Modulating Heparanase Activity: Tuning Sulfation Pattern and Glycosidic Linkage of Oligosaccharides

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Table of Contents

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1. General Information

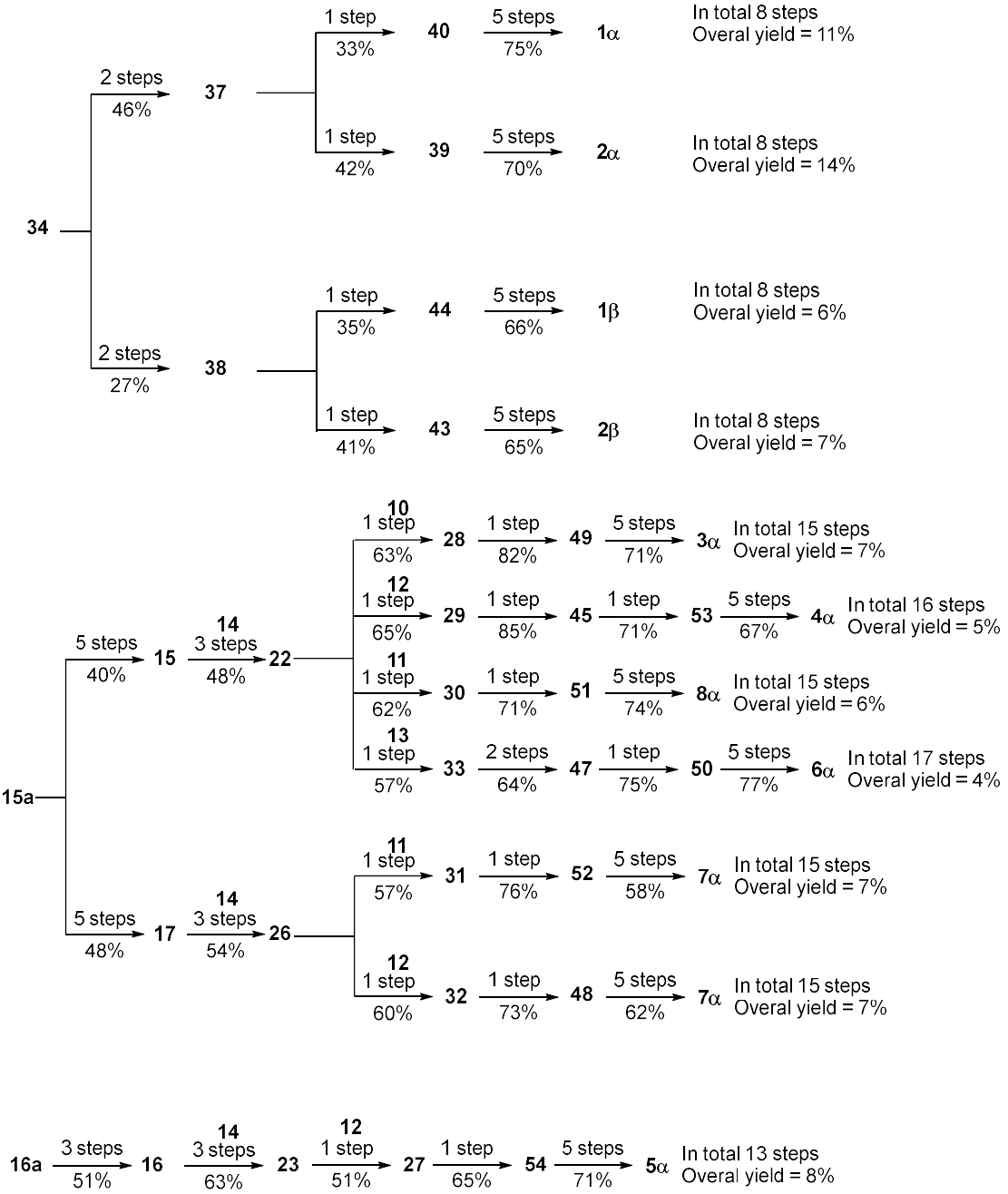
Methods and Reagents. All reactions were performed in oven-dried flasks fitted with septa under a positive pressure of nitrogen atmosphere. Organic solutions were concentrated using a Buchi rotary evaporator below 40 °C at 25 torr. Analytical thin-layer chromatography was routinely utilized to monitor the progress of the reactions and performed using pre-coated glass plates with 230-400 mesh silica gel impregnated with a fluorescent indicator (250 nm). Visualization was then achieved using UV light, iodine, or ceric ammonium molybdate. Flash column chromatography was performed using 40-63 μm silica gel (SiliaFlash F60 from Silicycle). Dry solvents were obtained from a SG Waters solvent system utilizing activated alumina columns under an argon pressure. All other commercial reagents were used as received from Sigma Aldrich, Alfa Aesar, Acros Organics, TCI, and Combi-Blocks, unless otherwise noted.

Instrumentation. All new compounds were characterized by Nuclear Magnetic Resonance (NMR) spectroscopy and High-Resolution Mass spectrometry (HRMS). All ^{1}H NMR spectra were recorded on either Agilent 400 or 600 MHz spectrometers. All ^{13}C NMR spectra were recorded on either Agilent 100 or 150 MHz spectrometer. Chemical shifts are expressed in parts per million (δ scale) referenced to the residual proton in the NMR solvent ($CDCl_3$: δ 7.26 ppm, δ 77.16 ppm). Data are presented as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, and bs = broad singlet), integration, and coupling constant in hertz (Hz).

High resolution mass spectra (HRMS) were recorded using a Micromass LCT Premier XE instrument (Waters) and were determined by electrospray ionization (ESI).

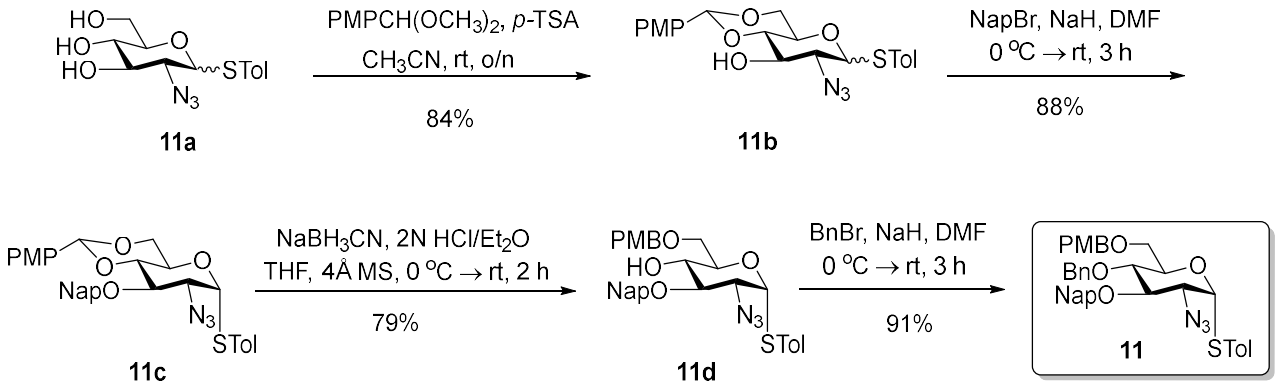
2. Modular Synthetic Scheme of Trisaccharides and Monosaccharide Building Blocks

2.1. Synthetic summary

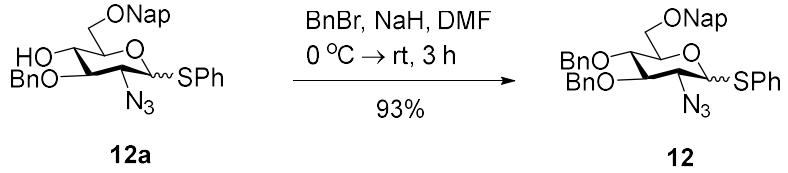


2.2. General Synthetic Schemes for Monosaccharide Building Blocks Preparation

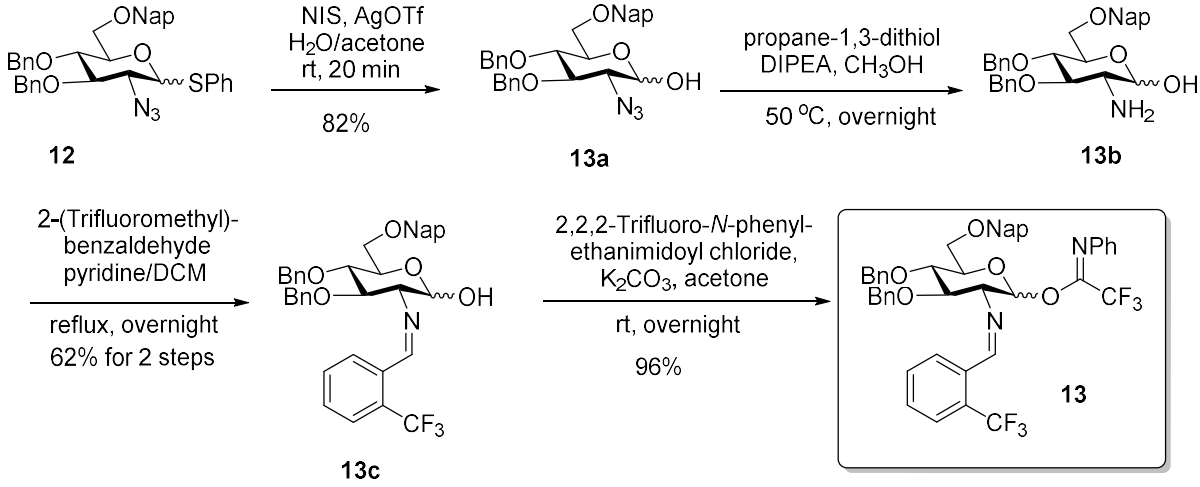
_p_-Tolyl 2-azido-2-deoxy-3-_O_-(2-naphthylmethyl)-4-_O_-benzyl-6-_O_-(_p_-methoxybenzyl)-1-thio-α-D-glucopyranoside (11):



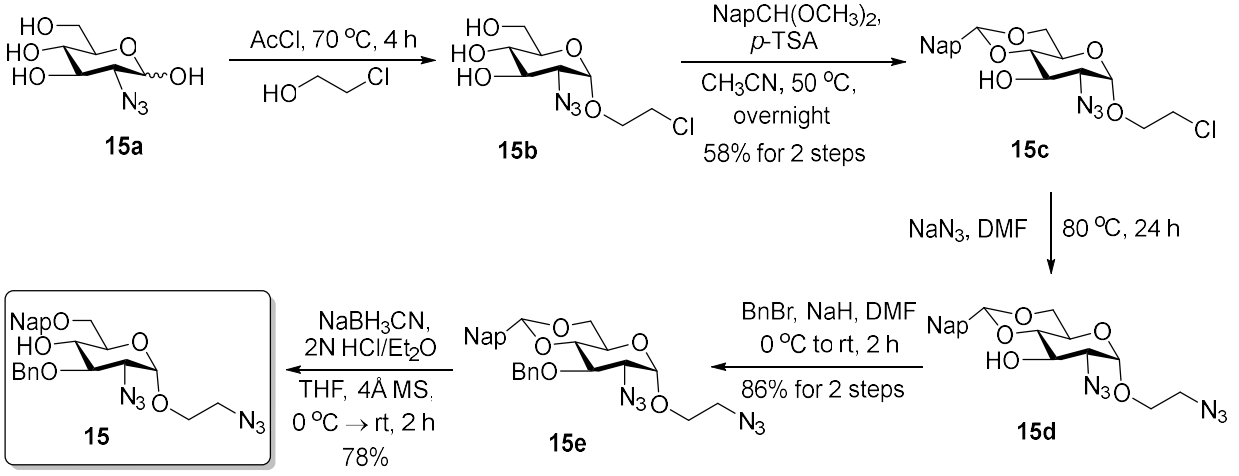
Phenyl 2-azido-2-deoxy-3,4-di-_O_-benzyl-6-_O_-(2-naphthylmethyl)-1-thio-D-glucopyranoside (12):



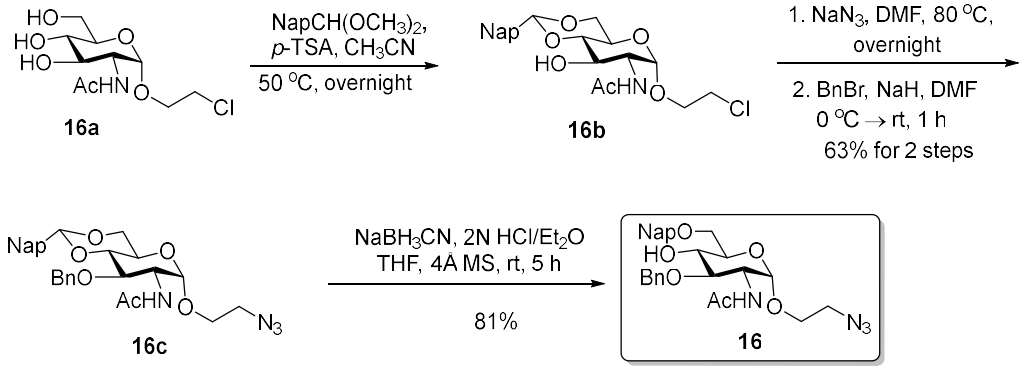
2-_N_-[(2-trifluoromethyl)benzylidene]-2-deoxy-3,4-di-_O_-benzyl-6-_O_-(2-naphthylmethyl)-D-glucopyranosyl _N_-phenyl-trifluoroacetimidate (13):



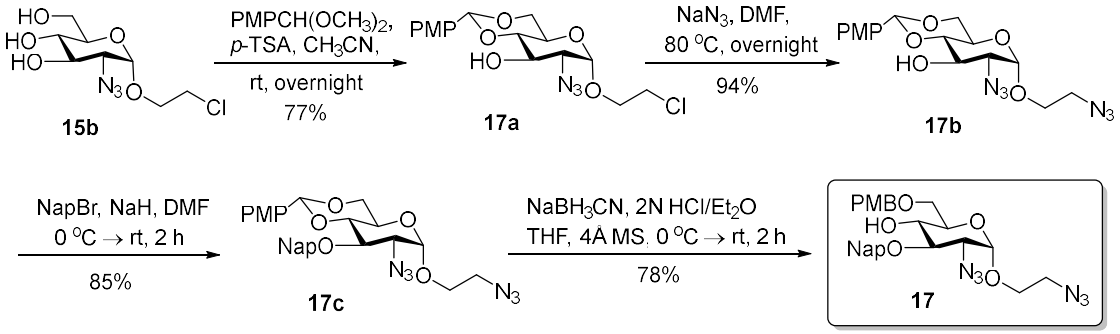
2-Azidoethyl 2-azido-2-deoxy-3-*O*-benzyl-6-*O*-(2-naphthylmethyl)-α-D-glucopyranoside (15):



2-Azidoethyl 2-acetamido-2-deoxy-3-*O*-benzyl-6-*O*-(2-naphthylmethyl)-α-D-glucopyranoside (16):

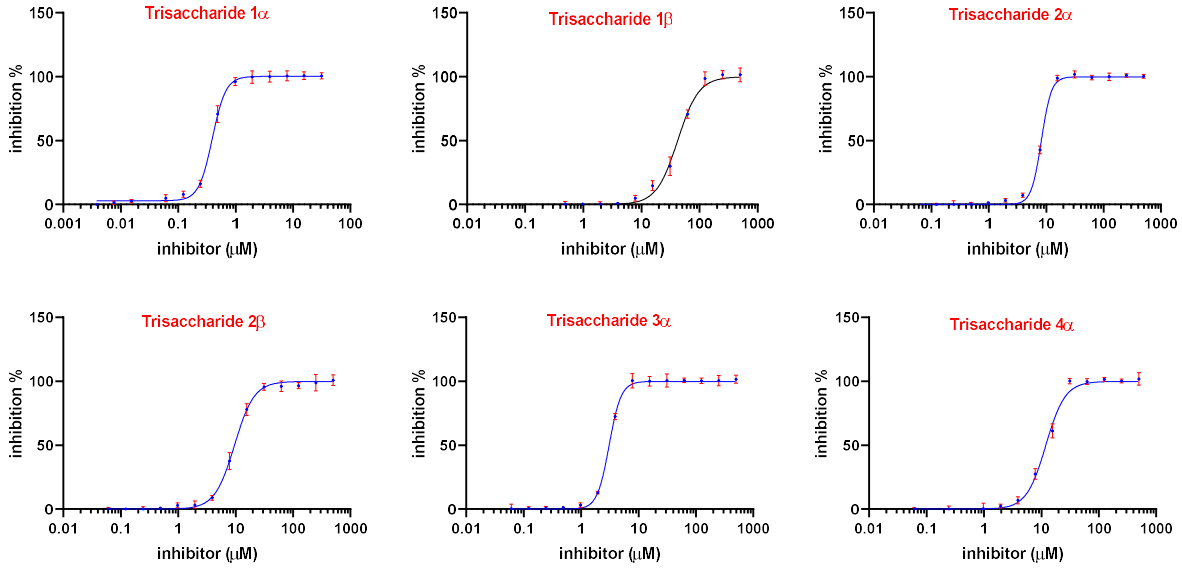


2- Azidoethyl 2-azido-2-deoxy-3-*O*-(2-naphthylmethyl)-6-*O*-(*p*-methoxybenzyl)-α-D-glucopyranoside (17):



3. TR-FRET Heparanase Inhibition Assay[1]

42 µL of HS trisaccharide solution in Milli-Q water (0.0038-500 µM) or just Milli-Q water (as a control), and 42 µL of heparanase (5.3 nM, R&D Systems) solution in pH 7.5 triz buffer (consisting of 20 mM TrisHCl, 0.15 M NaCl and 0.1% CHAPS) or just buffer as blank were added into microtubes and pre-incubated at 37 °C for 10 min bringing the [heparanase] to 0.5 nM. Next, 84 µL of biotin-heparan sulfate-Eu cryptate (Cisbio, Cat #: 61BHSKAA) (58.6 ng in pH 5.5 0.2 M NaOAc buffer) was added to the microtubes, and the resulting mixture was incubated for 60 min at 37 °C. The reaction mixture was stopped by adding 168 µL of Streptavidin-XLent! (Cisbio, Cat #: 611SAXLA) (1.0 µg/ml) solution in pH 7.5 dilution buffer made of 0.1 M NaH_2PO_4, 0.8 M KF, 0.1% BSA. After the mixture had been stirring at room temperature for 15 min, 100 µL (per well) of the reaction mixture was transferred to a 96 well microplate (Corning #3693 96 well, white polystyrene, half-area) in triplicates and HTRF emissions at 616 nm and 665 nm were measured by exciting at 340 nm using a SpectraMax iD5 Microplate Reader (Molecular Devices).



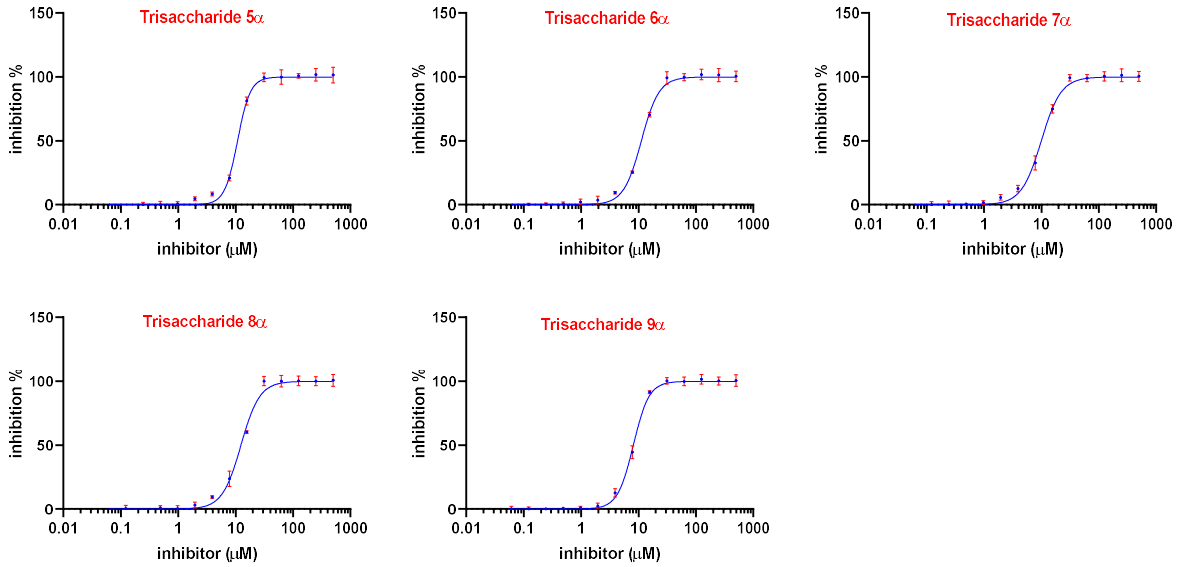


Figure S1. Inhibition of heparanase by different trisaccharide ligands.

4. Trisaccharides Hydrolysis Assay[2]

Micro centrifuge tubes were pretreated for 2h at 37 °C by using a solution of phosphate buffered saline comprising of 0.05% Tween 20 (PBST) and 4% bovine serum albumin. Tubes were then washed three times with PBST, dried, stored at 4 °C, and used for assay within a week. A 100 μM solution of fondaprinux or trisaccharide with heparanase enzyme (3 nM) in pH 5.0 sodium acetate buffer (40 mM) were incubated at 37 °C in a microtube. The enzymatic reaction was stopped at different time points by adding equal volumes of reaction mixture and freshly made 1.69 mM WST-1 (Toronto Research Chemicals) in 0.1 M NaOH into a new microtube and developed at 60 °C for 60 min. A 200 μl (per well) solution of developed reaction solution was transferred to a clear 96-well microplate in triplicates and absorbance was measured at 584 nm with a SpectraMax iD5 Microplate Reader (Molecular Devices). For the controls, exactly the same procedure was followed for a 100 μM solution of trisaccharides without heparanase enzyme to rationalize non-enzymatic autohydrolysis.

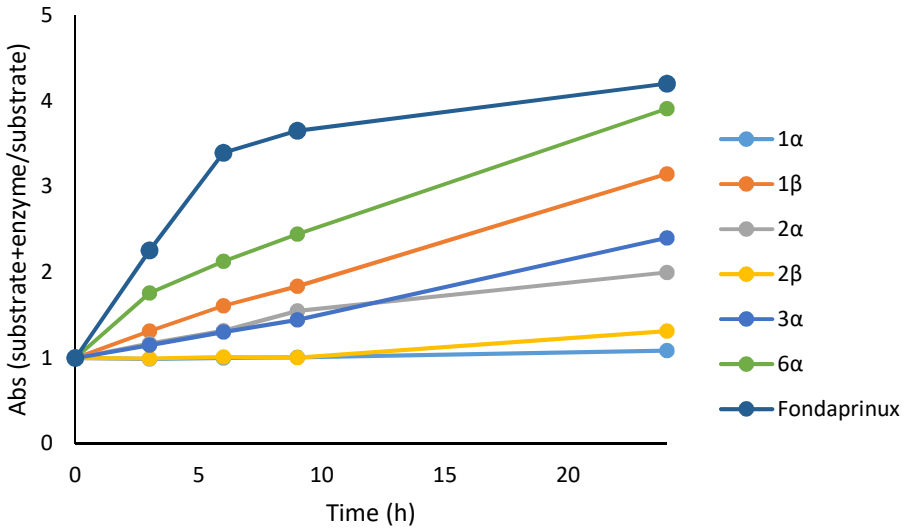


Figure S2. Hydrolytic potential of **1α**, **1β**, **2α, 2β, 3α** or **6α** with heparanase over time compared to fondaprinux.

5. Two-Stage Chromogenic Assay to Evaluate Anticoagulant Activity[2]

BIOPHEN Heparin AntiXa (2 stages) USP/EP (Cat #: A221005-USP) kits from Aniara and BIOPHEN Heparin Anti-IIa (2 stages) USP/EP (Cat #: A220005-USP) were utilized to assess FXa and FIIa activity, respectively.

Factor Xa activity:

All the reagents were reconstituted and prepared according to the manufacturer's instructions and incubated at 37 °C for 15 min. Different concentrations of heparin (0.002-500 nM; 40 µL) or trisaccharide **1α** (3.9-4000 nM; 40 µL) and ATIII (0.04 IU; 40 µL) were added to a deep-well block (Nunc 96 DeepWell 1.0 mL/well, clear), mixed, and incubated at 37 °C for 2 min. To the reaction mixture, FXa (0.32 µg; 40 µL) was added by multichannel pipette and was incubated at 37 °C for another 2 min (stage 1), then FXa specific chromogenic substrate (0.048 mmol; 40 µL) was added. The reaction was stopped by adding citric acid (240 µL; 20 g/L) exactly after 2 min. A 100 µL solution was then transferred to a clear 96-well microplate in triplicate, and absorbance at 405 nm was measured with a SpectraMax iD5 Microplate Reader (Molecular Devices). The sample blank was measured by mixing the reagents in reverse order from that of the test, i.e. citric

acid, FXa substrate, FXa, ATIII, and sample. The sample blank value was deducted from the absorbance measured for the corresponding assay.

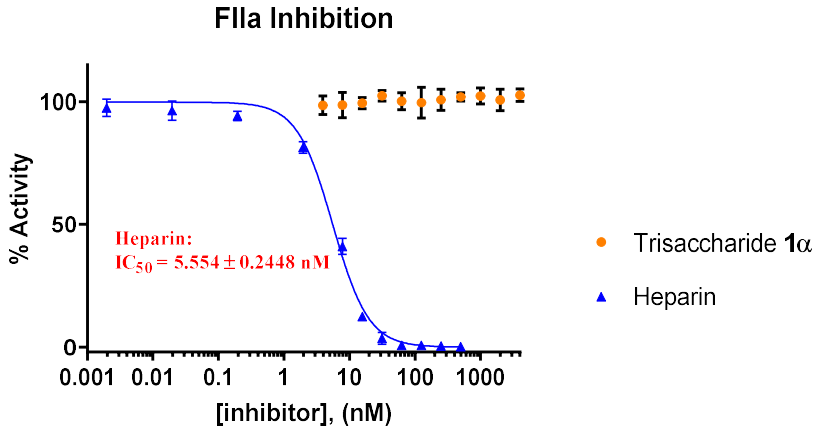


Figure S3. Inhibition of FXa by trisaccharide **1α** in comparison to heparin.

Factor IIa activity

All the reagents were reconstituted and prepared according to the manufacturer's instructions and incubated at 37 °C for 15 min. Different concentrations of heparin (0.002-500 nM; 40 µL) or trisaccharide **1α** (3.9-4000 nM; 40 µL) and ATIII (0.01 IU; 40 µL) were added to a deep-well block (Nunc 96 DeepWell 1.0 mL/well, clear), mixed, and incubated at 37 °C for 2 min. To the reaction mixture, FIIa (1.2 nkat; 40 µL) was added by multichannel pipette and was incubated at 37 °C for another 2 min (stage 1), then FIIa specific chromogenic substrate (0.048 mmol; 40 µL) was added. The reaction was stopped by adding citric acid (240 µL; 20 g/L) exactly after 2min. 100 µL was transferred to a 96-well microplate in triplicate, and absorbance at 405 nm was measured with SpectraMax iD5 Microplate Reader (Molecular Devices). The sample blank was measured by mixing the reagents in reverse order from that of the test, i.e. citric acid, FXa substrate, FXa, ATIII, and sample. The sample blank value was deducted from the absorbance measured for the corresponding assay.

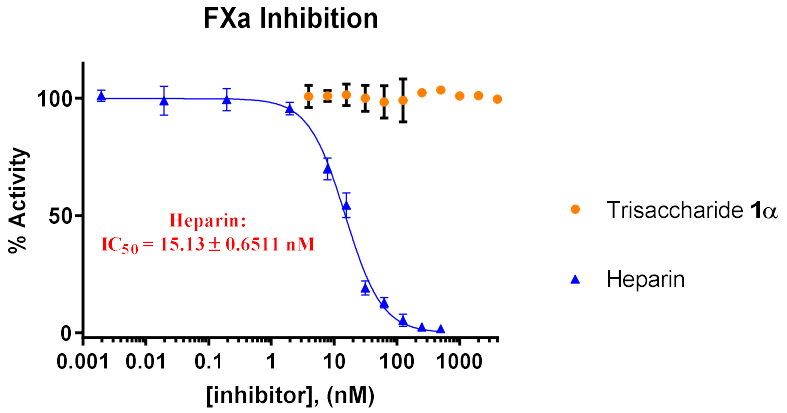
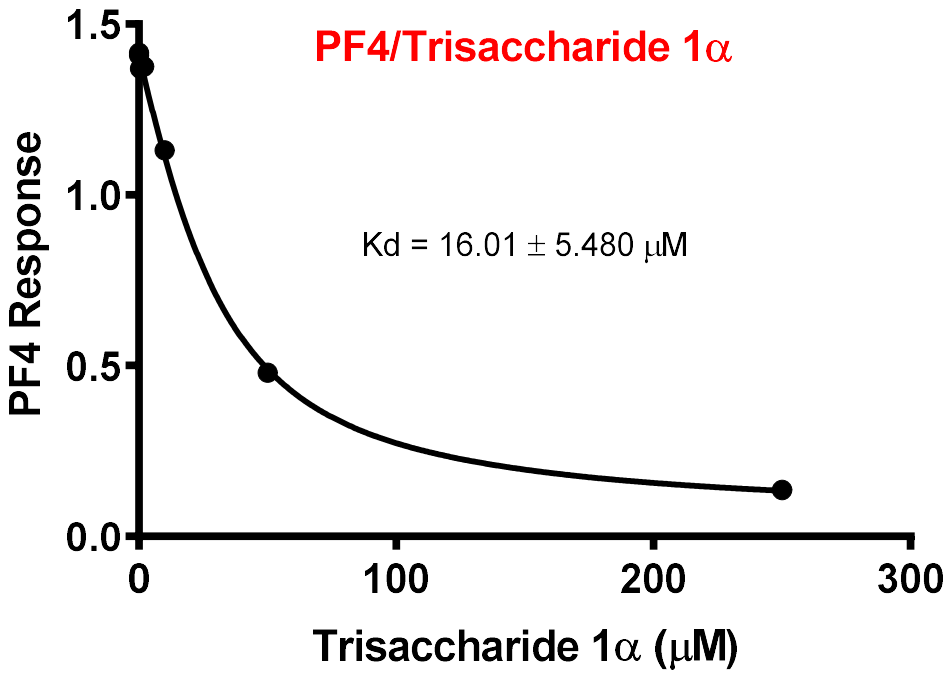
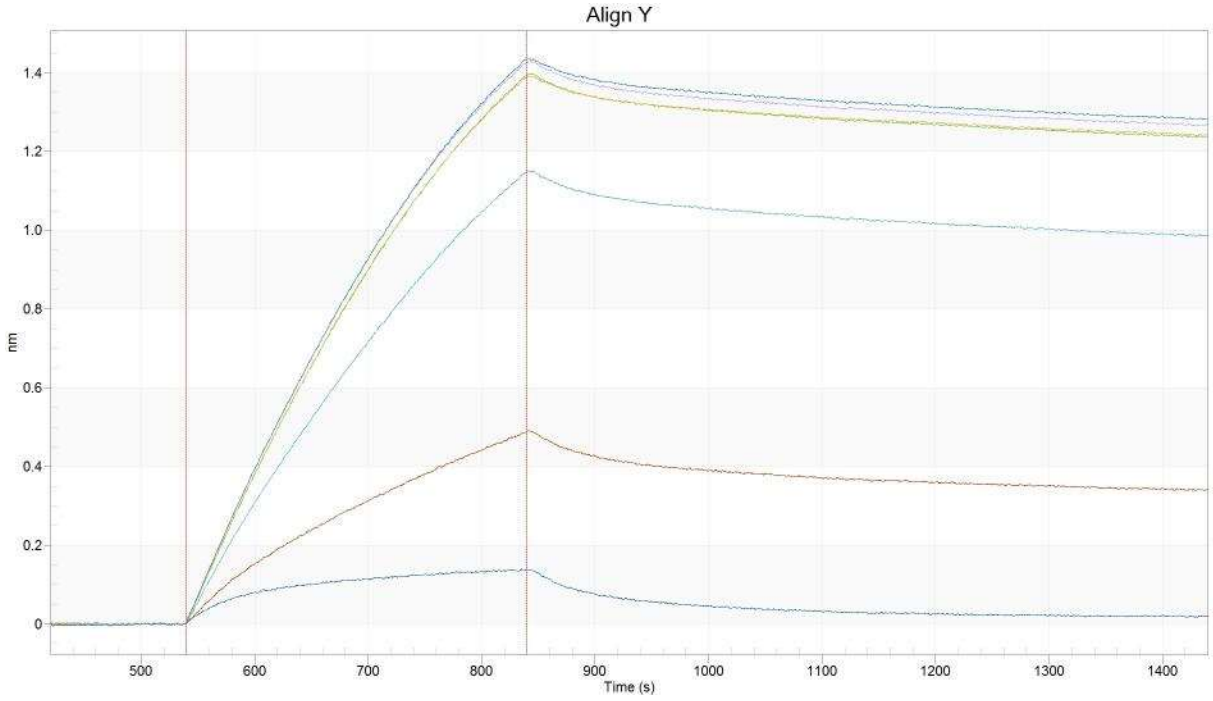


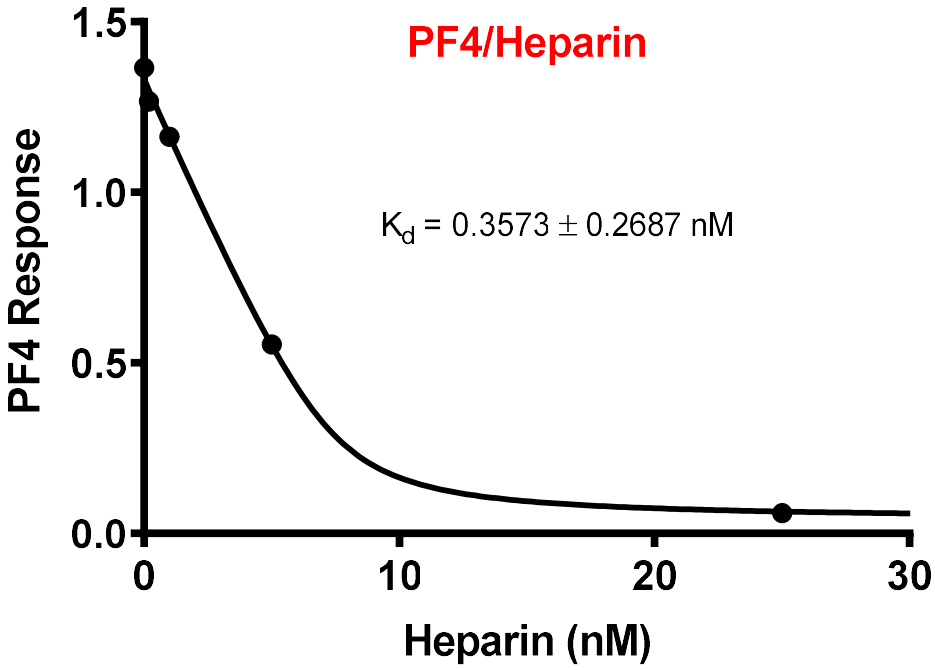
Figure S4. Inhibition of FIIa by trisaccharide **1α** in comparison to heparin.

6. Platelet Factor 4 Binding Test by Biolayer Interferometry (BLI) Assay[1]

BLI assays were performed on an Octet Red Instrument (fortéBIO) at 25 °C. Immobilization and binding analysis were carried out at 1000 rpm using HBS-EP buffer [10 mM HEPES, pH 7.4, 150 mM NaCl, 3.0 mM EDTA, and 0.005% (v/v) surfactant tween20]. A solution affinity assay, used to determine affinities of ligands by SPR analysis was adopted to BLI.[3] In this method PF 4 (100 nM; 100 µL) was mixed with various concentrations of trisaccharide **1α** (0.08-250 µM; 100 µL) or heparin (0.4-400 nM; 100 µL). Free protein in this equilibrium mixture is tested for binding against immobilized heparin (all proteins are carrier-free and purchased from R&D Systems). Heparin-biotin (Creative PEGworks, 18 kDa, 1 biotin per HP polymer), 5 µg /mL was immobilized on to streptavidin biosensors (fortéBio) for 5 min. Binding experiments were carried under conditions of mass transport. Binding was fitted to equation 1 of ref 10 using Graphpad Prism. BLI response was used in place of F and ligand (heparin / trisaccharide **1α**) concentration was used in place of [metal].


PF4/Trisaccharide 1α
Kd = 16.01 ± 5.480 μM
PF4 Response
1.5
1.0
0.5
0.0
0
100
200
300
Trisaccharide 1α (μM)


Align Y
nm
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0
500
600
700
800
900
1000
1100
1200
1300
1400
Time (s)



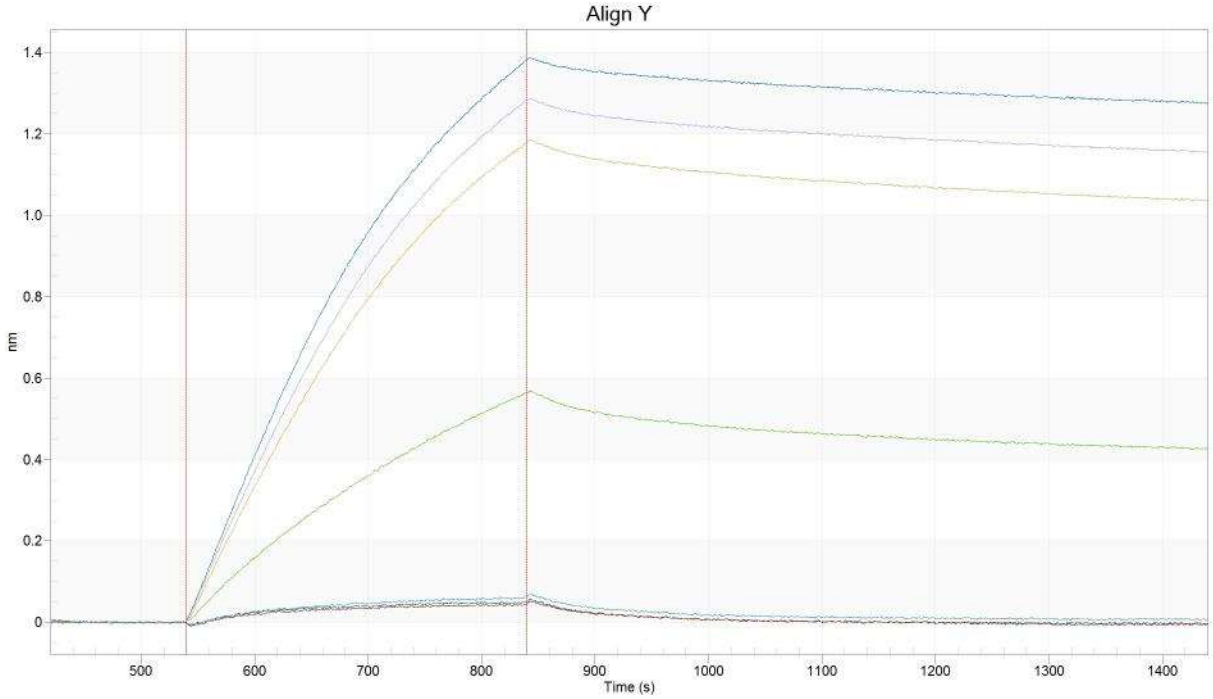


Figure S5. BLI assay to test binding affinity of PF4 and trisaccharide **1α** / heparin.

7. Inhibition of ECM Degradation Assay

Briefly, sulfate [^{35}S] labeled ECM coating the surface of 35 mm tissue culture dishes,[12] is incubated (5 h, 37ºC, pH 6.0, 1 ml final volume) with recombinant human heparanase (0.5 ng/ml) in the absence and presence of trisaccharide **1α**. The reaction mixture contains: 50 mM NaCl, 1 mM DTT, 1 mM $CaCl_2$, and 10 mM buffer Phosphate-Citrate, pH 6.0. To evaluate the occurrence of proteoglycan degradation, the incubation medium is collected and applied for gel filtration on Sepharose 6B columns (0.9 x 30 cm). Fractions (0.2 ml) are eluted with PBS and counted for radioactivity. The excluded volume (V_o) is marked by blue dextran and the total included volume (V_t) by phenol red. Degradation fragments of HS side chains, characterized as described,[13] are eluted from Sepharose 6B at $0.5 < Kav < 0.8$ (fractions 20-35). Results are best represented by the actual gel filtration pattern.[14-16] Each experiment was performed at least three times and the variation of elution positions (Kav values) did not exceed ±15%.

8. Inhibition of Heparanase in Pancreatic β Cells

Culture of mouse pancreatic β cell line MIN6 and detection of cellular mitochondrial ROS using mitochondrial ROS trackers. Mouse pancreatic β cell line (insulinoma) MIN6 was cultured as we previously described.[17] Specifically, MIN6 cells were cultured with high-glucose DMEM supplemented with 10% fetal bovine serum (FBS), 50μM β-mercaptoethanol, and antibotics. Before experiments, MIN6 cells were seeded in 4-well slide-chambers for immunofluorescence analyses. When the cells reached approximately 50% confluency, they were cultured in the conditioned medium from heparanase-producing CHO cells or control CHO cells. Heparanase-producing CHO cells were provided by Dr. Israel Vlodavsky (Technion, Israel). Meanwhile, the cells were treated with vehicle (PBS) or the trisaccharide 1α inhibitor for 24 h. The cells were then stained with mitochondrial ROS tracker probe using the concentration of 100 nM according to the manufacturer's instruction (Molecular Probes, Invitrogen). Mitochondrial ROS in the MIN6 cells were visualized by fluorescence microscopy, and the extents and intensities of fluorescent activities, which reflect intracellular mitochondrial ROS, were quantified using ImageJ software.
Quantitative real-time PCR (qPCR) - Total RNA from Min-6 cells was extracted using Trizol by following manufacture's instruction and complementary DNA was synthesized from 500 ng of total RNA to with High Capacity cDNA Reverse Transcription Kit (AB applied biosystems). The

abundance of mRNA was measured by q PCR analysis using the SYBR Green PCR Master mix (Applied Biosystems).

The sequences of the primers for SOD1: 5'-GCGGTGAACCAGTTGTGTTGTC-3' and 5'-CAGTCACATTGCCCAGGTCTCC-3'; and for ERO1α: 5'-ATGGACTGTGTTGGCTGCTT-3' and 5'-GCTTTCCGGCATATTTGCGA-3'.

9. Computational study

All molecular docking and MD simulations were conducted with the YASARA Structure software package (www.yasara.org). Detail procedures for the computation study were reported in previous article.[5]

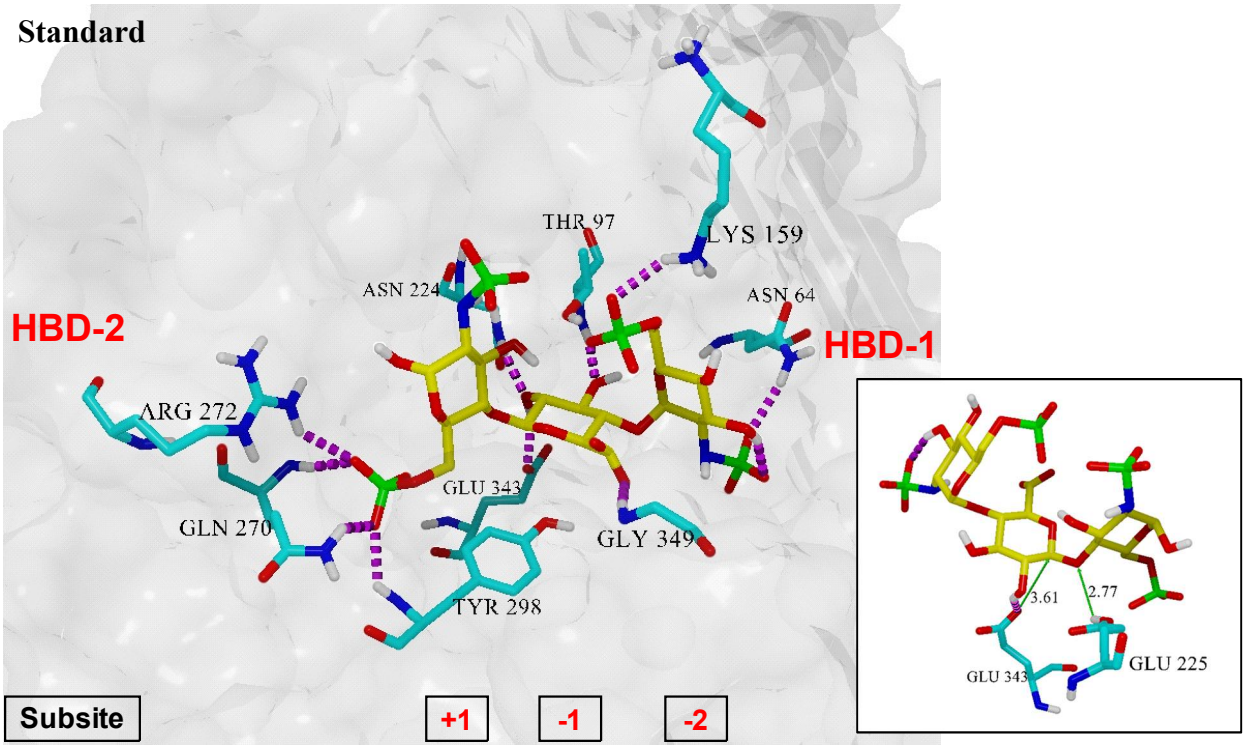
9.1. Computational Docking

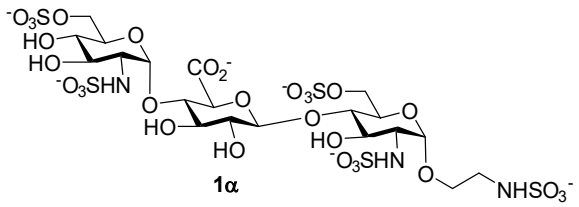
In the docking study, the enzyme structure was inherited from previously modified *apo* heparanase structure (PDB code: 5E8M).[5,6] The ligand saccharide backbone was obtained from Glycam GAGs builders (www.glycam.com), then the sulfation patterns and aliphatic portion was modified on GaussView 5.0.[7] The modified ligand pdb files were then subjected to energy minimization in YASARA, and saved in .yob format. Each ligand was docked separately into heparanase within the simulation cell (80 x 61 x 76 Å) using Autodock VINA[8] default parameters. The ligand and enzyme were subjected to 100 docking runs, where AMBER14 force field[9] was applied to the protein, and GLYCAM06[10] and GAFF/Am1BCC force filed was applied to each ligand. During docking study, the ligands and receptor binding pocket residues were kept flexible. The most populated clusters of the docking runs was subjected to MD simulation.

9.2. Docking Results for HS Trisaccharides

Figure S6: Snapshots of the first cluster when ligands docked into human heparanase and the distance between the catalytic residues and the scissile bond

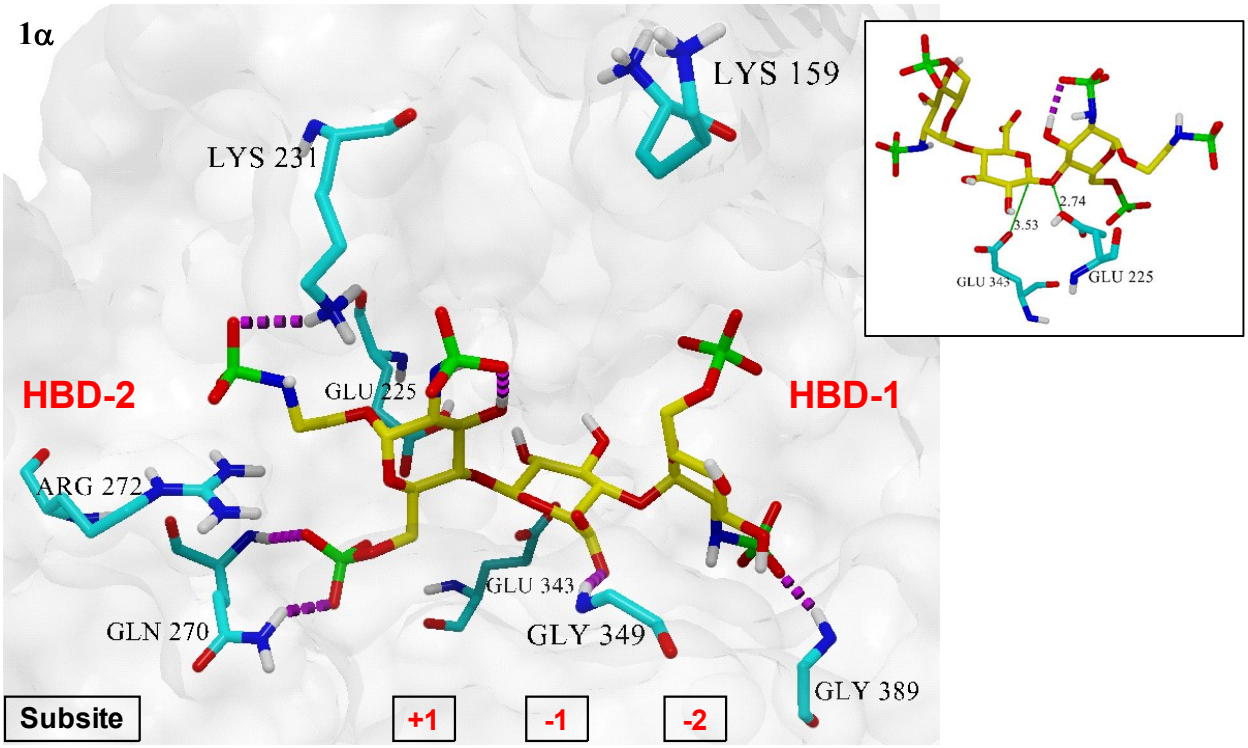
Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
001	016	000009.5228+-000000.5844	00000000104647.2293
002	002	000009.0810+-000000.0220	00000000220586.4078
003	002	000008.8185+-000000.0695	00000000343552.2965

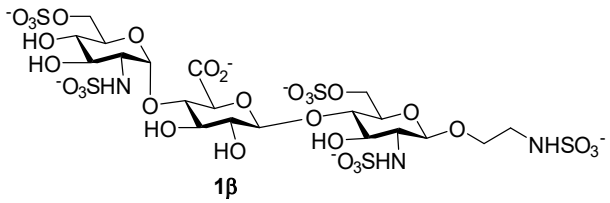




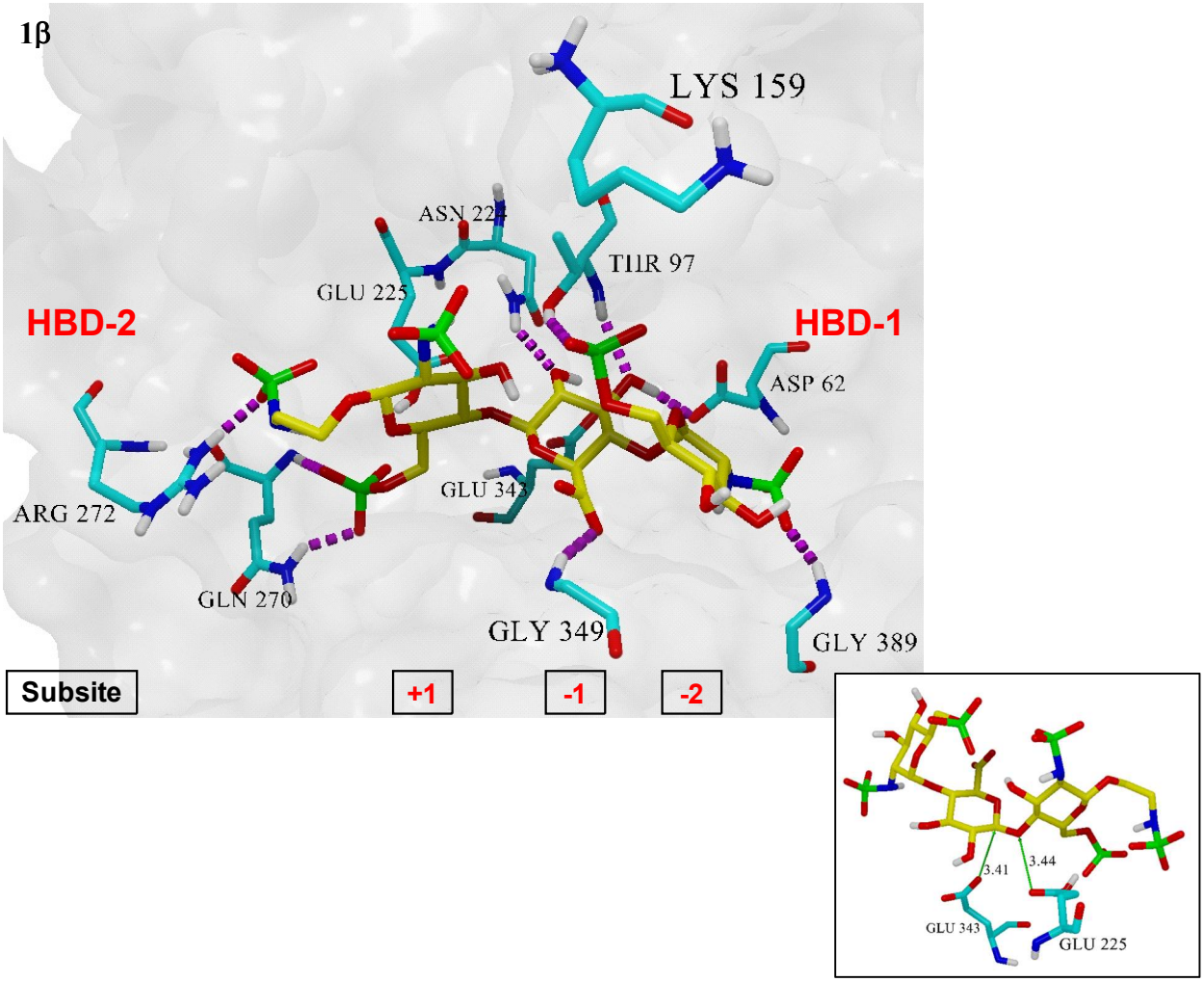
1α

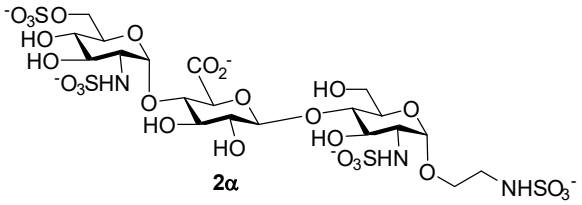
Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
001	010	000010.1993+-000000.7108	00000000033408.6388
002	003	000009.0003+-000000.2895	00000000252759.5868
003	001	000009.3710+-000000.0000	00000000135209.3990
004	004	000008.6332+-000000.3046	00000000469658.2439
005	001	000008.4040+-000000.0000	00000000691549.1274
006	001	000007.6410+-000000.0000	00000002506723.6529





Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
001	010	000009.2158+-000001.1214	00000000175699.1364
002	001	000008.5790+-000000.0000	00000000514691.4643
003	001	000007.9040+-000000.0000	00000001608148.1296
004	002	000007.4785+-000000.2465	00000003297765.3798
005	004	000007.3618+-000000.1473	00000004016040.0966
006	001	000007.3470+-000000.0000	00000004117274.1365
007	001	000007.1290+-000000.0000	00000005948464.8219

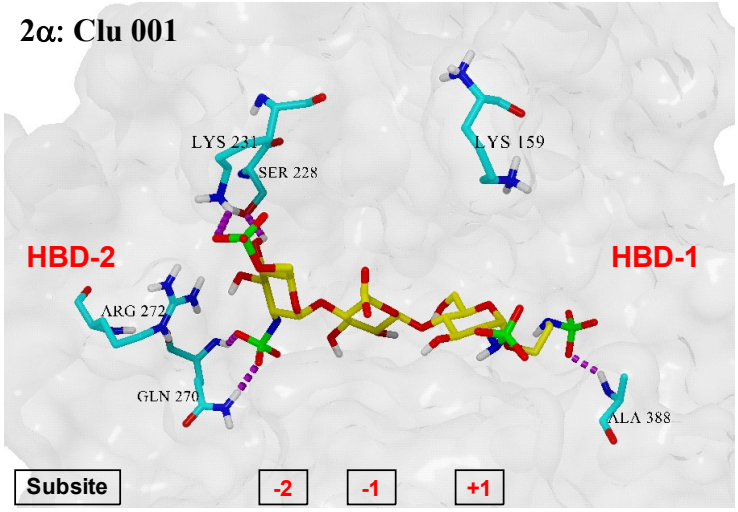


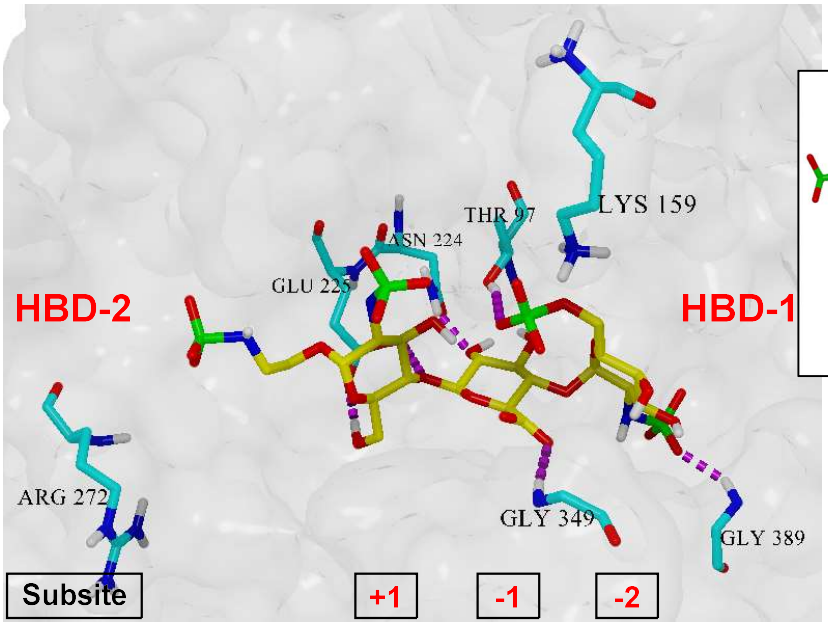


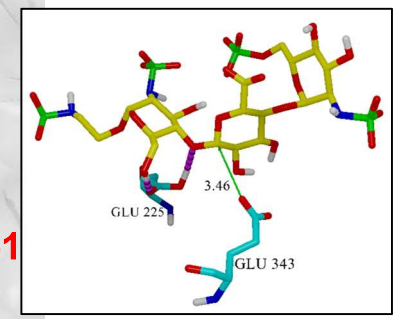
2α

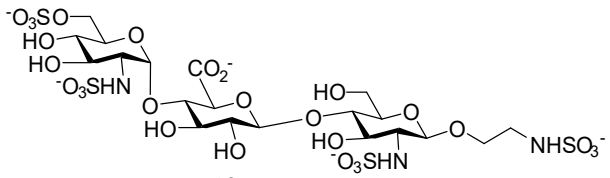
Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
001	007	000008.3014+-000000.7134	000000000822258.8077
002	006	000008.9397+-000000.2119	000000000280012.0893
003	001	000007.9780+-000000.0000	000000001419330.0346
004	002	000007.7145+-000000.1695	000000002214269.3720
005	002	000007.2910+-000000.5280	000000004525413.3424
006	001	000007.6090+-000000.0000	000000002645832.3817
007	001	000006.7540+-000000.0000	000000011201647.3126

2α: Clu 001





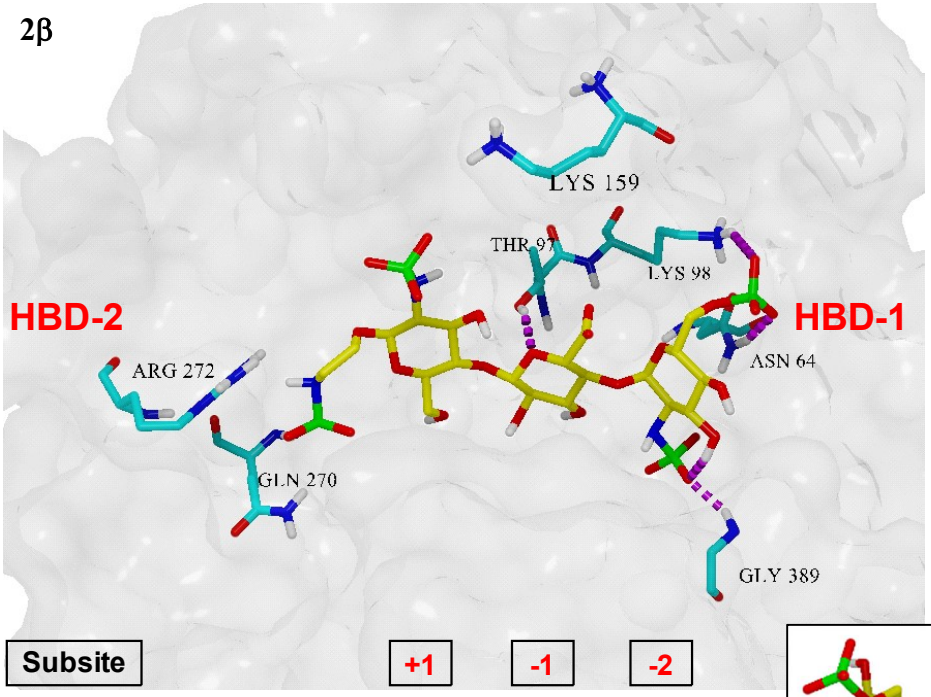


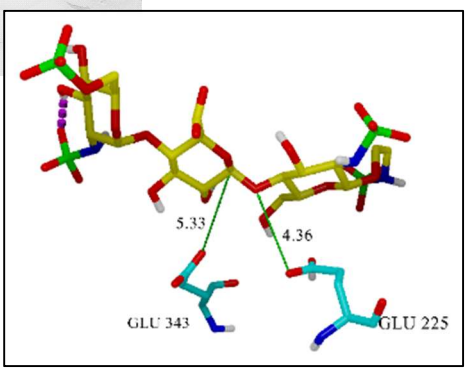


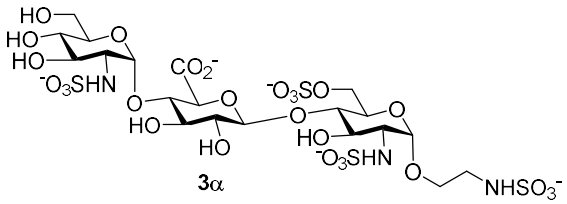
2β

Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
001	003	000008.9960+-000000.2861	00000000254615.0745
002	001	000009.2060+-000000.0000	00000000178629.3151
003	002	000008.4735+-000000.6395	00000000615006.0169
004	004	000007.4480+-000000.4922	00000003471971.8437
005	001	000007.8580+-000000.0000	00000001737978.0922
006	001	000006.8340+-000000.0000	00000009786818.2811
007	001	000006.7600+-000000.0000	00000011088781.6526
008	002	000006.6335+-000000.1095	00000013728057.8518
009	001	000006.6810+-000000.0000	00000012670426.9642
010	001	000006.6700+-000000.0000	00000012907865.2525
011	001	000006.6240+-000000.0000	00000013949950.6377
012	002	000006.4580+-000000.0050	00000018460848.0877

2β







3α

Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
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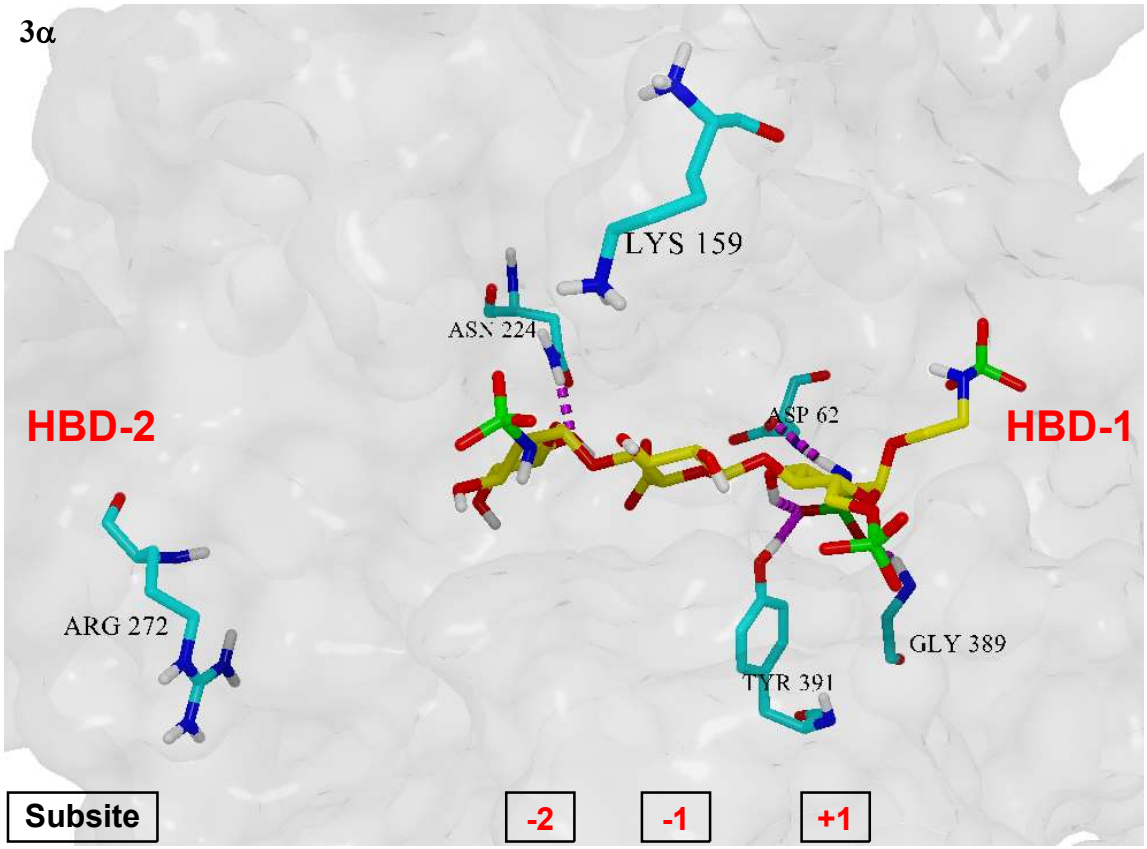
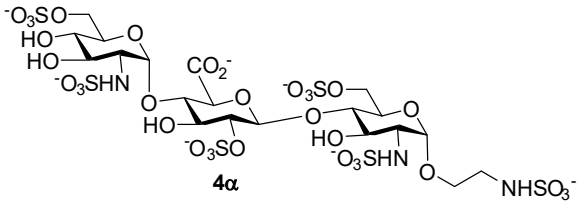
003	001	000007.5370+-000000.0000	00000002987711.9929
004	003	000007.0517+-000000.2647	00000006777830.8857
005	001	000007.0270+-000000.0000	00000007065967.1517
006	001	000006.9620+-000000.0000	00000007885277.8732
007	001	000006.8680+-000000.0000	00000009241009.1910
008	001	000006.8120+-000000.0000	00000010157056.5632
009	001	000006.7580+-000000.0000	00000011126282.5828
010	001	000006.7230+-000000.0000	00000011803346.6062





4α

Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
001	008	000009.8015+-000000.4618	00000000065380.5138
002	004	000009.3918+-000000.4370	00000000130556.0984
003	001	000009.4400+-000000.0000	00000000120345.5497
004	003	000008.3830+-000000.5373	00000000716500.3913
005	001	000007.6750+-000000.0000	00000002366922.2220
006	001	000007.5600+-000000.0000	00000002873953.7925
007	001	000007.5340+-000000.0000	00000003002879.7170
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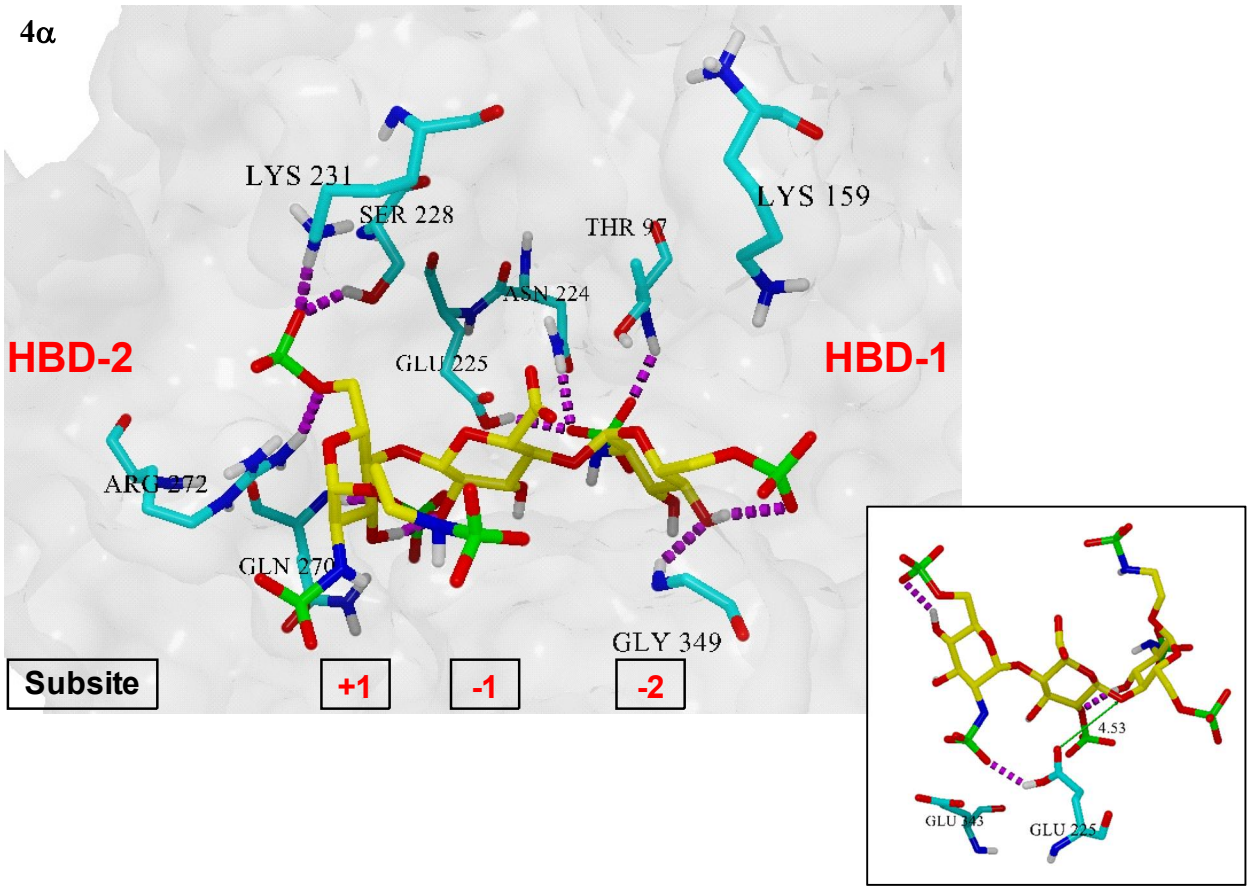

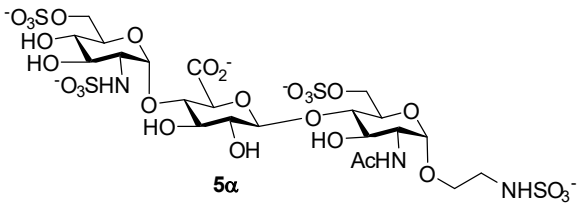

5α

Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
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003	003	000008.4820+-000000.4666	000000000606246.0493
004	005	000008.0832+-000000.2766	000000001188423.8783
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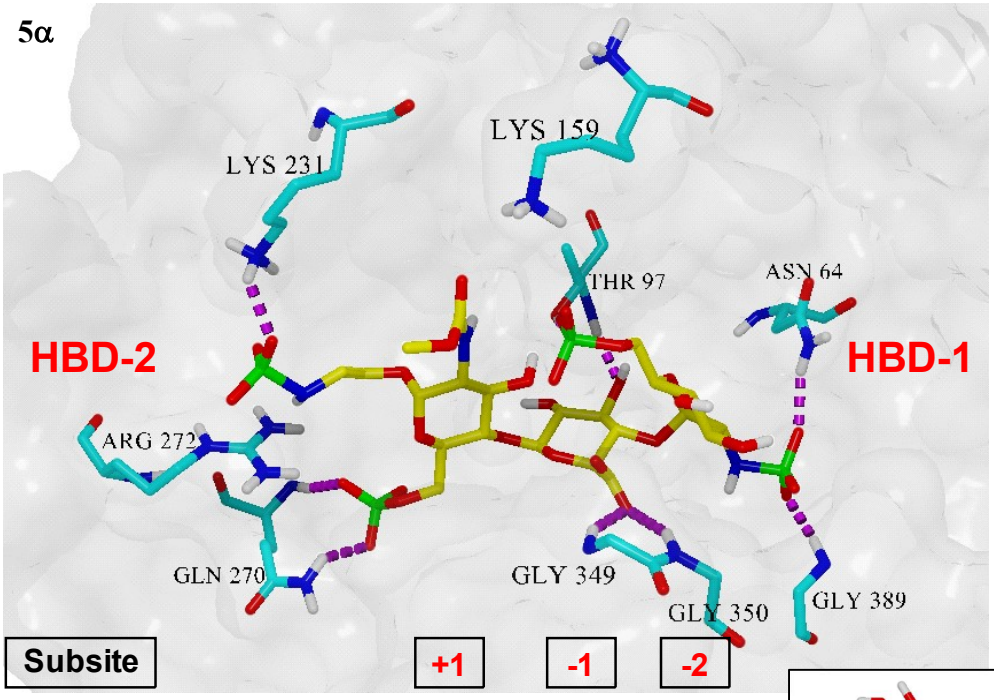

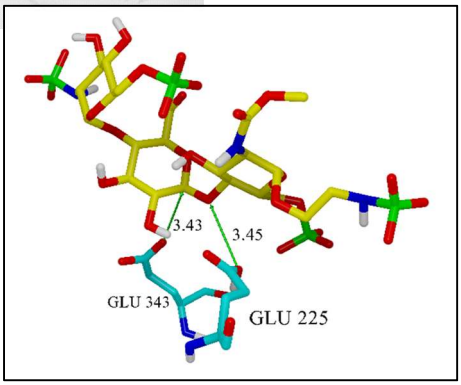

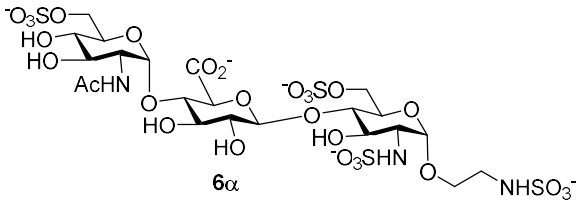

6α

Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
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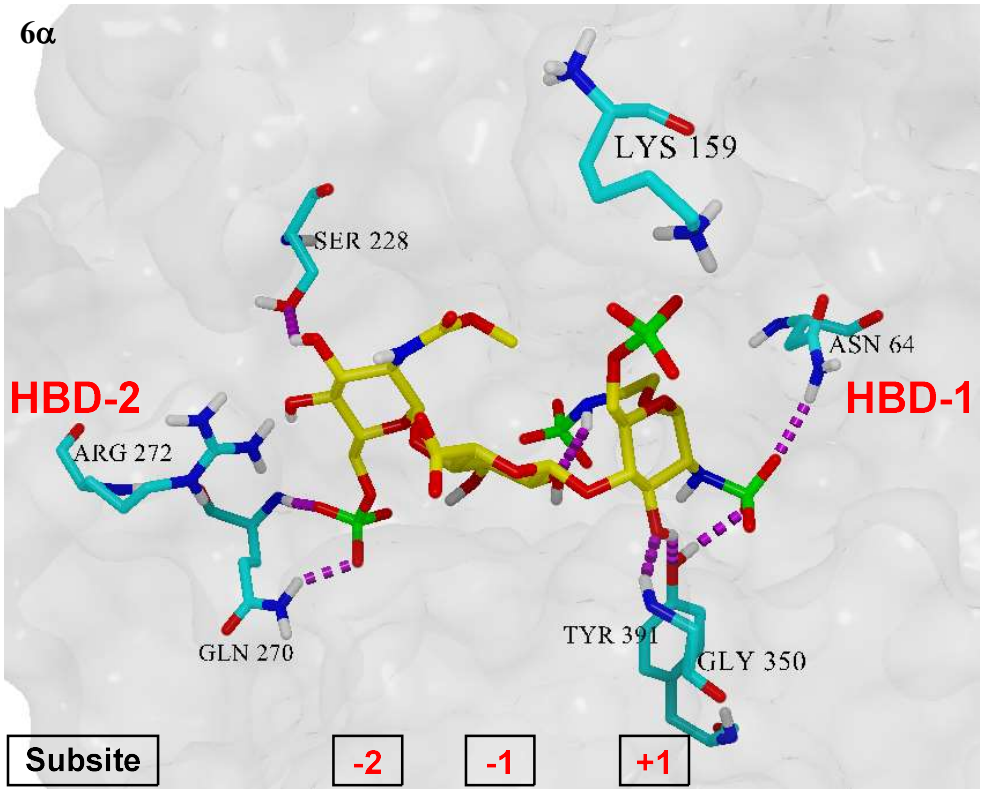
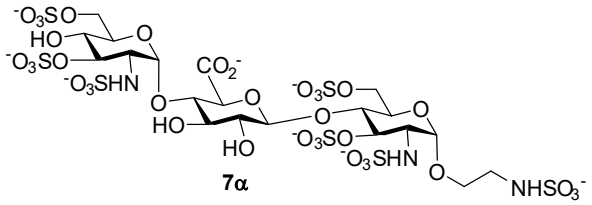
003	001	000008.4690+-000000.0000	00000000619695.0101
004	001	000008.3330+-000000.0000	00000000779590.4108
005	001	000008.2310+-000000.0000	00000000926047.3751
006	001	000008.2140+-000000.0000	00000000953003.3801
007	001	000008.0510+-000000.0000	00000001254799.6455
008	003	000007.6833+-000000.1532	00000002333864.8466
009	003	000007.2710+-000000.1149	00000004680781.2506
010	004	000006.9325+-000000.1604	00000008287826.7693
011	001	000006.8150+-000000.0000	00000010105752.6661
012	001	000006.7300+-000000.0000	00000011664715.3546
013	001	000006.6450+-000000.0000	00000013464171.2299





Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
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003	002	000008.5505+-000000.1175	00000000540055.0431
004	002	000007.8640+-000000.0630	00000001720465.8644
005	002	000007.6770+-000000.0990	00000002358946.4528
006	004	000007.2752+-000000.1354	00000004647324.9099
007	001	000007.3750+-000000.0000	00000003927224.4646
008	001	000007.3030+-000000.0000	00000004434678.3692
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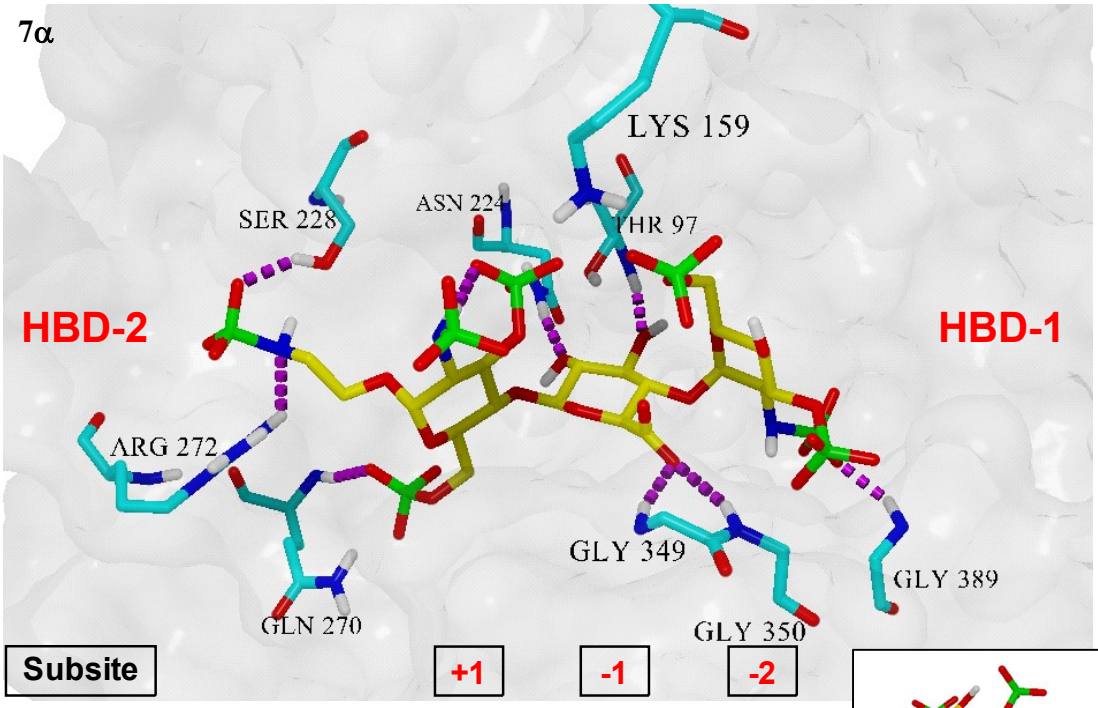
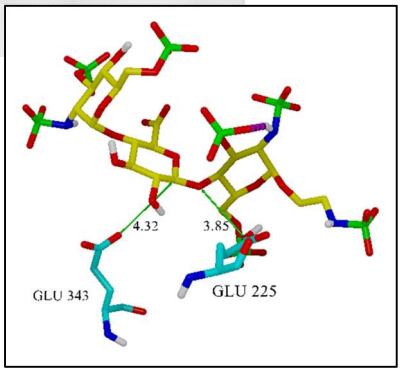
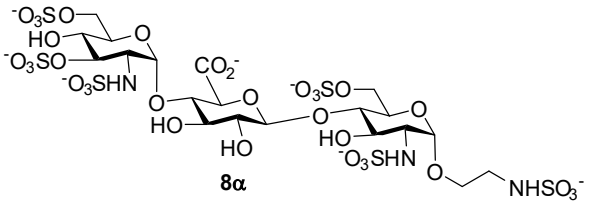
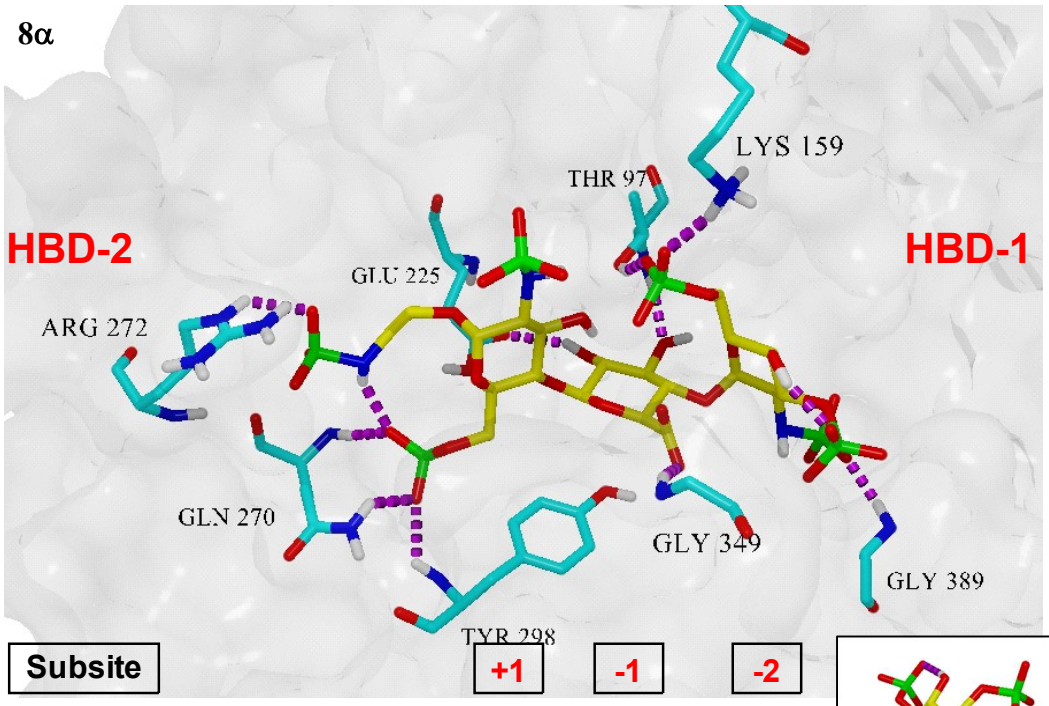


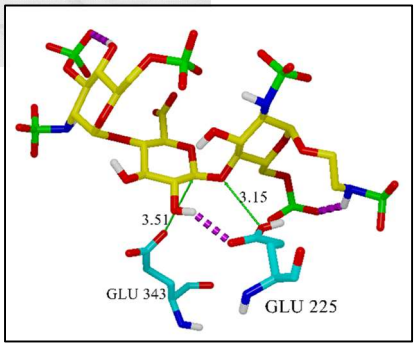


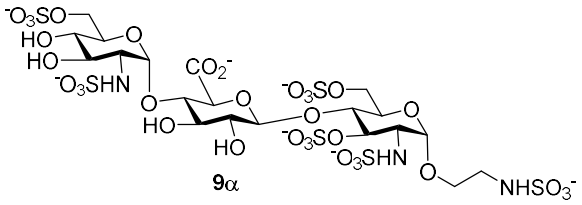


8α

Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
001	007	000010.2929+-000000.6847	00000000028528.6276
002	003	000009.9593+-000000.2828	00000000050090.5229
003	004	000009.7858+-000000.1594	00000000067141.8071
004	001	000009.7630+-000000.0000	00000000069770.1005
005	004	000009.5748+-000000.1096	00000000095864.3483
006	001	000009.5040+-000000.0000	00000000108023.3395

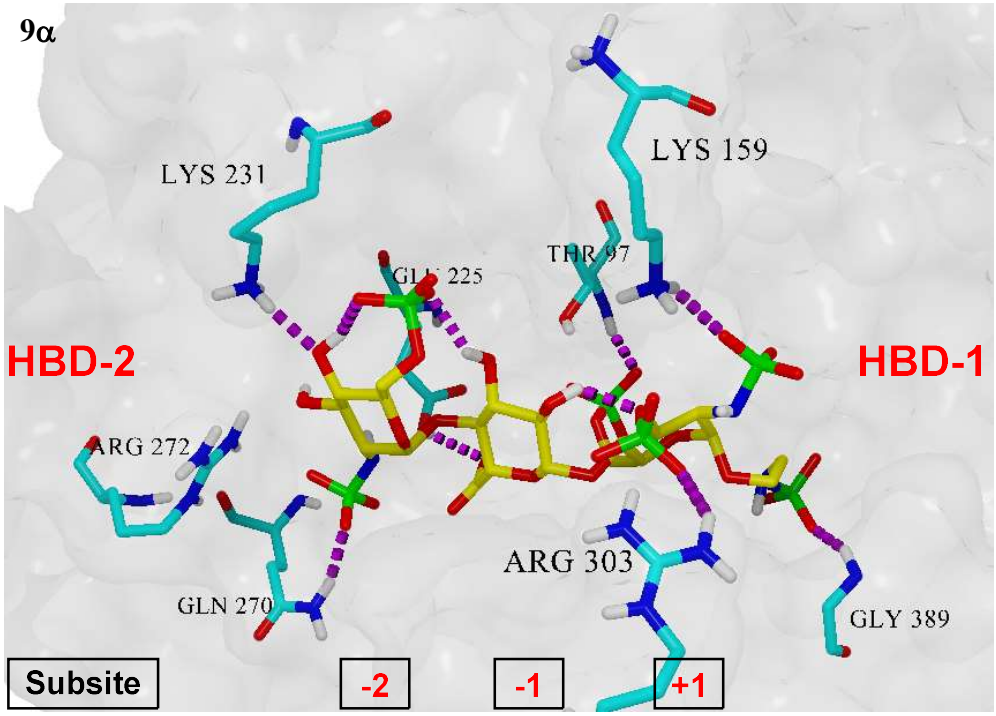






9α

Clu	Members	Bind.energy spread [kcal/mol]	Dissoc. constant [pM]
001	001	000010.2700+-000000.0000	00000000029650.7014
002	001	000009.2050+-000000.0000	00000000178931.1836
003	001	000008.6190+-000000.0000	00000000481090.4998
004	001	000008.1180+-000000.0000	00000001120631.2920
005	001	000007.9030+-000000.0000	00000001610864.4683
006	002	000007.6850+-000000.0140	00000002327309.3568
007	002	000007.4250+-000000.0720	00000003609402.5303
008	003	000007.3183+-000000.0573	00000004321382.5274
009	002	000007.2120+-000000.0790	00000005170894.4260
010	001	000007.2650+-000000.0000	00000004728423.9477
011	001	000007.2530+-000000.0000	00000004825169.0517
012	002	000007.1125+-000000.1095	00000006116453.2049
013	001	000007.1910+-000000.0000	00000005357457.2666
014	001	000007.0480+-000000.0000	00000006819908.8375



9.3. Molecular Dynamic Simulations

The most populated clusters (heparanase-ligand complexes) from the each docking run were subjected to MD simulation. Each enzyme-ligand complex was imported into YASARA and energy minimized in *vacuo*. Meanwhile, a simulation cell was formulated with at least 10 Å from all three sides of the enzyme-ligand complex (86 x 61x 59 Å for **1α** complex and 67 x 89 x 61 Å for **1β** complex). AMBER14 force field was applied with periodic boundary conditions. The force field parameters were assigned using YASARA's "AutoSMILES" feature. The simulations were run in a default physiological solution (pH 7.4, 0.9% m/v NaCl solution with TIP3P water,[11] water density set at 0.997 g/mL) at 298 K. The pK_a of titratable side chains was predicted and assigned to respective groups before running MD and Glu 225 was manually protonated.

MD simulations were performed for 25000 ps for both **1α** and **1β** complexes initially. As suspicious movements were found in **1α** complex at the end of the simulation, an extra 5000 ps were added to the duration for **1α** complex. The cutoff radius for long-rage electrostatics was set to 8 Å. The snapshots were saved every 25 ps for the whole simulation duration, which were then analyzed for the RMSD of the Cα and heavy atoms of ligand-protein complex, as well as the distance between the side chain carboxylic carbons of catalytic residues and the scissile bond (Glu225-O1, Glu343-C1).

Figure S7. RMSD of the Cα and heavy atoms of (A) **1α** and (B) **1β** complex

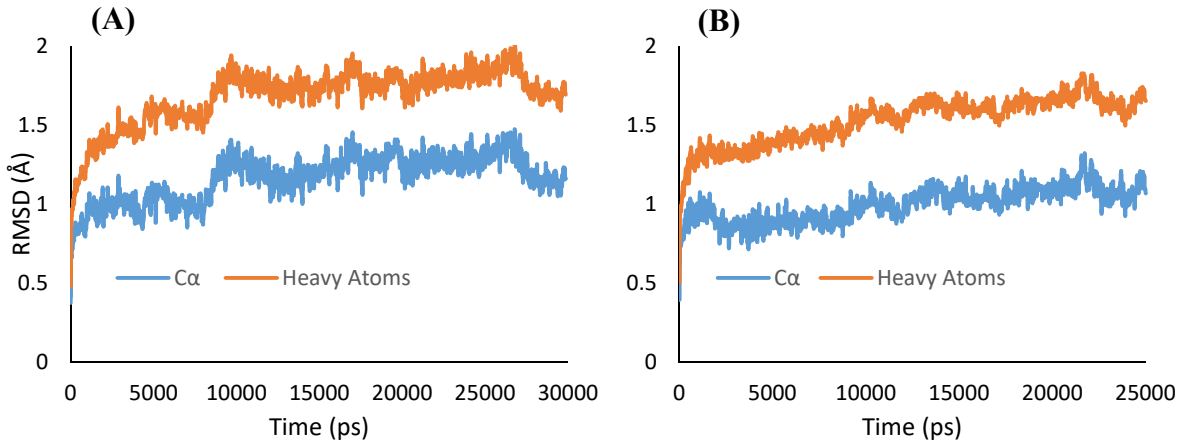


Figure S8. Evolution of distances between catalytic residues and the scissile bond during the simulation in (A) **1α** and (B) **1β** complex

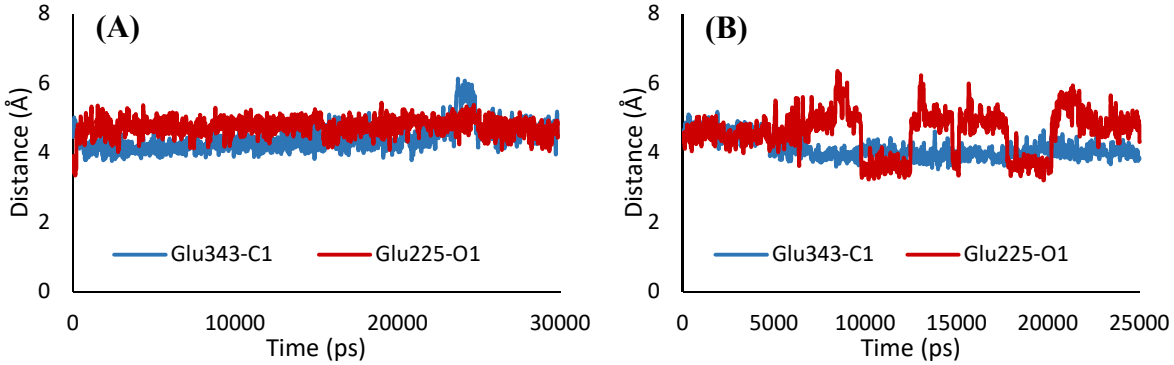
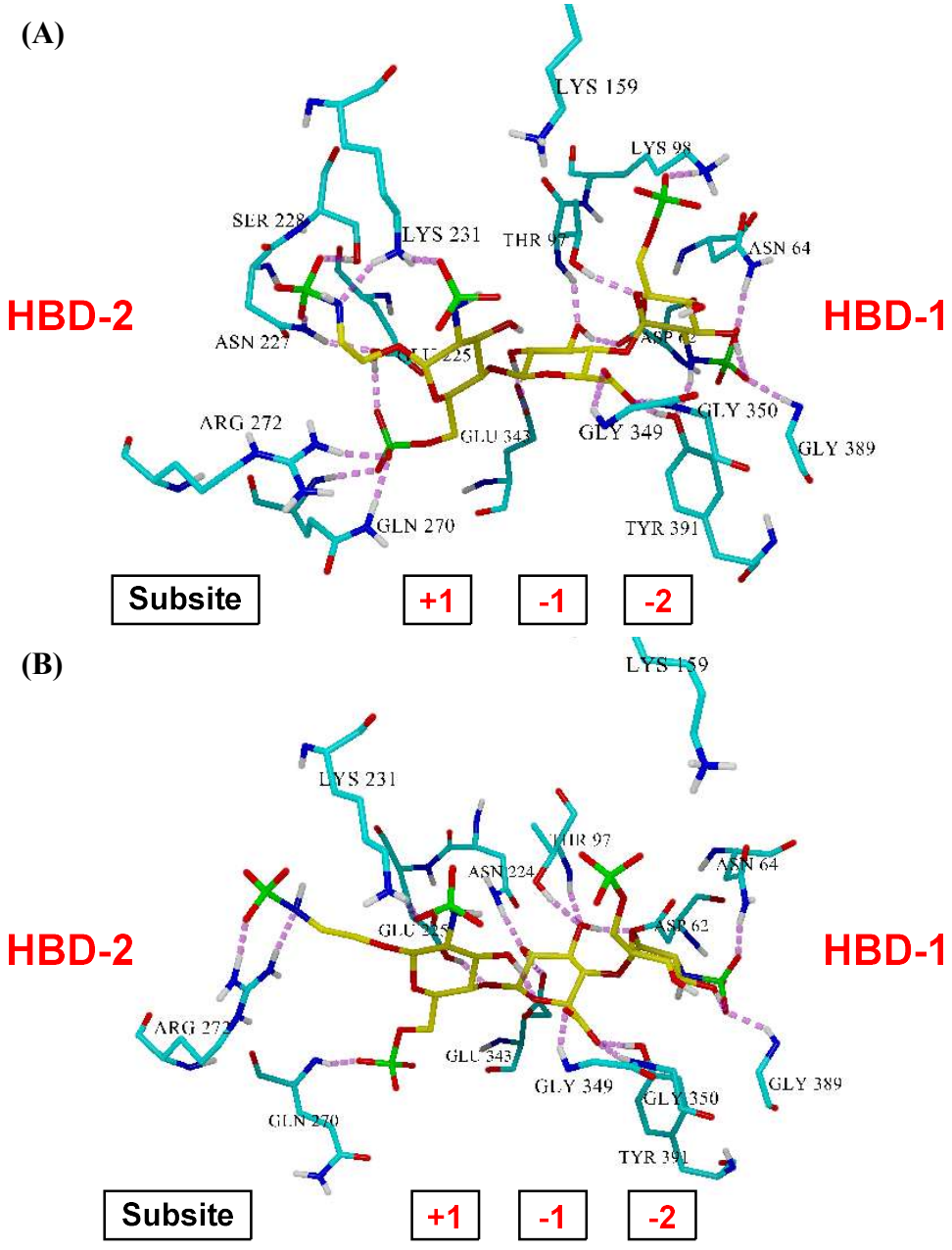


Figure S9. Example snapshots of hydrogen bonding pattern of (A) **1α** and (B) **1β** complex during MD simulation



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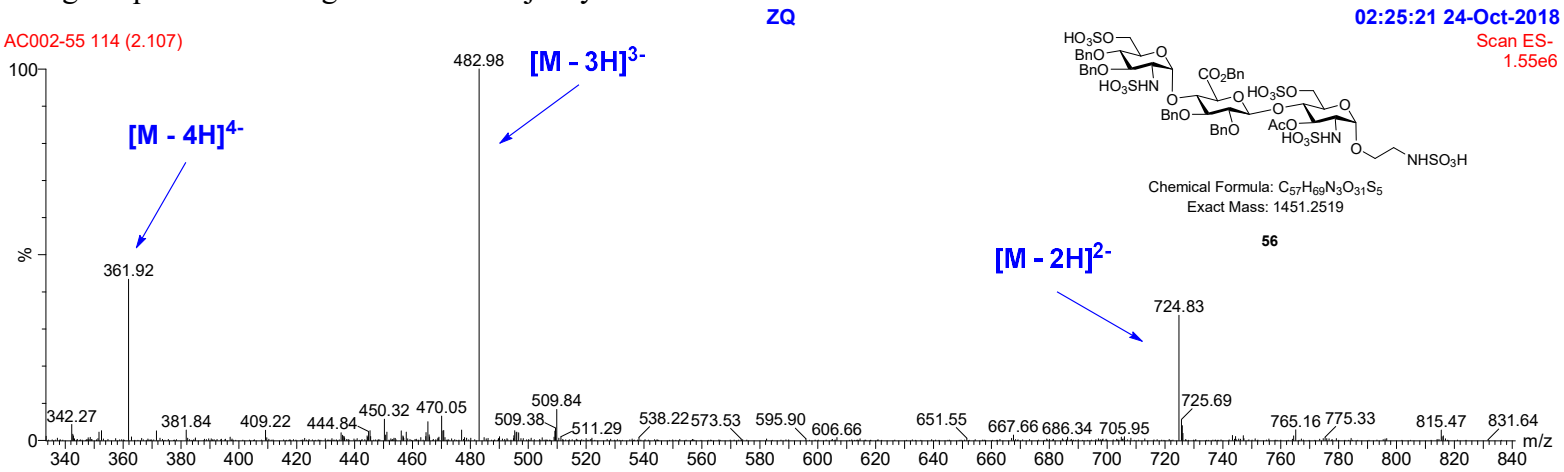
11. Mass Spectra

11.1. Detection for Microwave-Assisted Sulfonation Reaction

Using 3 equiv SO_3·TEA gave **56** as a majority



Using 5 equiv SO_3·TEA produced more **57** as side-product

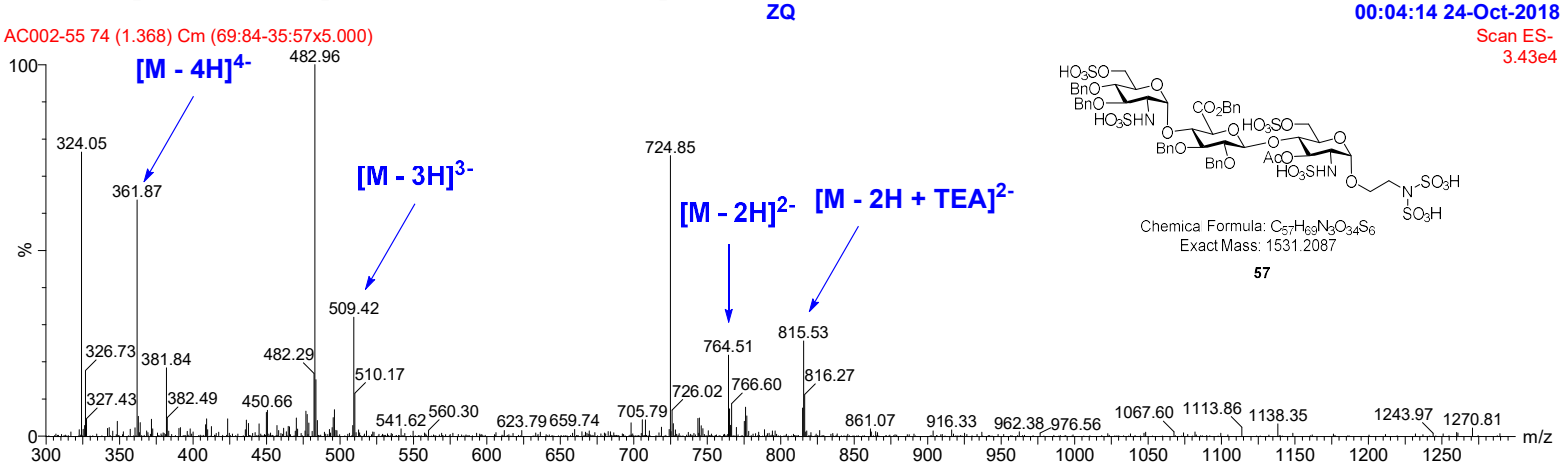
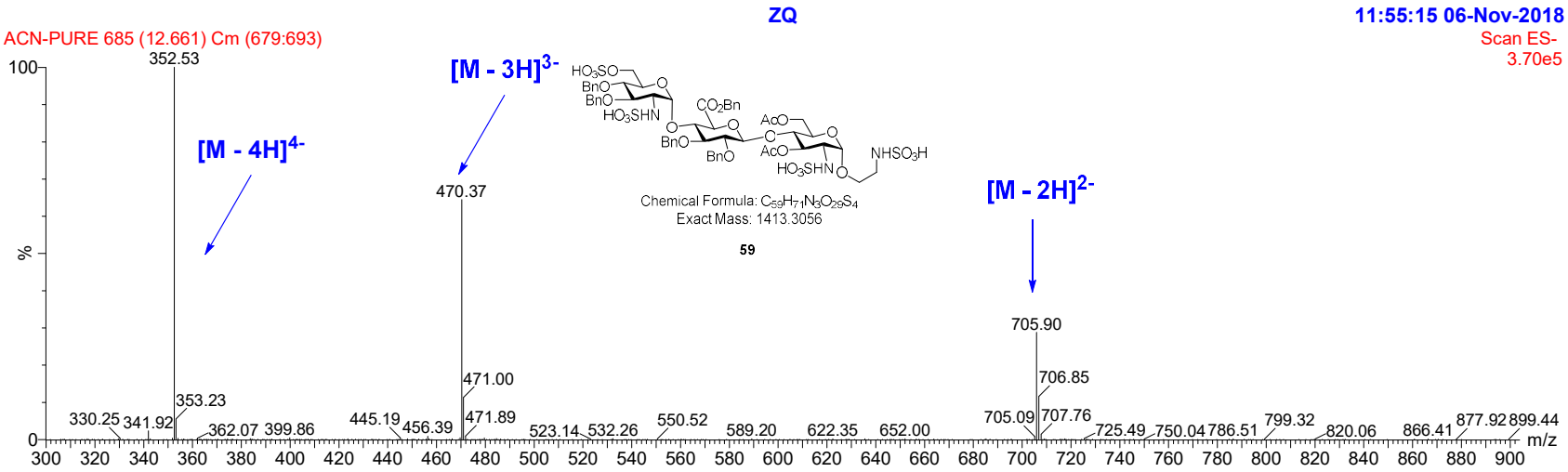


Figure S6. Microwave assisted sulfonation monitored by negative-mode mass, indicating 3 equiv SO_3·TEA was good.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Scan ES-
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[M - 3H]3-
[M - 4H]4-
[M - 2H]2-
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Exact Mass: 1413.3056
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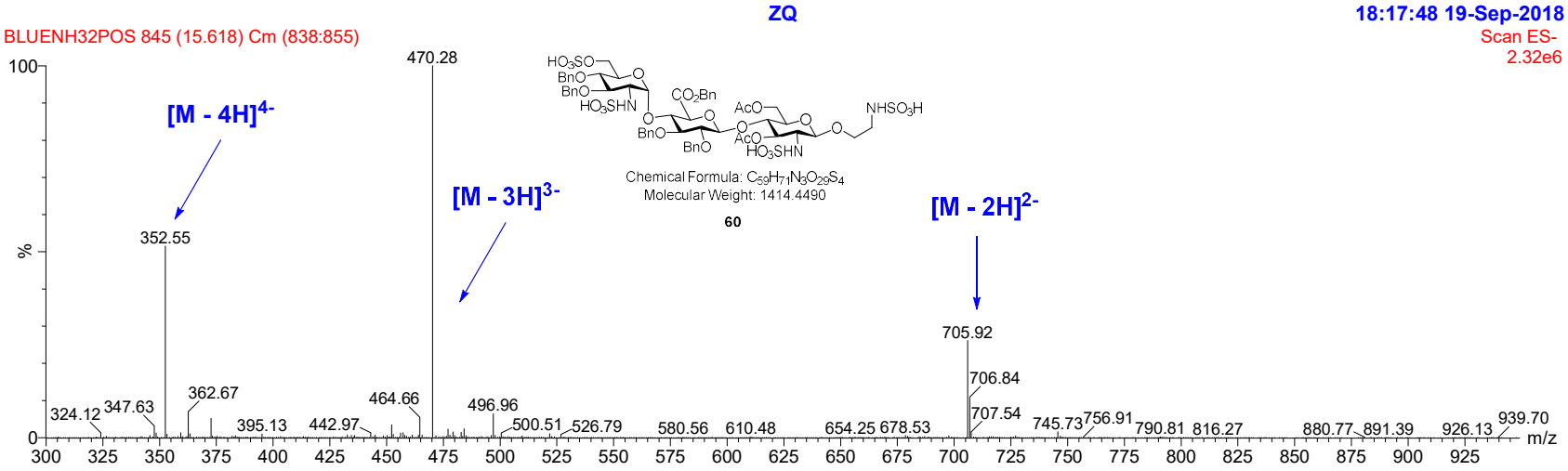
352.53
470.37
705.90
471.00
706.85
353.23
471.89
707.76
330.25 341.92 362.07 399.86 445.19 456.39 523.14 532.26 550.52 589.20 622.35 652.00 705.09 725.49 750.04 786.51 799.32 820.06 866.41 877.92 899.44
m/z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Scan ES-
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[M - 3H]3-
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707.54
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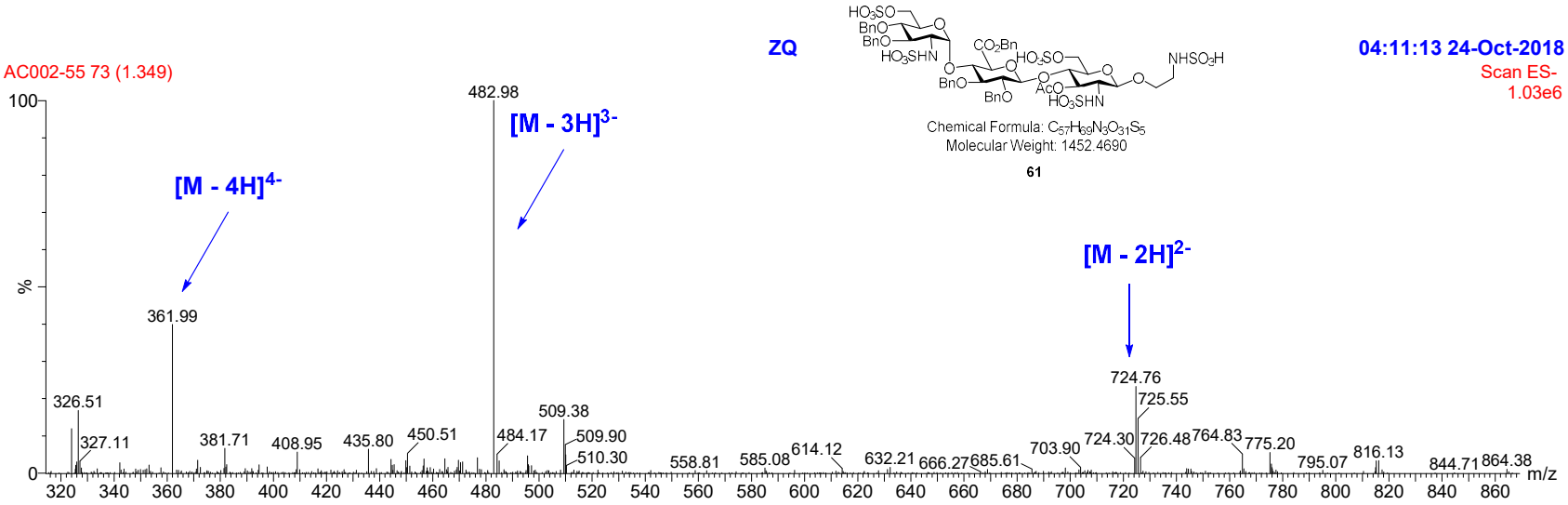
m/z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[M - 2H]2-
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361.99
724.76
725.55
509.38
509.90
510.30
484.17
450.51
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408.95
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326.51
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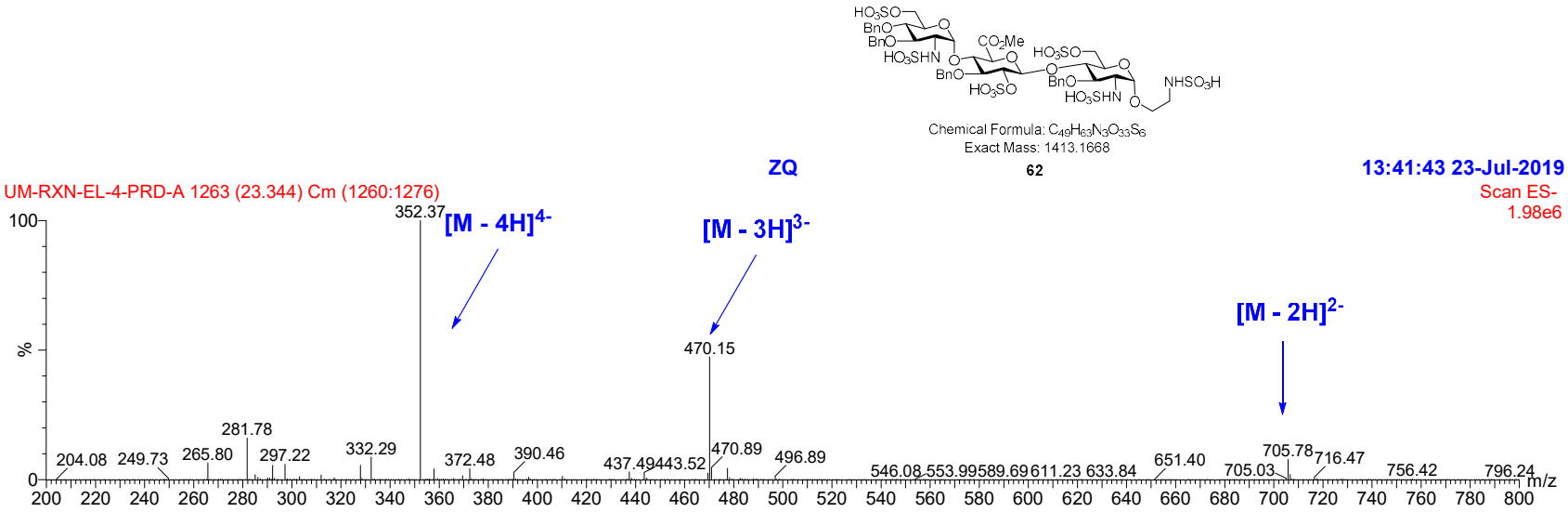
m/z
%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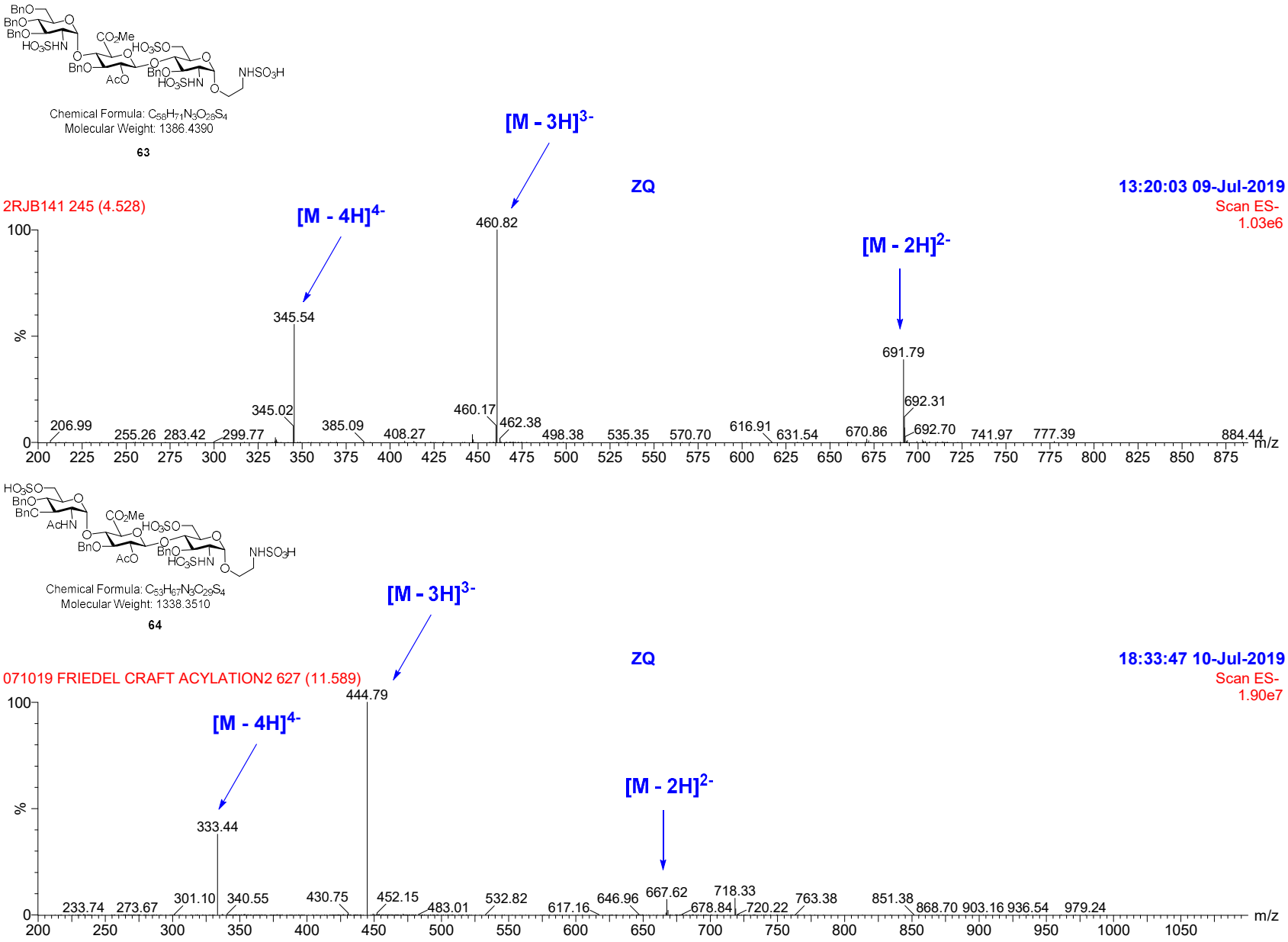
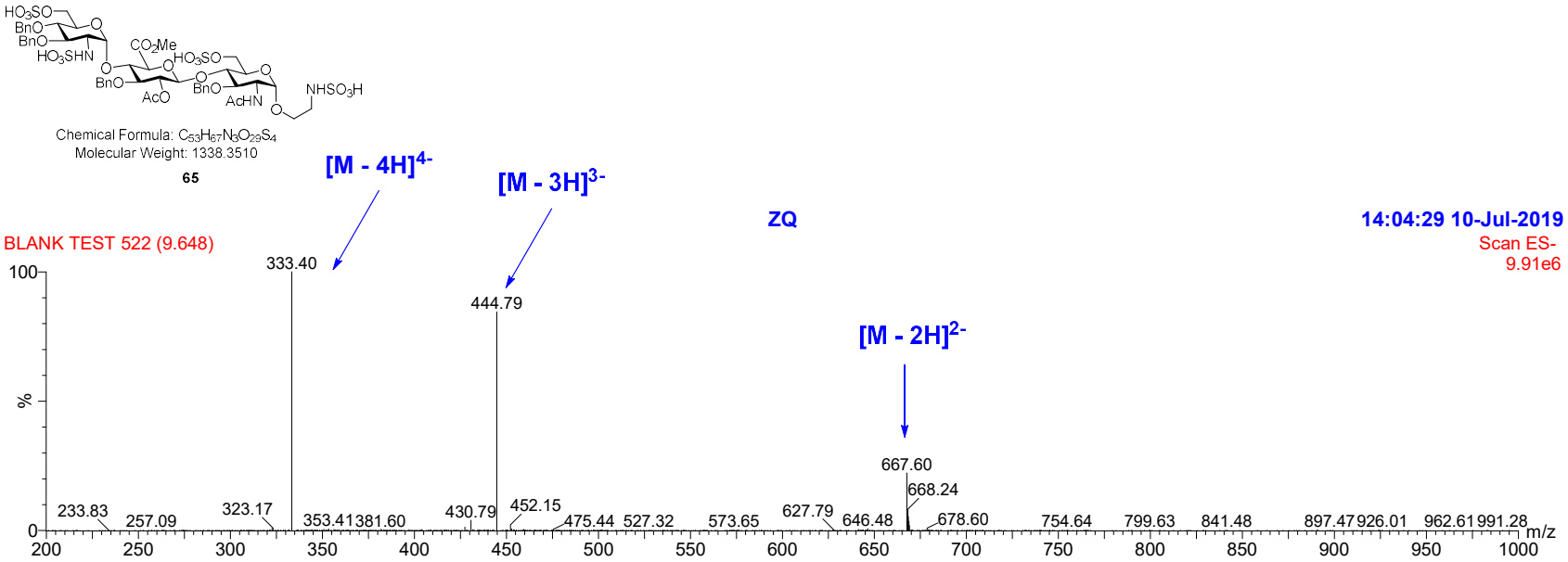
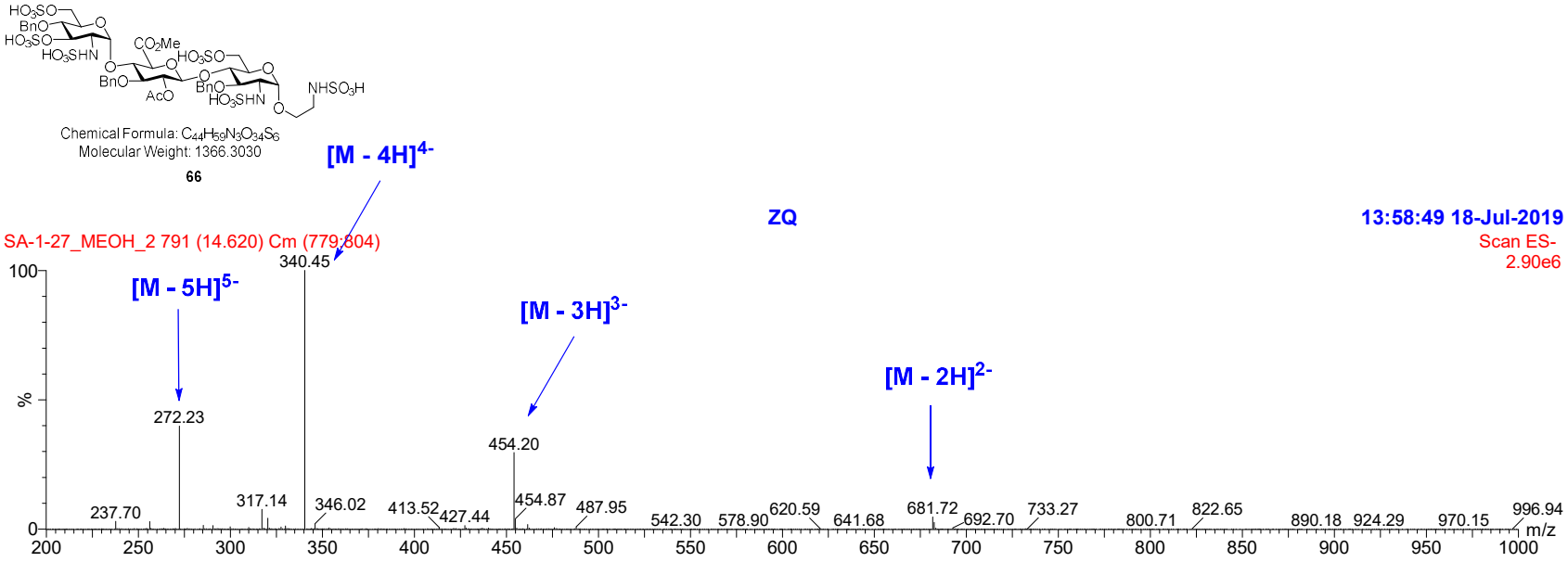
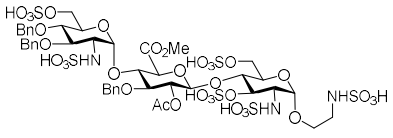
ZQ
13:41:43 23-Jul-2019
Scan ES-
1.98e6
[M - 4H]4-
[M - 3H]3-
[M - 2H]2-
352.37
470.15
281.78
204.08
249.73
265.80
297.22
332.29
372.48
390.46
437.49
443.52
470.89
496.89
546.08
553.99
589.69
611.23
633.84
651.40
705.03
705.78
716.47
756.42
796.24
m/z
%


Chemical Formula: C58H71N3O28S4
Molecular Weight: 1386.4390
63
[M - 3H]3-
ZQ
13:20:03 09-Jul-2019
2RJB141 245 (4.528)
Scan ES-
1.03e6
[M - 4H]4-
460.82
[M - 2H]2-
345.54
691.79
692.31
345.02
460.17
462.38
206.99
255.26
283.42
299.77
385.09
408.27
498.38
535.35
570.70
616.91
631.54
670.86
692.70
741.97
777.39
884.44
m/z
Chemical Formula: C53H67N3O29S4
Molecular Weight: 1338.3510
64
[M - 3H]3-
ZQ
18:33:47 10-Jul-2019
071019 FRIEDEL CRAFT ACYLATION2 627 (11.589)
Scan ES-
1.90e7
444.79
[M - 4H]4-
[M - 2H]2-
333.44
667.62
718.33
233.74
273.67
301.10
340.55
430.75
452.15
483.01
532.82
617.16
646.96
678.84
720.22
763.38
851.38
868.70
903.16
936.54
979.24
m/z

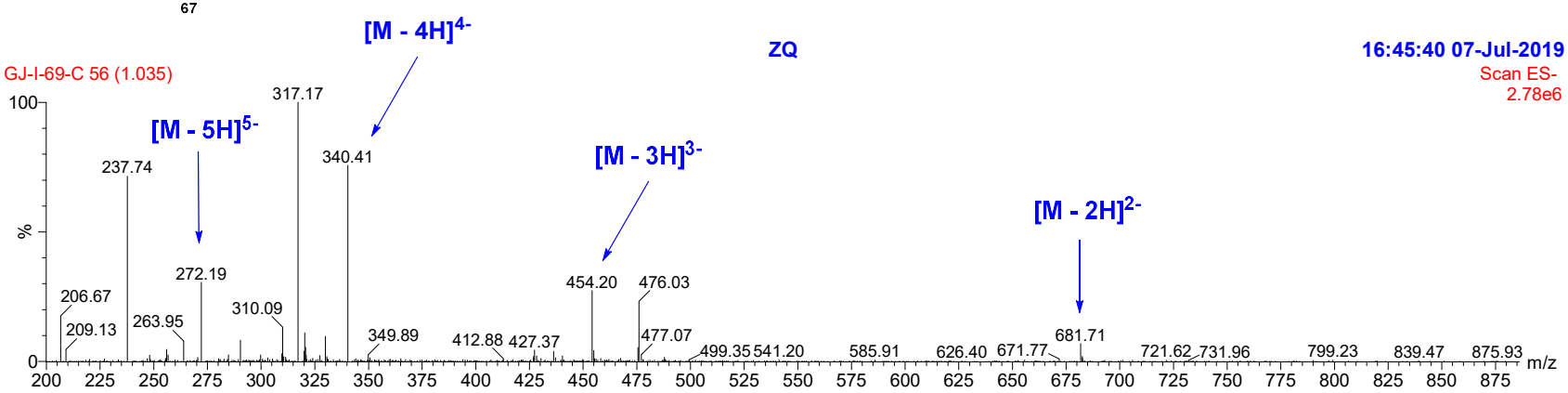

Chemical Formula: C53H67N3O29S4
Molecular Weight: 1338.3510
65
[M - 4H]4-
[M - 3H]3-
[M - 2H]2-
ZQ
14:04:29 10-Jul-2019
BLANK TEST 522 (9.648)
Scan ES-
9.91e6
333.40
444.79
667.60
668.24
233.83
257.09
323.17
353.41
381.60
430.79
452.15
475.44
527.32
573.65
627.79
646.48
678.60
754.64
799.63
841.48
897.47
926.01
962.61
991.28
m/z

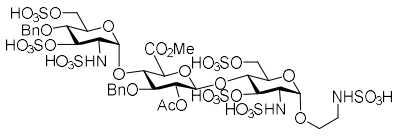

Chemical Formula: C44H59N3O34S6
Molecular Weight: 1366.3030
66
[M - 4H]4-
[M - 5H]5-
[M - 3H]3-
[M - 2H]2-
ZQ
13:58:49 18-Jul-2019
SA-1-27_MEOH_2 791 (14.620) Cm (779:804)
Scan ES-
2.90e6
340.45
272.23
454.20
237.70
317.14
346.02
413.52
427.44
454.87
487.95
542.30
578.90
620.59
641.68
681.72
692.70
733.27
800.71
822.65
890.18
924.29
970.15
996.94
m/z



Chemica Formula: $C_{44}H_{59}N_3C_{34}S_6$
Molecular Weight: 1366.3030

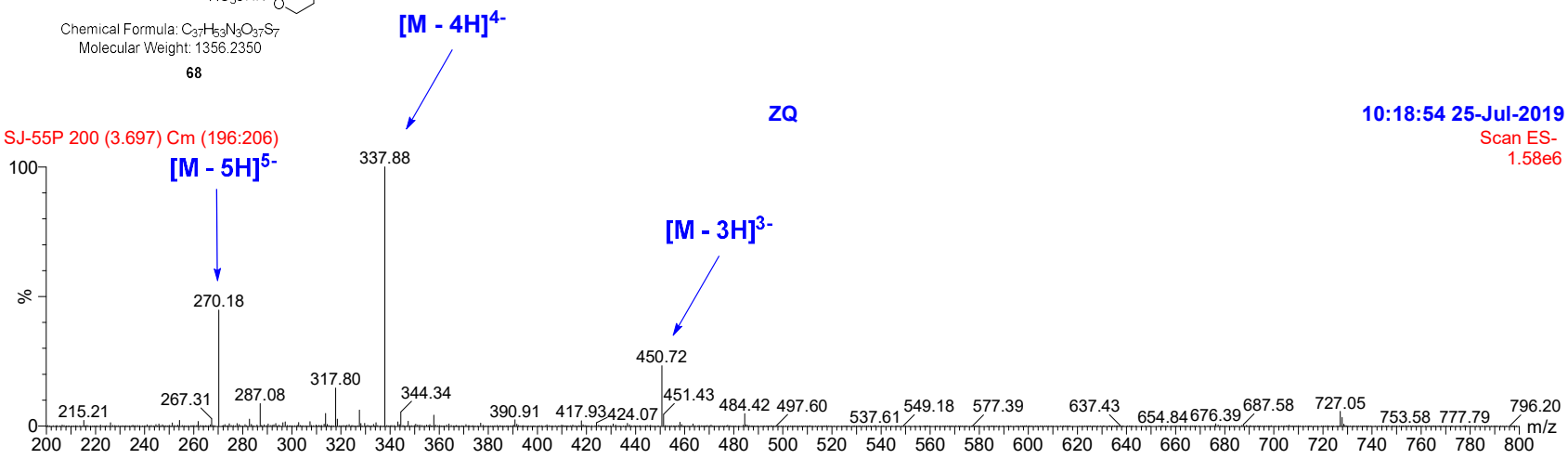
67



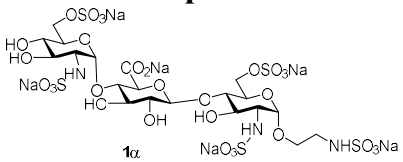


Chemical Formula: $C_{37}H_{53}N_3O_{37}S_7$
Molecular Weight: 1356.2350

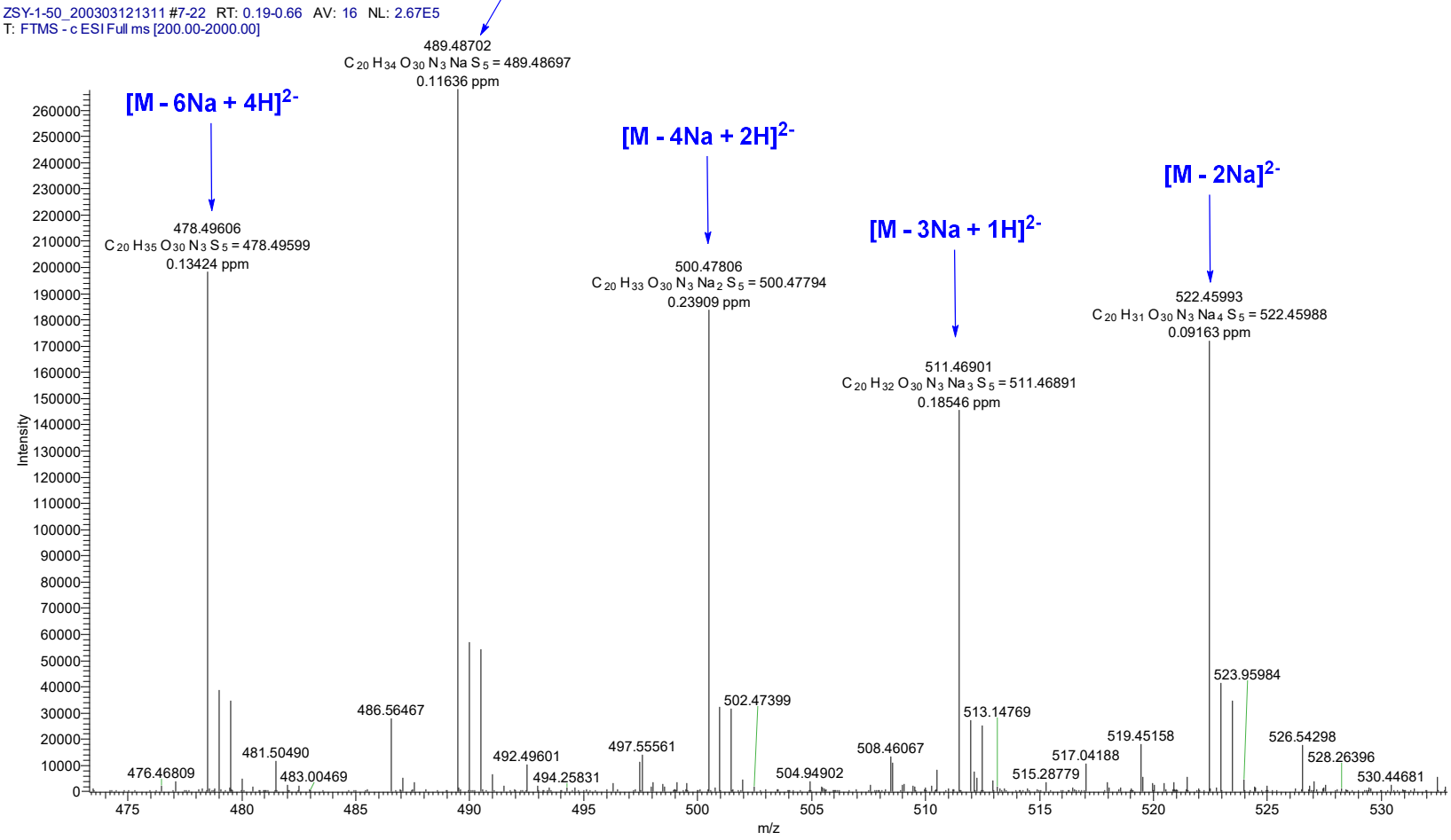
68

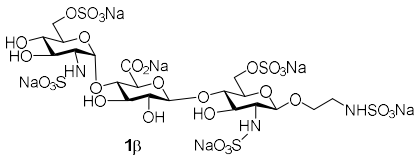


11.2. Mass Spectra for the Final HS Trisaccharides



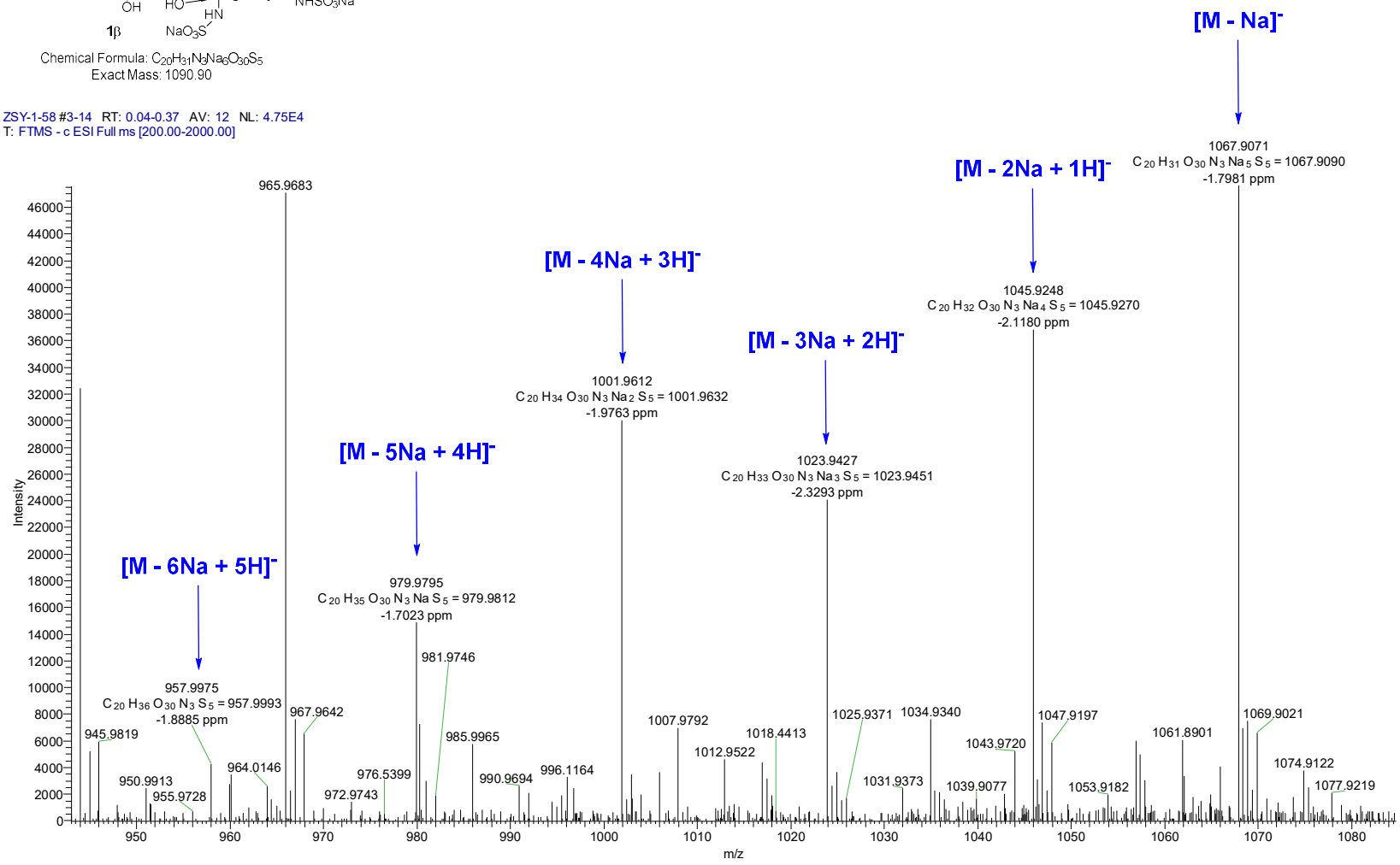
Chemical Formula: $C_{20}H_{31}N_3Na_6O_{30}S_5$
Exact Mass: 1090.90

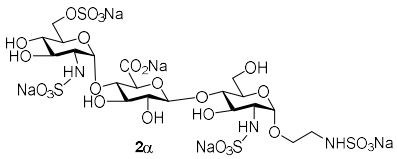




Chemical Formula: $C_{20}H_{31}N_3Na_6O_{30}S_5$
Exact Mass: 1090.90

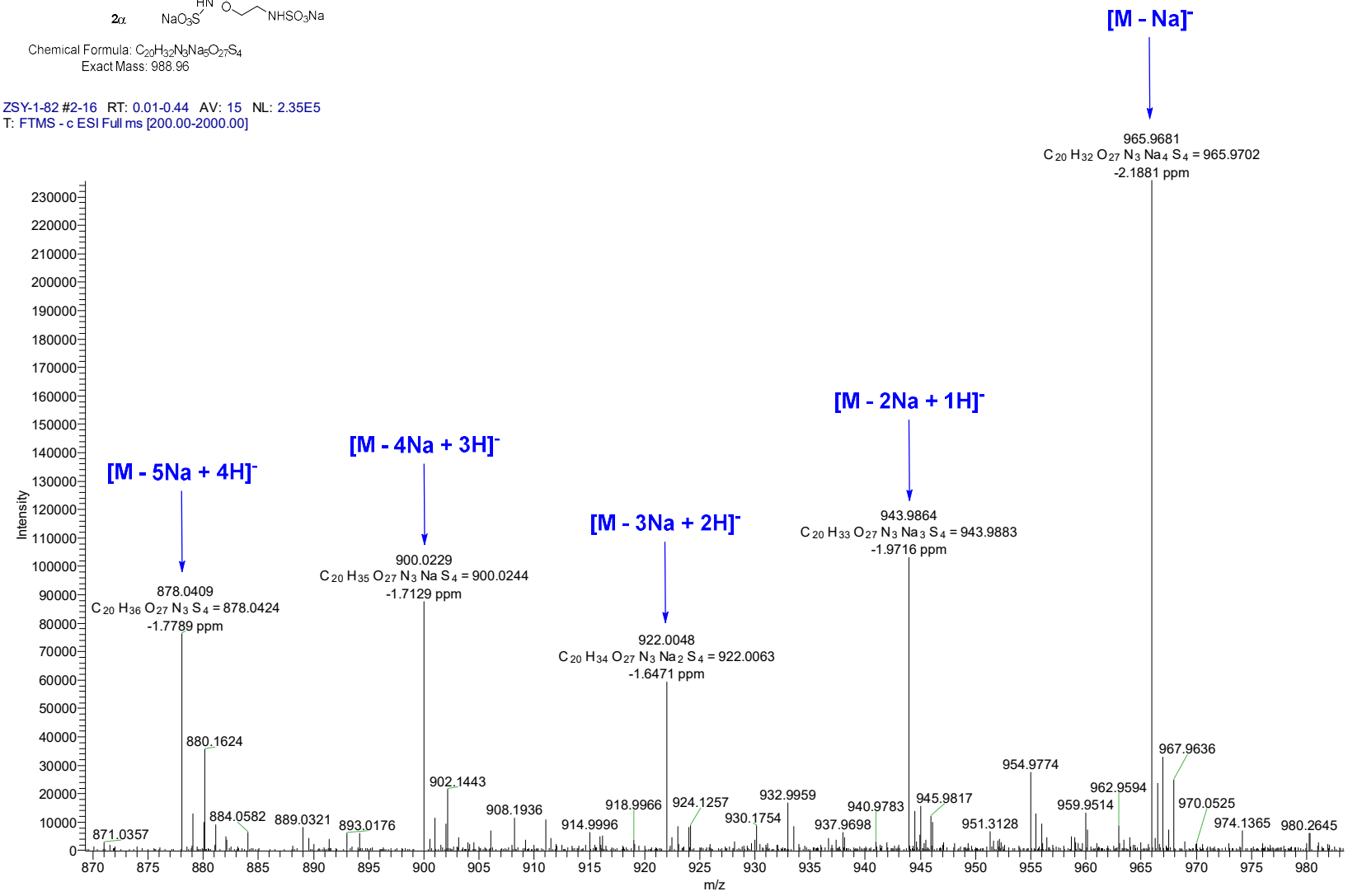
ZSY-1-58 #3-14 RT: 0.04-0.37 AV: 12 NL: 4.75E4
T: FTMS - c ESI Full ms [200.00-2000.00]

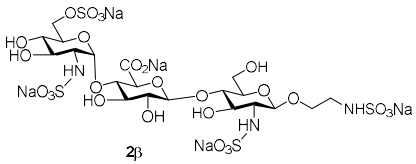




Chemical Formula: $C_{20}H_{32}N_3Na_5O_{27}S_4$
Exact Mass: 988.96

ZSY-1-82 #2-16 RT: 0.01-0.44 AV: 15 NL: 2.35E5
T: FTMS - c ESI Full ms [200.00-2000.00]

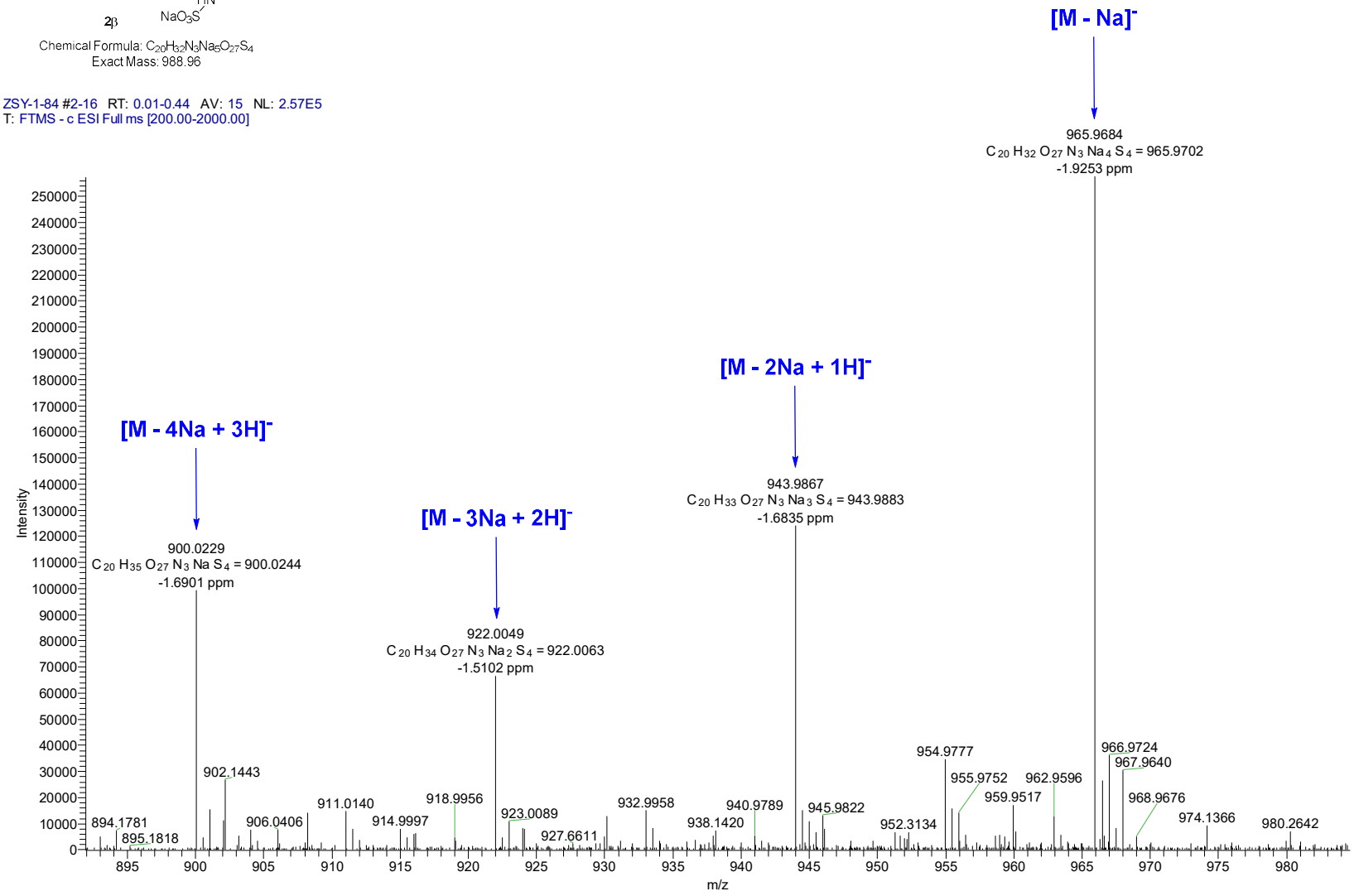


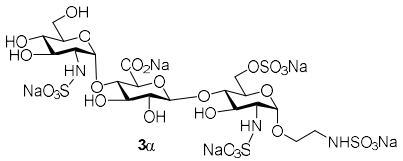


2β

Chemical Formula: $C_{20}H_{32}N_3Na_5O_{27}S_4$
Exact Mass: 988.96

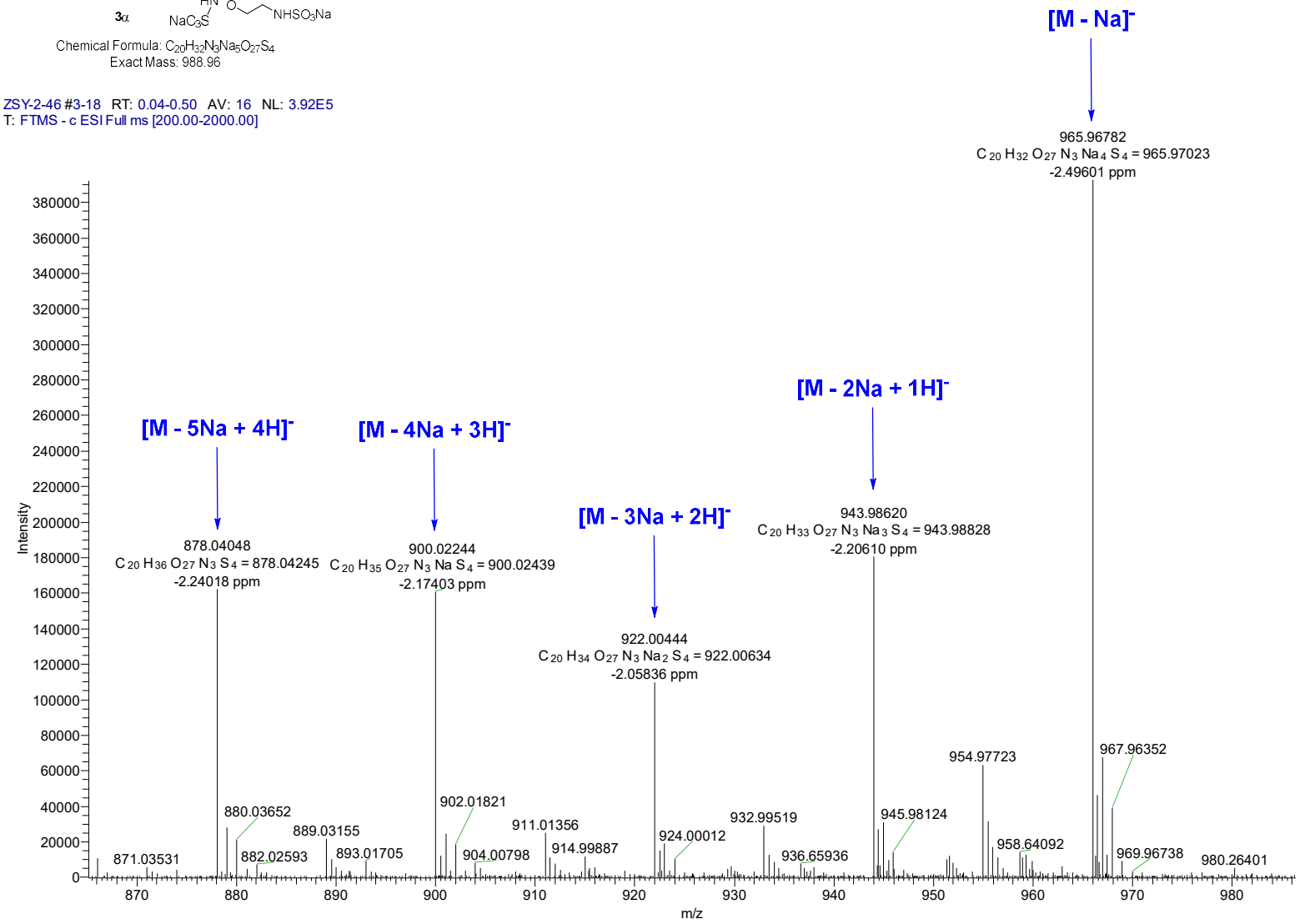
ZSY-1-84 #2-16 RT: 0.01-0.44 AV: 15 NL: 2.57E5
T: FTMS - c ESI Full ms [200.00-2000.00]

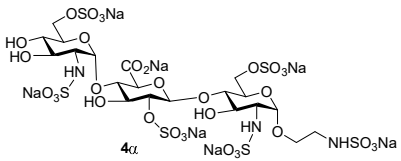




Chemical Formula: $C_{20}H_{32}N_3Na_5O_{27}S_4$
Exact Mass: 988.96

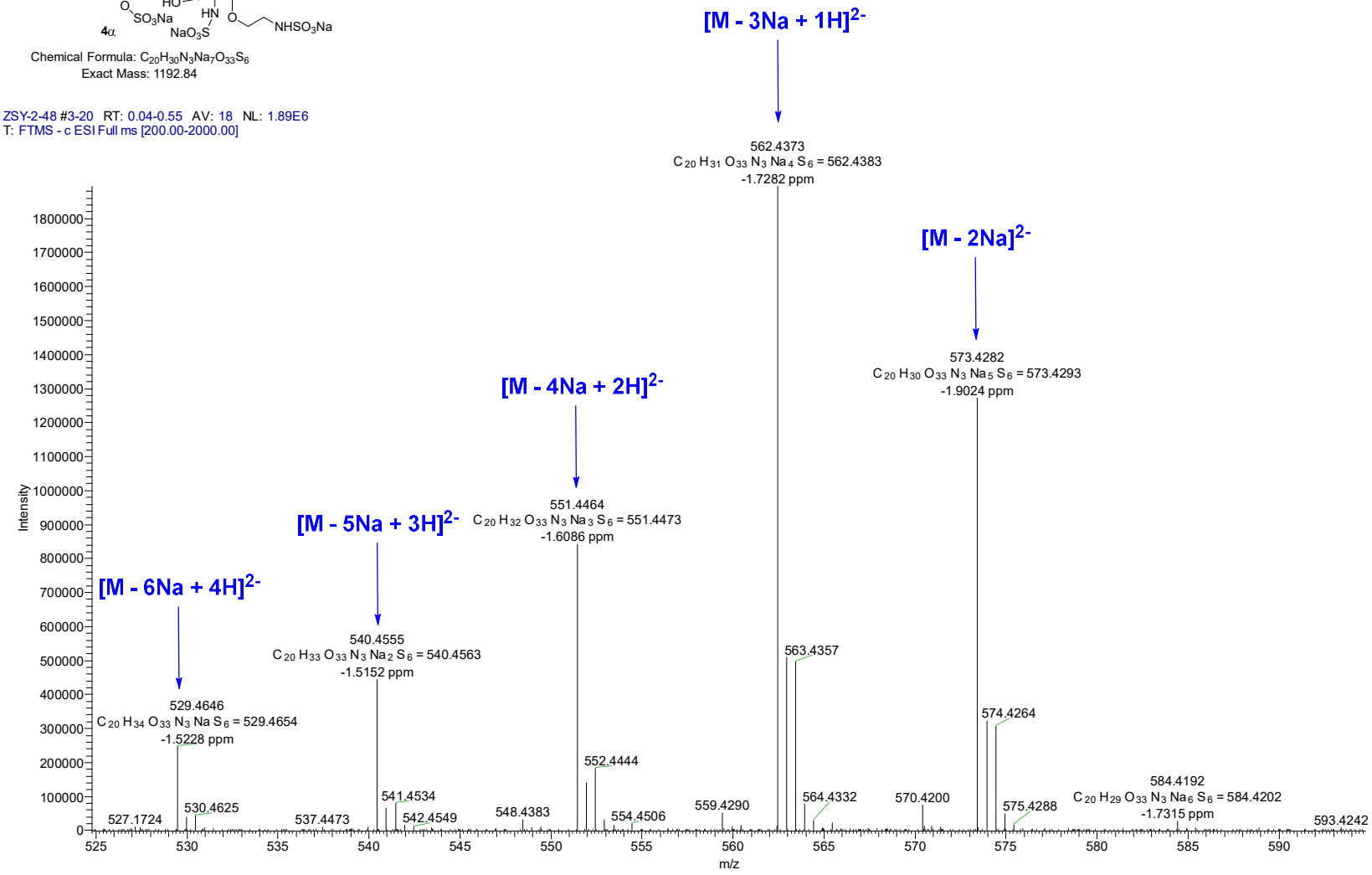
ZSY-2-46 #3-18 RT: 0.04-0.50 AV: 16 NL: 3.92E5
T: FTMS - c ESI Full ms [200.00-2000.00]

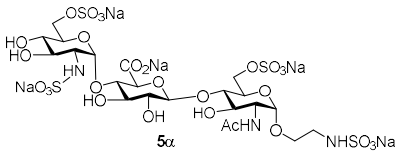




Chemical Formula: $C_{20}H_{30}N_3Na_7O_{33}S_6$
Exact Mass: 1192.84

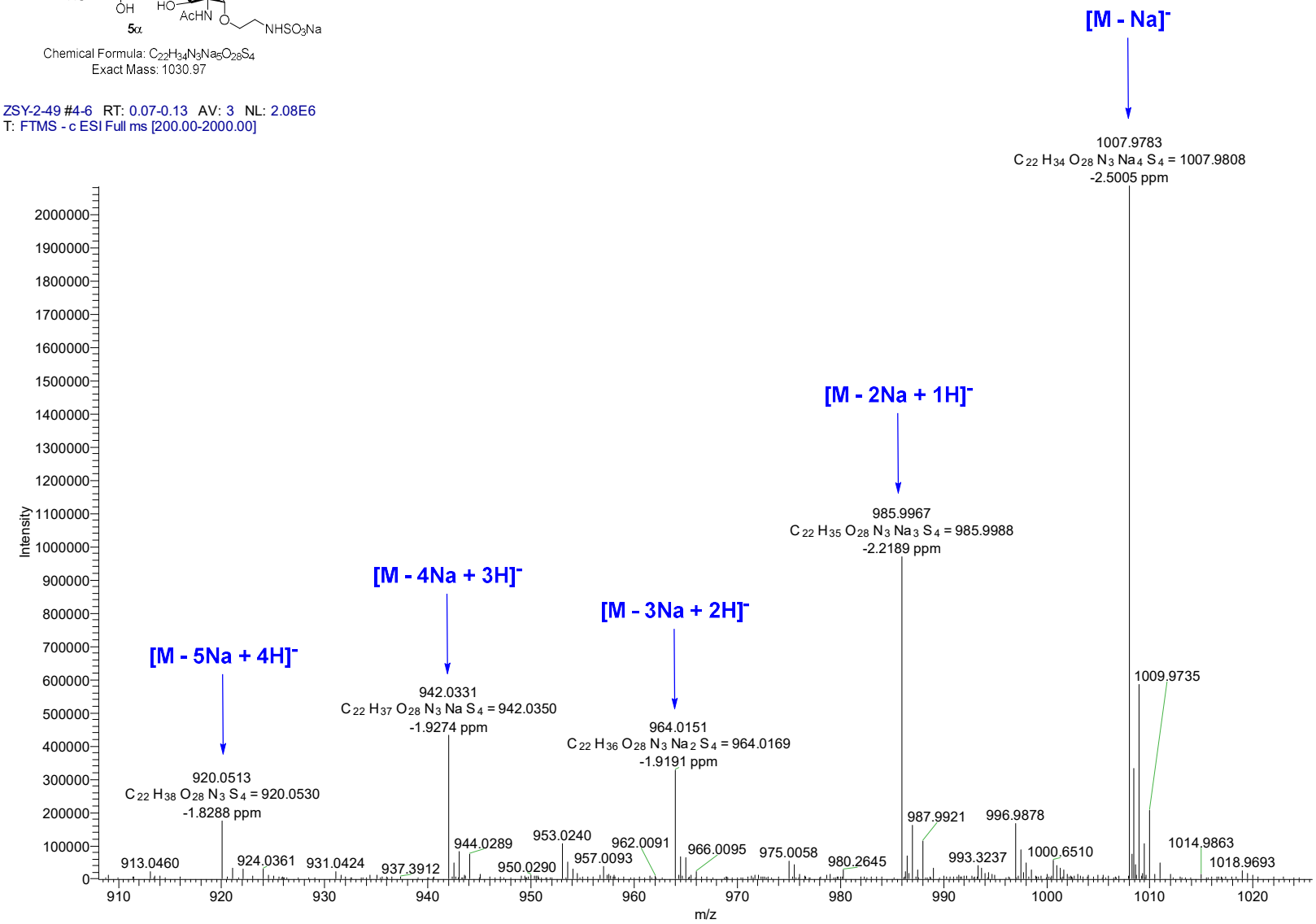
ZSY-2-48 #3-20 RT: 0.04-0.55 AV: 18 NL: 1.89E6
T: FTMS - c ESI Full ms [200.00-2000.00]

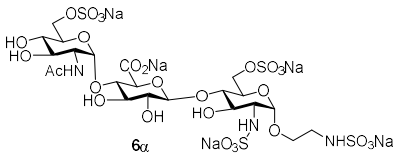




Chemical Formula: $C_{22}H_{34}N_3Na_5O_{26}S_4$
Exact Mass: 1030.97

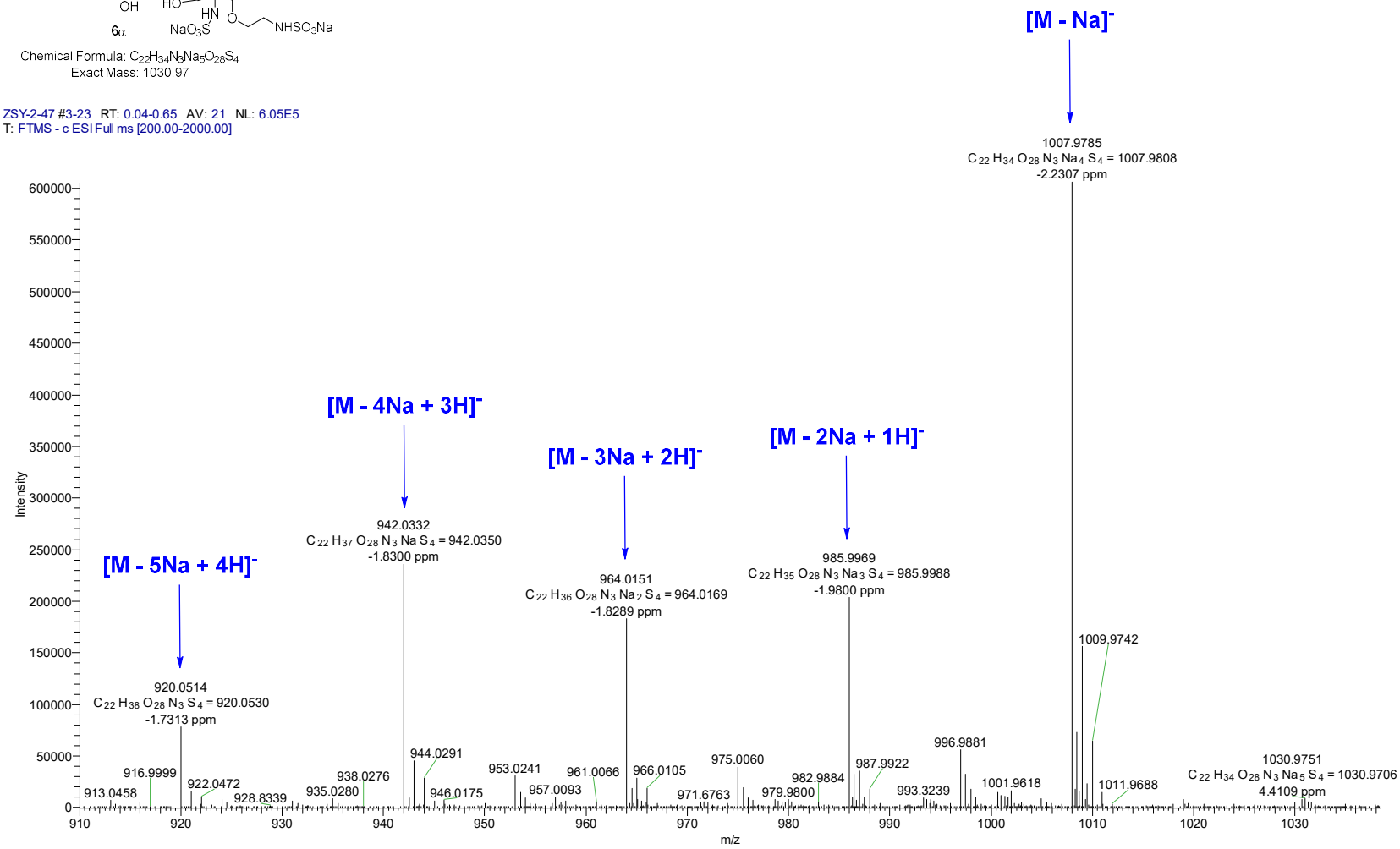
ZSY-2-49 #4-6 RT: 0.07-0.13 AV: 3 NL: 2.08E6
T: FTMS - c ESI Full ms [200.00-2000.00]

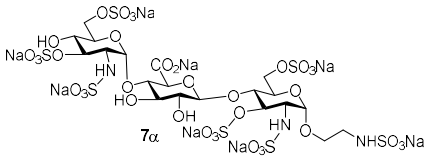




Chemical Formula: $C_{22}H_{34}N_3Na_5O_{28}S_4$
Exact Mass: 1030.97

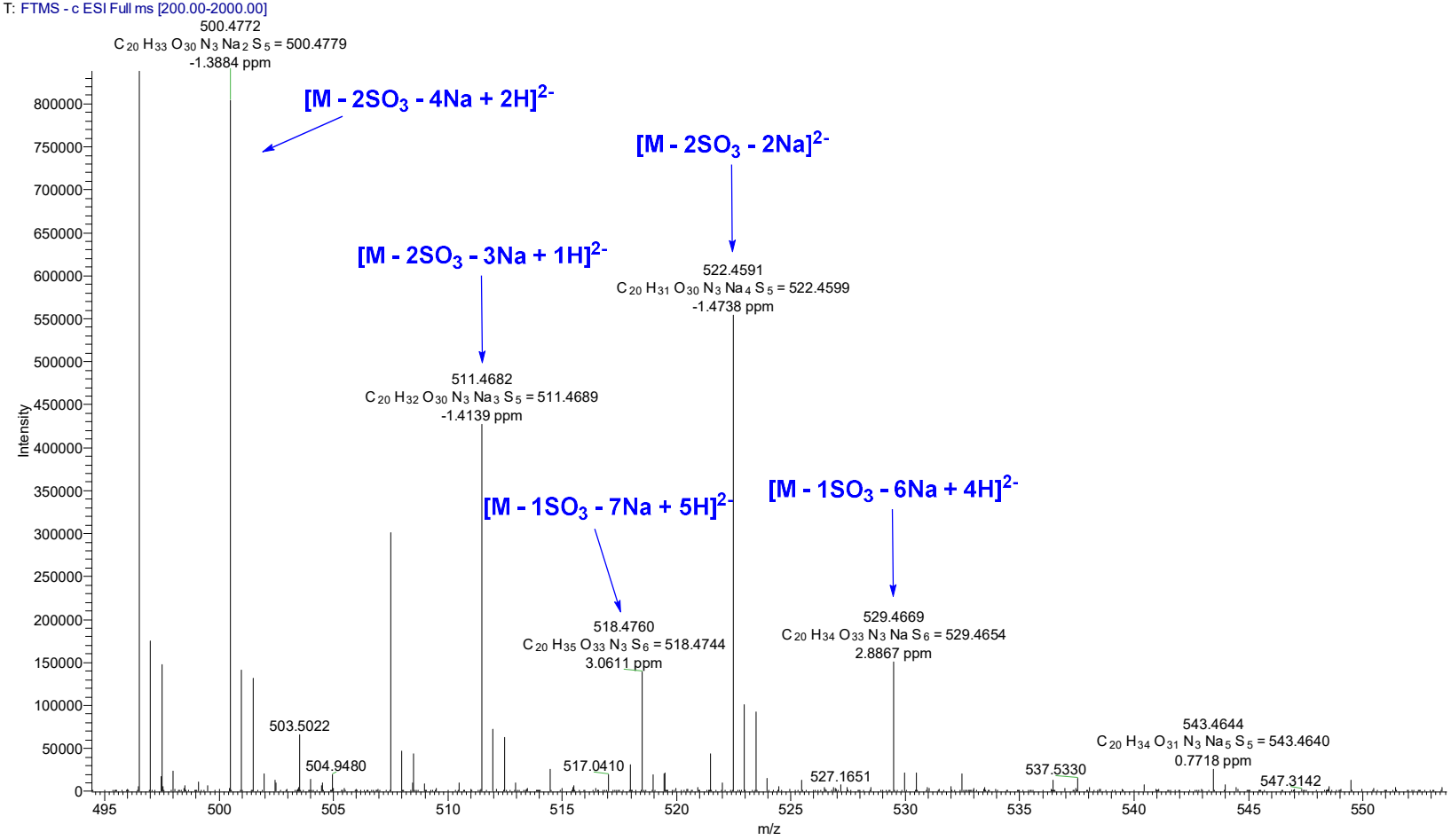
ZSY-2-47 #3-23 RT: 0.04-0.65 AV: 21 NL: 6.05E5
T: FTMS - c ESI Full ms [200.00-2000.00]

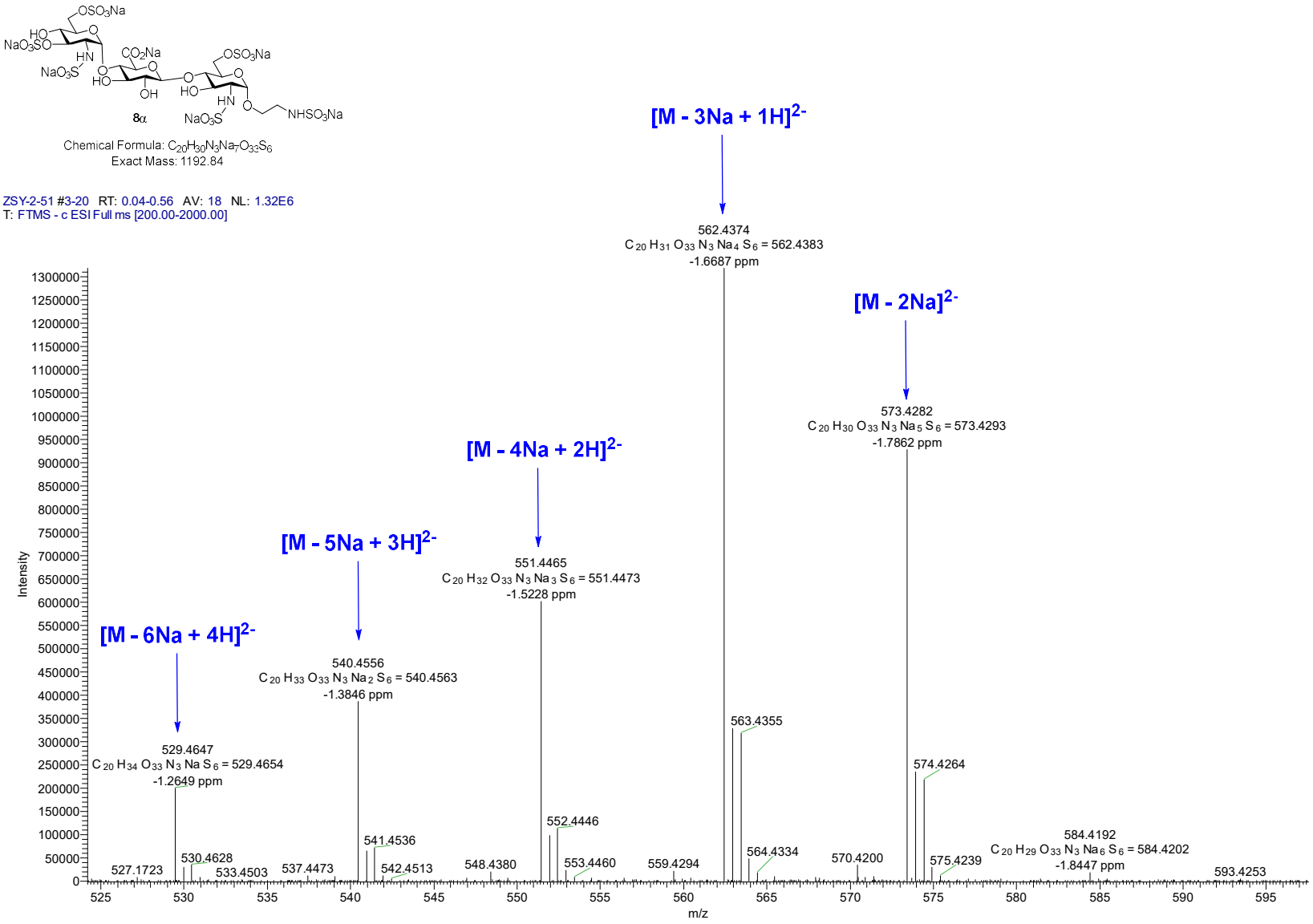


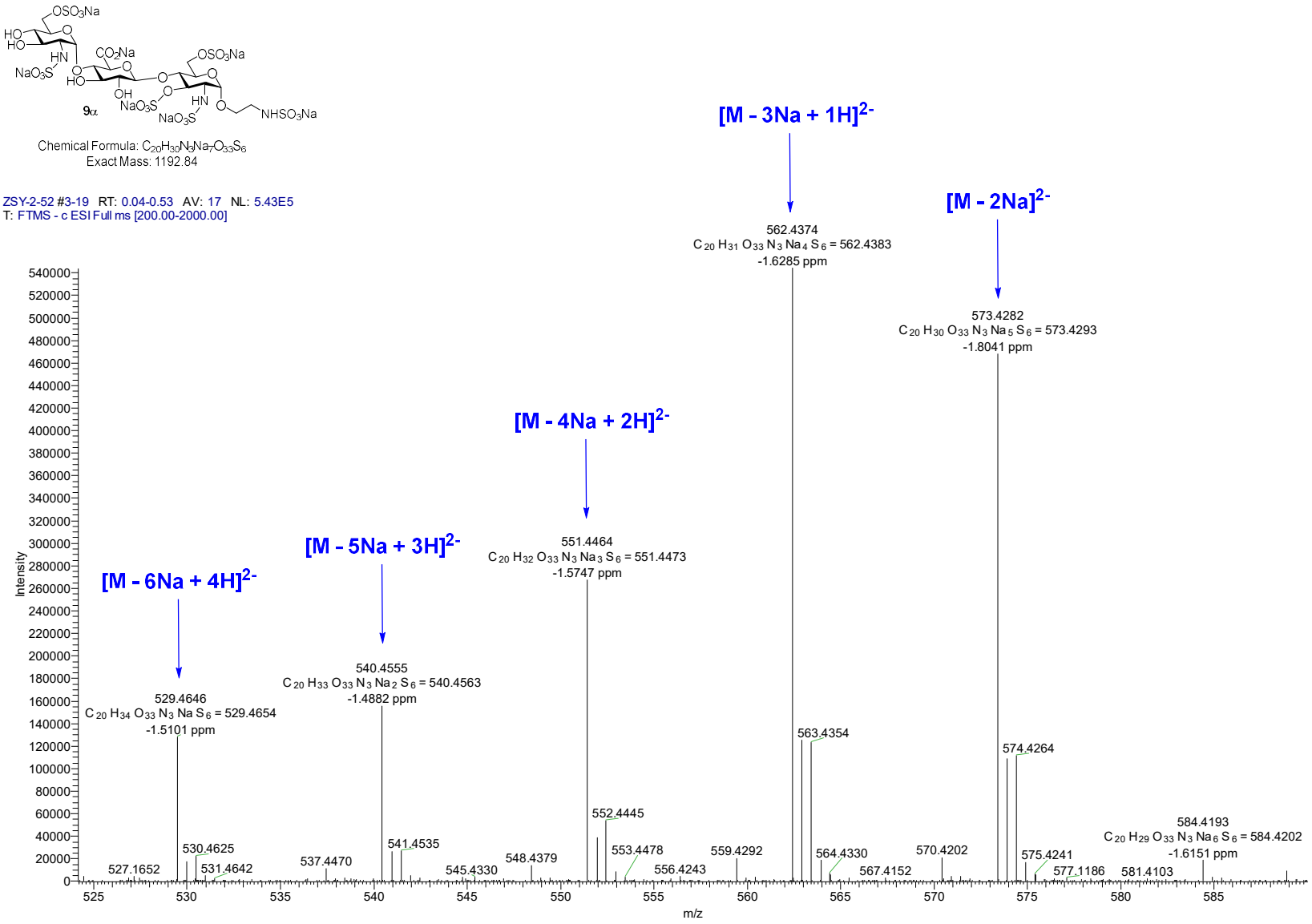


Chemical Formula: $C_{20}H_{23}N_3Na_8O_{36}S_7$
Exact Mass: 1294.78

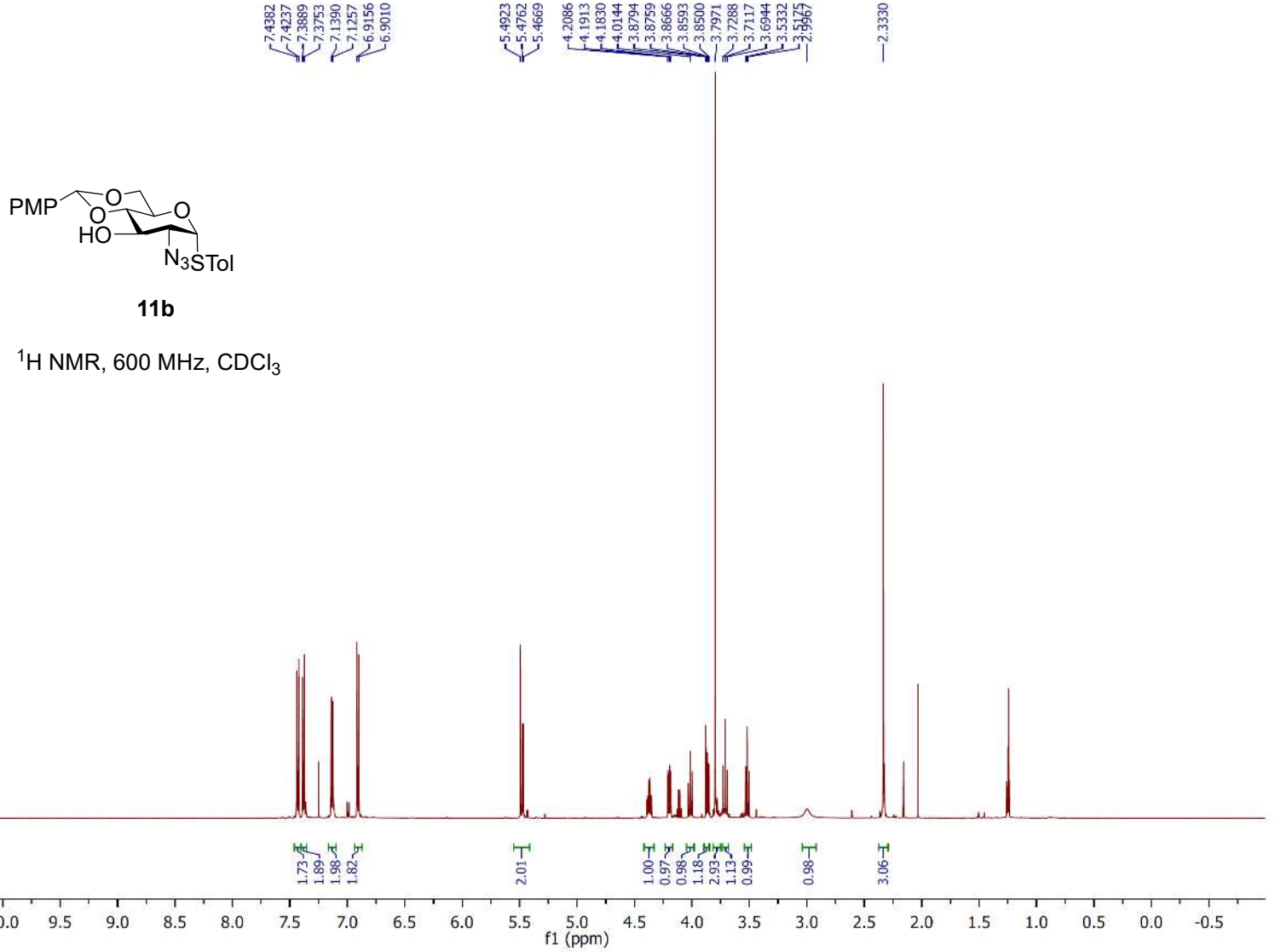
ZSY-2-50 #4-15 RT: 0.07-0.40 AV: 12 NL: 8.37E5
T: FTMS - c ESI Full ms [200.00-2000.00]

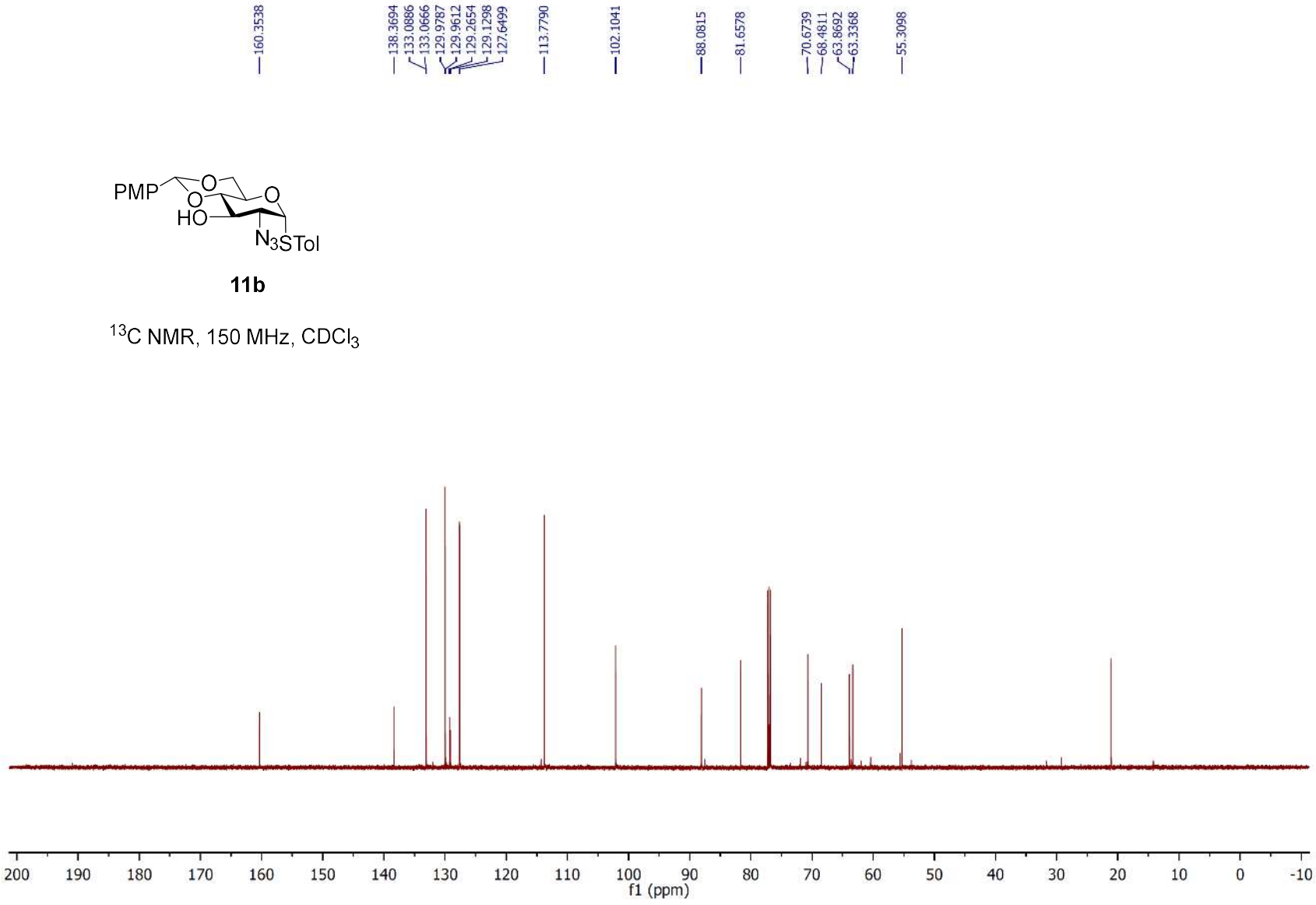



8α
Chemical Formula: C20H30N3Na7O33S6
Exact Mass: 1192.84
ZSY-2-51 #3-20 RT: 0.04-0.56 AV: 18 NL: 1.32E6
T: FTMS - c ESI Full ms [200.00-2000.00]
[M - 3Na + 1H]2-
562.4374
C20 H31 O33 N3 Na4 S6 = 562.4383
-1.6687 ppm
[M - 2Na]2-
573.4282
C20 H30 O33 N3 Na5 S6 = 573.4293
-1.7862 ppm
[M - 4Na + 2H]2-
551.4465
C20 H32 O33 N3 Na3 S6 = 551.4473
-1.5228 ppm
[M - 5Na + 3H]2-
540.4556
C20 H33 O33 N3 Na2 S6 = 540.4563
-1.3846 ppm
[M - 6Na + 4H]2-
529.4647
C20 H34 O33 N3 Na S6 = 529.4654
-1.2649 ppm
563.4355
574.4264
552.4446
541.4536
530.4628
527.1723
533.4503
537.4473
542.4513
548.4380
553.4460
559.4294
564.4334
570.4200
575.4239
584.4192
C20 H29 O33 N3 Na6 S6 = 584.4202
-1.8447 ppm
593.4253
Intensity
m/z


9α
Chemical Formula: C20H30N3Na7O33S6
Exact Mass: 1192.84
ZSY-2-52 #3-19 RT: 0.04-0.53 AV: 17 NL: 5.43E5
T: FTMS - c ESI Full ms [200.00-2000.00]
[M - 6Na + 4H]2-
[M - 5Na + 3H]2-
[M - 4Na + 2H]2-
[M - 3Na + 1H]2-
[M - 2Na]2-
529.4646
C20 H34 O33 N3 Na S6 = 529.4654
-1.5101 ppm
540.4555
C20 H33 O33 N3 Na2 S6 = 540.4563
-1.4882 ppm
551.4464
C20 H32 O33 N3 Na3 S6 = 551.4473
-1.5747 ppm
562.4374
C20 H31 O33 N3 Na4 S6 = 562.4383
-1.6285 ppm
573.4282
C20 H30 O33 N3 Na5 S6 = 573.4293
-1.8041 ppm
584.4193
C20 H29 O33 N3 Na6 S6 = 584.4202
-1.6151 ppm
Intensity
m/z

12. NMR Data


PMP
O
O
O
HO
N3STol
11b
1H NMR, 600 MHz, CDCl3
7.4382
7.4237
7.3889
7.3753
7.1390
7.1257
6.9156
6.9010
5.4923
5.4762
5.4669
4.2086
4.1913
4.1830
4.0144
3.8794
3.8759
3.8666
3.8593
3.8500
3.7971
3.7288
3.7117
3.6944
3.5332
2.3330
1.73
1.89
1.98
1.82
2.01
1.00
0.97
0.98
1.18
2.93
1.13
0.99
0.98
3.06
f1 (ppm)

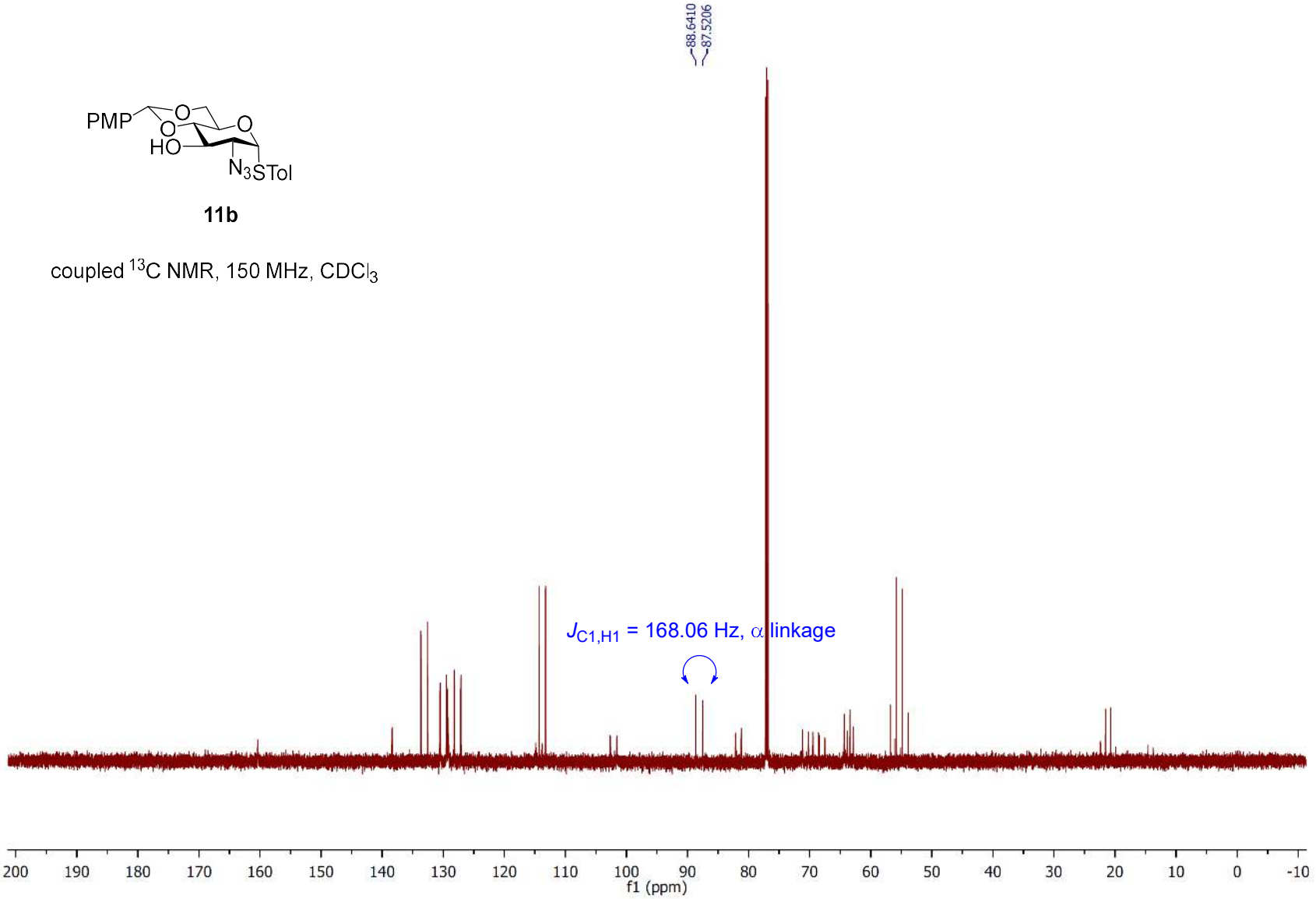


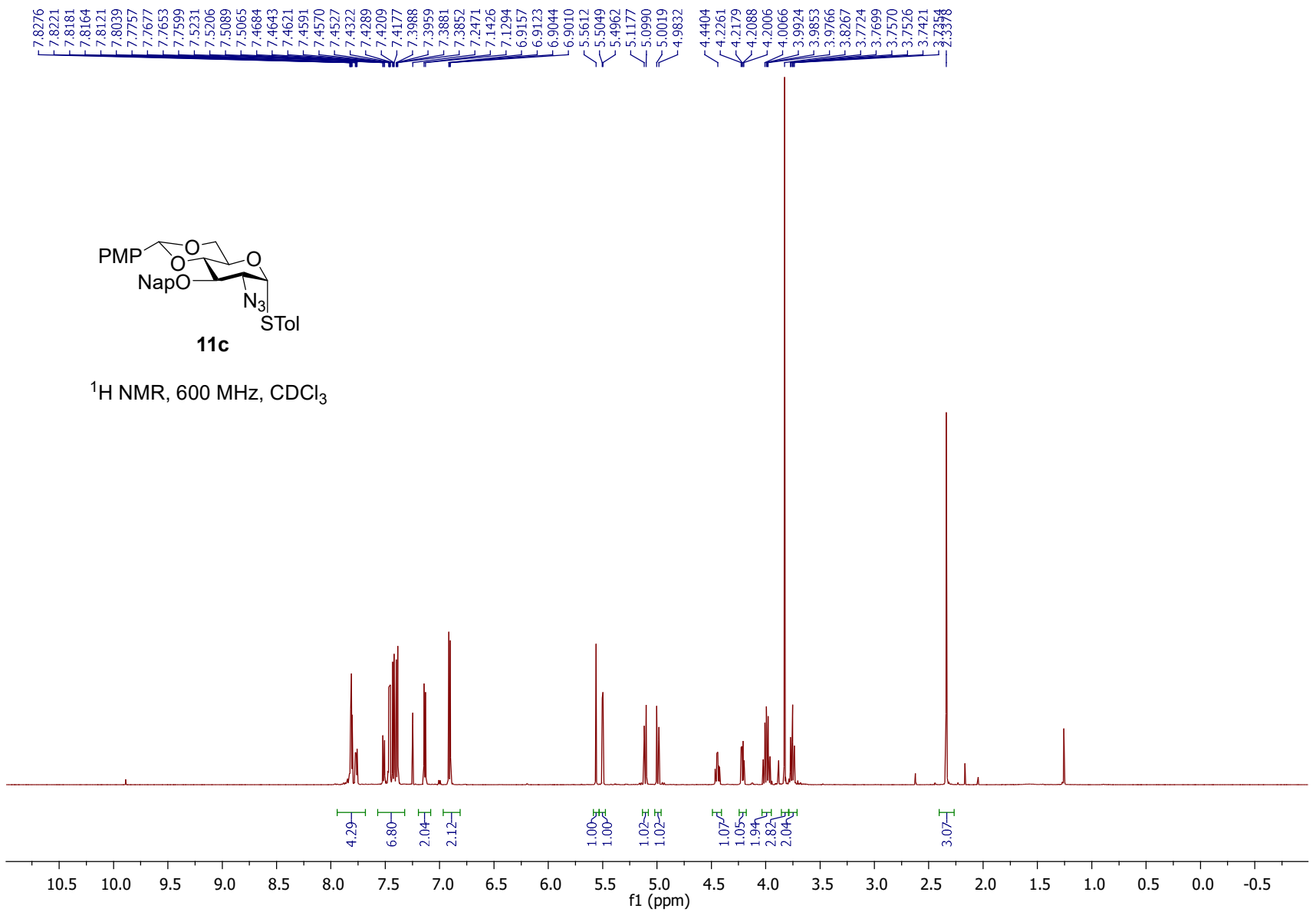
11b

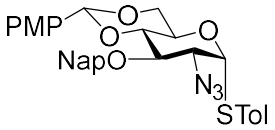
^{13}C NMR, 150 MHz, $CDCl_3$

11b

coupled ^{13}C NMR, 150 MHz, $CDCl_3$

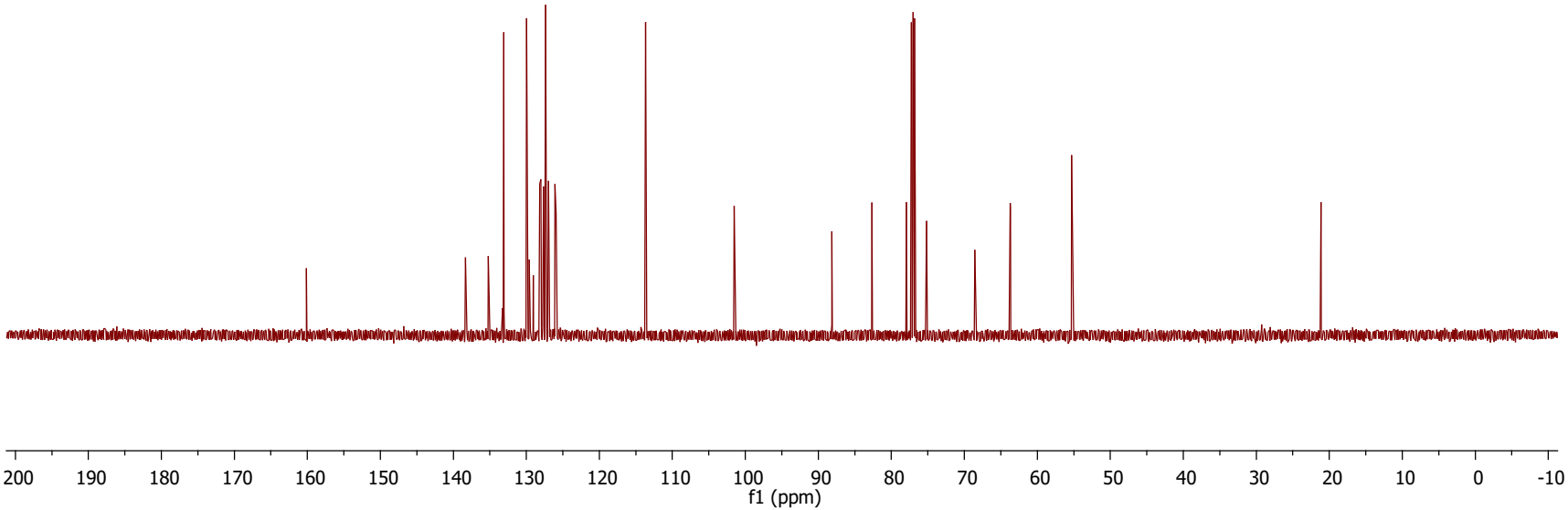


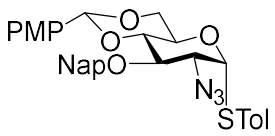

7.8276
7.8221
7.8181
7.8164
7.8121
7.8039
7.7757
7.7677
7.7653
7.7599
7.5231
7.5206
7.5089
7.5065
7.4684
7.4643
7.4621
7.4591
7.4570
7.4527
7.4322
7.4289
7.4209
7.4177
7.3988
7.3959
7.3881
7.3852
7.2471
7.1426
7.1294
6.9157
6.9123
6.9044
6.9010
5.5612
5.5049
5.4962
5.1177
5.0990
5.0019
4.9832
4.4404
4.2261
4.2179
4.2088
4.2006
4.0066
3.9924
3.9853
3.9766
3.8267
3.7724
3.7699
3.7570
3.7526
3.7421
3.7354
2.3378
PMP
O
O
O
NapO
N3
STol
11c
1H NMR, 600 MHz, CDCl3
4.29
6.80
2.04
2.12
1.00
1.00
1.02
1.02
1.07
1.05
1.94
2.82
2.04
3.07
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)



11c

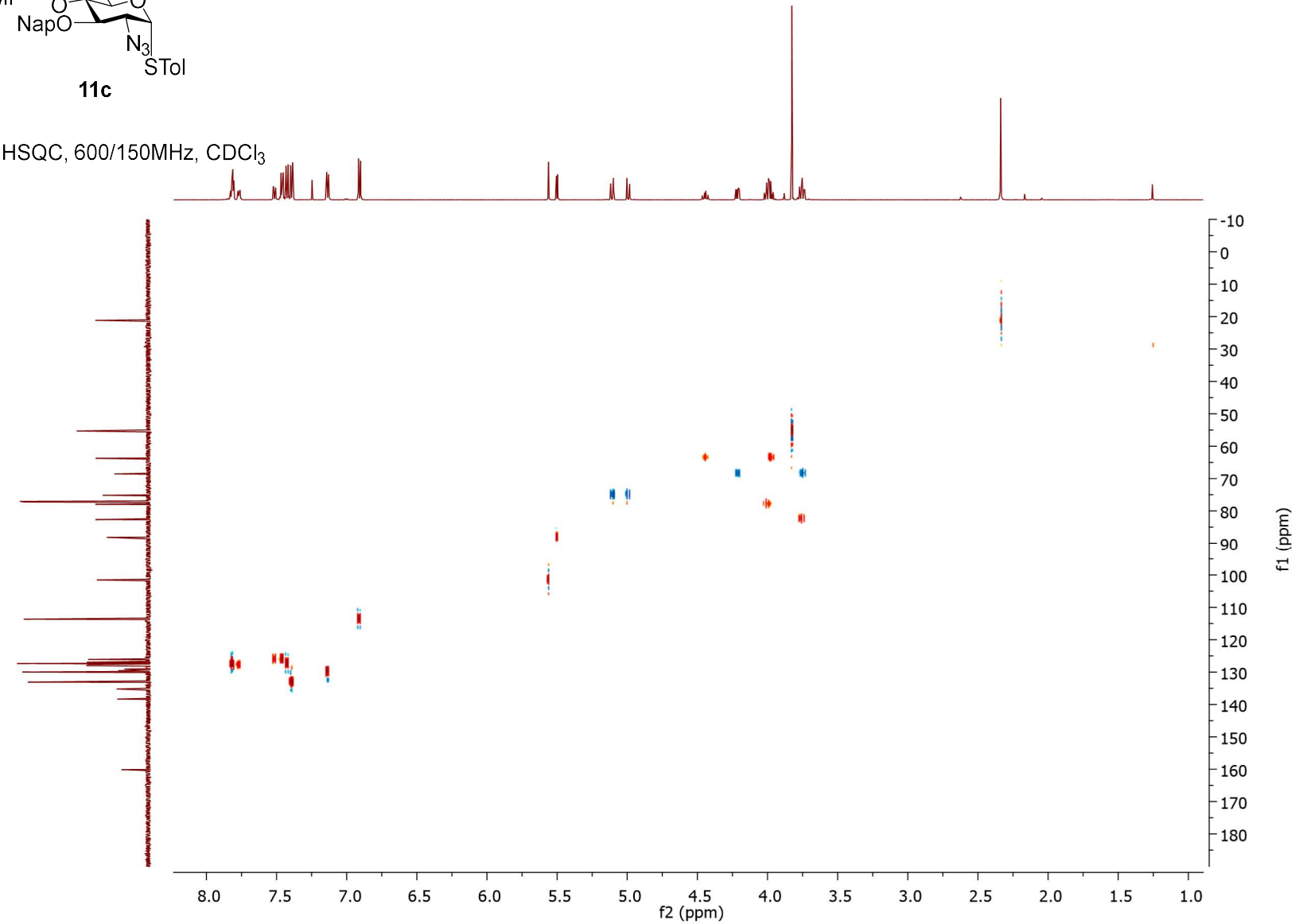
^{13}C NMR, 150 MHz, $CDCl_3$

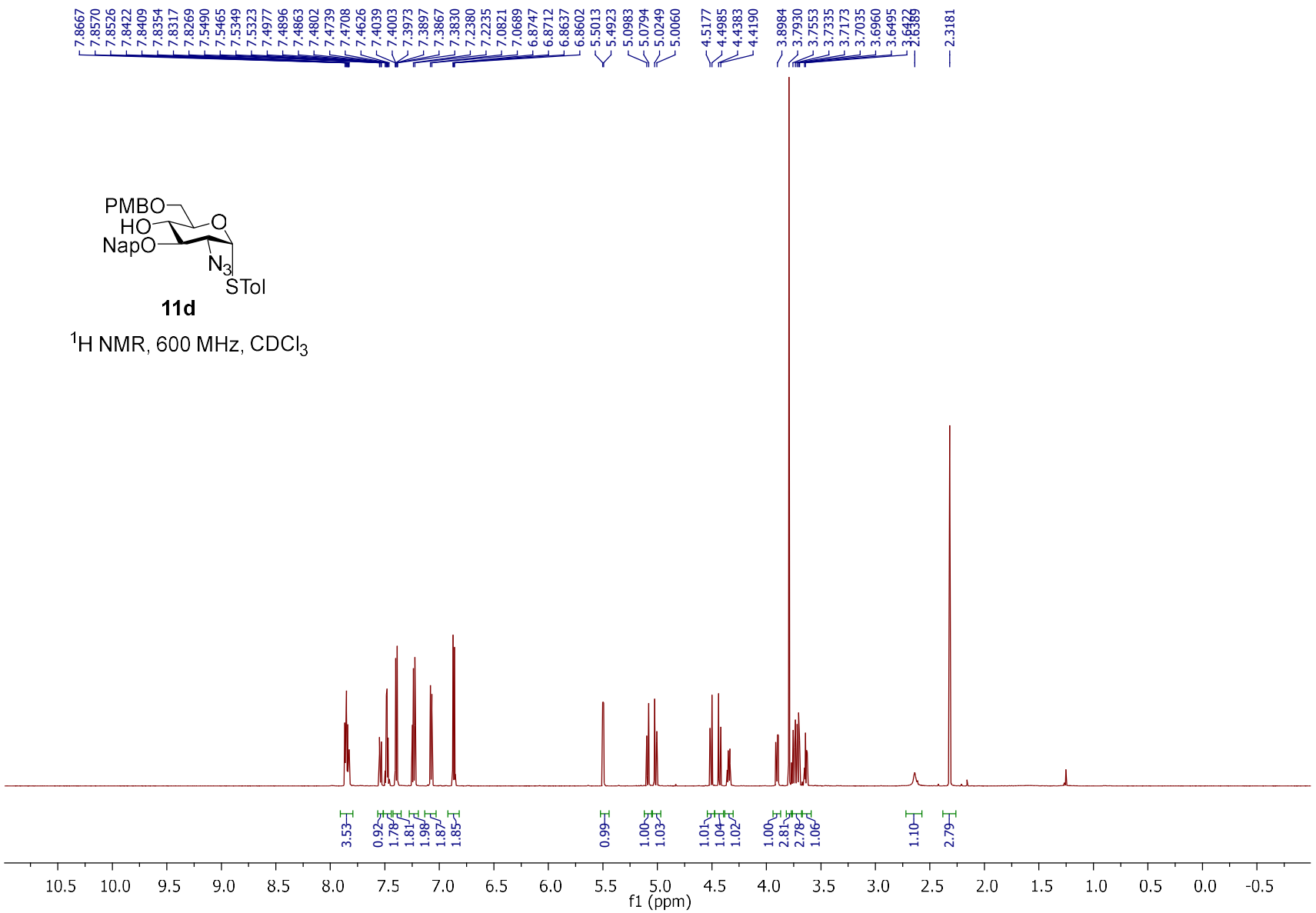


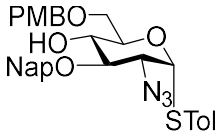


11c

^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

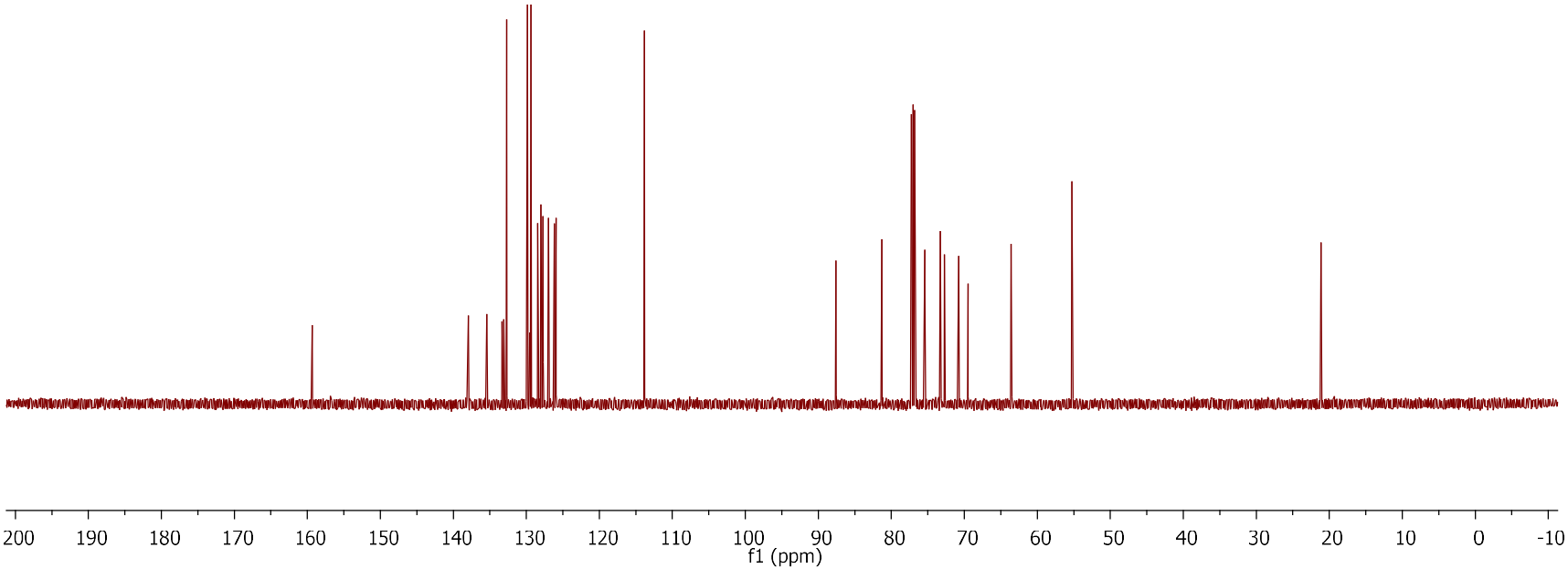


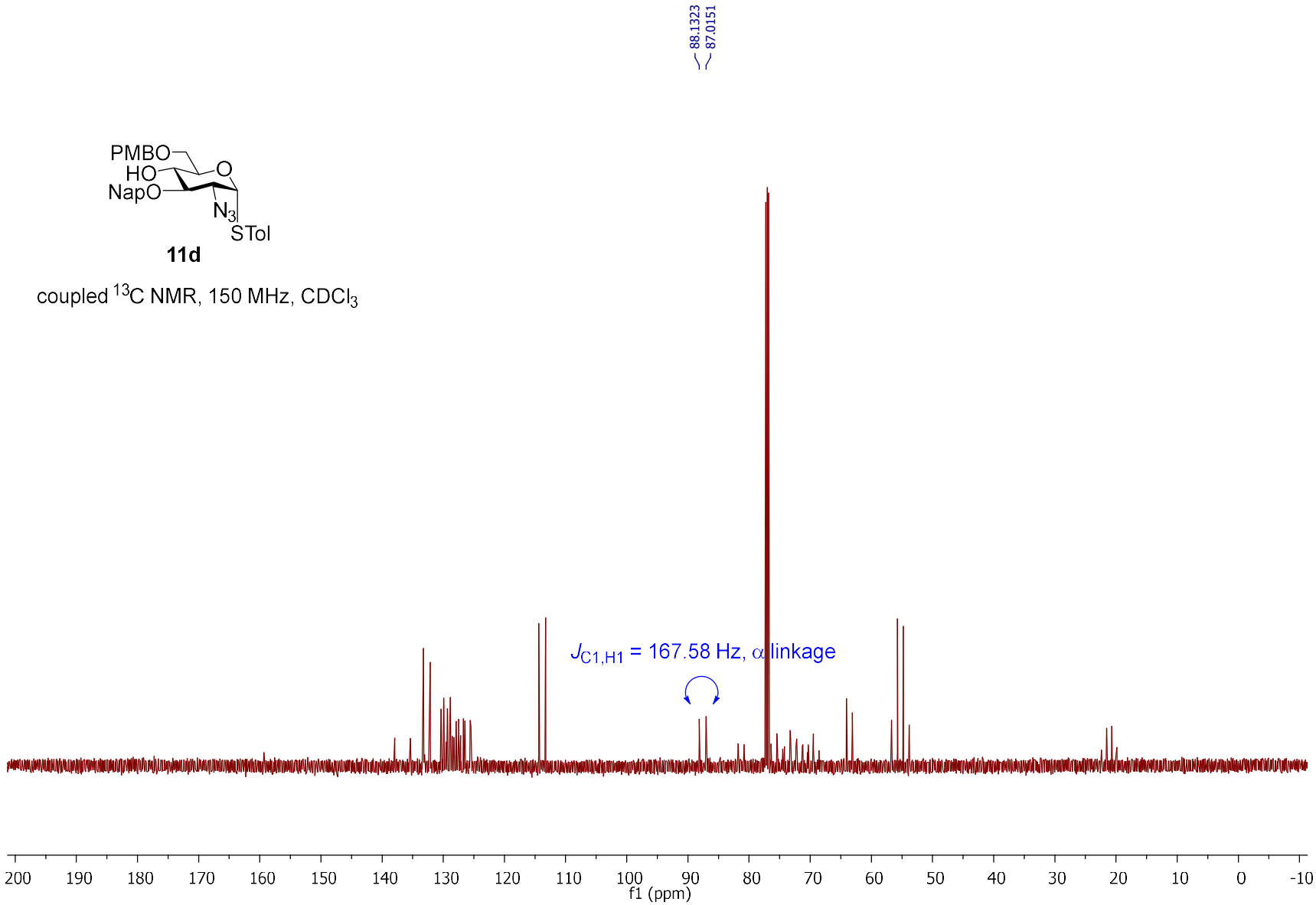

PMBO
HO
NapO
O
N3
STol
11d
1H NMR, 600 MHz, CDCl3
f1 (ppm)

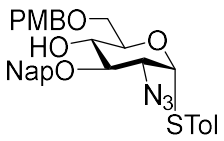


11d

^{13}C NMR, 150 MHz, $CDCl_3$

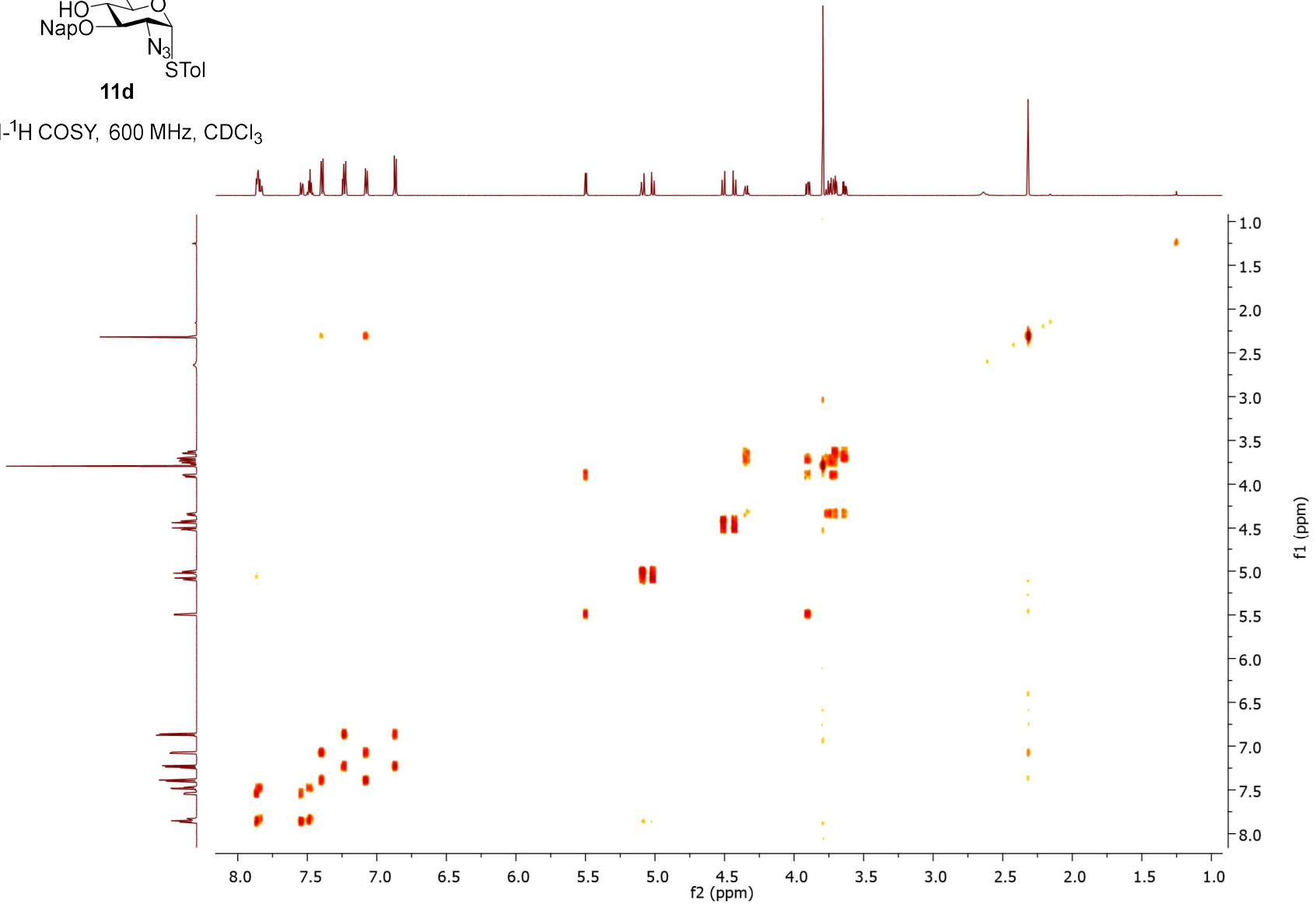


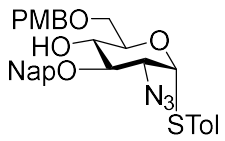

88.1323
87.0151
PMBO
HO
NapO
O
N3
STol
11d
coupled 13C NMR, 150 MHz, CDCl3
JC1,H1 = 167.58 Hz, α linkage
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)



11d

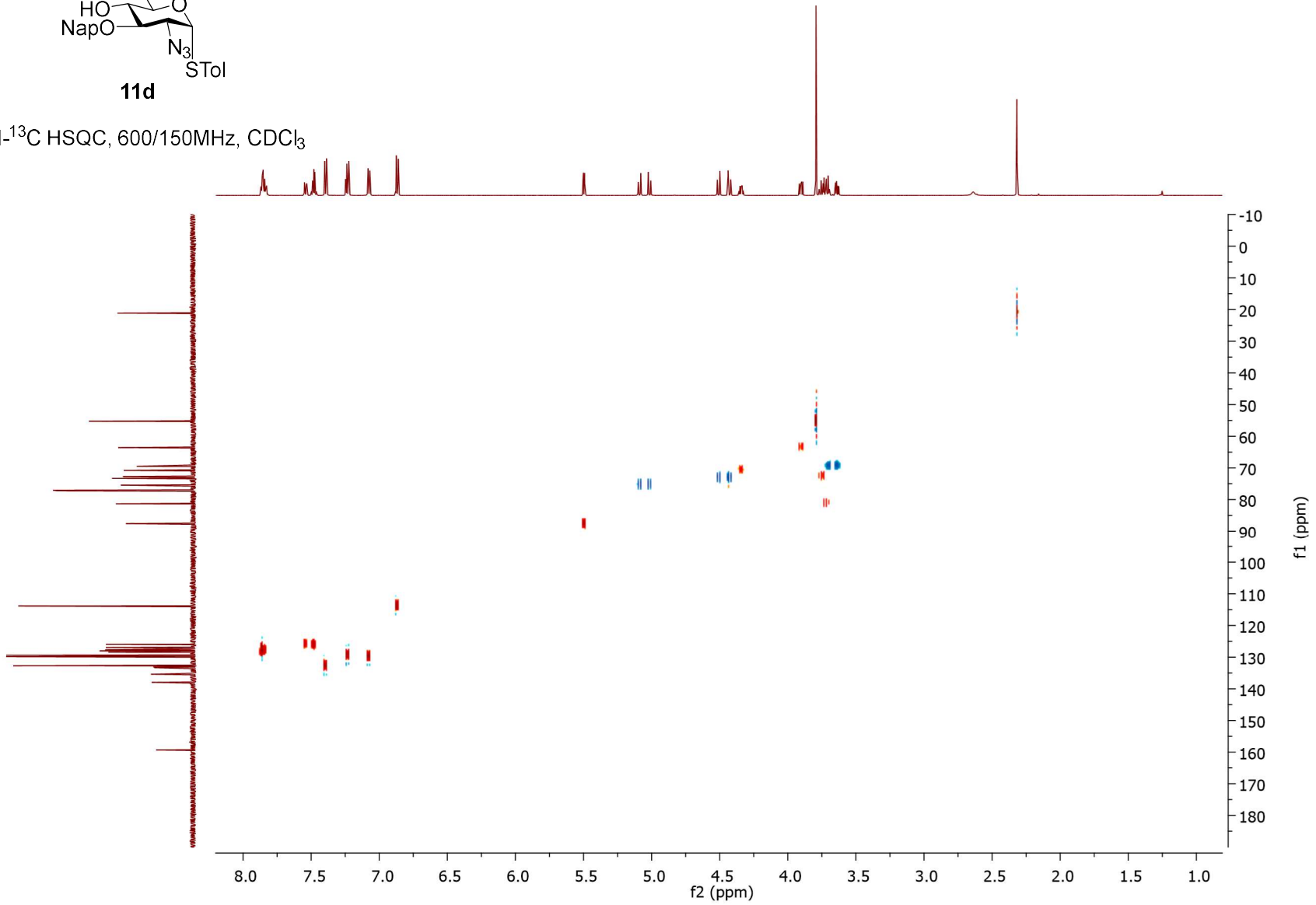
^{1}H-^{1}H COSY, 600 MHz, $CDCl_3$

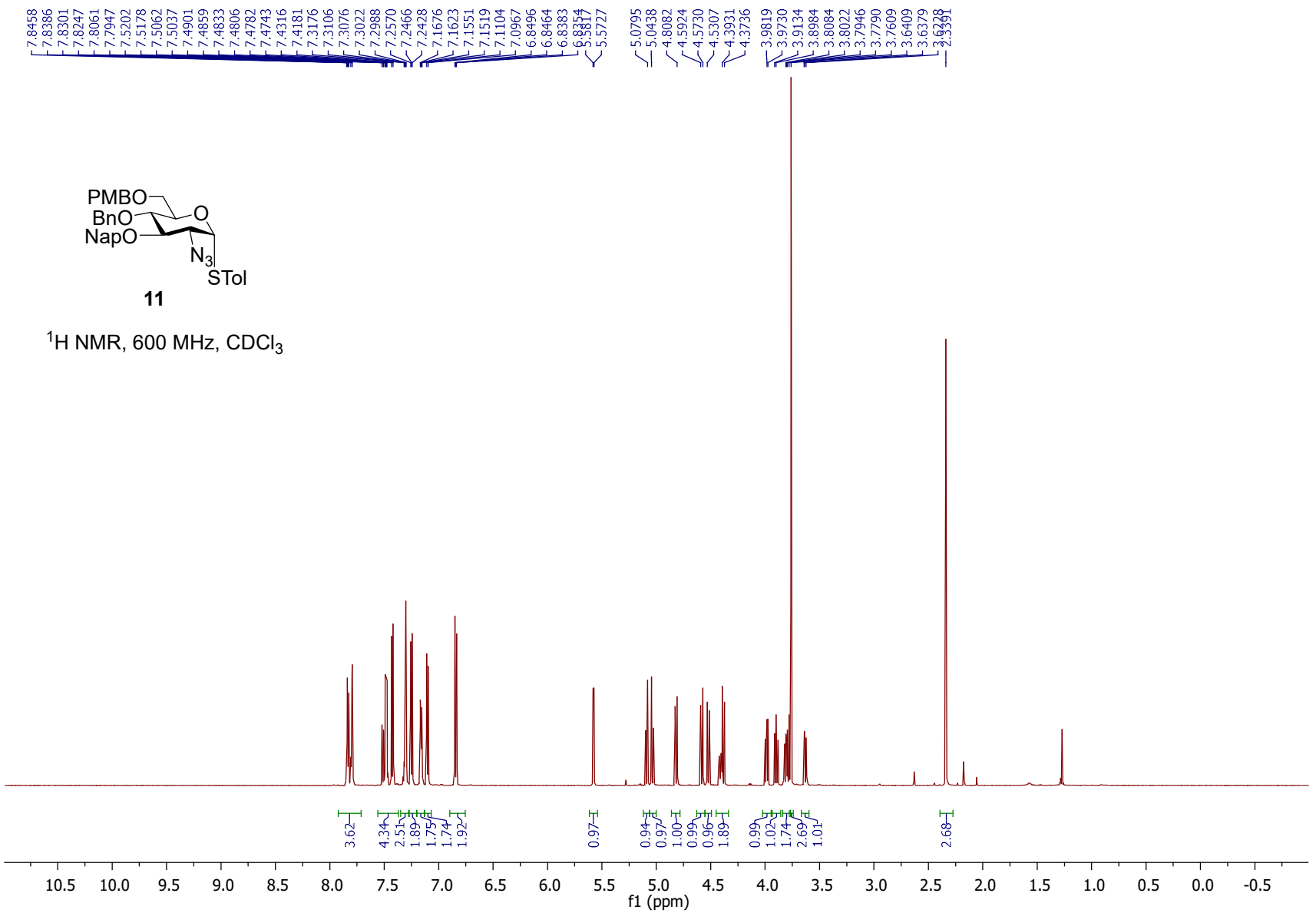


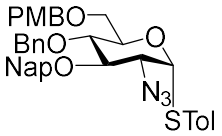


11d

^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

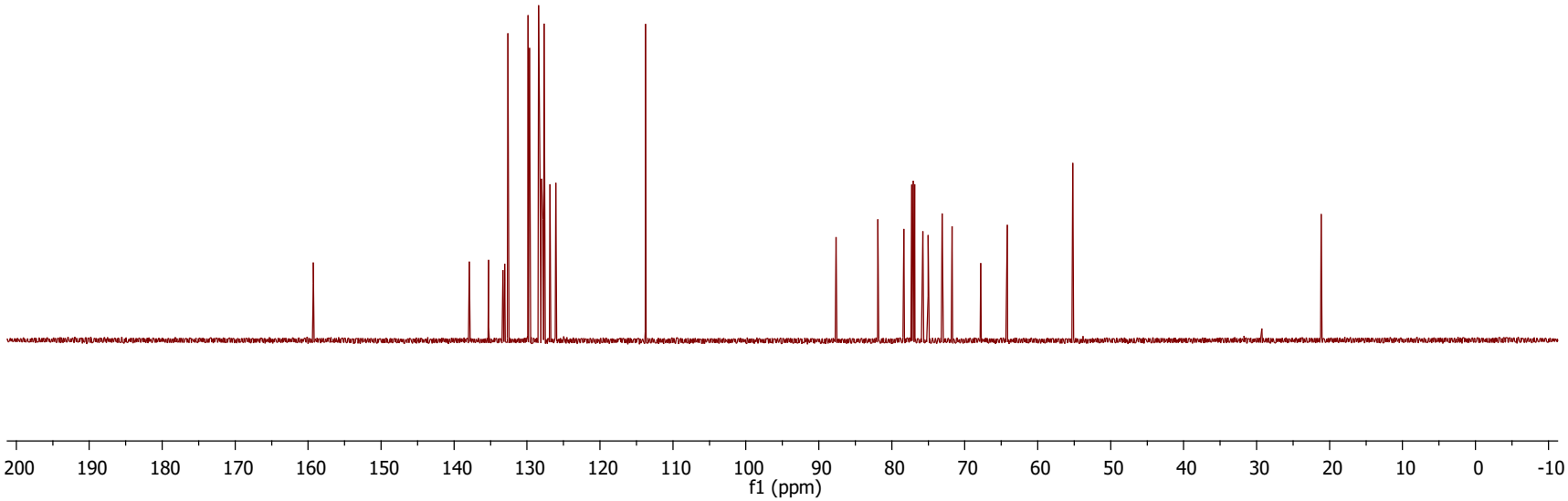


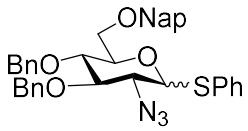

PMBO
BnO
NapO
O
N3
STol
11
1H NMR, 600 MHz, CDCl3
f1 (ppm)



11

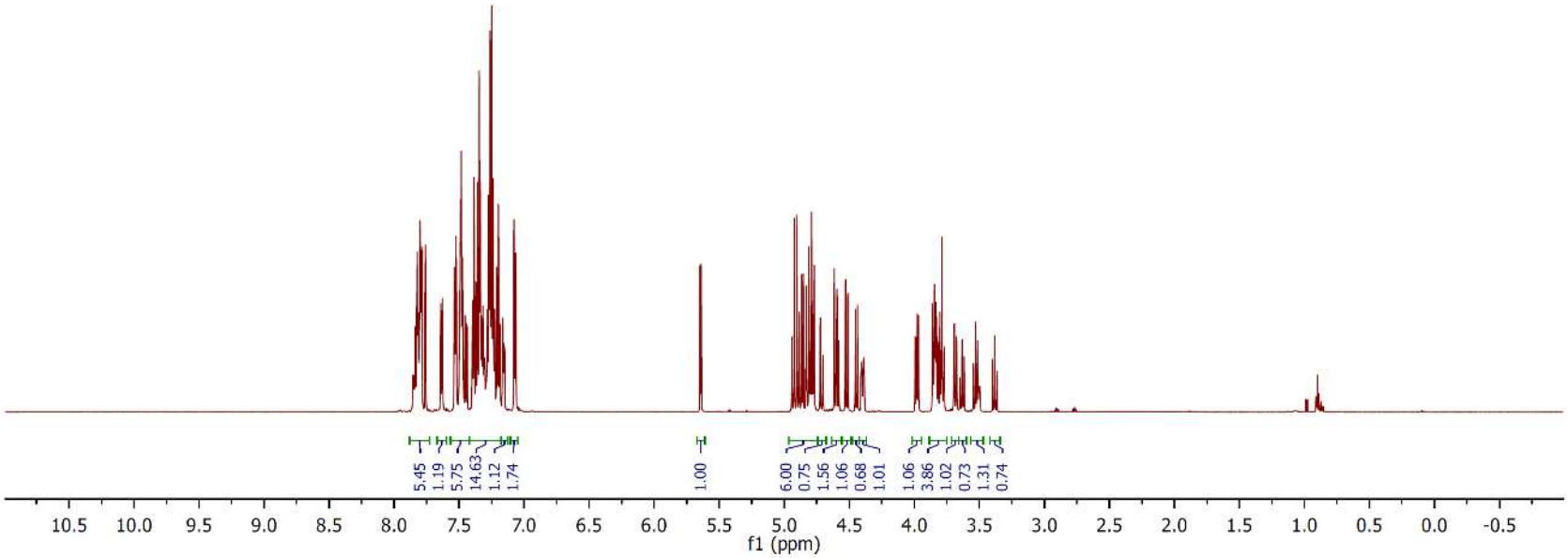
^{13}C NMR, 150 MHz, $CDCl_3$





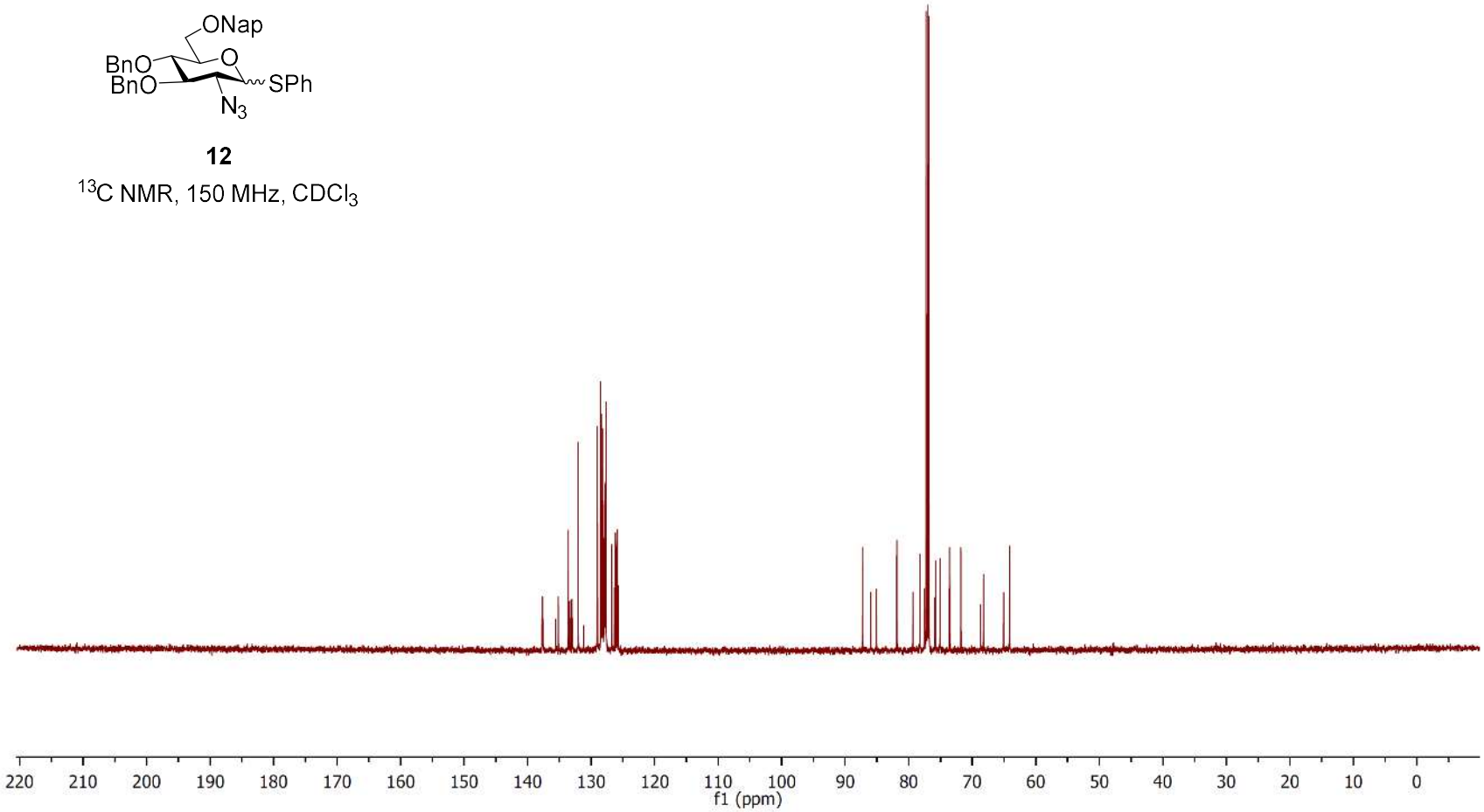
12

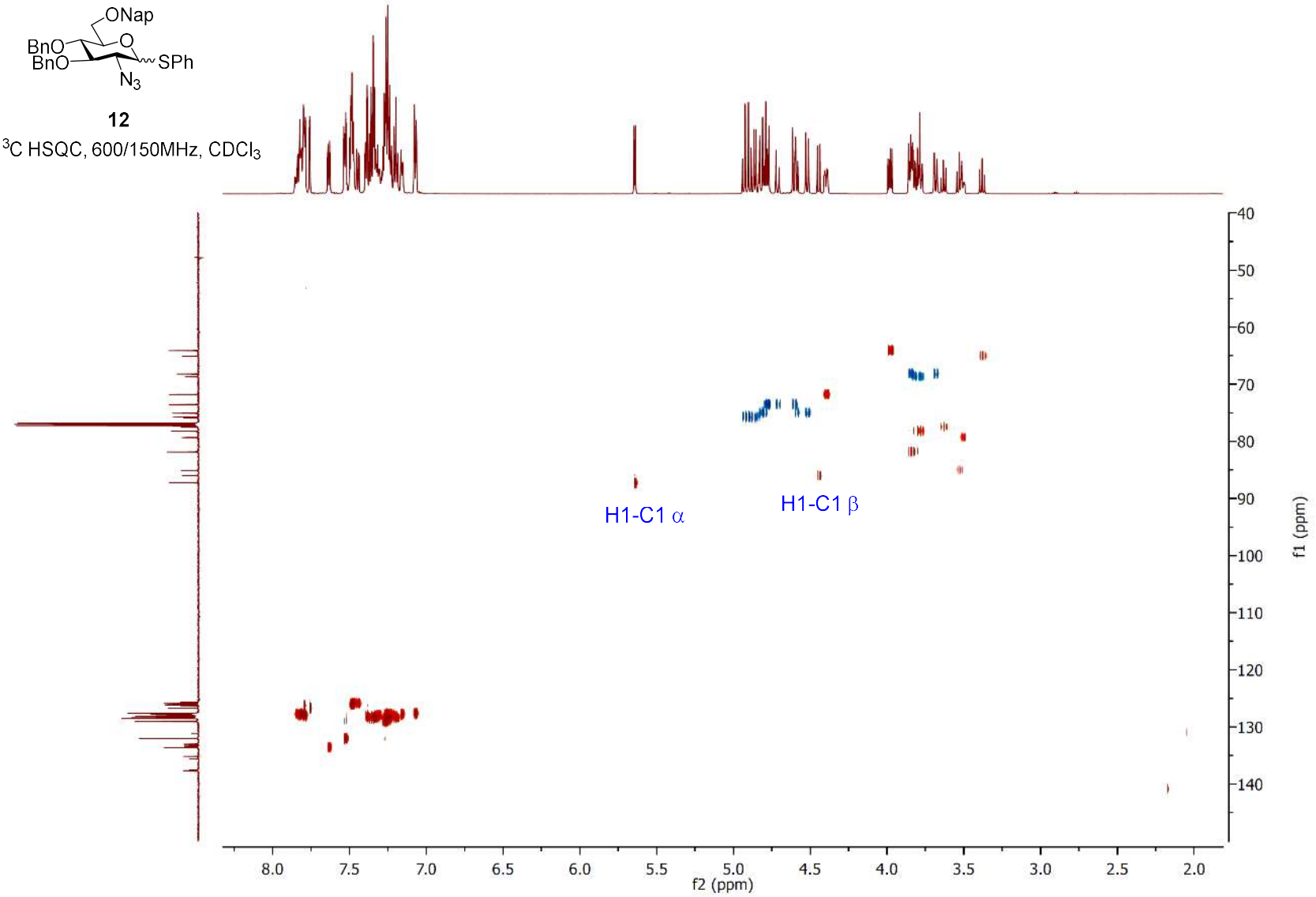
^{1}H NMR, 600 MHz, $CDCl_3$

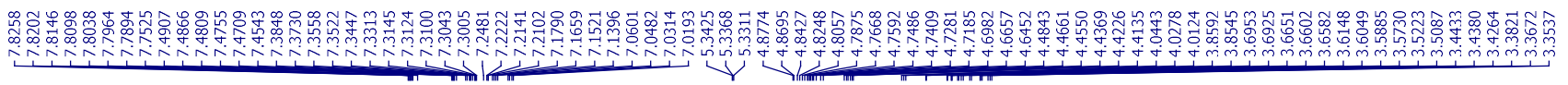


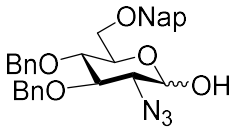
12

^{13}C NMR, 150 MHz, $CDCl_3$



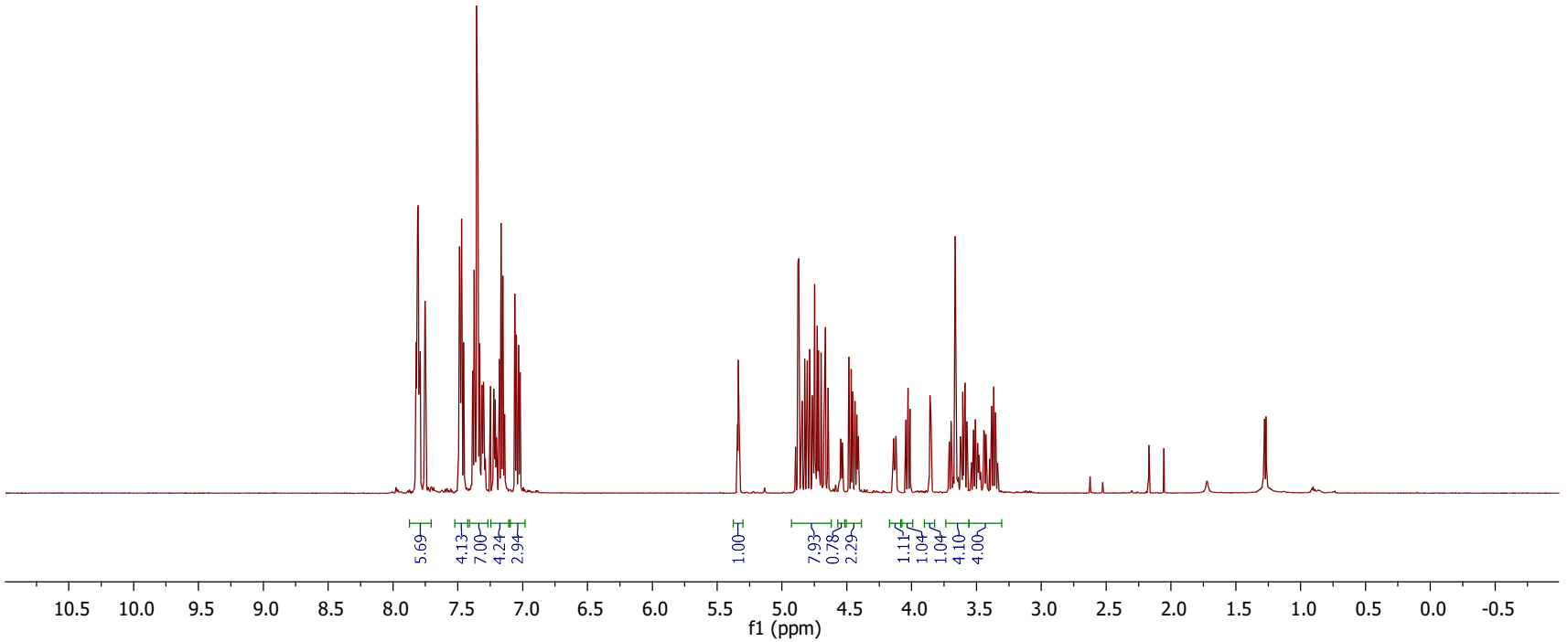

ONap
BnO
BnO
O
SPh
N3
12
1H-13C HSQC, 600/150MHz, CDCl3
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)

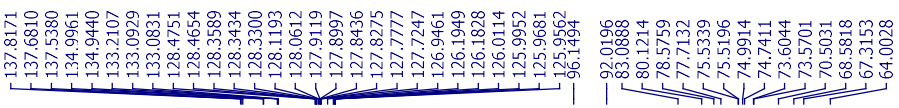


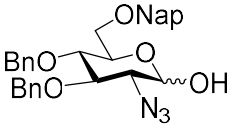


13a

^{1}H NMR, 600 MHz, $CDCl_3$

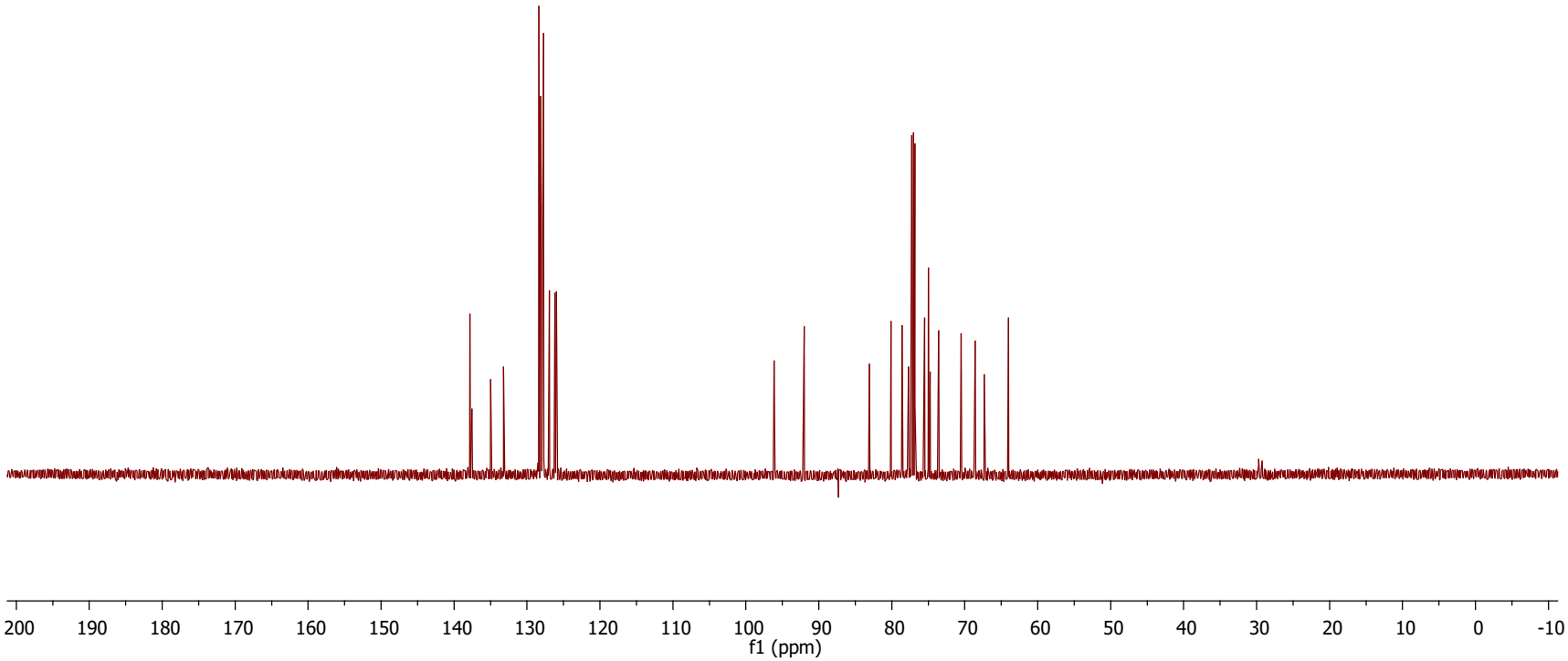


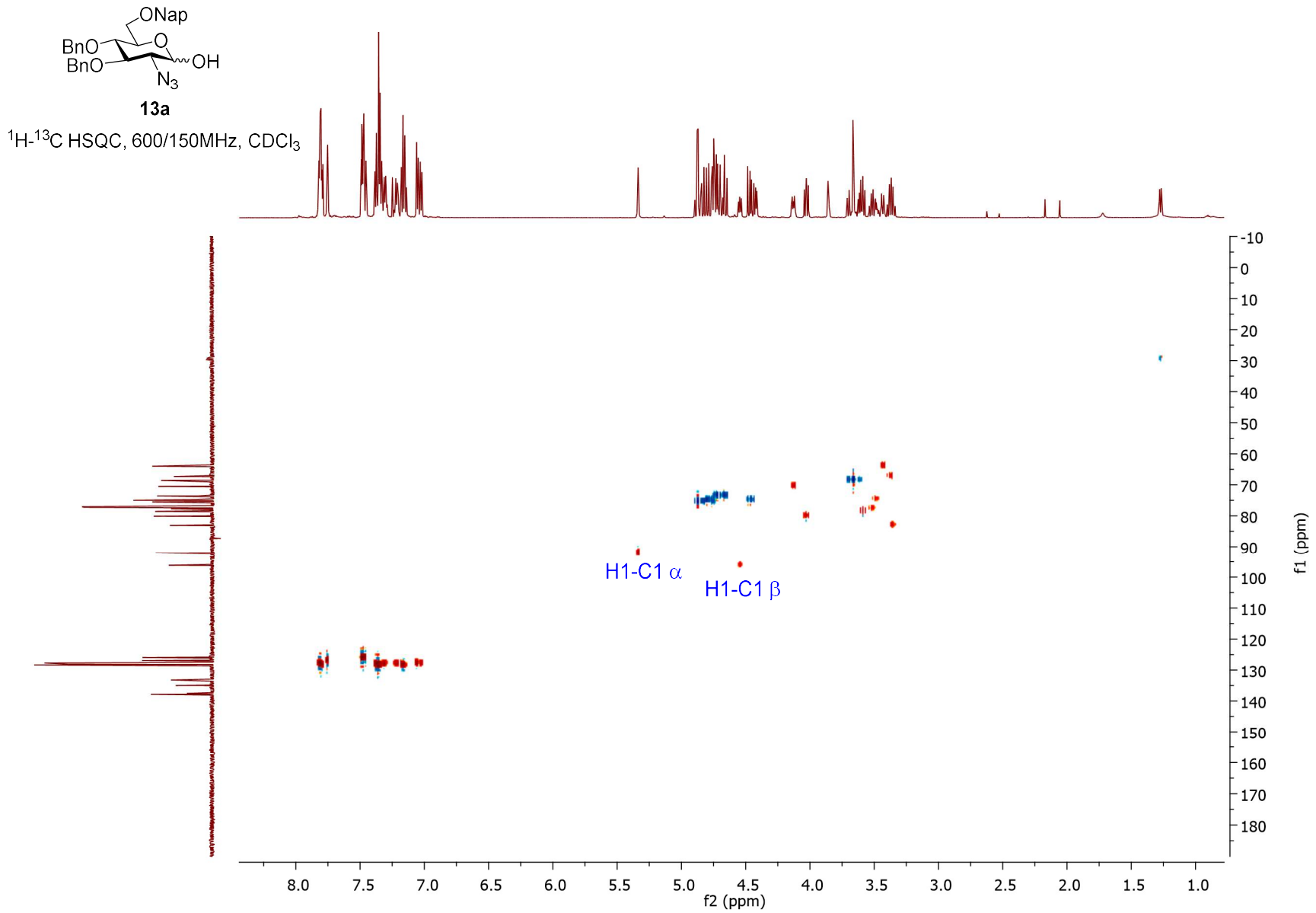


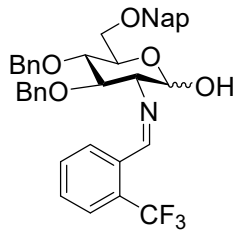


13a

^{13}C NMR, 150 MHz, $CDCl_3$

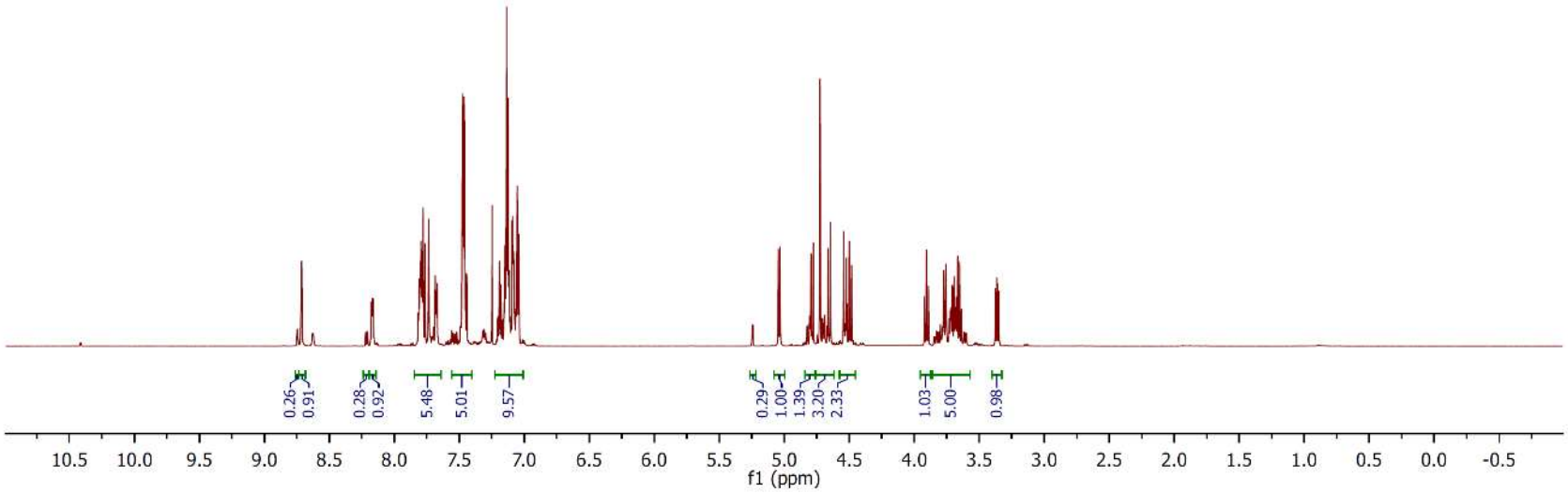


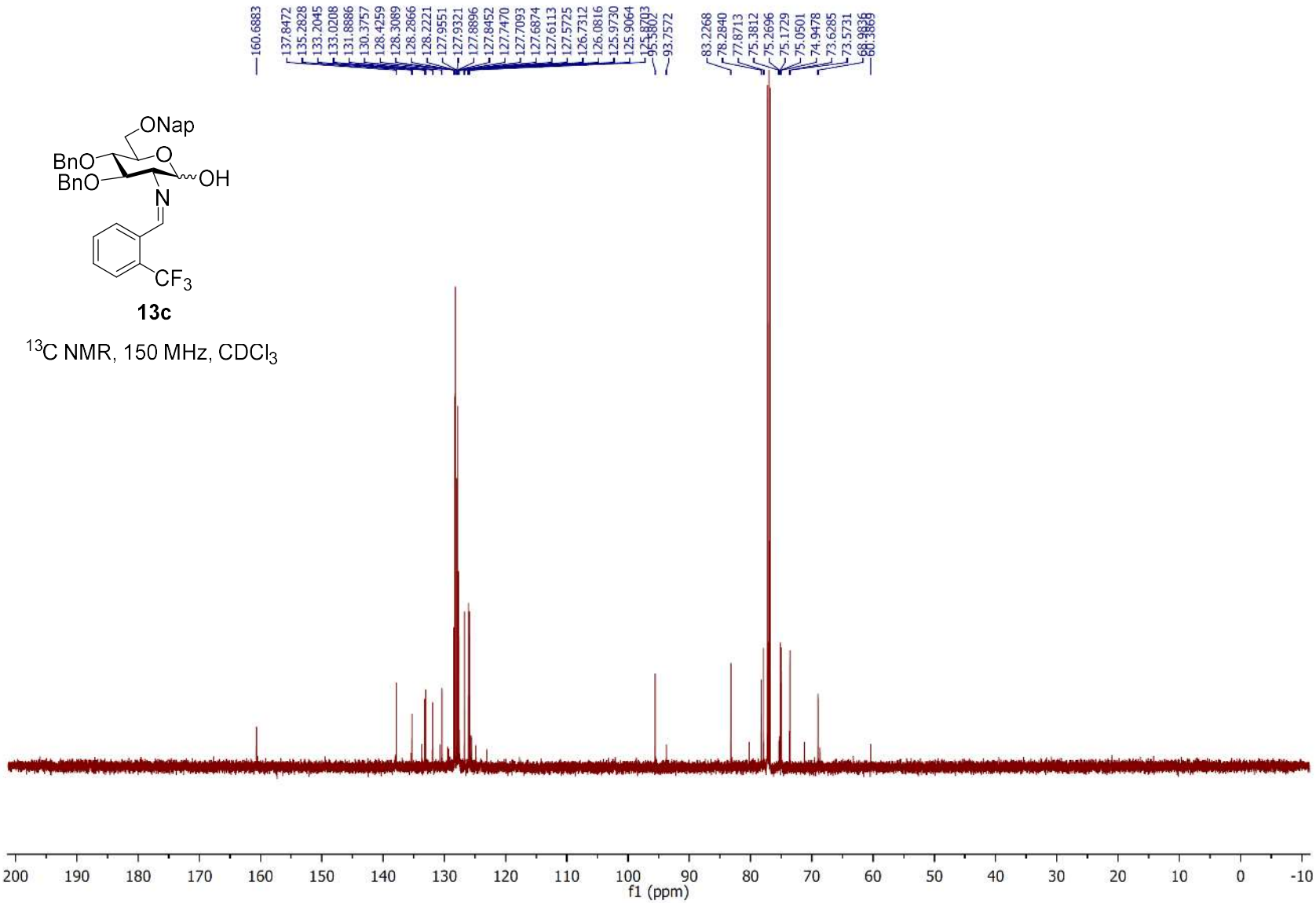

ONap
BnO
BnO
O
OH
N3
13a
1H-13C HSQC, 600/150MHz, CDCl3
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)

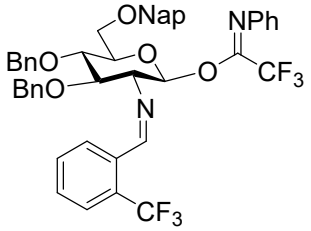


13c

^{1}H NMR, 600 MHz, $CDCl_3$

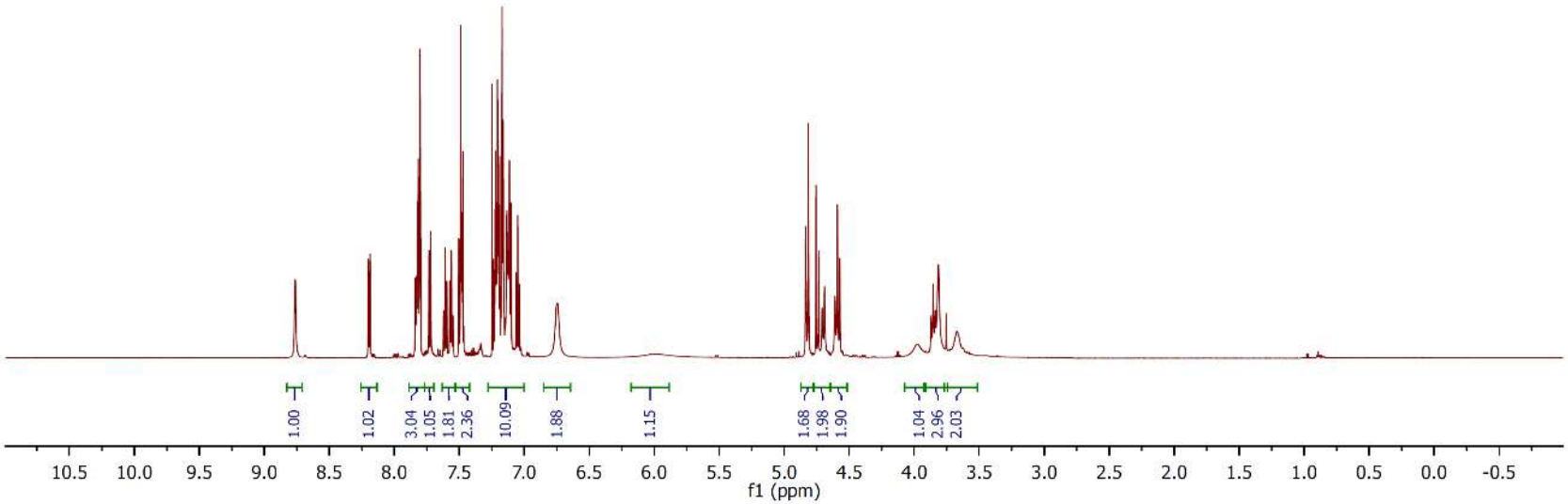


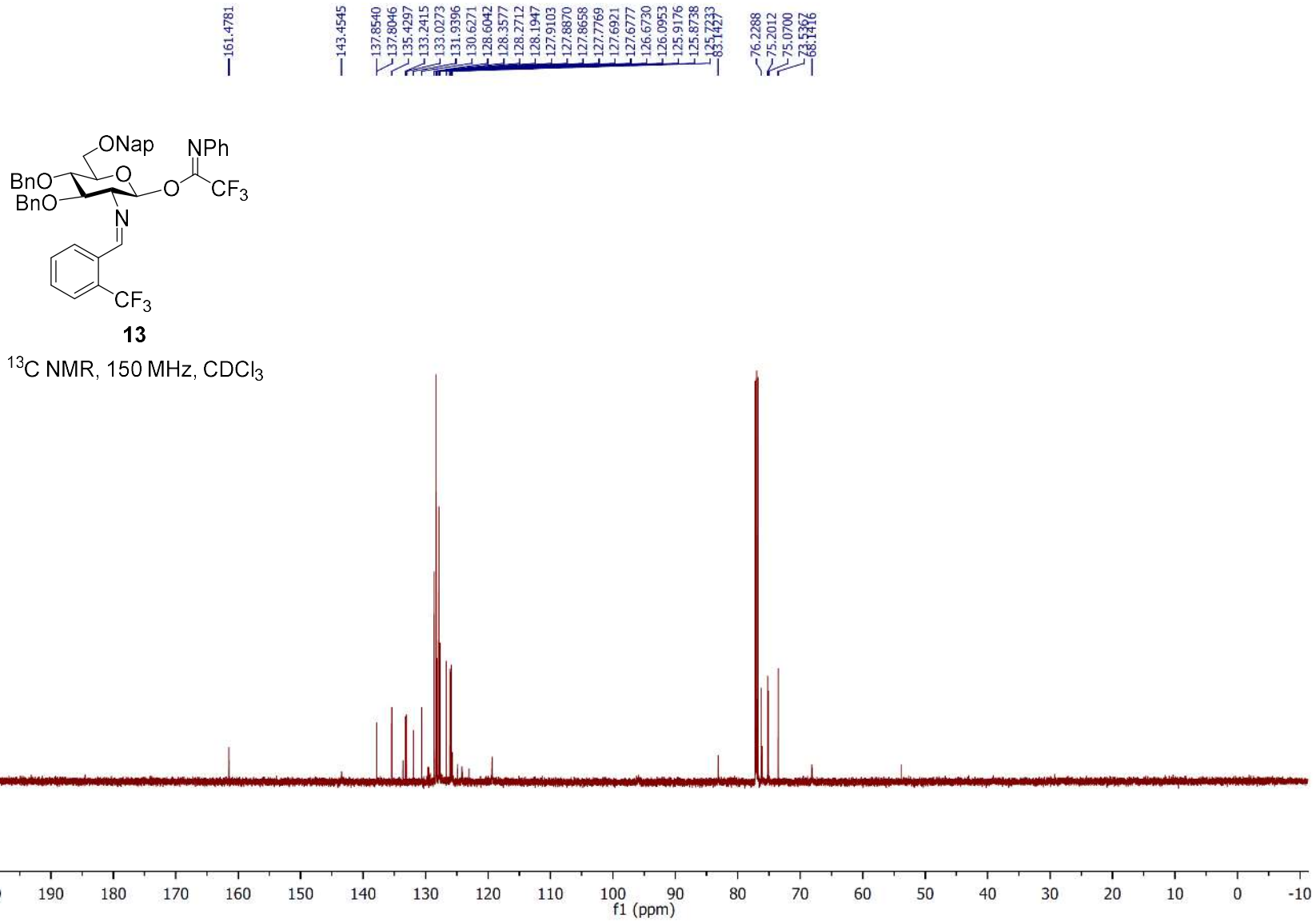

ONap
BnO
BnO
O
OH
N
CF3
13c
13C NMR, 150 MHz, CDCl3
160.6883
137.8472
135.2828
133.2045
133.0208
131.8886
130.3757
128.4259
128.3089
128.2866
128.2221
127.9551
127.9321
127.8896
127.8452
127.7470
127.7093
127.6874
127.6113
127.5725
126.7312
126.0816
125.9730
125.9064
95.5802
93.7572
83.2268
78.2840
77.8713
75.3812
75.2696
75.1729
75.0501
74.9478
73.6285
73.5731
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)



13

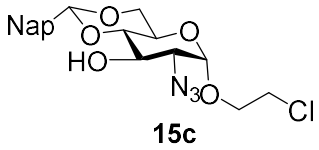
^{1}H NMR, 600 MHz, $CDCl_3$





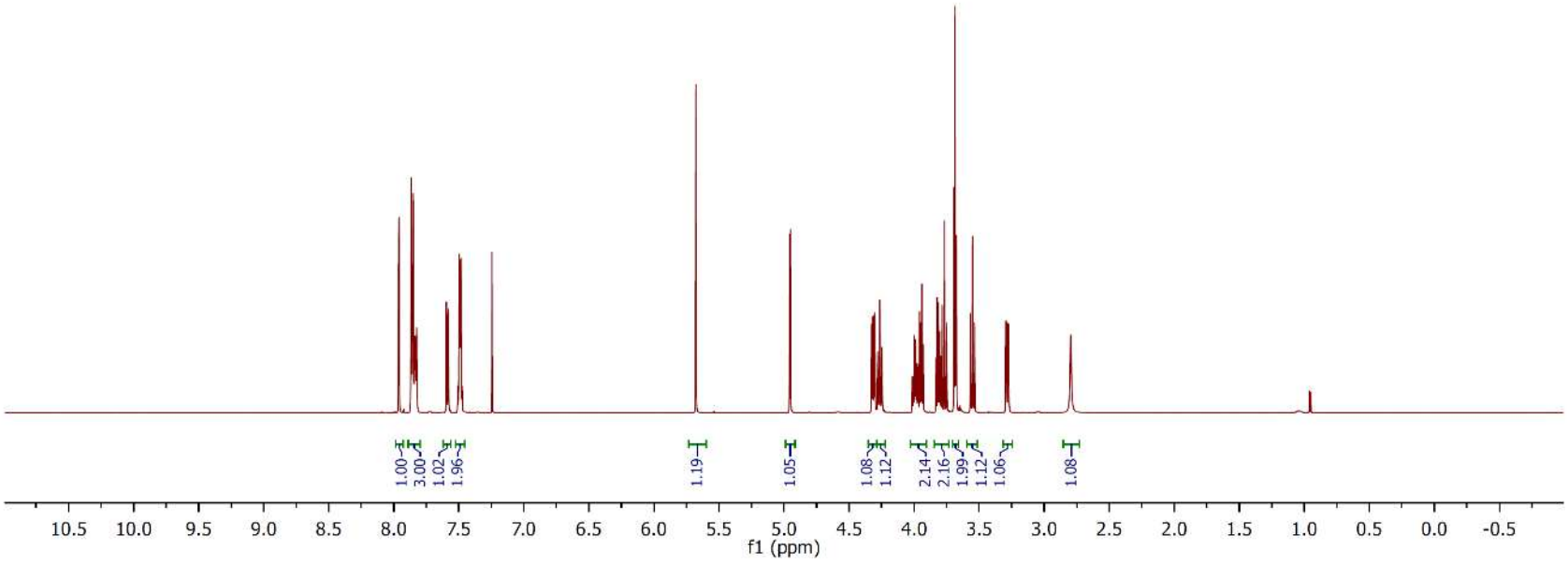
13

^{13}C NMR, 150 MHz, $CDCl_3$



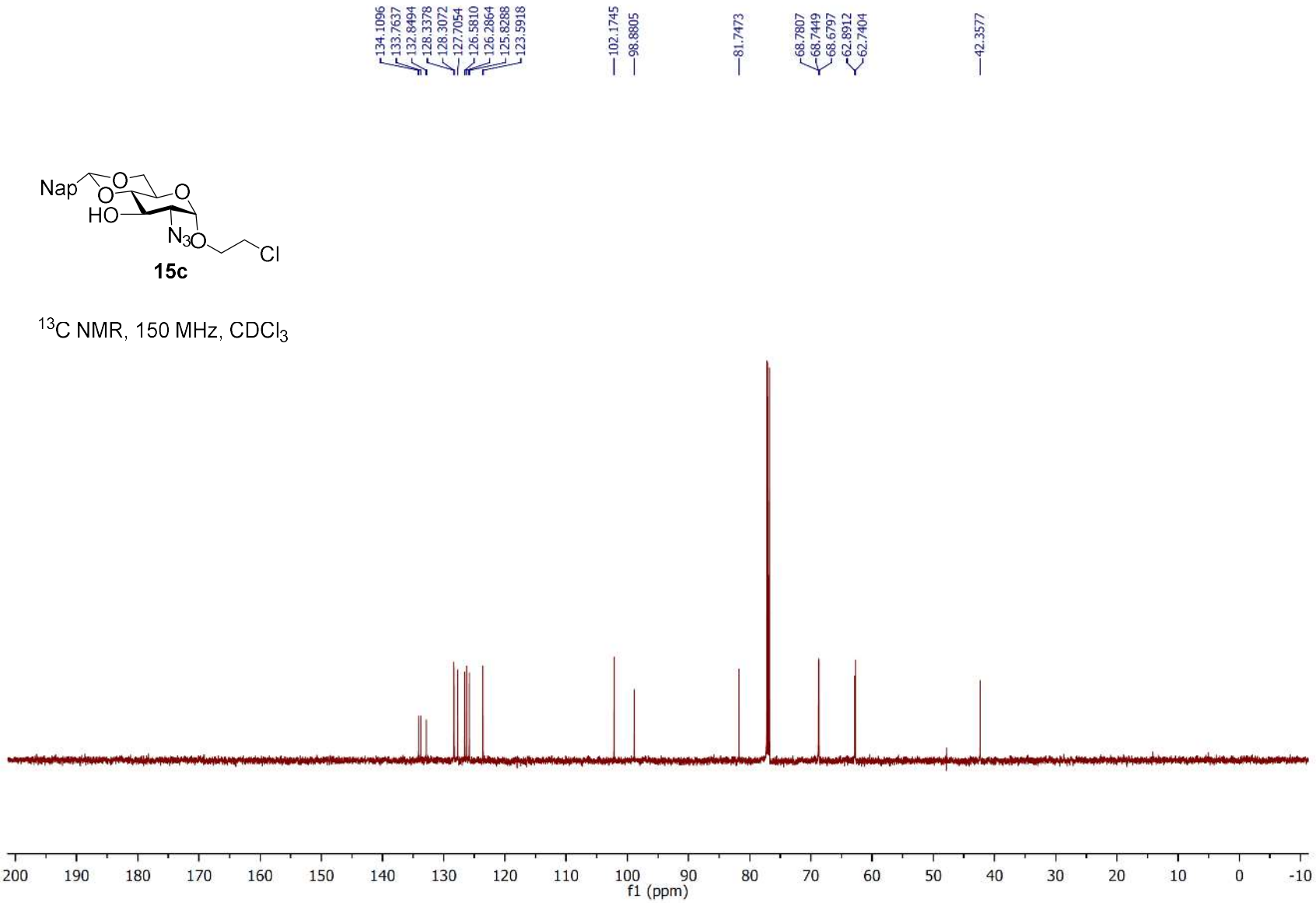
15c

^{1}H NMR, 600 MHz, $CDCl_3$



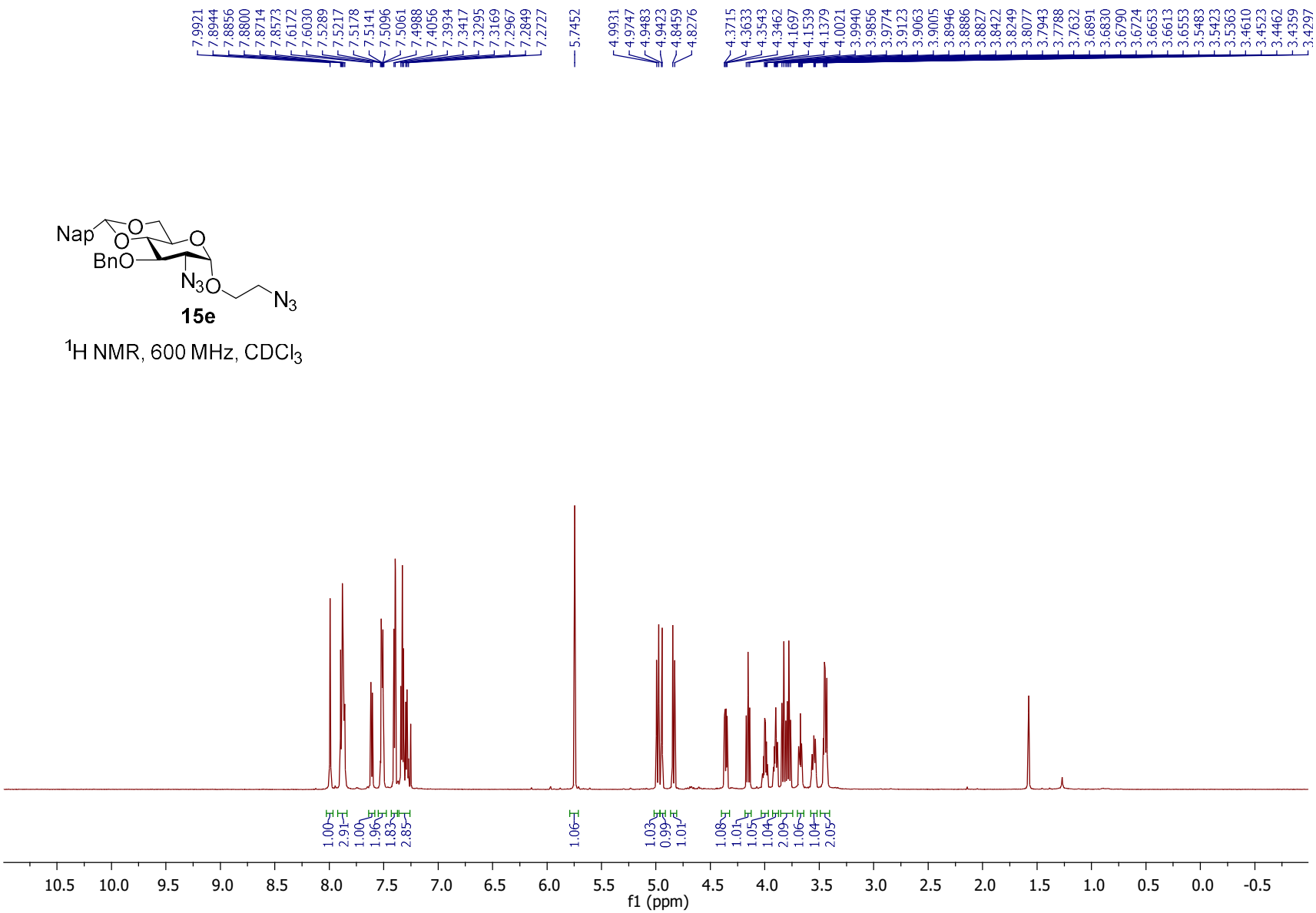
15c

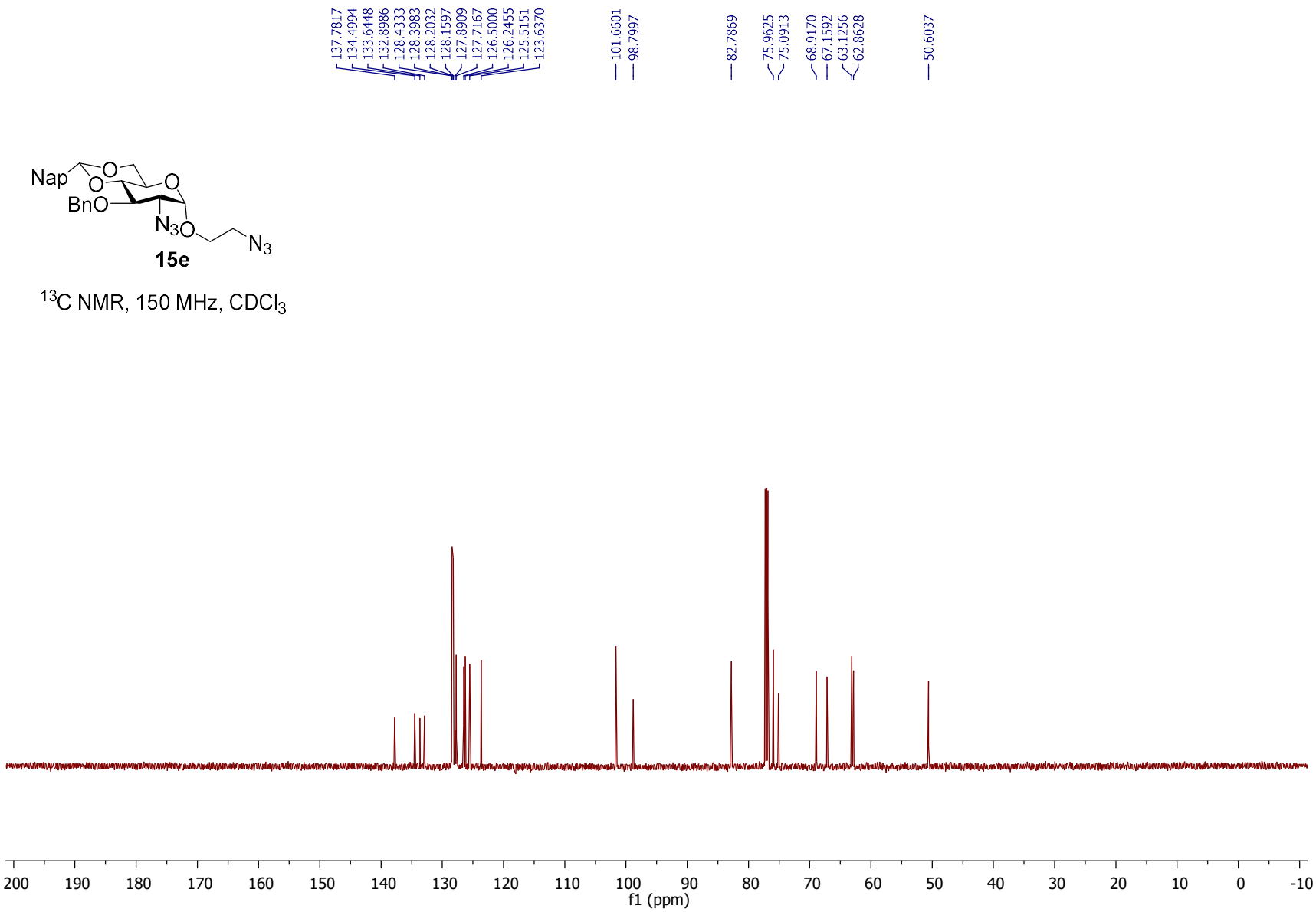
^{13}C NMR, 150 MHz, $CDCl_3$

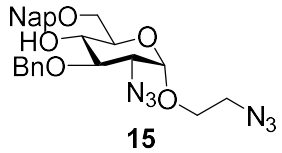


15e

^{1}H NMR, 600 MHz, $CDCl_3$

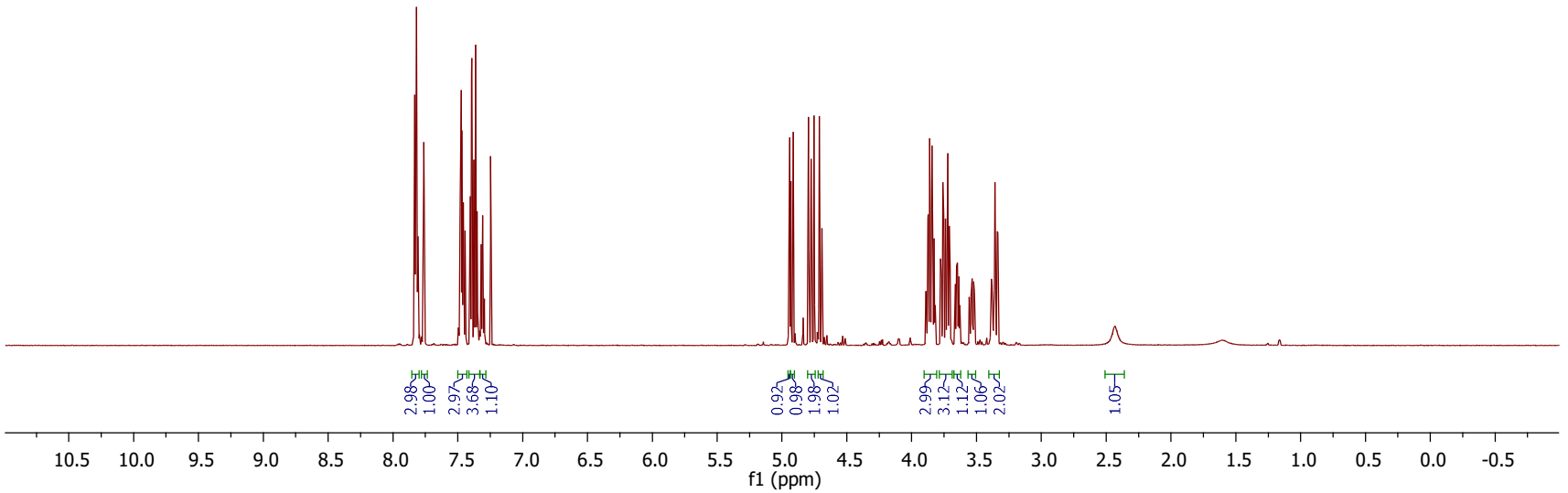


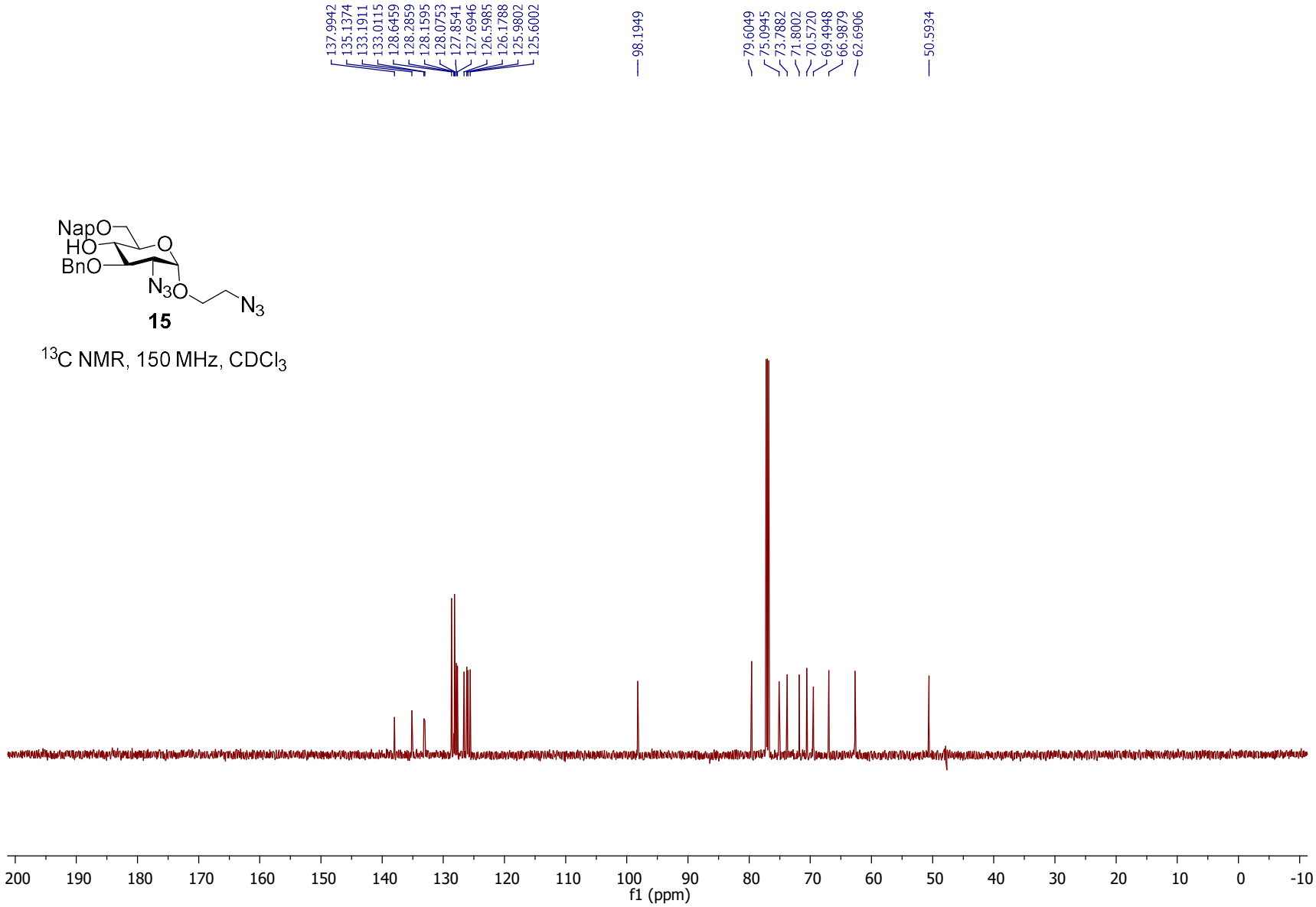

137.7817
134.4994
133.6448
132.8986
128.4333
128.3983
128.2032
128.1597
127.8909
127.7167
126.5000
126.2455
125.5151
123.6370
101.6601
98.7997
82.7869
75.9625
75.0913
68.9170
67.1592
63.1256
62.8628
50.6037
Nap
BnO
N3
O
N3
15e
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

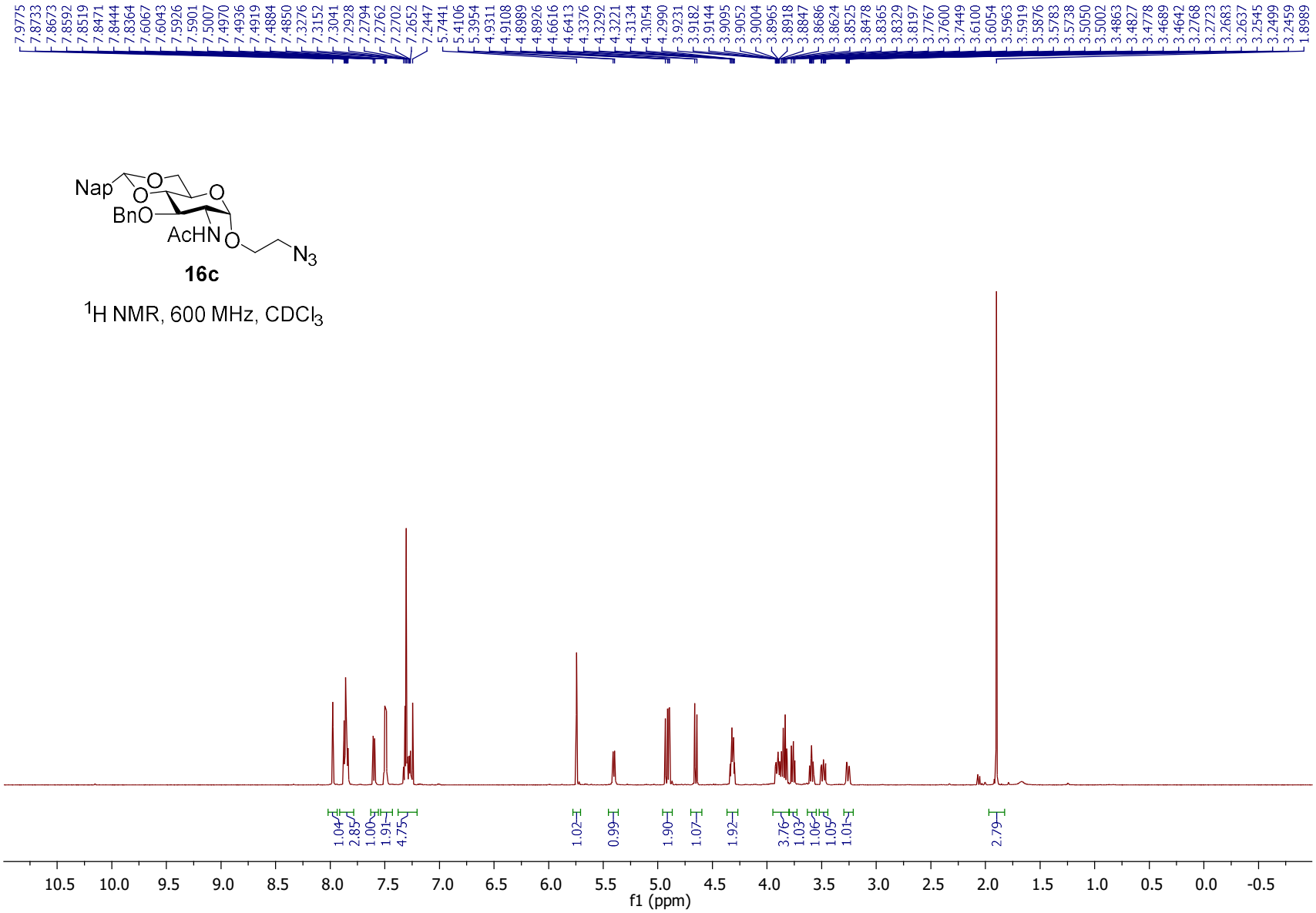


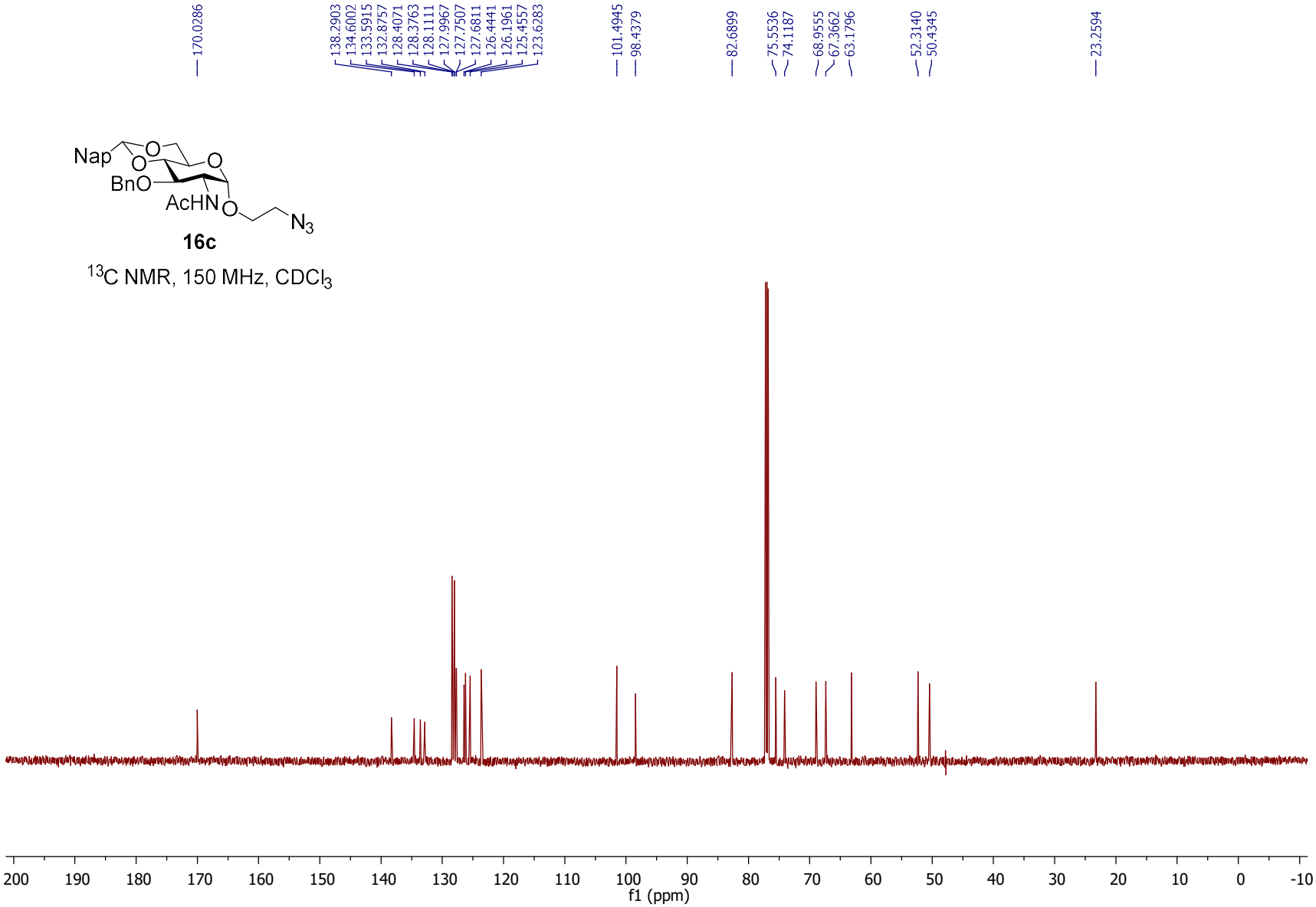
15

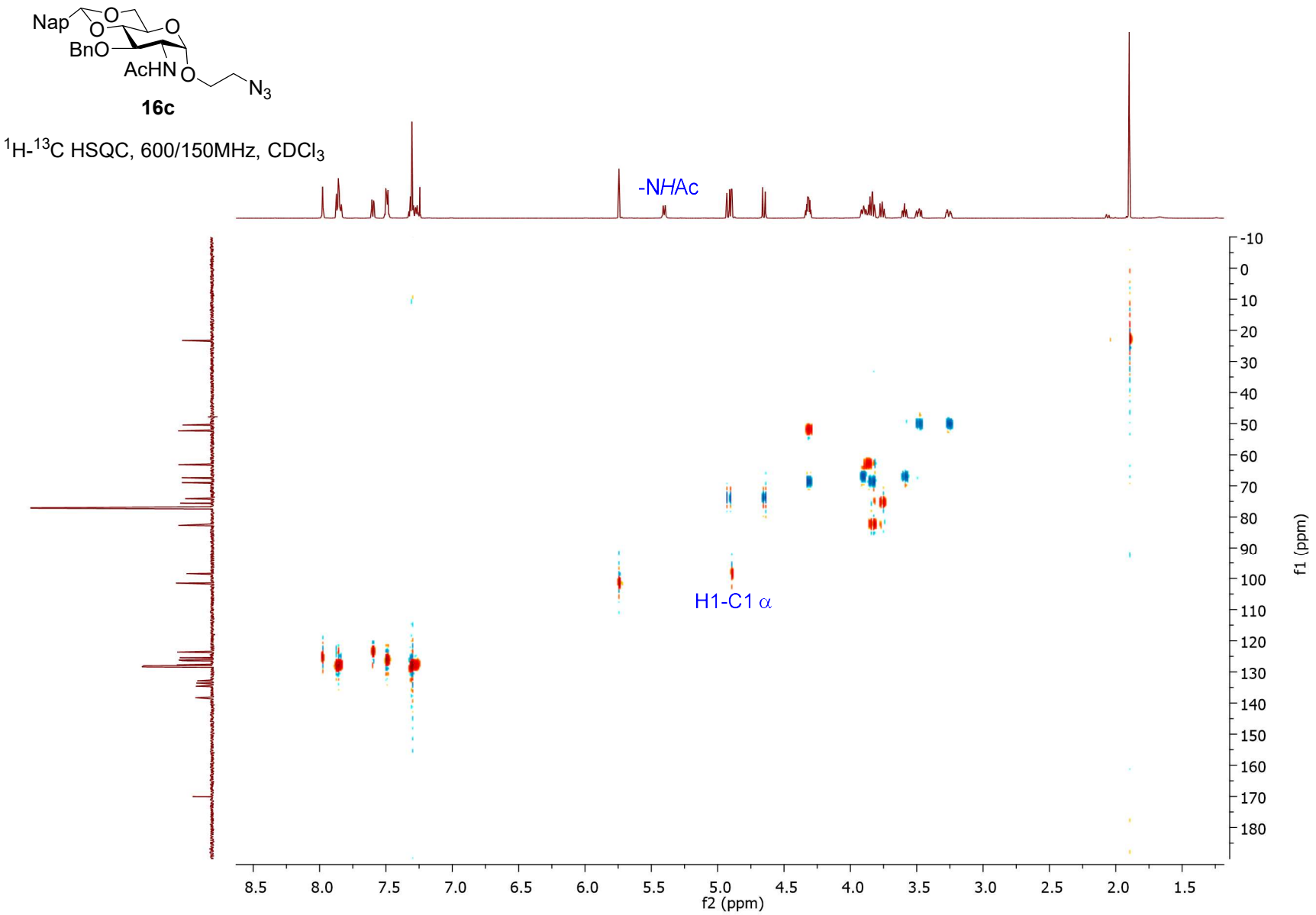
^{1}H NMR, 600 MHz, $CDCl_3$

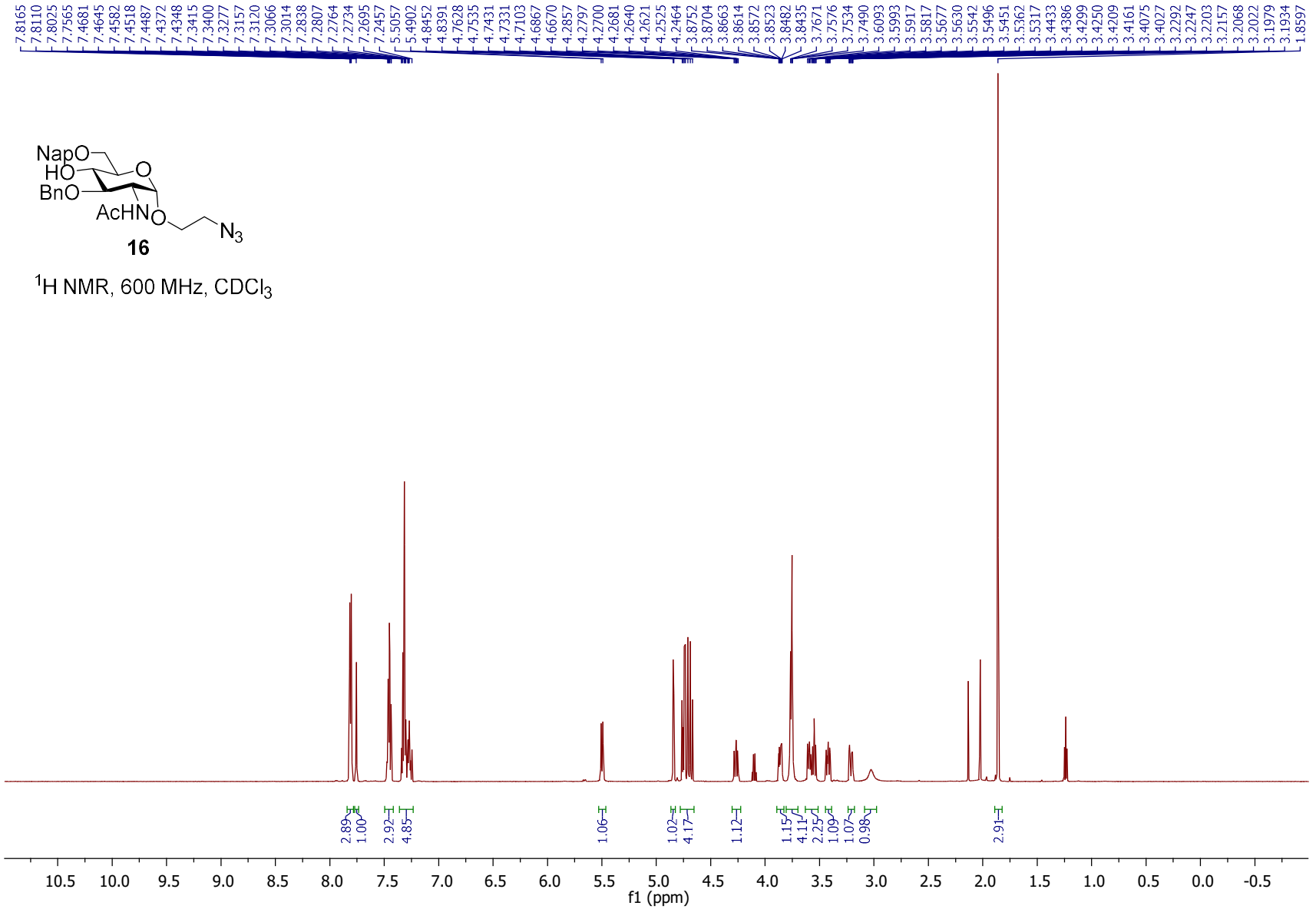


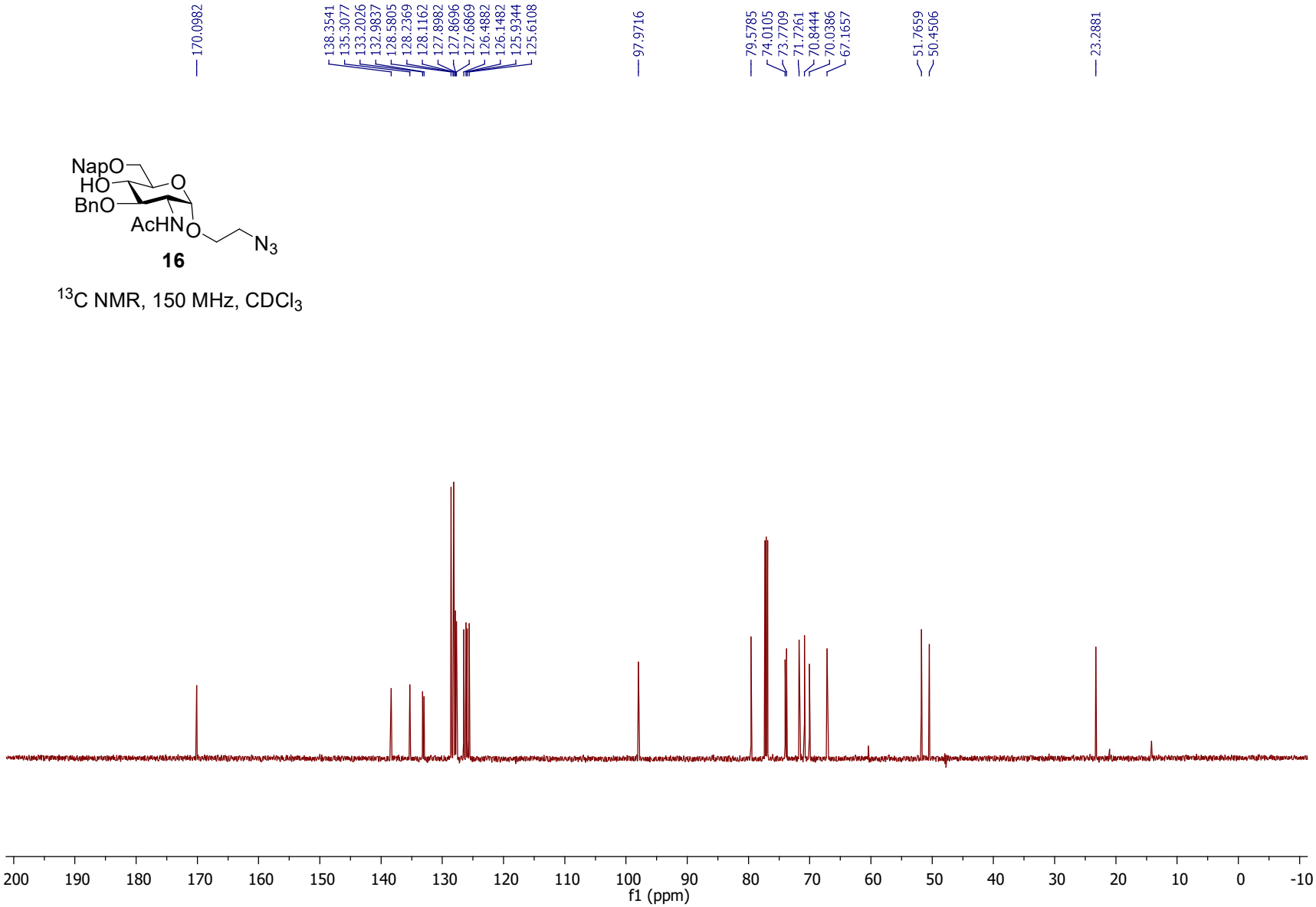

137.9942
135.1374
133.1911
133.0115
128.6459
128.2859
128.1595
128.0753
127.8541
127.6946
126.5985
126.1788
125.9802
125.6002
98.1949
79.6049
75.0945
73.7882
71.8002
70.5720
69.4948
66.9879
62.6906
50.5934
NapO
HO
BnO
O
N3O
N3
15
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


7.9775
7.8733
7.8673
7.8592
7.8519
7.8471
7.8444
7.8364
7.6067
7.6043
7.5926
7.5901
7.5007
7.4970
7.4936
7.4919
7.4884
7.4850
7.3276
7.3152
7.3041
7.2928
7.2794
7.2762
7.2702
7.2652
7.2447
5.7441
5.4106
5.3954
4.9311
4.9108
4.8989
4.8926
4.6616
4.6413
4.3376
4.3292
4.3221
4.3134
4.3054
4.2990
3.9231
3.9182
3.9144
3.9095
3.9052
3.9004
3.8965
3.8918
3.8847
3.8686
3.8624
3.8525
3.8478
3.8365
3.8329
3.8197
3.7767
3.7600
3.7449
3.6100
3.6054
3.5963
3.5919
3.5876
3.5783
3.5738
3.5050
3.5002
3.4863
3.4827
3.4778
3.4689
3.4642
3.2768
3.2723
3.2683
3.2637
3.2545
3.2499
3.2459
1.8989
Nap
O
O
O
BnO
AcHN
O
N3
16c
1H NMR, 600 MHz, CDCl3
1.04
2.85
1.00
1.91
4.75
1.02
0.99
1.90
1.07
1.92
3.76
1.03
1.06
1.05
1.01
2.79
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)


170.0286
138.2903
134.6002
133.5915
132.8757
128.4071
128.3763
128.1111
127.9967
127.7507
127.6811
126.4441
126.1961
125.4557
123.6283
101.4945
98.4379
82.6899
75.5536
74.1187
68.9555
67.3662
63.1796
52.3140
50.4345
23.2594
Nap
O
O
O
BnO
AcHN
O
N3
16c
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

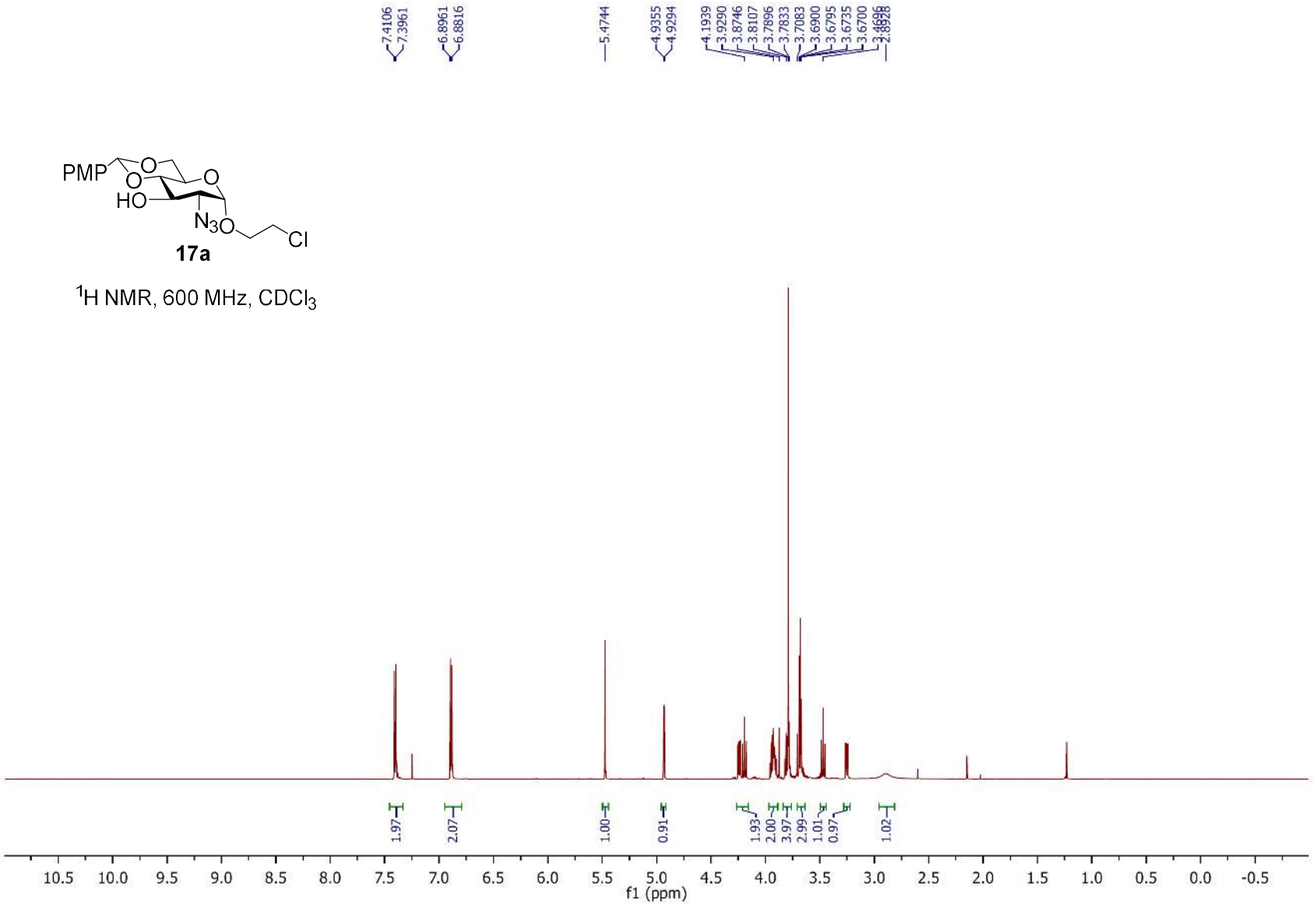

Nap
O
O
O
BnO
AcHN
O
N3
16c
1H-13C HSQC, 600/150MHz, CDCl3
-NHAc
H1-C1 α
f2 (ppm)
f1 (ppm)

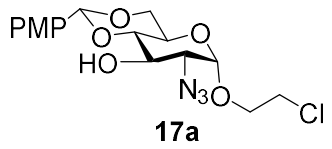

NapO
HO
BnO
AcHN
O
N3
16
1H NMR, 600 MHz, CDCl3
f1 (ppm)


170.0982
138.3541
135.3077
133.2026
132.9837
128.5805
128.2369
128.1162
127.8982
127.8696
127.6869
126.4882
126.1482
125.9344
125.6108
97.9716
79.5785
74.0105
73.7709
71.7261
70.8444
70.0386
67.1657
51.7659
50.4506
23.2881
NapO
HO
BnO
AcHN
O
O
N3
16
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

17a

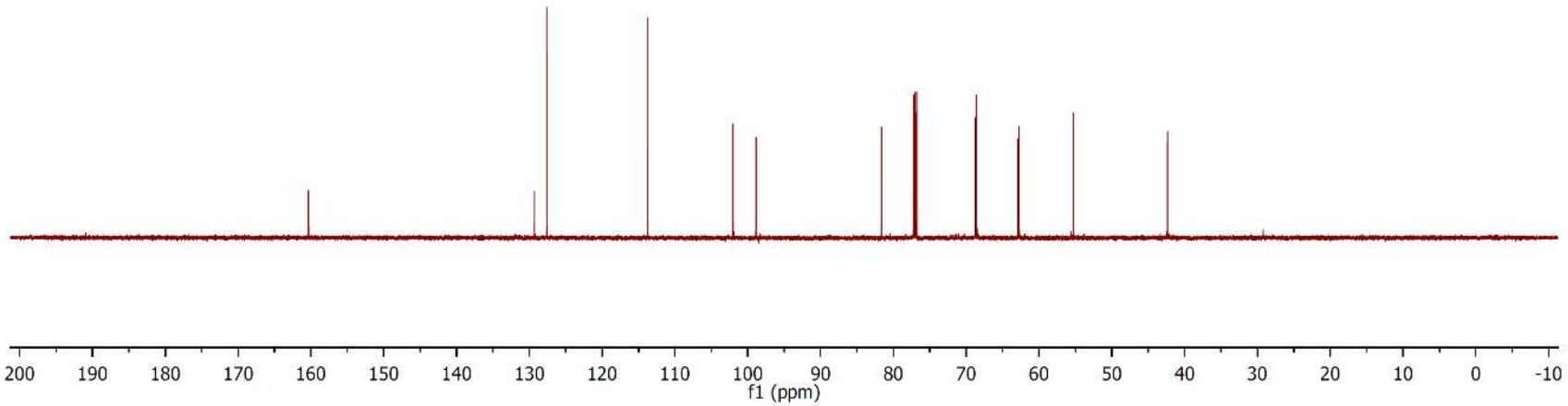
^{1}H NMR, 600 MHz, $CDCl_3$





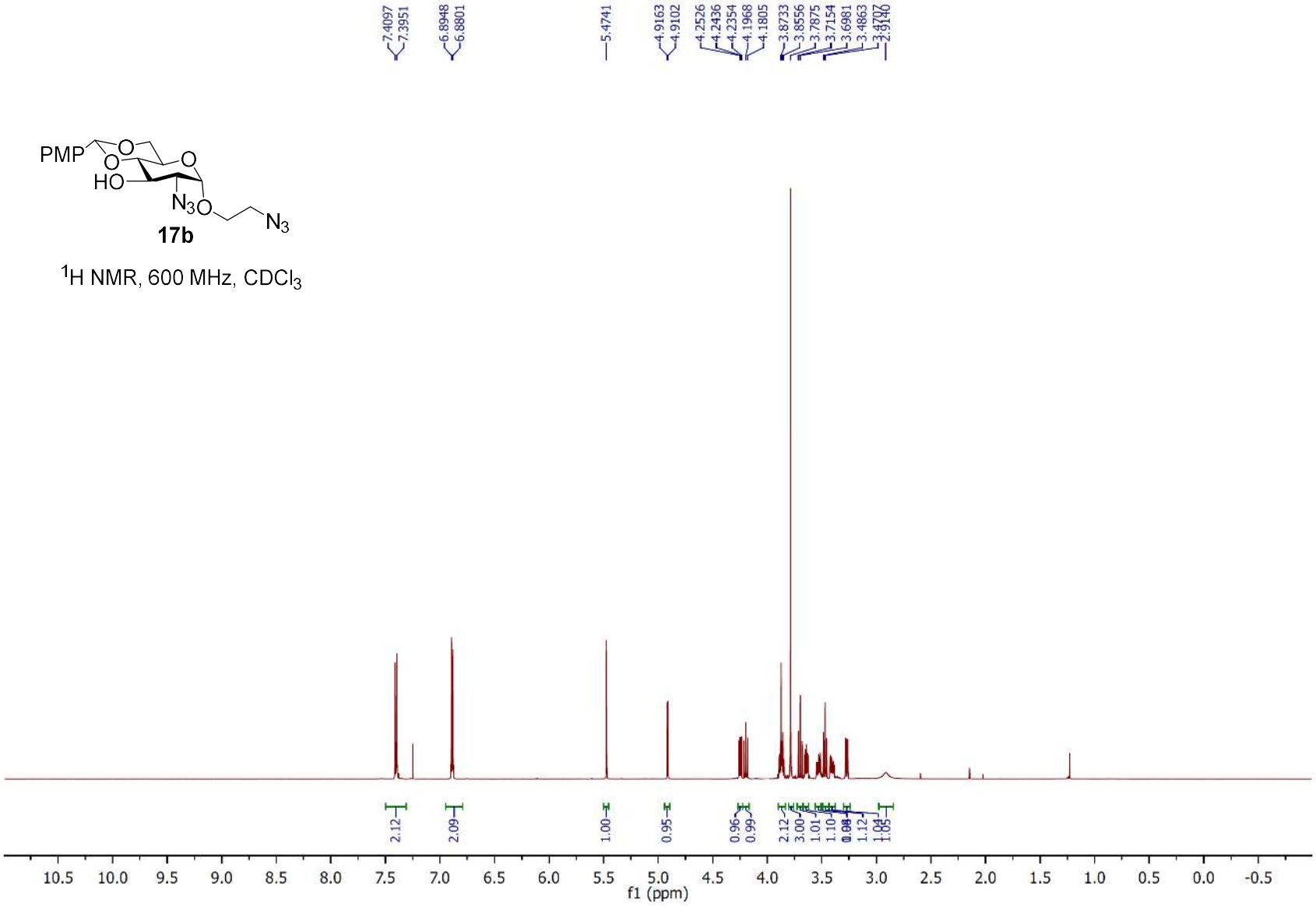
17a

^{13}C NMR, 150 MHz, $CDCl_3$



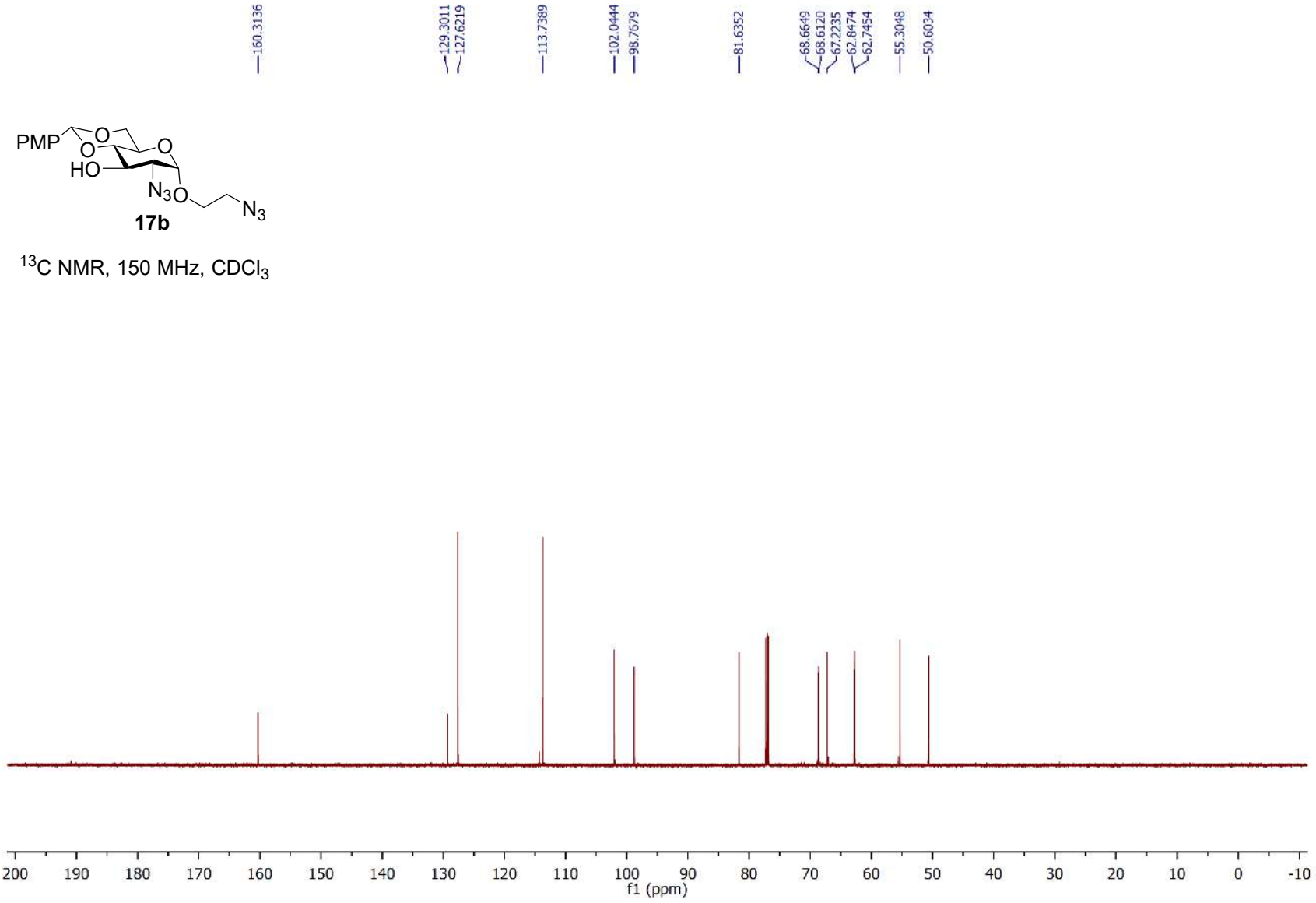
17b

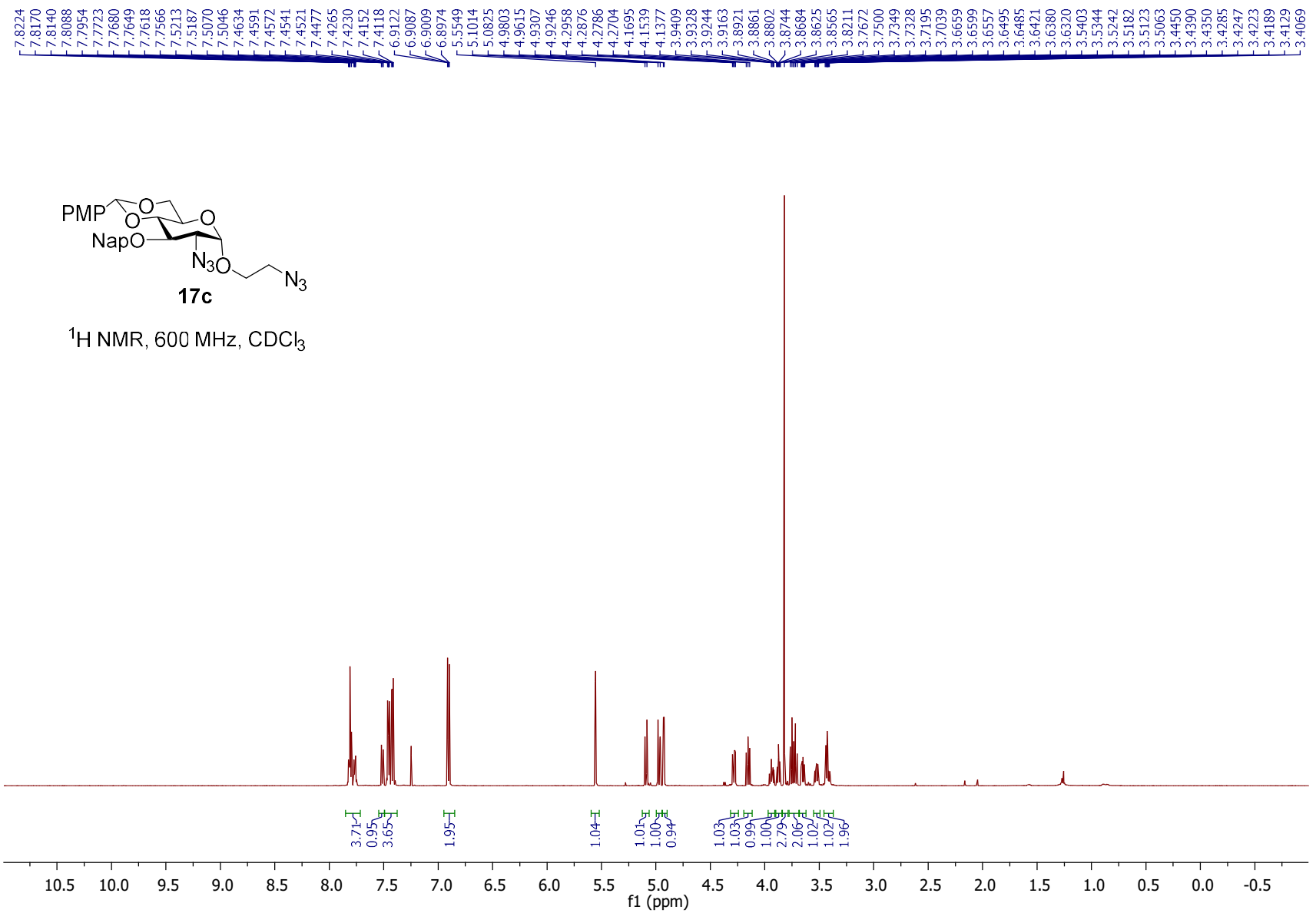
^{1}H NMR, 600 MHz, $CDCl_3$



17b

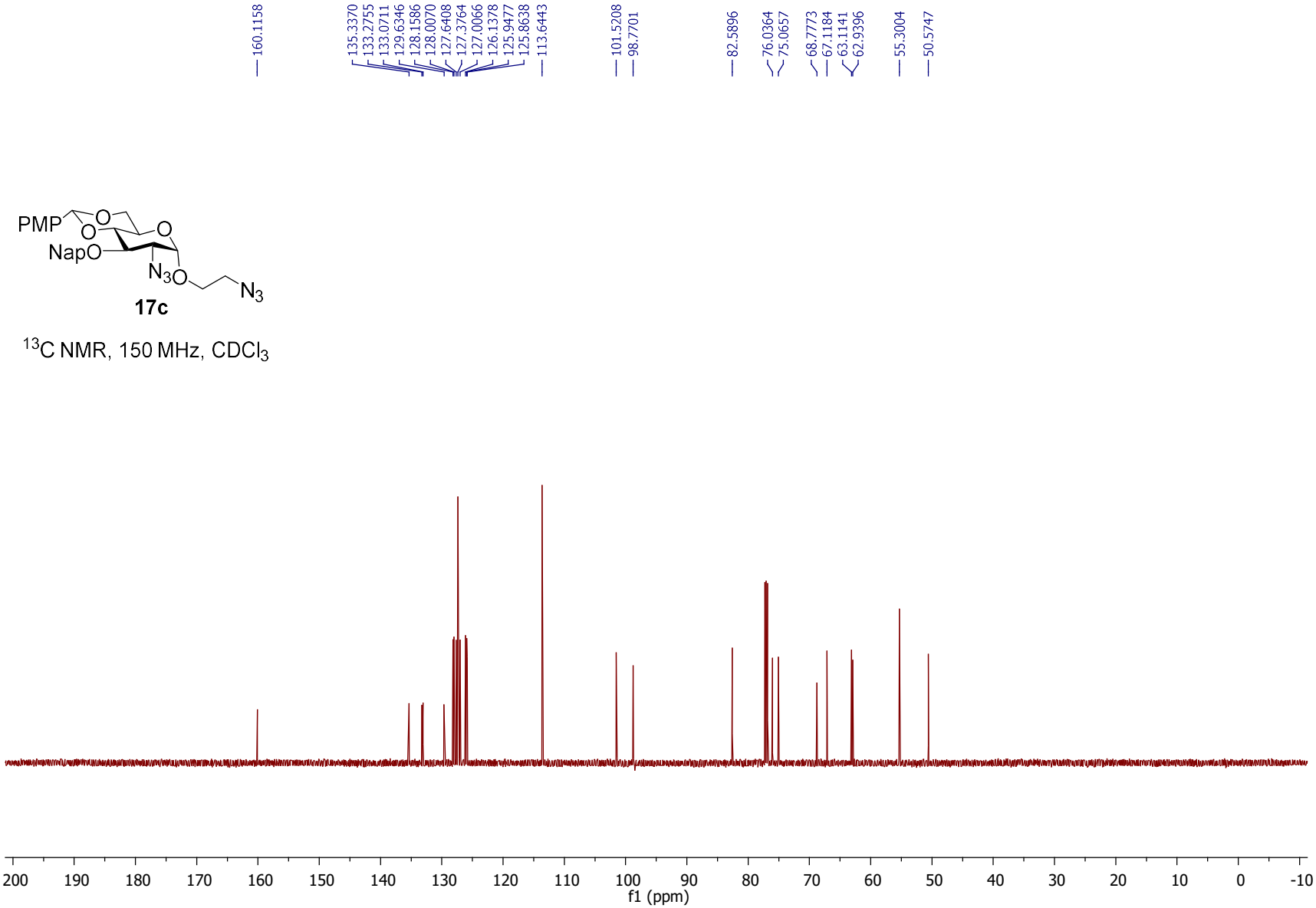
^{13}C NMR, 150 MHz, $CDCl_3$

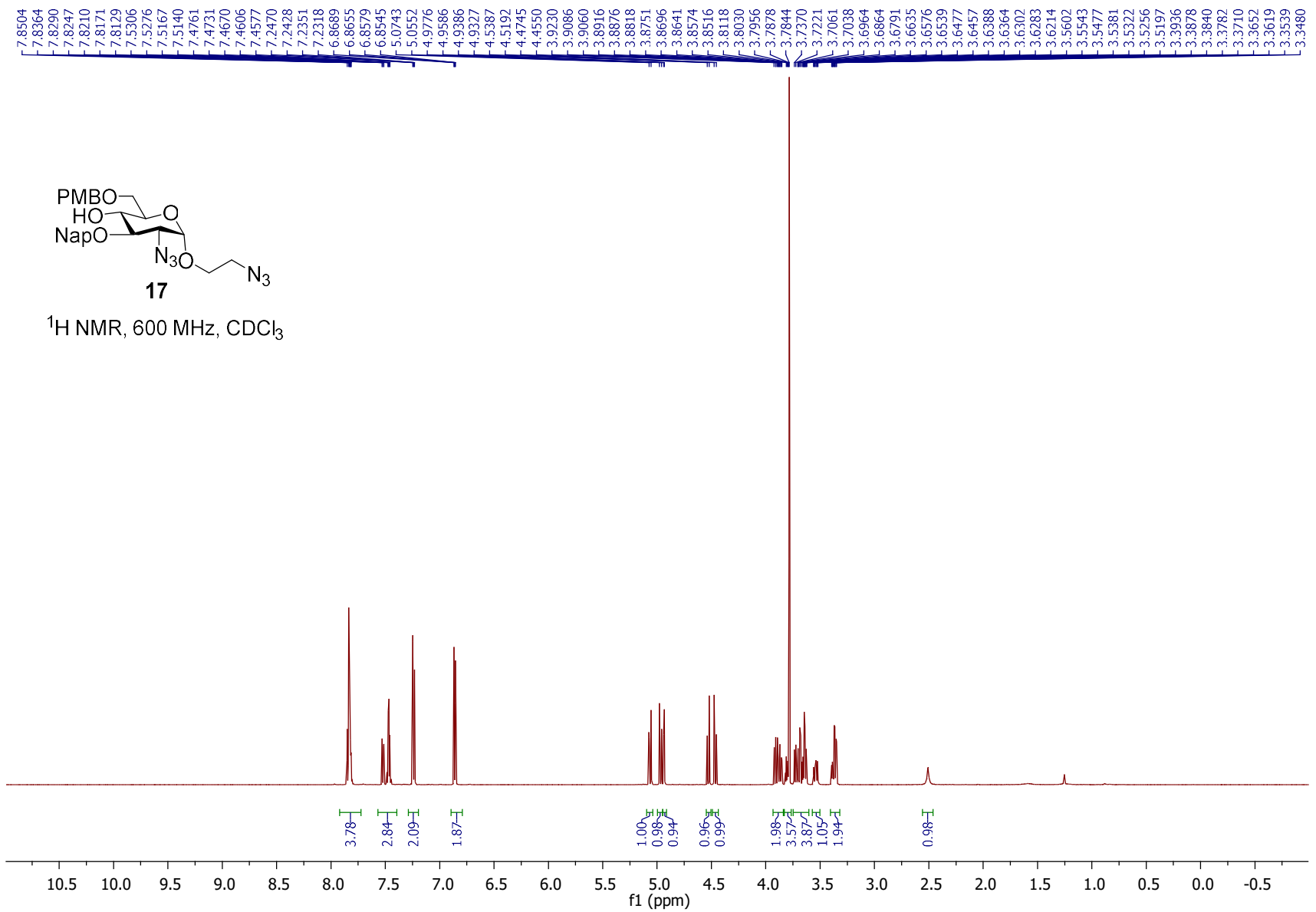



7.8224
7.8170
7.8140
7.8088
7.7954
7.7723
7.7680
7.7649
7.7618
7.7566
7.5213
7.5187
7.5070
7.5046
7.4634
7.4591
7.4572
7.4541
7.4521
7.4477
7.4265
7.4230
7.4152
7.4118
6.9122
6.9087
6.9009
6.8974
5.5549
5.1014
5.0825
4.9803
4.9615
4.9307
4.9246
4.2958
4.2876
4.2786
4.2704
4.1695
4.1539
4.1377
3.9409
3.9328
3.9244
3.9163
3.8921
3.8861
3.8802
3.8744
3.8684
3.8625
3.8565
3.8211
3.7672
3.7500
3.7349
3.7328
3.7195
3.7039
3.6659
3.6599
3.6557
3.6495
3.6485
3.6421
3.6380
3.6320
3.5403
3.5344
3.5242
3.5182
3.5123
3.5063
3.4450
3.4390
3.4350
3.4285
3.4247
3.4223
3.4189
3.4129
3.4069
PMP
NapO
N3O
N3
17c
1H NMR, 600 MHz, CDCl3
3.71
0.95
3.65
1.95
1.04
1.01
1.00
0.94
1.03
1.03
0.99
1.00
2.79
2.06
1.02
1.02
1.96
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)

17c

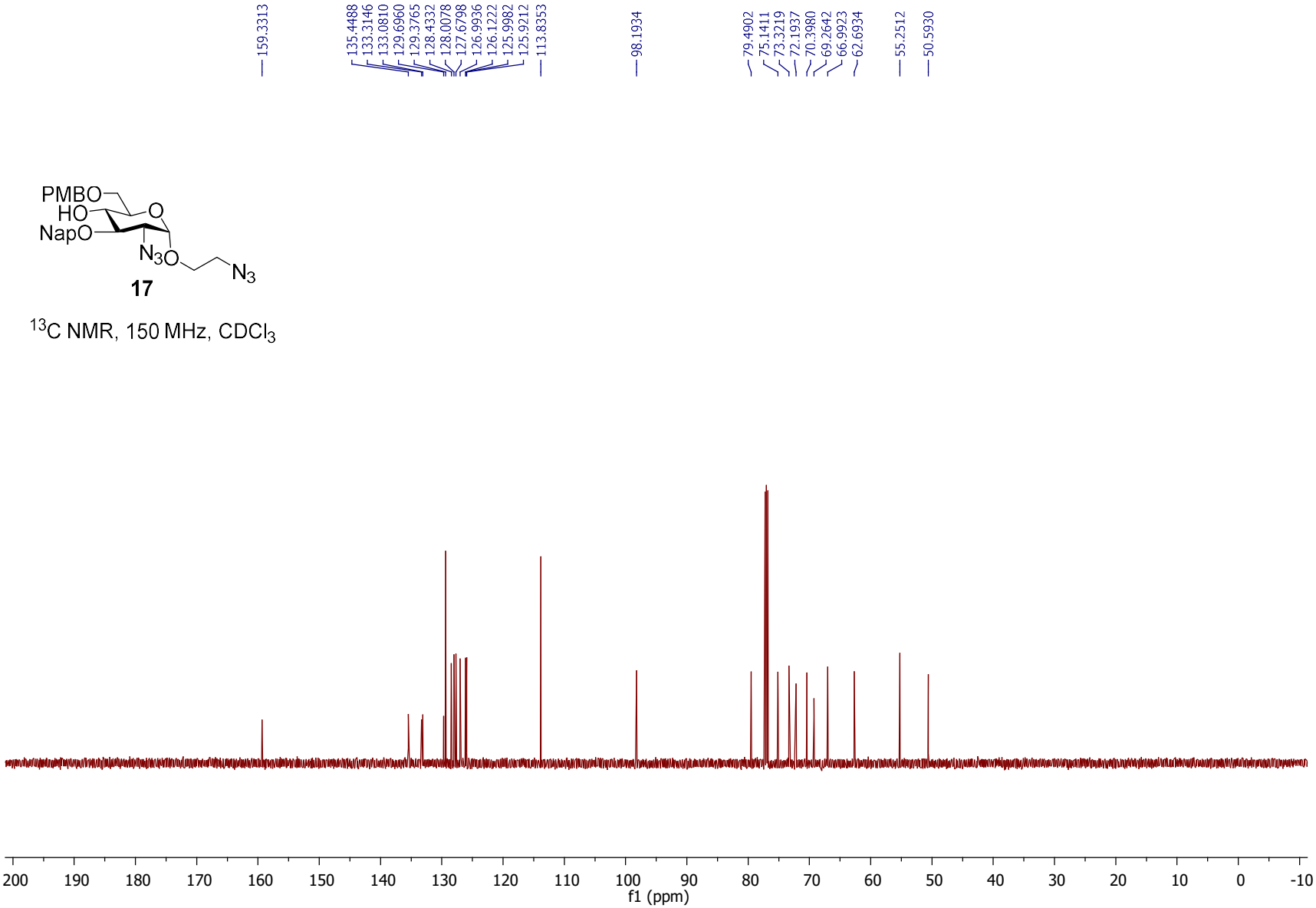
^{13}C NMR, 150 MHz, $CDCl_3$

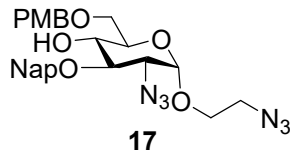



PMBO
HO
NapO
O
N3O
N3
17
1H NMR, 600 MHz, CDCl3
f1 (ppm)

17

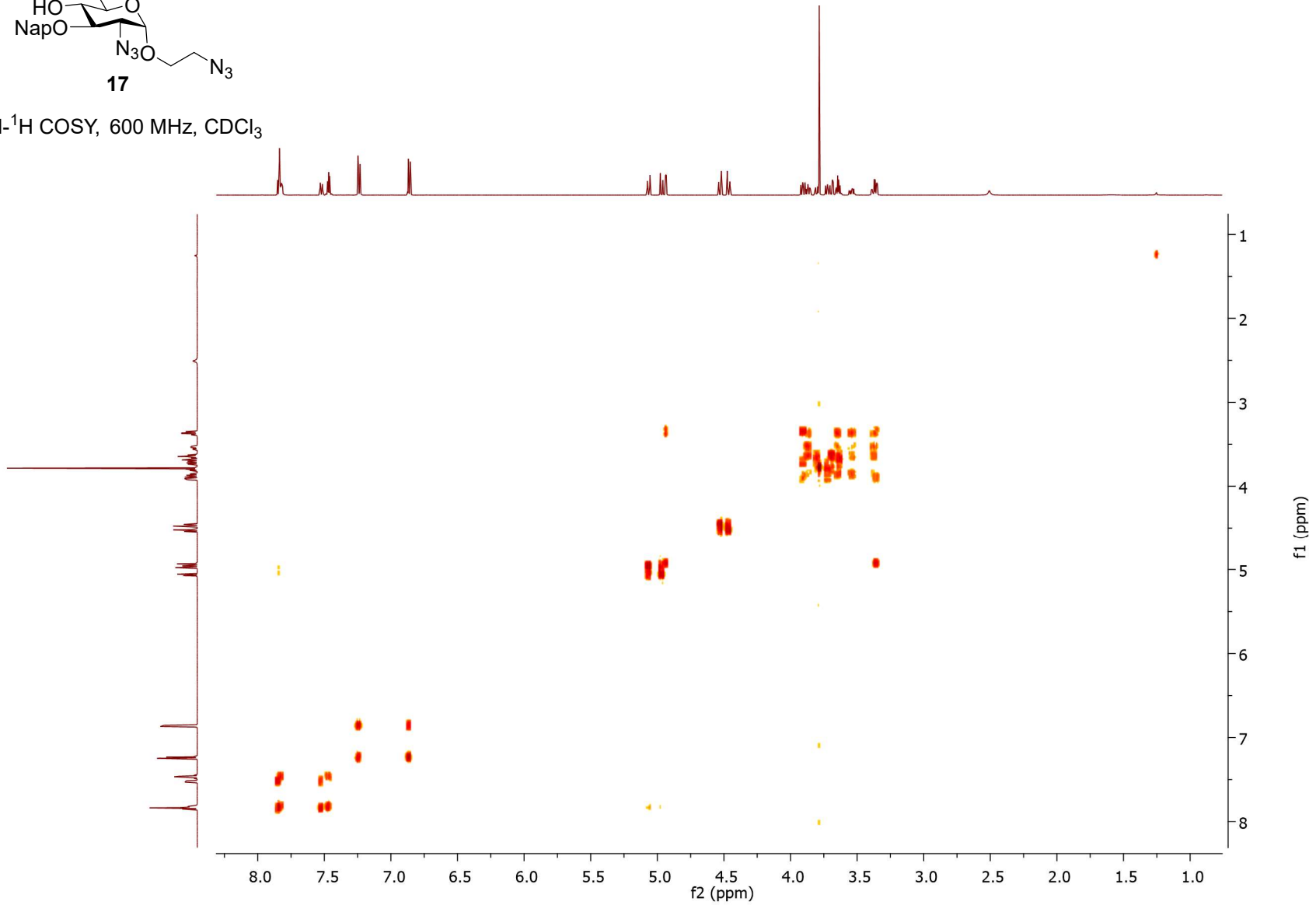
^{13}C NMR, 150 MHz, $CDCl_3$

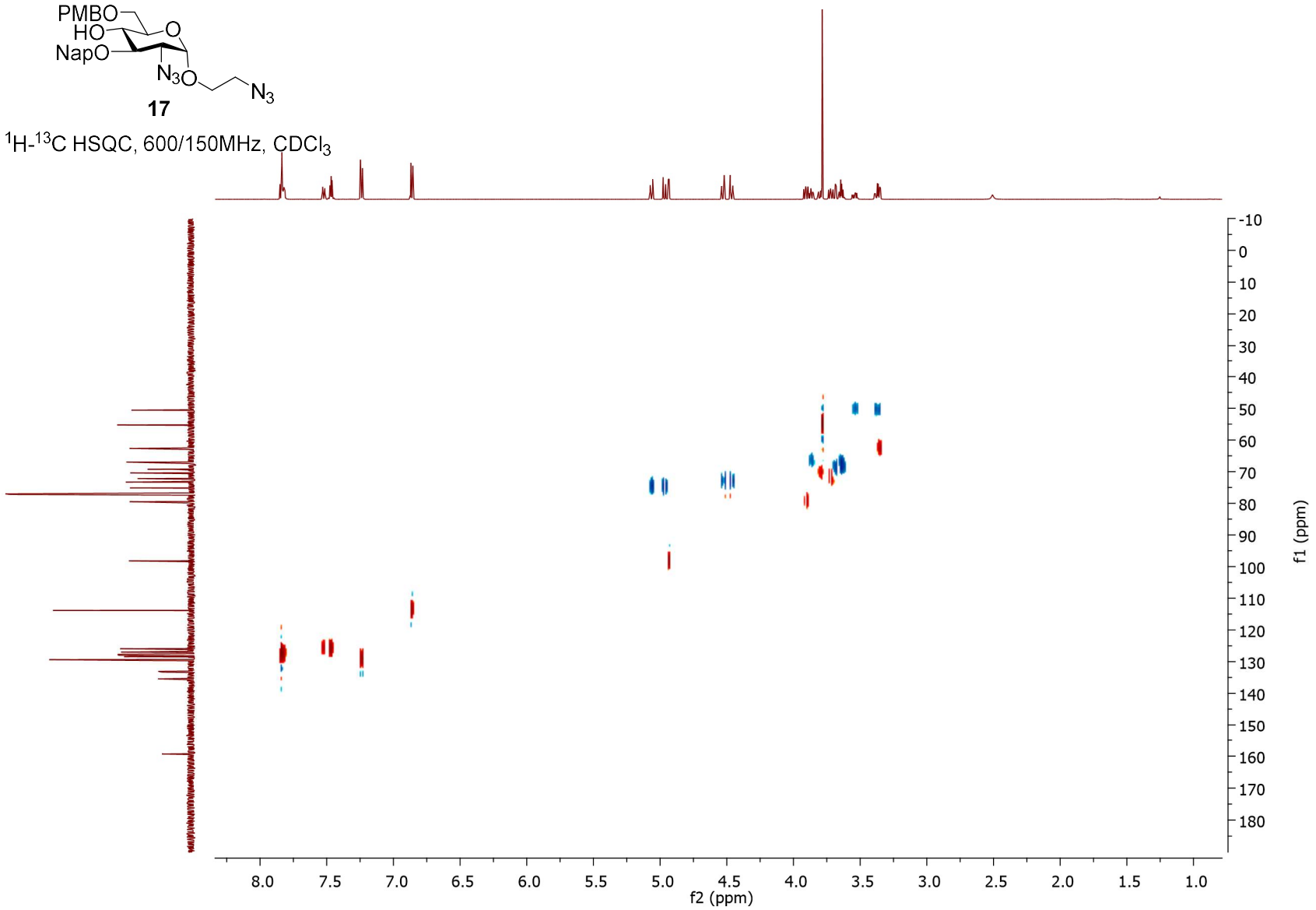


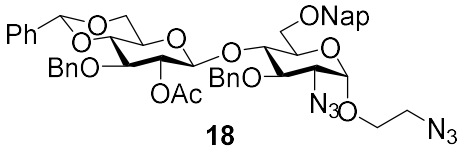


17

^{1}H-^{1}H COSY, 600 MHz, $CDCl_3$

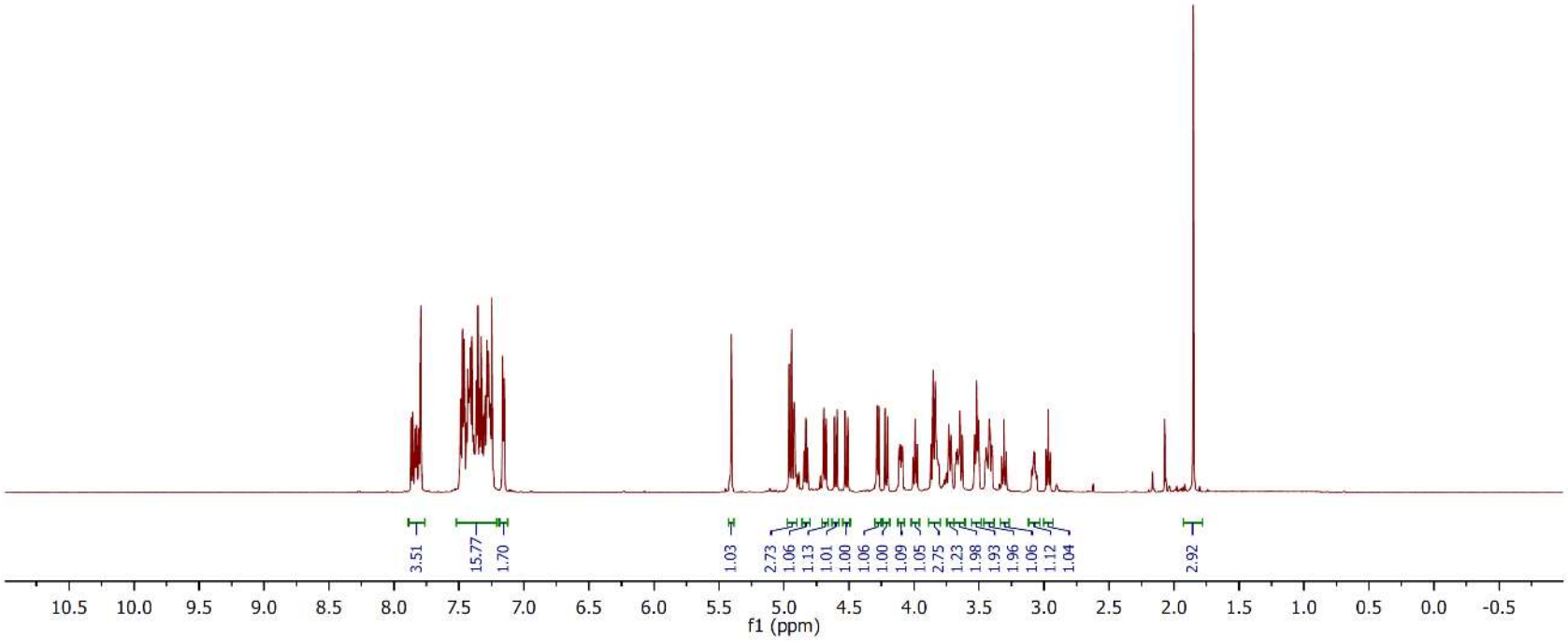


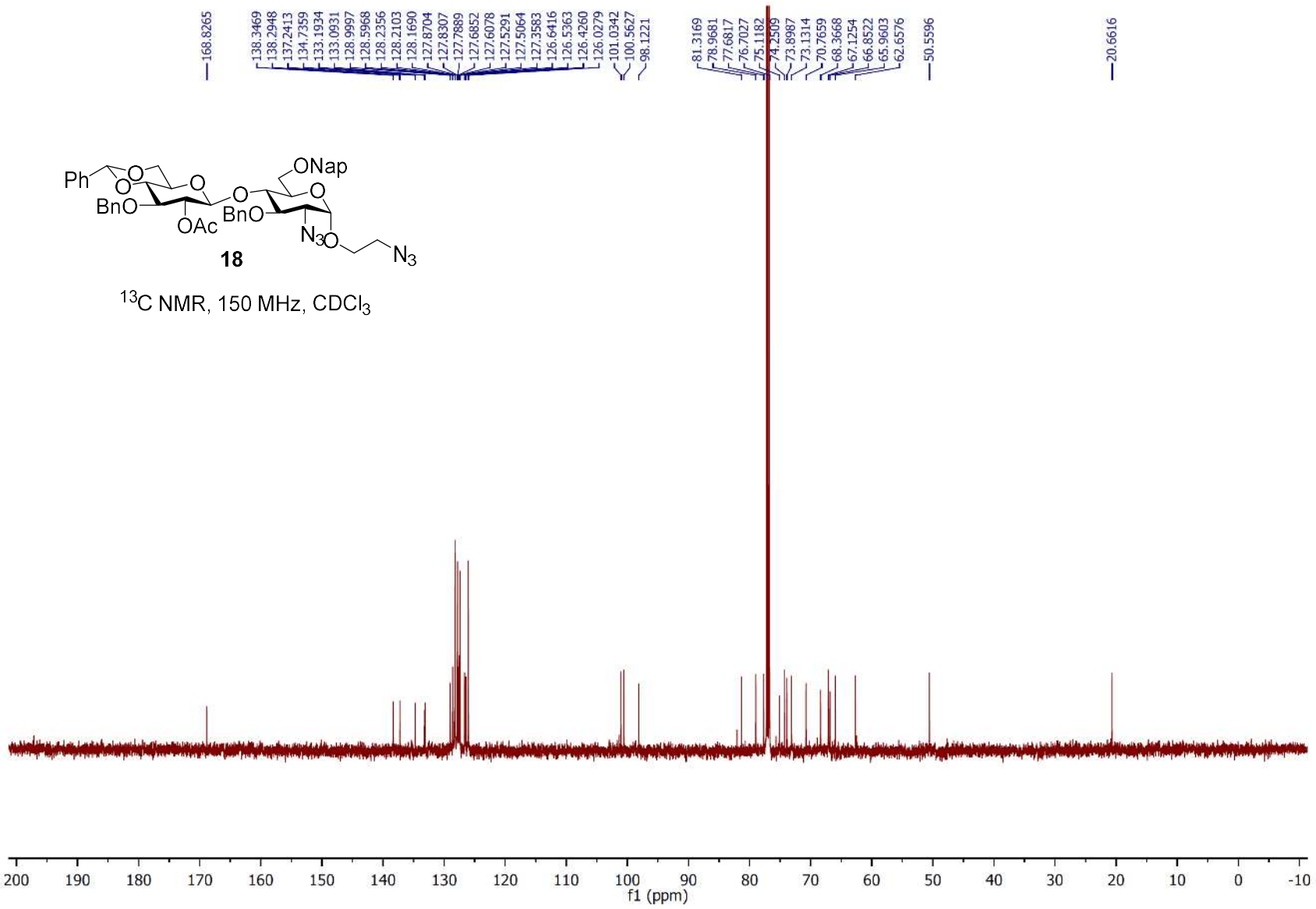

PMBO
HO
NapO
O
N3O
N3
17
1H-13C HSQC, 600/150MHz, CDCl3
f2 (ppm)
f1 (ppm)

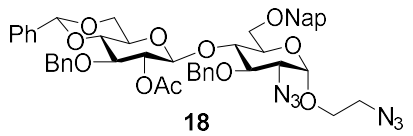


18

^{1}H NMR, 600 MHz, $CDCl_3$

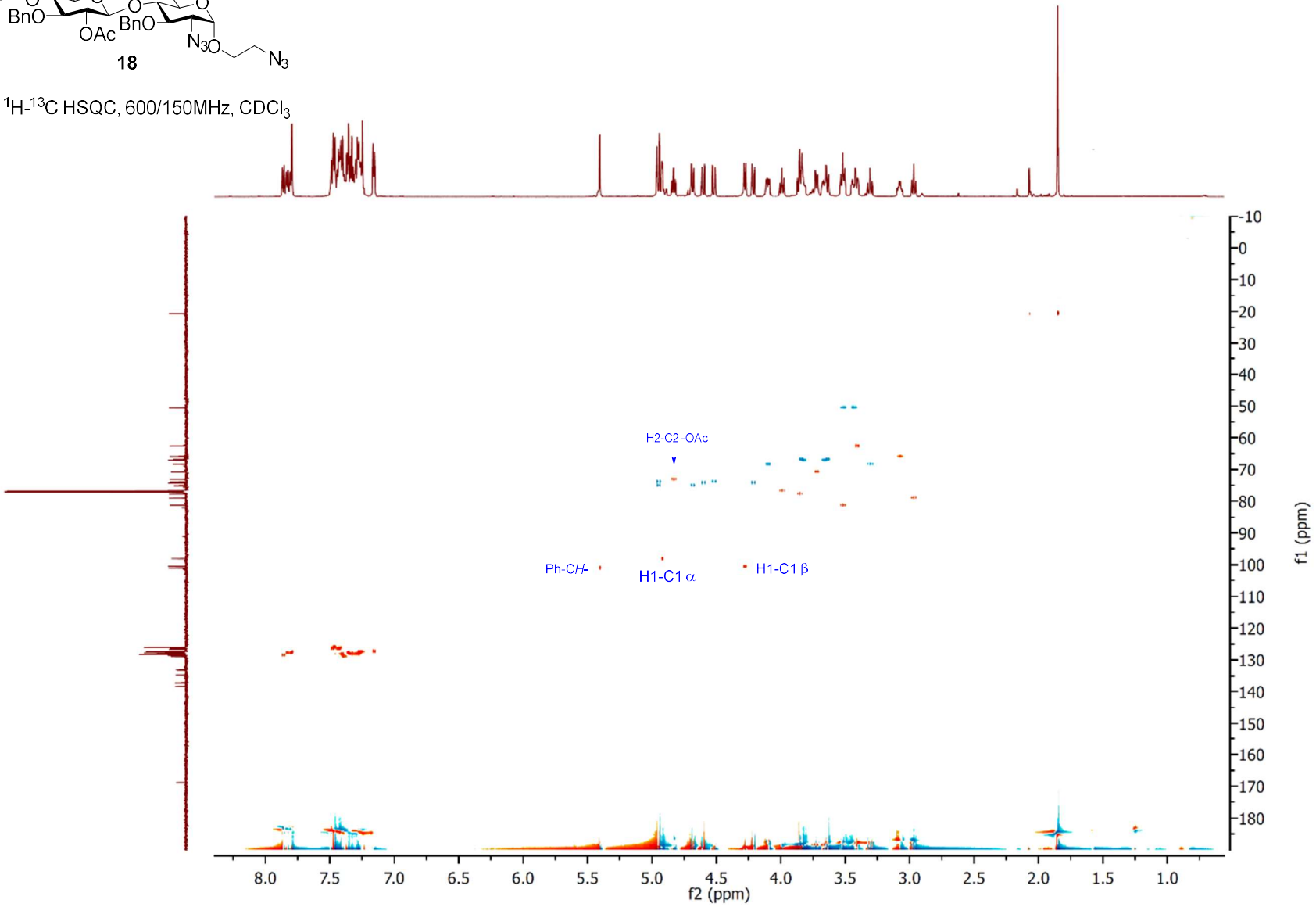


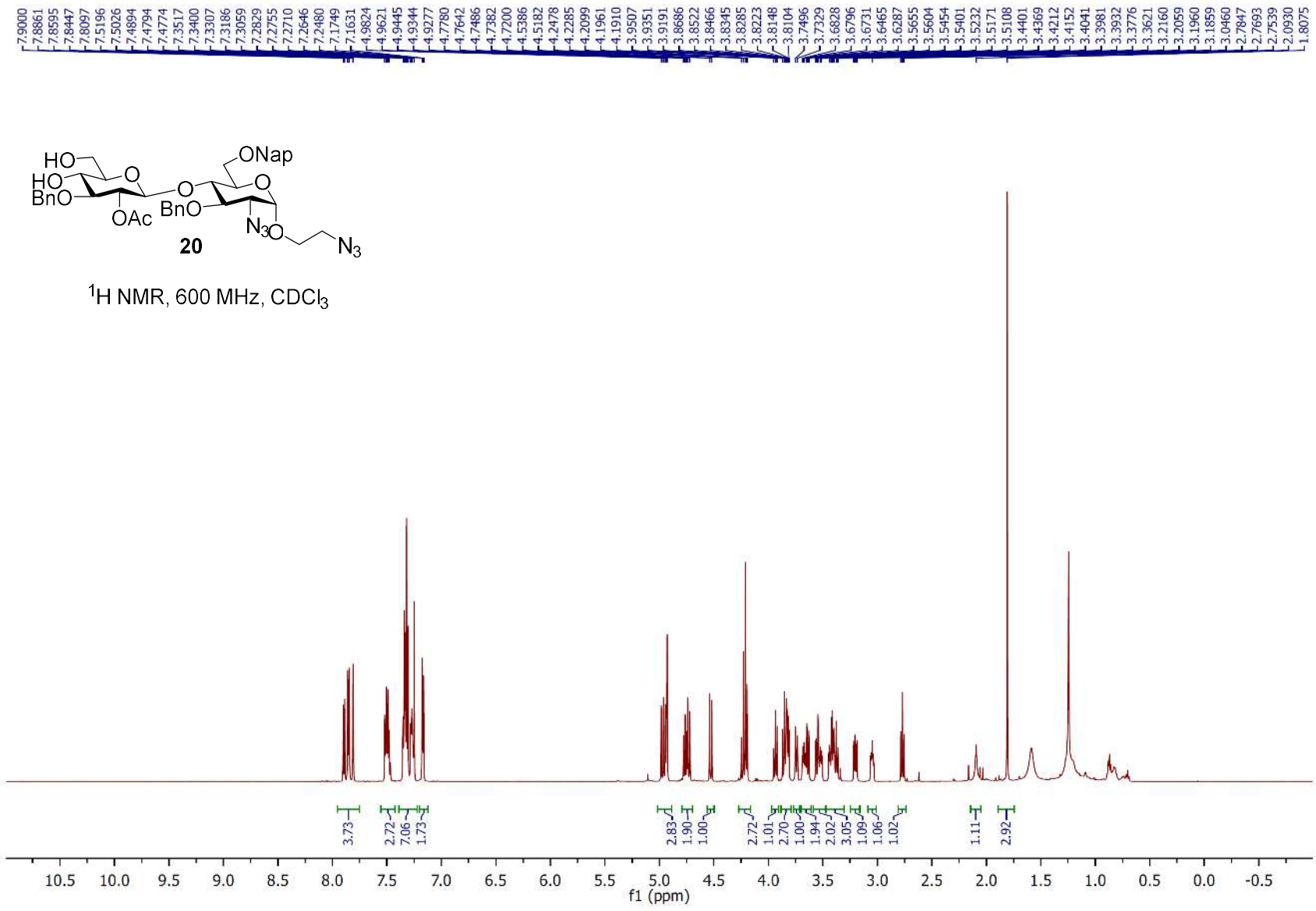

168.8265
138.3469
138.2948
137.2413
134.7359
133.1934
133.0931
128.9997
128.5968
128.2356
128.2103
128.1690
127.8704
127.8307
127.7889
127.6852
127.6078
127.5291
127.5064
127.3583
126.6416
126.5363
126.4260
126.0279
101.0342
100.5627
98.1221
81.3169
78.9681
77.6817
76.7027
75.1182
74.2509
73.8987
73.1314
70.7659
68.3668
67.1254
66.8522
65.9603
62.6576
50.5596
20.6616
Ph
O
O
O
BnO
OAc
BnO
O
ONap
O
N3
O
N3
18
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

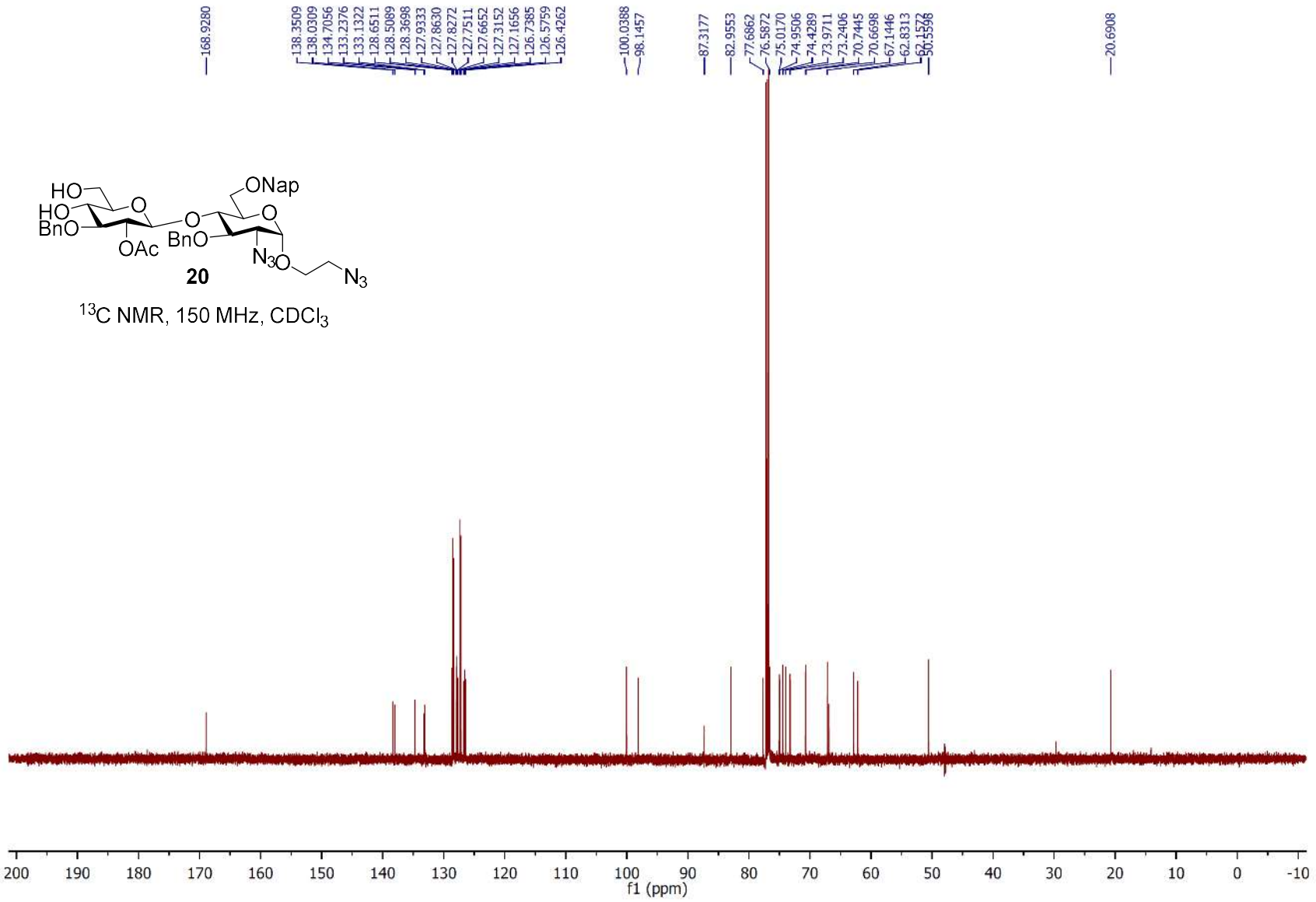


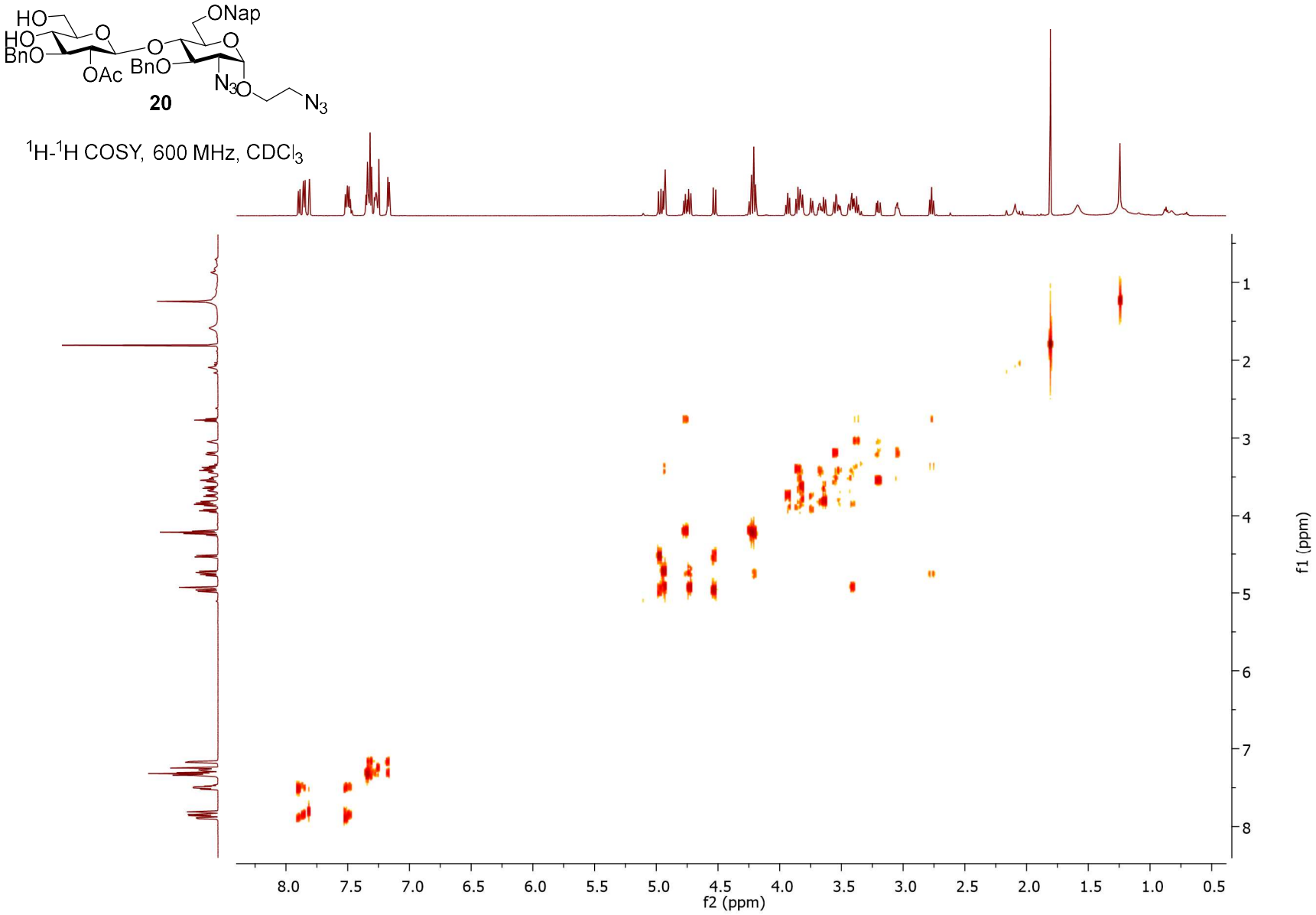
18

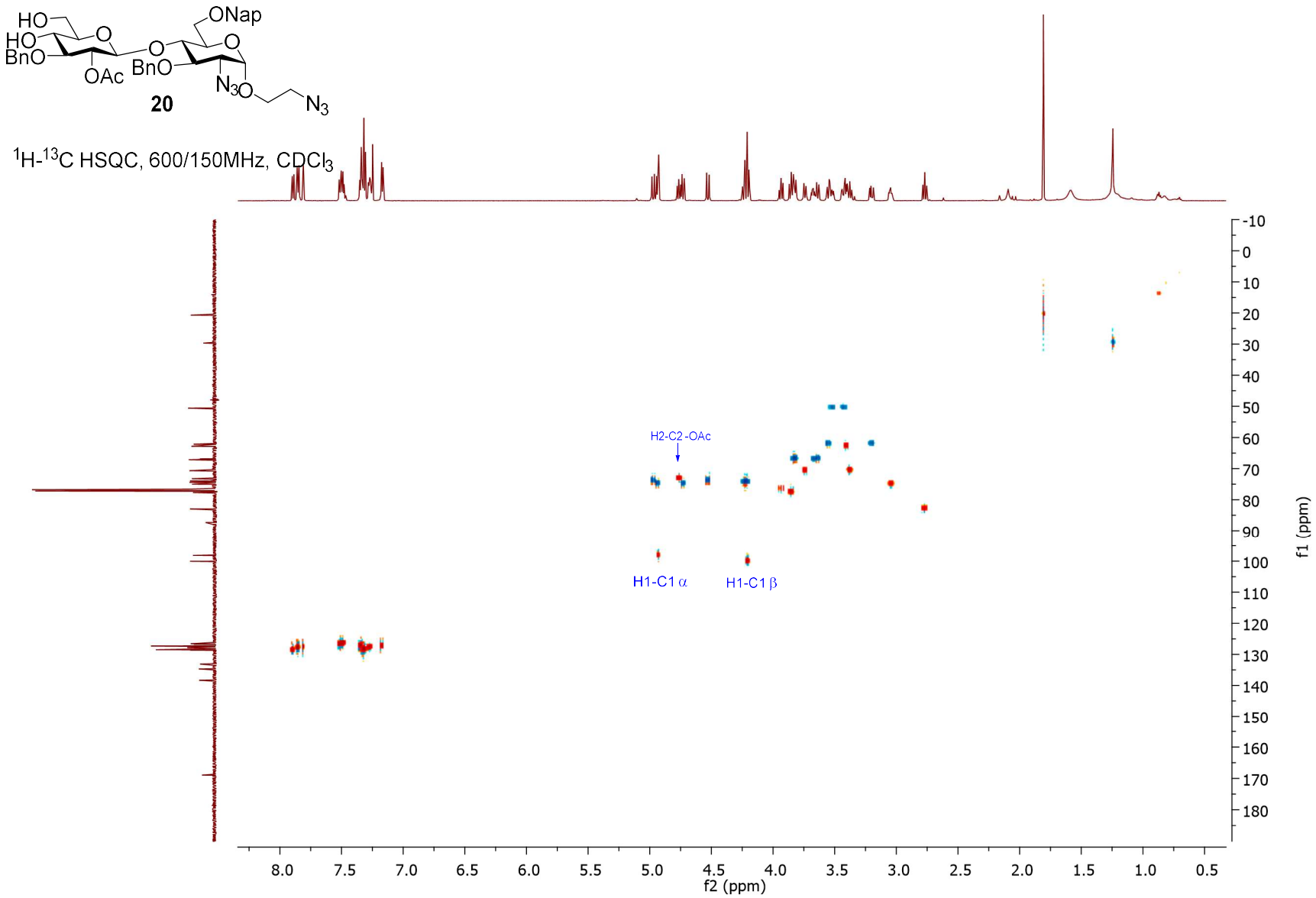
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

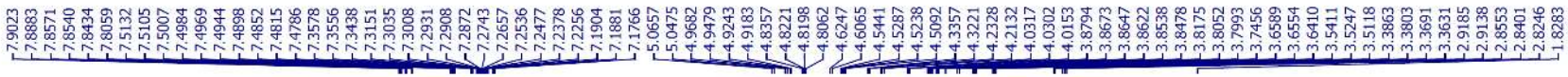


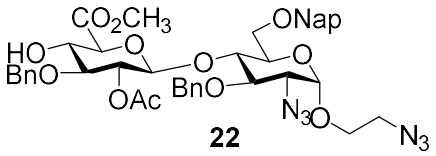

HO
HO
BnO
O
OAc
O
BnO
ONap
O
N3
O
N3
20
1H NMR, 600 MHz, CDCl3
f1 (ppm)


168.9280
138.3509
138.0309
134.7056
133.2376
133.1322
128.6511
128.5089
128.3698
127.9333
127.8630
127.8272
127.7511
127.6652
127.3152
127.1656
126.7385
126.5759
126.4262
100.0388
98.1457
87.3177
82.9553
77.6862
76.5872
75.0170
74.9506
74.4289
73.9711
73.2406
70.7445
70.6698
67.1446
62.8313
62.1572
50.5598
20.6908
HO
HO
BnO
O
OAc
O
BnO
ONap
O
N3
O
N3
20
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


HO
HO
BnO
O
OAc
O
BnO
ONap
O
N3O
N3
20
1H-1H COSY, 600 MHz, CDCl3
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
f2 (ppm)
1
2
3
4
5
6
7
8
f1 (ppm)

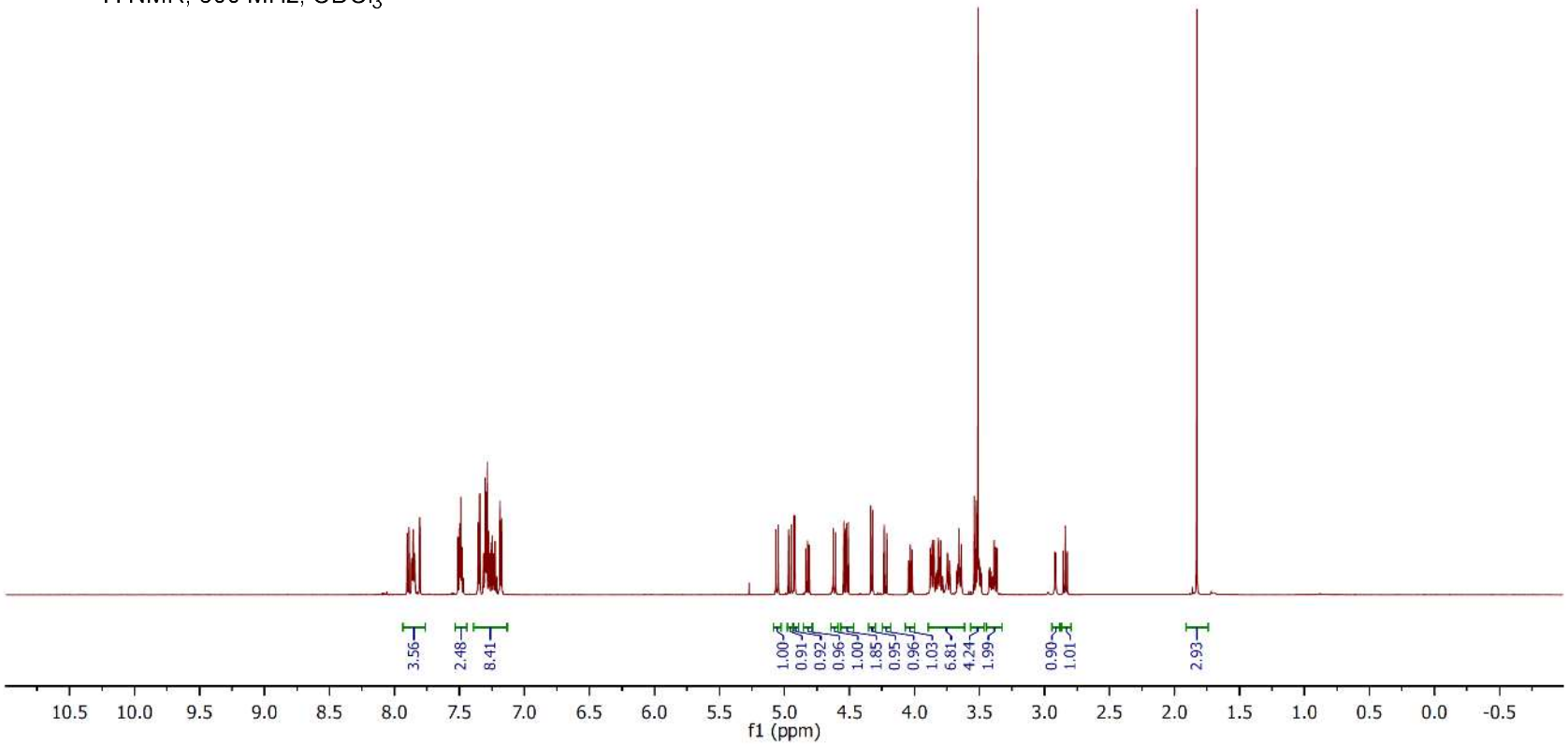

HO
HO
BnO
O
OAc
O
BnO
ONap
O
N3O
N3
20
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2-OAc
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)

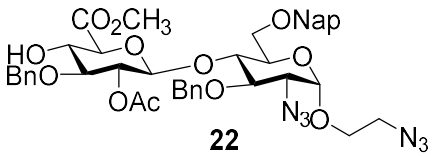




22

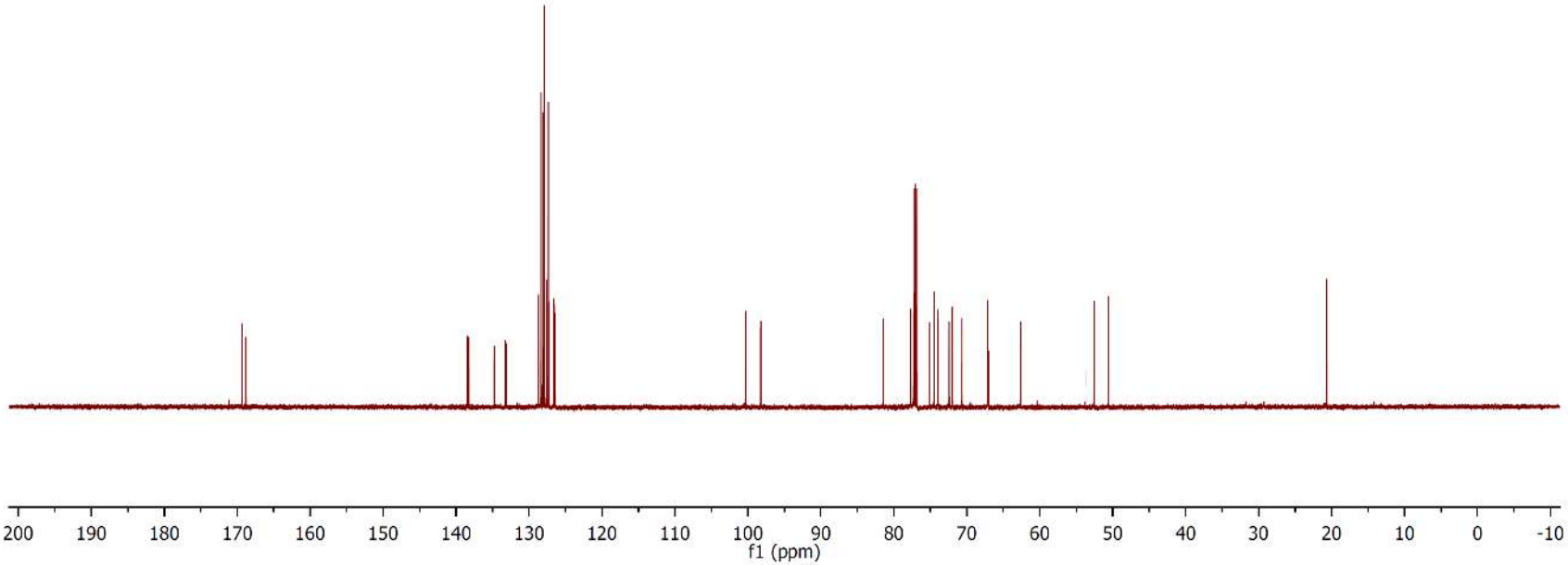
^{1}H NMR, 600 MHz, $CDCl_3$

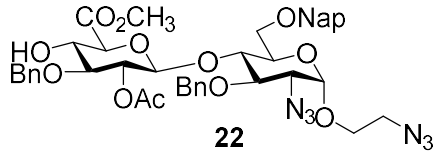




22

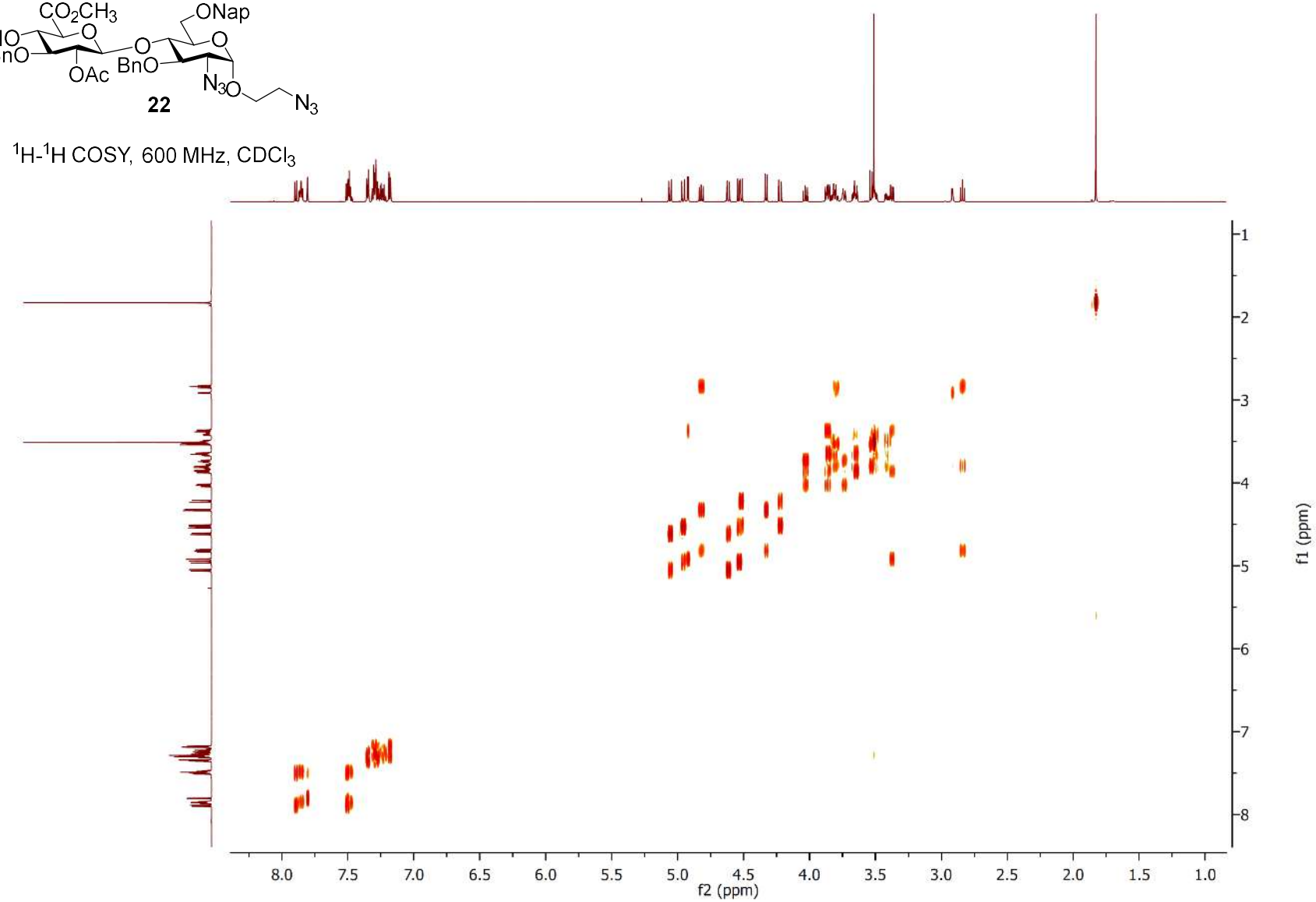
^{13}C NMR, 150 MHz, $CDCl_3$

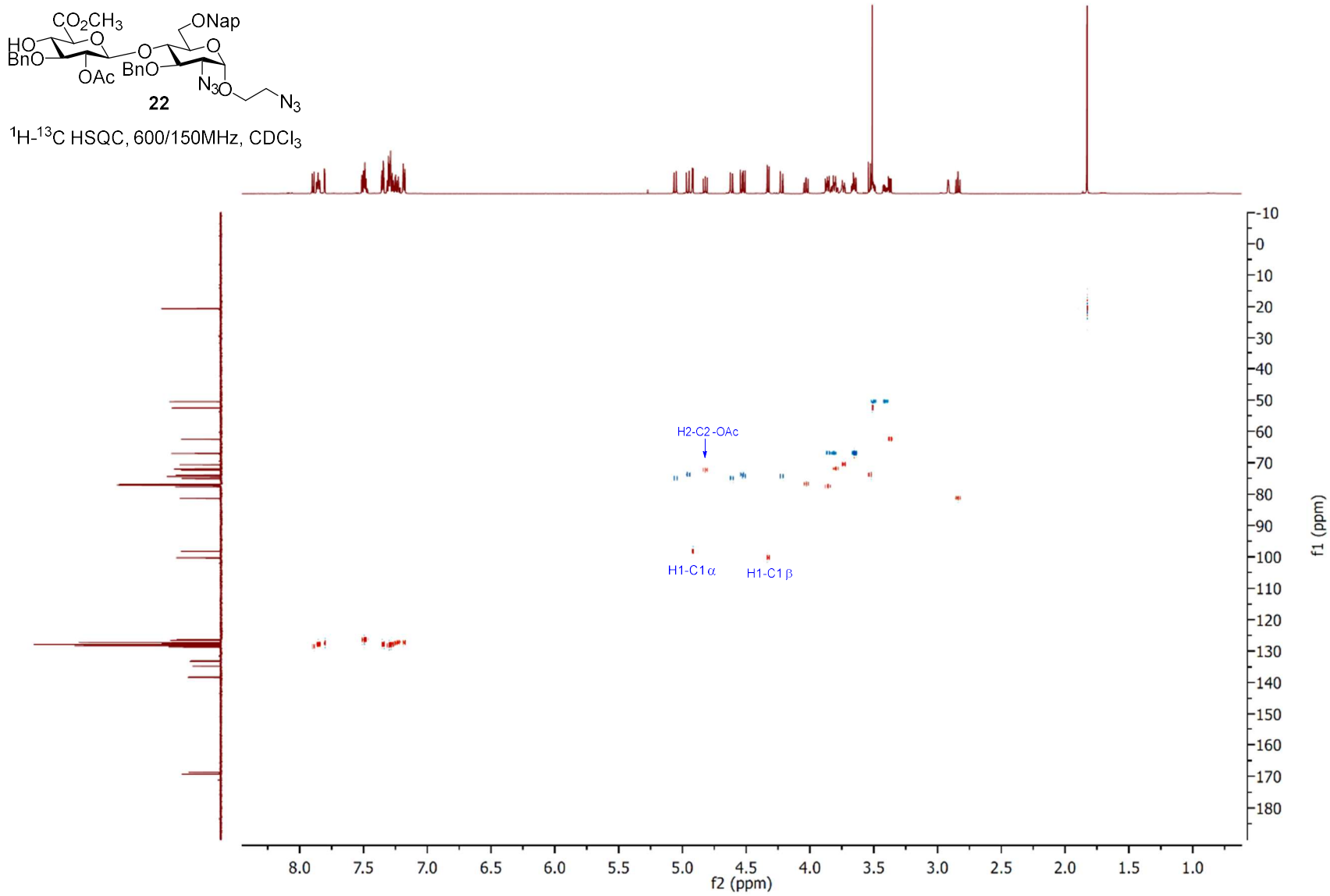


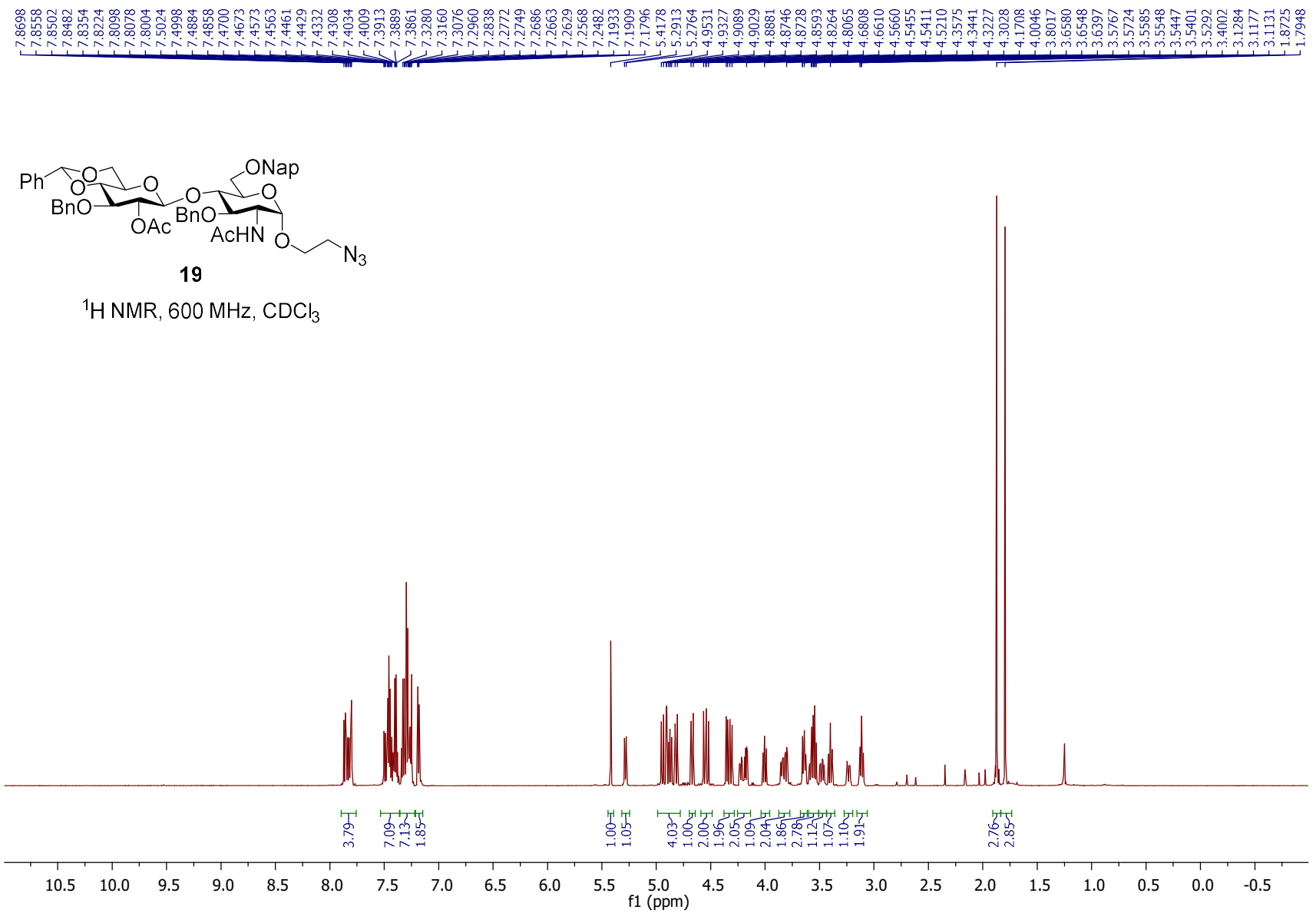


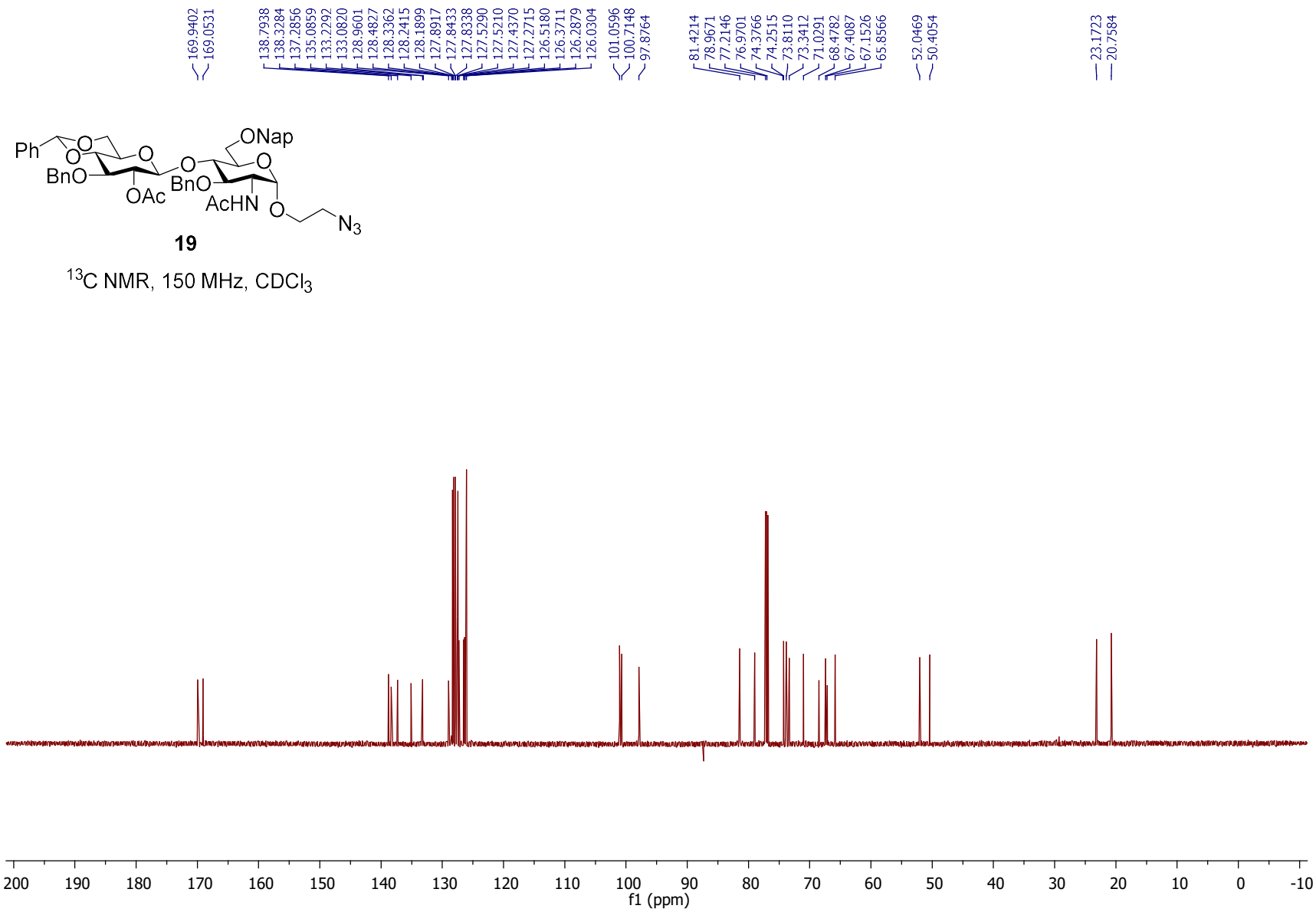
22

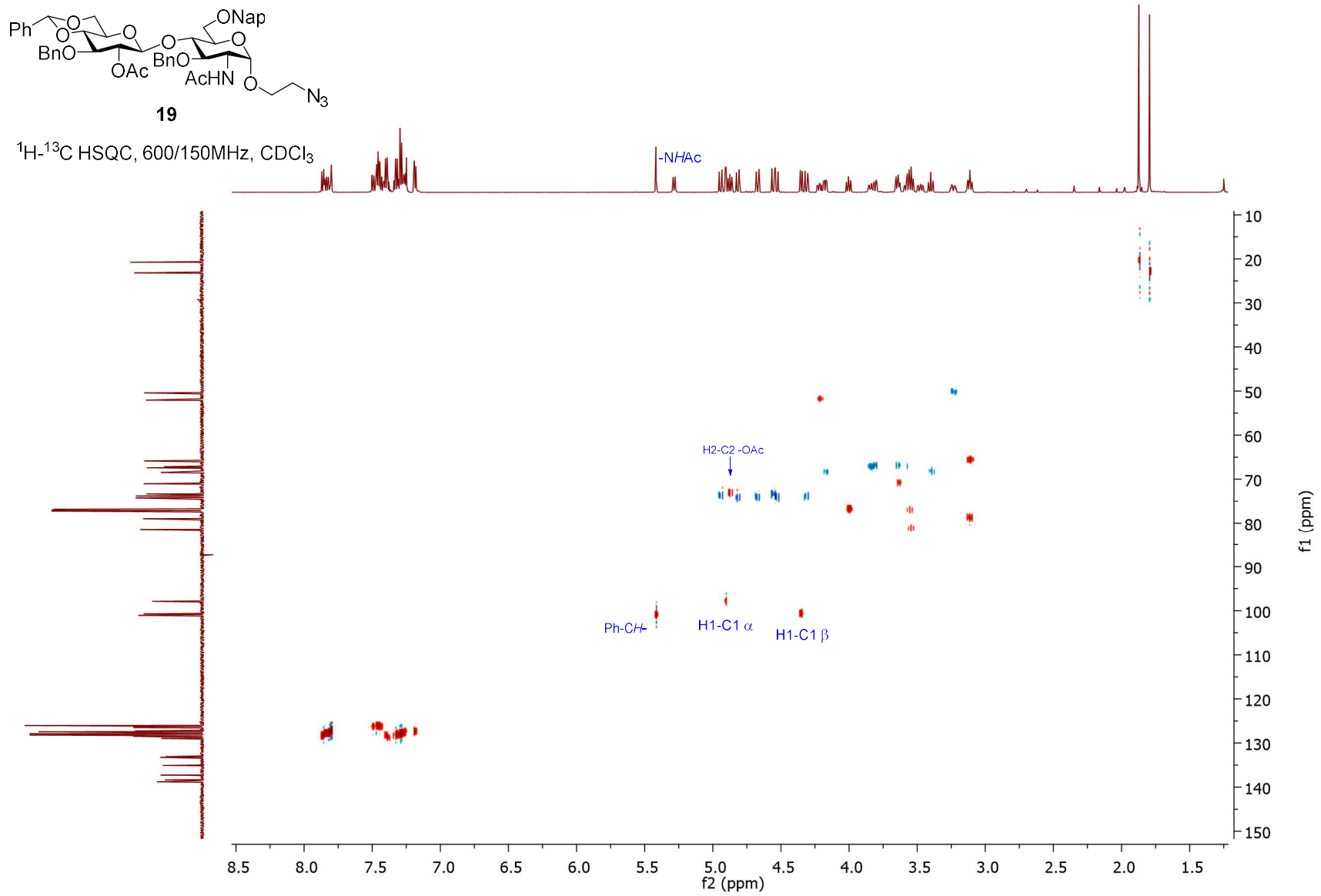
^{1}H-^{1}H COSY, 600 MHz, $CDCl_3$

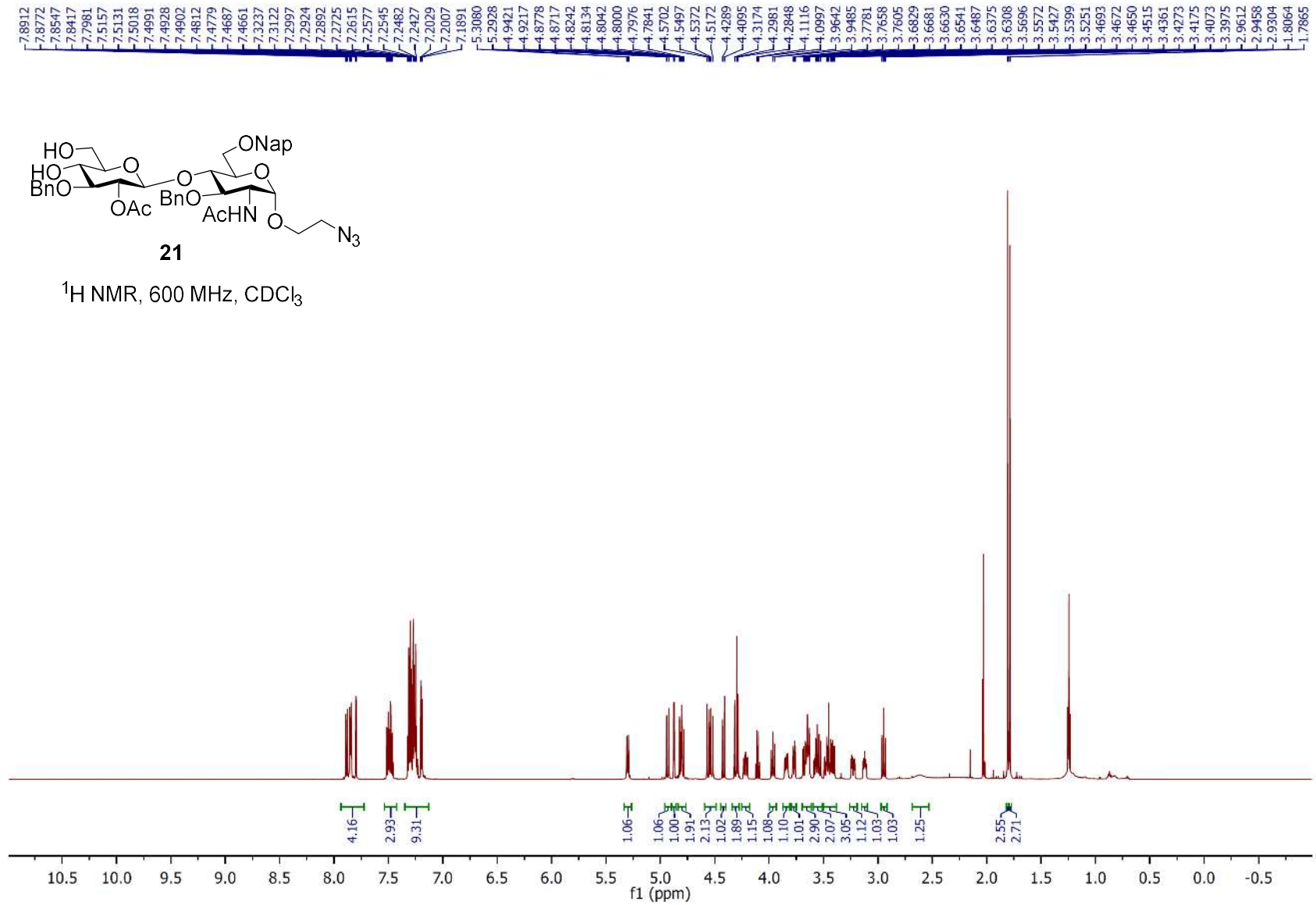



CO2CH3
ONap
HO
BnO
OAc
BnO
N3O
N3
22
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2-OAc
H1-C1α
H1-C1β
f2 (ppm)
f1 (ppm)


7.8698
7.8558
7.8502
7.8482
7.8354
7.8224
7.8098
7.8078
7.8004
7.5024
7.4998
7.4884
7.4858
7.4700
7.4673
7.4573
7.4563
7.4461
7.4429
7.4332
7.4308
7.4034
7.4009
7.3913
7.3889
7.3861
7.3280
7.3160
7.3076
7.2960
7.2838
7.2772
7.2749
7.2686
7.2663
7.2629
7.2568
7.2482
7.1933
7.1909
7.1796
5.4178
5.2913
5.2764
4.9531
4.9327
4.9089
4.9029
4.8881
4.8746
4.8728
4.8593
4.8264
4.8065
4.6808
4.6610
4.5660
4.5455
4.5411
4.5210
4.3575
4.3441
4.3227
4.3028
4.1708
4.0046
3.8017
3.6580
3.6548
3.6397
3.5767
3.5724
3.5585
3.5548
3.5447
3.5401
3.5292
3.4002
3.1284
3.1177
3.1131
1.8725
1.7948
Ph
BnO
OAc
BnO
AcHN
ONap
N3
19
1H NMR, 600 MHz, CDCl3
3.79
7.09
7.13
1.85
1.00
1.05
4.03
1.00
2.00
1.96
2.05
1.09
2.04
1.86
2.78
1.12
1.07
1.10
1.91
2.76
2.85
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)

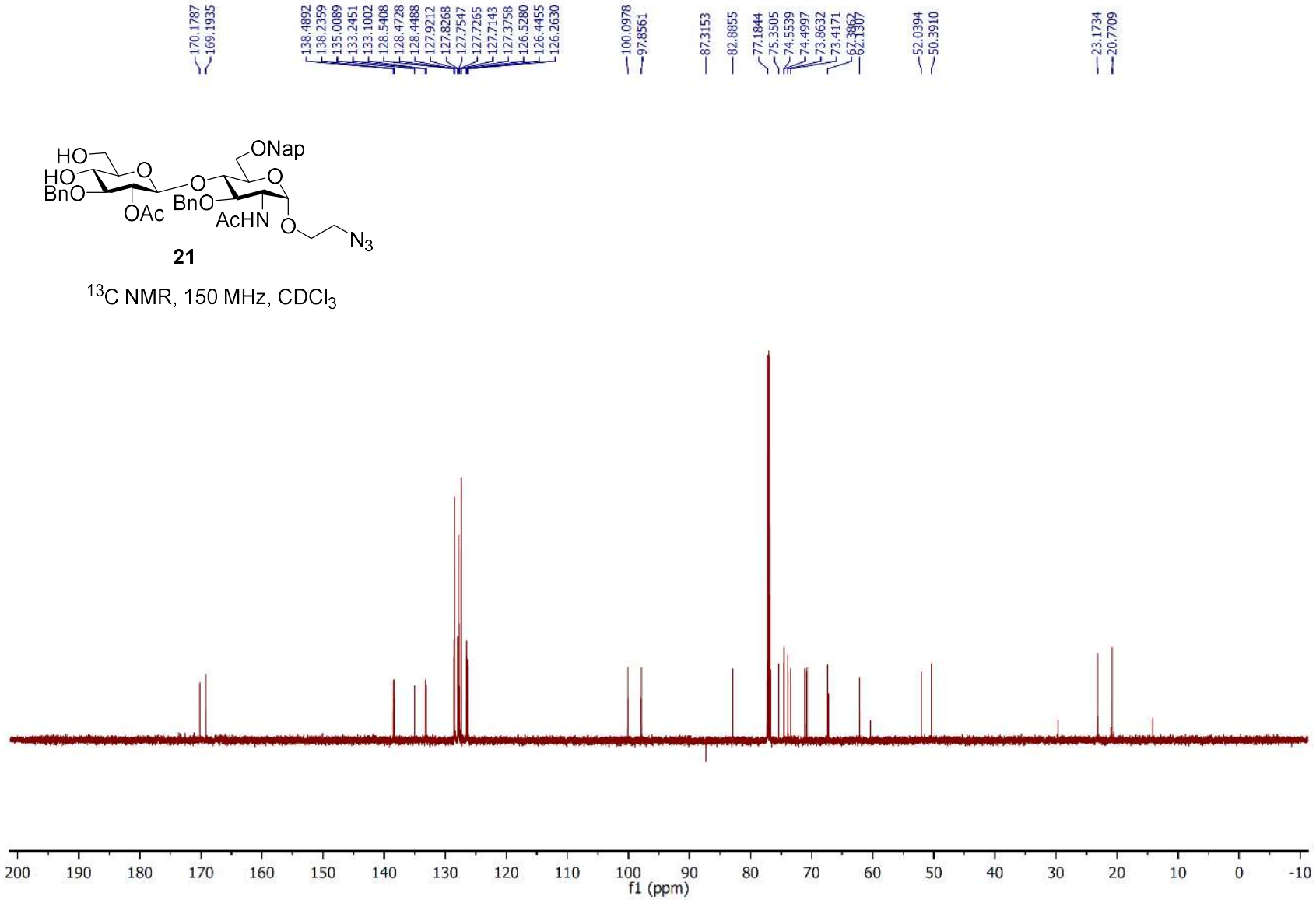

169.9402
169.0531
138.7938
138.3284
137.2856
135.0859
133.2292
133.0820
128.9601
128.4827
128.3362
128.2415
128.1899
127.8917
127.8433
127.8338
127.5290
127.5210
127.4370
127.2715
126.5180
126.3711
126.2879
126.0304
101.0596
100.7148
97.8764
81.4214
78.9671
77.2146
76.9701
74.3766
74.2515
73.8110
73.3412
71.0291
68.4782
67.4087
67.1526
65.8566
52.0469
50.4054
23.1723
20.7584
Ph
BnO
OAc
BnO
AcHN
ONap
N3
19
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

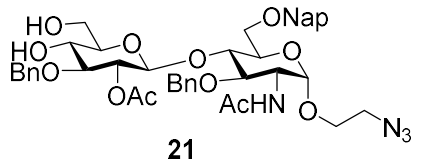

ONap
Ph
BnO
OAc
BnO
AcHN
N3
19
1H-13C HSQC, 600/150MHz, CDCl3
-NHAc
H2-C2 -OAc
Ph-CH-
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)



21

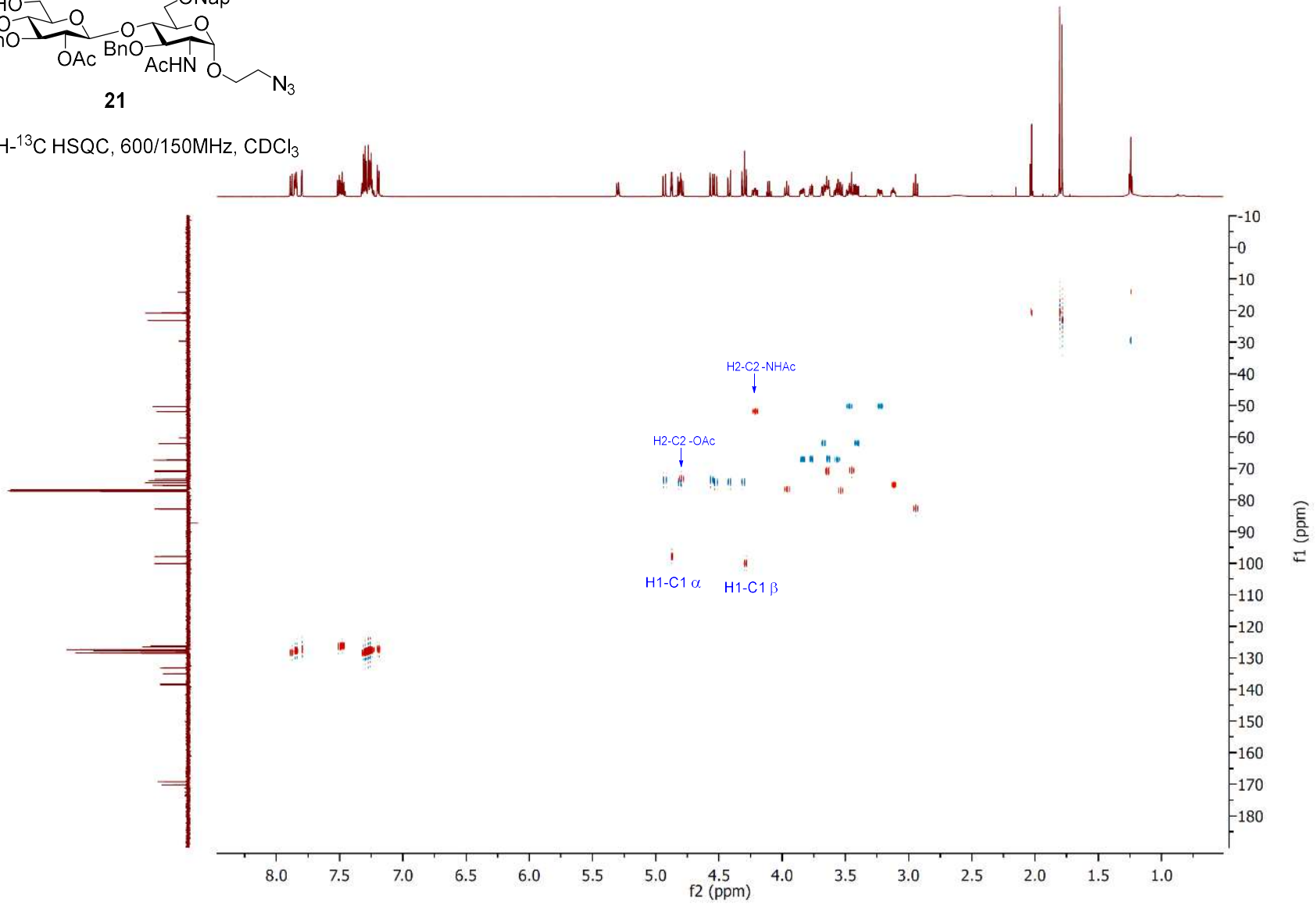
^{1}H NMR, 600 MHz, $CDCl_3$

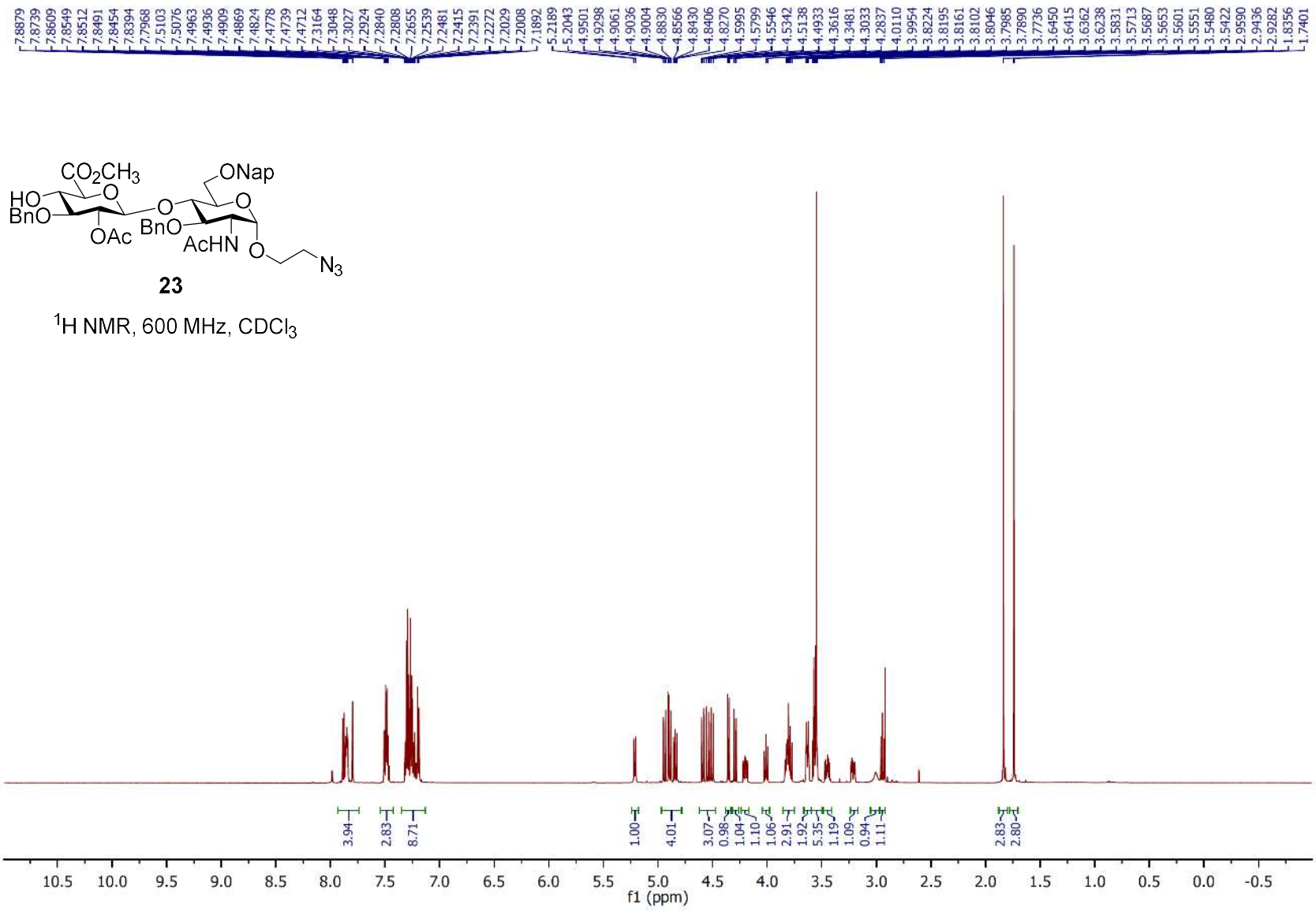

170.1787
169.1935
138.4892
138.2359
135.0089
133.2451
133.1002
128.5408
128.4728
128.4488
127.9212
127.8268
127.7547
127.7265
127.7143
127.3758
126.5280
126.4455
126.2630
100.0978
97.8561
87.3153
82.8855
77.1844
75.3505
74.5539
74.4997
73.8632
73.4171
67.3862
62.1307
52.0394
50.3910
23.1734
20.7709
HO
HO
BnO
O
OAc
O
BnO
AcHN
ONap
O
O
O
N3
21
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)



21

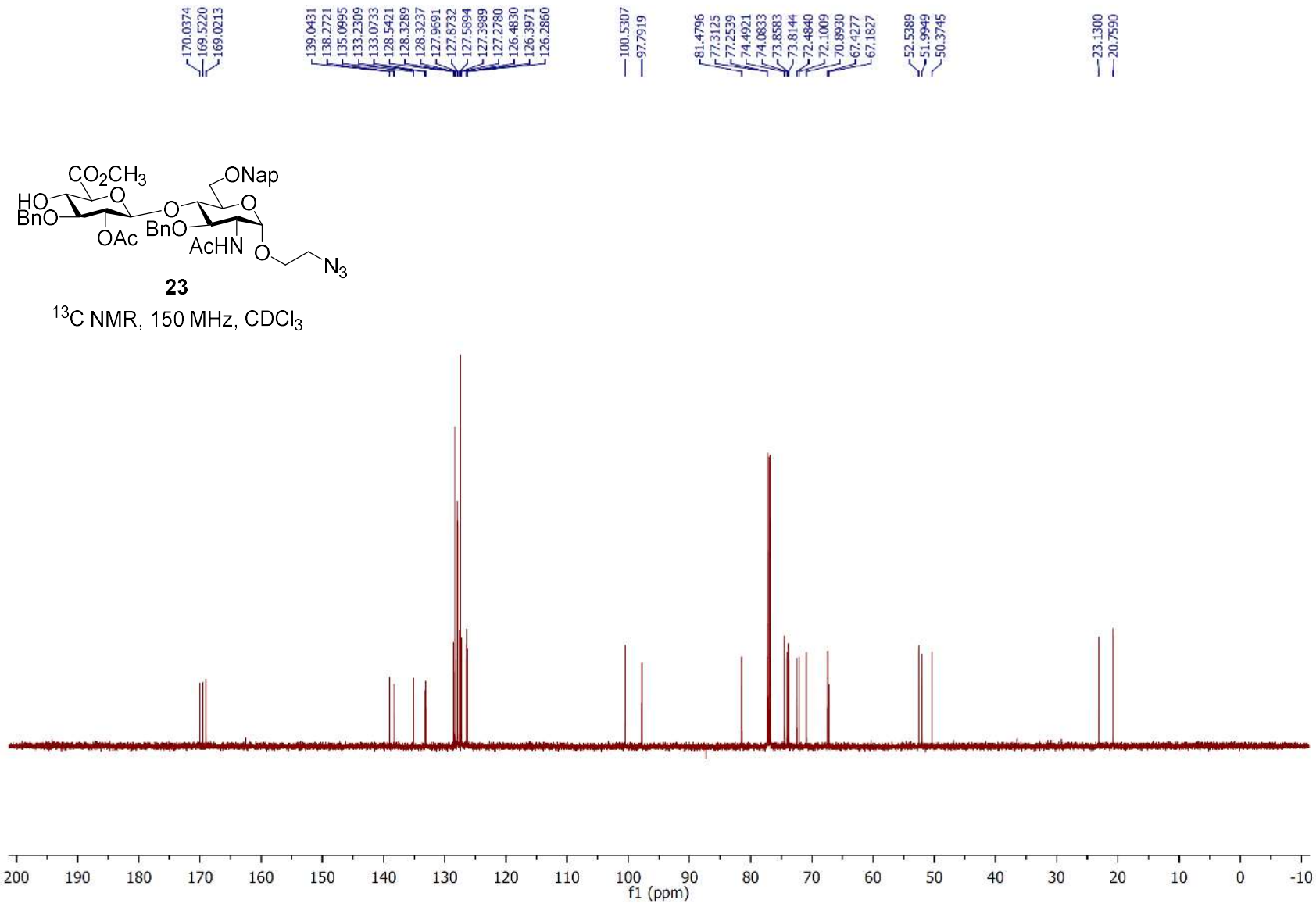
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

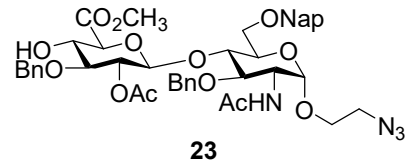



7.8879
7.8739
7.8609
7.8549
7.8512
7.8491
7.8454
7.8394
7.7968
7.5103
7.5076
7.4963
7.4936
7.4909
7.4869
7.4824
7.4778
7.4739
7.4712
7.3164
7.3048
7.3027
7.2924
7.2840
7.2808
7.2655
7.2539
7.2481
7.2415
7.2391
7.2272
7.2029
7.2008
7.1892
5.2189
5.2043
4.9501
4.9298
4.9061
4.9036
4.9004
4.8830
4.8566
4.8430
4.8406
4.8270
4.5995
4.5799
4.5546
4.5342
4.5138
4.4933
4.3616
4.3481
4.3033
4.2837
4.0110
3.9954
3.8224
3.8195
3.8161
3.8102
3.8046
3.7985
3.7890
3.7736
3.6450
3.6415
3.6362
3.6238
3.5831
3.5713
3.5687
3.5653
3.5601
3.5551
3.5480
3.5422
2.9590
2.9436
2.9282
1.8356
1.7401
CO2CH3
ONap
HO
BnO
OAc
BnO
AcHN
N3
23
1H NMR, 600 MHz, CDCl3
3.94
2.83
8.71
1.00
4.01
3.07
0.98
1.04
1.10
1.06
2.91
1.92
5.35
1.19
1.09
0.94
1.11
2.83
2.80
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)

23

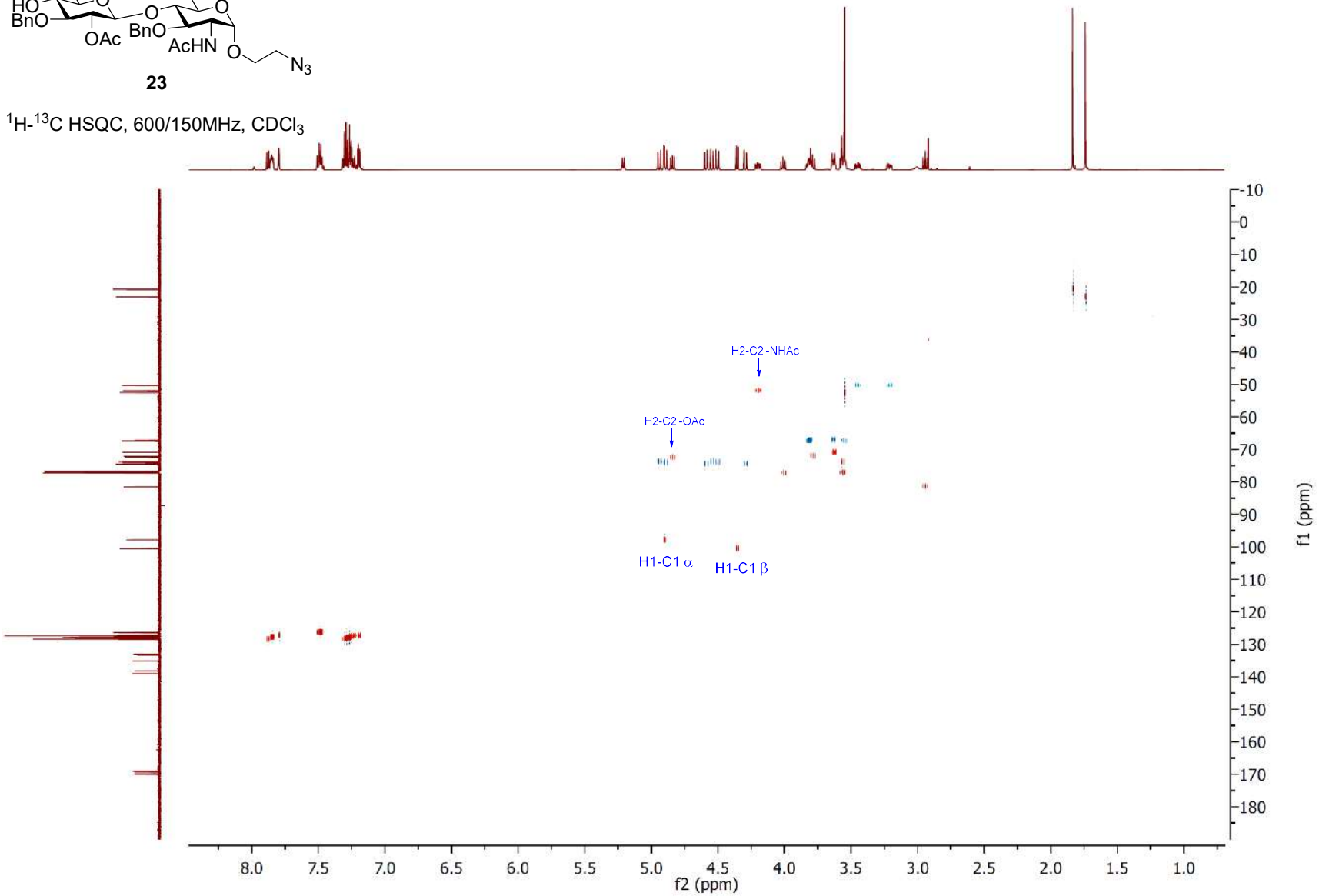
^{13}C NMR, 150 MHz, $CDCl_3$

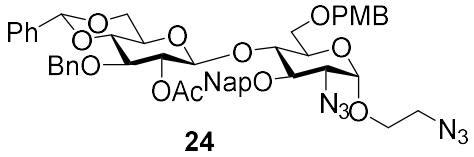




23

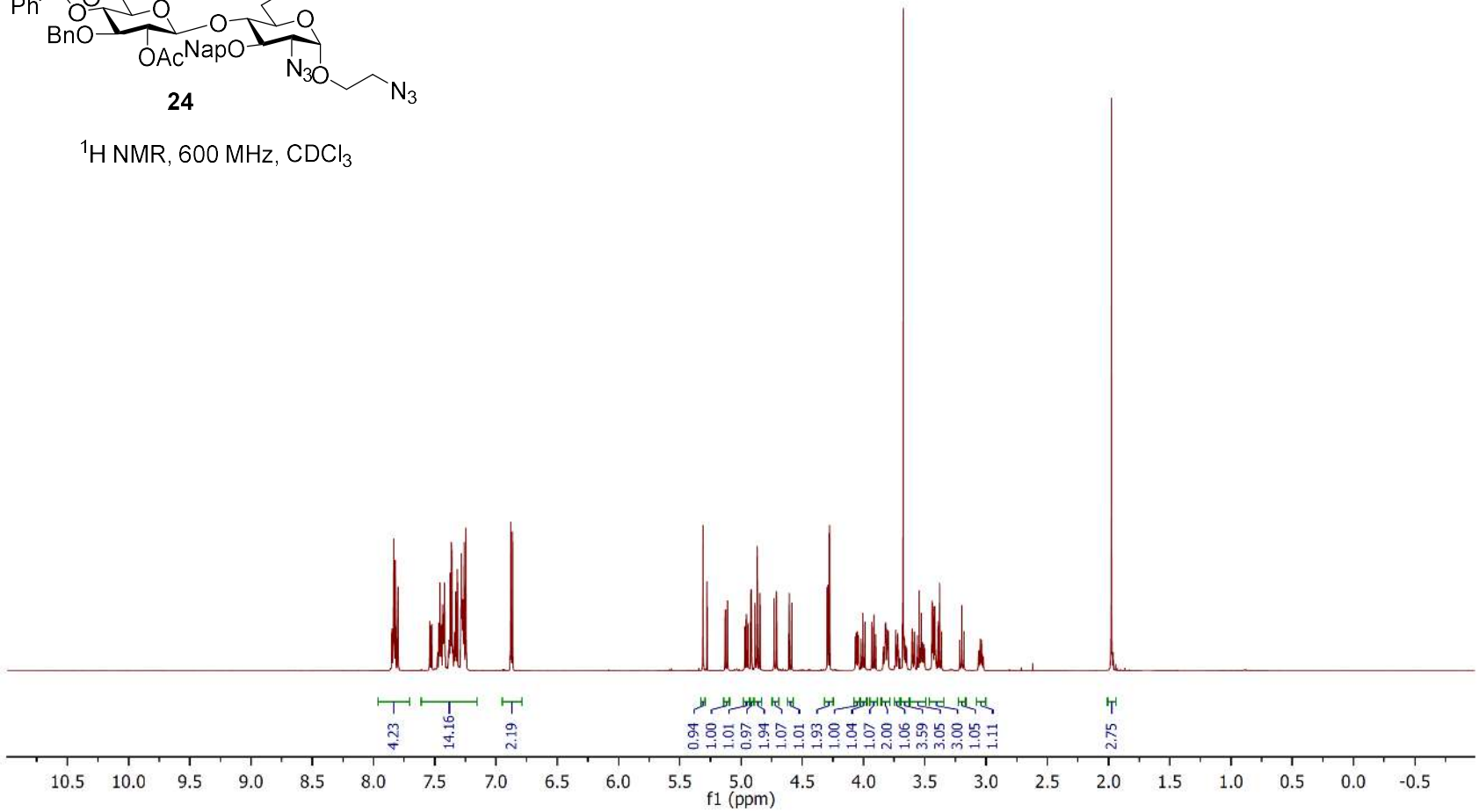
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

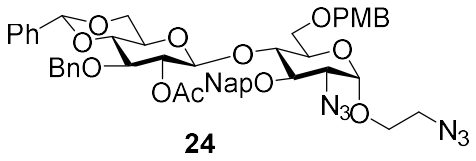




24

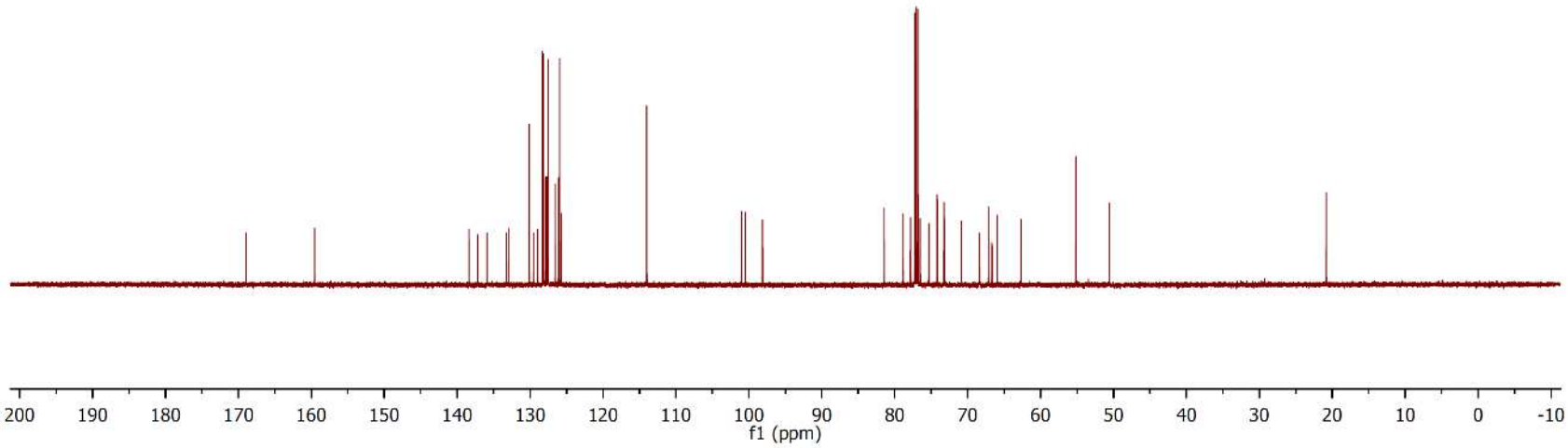
^{1}H NMR, 600 MHz, $CDCl_3$

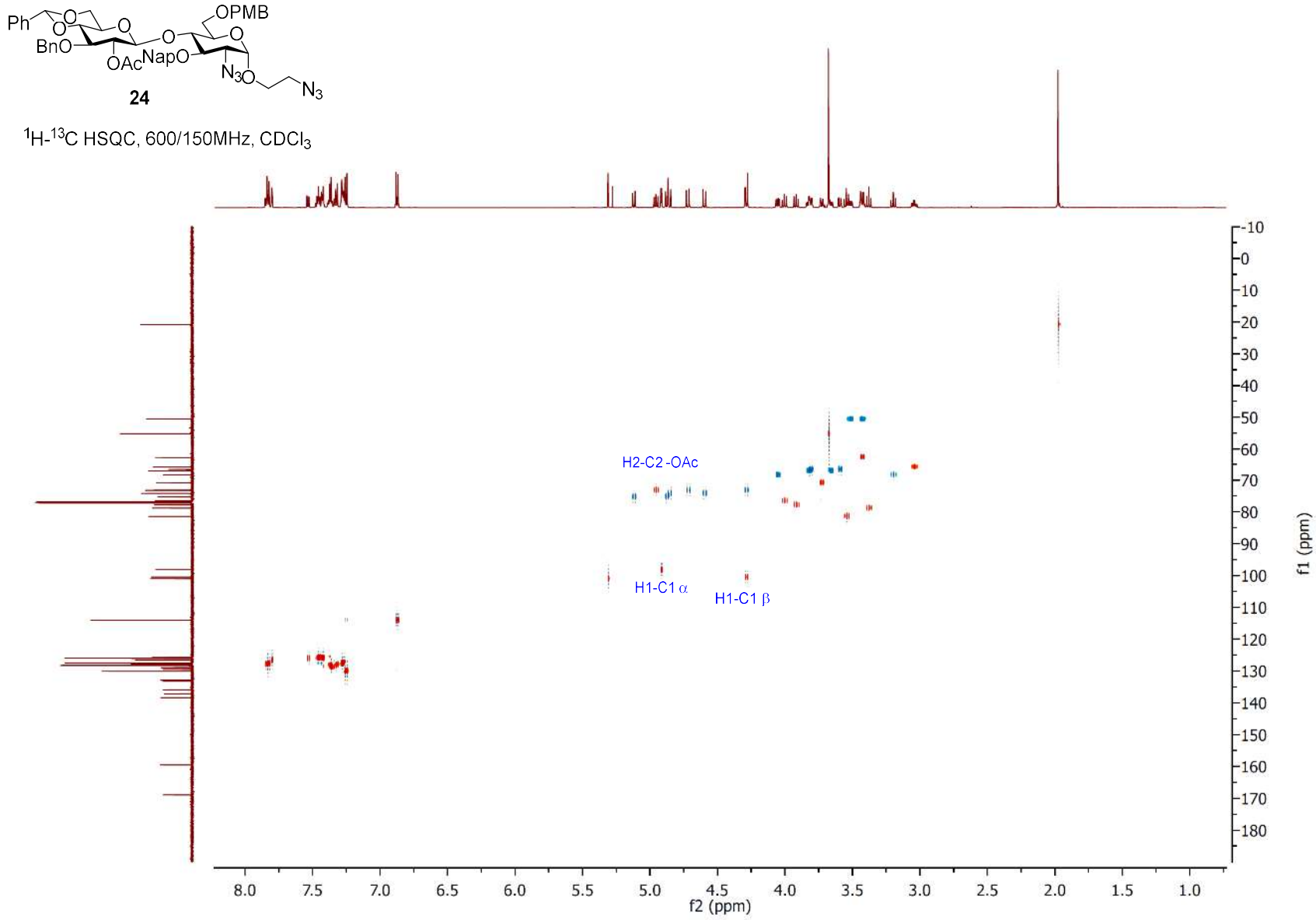


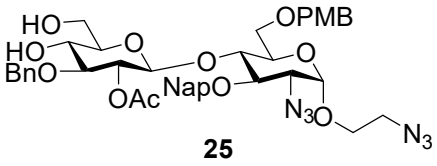


24

^{13}C NMR, 150 MHz, $CDCl_3$

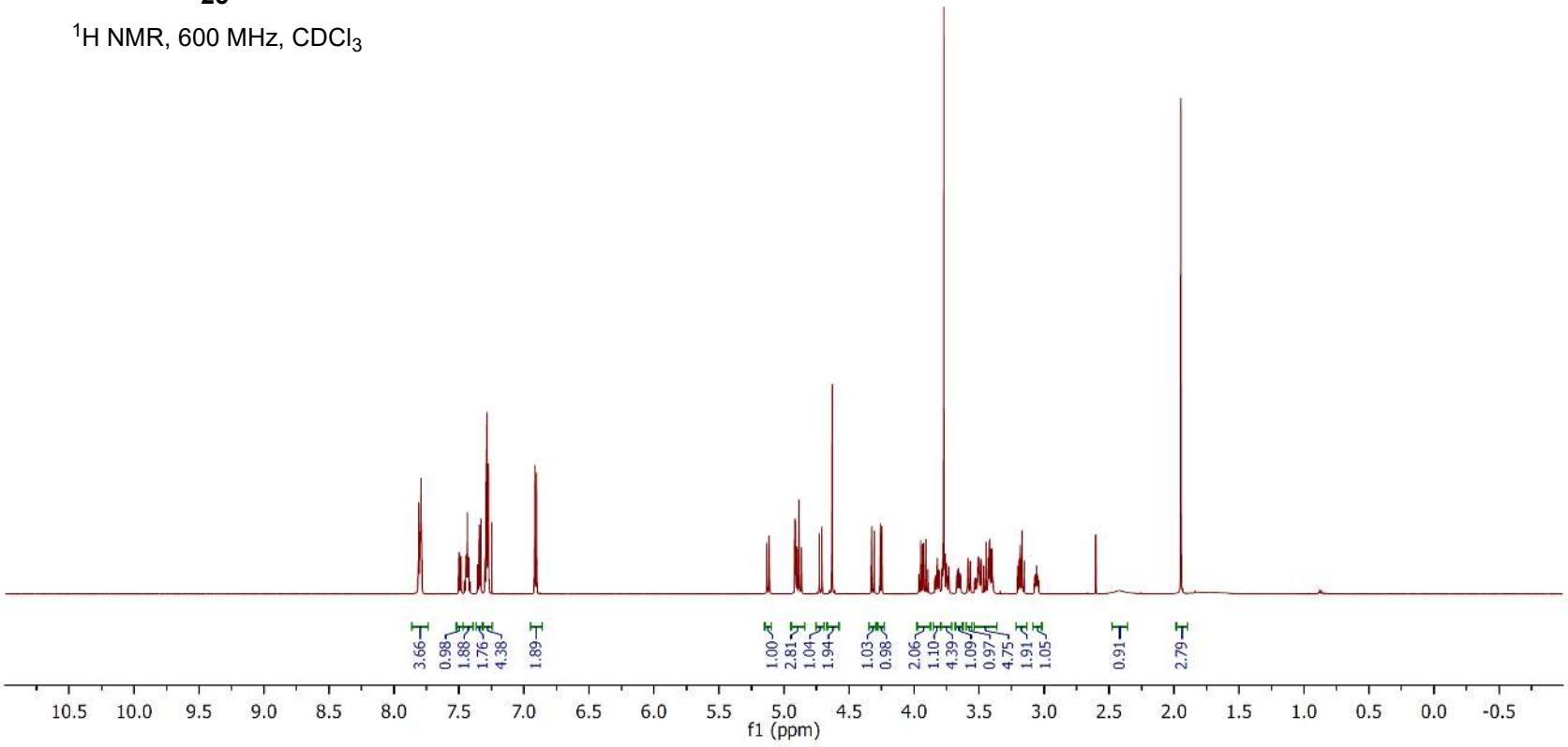



Ph
BnO
OAc
NapO
OPMB
N3O
N3
24
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2 -OAc
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)



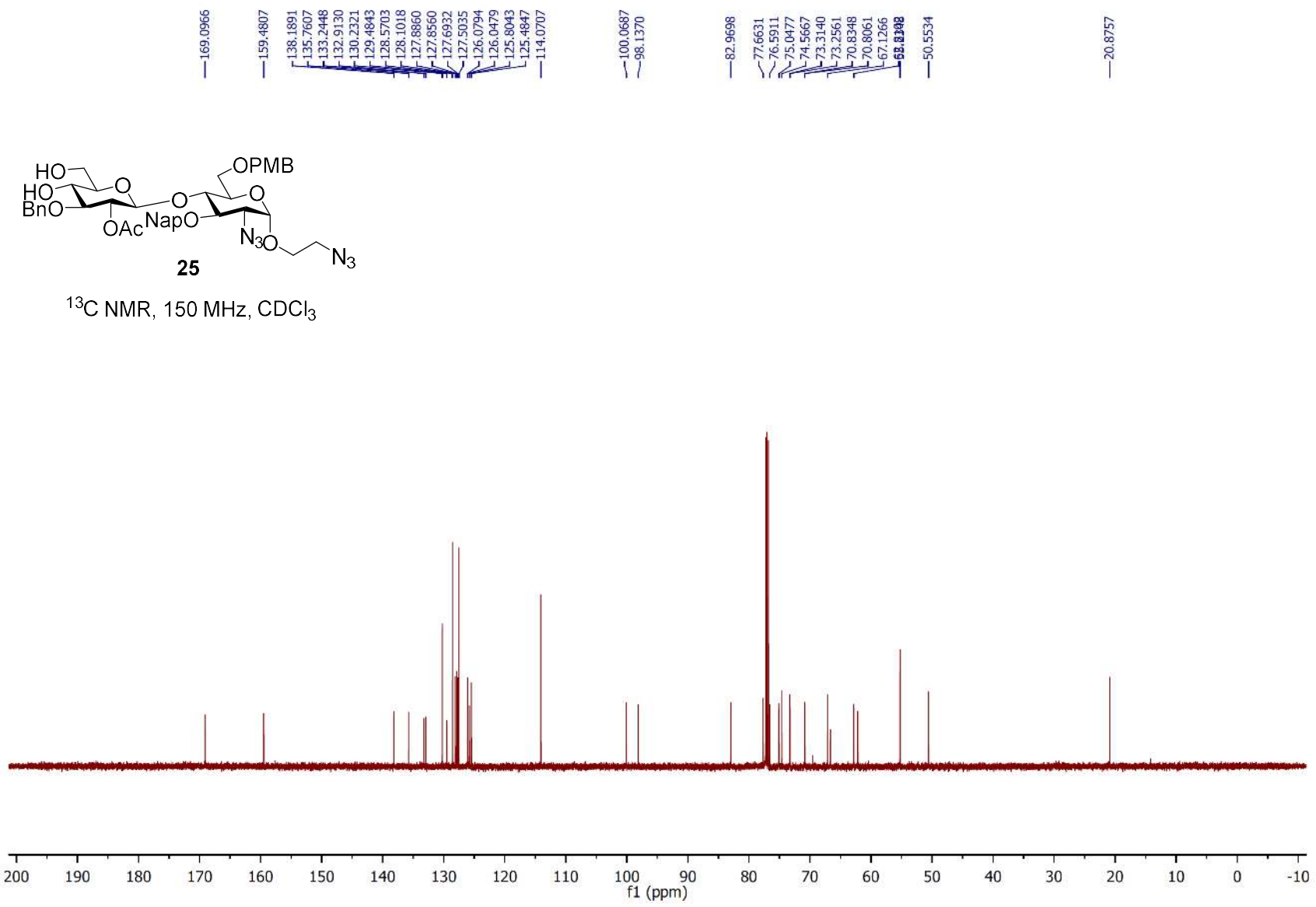
25

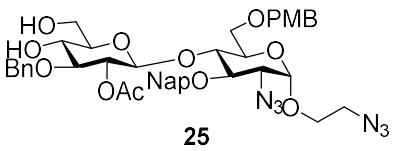
^{1}H NMR, 600 MHz, $CDCl_3$



25

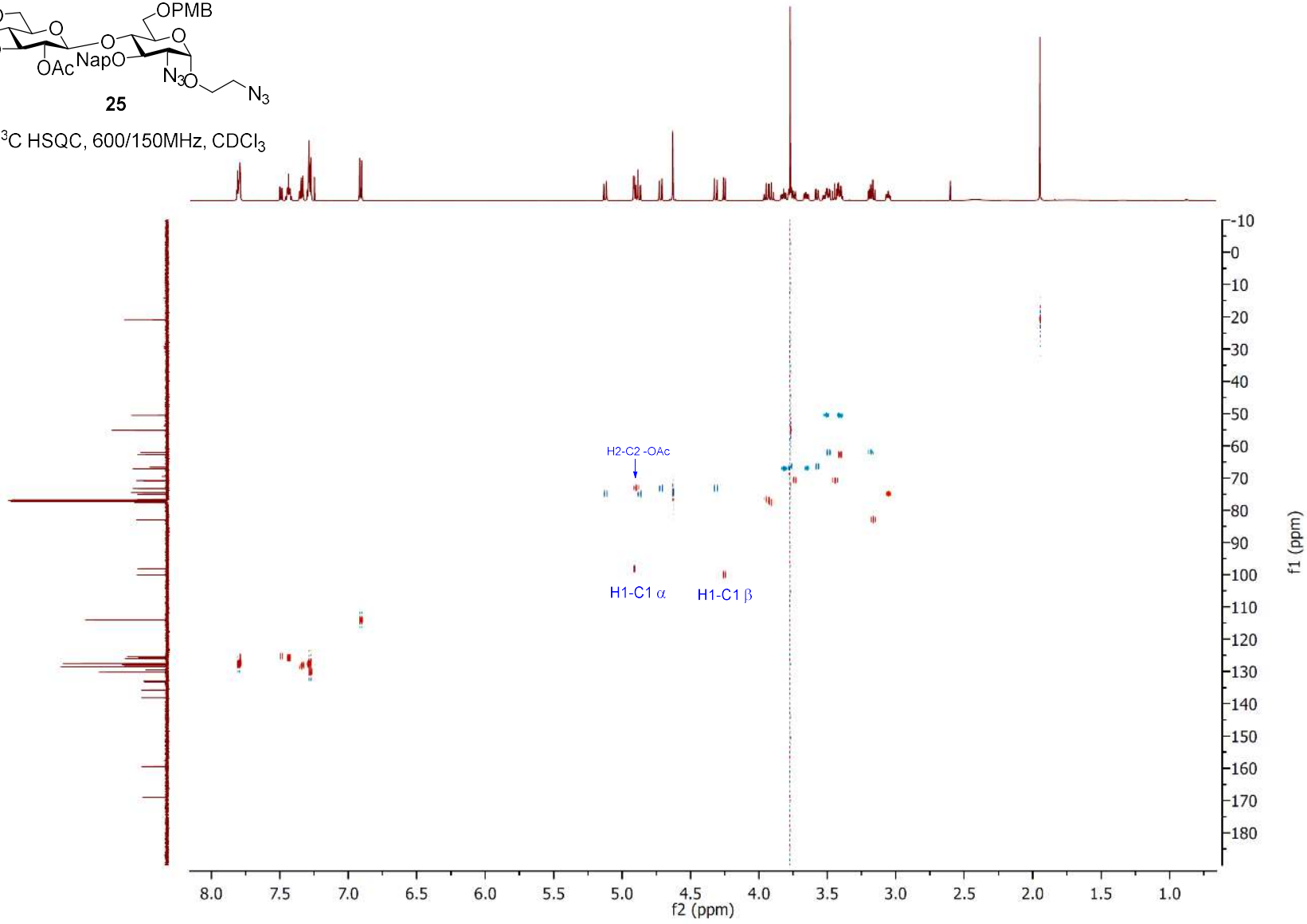
^{13}C NMR, 150 MHz, $CDCl_3$

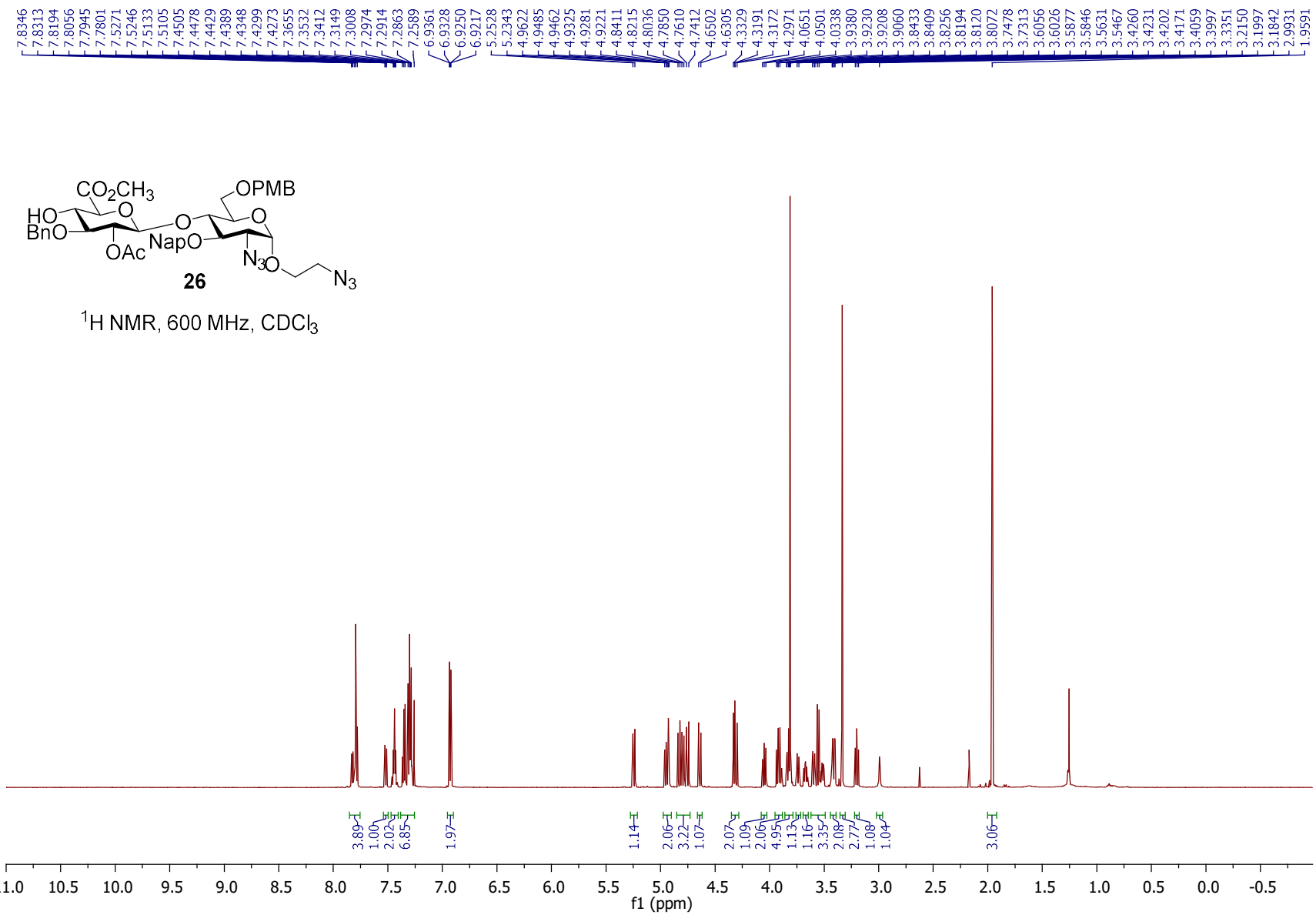


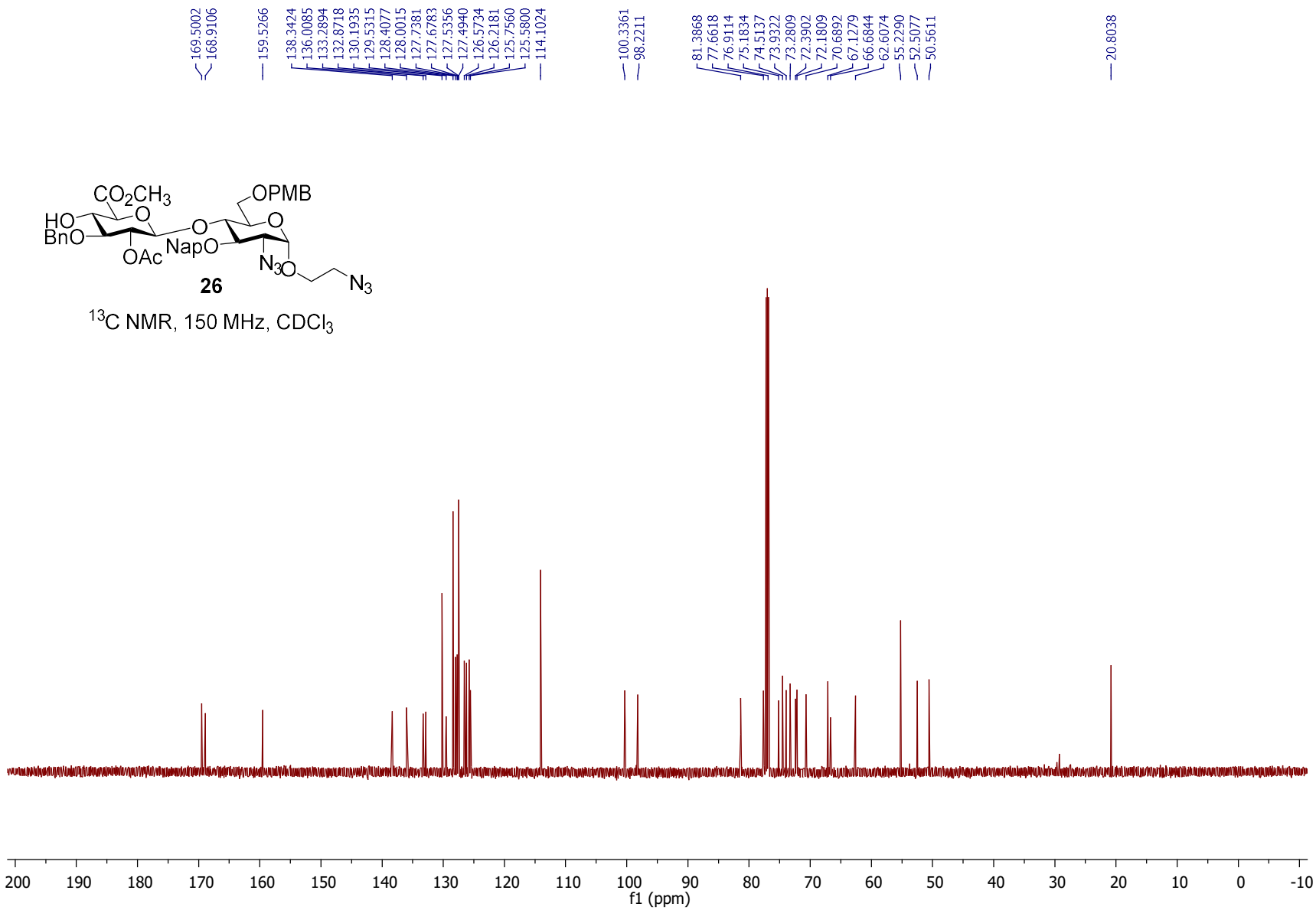


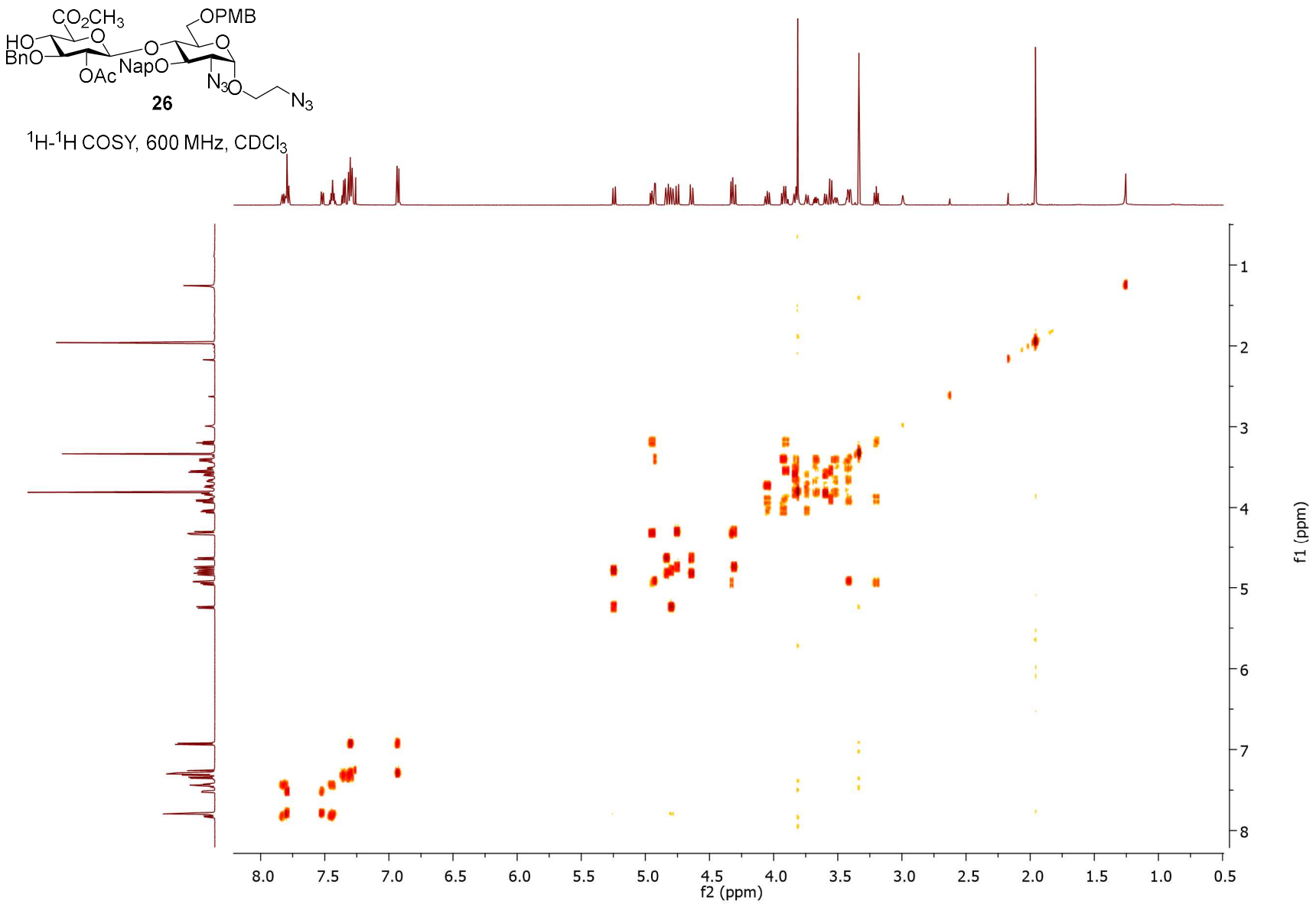
25

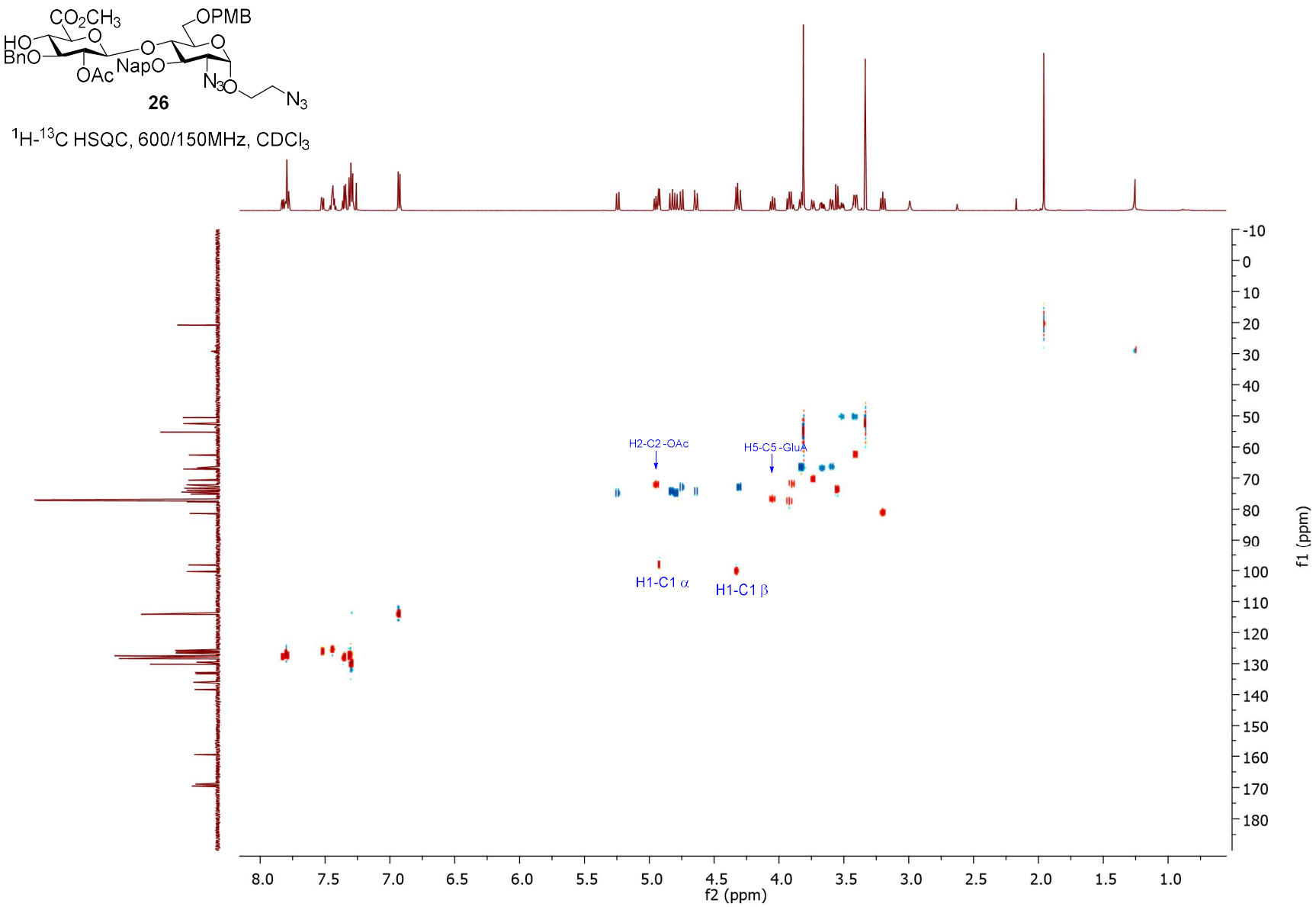
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

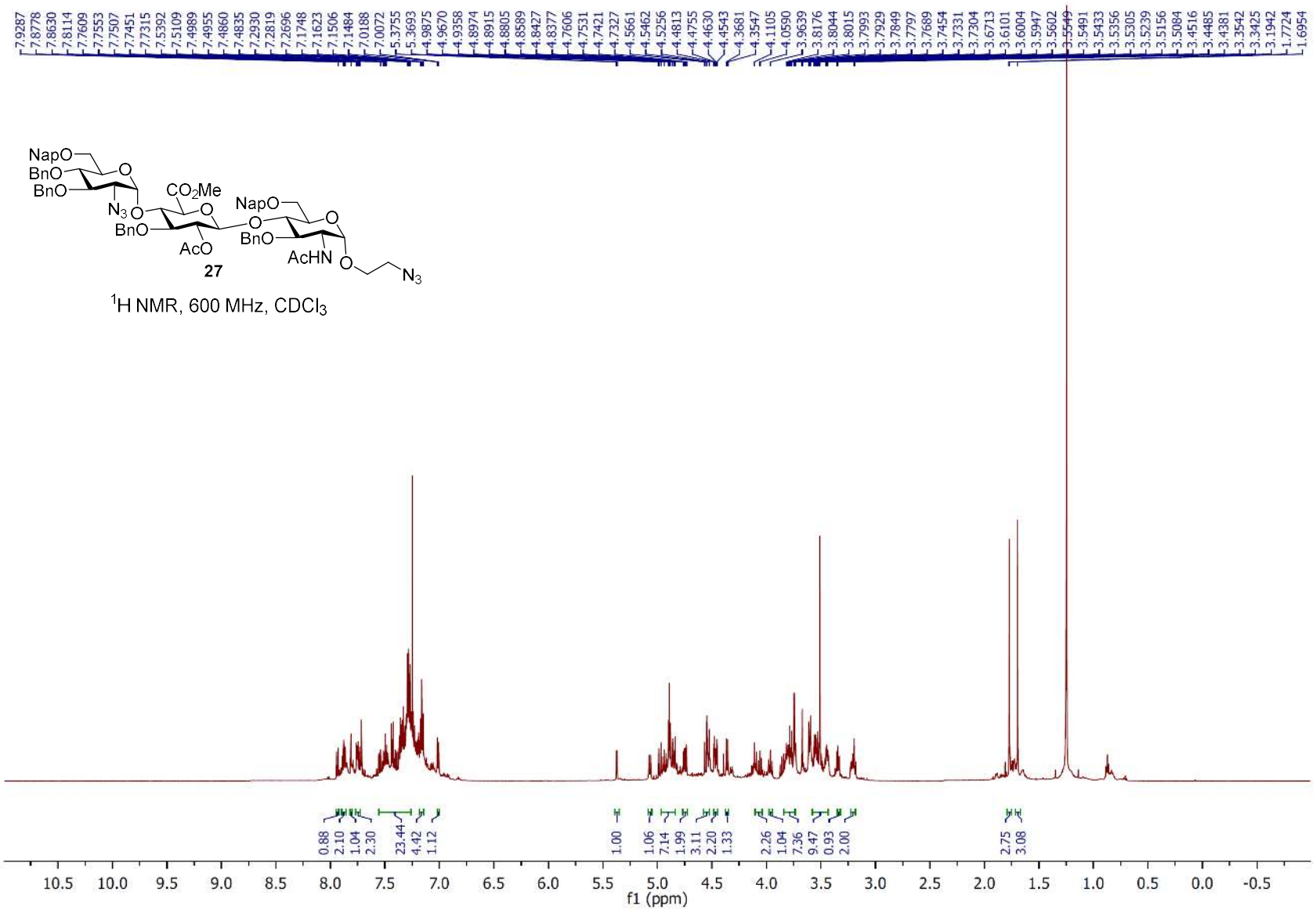


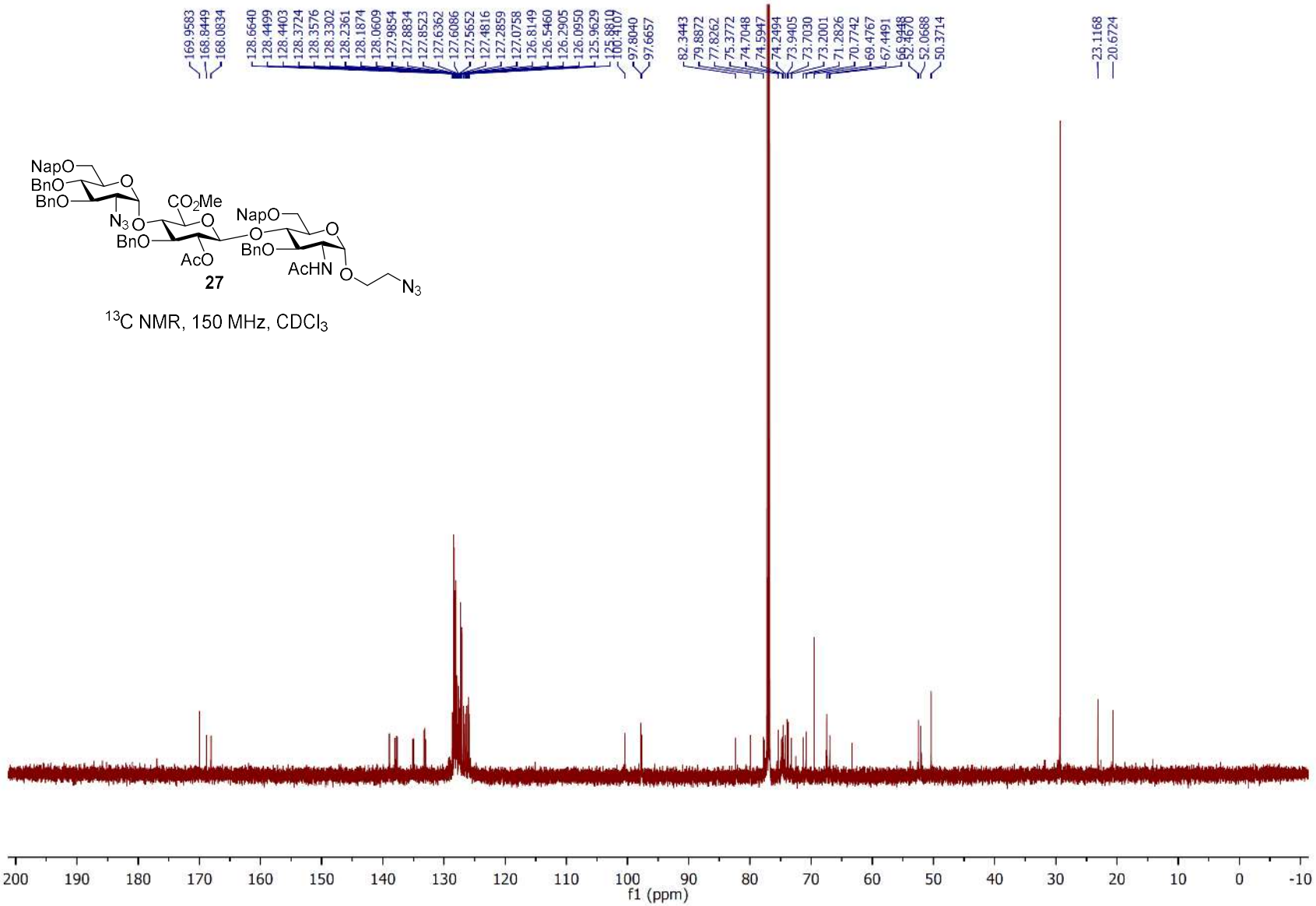

7.8346
7.8313
7.8194
7.8056
7.7945
7.7801
7.5271
7.5246
7.5133
7.5105
7.4505
7.4478
7.4429
7.4389
7.4348
7.4299
7.4273
7.3655
7.3532
7.3412
7.3149
7.3008
7.2974
7.2914
7.2863
7.2589
6.9361
6.9328
6.9250
6.9217
5.2528
5.2343
4.9622
4.9485
4.9462
4.9325
4.9281
4.9221
4.8411
4.8215
4.8036
4.7850
4.7610
4.7412
4.6502
4.6305
4.3329
4.3191
4.3172
4.2971
4.0651
4.0501
4.0338
3.9380
3.9230
3.9208
3.9060
3.8433
3.8409
3.8256
3.8194
3.8120
3.8072
3.7478
3.7313
3.6056
3.6026
3.5877
3.5846
3.5631
3.5467
3.4260
3.4231
3.4202
3.4171
3.4059
3.3997
3.3351
3.2150
3.1997
3.1842
2.9931
1.9591
CO2CH3
OPMB
HO
BnO
OAc
NapO
N3
O
N3
26
1H NMR, 600 MHz, CDCl3
3.89
1.00
2.02
6.85
1.97
1.14
2.06
3.22
1.07
2.07
1.09
2.06
4.95
1.13
1.16
3.35
2.08
2.77
1.08
1.04
3.06
11.0
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)

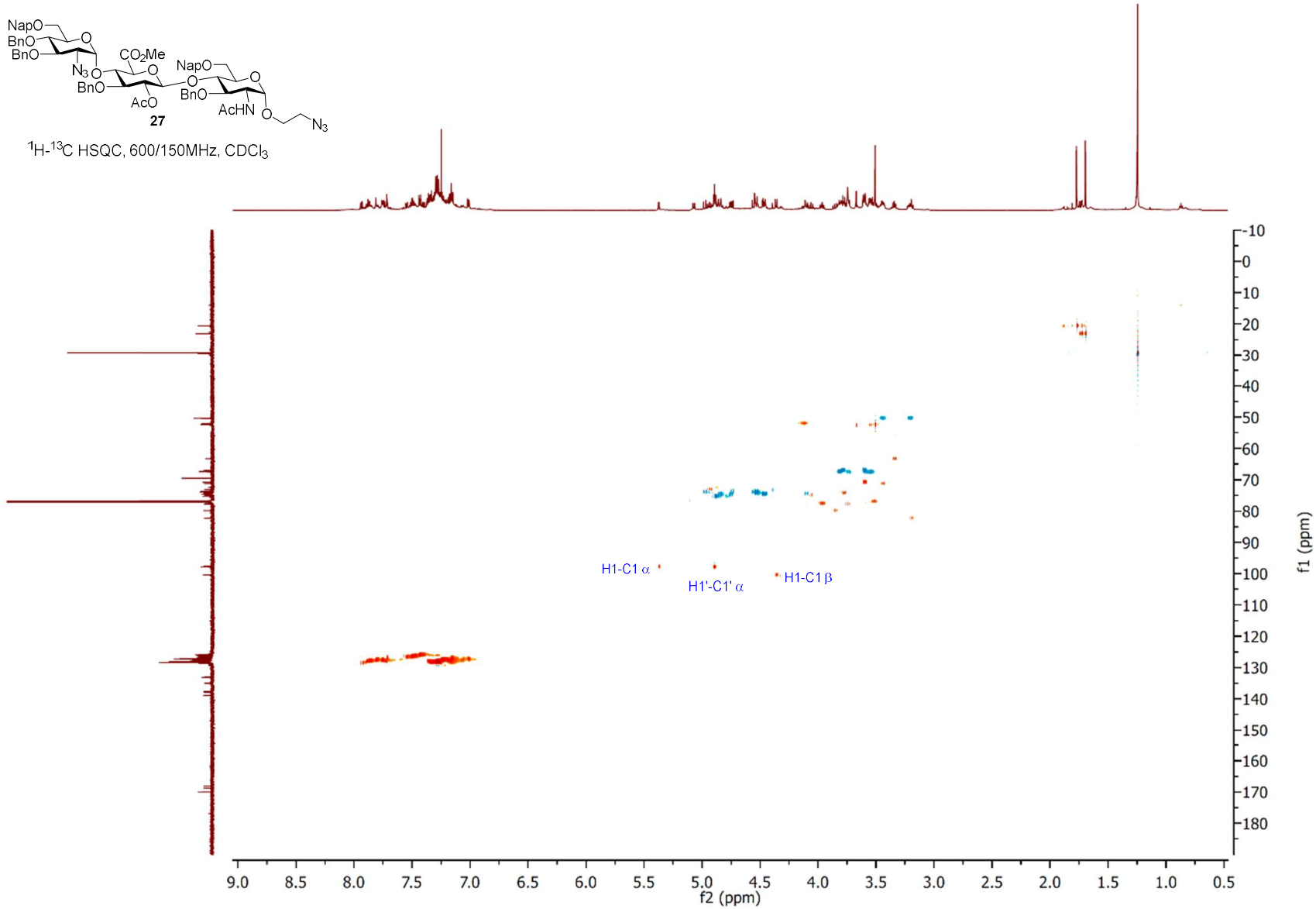

169.5002
168.9106
159.5266
138.3424
136.0085
133.2894
132.8718
130.1935
129.5315
128.4077
128.0015
127.7381
127.6783
127.5356
127.4940
126.5734
126.2181
125.7560
125.5800
114.1024
100.3361
98.2211
81.3868
77.6618
76.9114
75.1834
74.5137
73.9322
73.2809
72.3902
72.1809
70.6892
67.1279
66.6844
62.6074
55.2290
52.5077
50.5611
20.8038
CO2CH3
OPMB
HO
BnO
OAc
NapO
N3
N3
26
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

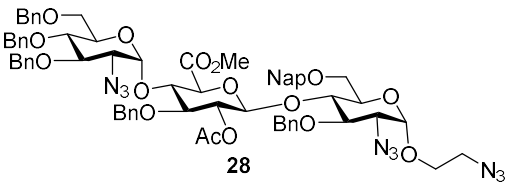

CO2CH3
OPMB
HO
BnO
O
NapO
OAc
N3O
N3
26
1H-1H COSY, 600 MHz, CDCl3
f2 (ppm)
f1 (ppm)


CO2CH3
OPMB
HO
BnO
OAc
NapO
N3
N3
26
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2-OAc
H5-C5-GluA
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)


NapO
BnO
BnO
N3
CO2Me
BnO
AcO
NapO
BnO
AcHN
N3
27
1H NMR, 600 MHz, CDCl3
f1 (ppm)

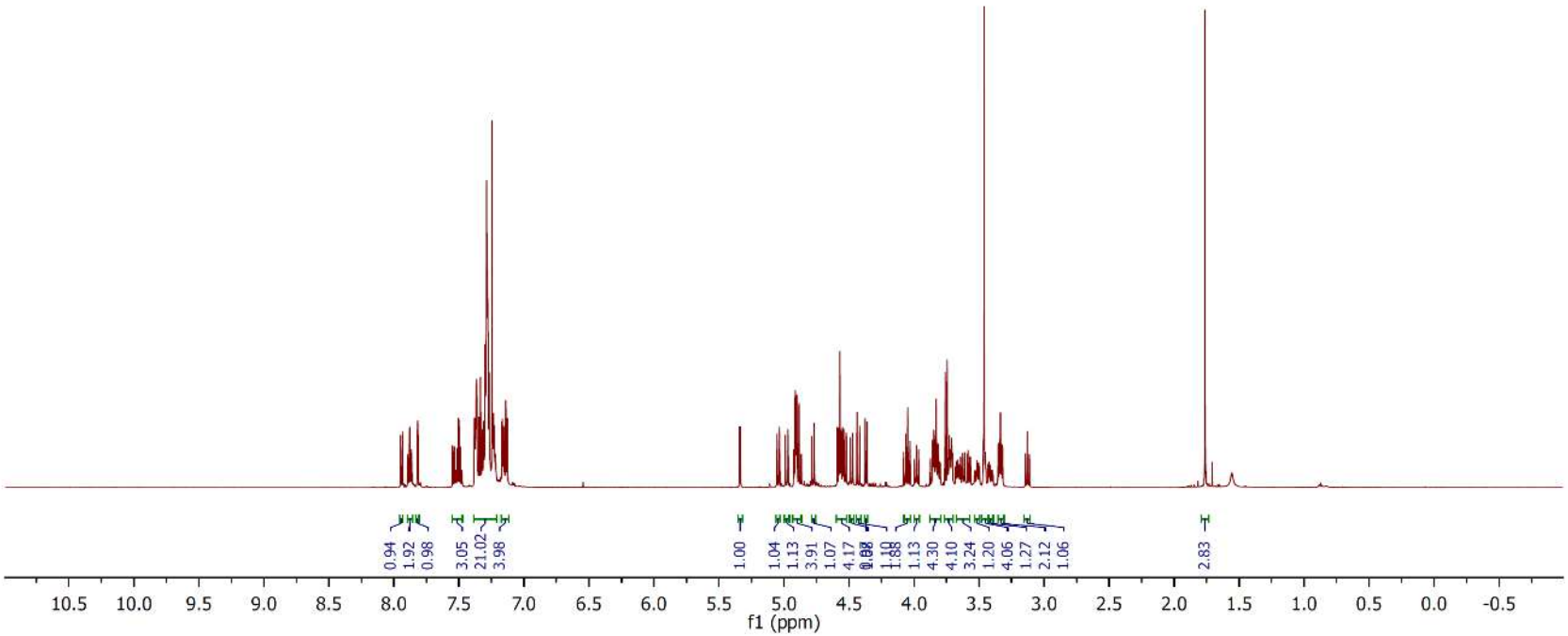

NapO
BnO
BnO
N3
O
CO2Me
O
BnO
AcO
NapO
O
BnO
AcHN
O
N3
27
13C NMR, 150 MHz, CDCl3
169.9583
168.8449
168.0834
128.6640
128.4499
128.4403
128.3724
128.3576
128.3302
128.2361
128.1874
128.0609
127.9854
127.8834
127.8523
127.6362
127.6086
127.5652
127.4816
127.2859
127.0758
126.8149
126.5460
126.2905
126.0950
125.9629
97.8040
97.6657
82.3443
79.8872
77.8262
75.3772
74.7048
74.5947
74.2494
73.9405
73.7030
73.2001
71.2826
70.7742
69.4767
67.4491
52.0688
50.3714
23.1168
20.6724
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

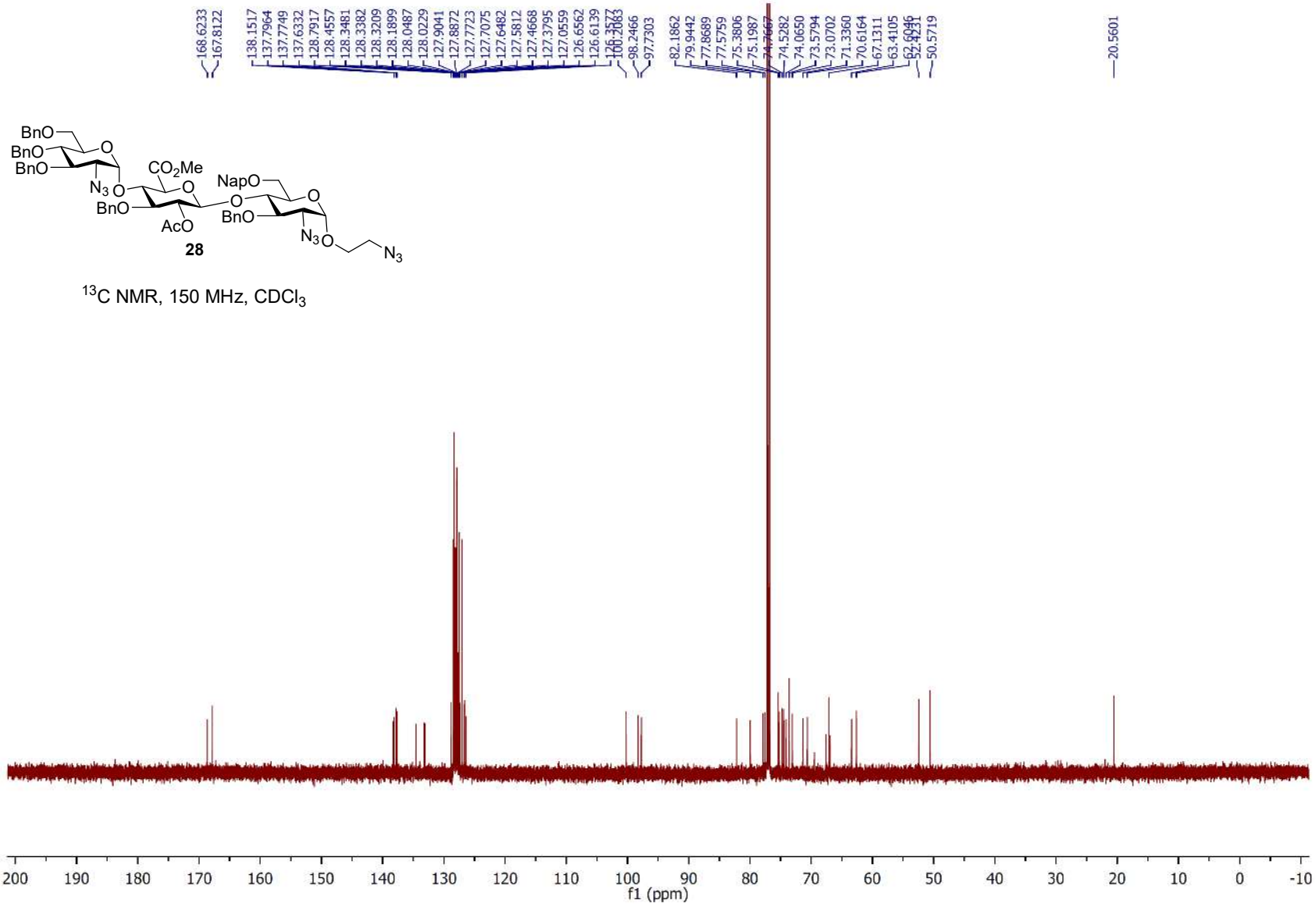

NapO
BnO
BnO
N3
CO2Me
O
BnO
AcO
NapO
BnO
AcHN
N3
27
1H-13C HSQC, 600/150MHz, CDCl3
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)

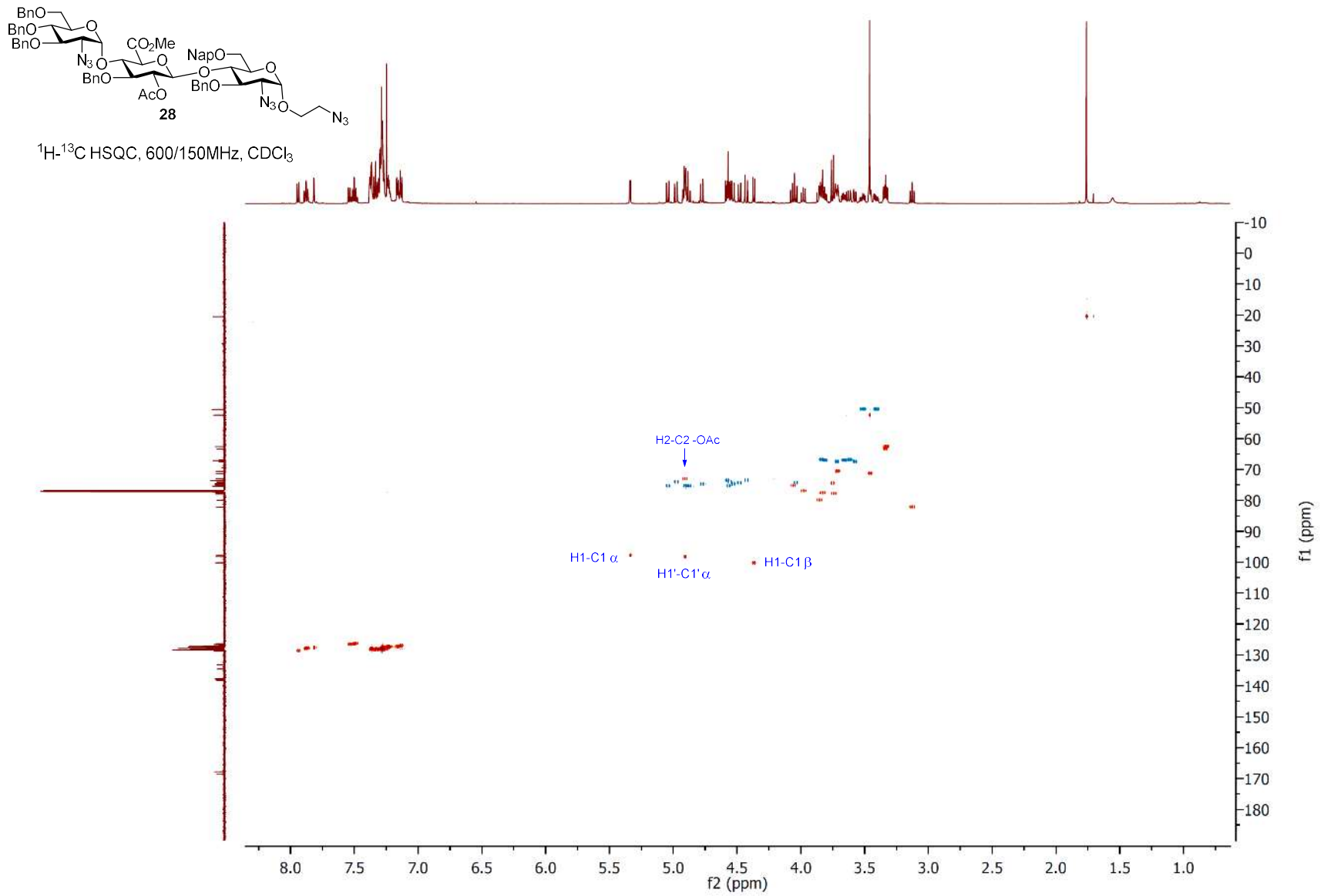


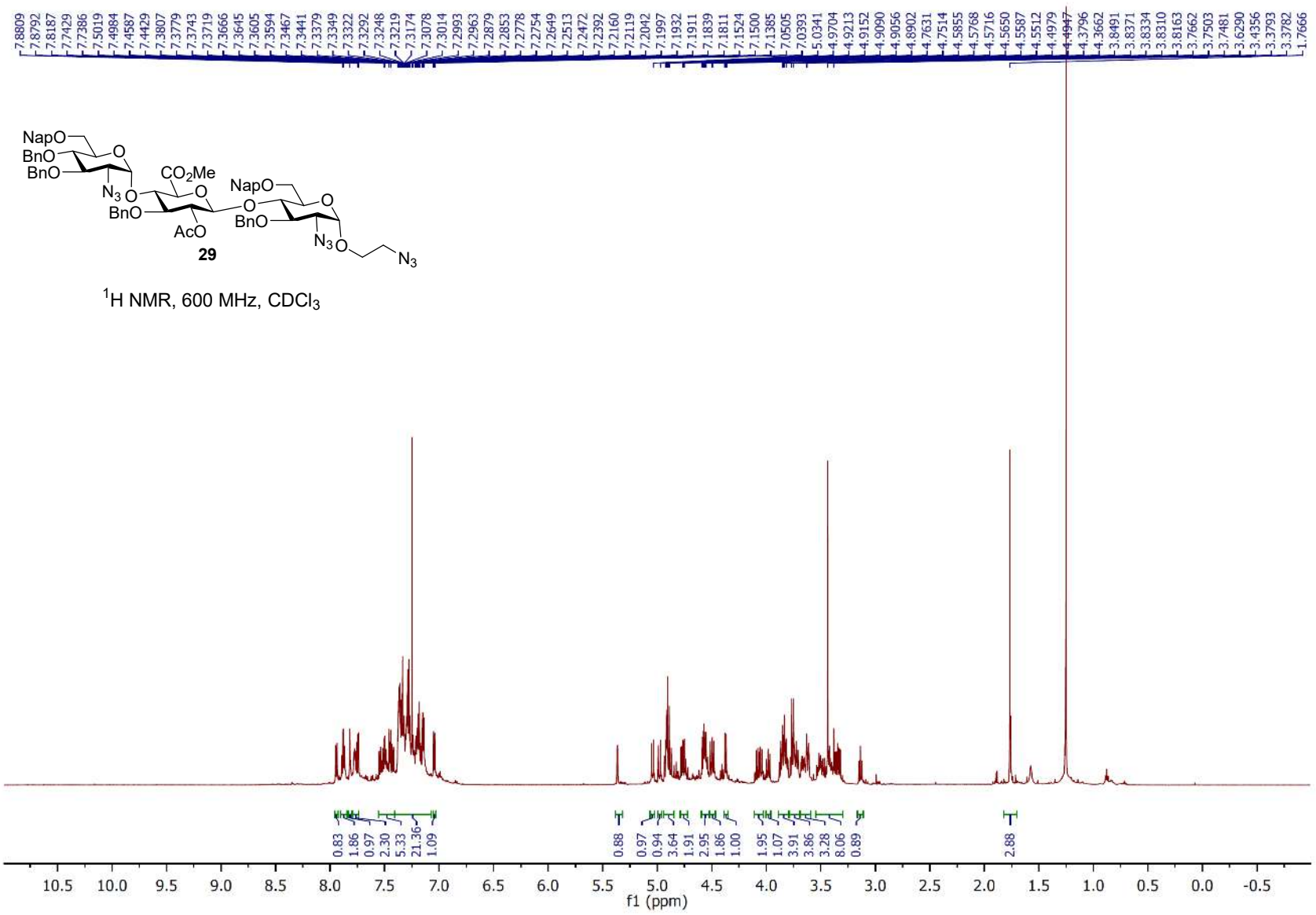
28

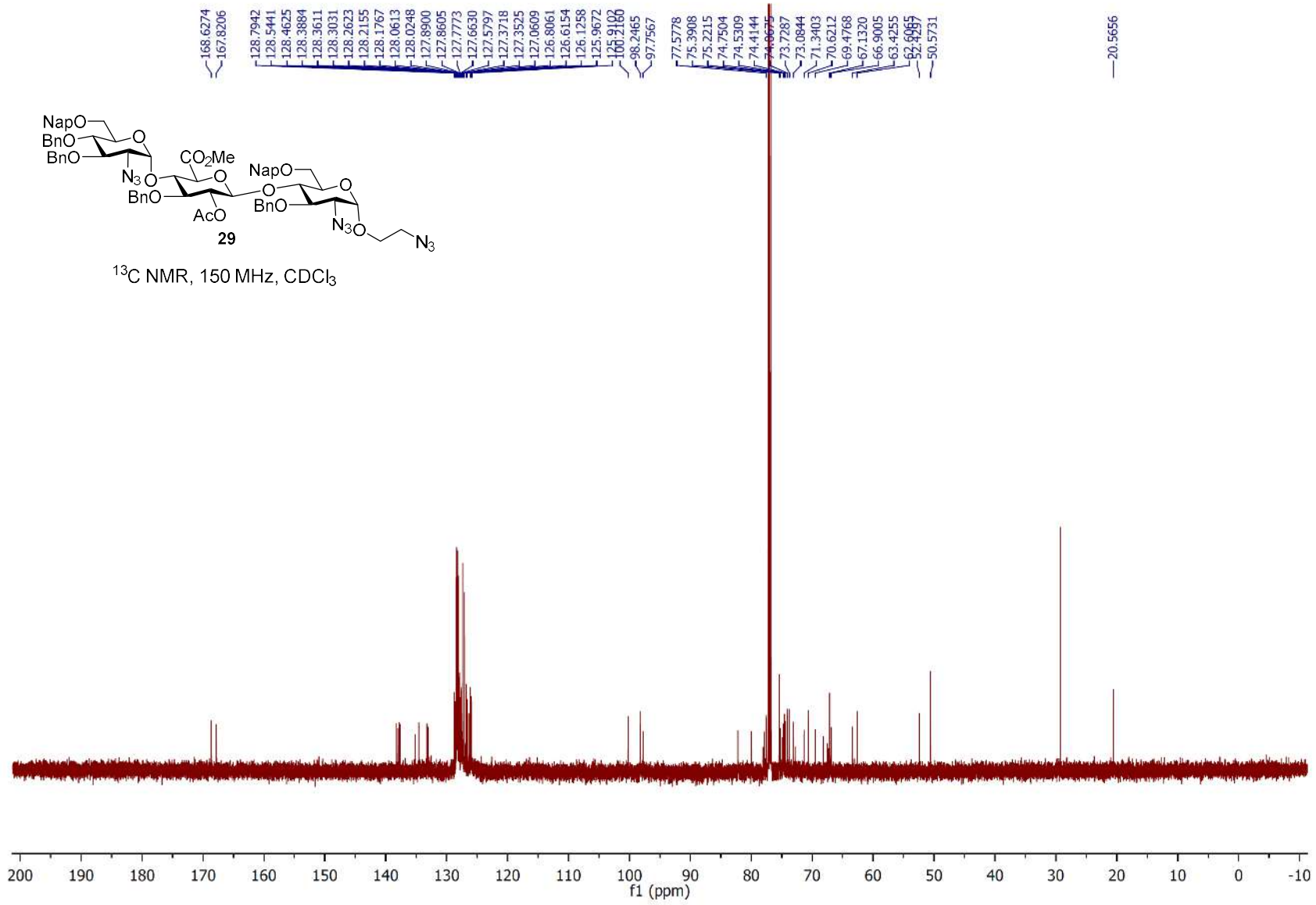
^{1}H NMR, 600 MHz, $CDCl_3$

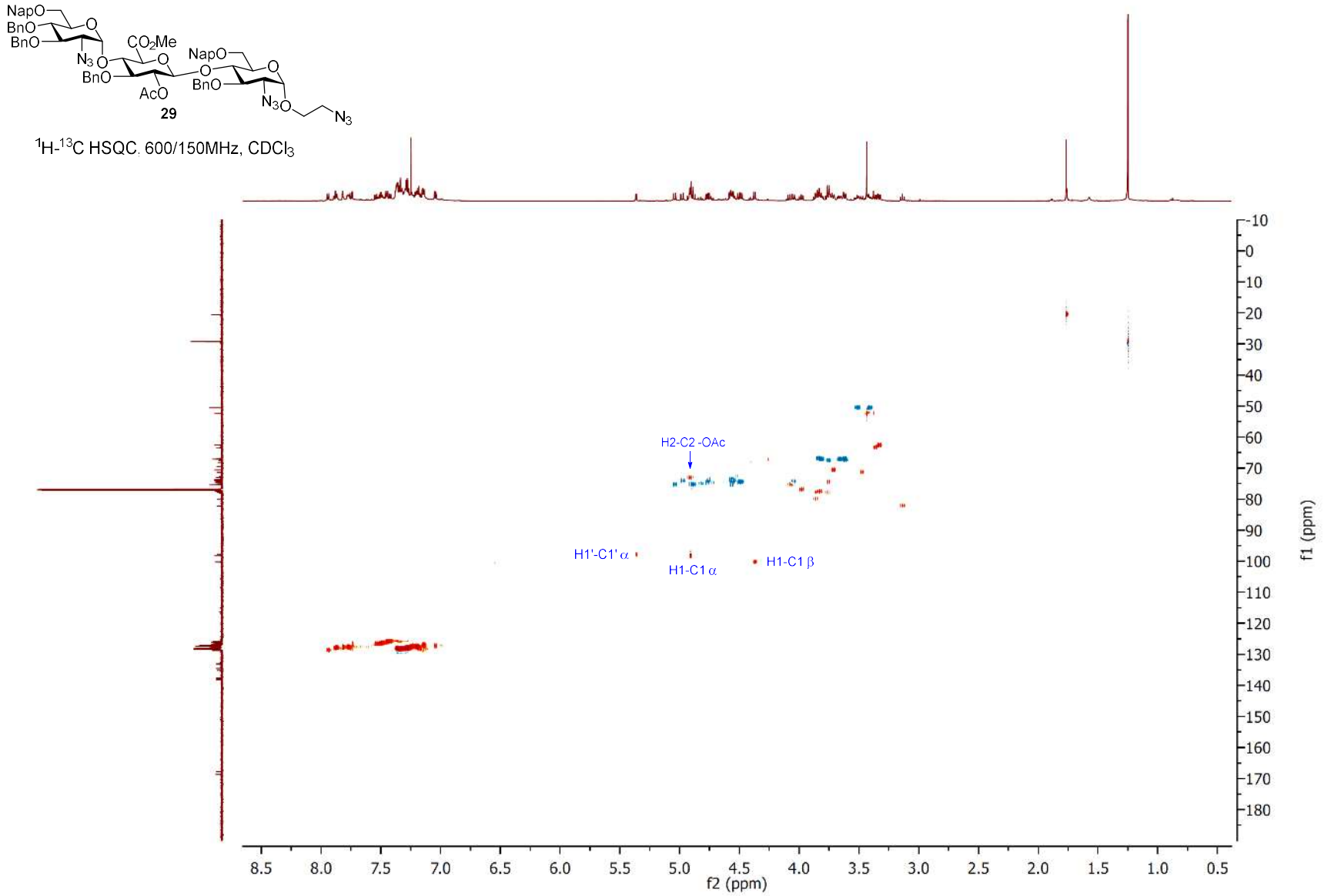


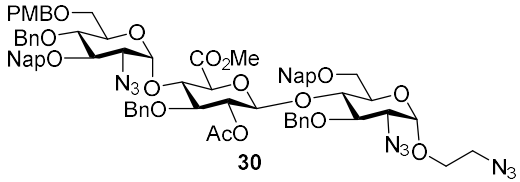

168.6233
167.8122
138.1517
137.7964
137.7749
137.6332
128.7917
128.4557
128.3481
128.3382
128.3209
128.1899
128.0487
128.0229
127.9041
127.8872
127.7723
127.7075
127.6482
127.5812
127.4668
127.3795
127.0559
126.6562
126.6139
98.2466
97.7303
82.1862
79.9442
77.8689
77.5759
75.3806
75.1987
74.5282
74.0650
73.5794
73.0702
71.3360
70.6164
67.1311
63.4105
50.5719
20.5601
BnO
BnO
BnO
O
N3
O
CO2Me
O
BnO
AcO
NapO
O
BnO
N3
O
N3
28
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


BnO
BnO
BnO
O
N3
O
BnO
CO2Me
O
AcO
NapO
O
BnO
O
N3
O
N3
28
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2 -OAc
H1-C1 α
H1'-C1'α
H1-C1 β
f2 (ppm)
f1 (ppm)


NapO
BnO
BnO
O
N3
O
CO2Me
O
BnO
AcO
NapO
O
O
BnO
N3
O
N3
29
1H NMR, 600 MHz, CDCl3
f1 (ppm)

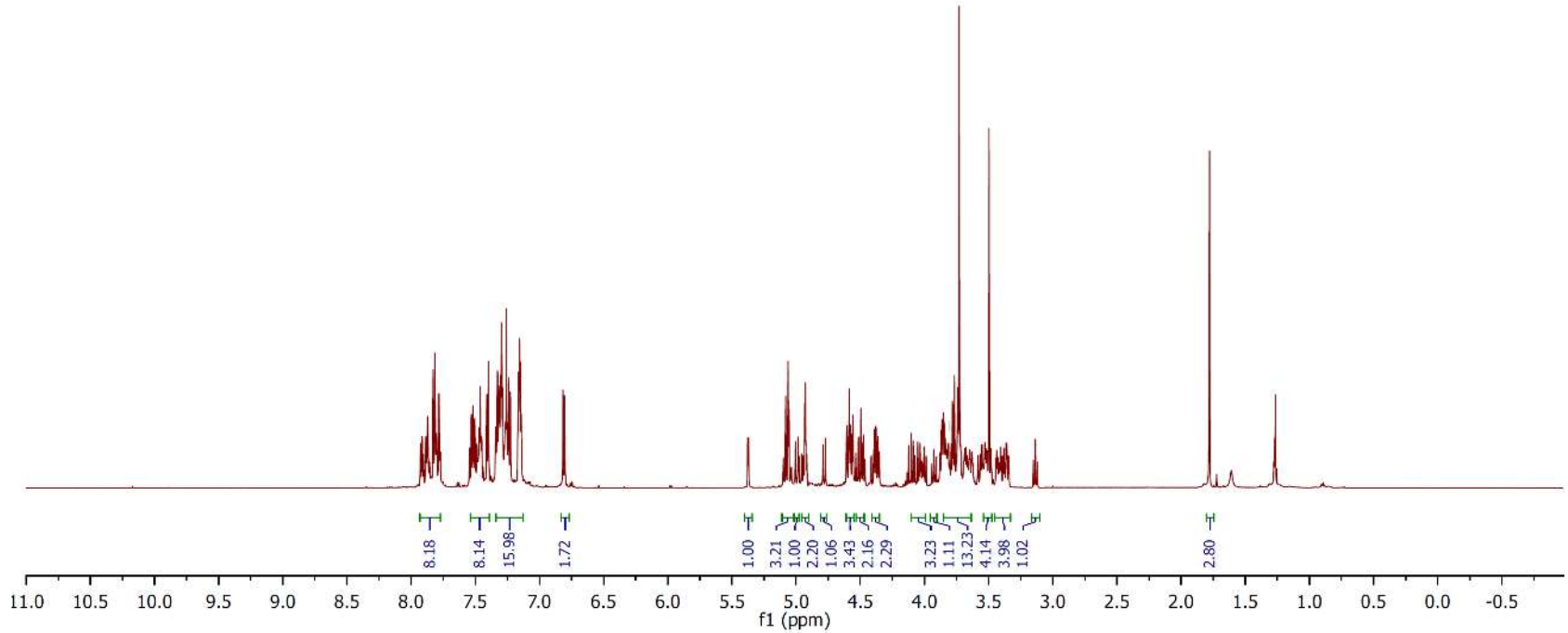

168.6274
167.8206
128.7942
128.5441
128.4625
128.3884
128.3611
128.3031
128.2623
128.2155
128.1767
128.0613
128.0248
127.8900
127.8605
127.7773
127.6630
127.5797
127.3718
127.3525
127.0609
126.8061
126.6154
126.1258
125.9672
98.2465
97.7567
77.5778
75.3908
75.2215
74.7504
74.5309
74.4144
73.7287
73.0844
71.3403
70.6212
69.4768
67.1320
66.9005
63.4255
52.4257
50.5731
20.5656
NapO
BnO
BnO
O
N3
CO2Me
O
BnO
AcO
29
NapO
O
BnO
N3
O
N3
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

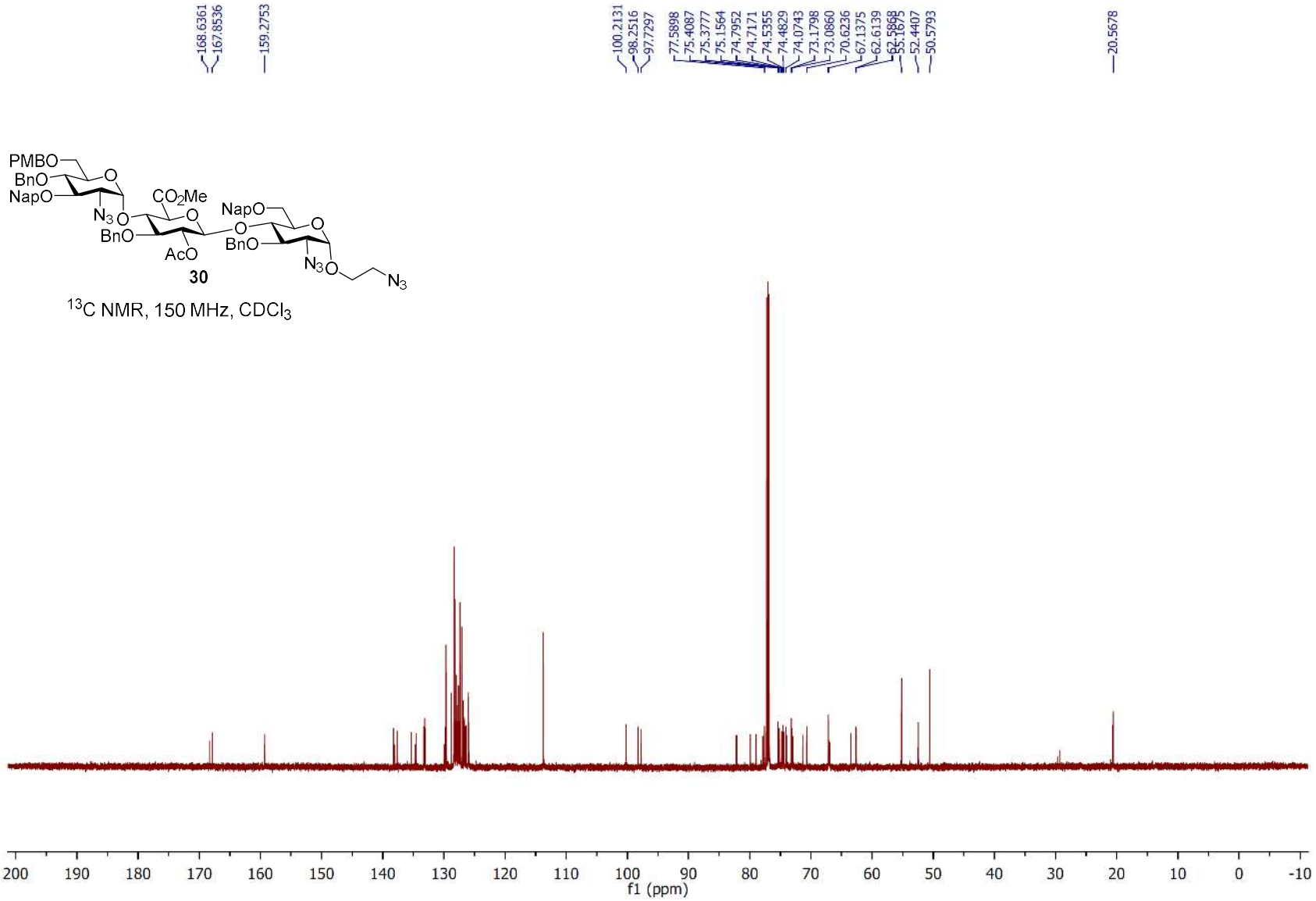

NapO
BnO
BnO
N3
O
CO2Me
O
BnO
AcO
NapO
O
BnO
N3
O
N3
29
1H-13C HSQC. 600/150MHz, CDCl3
H2-C2 -OAc
H1'-C1' α
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)

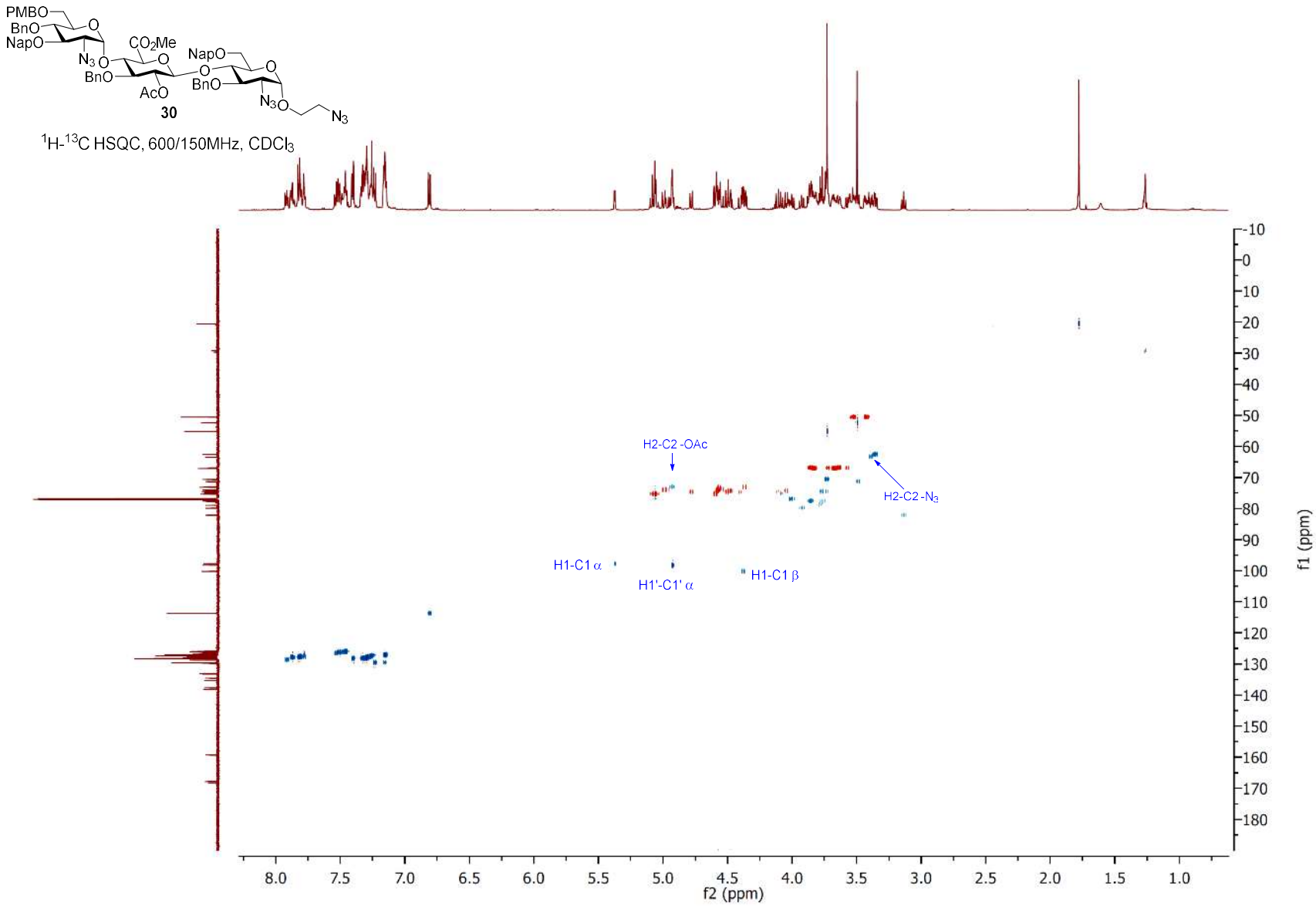


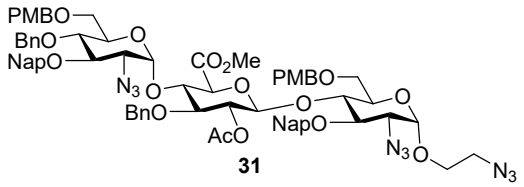
30

^{1}H NMR, 600 MHz, $CDCl_3$



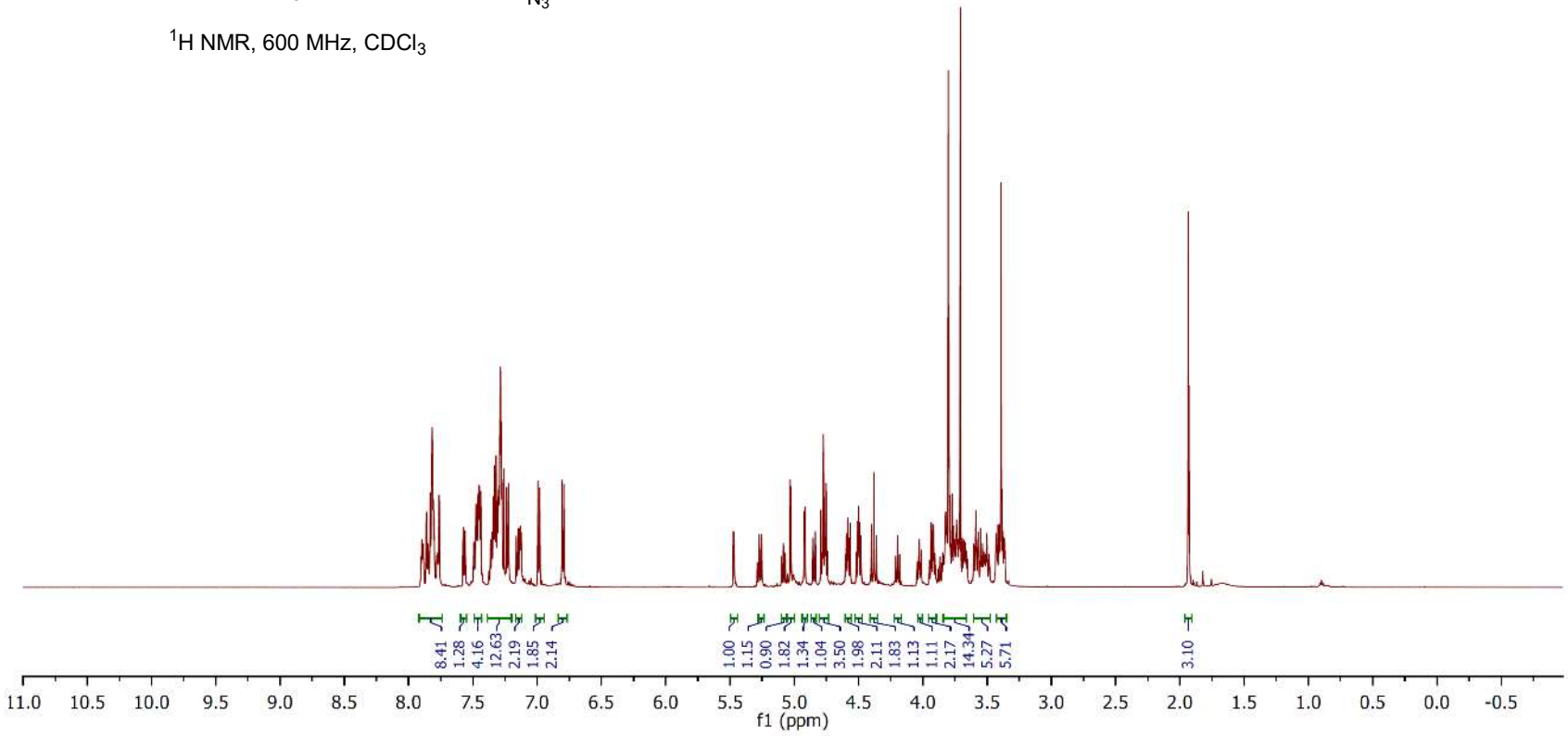

168.6361
167.8536
159.2753
100.2131
98.2516
97.7297
77.5898
75.4087
75.3777
75.1564
74.7952
74.7171
74.5355
74.4829
74.0743
73.1798
73.0860
70.6236
67.1375
62.6139
62.5868
55.1675
52.4407
50.5793
20.5678
PMBO
BnO
NapO
N3
O
CO2Me
BnO
AcO
NapO
BnO
N3
N3
30
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


PMBO
BnO
NapO
N3
CO2Me
AcO
BnO
NapO
BnO
N3
N3
30
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2 -OAc
H2-C2 -N3
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)



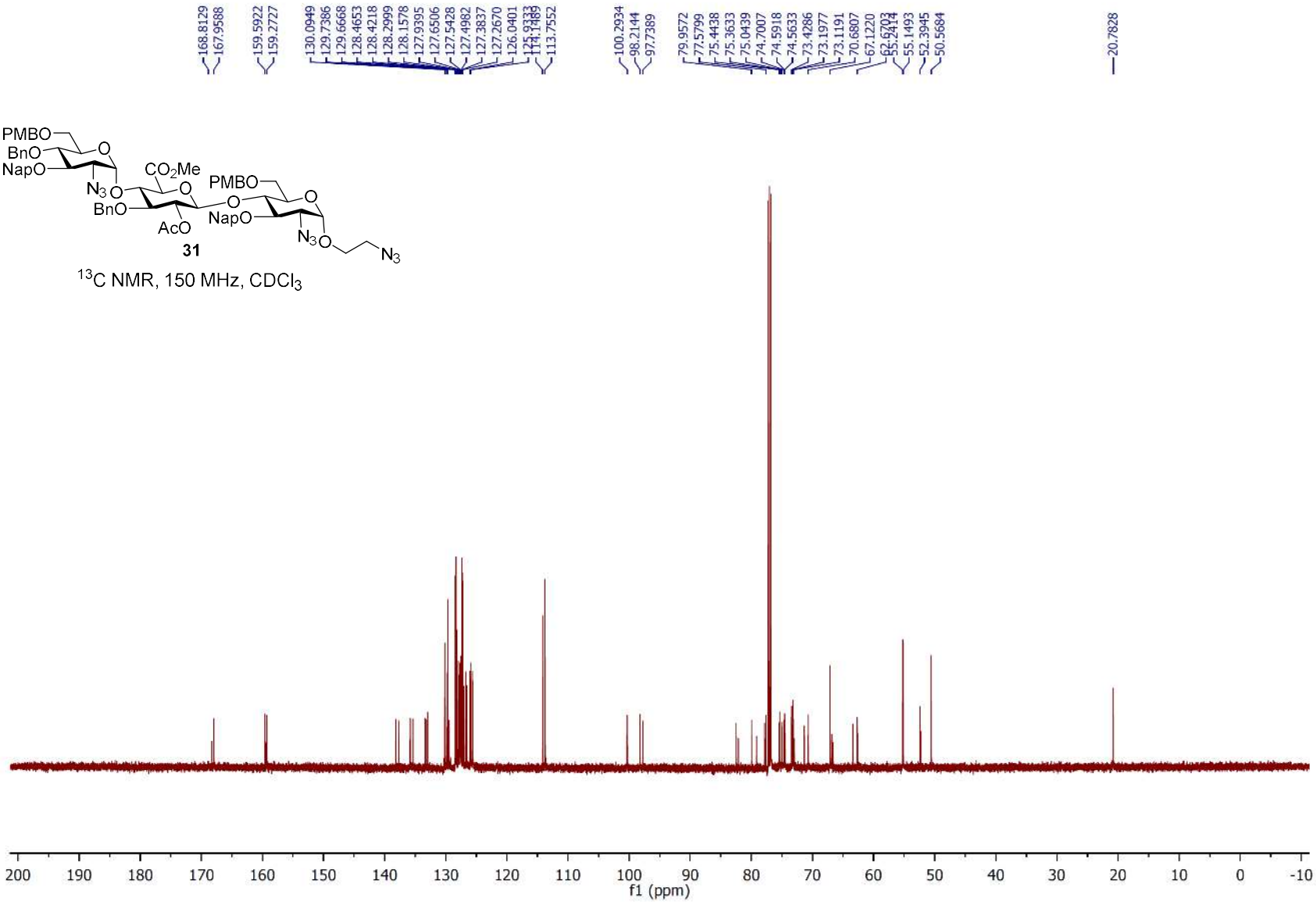
31

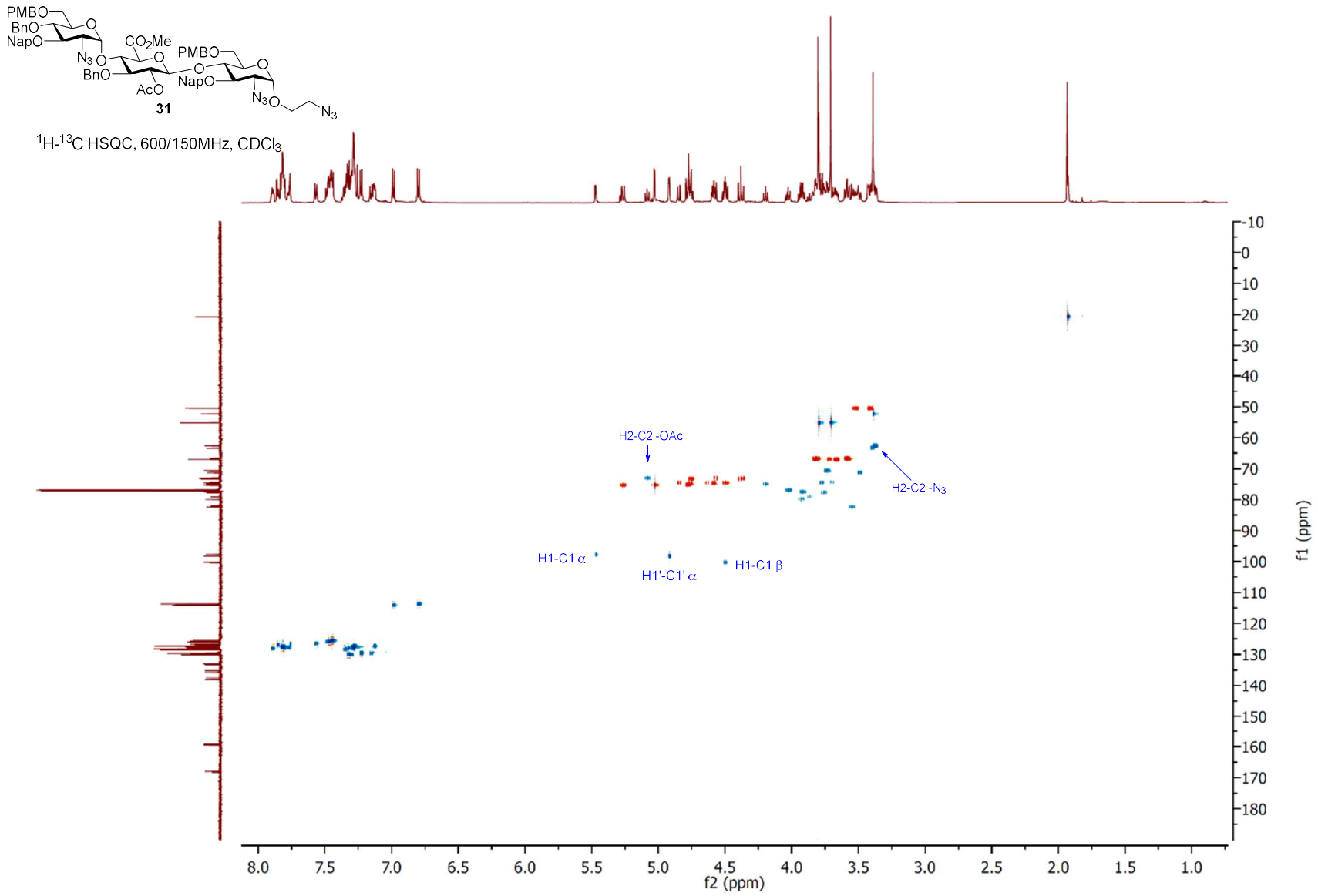
^{1}H NMR, 600 MHz, $CDCl_3$

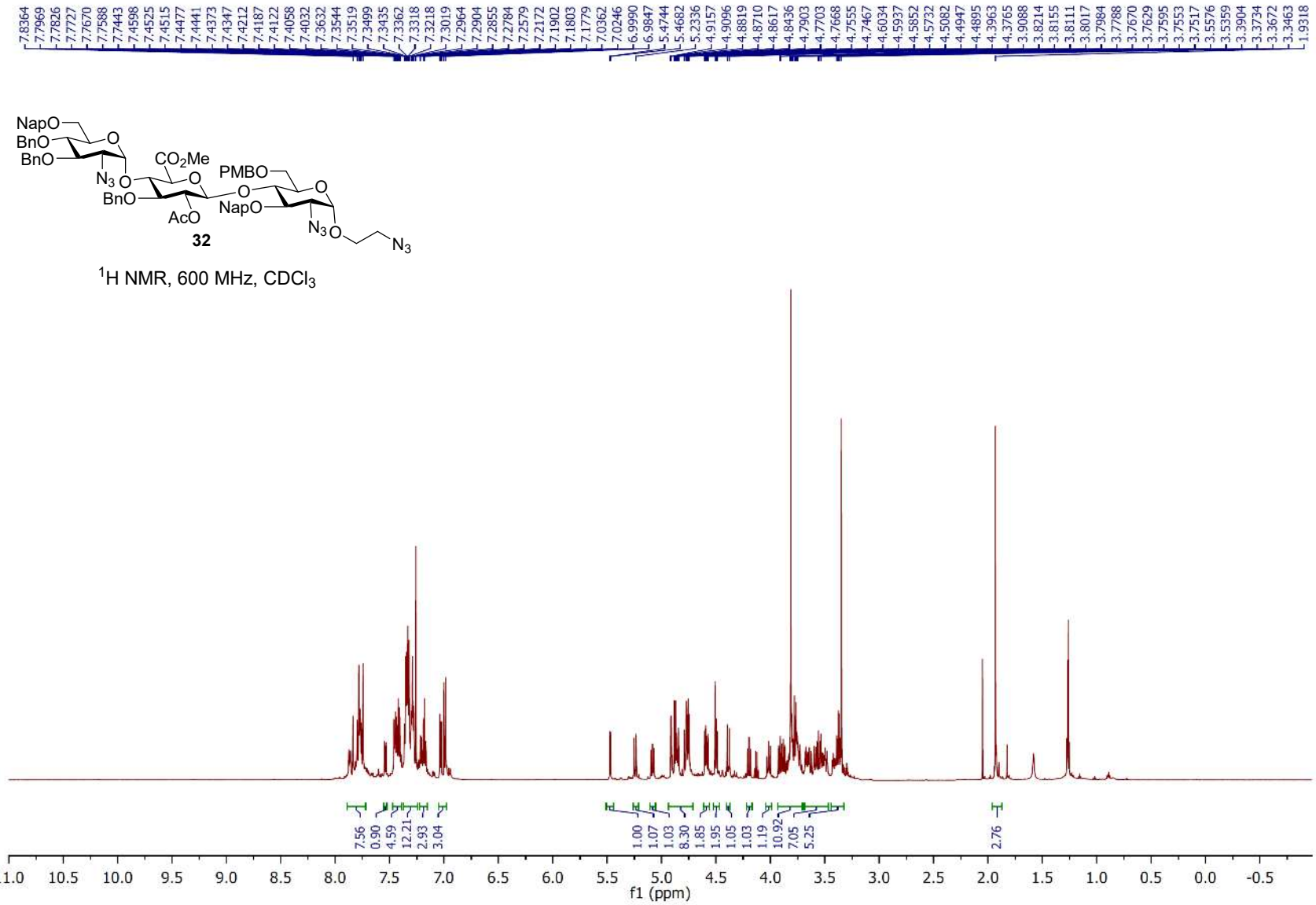


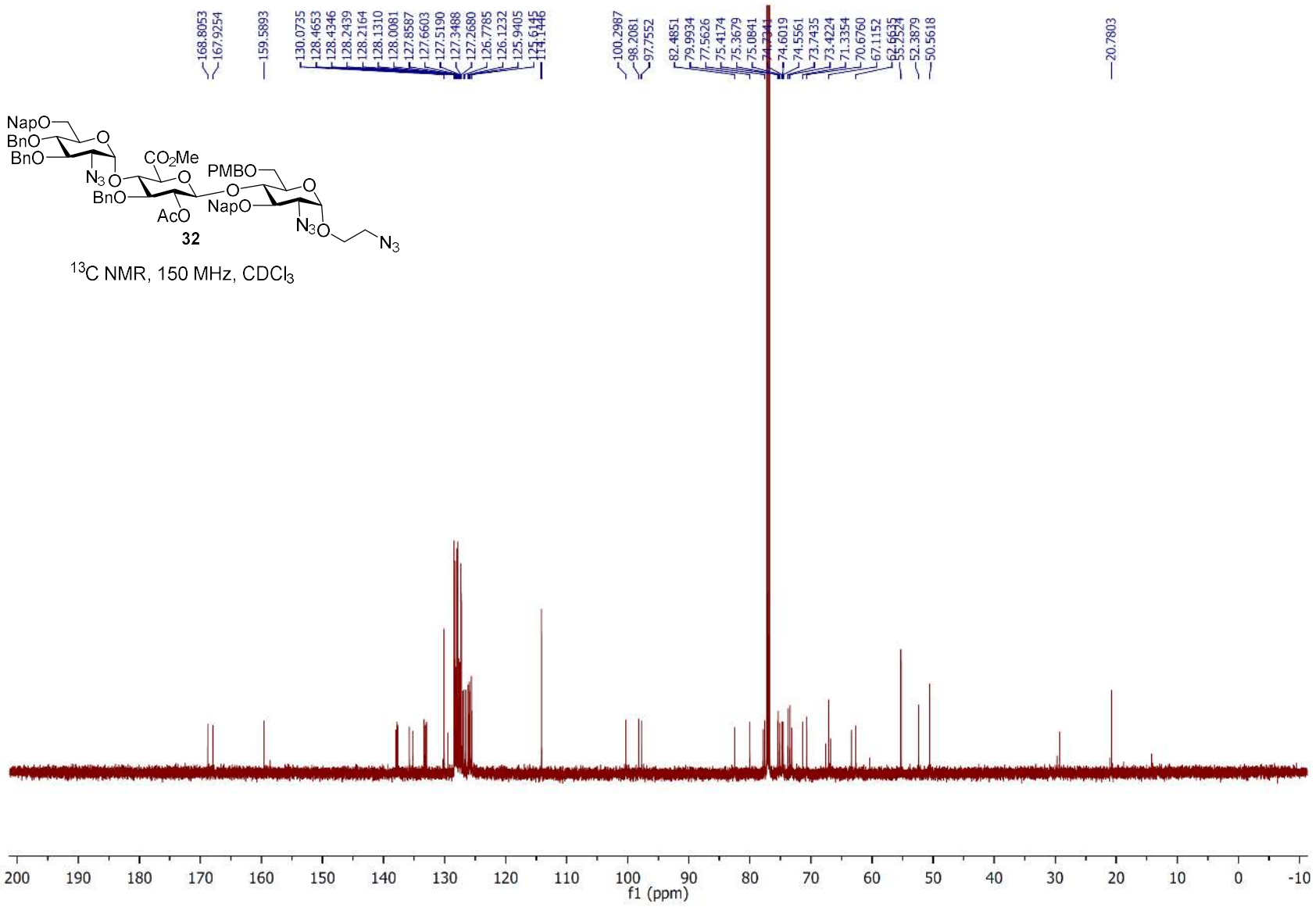
31

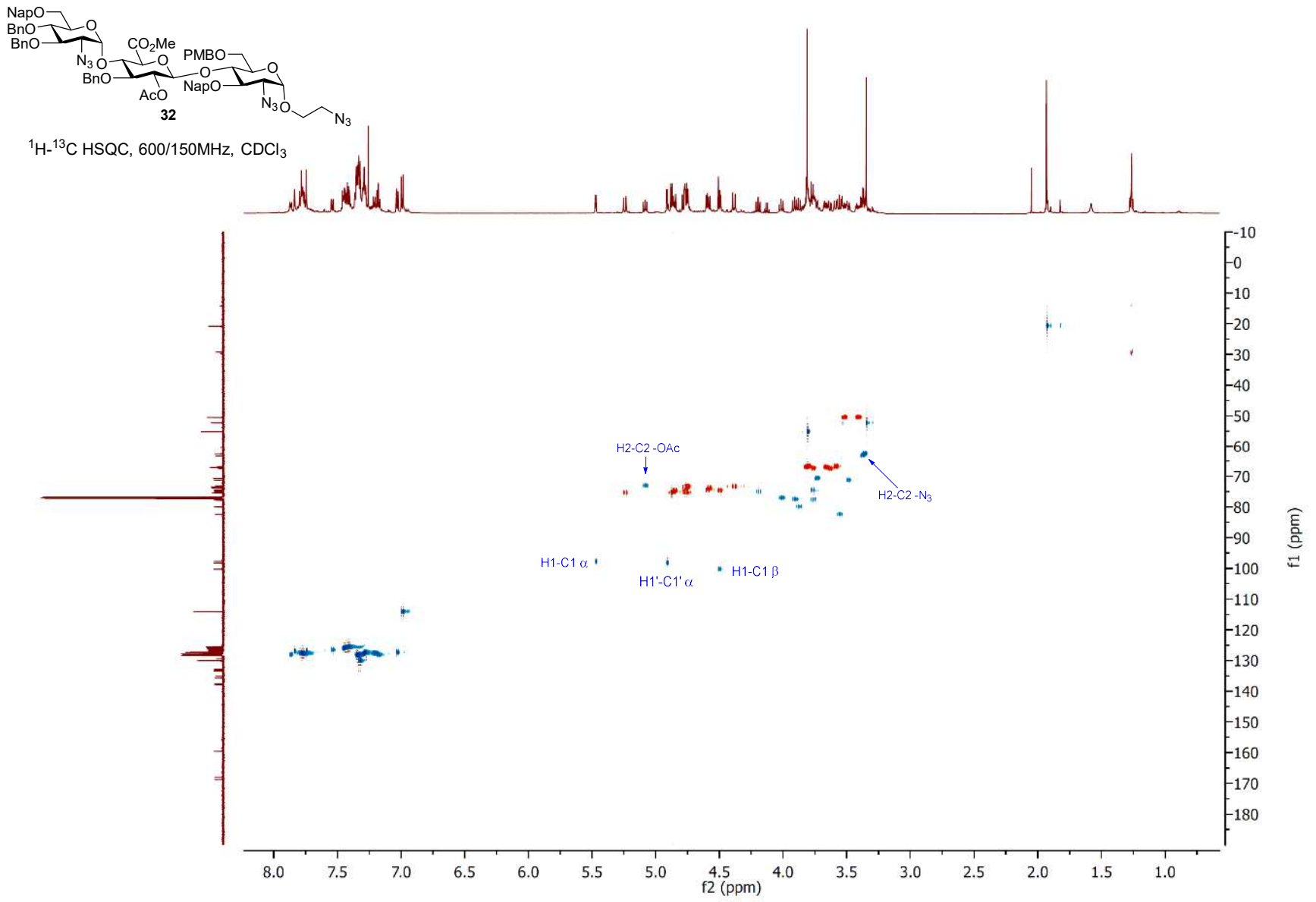
^{13}C NMR, 150 MHz, $CDCl_3$

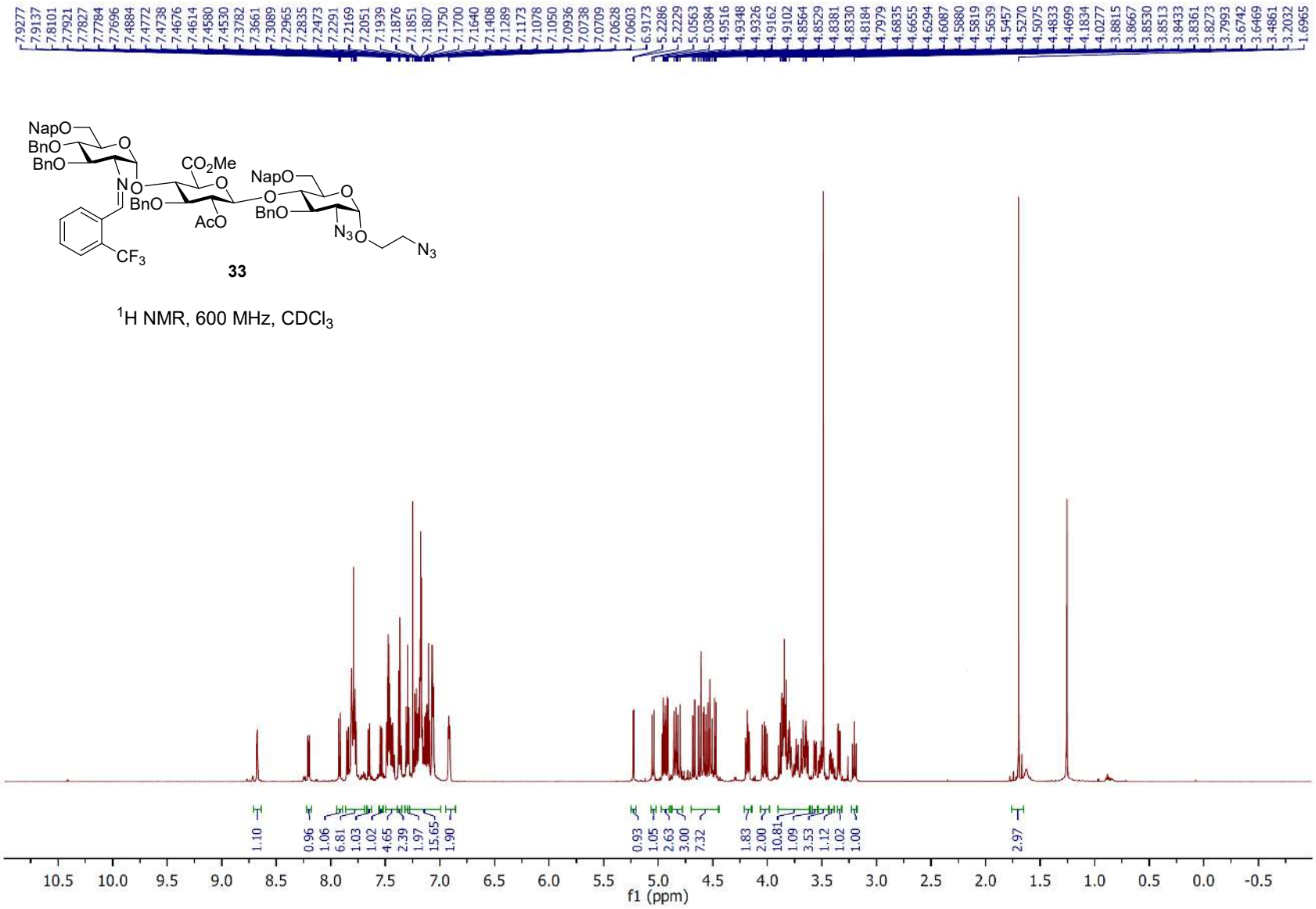


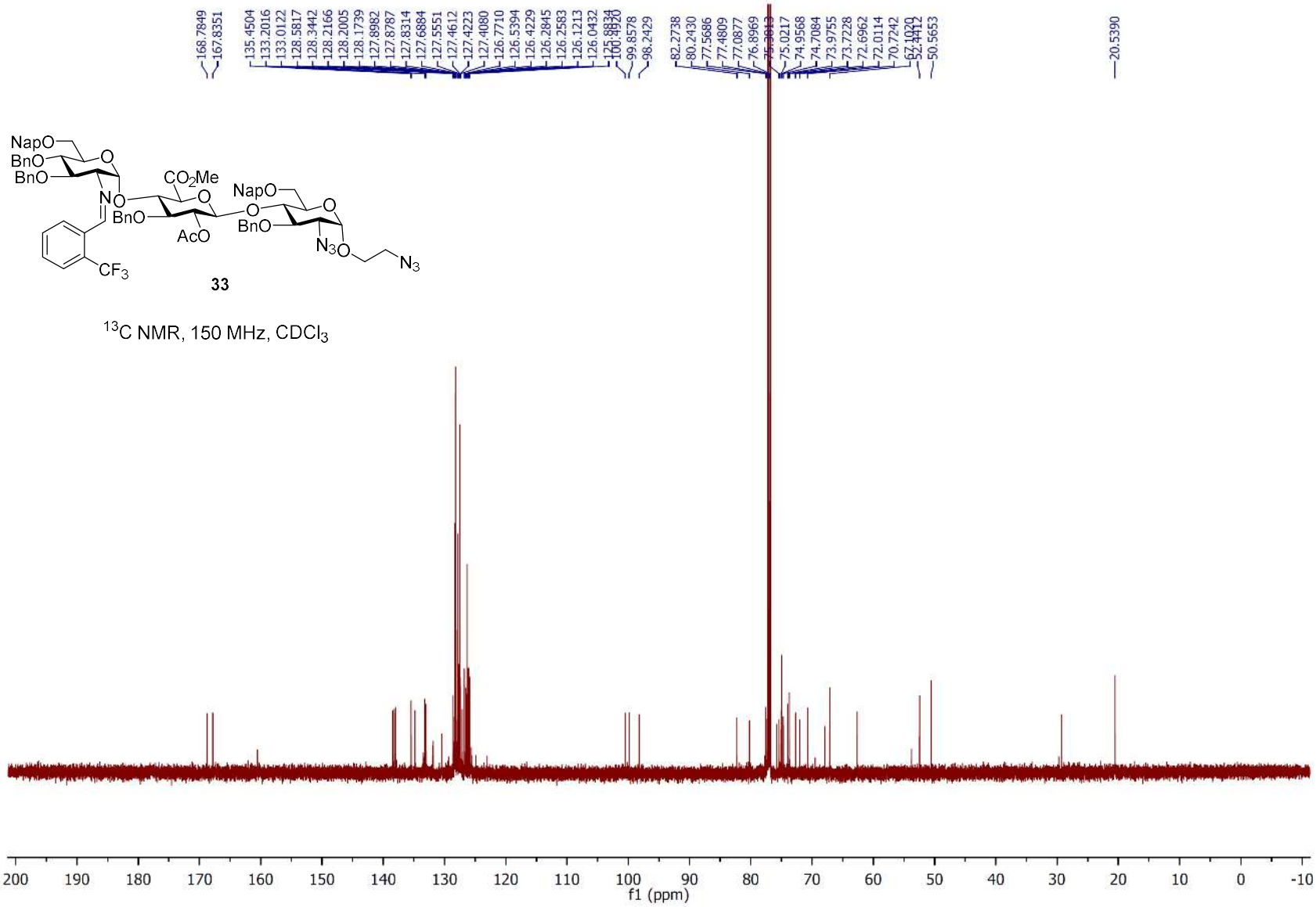

PMBO
BnO
NapO
N3
CO2Me
O
PMBO
BnO
AcO
NapO
N3
N3
31
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2 -OAc
H2-C2 -N3
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)

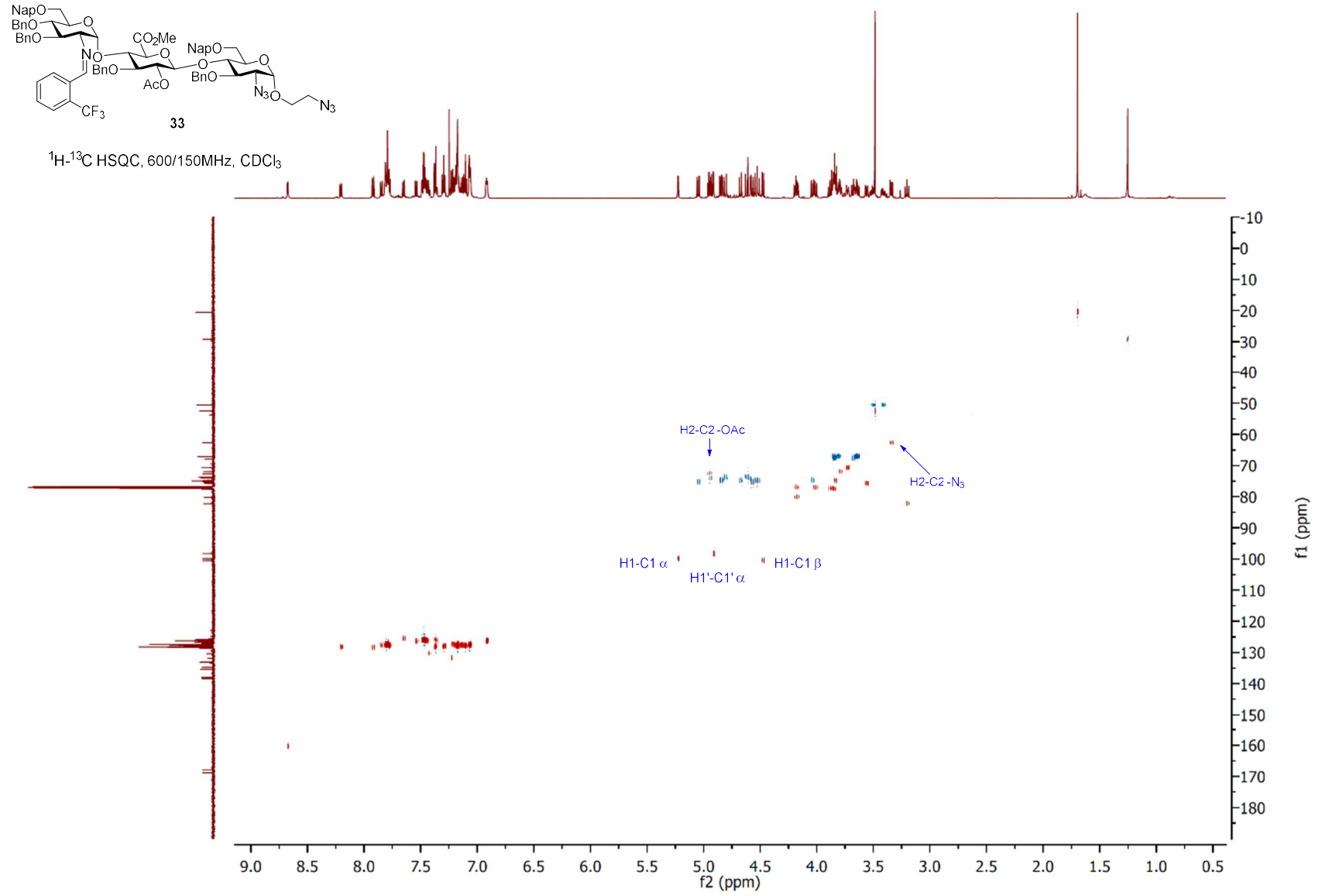

7.8364
7.7969
7.7826
7.7727
7.7670
7.7588
7.7443
7.4598
7.4525
7.4515
7.4477
7.4441
7.4373
7.4347
7.4212
7.4187
7.4122
7.4058
7.4032
7.3632
7.3544
7.3519
7.3499
7.3435
7.3362
7.3318
7.3218
7.3019
7.2964
7.2904
7.2855
7.2784
7.2579
7.2172
7.1902
7.1803
7.1779
7.0362
7.0246
6.9990
6.9847
5.4744
5.4682
5.2336
4.9157
4.9096
4.8819
4.8710
4.8617
4.8436
4.7903
4.7703
4.7668
4.7555
4.7467
4.6034
4.5937
4.5852
4.5732
4.5082
4.4947
4.4895
4.3963
4.3765
3.9088
3.8214
3.8155
3.8111
3.8017
3.7984
3.7788
3.7670
3.7629
3.7595
3.7553
3.7517
3.5576
3.5359
3.3904
3.3734
3.3672
3.3463
1.9318
NapO
BnO
BnO
O
N3
O
CO2Me
O
BnO
AcO
PMBO
O
O
NapO
N3
O
N3
32
1H NMR, 600 MHz, CDCl3
7.56
0.90
4.59
12.21
2.93
3.04
1.00
1.07
1.03
8.30
1.85
1.95
1.05
1.03
1.19
10.92
7.05
5.25
2.76
11.0
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)

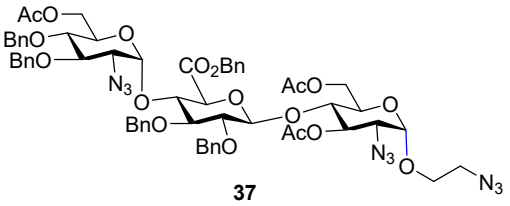

NapO
BnO
BnO
O
N3
O
CO2Me
O
BnO
AcO
PMBO
O
NapO
O
N3
O
N3
32
13C NMR, 150 MHz, CDCl3
168.8053
167.9254
159.5893
130.0735
128.4653
128.4346
128.2439
128.2164
128.1310
128.0081
127.8587
127.6603
127.5190
127.3488
127.2680
126.7785
126.1232
125.9405
100.2987
98.2081
97.7552
82.4851
79.9934
77.5626
75.4174
75.3679
75.0841
74.6019
74.5561
73.7435
73.4224
71.3354
70.6760
67.1152
62.6635
55.2524
52.3879
50.5618
20.7803
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


NapO
BnO
BnO
O
N3
O
CO2Me
O
BnO
AcO
PMBO
O
NapO
O
N3
O
N3
32
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2 -OAc
H2-C2 -N3
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)


7.9277
7.9137
7.8101
7.7921
7.7827
7.7784
7.7696
7.4884
7.4772
7.4738
7.4676
7.4614
7.4580
7.4530
7.3782
7.3661
7.3089
7.2965
7.2835
7.2473
7.2291
7.2169
7.2051
7.1939
7.1876
7.1851
7.1807
7.1750
7.1700
7.1640
7.1408
7.1289
7.1173
7.1078
7.1050
7.0936
7.0738
7.0709
7.0628
7.0603
6.9173
5.2286
5.2229
5.0563
5.0384
4.9516
4.9348
4.9326
4.9162
4.9102
4.8564
4.8529
4.8381
4.8330
4.8184
4.7979
4.6835
4.6655
4.6294
4.6087
4.5880
4.5819
4.5639
4.5457
4.5270
4.5075
4.4833
4.4699
4.1834
4.0277
3.8815
3.8667
3.8530
3.8513
3.8433
3.8361
3.8273
3.7993
3.6742
3.6469
3.4861
3.2032
1.6965
NapO
BnO
BnO
O
N
O
BnO
CO2Me
O
AcO
NapO
O
O
BnO
N3
O
N3
CF3
33
1H NMR, 600 MHz, CDCl3
1.10
0.96
1.06
6.81
1.03
1.02
4.65
2.39
1.97
15.65
1.90
0.93
1.05
2.63
3.00
7.32
1.83
2.00
10.81
1.09
3.53
1.12
1.02
1.00
2.97
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)

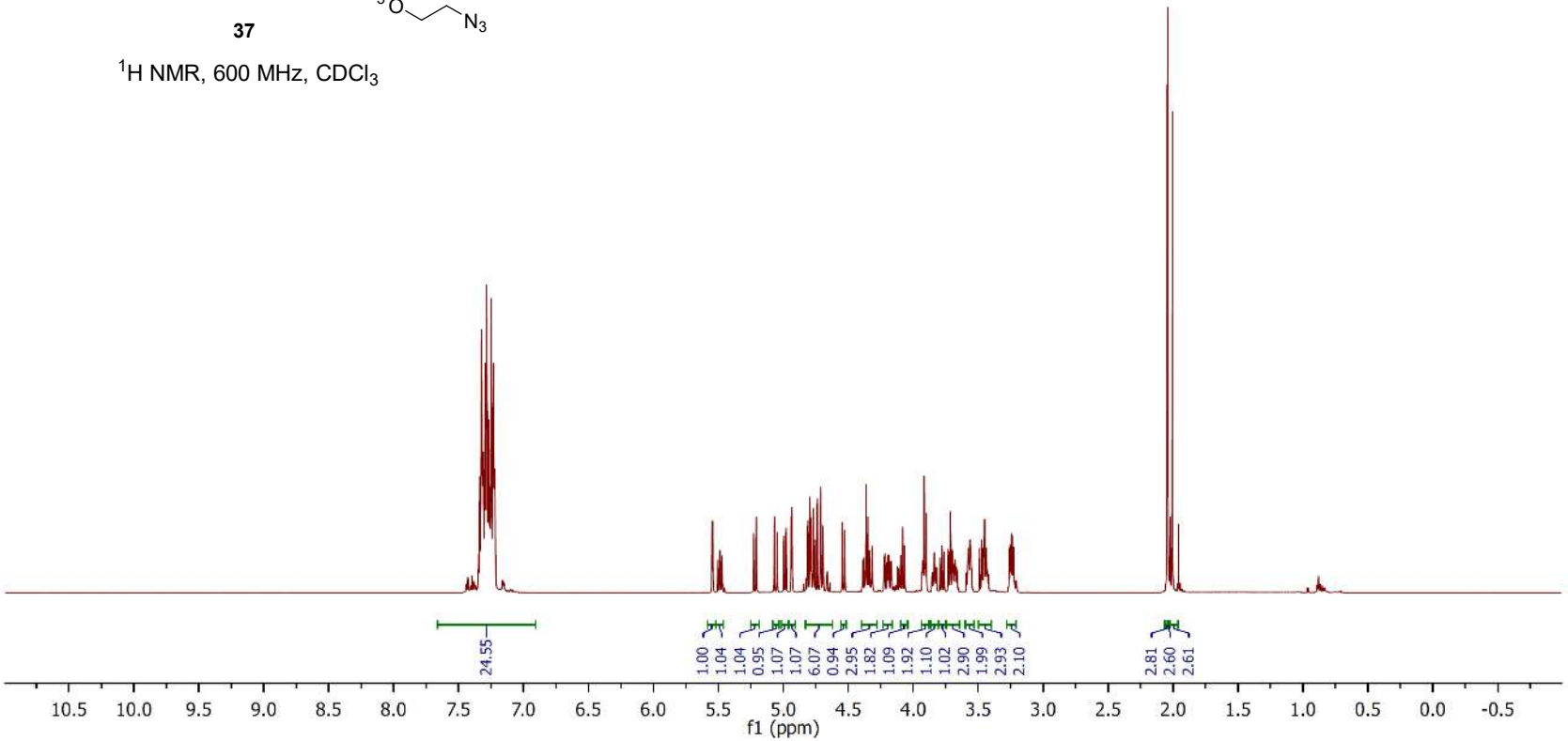

NapO
BnO
BnO
O
N
O
BnO
CO2Me
O
AcO
NapO
O
BnO
O
N3O
N3
CF3
33
13C NMR, 150 MHz, CDCl3
f1 (ppm)

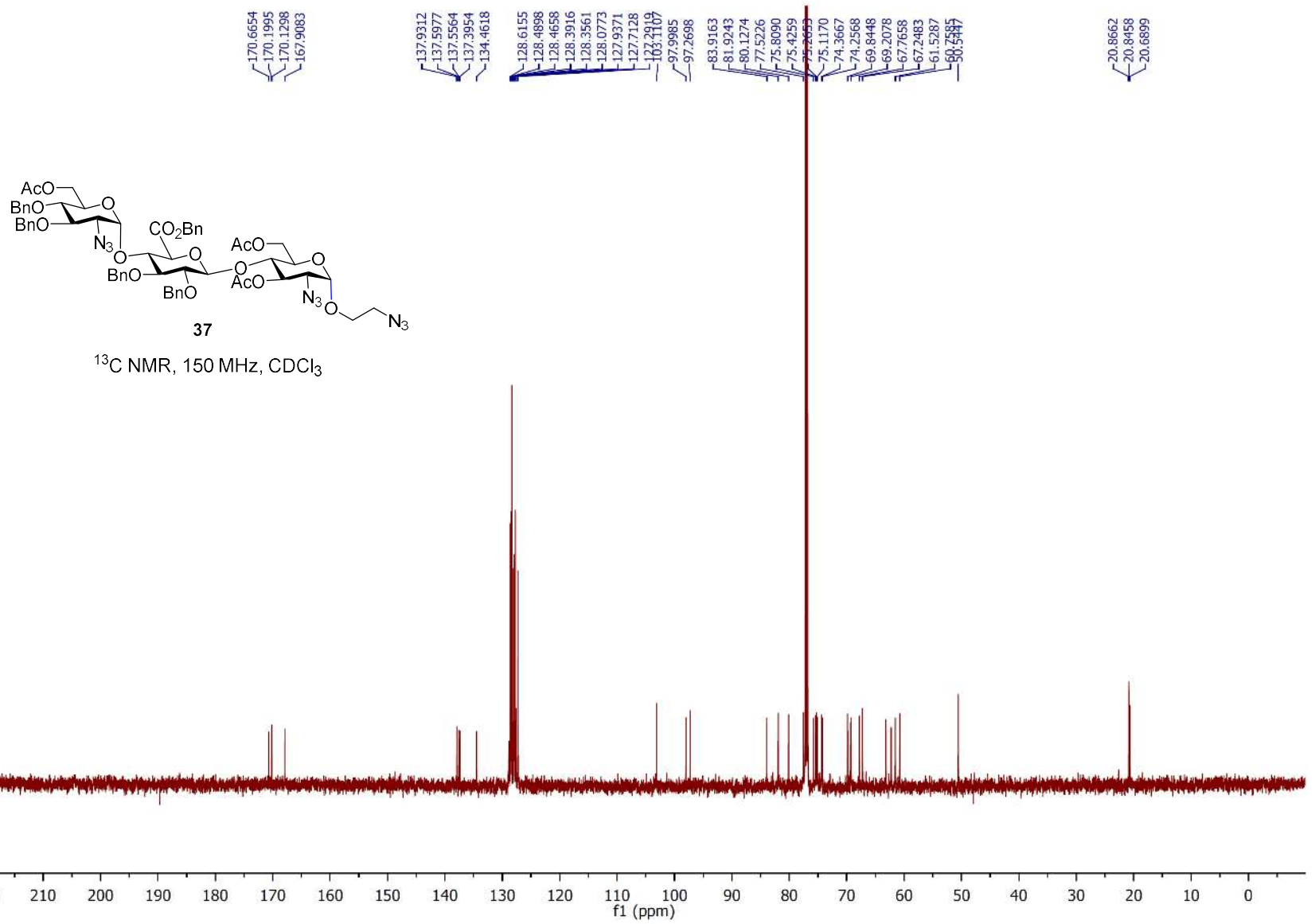

NapO
BnO
BnO
O
N
BnO
CO2Me
O
AcO
NapO
O
BnO
O
N3O
N3
CF3
33
1H-13C HSQC, 600/150MHz, CDCl3
H2-C2 -OAc
H2-C2 -N3
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)

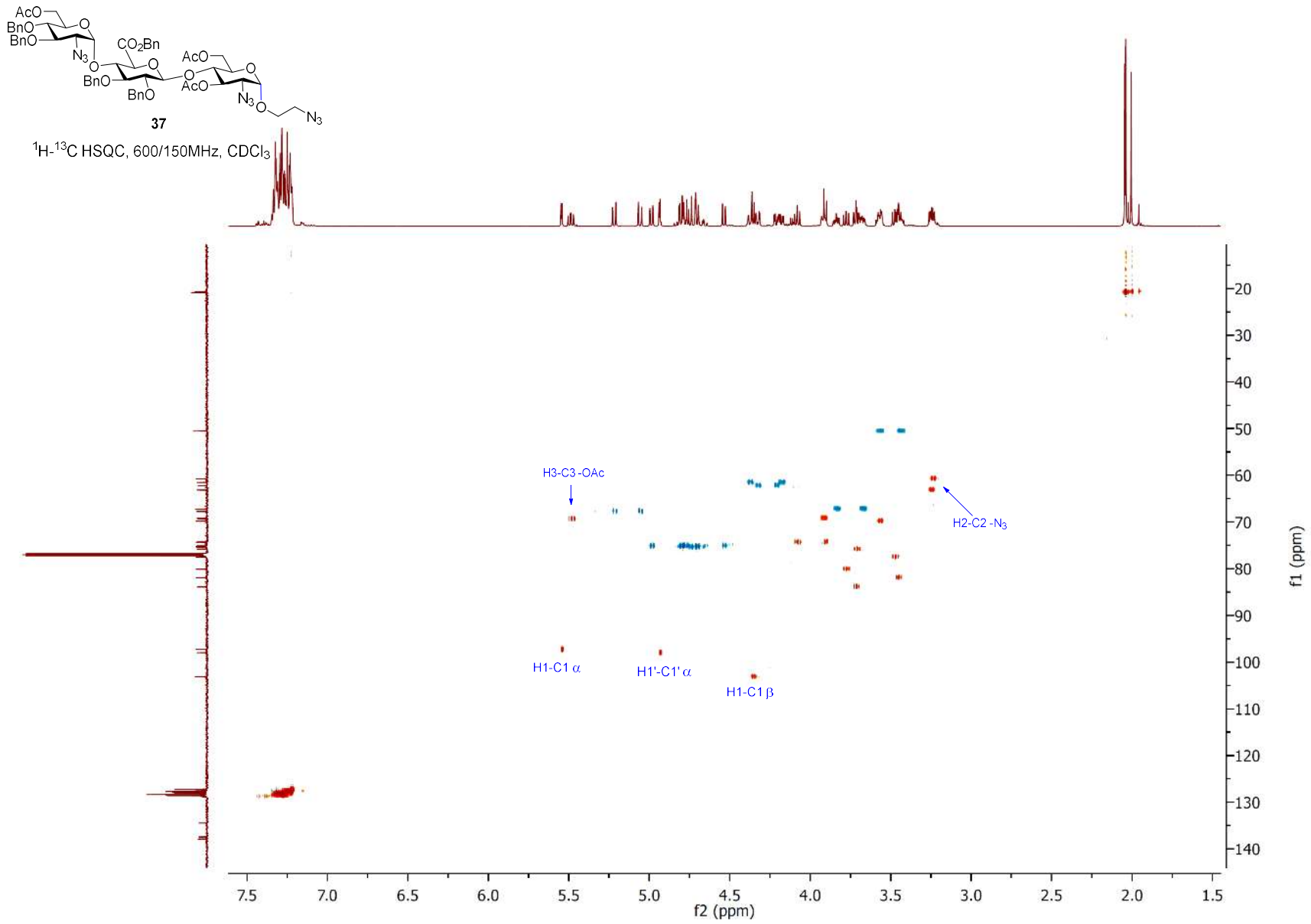


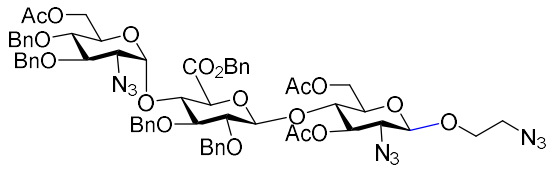
37

^{1}H NMR, 600 MHz, $CDCl_3$



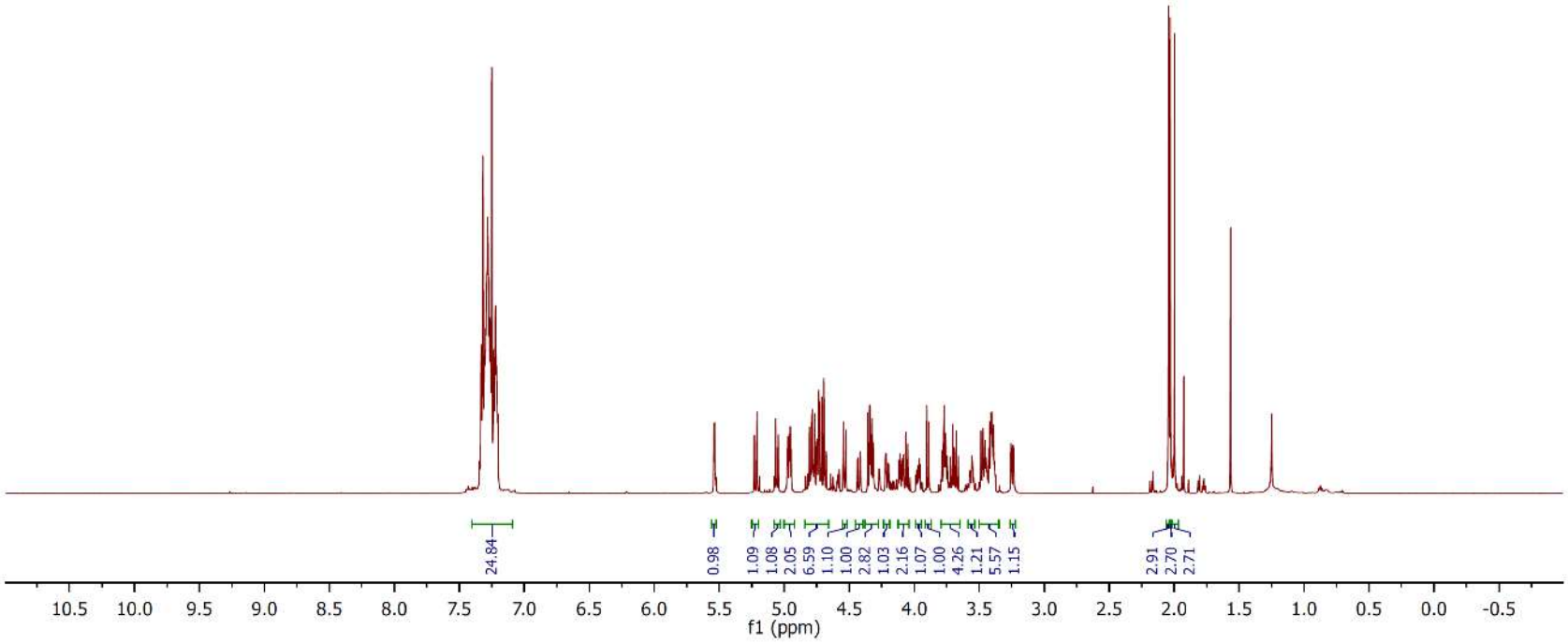

AcO
BnO
BnO
O
N3
CO2Bn
O
BnO
BnO
O
AcO
AcO
O
N3
O
N3
37
13C NMR, 150 MHz, CDCl3
170.6654
170.1995
170.1298
167.9083
137.9312
137.5977
137.5564
137.3954
134.4618
128.6155
128.4898
128.4658
128.3916
128.3561
128.0773
127.9371
127.7128
97.9985
97.2698
83.9163
81.9243
80.1274
77.5226
75.8090
75.4259
75.1170
74.3667
74.2568
69.8448
69.2078
67.7658
67.2483
61.5287
20.8662
20.8458
20.6899
f1 (ppm)

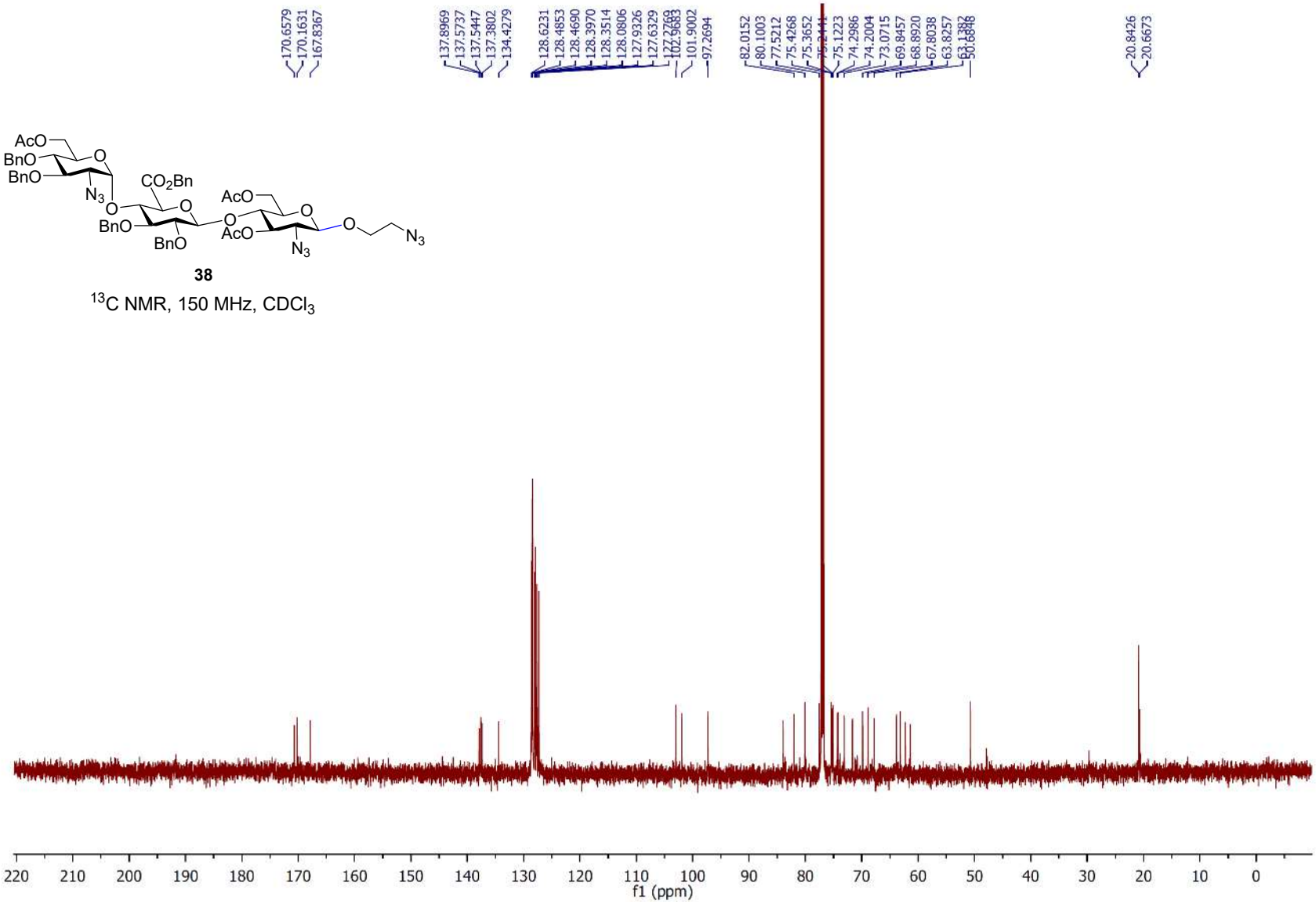

AcO
BnO
BnO
N3
O
CO2Bn
O
BnO
BnO
AcO
O
AcO
N3
O
N3
37
1H-13C HSQC, 600/150MHz, CDCl3
H3-C3 -OAc
H2-C2 -N3
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
20
30
40
50
60
70
80
90
100
110
120
130
140



38

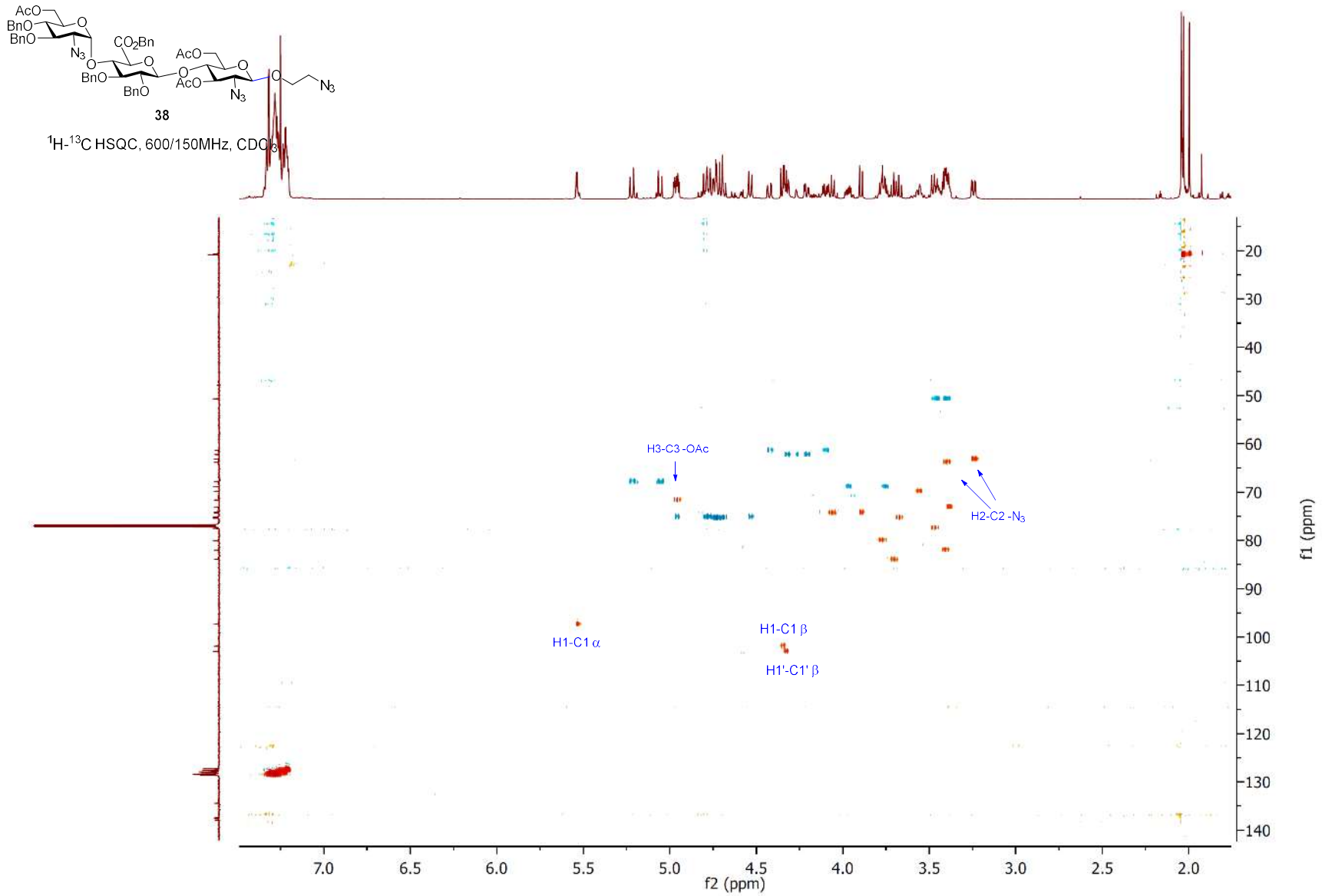
^{1}H NMR, 600 MHz, $CDCl_3$

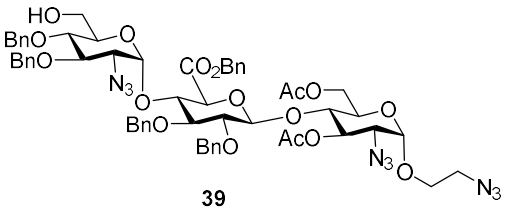




38

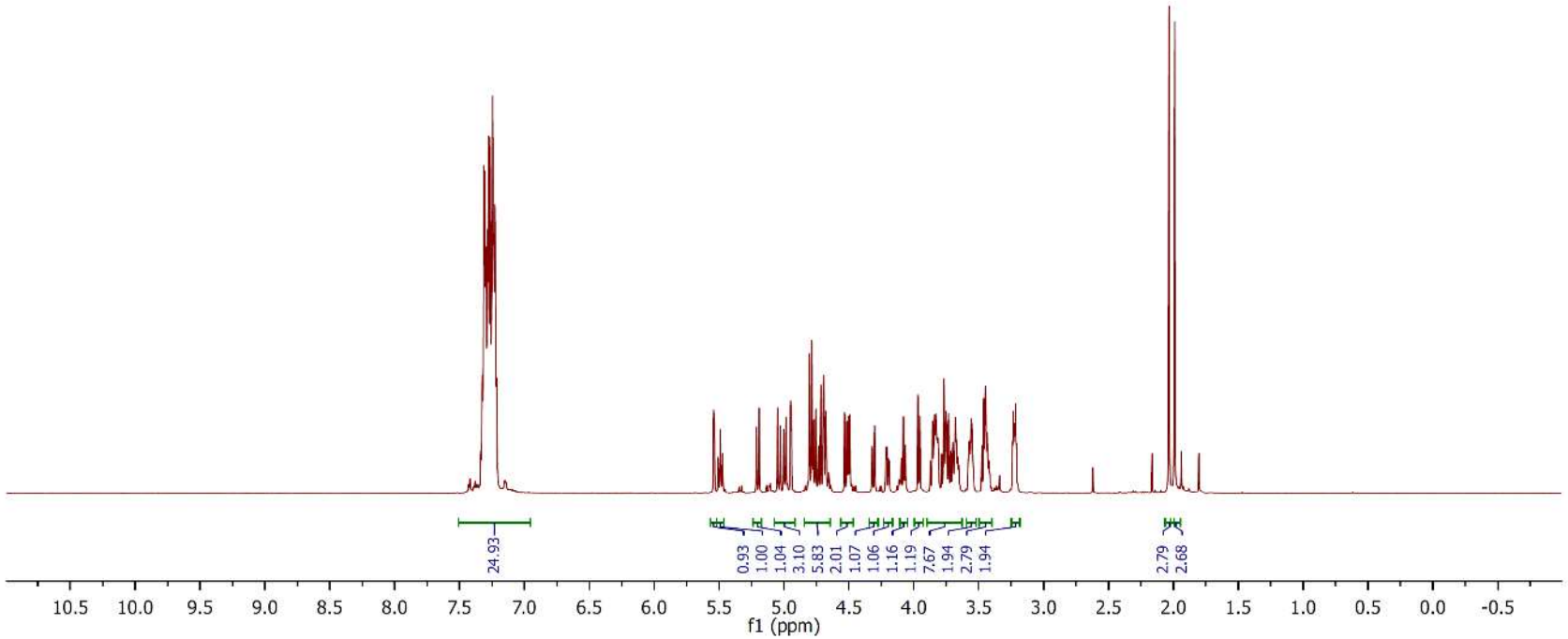
^{13}C NMR, 150 MHz, $CDCl_3$

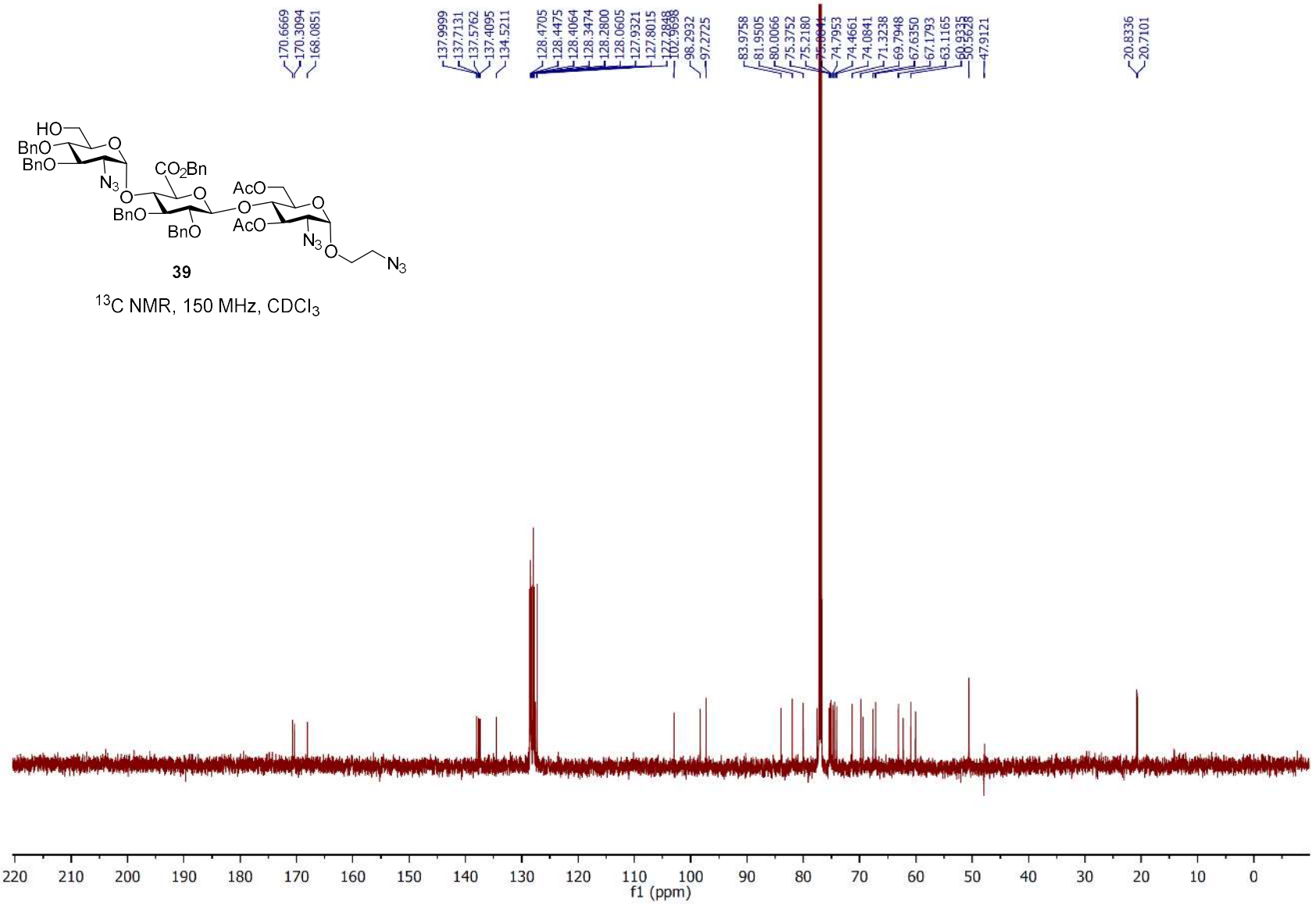

38
^{1}H-^{13}C HSQC, 600/150MHz, CDCl$_3$
H3-C3 -OAc
H2-C2 -N$_3$
H1-C1 α
H1-C1 β
H1'-C1' β
f2 (ppm)
f1 (ppm)

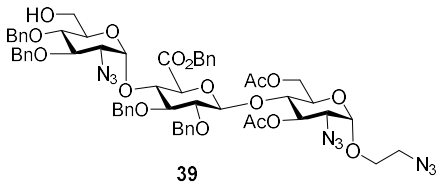


39

^{1}H NMR, 600 MHz, $CDCl_3$

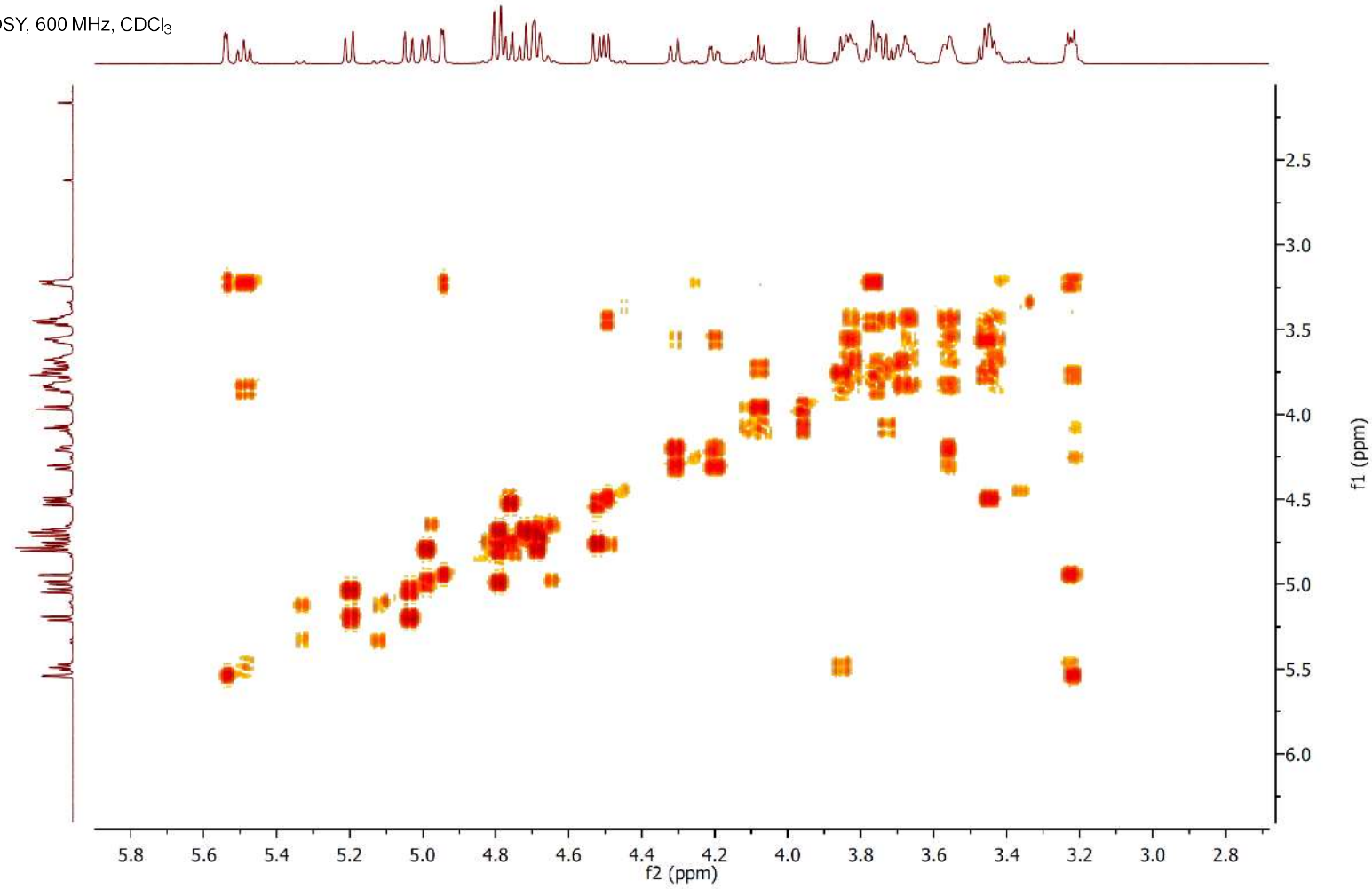


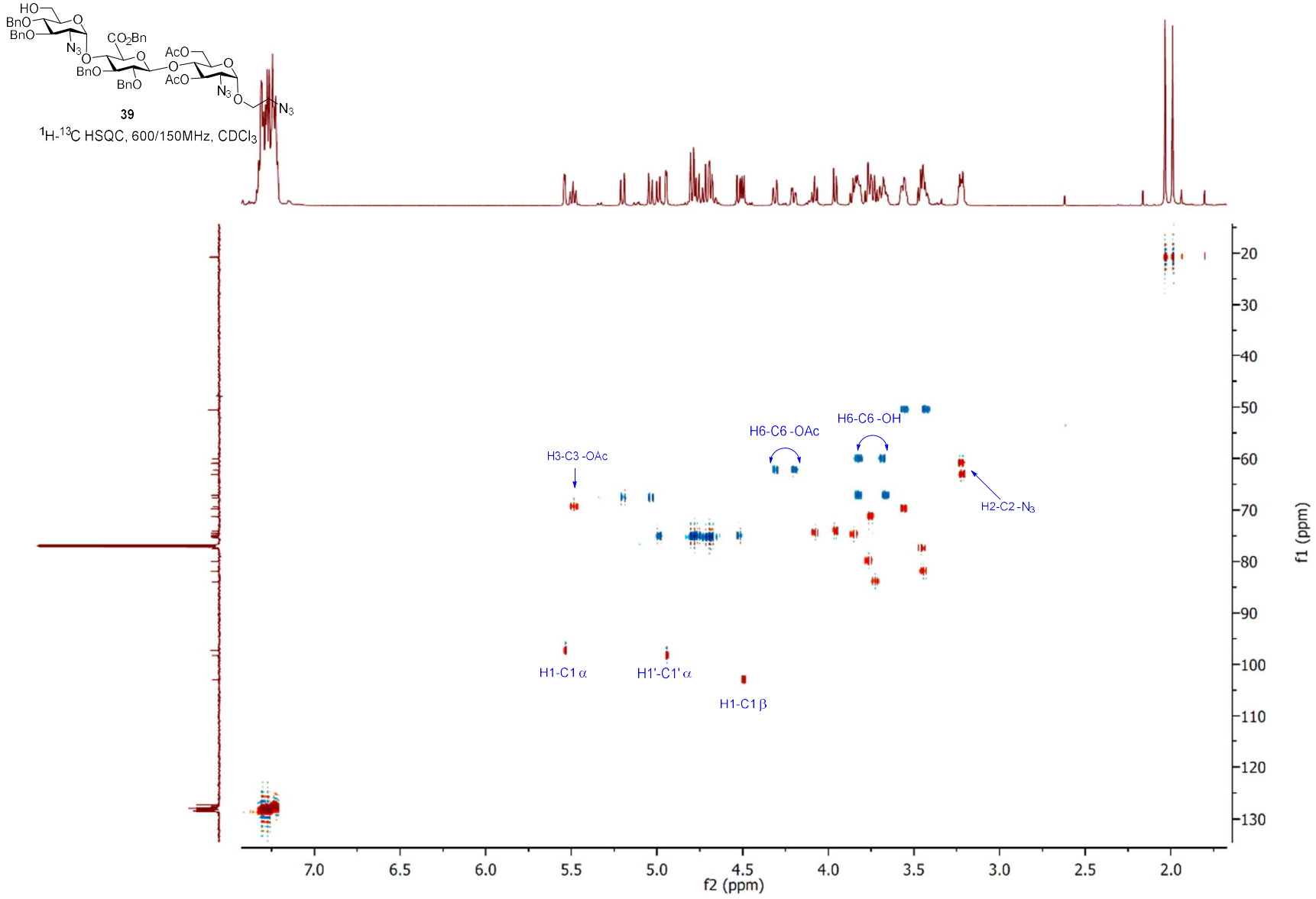

HO
BnO
BnO
O
N3
CO2Bn
O
AcO
O
BnO
BnO
O
AcO
N3
O
N3
39
13C NMR, 150 MHz, CDCl3
170.6669
170.3094
168.0851
137.9999
137.7131
137.5762
137.4095
134.5211
128.4705
128.4475
128.4064
128.3474
128.2800
128.0605
127.9321
127.8015
98.2932
97.2725
83.9758
81.9505
80.0066
75.3752
75.2180
74.7953
74.4661
74.0841
71.3238
69.7948
67.6350
67.1793
63.1165
47.9121
20.8336
20.7101
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)



39

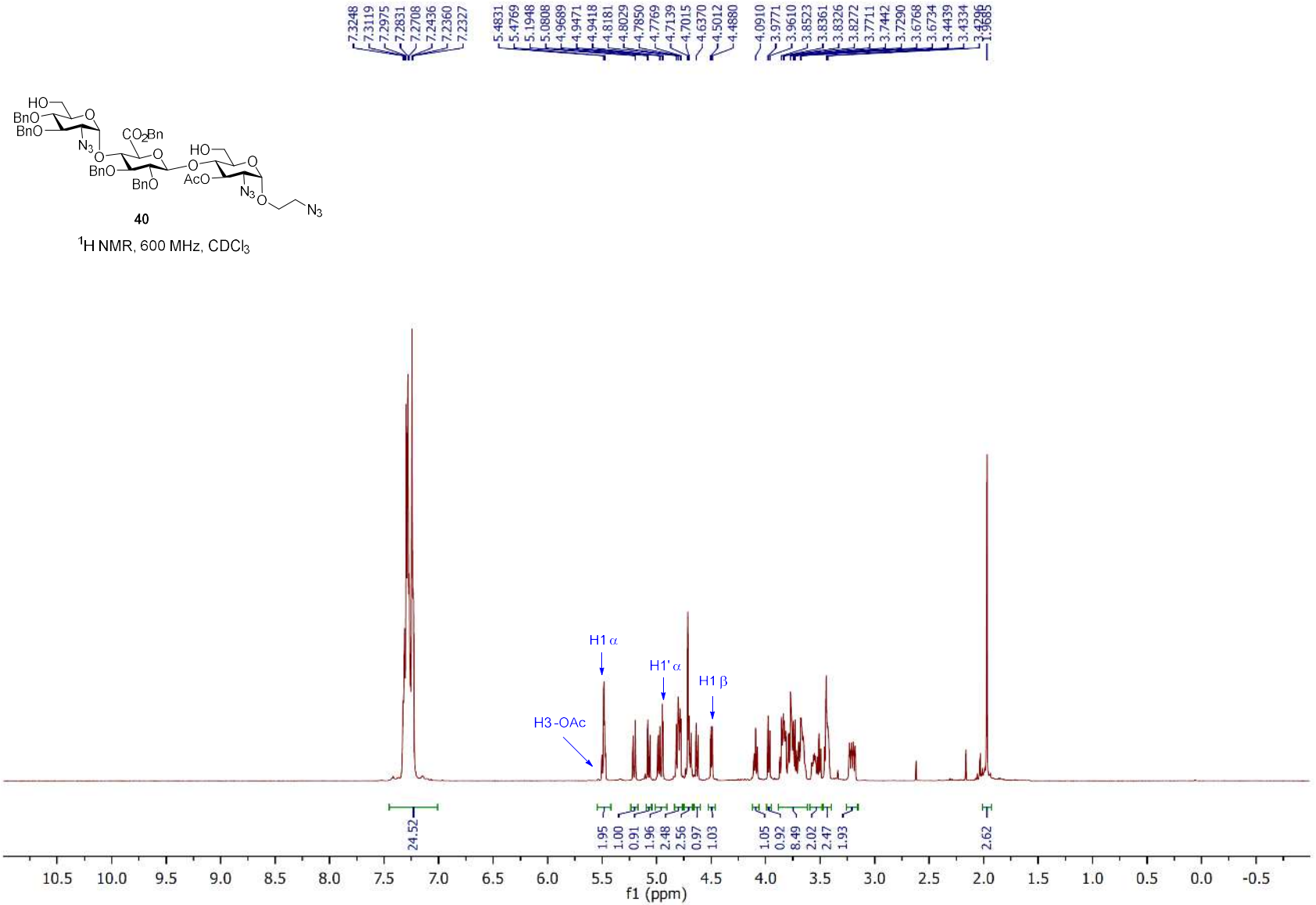
^{1}H-^{1}H COSY, 600 MHz, $CDCl_3$

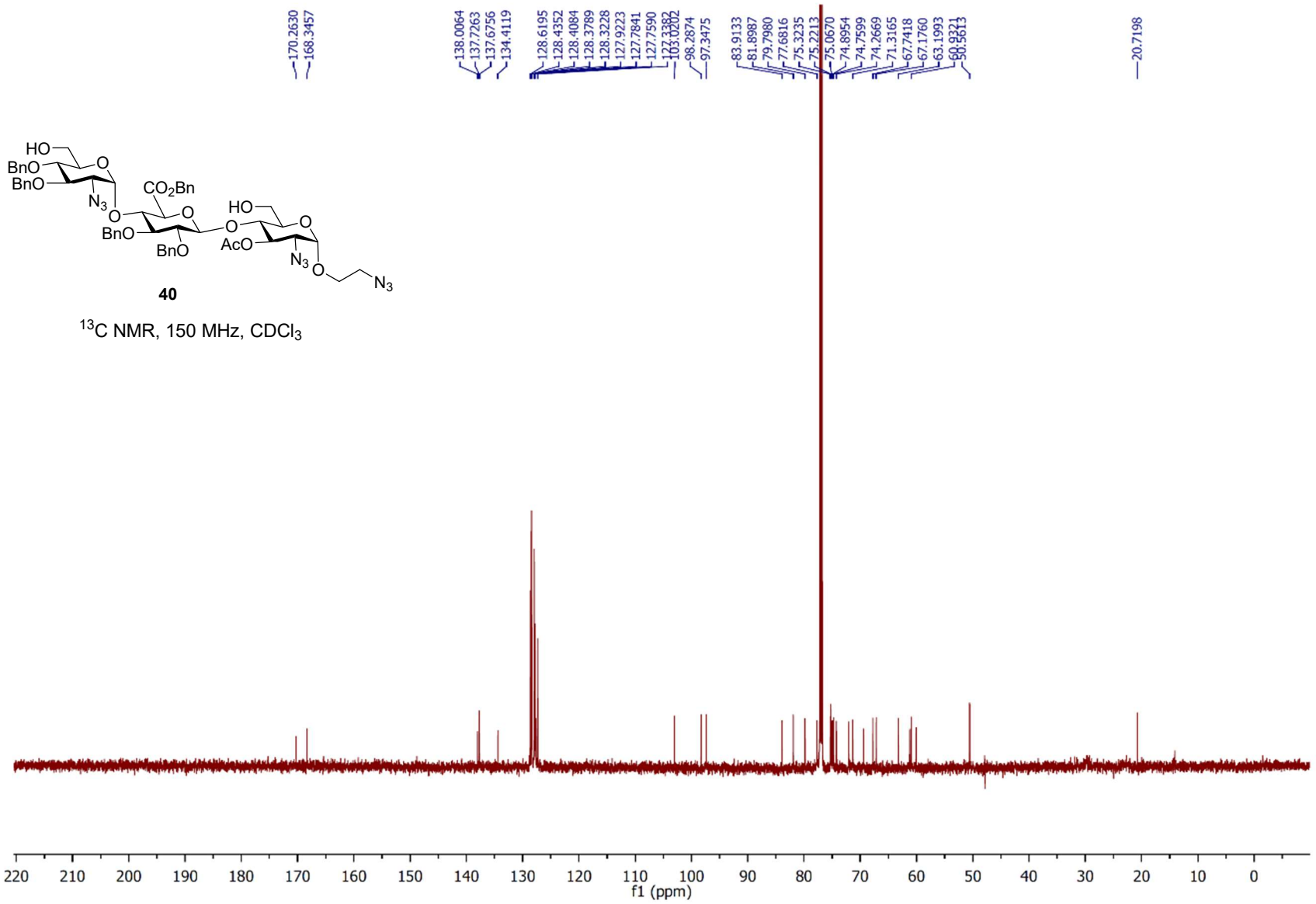


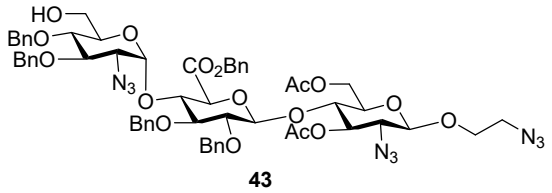

39
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$
H3-C3 -OAc
H6-C6 -OAc
H6-C6 -OH
H2-C2 -N_3
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)

40

^{1}H NMR, 600 MHz, $CDCl_3$

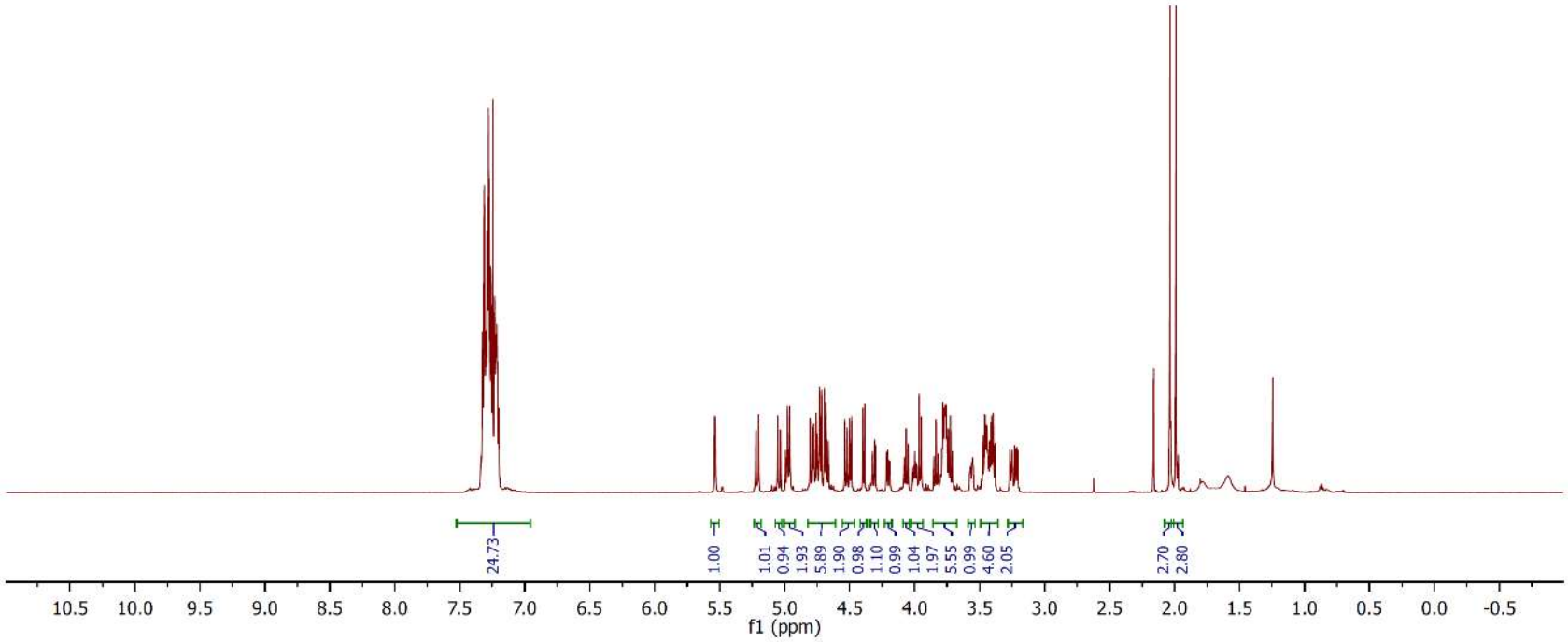


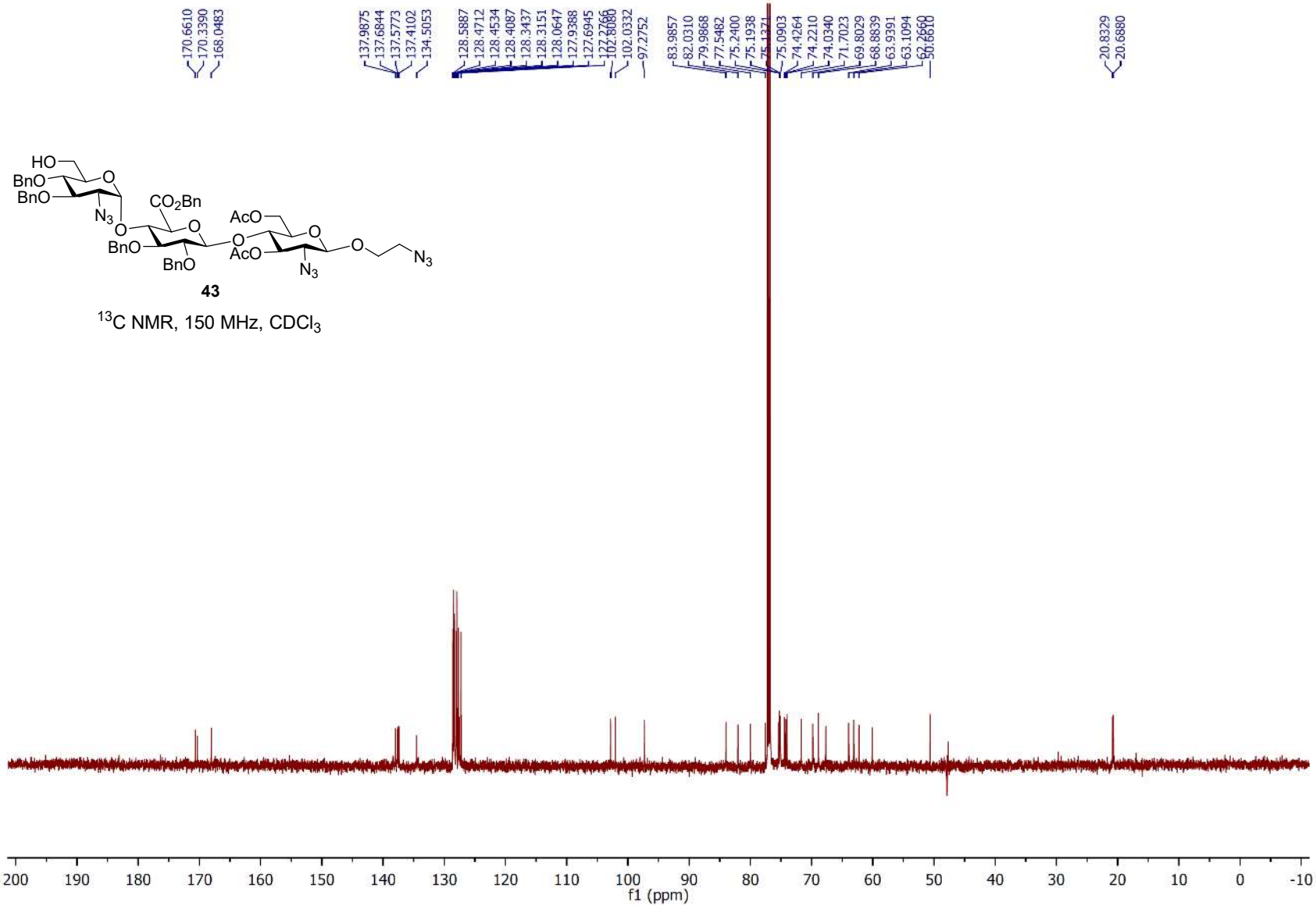

170.2630
168.3457
138.0064
137.7263
137.6756
134.4119
128.6195
128.4352
128.4084
128.3789
128.3228
127.9223
127.7841
127.7590
98.2874
97.3475
83.9133
81.8987
79.7980
77.6816
75.3235
75.2213
75.0670
74.8954
74.7599
74.2669
71.3165
67.7418
67.1760
63.1993
20.7198
HO
BnO
BnO
O
N3
CO2Bn
O
BnO
BnO
O
HO
O
O
AcO
N3
O
N3
40
13C NMR, 150 MHz, CDCl3
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)



43

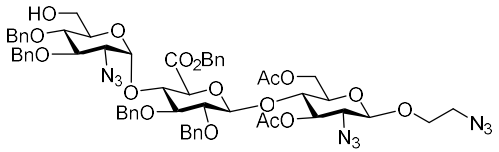
^{1}H NMR, 600 MHz, $CDCl_3$





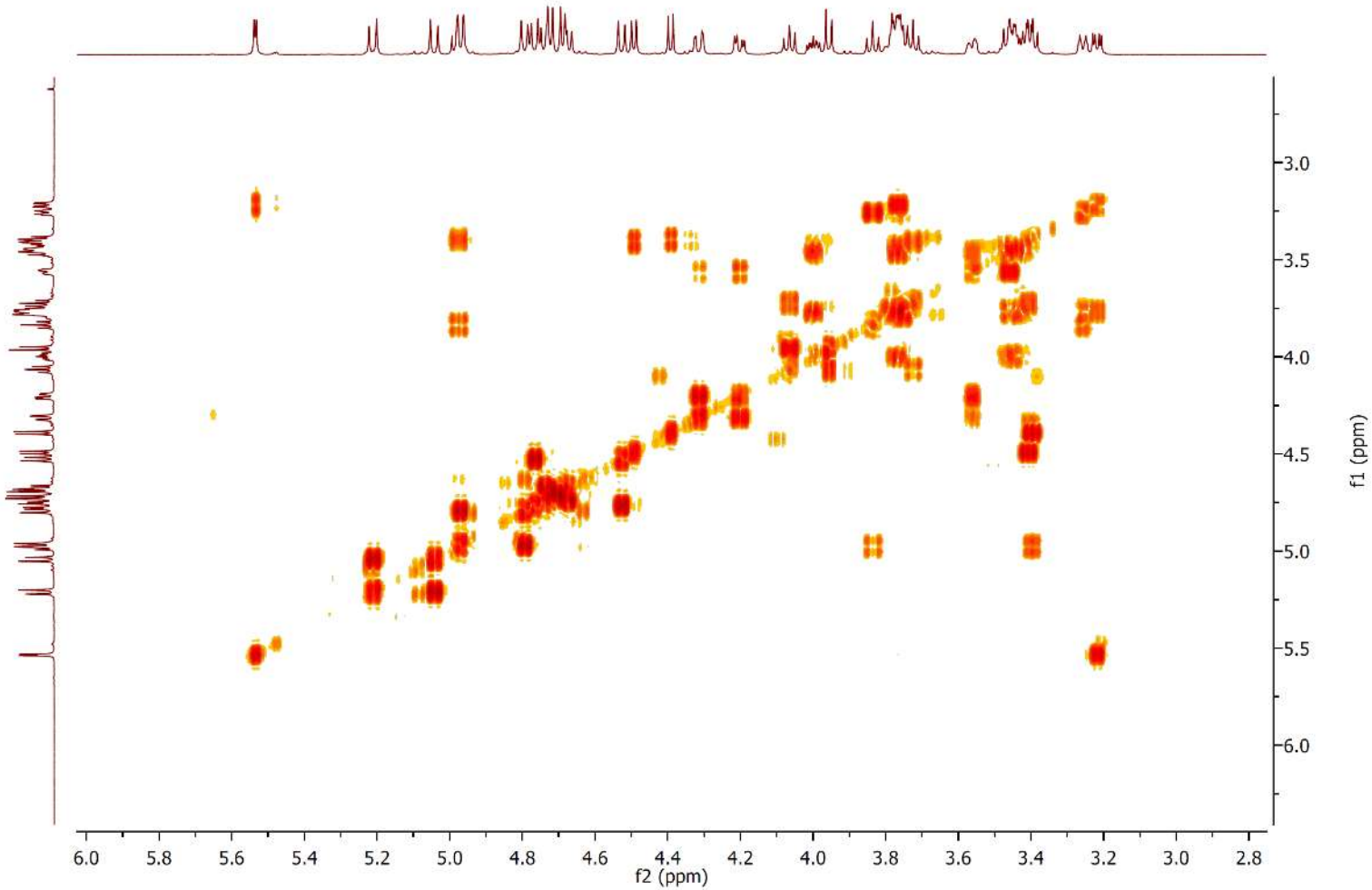
43

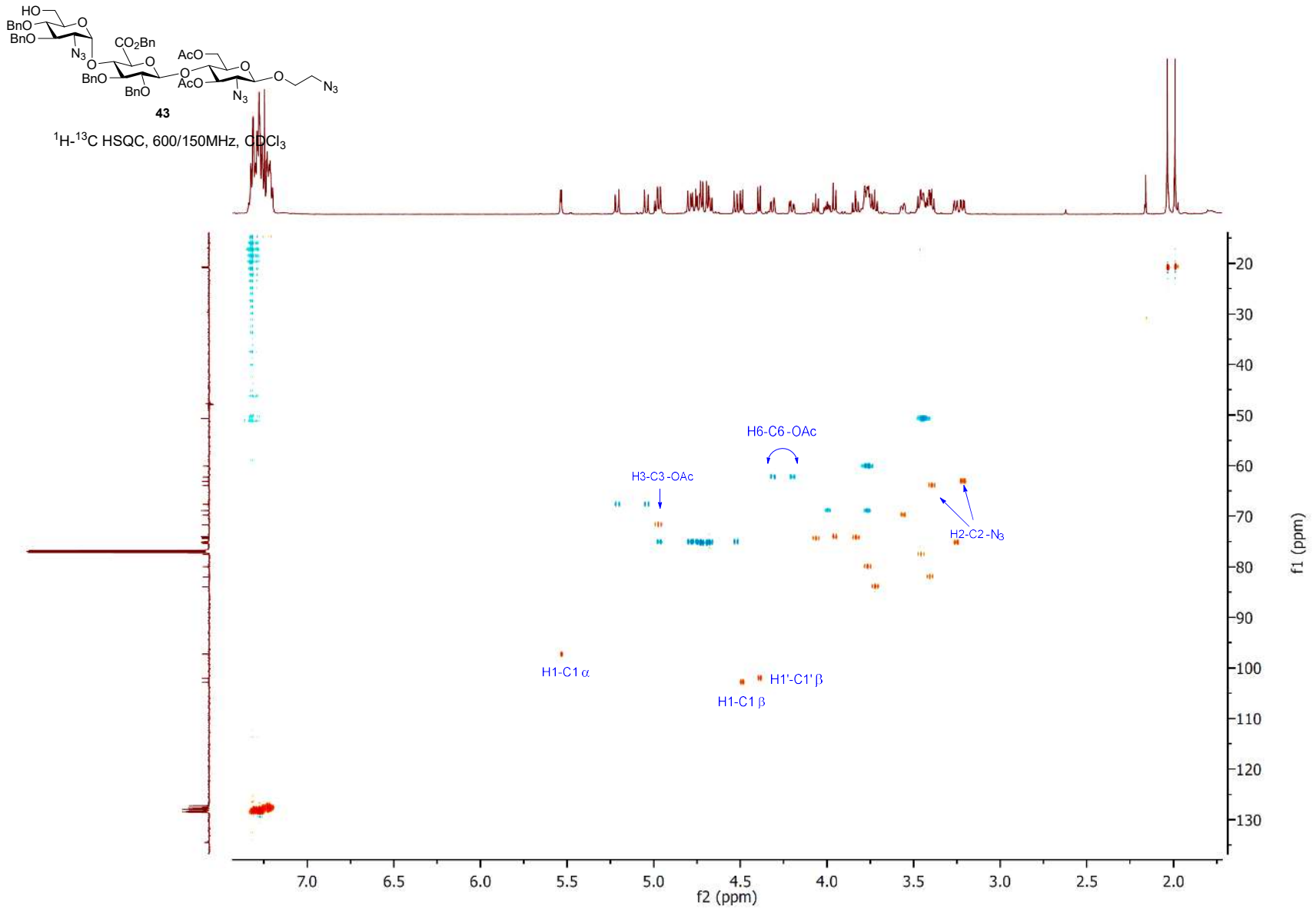
^{13}C NMR, 150 MHz, $CDCl_3$

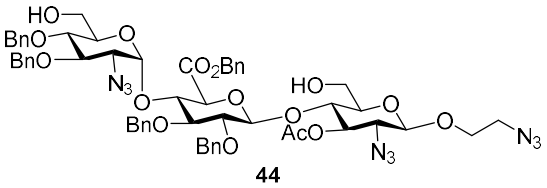


43

^{1}H-^{1}H COSY, 600 MHz, $CDCl_3$

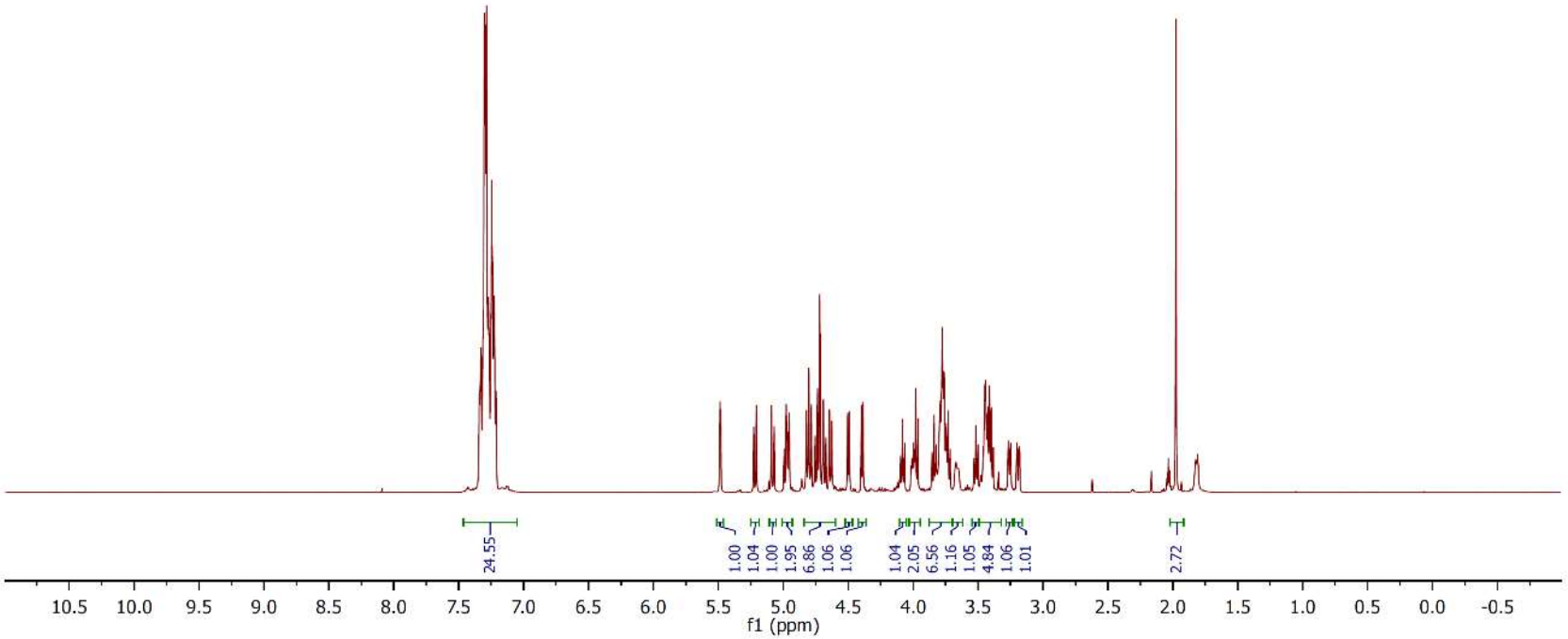


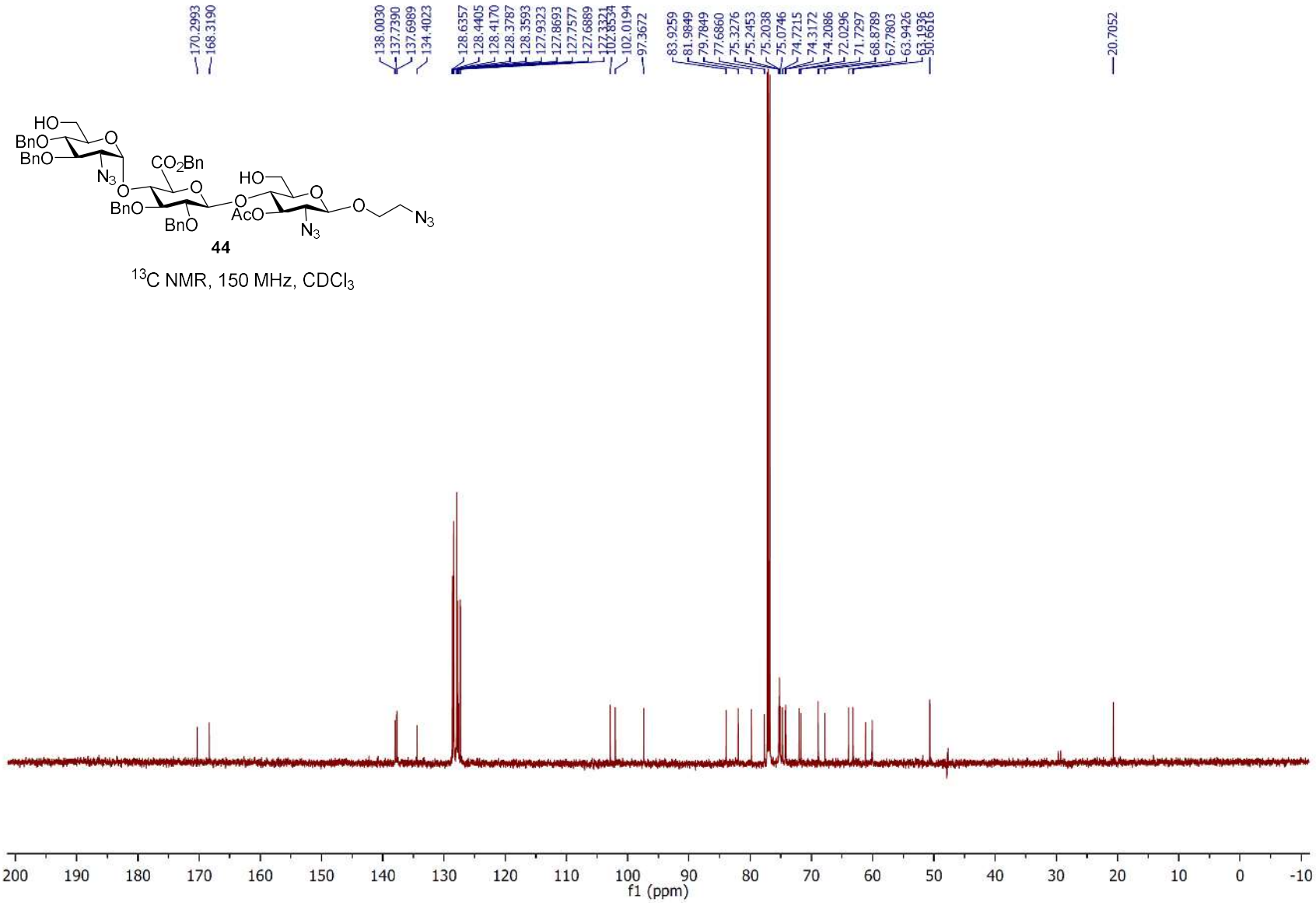

HO
BnO
BnO
O
N3
CO2Bn
O
BnO
BnO
O
AcO
AcO
O
N3
O
N3
43
1H-13C HSQC, 600/150MHz, CDCl3
H6-C6-OAc
H3-C3-OAc
H2-C2-N3
H1-C1 α
H1'-C1' β
H1-C1 β
20
30
40
50
60
70
80
90
100
110
120
130
f1 (ppm)
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
f2 (ppm)

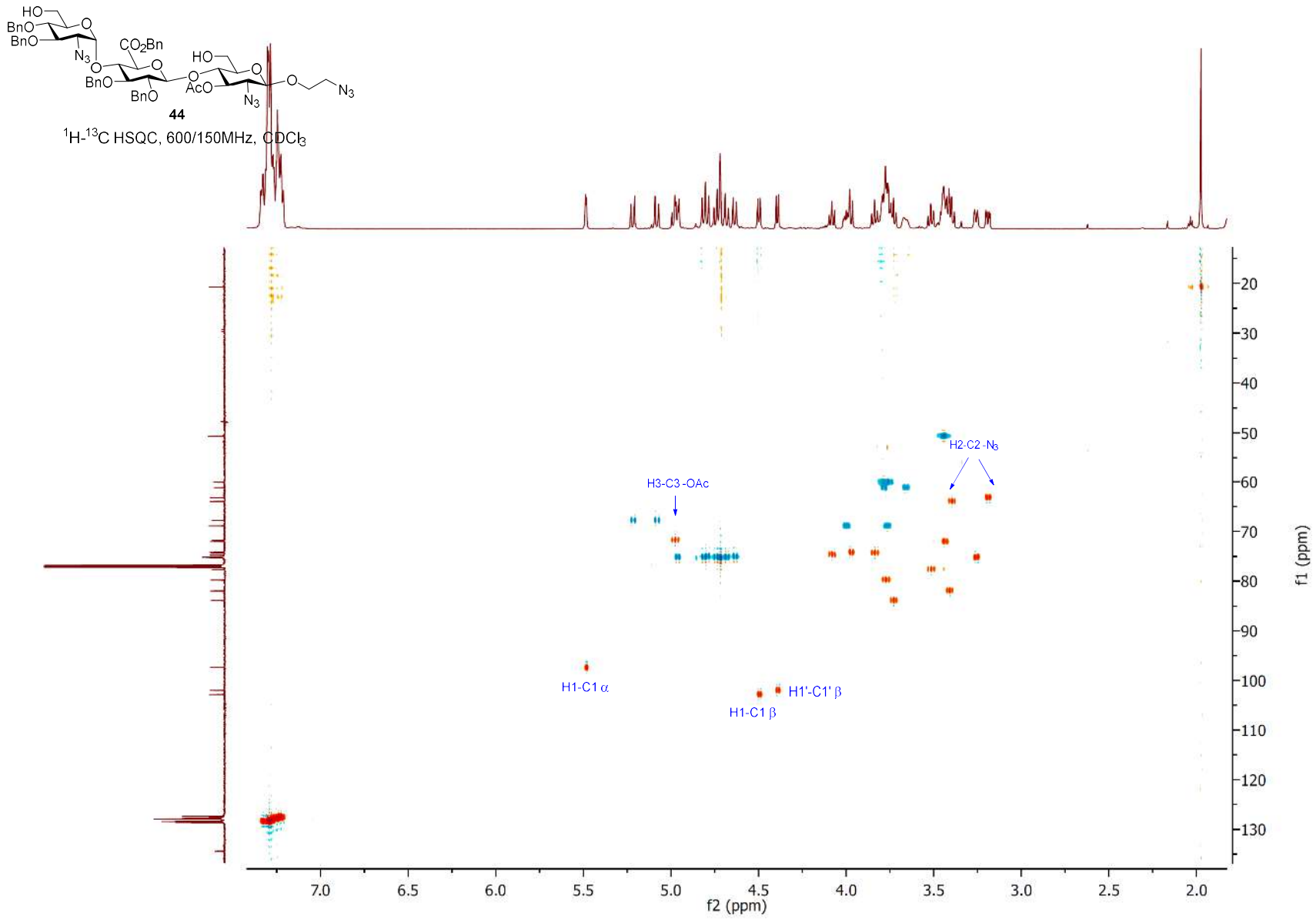


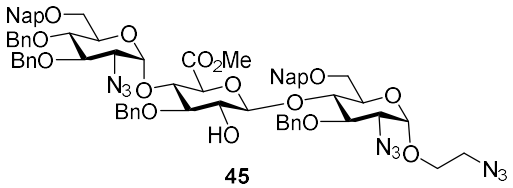
44

^{1}H NMR, 600 MHz, $CDCl_3$



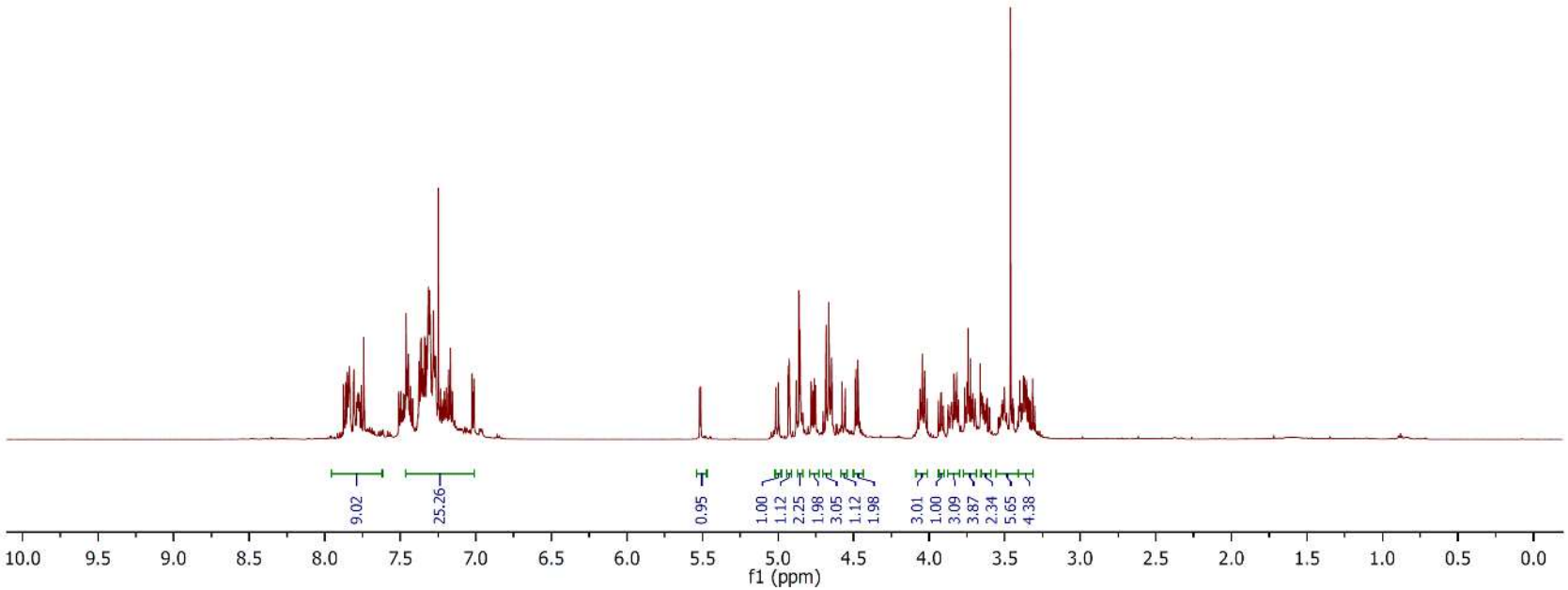

170.2993
168.3190
138.0030
137.7390
137.6989
134.4023
128.6357
128.4405
128.4170
128.3787
128.3593
127.9323
127.8693
127.7577
127.6889
102.0194
97.3672
83.9259
81.9849
79.7849
77.6860
75.3276
75.2453
75.2038
75.0746
74.7215
74.3172
74.2086
72.0296
71.7297
68.8789
67.7803
63.9426
20.7052
HO
BnO
BnO
O
N3
CO2Bn
O
BnO
BnO
O
HO
O
AcO
N3
O
N3
44
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

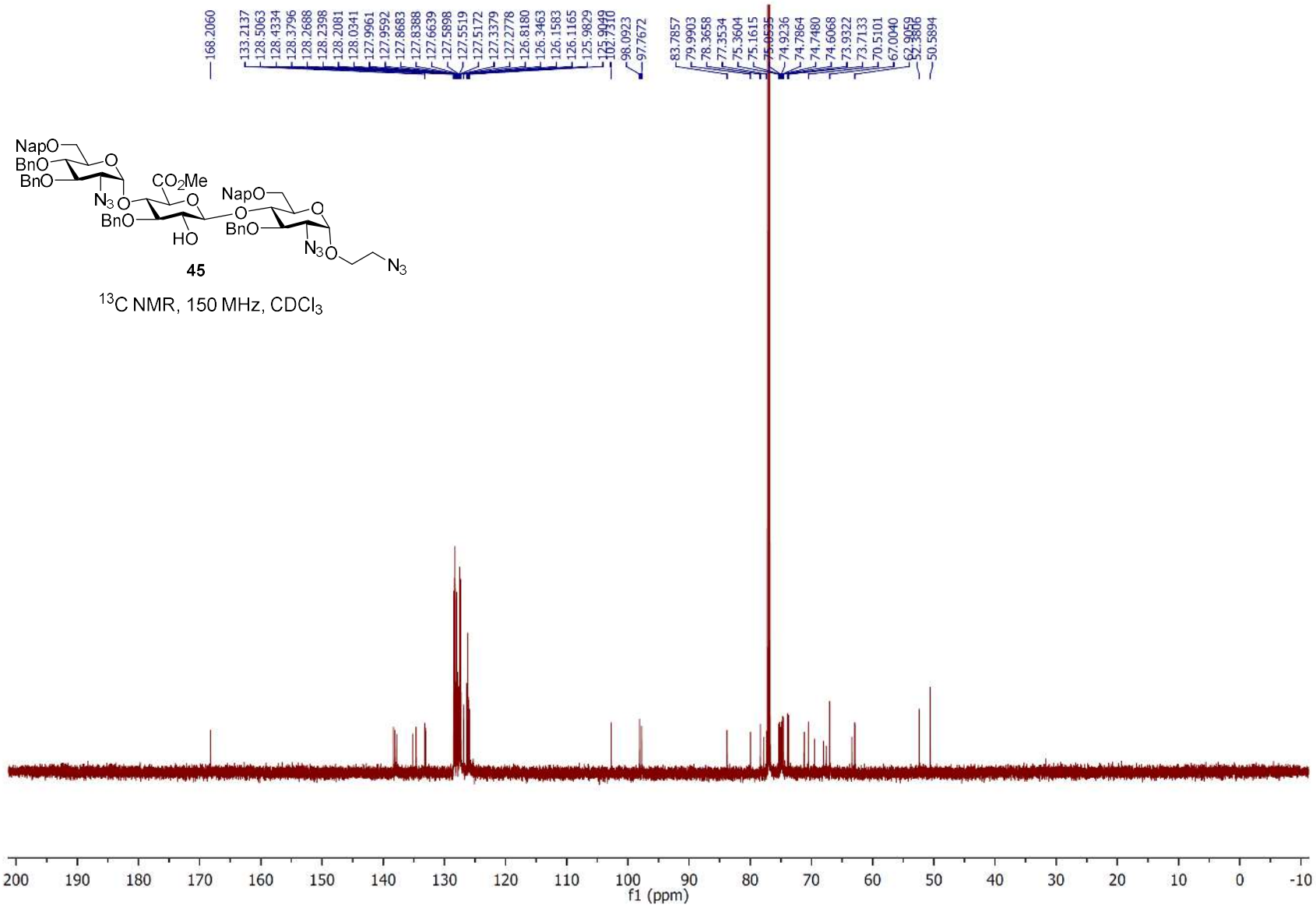

HO
BnO
BnO
O
N3
CO2Bn
O
BnO
BnO
HO
O
O
AcO
N3
O
N3
44
1H-13C HSQC, 600/150MHz, CDCl3
H3-C3-OAc
H2-C2-N3
H1-C1 α
H1-C1 β
H1'-C1' β
f2 (ppm)
f1 (ppm)

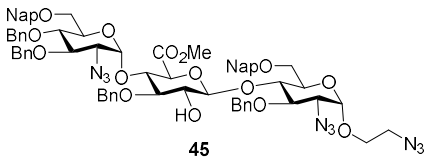


45

^{1}H NMR, 600 MHz, $CDCl_3$

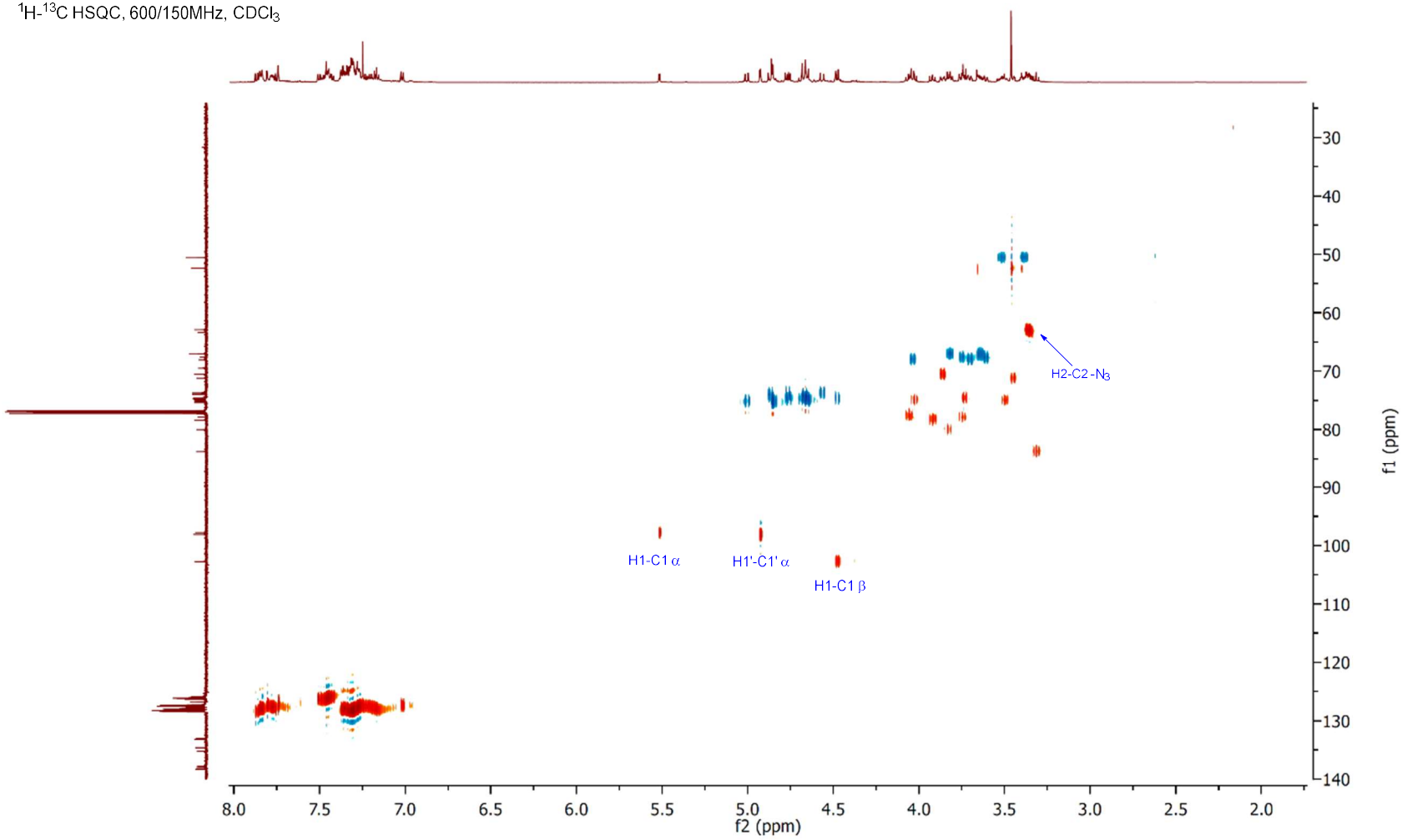


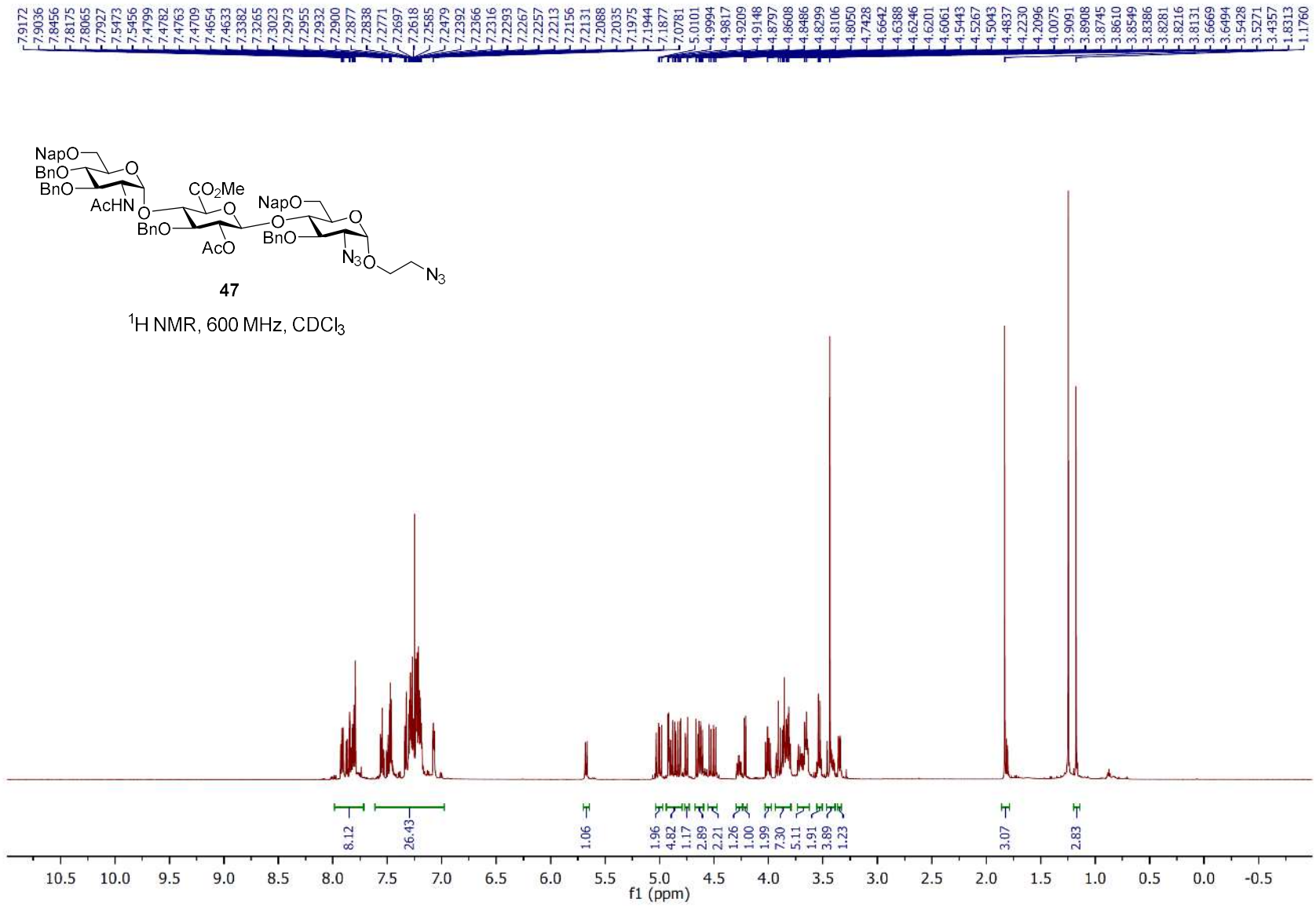

NapO
BnO
BnO
O
N3
O
CO2Me
O
BnO
HO
NapO
O
O
BnO
N3
O
N3
45
13C NMR, 150 MHz, CDCl3
168.2060
133.2137
128.5063
128.4334
128.3796
128.2688
128.2398
128.2081
128.0341
127.9961
127.9592
127.8683
127.8388
127.6639
127.5898
127.5519
127.5172
127.3379
127.2778
126.8180
126.3463
126.1583
126.1165
125.9829
102.7310
98.0923
97.7672
83.7857
79.9903
78.3658
77.3534
75.3604
75.1615
74.9236
74.7864
74.7480
74.6068
73.9322
73.7133
70.5101
67.0040
50.5894
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

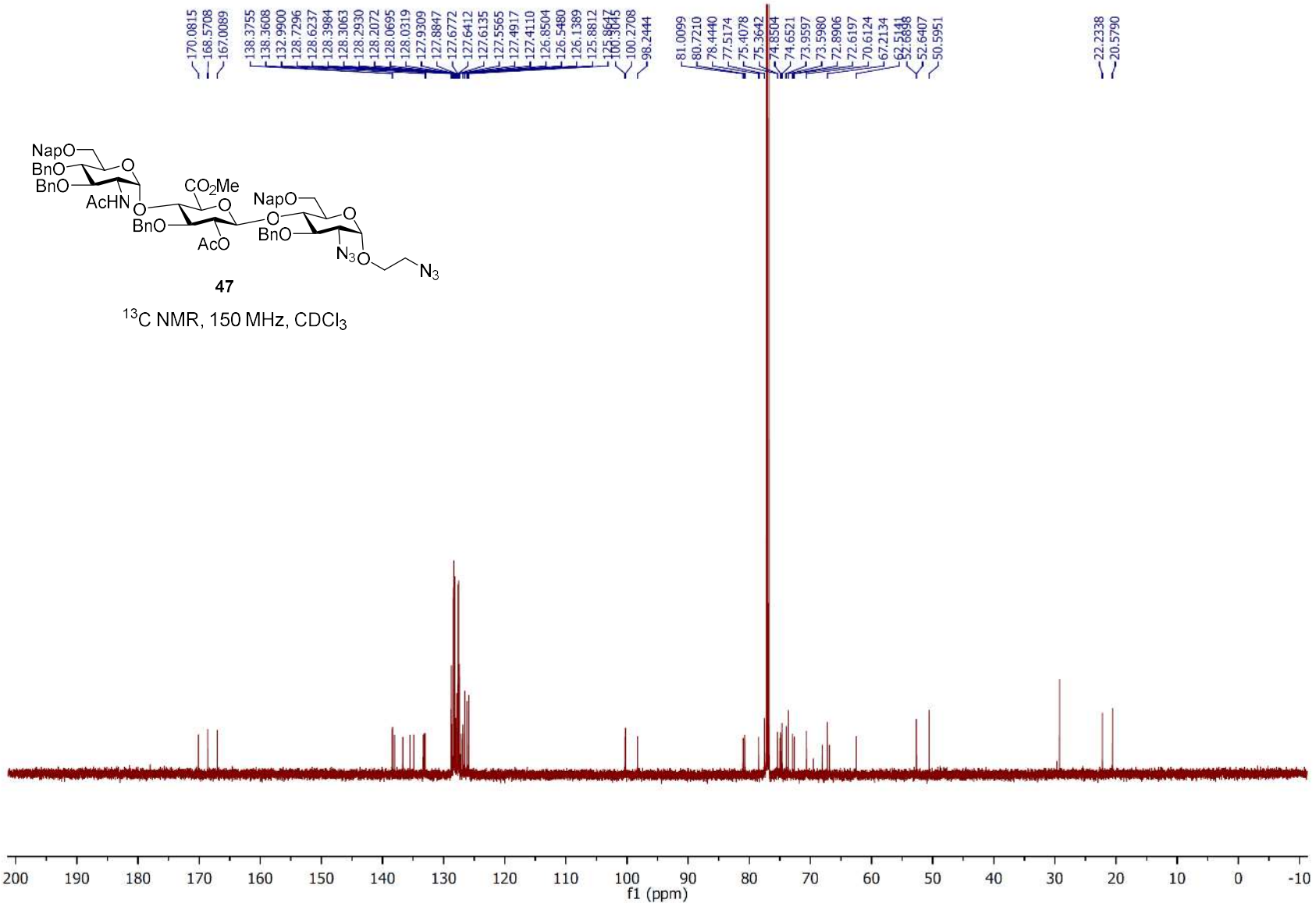


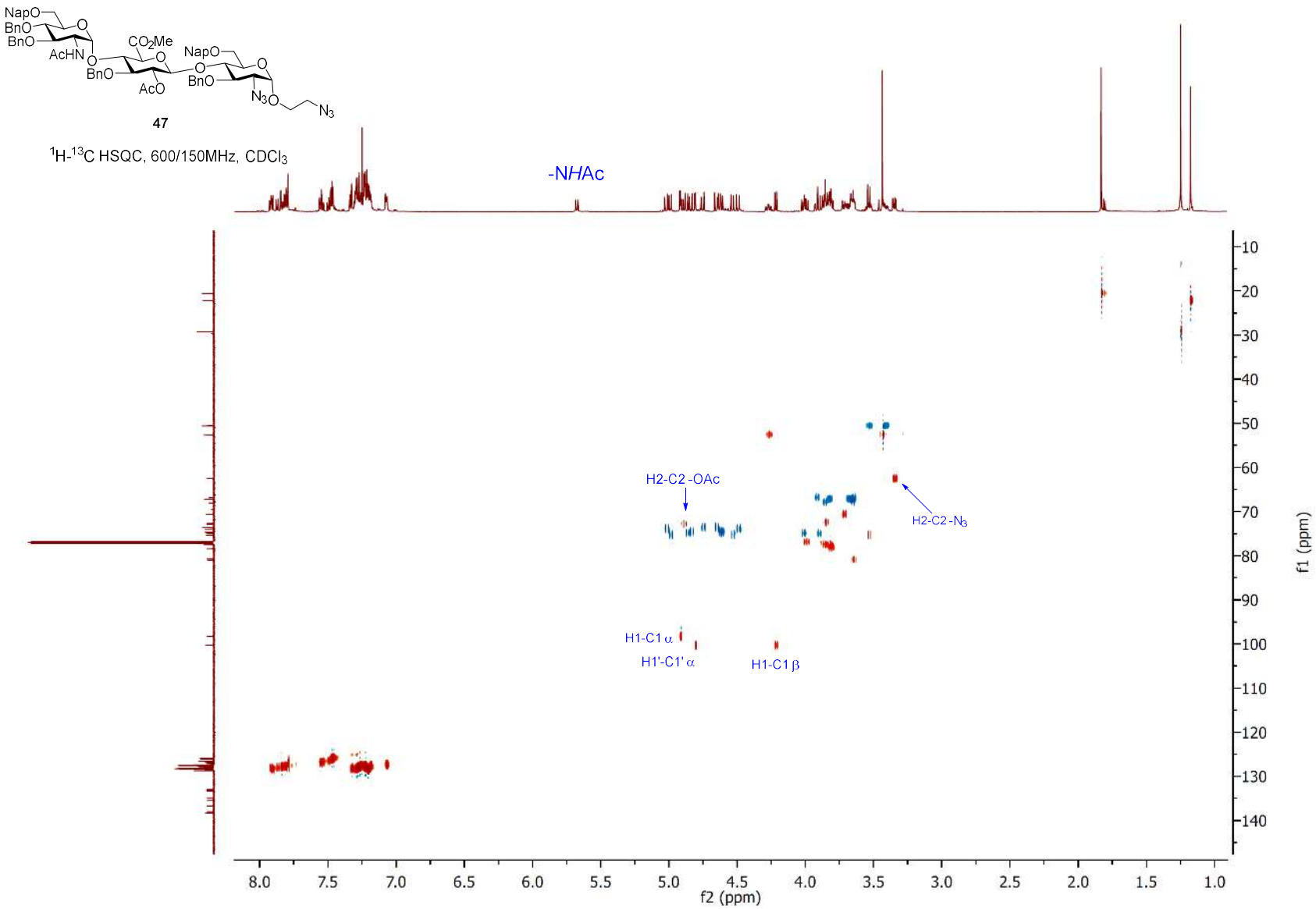
45

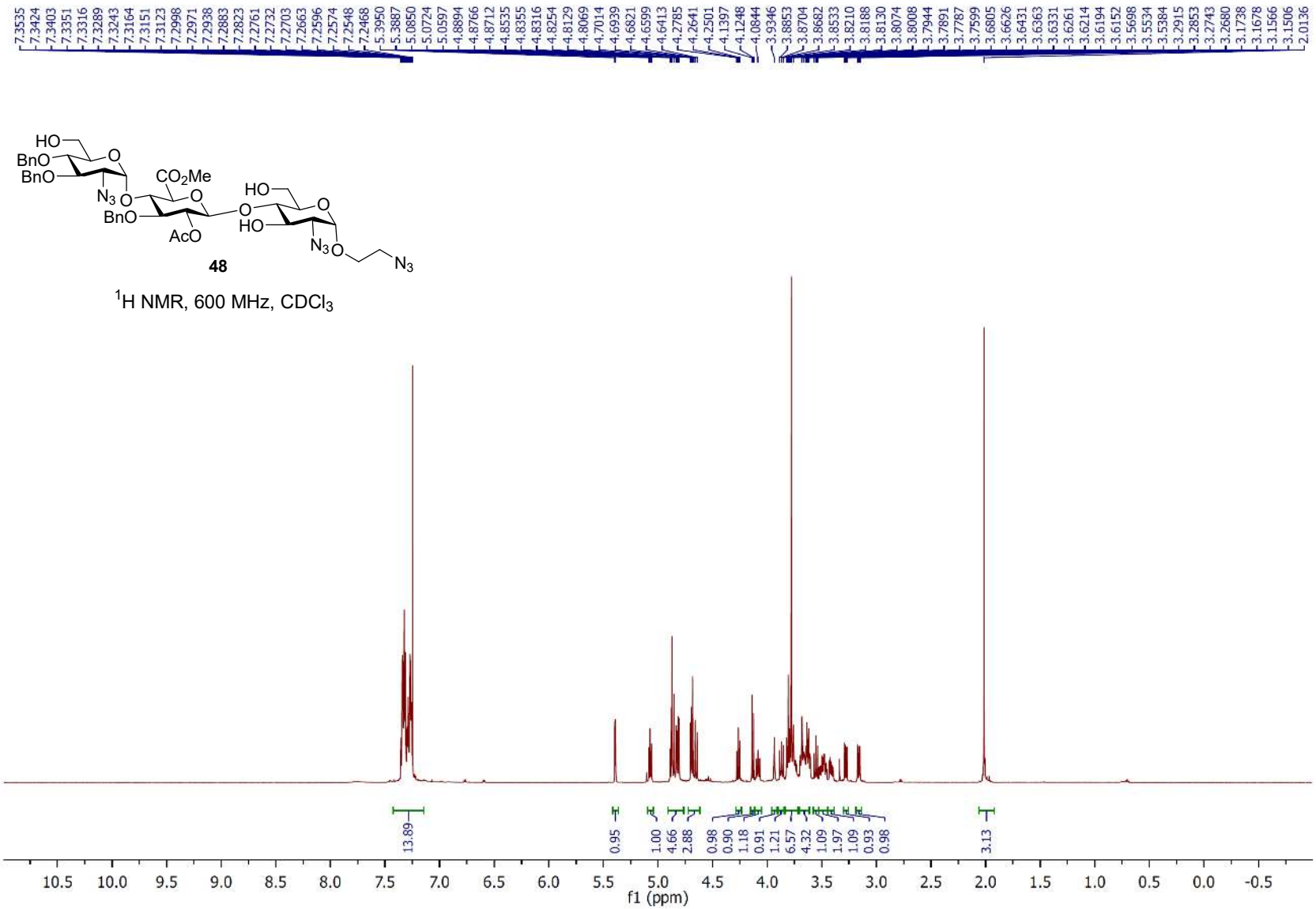
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

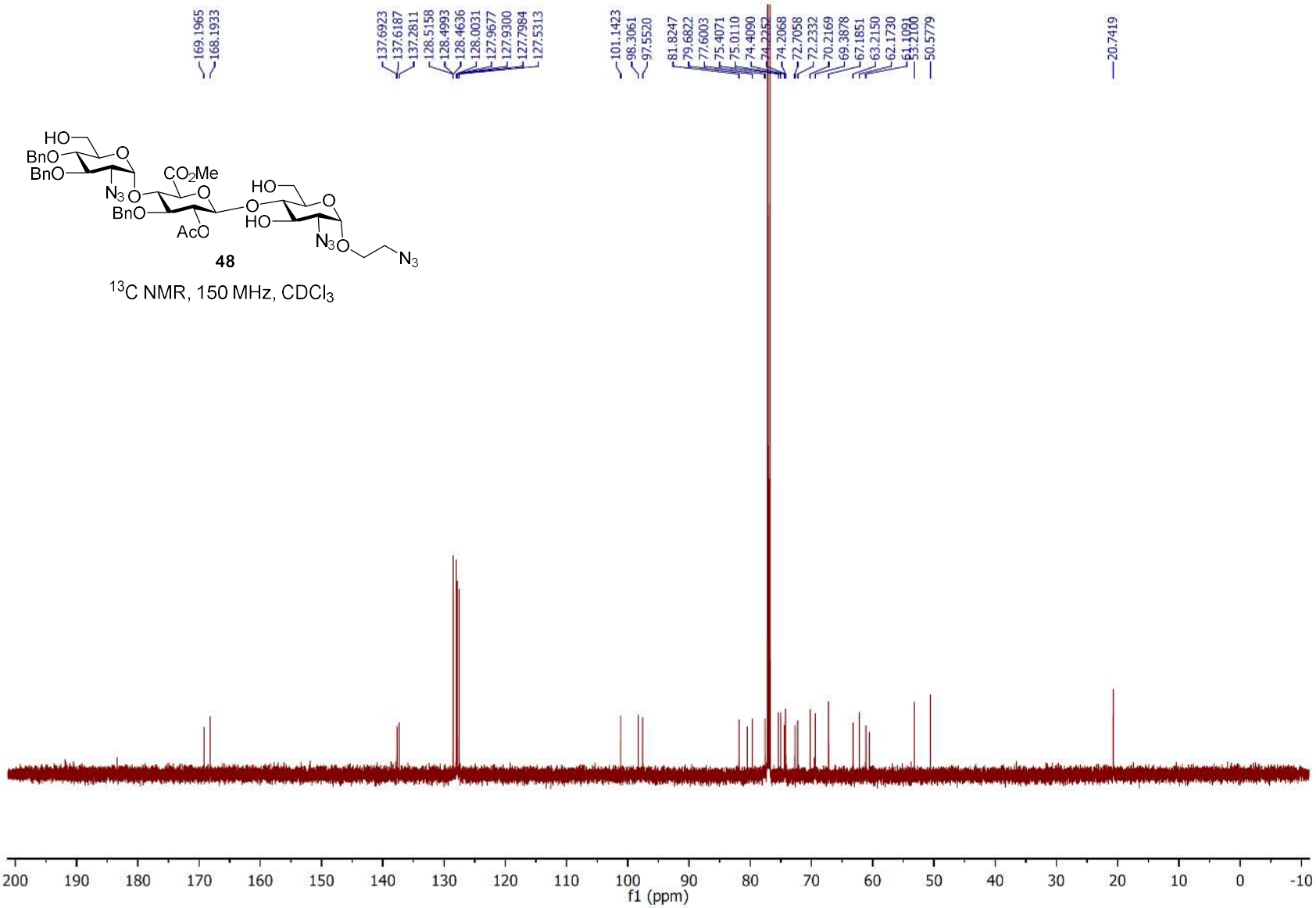


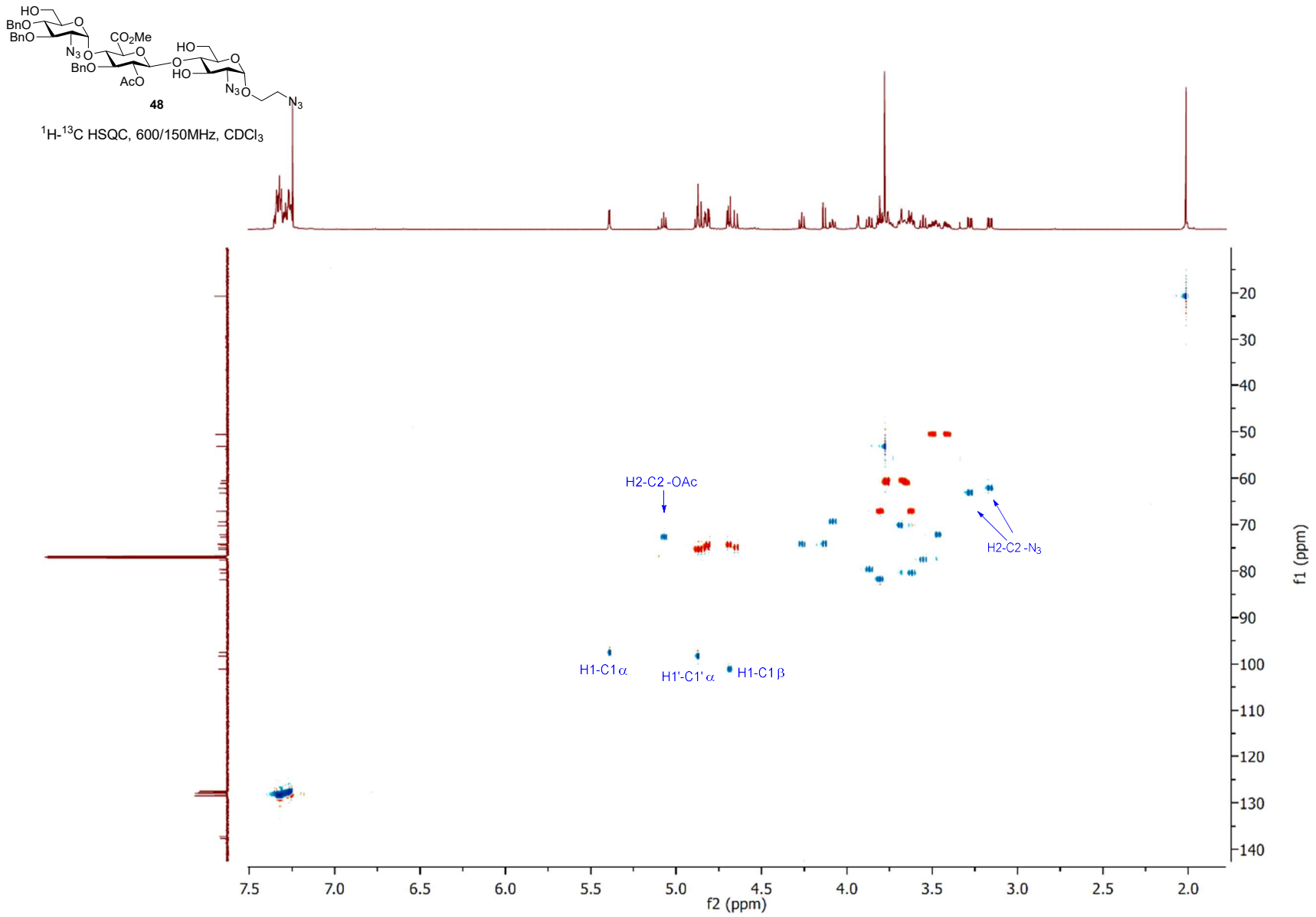

NapO
BnO
BnO
AcHN
O
BnO
AcO
CO2Me
NapO
BnO
N3O
N3
47
1H NMR, 600 MHz, CDCl3
f1 (ppm)

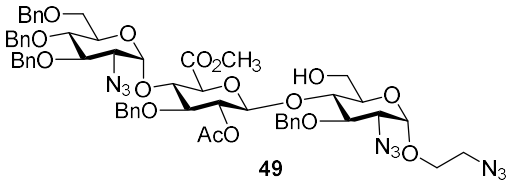

170.0815
168.5708
167.0089
138.3755
138.3608
132.9900
128.7296
128.6237
128.3984
128.3063
128.2930
128.2072
128.0695
128.0319
127.9309
127.8847
127.6772
127.6412
127.6135
127.5565
127.4917
127.4110
126.8504
126.5480
126.1389
125.8812
100.2708
98.2444
81.0099
80.7210
78.4440
77.5174
75.4078
75.3642
74.8504
74.6521
73.9597
73.5980
72.8906
72.6197
70.6124
67.2134
52.6407
50.5951
22.2338
20.5790
NapO
BnO
BnO
AcHN
CO2Me
BnO
AcO
NapO
BnO
N3
N3
47
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


NapO
BnO
BnO
AcHN
CO2Me
BnO
AcO
NapO
BnO
N3O
N3
47
1H-13C HSQC, 600/150MHz, CDCl3
-NHAc
H2-C2-OAc
H2-C2-N3
H1-C1α
H1'-C1'α
H1-C1β
f2 (ppm)
f1 (ppm)


7.3535
7.3424
7.3403
7.3351
7.3316
7.3289
7.3243
7.3164
7.3151
7.3123
7.2998
7.2971
7.2938
7.2883
7.2823
7.2761
7.2732
7.2703
7.2663
7.2596
7.2574
7.2548
7.2468
5.3950
5.3887
5.0850
5.0724
5.0597
4.8894
4.8766
4.8712
4.8535
4.8355
4.8316
4.8254
4.8129
4.8069
4.7014
4.6939
4.6821
4.6599
4.6413
4.2785
4.2641
4.2501
4.1397
4.1248
4.0844
3.9346
3.8853
3.8704
3.8682
3.8533
3.8210
3.8188
3.8130
3.8074
3.8008
3.7944
3.7891
3.7787
3.7599
3.6805
3.6626
3.6431
3.6363
3.6331
3.6261
3.6214
3.6194
3.6152
3.5698
3.5534
3.5384
3.2915
3.2853
3.2743
3.2680
3.1738
3.1678
3.1566
3.1506
2.0136
HO
BnO
BnO
O
N3
O
CO2Me
O
BnO
AcO
O
HO
HO
O
N3
O
N3
48
1H NMR, 600 MHz, CDCl3
13.89
0.95
1.00
4.66
2.88
0.98
0.90
1.18
0.91
1.21
6.57
4.32
1.09
1.97
1.09
0.93
0.98
3.13
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
f1 (ppm)

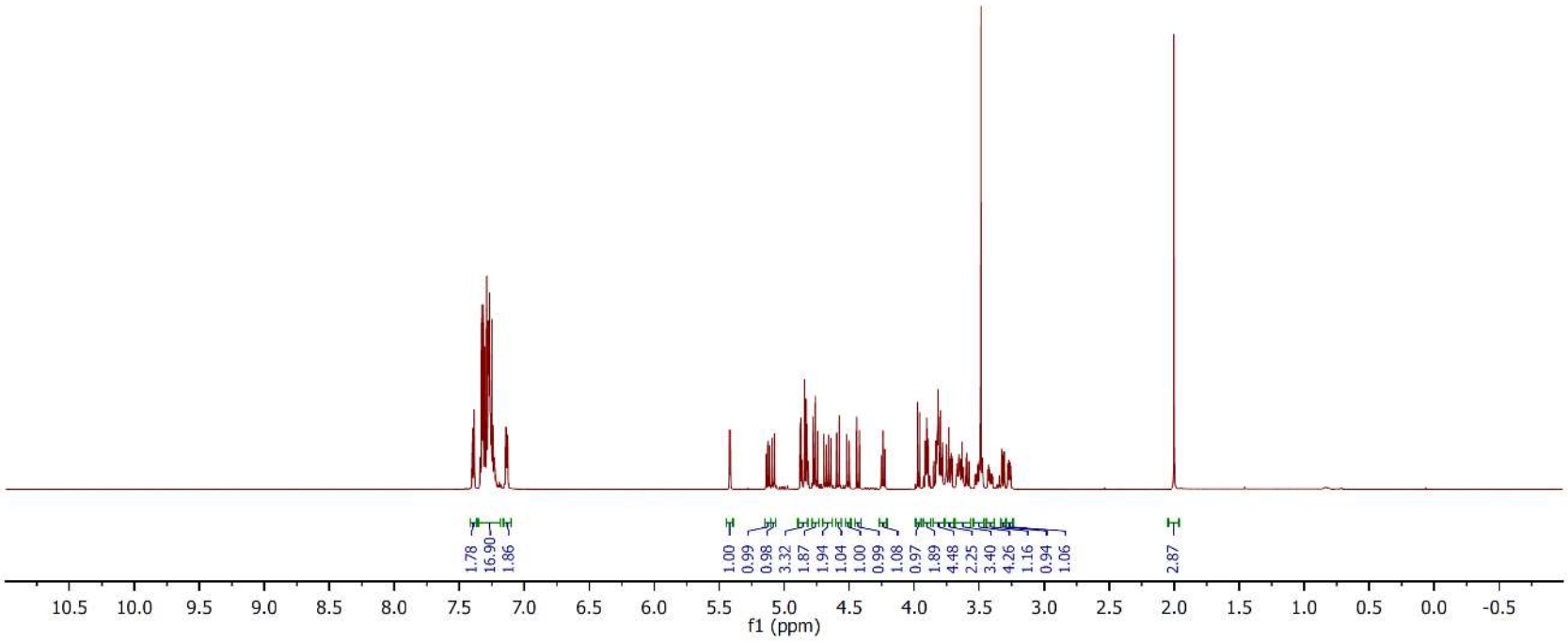

169.1965
168.1933
137.6923
137.6187
137.2811
128.5158
128.4993
128.4636
128.0031
127.9677
127.9300
127.7984
127.5313
101.1423
98.3061
97.5520
81.8247
79.6822
77.6003
75.4071
75.0110
74.4090
74.2252
74.2068
72.7058
72.2332
70.2169
69.3878
67.1851
63.2150
62.1730
51.1091
50.5779
20.7419
HO
BnO
BnO
O
CO2Me
N3
O
O
BnO
AcO
O
HO
HO
O
N3
O
N3
48
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

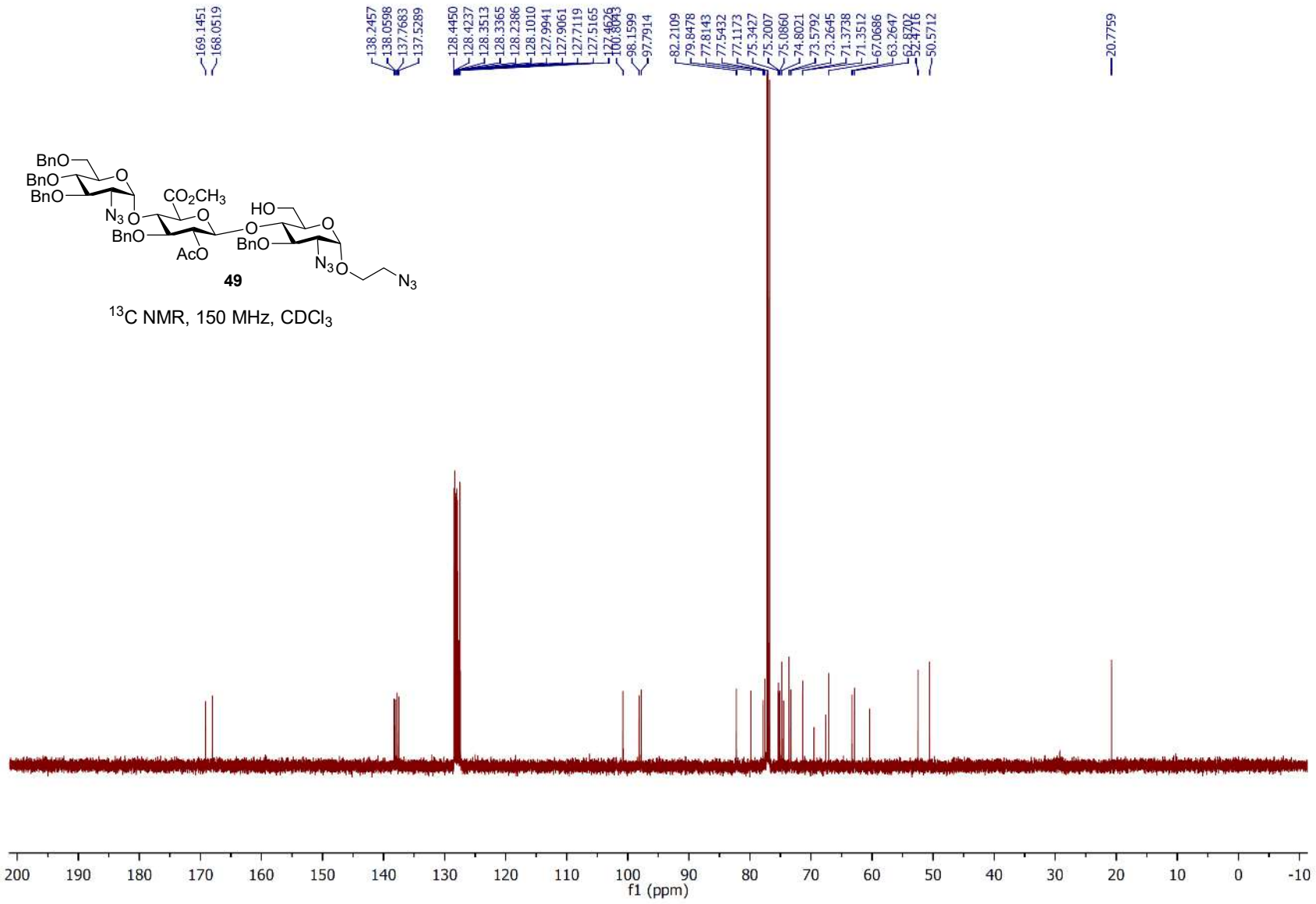

48
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$
H2-C2 -OAc
H2-C2 -N_3
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)

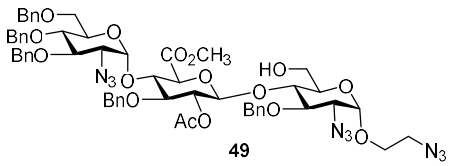


49

^{1}H NMR, 600 MHz, $CDCl_3$

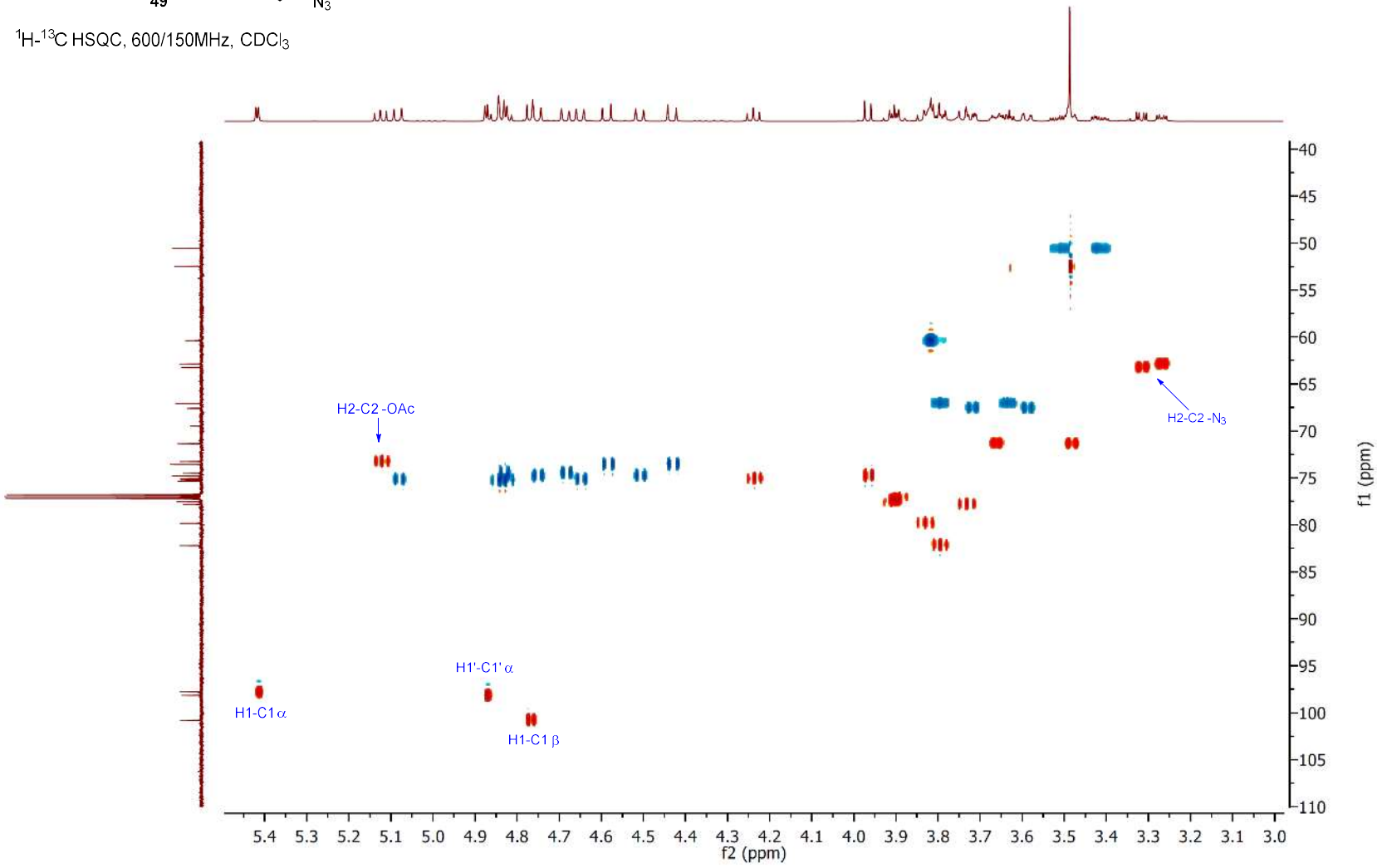



169.1451
168.0519
138.2457
138.0598
137.7683
137.5289
128.4450
128.4237
128.3513
128.3365
128.2386
128.1010
127.9941
127.9061
127.7119
127.5165
127.4626
100.8043
98.1599
97.7914
82.2109
79.8478
77.8143
77.5432
77.1173
75.3427
75.2007
75.0860
74.8021
73.5792
73.2645
71.3738
71.3512
67.0686
63.2647
62.8702
52.4716
50.5712
20.7759
BnO
BnO
BnO
O
N3
O
CO2CH3
O
BnO
AcO
HO
O
O
BnO
N3
O
N3
49
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)



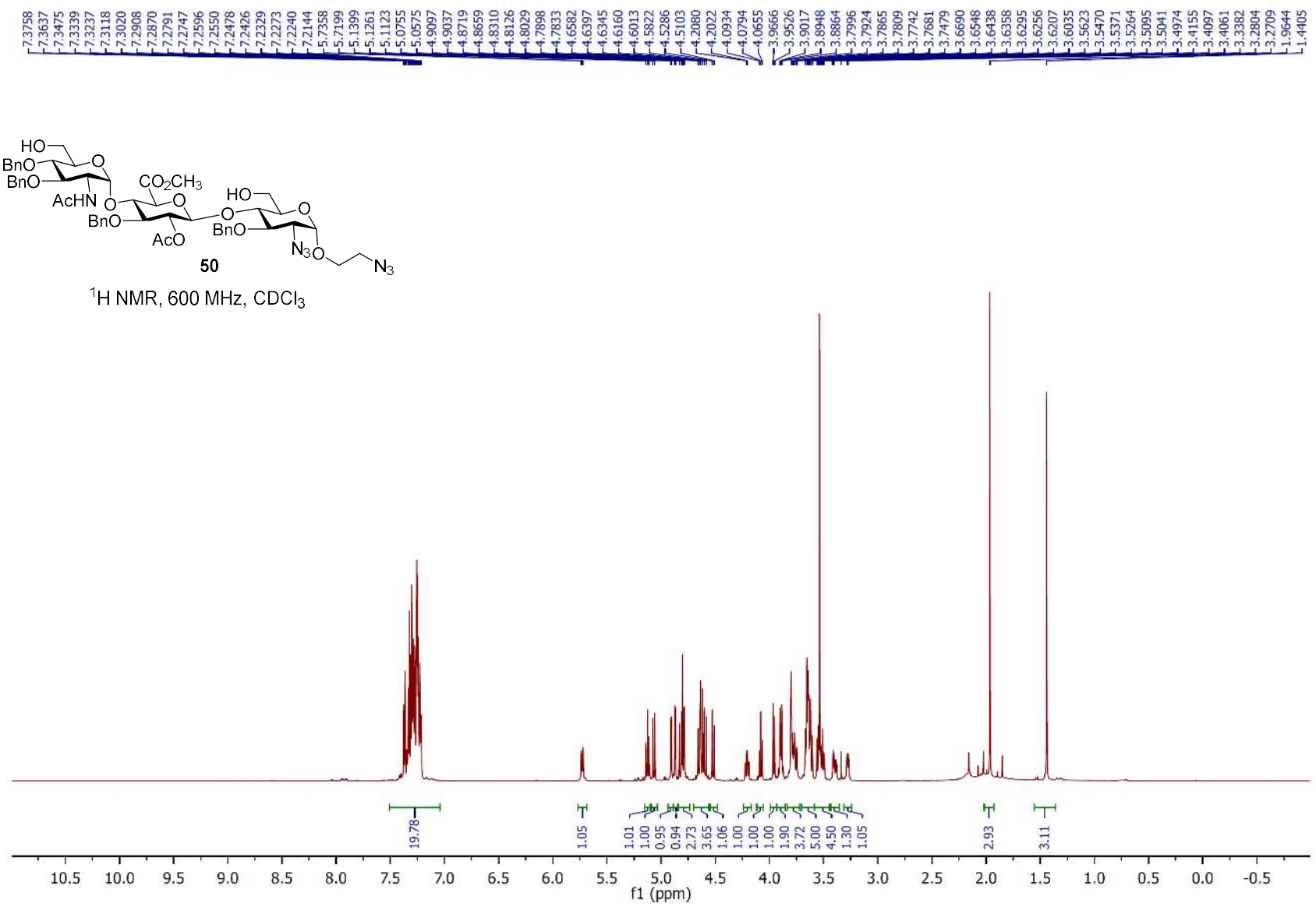
49

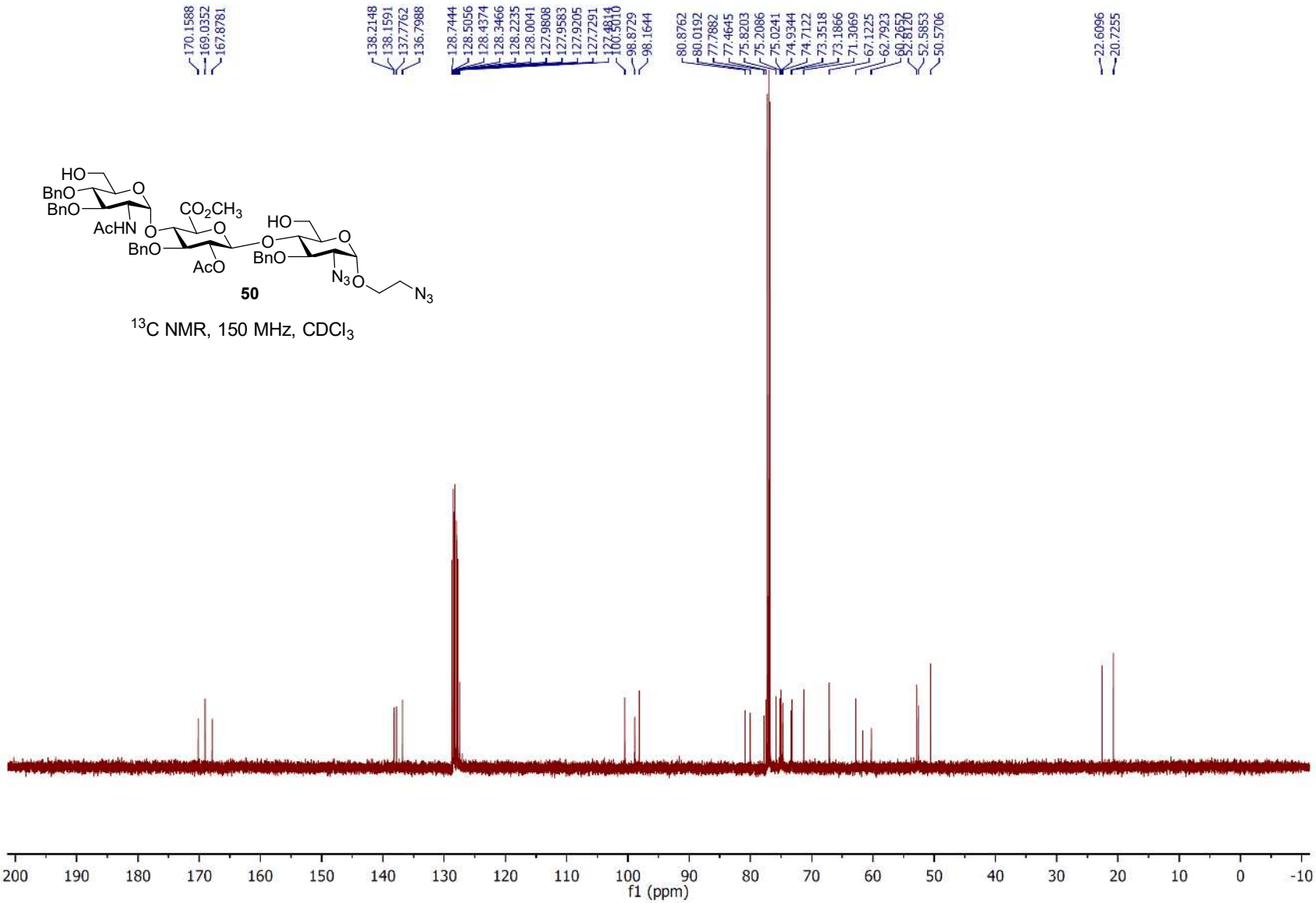
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

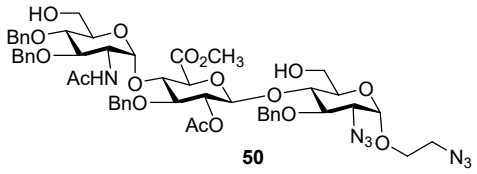


50

^{1}H NMR, 600 MHz, $CDCl_3$

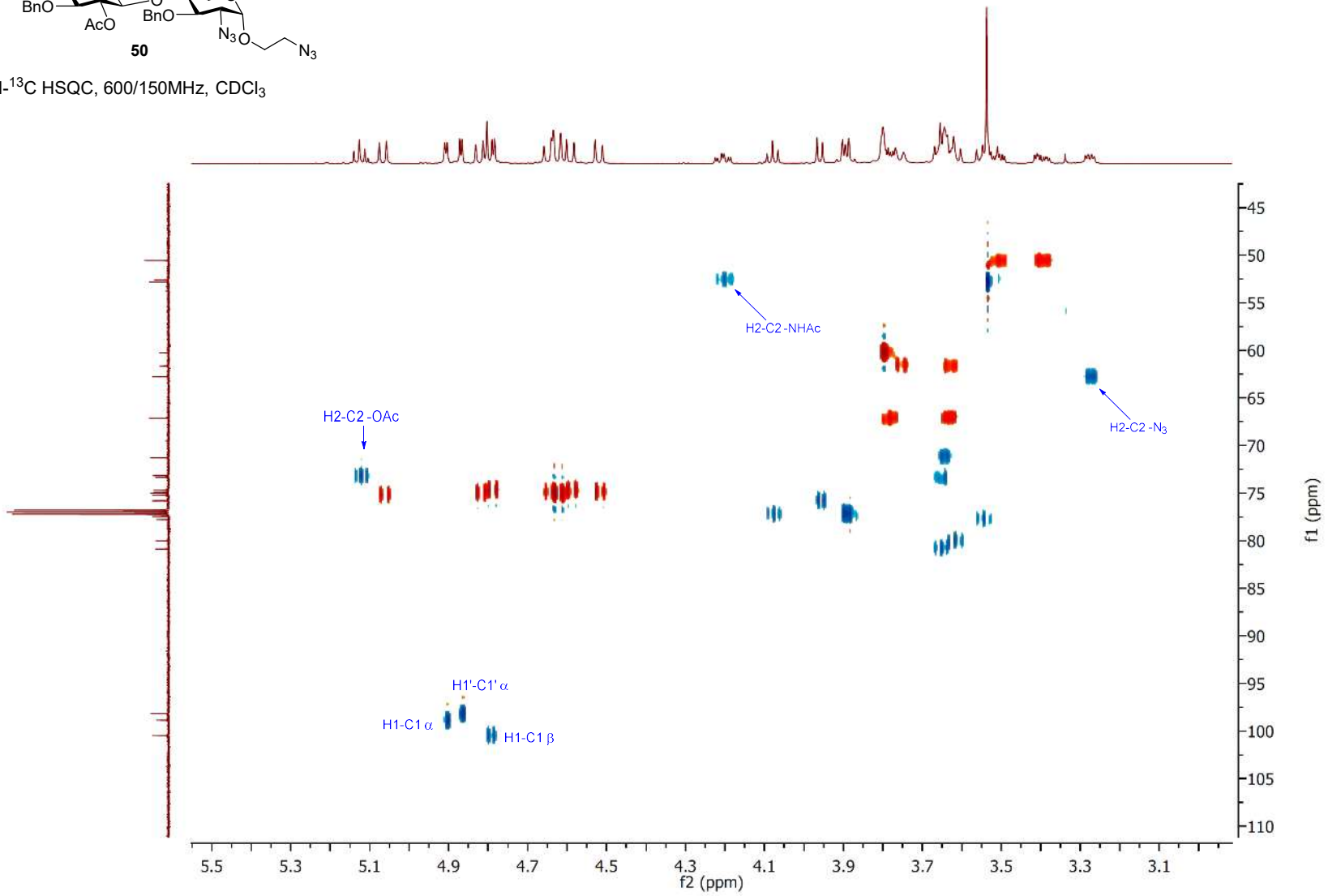


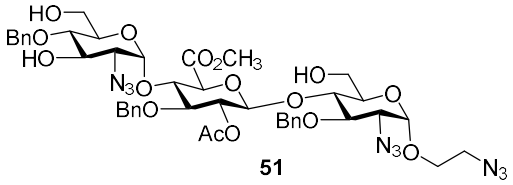

170.1588
169.0352
167.8781
138.2148
138.1591
137.7762
136.7988
128.7444
128.5056
128.4374
128.3466
128.2235
128.0041
127.9808
127.9583
127.9205
127.7291
127.4814
100.5010
98.8729
98.1644
80.8762
80.0192
77.7882
77.4645
75.8203
75.2086
75.0241
74.9344
74.7122
73.3518
73.1866
71.3069
67.1225
62.7923
52.5853
50.5706
22.6096
20.7255
HO
BnO
BnO
AcHN
O
CO2CH3
BnO
AcO
HO
BnO
N3
N3
50
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)



50

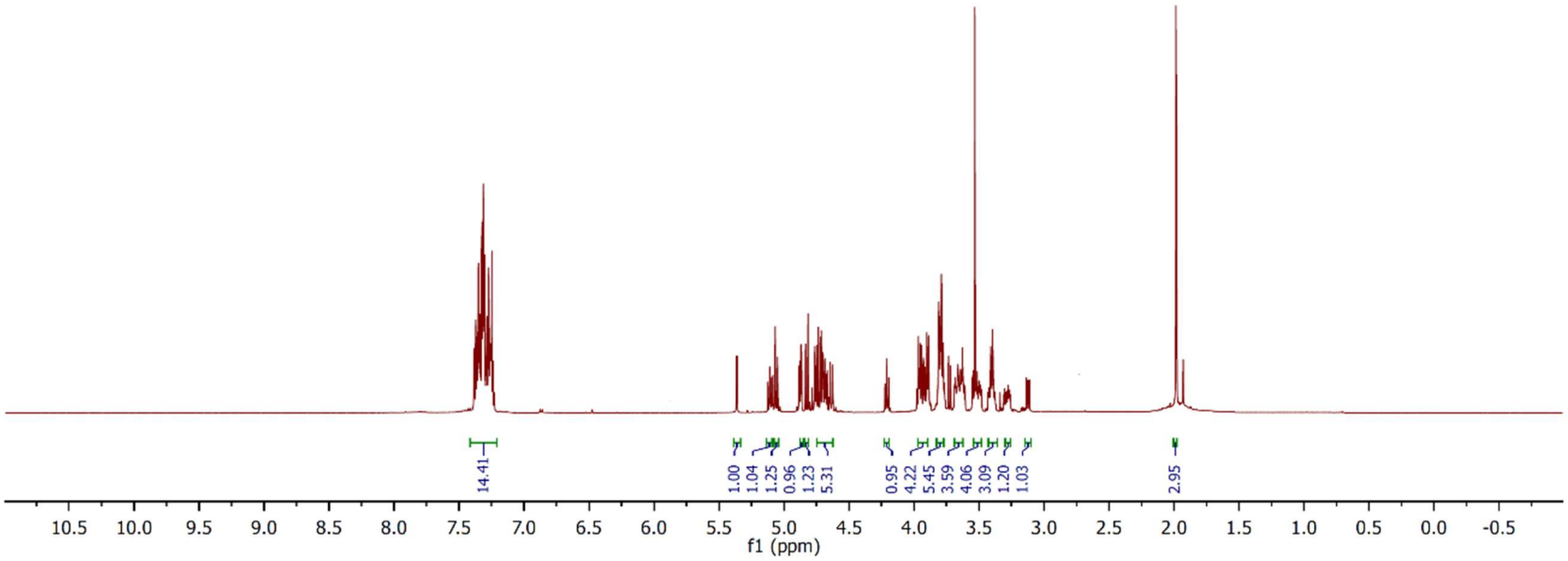
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

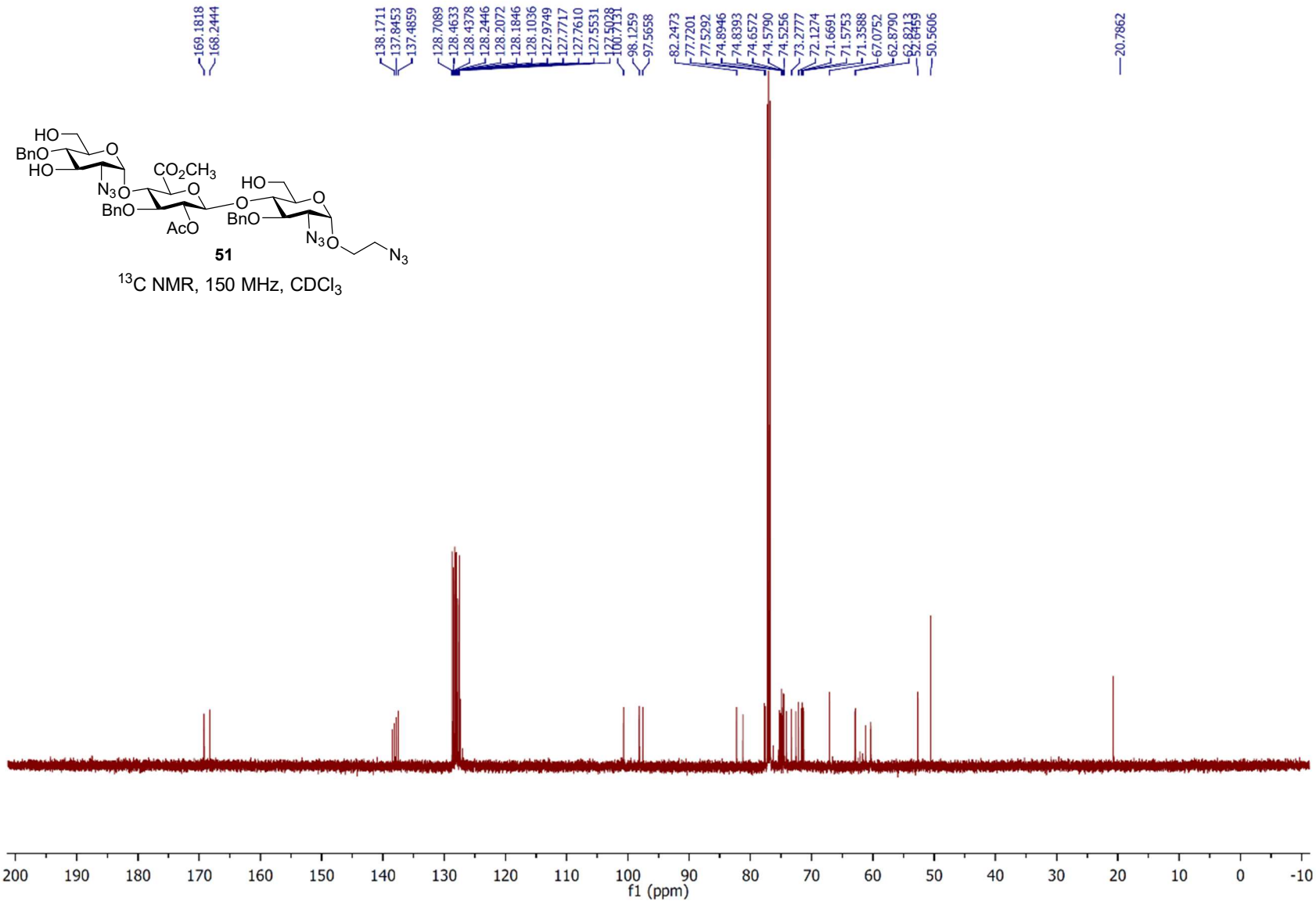


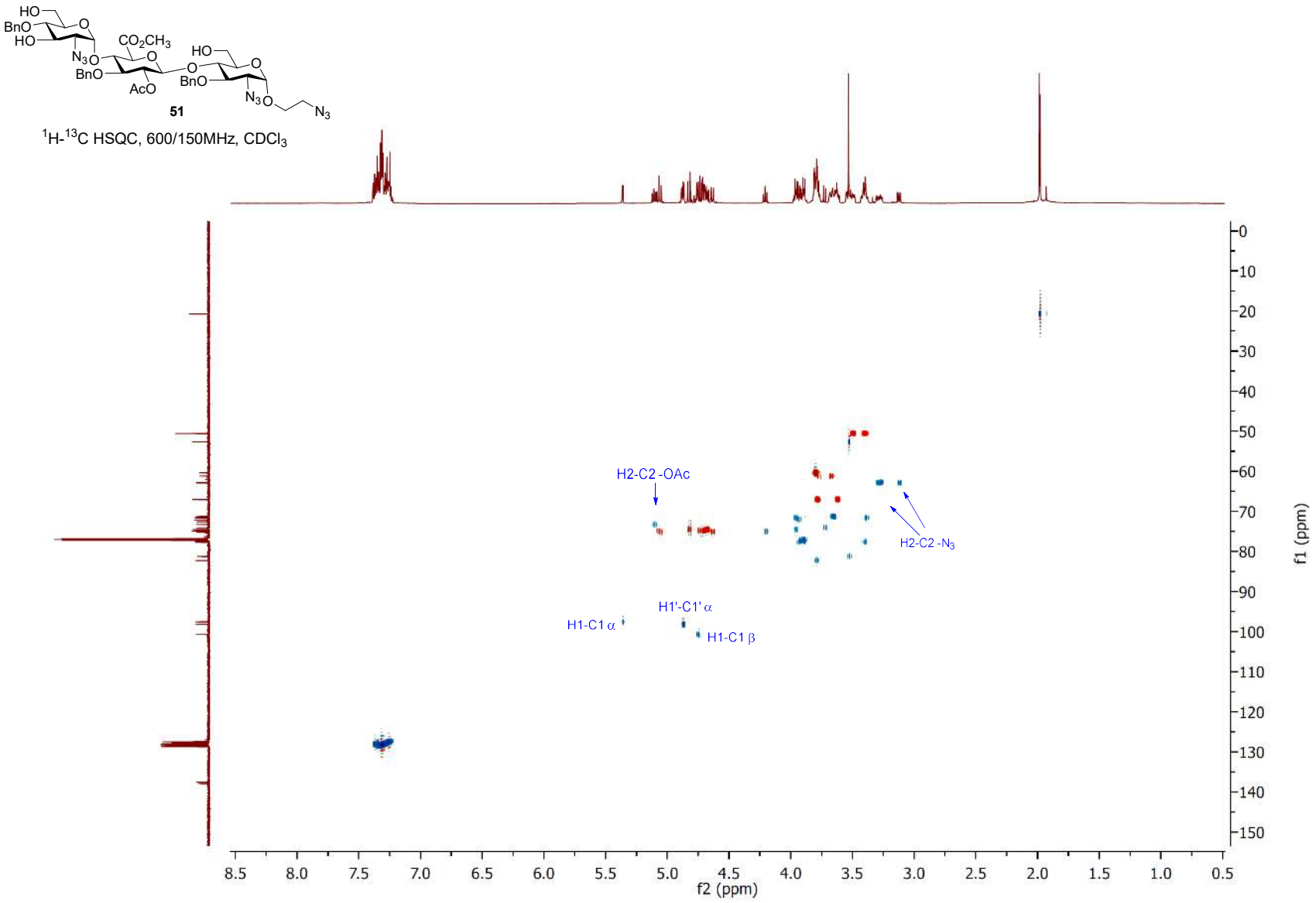


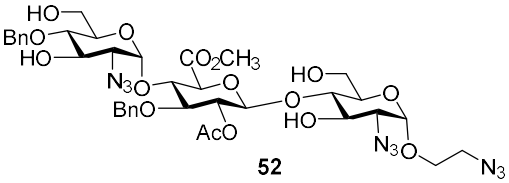
51

^{1}H NMR, 600 MHz, $CDCl_3$



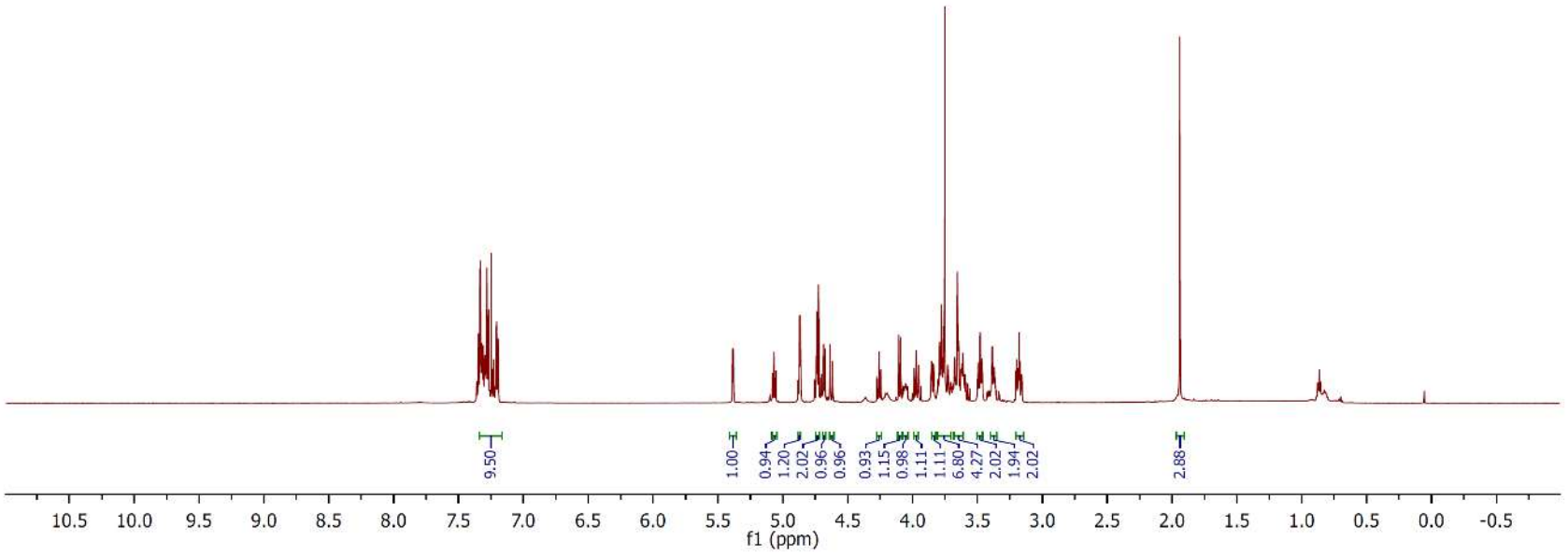

169.1818
168.2444
138.1711
137.8453
137.4859
128.7089
128.4633
128.4378
128.2446
128.2072
128.1846
128.1036
127.9749
127.7717
127.7610
127.5531
127.5028
98.1259
97.5658
82.2473
77.7201
77.5292
74.8946
74.8393
74.6572
74.5790
74.5256
73.2777
72.1274
71.6691
71.5753
71.3588
67.0752
62.8790
52.8459
50.5606
20.7862
HO
BnO
HO
O
N3O
CO2CH3
O
BnO
AcO
O
HO
O
BnO
N3O
N3
51
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

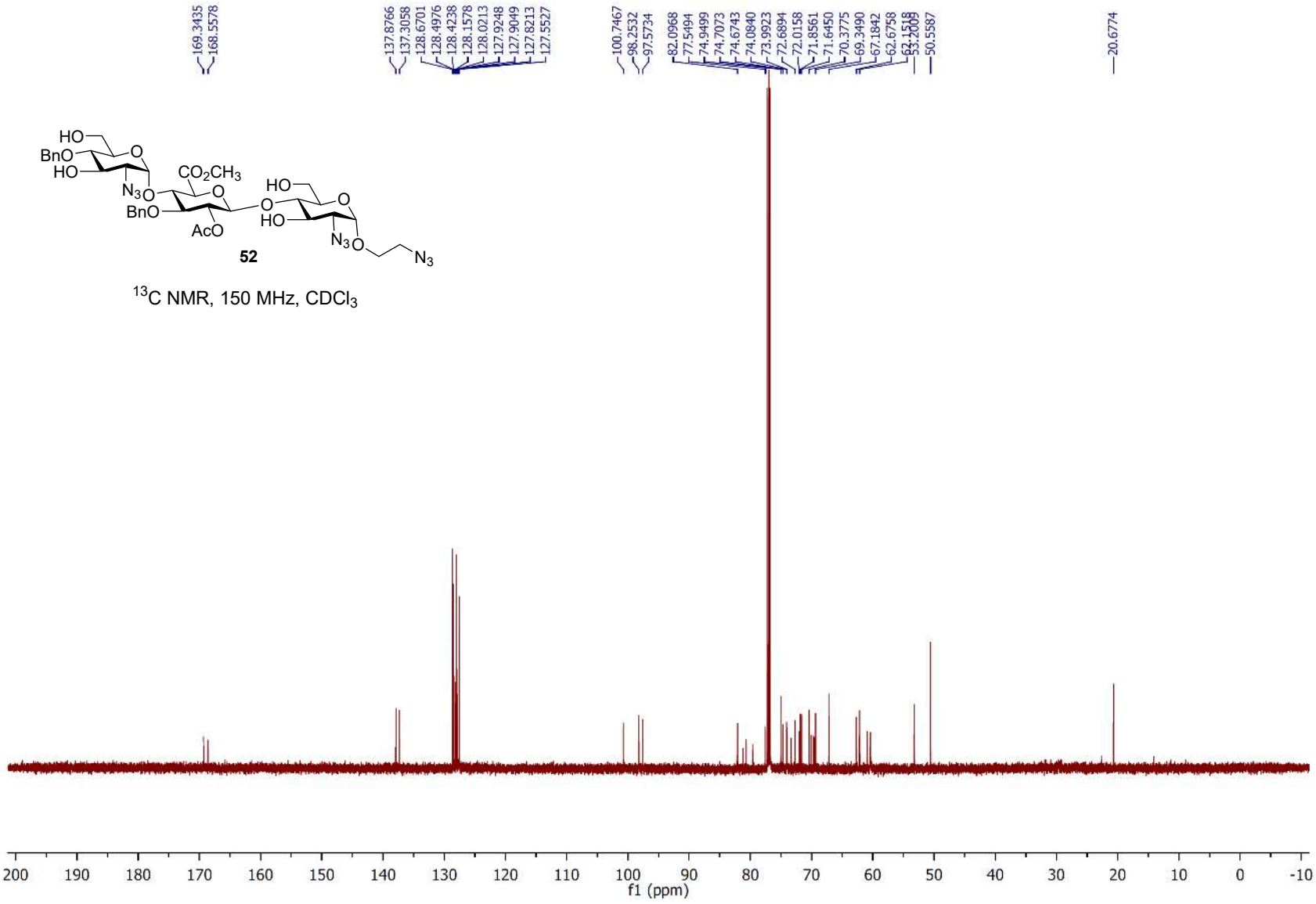

51
^{1}H-^{13}C HSQC, 600/150MHz, CDCl$_3$
H2-C2 -OAc
H2-C2 -N$_3$
H1'-C1' α
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)

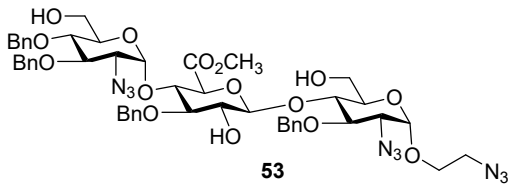


52

^{1}H NMR, 600 MHz, $CDCl_3$

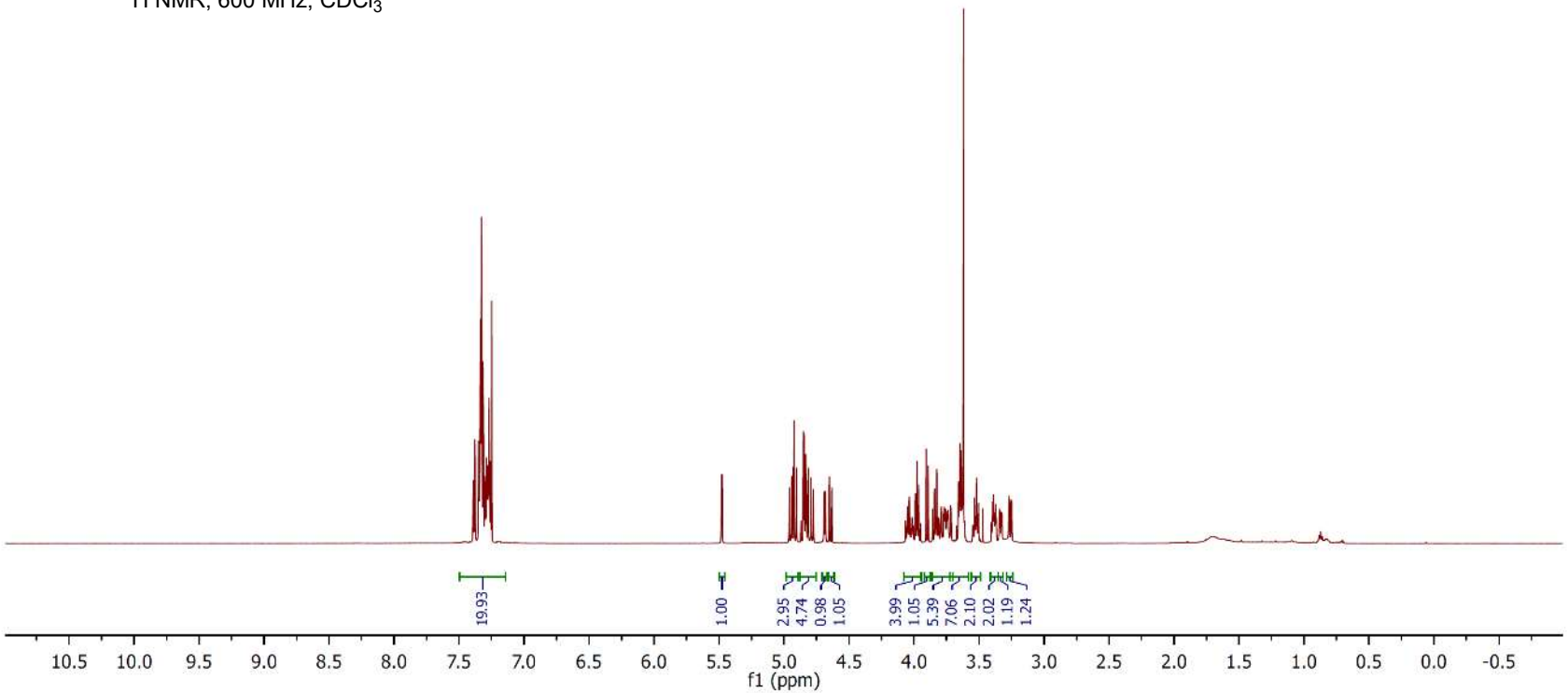


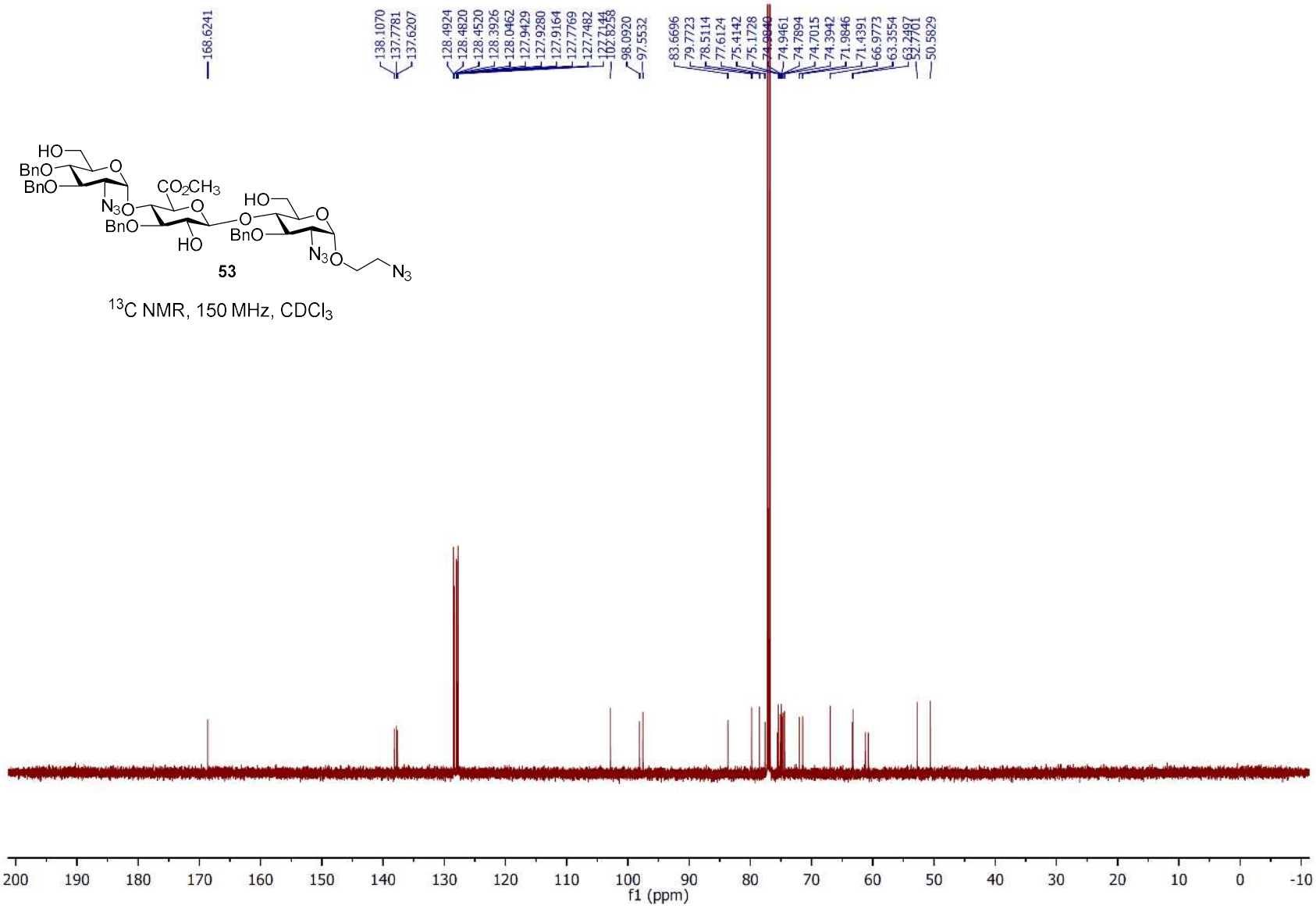

169.3435
168.5578
137.8766
137.3058
128.6701
128.4976
128.4238
128.1578
128.0213
127.9248
127.9049
127.8213
127.5527
100.7467
98.2532
97.5734
82.0968
77.5494
74.9499
74.7073
74.6743
74.0840
73.9923
72.6894
72.0158
71.8561
71.6450
70.3775
69.3490
67.1842
62.6758
53.2009
50.5587
20.6774
HO
BnO
HO
O
N3O
CO2CH3
O
BnO
AcO
O
HO
HO
O
N3O
N3
52
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

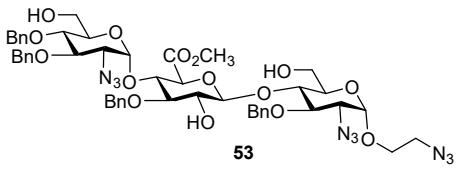


53

^{1}H NMR, 600 MHz, $CDCl_3$

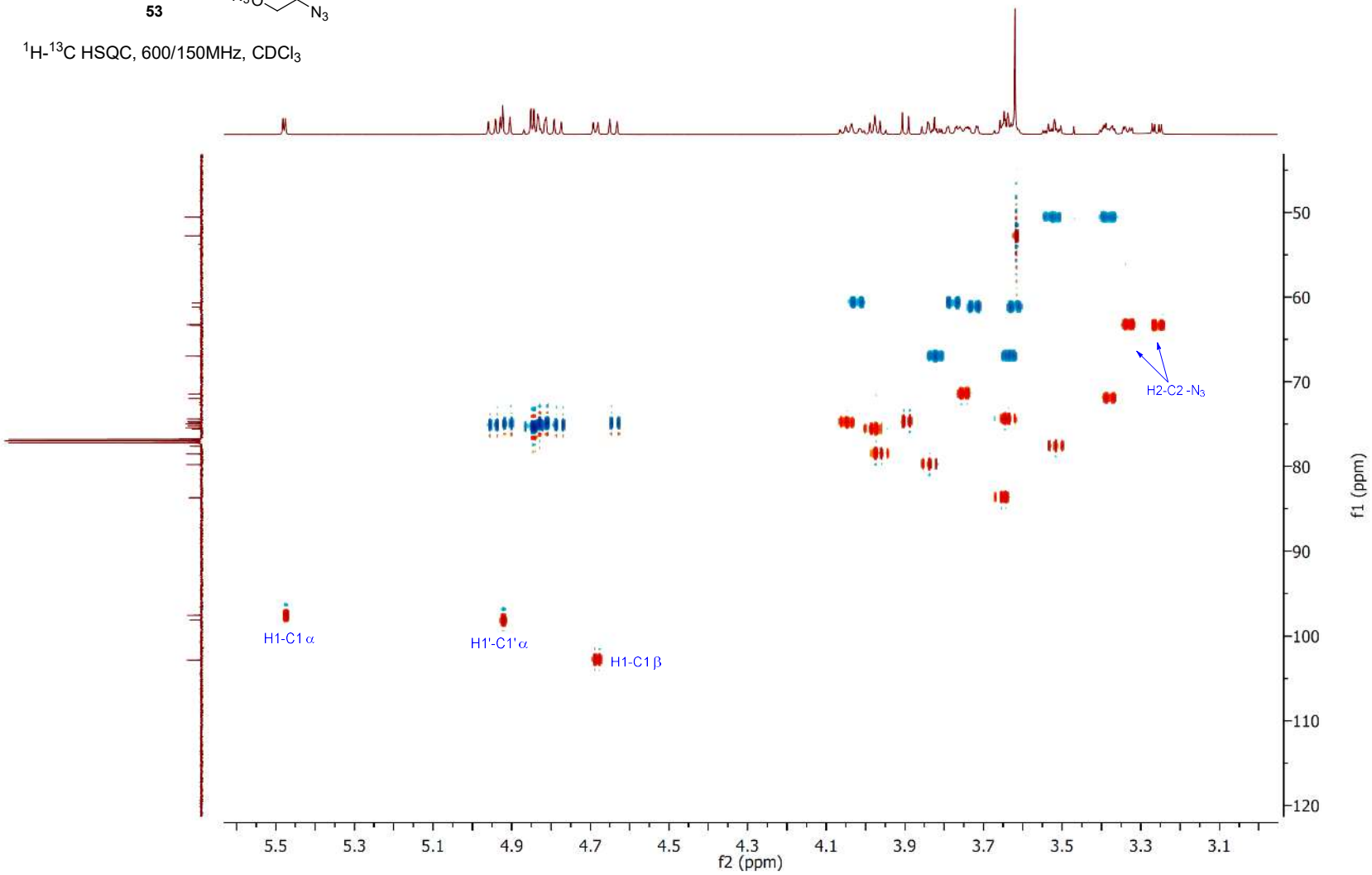


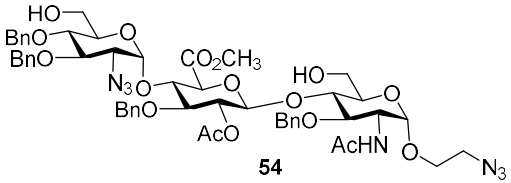

168.6241
138.1070
137.7781
137.6207
128.4924
128.4820
128.4520
128.3926
128.0462
127.9429
127.9280
127.9164
127.7769
127.7482
127.7144
127.6258
98.0920
97.5532
83.6696
79.7723
78.5114
77.6124
75.4142
75.1728
74.9461
74.7894
74.7015
74.3942
71.9846
71.4391
66.9773
63.3554
63.2497
52.7701
50.5829
HO
BnO
BnO
O
N3O
CO2CH3
O
BnO
HO
O
HO
O
BnO
N3O
N3
53
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)



53

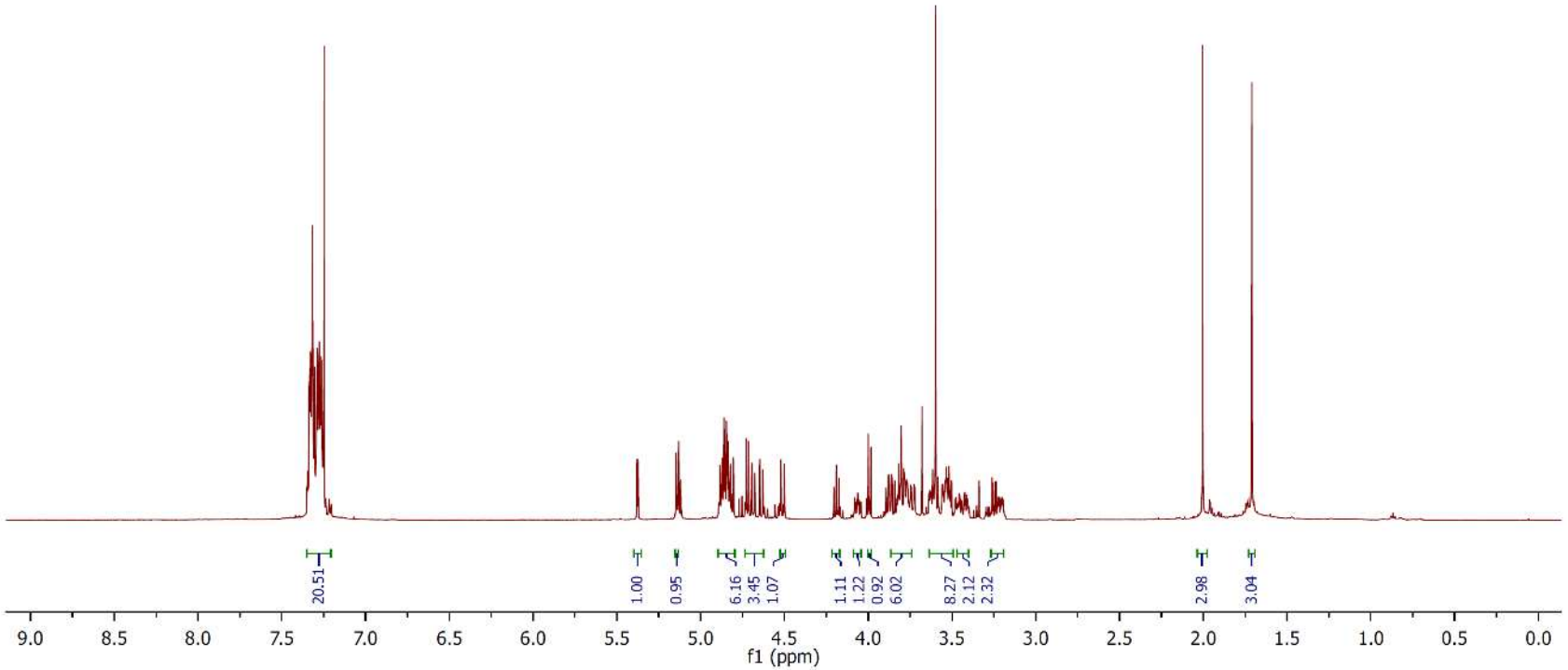
^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

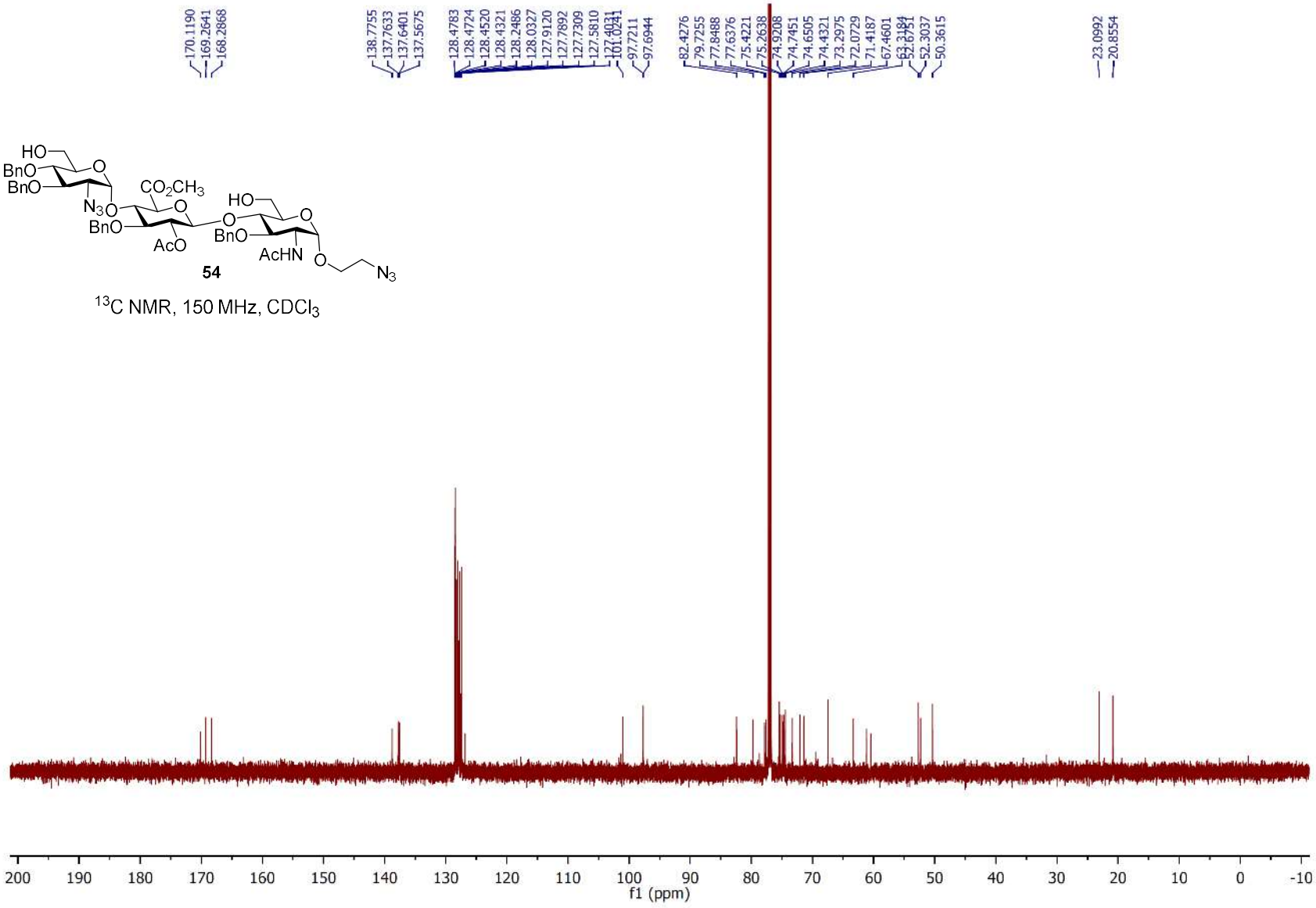


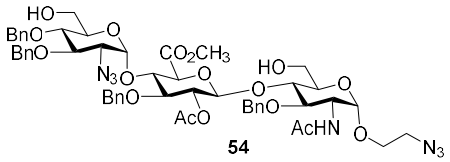


54

^{1}H NMR, 600 MHz, $CDCl_3$

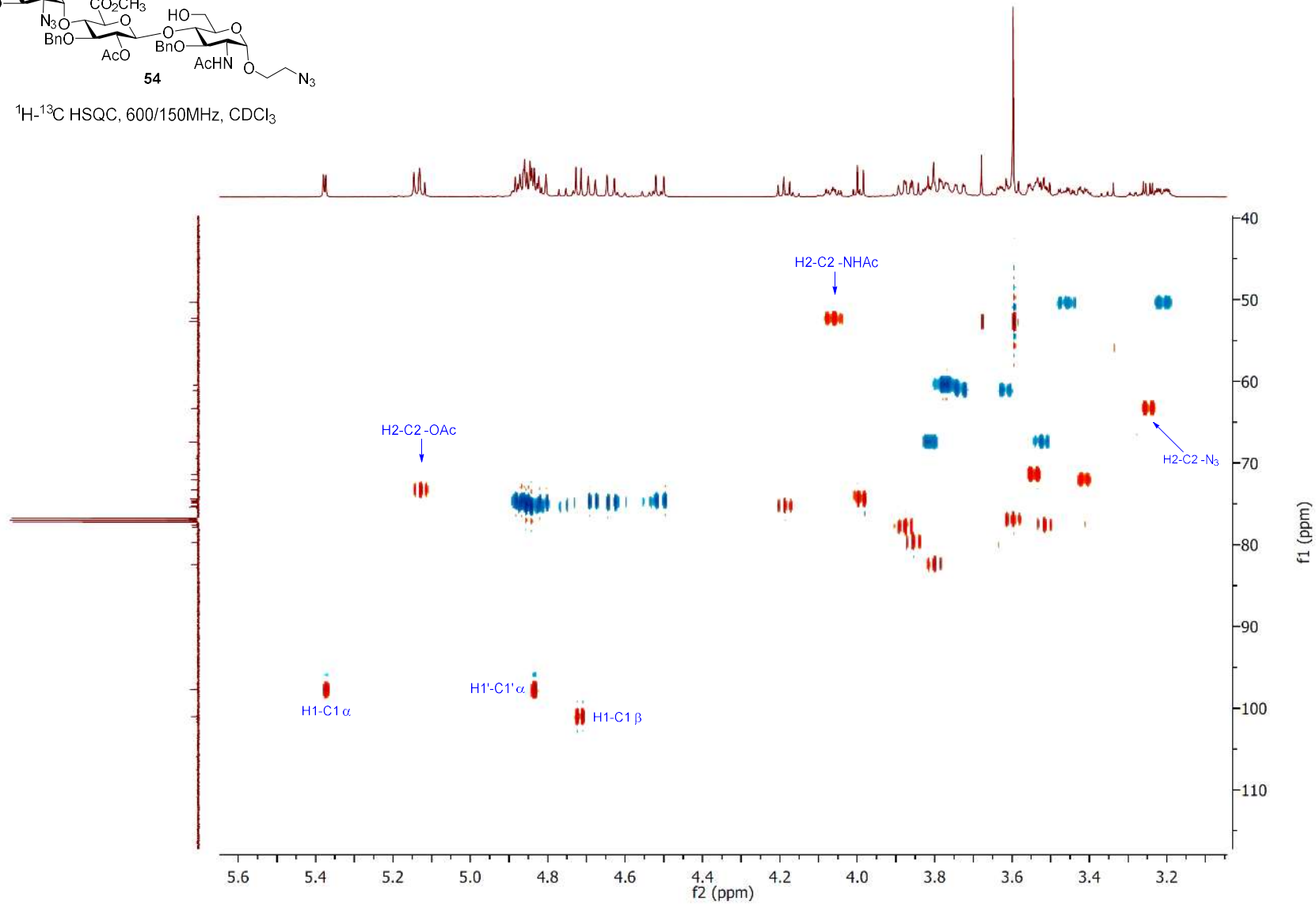


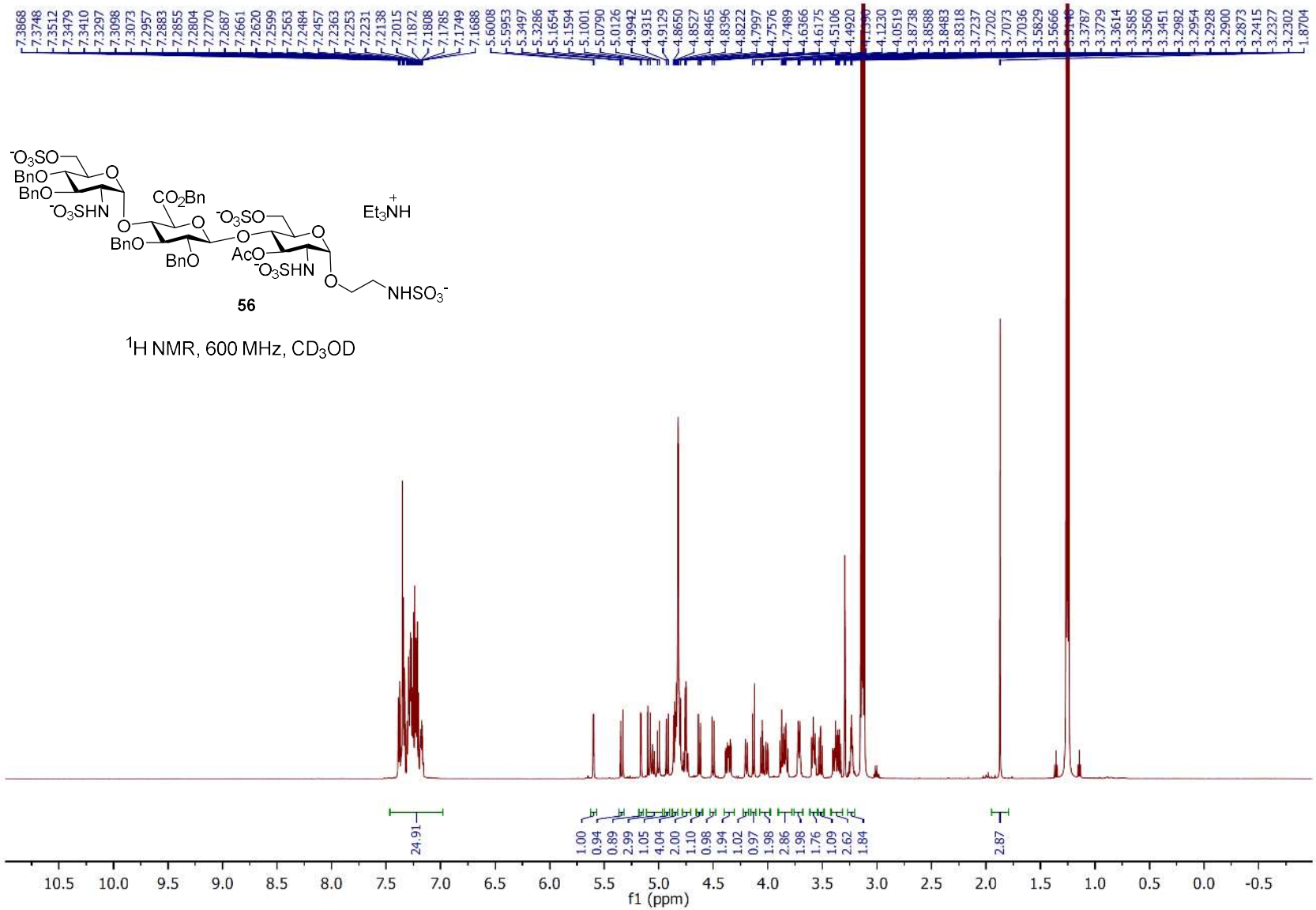

170.1190
169.2641
168.2868
138.7755
137.7633
137.6401
137.5675
128.4783
128.4724
128.4520
128.4321
128.2486
128.0327
127.9120
127.7892
127.7309
127.5810
101.0241
97.7211
97.6944
82.4276
79.7255
77.8488
77.6376
75.4221
75.2638
74.9208
74.7451
74.6505
74.4321
73.2975
72.0729
71.4187
67.4601
52.3037
50.3615
23.0992
20.8554
HO
BnO
BnO
O
N3
CO2CH3
O
BnO
AcO
O
HO
BnO
O
AcHN
O
N3
54
13C NMR, 150 MHz, CDCl3
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

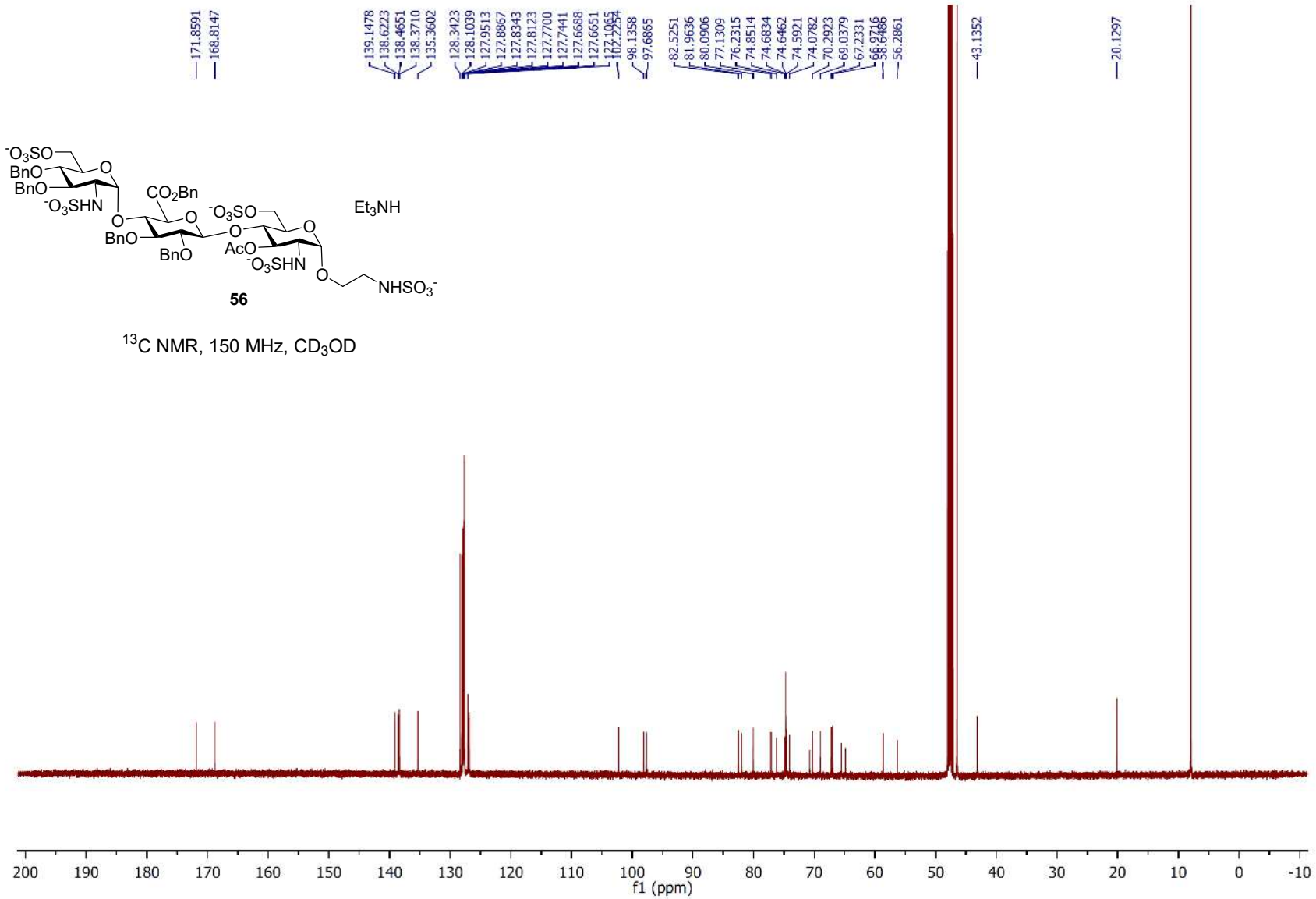


54

^{1}H-^{13}C HSQC, 600/150MHz, $CDCl_3$

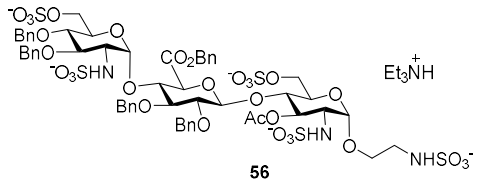



$^{-}O_3SO$
BnO
BnO
$^{-}O_3SHN$
CO_2Bn
BnO
BnO
$^{-}O_3SO$
AcO
$^{-}O_3SHN$
Et_3NH^{+}
$NHSO_3^{-}$
56
^{1}H NMR, 600 MHz, CD_3OD
f1 (ppm)



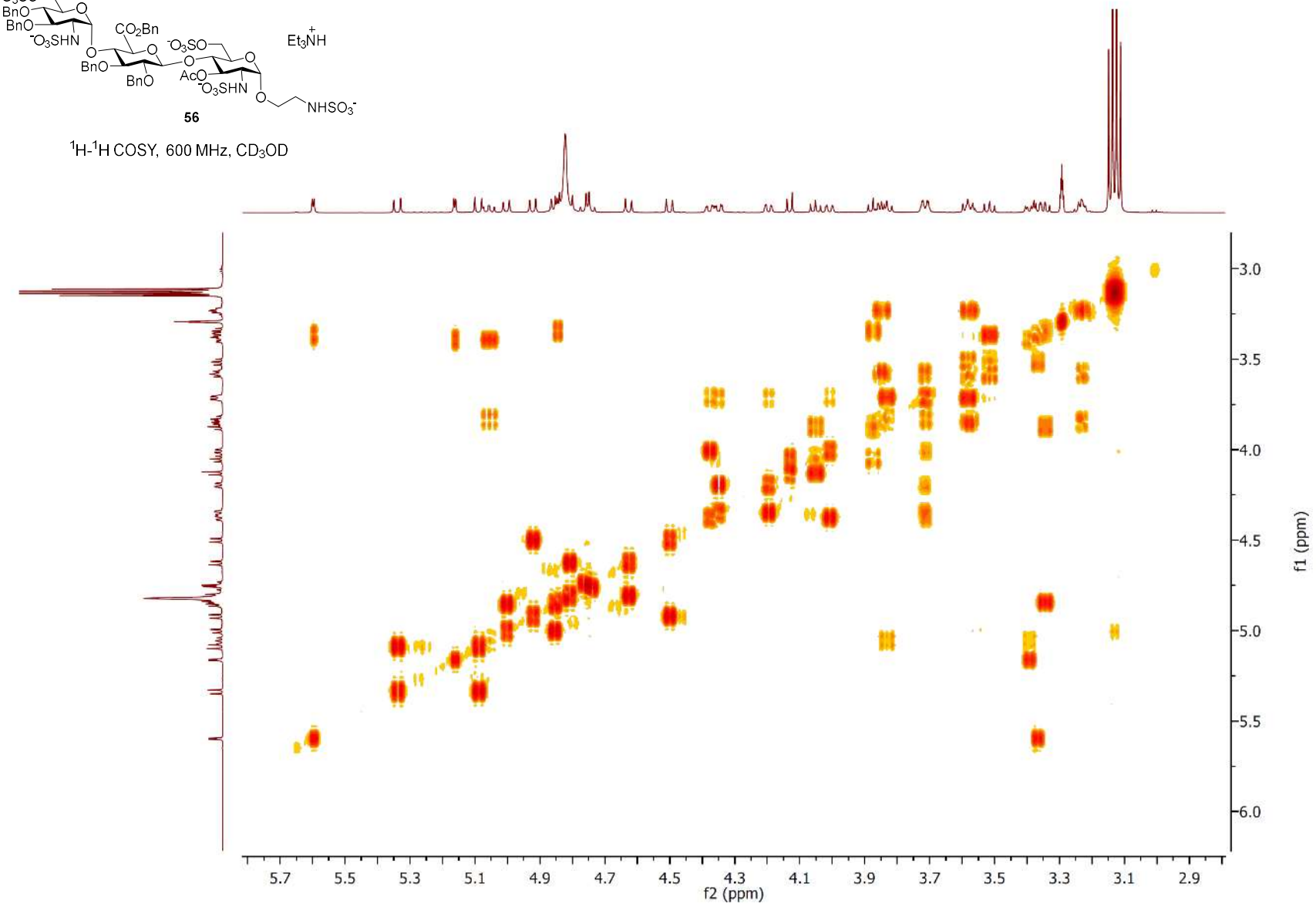
56

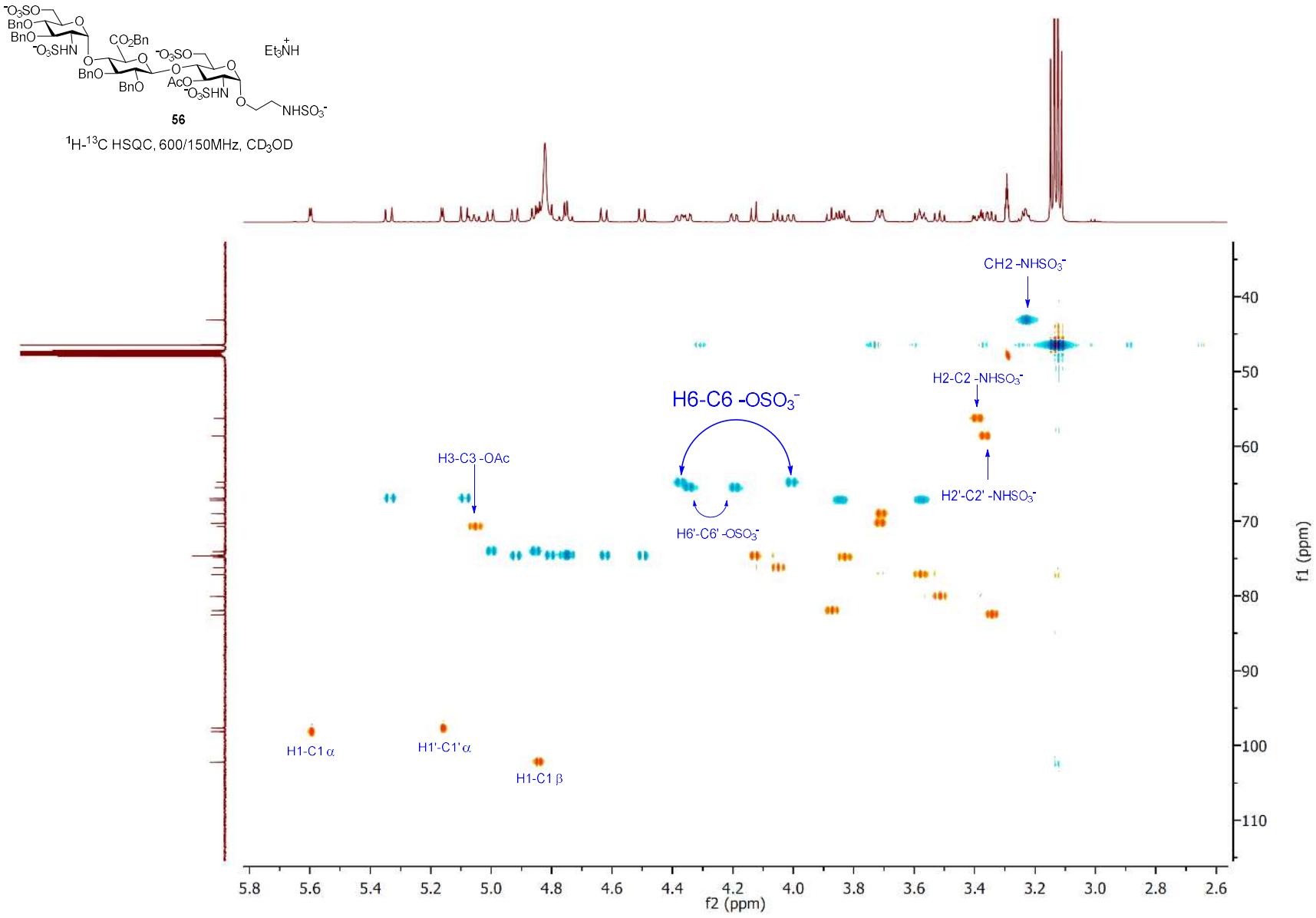
^{13}C NMR, 150 MHz, CD_3OD

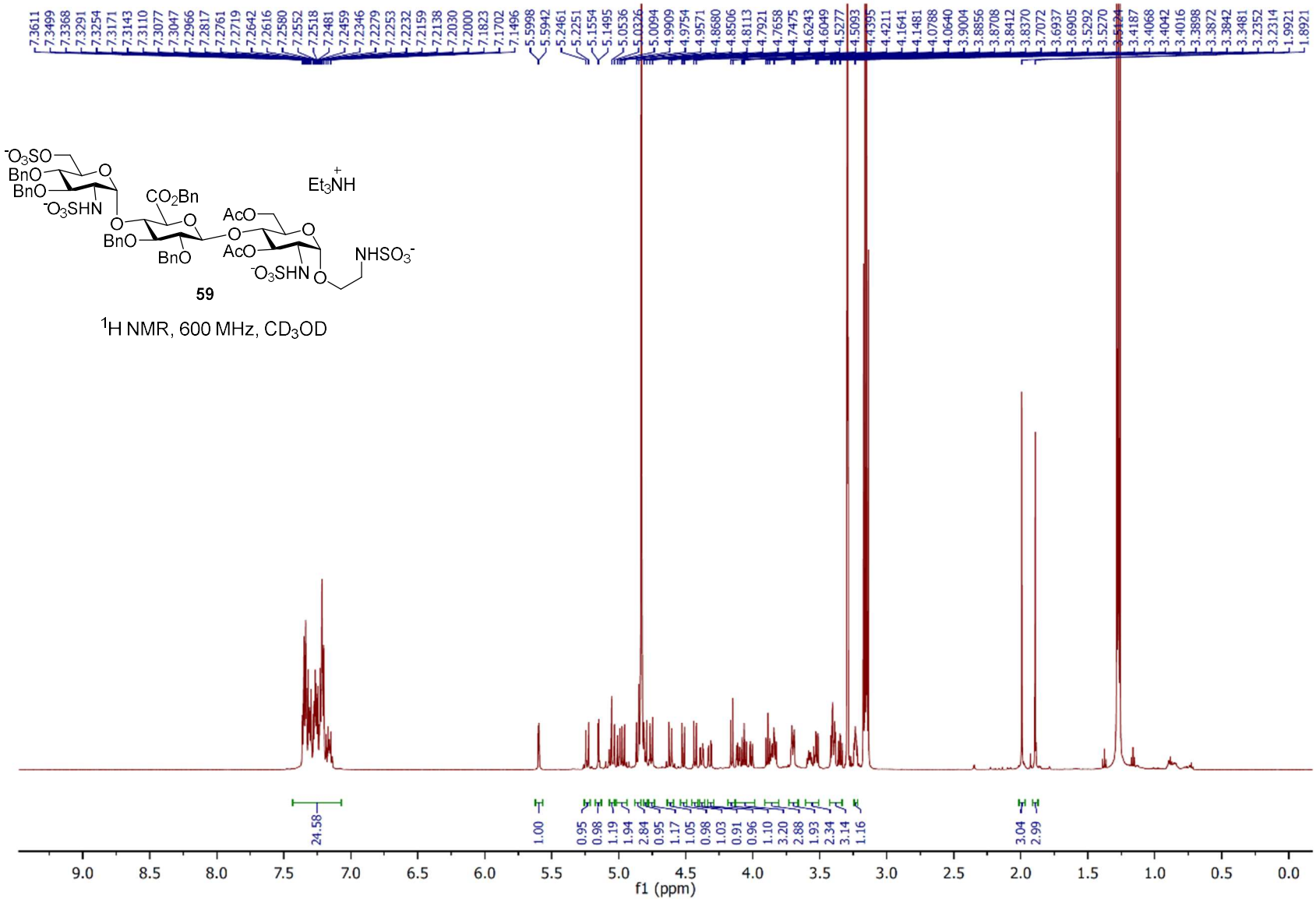


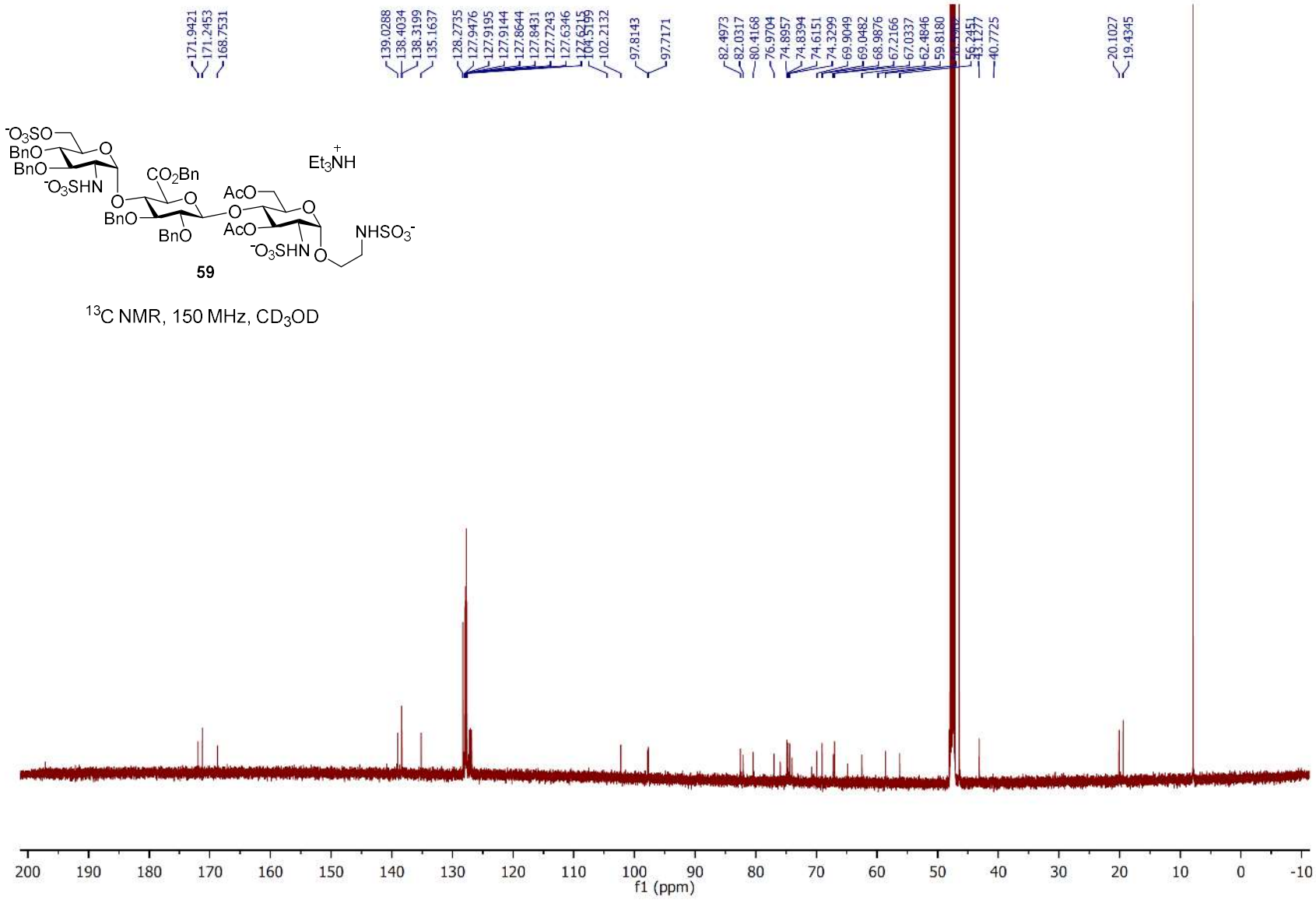
56

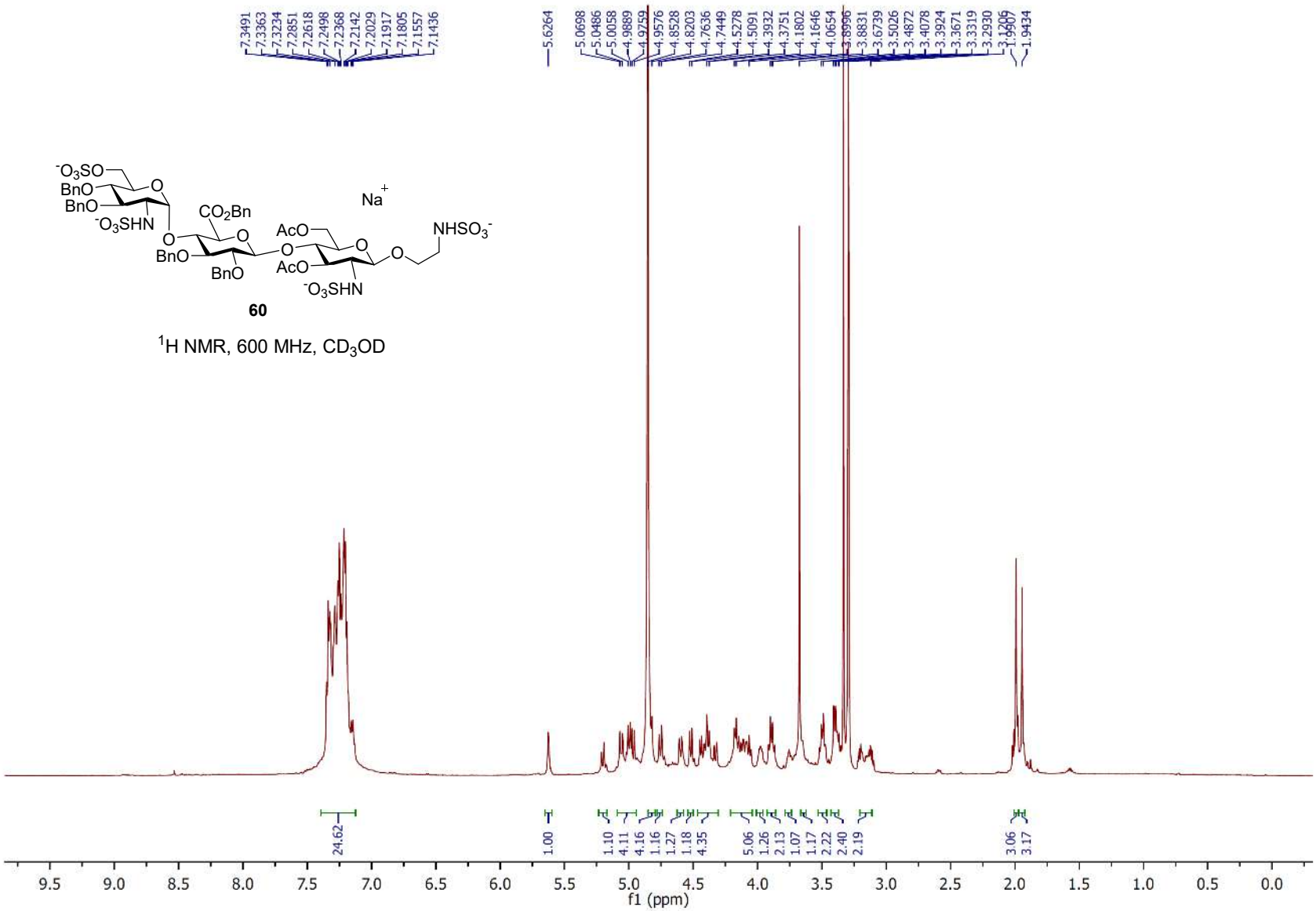
^{1}H-^{1}H COSY, 600 MHz, CD_3OD

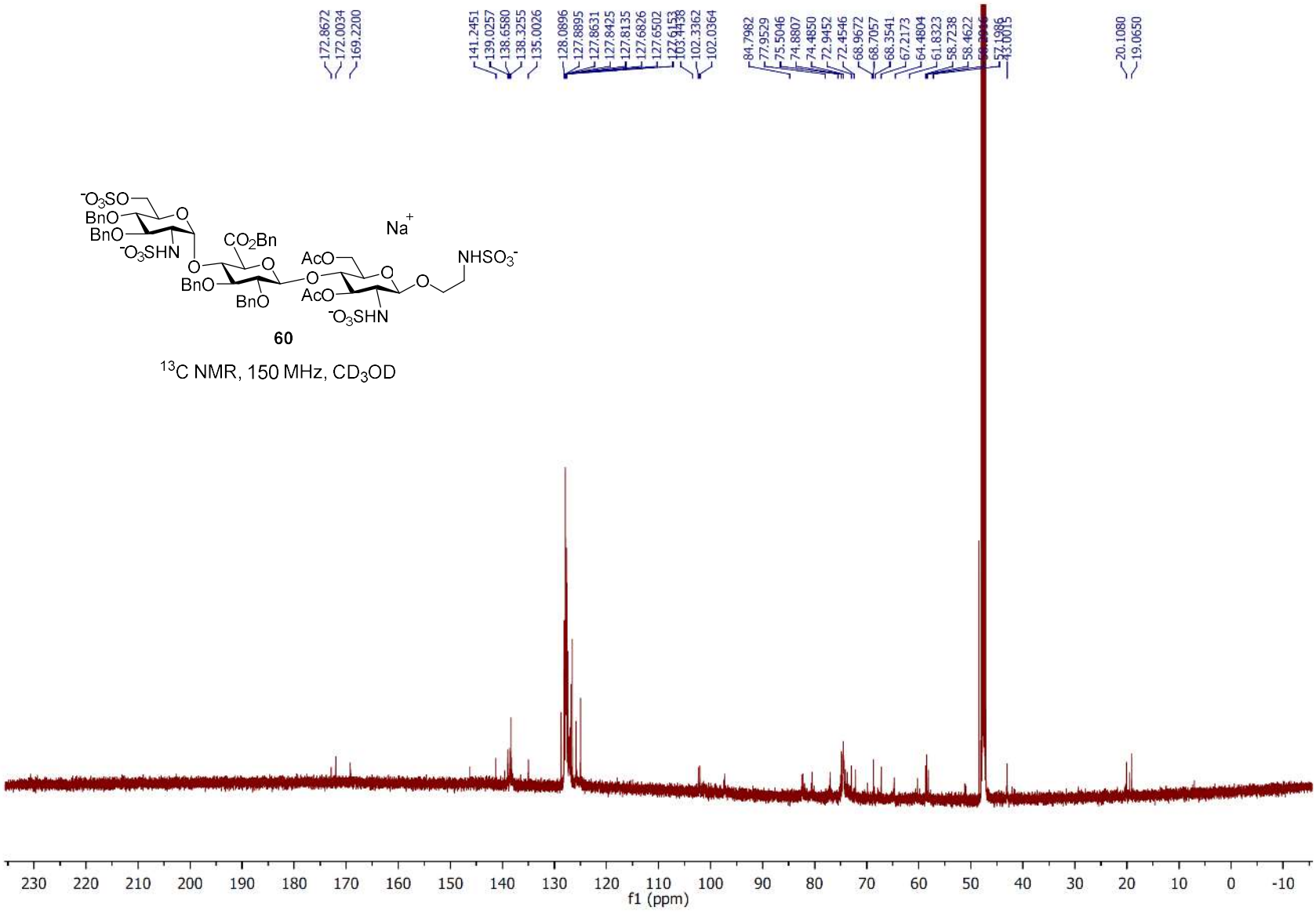


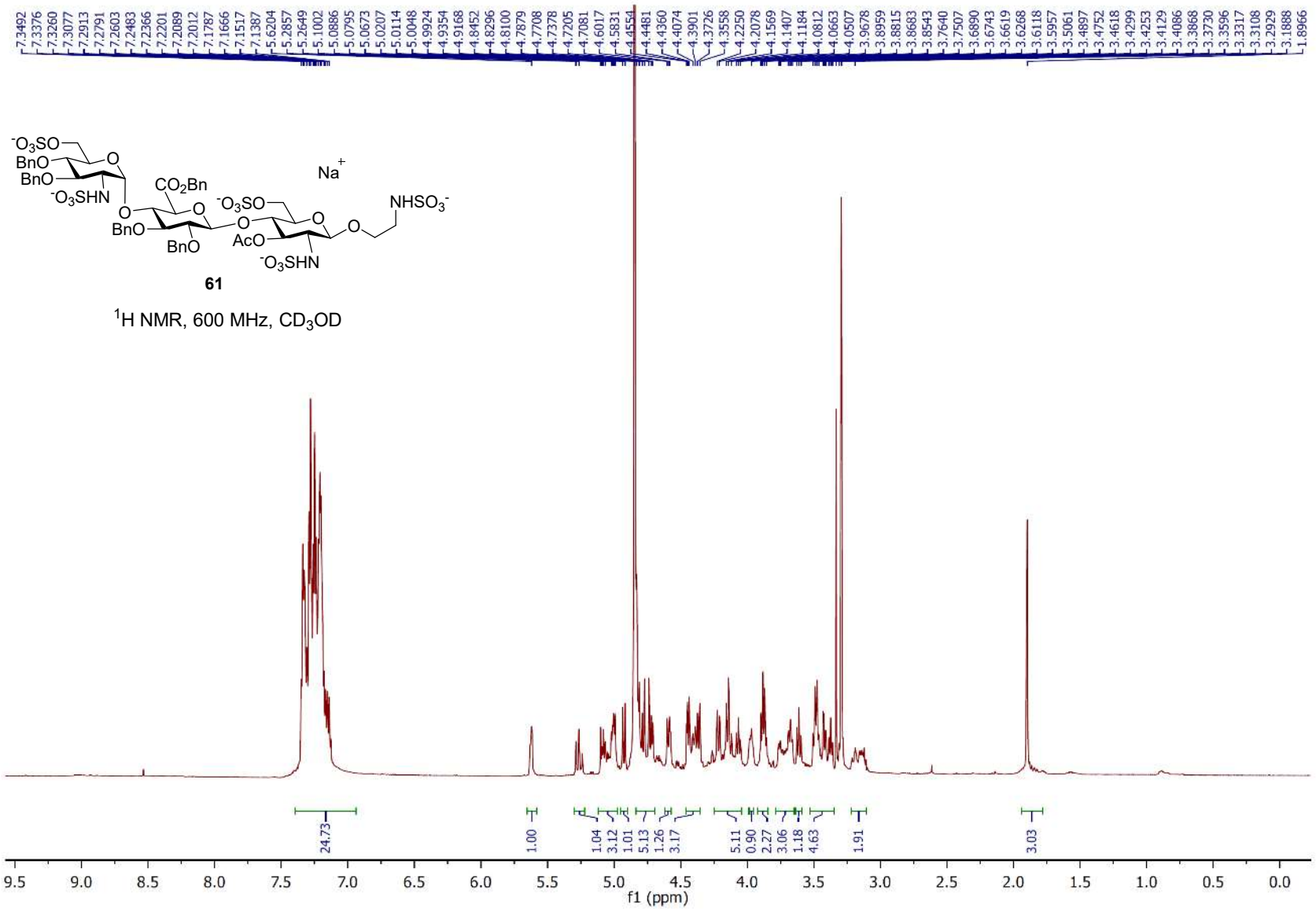

56
^{1}H-^{13}C HSQC, 600/150MHz, CD$_3$OD
CH2 -NHSO$_3^-$
H2-C2 -NHSO$_3^-$
H6-C6 -OSO$_3^-$
H3-C3 -OAc
H6'-C6' -OSO$_3^-$
H2'-C2' -NHSO$_3^-$
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)

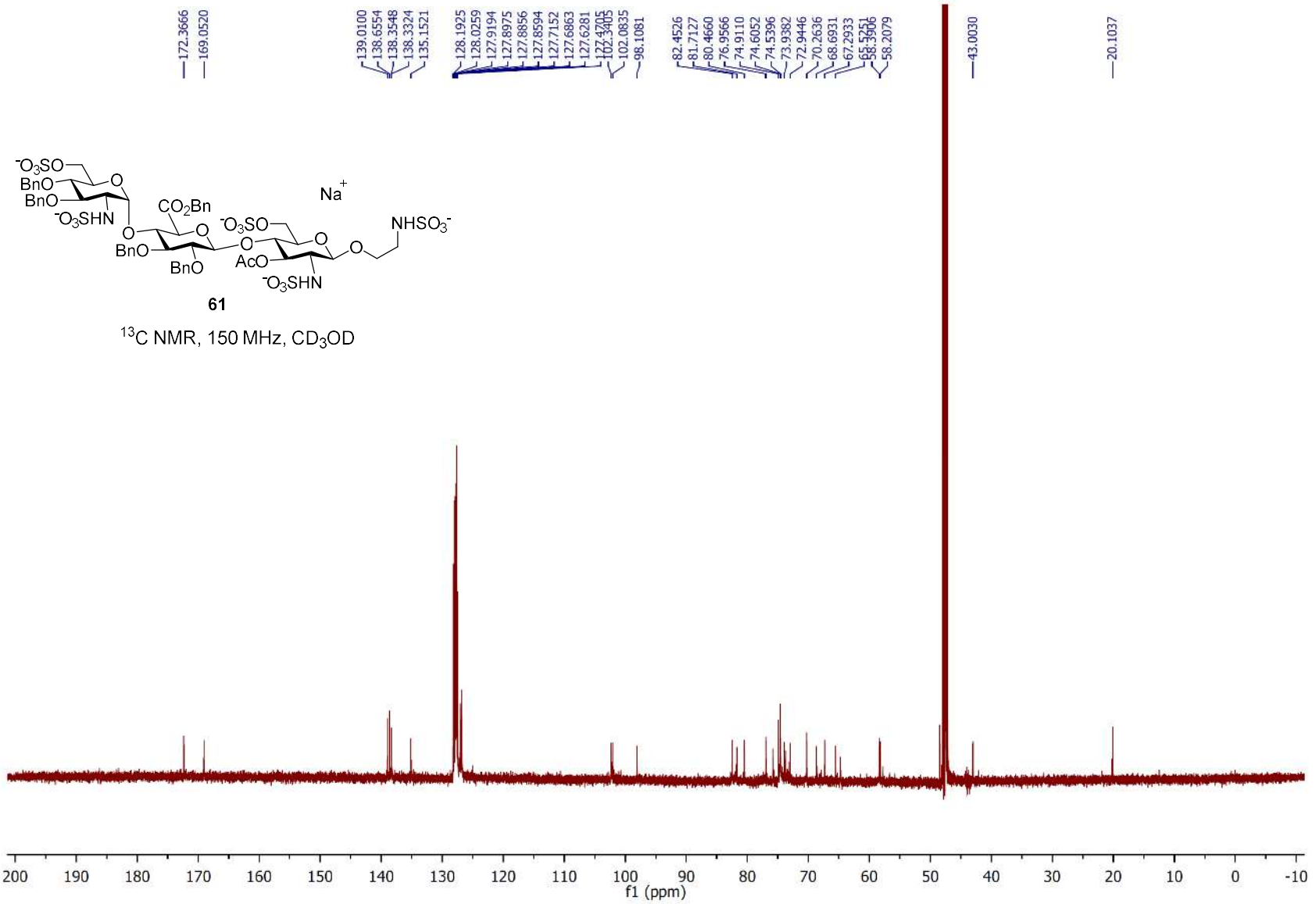

59
1H NMR, 600 MHz, CD3OD
f1 (ppm)

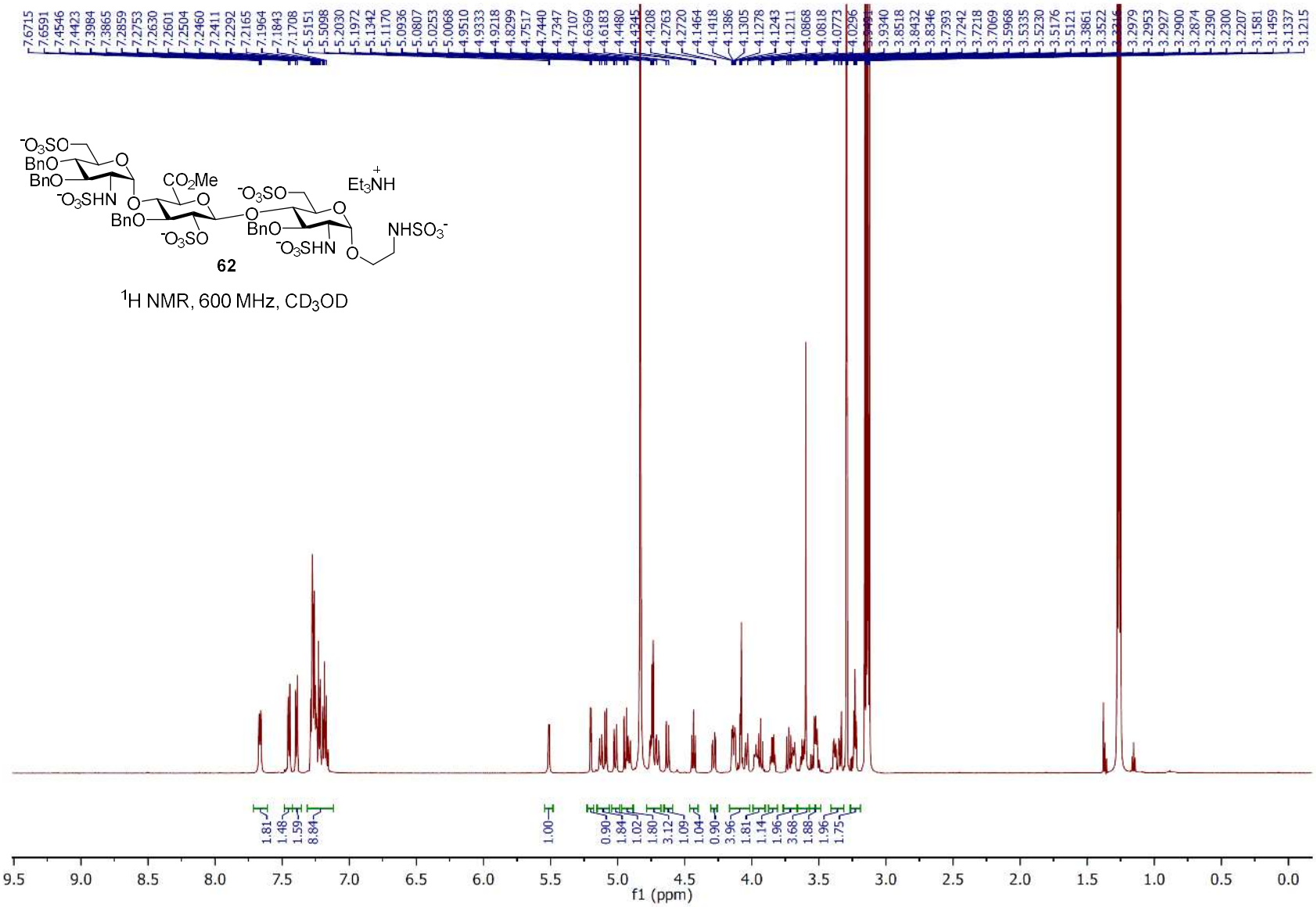

171.9421
171.2453
168.7531
139.0288
138.4034
138.3199
135.1637
128.2735
127.9476
127.9195
127.9144
127.8644
127.8431
127.7243
127.6346
102.2132
97.8143
97.7171
82.4973
82.0317
80.4168
76.9704
74.8957
74.8394
74.6151
74.3299
69.9049
69.0482
68.9876
67.2166
67.0337
62.4846
59.8180
56.2451
43.1277
40.7725
20.1027
19.4345
59
13C NMR, 150 MHz, CD3OD
f1 (ppm)

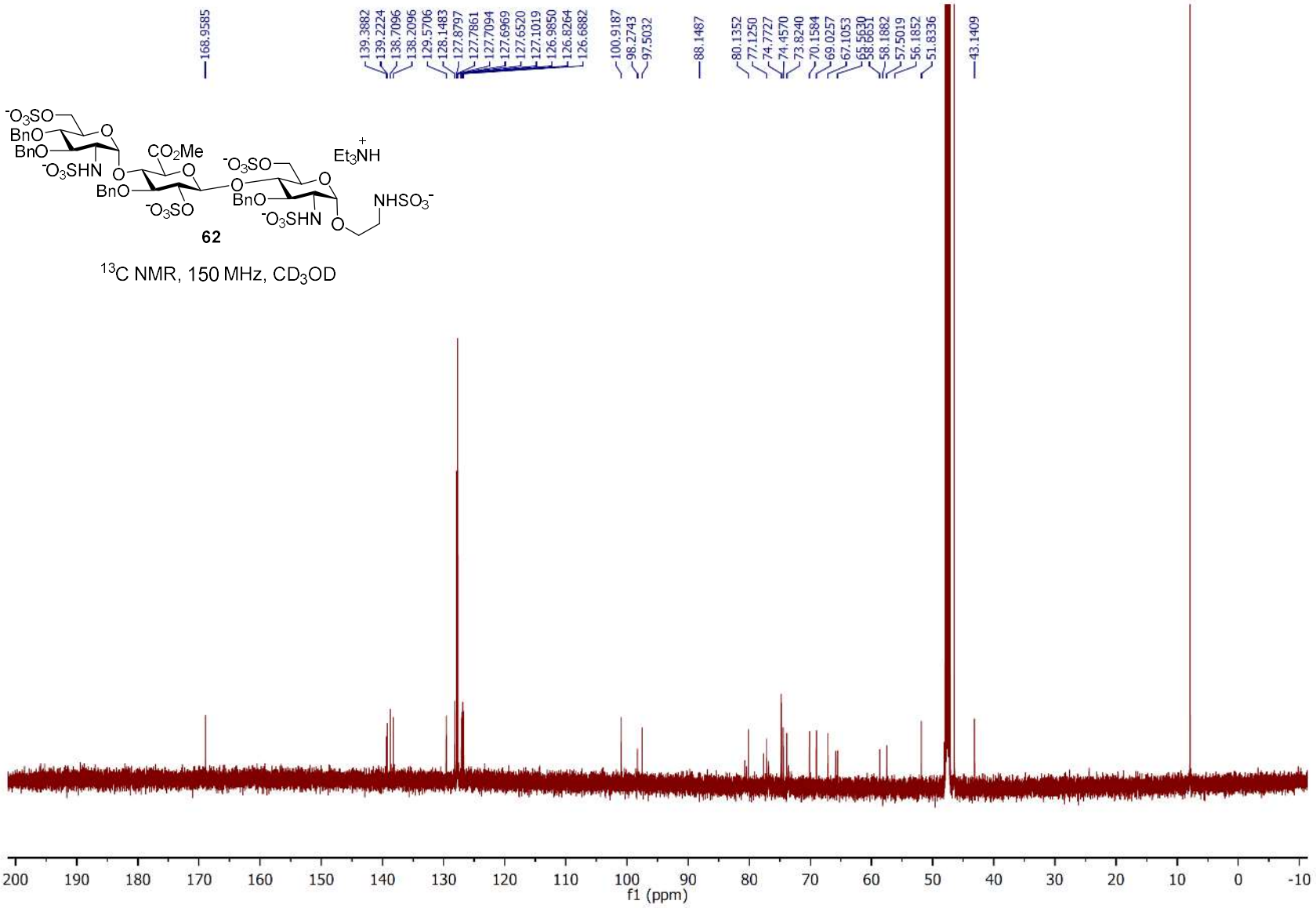

7.3491
7.3363
7.3234
7.2851
7.2618
7.2498
7.2368
7.2142
7.2029
7.1917
7.1805
7.1557
7.1436
5.6264
5.0698
5.0486
5.0058
4.9889
4.9759
4.9576
4.8528
4.8203
4.7636
4.7449
4.5278
4.5091
4.3932
4.3751
4.1802
4.1646
4.0654
3.8996
3.8831
3.6739
3.5026
3.4872
3.4078
3.3924
3.3671
3.3319
3.2930
1.9434
$^{-}O_3SO$
BnO
BnO
$^{-}O_3SHN$
CO_2Bn
AcO
AcO
$^{-}O_3SHN$
$NHSO_3^{-}$
Na^{+}
60
^{1}H NMR, 600 MHz, CD_3OD
24.62
1.00
1.10
4.11
4.16
1.16
1.27
1.18
4.35
5.06
1.26
2.13
1.07
1.17
2.22
2.40
2.19
3.06
3.17
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

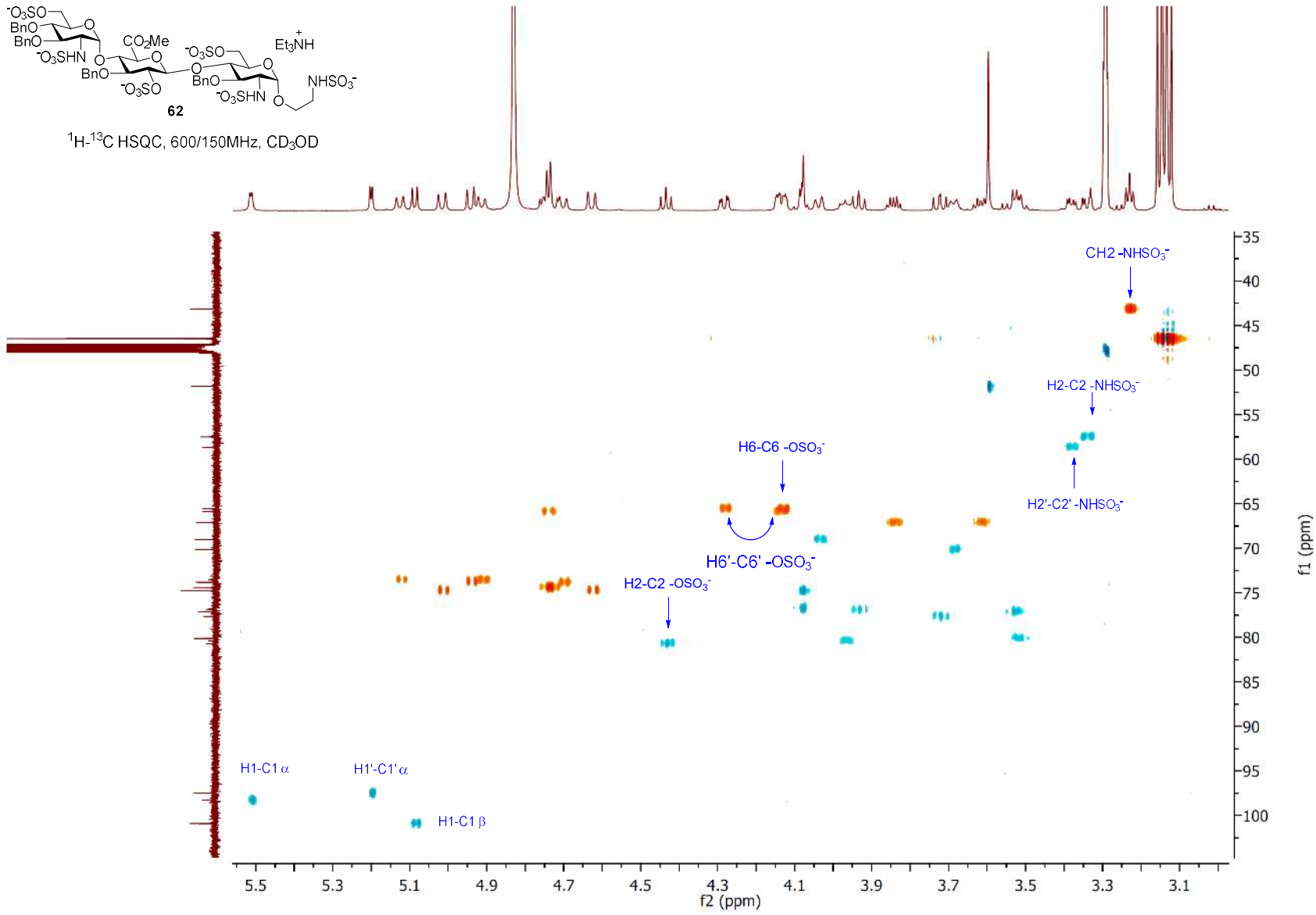

172.8672
172.0034
169.2200
141.2451
139.0257
138.6580
138.3255
135.0026
128.0896
127.8895
127.8631
127.8425
127.8135
127.6826
127.6502
127.6153
103.3438
102.3362
102.0364
84.7982
77.9529
75.5046
74.8807
74.4850
72.9452
72.4546
68.9672
68.7057
68.3541
67.2173
64.4804
61.8323
58.7238
58.4622
57.1986
43.0015
20.1080
19.0650
$^{-}O_3SO$
BnO
BnO
$^{-}O_3SHN$
CO_2Bn
BnO
BnO
AcO
AcO
$^{-}O_3SHN$
Na^+
$NHSO_3^-$
60
^{13}C NMR, 150 MHz, CD_3OD
230
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

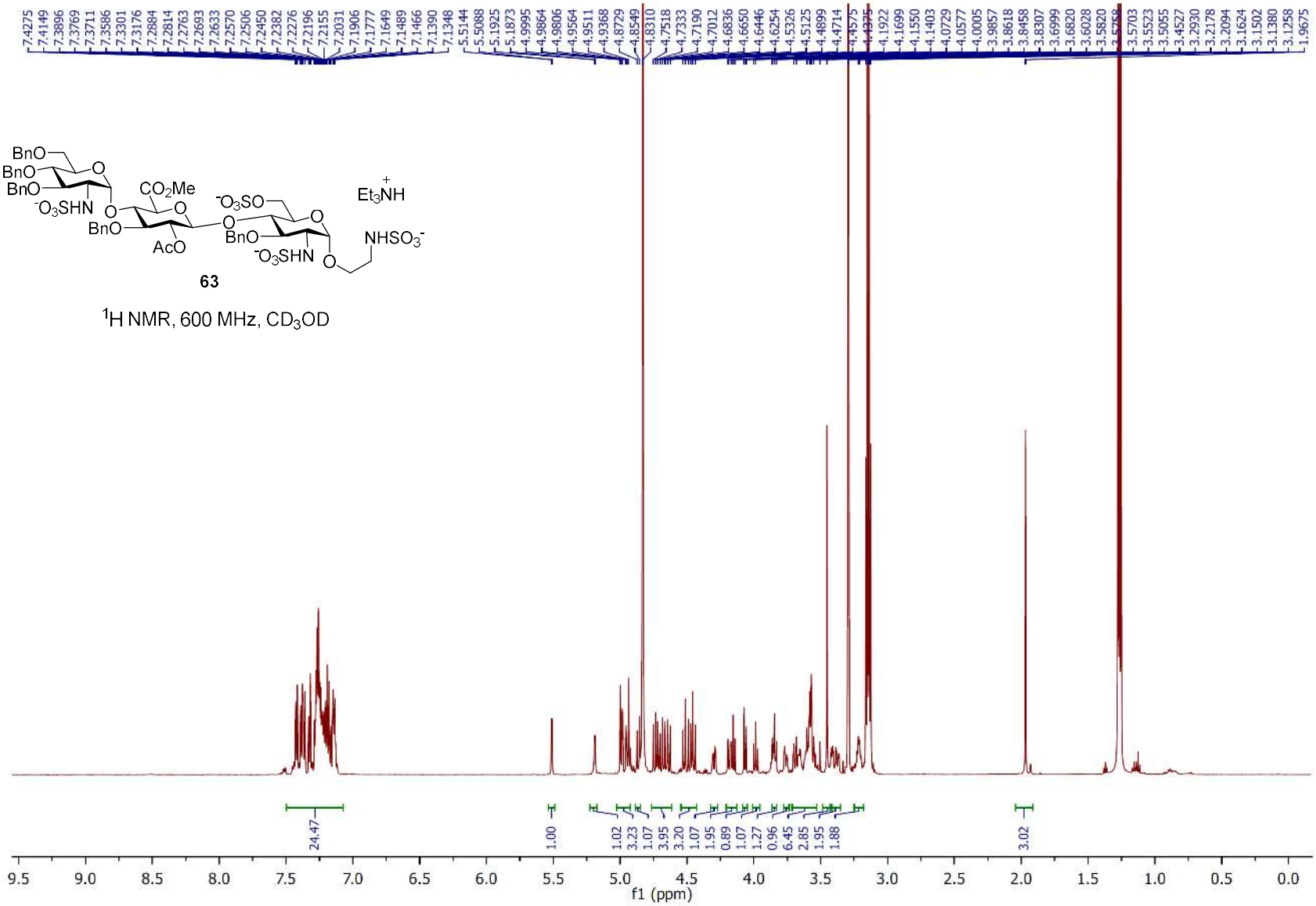

61
1H NMR, 600 MHz, CD3OD
f1 (ppm)

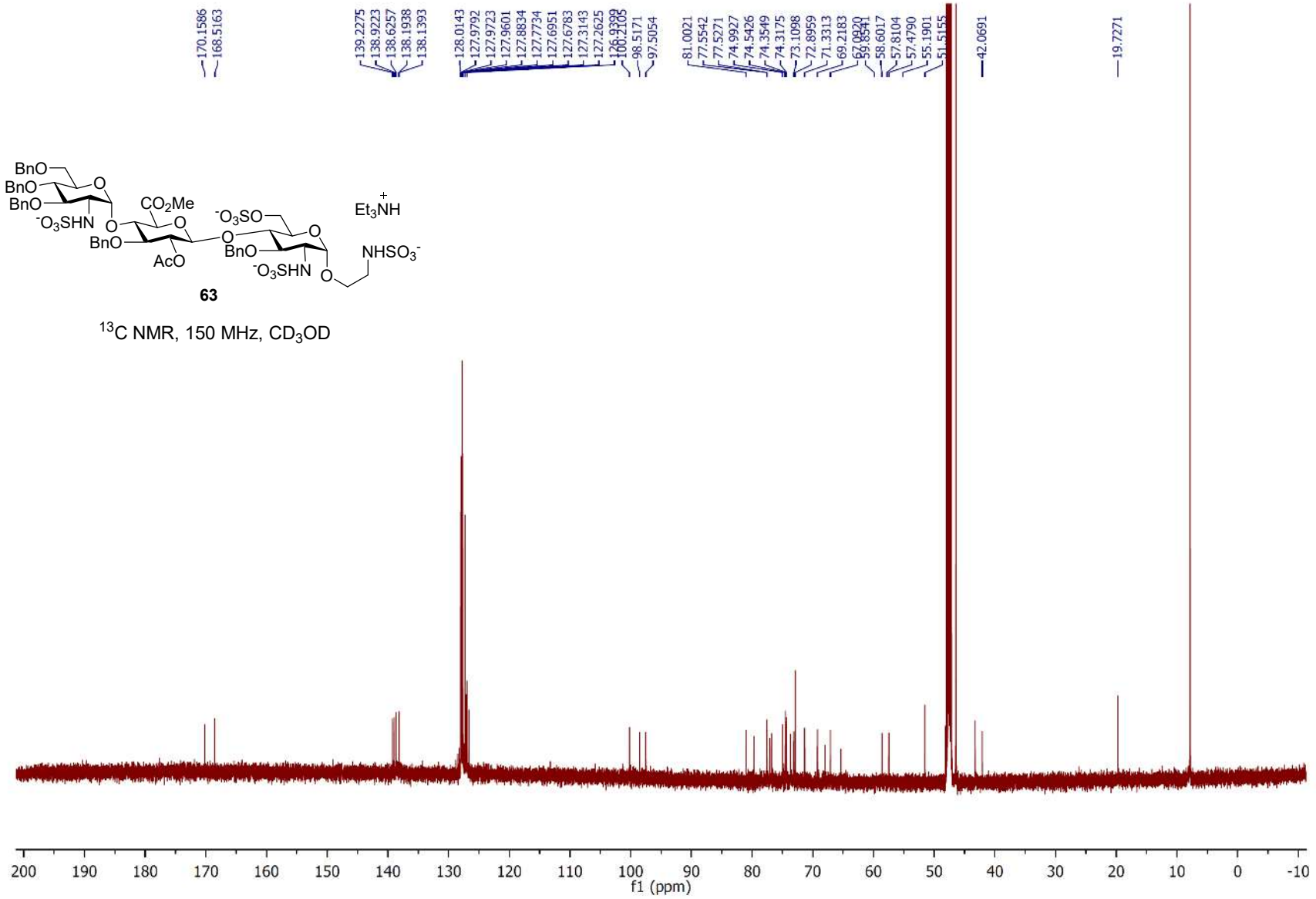

172.3666
169.0520
139.0100
138.6554
138.3548
138.3324
135.1521
128.1925
128.0259
127.9194
127.8975
127.8856
127.8594
127.7152
127.6863
127.6281
127.4705
127.3405
102.0835
98.1081
82.4526
81.7127
80.4660
76.9566
74.9110
74.6052
74.5396
73.9382
72.9446
70.2636
68.6931
67.2933
65.5251
58.3906
58.2079
43.0030
20.1037
Na+
61
13C NMR, 150 MHz, CD3OD
f1 (ppm)

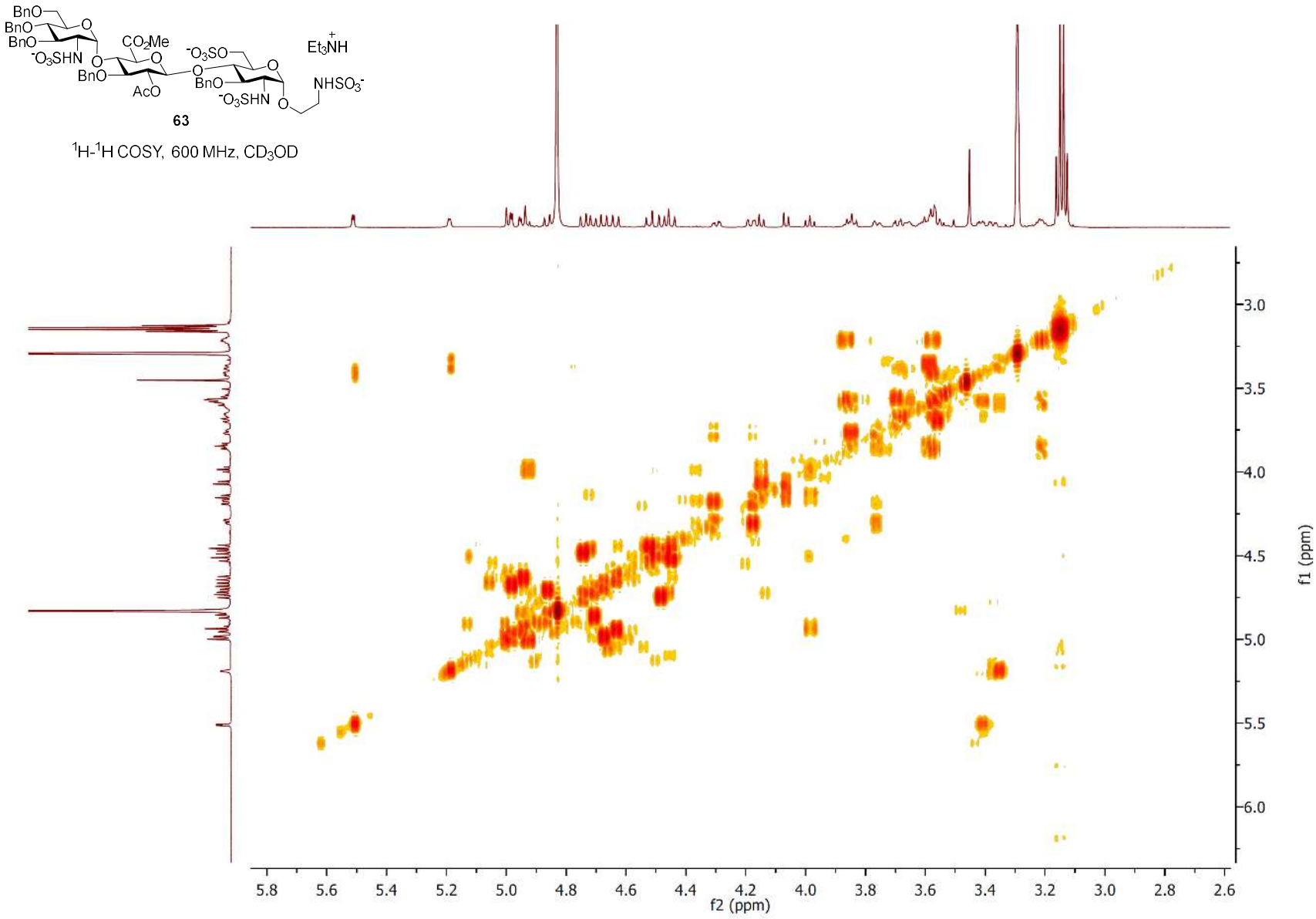

62
1H NMR, 600 MHz, CD3OD
f1 (ppm)

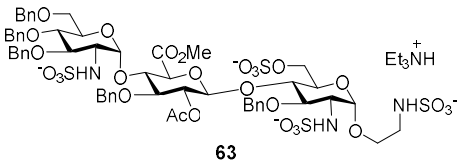

168.9585
139.3882
139.2224
138.7096
138.2096
129.5706
128.1483
127.8797
127.7861
127.7094
127.6969
127.6520
127.1019
126.9850
126.8264
126.6882
100.9187
98.2743
97.5032
88.1487
80.1352
77.1250
74.7727
74.4570
73.8240
70.1584
69.0257
67.1053
58.8831
58.1882
57.5019
56.1852
51.8336
43.1409
62
13C NMR, 150 MHz, CD3OD
f1 (ppm)


62
^{1}H-^{13}C HSQC, 600/150MHz, CD$_3$OD
CH2 -NHSO$_3^-$
H2-C2 -NHSO$_3^-$
H6-C6 -OSO$_3^-$
H2'-C2' -NHSO$_3^-$
H6'-C6' -OSO$_3^-$
H2-C2 -OSO$_3^-$
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)


BnO
BnO
BnO
O
$^{-}O_3SHN$
CO_2Me
O
BnO
AcO
O
$^{-}O_3SO$
O
BnO
$^{-}O_3SHN$
O
Et_3NH^{+}
$NHSO_3^{-}$
63
^{1}H NMR, 600 MHz, CD_3OD
24.47
1.00
1.02
3.23
1.07
3.95
3.20
1.07
1.95
0.89
1.07
1.27
0.96
6.45
2.85
1.95
1.88
3.02
f1 (ppm)

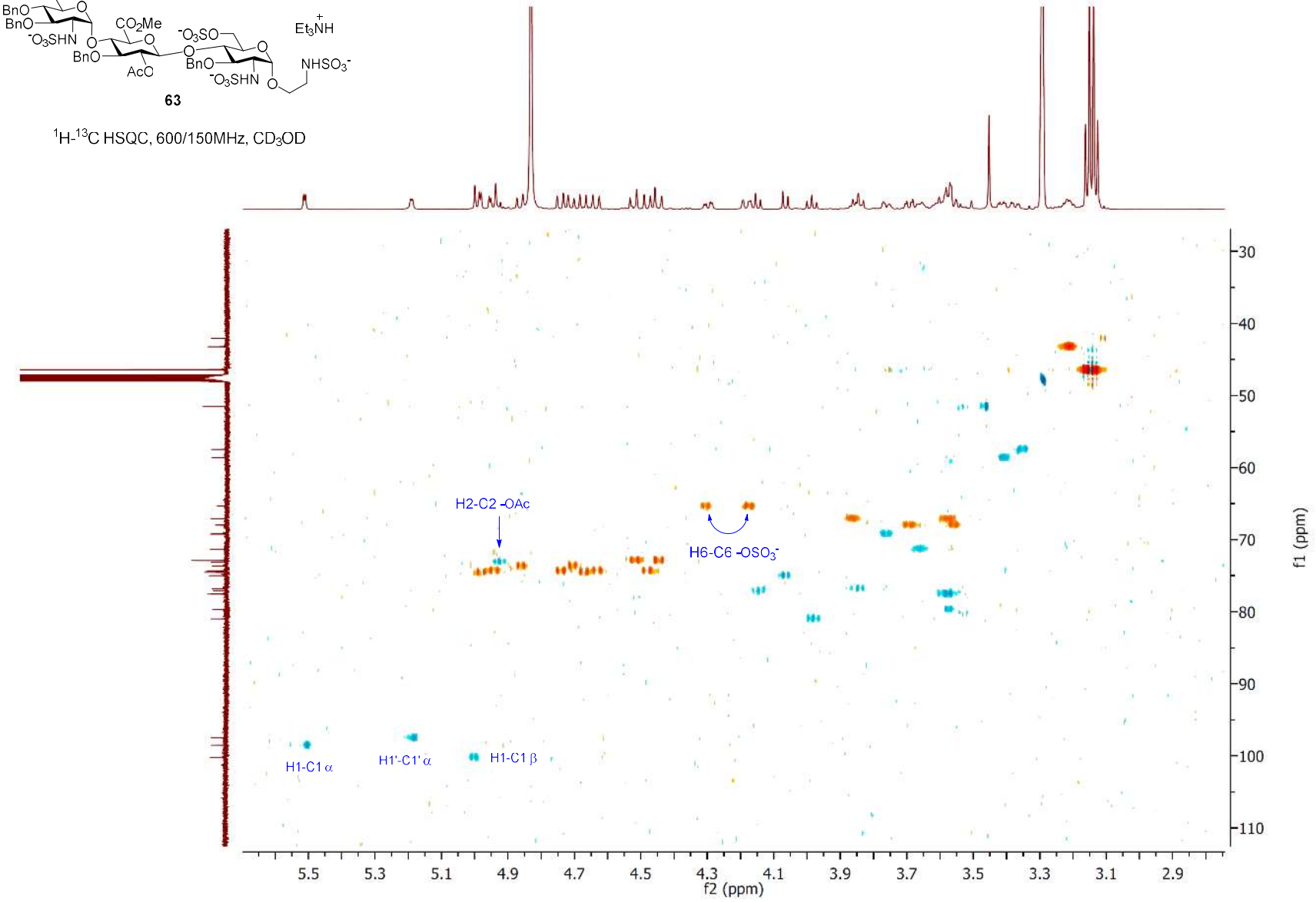

170.1586
168.5163
139.2275
138.9223
138.6257
138.1938
138.1393
128.0143
127.9792
127.9723
127.9601
127.8834
127.7734
127.6951
127.6783
127.3143
127.2625
100.2105
98.5171
97.5054
81.0021
77.5542
77.5271
74.9927
74.5426
74.3549
74.3175
73.1098
72.8959
71.3313
69.2183
58.6017
57.8104
57.4790
55.1901
51.5155
42.0691
19.7271
BnO
BnO
BnO
$^{-}O_3SHN$
O
CO_2Me
O
BnO
AcO
O
$^{-}O_3SO$
O
BnO
$^{-}O_3SHN$
O
Et_3NH^{+}
$NHSO_3^{-}$
63
^{13}C NMR, 150 MHz, CD_3OD
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

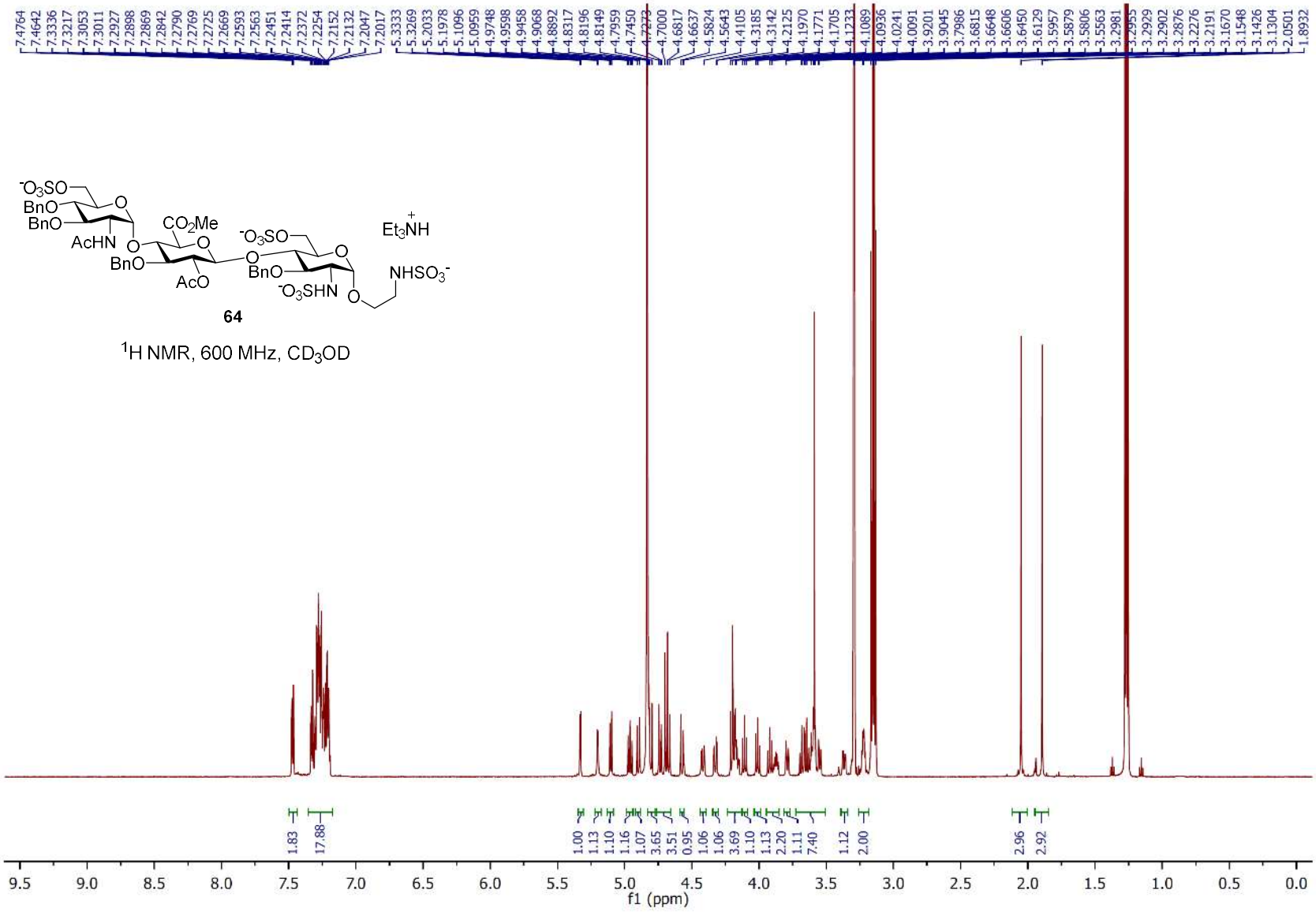

BnO
BnO
BnO
$^{-}O_3SHN$
CO_2Me
AcO
$^{-}O_3SO$
BnO
$^{-}O_3SHN$
$Et_3\overset{+}{N}H$
$NHSO_3^{-}$
63
^1H-^1H COSY, 600 MHz, CD_3OD
5.8
5.6
5.4
5.2
5.0
4.8
4.6
4.4
4.2
4.0
3.8
3.6
3.4
3.2
3.0
2.8
2.6
f2 (ppm)
3.0
3.5
4.0
4.5
5.0
5.5
6.0
f1 (ppm)

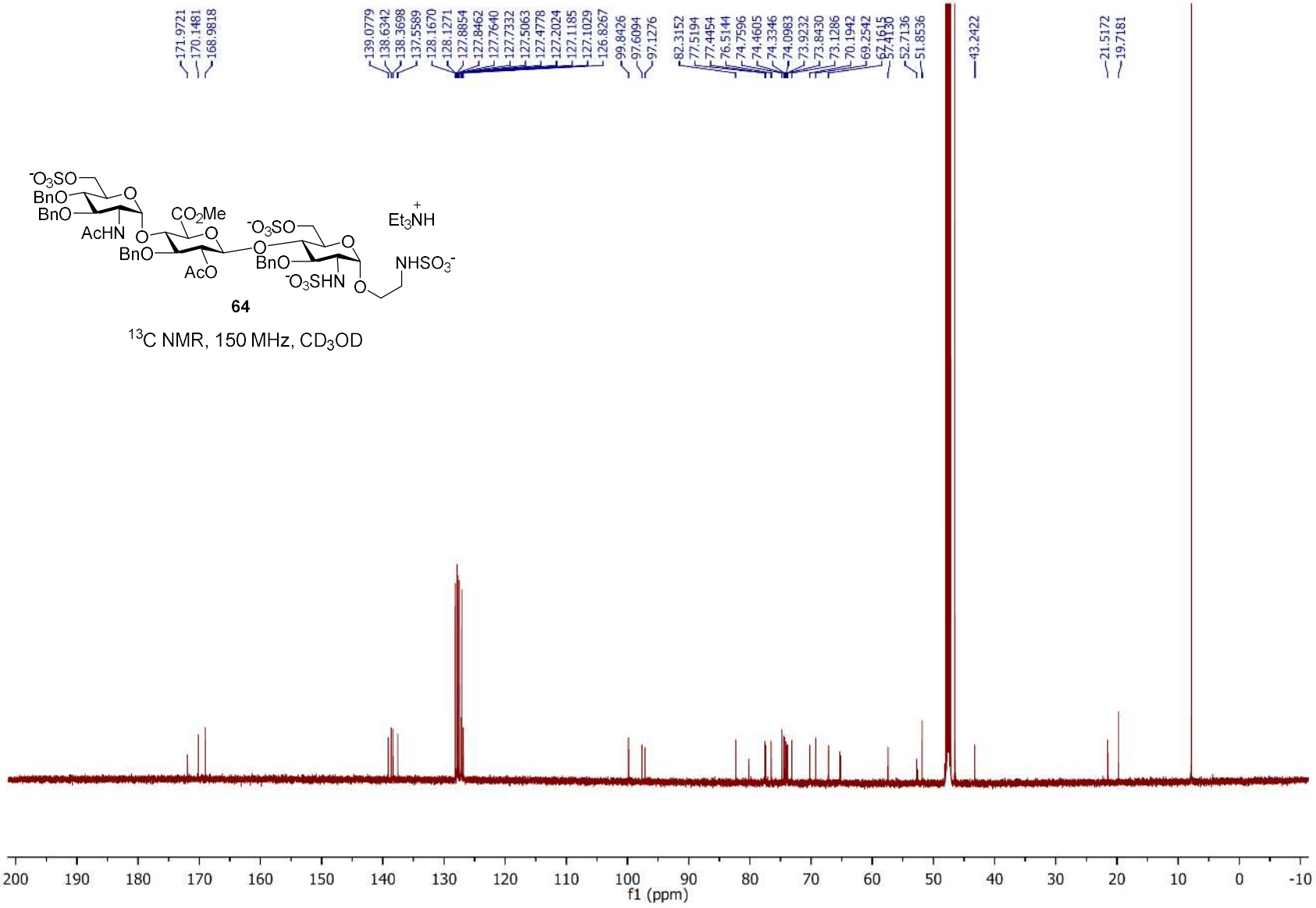


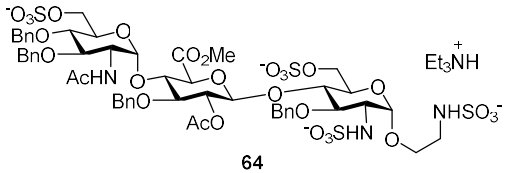
63

^{1}H-^{13}C HSQC, 600/150MHz, CD_3OD



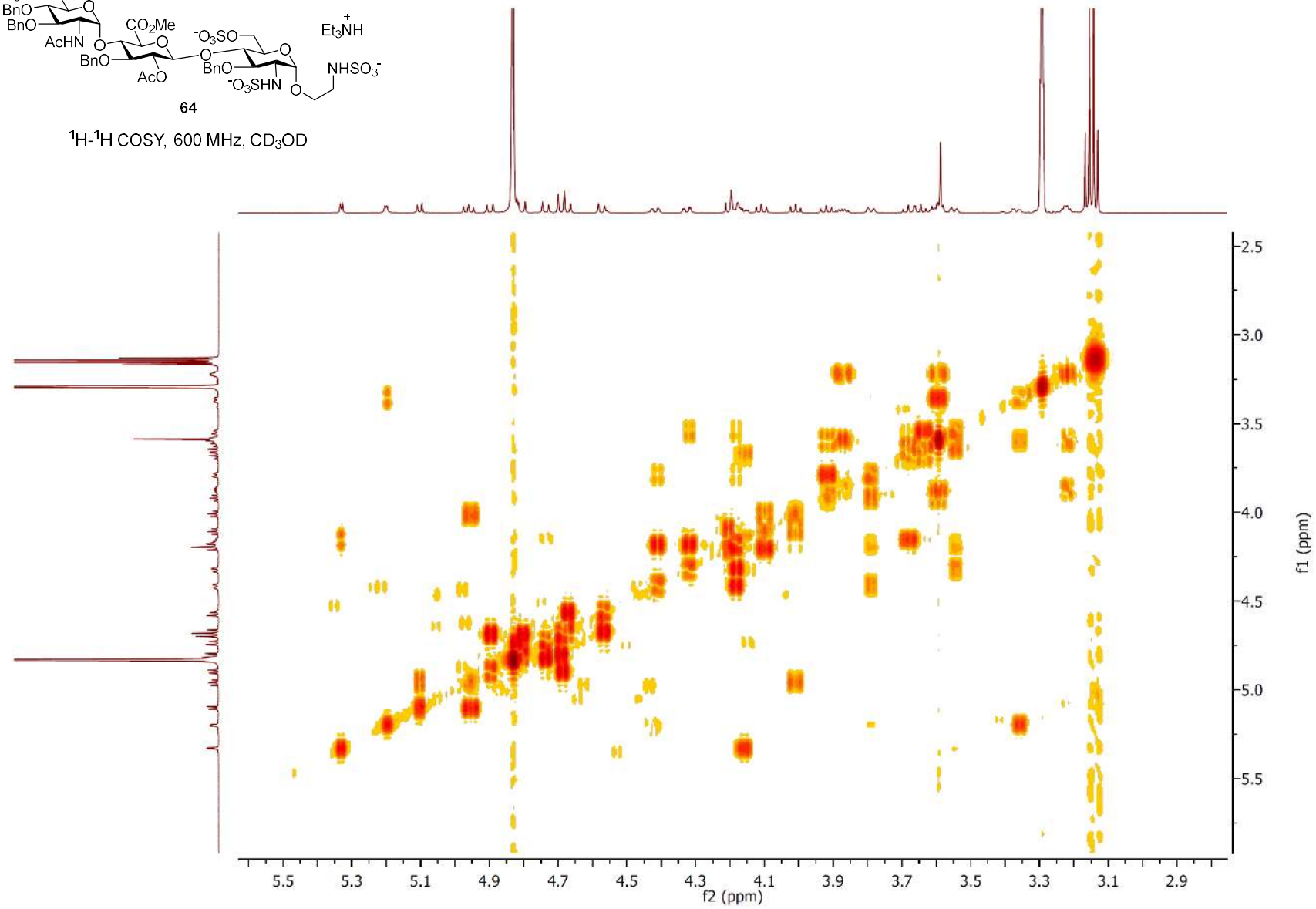

64
1H NMR, 600 MHz, CD3OD
f1 (ppm)

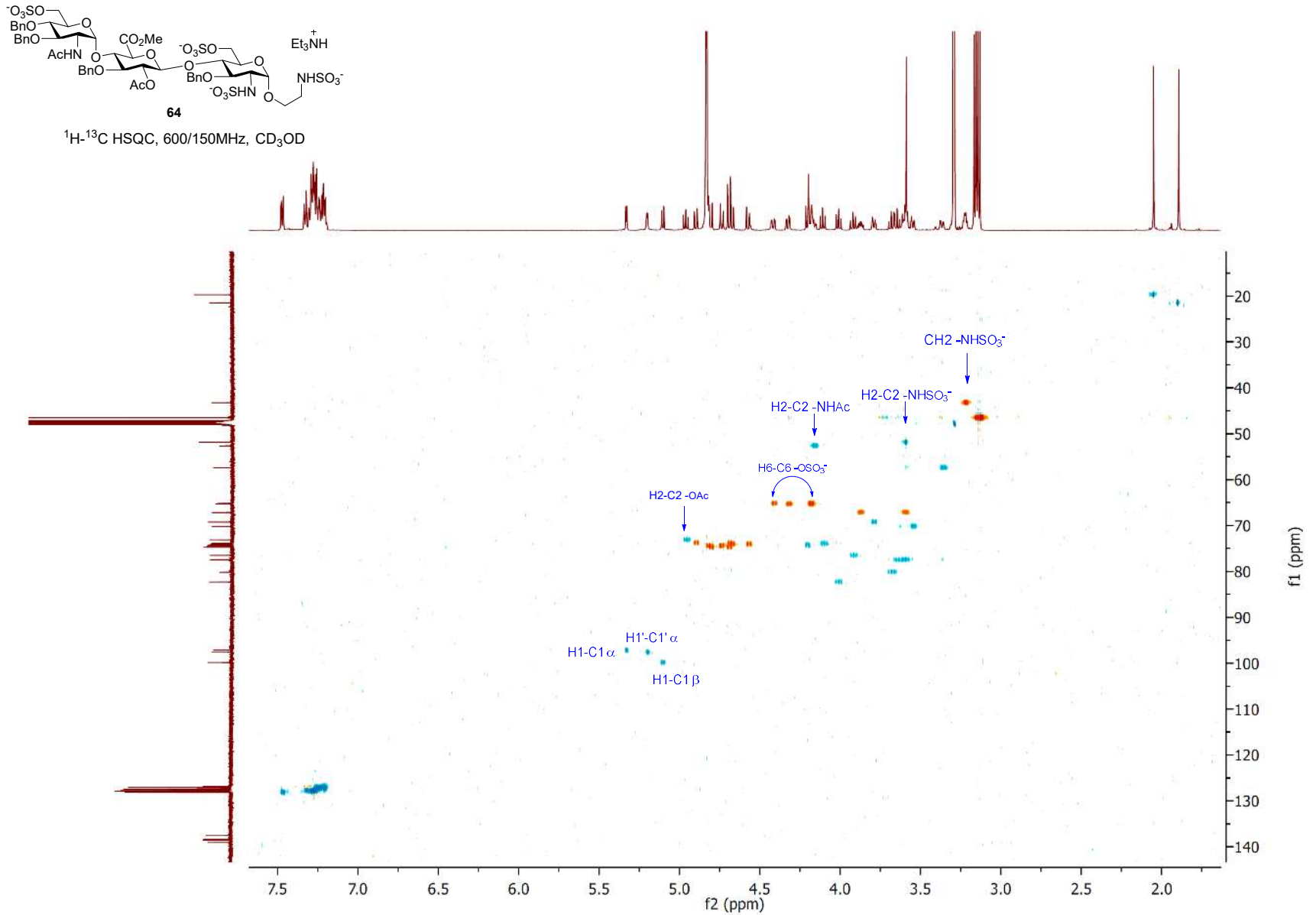

171.9721
170.1481
168.9818
139.0779
138.6342
138.3698
137.5589
128.1670
128.1271
127.8854
127.8462
127.7640
127.7332
127.5063
127.4778
127.2024
127.1185
127.1029
126.8267
99.8426
97.6094
97.1276
82.3152
77.5194
77.4454
76.5144
74.7596
74.4605
74.3346
74.0983
73.9232
73.8430
73.1286
70.1942
69.2542
67.1615
57.4130
52.7136
51.8536
43.2422
21.5172
19.7181
64
13C NMR, 150 MHz, CD3OD
f1 (ppm)

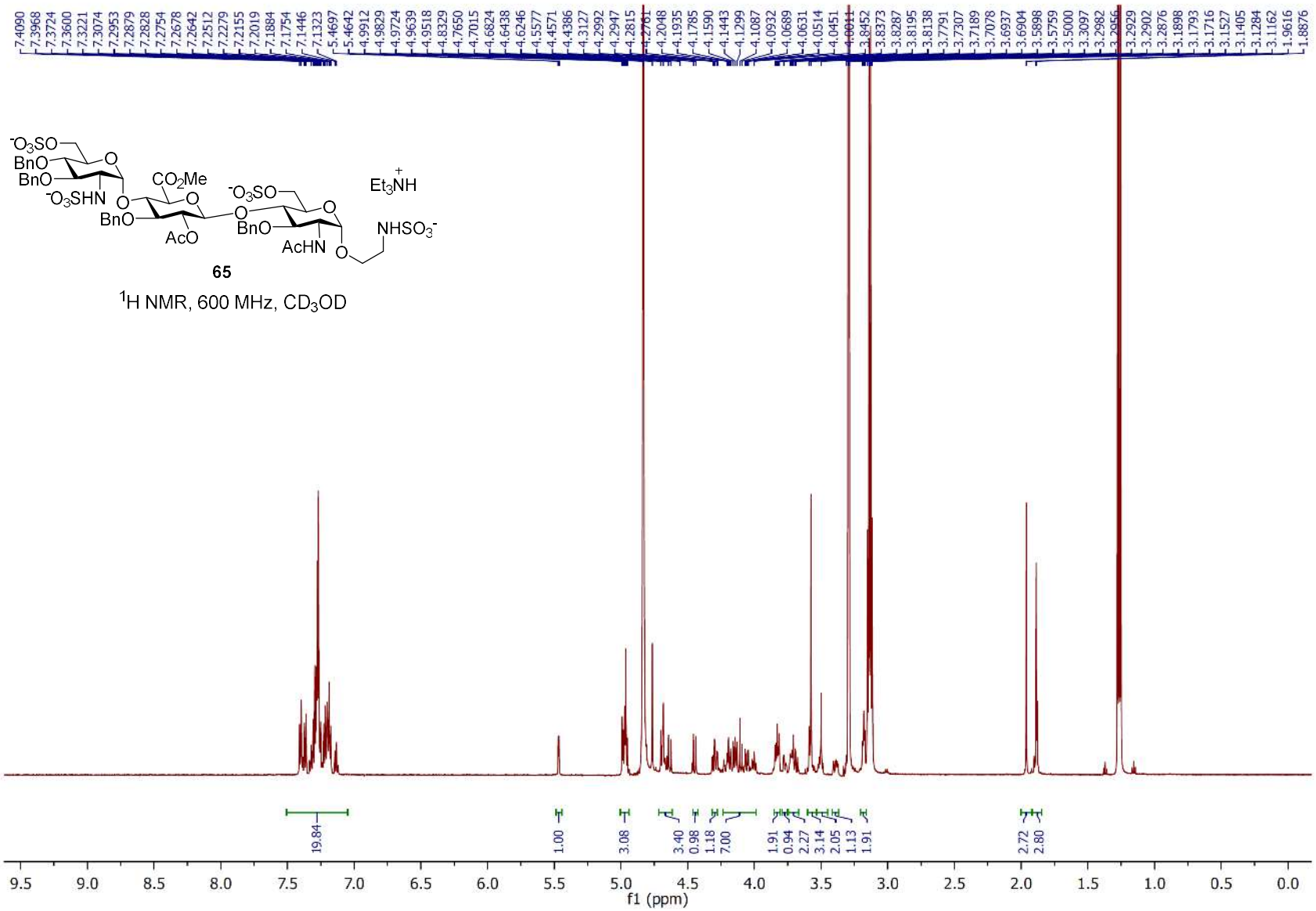


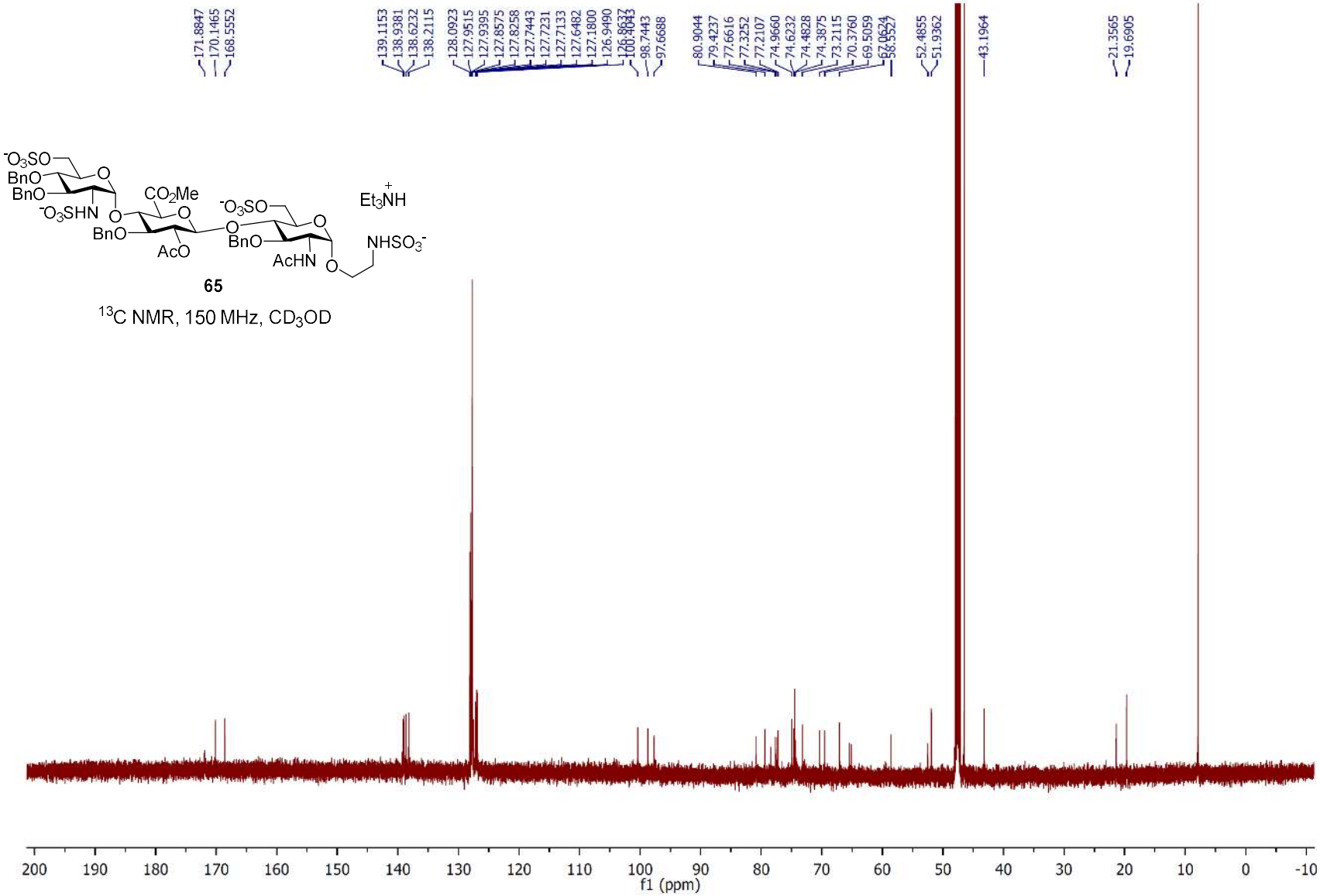
64

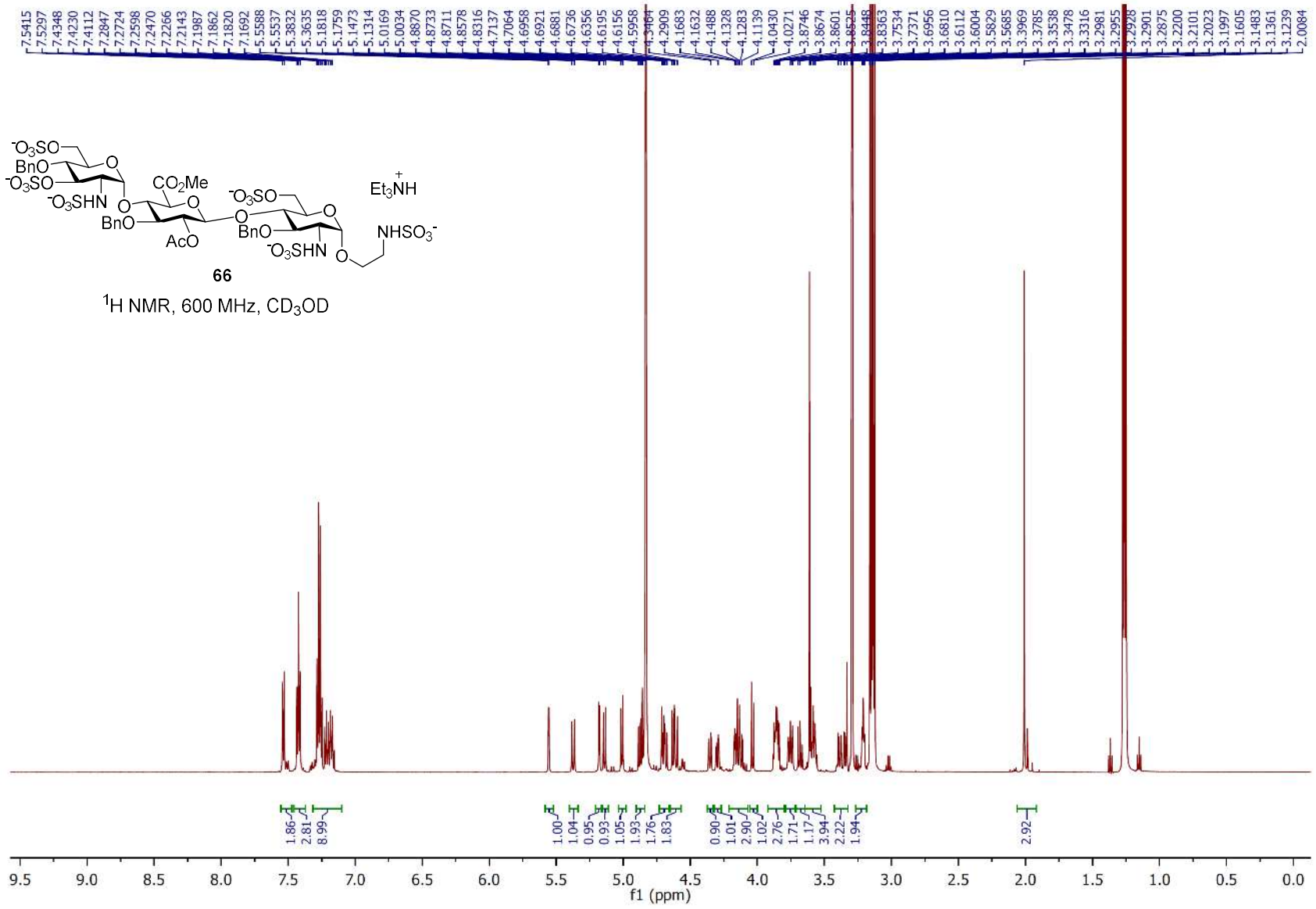
^{1}H-^{1}H COSY, 600 MHz, CD_3OD

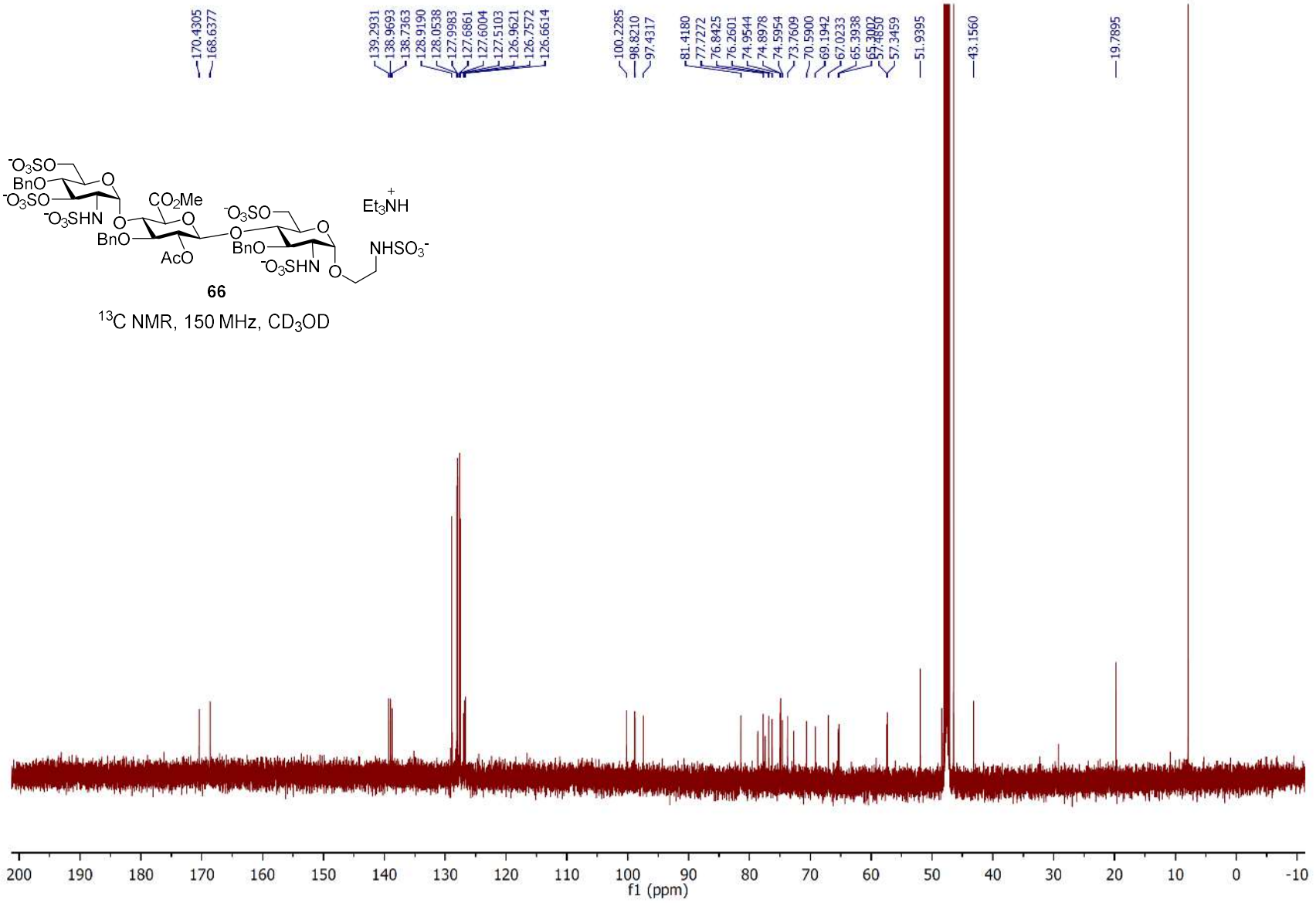


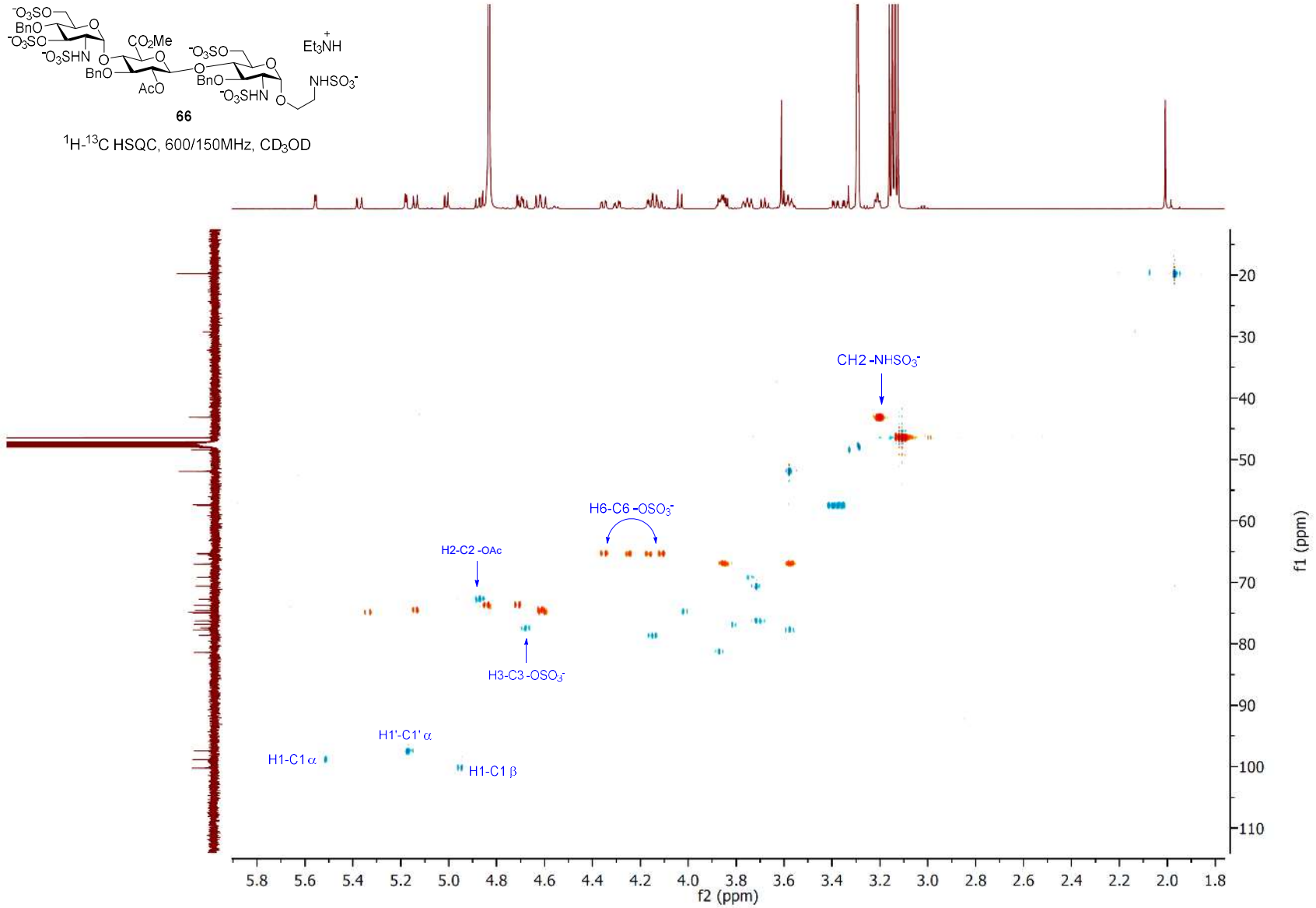

64
^{1}H-^{13}C HSQC, 600/150MHz, CD_3OD
CH2 -NHSO3-
H2-C2 -NHSO3-
H2-C2 -NHAc
H6-C6 -OSO3-
H2-C2 -OAc
H1'-C1' α
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)

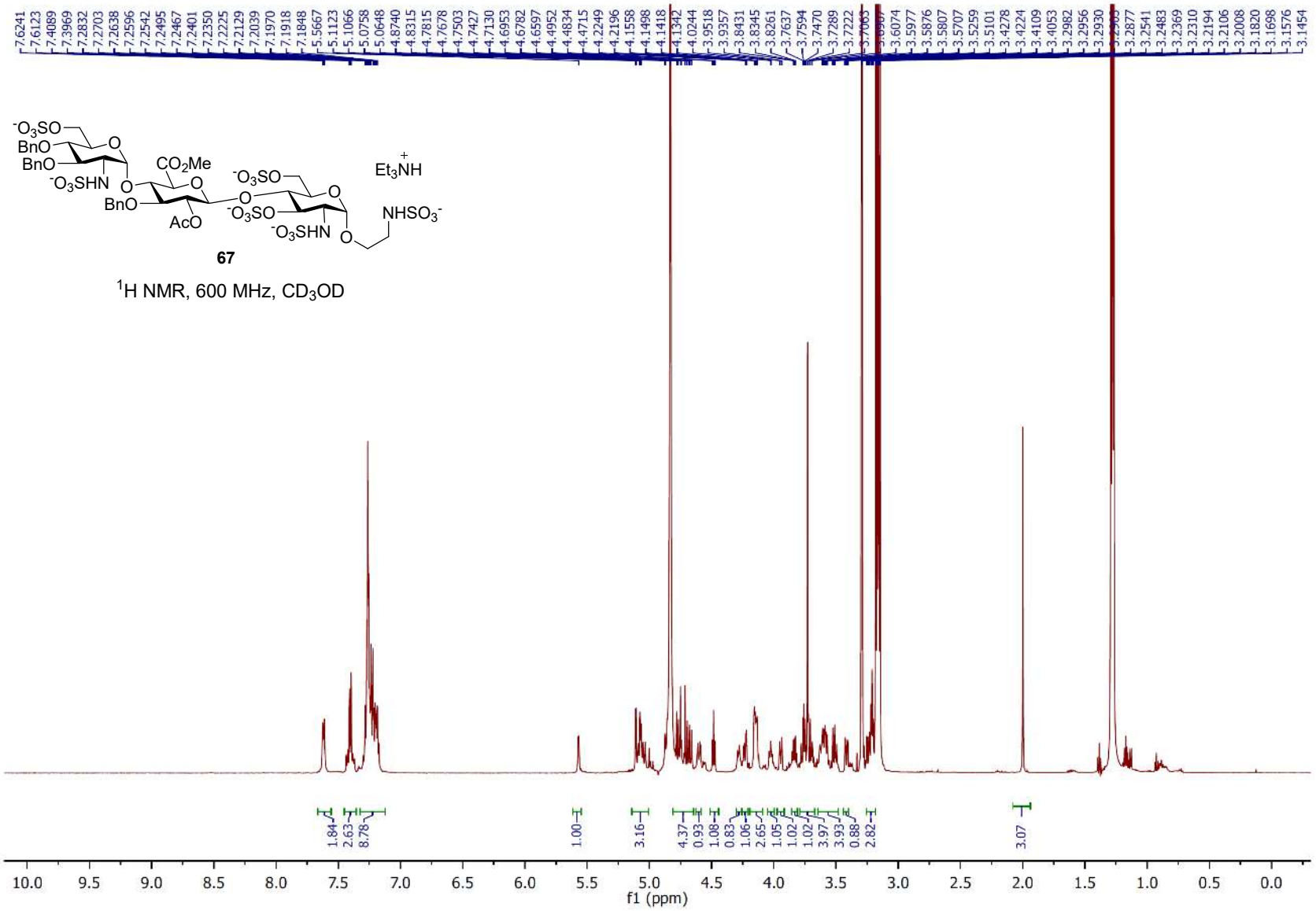

65
1H NMR, 600 MHz, CD3OD
f1 (ppm)

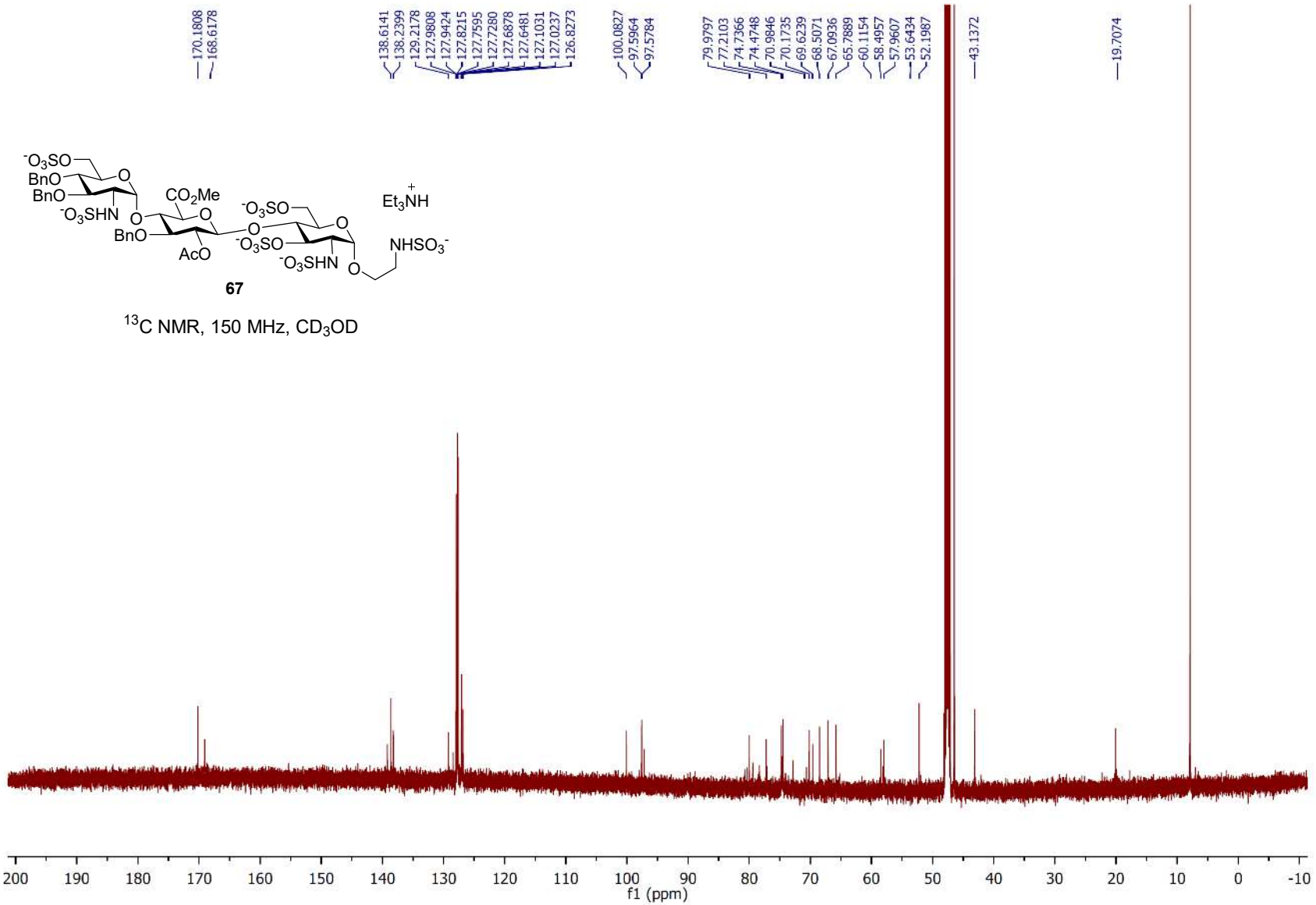

171.8847
170.1465
168.5552
139.1153
138.9381
138.6232
138.2115
128.0923
127.9515
127.9395
127.8575
127.8258
127.7443
127.7231
127.7133
127.6482
127.1800
126.9490
98.7443
97.6688
80.9044
79.4237
77.6616
77.3252
77.2107
74.9660
74.6232
74.4828
74.3875
73.2115
70.3760
69.5059
52.4855
51.9362
43.1964
21.3565
19.6905
65
13C NMR, 150 MHz, CD3OD
f1 (ppm)


66
1H NMR, 600 MHz, CD3OD
f1 (ppm)


170.4305
168.6377
139.2931
138.9693
138.7363
128.9190
128.0538
127.9983
127.6861
127.6004
127.5103
126.9621
126.7572
126.6614
100.2285
98.8210
97.4317
81.4180
77.7272
76.8425
76.2601
74.9544
74.8978
74.5954
73.7609
70.5900
69.1942
67.0233
65.3938
65.3002
57.4850
57.3459
51.9395
43.1560
19.7895
$^{-}O_3SO$
BnO
$^{-}O_3SO$
$^{-}O_3SHN$
CO_2Me
BnO
AcO
$^{-}O_3SO$
BnO
$^{-}O_3SHN$
$Et_3\overset{+}{N}H$
$NHSO_3^{-}$
66
^{13}C NMR, 150 MHz, CD_3OD
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

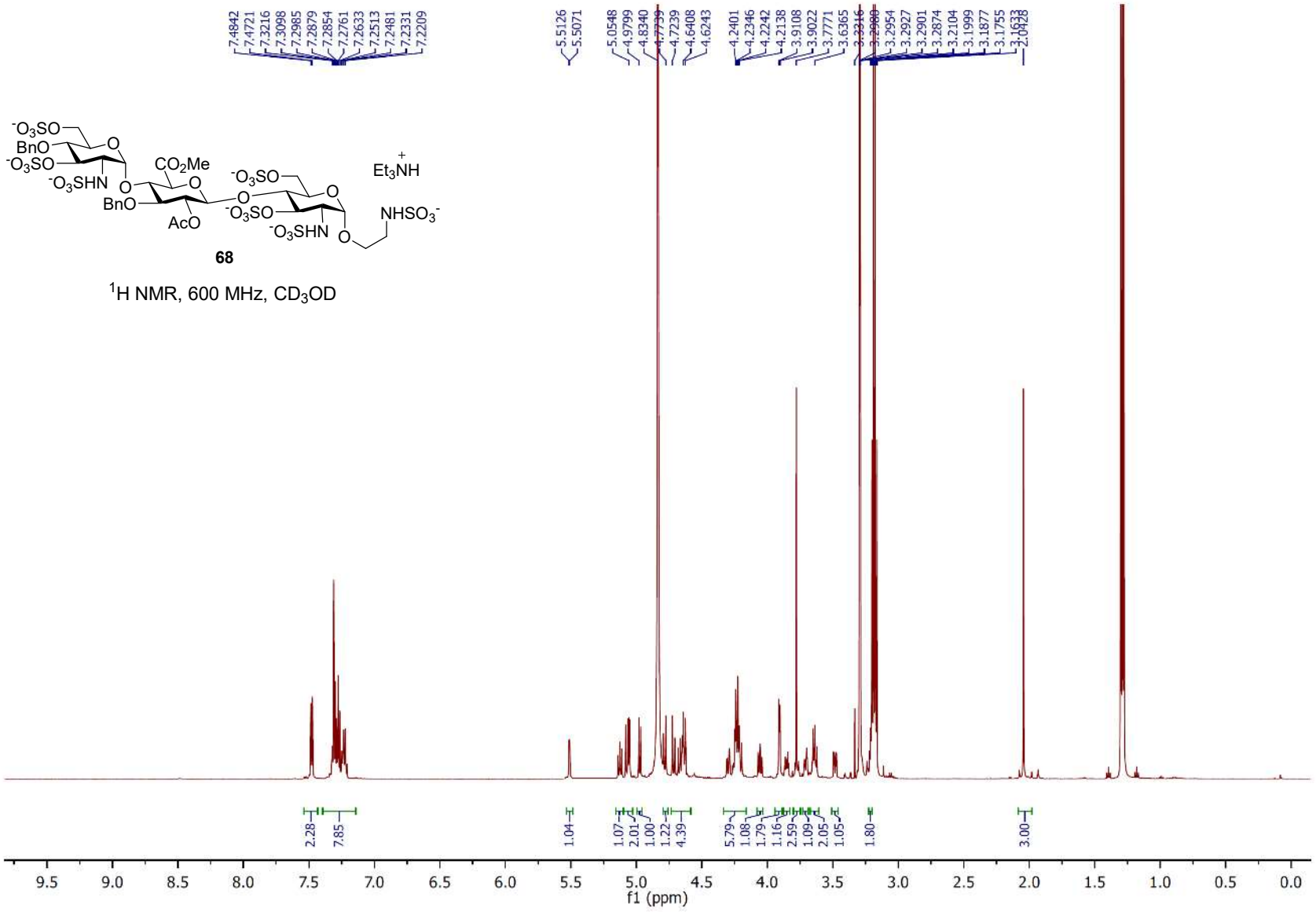

66
1H-13C HSQC, 600/150MHz, CD3OD
CH2 -NHSO3-
H6-C6 -OSO3-
H2-C2 -OAc
H3-C3 -OSO3-
H1'-C1' α
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)

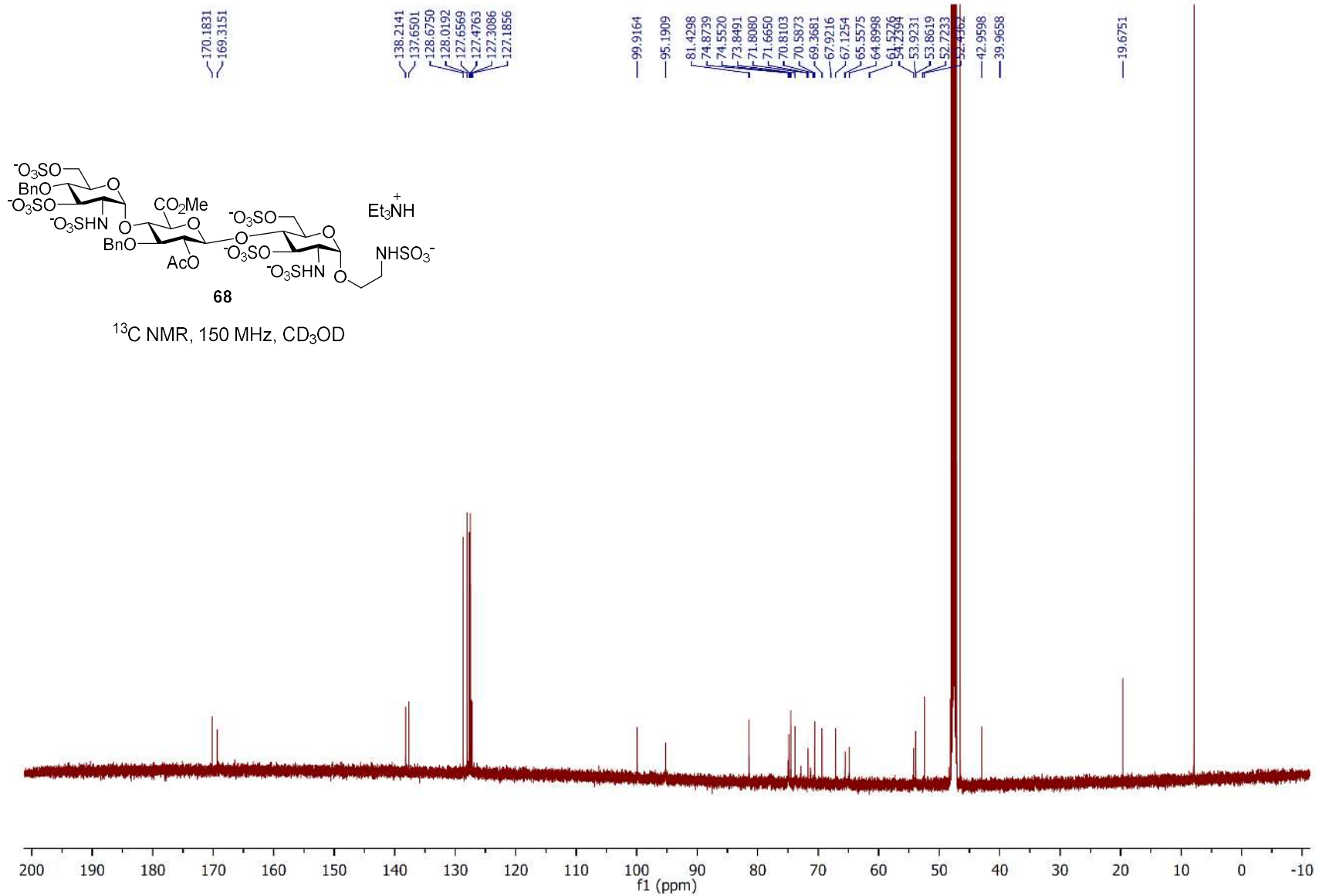

$^{-}O_3SO$
BnO
BnO
$^{-}O_3SHN$
CO_2Me
BnO
AcO
$^{-}O_3SO$
$^{-}O_3SO$
$^{-}O_3SHN$
Et_3NH^{+}
$NHSO_3^{-}$
67
^1H NMR, 600 MHz, CD_3OD
f1 (ppm)

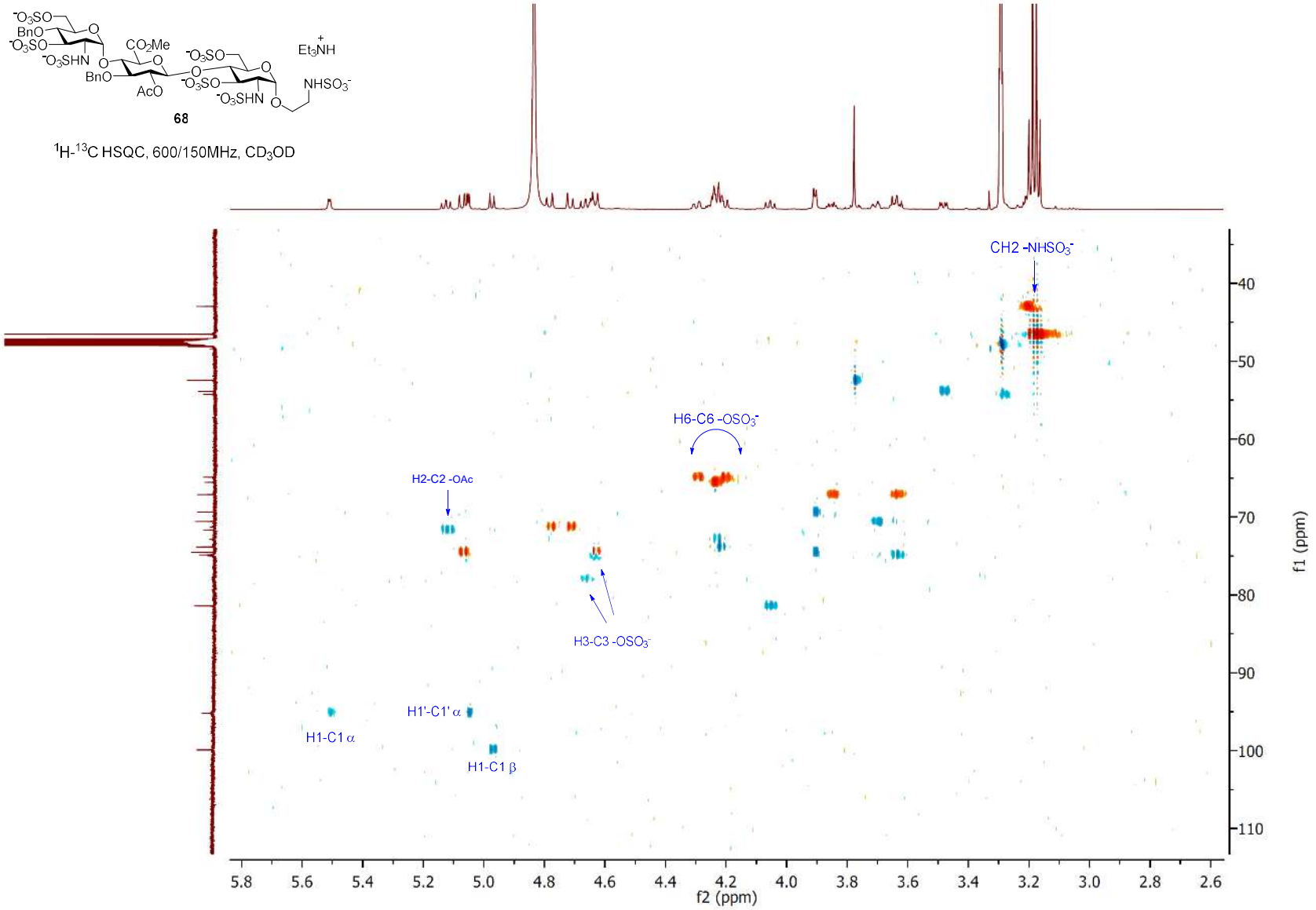


67

^{13}C NMR, 150 MHz, CD_3OD

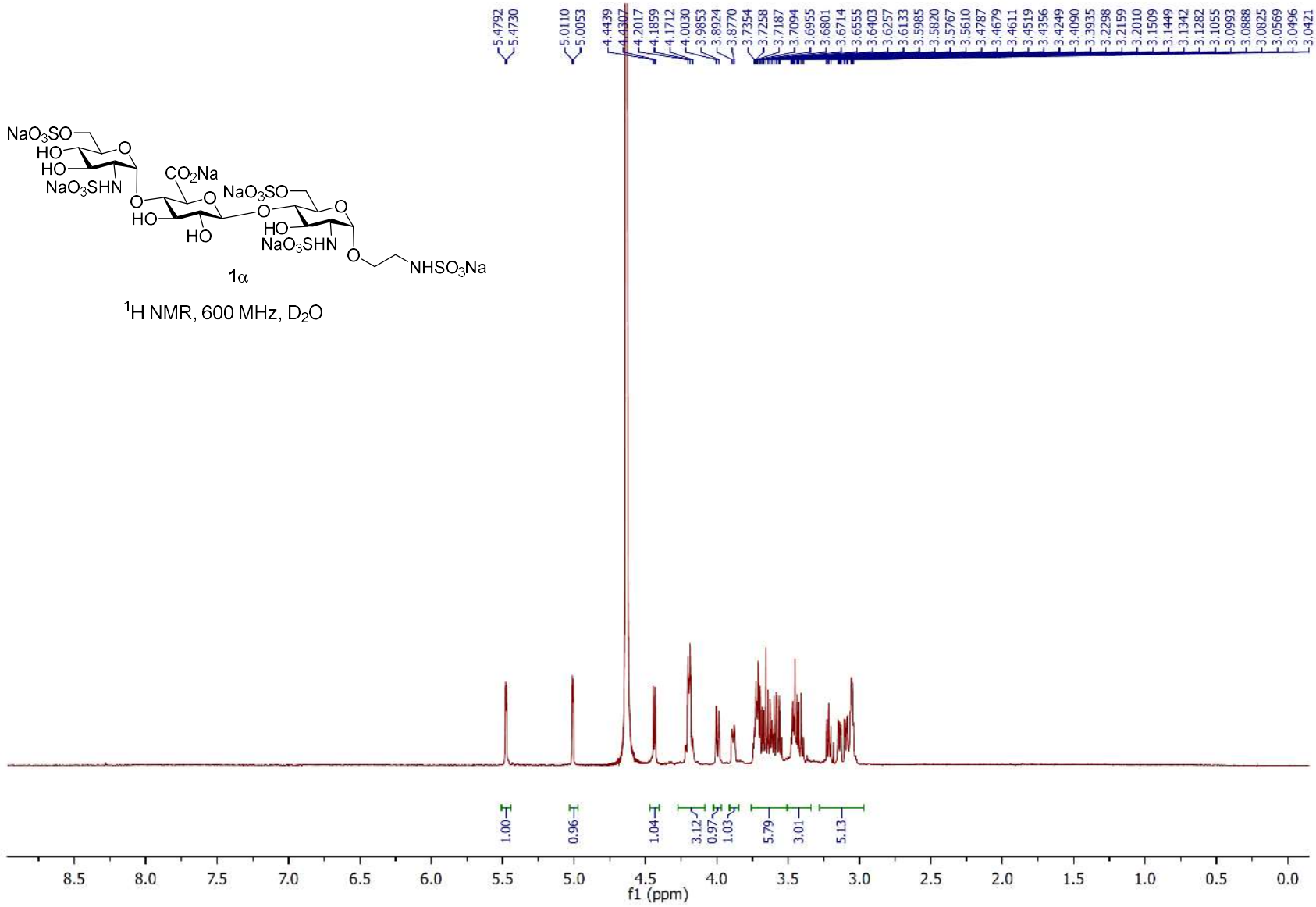

68
1H NMR, 600 MHz, CD3OD
f1 (ppm)

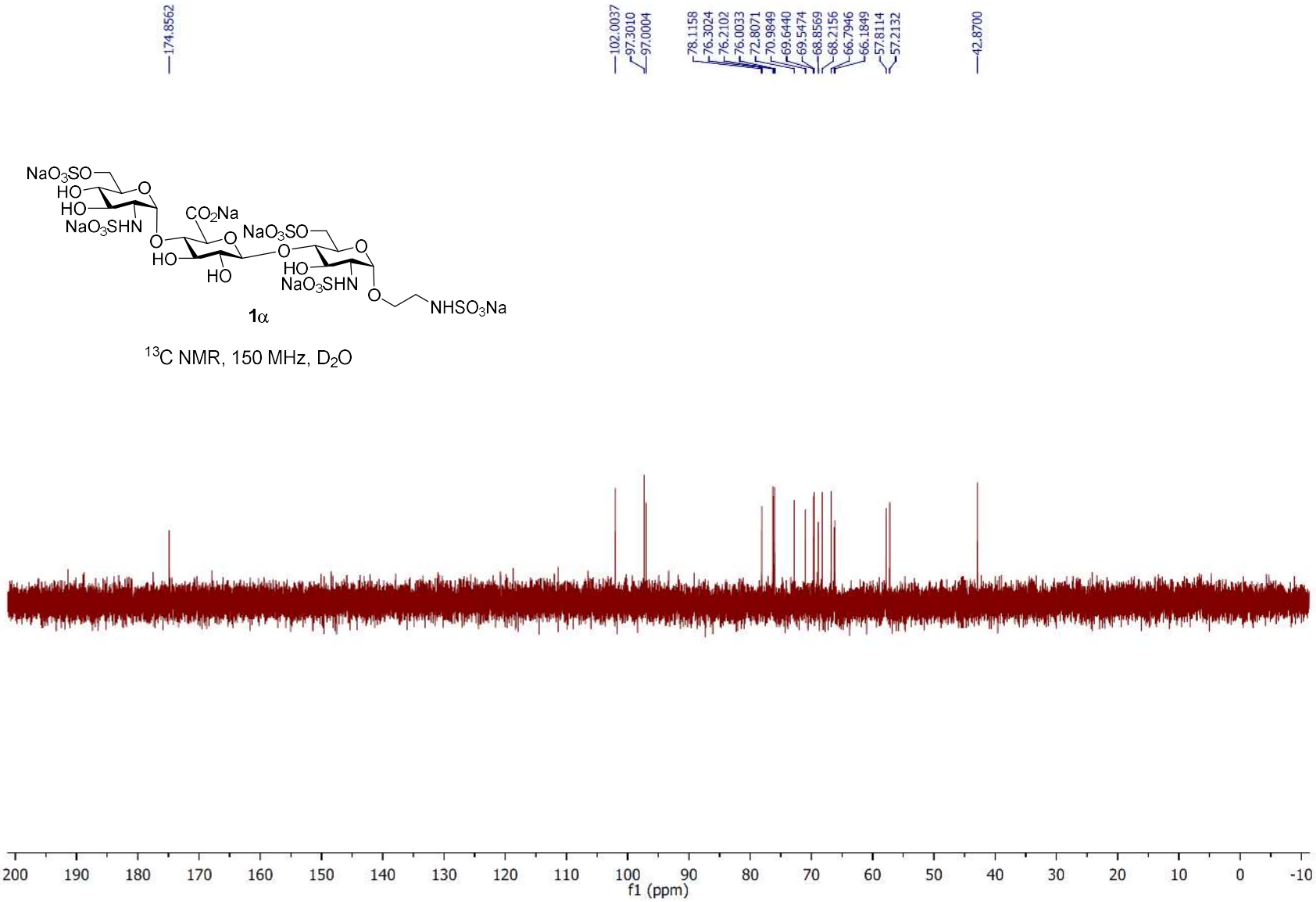

170.1831
169.3151
138.2141
137.6501
128.6750
128.0192
127.6569
127.4763
127.3086
127.1856
99.9164
95.1909
81.4298
74.8739
74.5520
73.8491
71.8080
71.6650
70.8103
70.5873
69.3681
67.9216
67.1254
65.5575
64.8998
61.5276
54.2394
53.9231
53.8619
52.7233
52.4362
42.9598
39.9658
19.6751
$^{-}O_3SO$
BnO
$^{-}O_3SO$
$^{-}O_3SHN$
CO_2Me
AcO
BnO
$^{-}O_3SO$
$^{-}O_3SO$
$^{-}O_3SHN$
Et_3NH^{+}
$NHSO_3^{-}$
68
^{13}C NMR, 150 MHz, CD_3OD
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

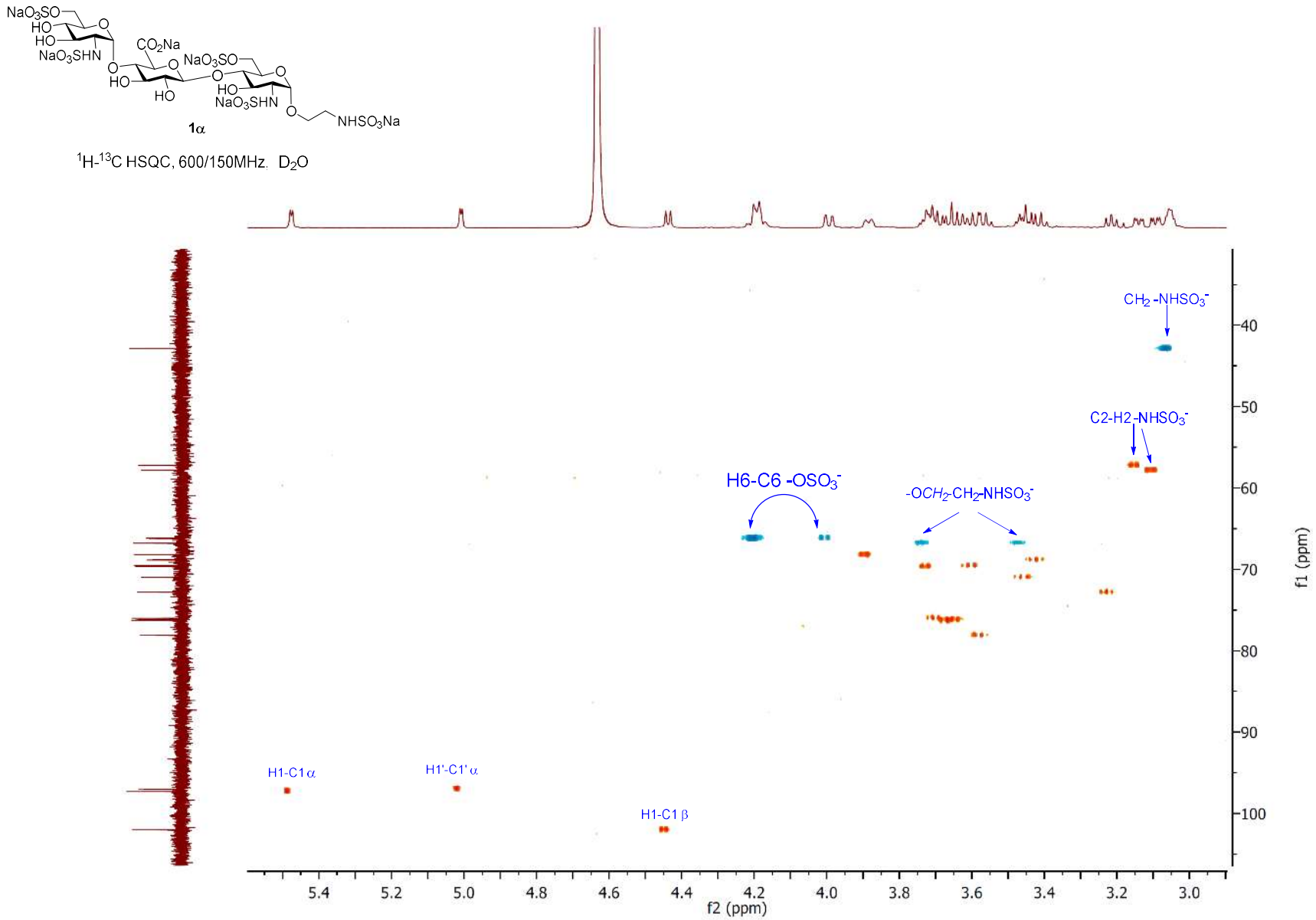

68
^{1}H-^{13}C HSQC, 600/150MHz, CD$_3$OD
CH2 -NHSO$_3^-$
H6-C6 -OSO$_3^-$
H2-C2 -OAc
H3-C3 -OSO$_3^-$
H1'-C1' α
H1-C1 α
H1-C1 β
f2 (ppm)
f1 (ppm)

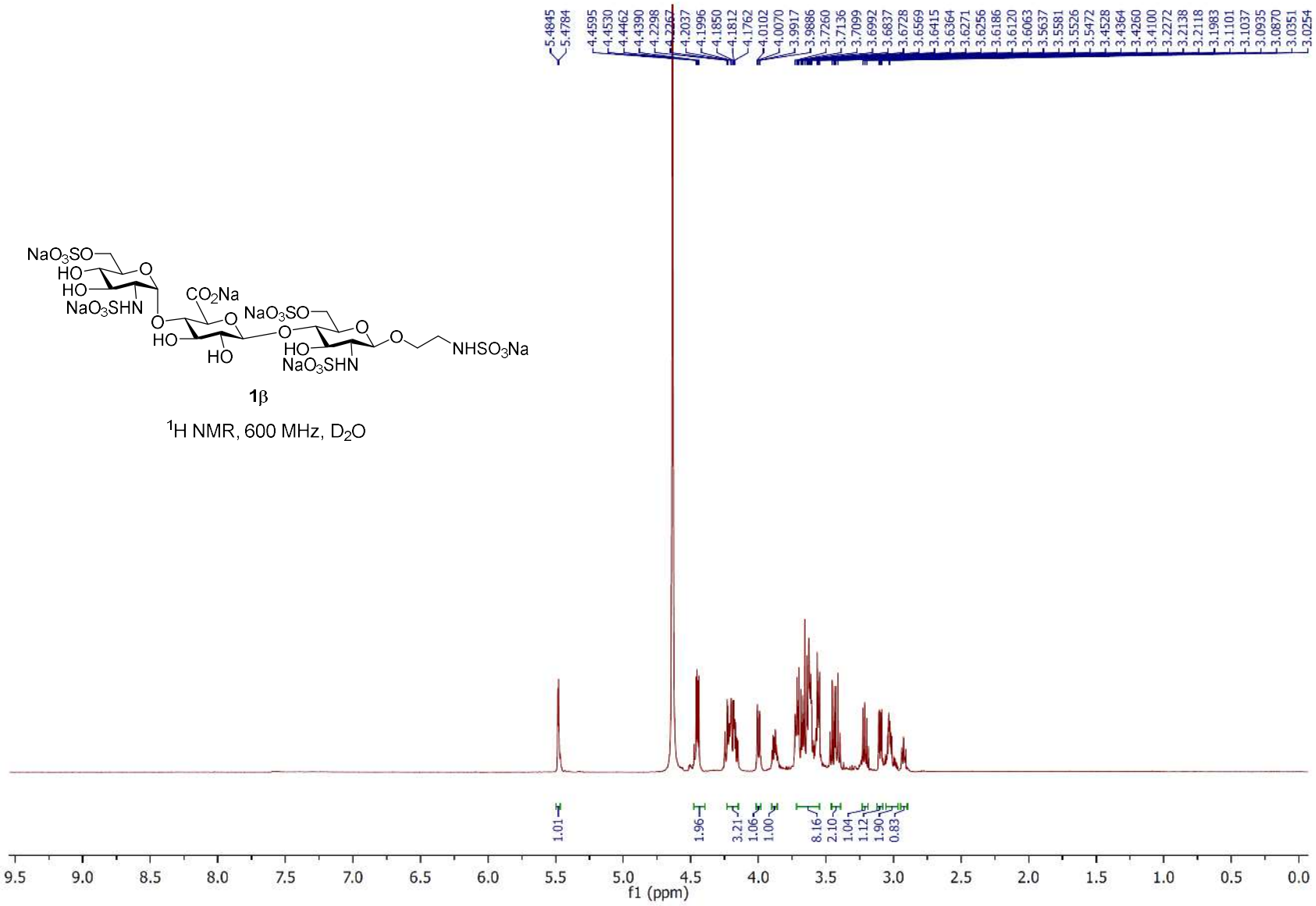
1α

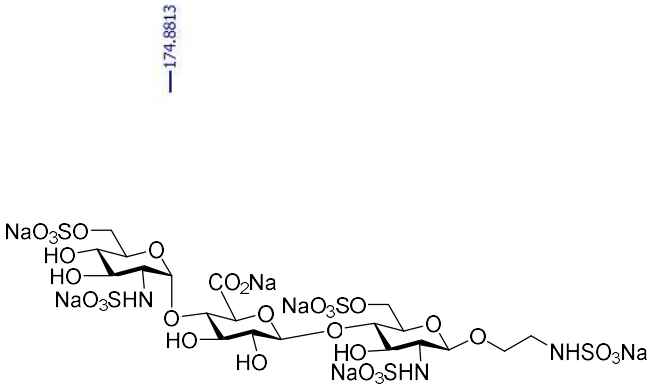
^{1}H NMR, 600 MHz, D_2O




174.8562
102.0037
97.3010
97.0004
78.1158
76.3024
76.2102
76.0033
72.8071
70.9849
69.6440
69.5474
68.8569
68.2156
66.7946
66.1849
57.8114
57.2132
42.8700
NaO3SO
HO
HO
NaO3SHN
O
CO2Na
NaO3SO
HO
HO
HO
NaO3SHN
NHSO3Na
1α
13C NMR, 150 MHz, D2O
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

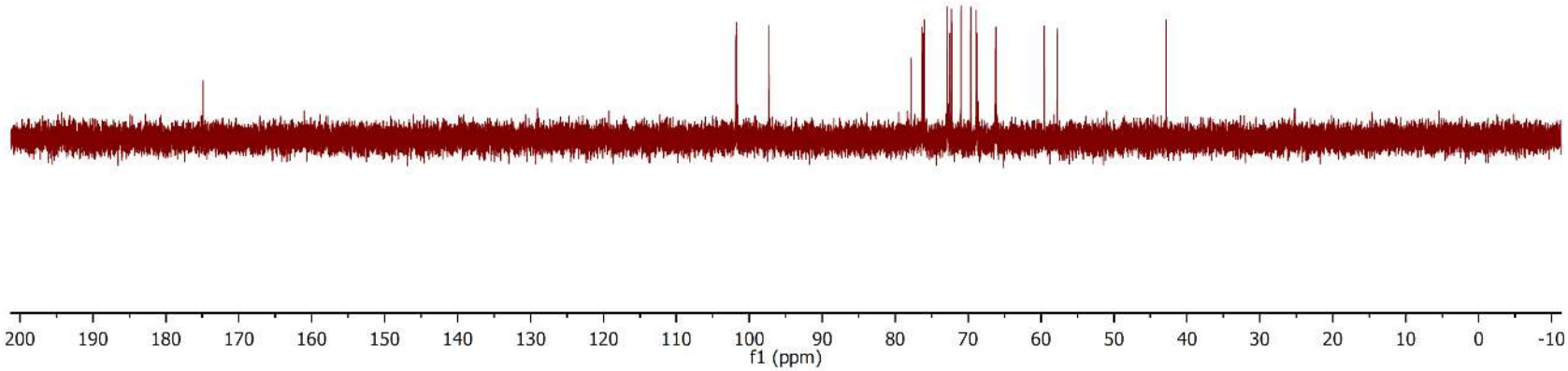

NaO3SO
HO
HO
NaO3SHN
CO2Na
NaO3SO
HO
HO
NaO3SHN
NHSO3Na
1α
1H-13C HSQC, 600/150MHz. D2O
CH2-NHSO3-
C2-H2-NHSO3-
H6-C6 -OSO3-
-OCH2-CH2-NHSO3-
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)

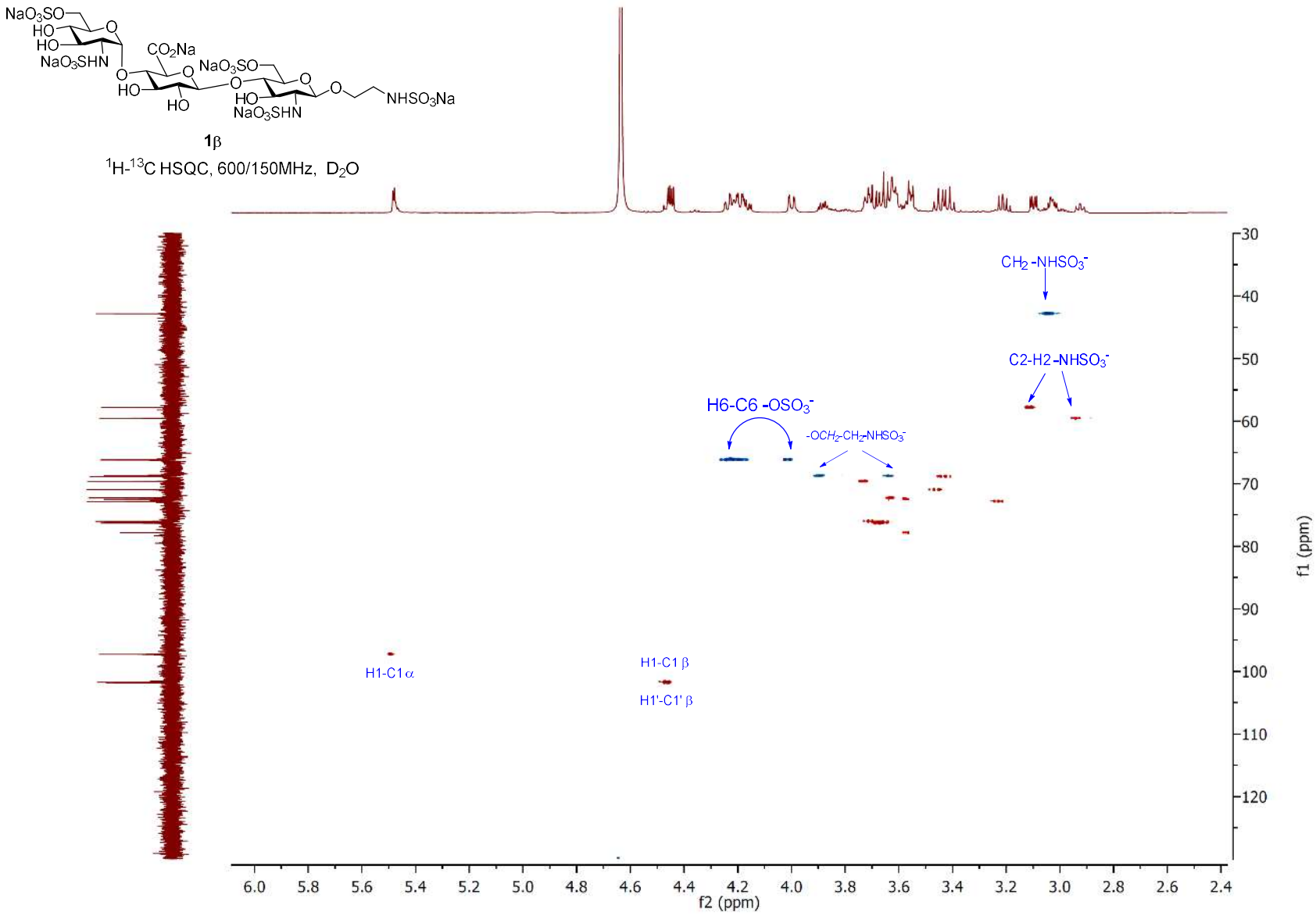

NaO3SO
HO
HO
NaO3SHN
O
CO2Na
O
NaO3SO
O
HO
HO
HO
NaO3SHN
O
O
NHSO3Na
1β
1H NMR, 600 MHz, D2O
5.4845
5.4784
4.4595
4.4530
4.4462
4.4390
4.2298
4.2267
4.2037
4.1996
4.1850
4.1812
4.1762
4.0102
4.0070
3.9917
3.9886
3.7260
3.7136
3.7099
3.6992
3.6837
3.6728
3.6569
3.6415
3.6364
3.6271
3.6256
3.6186
3.6120
3.6063
3.5637
3.5581
3.5526
3.5472
3.4528
3.4364
3.4260
3.4100
3.2272
3.2138
3.2118
3.1983
3.1101
3.1037
3.0935
3.0870
3.0351
3.0254
1.01
1.96
3.21
1.06
1.00
8.16
2.10
1.04
1.12
1.90
0.83
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

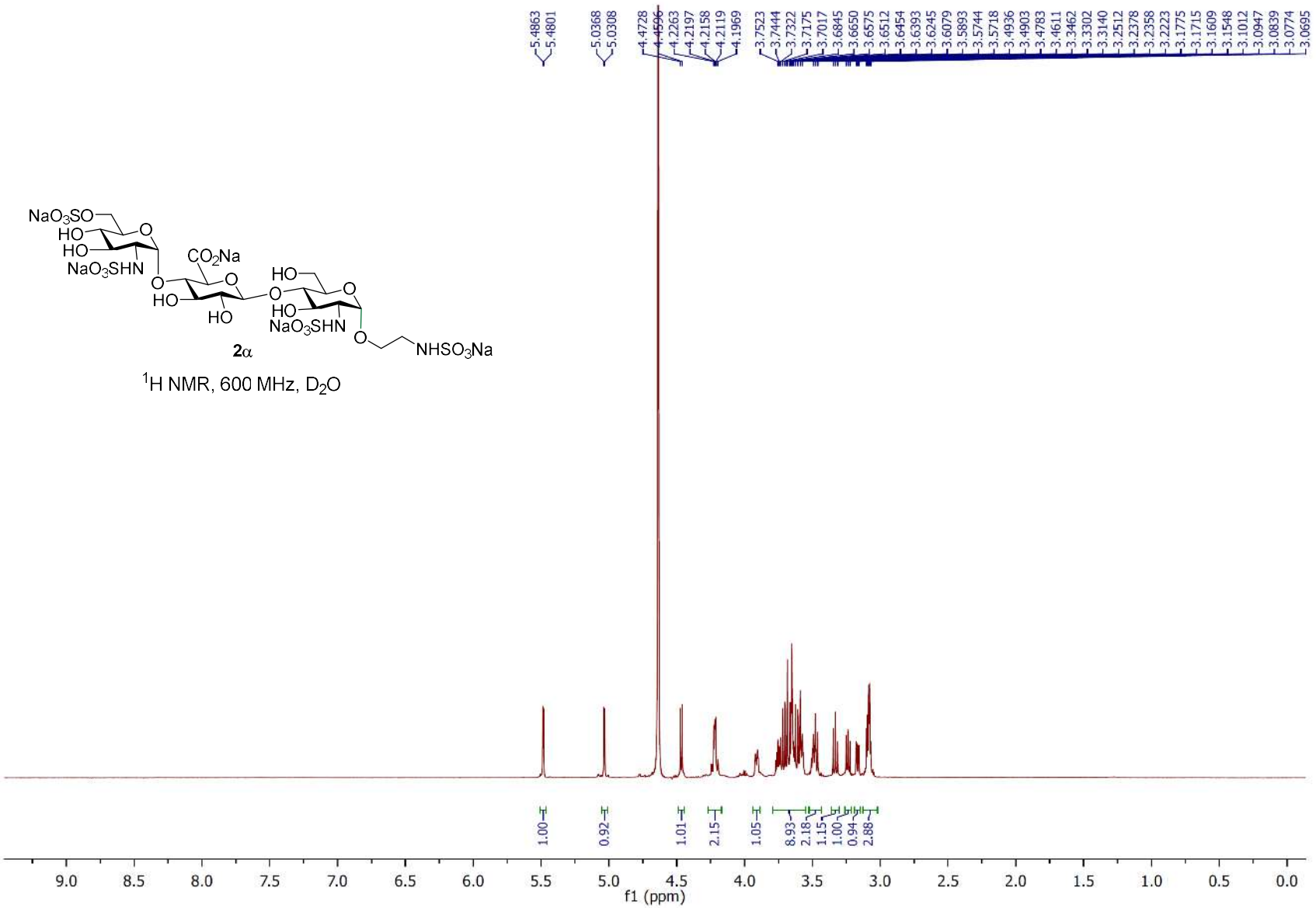


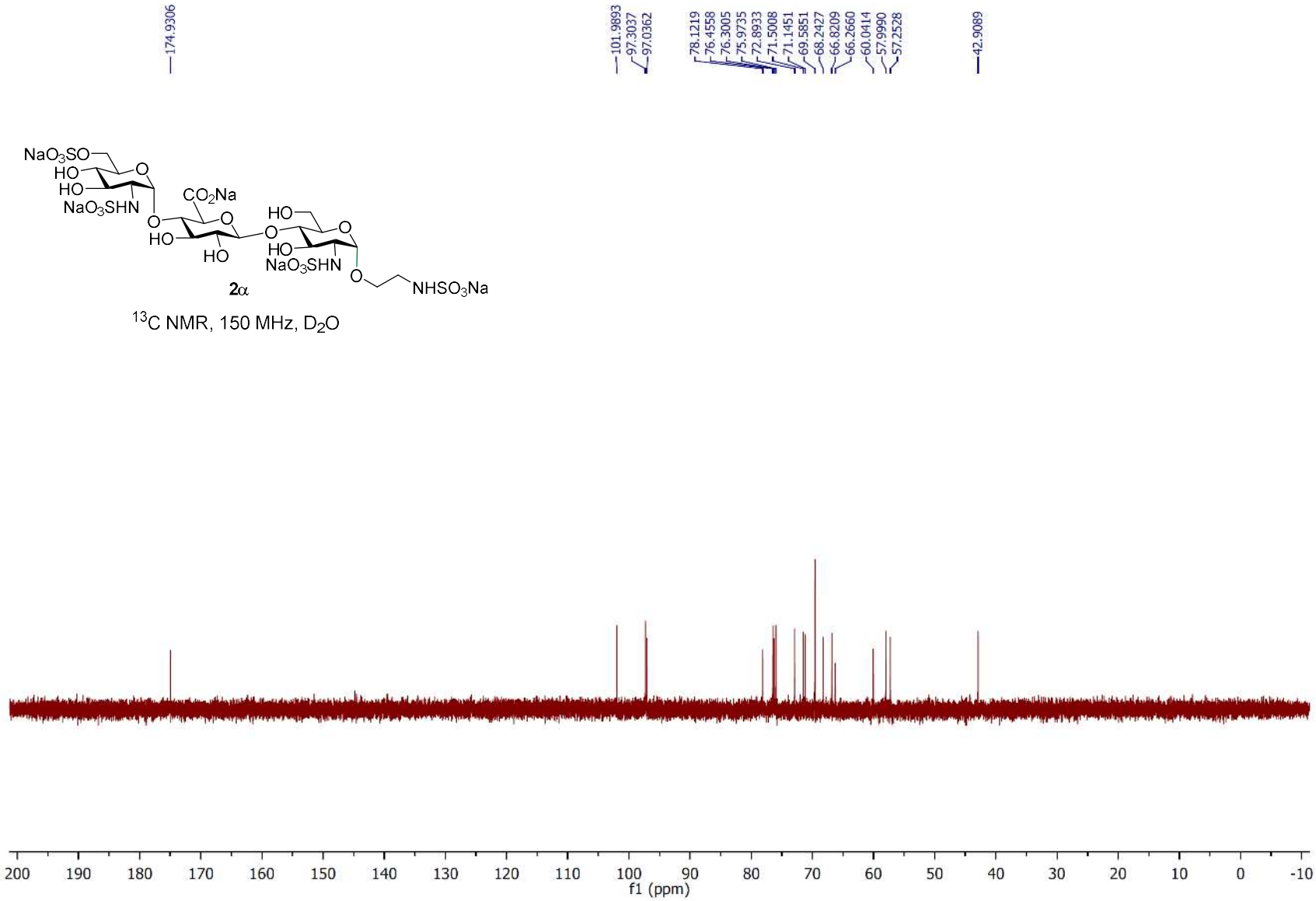
1β

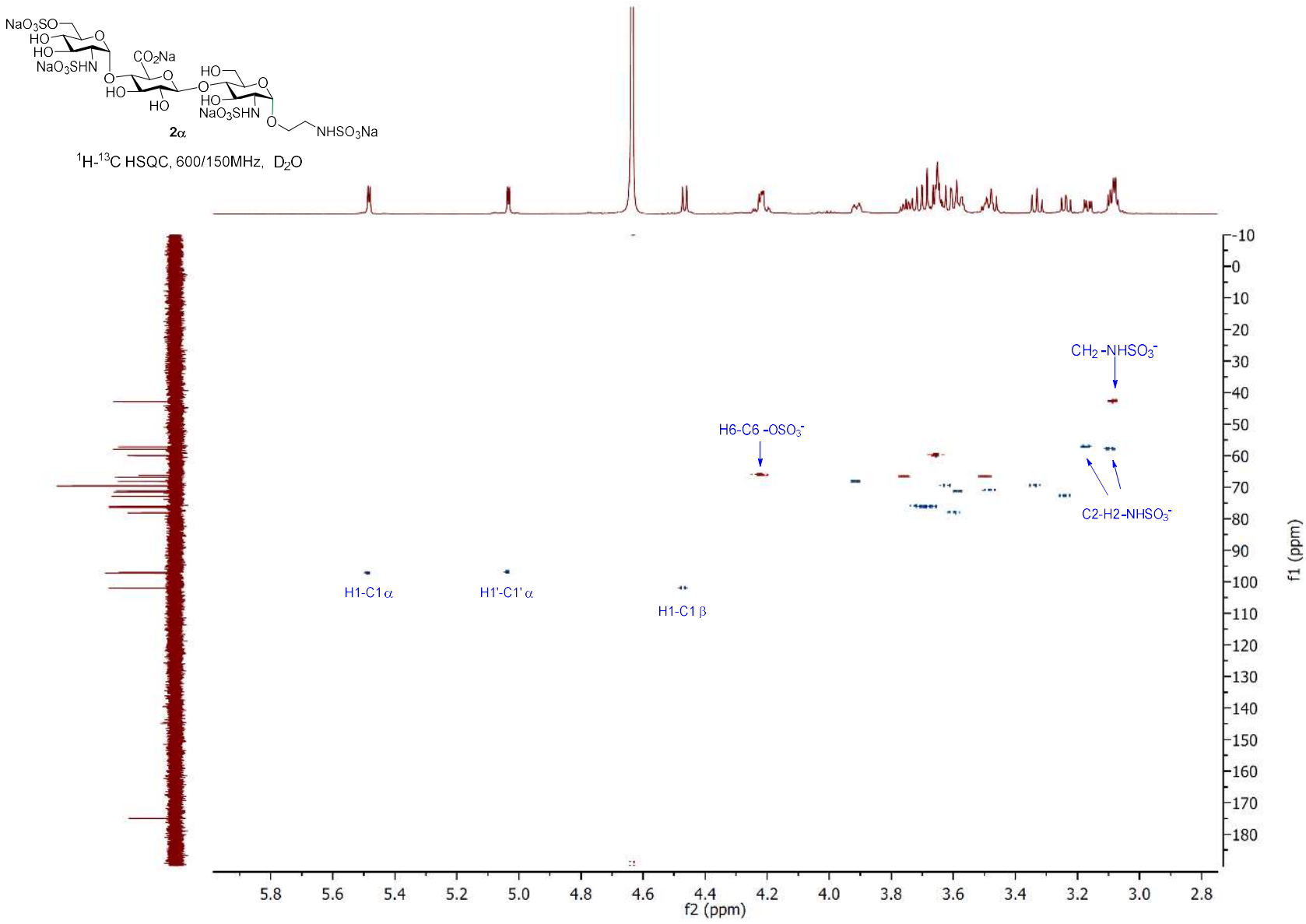
^{13}C NMR, 150 MHz, D_2O

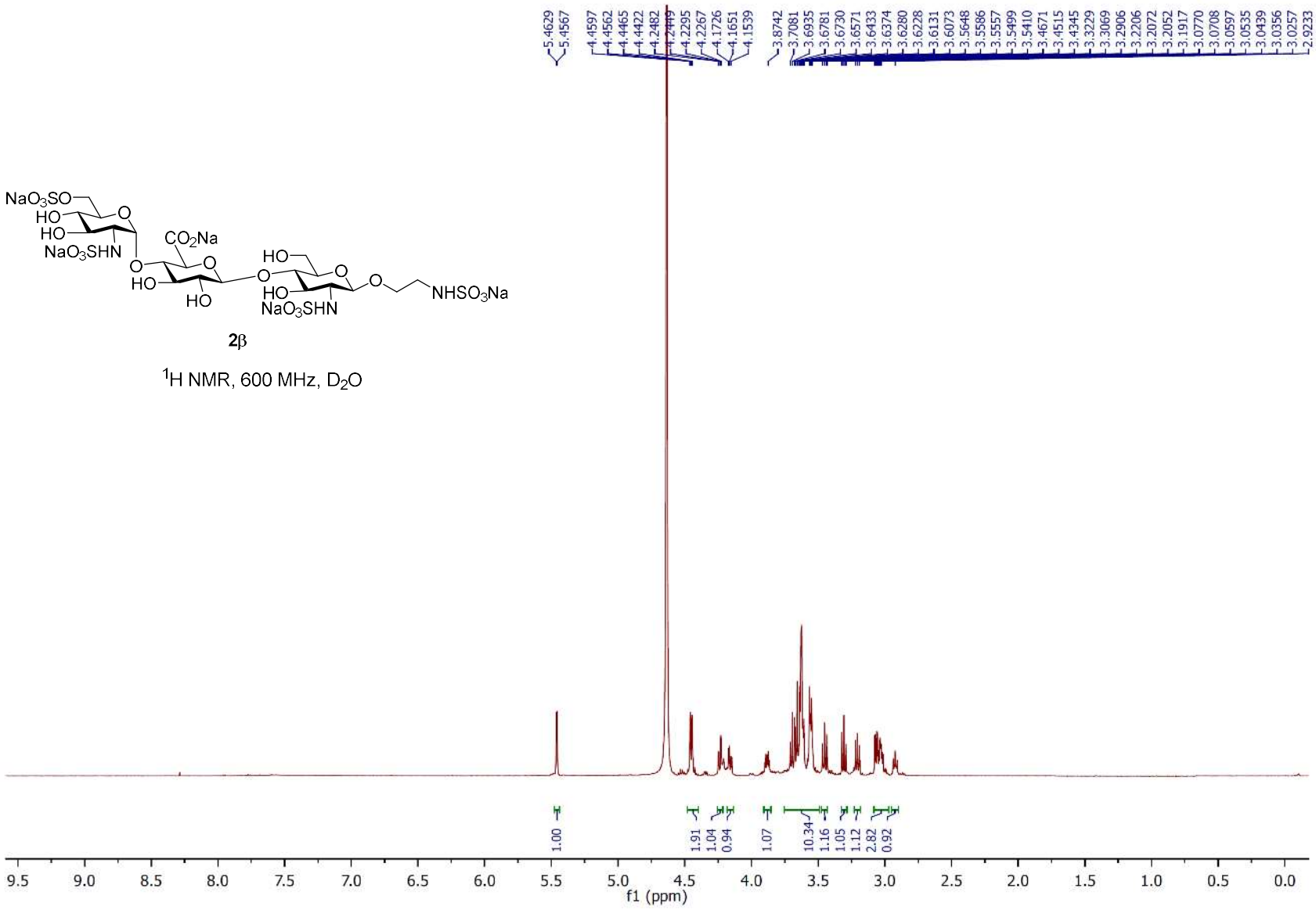


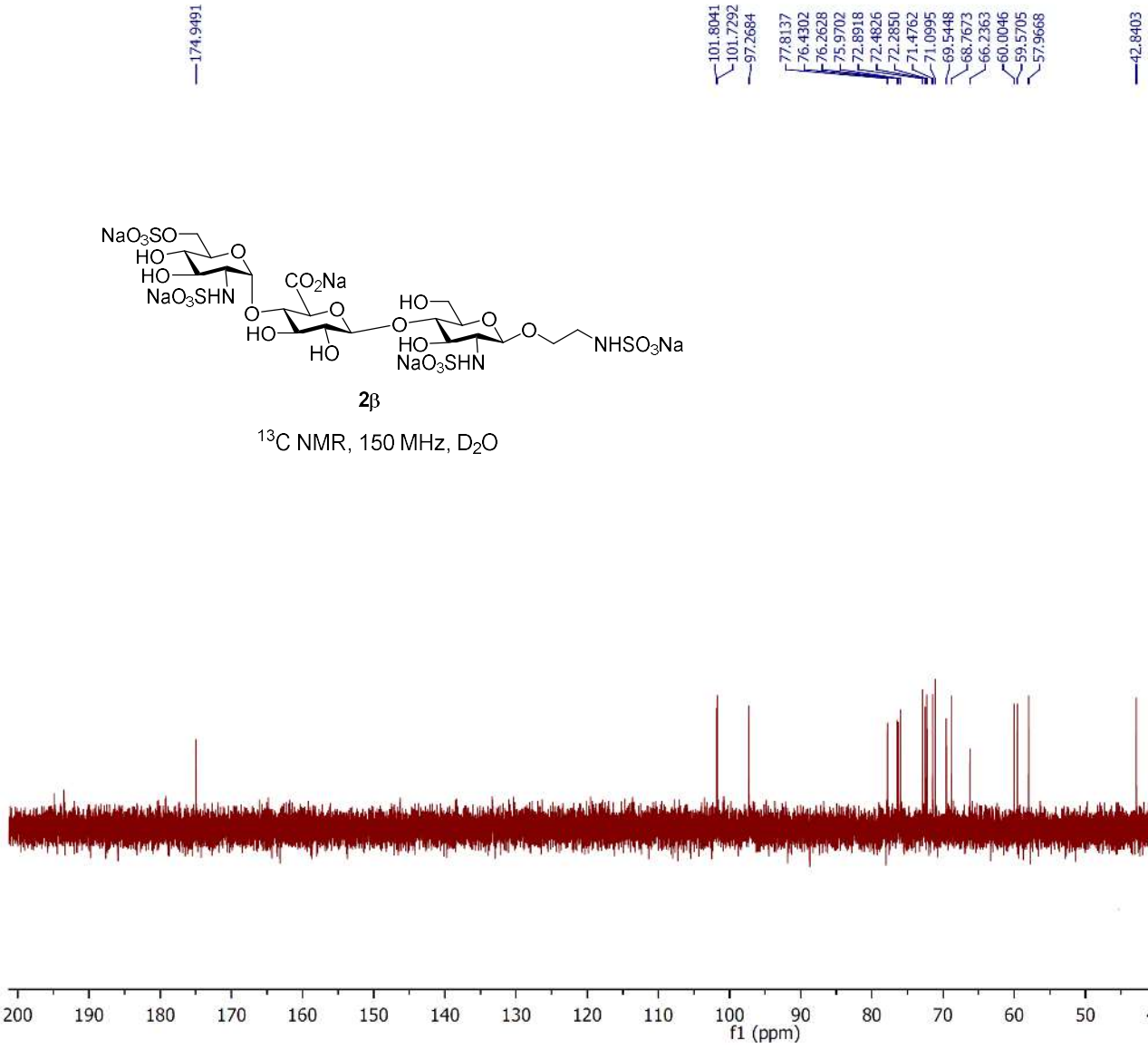

1β
1H-13C HSQC, 600/150MHz, D2O
CH2-NHSO3-
C2-H2-NHSO3-
H6-C6 -OSO3-
-OCH2-CH2-NHSO3-
H1-C1α
H1-C1β
H1'-C1'β
f2 (ppm)
f1 (ppm)

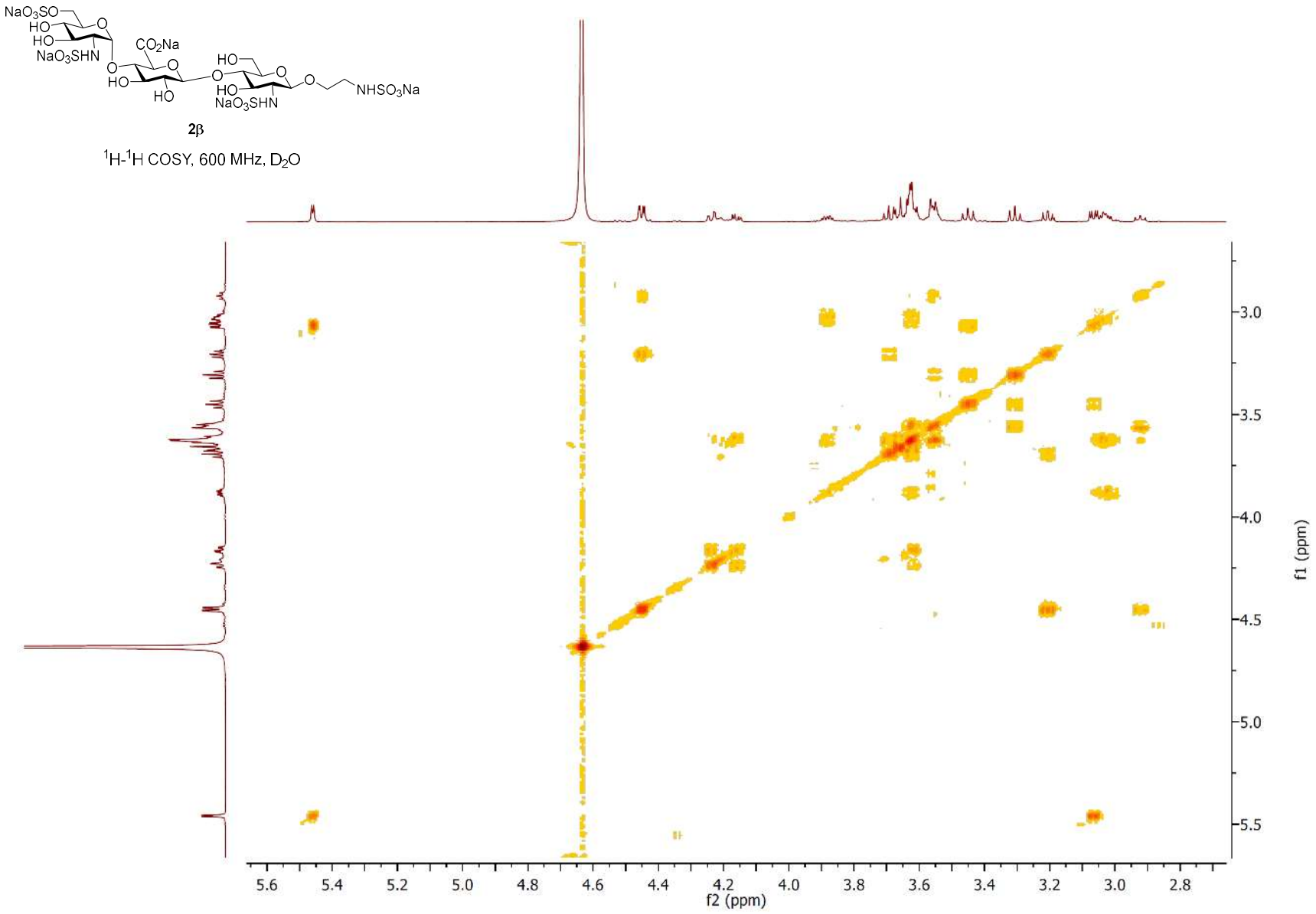

NaO3SO
HO
HO
NaO3SHN
O
CO2Na
O
HO
HO
O
HO
HO
NaO3SHN
O
NHSO3Na
2α
1H NMR, 600 MHz, D2O
5.4863
5.4801
5.0368
5.0308
4.4728
4.4696
4.2263
4.2197
4.2158
4.2119
4.1969
3.7523
3.7444
3.7322
3.7175
3.7017
3.6845
3.6650
3.6575
3.6512
3.6454
3.6393
3.6245
3.6079
3.5893
3.5744
3.5718
3.4936
3.4903
3.4783
3.4611
3.3462
3.3302
3.3140
3.2512
3.2378
3.2358
3.2223
3.1775
3.1715
3.1609
3.1548
3.1012
3.0947
3.0839
3.0774
3.0695
1.00
0.92
1.01
2.15
1.05
8.93
2.18
1.15
1.00
0.94
2.88
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

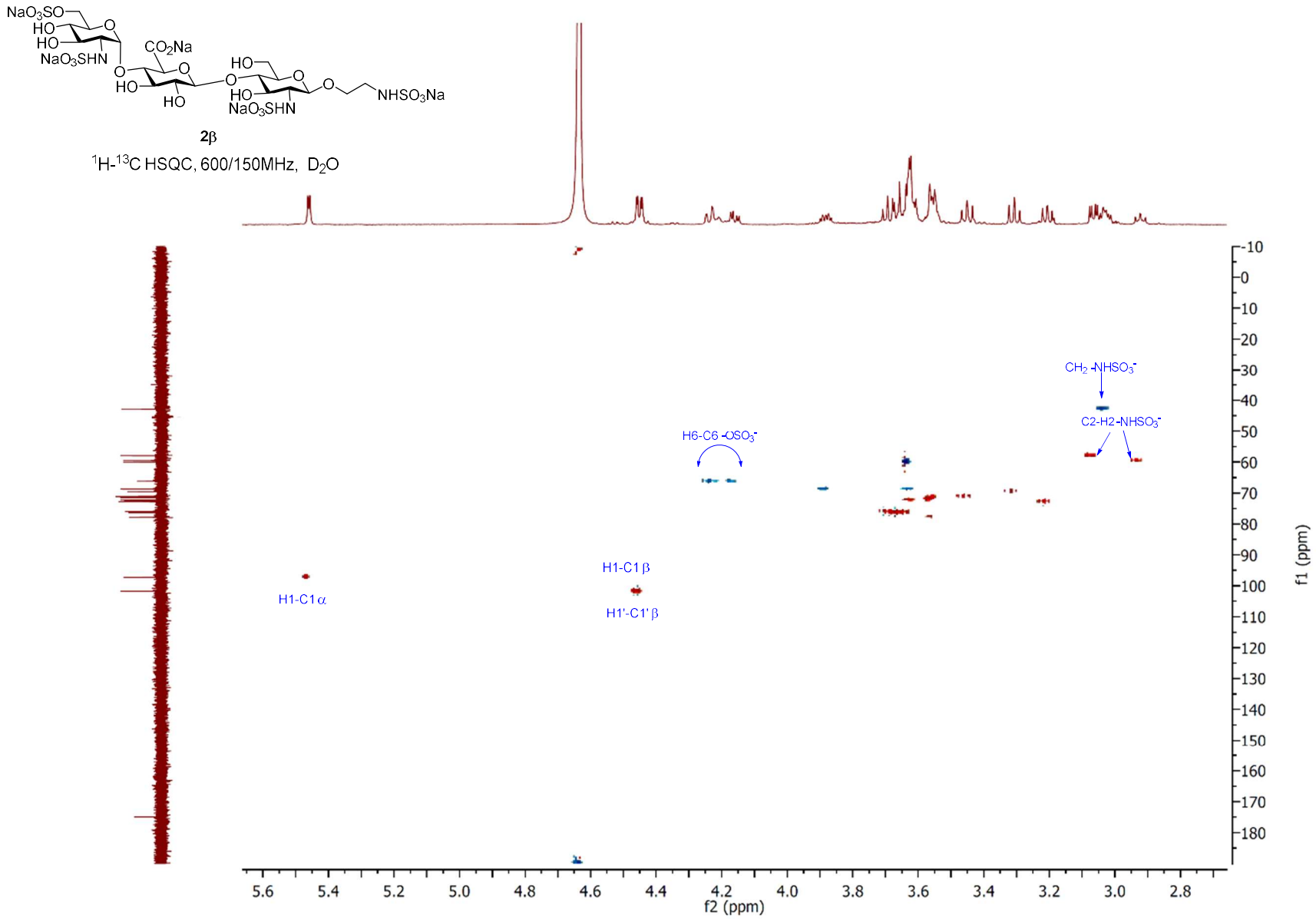

174.9306
101.9893
97.3037
97.0362
78.1219
76.4558
76.3005
75.9735
72.8933
71.5008
71.1451
69.5851
68.2427
66.8209
66.2660
60.0414
57.9990
57.2528
42.9089
NaO3SO
HO
HO
NaO3SHN
CO2Na
HO
HO
HO
HO
NaO3SHN
NHSO3Na
2α
13C NMR, 150 MHz, D2O
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

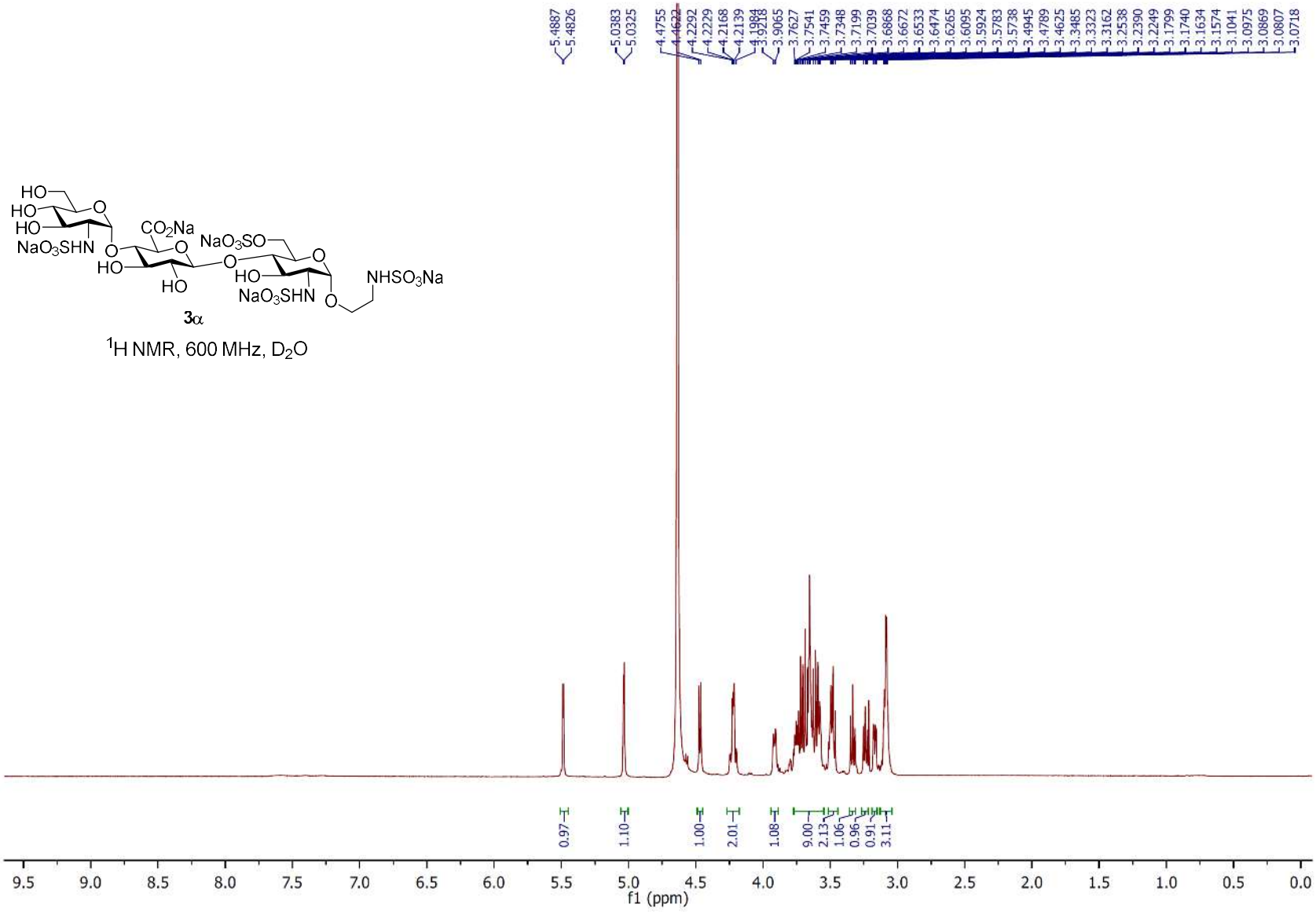

2α
^{1}H-^{13}C HSQC, 600/150MHz, D$_2$O
H1-C1α
H1'-C1'α
H1-C1β
H6-C6 -OSO$_3^-$
CH$_2$ -NHSO$_3^-$
C2-H2-NHSO$_3^-$
f2 (ppm)
f1 (ppm)

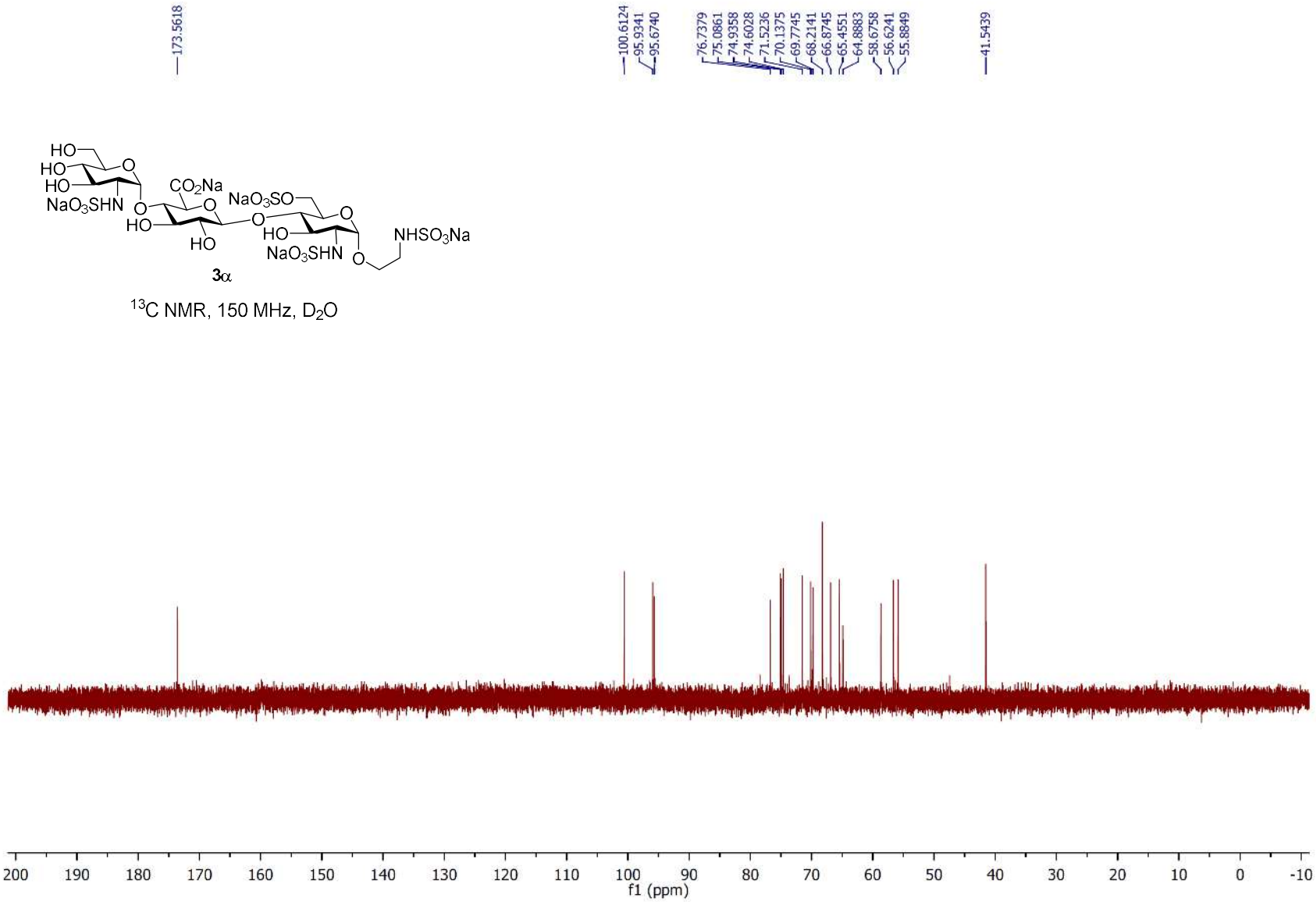

NaO3SO
HO
HO
NaO3SHN
O
CO2Na
HO
HO
HO
HO
NaO3SHN
NHSO3Na
2β
1H NMR, 600 MHz, D2O
5.4629
5.4567
4.4597
4.4562
4.4465
4.4422
4.2482
4.2449
4.2295
4.2267
4.1726
4.1651
4.1539
3.8742
3.7081
3.6935
3.6781
3.6730
3.6571
3.6433
3.6374
3.6280
3.6228
3.6131
3.6073
3.5648
3.5586
3.5557
3.5499
3.5410
3.4671
3.4515
3.4345
3.3229
3.3069
3.2906
3.2206
3.2072
3.2052
3.1917
3.0770
3.0708
3.0597
3.0535
3.0439
3.0356
3.0257
2.9233
1.00
1.91
1.04
0.94
1.07
10.34
1.16
1.05
1.12
2.82
0.92
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

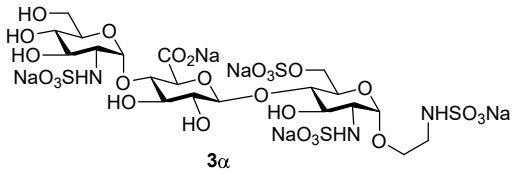

174.9491
101.8041
101.7292
97.2684
77.8137
76.4302
76.2628
75.9702
72.8918
72.4826
72.2850
71.4762
71.0995
69.5448
68.7673
66.2363
60.0046
59.5705
57.9668
42.8403
NaO3SO
HO
HO
NaO3SHN
CO2Na
HO
HO
HO
NaO3SHN
NHSO3Na
2β
13C NMR, 150 MHz, D2O
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


NaO3SO
HO
HO
NaO3SHN
CO2Na
HO
HO
HO
HO
NaO3SHN
NHSO3Na
2β
1H-1H COSY, 600 MHz, D2O
5.6
5.4
5.2
5.0
4.8
4.6
4.4
4.2
4.0
3.8
3.6
3.4
3.2
3.0
2.8
f2 (ppm)
3.0
3.5
4.0
4.5
5.0
5.5
f1 (ppm)


2β
1H-13C HSQC, 600/150MHz, D2O
H6-C6 -OSO3-
CH2 -NHSO3-
C2-H2-NHSO3-
H1-C1α
H1-C1β
H1'-C1'β
f2 (ppm)
f1 (ppm)

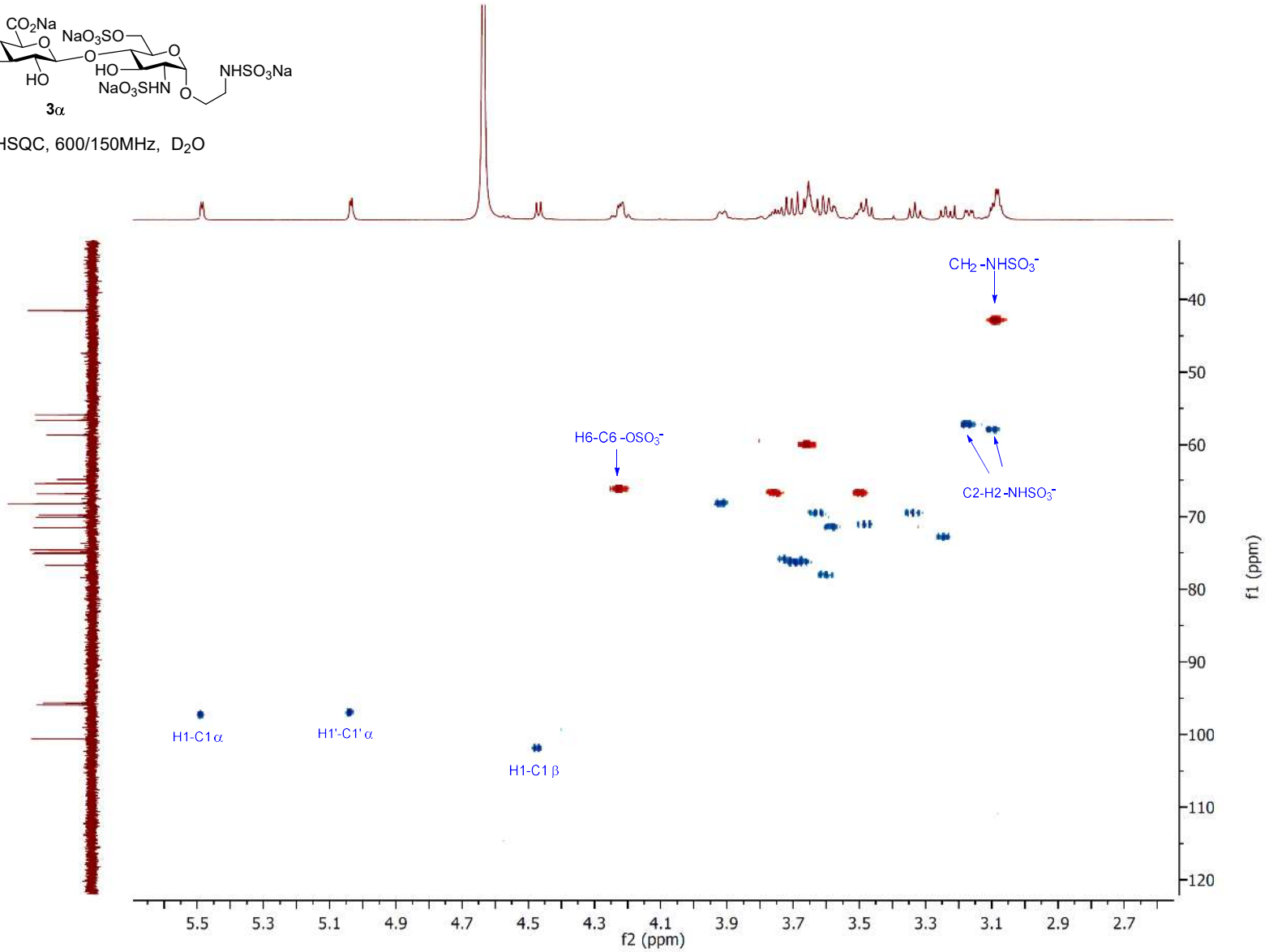

HO
HO
HO
NaO3SHN
O
CO2Na
O
HO
HO
NaO3SO
O
HO
NaO3SHN
O
NHSO3Na
3α
1H NMR, 600 MHz, D2O
5.4887
5.4826
5.0383
5.0325
4.4755
4.4622
4.2292
4.2229
4.2168
4.2139
4.1984
3.9218
3.9065
3.7627
3.7541
3.7459
3.7348
3.7199
3.7039
3.6868
3.6672
3.6533
3.6474
3.6265
3.6095
3.5924
3.5783
3.5738
3.4945
3.4789
3.4625
3.3485
3.3323
3.3162
3.2538
3.2390
3.2249
3.1799
3.1740
3.1634
3.1574
3.1041
3.0975
3.0869
3.0807
3.0718
0.97
1.10
1.00
2.01
1.08
9.00
2.13
1.06
0.96
0.91
3.11
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

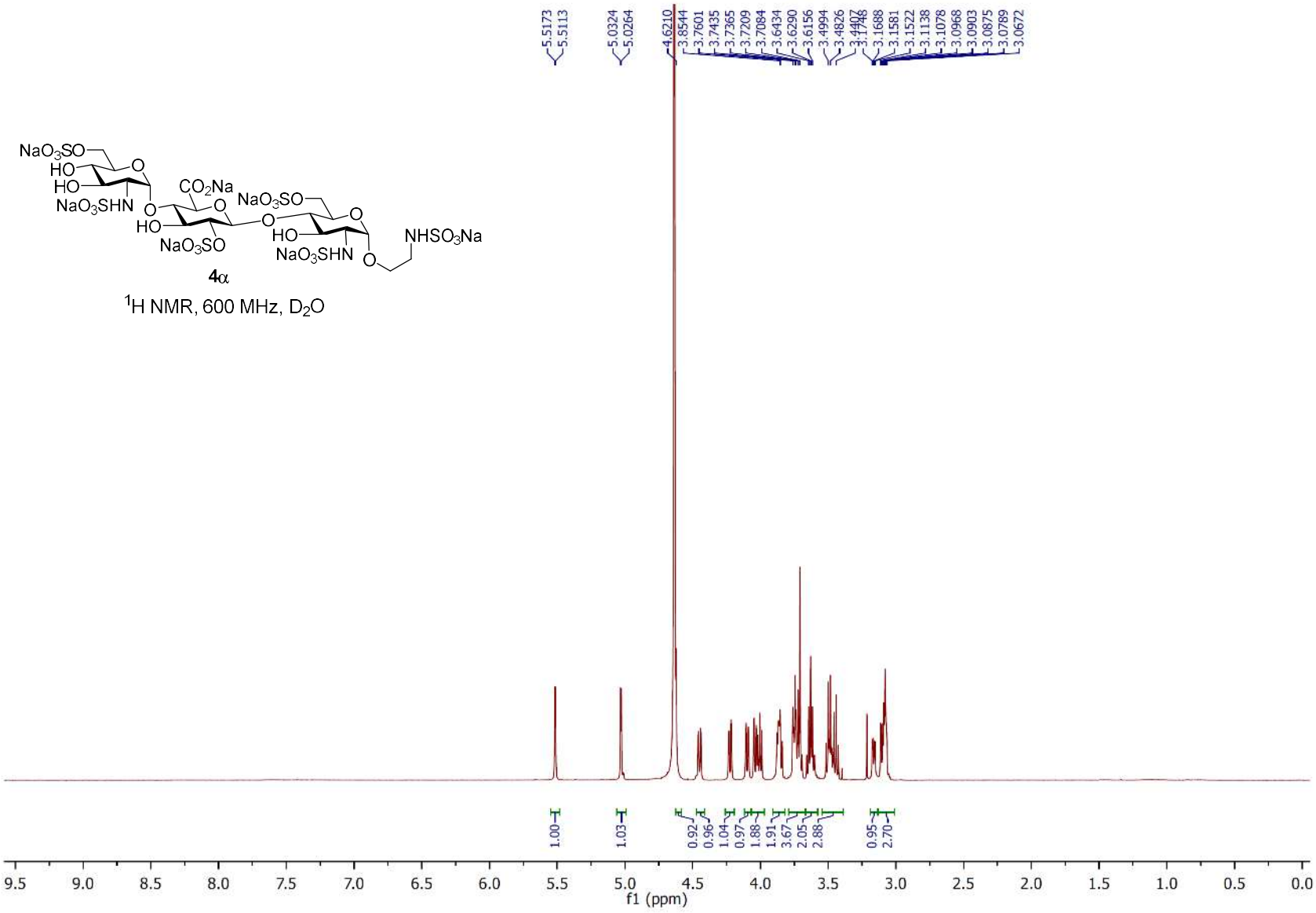

173.5618
100.6124
95.9341
95.6740
76.7379
75.0861
74.9358
74.6028
71.5236
70.1375
69.7745
68.2141
66.8745
65.4551
64.8883
58.6758
56.6241
55.8849
41.5439
HO
HO
HO
NaO3SHN
O
CO2Na
O
HO
HO
NaO3SO
O
O
HO
NaO3SHN
O
NHSO3Na
3α
13C NMR, 150 MHz, D2O
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

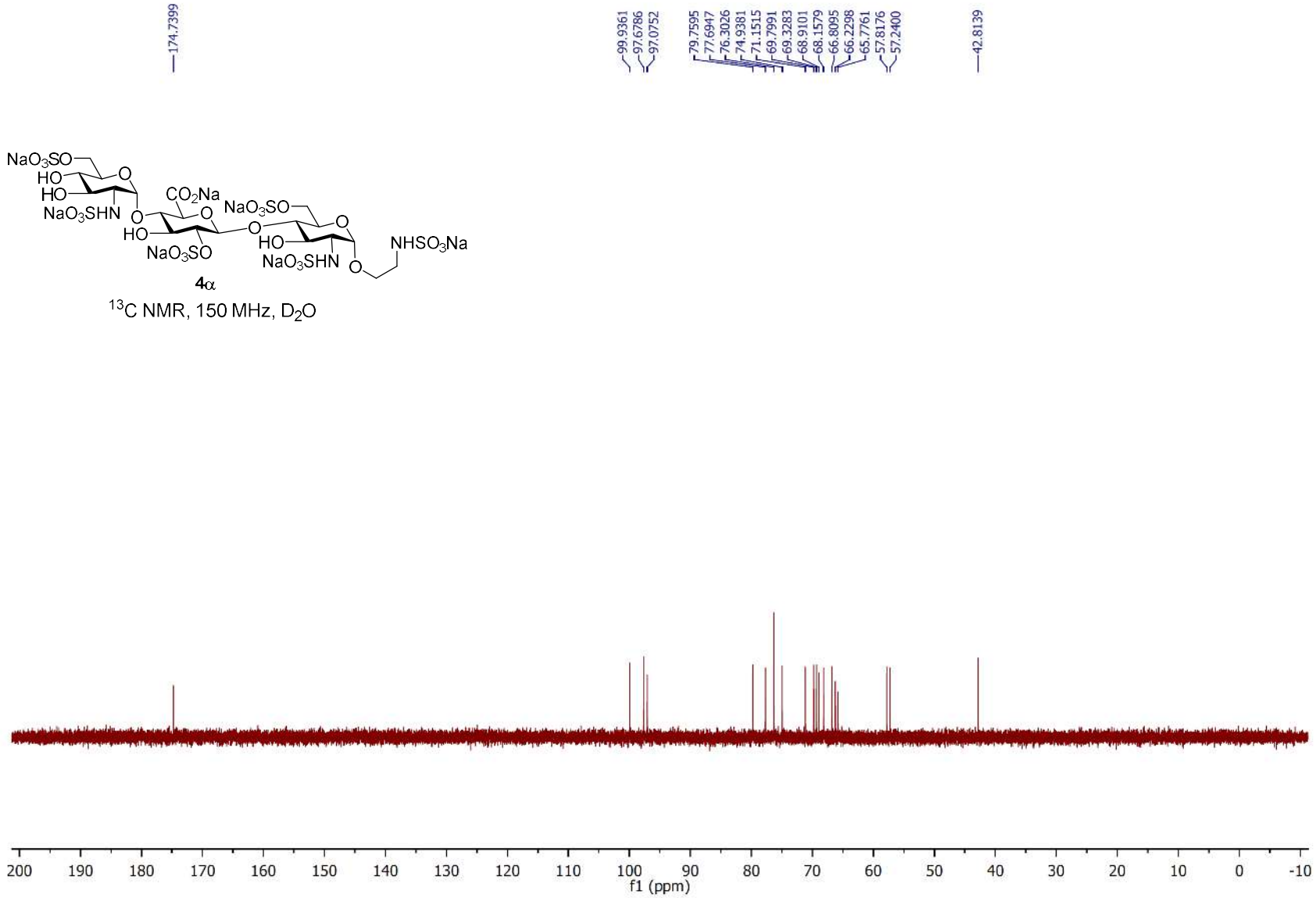


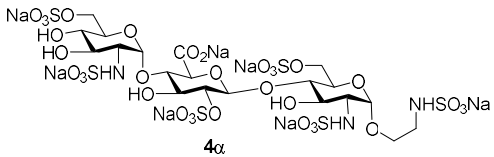
3α

^{1}H-^{13}C HSQC, 600/150MHz, D_2O



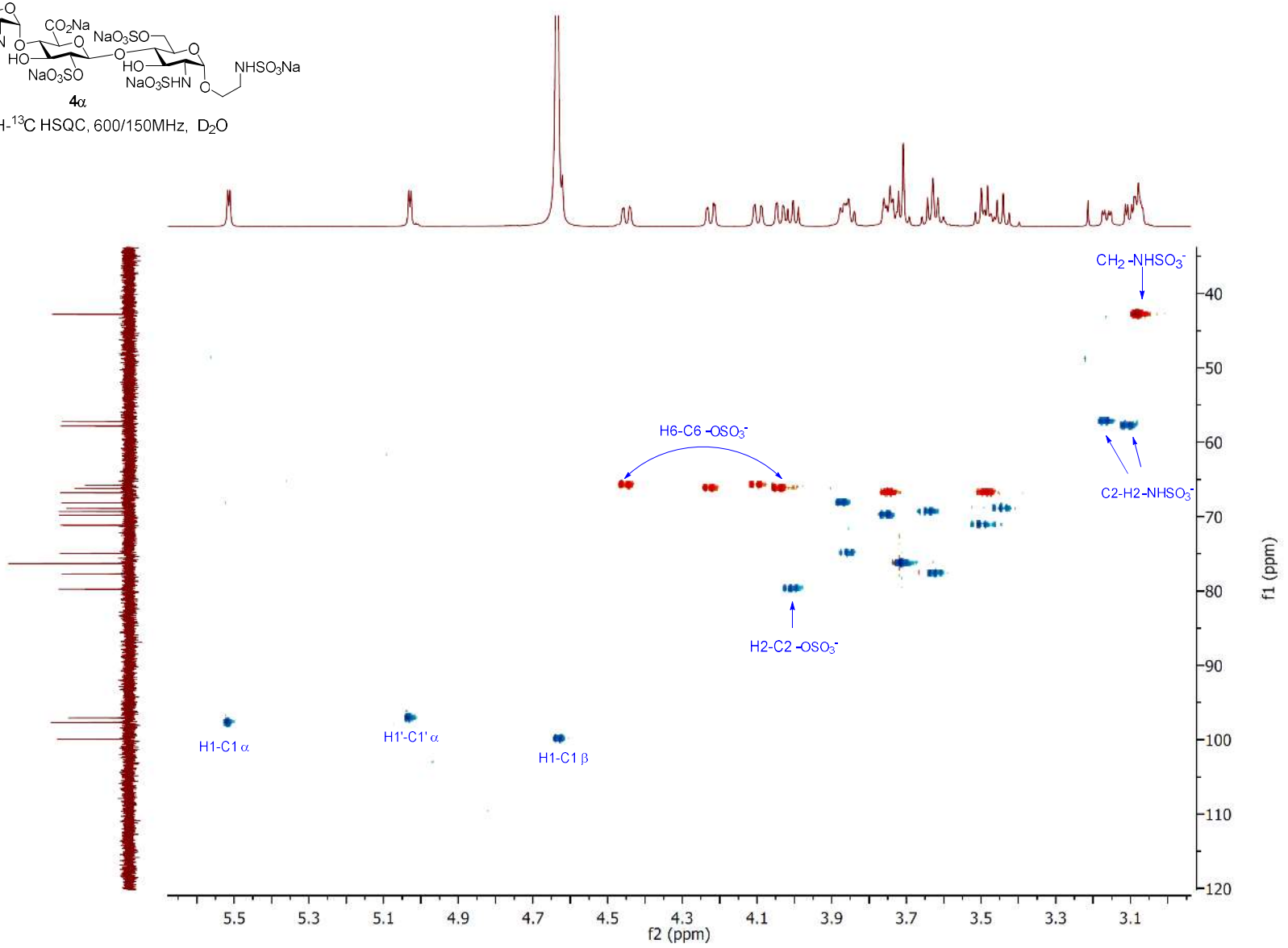

NaO3SO
HO
HO
NaO3SHN
O
CO2Na
O
HO
NaO3SO
O
NaO3SO
O
HO
NaO3SHN
O
NHSO3Na
4α
1H NMR, 600 MHz, D2O
5.5173
5.5113
5.0324
5.0264
4.6210
3.8544
3.7601
3.7435
3.7365
3.7209
3.7084
3.6434
3.6290
3.6156
3.4994
3.4826
3.1688
3.1581
3.1522
3.1138
3.1078
3.0968
3.0903
3.0875
3.0789
3.0672
1.00
1.03
0.92
0.96
1.04
0.97
1.88
1.91
3.67
2.05
2.88
0.95
2.70
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)


174.7399
99.9361
97.6786
97.0752
79.7595
77.6947
76.3026
74.9381
71.1515
69.7991
69.3283
68.9101
68.1579
66.8095
66.2298
65.7761
57.8176
57.2400
42.8139
NaO3SO
HO
HO
NaO3SHN
CO2Na
NaO3SO
HO
NaO3SO
HO
NaO3SHN
NHSO3Na
4α
13C NMR, 150 MHz, D2O
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)



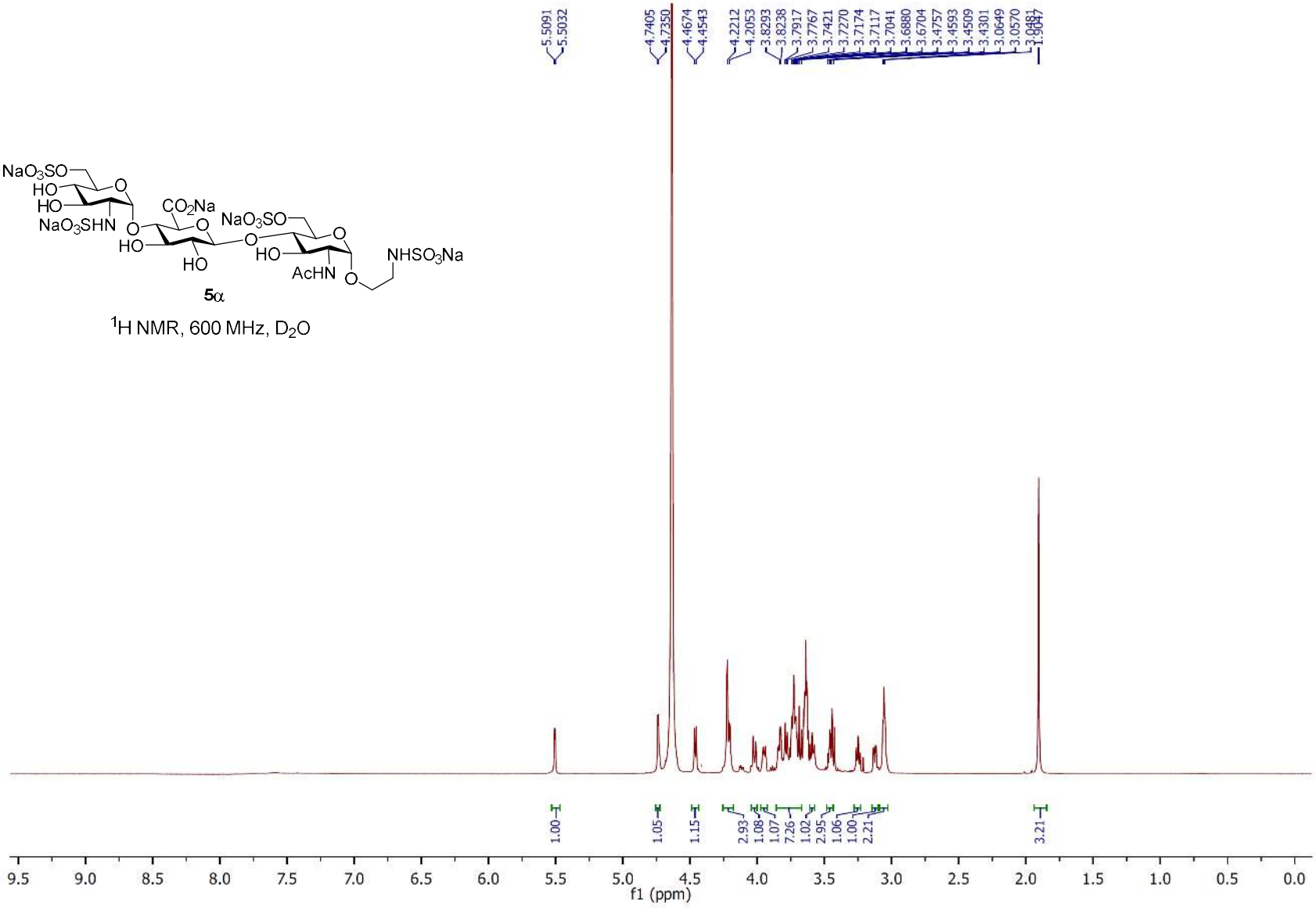
4α

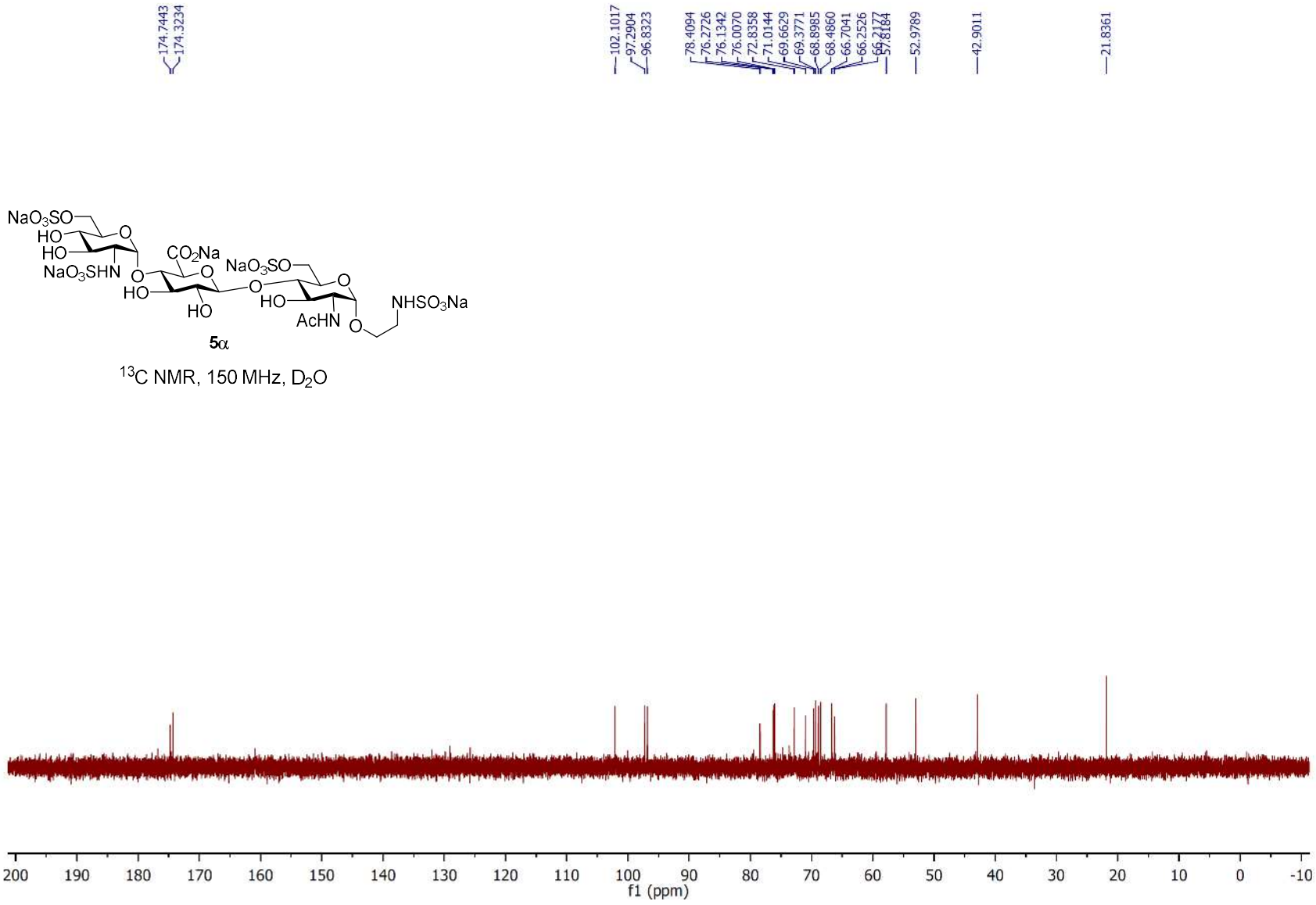
^{1}H-^{13}C HSQC, 600/150MHz, D_2O



5α

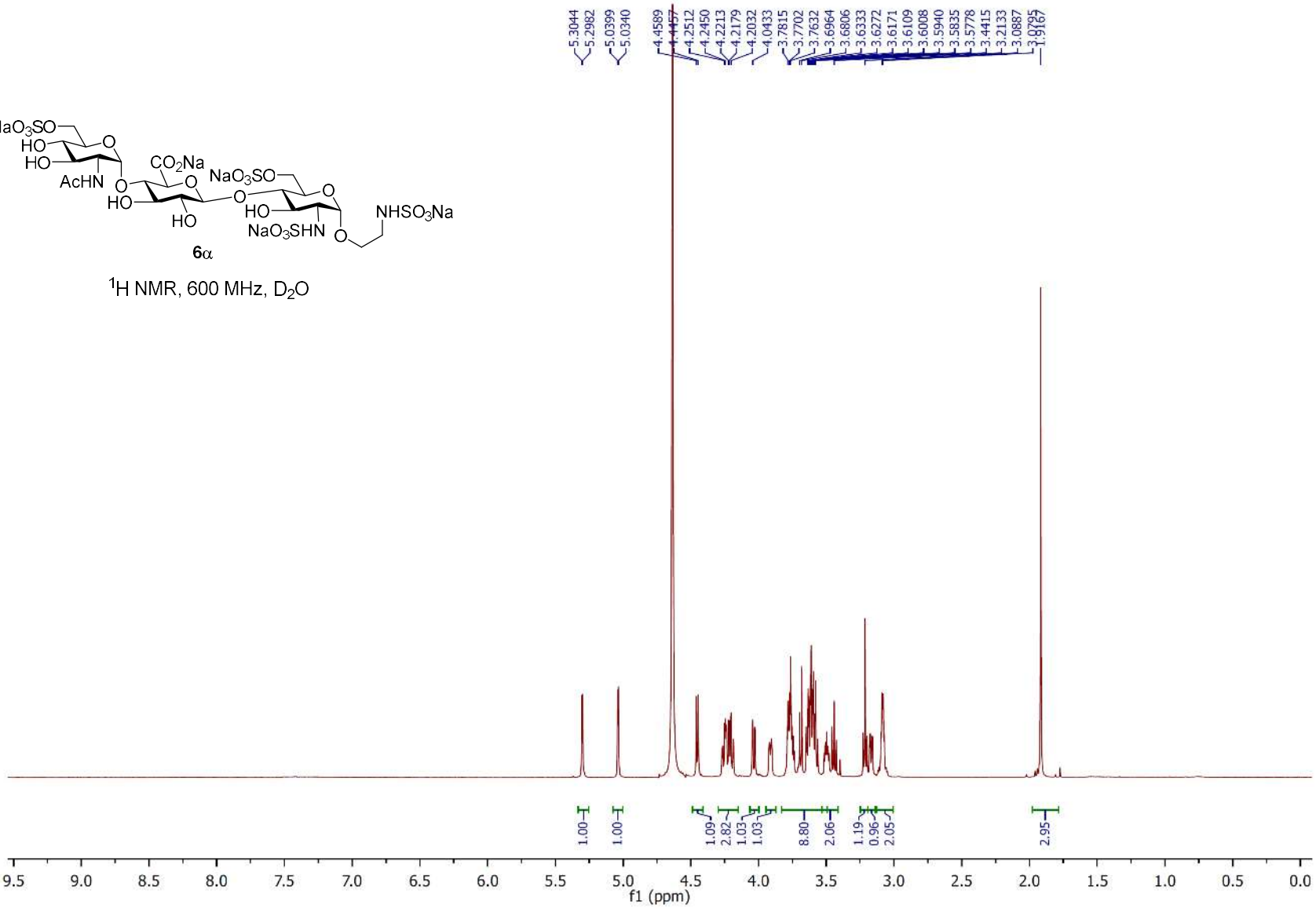
^{1}H NMR, 600 MHz, D_2O

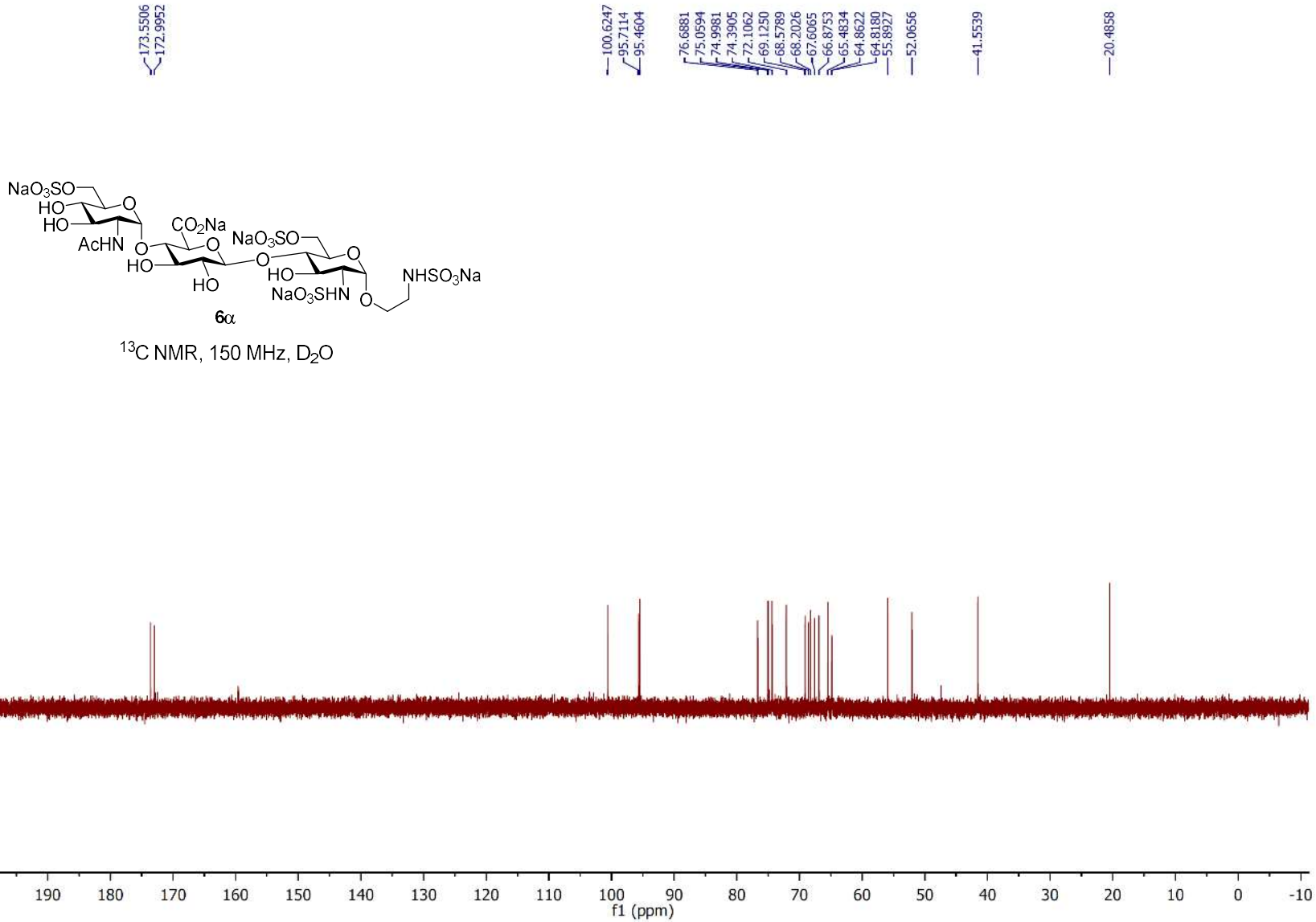


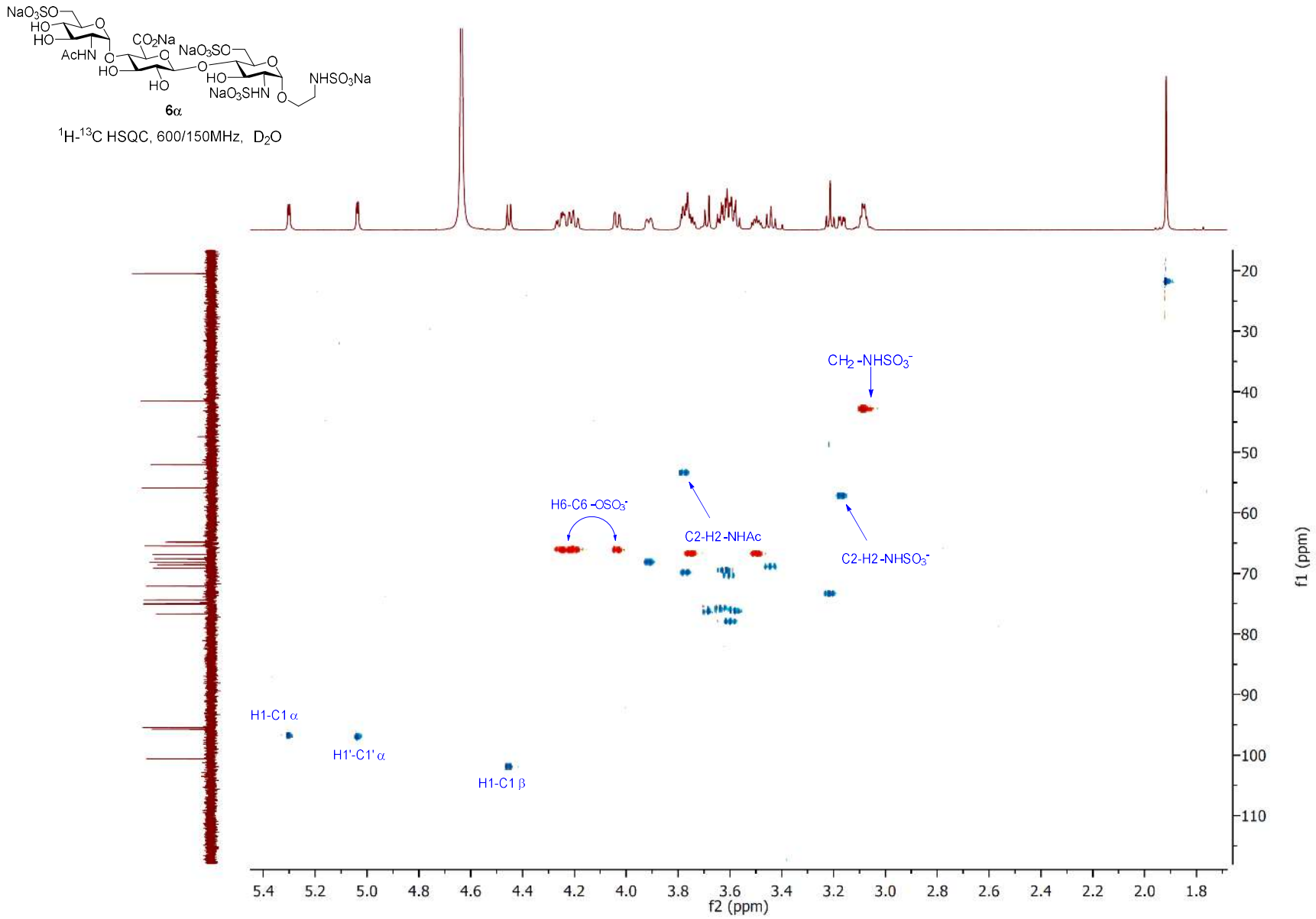

174.7443
174.3234
102.1017
97.2904
96.8323
78.4094
76.2726
76.1342
76.0070
72.8358
71.0144
69.6629
69.3771
68.8985
68.4860
66.7041
66.2526
66.2177
57.8184
52.9789
42.9011
21.8361
NaO3SO
HO
HO
NaO3SHN
O
CO2Na
O
HO
HO
O
NaO3SO
O
HO
AcHN
O
NHSO3Na
5α
13C NMR, 150 MHz, D2O
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

6α

^{1}H NMR, 600 MHz, D_2O

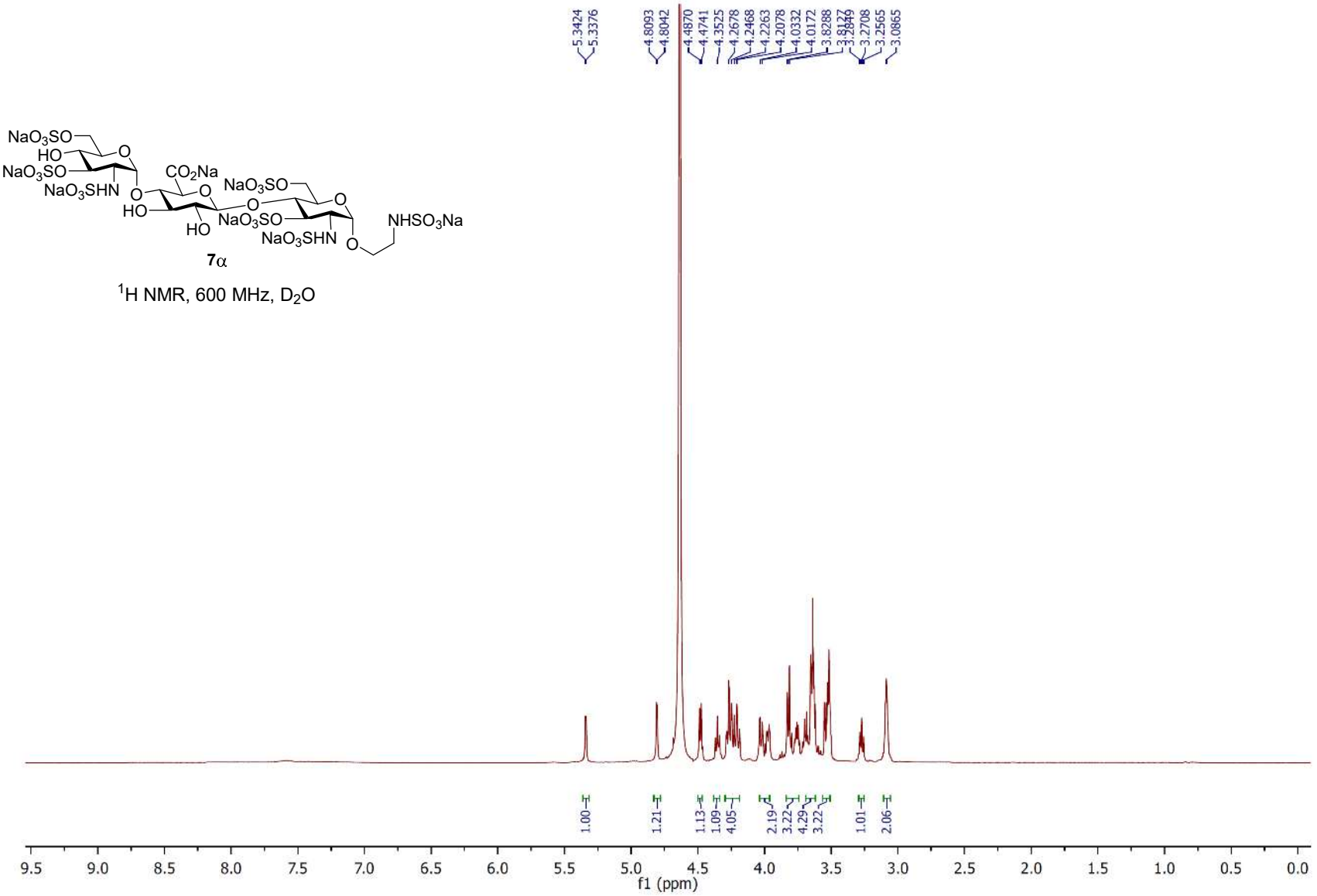



173.5506
172.9952
100.6247
95.7114
95.4604
76.6881
75.0594
74.9981
74.3905
72.1062
69.1250
68.5789
68.2026
67.6065
66.8753
65.4834
64.8622
64.8180
55.8927
52.0656
41.5539
20.4858
NaO3SO
HO
HO
AcHN
O
CO2Na
O
HO
HO
O
NaO3SO
O
HO
NaO3SHN
O
NHSO3Na
6α
13C NMR, 150 MHz, D2O
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


6α
1H-13C HSQC, 600/150MHz, D2O
CH2-NHSO3-
H6-C6-OSO3-
C2-H2-NHAc
C2-H2-NHSO3-
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)

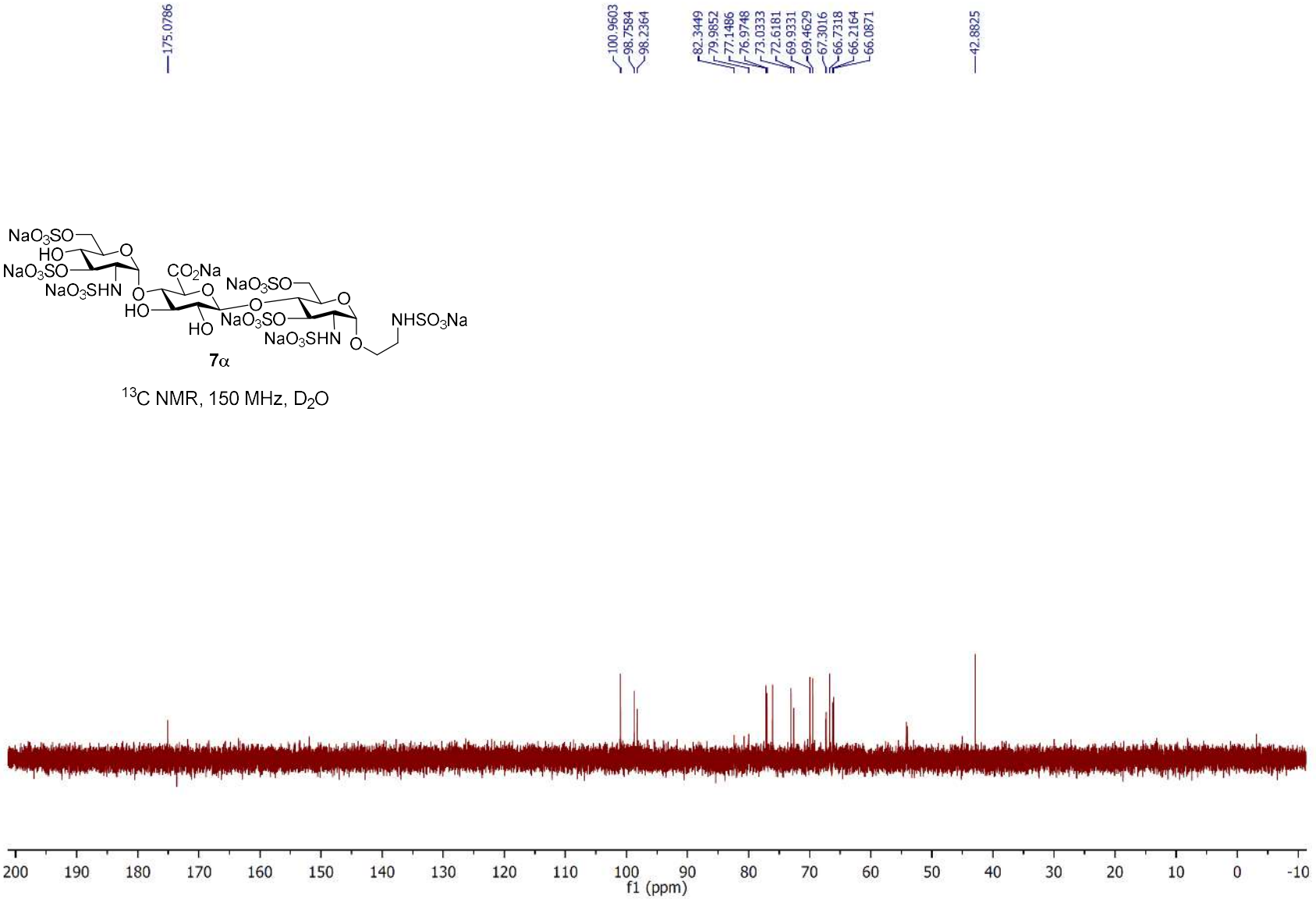
7α

^{1}H NMR, 600 MHz, D_2O



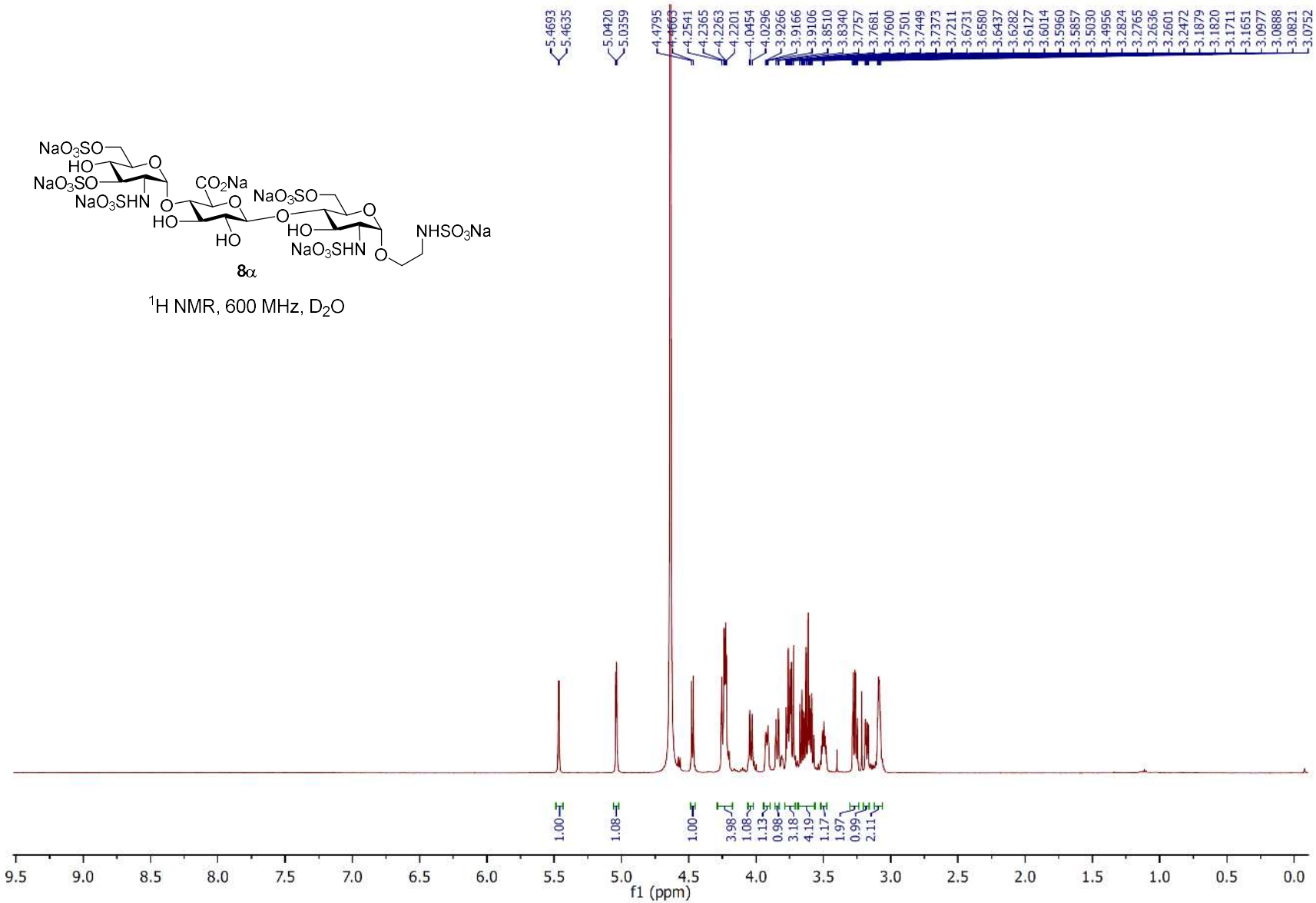
7α

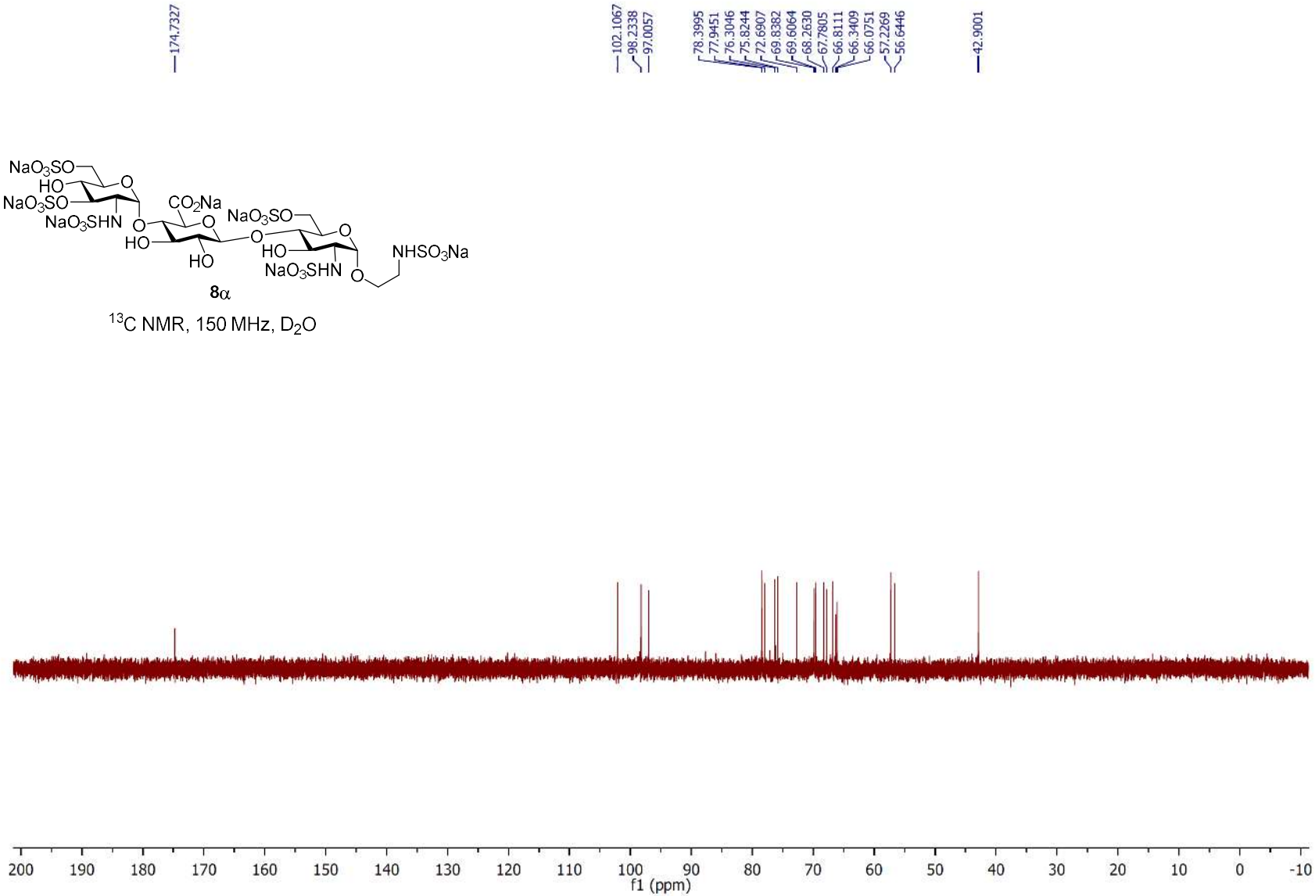
^{13}C NMR, 150 MHz, D_2O



8α

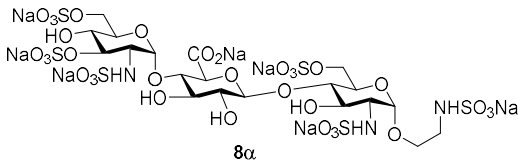
^{1}H NMR, 600 MHz, D_2O





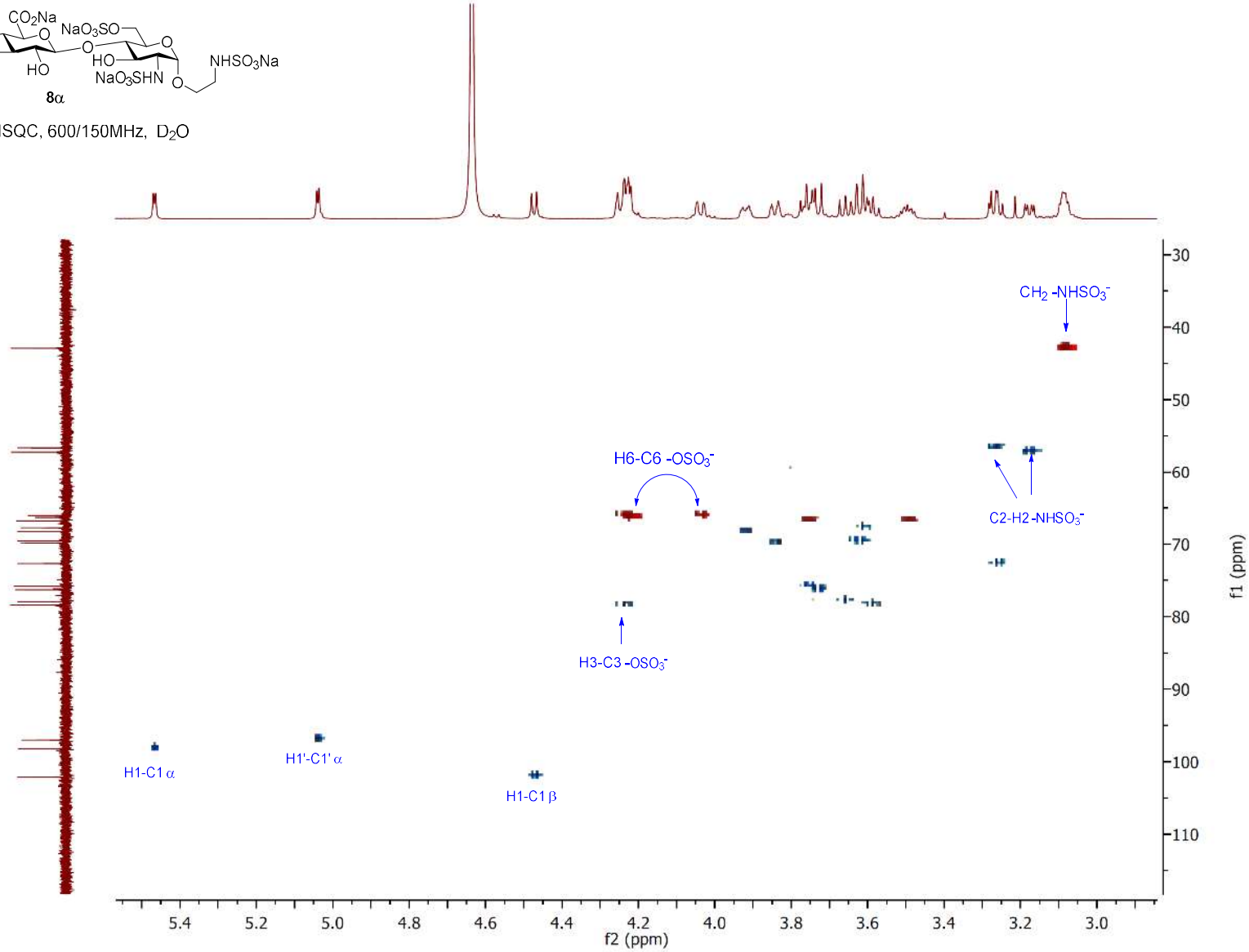
8α

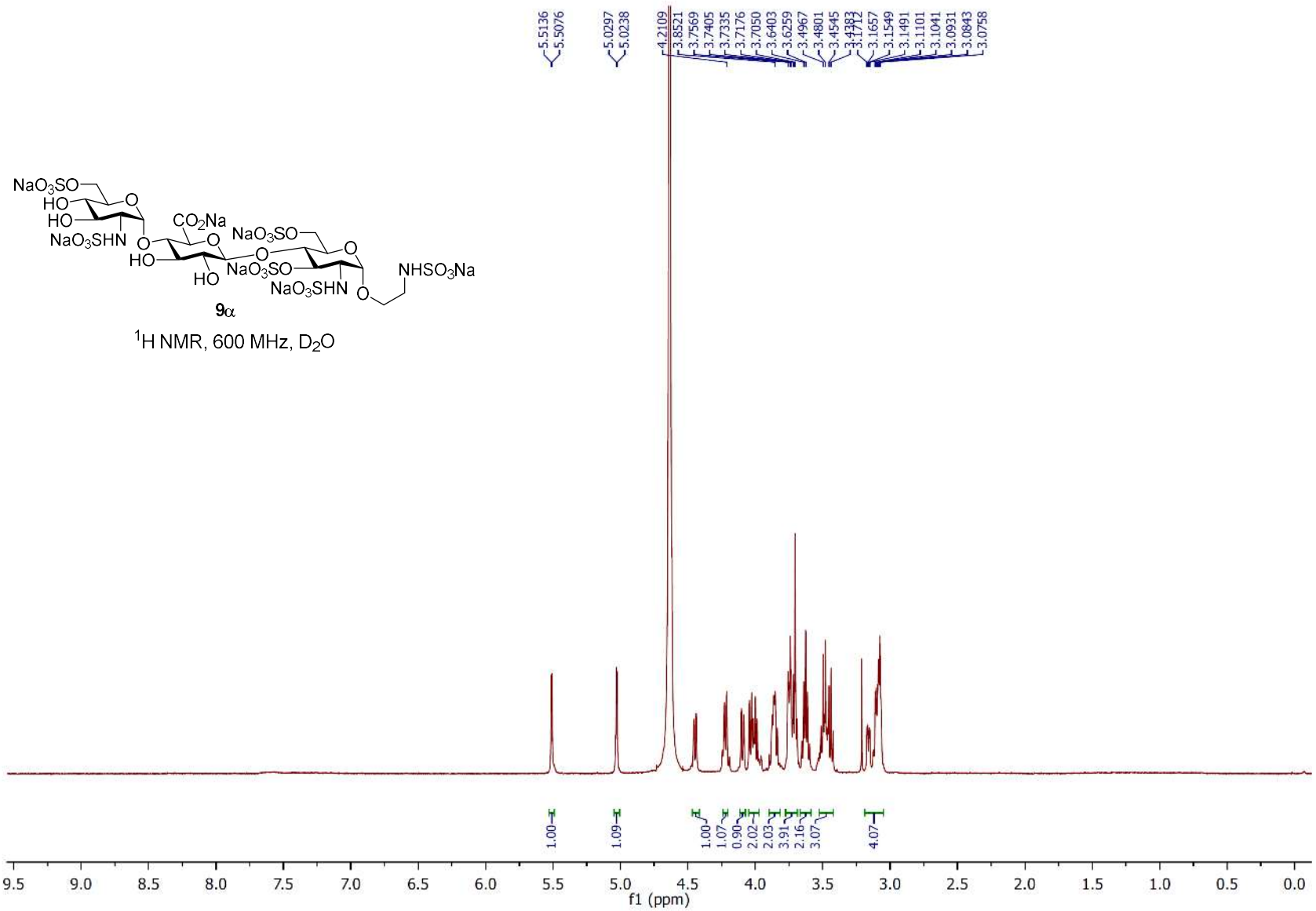
^{13}C NMR, 150 MHz, D_2O

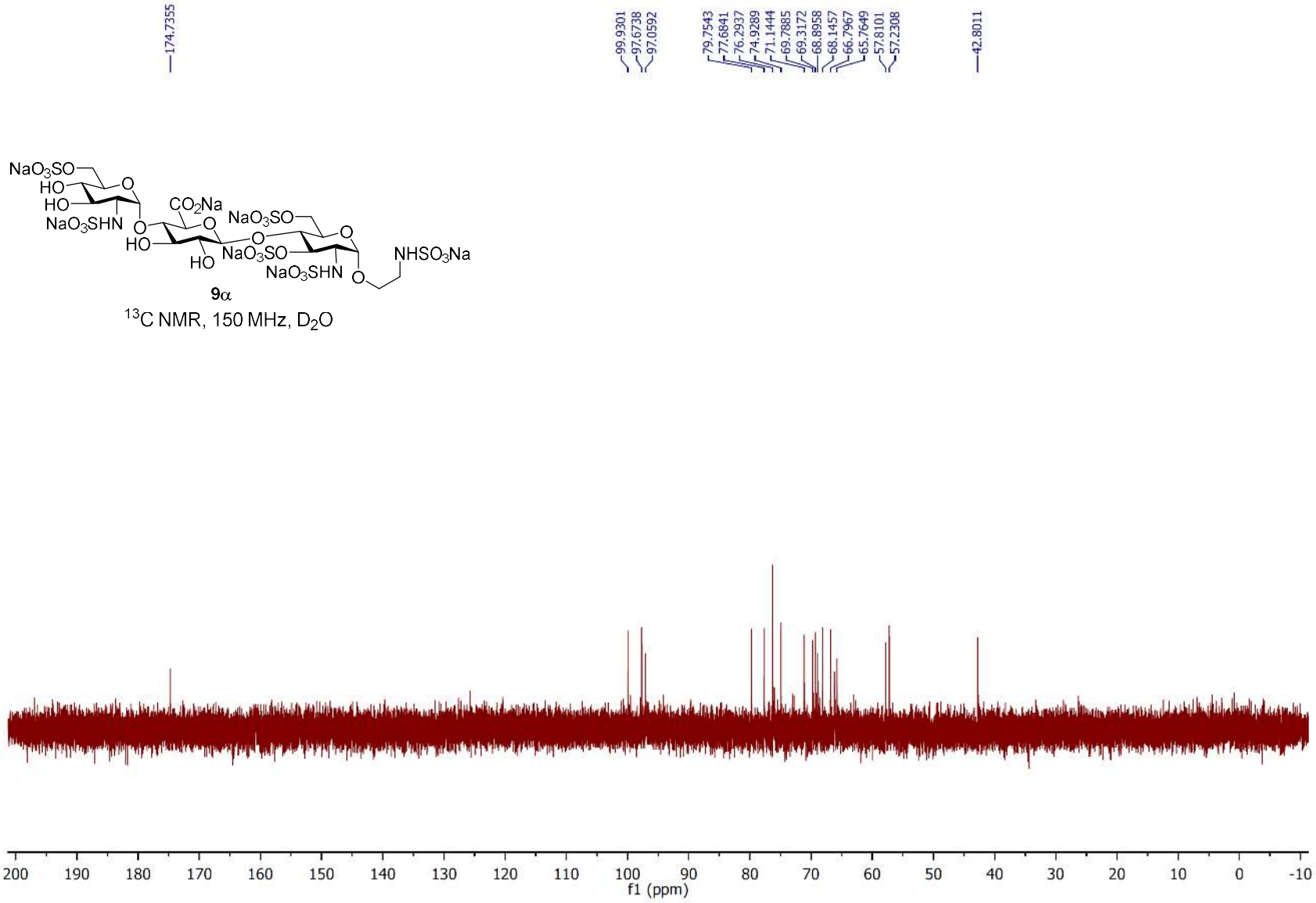


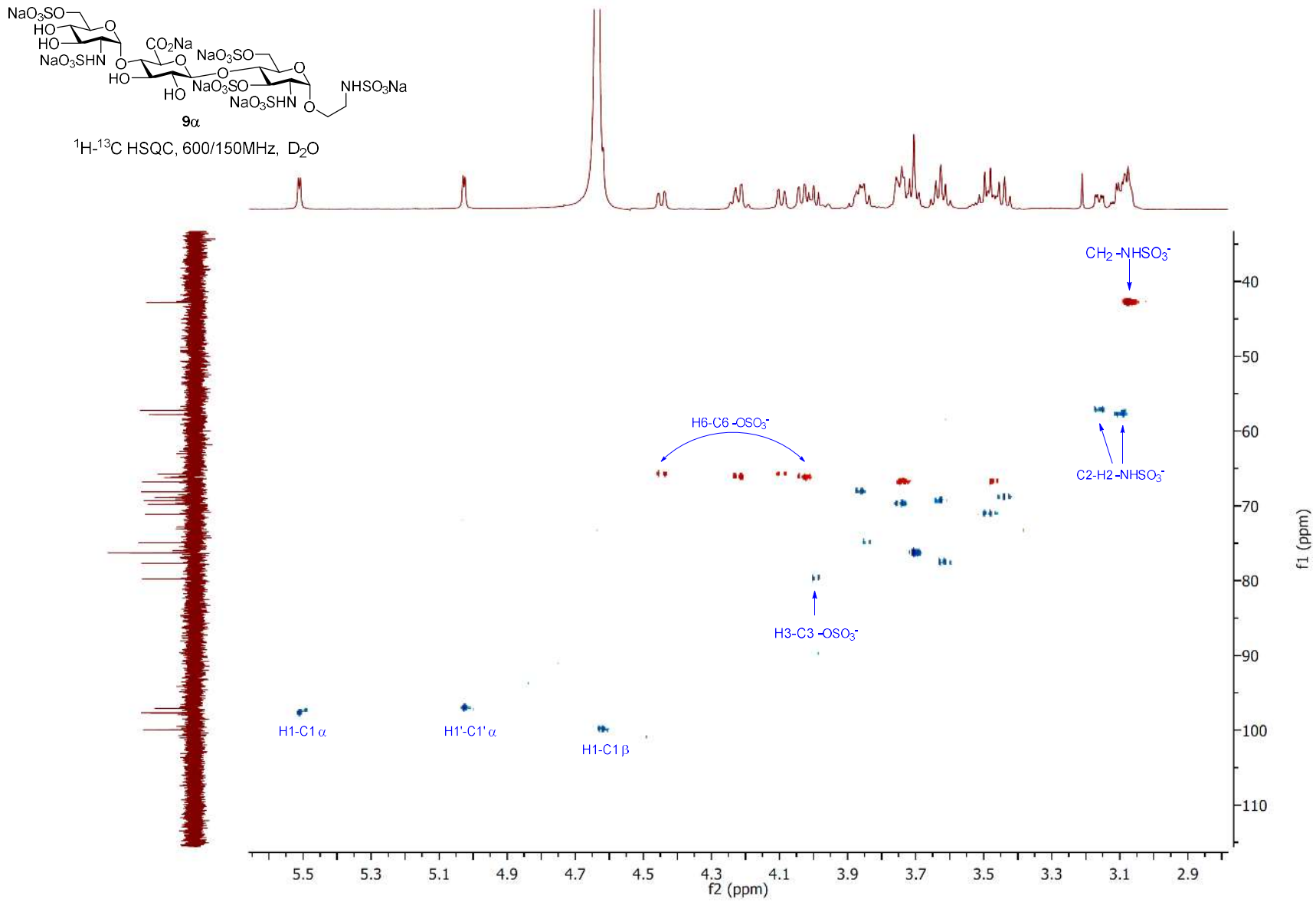
8α

^{1}H-^{13}C HSQC, 600/150MHz, D_2O




NaO3SO
HO
HO
NaO3SHN
O
CO2Na
O
HO
HO
NaO3SO
O
NaO3SO
NaO3SHN
O
NHSO3Na
9α
1H NMR, 600 MHz, D2O
5.5136
5.5076
5.0297
5.0238
4.2109
3.8521
3.7569
3.7405
3.7335
3.7176
3.7050
3.6403
3.6259
3.4967
3.4801
3.4545
3.1657
3.1549
3.1491
3.1101
3.1041
3.0931
3.0843
3.0758
1.00
1.09
1.00
1.07
0.90
2.02
2.03
3.91
2.16
3.07
4.07
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)


174.7355
99.9301
97.6738
97.0592
79.7543
77.6841
76.2937
74.9289
71.1444
69.7885
69.3172
68.8958
68.1457
66.7967
65.7649
57.8101
57.2308
42.8011
NaO3SO
HO
HO
NaO3SHN
CO2Na
NaO3SO
HO
NaO3SO
HO
NaO3SHN
NHSO3Na
9α
13C NMR, 150 MHz, D2O
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


9α
^{1}H-^{13}C HSQC, 600/150MHz, D$_2$O
CH$_2$ -NHSO$_3^-$
H6-C6 -OSO$_3^-$
C2-H2 -NHSO$_3^-$
H3-C3 -OSO$_3^-$
H1-C1 α
H1'-C1' α
H1-C1 β
f2 (ppm)
f1 (ppm)