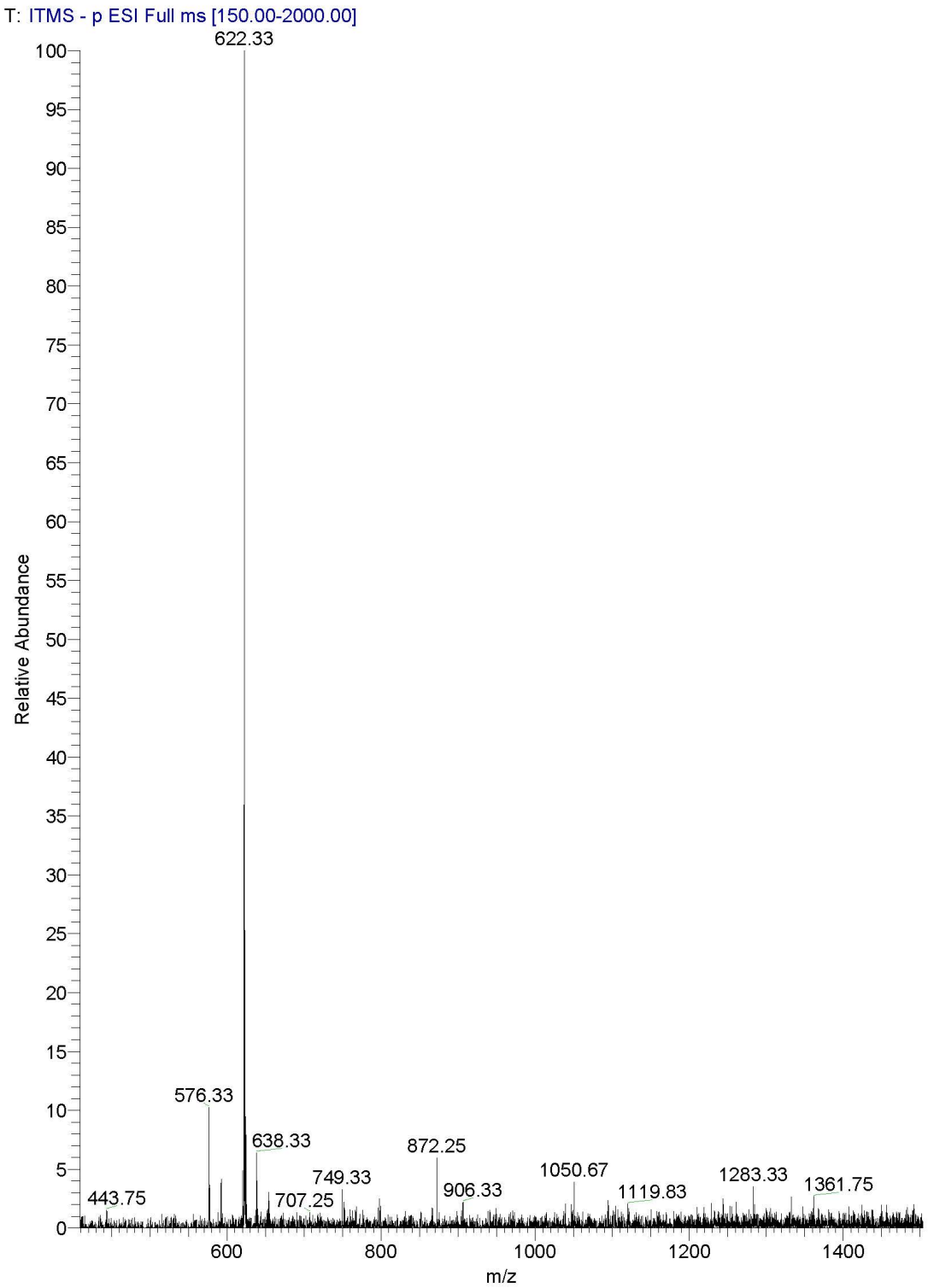
NMR and ESI-MS spectra of compound 11



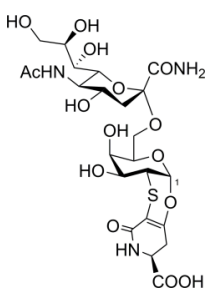
^{1}H at 500 MHz in CD_3OD - 25 deg



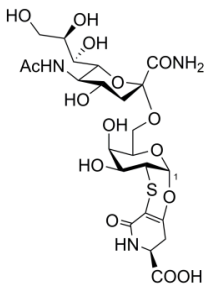
NMR and ESI-MS spectra of compound 12



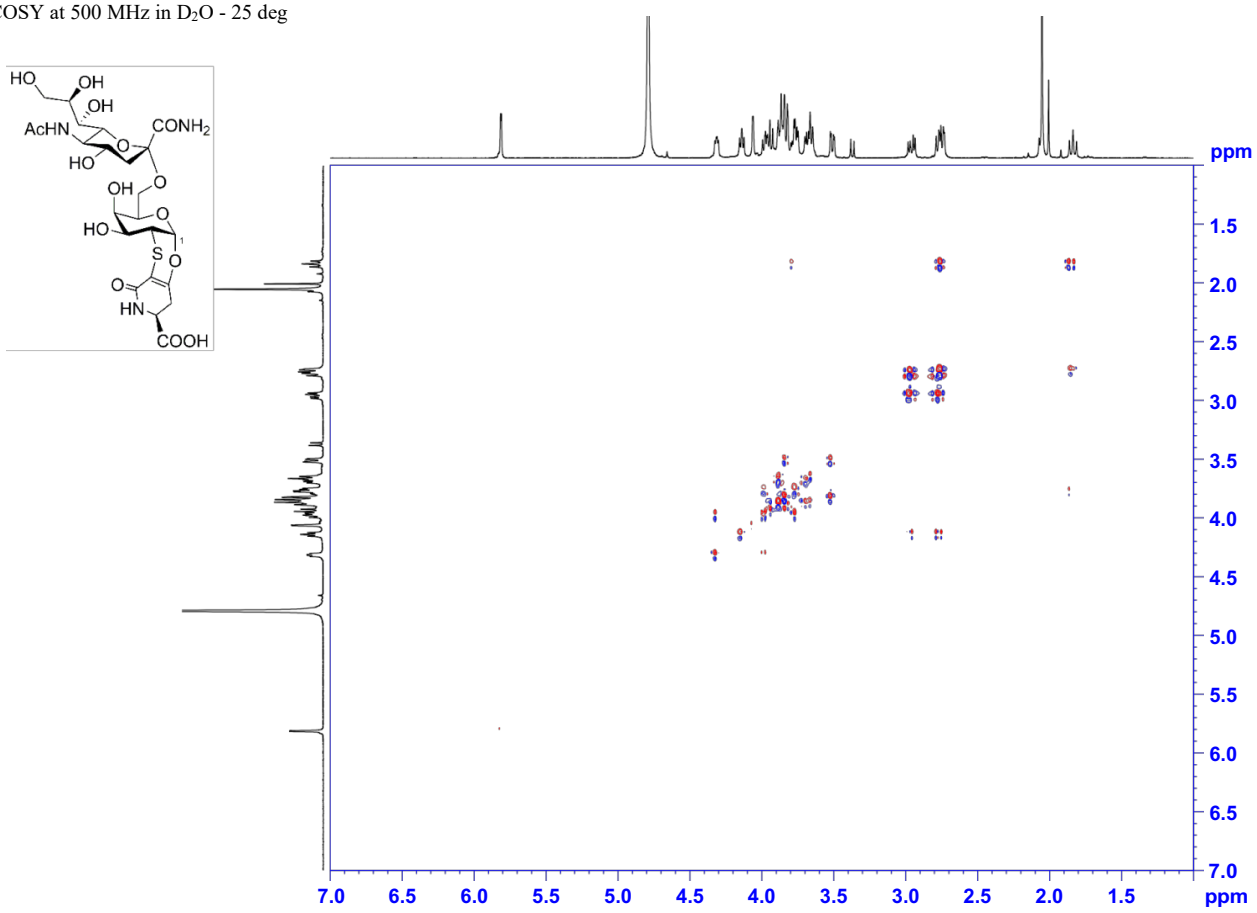
^{1}H at 500 MHz in D_2O - 25 deg



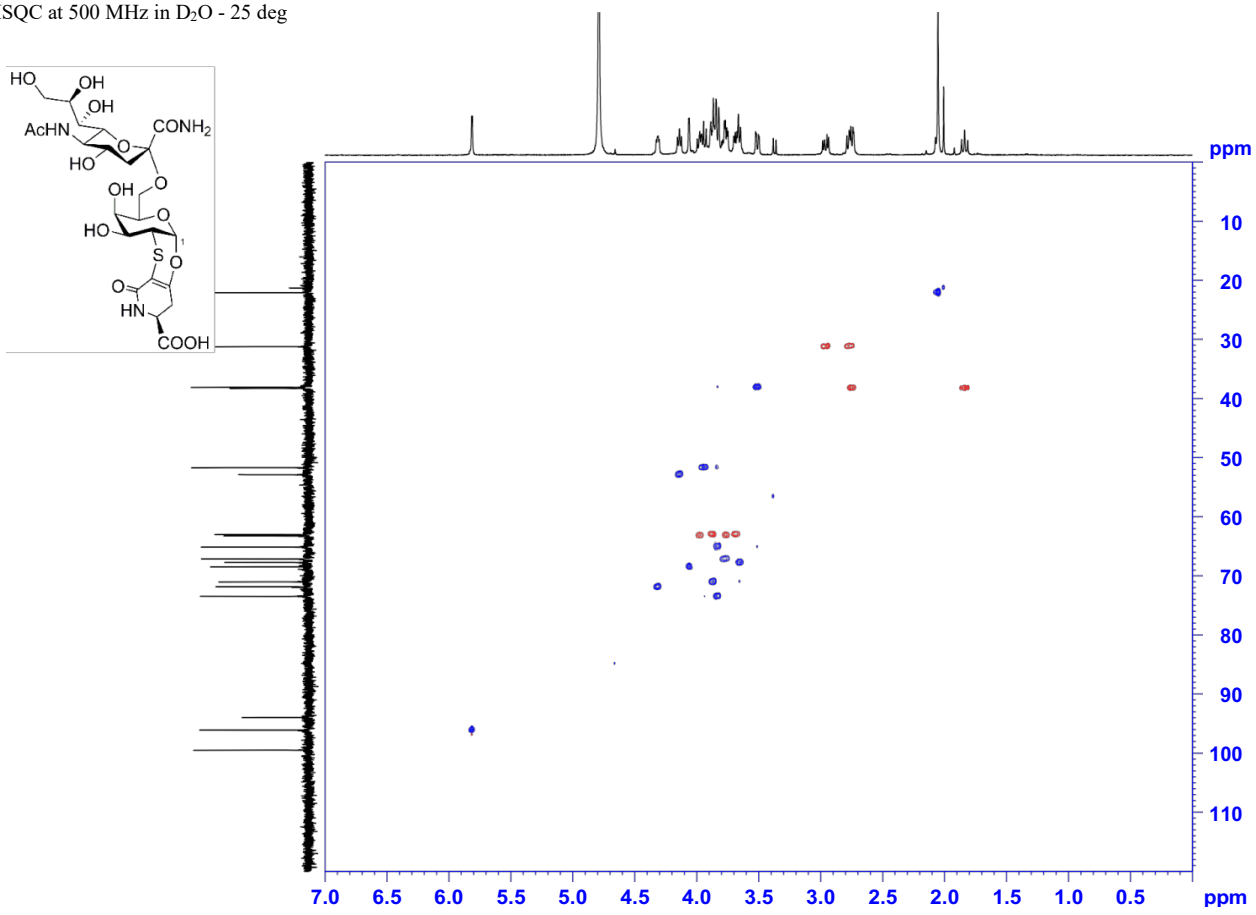
^{13}C at 125 MHz in D_2O - 25 deg



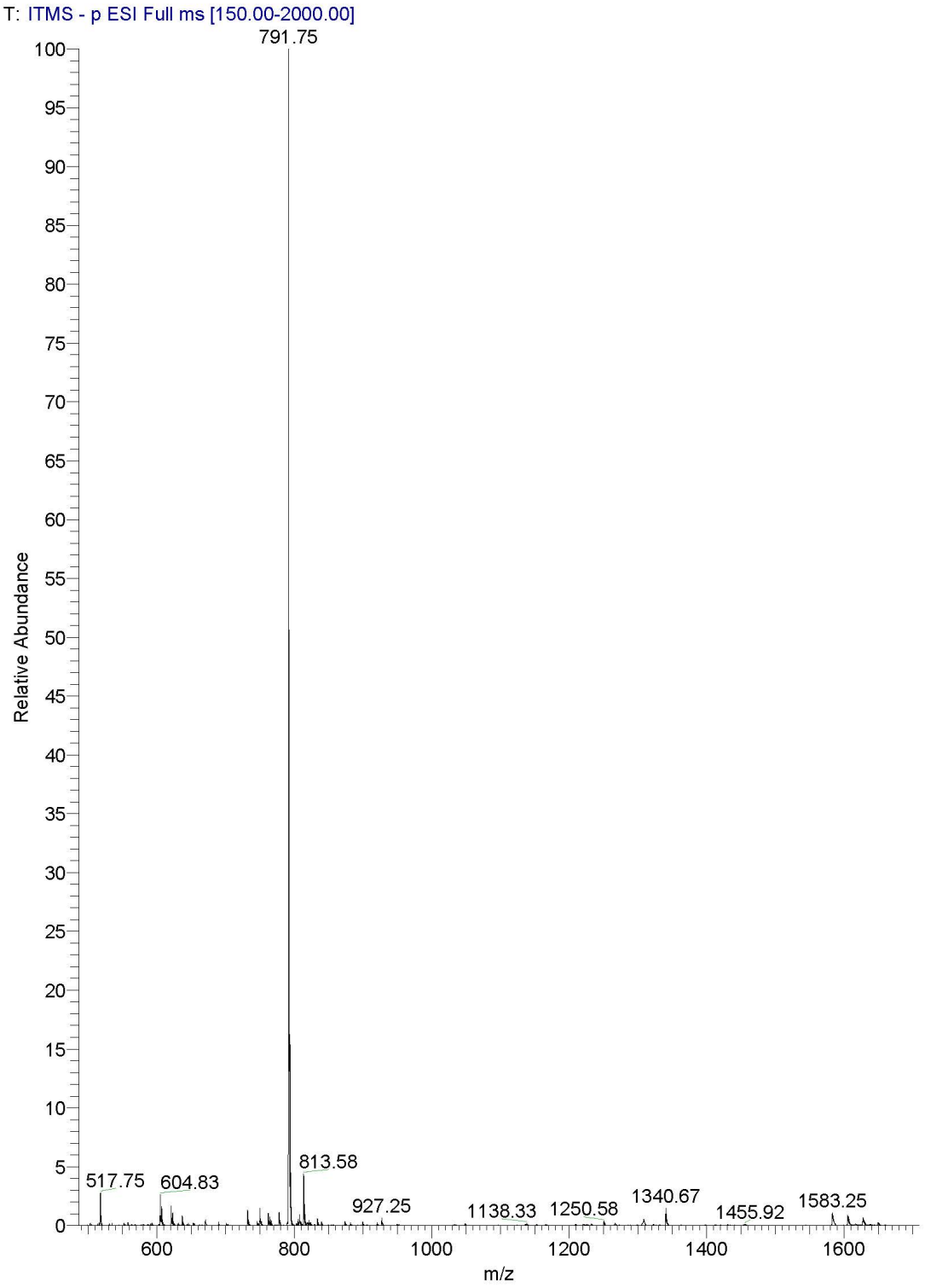
COSY at 500 MHz in D_2O - 25 deg



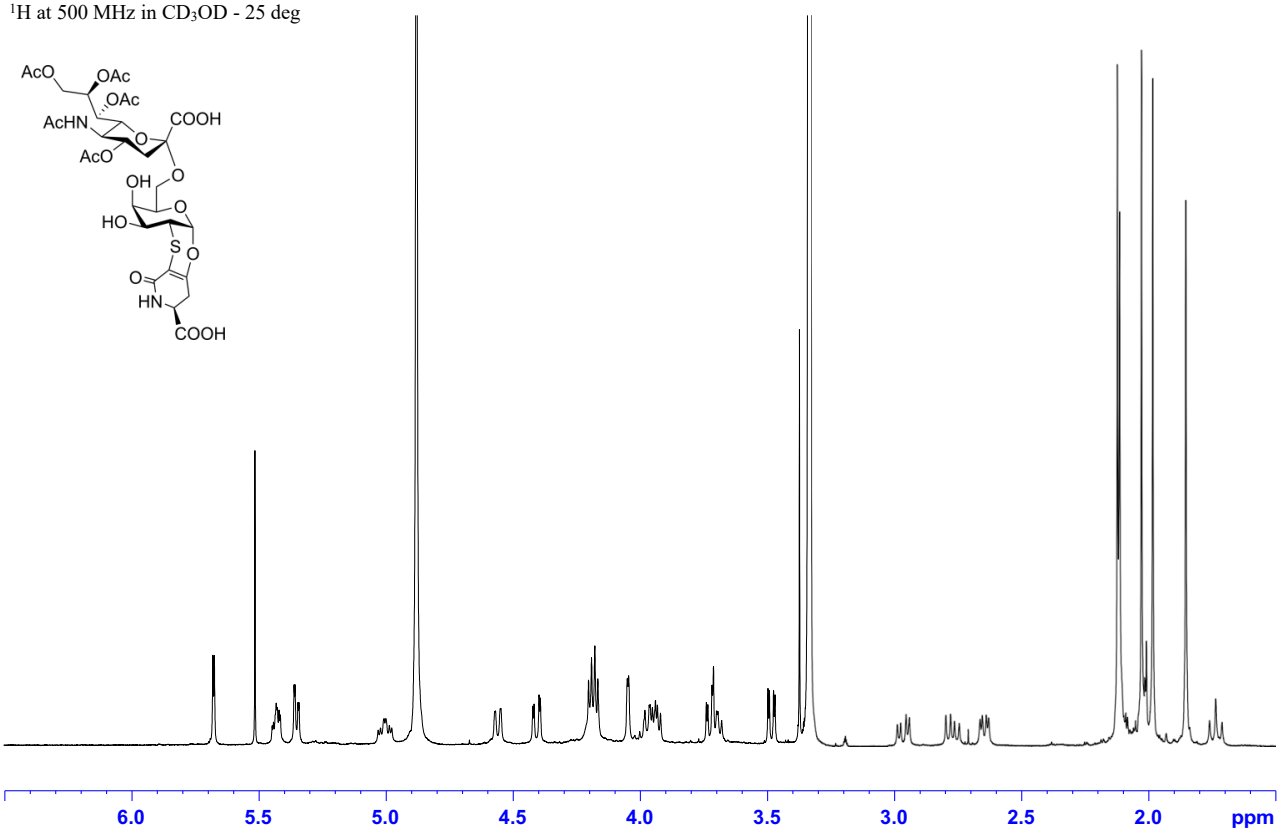
HSQC at 500 MHz in D_2O - 25 deg



NMR and ESI-MS spectra of compound 13



^{1}H at 500 MHz in CD_3OD - 25 deg



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