Supporting Information for:

A sulfonamide sialoside analogue for targeting Siglec-8 and -F on immune cells.

Corwin M. Nycholat,[1†] Shiteng Duan,[1†] Eva Knuplez,[2] Charli Worth,[1] Mila Elich,[1] Anzhi Yao,[1] Jeremy O'Sullivan,[2] Ryan McBride,[1] Yadong Wei,[3] Steve M. Fernandes,[4] Zhou Zhu,[3,5] Ronald L. Schnaar,[4] Bruce S. Bochner,[2] James C. Paulson[1*]

[1]Department of Molecular Medicine, The Scripps Research Institute, La Jolla, CA 92037, USA

[2]Department of Medicine, Division of Allergy and Immunology, Northwestern University Feinberg School of Medicine, Chicago, IL 60611, USA

[3]Section of Allergy and Clinical Immunology, Yale University School of Medicine, New Haven, CT 06511, USA

[4]Department of Pharmacology and Molecular Sciences, Johns Hopkins University School of Medicine, Baltimore, MD 21205, USA

[5]Department of Molecular Microbiology and Immunology, Department of Pediatrics, Brown University Alpert Medical School, Providence, RI 02912, USA

[*]To whom correspondence should be addressed. Tel: +1-858-784-9634; e-mail: jpaulson@scripps.edu

[†]These authors contributed equally to this work.

Table of Contents

A. General Methods

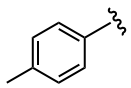
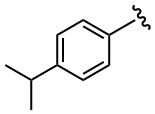
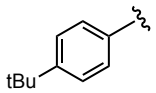
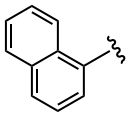
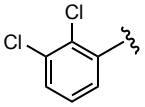
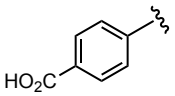
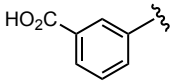
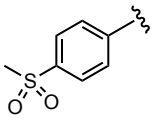
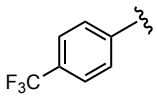
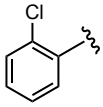
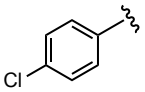
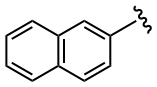
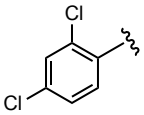
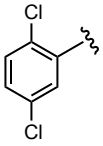
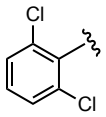
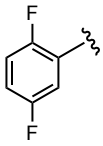
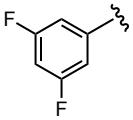
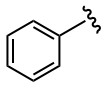
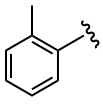
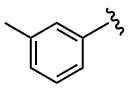
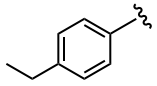
Chemical and biological reagents were obtained from Fisher Scientific or Sigma-Aldrich. 1,2-distearoyl-*sn*-glycero-3-phosphocholine (DSPC) and cholesterol were obtained from Avanti Polar Lipids (Alabaster, AL). 3-(*N*-succinimidyloxyglutaryl) aminopropyl polyethyleneglycol-carbamyl distearoyl phosphatidyl-ethanolamine (DSPE-PEG-NHS) and polyethyleneglycol-distearoyl phosphoethanolamine (DSPE-PEG) were obtained from NOF America Corporation (White Plains, NY). 5-Acetamido-9-amino-3,5,9-trideoxy-D-glycero-D-galacto-2-nonulopyranosyl-onate (9-amino-Neu5Ac) (i) and Galβ1-4GlcNAc-ethyl azide (LacNAc-ethyl azide) (ii), and 9-amino Neu5Acα2,3LacNAc-ethyl azide (iii) were prepared as previously described.[1-3] 6′-O-sulfo Galβ1-4GlcNAc-ethyl azide (v) was provided by Prof. Nicolai Bovin (Shemyakin-Ovchinnikov Institute of Bioorganic Chemistry, Moscow). Plasmid encoding *Pasteurella multocida* α2-3-sialyltransferase 1 (PmST1) was obtained from Prof. Peng Wu (Albert Einstein College of Medicine, NY) and expressed and purified as previously reported.[3] Plasmid encoding *Photobacterium damsela* α2-6-sialyltransferase (Pd2,6ST) were obtained from Prof. Xi Chen (University of California, Davis) and expressed and purified as described previously.[4] Enzymatic reactions were monitored by analytical thin-layer chromatography (TLC) performed on glass plates coated with Silica gel 60-F_{254} (E. Merck) and visualized under UV or by treatment with 10% sulfuric acid in ethanol followed by heating. Siglec-F Fc chimera were obtained commercially from R&D Systems. Siglec-8 COMP (cartilage oligomeric matrix protein) and Fc fusion proteins were obtained from Prof. Ron Schnaar (Johns Hopkins University).[5] Goat anti-human-IgG-RPE was obtained from Jackson ImmunoResearch. Alexa Fluor® 488 anti-His Tag antibody was obtained from BioLegend®. Chinese hamster ovary (CHO) cell lines expressing human or murine siglecs were maintained as previously described.[6-8] BW5147 cells expressing Siglec-G and BWZ cells expressing Siglec-H were maintained as previously described.[3, 9] Siglec-G KO mice were a generous gift from Dr. Paul Crocker, University of Dundee, UK. Siglec-F KO mice were a generous gift from Dr. Ajit Varki, UCSD. ^{1}H NMR spectra were obtained on a *Bruker* DRX-600 (600 MHz) spectrometer and are reported in parts per million (δ) relative to HOD (4.78 ppm, D_2O).

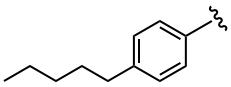
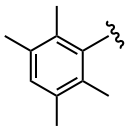
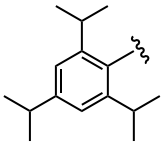
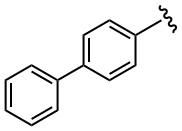
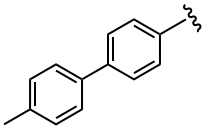
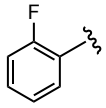
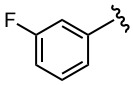
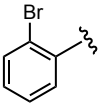
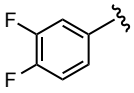
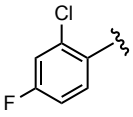
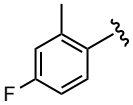
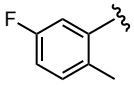
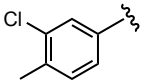
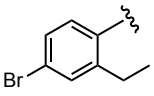
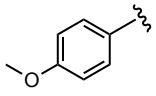
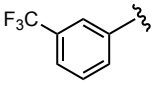
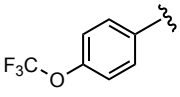
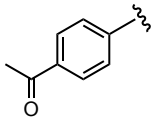
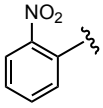
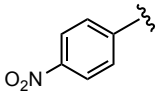
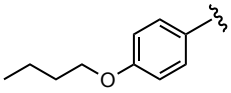
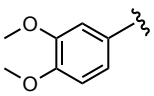
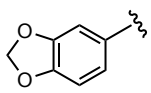
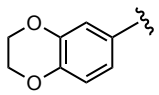
Coupling constants (*J*) are reported in Hertz. ^{13}C NMR spectra were obtained on a *Bruker* DRX-600 (150 MHz) spectrometer and are reported in parts per million (δ). Mass spectrometry data were acquired with an LC/MSD TOF (Agilent Technologies, Foster City, California, USA) for ESI-TOF high resolution mass spectrometry (HRMS) data.

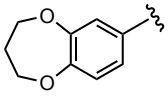
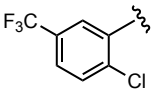
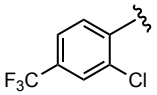
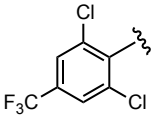
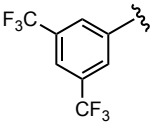
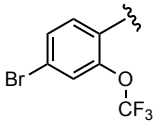
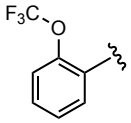
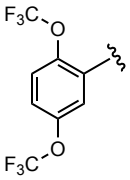
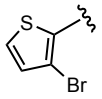
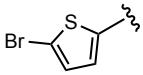
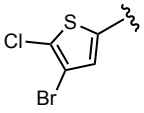
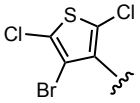
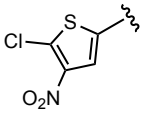
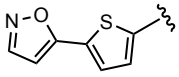
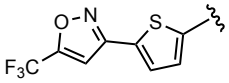
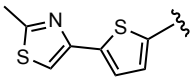
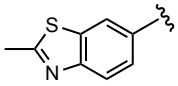
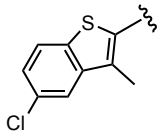
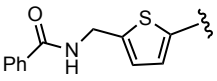
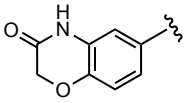
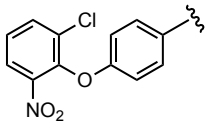
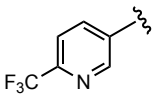
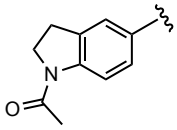
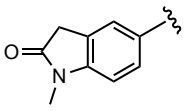
B. Glycan Microarray screening

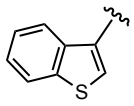
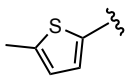
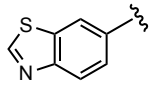
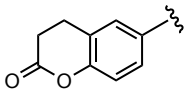
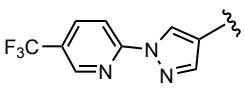
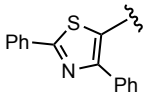
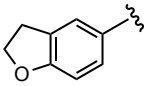
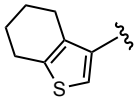
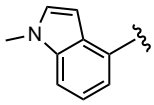
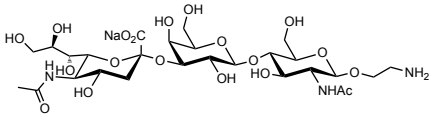
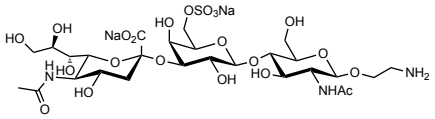
Protocols for the glycan microarray analysis of recombinant Siglecs were as previously described.[10] Glycan arrays were custom printed on *N*-hydroxy succinimide activated glass slides (Slide-H, Schott) as previously described using a MicroGridII (Digilab) contact microarray robot equipped with Stealth4B microarray pins (Telechem). The glycans were printed starting from 100 μM in five, 5-fold dilutions (4 replicates each). Briefly, each Siglec was pre-complexed prior to applying to the glass slide. Recombinant mouse Siglec-F or Siglec-8 Fc chimeras (10 μg/ml, R&D Systems) were mixed with goat anti-human-IgG-RPE (5 μg/ml, Jackson ImmunoResearch). Recombinant human Siglec-8 COMP (10 μg/ml) was mixed with Alexa Fluor® 488 anti-His Tag Antibody (5 μg/ml, BioLegend®). These prepared mixtures of complexes were incubated for 15 min on ice, diluted to 100 μl with PBS-T (phosphate buffered saline containing 0.05% Tween 20) and incubated on the array surface in a humidified chamber for 1 hour. Slides were subsequently washed by successive rinses with PBS-T, PBS and deionized H_2O. Washed arrays were dried by centrifugation and immediately scanned for Alexa Fluor 488 or RPE signal on a Perkin-Elmer Proscanarray Express. Fluorescent signal intensity was measured using Imagene (Biodiscovery) and raw signal data were calculated for mean intensity.

Table S1. Sulfonamide sialoside analog library. Shown are the substituents (R, Scheme 1) of the α2-3 sialosides (**1**-**78**) and α2-6 sialosides (**79**-**156**). Shown also are the control glycans, Neu5Acα2-3-Galβ1-4GlcNAc (**C1**) and 6′-*O*-sulfo Neu5Acα2-3-Galβ1-4GlcNAc (**C2**).

#	R =	#	R =	#	R =
1, 79		**2, 80**		**3, 81**	
4, 82		**5, 83**		**6, 84**	
7, 85		**8, 86**		**9, 87**	
10, 88		**11, 89**		**12, 90**	
13, 91		**14, 92**		**15, 93**	
16, 94		**17, 95**		**18, 96**	
19, 97		**20, 98**		**21, 99**	

#	R =	#	R =	#	R =
22, 100		**23, 101**		**24, 102**	
25, 103		**26, 104**		**27, 105**	
28, 106		**29, 107**		**30, 108**	
31, 109		**32, 110**		**33, 111**	
34, 112		**35, 113**		**36, 114**	
37, 115		**38, 116**		**39, 117**	
40, 118		**41, 119**		**42, 120**	
43, 121		**44, 122**		**45, 123**	

#	R =	#	R =	#	R =
46, 124		**47, 125**		**48, 126**	
49, 127		**50, 128**		**51, 129**	
52, 130		**53, 131**		**54, 132**	
55, 133		**56, 134**		**57, 135**	
58, 136		**59, 137**		**60, 138**	
61, 139		**62, 140**		**63, 141**	
64, 142		**65, 143**		**66, 144**	
67, 145		**68, 146**		**69, 147**	

#	R =	#	R =	#	R =
70, 148		**71, 149**		**72, 150**	
73, 151		**74, 152**		**75, 153**	
76, 154		**77, 155**		**78, 156**	
C1		**C2**			

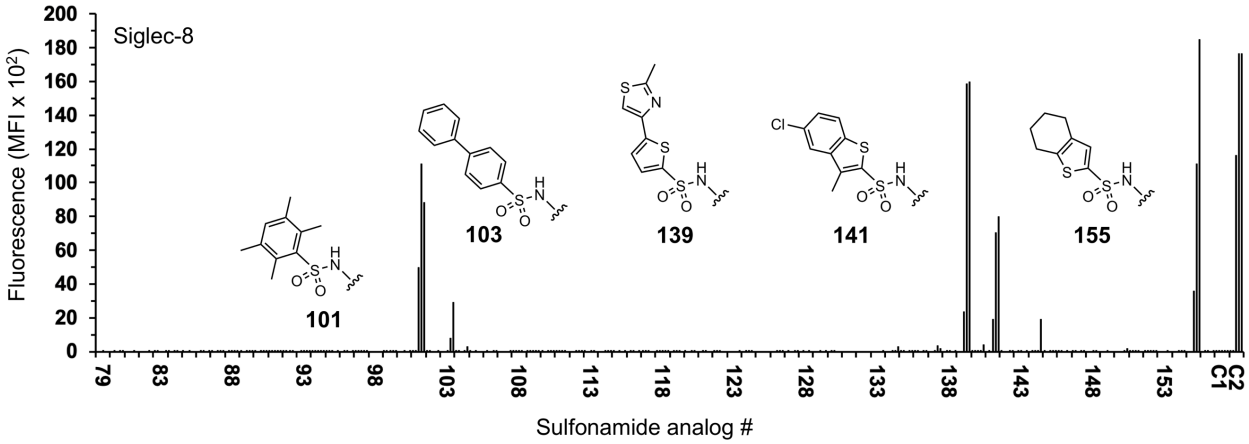


Figure S1. Binding of recombinant Siglec-8 to the α2,6-sialyl sulfonamide analog array. Human Siglec-8 Fc was pre-complexed with goat anti-human IgG-RPE (R-phycoerythrin). The complexed proteins were overlaid onto the printed array, washed then scanned for fluorescence. Analogs **79**-**156** correspond to the groups listed in Table S1. Shown is relative fluorescence of Siglec-8 binding. Each glycan was printed at 100, 20, 4, 0.8, and 0.16 μM in 4 replicates each (increasing concentration from left to right). The controls are Neu5Acα2-3Galβ1-4GlcNAc (**C1**) and 6′-*O*-sulfo Neu5Acα2-3Galβ1-4GlcNAc (**C2**).

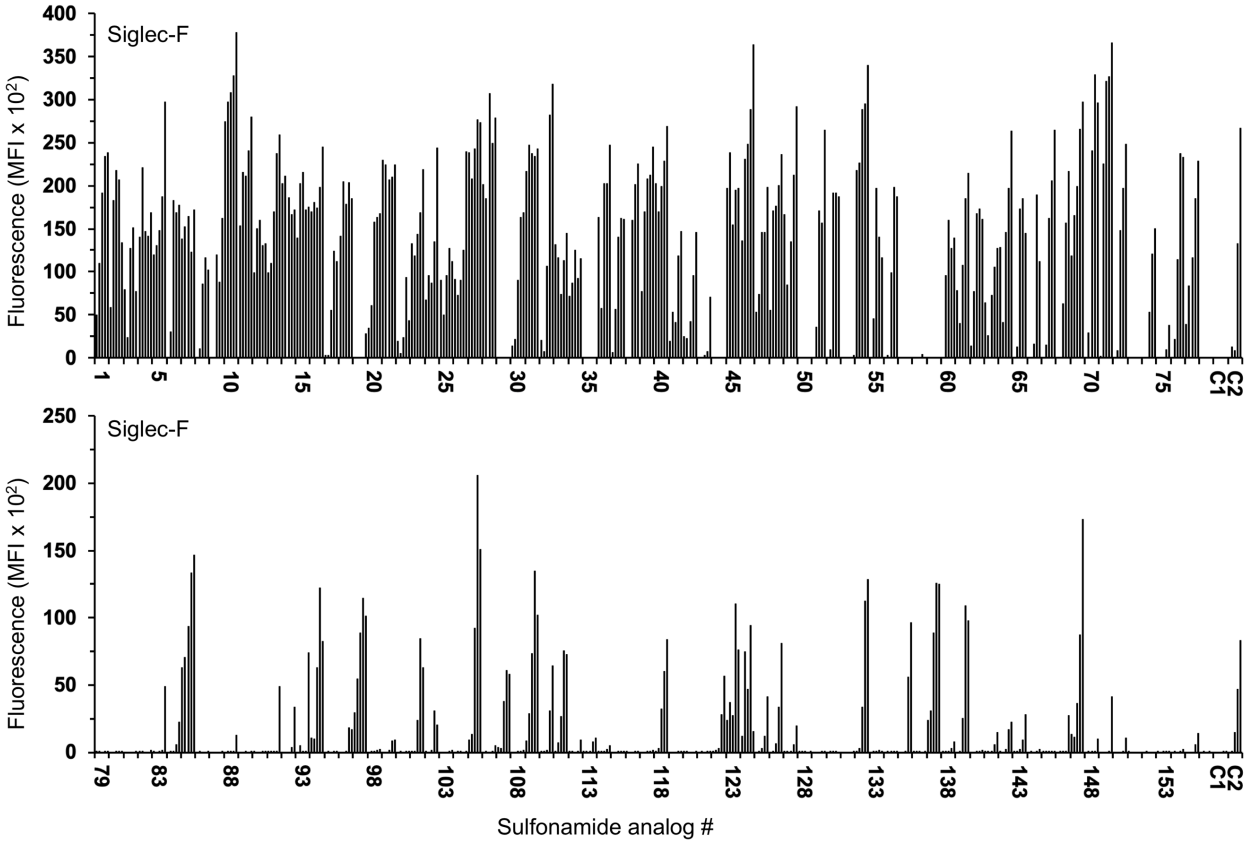
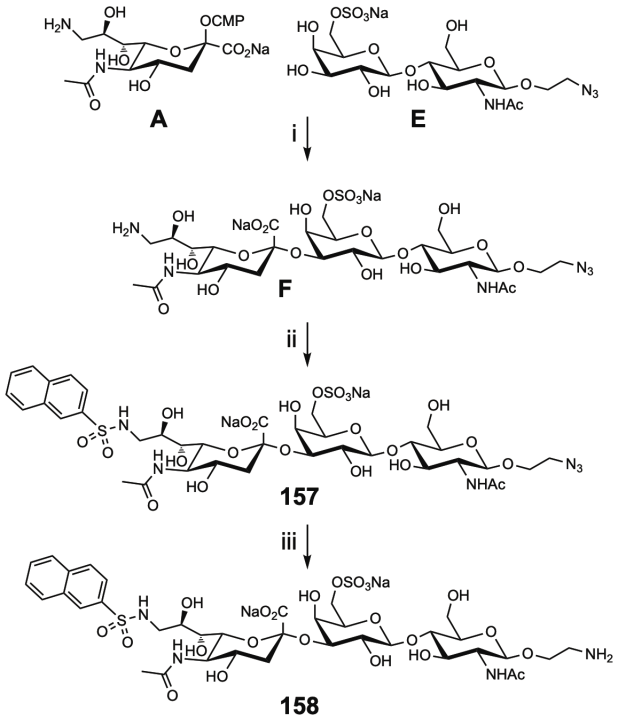


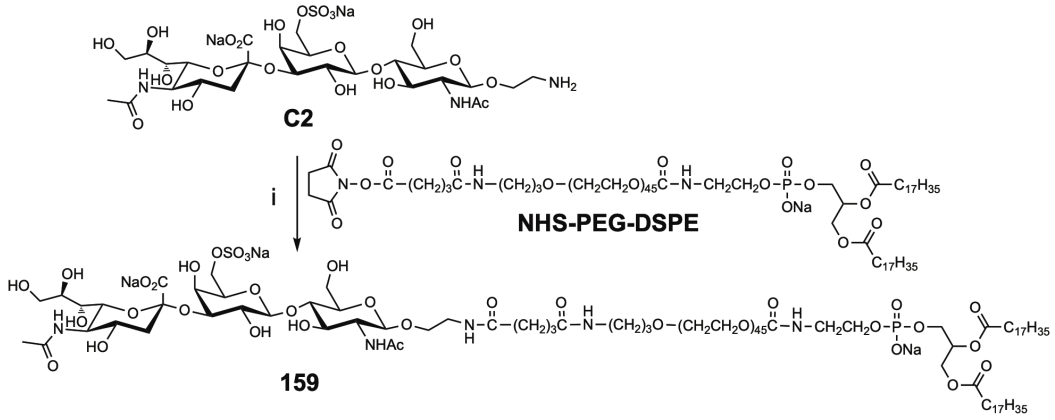
Figure S2. Binding of recombinant Siglec-F to the α2,3- and α2,6-sialyl sulfonamide analog arrays. Murine Siglec-F Fc was pre-complexed with goat anti-human IgG-RPE (R-phycoerythrin). The complexed proteins were overlaid onto the printed array, washed then scanned for fluorescence. Analogs **1-78** (α2,3-sialyl sulfonamide analogs) and **79-156** (α2,6-sialyl sulfonamide analogs) correspond to the groups listed in Table S1. Shown is relative fluorescence of Siglec-F binding. Each glycan was printed at 100, 20, 4, 0.8, and 0.16 μM in 4 replicates each (increasing concentration from left to right). The controls are Neu5Acα2-3Galβ1-4GlcNAc (**C1**) and 6′-*O*-sulfo Neu5Acα2-3Galβ1-4GlcNAc (**C2**).

Scheme S1. Synthesis of 6′-*O*-sulfo sulfonamide analog **158**.[a]



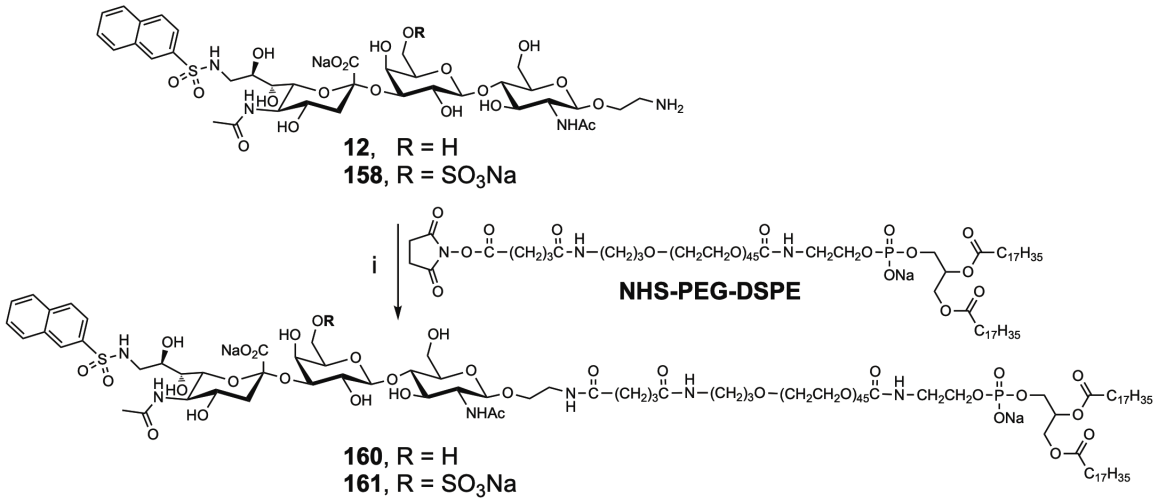
[a]Reagents and conditions: (i) *Pasteurella multocida* α2,3-sialyltransferase; (ii) 2-naphthyl sulfonyl chloride (**12**, see Table S1, 2 eq.), DIEA (5 eq.), CH_3OH; (iii) PMe_3 (2 eq.), THF, H_2O (pH 9).

Scheme S2. Synthesis of glyco-PEG-lipid conjugate **159**.[a]



[a]Reagents and conditions: (i) DIEA (2 eq.), $CHCl_3$, DMSO

Scheme S3. Synthesis of glyco-PEG-lipid conjugates **160** and **161**.[a]



[a]Reagents and conditions: (i) DIEA (2 eq.), $CHCl_3$, DMSO

C. Liposome formulation and *in vitro* cell binding assays.

Liposomes were prepared as previously described.[7] Glycans **C2**, **12**, and **158** were coupled as described in synthetic methods to DSPE-PEG-NHS (PEG chain molecular weight 2000). Fluorescent labeled liposomes were prepared by incorporating Alexa Fluor647-PEG-DSPE (0.1 mol%). Targeted liposomes containing 2 mol% Glyco-PEG-DSPE **159**, **160** or **161** were composed of DSPC:cholesterol:PEG-DSPE:Glyco-PEG-DSPE:AlexaFluor647-PEG-DSPE in a 57:38:2.9:2:0.1 molar ratio. Non-targeted liposomes were composed of DSPC:cholesterol:PEG-DSPE:AlexaFluor647-PEG-DSPE in a 57:38:4.9:0.1 molar ratio. For liposome preparation, lipids dissolved in $CHCl_3$ (DSPC, and cholesterol) and DMSO (Glyco-PEG-DSPE, PEG-DSPE, and Alexa Fluor647-PEG-DSPE) were mixed and concentrated under reduced pressure followed by lyophilization. The lipid mixtures were hydrated in PBS to achieve a final liposome concentration of 1 mM (total phospholipids). Liposomes were extruded through polycarbonate membrane filters (Millipore) with controlled pore sizes of 0.8, 0.2, and 0.1 μM.

For the binding assay CHO or BW cells expressing Siglecs (~10^5 cells) or splenocytes from WT or Siglec-G KO mice (~$2x10^6$ cells) were suspended in media (RPMI1640 + 10 % fetal bovine serum) then incubated with fluorescent targeted (**159**, **160** or **161**) or non-targeted liposomes (20 μM final total lipid) at 37 °C for 1 hour. Cells were then washed with excess FACS buffer (HBSS containing 0.1% BSA and 2 mM EDTA) then liposome binding was analyzed by flow cytometry.

D. Generation of mice with Siglec-8+/F- Eosinophils.

C57BL/6 mice genetically engineered to express Siglec-8 only on eosinophils were generated as previously described (so-called *SIGLEC8*^{Eo} mice).[11] Because these mice, like their wildtype counterparts, endogenously express Siglec-F on their eosinophils and other cells,[12-14] another novel strain of mice was generated by cross-breeding *SIGLEC8*^{Eo} mice with C57BL/6 mice genetically engineered to lack Siglec-F,[15] (generously provided by Dr. Ajit Varki, University of California San Diego) to generate mice that lacked Siglec-F on all cells including eosinophils, but whose

eosinophils expressed Siglec-8 (Siglec-8^{+}Siglec-F^{-} mice). The presence or absence of Siglec-8 and Siglec-F was routinely confirmed both genetically and via flow cytometry.

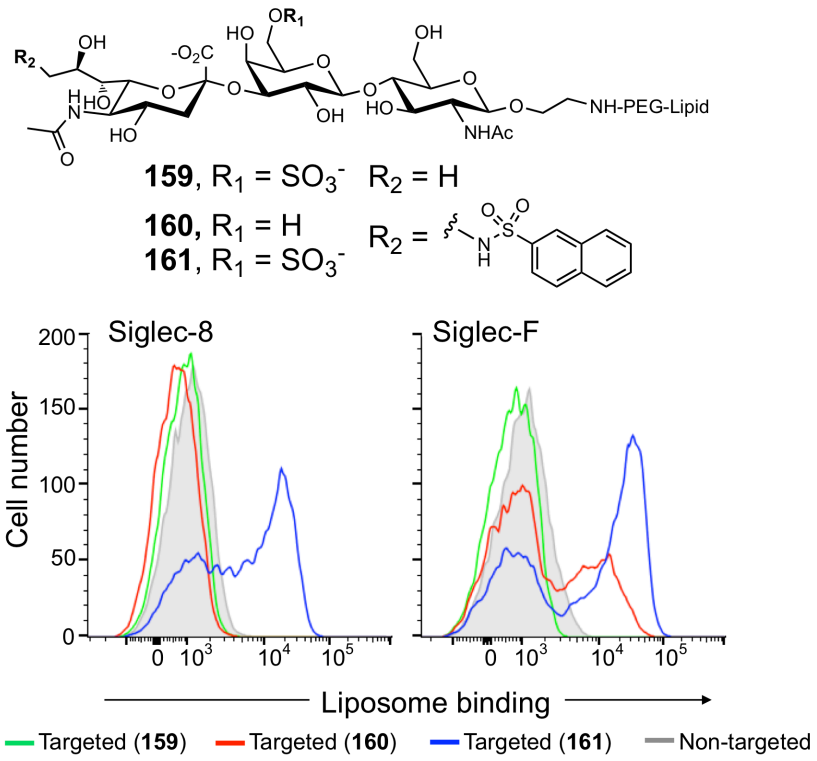


Figure S3. *In vitro* binding/uptake of fluorescent targeted liposomes (**159-161**) to Siglec-8 and -F expressing CHO cells. Cells were treated with 20 μM fluorescent targeted liposomes (2 mol% of **159-161**) at 37 °C for 1 hour. Liposome binding was assessed by flow cytometry.

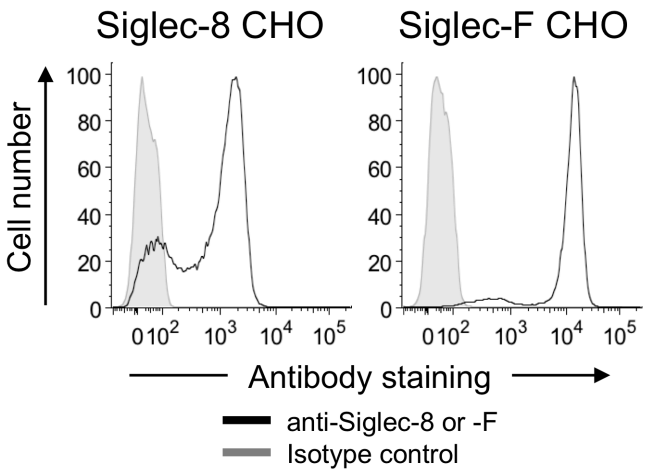


Figure S4. Antibody staining of Siglec-8 and -F expressing CHO cells analyzed by flow cytometry. Anti-Siglec-8 and -F antibodies and isotype controls were PE labeled and used at 2 μg/ml.

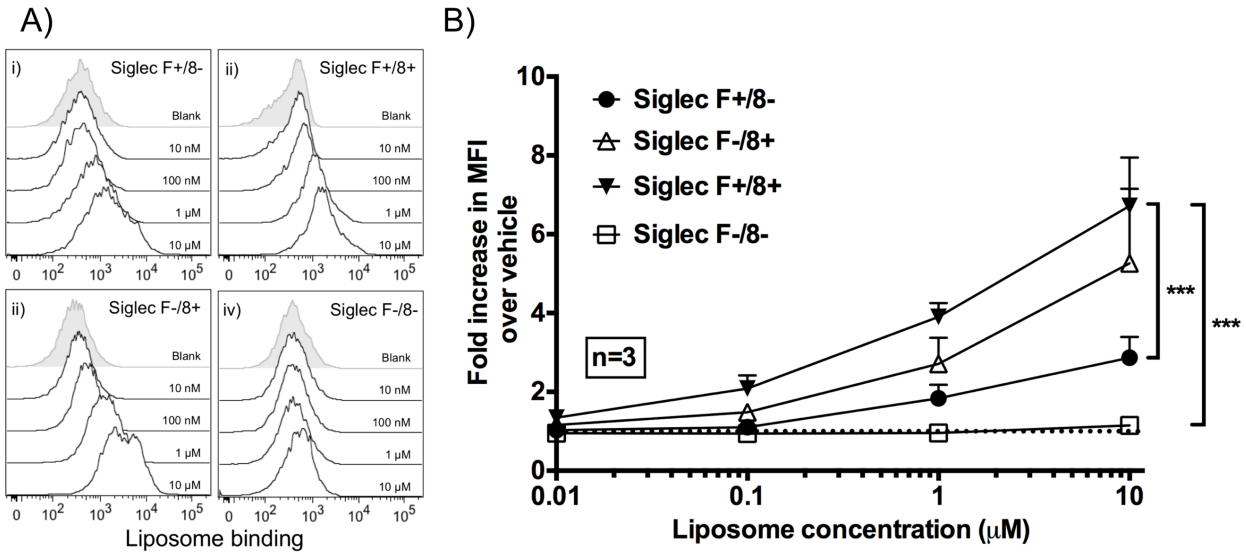


Figure S5. A) Representative flow cytometry analysis of *in vitro* binding of targeted liposomes to murine bone marrow derived eosinophils (bmEos) prepared from (i) wild-type C57BL/6 mice (Siglec-F^+/Siglec-8^-), (ii) Siglec-F^-/Siglec-8^+ mice, (iii) Siglec-F^+/Siglec-8^+ mice, and (iv) Siglec-F null mice (Siglec-F^-/Siglec-8^-). B) Fold increase in mean fluorescence intensity (MFI) of targeted liposome vs non-targeted liposomes binding to bmEos. Eosinophils were incubated at 37 °C with targeted (**161**) or non-targeted liposomes for 24 hours. Statistical significance was analyzed by 2-way ANOVA followed by Tukey's test: * P <0.05; ** P <0.01; *** P <0.001; **** P <0.0001; n.s., not significant (P >0.05).

E. *In vitro* liposome binding to murine bone marrow derived eosinophils.

Murine bone marrow derived Eosinophils were generated as previously described.[16] Eosinophils were prepared from wild-type C57BL/6 mice (Siglec-F+/Siglec-8-), Siglec-F null mice (Siglec-F-/Siglec-8-),[15] Siglec-F+/Siglec-8+ mice,[11] and Siglec-F-/Siglec-8+ mice. Mature Eosinophils (day 14) were used for the liposome binding assays. Cells were cultured in RPMI1640 media containing 20% fetal bovine serum (FBS), penicillin (100 IU/ml), streptomycin (10 μg/ml), glutamine (2 mM), HEPES (25 mM), non-essential amino acid (1x), sodium pyruvate (1 mM), and β-mercaptoethanol (50 μM). Cells (~200,000 cells/well) in media (180 μl) were transferred to a 96 well plate then incubated at 37 °C for 24 hours. For the liposome binding assay, cells (n=3) were

treated with 20 μl of either targeted (+ 2 mol% ^{NSA}Neu5Ac, **160**) or non-targeted (-^{NSA}Neu5Ac) fluorescent (AF647) liposomes at final total lipid concentrations of 10, 1, 0.1, and 0.01 μM. Control cells were treated with media (20 μl). The cells were then incubated at 37 °C for 24 hours. The cells were washed with PBS, collected then fluorescent liposome binding was analyzed by FACS analysis (LSR-II). Statistical significance was determined by 2-way ANOVA followed by Tukey's test. All p values were determined using Prism (Version 6.0f). A P <0.05 was considered significant.

F. *In vivo* liposome binding to murine eosinophils.

C57BL/6 (wild type) were maintained in pathogen-free conditions at The Scripps Research Institute breeding facility and were used in accordance to the guidelines of the Institutional Animal Care Committee at the National Institutes of Health. For the *in vivo* binding assay, wild type were injected *via* the tail vein with 200 μl of 1 mM fluorescent targeted liposomes (**160**) or non-targeted liposomes. After 1 hour, the spleens were harvested then the binding of liposomes to immune cells was analyzed by flow cytometry. Cells were stained with antibodies to identify Eosinophils ($CD11b^+CCR3^+$), B cells ($CD19^+$), and T cells ($CD4^+$ or $CD8^+$).

For Figure 4B, WT mice were intravenously given 200 μl of 3 mM targeted (5 mol%, 120 μg, **160**), 3 mM non-targeted liposomes, anti-Siglec-F antibody (100 μg) or PBS. Mice were retro-orbitally bled, and eosinophil frequencies in the blood were analyzed 1- and 3-days post injection by flow cytometry. Eosinophil frequency was determined by dividing cells that were $CD11b^+CCR3^+$ by live immune cells (PI^-CD45^+). *** P <0.001; N.S., not significant (P >0.05) determined by 1-way ANOVA followed by Tukey's test.

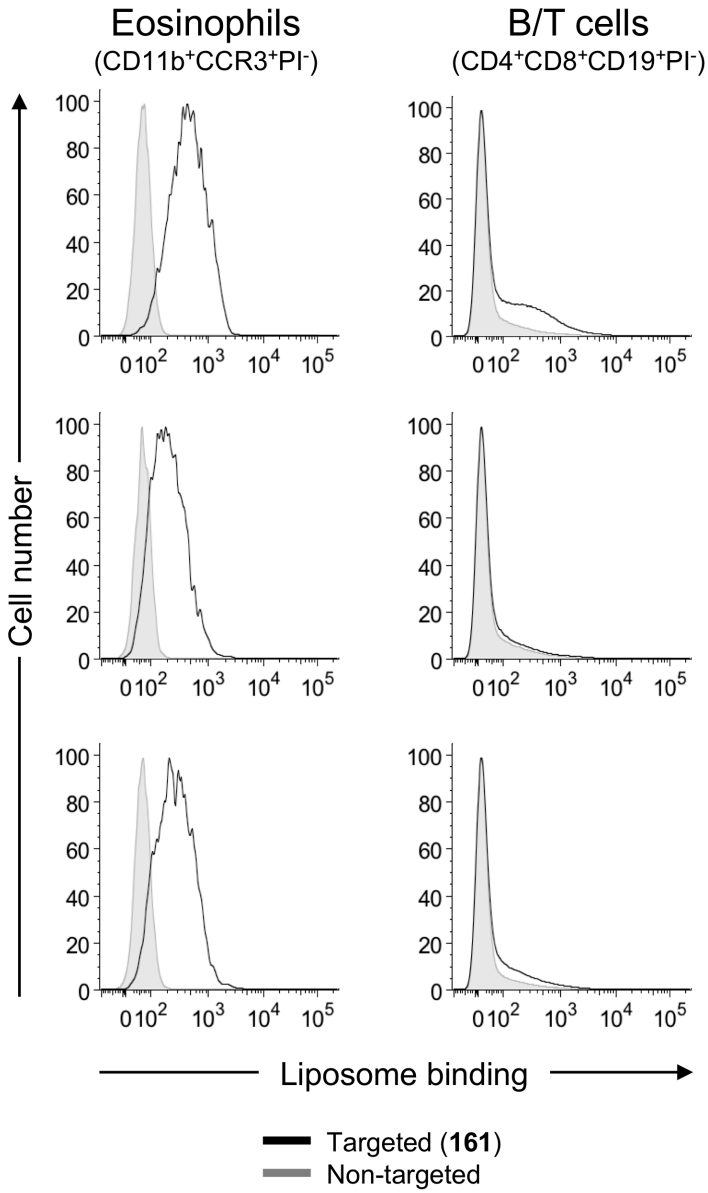


Figure S6. Targeted liposomes (**161**) bind eosinophils *in vivo*. WT mice were intravenously given either targeted (**161**, n=3) or non-targeted (n=3) fluorescent liposomes. After one hour, splenocytes were harvested, stained with antibodies then analyzed by flow cytometry. Binding of targeted (black line) or non-targeted (grey) to eosinophils ($CD11b^+CCR3^+PI^-$, *left*) or B/T cells ($CD4^+CD8^+CD19^+PI^-$, *right*) were overlaid. The top histograms from each column were shown as representative data in Figure 4A.

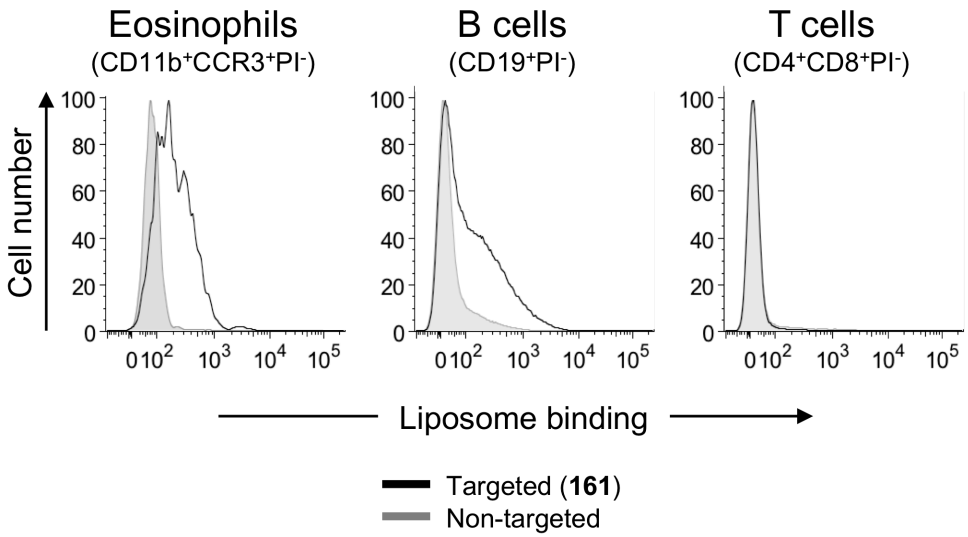


Figure S7. Targeted liposome (**161**) binding to splenocytes *in vivo* after 24 hours. WT mice were intravenously given either targeted (2 mol% **161**, n=4) or non-targeted (n=4) fluorescent liposomes (200 μl of 1 mM liposomes). After 24 hours, splenocytes were harvested, stained with antibodies then analyzed by flow cytometry. Binding of targeted (black line) or non-targeted (grey) to eosinophils ($CD11b^+CCR3^+PI^-$, *left*), B cells ($CD19^+PI^-$, *middle*) or T cells ($CD4^+CD8^+PI^-$, *right*) were overlaid.

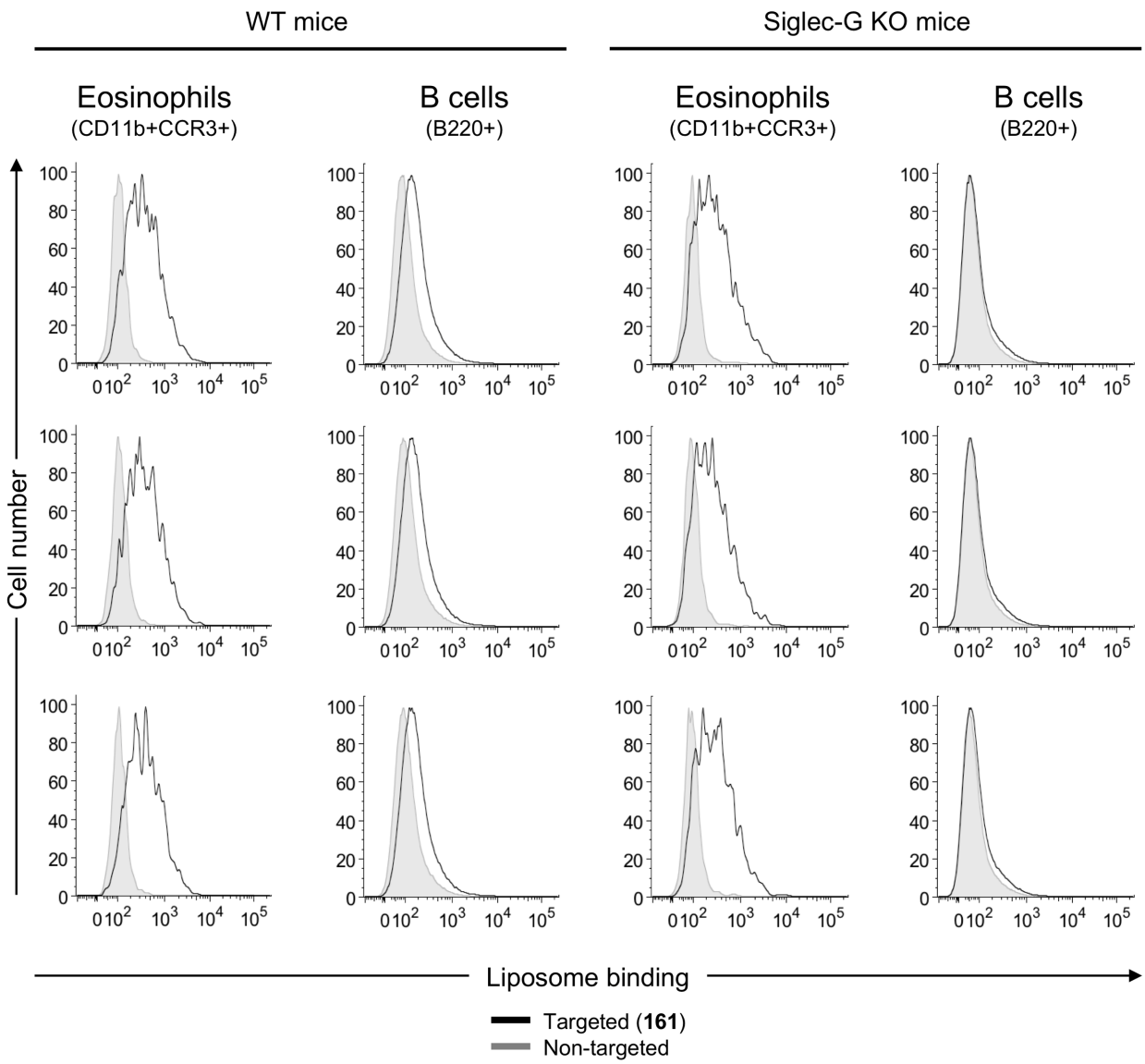
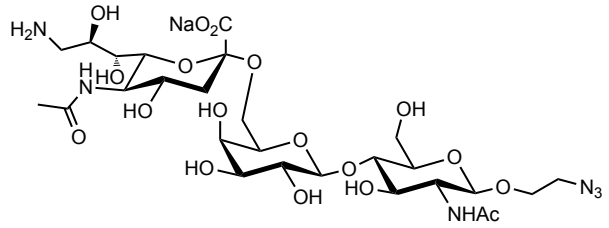


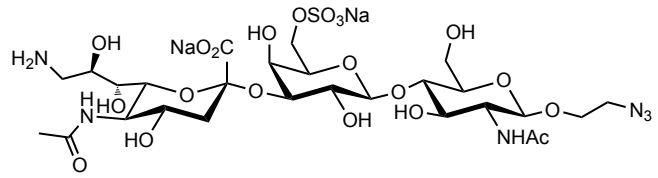
Figure S8. Targeted liposomes (**161**) bind murine B cells via Siglec-G. Splenocytes from WT or Siglec-G KO mice were incubated with targeted (**161**) or non-targeted fluorescent liposomes in triplicates, followed by antibody staining and flow cytometry analysis. Binding of targeted (black line) or non-targeted (grey) liposomes to either eosinophils ($CD11b^+CCR3^+$) or B cells ($B220^+$) from WT or Siglec-G KO mice were overlaid.

G. Synthetic methods.



Synthesis of 2-azidoethyl (5-acetamido-9-amino-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (D)

Routine preparation of compound (**D**) was as follows. Glycosyl acceptor LacNAc-ethyl azide **B** (1 eq.) and glycosyl donor CMP 9-amino Neu5Ac **A** (2 eq.) were dissolved in Tris-HCl buffer (100mM, pH 9.0, 25 mL/mmol acceptor) containing $MgCl_2$ (20mM). α2,6-sialyltransferase Pd2,6ST (~30 U/mmol donor) was added then the reaction was mixed at 37 °C. The pH was monitored and adjusted (to pH 8.5-9.0) as needed using 1.0 M NaOH (aq.). After, 20 hours the completed reaction was filtered through a pipette column (1 cm) of Dowex® resin (50WX8, Na^+ form). The product was then purified by size exclusion chromatography (Sephadex® G-15, H_2O eluent) and isolated in 70-80% yield. R_f 0.21 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 4.65 (1H, d, J = 8.2 Hz), 4.47 (1H, d, J = 7.9 Hz), 4.11-4.05 (2H, m), 4.02-3.97 (2H, m), 3.95 (1H, d, J = 3.2 Hz), 3.86-3.73 (7H, m), 3.70-3.62 (4H, m), 3.56-3.49 (4H, m), 3.46-3.42 (2H, m), 3.04 (1H, dd, J = 9.6, 13.1 Hz), 2.69 (1H, dd, J = 4.6, 12.4 Hz), 2.08 (3H, s), 2.04 (3H, s), 1.73 (1H, dd, J = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.2, 173.0, 103.0, 100.3, 99.8, 80.2, 74.0, 73.1, 72.0, 71.9, 70.3, 69.8, 68.3, 68.1, 67.9, 67.5, 62.8, 59.8, 54.4, 51.4, 49.9, 42.0, 39.6, 21.9, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{27}H_{46}N_6O_{18}$ 743.2941, found 743.2938.



Synthesis of 2-azidoethyl (5-acetamido-9-amino-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-6-*O*-sulfo-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (F)

Routine preparation of compound (**F**) was as follows. 6′-*O*-sulfo LacNAc-ethyl azide **E** (16.0 mg, 28.9 μmol) and CMP 9-amino Neu5Ac **A** (28.5 mg, 43.3 μmol, 1.5 eq.) were dissolved in Tris-HCl buffer (100mM, pH 9.0, ~40 mL/mmol acceptor) containing $MgCl_2$ (20mM). The α2,3-sialyltransferase PmST1 (~40 U/mmol donor) was added then the reaction was mixed at 37 °C. The pH was monitored and adjusted (to pH 8.5-9.0) as needed using 1.0 M NaOH (aq.). After, 20 hours the completed reaction was filtered through a pipette column (1 cm) of Dowex® resin (50WX8, Na^+ form). The product was then purified by size exclusion chromatography (Sephadex® G-15, H_2O eluent) and isolated in 70-80% yield with a purity >90%. R_f 0.16 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 4.54-4.53 (2H, m), 4.14-4.05 (3H, m), 4.01-3.89 (5H, m), 3.79-3.61 (7H, m), 3.58-3.50 (3H, m), 3.47 (1H, dd, J = 1.2, 8.7 Hz), 3.41 (1H, ddd, J = 2.9, 7.5, 13.8 Hz), 3.37-3.32 (2H, m), 2.92 (1H, dd, J = 9.8, 13.0 Hz), 2.66 (1H, dd, J = 4.6, 12.5 Hz), 1.97 (3H, s), 1.96 (3H, s), 1.75 (1H, dd, J = 12.0, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.2, 173.4, 102.0, 100.5, 99.8, 78.7, 74.7, 74.3, 72.1_3, 72.0_6, 71.8, 69.6, 68.9, 68.2, 67.8_1, 67.7_7, 66.9, 66.7, 59.8, 54.6, 51.1, 49.9, 41.8, 38.7, 21.8, 21.6; **HRMS** (ESI-TOF high acc.) *m/z* $[M + Na]^+$ expected for $C_{27}H_{46}N_6O_{21}S$ 845.2329, found 845.2324.

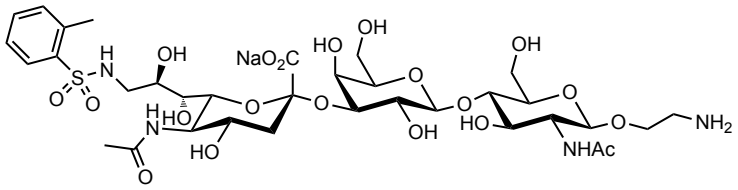
General procedure (A) for synthesis of 9-*N*-sulfonyl-subsituted sialoside analogs (1-156)

For the initial screen of sialoside analog glycan arrays, 9-*N*-sulfonyl (sulfonamide) substituted sialic acid analogs (**1-156**) were prepared as follows. Analogs **1-78** were synthesized from 9-amino Neu5Acα2,3LacNAc (**C**) and analogs **79-156** were synthesized from 9-amino Neu5Acα2,6LacNAc (**D**). Aqueous stock solutions of (**C**) or (**D**) were prepared then used to

aliquot 0.5 mg samples of each glycan into 2 ml glass vials. Samples were then lyophilized to white amorphous solids. In parallel, compounds iii or iv (0.50 mg, 0.65 μmol) were dissolved in anhydrous methanol (250 μl) containing *N*,*N*-diisopropylethylamine (5 eq.). Substituted sulfonyl chlorides (1.3 μmol, 2 eq.; **1**-**156**, see Table S1) were then added and the reactions were kept at room temperature (22 °C) for 2 hours. The reactions were monitored by TLC (EtOAc-CH_3OH-AcOH-H_2O, 6:3:3:2). The completed reactions were then concentrated under reduced pressure. The remaining residues were dissolved in deionized H_2O (1 ml) with pH adjusted to 9 with 1 mM NaOH. In parallel to each reaction was added trimethyl phosphine (5 eq., 1 M in THF) and the reactions were kept at room temperature for ~20 hours. The reactions were monitored by TLC (EtOAc-CH_3OH-AcOH-H_2O, 6:3:3:2). The completed reactions were then concentrated under reduced pressure then lyophilized to a white amorphous solid. Sulfonamide analogs were used as without purification. Aqueous stock solutions (200 μM) of each analog were prepared for glycan microarray printing.

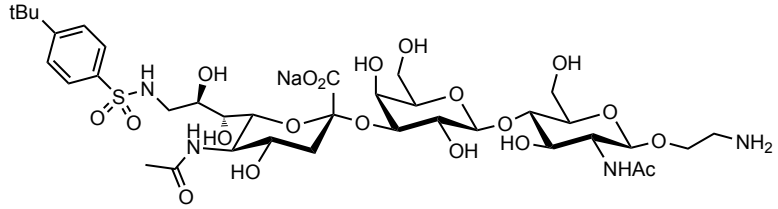
General procedure (B) for re-synthesis of 9-*N*-sulfonyl-subsituted sialoside analogs

The re-synthesis of α2,3- (**2**, **7**, **12**, **13**, **25**, **30**, **43**, **50**, **54**, **61**, **65**, and **76**) and α2,6- (**82**, **89**, **97**, **106**, **110**, **126**, **129**, **150**, and **155**) sulfonamide analogs (2.0 mg scale each) was accomplished as described above. Following reduction of ethyl azides using trimethylphosphine the reactions were concentrated under reduced pressure then dissolved in H_2O and passed through Dowex® resin (50WX8, Na^+ form). The filtrates were then passed through equilibrated Waters C-18 Sep-Pak® cartridges (500 mg). The columns were washed with deionized H_2O then the products were eluted with 30-50% methanol-H_2O solutions. Fractions containing product were combined and the samples lyophilized. Typical isolated yields for the 9-*N*-sulfonyl substituted analogs were 75-85%.



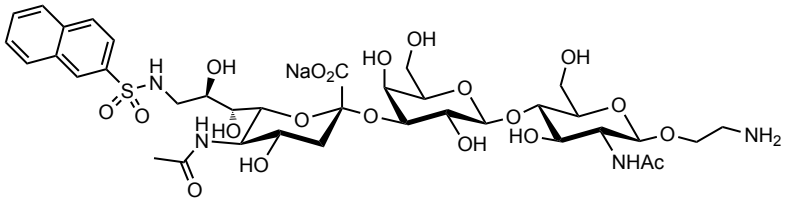
2-aminoethyl (5-acetamido-9-(2-methyl benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (2)

Prepared using general procedure **B**. R_f 0.11 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.96 (1H, d, *J* = 7.9 Hz), 7.62 (1H, dd, *J* = 7.4, 7.5 Hz), 7.50 (1H, d, *J* = 7.5 Hz), 7.46 (1H, dd, *J* = 7.6, 7.7 Hz), 4.58 (1H, d, *J* = 8.4 Hz), 4.48 (1H, d, *J* = 7.8 Hz), 4.07 (1H, dd, *J* = 3.1, 10.1 Hz), 4.05 (1H, ddd, *J* = 3.5, 7.6, 7.9 Hz), 4.00 (1H, dd, *J* = 1.9, 12.3 Hz), 3.93 (1H, d, *J* = 2.9 Hz), 3.92-3.88 (2H, m), 3.85 (1H, dd, *J* = 5.1, 12.3 Hz), 3.82-3.79 (2H, m), 3.78-3.67 (6H, m), 3.63-3.61 (2H, m), 3.57 (1H, dd, *J* = 8.0, 9.6 Hz), 3.50 (1H, dd, *J* = 1.2, 8.7 Hz), 3.37 (1H, dd, *J* = 2.5, 13.9 Hz), 3.23 (1H, ddd, *J* = 3.5, 5.7, 13.7 Hz), 3.18 (1H, ddd, *J* = 3.5, 7.4, 13.5 Hz), 3.05 (1H, dd, *J* = 7.5, 13.9 Hz), 2.75 (1H, dd, *J* = 4.6, 12.5 Hz), 2.65 (3H, s), 2.06 (3H, s), 2.03 (3H, s), 1.78 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 175.1, 174.9, 173.4, 137.1, 136.6, 133.5, 132.9, 128.7, 126.4, 102.6, 101.0, 99.8, 78.3, 75.5, 75.2, 74.7, 72.8, 72.2, 70.3, 69.3, 69.2, 68.2, 67.4, 65.9, 61.0, 60.0, 54.8, 51.7, 44.9, 39.7, 39.4, 22.1, 22.0, 19.3; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{34}H_{54}N_4O_{20}S$ 871.3125, found 871.3122.



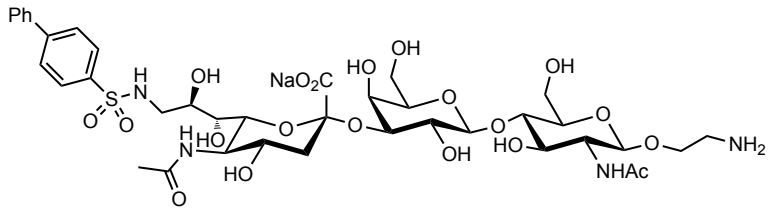
2-aminoethyl (5-acetamido-9-(4-*t*-butyl benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (7)

Prepared using general procedure **B**. R_f 0.16 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.85 (2H, d, *J* = 8.5 Hz), 7.74 (2H, d, *J* = 8.5 Hz), 4.57 (1H, d, *J* = 8.3 Hz), 4.54 (1H, d, *J* = 7.8 Hz), 4.11 (1H, dd, *J* = 2.9, 9.8 Hz), 4.05-3.99 (2H, m), 3.96-3.93 (2H, m), 3.86-3.53 (16H, m), 3.27 (1H, dd, *J* = 2.4, 13.4 Hz), 3.16 (1H, ddd, *J* = 3.7, 5.8, 13.8 Hz), 3.11 (1H, ddd, *J* = 3.7, 7.1, 13.6 Hz), 2.98 (1H, dd, *J* = 7.2, 13.5 Hz), 2.76 (1H, dd, *J* = 4.5, 12.4 Hz), 2.05 (3H, s), 2.03 (3H, s), 1.79 (1H, dd, *J* = 12.1, 12.2 Hz), 1.36 (9H, s); **^{13}C NMR** (D_2O, 150 MHz) δ 175.1, 174.8, 173.7, 157.8, 135.0, 126.6, 102.5, 101.0, 99.8, 78.0, 75.6, 75.2, 74.7, 72.8, 72.2, 70.2, 69.3, 69.1, 68.2, 67.4, 66.8, 61.0, 60.0, 54.9, 51.7, 45.1, 39.7, 39.5, 34.6, 30.2, 22.1, 22.0; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{37}H_{60}N_4O_{20}S$ 913.3594, found 913.3593.



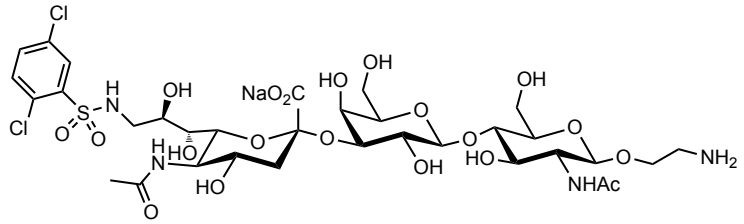
2-aminoethyl (5-acetamido-9-(2-naphthyl sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (12)

Prepared using general procedure **B**. R_f 0.15 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.56 (1H, s), 8.18 (1H, d, *J* = 8.7 Hz), 8.15 (1H, d, *J* = 8.1 Hz), 8.08 (1H, d, *J* = 8.1 Hz), 7.92 (1H, dd, *J* = 2.0, 8.7 Hz), 7.78 (1H, dd, *J* = 7.1, 7.2 Hz), 7.74 (1H, dd, *J* = 7.3, 7.6 Hz), 4.53 (1H, d, *J* = 8.4 Hz), 4.30 (1H, d, *J* = 7.8 Hz), 4.02 (1H, dd, *J* = 3.0, 9.8 Hz), 3.98 (1H, ddd, *J* = 3.9, 6.7, 10.9 Hz), 3.95-3.90 (3H, m), 3.83-3.64 (9H, m), 3.58-3.51 (5H, m), 3.39 (1H, dd, *J* = 2.7, 13.8 Hz), 3.09 (1H, dd, *J* = 7.5, 13.8 Hz), 3.03 (1H, ddd, *J* = 3.8, 6.1, 13.7 Hz), 2.98 (1H, ddd, *J* = 4.1, 6.7, 13.6 Hz), 2.75 (1H, dd, *J* = 4.6, 12.5 Hz), 2.05 (3H, s), 2.01 (3H, s), 1.77 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.3, 173.2, 135.2, 134.3, 131.4, 129.5, 128.9, 128.8, 127.7, 127.5, 121.2, 102.1, 100.7, 99.3, 78.0, 75.1, 74.6, 74.2, 72.3, 71.7, 70.0, 68.8, 68.7, 68.3, 67.7, 66.9, 62.0, 61.2, 60.4, 59.6, 54.5, 51.3, 44.7, 39.3, 21.7, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{37}H_{54}N_4O_{20}S$ 907.3125, found 907.3122.



2-aminoethyl (5-acetamido-9-(4-phenyl benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (13)

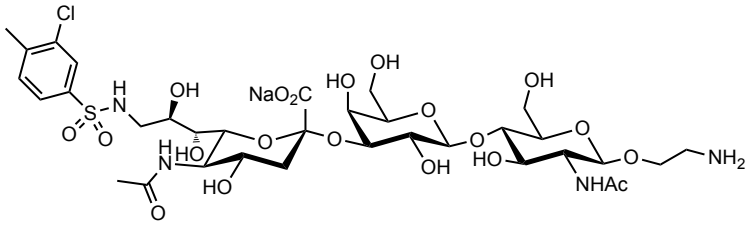
Prepared using general procedure **B**. R_f 0.16 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.01 (2H, d, *J* = 8.4 Hz), 7.94 (2H, d, *J* = 8.4 Hz), 7.79 (2H, d, *J* = 7.5 Hz), 7.59 (2H, dd, *J* = 7.5, 7.7 Hz), 7.52 (1H, dd, *J* = 7.3, 7.4 Hz), 4.54 (1H, d, *J* = 8.4 Hz), 4.45 (1H, d, *J* = 7.8 Hz), 4.06 (1H, dd, *J* = 3.0, 9.9 Hz), 3.99-3.92 (4H, m), 3.85-3.76 (4H, m), 3.72-3.62 (7H, m), 3.59-3.54 (3H, m), 3.37 (1H, dd, *J* = 2.5, 13.6 Hz), 3.10-3.01 (3H, m), 2.76 (1H, dd, *J* = 4.5, 12.4 Hz), 2.05 (3H, s), 2.03 (3H, s), 1.78 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 175.1, 174.8, 173.7, 145.5, 138.8, 137.2, 129.3, 128.8, 127.9, 127.3_2, 127.3_0, 102.5, 101.1, 99.8, 78.2, 75.5, 75.1, 74.7, 72.8, 72.2, 70.4, 69.3, 69.2, 68.2, 67.7, 67.4, 60.9, 60.0, 54.9, 51.7, 45.2, 39.7, 39.6, 22.1, 22.0; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{39}H_{56}N_4O_{20}S$ 933.3281, found 933.3267.



2-aminoethyl (5-acetamido-9-(2,5-dichloro benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (25)

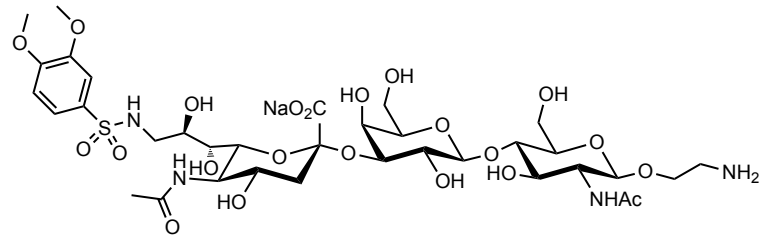
Prepared using general procedure **B**. R_f 0.15 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.13 (1H, d, *J* = 2.3 Hz), 7.68 (1H, dd, *J* = 2.3, 8.6 Hz), 7.66 (1H, d, *J* = 8.6 Hz), 4.59 (1H, d, *J* = 8.4 Hz), 4.50 (1H, d, *J* = 7.8 Hz), 4.08-4.05 (2H, m), 4.02 (1H, dd, *J* = 1.9,

12.3 Hz), 3.94 (1H, d, *J* = 2.9 Hz), 3.93-3.86 (3H, m), 3.83-3.55 (11H, m), 3.50 (1H, dd, *J* = 1.1, 8.8 Hz), 3.42 (1H, dd, *J* = 2.3, 14.1 Hz), 3.25 (1H, ddd, *J* = 3.4, 5.6, 13.6 Hz), 3.20 (1H, ddd, *J* = 3.5, 9.6, 13.5 Hz), 3.13 (1H, dd, *J* = 7.3, 14.2 Hz), 2.75 (1H, dd, *J* = 4.6, 12.5 Hz), 2.06 (3H, s), 2.05 (3H, s), 1.77 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 175.1, 174.9, 173.7, 137.5, 134.3, 133.1, 132.9, 130.5, 129.3, 102.7, 101.0, 99.7, 78.3, 75.5, 75.3, 74.7, 72.7, 72.2, 70.4, 69.3, 69.1, 68.1, 67.3, 65.8, 61.1, 60.0, 54.9, 51.7, 45.2, 39.7, 39.4, 22.1, 22.0; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{33}H_{50}Cl_2N_4O_{20}S$ 925.2189, found 925.2191.



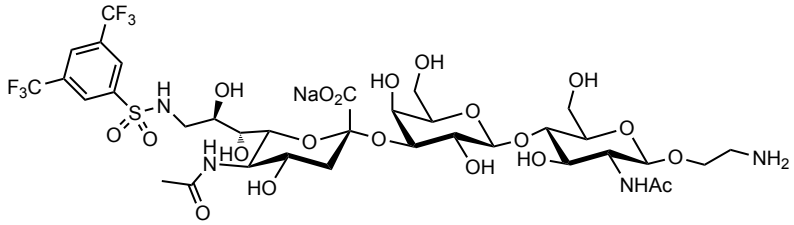
2-aminoethyl (5-acetamido-9-(3-chloro-4-methyl benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (30)

Prepared using general procedure **B**. R_f 0.15 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.94 (1H, d, *J* = 2.0 Hz), 7.73 (1H, dd, *J* = 2.0, 8.1 Hz), 7.57 (1H, d, *J* = 8.1 Hz), 4.58 (1H, d, *J* = 8.4 Hz), 4.50 (1H, d, *J* = 7.8 Hz), 4.09-4.05 (2H, m), 4.01 (1H, dd, *J* = 1.8, 12.3 Hz), 3.94 (1H, d, *J* = 2.6 Hz), 3.93-3.89 (2H, m), 3.86-3.80 (3H, m), 3.77-3.63 (8H, m), 3.57 (1H, dd, *J* = 7.8, 9.8 Hz), 3.52 (1H, dd, *J* = 1.8, 8.7 Hz), 3.32 (1H, dd, *J* = 2.5, 13.6 Hz), 3.25 (1H, ddd, *J* = 3.5, 5.6, 13.7 Hz), 3.20 (1H, ddd, *J* = 3.7, 7.6, 13.5 Hz), 3.03 (1H, dd, *J* = 7.3, 13.6 Hz), 2.76 (1H, dd, *J* = 4.5, 12.4 Hz), 2.48 (3H, s), 2.06 (3H, s), 2.03 (3H, s), 1.79 (1H, dd, *J* = 12.1, 12.1 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.4, 173.2, 142.1, 136.7, 134.4, 131.5, 126.6, 124.5, 102.1, 100.5, 99.4, 77.8, 75.1, 74.8, 74.3, 72.3, 71.7, 69.8, 68.8, 68.6, 67.7, 67.0, 65.2, 60.6, 59.6, 54.4, 51.2, 44.6, 39.2, 38.9, 21.7, 21.5, 19.0; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{34}H_{53}ClN_4O_{20}S$ 905.2735, found 905.2730.



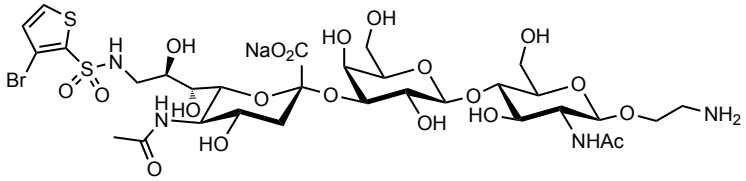
2-aminoethyl (5-acetamido-9-(3,4-di-methoxy benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (43)

Prepared using general procedure **B**. R_f 0.13 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.57 (1H, dd, *J* = 2.1, 8.5 Hz), 7.47 (1H, d, *J* = 2.0 Hz), 7.22 (1H, d, *J* = 8.6 Hz), 4.58 (1H, d, *J* = 8.4 Hz), 4.51 (1H, d, *J* = 7.8 Hz), 4.09 (1H, dd, *J* = 3.0, 9.8 Hz), 4.06 (1H, ddd, *J* = 3.4, 7.6, 11.2 Hz), 4.01 (1H, dd, *J* = 1.9, 12.3 Hz), 3.96 (3H, s), 3.95 (3H, s), 3.94-3.89 (3H, m), 3.87-3.79 (3H, m), 3.75-3.62 (8H, m), 3.57 (1H, dd, *J* = 8.0, 9.7 Hz), 3.52 (1H, dd, *J* = 1.3, 8.7 Hz), 3.29 (1H, dd, *J* = 2.6, 13.6 Hz), 3.25 (1H, ddd, *J* = 4.0, 6.1, 13.4 Hz), 3.20 (1H, ddd, *J* = 3.7, 7.5, 13.5 Hz), 2.99 (1H, dd, *J* = 7.4, 13.6 Hz), 2.76 (1H, dd, *J* = 4.6, 12.5 Hz), 2.06 (3H, s), 2.03 (3H, s), 1.79 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.4, 173.3, 151.8, 148.1, 129.6, 120.9, 111.0, 109.1, 102.1, 100.5, 99.4, 77.7, 75.1, 74.7, 74.2, 72.3, 71.7, 69.8, 68.8, 68.7, 67.7, 67.0, 65.2, 60.6, 59.5, 55.6, 55.5, 54.4, 51.2, 44.6, 39.2, 38.9, 21.7, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{35}H_{56}N_4O_{22}S$ 917.3180, found 917.3177.



2-aminoethyl (5-acetamido-9-(3,5-di-trifluoromethyl benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (50)

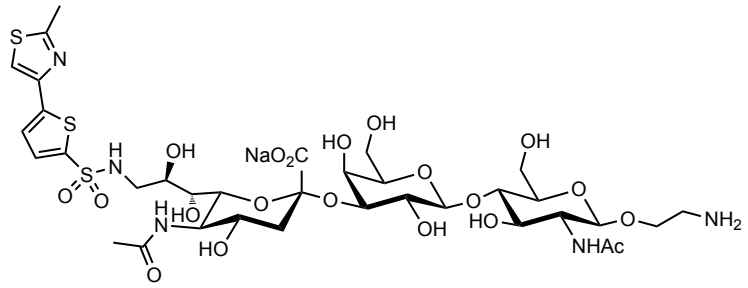
Prepared using general procedure **B**. R_f 0.24 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.49 (2H, s), 8.42 (1H, s), 4.58 (1H, d, *J* = 8.4 Hz), 4.54 (1H, d, *J* = 7.8 Hz), 4.09 (1H, dd, *J* = 3.0, 9.8 Hz), 4.05 (1H, ddd, *J* = 3.5, 7.3, 11.4 Hz), 4.02 (1H, dd, *J* = 1.7, 12.2 Hz), 3.95-3.93 (2H, m), 3.90-3.66 (10H, m), 3.64-3.60 (2H, m), 3.58 (1H, dd, *J* = 8.0, 9.6 Hz), 3.53 (1H, dd, *J* = 1.3, 8.9 Hz), 3.34 (1H, dd, *J* = 2.5, 13.6 Hz), 3.21 (1H, ddd, *J* = 4.0, 6.3, 14.0 Hz), 3.16 (1H, ddd, *J* = 3.6, 7.4, 13.5 Hz), 3.08 (1H, dd, *J* = 7.1, 13.7 Hz), 2.76 (1H, dd, *J* = 4.7, 12.5 Hz), 2.06 (3H, s), 2.03 (3H, s), 1.79 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.7, 174.4, 173.3, 140.8, 131.8, 131.6, 127.1, 126.9, 123.1, 121.3, 102.1, 100.5, 99.4, 77.6, 75.1, 74.8, 74.3, 72.3, 71.8, 69.8, 68.9, 68.6, 67.7, 67.0, 65.8, 60.6, 59.5, 54.4, 51.2, 44.7, 39.2, 39.0, 21.7, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{35}H_{50}F_6N_4O_{20}S$ 993.2716, found 993.2717.



2-aminoethyl (5-acetamido-9-(3-bromo thiophene-2-sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (54)

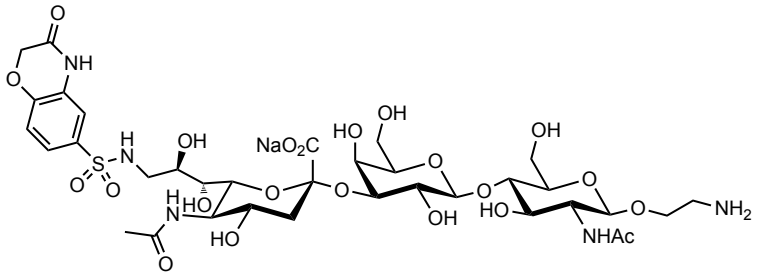
Prepared using general procedure **B**. R_f 0.16 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.82 (1H, d, *J* = 5.2 Hz), 7.30 (1H, d, *J* = 5.3 Hz), 4.59 (1H, d, *J* = 8.4 Hz), 4.54 (1H, d, *J* = 7.9 Hz), 4.09 (1H, dd, *J* = 3.1, 9.8 Hz), 4.07 (1H, ddd, *J* = 3.5, 7.6, 11.3 Hz), 4.03 (1H, dd, *J* = 2.3, 12.4 Hz), 3.95-3.87 (4H, m), 3.83-3.66 (8H, m), 3.64-3.57 (3H, m), 3.52 (1H, dd, *J* = 1.8, 8.7 Hz), 3.45 (1H, dd, *J* = 2.7, 14.0 Hz), 3.25 (1H, ddd, *J* = 4.6, 5.5, 13.7 Hz), 3.20 (1H, ddd, *J* = 3.9, 7.7, 13.4 Hz), 3.16 (1H, dd, *J* = 7.4, 14.0 Hz), 2.75 (1H, dd, *J* = 4.7, 12.5 Hz), 2.06 (3H, s), 2.05 (3H, s), 1.78 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.4, 173.3, 133.8, 132.5, 132.1, 113.2, 102.1, 100.5, 99.3, 77.7, 75.0, 74.8, 74.2, 72.3, 71.8, 69.8, 68.9, 68.7,

67.7, 66.9, 65.3, 60.6, 59.6, 54.4, 51.3, 44.8, 39.2, 39.0, 21.7, 21.6; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{31}H_{49}BrN_4O_{20}S_2$ 941.1638, found 941.1638.



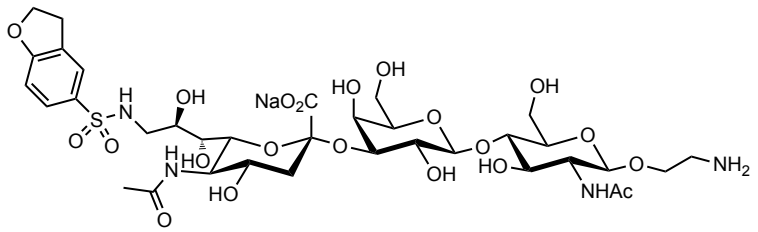
2-aminoethyl (5-acetamido-9-(5-(2-methyl-1,3-thiazol-4-yl) thiophene-2-sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (61)

Prepared using general procedure **B**. R_f 0.15 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.77 (1H, s), 7.71 (1H, d, *J* = 4.0 Hz), 7.51 (1H, d, *J* = 3.9 Hz), 4.56 (1H, d, *J* = 8.4 Hz), 4.44 (1H, d, *J* = 7.9 Hz), 4.07-4.03 (2H, m), 3.99 (1H, dd, *J* = 2.3, 12.3 Hz), 3.96 (1H, ddd, *J* = 2.7, 8.1, 13.1 Hz), 3.94 (1H, d, *J* = 3.2 Hz), 3.89 (1H, ddd, *J* = 3.8, 6.0, 12.0 Hz), 3.84-3.77 (3H, m), 3.72-3.55 (10H, m), 3.46 (1H, dd, *J* = 2.9, 13.8 Hz), 3.25-2.18 (2H, m), 3.15 (1H, dd, *J* = 7.2, 13.7 Hz), 2.75$_3$ (1H, dd, *J* = 4.7 Hz), 2.74$_7$ (3H, s), 2.05 (3H, s), 2.02 (3H, s), 1.78 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.4, 173.2, 169.3, 145.8, 143.6, 136.9, 133.2, 123.8, 115.5, 102.1, 100.5, 99.4, 77.8, 75.1, 74.7, 74.3, 72.3, 71.7, 70.0, 68.8, 68.7, 67.7, 66.9, 65.5, 60.5, 59.6, 54.4, 51.3, 44.9, 39.3, 39.0, 21.7, 21.5, 17.3; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{35}H_{53}N_5O_{20}S_3$ 960.2519, found 960.2516.



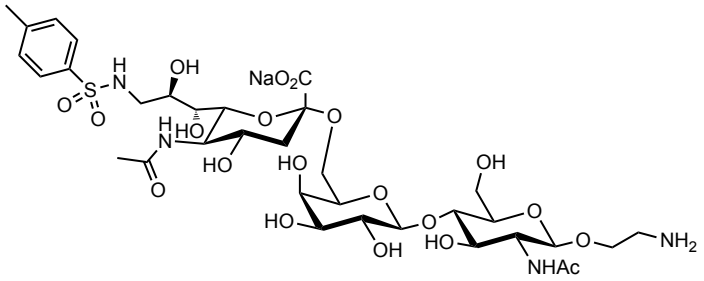
2-aminoethyl (5-acetamido-9-(3-oxo-3,4-dihydro-2H-1,4-benzoxazine-6-sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (65)

Prepared using general procedure **B**. R_f 0.16 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.57 (1H, dd, *J* = 2.2, 8.7 Hz), 7.45 (1H, d, *J* = 2.2 Hz), 7.21 (1H, d, *J* = 8.5 Hz), 4.70 (2H, m, HSQC correlation), 4.58 (1H, d, *J* = 8.4 Hz), 4.50 (1H, d, *J* = 7.8 Hz), 4.09 (1H, dd, *J* = 3.1, 10.0 Hz), 4.06 (1H, ddd, *J* = 3.4, 7.5, 11.2 Hz), 4.00 (1H, dd, *J* = 2.3, 12.4 Hz), 3.94-3.56 (15H, m), 3.52 (1H, dd, *J* = 1.7, 8.6 Hz), 3.30 (1H, dd, *J* = 2.8, 13.6 Hz), 3.23 (1H, ddd, *J* = 4.5, 9.4, 13.6 Hz), 3.19 (1H, ddd, *J* = 3.6, 7.5, 13.6 Hz), 3.02 (1H, dd, *J* = 7.4, 13.6 Hz), 2.76 (1H, dd, *J* = 4.7, 12.5 Hz), 2.06 (3H, s), 2.03 (3H, s), 1.78 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.4, 173.3, 166.7, 146.7, 131.7, 126.0, 123.0, 116.8, 114.5, 102.1, 100.5, 99.3, 77.7, 75.1, 74.8, 74.3, 72.3, 71.7, 69.8, 68.8, 68.7, 67.7, 66.9, 66.1, 65.4, 60.6, 59.6, 54.4, 51.3, 44.6, 39.2, 39.0, 21.7, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{35}H_{53}N_5O_{22}S$ 928.2976, found 928.2977.



2-aminoethyl (5-acetamido-9-(2,3-dihydrobenzo[b] furan-5-sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (76)

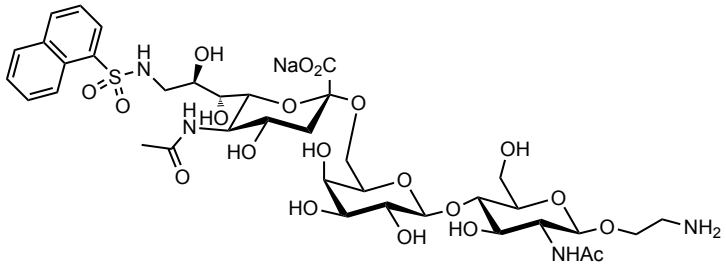
Prepared using general procedure **B**. R_f 0.13 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.78 (1H, d, *J* = 2.1 Hz), 7.71 (1H, dd, *J* = 2.1, 8.6 Hz), 6.99 (1H, d, *J* = 8.5 Hz), 4.73 (2H, dd, *J* = 8.8, 8.8 Hz), 4.58 (1H, d, *J* = 8.4 Hz), 4.52 (1H, d, *J* = 7.9 Hz), 4.09 (1H, dd, *J* = 3.1, 10.0 Hz), 4.06 (1H, ddd, *J* = 3.5, 7.6, 11.6 Hz), 4.01 (1H, dd, *J* = 2.3, 12.4 Hz), 3.95-3.62 (14H, m), 3.58 (1H, dd, *J* = 7.8, 9.8 Hz), 3.52 (1H, dd, *J* = 1.8, 8.7 Hz), 3.33 (2H, dd, *J* = 8.8, 8.8 Hz), 3.27 (1H, dd, *J* = 2.9, 11.1 Hz), 3.25 (1H, ddd, *J* = 3.5, 5.9, 13.6 Hz), 3.20 (1H, ddd, *J* = 3.6, 7.5, 13.6 Hz), 2.97 (1H, dd, *J* = 7.4, 13.5 Hz), 2.76 (1H, dd, *J* = 4.7, 12.5 Hz), 2.06 (3H, s), 2.03 (3H, s), 1.79 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.4, 173.3, 163.0, 129.3, 129.2, 127.8, 123.8, 109.1, 102.1, 100.5, 99.3, 77.7, 75.1, 74.8, 74.2, 72.4, 72.3, 71.7, 69.8, 68.9, 68.7, 67.7, 67.0, 65.3, 60.6, 59.5, 54.4, 51.2, 44.6, 39.2, 39.0, 28.0, 21.7, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{35}H_{54}N_4O_{21}S$ 899.3074, found 899.3080.



2-aminoethyl (5-acetamido-9-(4-methyl benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (82)

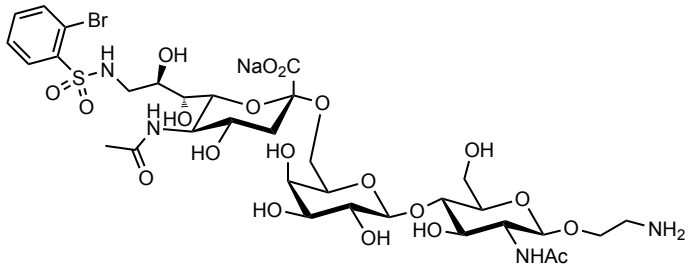
Prepared using general procedure **B**. R_f 0.13 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.79 (2H, d, *J* = 8.3 Hz), 7.49 (2H, d, *J* = 8.2 Hz), 4.61 (1H, d, *J* = 7.9 Hz), 4.46 (1H, d, *J* = 7.9 Hz), 4.06 (1H, ddd, *J* = 3.6, 7.2, 11.1 Hz), 4.01 (1H, dd, *J* = 1.6, 12.4 Hz), 3.96 (1H, dd, *J* = 9.7 Hz), 3.93 (1H, d, *J* = 3.6 Hz), 3.90-3.62 (12H, m), 3.58-3.53 (2H, m), 3.46 (1H, dd, *J* = 1.4, 9.0 Hz), 3.26 (1H, dd, *J* = 2.6, 13.5 Hz), 3.20 (1H, ddd, *J* = 3.6, 5.9, 13.7 Hz), 3.16 (1H, ddd, *J* = 3.8, 7.4, 13.5 Hz), 3.01 (1H, dd, *J* = 7.1, 13.5 Hz), 2.66 (1H, dd, *J* = 4.6, 12.4 Hz), 2.46 (3H, s), 2.06 (3H, s), 2.03 (3H, s), 1.69 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150

MHz) δ 174.5, 174.4, 173.0, 144.5, 134.5, 129.6, 126.3, 102.9, 100.3, 99.7, 80.0, 74.0, 73.2, 72.0, 71.9, 71.8, 70.2, 69.4, 68.8, 67.9, 67.7, 65.8, 62.9, 59.8, 54.3, 51.4, 44.7, 39.6, 39.0, 21.8, 21.5, 20.2; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{34}H_{54}N_4O_{20}S$ 871.3125, found 871.3126.



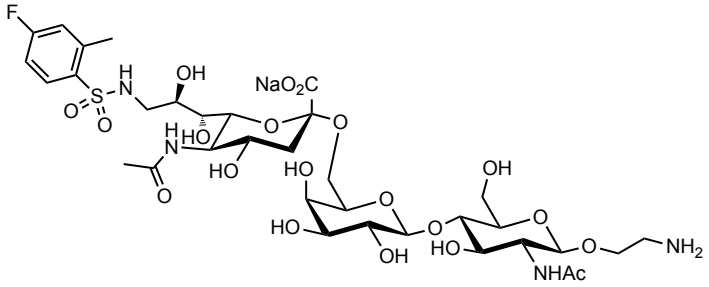
2-aminoethyl (5-acetamido-9-(1-naphthyl sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (89)

Prepared using general procedure **B**. R_f 0.14 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.62 (1H, d, *J* = 8.6 Hz), 8.29-8.27 (2H, m), 8.14 (1H, d, *J* = 8.2 Hz), 7.81 (1H, dd, *J* = 7.3, 7.7 Hz), 7.74 (1H, dd, *J* = 7.5, 7.5 Hz), 7.70 (1H, dd, *J* = 7.8, 7.9 Hz), 4.57 (1H, d, *J* = 8.4 Hz), 4.45 (1H, d, *J* = 7.9 Hz), 4.05 (1H, ddd, *J* = 3.5, 7.3, 11.4 Hz), 4.00 (1H, dd, *J* = 2.3, 12.4 Hz), 3.93-3.51 (16H, m), 3.40 (1H, dd, *J* = 1.7, 9.0 Hz), 3.32 (1H, dd, *J* = 2.8, 13.9 Hz), 3.21 (1H, ddd, *J* = 3.7, 6.1, 13.8 Hz), 3.17 (1H, ddd, *J* = 3.7, 7.4, 13.6 Hz), 3.05 (1H, dd, *J* = 7.1, 13.9 Hz), 2.64 (1H, dd, *J* = 4.7, 12.5 Hz), 2.03 (3H, s), 1.98 (3H, s), 1.66 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.4_4, 174.4_1, 172.9, 134.5, 133.7, 132.7, 129.0_4, 128.9_5, 128.2, 126.9, 126.8, 124.0, 123.1, 102.9, 100.3, 99.7, 80.0, 74.0, 73.2, 71.9_4, 71.8_9, 71.7, 70.2, 69.5, 68.9, 68.0, 67.6, 65.5, 63.0, 59.7, 54.2, 51.4, 44.7, 39.6, 39.0, 21.8, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{37}H_{54}N_4O_{20}S$ 907.3125, found 907.3128.



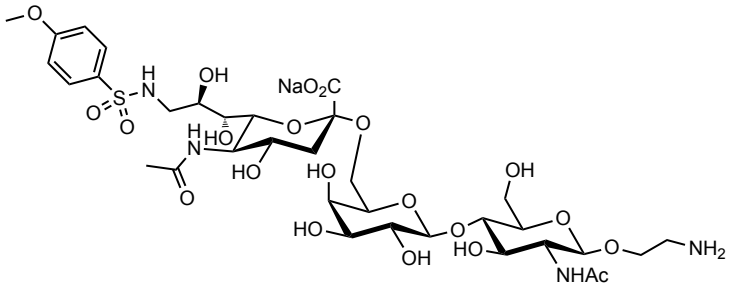
2-aminoethyl (5-acetamido-9-(2-bromo benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (97)

Prepared using general procedure **B**. R_f 0.14 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.12 (1H, dd, *J* = 1.8, 7.6 Hz), 7.93 (1H, dd, *J* = 1.0, 7.5 Hz), 7.62-7.57 (2H, m), 4.62 (1H, d, *J* = 7.9 Hz), 4.47 (1H, d, *J* = 7.9 Hz), 4.07 (1H, ddd, *J* = 3.4, 7.3, 11.6 Hz), 4.01 (1H, dd, *J* = 1.8, 12.3 Hz), 3.95-3.53 (16H, m), 3.42-3.38 (2H, m), 3.25 (1H, ddd, *J* = 3.6, 7.5, 13.5 Hz), 3.21 (1H, ddd, *J* = 3.6, 7.5, 13.5 Hz), 3.10 (1H, dd, *J* = 7.3, 14.2 Hz), 2.65 (1H, dd, *J* = 4.6, 12.4 Hz), 2.05 (3H, s), 2.04 (3H, s), 1.66 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.4, 172.9, 136.9, 135.0, 134.2, 130.7, 127.7, 118.6, 102.9, 100.3, 99.7, 79.8, 74.0, 73.2, 72.0, 71.9, 71.7, 70.3, 69.5, 68.9, 67.9, 67.6, 65.1, 62.9, 59.7, 54.3, 51.4, 44.8, 39.7, 38.9, 21.8, 21.6; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{33}H_{51}BrN_4O_{20}S$ 935.2073, found 935.2068.



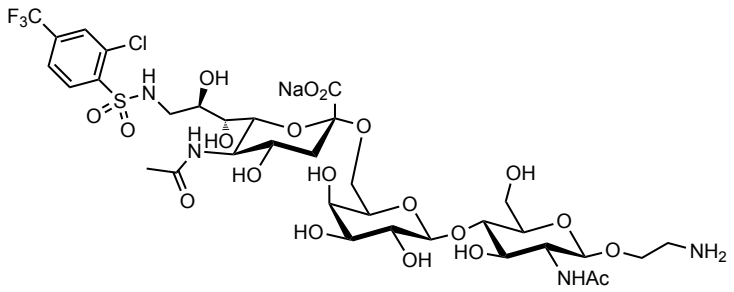
2-aminoethyl (5-acetamido-9-(4-fluoro-2-methyl benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (106)

Prepared using general procedure **B**. R_f 0.14 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.00 (1H, dd, *J* = 5.6, 8.9 Hz), 7.27 (1H, dd, *J* = 2.7, 9.7 Hz), 7.17 (1H, ddd, *J* = 2.7, 8.5, 8.5 Hz), 4.61 (1H, d, *J* = 7.5 Hz); 4.47 (1H, d, *J* = 7.9 Hz), 4.07 (1H, ddd, *J* = 3.5, 7.3, 11.6 Hz), 4.01 (1H, dd, *J* = 2.2, 12.4 Hz), 3.95-3.61 (14H, m), 3.59 (1H, dd, *J* = 8.0, 9.7 Hz), 3.54 (1H, dd, *J* = 3.5, 10.1 Hz), 3.39 (1H, dd, *J* = 1.0, 9.0 Hz), 3.33 (1H, dd, *J* = 2.4, 14.0 Hz), 3.24 (1H, ddd, *J* = 3.5, 6.0, 13.7 Hz), 3.19 (1H, ddd, *J* = 3.6, 7.5, 13.7 Hz), 3.03 (1H, dd, *J* = 7.5, 14.0 Hz), 2.66 (1H, dd, *J* = 4.6, 12.7 Hz), 2.64 (3H, s), 2.06 (3H, s), 2.03 (3H, s), 1.67 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.4, 172.9, 164.5 ($J_{C,F}$ = 252.3 Hz), 140.4 ($J_{C,F}$ = 9.4 Hz), 132.1, 131.6 ($J_{C,F}$ = 10.3 Hz), 119.1 ($J_{C,F}$ = 22.6 Hz), 112.8 ($J_{C,F}$ = 22.2 Hz), 102.9, 100.3, 99.7, 79.8, 74.0, 73.2, 71.9$_4$, 71.9$_0$, 71.7, 70.3, 69.4, 68.9, 67.9, 67.6, 65.3, 62.9, 59.7, 54.3, 51.4, 44.5, 39.7, 39.0, 21.8, 21.5, 18.9; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{34}H_{53}FN_4O_{20}S$ 889.3031, found 889.3032.



2-aminoethyl (5-acetamido-9-(4-methoxy benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (110)

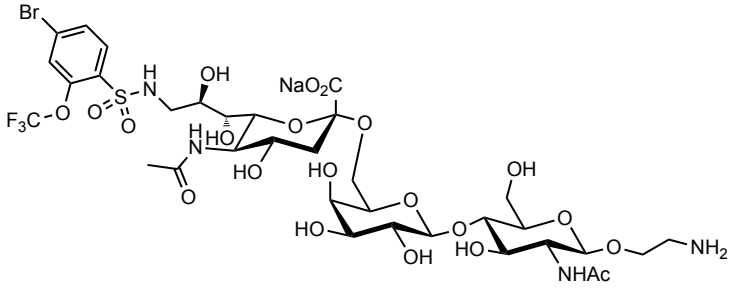
Prepared using general procedure **B**. R_f 0.13 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.87 (2H, d, *J* = 8.9 Hz), 7.19 (2H, d, *J* = 8.9 Hz), 4.62 (1H, d, *J* = 7.8 Hz), 4.46 (1H, d, *J* = 7.9 Hz), 4.07 (1H, ddd, *J* = 3.5, 7.3, 11.6 Hz), 4.01 (1H, dd, *J* = 2.2, 12.4 Hz), 3.97-3.75 (9H, m), 3.93 (3H, s), 3.69-3.62 (5H, m), 3.56 (1H, dd, *J* = 7.9, 10.0 Hz), 3.54 (1H, dd, *J* = 3.6, 10.2 Hz), 3.46 (1H, dd, *J* = 1.8, 8.9 Hz), 3.28-3.22 (2H, m), 3.19 (1H, ddd, *J* = 3.6, 7.3, 13.6 Hz), 3.00 (1H, dd, *J* = 7.2, 13.5 Hz), 2.66 (1H, dd, *J* = 4.7, 12.5 Hz), 2.06 (3H, s), 2.03 (3H, s), 1.69 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.4, 173.0, 129.4, 128.6, 114.3, 102.9, 100.3, 99.7, 80.0, 74.0, 73.2, 72.0, 71.9, 71.8, 70.2, 69.4, 68.9, 67.9, 67.7, 65.3, 62.9, 59.7, 55.3, 54.2, 53.9, 51.4, 44.7, 39.7, 39.0, 21.8, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* [M + H]$^+$ expected for $C_{34}H_{54}N_4O_{21}S$ 887.3074, found 887.3072.



2-aminoethyl (5-acetamido-9-(2-chloro-4-trifluoromethyl benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (126)

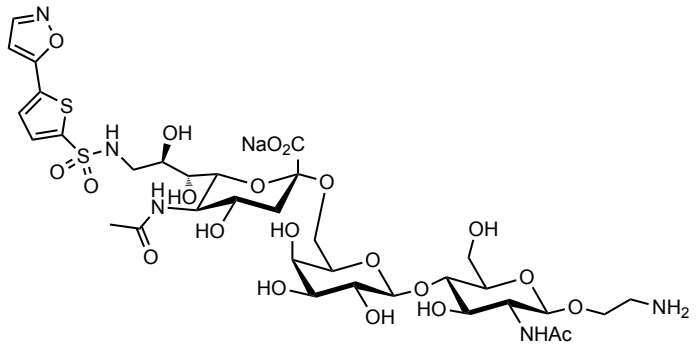
Prepared using general procedure **B**. R_f 0.20 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.24 (1H, d, *J* = 8.3 Hz), 8.08 (1H, s), 7.87 (1H, d, *J* = 8.1 Hz), 4.61 (1H, d, *J* = 8.0 Hz), 4.47 (1H, d, *J* = 7.9 Hz), 4.06 (1H, ddd, *J* = 3.5, 7.3, 11.4 Hz), 4.01 (1H, dd, *J* = 1.8, 12.2 Hz), 3.95-3.56 (15H, m), 3.52 (1H, dd, *J* = 3.6, 10.1 Hz), 3.43-3.40 (2H, m), 3.23 (1H, ddd, *J* = 3.5, 6.0, 13.7 Hz), 3.19 (1H, ddd, *J* = 3.7, 7.5, 13.6 Hz), 3.14 (1H, dd, *J* = 7.3, 14.3 Hz), 2.65 (1H, dd, *J* = 4.8, 12.5 Hz), 2.05 (3H, s), 2.04 (3H, s), 1.66 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.6, 174.4, 172.8, 139.4, 134.6 (J_{CF} = 33.5 Hz), 131.2 (J_{CF} = 23.1 Hz), 128.5, 124.1, 123.1, 121.3, 102.9, 100.3, 99.7, 79.8, 74.0, 73.2, 72.0, 71.9, 71.7, 70.3, 69.7, 68.8, 67.9,

67.6, 65.3, 62.8, 59.7, 54.2, 51.4, 44.9, 39.7, 39.0, 21.8, 21.6; **HRMS** (ESI-TOF high acc.) *m/z* [M + H]$^+$ expected for $C_{34}H_{50}ClF_3N_4O_{20}S$ 959.2452, found 959.2454.



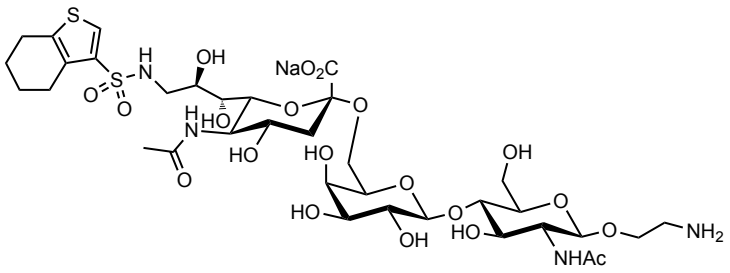
2-aminoethyl (5-acetamido-9-(4-bromo-2-trifluoromethoxy benzene sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (129)

Prepared using general procedure **B**. R_f 0.18 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.91 (1H, d, *J* = 8.5 Hz), 7.87 (1H, s), 7.76 (1H, dd, *J* = 1.7, 8.5 Hz), 4.62 (1H, d, *J* = 7.8 Hz), 4.46 (1H, d, *J* = 7.9 Hz), 4.06 (1H, ddd, *J* = 3.5, 7.3, 11.6 Hz), 4.01 (1H, dd, *J* = 2.1, 13.2 Hz), 3.96-3.62 (14H, m), 3.57 (1H, dd, *J* = 7.9, 10.0 Hz), 3.54 (1H, dd, *J* = 3.4, 10.3 Hz), 3.43-3.39 (2H, m), 3.23 (1H, ddd, *J* = 3.5, 5.7, 13.7 Hz), 3.18 (1H, ddd, *J* = 3.7, 7.4, 13.7 Hz), 3.13 (1H, dd, *J* = 7.3, 14.1 Hz), 2.65 (1H, dd, *J* = 4.7, 12.5 Hz), 2.06 (3H, s), 2.03 (3H, s), 1.68 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.4, 172.9, 145.2, 131.0, 129.9, 129.6, 127.9, 123.5, 102.9, 100.3, 99.7, 80.0, 74.0, 73.3, 72.0, 71.9, 71.8, 70.2, 69.5, 68.9, 68.0, 67.6, 65.6, 63.0, 59.8, 54.2, 51.4, 44.9, 39.7, 39.0, 21.8, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* [M + H]$^+$ expected for $C_{34}H_{50}BrF_3N_4O_{21}S$ 1019.1896, found 1019.1891.



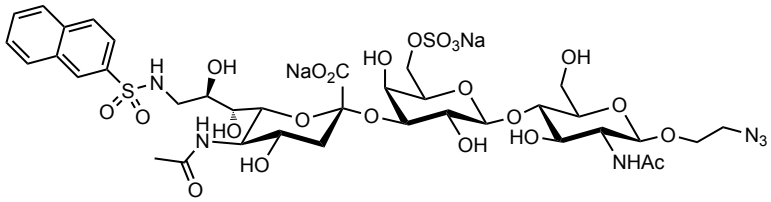
2-aminoethyl (5-acetamido-9-(1,3-benzothiazole-6-sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (150)

Prepared using general procedure **B**. R_f 0.08 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 9.51 (1H, s), 8.71 (1H, s), 8.31 (1H, d, *J* = 8.7 Hz), 8.06 (1H, dd, *J* = 1.7, 8.7 Hz), 4.59 (1H, d, *J* = 8.2 Hz), 4.45 (1H, d, *J* = 7.9 Hz), 4.05 (1H, ddd, *J* = 3.5, 7.2, 11.6 Hz), 4.00 (1H, dd, *J* = 1.8, 12.4 Hz), 3.94-3.61 (14H, m), 3.56 (1H, dd, *J* = 8.0, 9.5 Hz), 3.52 (1H, dd, *J* = 3.3, 10.2 Hz), 3.45 (1H, d, *J* = 9.0 Hz), 3.32 (1H, dd, *J* = 2.5, 13.6 Hz), 3.27 (1H, ddd, *J* = 3.5, 5.7, 13.6 Hz), 3.18 (1H, ddd, *J* = 3.7, 7.4, 13.6 Hz), 3.08 (1H, dd, *J* = 7.2, 13.6 Hz), 2.65 (1H, dd, *J* = 4.7, 12.4 Hz), 2.04 (3H, s), 2.01 (3H, s), 1.67 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.4, 172.9, 161.0, 154.2, 135.2, 133.7, 123.9, 123.1, 122.1, 102.9, 100.3, 99.7, 80.0, 74.0, 73.2, 72.0, 71.9, 71.8, 70.2, 69.5, 68.9, 67.9, 67.6, 65.5, 62.9, 59.7, 54.2, 51.4, 44.8, 39.6, 39.0, 21.8, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{34}H_{51}N_5O_{20}S_2$ 914.2641, found 914.2641.



2-aminoethyl (5-acetamido-9-(4,5,6,7-tetrahydro benzo[b] thiophene-2-sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→6)-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (155)

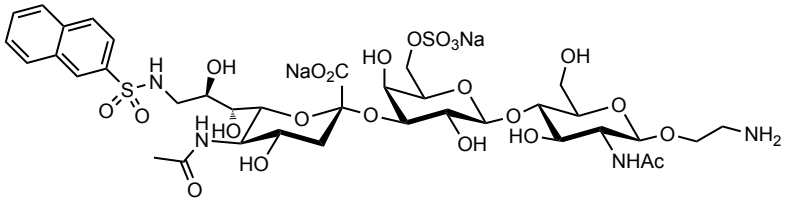
Prepared using general procedure **B**. R_f 0.14 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 7.35 (1H, s), 4.53 (1H, d, *J* = 7.4 Hz), 4.37 (1H, d, *J* = 7.9 Hz), 3.98 (1H, ddd, *J* = 3.1, 6.9, 11.6 Hz), 3.93-3.41 (19H, m), 3.23 (1H, d, *J* = 11.4 Hz), 3.16-31.0 (2H, m), 2.98 (1H, dd, *J* = 7.0, 13.4 Hz), 2.74-2.73 (2H, m), 2.58 (1H, dd, *J* = 4.5, 12.5 Hz), 2.57-2.55 (2H, m), 1.98 (3H, s), 1.95 (3H, s), 1.77-1.75 (2H, m), 1.71-1.70 (2H, m), 1.61 (1H, dd, *J* = 12.1, 12.1 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.4, 173.0, 144.8, 136.3, 133.5, 132.8, 102.9, 100.3, 99.7, 80.0, 74.0, 73.2, 72.0, 71.8, 70.2, 69.4, 68.9, 67.9, 67.7, 65.4, 62.9, 59.8, 54.2, 51.4, 44.9, 39.6, 39.0, 24.4, 24.3, 22.1, 21.8, 21.6, 21.4; **HRMS** (ESI-TOF high acc.) *m/z* $[M + H]^+$ expected for $C_{35}H_{56}N_4O_{20}S_2$ 917.3002, found 917.3011.



2-azidoethyl (5-acetamido-9-(2-naphthyl sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-6-*O*-sulfo-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (157)

Compound **F** (18.8 mg, 21.7 μmol) was dissolved in anhydrous methanol (1 ml) containing *N*,*N*-diisopropylethylamine (18.9 μl, 109 μmol, 5 eq.). 2-Naphthyl sulfonyl chloride (9.8 mg, 43.4 μmol, 2 eq.) was then added and the reaction was kept at room temperature (22 °C) for 2 hours.

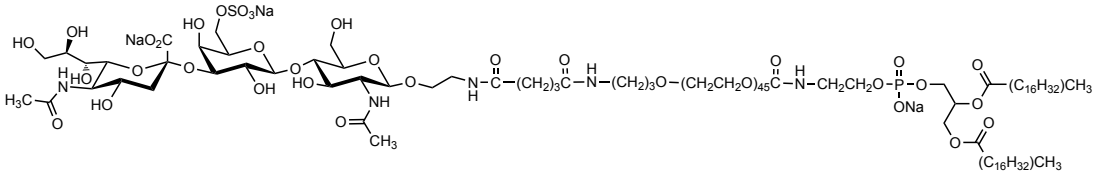
The reaction was monitored by TLC (EtOAc-CH_3OH-AcOH-H_2O, 6:3:3:2). The completed reaction was then concentrated under reduced pressure. The residue was dissolved in deionized H_2O (1 ml) and passed through Dowex® resin (50WX8, Na^+ form). The fractions containing product were combined then passed through an equilibrated Waters C-18 Sep-Pak® cartridge (500 mg). The column was washed with deionized H_2O then the product was eluted with 30-50% methanol-H_2O solutions. Fractions containing product were combined and the sample lyophilized to give a white amorphous solid (18.3 mg, 80%). R_f 0.58 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.59 (1H, s), 8.20 (1H, d, *J* = 8.8 Hz), 8.17 (1H, d, *J* = 8.0 Hz), 8.11 (1H, d, *J* = 8.1 Hz), 7.94 (1H, dd, *J* = 1.4, 8.6 Hz), 7.80 (1H, dd, *J* = 6.9, 8.0 Hz), 7.77 (1H, dd, *J* = 7.3, 7.4 Hz), 4.56 (1H, d, *J* = 8.5 Hz), 4.15-4.02 (4H, m), 3.94-3.75 (7H, m), 3.70-3.61 (5H, m), 3.56-3.42 (6H, m), 3.28 (1H, dd, *J* = 9.1, 9.3 Hz), 3.14 (1H, dd, *J* = 7.8, 13.9 Hz), 2.74 (1H, dd, *J* = 4.5, 12.5 Hz), 2.05 (3H, s), 2.02 (3H, s), 1.75 (1H, dd, *J* = 12.1, 12.2 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.2, 173.1, 135.6, 134.2, 131.4, 129.5, 129.1, 128.8, 127.7, 127.6, 127.5, 121.2, 102.1, 100.5, 99.3, 79.2, 74.8, 74.2, 72.3, 72.2, 71.5, 70.2, 68.8, 68.5, 68.2, 67.7, 66.7, 66.6, 61.1, 59.9, 54.4, 51.3, 49.9, 44.6, 39.3, 21.8, 21.6.



2-aminoethyl (5-acetamido-9-(2-naphthyl sulfonamido)-3,5,9-trideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonic acid)-(2→3)-6-*O*-sulfo-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranoside) (158)

Compound **157** (10 mg, 9.5 μmol) was dissolved in deionized H_2O (1 ml) with pH adjusted to 9 with 1 mM NaOH (aq.). Trimethyl phosphine (5 eq., 1 M in THF) was added then the reaction was mixed at room temperature for ~20 hours. The reaction was monitored by TLC (EtOAc-CH_3OH-AcOH-H_2O, 6:3:3:2). The completed reactions were then concentrated under reduced pressure to

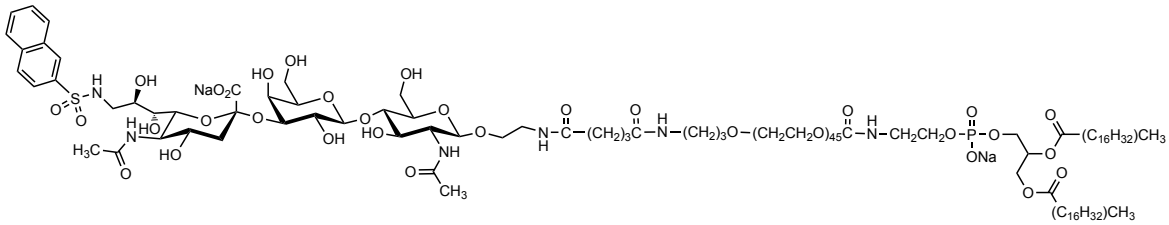
removed residual THF and excess trimethylphosphine. The remaining aqueous solution was then lyophilized to a white amorphous solid. The residue was dissolved in H_2O then passed through an equilibrated Waters C-18 Sep-Pak® cartridge (500 mg). The column was washed with deionized H_2O then the product was eluted with 30-50% methanol-H_2O solutions. Fractions containing product were combined and the sample lyophilized to a white amorphous solid (9.3 mg, 95%). R_f 0.15 (EtOAc-MeOH-AcOH-H_2O, 6:3:3:2); **^{1}H NMR** (D_2O, 600 MHz) δ 8.58 (1H, s), 8.19 (1H, d, J = 8.8 Hz), 8.16 (1H, d, J = 8.0 Hz), 8.10 (1H, d, J = 8.0 Hz), 7.94 (1H, dd, J = 1.7, 8.7 Hz), 7.79 (1H, d, J = 6.9, 8.0 Hz), 7.76 (1H, d, J = 6.9, 7.9 Hz), 4.53 (1H, d, J = 8.4 Hz), 4.14-4.09 (3H, m), 4.02 (1H, ddd, J = 3.6, 7.1, 11.0 Hz), 3.95-3.88 (4H, m), 3.86-3.77 (3H, m), 3.72-3.65 (4H, m), 3.62 (1H, dd, J = 4.3, 7.7 Hz), 3.59-3.54 (2H, m), 3.49 (1H, dd, J = 8.1, 9.5 Hz), 3.45 (1H, dd, J = 2.7, 13.9 Hz), 3.35 (1H, dd, J = 9.1, 9.3 Hz), 3.17-3.09 (3H, m), 2.75 (1H, dd, J = 4.6, 12.5 Hz), 2.05 (3H, s), 2.02 (3H, s), 1.75 (1H, dd, J = 12.1, 12.3 Hz); **^{13}C NMR** (D_2O, 150 MHz) δ 174.5, 174.2, 173.1, 135.6, 134.2, 131.4, 129.5, 129.0, 128.8, 127.7, 127.6, 127.5, 121.2, 102.1, 100.8, 99.3, 79.1, 74.8, 74.1, 72.3, 72.2, 71.4, 70.2, 69.5, 68.8, 68.5, 67.7, 66.7, 66.6, 59.9, 54.5, 51.3, 44.7, 39.4, 39.3, 21.7, 21.5; **HRMS** (ESI-TOF high acc.) *m/z* $[M + Na]^+$ expected for $C_{37}H_{54}N_4O_{23}S_2$ 1009.2512, found 1009.2503.



Synthesis of glyco-pegylated lipid conjugate (159)

Compound **C2^{17}** (6.4 mg, 7.4 μmol, 2.0 eq.) and DSPE-PEG-NHS (11.3 mg, 3.7 μmol, 1 eq.) were dissolved in a mixture of anhydrous CH_2Cl_2 (0.25 ml) and anhydrous DMSO (0.25 ml) in a flame dried pear shape flask (5 ml). *N*,*N*-diisopropylethylamine (1.3 μl, 7.4 μmol, 2 eq.) was then added and the reaction was mixed at room temperature (22 °C) under an inert N_2 (g) atmosphere. The reaction was monitored by TLC ($CHCl_3$-MeOH-H_2O, 80:18:2) using an Iodine/silica chamber for staining. After 20 hours, the reaction appeared complete by TLC and was concentrated under

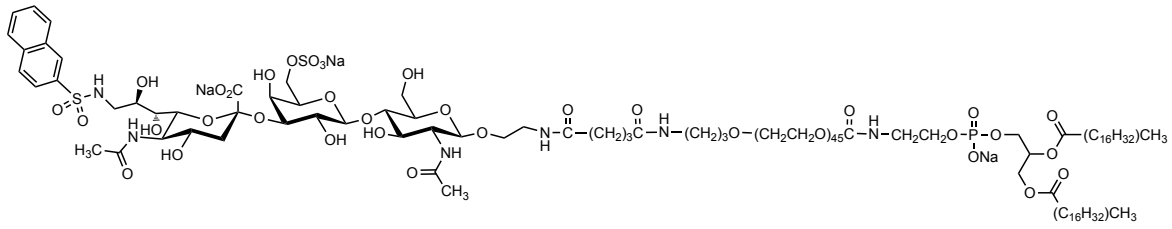
reduced pressure to remove CH_2Cl_2. The remaining DMSO solution was diluted with H_2O then lyophilized. The residue was then dissolved in deionized H_2O (10 ml) and dialyzed against deionized H_2O (3 x 3 l) using a Thermo Scientific Slide-A-Lyzer dialysis cassette (10,000 MWCO). The aqueous solution was removed from the cassette then lyophilized to provide **159** as a white amorphous solid (13.6 mg, 97%, ~78% coupling efficiency). The coupling efficiency was estimated by ^{1}H-NMR analysis. The product was used directly for the formulation of liposomes. **^{1}H NMR** (DMSO, 600 MHz) δ 1.87-1.84 ($6H_{expected}$, $4.7H_{observed}$, m, NHAc *CH_3* x2), 0.86 (6H, m, DSPE CH_2*CH_3* x2).



Synthesis of glyco-pegylated lipid conjugate (160)

Compound **12** (6.5 mg, 7.0 μmol, 1.5 eq.) and DSPE-PEG-NHS (14.3 mg, 4.7 μmol, 1 eq.) were dissolved in a mixture of anhydrous CH_2Cl_2 (0.5 ml) and anhydrous DMSO (0.5 ml) in a flame dried pear shape flask (5 ml). *N*,*N*-diisopropylethylamine (4.1 μl, 23 μmol, 5 eq.) was then added and the reaction was mixed at room temperature (22 °C) under an inert N_2 (g) atmosphere. The reaction was monitored by TLC ($CHCl_3$-MeOH-H_2O, 80:18:2) using an Iodine/silica chamber for staining. After 20 hours, the reaction appeared complete by TLC and was concentrated under reduced pressure to remove CH_2Cl_2. The remaining DMSO solution was diluted with H_2O then lyophilized. The residue was then dissolved in deionized H_2O (10 ml) and dialyzed against deionized H_2O (3 x 3 l) using a Thermo Scientific Slide-A-Lyzer dialysis cassette (10,000 MWCO). The aqueous solution was removed from the cassette then lyophilized to provide **159** as a white amorphous solid (18 mg, 99%, ~98% coupling efficiency). The coupling efficiency was estimated by ^{1}H-NMR analysis. The product was used directly for the formulation of liposomes.

^{1}H NMR (DMSO, 600 MHz) δ 8.43 ($1H_{expected}$, $0.98H_{observed}$, s, 2-naphthyl $Ar_{H\text{-}1}$), 0.85 (6H, m, DSPE CH_2CH_3 x2).



Synthesis of glyco-pegylated lipid conjugate (161)

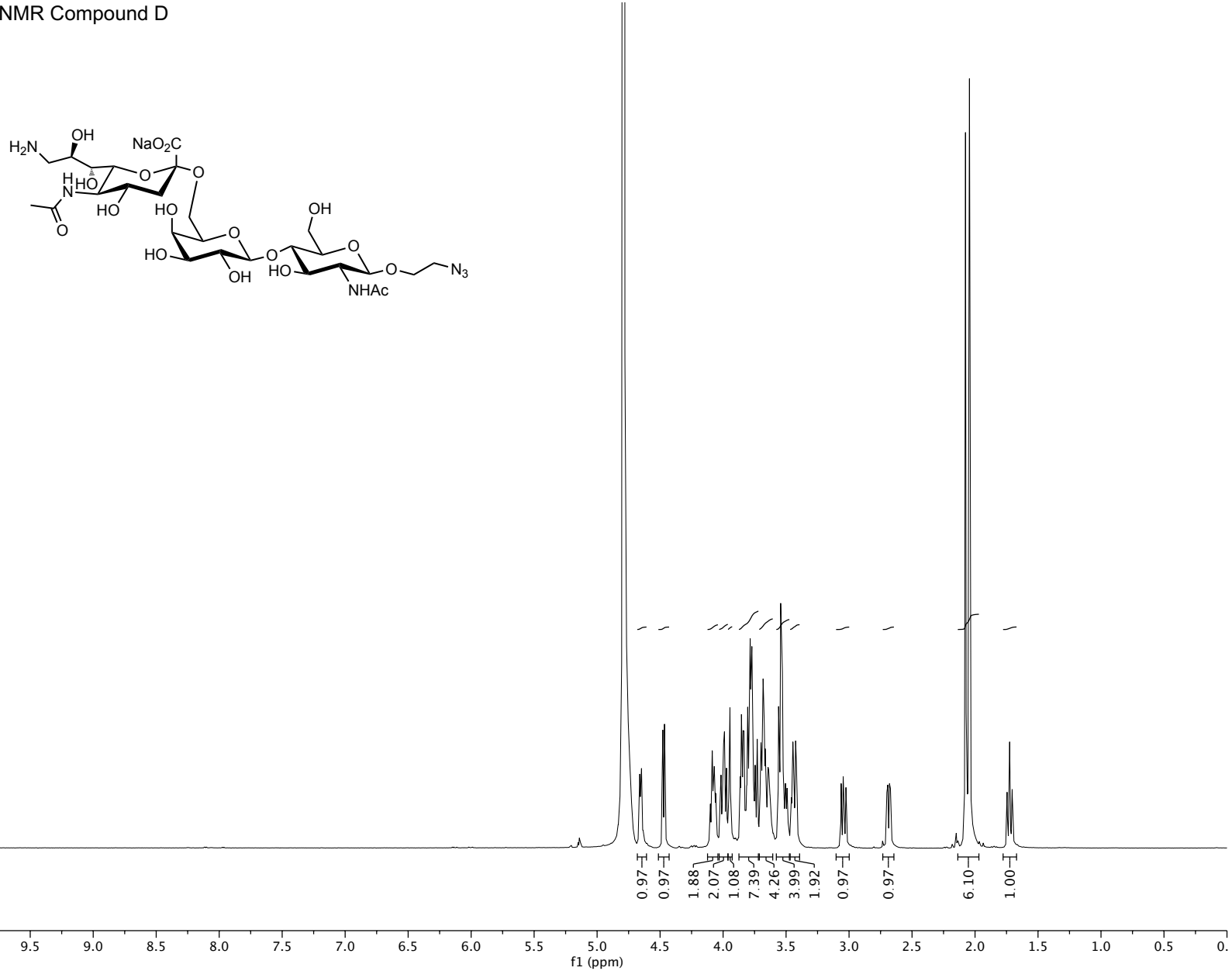
Compound **158** (7.5 mg, 7.3 μmol, 1.5 eq.) and DSPE-PEG-NHS (14.9 mg, 4.9 μmol, 1 eq.) were dissolved in a mixture of anhydrous CH_2Cl_2 (0.5 ml) and anhydrous DMSO (0.5 ml) in a flame dried pear shape flask (5 ml). *N*,*N*-diisopropylethylamine (1.7 μl, 9.7 μmol, 2 eq.) was then added and the reaction was mixed at room temperature (22 °C) under an inert N_2 (g) atmosphere. The reaction was monitored by TLC ($CHCl_3$-MeOH-H_2O, 80:18:2) using an Iodine/silica chamber for staining. After 20 hours, the reaction appeared complete by TLC and was concentrated under reduced pressure to remove CH_2Cl_2. The remaining DMSO solution was diluted with H_2O then lyophilized. The residue was then dissolved in deionized H_2O (10 ml) and dialyzed against deionized H_2O (3 x 3 l) using a Thermo Scientific Slide-A-Lyzer dialysis cassette (10,000 MWCO). The aqueous solution was removed from the cassette then lyophilized to provide **160** as a white amorphous solid (19.2 mg, 99%, ~99% coupling efficiency). The coupling efficiency was estimated by ^{1}H-NMR analysis. The product was used directly for the formulation of liposomes.
^{1}H NMR (DMSO, 600 MHz) δ 8.44 ($1H_{expected}$, $0.99H_{observed}$, s, 2-naphthyl $Ar_{H\text{-}1}$), 0.86 (6H, m, DSPE CH_2CH_3 x2).

H. References.

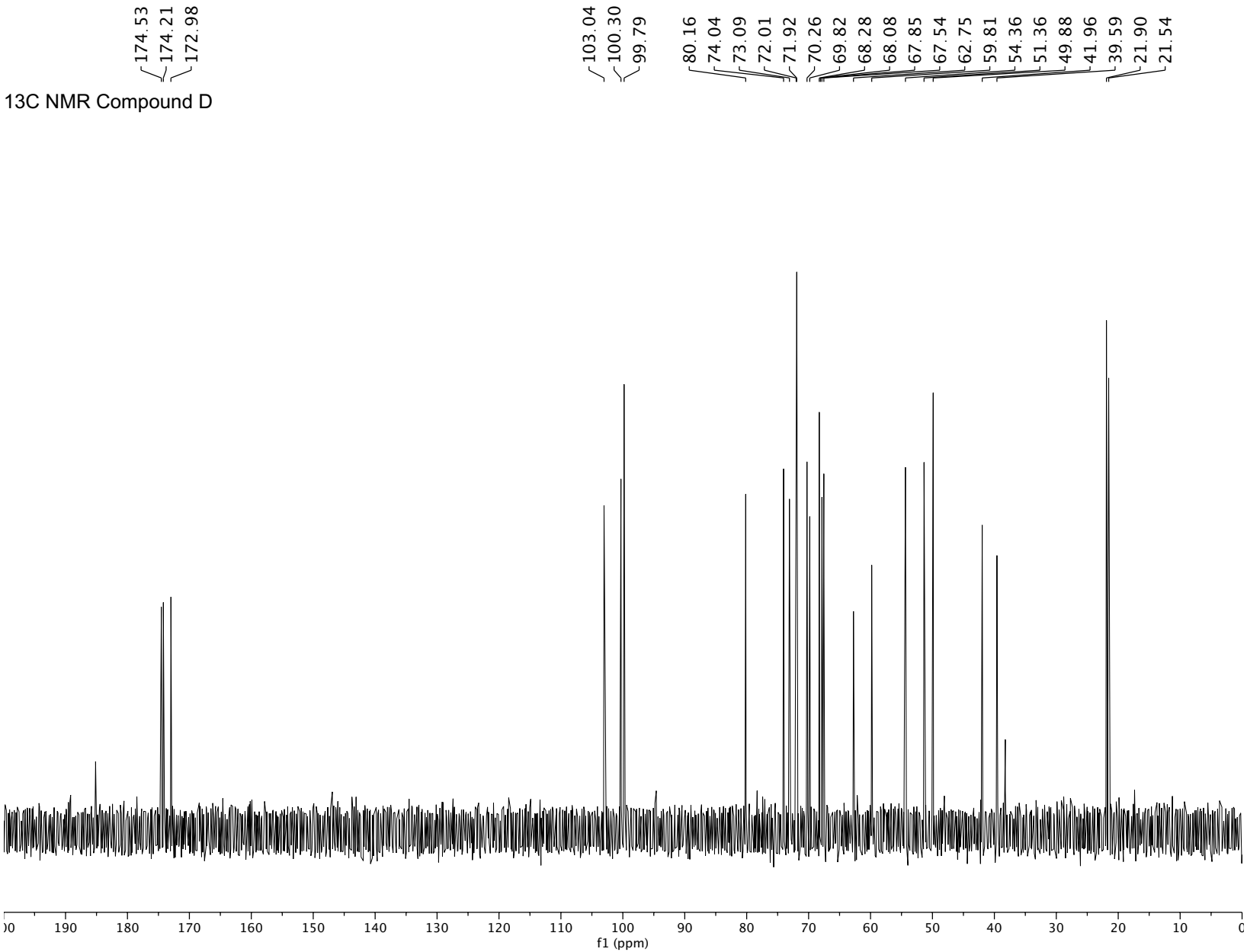
(1) Han, S.; Collins, B. E.; Bengtson, P.; Paulson, J. C., Homomultimeric complexes of CD22 in B cells revealed by protein-glycan cross-linking. *Nat Chem Biol* **2005,** *1* (2), 93-97.
(2) Blixt, O.; Brown, J.; Schur, M. J.; Wakarchuk, W.; Paulson, J. C., Efficient preparation of natural and synthetic galactosides with a recombinant beta-1,4-galactosyltransferase-/UDP-4 '-gal epimerase fusion protein. *J Org Chem* **2001,** *66* (7), 2442-2448.
(3) Nycholat, C. M.; Rademacher, C.; Kawasaki, N.; Paulson, J. C., In Silico-Aided Design of a Glycan Ligand of Sialoadhesin for in Vivo Targeting of Macrophages. *J Am Chem Soc* **2012,** *134* (38), 15696-15699.
(4) Yu, H.; Huang, S. S.; Chokhawala, H.; Sun, M. C.; Zheng, H. J.; Chen, X., Highly efficient chemoenzymatic synthesis of naturally occurring and non-natural alpha-2,6-linked sialosides: A P. damsela alpha-2,6-sialyltransferase with extremely flexible donor-substrate specificity. *Angew Chem Int Edit* **2006,** *45* (24), 3938-3944.
(5) Gonzalez-Gil, A.; Porell, R. N.; Fernandes, S. M.; Wei, Y. D.; Yu, H. F.; Carroll, D. J.; McBride, R.; Paulson, J. C.; Tiemeyer, M.; Aoki, K.; Bochner, B. S.; Schnaar, R. L., Sialylated keratan sulfate proteoglycans are Siglec-8 ligands in human airways. *Glycobiology* **2018,** *28* (10), 786-801.
(6) Tateno, H.; Li, H. Y.; Schur, M. J.; Bovin, N.; Crocker, P. R.; Wakarchuk, W. W.; Paulson, J. C., Distinct endocytic mechanisms of CD22 (Siglec-2) and Siglec-F reflect roles in cell signaling and innate immunity. *Mol Cell Biol* **2007,** *27* (16), 5699-5710.
(7) Chen, W. C.; Completo, G. C.; Sigal, D. S.; Crocker, P. R.; Saven, A.; Paulson, J. C., In vivo targeting of B-cell lymphoma with glycan ligands of CD22. *Blood* **2010,** *115* (23), 4778-4786.
(8) Chen, W. H. C.; Kawasaki, N.; Nycholat, C. M.; Han, S. F.; Pilotte, J.; Crocker, P. R.; Paulson, J. C., Antigen Delivery to Macrophages Using Liposomal Nanoparticles Targeting Sialoadhesin/CD169. *PloS one* **2012,** *7* (6).
(9) Pfrengle, F.; Macauley, M. S.; Paulson, J. C., Investigating the inhibitory function of Siglec-G on B cells using antigenic liposomes decorated with carbohydrate ligands. *Abstr Pap Am Chem S* **2012,** *243*.
(10) Blixt, O.; Han, S. F.; Liao, L.; Zeng, Y.; Hoffmann, J.; Futakawa, S.; Paulson, J. C., Sialoside analogue arrays for rapid identification of high affinity siglec ligands. *J Am Chem Soc* **2008,** *130* (21), 6680-+.
(11) O'Sullivan, J. A.; Wei, Y.; Carroll, D. J.; Moreno-Vinasco, L.; Cao, Y.; Zhang, F.; Lee, J. J.; Zhu, Z.; Bochner, B. S., Frontline Science: Characterization of a novel mouse strain expressing human Siglec-8 only on eosinophils. *J Leukoc Biol* **2018,** *104* (1), 11-19.
(12) Angata, T.; Hingorani, R.; Varki, N. M.; Varki, A., Cloning and characterization of a novel mouse Siglec, mSiglec-F: differential evolution of the mouse and human (CD33) Siglec-3-related gene clusters. *J. Biol. Chem.* **2001,** *276* (48), 45128-45136.
(13) Zhang, J. Q.; Biedermann, B.; Nitschke, L.; Crocker, P. R., The murine inhibitory receptor mSiglec-E is expressed broadly on cells of the innate immune system whereas mSiglec-F is restricted to eosinophils. *Eur. J. Immunol.* **2004,** *34* (4), 1175-1184.
(14) Tateno, H.; Crocker, P. R.; Paulson, J. C., Mouse Siglec-F and human Siglec-8 are functionally convergent paralogs that are selectively expressed on eosinophils and recognize 6'-sulfo-sialyl Lewis X as a preferred glycan ligand. *Glycobiology* **2005,** *15* (11), 1125-1135.
(15) Zhang, M.; Angata, T.; Cho, J. Y.; Miller, M.; Broide, D. H.; Varki, A., Defining the in vivo function of Siglec-F, a CD33-related Siglec expressed on mouse eosinophils. *Blood* **2007,** *109* (10), 4280-4287.
(16) Dyer, K. D.; Moser, J. M.; Czapiga, M.; Siegel, S. J.; Percopo, C. M.; Rosenberg, H. F., Functionally competent eosinophils differentiated ex vivo in high purity from normal mouse bone marrow. *J Immunol* **2008,** *181* (6), 4004-4009.

(17) Cheng, C. W.; Chou, C. C.; Hsieh, H. W.; Tu, Z.; Lin, C. H.; Nycholat, C.; Fukuda, M.; Khoo, K. H., Efficient Mapping of Sulfated Glycotopes by Negative Ion Mode nanoLC-MS/MS-Based Sulfoglycomic Analysis of Permethylated Glycans. *Anal Chem* **2015,** *87* (12), 6380-6388.

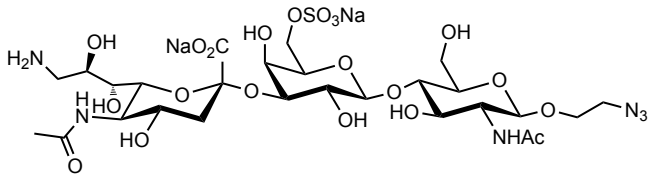
1H NMR Compound D

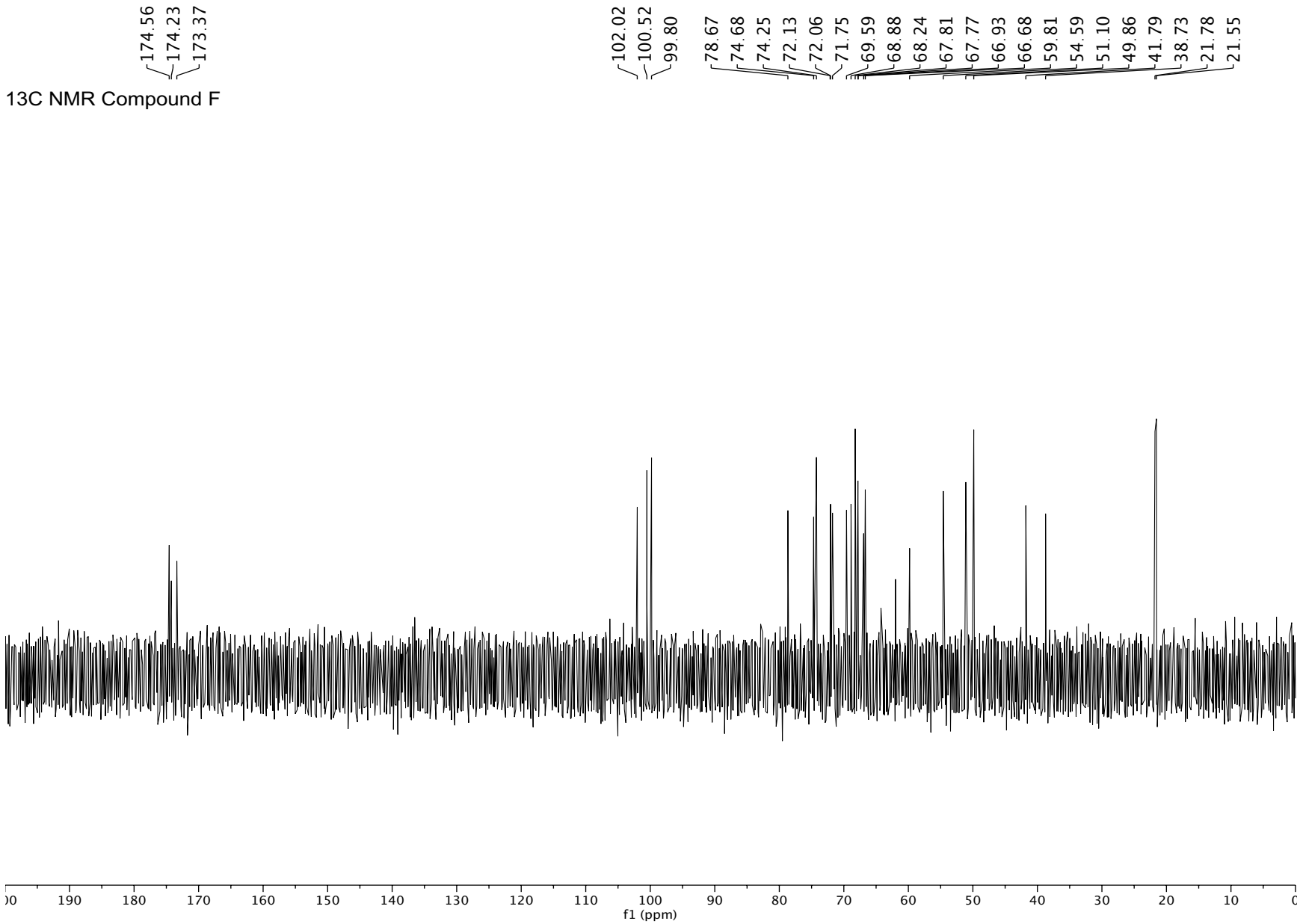


13C NMR Compound D



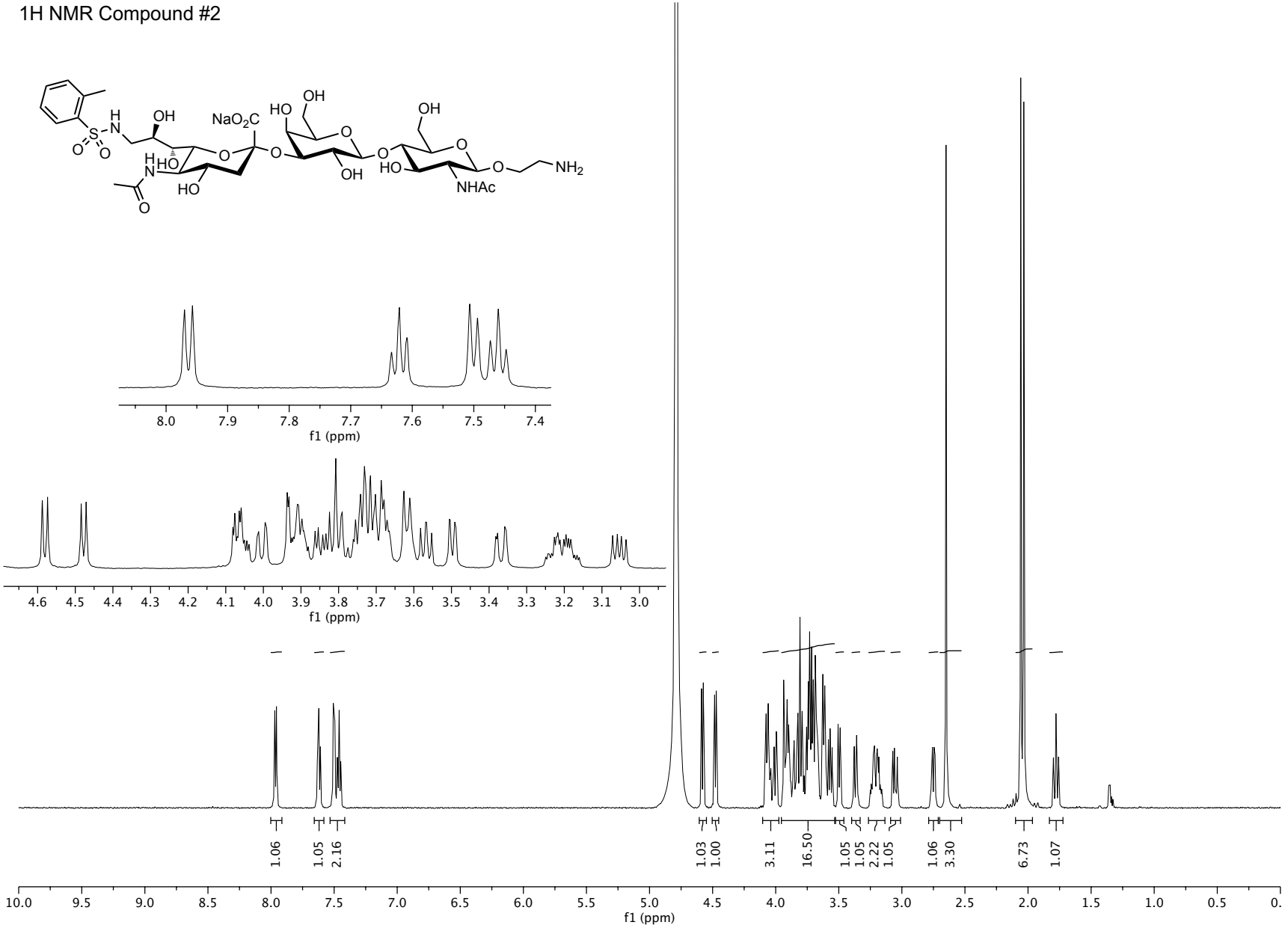
1H NMR Compound F

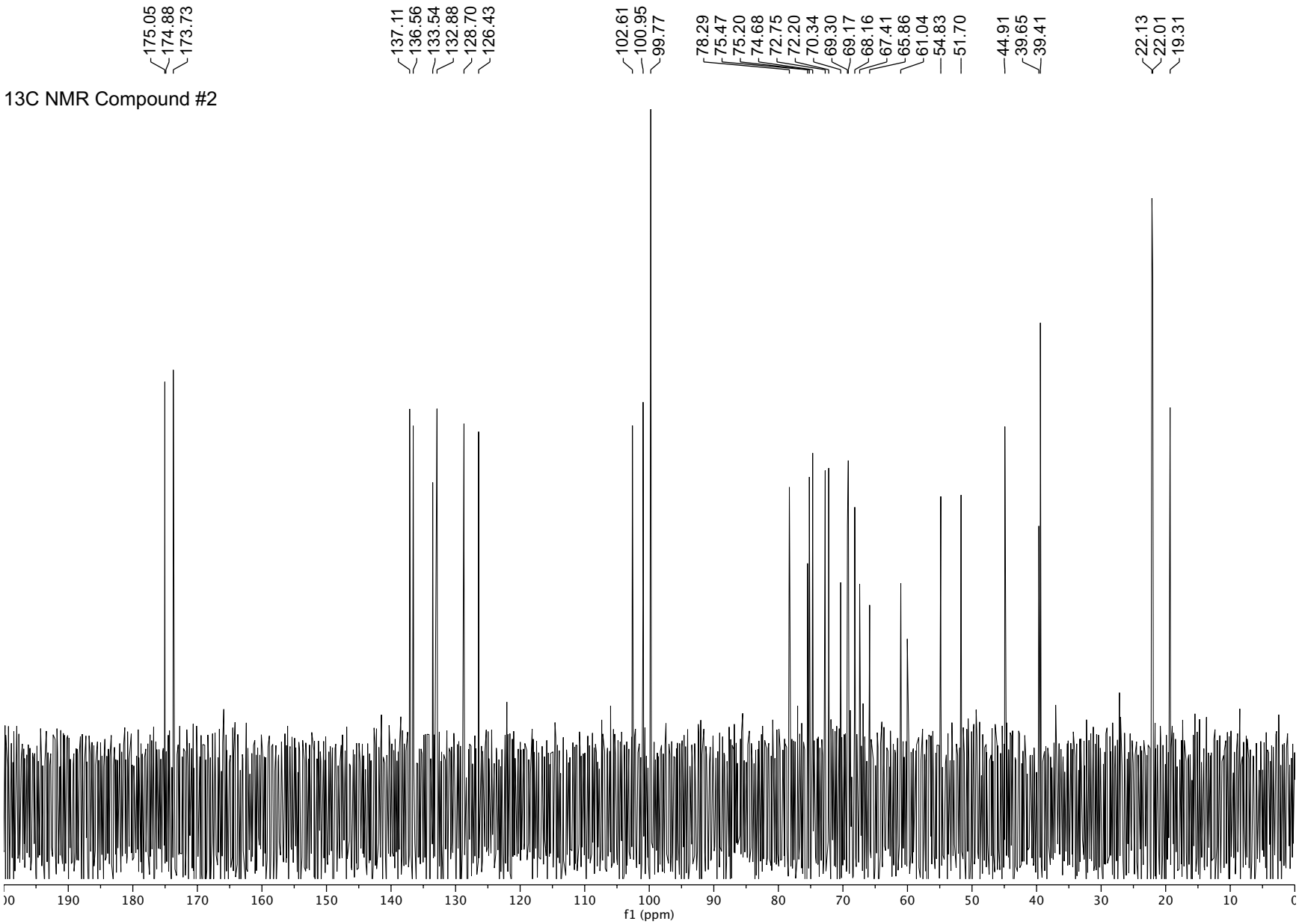




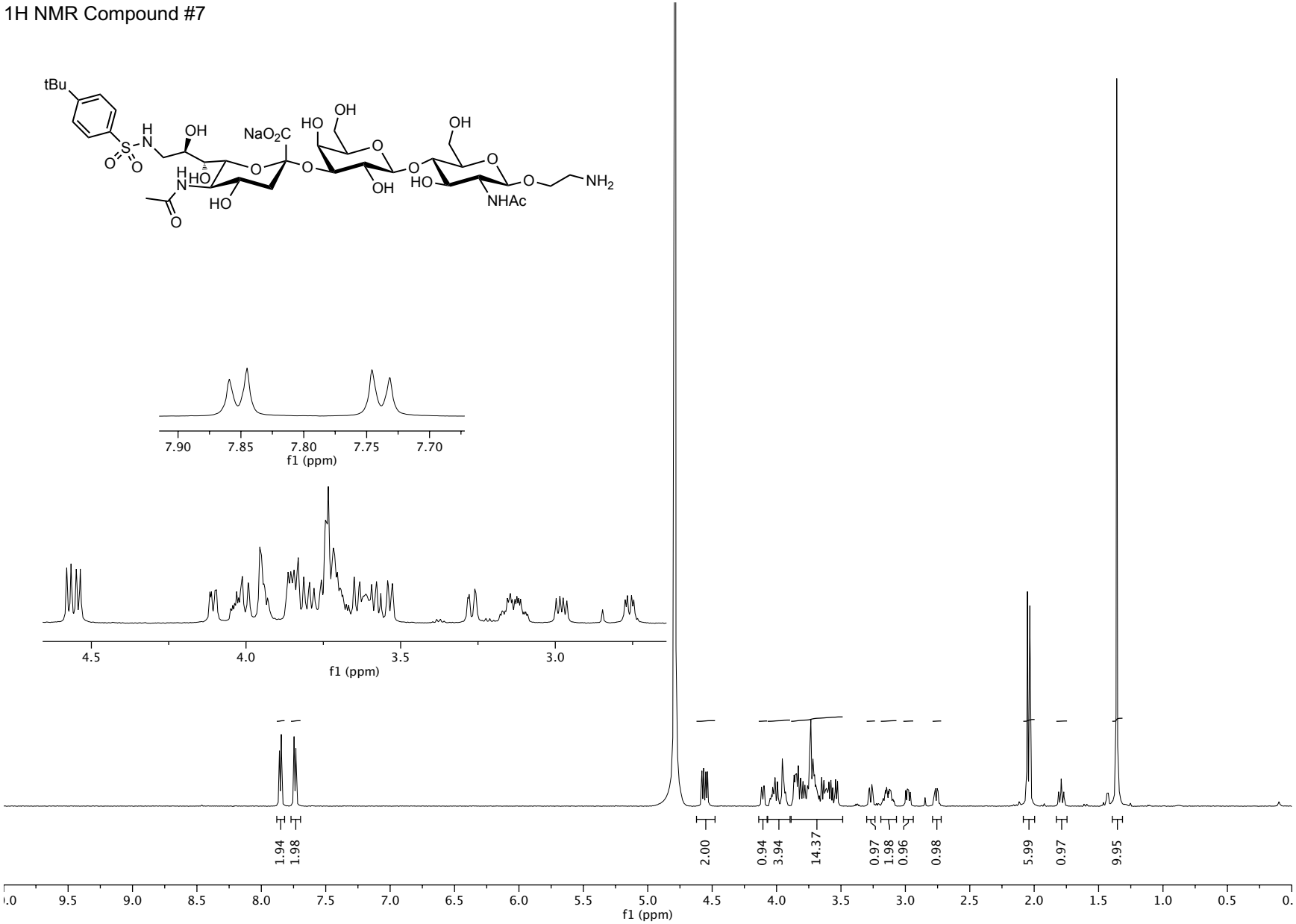
13C NMR Compound F

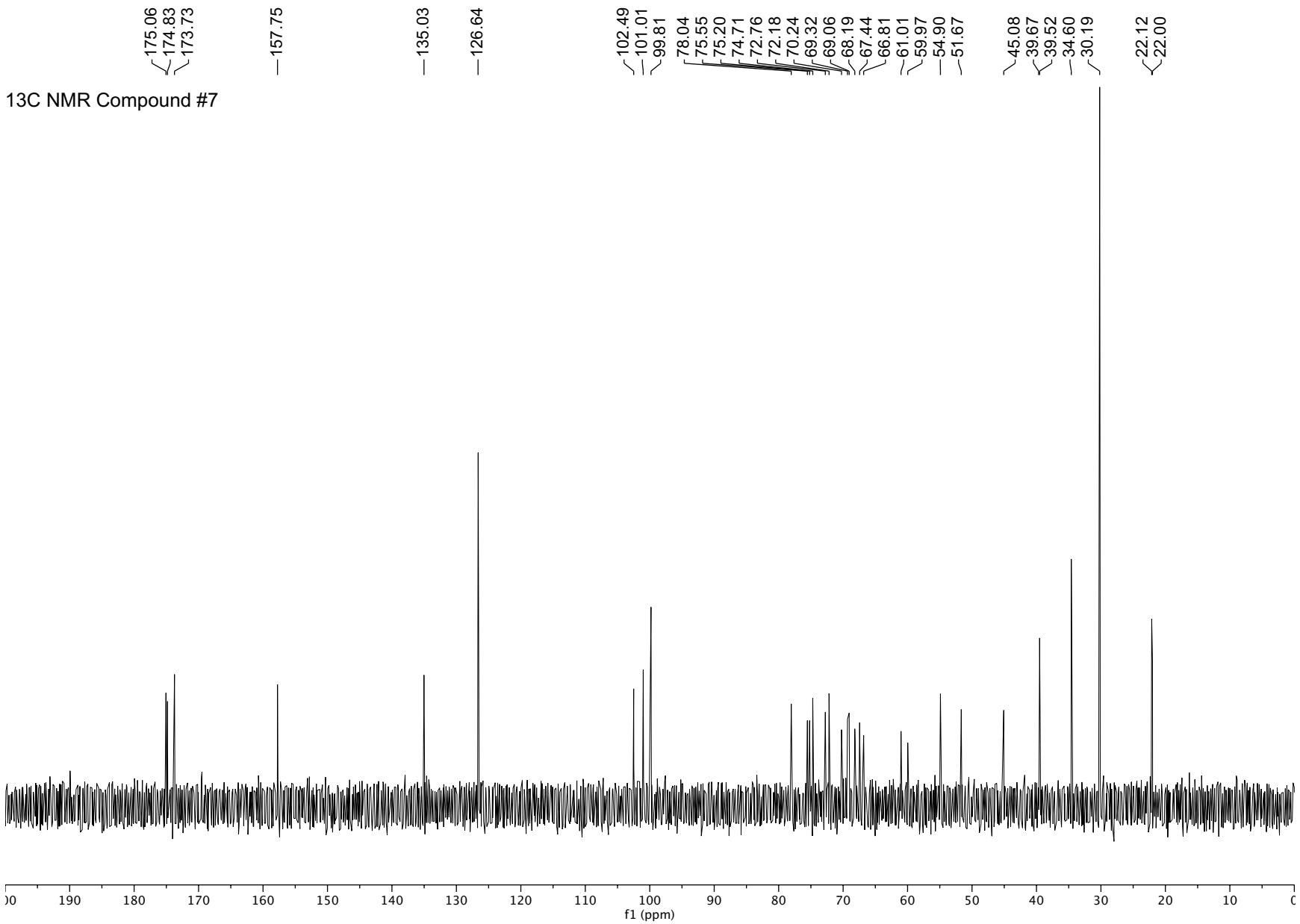
1H NMR Compound #2



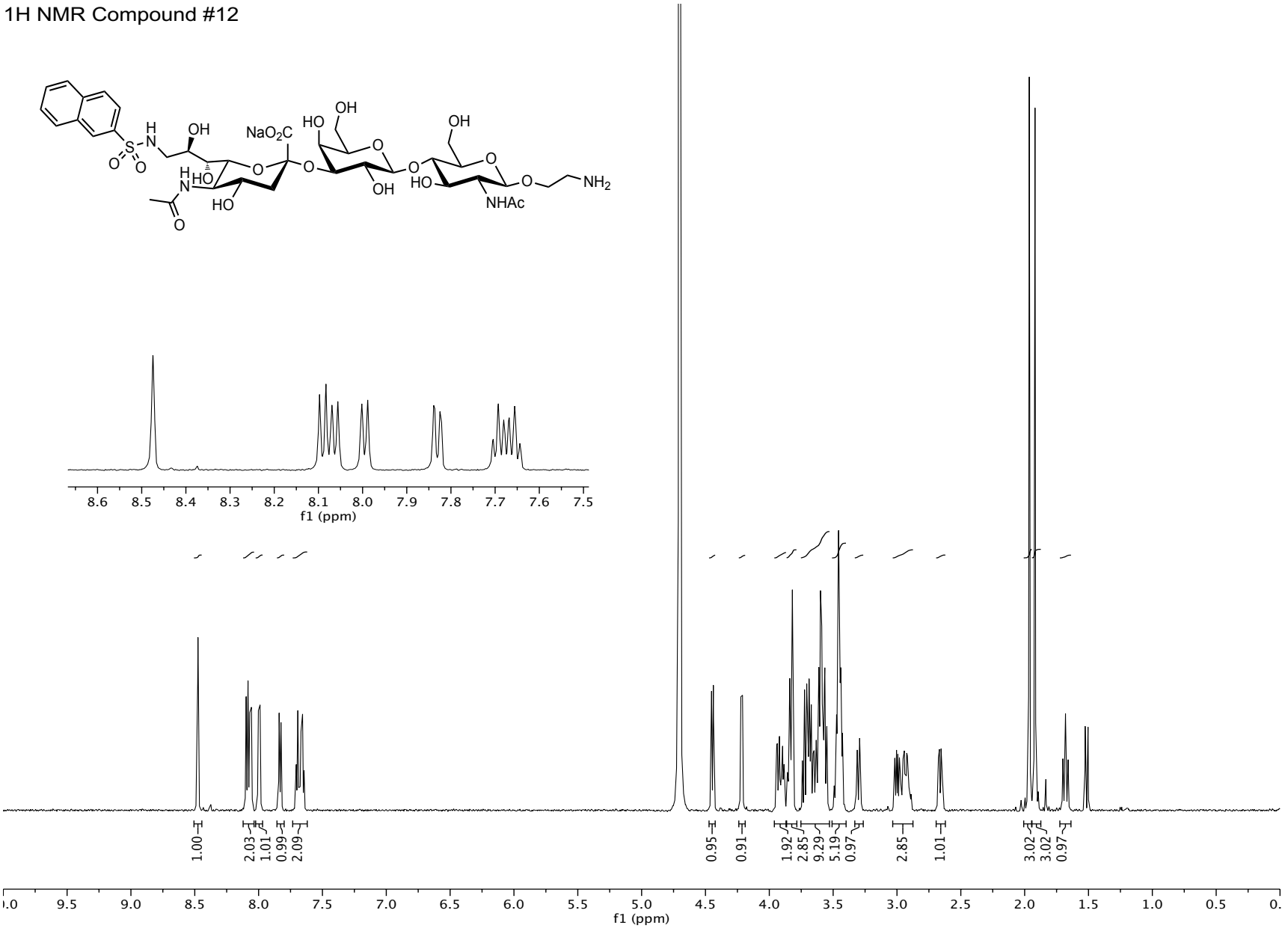

175.05
174.88
173.73
137.11
136.56
133.54
132.88
128.70
126.43
102.61
100.95
99.77
78.29
75.47
75.20
74.68
72.75
72.20
70.34
69.30
69.17
68.16
67.41
65.86
61.04
54.83
51.70
44.91
39.65
39.41
22.13
22.01
19.31
13C NMR Compound #2
f1 (ppm)

1H NMR Compound #7



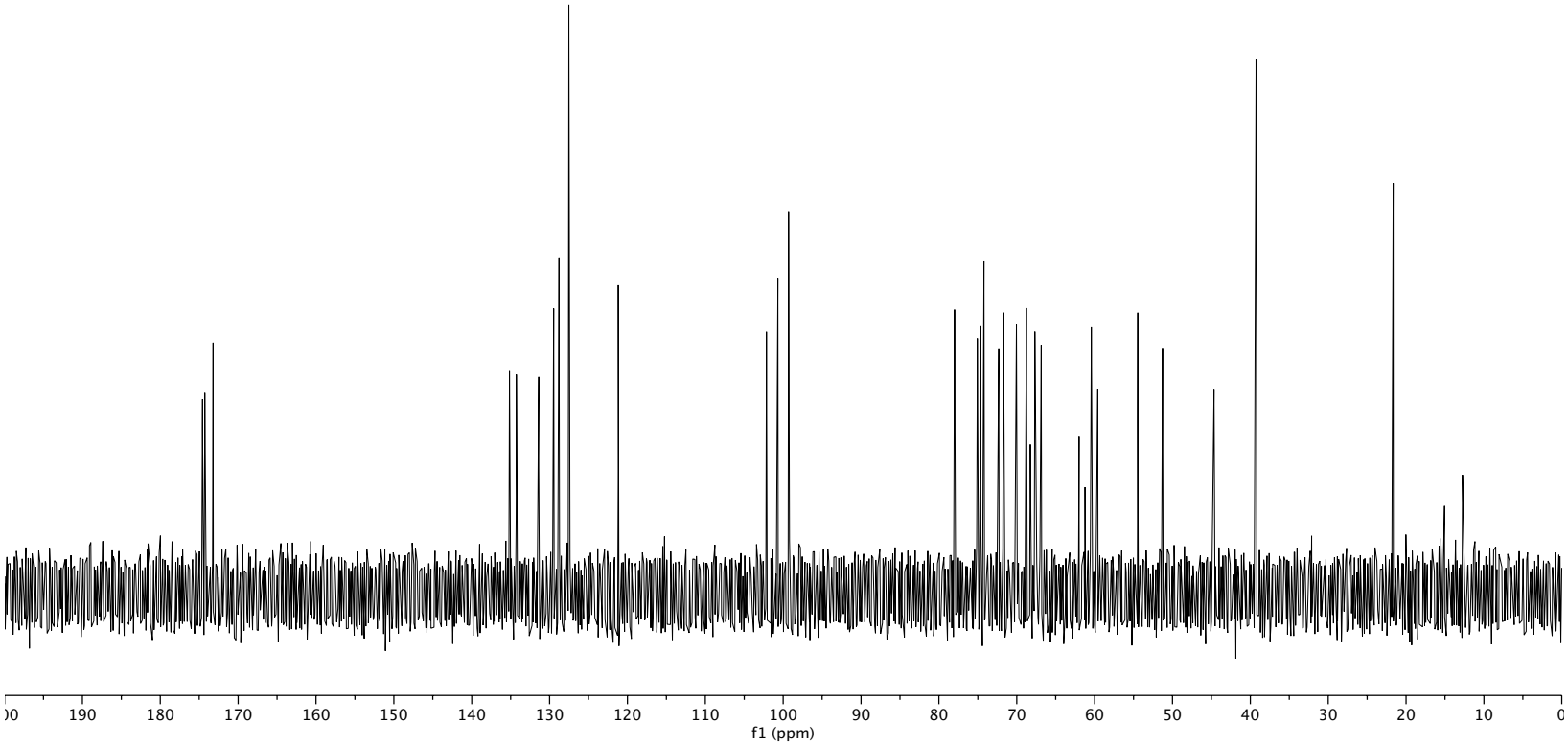

175.06
174.83
173.73
157.75
135.03
126.64
102.49
101.01
99.81
78.04
75.55
75.20
74.71
72.76
72.18
70.24
69.32
69.06
68.19
67.44
66.81
61.01
59.97
54.90
51.67
45.08
39.67
39.52
34.60
30.19
22.12
22.00
13C NMR Compound #7
f1 (ppm)

1H NMR Compound #12

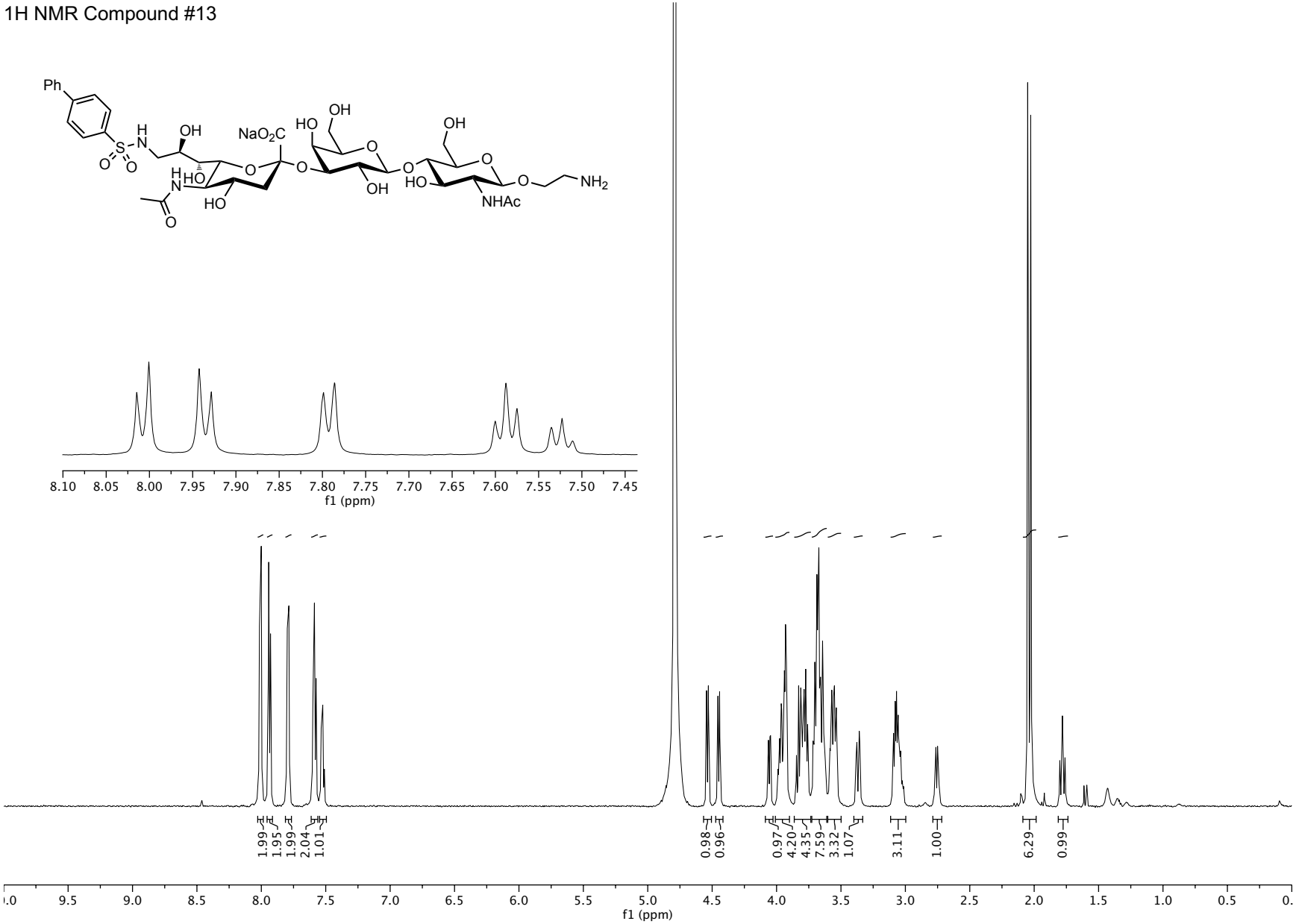


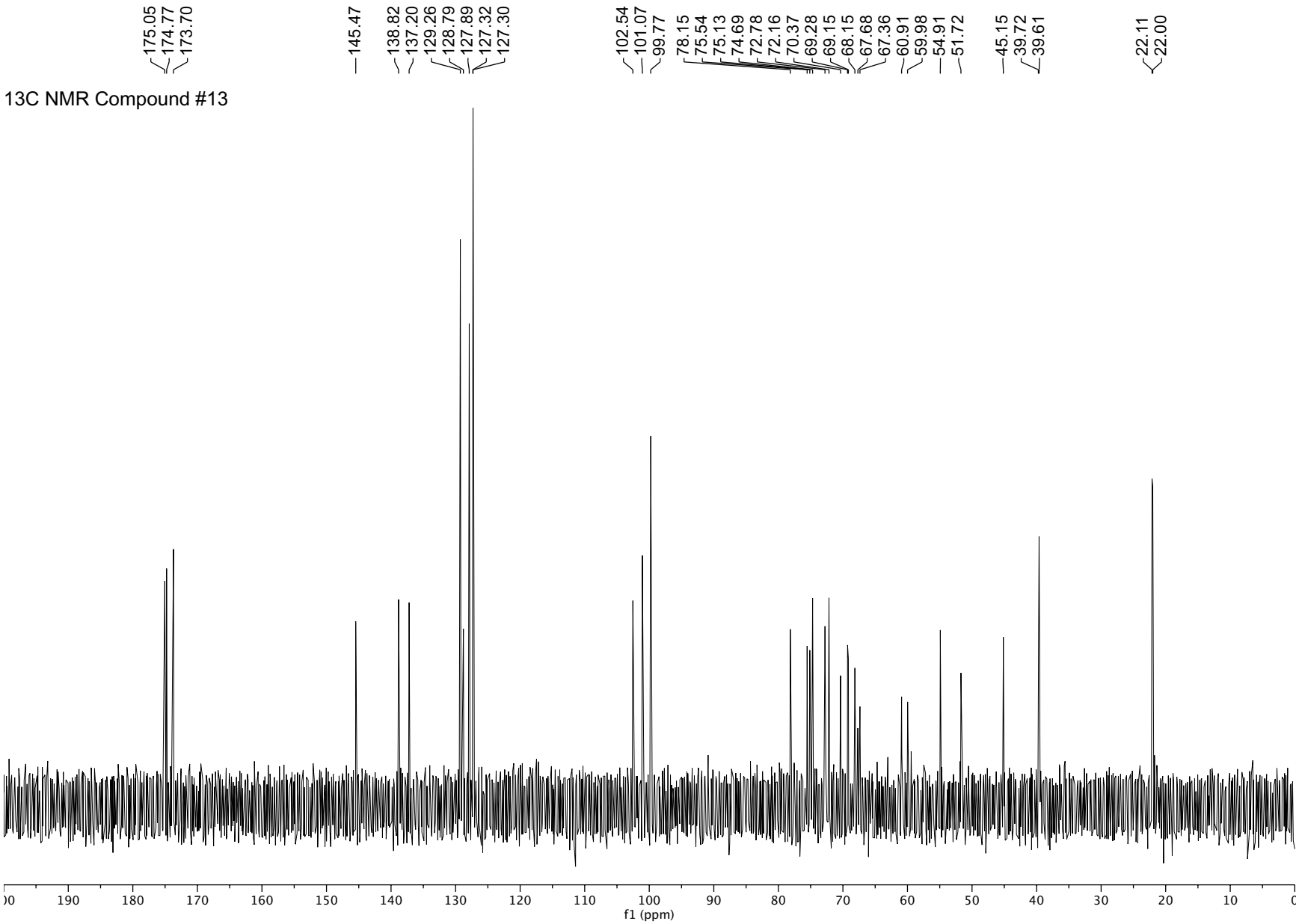
174.57
174.28
173.22
135.15
134.26
131.40
129.47
128.89
128.79
127.69
127.52
121.18
102.14
100.68
99.30
77.99
75.06
74.62
74.21
72.31
71.70
70.04
68.77
68.71
68.26
67.67
66.86
61.99
61.24
60.42
59.60
54.46
51.27
44.66
39.27
21.66
21.52

13C NMR Compound #12

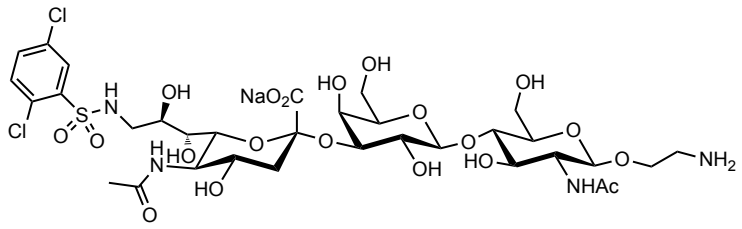


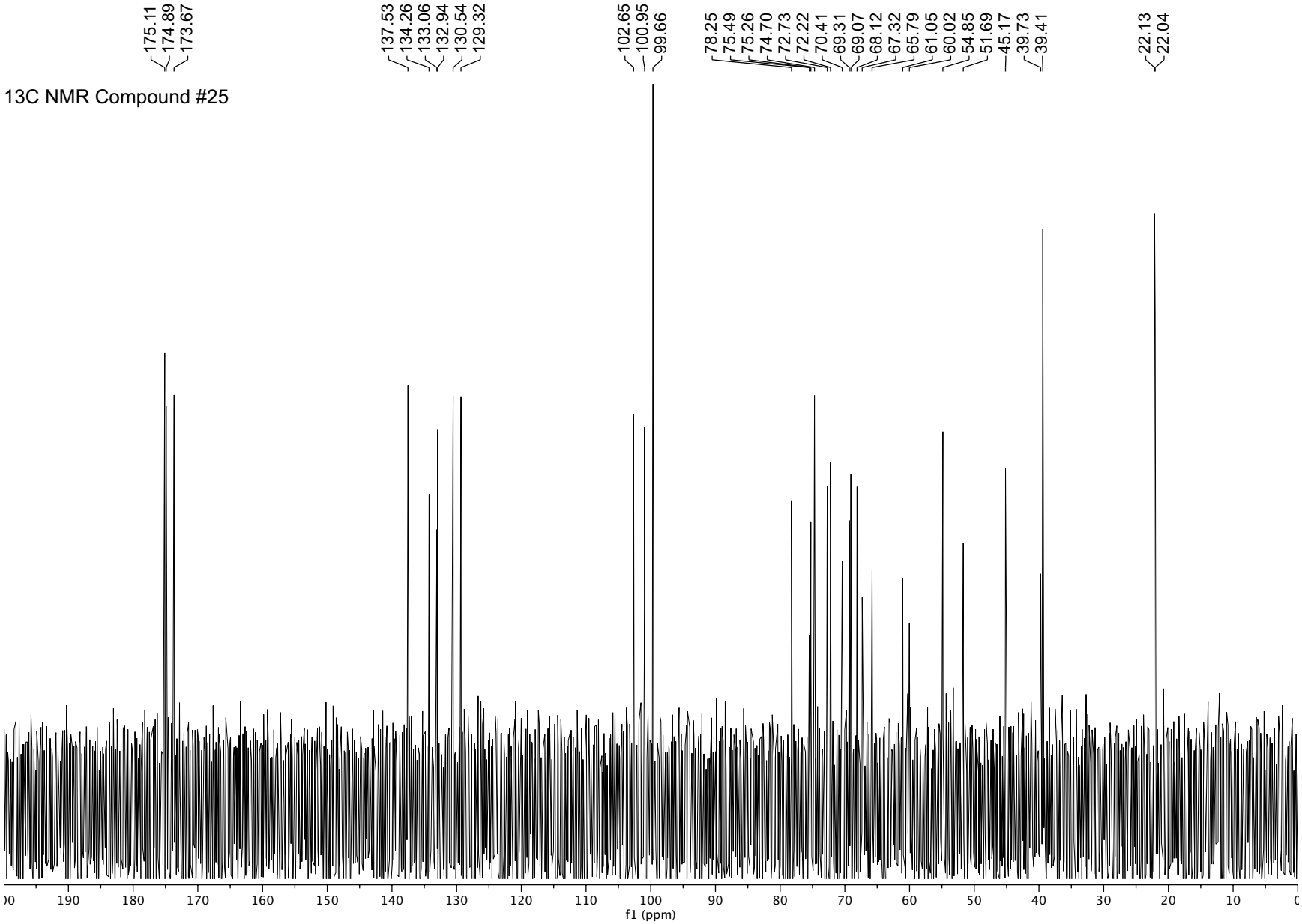
1H NMR Compound #13



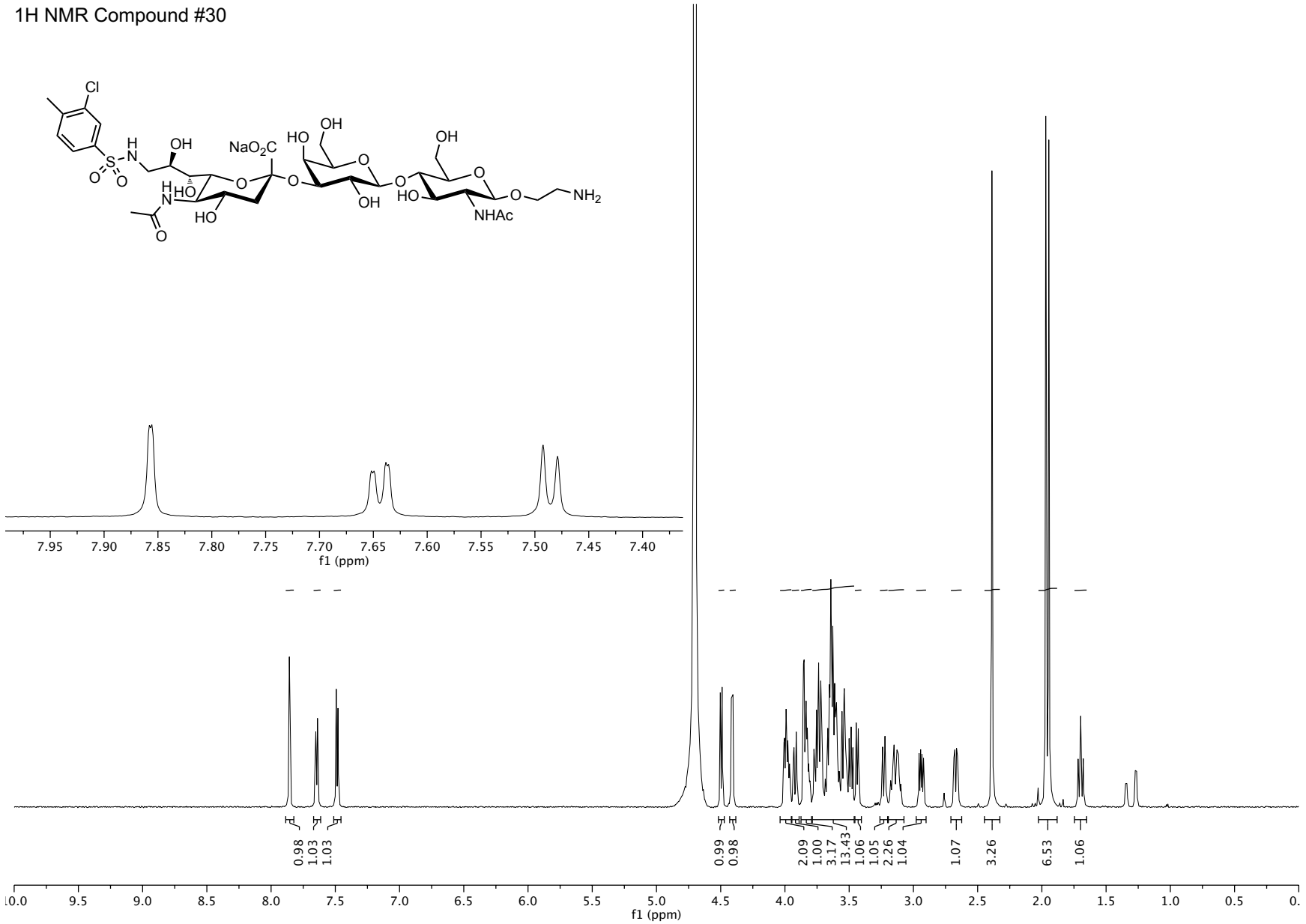

175.05
174.77
173.70
145.47
138.82
137.20
129.26
128.79
127.89
127.32
127.30
102.54
101.07
99.77
78.15
75.54
75.13
74.69
72.78
72.16
70.37
69.28
69.15
68.15
67.68
67.36
60.91
59.98
54.91
51.72
45.15
39.72
39.61
22.11
22.00
13C NMR Compound #13
f1 (ppm)

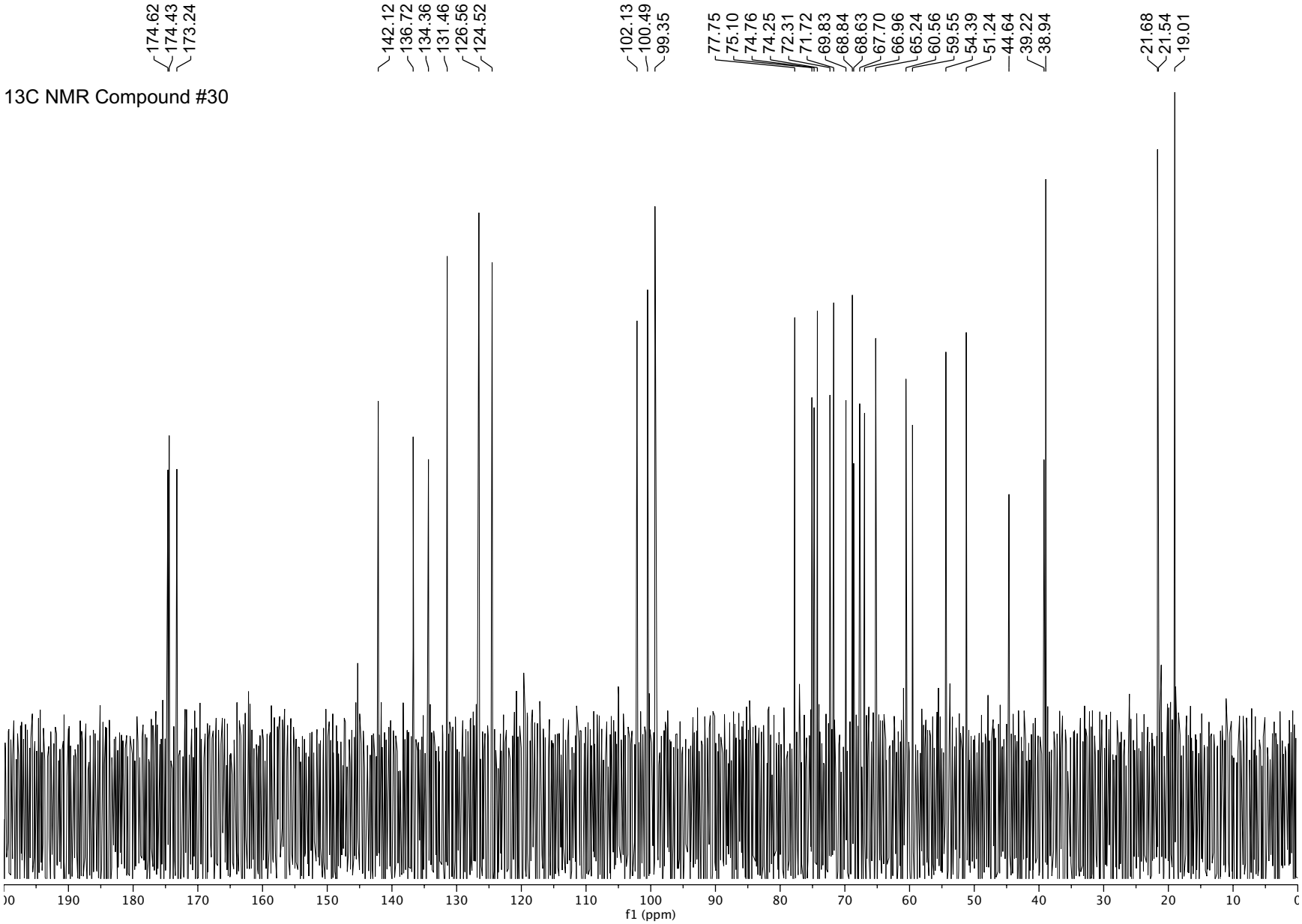
1H NMR Compound #25



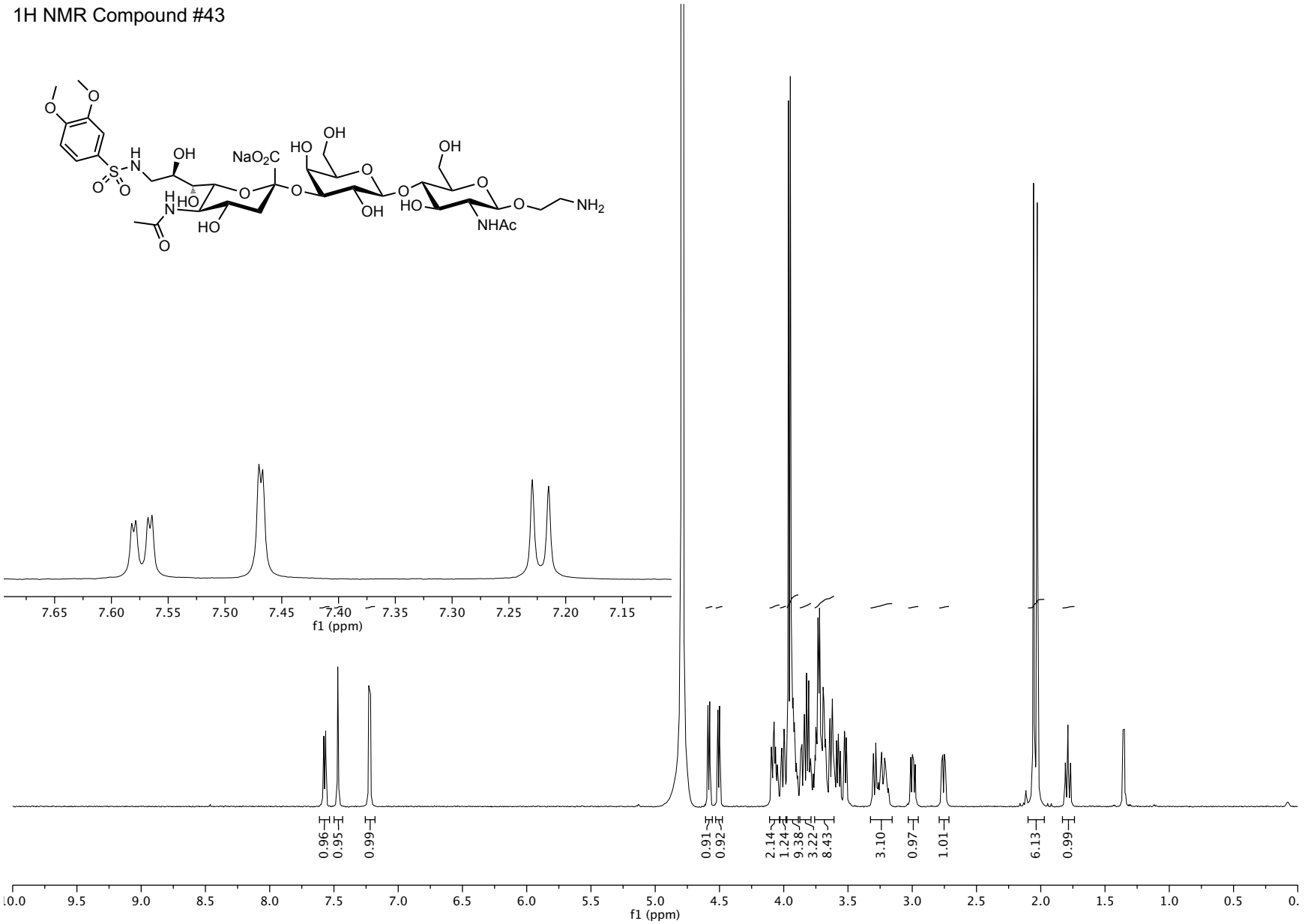

175.11
174.89
173.67
137.53
134.26
133.06
132.94
130.54
129.32
102.65
100.95
99.66
78.25
75.49
75.26
74.70
72.73
72.22
70.41
69.31
69.07
68.12
67.32
65.79
61.05
60.02
54.85
51.69
45.17
39.73
39.41
22.13
22.04
13C NMR Compound #25
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

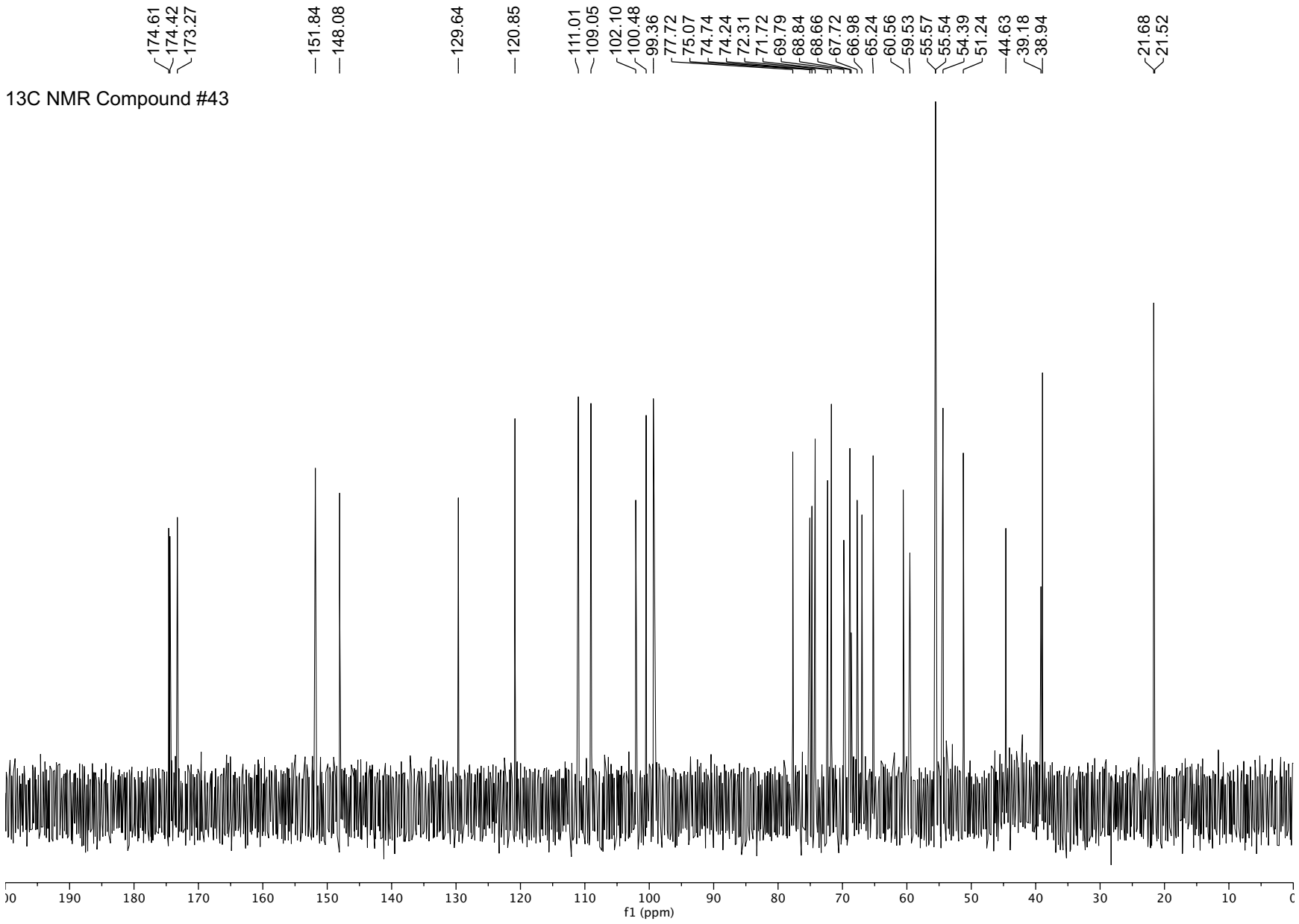
1H NMR Compound #30



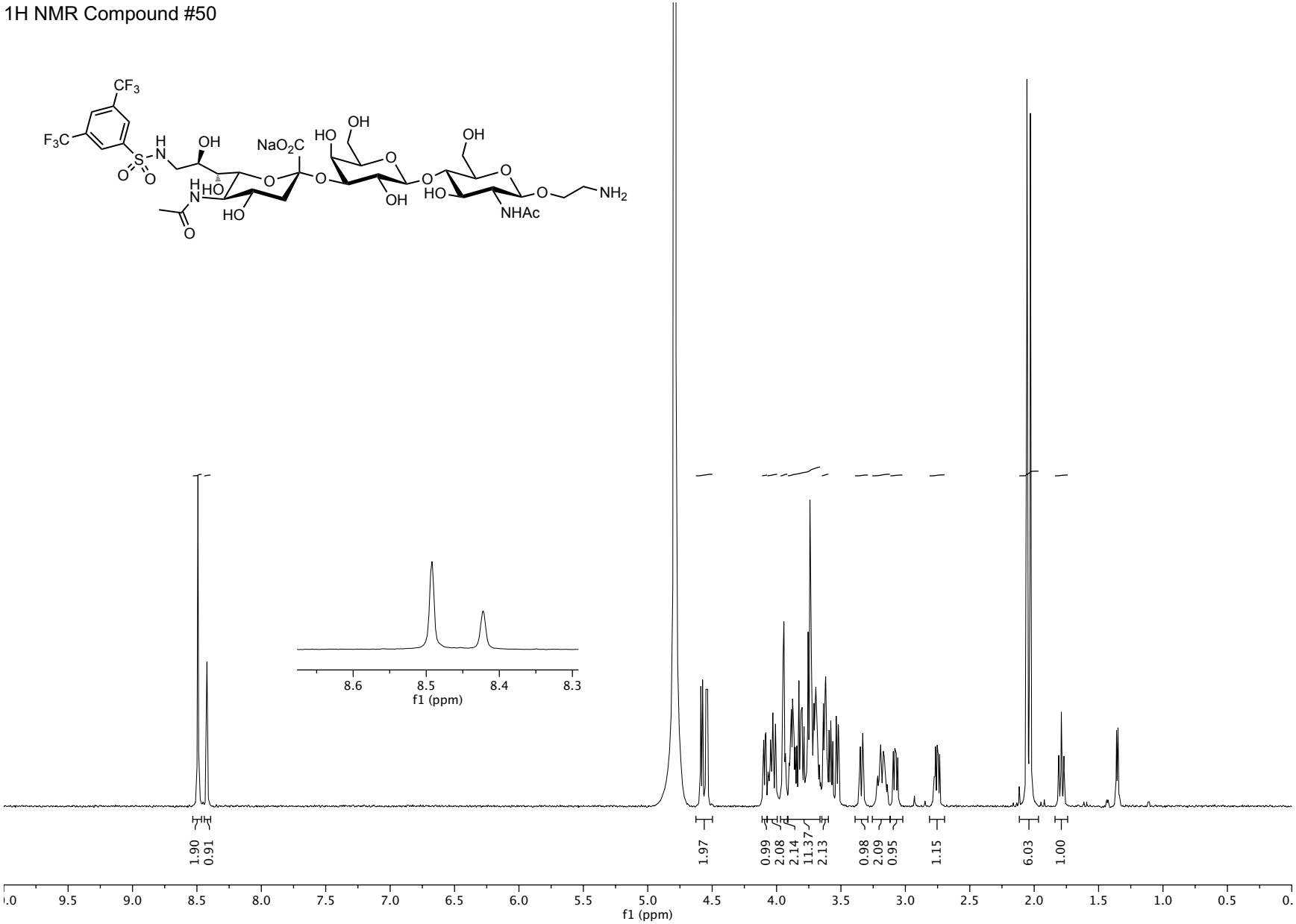

174.62
174.43
173.24
142.12
136.72
134.36
131.46
126.56
124.52
102.13
100.49
99.35
77.75
75.10
74.76
74.25
72.31
71.72
69.83
68.84
68.63
67.70
66.96
65.24
60.56
59.55
54.39
51.24
44.64
39.22
38.94
21.68
21.54
19.01
13C NMR Compound #30
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

1H NMR Compound #43

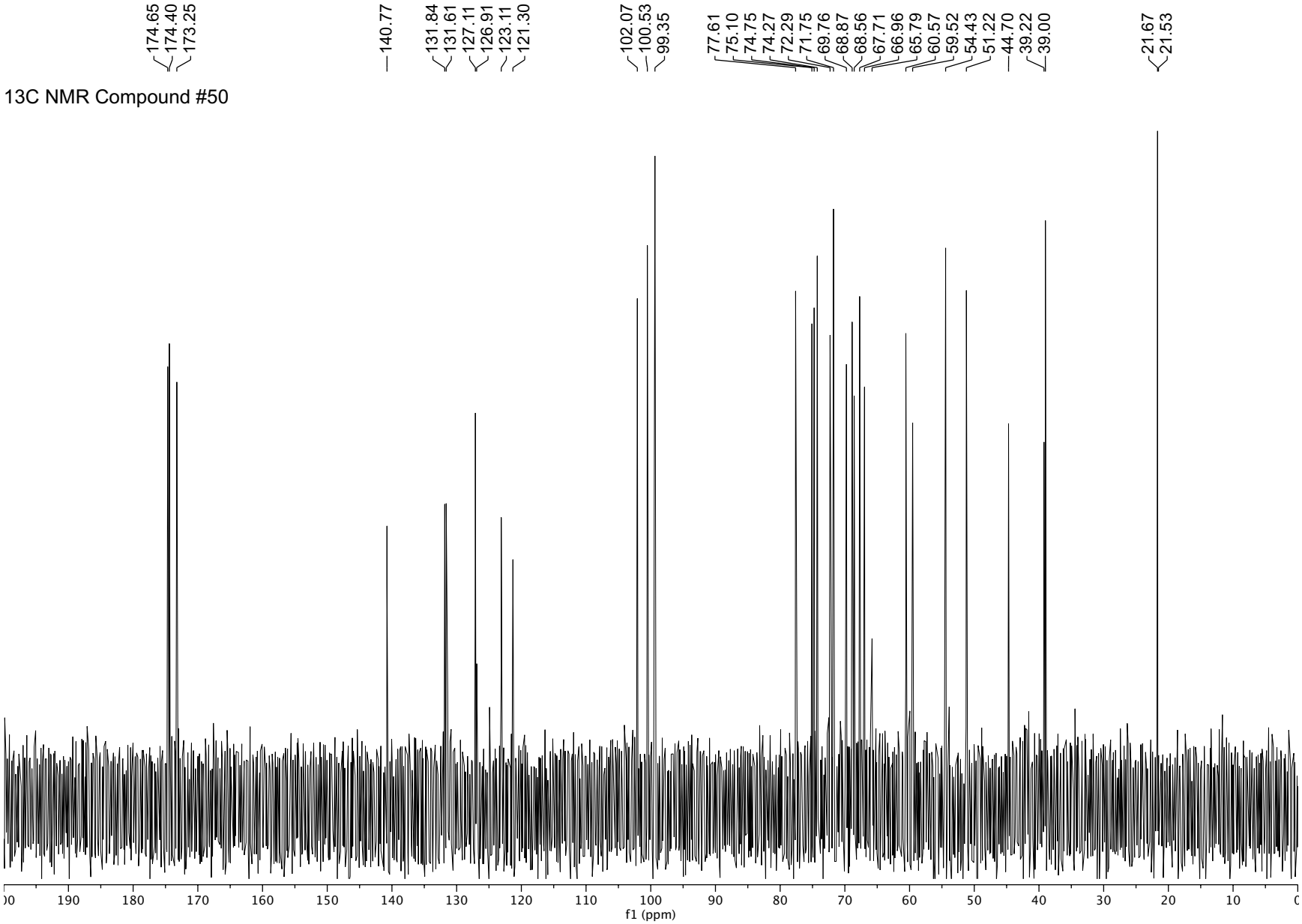



174.61
174.42
173.27
151.84
148.08
129.64
120.85
111.01
109.05
102.10
100.48
99.36
77.72
75.07
74.74
74.24
72.31
71.72
69.79
68.84
68.66
67.72
66.98
65.24
60.56
59.53
55.57
55.54
54.39
51.24
44.63
39.18
38.94
21.68
21.52
13C NMR Compound #43
f1 (ppm)

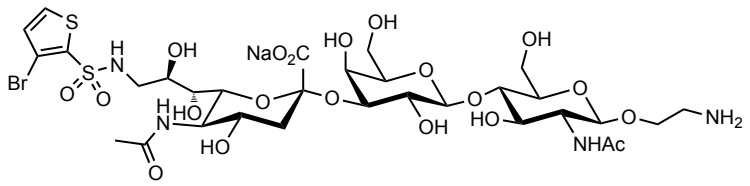
1H NMR Compound #50

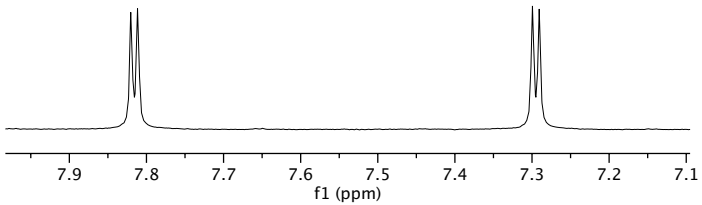


13C NMR Compound #50

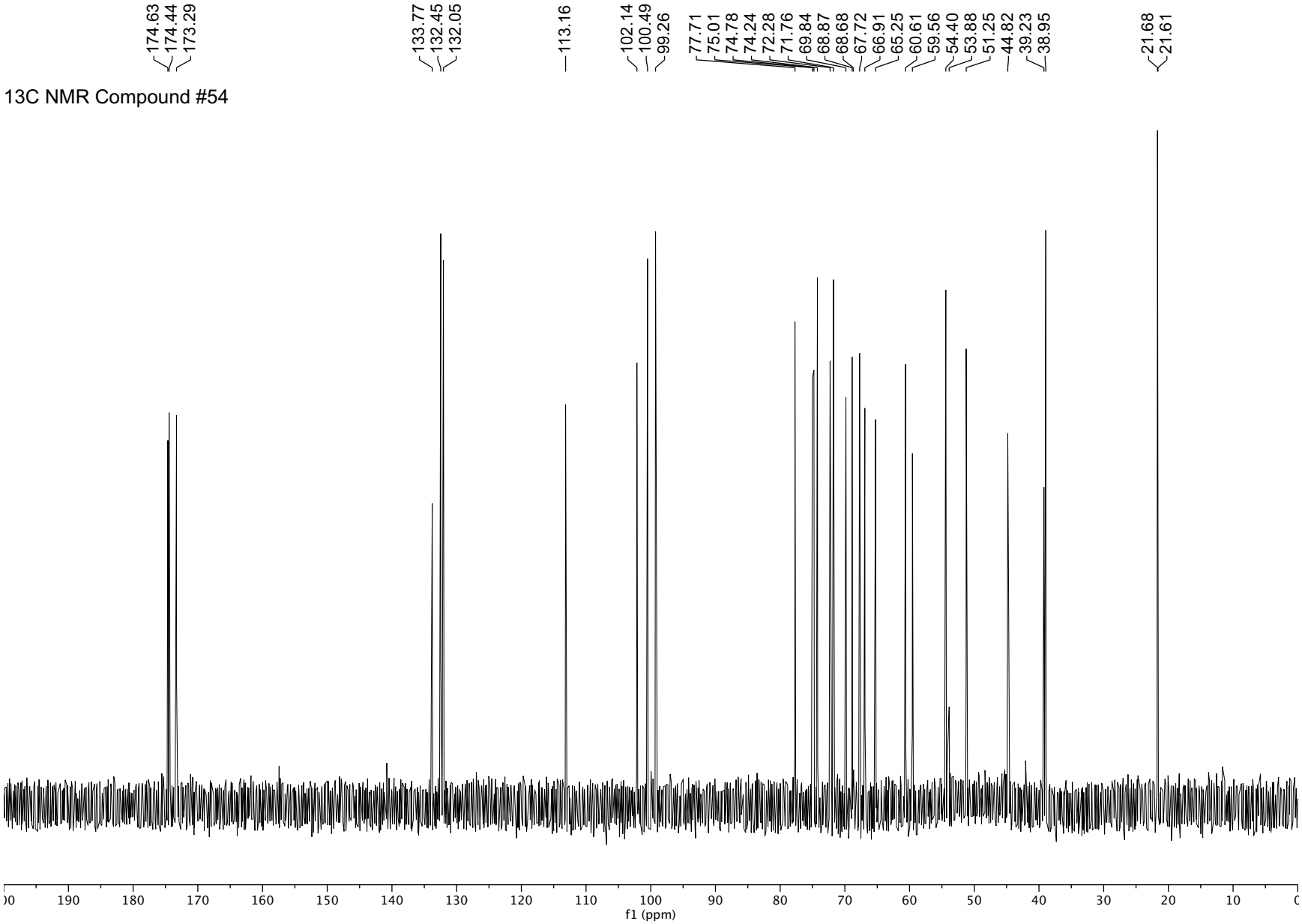


1H NMR Compound #54

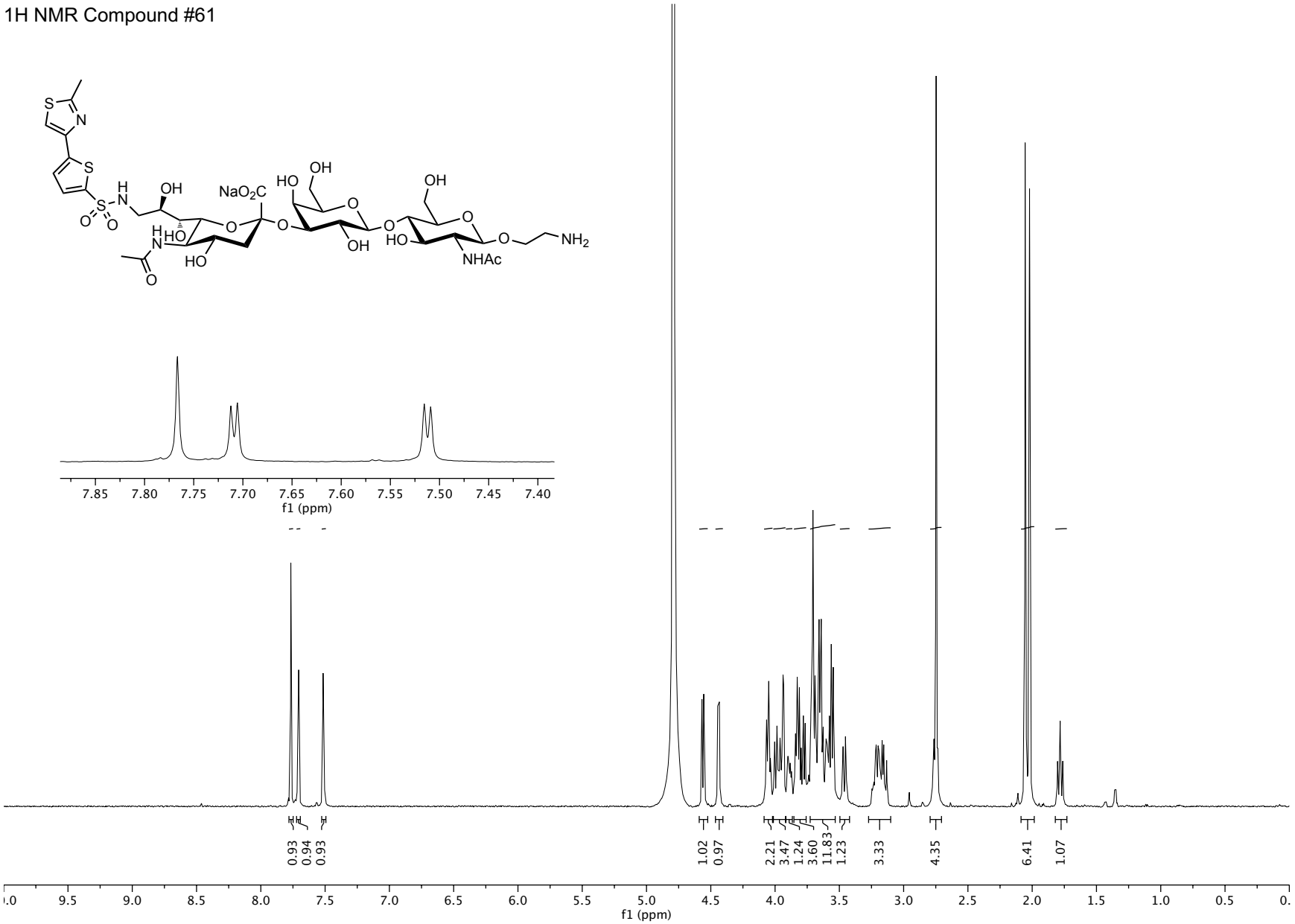


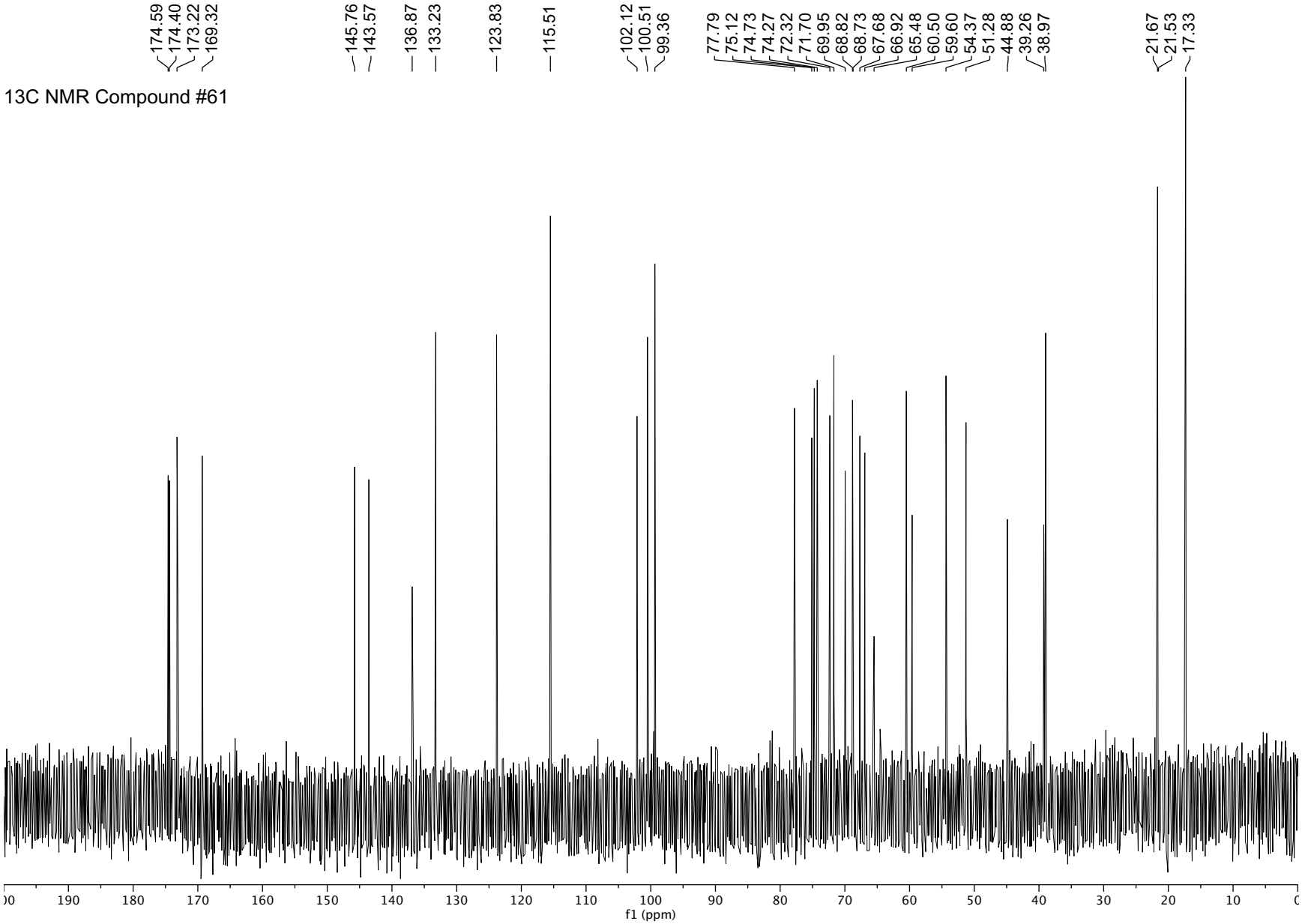


13C NMR Compound #54

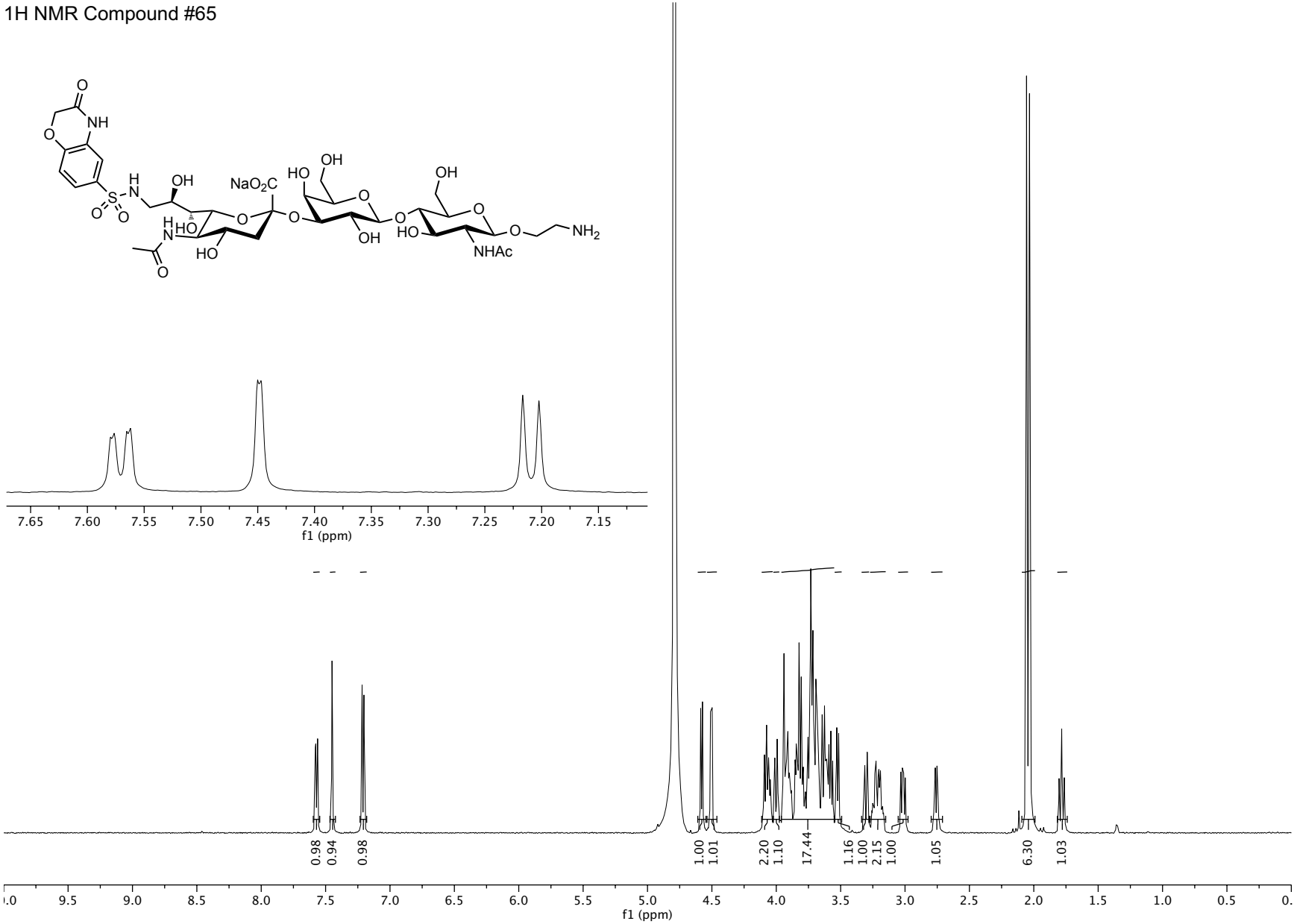


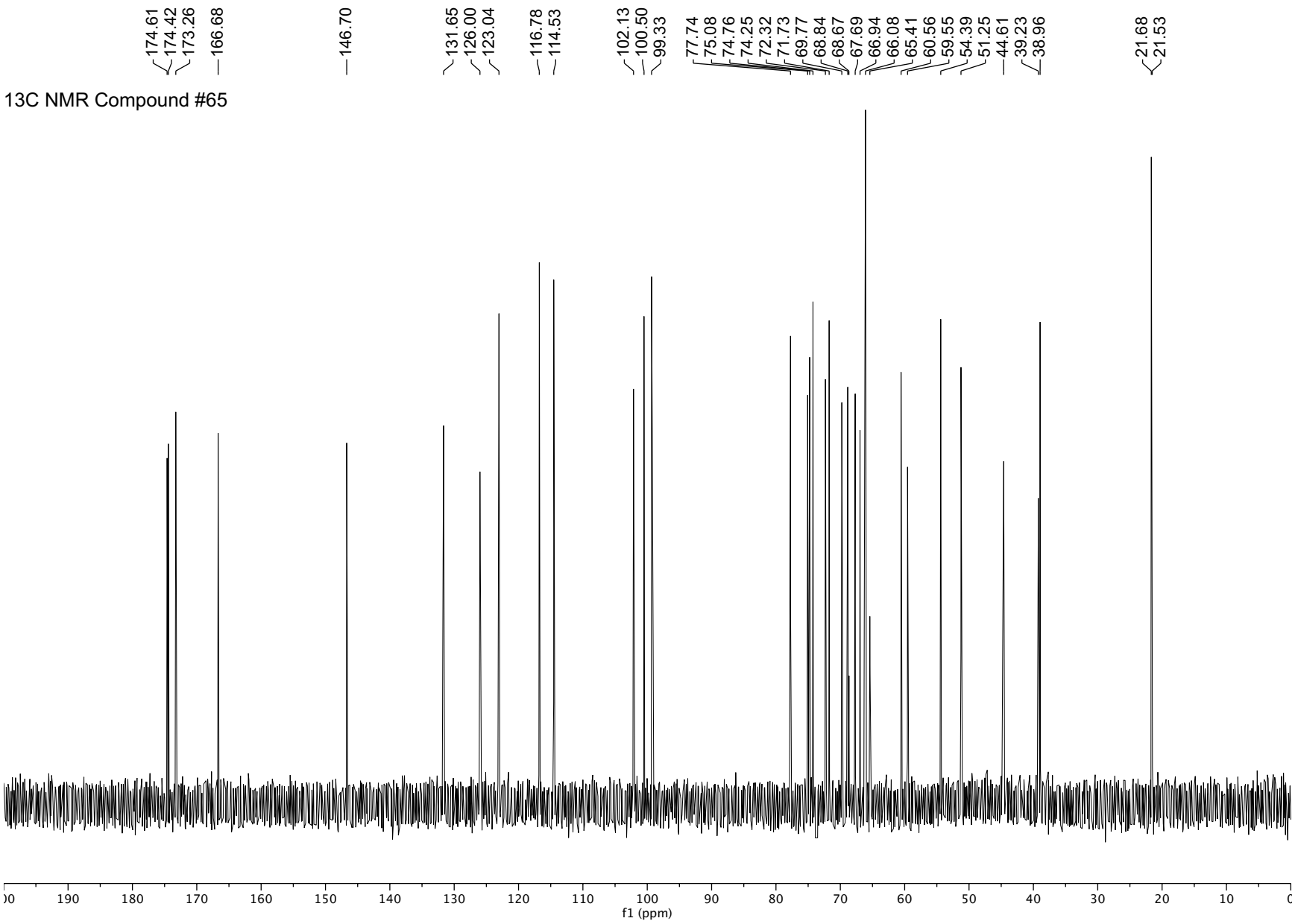
1H NMR Compound #61



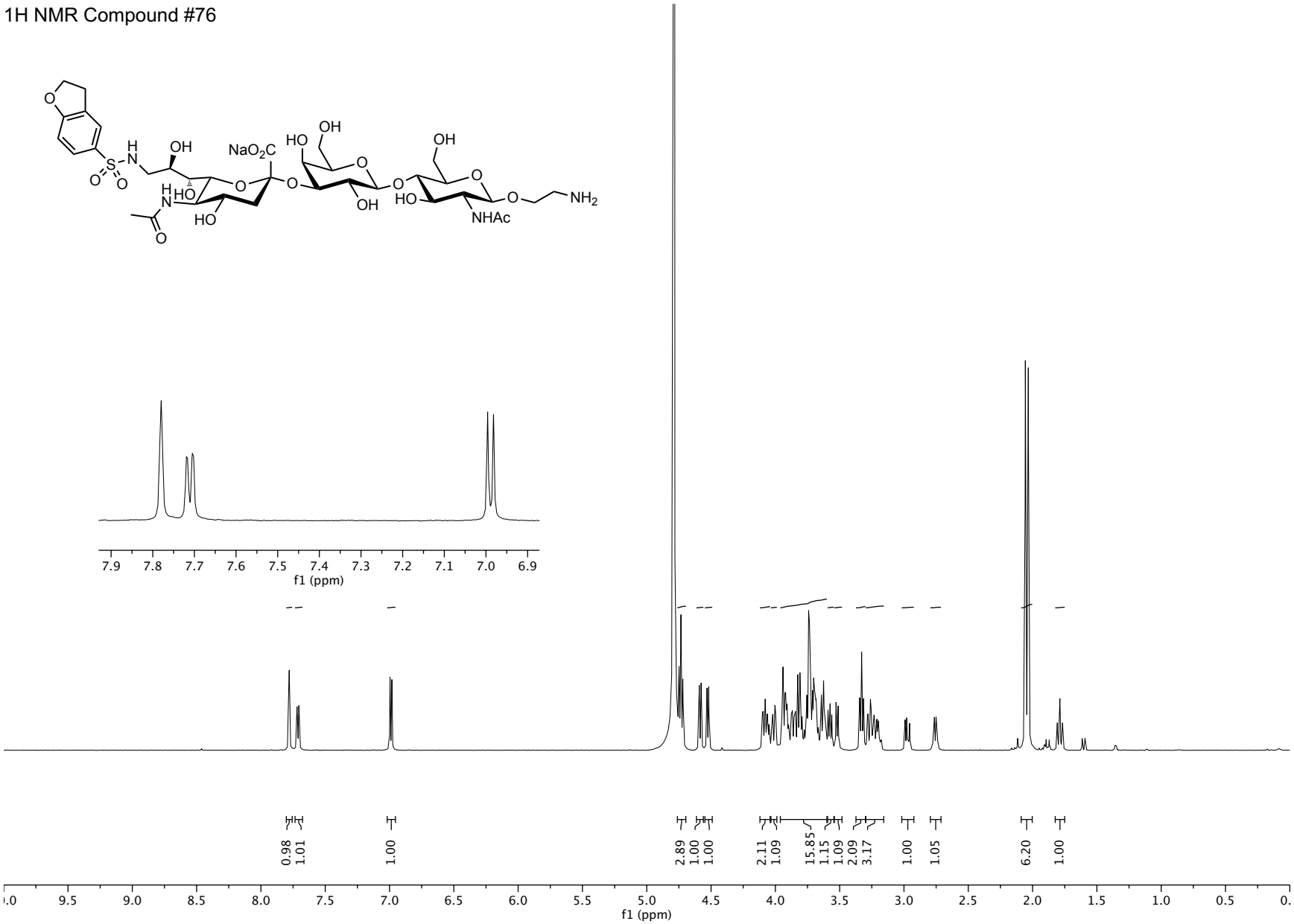

174.59
174.40
173.22
169.32
145.76
143.57
136.87
133.23
123.83
115.51
102.12
100.51
99.36
77.79
75.12
74.73
74.27
72.32
71.70
69.95
68.82
68.73
67.68
66.92
65.48
60.50
59.60
54.37
51.28
44.88
39.26
38.97
21.67
21.53
17.33
13C NMR Compound #61
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

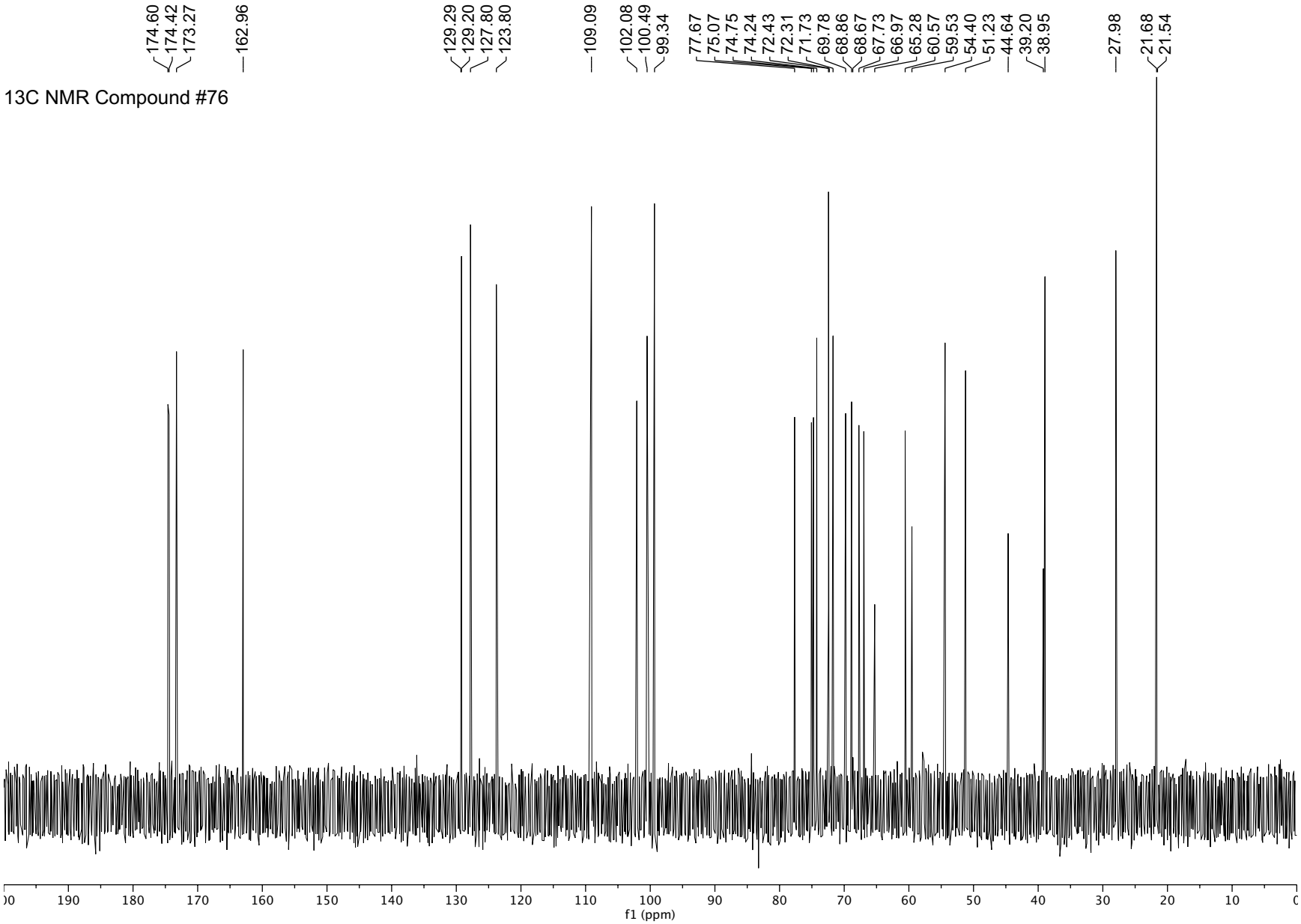
1H NMR Compound #65



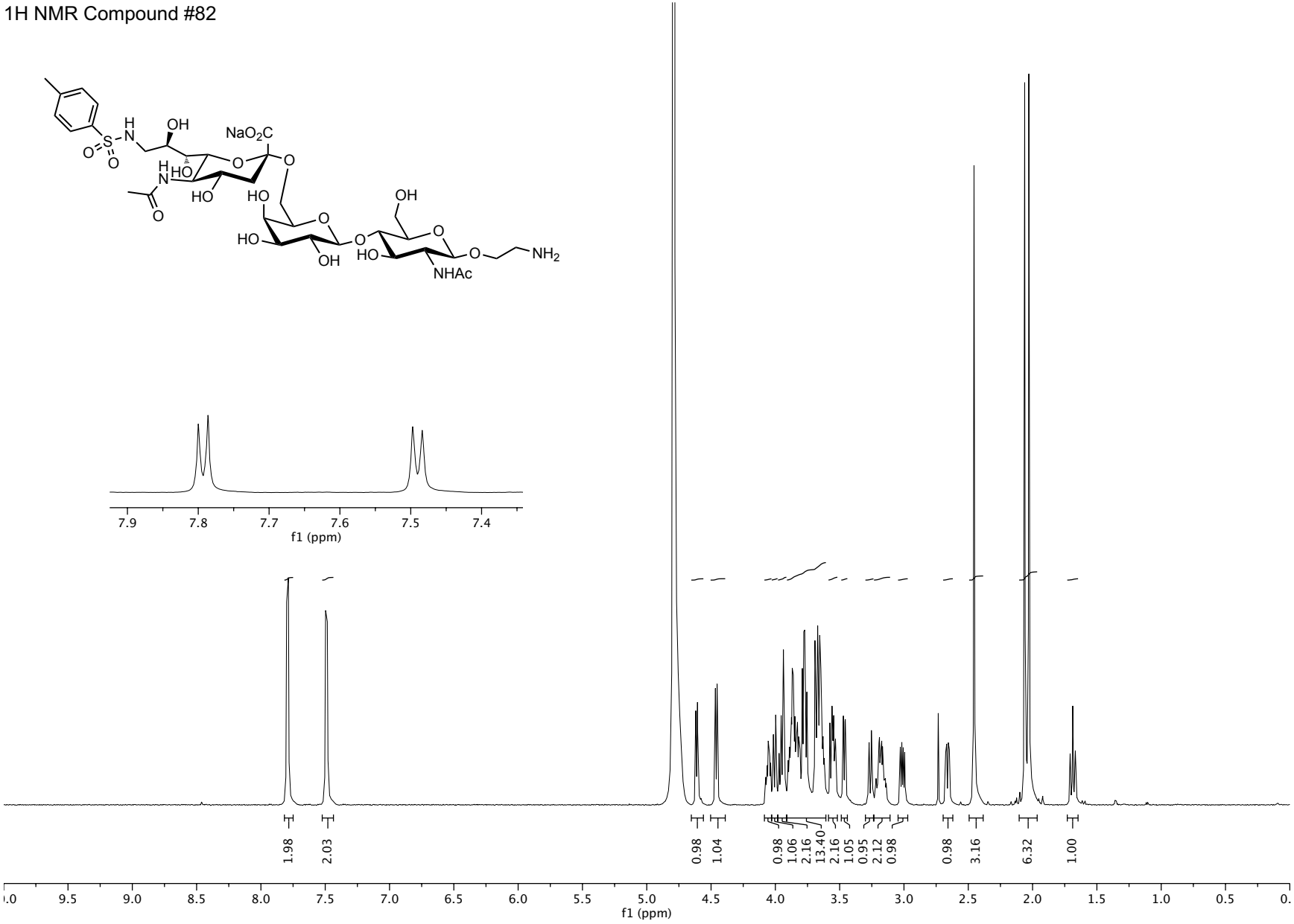

174.61
174.42
173.26
166.68
146.70
131.65
126.00
123.04
116.78
114.53
102.13
100.50
99.33
77.74
75.08
74.76
74.25
72.32
71.73
69.77
68.84
68.67
67.69
66.94
66.08
65.41
60.56
59.55
54.39
51.25
44.61
39.23
38.96
21.68
21.53
13C NMR Compound #65
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

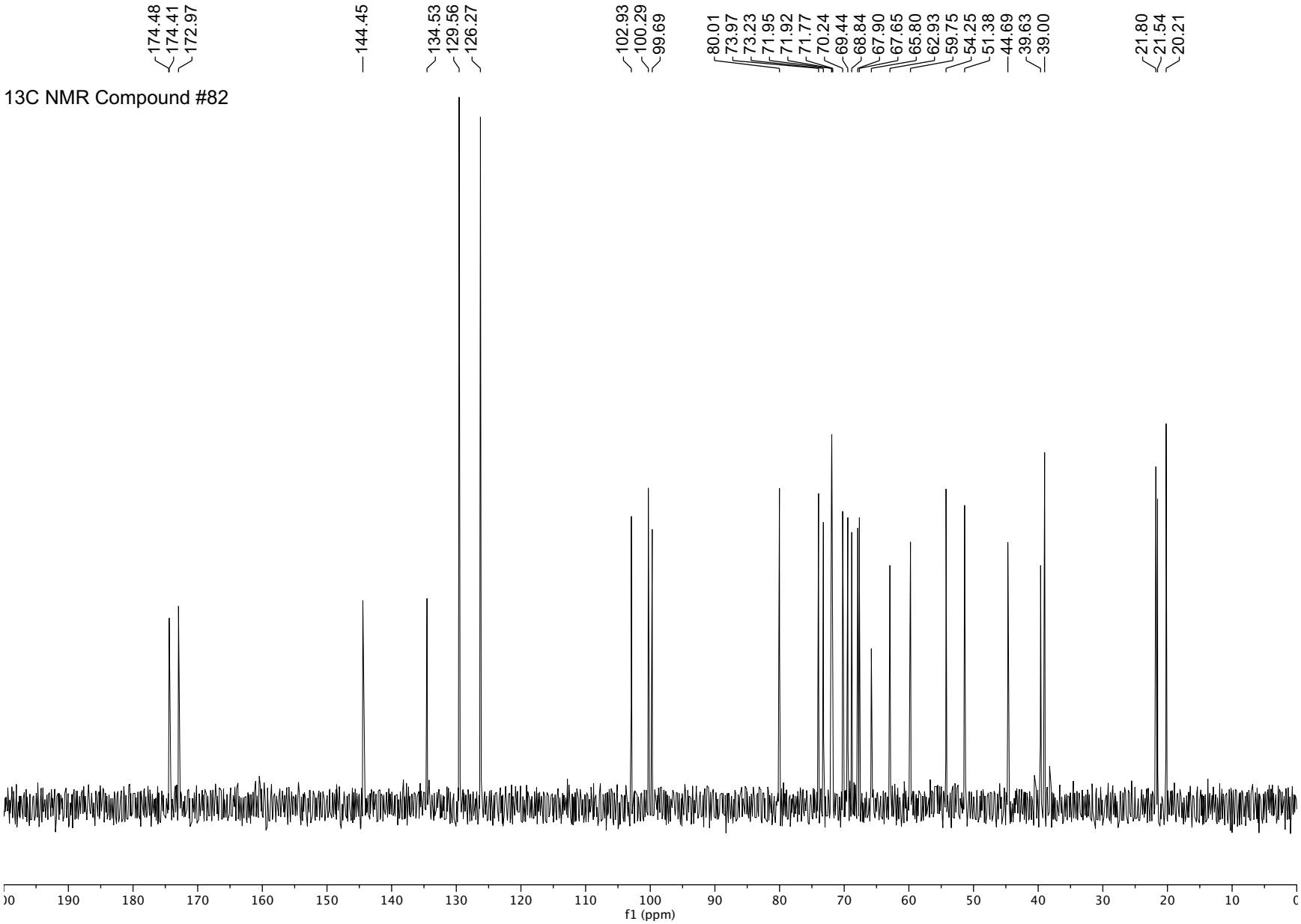
1H NMR Compound #76



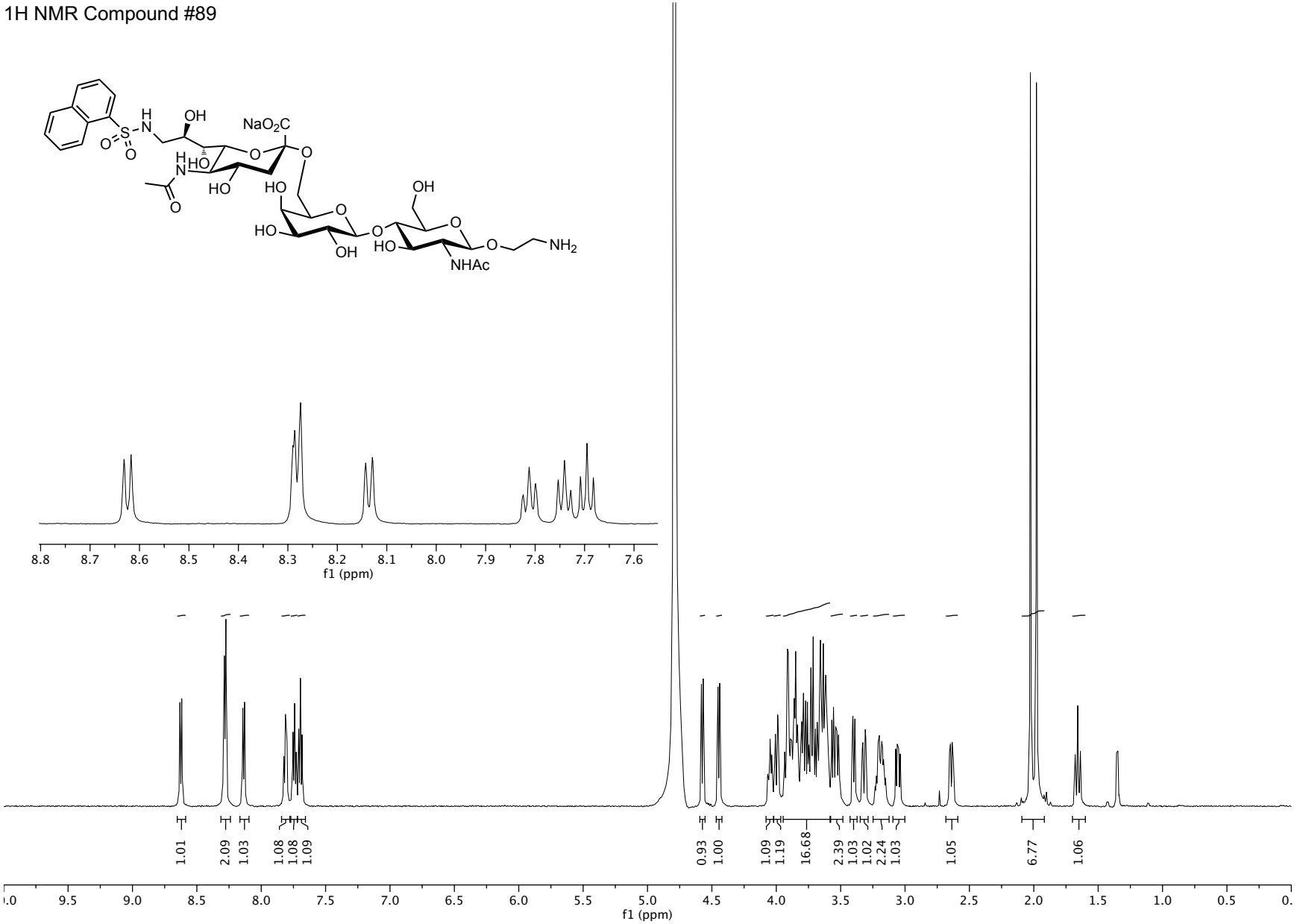

174.60
174.42
173.27
162.96
129.29
129.20
127.80
123.80
109.09
102.08
100.49
99.34
77.67
75.07
74.75
74.24
72.43
72.31
71.73
69.78
68.86
68.67
67.73
66.97
65.28
60.57
59.53
54.40
51.23
44.64
39.20
38.95
27.98
21.68
21.54
13C NMR Compound #76
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

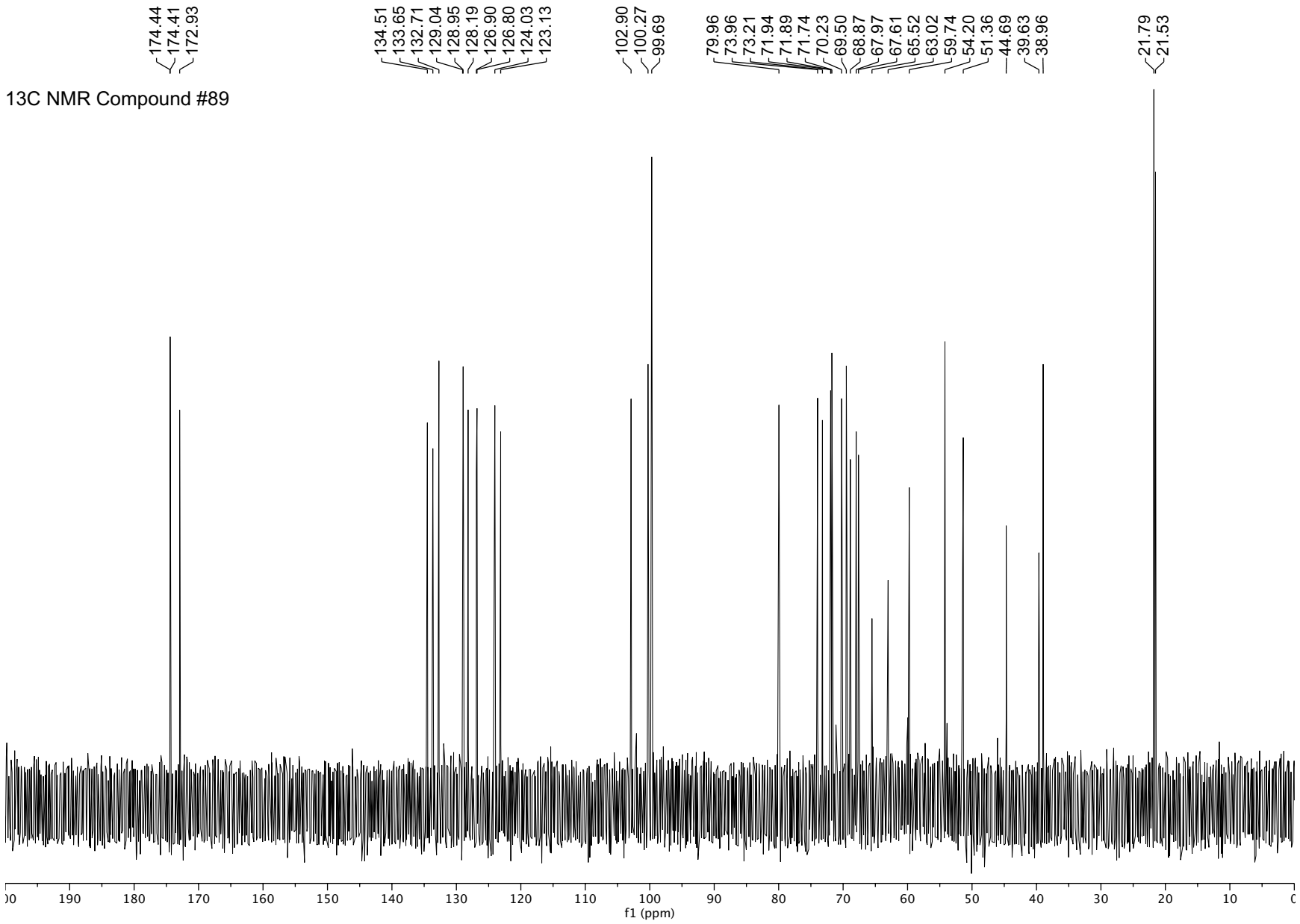
1H NMR Compound #82



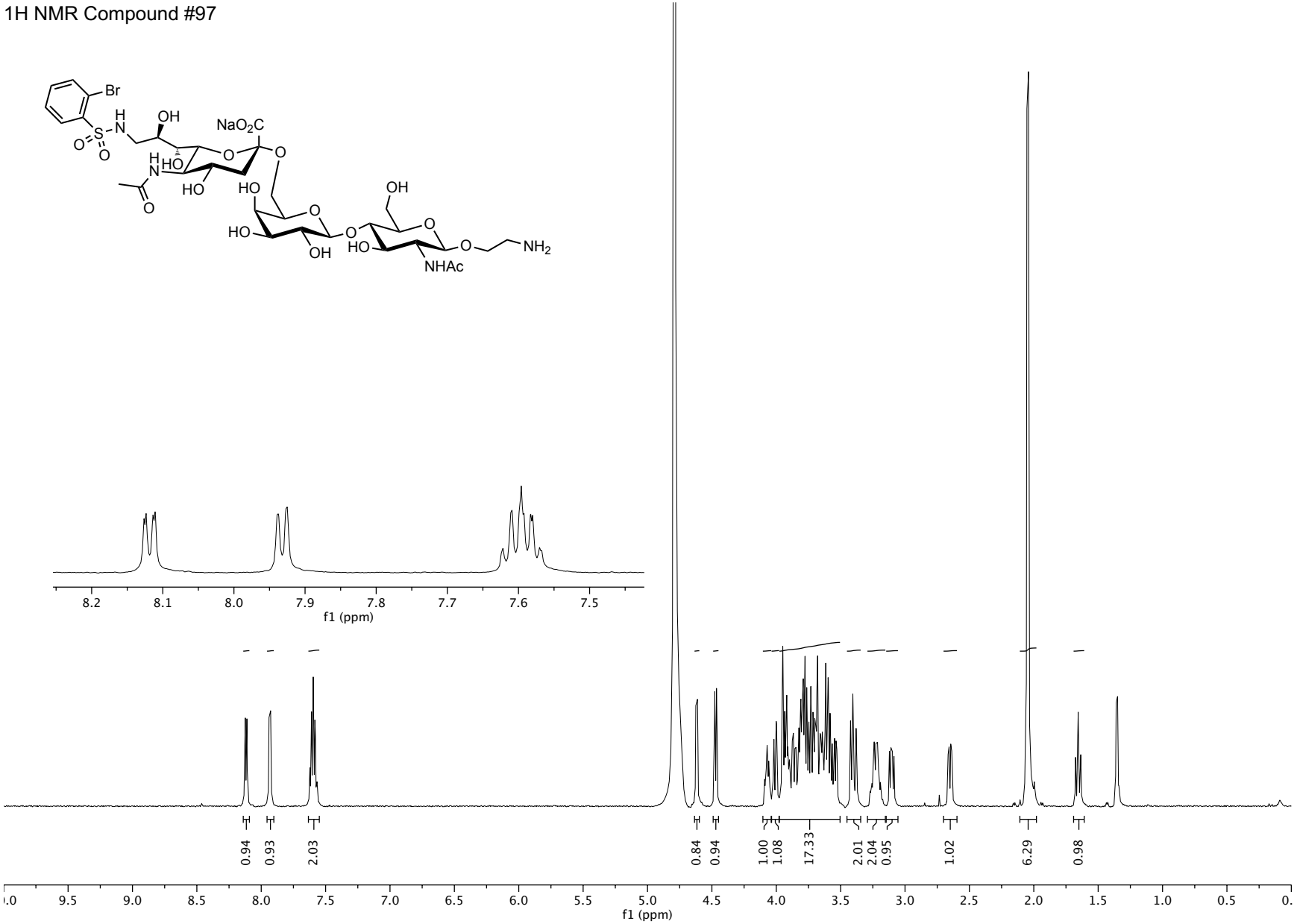

174.48
174.41
172.97
144.45
134.53
129.56
126.27
102.93
100.29
99.69
80.01
73.97
73.23
71.95
71.92
71.77
70.24
69.44
68.84
67.90
67.65
65.80
62.93
59.75
54.25
51.38
44.69
39.63
39.00
21.80
21.54
20.21
13C NMR Compound #82
f1 (ppm)

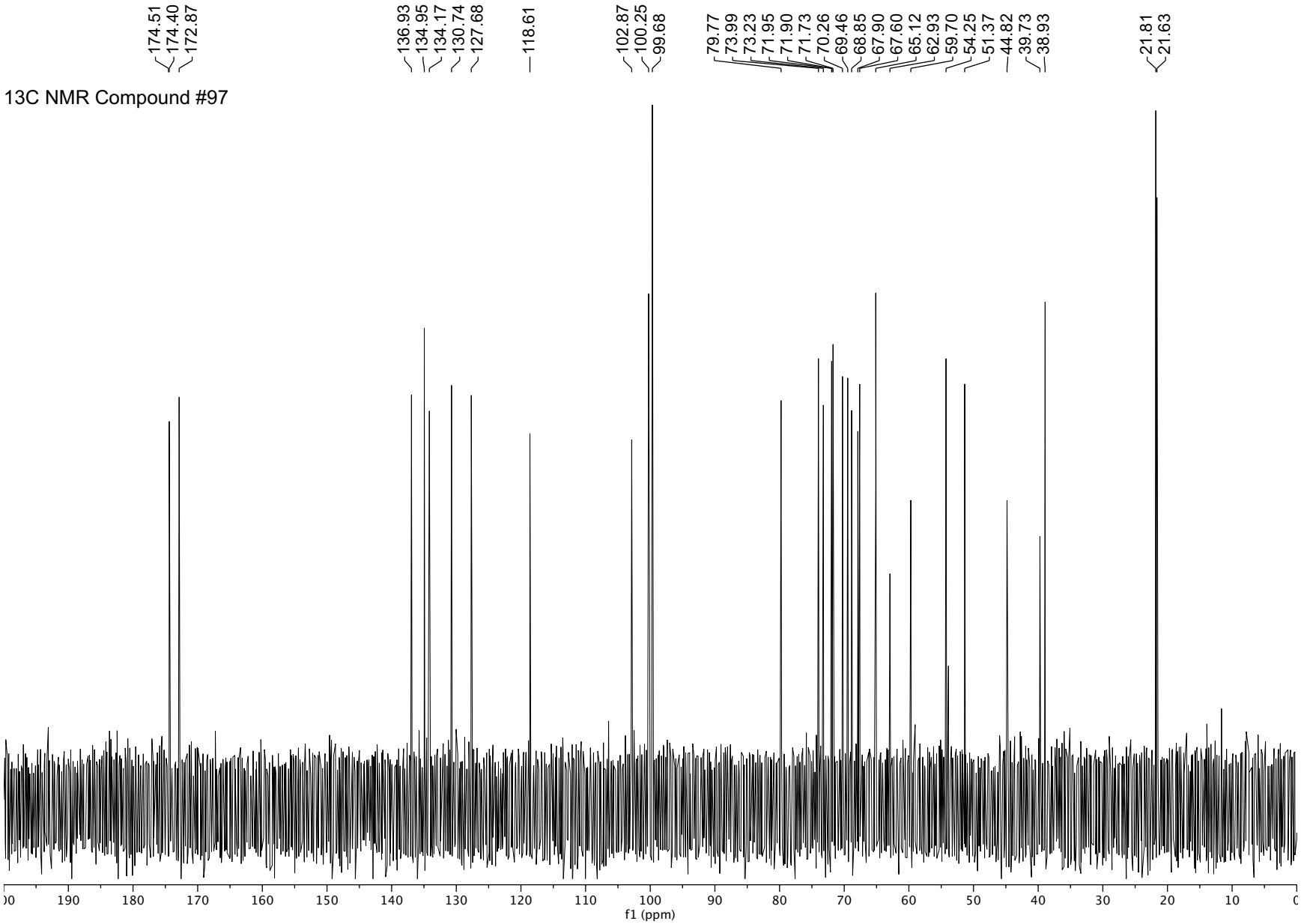
1H NMR Compound #89



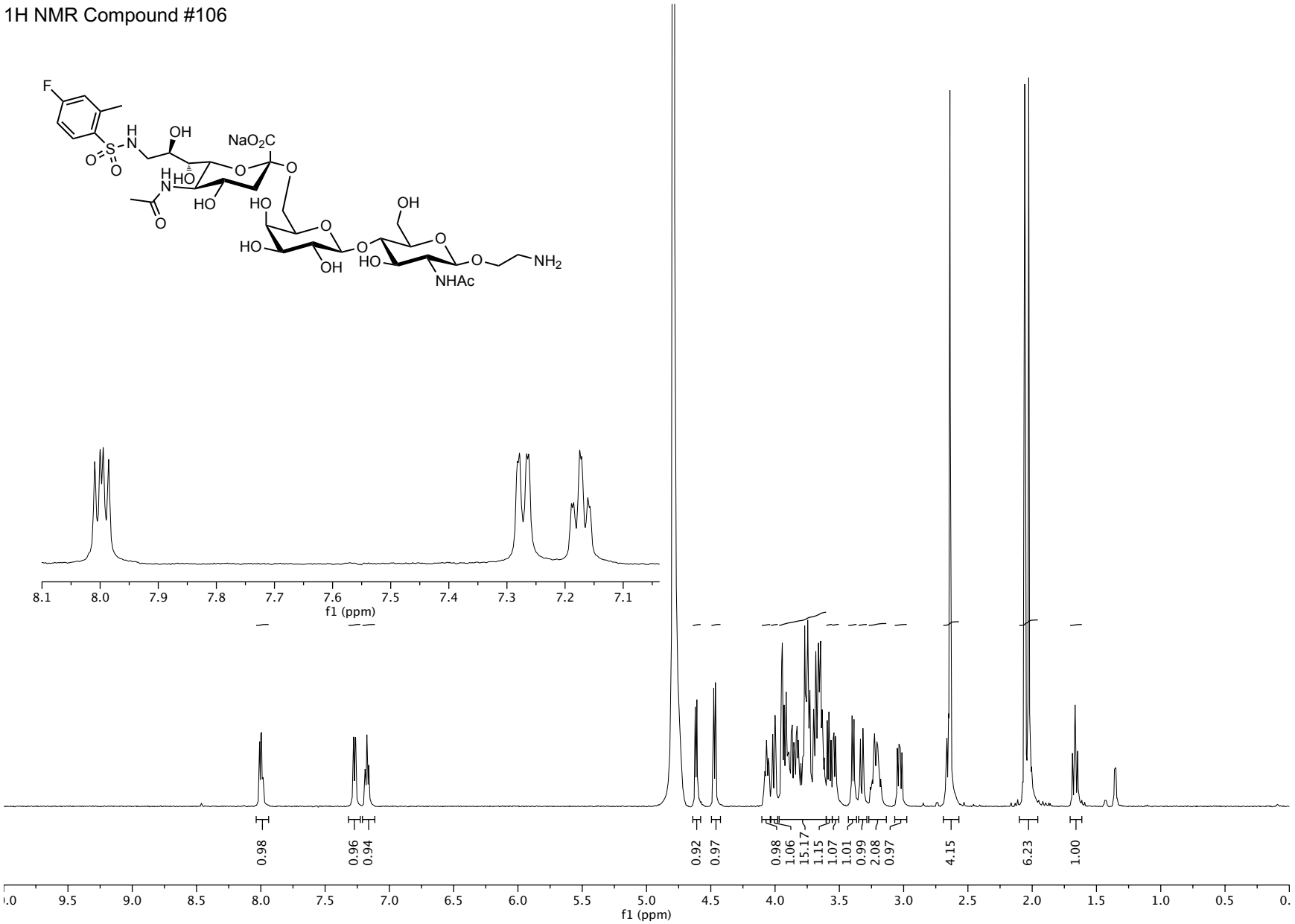

174.44
174.41
172.93
134.51
133.65
132.71
129.04
128.95
128.19
126.90
126.80
124.03
123.13
102.90
100.27
99.69
79.96
73.96
73.21
71.94
71.89
71.74
70.23
69.50
68.87
67.97
67.61
65.52
63.02
59.74
54.20
51.36
44.69
39.63
38.96
21.79
21.53
13C NMR Compound #89
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

1H NMR Compound #97

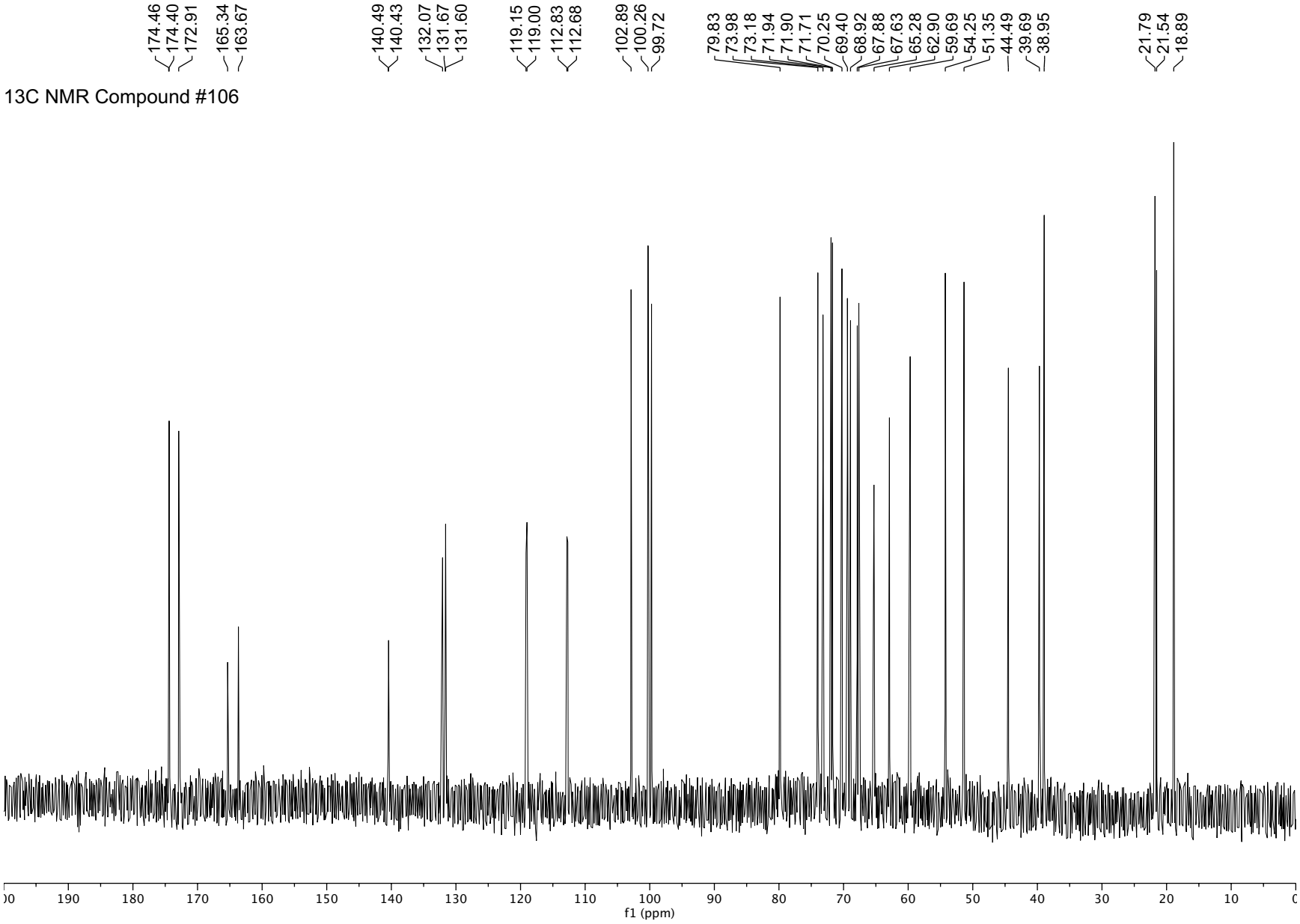



174.51
174.40
172.87
136.93
134.95
134.17
130.74
127.68
118.61
102.87
100.25
99.68
79.77
73.99
73.23
71.95
71.90
71.73
70.26
69.46
68.85
67.90
67.60
65.12
62.93
59.70
54.25
51.37
44.82
39.73
38.93
21.81
21.63
13C NMR Compound #97
f1 (ppm)

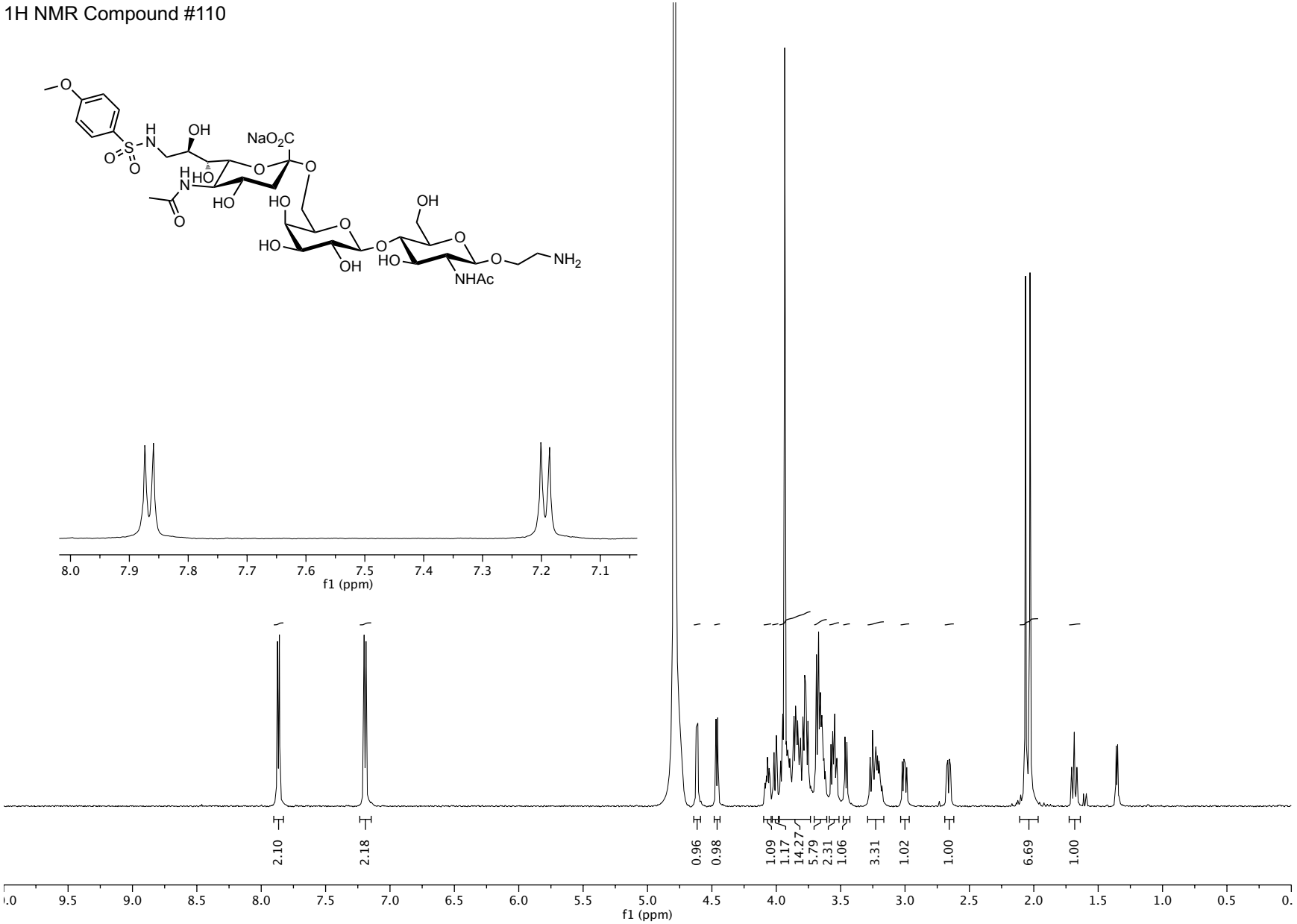
1H NMR Compound #106

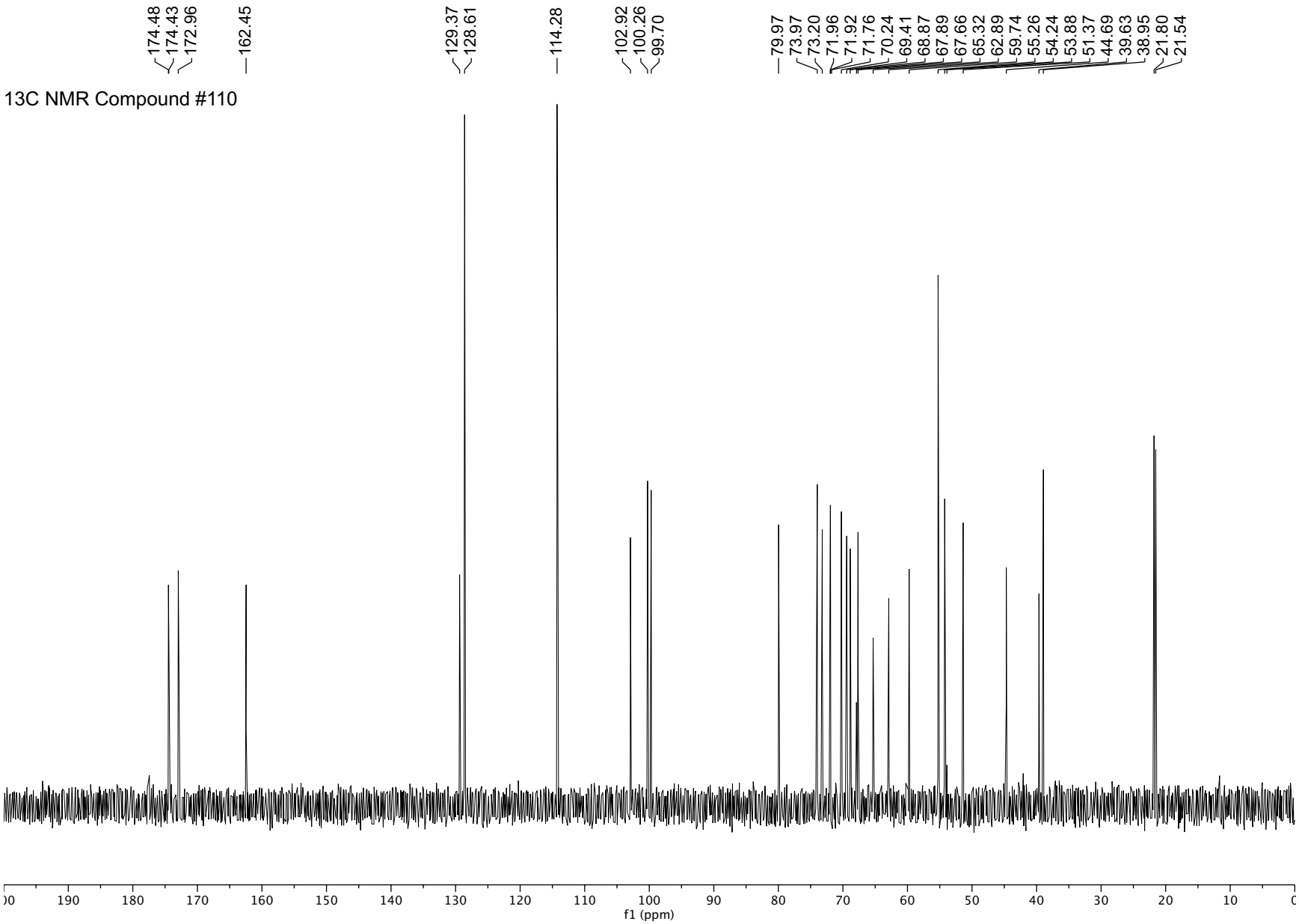


13C NMR Compound #106

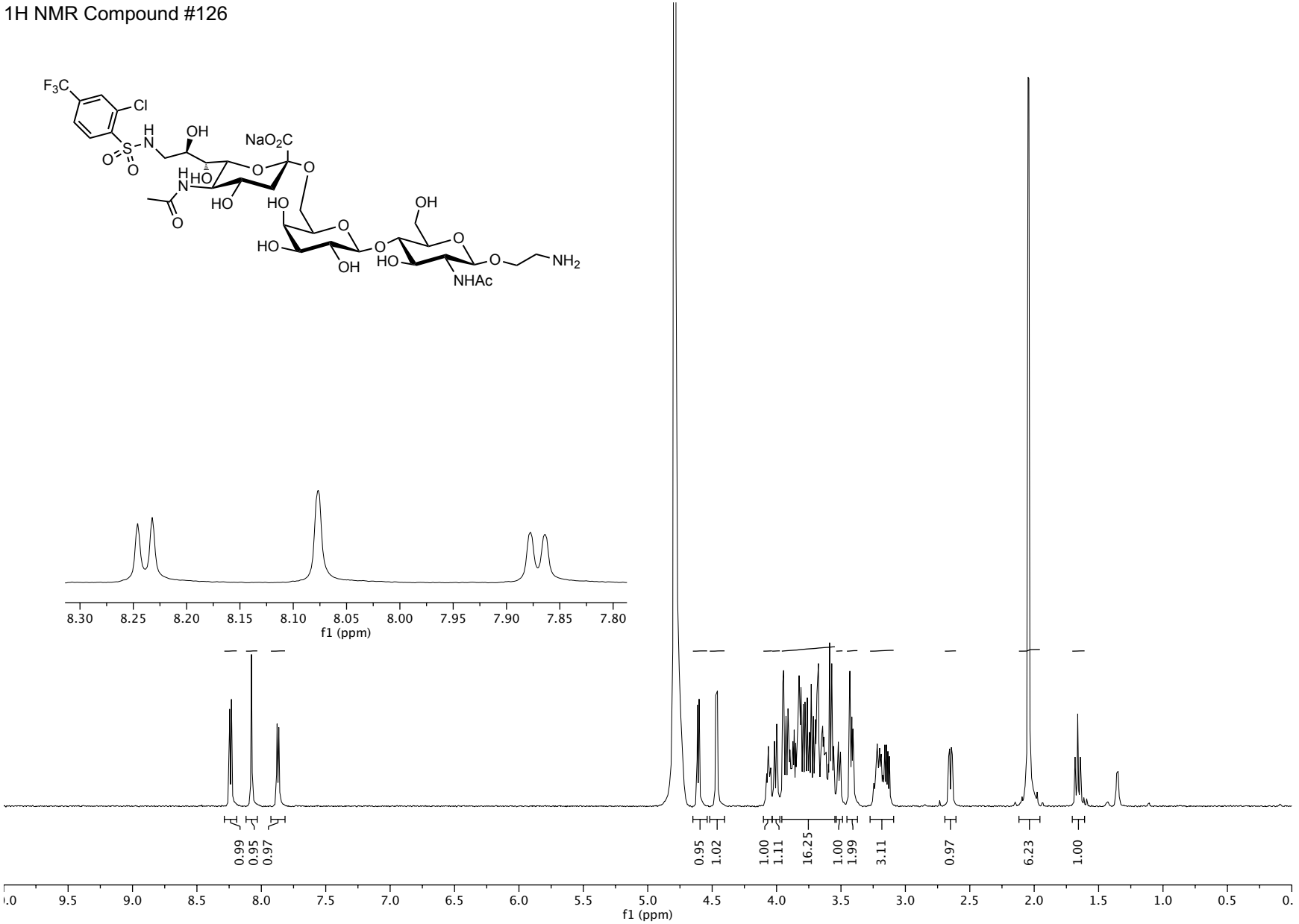


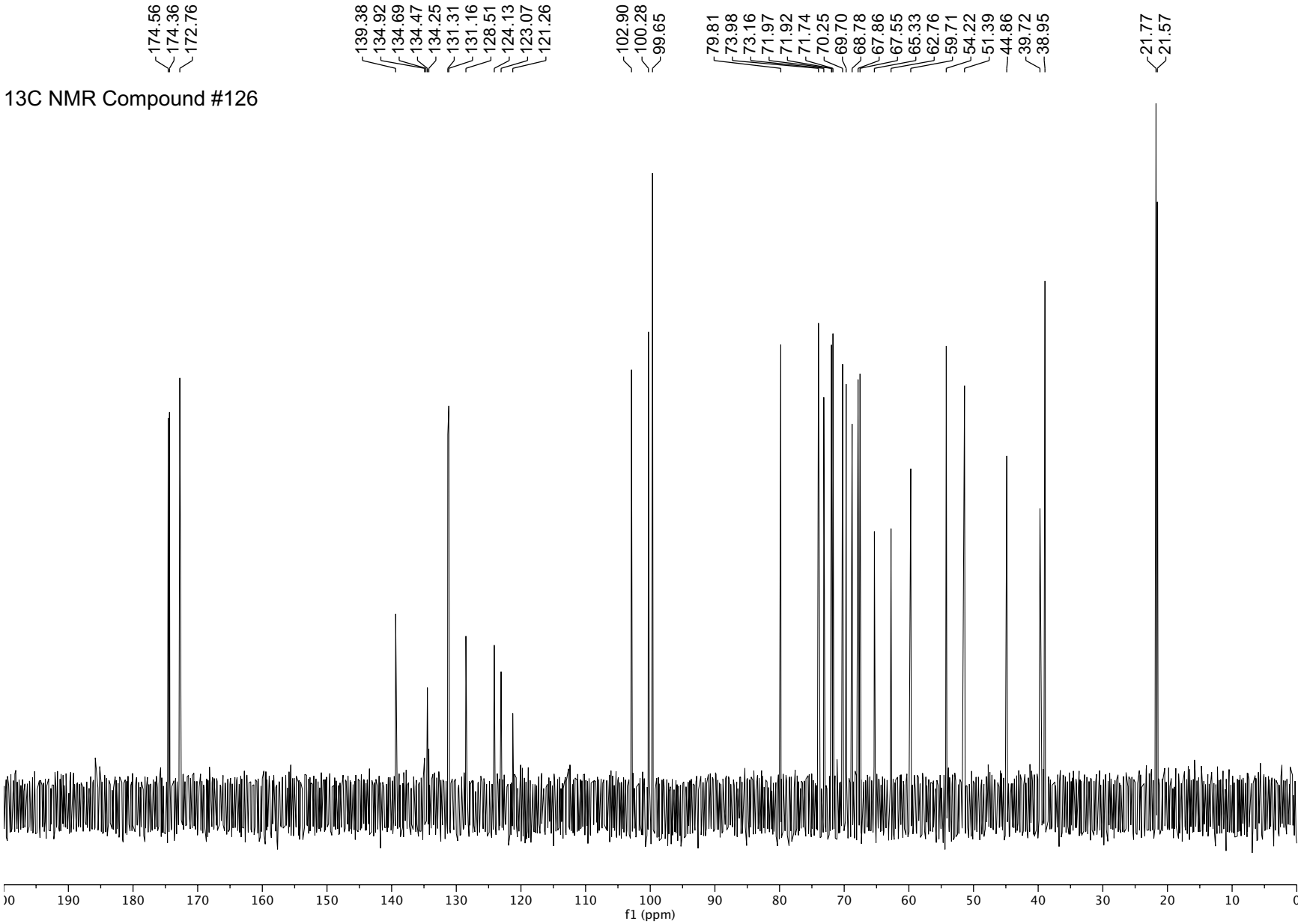
1H NMR Compound #110



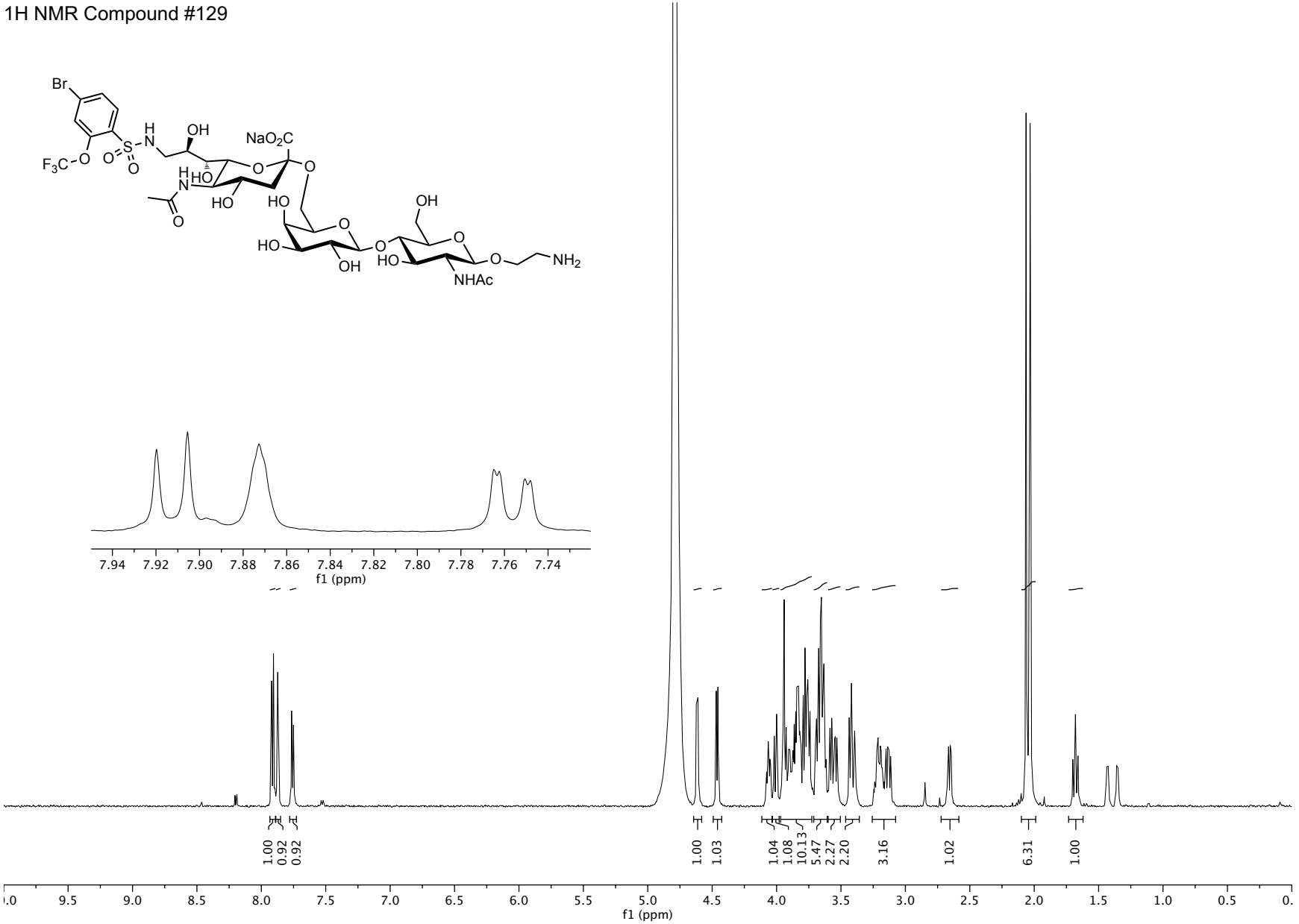

174.48
174.43
172.96
162.45
129.37
128.61
114.28
102.92
100.26
99.70
79.97
73.97
73.20
71.96
71.92
71.76
70.24
69.41
68.87
67.89
67.66
65.32
62.89
59.74
55.26
54.24
53.88
51.37
44.69
39.63
38.95
21.80
21.54
13C NMR Compound #110
f1 (ppm)

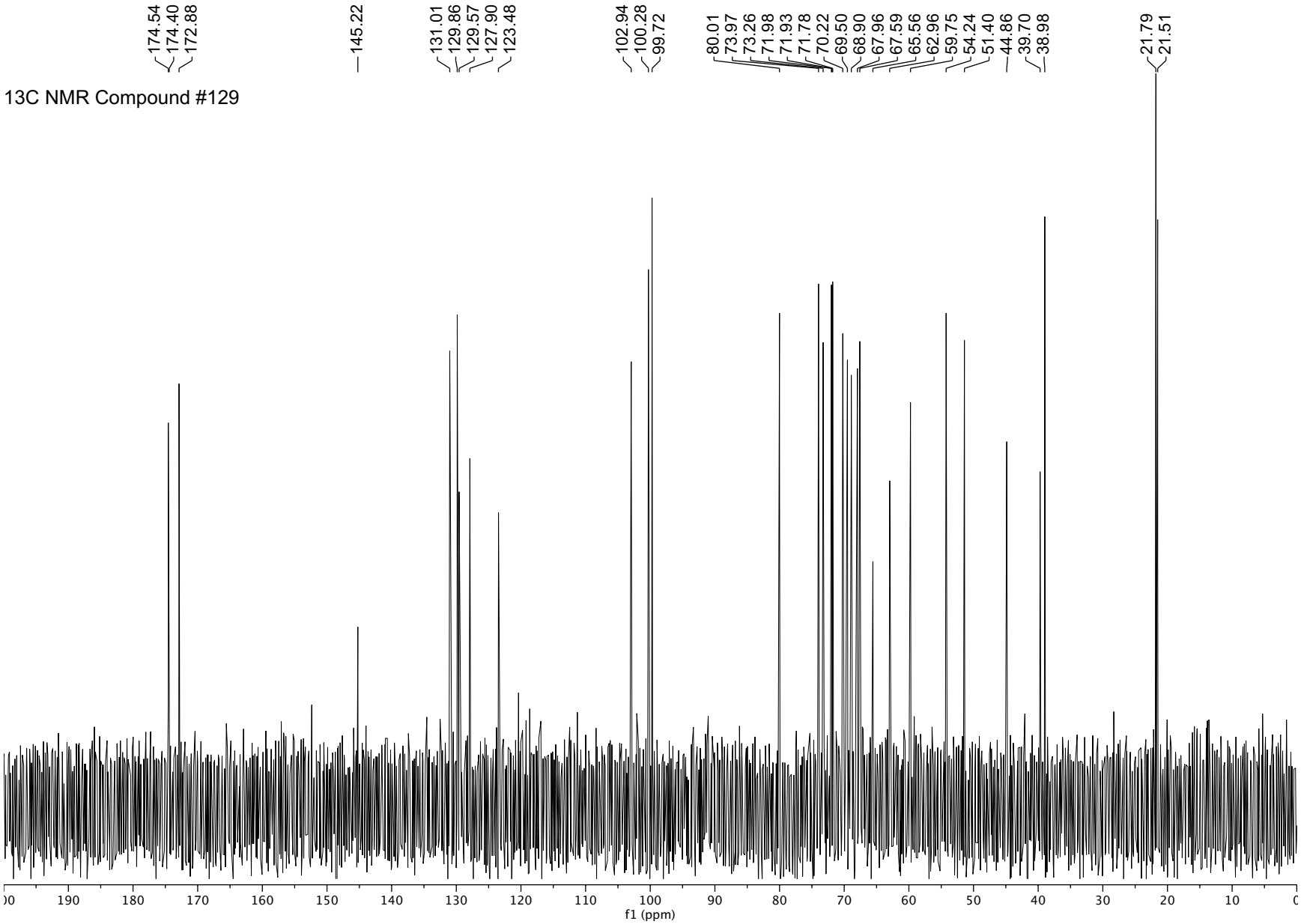
1H NMR Compound #126




174.56
174.36
172.76
139.38
134.92
134.69
134.47
134.25
131.31
131.16
128.51
124.13
123.07
121.26
102.90
100.28
99.65
79.81
73.98
73.16
71.97
71.92
71.74
70.25
69.70
68.78
67.86
67.55
65.33
62.76
59.71
54.22
51.39
44.86
39.72
38.95
21.77
21.57
13C NMR Compound #126
f1 (ppm)

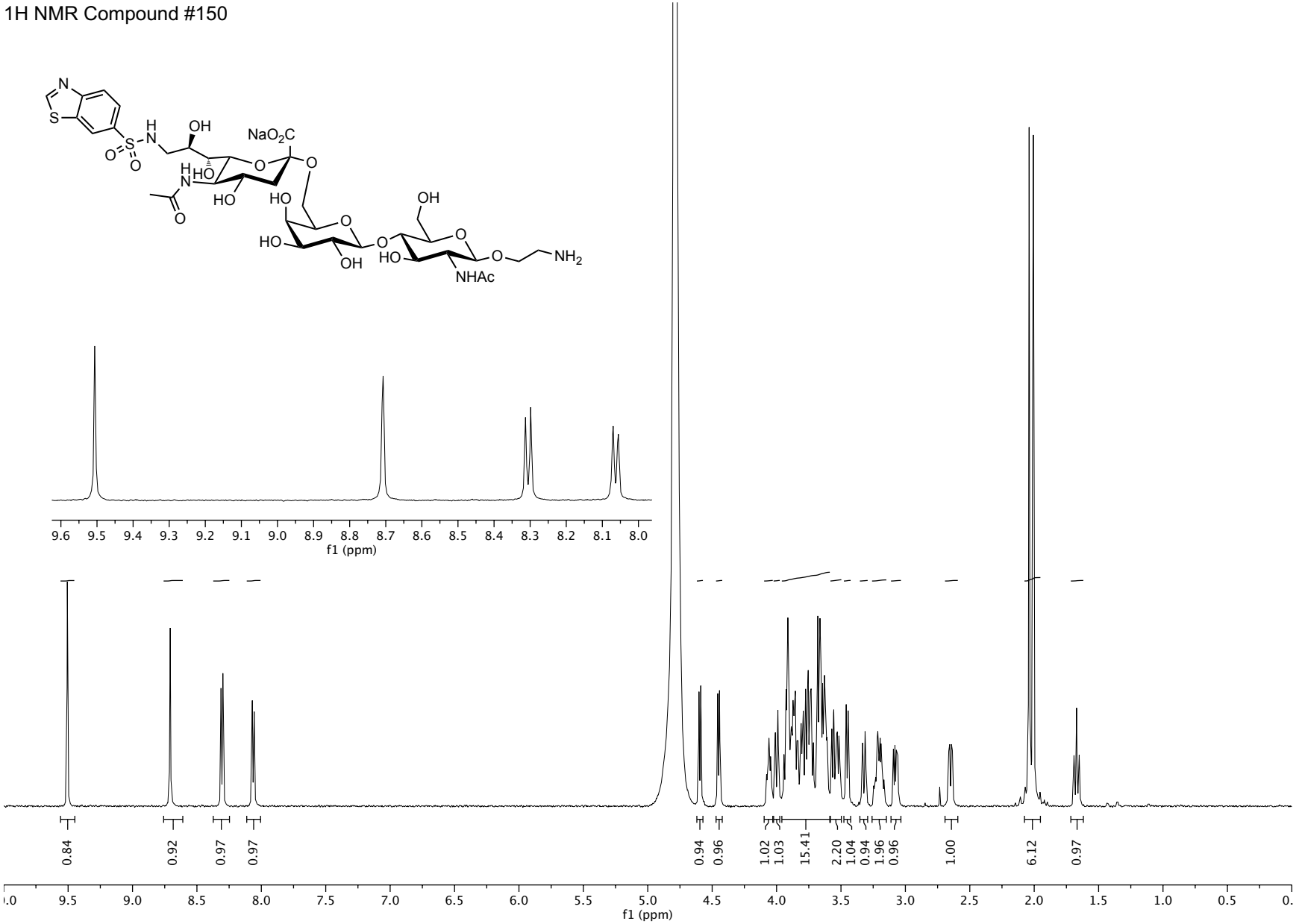
1H NMR Compound #129

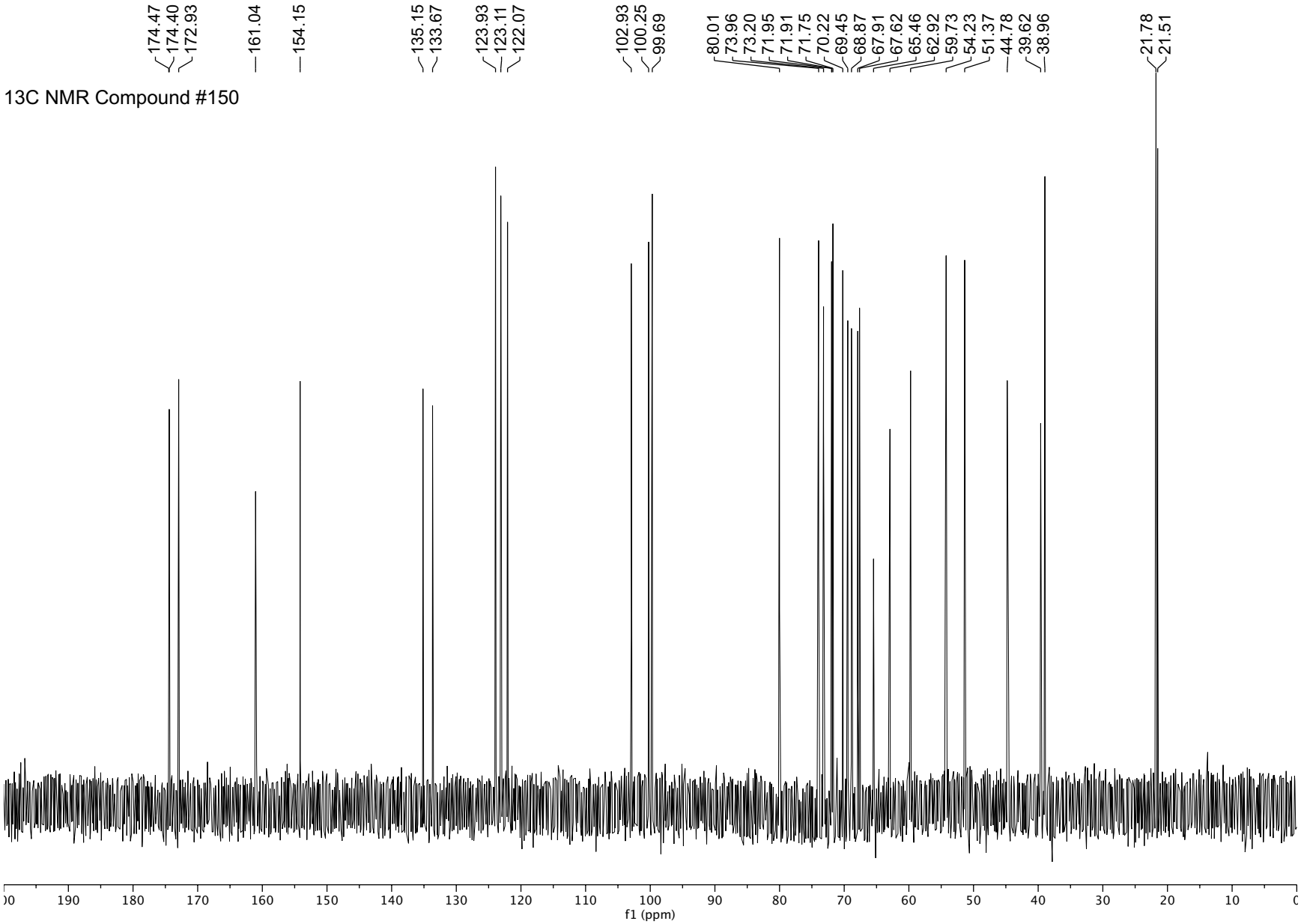




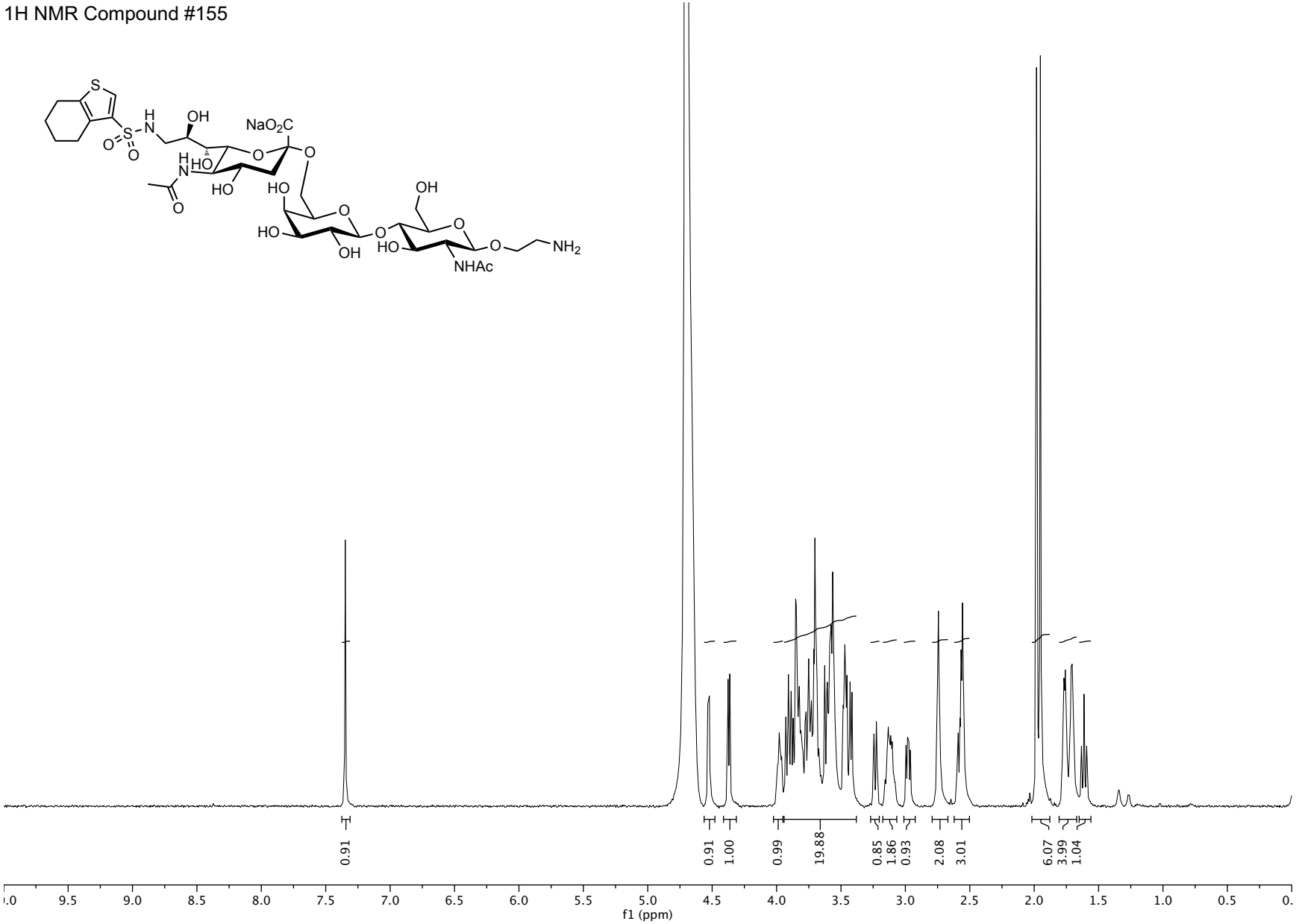
13C NMR Compound #129

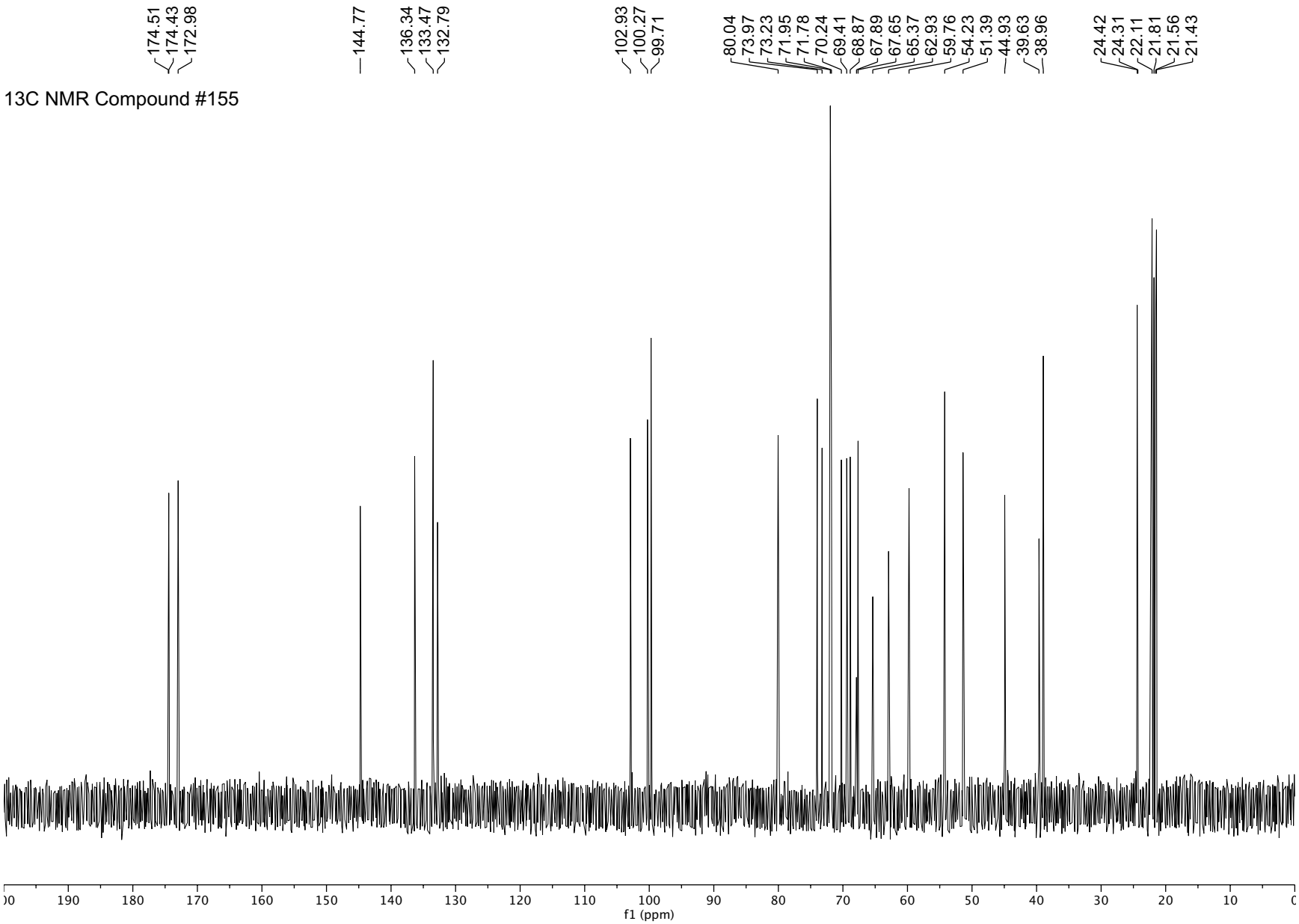
1H NMR Compound #150



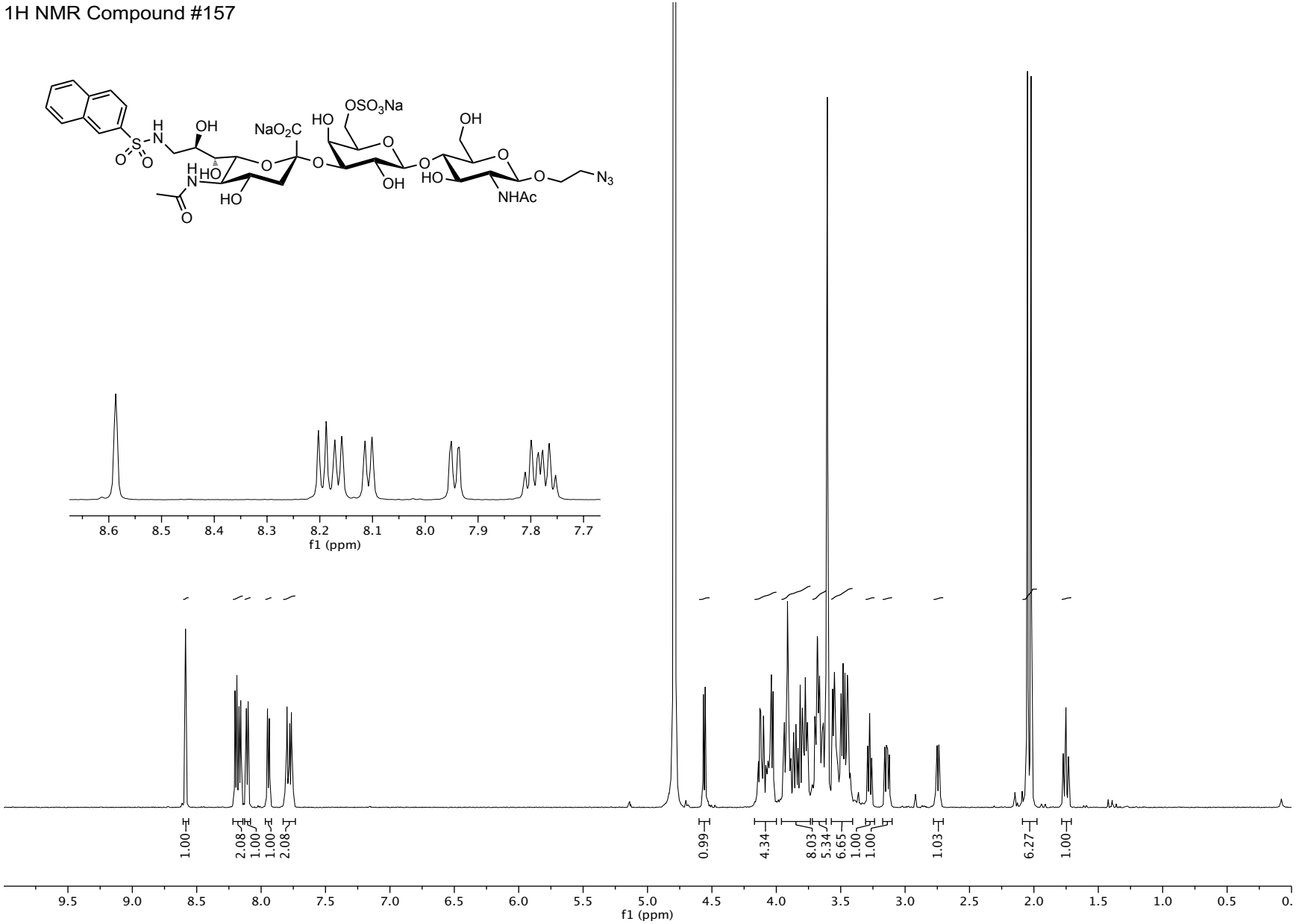

174.47
174.40
172.93
161.04
154.15
135.15
133.67
123.93
123.11
122.07
102.93
100.25
99.69
80.01
73.96
73.20
71.95
71.91
71.75
70.22
69.45
68.87
67.91
67.62
65.46
62.92
59.73
54.23
51.37
44.78
39.62
38.96
21.78
21.51
13C NMR Compound #150
f1 (ppm)

1H NMR Compound #155



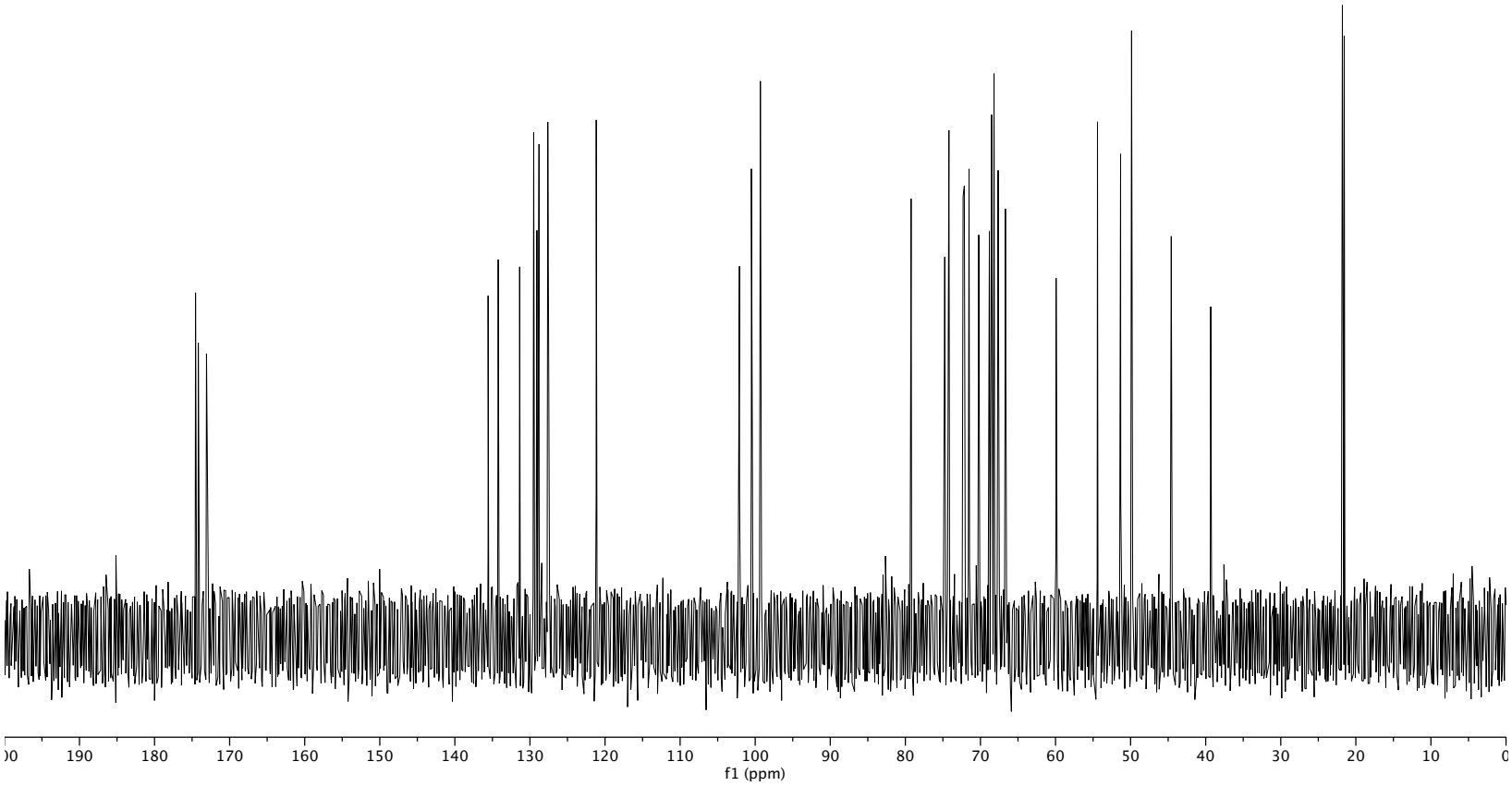

174.51
174.43
172.98
144.77
136.34
133.47
132.79
102.93
100.27
99.71
80.04
73.97
73.23
71.95
71.78
70.24
69.41
68.87
67.89
67.65
65.37
62.93
59.76
54.23
51.39
44.93
39.63
38.96
24.42
24.31
22.11
21.81
21.56
21.43
13C NMR Compound #155
f1 (ppm)

1H NMR Compound #157

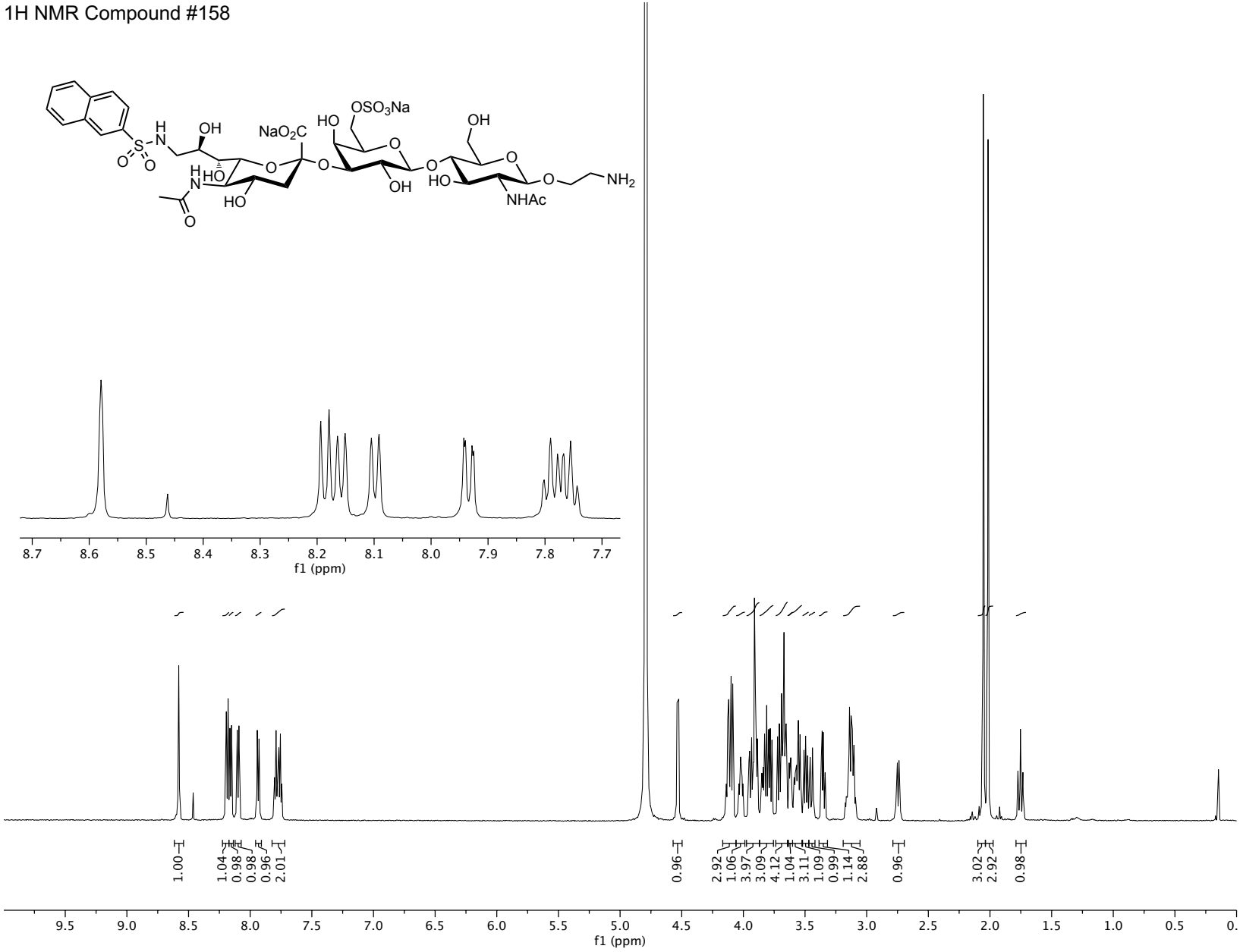


174.54
174.17
173.08
135.57
134.23
131.37
129.50
129.08
128.79
127.69
127.62
127.51
121.17
102.10
100.51
99.31
79.23
74.75
74.20
72.31
72.15
71.52
70.23
68.81
68.51
68.19
67.65
66.67
66.57
59.92
54.41
51.33
49.87
44.59
39.34
21.79
21.55

13C NMR Compound #157

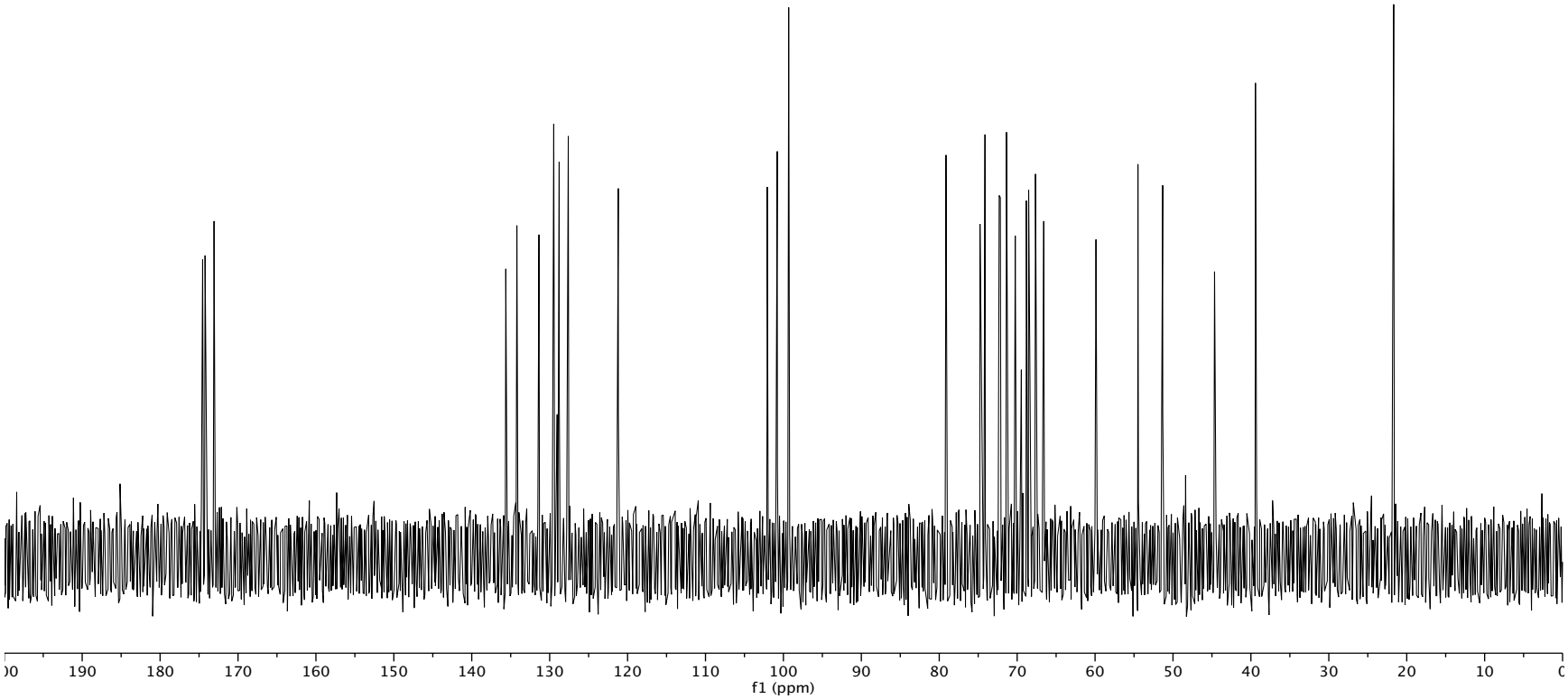


1H NMR Compound #158

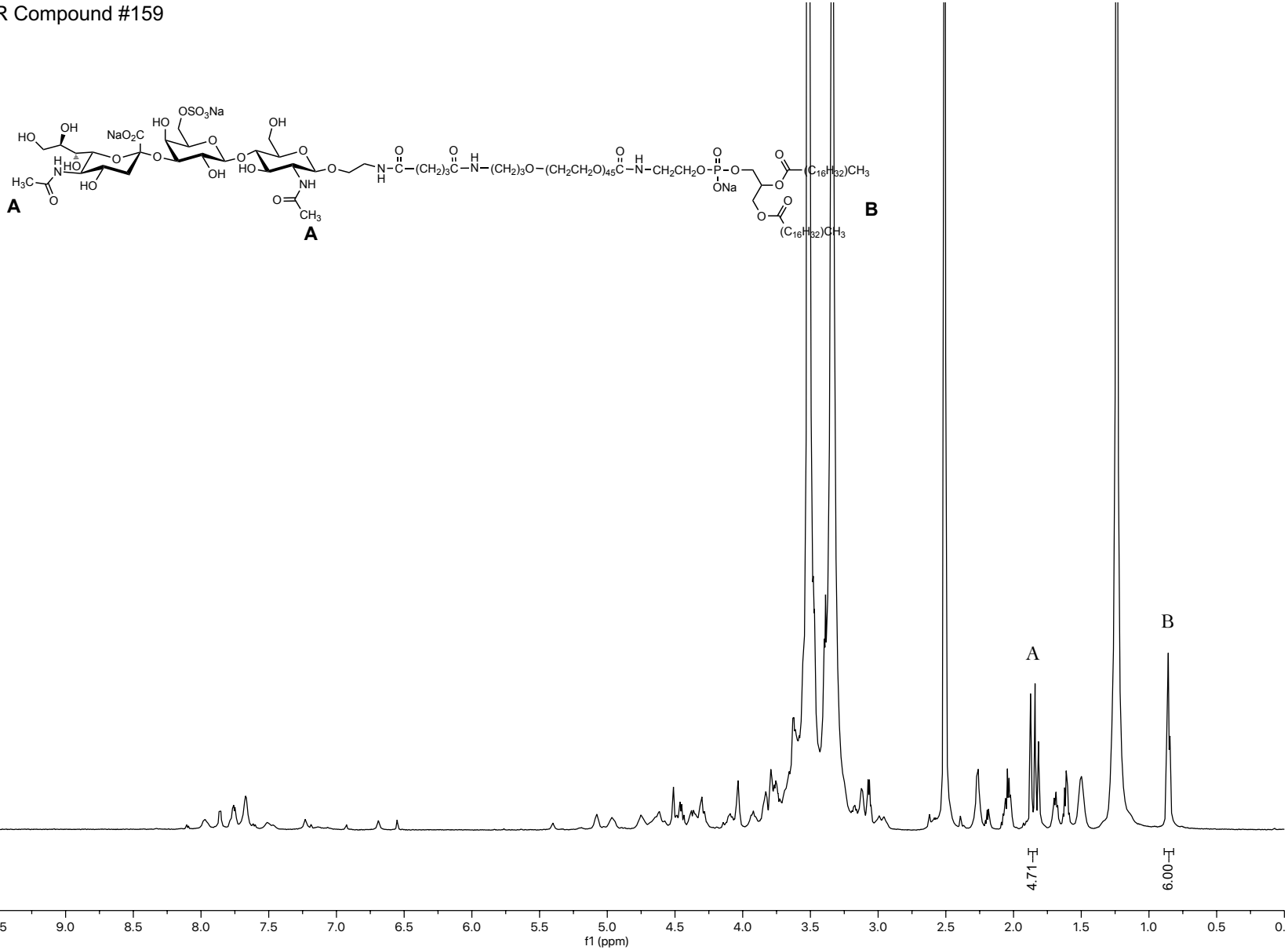


174.54
174.23
173.08
135.63
134.21
131.37
129.48
129.03
128.78
127.65
127.61
127.49
121.19
102.08
100.79
99.32
79.13
74.76
74.13
72.31
72.16
71.37
70.24
69.47
68.82
68.52
67.65
66.73
66.59
59.88
54.49
51.32
44.67
39.42
39.32
21.67
21.54

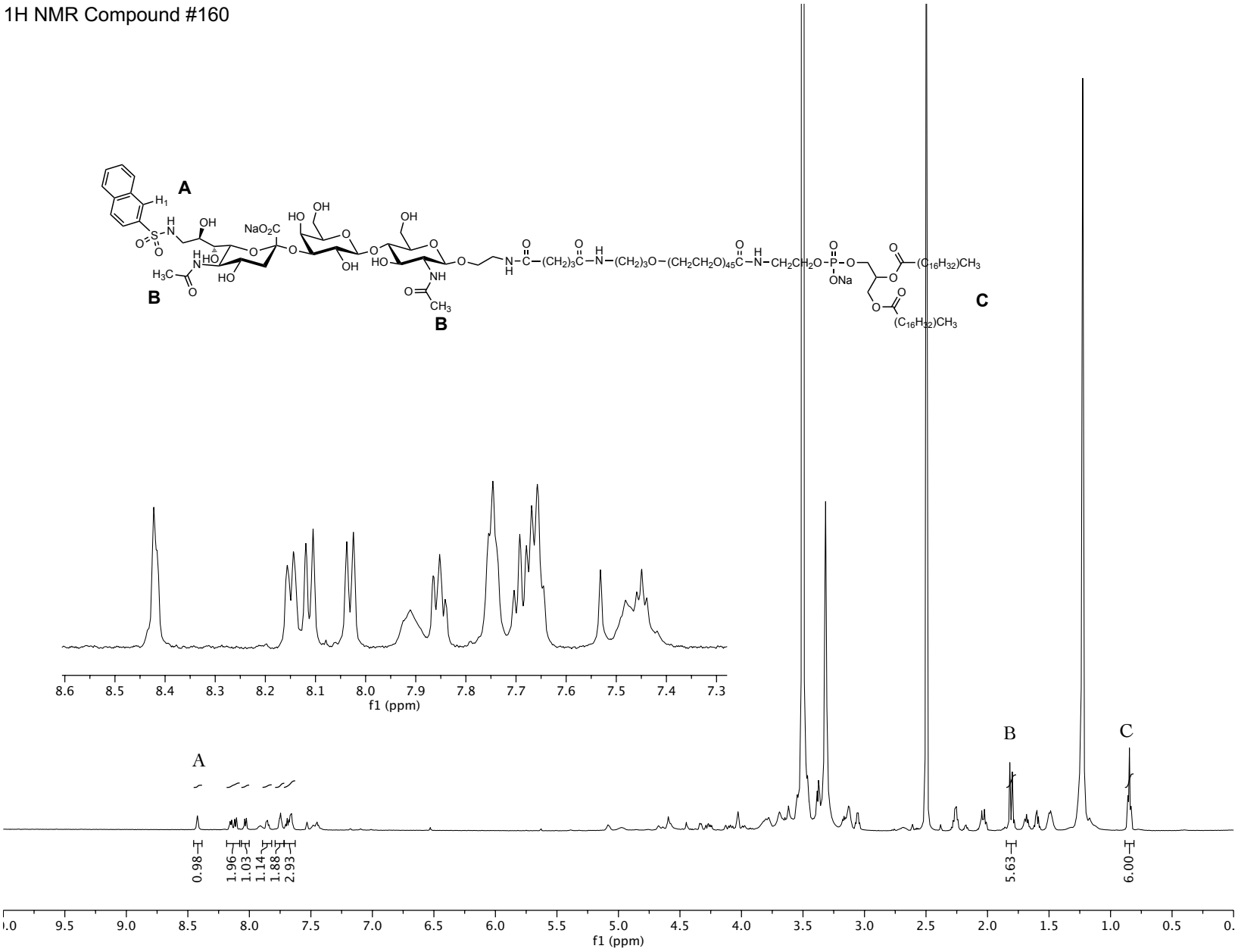
13C NMR Compound #158



1H NMR Compound #159



1H NMR Compound #160



1H NMR Compound #161