Reaction of N-Acetylneuraminic Acid Derivatives with Perfluorinated Anhydrides: a Short Access to N-Perfluoracylated Glycals with Antiviral Properties

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Supporting Information

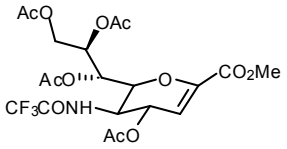
1. General Methods

Solvents were dried using standard methods and distilled before use. The reactions are thermostated by block heater -BBA- Grant Boekel apparatus. The progress of all reactions was monitored by thin-layer chromatography (TLC) carried out on 0.25 mm E. Merck silica gel plates (60 F_{254}) using UV light, 50% sulphuric acid, anisaldehyde/H_2SO_4/EtOH solution or 0.2% ninhydrin in ethanol and heat as developing agent. The flash chromatography was performed with normal phase silica gel (E. Merck 230-400 mesh silica gel), following the general protocol of Still[1]. GLC was performed by Hewlett 5890 PACKARD Series II using HP-5 30 m x 0.32 mm, 0.25 μm film-thickness column. Melting points were measured on a SMP3 mp apparatus (Stuart Scientific, USA) and are not corrected. NMR spectra were recorded at 25°C on a Bruker AM-500 spectrometer operating at 500.13 MHz for ^1H and 125.76 MHz for ^{13}C. The chemical shifts are reported in ppm and coupling constants are given in Hz, relative to CD_3OD signal fixed at 3.31 ppm for ^1H spectra and to CD_3OD signal fixed at 49.05 ppm for ^{13}C spectra, relative to $CDCl_3$ signal fixed at 7.26 ppm for ^1H spectra and to $CDCl_3$ signal fixed at 77.00, relative to CD_3CN signal fixed at 1.94 ppm for ^1H spectra and to CD_3CN signal fixed at 1.24 ppm for ^{13}C spectra. Proton and carbon assignments were established, if necessary, with ^1H-^1H and ^1H-^{13}C correlated NMR experiments. Data for ^1H NMR are recovered as follows: chemical shift (ppm), multiplicity (s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; br , broad), coupling constant(s) in Hz, number of protons, assignment of proton(s). In some cases, reported in the text, the ^1H NMR inspection was performed on the total reaction mixture using CD_3CN as a solvent. Optical rotations were taken on a Perkin-Elmer 241 polarimeter equipped with a 1 dm tube; $[\alpha]_D$ values are given in 10^{-1}deg cm^2 g^{-1} and the concentrations are given in g/100 mL.
Infrared (IR) spectra were recorded for CH_2Cl_2 solution using a Perkin-Elmer 1420 instrument. Mass spectrometry was performed using Finnigan LCQ_{Deca} quadrupole ion-trap mass spectrometer equipped with an ESI ion source (Finnigan ThermoQuest, San Jose, CA, USA). The spectra were collected in continuous flow mode by connecting the infusion pump directly to the ESI source. Solutions of compounds were infused at a flow rate of 5 μL/min. The spray voltage was set at 5.0 kV in the positive and at 4.5 kV in the negative ion mode with a capillary temperature of 220 °C. Full-scan mass spectra were recorded by scanning a m/z range of 100-2000.
Work-up refers to successive washing of the organic layer with an ice cold aqueous $NaHCO_3$ saturated solution and water, to drying over Na_2SO_4, and evaporation of the solvent under reduced pressure.

2. Preparation of *N*-Perfluoroacylneuraminic Acid Glycals: General Procedure

The acylamide (0.2 mmol) dissolved in CH_3CN (0.60 mL) was reacted with the appropriate perfluorinated anhydride (0.6-1.4 mmol, 3-7 molar equivalents) at 135 °C for 5-15 min in a sealed tube. Then the reaction mixture was cooled, added of methanol (0.20 mL) and evaporated under reduced pressure to afford a crude residue which, after usual work-up, and rapid chromatography, using the designed solvent system, afforded the appropriate glycal. This procedure was used both for the exclusive *N*-transacylation and for the *N*-transacylation coupled with elimination reaction.

2.1 Glycal 5: 4,7,8,9-tetra-*O*-Acetyl-2,3-dehydro-2-deoxy-5-*N*-(2,2,2-trifluoroacetyl)-β-neuraminic acid methyl ester



5

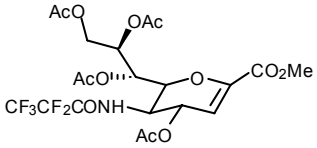
a) *By N-transacylation from the glycal* **3**

Starting with the glycal **3**[2] (95 mg; 0.20 mmol), glycal **5** was obtained (83 mg, 79 % yield), after a 10 min reaction with TFAA and flash chromatography (eluting with hexane/AcOEt; 6:4, v/v). Glycal **5** showed: m.p. 115-116°C; $[\alpha]_D^{20}$ = + 50.8 (c = 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.21 (d, $J_{NH,2}$ = 9.0 Hz, 1H; N-H), 5.97 (d, $J_{3,4}$ = 2.6 Hz, 1H; H-3), 5.65 (dd, $J_{4,5}$ = 8.0, $J_{4,3}$ = 2.6 Hz, 1H; H-4), 5.47 (dd, $J_{7,8}$ = 4.3, $J_{7,6}$ = 3.0 Hz, 1H; H-7), 5.29 (m, 1H; H-8), 4.71 (dd, $J_{9a,9b}$ = 12.4, $J_{9a,8}$ = 2.6 Hz, 1H; H-9a), 4.49 (dd, $J_{6,5}$ = 9.7, $J_{6,7}$ = 3.0 Hz, 1H; H-6), 4.34 (m, 1H; H-5), 4.18 (dd, $J_{9b,9a}$ = 12.4, $J_{9b,8}$ = 7.3 Hz, 1H; H-9b), 3.81 (s, 3H; $COOCH_3$); 2.11 (s, 3H; CH_3COO at C-7), 2.06 (overlapping, 6H; 2XCH_3COO), 2.04 (s, 3H; CH_3COO); ^{13}C NMR ($CDCl_3$) δ 170.7 (3C, $CH_3\underline{C}OO$ at C-4, $CH_3\underline{C}OO$ at C-8 and $CH_3\underline{C}OO$ at C-9), 170.0 ($CH_3\underline{C}OO$ at C-7), 161.3 (C-1), 157.4 (q, $J_{C\text{-}F}$ = 38 Hz; $\underline{C}OCF_3$), 145.3 (C-2), 120.0-110.0 (1C, $\underline{C}F_3$), 107.7 (C-3), 75.9 (C-6), 71.3 (C-8), 67.4 (C-4 and C-7), 61.8 (C-9), 52.7 ($COO\underline{C}H_3$), 47.8 (C-5), 20.8 ($\underline{C}H_3COO$), 20.6 (2C, $\underline{C}H_3COO$), 20.5 ($\underline{C}H_3COO$); IR 1750,1724 1660 cm^{-1}; MS (ESI positive) *m/z* 550.2 $[M+Na]^+$, 1077.8 $[2M+Na]^+$.
Anal. Calcd for $C_{20}H_{24}F_3NO_{12}$: C, 45.55; H, 4.59; N, 2.66. Found: C, 45.65; H, 4.49; N, 2.53.

b) *By N-transacylation-elimination from 4,7,8,9-tetra-O-acetyl-5-N-acetyl-β-neuraminic acid methyl ester* **8**

Starting with 4,7,8,9-tetra-*O*-acetyl-5-*N*-acetyl-β-neuraminic acid methyl ester **8**[3] (107 mg, 0.2 mmol), after a 15 min reaction, with TFAA and flash chromatography (eluting with hexane/AcOEt; 6:4, v/v) glycal **5** was obtained (77 mg, 73% yield). Glycal **5** showed: m.p. 114-116°C; $[\alpha]_D^{20}$ = + 50.3 (c = 1, $CHCl_3$). All physico-chemical properties were superimposable to those of the compound described above.
Anal. Calcd for $C_{20}H_{24}F_3NO_{12}$: C, 45.55; H, 4.59; N, 2.66. Found: C, 45.58; H, 4.45; N, 2.62.

2.2 Glycal 6: 4,7,8,9-tetra-*O*-Acetyl-2,3-dehydro-2-deoxy-5-*N*-(2,2,3,3,3-pentafluoropropyonil) -β-neuraminic acid methyl ester



6

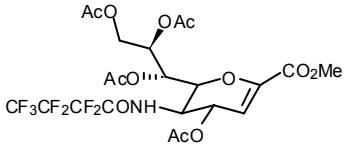
a) *By N-transacylation, from the glycal* **3**

Starting with the glycal **3**[2] (95 mg; 0.20 mmol), compound **6** (90 mg, 78% yield)was obtained, after a 10 min reaction with PFPAA (0.16 mL, 0.8 mmol), and flash chromatography (eluting with hexane/AcOEt; 6:4, v/v). Glycal **6** showed: m.p. 120-122°C; $[\alpha]_D^{20}$ = + 55.3 (c = 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.10 (d, $J_{NH,2}$ = 9.3 Hz, 1H; N-H), 5.98 (d, $J_{3,4}$ = 2.8 Hz, 1H; H-3), 5.70 (dd, $J_{4,5}$ = 8.0, $J_{4,3}$ = 2.8 Hz, 1H; H-4), 5.43 (dd, $J_{7,8}$ = 4.5, $J_{7,6}$ = 3.0 Hz, 1H; H-7), 5.31 (m, 1H; H-8), 4.67

(dd, $J_{9a,9b}$ = 12.4, $J_{9a,8}$ = 2.9 Hz, 1H ;H-9a), 4.51 (dd, $J_{6,5}$ = 9.7, $J_{6,7}$ = 3.0 Hz, 1H; H-6), 4.37 (m, 1H; H-5), 4.19 (dd, $J_{9b,9a}$ = 12.4, $J_{9b,8}$ = 6.8 Hz, 1H; H-9b), 3.84 (s, 3H; $COOCH_3$); 2.13 (s, 3H; CH_3COO), 2.08 (s, 3H; CH_3COO), 2.06 (s, 3H; CH_3COO), 2.05 (s, 3H; CH_3COO); ^{13}C NMR ($CDCl_3$) δ 170.7-170.5 (3C, $CH_3\underline{C}OO$ at C-4, $CH_3\underline{C}OO$ at C-8 and $CH_3\underline{C}OO$ at C-9), 170.0 ($CH_3\underline{C}OO$ at C-7), 161.3 (C-1), 158.1 (t, $J_{C\text{-}F}$ = 26 Hz; $\underline{C}OCF_2CF_3$), 145.2 (C-2) 120.0- 112.0 (2C, $\underline{C}F_2\underline{C}F_3$), 107.7 (C-3), 75.6 (C-6), 70.9 (C-8), 67.3 (C-4 and C-7), 61.8 (C-9), 52.2 ($COO\underline{C}H_3$), 47.7 (C-5), 20.8 ($\underline{C}H_3COO$), 20.6 ($\underline{C}H_3COO$), 20.5 (2C,$\underline{C}H_3COO$); IR 1747, 1728, 1661cm^{-1}; MS (ESI positive) *m/z* 600.1 $[M+Na]^+$, 1176.4 $[2M+Na]^+$.
Anal. Calcd for $C_{21}H_{24}F_5NO_{12}$: C, 43.68; H, 4.19; N, 2.43. Found: C, 43.50; H, 4.35; N, 2.50.

b) *By N-transacylation-elimination from 4,7,8,9-tetra-O-acetyl-N-acetyl-β-neuraminic acid methyl ester* **8**
Starting from 4,7,8,9-tetra-*O*-acetyl-5-*N*-acetyl-β-neuraminic acid methyl ester **8**[3] (0.107 g, 0.2 mmol) and PFPAA (0.16 mL, 0.8 mmol) after a reaction for 15 min at 135°C the glycal **6** was obtained (86.6 mg, 75% yield) after column chromatography (eluting with hexane/AcOEt; 6:4, v/v). Glycal **6** showed: m.p. 120-122°C; $[\alpha]_D^{20}$ = + 55.3 (c = 1, $CHCl_3$) with all physico-chemical properties superimposable to those of the compound described above. Anal. Calcd for Anal. Calcd for $C_{21}H_{24}F_5NO_{12}$: C, 43.68; H, 4.19; N, 2.43. Found: C, 43.80; H, 4.10; N, 2.38.

2.3 Glycal 7: 4,7,8,9-tetra-*O*-Acetyl-2,3-dehydro-2-deoxy-5-*N*-(2,2,3,3,4,4,4 heptafluorobutanoyl) -β-neuraminic acid methyl ester



7

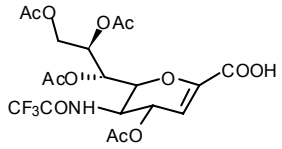
a) *By N-transacylation, from the glycal* **3**

Starting with the glycal **3**[2] (95 mg; 0.20 mmol), compound **7** was obtained (102 mg, 81% yield), after a 10 min reaction with HFBAA (0.16 mL, 0.8 mmol) and flash chromatography (eluting with hexane/AcOEt; 6:4, v/v). Glycal **7** showed: m.p. 115-116°C; $[\alpha]_D^{20}$ = + 49.1 (c = 1, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 7.21 (d, $J_{NH,2}$ = 9.0 Hz, 1H; N-H), 5.97 (d, $J_{3,4}$ = 2.6 Hz, 1H; H-3), 5.70 (dd, $J_{4,5}$ = 9.0, $J_{4,3}$ = 2.6 Hz, 1H; H-4), 5.43 (dd, $J_{7,8}$ = 4.4, $J_{7,6}$ = 2.8 Hz, 1H; H-7), 5.30 (m, 1H; H-8), 4.66 (dd, $J_{9a,9b}$ = 12.4, $J_{9a,8}$ = 2.6 Hz, 1H ; H-9a), 4.53 (dd, $J_{6,5}$ = 9.8, $J_{6,7}$ = 2.8 Hz, 1H ; H-6), 4.35 (q app., $J_{5,4}$ = $J_{5,6}$ = $J_{5,NH}$ = 9.0 Hz, 1H; H-5), 4.19 (dd, $J_{9b,9a}$ = 12.4, $J_{9b,8}$ = 6.8 Hz, 1H; H-9b), 3.81 (s, 3H; $COOCH_3$); 2.14 (s, 3H; CH_3COO at C-7), 2.07 (s, 3H; CH_3COO), 2.06 (s, 3H; CH_3COO), 2.00 (s, 3H; CH_3COO); ^{13}C NMR ($CDCl_3$) δ 170.6 (3C, $CH_3\underline{C}OO$ at C-4, $CH_3\underline{C}OO$ at C-8 and $CH_3\underline{C}OO$ at C-9), 170.1 ($CH_3\underline{C}OO$ at C-7), 161.2 (C-1), 157.8 (t, $J_{C\text{-}F}$ = 27 Hz; $\underline{C}OCF_2CF_2CF_3$), 145.1 (C-2), 122.0- 109.0 (3C, $\underline{C}F_2\underline{C}F_2\underline{C}F_3$), 107.3 (C-3), 75.6 (C-6), 70.9 (C-8), 67.4 (C-4 and C-7), 61.8 (C-9), 52.7 ($COO\underline{C}H_3$), 47.8 (C-5), 20.8 ($\underline{C}H_3COO$), 20.6 (2C, $\underline{C}H_3COO$), 20.5 ($\underline{C}H_3COO$); IR 1746, 1729, 1668cm^{-1}; MS (ESI positive) *m/z* 650.0 $[M+Na]^+$, 1276.2 $[2M+Na]^+$.
Anal. Calcd for $C_{22}H_{24}F_7NO_{12}$: C, 42.11; H, 3.86; N, 2.23. Found: C, 42.20; H, 3.70; N, 2.40.

b) *By N-transacylation-elimination from 4,7,8,9-tetra-O-acetyl-5-N-acetyl-β-neuraminic acid methyl ester* **8**
Starting from 4,7,8,9-tetra-*O*-acetyl-5-*N*-acetyl-β-neuraminic acid methyl ester **8**[3] (0.107 g, 0.2 mmol) and PFPAA (0.16 mL, 0.8 mmol) after a reaction for 15 min at 135°C the glycal **7** was obtained (86.6 mg, 75% yield) after column chromatography (eluting with hexane/AcOEt; 6:4, v/v).

Glycal **7** showed: m.p. 114-117°C; $[\alpha]_D^{20}$ = + 50.3 (c = 1, $CHCl_3$) with all physico-chemical properties superimposable to those of the compound described above. Anal. Calcd for $C_{22}H_{24}F_7NO_{12}$: C, 42.11; H, 3.86; N, 2.23. Found: C, 42.08; H, 3.90; N, 2.35.

2.4 Glycal 13: 4,7,8,9-tetra-*O*-Acetyl-2,3-dehydro-2-deoxy-5-*N*-(2,2,2-trifluoroacetyl) -β-neuraminic acid

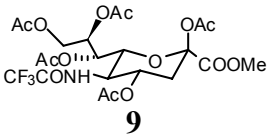


13

Starting from 4,7,8,9-tetra-*O*-acetyl-5-*N*-acetyl-β-neuraminic acid **12**[4] (0.107 g, 0.2 mmol) and TFAA (0.11 mL, 0.8 mmol) after a reaction for 15 min at 135°C, the glycal **13** was obtained as a white solid (73 mg, 71% yield) after flash chromatography (eluting with AcOEt/MeOH; 85:15, v/v;). Glycal **13** showed: m.p. 115-116°C; $[\alpha]_D^{20}$ = + 50.8 (c = 1, CH_3OH); ^{1}H NMR (CD_3OD) δ 5.75 (d, $J_{3,4}$ = 2.5 Hz, 1H; H-3), 5.62 (dd, $J_{4,5}$ = 8.6, $J_{4,3}$ = 2.5 Hz, 1H; H-4), 5.49 (m, 1H; H-8), 5.43 (dd, $J_{7,8}$ = 7.0, $J_{7,6}$ = 2.1 Hz, 1H; H-7), 4.55 (dd, $J_{9a,9b}$ = 12.5, $J_{9a,8}$ = 2.7 Hz, 1H ; H-9a), 4.42 (dd, $J_{6,5}$ = 10.6, $J_{6,7}$ = 2.1 Hz, 1H ; H-6), 4.24-4.19 (overlapping, 2H, H-5 and H-9b), 2.08 (s, 3H; CH_3COO), 2.04 (s, 3H; CH_3COO), 2.01 (overlapping, 6H; CH_3COO); ^{13}C NMR (CD_3OD) δ 172.6 ($CH_3\underline{C}OO$ at C-9), 172.2 ($CH_3\underline{C}OO$ at C-4), 171.7 ($CH_3\underline{C}OO$ at C-8), 171.4 ($CH_3\underline{C}OO$ at C-7), 169.2 (C-1), 159.2 (q, $J_{C\text{-}F}$ = 38 Hz; $\underline{C}OCF_3$), 151.6 (C-2), 120.0-116.0 (1C, $\underline{C}F_3$), 105.3 (C-3), 76.4 (C-6), 71.4 (C-8), 71.1 (C-4), 68.7 (C-7), 63.1 (C-9), 48.5 (C-5), 20.8 ($\underline{C}H_3COO$), 20.7 (3C, $\underline{C}H_3COO$);); IR 1741, 1730, 1662cm^{-1}; MS (ESI negative) *m/z* 511.8 $[M\text{-}H]^-$.
Anal. Calcd for $C_{20}H_{24}F_3NO_{12}$: C, 45.55; H, 4.59; N, 2.66. Found: C, 45.44; H, 4.60; N, 2.50.

3. Saturated *N*-Perfluoroacylneuraminic Acid: General Procedure

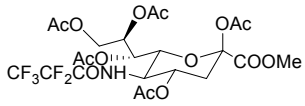
3.1 Compound 9: 2,4,7,8,9-Penta-*O*-Acetyl-5-*N*-(2,2,2-trifluoroacetyl)-β-neuraminic acid methyl ester



9

Compound **9**, as a glass, showed: $[\alpha]_D^{20}$ = - 22.3 (c = 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 6.60 (d, $J_{NH,2}$ = 9.2 Hz, 1H; N-H), 5.43 (m, 1H; H-4), 5.37 (m, 1H; H-7), 5.12 (m, 1H; H-8), 4.51 (d, $J_{9a,9b}$ = 12.5, 1H ; H-9a), 4.31 (br d, $J_{6,5}$ = 10.5, 1H ; H-6), 4.17 (dd, $J_{9b,9a}$ = 12.5, $J_{9b,8}$ = 5.8 Hz, 1H; H-9b), 4.08 (m, 1H; H-5), 3.83 (s, 3H; $COOCH_3$); 2.62 (br d, $J_{3a,3b}$ = 13.4 Hz, 1H; H-3a) 2.18 (overlapping, 6H; 2XCH_3COO), 2.16-2.02 (overlapping, 4H; CH_3COO and H-3b), 2.06 (overlapping, 6H; 2XCH_3COO); ^{13}C NMR ($CDCl_3$) δ 171.1, 170.8, 170.6, 170.1, 168.2, 166.1, 157.4 (q, $J_{C\text{-}F}$ = 38 Hz; $\underline{C}OCF_3$), 116.6 (1C, $J_{C\text{-}F}$ = 287 Hz; $\underline{C}F_3$), 97.3, 71.8, 71.7, 67.7, 67.5, 62.0, 53.3, 49.9, 35.8, 20.9, 20.7, 20.6, 20.5; IR 1752, 1725, 1671cm^{-1}; MS (ESI positive) *m/z* 610.0 $[M+Na]^+$, 1197.1 $[2M+Na]^+$.
Anal. Calcd for $C_{22}H_{28}F_3NO_{14}$: C, 45.98; H, 4.80; N, 2.38. Found: C, 45.94; H, 4.78; N, 2.35.

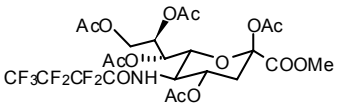
3.2 Compound 10: 2,4,7,8,9-Penta-*O*-Acetyl-5-*N*-(2,2,3,3,3-pentafluoropropyonil)-β-neuraminic acid methyl ester



10

Compound **10,** as a glass, showed: $[\alpha]_D^{20}$ = - 18.5 (c = 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.00 (d, $J_{NH,2}$ = 9.2 Hz, 1H; N-H), 5.42 (ddd, $J_{4,3b}$ = $J_{4,5}$ = 10.8, $J_{4,3a}$ = 4.8 Hz, 1H; H-4), 5.32 (m, 1H; H-7), 5.04 (br s, 1H; H-8), 4.51 (d, $J_{9a,9b}$ = 12.5, 1H ; H-9a), 4.30 (br d, $J_{6,5}$ = 10.6, 1H ; H-6), 4.16 (dd, $J_{9b,9a}$ = 12.5, $J_{9b,8}$ = 6.5 Hz, 1H; H-9b), 4.08 (m, 1H; H-5), 3.85 (s, 3H; $COOCH_3$); 2.58 (dd, $J_{3a,3b}$ = 13.4, $J_{3a,4}$ = 4.8 Hz, 1H; H-3a) 2.15 (overlapping, 6H; 2XCH_3COO), 2.09-2.05 (overlapping, 4H; CH_3COO and H-3b), 2.03 (s, 3H; CH_3COO), 2.02 (s, 3H; CH_3COO); ^{13}C NMR ($CDCl_3$) δ 170.8, 170.6, 170.5, 170.1, 168.2, 166.1, 158.1 ($\underline{C}OCF_2CF_3$), 116.6-110.0 (2C, $\underline{C}F_2\underline{C}F_3$), 97.1, 71.6, 71.4, 67.6, 67.3, 61.9, 53.3, 50.1, 35.9, 20.9, 20.7, 20.6, 20.5; IR 1747, 1718, 1669cm^{-1}; MS (ESI positive) *m/z* 610.0 $[M+Na]^+$, 1298.3 $[2M+Na]^+$.
Anal. Calcd for $C_{23}H_{28}F_5NO_{14}$: C, 43.34; H, 4.43; N, 2.20. Found: C, 43.28; H, 4.47; N, 2.23.

3.3 Compound 11: 2,4,7,8,9-Penta-*O*-Acetyl-5-*N*-(2,2,3,3,4,4,4-heptafluorobutanoyl)-β-neuraminic acid methyl ester



11

Compound **11** showed: $[\alpha]_D^{20}$ = - 22.5 (c = 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 6.83 (d, $J_{NH,2}$ = 9.1 Hz, 1H; N-H), 5.48 (ddd, $J_{4,3b}$ = $J_{4,5}$ = 11.0, $J_{4,3a}$ = 5.0 Hz, 1H; H-4), 5.28 (dd, $J_{7,8}$ = 5.7, $J_{7,6}$ = 1.7 Hz 1H; H-7), 5.09 (ddd, $J_{8,9b}$ = $J_{8,7}$ = 5.7, $J_{8,9a}$ = 2.4 Hz 1H; H-8), 4.47 (dd, $J_{9a,9b}$ = 12.5, $J_{9b,8}$ = 5.7 Hz Hz, 1H ; H-9a), 4.36 (dd, $J_{6,5}$ = 10.5, $J_{6,7}$ =1.7 Hz, 1H ; H-6), 4.14 (dd, $J_{9b,9a}$ = 12.5, $J_{9b,8}$ = 5.7 Hz, 1H; H-9b), 3.99 (m, 1H; H-5), 3.80 (s, 3H; $COOCH_3$); 2.62 (dd, $J_{3a,3b}$ = 13.5, $J_{3a,4}$ = 5.0 Hz, 1H; H-3a) 2.16 (overlapping, 6H; 2XCH_3COO), 2.06 (s, 3H; CH_3COO), 2.05-2.00 (overlapping, 7H; 2XCH_3COO and H-3b); ^{13}C NMR ($CDCl_3$) δ 170.5, 170.4, 170.3, 170.12, 168.2, 166.1, 157.9 (1C, $\underline{C}OCF_2CF_2CF_3$), 120.0-108.0 (3C, $CO\underline{C}F_2\underline{C}F_2\underline{C}F_3$), 97.1, 71.3, 70.9, 67.6, 67.1, 61.8, 53.3, 50.6, 35.9, 20.7, 20.6 (3C) 20.5; IR 1752, 1715, 1669cm^{-1}; MS (ESI positive) *m/z* 710.6 $[M+Na]^+$.
Anal. Calcd for $C_{24}H_{28}F_7NO_{14}$: C, 41.93; H, 4.11; N, 2.04. Found: C, 41.89; H, 4.08; N, 2.07.

4. 1,7-Lactonization and N-transacylation of sialic acids by action of HFBAA

4. 1 Treatment of Neu5Ac (1) with HFBAA

The reaction was performed treating Neu5Ac **1** (30 mg, 0.1mmol), dissolved in CD_3CN (0.300 mL), with HFBAA (0.034 mL, 1.4 mmol) at 135°C for 15 min, and the reaction mixture was subjected to NMR analyses. The ^{1}H-NMR spectrum showed the absence of any olefinic signal between 5.6-6.5 ppm, attributable to the proton at C-3 of sialic glycals. On the contrary it showed diagnostic[5] signals for the presence of a 1,7-lactone (detailed in the following). ^{13}C analyses of the reaction mixture,

performed after evaporation of CD_3CN and dilution of the reaction mixture with $CDCl_3$, confirmed the presence of the lactone ring and of the perfluorinated amide group in place of the starting acetyl group.

Compound **14** showed:

^{1}H NMR (CD_3CN) δ 8.23 (d, $J_{NH,5}$ = 7.6 Hz, 1H; N-H), 5.80 (dt, $J_{8,7} = J_{8,9b}$ = 5.7, $J_{8,9a}$ = 2.6 Hz, 1H; H-8), 5.61 (br s, 1H; H-4), 5.04 (d, $J_{8,7}$ = 5.7 Hz, 1H; H-7), 4.96 (dd, $J_{9a,9b}$ = 13.0, $J_{9a,8}$ = 2.6 Hz, 1H; H-9a), 4.75 (dd, $J_{9b,9a}$ = 13.0, $J_{9b,8}$ = 5.7 Hz, 1H; H-9b), 4.66 (br s, 1H; H-6), 4.54 (br d, $J_{5,NH}$ = 7.6 Hz, 1H; H-5), 2.77 (dd, $J_{3a,3b}$ = 15.4, $J_{3a,4}$ = 4.1 Hz, 1H; H-3a), 2.51 (br d, $J_{3b,3a}$ = 15.4 Hz, 1H; H-3b);

^{1}H NMR ($CDCl_3$) δ 7.34 (d, $J_{NH,5}$ = 7.8 Hz, 1H; N-H), 5.69 (br d, $J_{8,7}$= 9.0 Hz, 1H; H-8), 5.53 (br s, 1H; H-4), 5.04 (br d, $J_{9a,9b}$ = 13.0, 1H; H-9a), 4.85 (d, $J_{8,7}$ = 9.0 Hz, 1H; H-7), 4.77 (dd, $J_{9b,9a}$ = 13.0, $J_{9b,8}$ = 2.8 Hz, 1H; H-9b), 4.50 (br d, $J_{5,NH}$ = 7.8 Hz, 1H; H-5), 4.32 (br s, 1H; H-6), 2.62-2.59 (m, 2H, H-3a and 3b); ^{13}C NMR ($CDCl_3$) 160.1 (C-1), 158-100 (overlapping $\underline{C}O\underline{C}F_2\underline{C}F_2\underline{C}F_3$), 93.63 (C-2), 74.9 (C-7), 72.8 (C-8), 71.6 (C-6), 69.9 (C-4), 63.3 (C-9), 48.6 (C-5), 32.7 (C-3).

All attempt to isolate the formed lactone were unsuccessful.

4. 2 Treatment of Neu5,9Ac$_2$ (15) and of Neu5,8,9Ac$_3$ (16) with HFBAA.

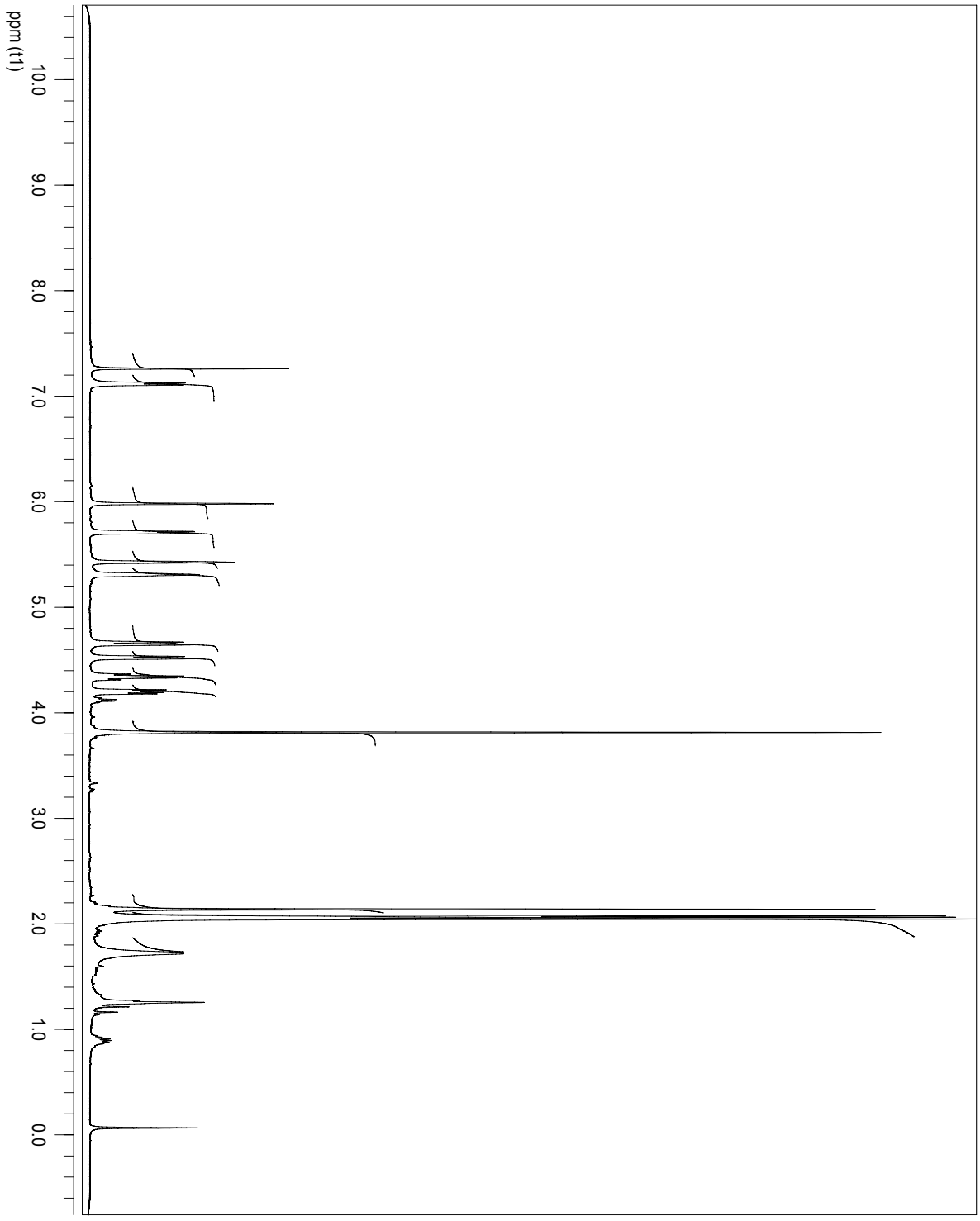
The reaction was performed treating Neu5,9Ac$_2$ **15** (35 mg, 0.1mmol), dissolved in CD_3CN (0.300 mL), with HFBAA (0.034 mL, 1.4 mmol) at 135°C for 15 min, and the reaction mixture was subjected to NMR analyses. The ^{1}H-NMR spectrum showed the absence of any olefinic signal between 5.6-6.5 ppm, attributable to the proton at C-3 of sialic glycals. On the contrary it showed diagnostic[5] signals for the presence of a 1,7-lactone (detailed in the following).

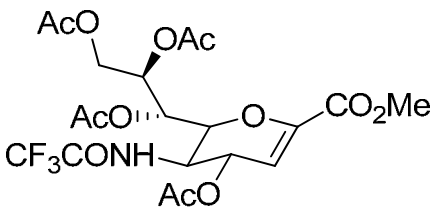
Compound **17** showed: ^{1}H NMR ($CDCl_3$) δ 7.11 (d, $J_{NH,5}$ = 8.4 Hz, 1H; N-H), 5.58 (br m, 1H; H-8), 5.51 (br s, 1H; H-8), 4.86-4.80 (overlapping, 2H; H-7 and H-9a), 4.45-4.35 (overlapping, 2H; H-5 and H-6), 4.30 (dd, $J_{9b,9a}$ = 12.8, $J_{9b,8}$ = 3.5 Hz, 1H; H-9b), 2.62-2.59 (m, 2H, H-3a and 3b), 2.12 (s, 3H, $COCH_3$ at C-9).

The reaction was performed treating Neu5,8,9Ac$_3$ **16** (40 mg, 0.1mmol), dissolved in CD_3CN (0.300 mL), with HFBAA (0.034 mL, 1.4 mmol) at 135°C for 15 min, and the reaction mixture was subjected to NMR analyses. The ^{1}H-NMR spectrum showed the absence of any olefinic signal between 5.6-6.5 ppm, attributable to the proton at C-3 of sialic glycals. On the contrary it showed diagnostic[5] signals for the presence of a 1,7-lactone (detailed in the following).

Compound **18** showed: ^{1}H NMR ($CDCl_3$) δ 7.15 (d, $J_{NH,5}$ = 7.7 Hz, 1H; N-H), 5.52 (br d, 1H; H-4), 5.44 (m, 1H; H-8), 4.76-4.71 (overlapping, $J_{8,7}$ = 8.3 Hz, 2H; H-7 and H-9a), 4.47 (br s, 1H; H-6), 4.39 (br d, $J_{5,NH}$ = 7.7 Hz, 1H; H-5), 4.32 (dd, $J_{9b,9a}$ = 12.8, $J_{9b,8}$ = 3.5 Hz, 1H; H-9b), 2.62 (brd, $J_{3a,3b}$ = 15.4, 1H; H-3a), 2.51 (dd, $J_{3a,3b}$ = 15.4, $J_{3b,4}$ = 4.2 Hz, 1H; H-3a), 2.14 (s, 3H, $COCH_3$), 2.11(s, 3H, $COCH_3$).

All attempt to isolate the formed lactone were unsuccessful.





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Temperature:
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Number of Scans:
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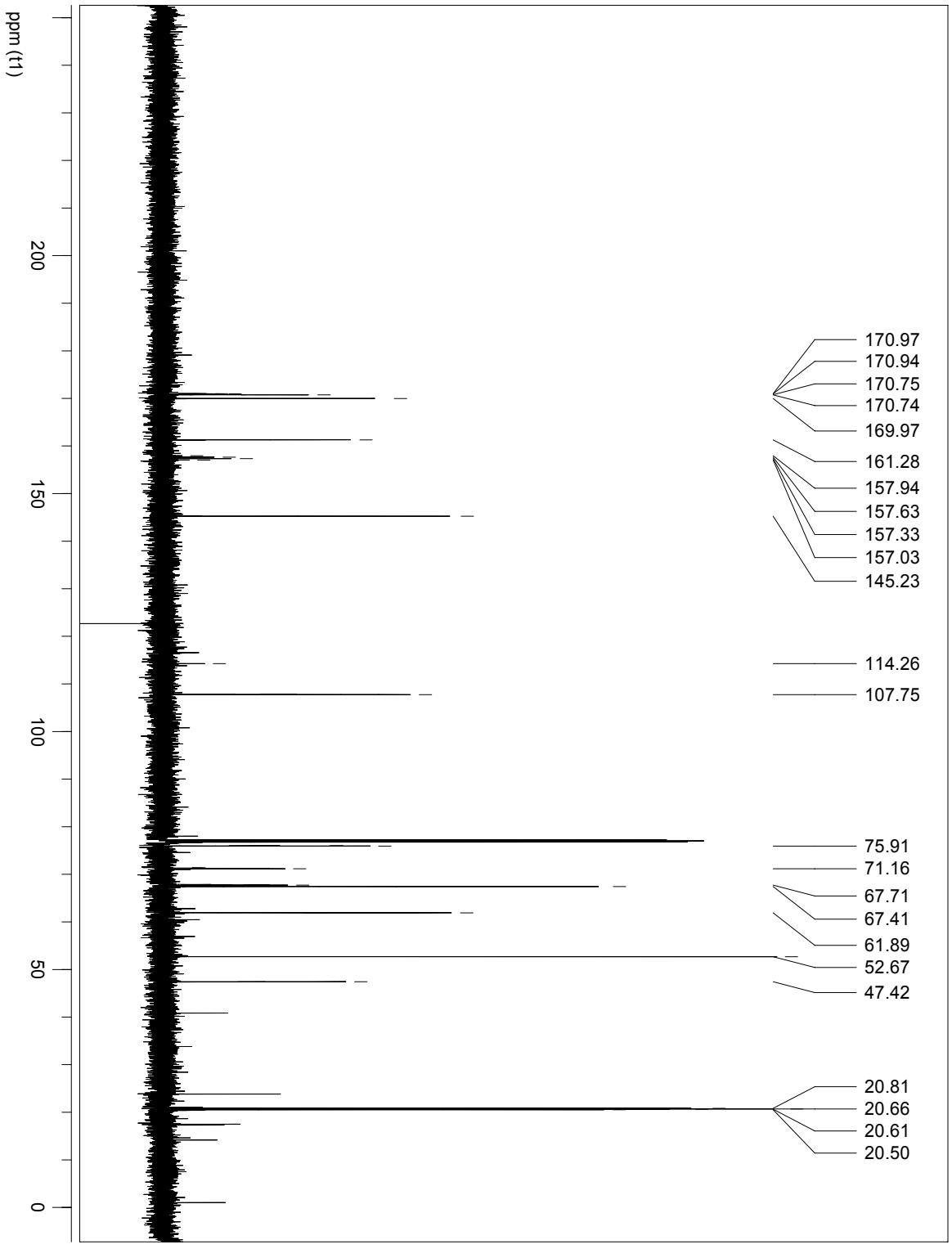

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Temperature:
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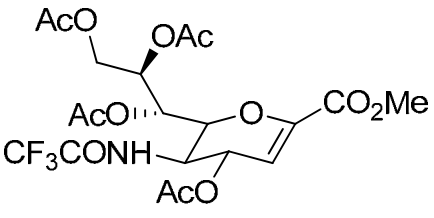
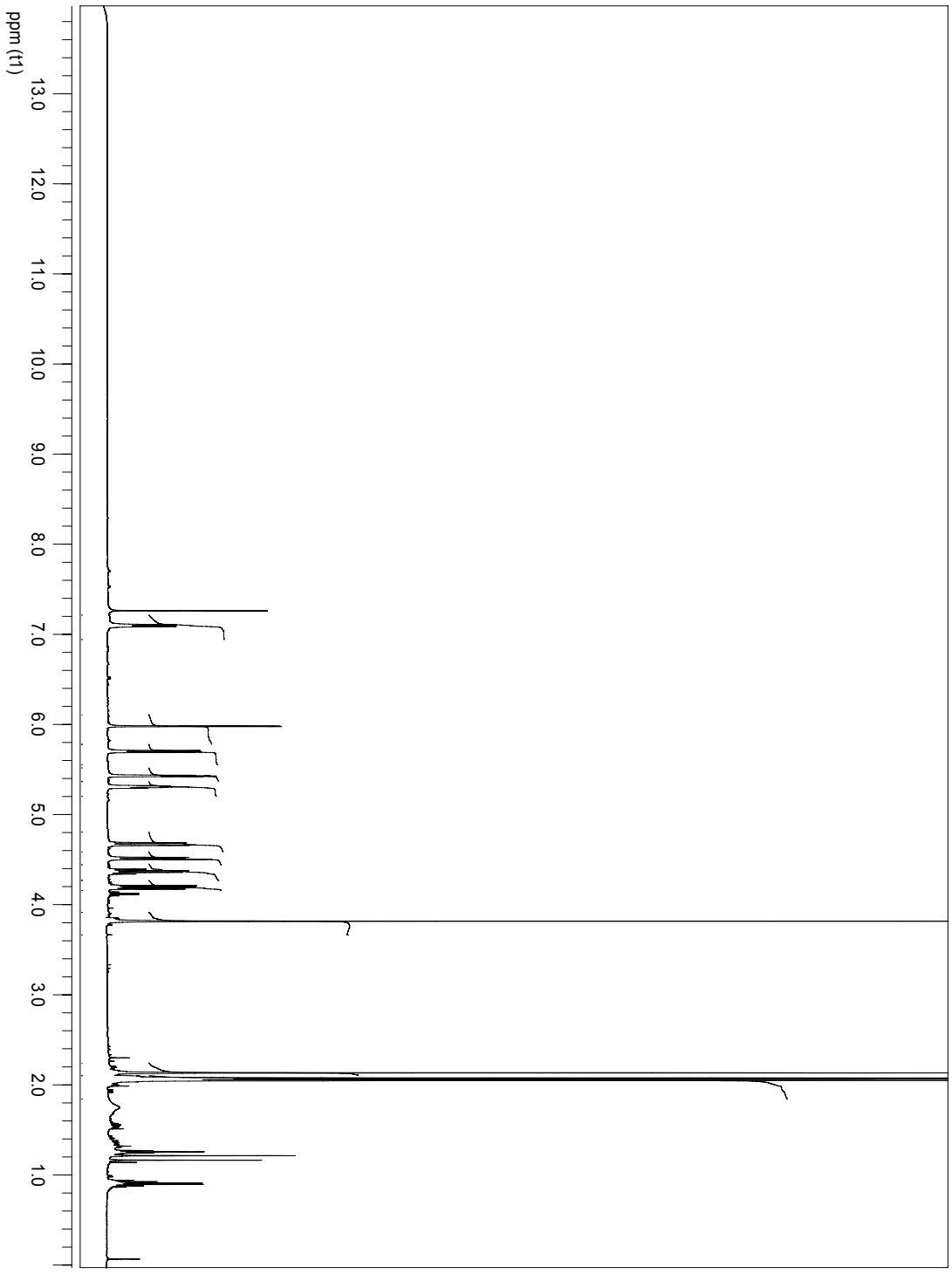
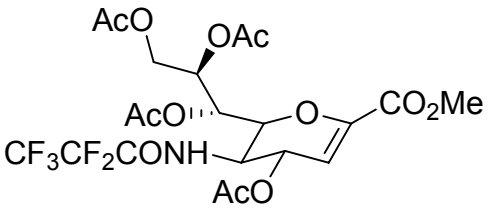


Glycal 5





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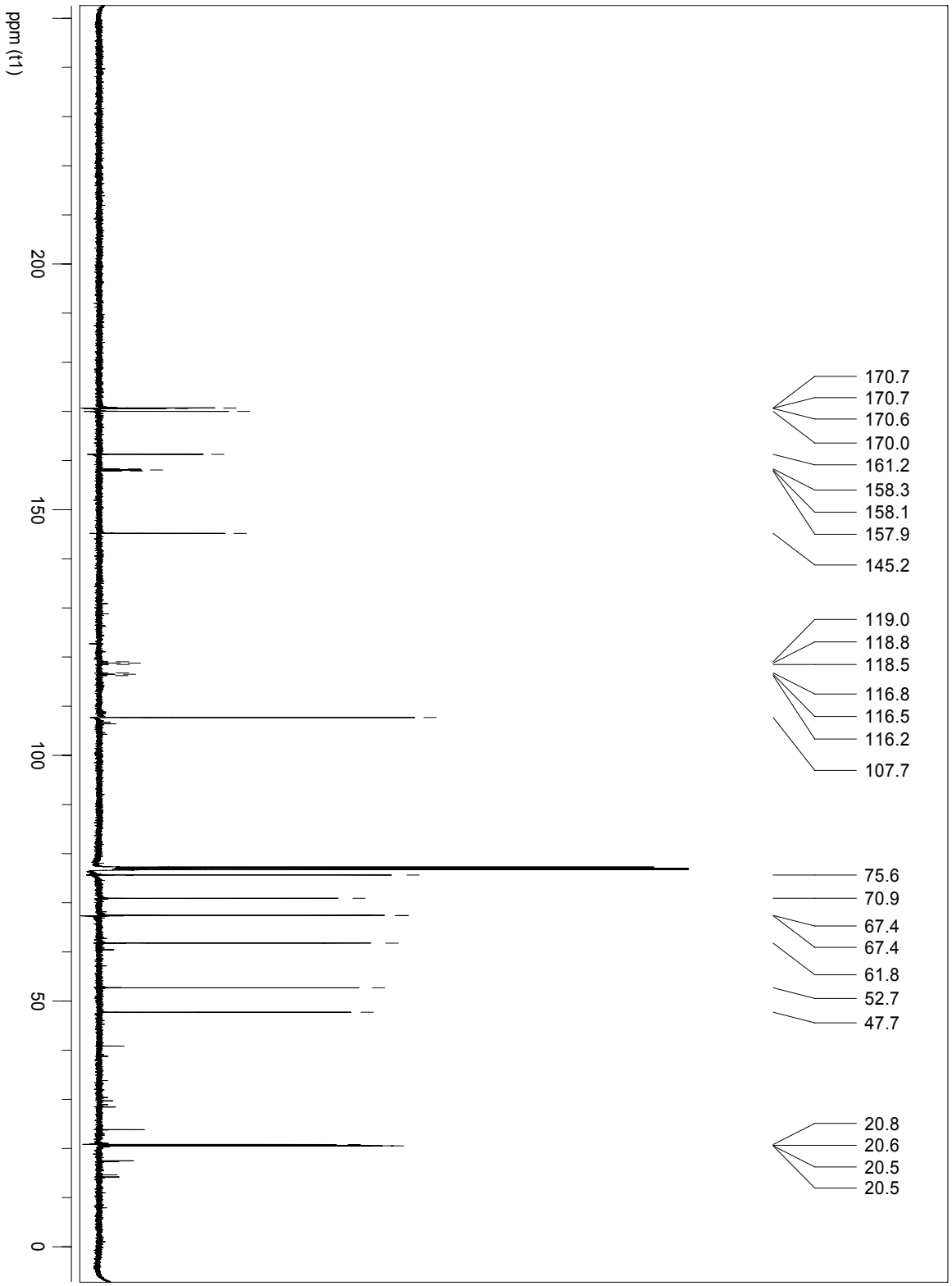

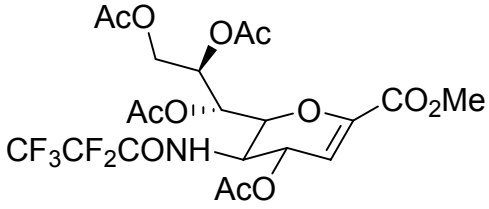

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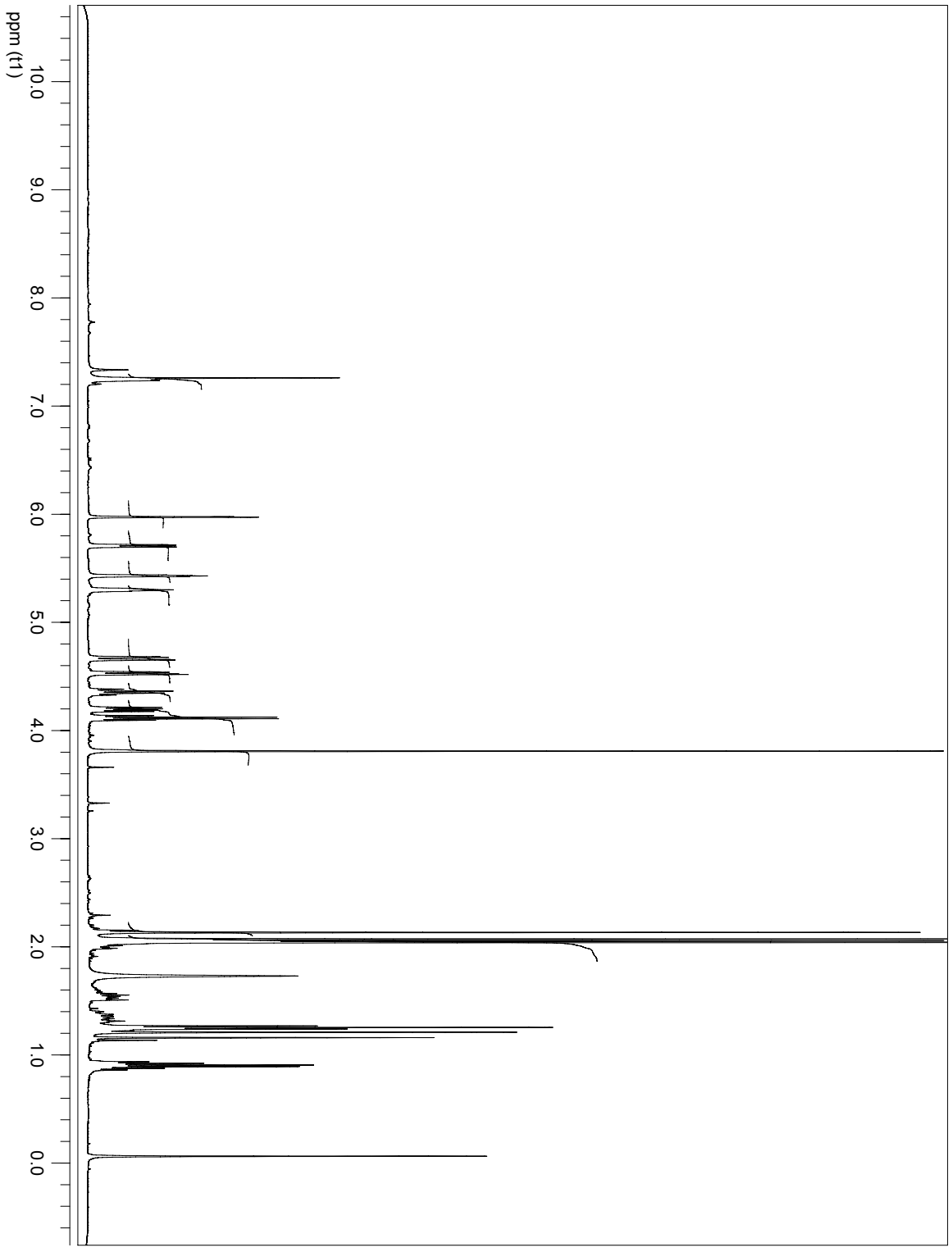

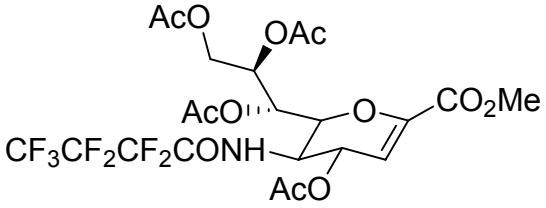

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Pulse Program:
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Temperature:
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Number of Scans:
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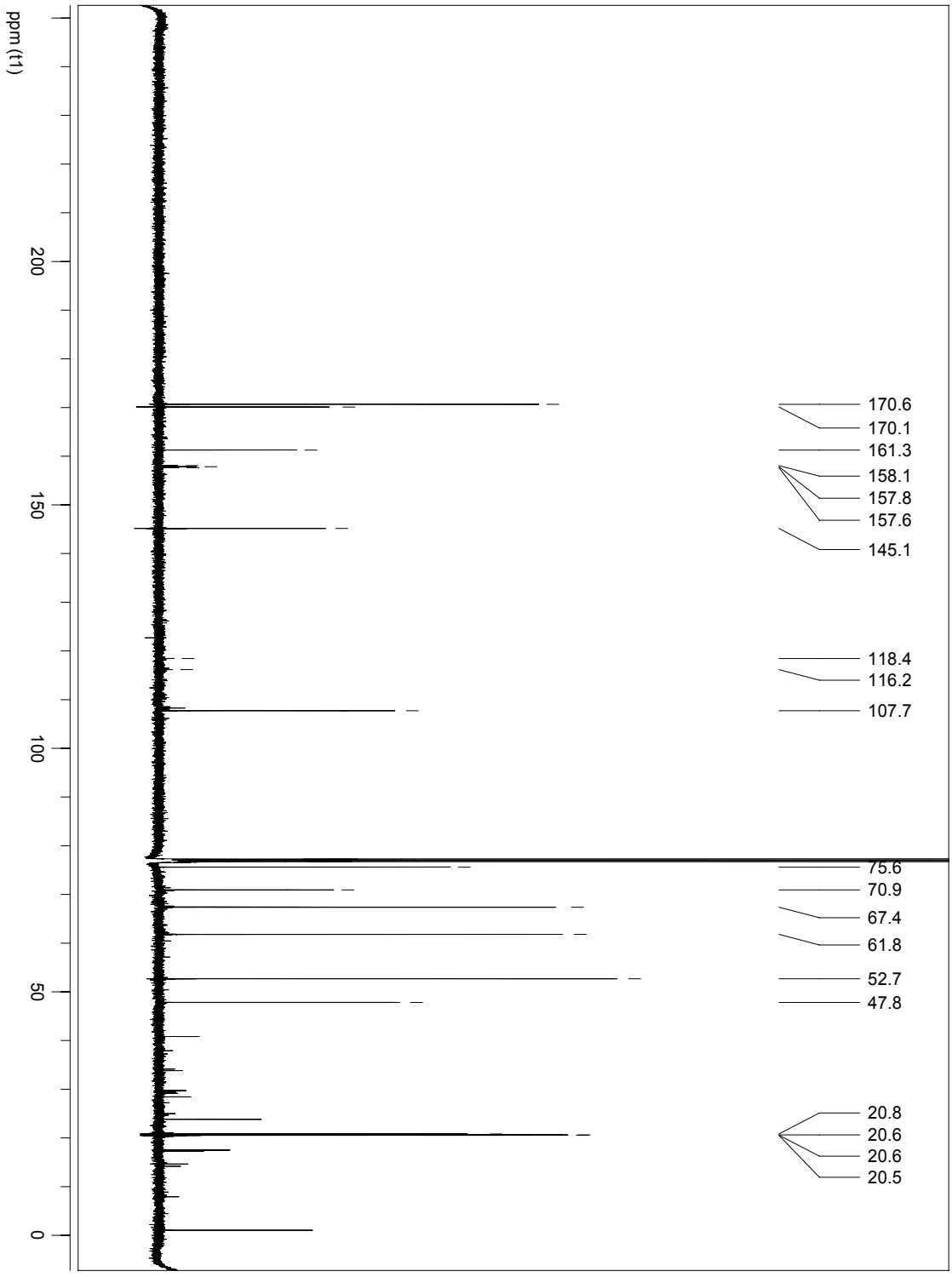

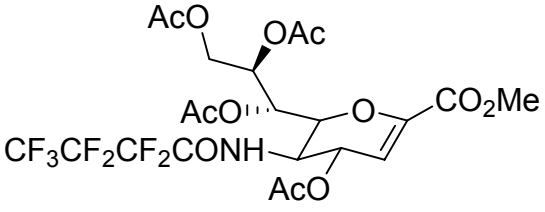

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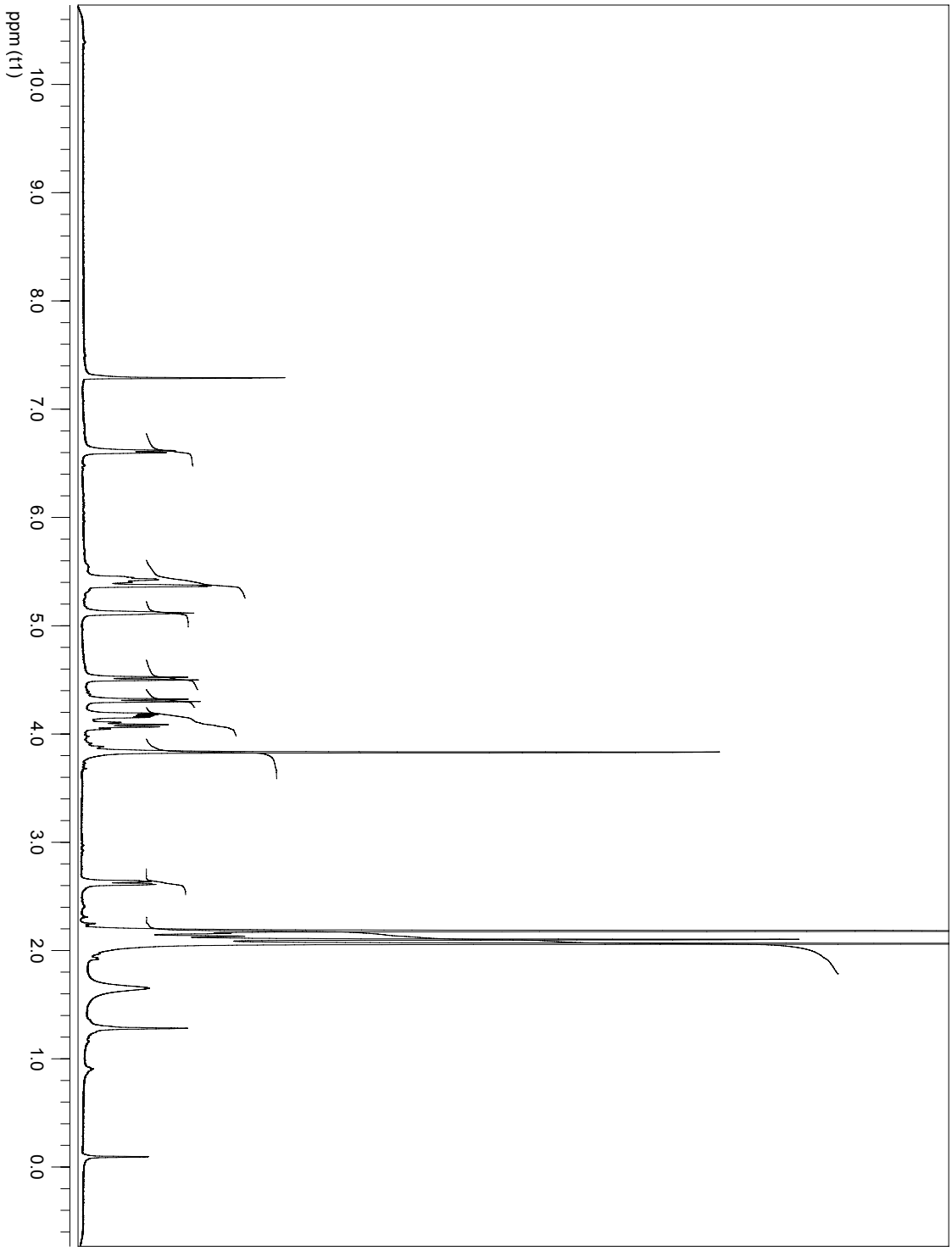

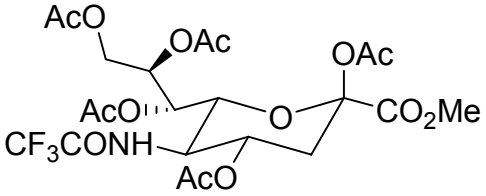

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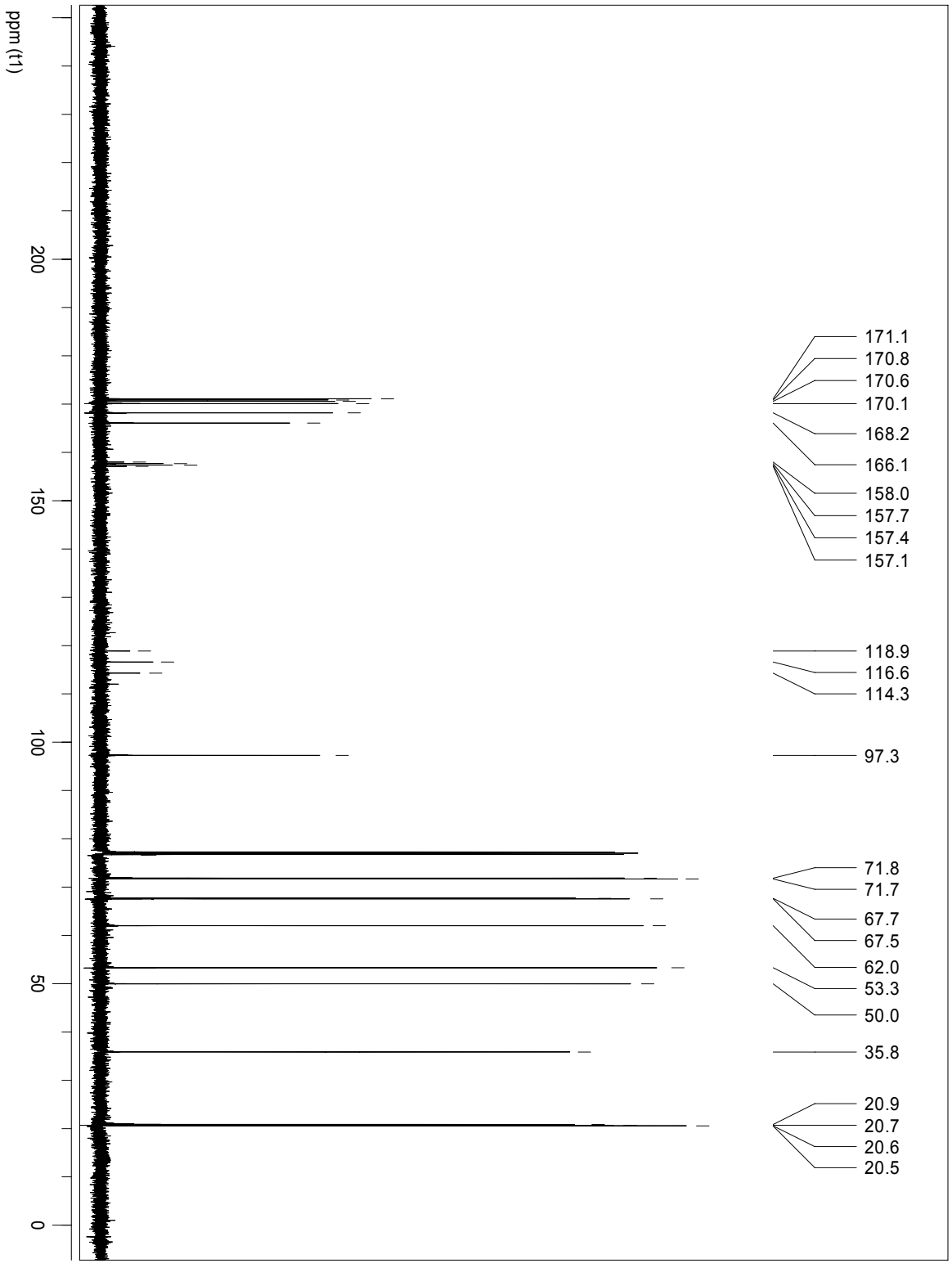

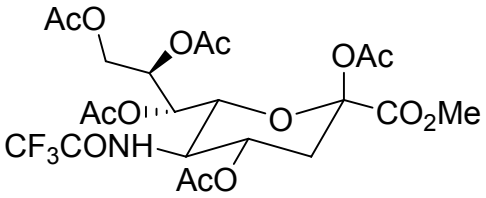

Compound 9

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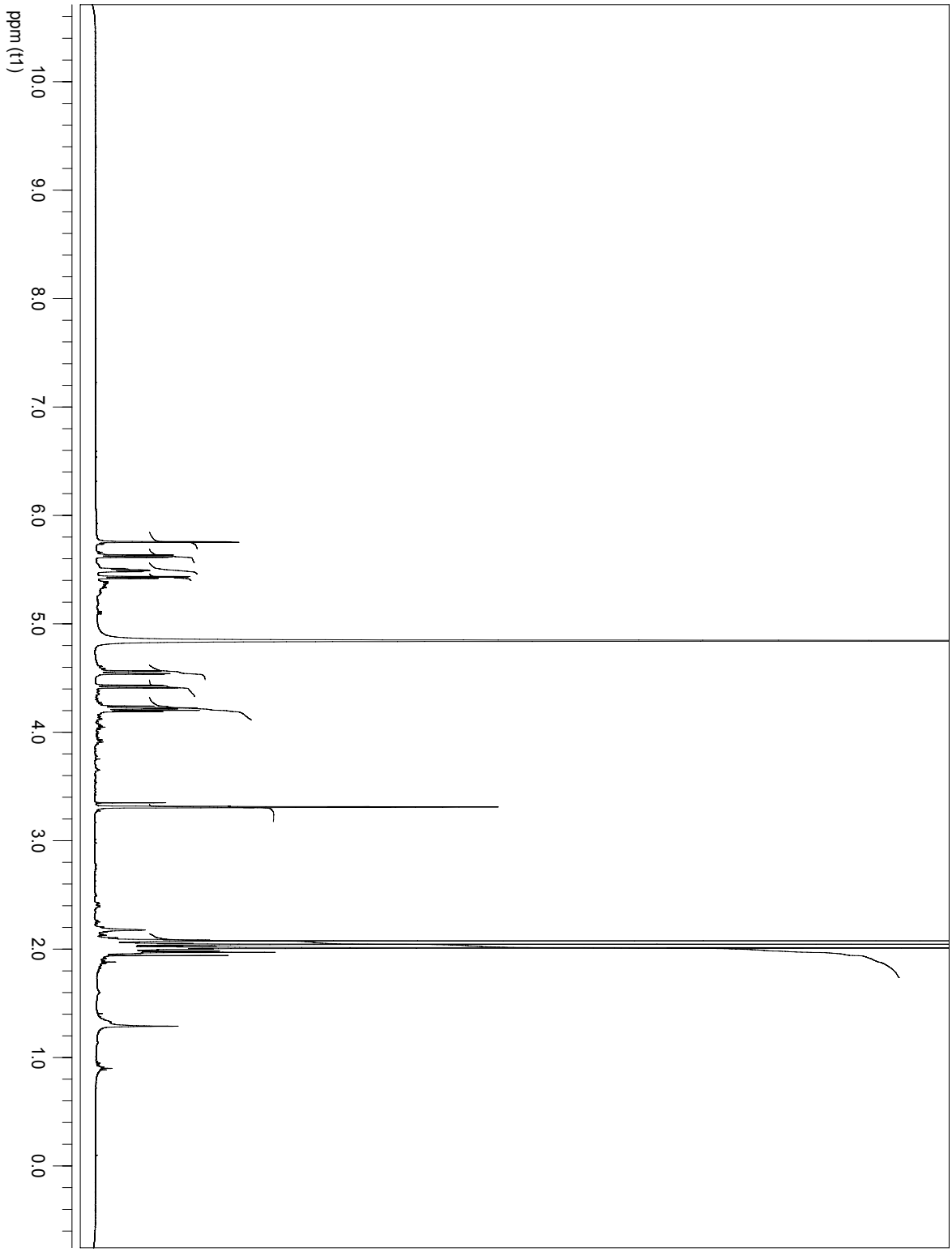

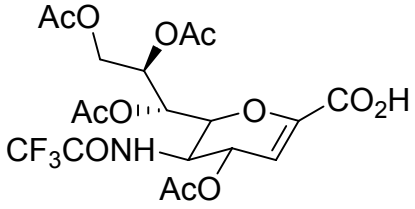

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Temperature:
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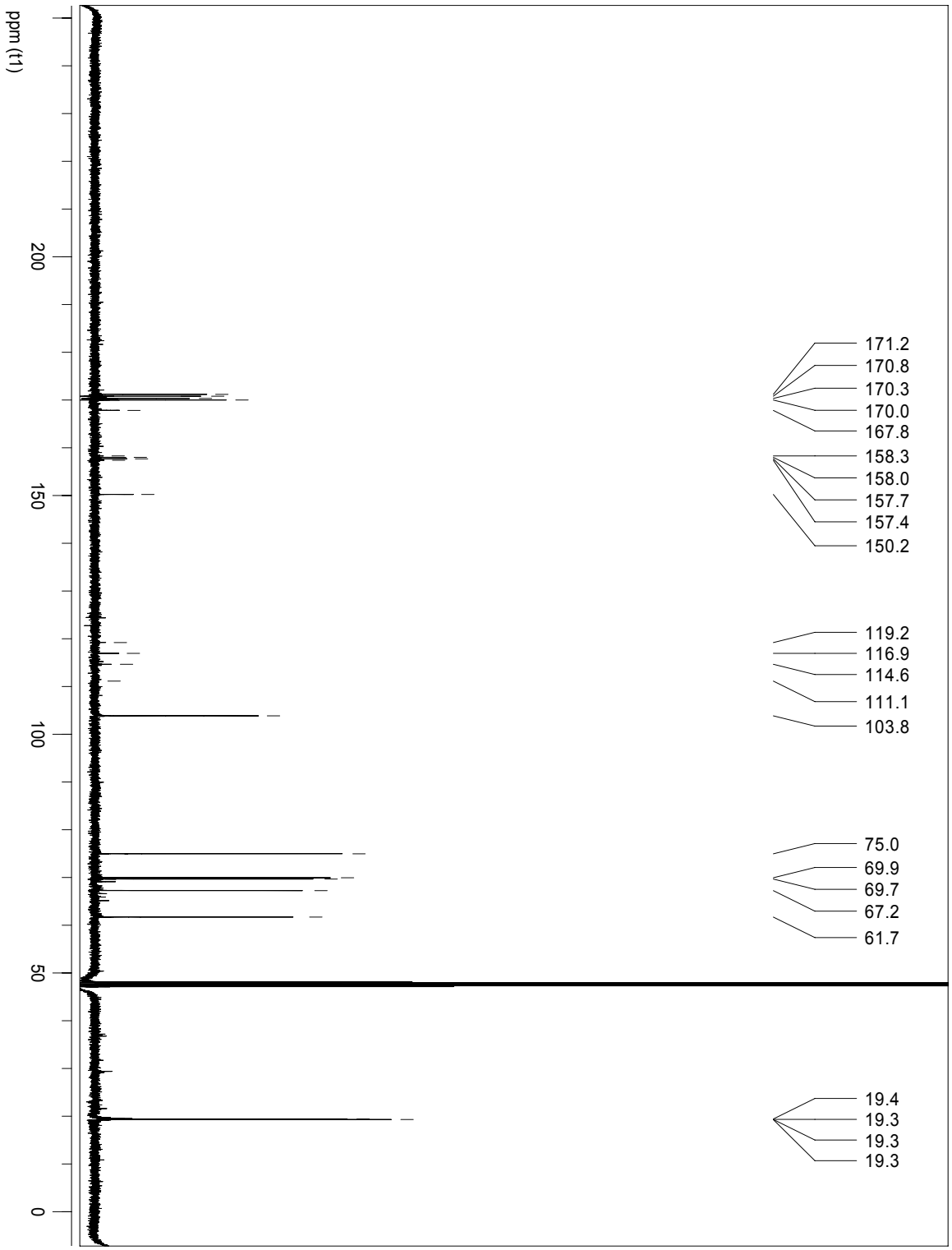

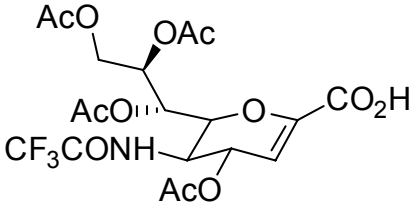

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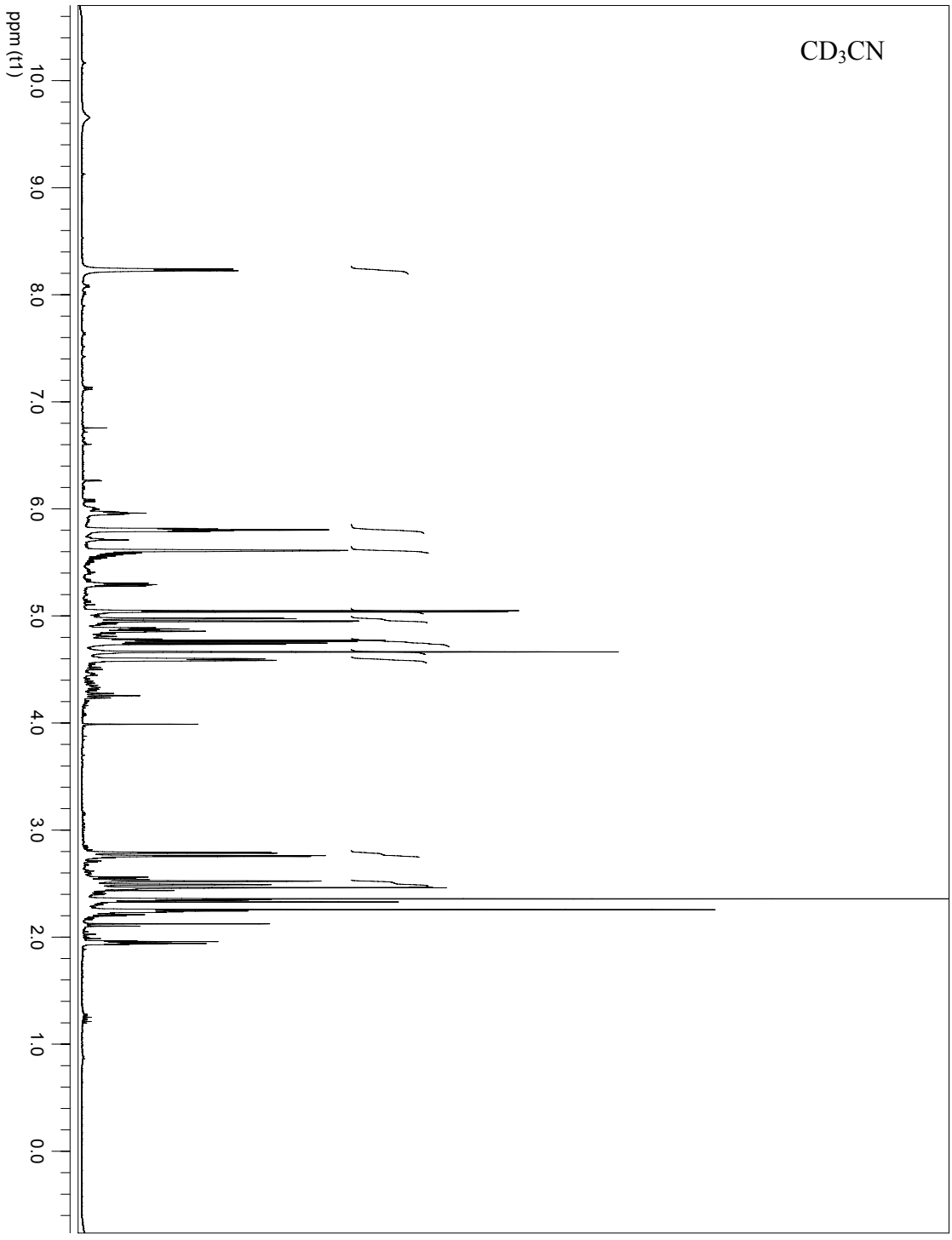
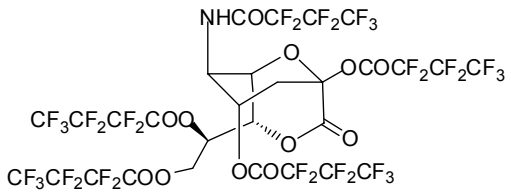


Treatment of Neu5Ac **1** with HFBAA



crude product of reaction

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Temperature:
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Number of Scans:
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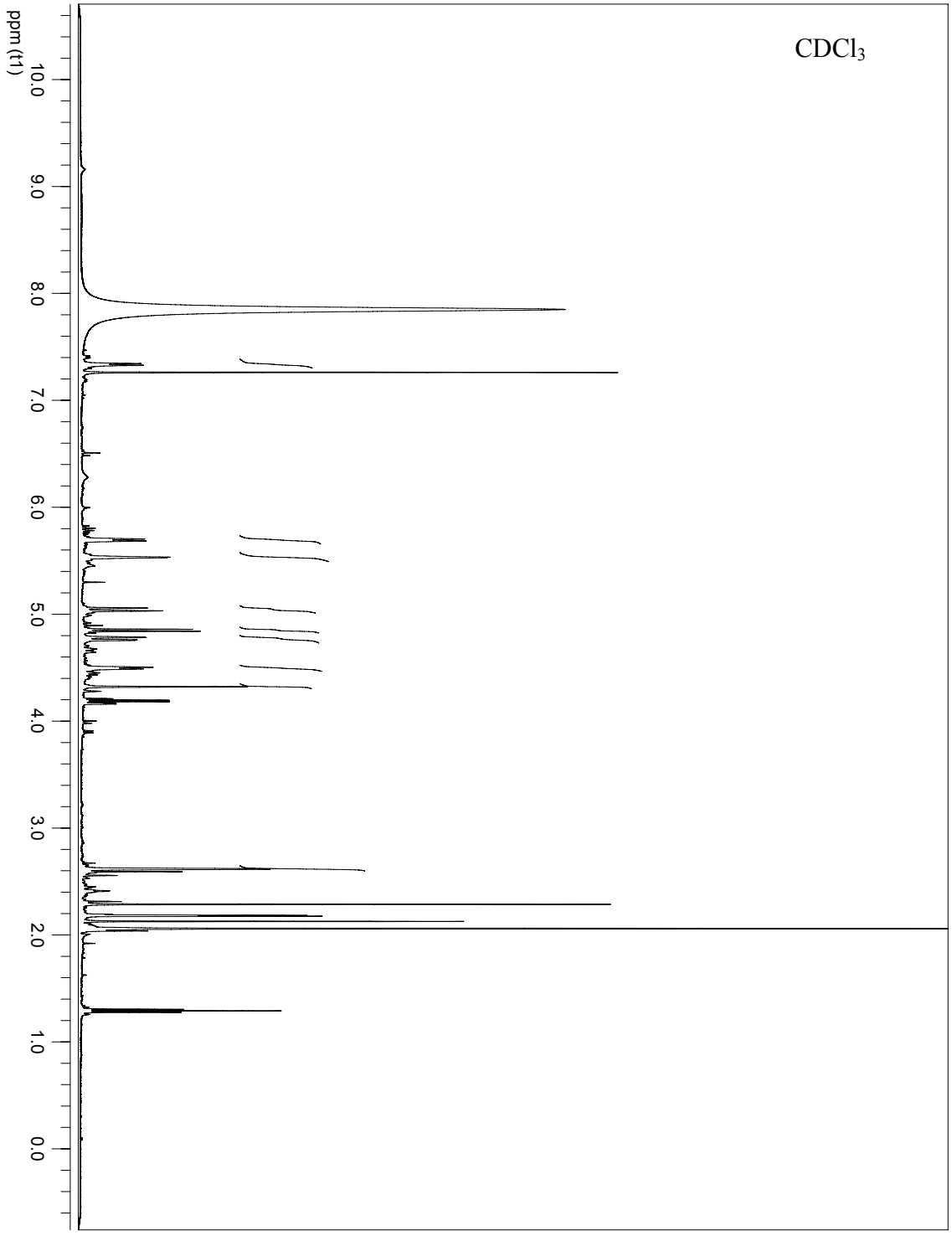
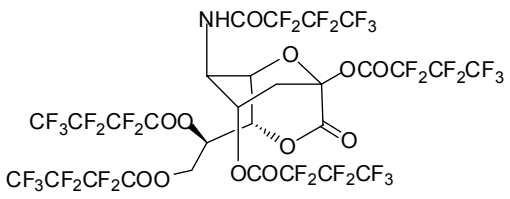


Treatment of Neu5Ac **1** with HFBAA



crude product of reaction

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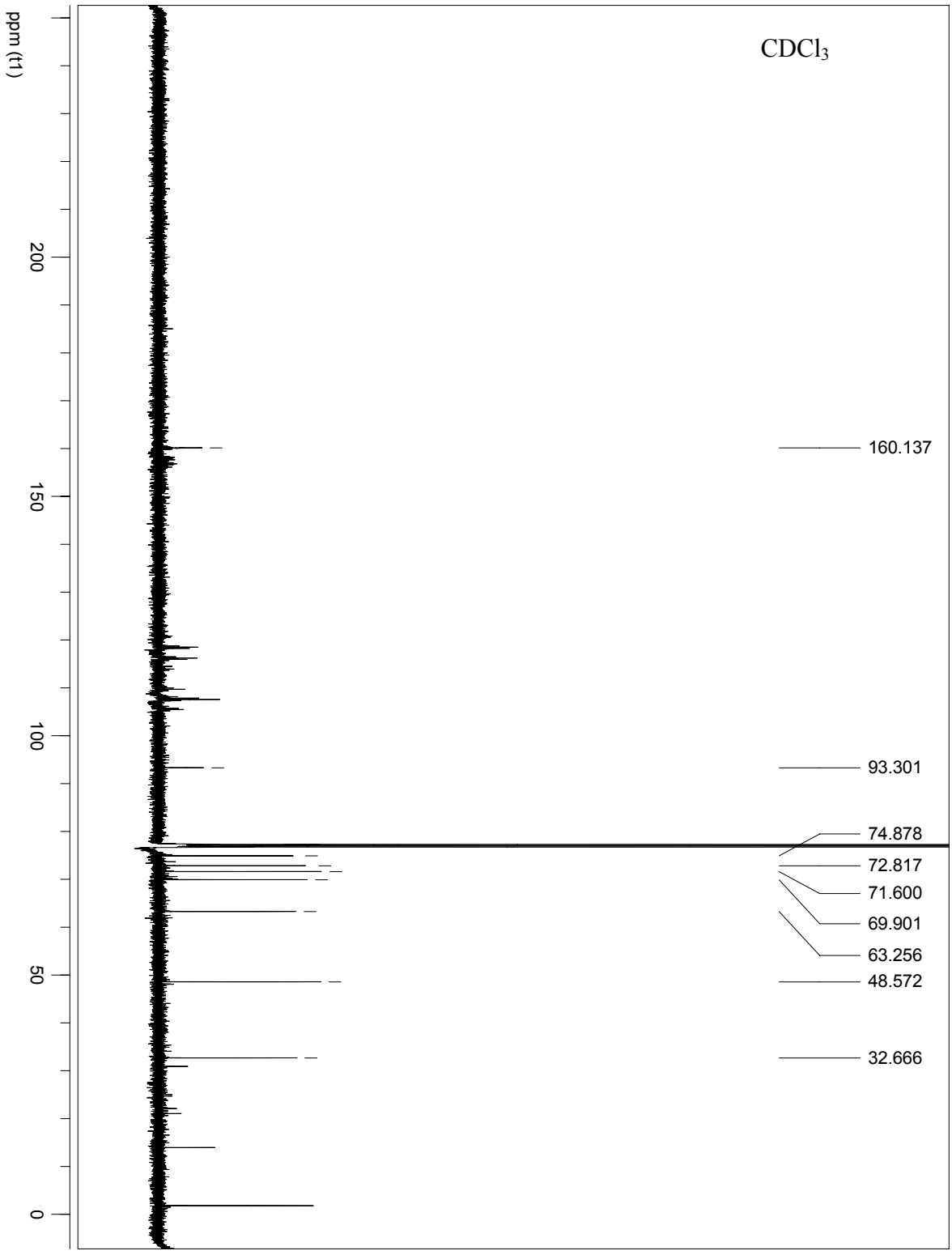
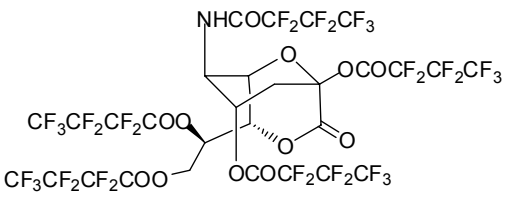
Fri Jul 18 10:16:53 AM



Treatment of Neu5Ac **1** with HFBAA



crude product of reaction

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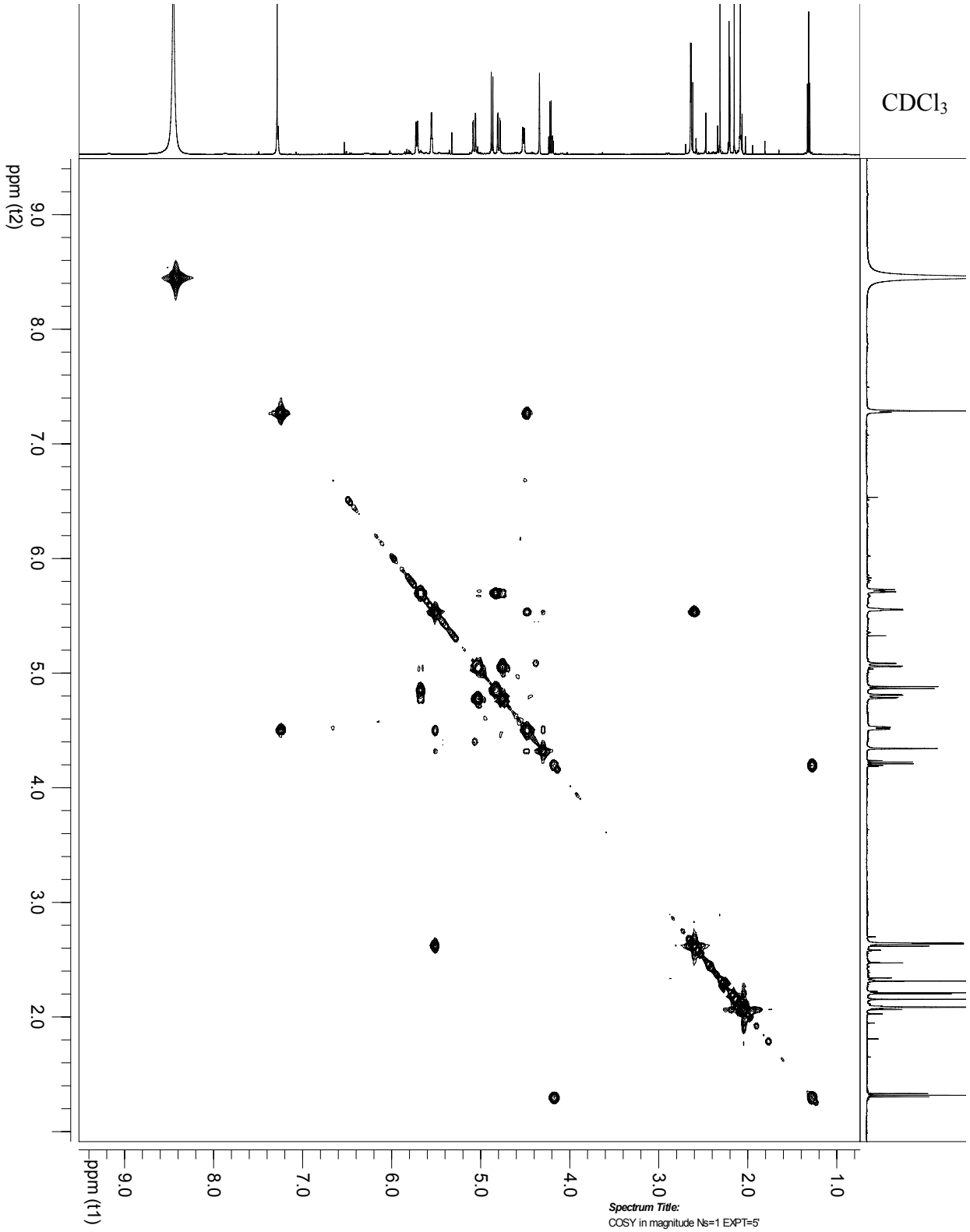

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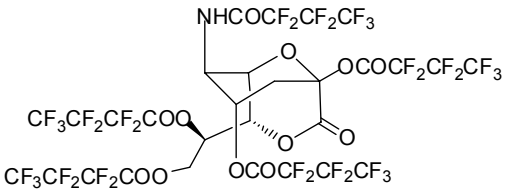
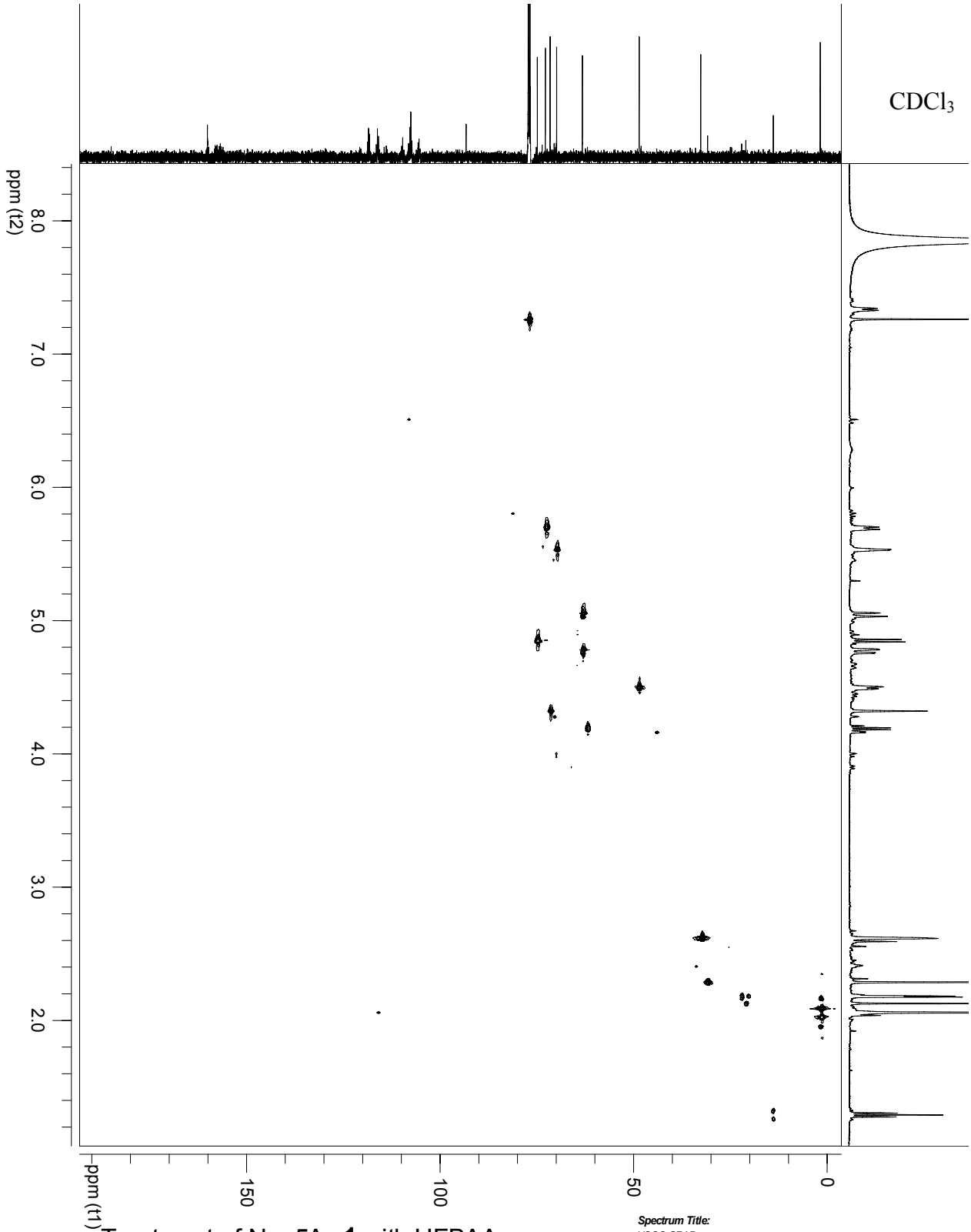
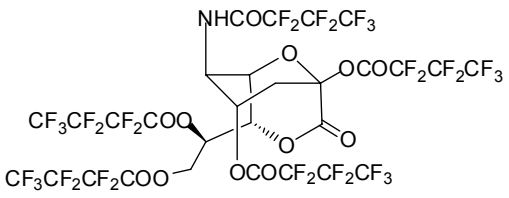
Treatment of Neu5Ac **1** with HFBAA



crude product of reaction



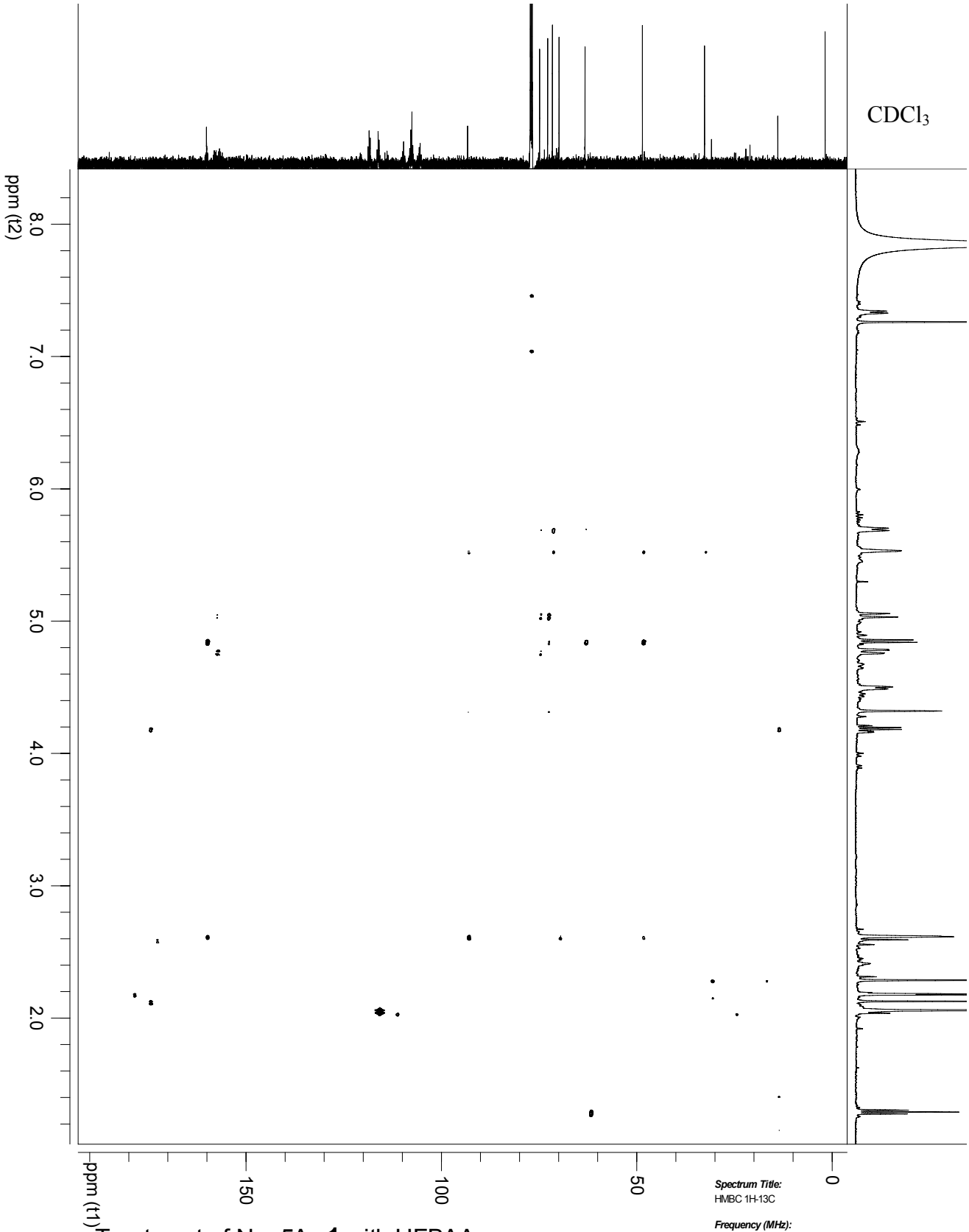
Treatment of Neu5Ac **1** with HFBAA



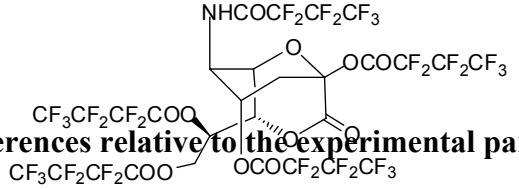
crude product of reaction

Spectrum Title:
HSQC GRAD

Frequency (MHz):
(f2) 500.132 (f1) 125.770
Original Points Count:
(f2) 512 (f1) 512
Actual Points Count:
(f2) 512 (f1) 512
Acquisition Time (sec):
(f2) 0.1389 (f1) 0.0207
Spectral Width (ppm):
(f2) 7.373 (f1) 196.808
Pulse Program:
HSQCETGP
Temperature:
299.2
Number of Scans:
4



Treatment of Neu5Ac **1** with HFBAA



crude product of reaction

5. References relative to the experimental part

[1] W.C Still, M. Kahn, A. Mitra, *J. Org. Chem.* **1978**, *43*, 2923-2925.
[2] Ogura, Haruo; Fujita, Hideshi; Furuhata, Kimio; Itoh, Masayoshi; Shitori, Yoshiyasu; Chemical and Pharmaceutical Bulletin; English; 34; 4; 1986; 1479 - 1484; ISSN: 0009-2363. *Chem Pharm.Bull.* **1986,** *34*, 1479-1484.
[3] H. Ogura, K. Furuhata, M. Itoh, Y. Shitori, *Carbohydr. Res.* **1986**, *15*8, 37-51.
[4] N.Sugyama, K. Sugai, N.Yamada, M.Goto, C.Ban, K. Furuhata, H.Takayanagi and H.Ogura, *Chem Pharm.Bull.* **1988,** *36*, 1147-1152.