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Supplementary Information

for

Synthesis and Immunological Studies of Group A Streptococcus Cell-Wall Oligosaccharide-Streptococcal C5a Peptidase Conjugates as Bivalent Vaccines

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I. Experimental Section

General Methods. Chemicals and materials were obtained from commercial sources and were used as received without additional purification unless otherwise noted. Molecular sieves (MS) 4Å were flame-dried under high vacuum and cooled to room temperature under a nitrogen atmosphere before use. Optical rotations were determined at 25 °C with an automatic polarimeter. ^{1}H and ^{13}C NMR spectra were recorded with a 600 MHz spectrometer for solutions in $CDCl_3$ or D_2O, and chemical shifts (δ) were given in ppm downfield from internal Me_4Si or with DHO signal as a reference. Chemical shifts and coupling constants were obtained from a first-order analysis of one-dimensional spectra and assignments of proton and carbon resonances were based on ^{1}H-^{1}H COSY and ^{13}C-^{1}H HSQC experiments. High-resolution mass spectra (HRMS) were performed on an LTQ Orbitrap spectrometer using electrospray ionization (ESI) mode. MALDI-TOF mass spectra were obtained from samples dispersed in a 2,5-dihydroxybenzoic acid (DHB) or 3,5-Dimethoxy-4-hydroxycinnamic acid (SA) matrix. Thin-layer chromatography (TLC) was performed on silica gel GF_{254} plates and visualized by charring with 30% H_2SO_4 in MeOH or by a UV detector. Column chromatography was performed with Silica gel (100−200 mesh), or size-exclusion gel (Bio Gel P-2). Solutions were concentrated at <60 °C under diminished pressure.

Isopropyl 2,3-*O*-isopropylidene-1-thio-α-L-rhamnopyranoside (13)

To a mixture of **12** (5.0 g, 22.5 mmol) and 2,2-dimethoxypropane (2.9 mL, 23.6 mmol) in anhydrous acetone (25 mL) was added a catalytic amount of *p*-TsOH (100 mg). The reaction mixture was stirred for 1 h at rt, then neutralized with trimethylamine and concentrated. The crude product was purified by silica gel column chromatography (petroleum ether-ethyl

acetate 2:1) to give **13** (5.43 g, 92%) as a white solid. $[\alpha]_D^{25} = -162°$ (c 0.5, $CHCl_3$); ^1H NMR (600 MHz, $CDCl_3$): δ 5.56 (s, 1H, H-1^{Rha}), 4.16 (d, J = 6.0 Hz, 1H, H-2^{Rha}), 4.04–3.95 (m, 2H, H-3^{Rha}, H-5^{Rha}), 3.45–3.39 (m, 1H, H-4^{Rha}), 3.07–3.00 (m, 1H, -C*H*$(CH_3)_2$), 2.44 (d, J = 4.2 Hz, 1H, -O*H*), 1.53 (s, 3H, -C(CH_3)$_2$), 1.34 (s, 3H, -C(CH_3)$_2$), 1.33 (d, J = 6.0 Hz, 3H, -CH(CH_3)$_2$), 1.29, 1.28 (2 d, J = 6.6 Hz, 2×3H, H-6^{Rha}, -CH(CH_3)$_2$); ^{13}C NMR (150 MHz, $CDCl_3$): δ 109.49, 79.18, 78.39, 76.87, 75.37, 65.99, 34.69, 28.17, 26.35, 23.41, 23.21, 17.19; MALDI-TOF MS (positive ion): Calcd for $C_{12}H_{22}O_4SNa$ $[M+Na]^+$: 285.11; found m/z: 285.29.

Isopropyl 4-*O*-benzoyl-2,3-*O*-isopropylidene-1-thio-α-L-rhamnopyranoside (14). A solution of benzoyl chloride (3.0 mL, 26.3 mmol) in CH_2Cl_2 (5 mL) was added dropwise to a solution of **13** (5.30 g, 20.2 mmol) in anhydrous CH_2Cl_2 (20 mL) containing 10 mL of pyridine at 0 °C. The mixture was stirred for 3 h with the reaction temperature slowly warming up to rt. The reaction mixture was then diluted with CH_2Cl_2 (100 mL), and washed with 1 M HCl and brine. The organic layer was dried over anhydrous Na_2SO_4, filtered and concentrated. The resulting residue was purified by flash column chromatography (petroleum ether-ethyl acetate 10:1) to afford **14** (7.11 g, 96%) as a colorless syrup. $[\alpha]_D^{25} = -124°$ (c 0.5, $CHCl_3$); ^1H NMR (600 MHz, $CDCl_3$): δ 8.06 (d, J = 7.8 Hz, 2H, Ar*H*), 7.56 (t, J = 7.8 Hz, 1H, Ar*H*), 7.40 (t, J = 7.8 Hz, 2H, Ar*H*), 5.65 (s, 1H, H-1^{Rha}), 5.17 (dd, J = 9.6, 7.8 Hz, 1H, H-4^{Rha}), 4.32–4.22 (m, 3H, H-2^{Rha}, H-3^{Rha}, H-5^{Rha}), 3.12–3.05 (m, 1H, -C*H*$(CH_3)_2$), 1.63 (s, 3H, -C(CH_3)$_2$), 1.37 (d, J = 6.6 Hz, 3H, -CH(CH_3)$_2$), 1.34 (s, 3H, C(CH_3)$_2$), 1.32 (d, J = 6.6 Hz, 3H, -CH(CH_3)$_2$), 1.21 (d, J = 6.6 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 165.74, 133.16, 129.81 (2C), 129.77, 128.35 (2C), 109.87, 79.34,

76.94, 75.65, 75.45, 64.77, 34.90, 27.79, 26.51, 23.44, 23.27, 17.03; MALDI-TOF MS (positive ion): Calcd for $C_{19}H_{26}O_5SNa$ $[M+Na]^+$: 389.14; found m/z: 389.36.

Isopropyl 4-*O*-benzoyl-1-thio-α-L-rhamnopyranoside (9). A solution of **14** (6.50 g, 17.8 mmol) in CH_2Cl_2 (50 mL) was added 90% aqueous TFA (10 mL) at rt. The reaction mixture was stirred for 3 h, and then co-evaporated with toluene (3×50 mL). The resulting residue was purified by flash column chromatography (petroleum ether-ethyl acetate 1:1) to yield **9** (5.27 g, 91%) as a white solid. $[\alpha]_D^{25}$ = –98° (c 2, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.06 (d, J = 8.4 Hz, 2H, Ar*H*), 7.60 (t, J = 7.8 Hz, 1H, Ar*H*), 7.46 (t, J = 7.8 Hz, 2H, Ar*H*), 5.41 (s, 1H, H-1^{Rha}), 5.04 (t, J = 9.6 Hz, 1H, H-4^{Rha}), 4.42–4.34 (m, 1H, H-5^{Rha}), 4.12–4.06 (m, 1H, H-2^{Rha}), 4.02–3.94 (m, 1H, H-3^{Rha}), 3.32–3.26 (m, 1H, -O*H*), 3.14–3.04 (m, 1H, -C*H*$(CH_3)_2$), 2.83 (m, 1H, -O*H*), 1.35 (d, J = 6.6 Hz, 1H, -CH(C*H*$_3)_2$), 1.34 (d, J = 6.6 Hz, 1H, -CH(C*H*$_3)_2$), 1.29 (d, J = 6.6 Hz, 1H, H-6^{Rha}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 167.50, 133.52, 129.90 (2C), 129.36, 128.46 (2C), 83.26, 76.49, 72.71, 70.93, 66.28, 35.75, 23.75, 23.56, 17.44; MALDI-TOF MS (positive ion): Calcd for $C_{16}H_{22}O_5SNa$ $[M+Na]^+$: 349.11; found m/z: 349.37.

Isopropyl 2,4-di-*O*-benzoyl-1-thio-α-L-rhamnopyranoside (10). To a stirred solution of **9** (1.30 g, 3.99 mmol) in dry CH_2Cl_2 (10 mL) was added trimethyl orthobenzoate (1.53 mL, 7.98 mmol) at rt. The mixture was treated with a catalytic amount of (±)-10-camphorsulfonic acid for 1 h under nitrogen atmosphere, and then neutralized with trimethylamine and concentrated. To the residue generated above was added 80% HOAc, and after stirring 30 min, the mixture was concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 3:1) to yield **10** (1.42 g, 83%) as a white solid.

$[\alpha]_D^{25}$ = –25°(c 0.5, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.12 (d, J = 8.4 Hz, 2H, Ar*H*), 8.08 (d, J = 8.4 Hz, 2H, Ar*H*), 7.63–7.58 (m, 2H, Ar*H*), 7.52–7.45 (m, 4H, Ar*H*), 5.50 (d, J = 1.2 Hz, 1H, H-1^{Rha}), 5.47 (dd, J = 3.6, 1.8 Hz, 1H, H-2^{Rha}), 5.28 (t, J = 9.6 Hz, 1H, H-4^{Rha}), 4.46–4.38 (m, 1H, H-5^{Rha}), 4.27–4.20 (m, 1H, H-3^{Rha}), 3.18-3.09 (m, 1H, -C*H*$(CH_3)_2$), 2.49 (d, J = 7.8 Hz, 1H, -O*H*), 1.36 (d, J = 6.6 Hz, 6H, -CH(C*H*$_3$)$_2$) , 1.32 (d, J = 6.0 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 167.15, 166.01, 133.54, 129.89, 129.86, 129.41, 129.36, 128.58, 128.52, 81.65, 75.86, 75.47, 69.73, 66.92, 36.61, 23.83, 23.63, 17.55; MALDI-TOF MS (positive ion): Calcd for $C_{23}H_{26}O_6SNa$ $[M+Na]^+$: 453.13; found m/z: 453.45; Calcd for $C_{23}H_{26}O_6SK$ $[M+K]^+$: 469.11; found m/z: 469.44.

Isopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-1-thio-α-L-rhamnopyranoside (15) and Isopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→2)-4-*O*-benzoyl-1-thio-α-L-rhamnopyranoside (15a). After a mixture of **11** (2.52 g, 4.35 mmol), **9** (1.35 g, 4.14 mmol), and 4 Å molecular sieves in dry CH_2Cl_2 (20 mL) were stirred at rt for 30 min under a N_2 atmosphere, TMSOTf (79 μL, 0.435 mmol) were added dropwise at -15 ºC. The reaction mixture was stirred under these conditions for 1 h, and neutralized with triethylamine, filtered through a Celite pad, and concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 1:1) to yield **15** (1.73 g, 56%) and its β-1,2-linked regioisomer **15a** (583 mg, 19%) as a white foamy solid. Physical data of **15**: $[\alpha]_D^{25}$ = –86° (c 0.5, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.58 (d, J = 7.2 Hz, 2H, Ar*H*), 7.48–7.38 (m, 4H, Ar*H*), 7.23–7.11 (m, 3H, Ar*H*), 5.66 (dd, J = 10.8, 9.6 Hz, 1H, H-3^{GlcN}), 5.51 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 5.41 (s, 1H, H-1^{Rha}), 5.21 (t, J = 9.6 Hz, 1H, H-4^{Rha}), 5.06 (t, J = 9.6 Hz, 1H, H-4^{GlcN}), 4.31 (dd, J = 10.8, 8.4 Hz, 1H, H-2^{GlcN}),

4.27 (br s, 1H, H-2^{Rha}), 4.23–4.16 (m, 3H, H-5^{Rha}, H-6a^{GlcN}, H-6b^{GlcN}), 3.98 (dd, *J* = 9.6, 3.6 Hz, 1H, H-3^{Rha}), 3.90 (ddd, *J* = 9.6, 5.4, 3.0 Hz, 1H, H-5^{GlcN}), 3.09–3.00 (m, 1H, -C*H*(CH$_3$)$_2$), 2.95 (br s, 1H, -O*H*), 2.13, 2.00, 1.74 (3 s, 3×3H, 3×-*C*H$_3$CO), 1.32, 1.30 (2 d, *J* = 6.6 Hz, 2×3H, -CH(C*H*$_3$)$_2$), 1.06 (d, *J* = 6.0 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, CDCl$_3$): *δ* 170.71, 169.95, 169.38, 164.80, 133.85, 132.74, 129.43, 129.02, 128.14, 98.73 (C-1^{GlcN}), 82.42 (C-1^{Rha}), 80.16 (C-3^{Rha}), 72.34 (C-4^{Rha}), 71.93 (2C, C-2^{Rha}, C-5^{GlcN}), 70.27 (C-3^{GlcN}), 68.81 (C-4^{GlcN}), 66.53 (C-5^{Rha}), 62.04 (C-6^{GlcN}), 54.32 (C-2^{GlcN}), 35.46 (-*C*H(CH$_3$)$_2$), 23.60 (-CH(*C*H$_3$)$_2$), 23.49 (-CH(*C*H$_3$)$_2$), 20.78, 20.59, 20.29 (3C, 3×-*C*H$_3$CO), 17.22 (C-6^{Rha}); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for C$_{36}$H$_{45}$O$_{14}$N$_2$S [M+NH$_4$]$^+$: 761.2586; found m/z: 761.2591. Physical data of **15a**: [α]$_D^{25}$ = -78° (c 0.5, CHCl$_3$); ^{1}H NMR (600 MHz, CDCl$_3$): *δ* 7.92–7.84 (br s, 2H, Ar*H*), 7.82 (d, 2H, *J* = 7.8 Hz, Ar*H*), 7.72–7.67 (m, 2H, Ar*H*), 7.52 (t, *J* = 7.8 Hz, 1H, Ar*H*), 7.52 (t, *J* = 7.8 Hz, 2H, Ar*H*), 5.88 (dd, *J* = 10.8, 9.6 Hz, 1H, H-3^{GlcN}), 5.54 (d, *J* = 8.4 Hz, 1H, H-1^{GlcN}), 5.43 (s, 1H, H-1^{Rha}), 5.15 (t, *J* = 9.6 Hz, 1H, H-4^{GlcN}), 4.65 (t, *J* = 9.6 Hz, 1H, H-4^{Rha}), 4.47 (dd, *J* = 10.8, 8.4 Hz, 1H, H-2^{GlcN}), 4.29 (dd, *J* = 12.6, 5.4 Hz, 1H, H-6a^{GlcN}), 4.21–4.14 (m, 2H, H-5^{Rha}, H-6b^{GlcN}), 4.01 (d, *J* = 2.4 Hz, 1H, H-2^{Rha}), 3.93–3.89 (m, 1H, H-5^{GlcN}), 3.83–3.78 (m, 1H, H-3^{Rha}), 3.11–3.02 (m, 1H, -C*H*(CH$_3$)$_2$), 2.32 (d, *J* = 7.2 Hz, 1H, -O*H*), 2.10, 2.03, 1.88 (3 s, 3×3H, 3×-*C*H$_3$CO), 1.31, 1.30 (2 d, *J* = 6.6 Hz, 2×3H, -CH(C*H*$_3$)$_2$), 1.17 (d, *J* = 6.0 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, CDCl$_3$): *δ* 170.65, 170.13, 169.50, 166.94, 134.02, 133.32, 129.63, 129.23, 128.33, 100.16 (C-1^{GlcN}), 83.11 (C-1^{Rha}), 82.06 (C-2^{Rha}), 76.01 (C-4^{Rha}), 71.93 (C-5^{GlcN}), 70.60 (C-3^{Rha}), 70.43 (C-3^{GlcN}), 69.05 (C-4^{GlcN}), 66.52 (C-5^{Rha}), 62.14 (C-6^{GlcN}), 54.50 (C-2^{GlcN}), 36.15 (-*C*H(CH$_3$)$_2$), 23.75 (-CH(*C*H$_3$)$_2$), 23.63 (-CH(*C*H$_3$)$_2$), 20.78, 20.65, 20.52 (3C, 3×-*C*H$_3$CO), 17.37 (C-6^{Rha}); MALDI-TOF MS (positive ion): Calcd for C$_{36}$H$_{41}$O$_{14}$NSNa [M+Na]$^+$: 766.21; found m/z: 766.65.

Isopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-2-chloroacetyl-1-thio-α-L-rhamnopyranoside (16). Chloroacetyl chloride (0.8 mL, 11.6 mmol) in dry CH_2Cl_2 (5 ml) was introduced by dripping slowly at 0 °C to a stirred solution of **15** (1.65 g, 2.22 mmol) in CH_2Cl_2 (15 mL) and pyridine (5 mL). The reaction mixture was stirred for 2 h at 0 °C, and then diluted with ethyl acetate, washed with 1 M HCl and water. The organic phase was dried over Na_2SO_4, filtered, and concentrated. The residue was purified by column chromatography (petroleum ether-ethyl acetate 3:2) to give **16** (1.69 g, 93%) as a white foamy solid. $[\alpha]_D^{25} = -4°$ (c 1, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.59 (d, J = 8.4 Hz, 2H, Ar*H*), 7.53–7.36 (m, 4H, ArH), 7.23–7.11 (m, 3H, Ar*H*), 5.63 (t, J = 9.6 Hz, 1H, H-3^{GlcN}), 5.45 (br s, 1H, H-2^{Rha}), 5.44 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 5.30 (s, 1H, H-1^{Rha}), 5.14 (t, J = 9.6 Hz, 1H, H-4^{Rha}), 5.05 (t, J = 9.6 Hz, 1H, H-4^{GlcN}), 4.32–4.15 (m, 5H, -ClC*H*$_2$CO, H-2^{GlcN}, H-6a^{GlcN}, H-5^{Rha}), 4.15–4.07 (m, 2H, H-6b^{GlcN}, H-3^{Rha}), 3.85 (dd, J = 9.6, 3.6 Hz, 1H, H-5^{GlcN}), 3.10–3.02 (m, 1H, -C*H*(CH$_3$)$_2$), 2.14, 1.97, 1.73 (3 s, 3×3H, 3×-C*H*$_3$CO), 1.30, 1.31 (2 d, J = 6.6 Hz, 2×3H, -CH(C*H*$_3$)$_2$), 1.06 (d, J = 6.0 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.77, 169.96, 169.34, 166.85, 164.77, 133.82, 132.96, 130.67, 129.48, 128.76, 128.20, 123.14, 98.59 (C-1^{GlcN}), 81.04 (C-1^{Rha}), 76.35 (C-3^{Rha}), 75.33 (C-2^{Rha}), 72.25 (C-4^{Rha}), 71.75 (C-5^{GlcN}), 70.23 (C-3^{GlcN}), 68.54 (C-4^{GlcN}), 66.94 (C-5^{Rha}), 61.78 (C-6^{GlcN}), 54.39 (C-2^{GlcN}), 41.05 (-ClCH$_2$CO), 36.48 (-*C*H(CH$_3$)$_2$), 23.69 (-CH(*C*H$_3$)$_2$), 23.49 (-CH(*C*H$_3$)$_2$), 20.81, 20.58, 20.30 (3C, 3×-CH$_3$CO), 17.21 (C-6^{Rha}); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{38}H_{46}O_{15}N_2ClS$ $[M+NH_4]^+$: 837.2302; found m/z: 837.2310.

3-Azidopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-2-chloroacetyl-α-L-rhamnopyranoside (17). A solution of **16** (500 mg, 0.61 mmol), 3-azido-1-propane (123 mg, 1.22 mmol), and 4 Å molecular sieves in dry CH_2Cl_2 (5 mL) were stirred at rt for 20 min under a N_2 atmosphere. Then, NIS (206 mg, 0.916 mmol) and TMSOTf (16.6 μL, 91.6 μmol) were sequentially added at 0 ºC. After stirring for 1 h, the reaction mixture was neutralized with triethylamine, filtered through a Celite pad and concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 3:2) to yield **17** (438 mg, 85%) as a white foamy solid. $[\alpha]_D^{25}$ = +18° (c 0.5, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.59 (d, J = 7.8 Hz, 2H, Ar*H*), 7.51–7.38 (m, 4H, Ar*H*), 7.30–7.20 (m, 3H, Ar*H*), 5.64 (t, J = 10.2 Hz, 1H, H-3^{GlcN}), 5.49 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 5.35 (br s, 1H, H-2^{Rha}), 5.15 (t, J = 9.6 Hz, 1H, H-4^{Rha}), 5.11 (t, J = 9.6 Hz, 1H, H-4^{GlcN}), 4.78 (s, 1H, H-1^{Rha}), 4.34–4.11 (m, 6H, H-3^{Rha}, H-2^{GlcN}, -ClC*H*$_2$CO, H-6a^{GlcN}, H-6b^{GlcN}), 3.87–3.74 (m, 3H, H-5^{GlcN}, H-5^{Rha}, -OC*H*$_2$CH$_2$-), 3.57–3.50 (m, 1H, -OC*H*$_2$CH$_2$-), 3.45 (t, J = 6.6 Hz, 2H, -C*H*$_2$N$_3$), 2.14, 1.98, 1.74 (3 s, 3×3H, 3×-C*H*$_3$CO), 1.96–1.85 (m, 2H, -OCH$_2$C*H*$_2$CH$_2$N$_3$), 1.08 (d, J = 6.6 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.69, 170.00, 169.30, 166.91, 164.75, 133.84, 132.93, 129.43, 128.81, 128.21, 98.48 (C-1^{GlcN}), 96.92 (C-1^{Rha}), 75.79 (C-3^{Rha}), 72.83 (C-2^{Rha}), 71.94 (H-4^{Rha}), 71.69 (C-5^{GlcN}), 70.30 (C-3^{GlcN}), 68.36 (C-4^{GlcN}), 66.30 (C-5^{Rha}), 64.95 (-O*C*H$_2$CH$_2$-), 61.31 (C-6^{GlcN}), 54.45 (C-2^{GlcN}), 48.35 (-*C*H$_2$N$_3$), 41.01 (-Cl*C*H$_2$CO), 28.69 (-OCH$_2$*C*H$_2$CH$_2$N$_3$), 20.72, 20.58, 20.30 (3C, 3×-*C*H$_3$CO), 17.31 (C-6^{Rha}); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{38}H_{41}O_{16}N_4Cl$ $[M+Na]^+$: 867.2098; found m/z: 867.2164.

3-Azidopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-α-L-rhamnopyranoside (6). To a solution of **17** (350 mg, 0.415 mmol) in methanol (20 mL) and CH_2Cl_2 (5 mL) was added

thiourea (158 mg, 2.08 mmol) and 2,6-lutidine (49 μL, 0.415 mmol) at rt. The reaction mixture was stirred at 70 ºC for overnight, and then cooled down to rt, and diluted with ethyl acetate, washed with 1 M HCl and water. The organic phase was dried over Na_2SO_4 and concentrated. The residue was purified by column chromatography (petroleum ether-ethyl acetate 1:1) to give **6** (293 mg, 92%) as a white foamy solid. $[\alpha]_D^{25}$ = +25° (c 0.5, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.56 (d, J = 8.4 Hz, 2H, Ar*H*), 7.46–7.26 (m, 5H, Ar*H*), 7.13 (t, J =7.8 Hz, 2H, Ar*H*), 5.65 (dd, J = 10.2, 9.6 Hz, 1H, H-3^{GlcN}), 5.54 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 5.19 (t, J = 9.6 Hz, 1H, H-4^{Rha}), 5.09 (t, J = 9.6 Hz, 1H, H-4^{GlcN}), 4.84 (s, 1H, H-1^{Rha}), 4.31 (dd, J = 10.2, 8.4 Hz, 1H, H-2^{GlcN}), 4.24–4.19 (m, 2H, H-6a^{GlcN}, H-6b^{GlcN}), 4.15 (br s, 1H, H-2^{Rha}), 4.00 (dd, J = 9.6, 3.0 Hz, 1H, H-3^{Rha}), 3.93–3.86 (m, 1H, H-5^{GlcN}), 3.82–3.72 (m, 2H, H-5^{Rha}, -OC*H*$_2$CH$_2$-), 3.54–3.47 (m, 1H, -OC*H*$_2$CH$_2$-), 3.42 (t, J = 6.6 Hz, 2H, -C*H*$_2$N$_3$), 2.87 (s, 1H, -O*H*), 2.10, 1.99, 1.74 (3 s, 3×3H, 3×-C*H*$_3$CO), 1.95–1.83 (m, 2H, -OCH$_2$C*H*$_2$CH$_2$N$_3$), 1.06 (d, J = 6.0 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.63, 169.98, 169.34, 164.75, 133.87, 132.69, 129.33, 129.02, 128.12, 99.12 (C-1^{Rha}), 98.49 (C-1^{GlcN}), 79.83 (C-3^{Rha}), 71.88 (C-4^{Rha}), 71.85 (C-5^{GlcN}), 70.31 (C-3^{GlcN}), 69.96 (C-2^{Rha}), 68.72 (C-4^{GlcN}), 66.13 (H-5^{Rha}), 64.51 (-O*C*H$_2$CH$_2$-), 61.82 (H-6^{GlcN}), 54.31 (C-2^{GlcN}), 48.45 (-*C*H$_2$N$_3$), 28.74 (-OCH$_2$*C*H$_2$CH$_2$N$_3$), 20.68, 20.58, 20.28 (3C, 3×-*C*H$_3$CO), 17.33 (C-6^{Rha}); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{36}H_{44}O_{15}N_5$ $[M+NH_4]^+$: 786.2828; found m/z: 786.2837.

3-Azidopropyl 2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→2)-[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranoside (18). After a mixture of **8** (169 mg, 0.390 mmol), **6** (200 mg, 0.26 mmol), and 4 Å molecular sieves in dry CH_2Cl_2 (3 mL) were stirred at rt for 20 min under a N2 atmosphere, TMSOTf

(7.1 μL, 39 μmol) were added at 0 °C. The reaction mixture was stirred under these conditions for 1 h, and then neutralized with triethylamine, filtered through a Celite pad, and concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 3:2) to yield **18** (239 mg, 88%) as a white foamy solid. $[\alpha]_D^{25}$ = –18° (c 1, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.65 (br s, 1H, Ar*H*), 7.56 (d, *J* = 7.8 Hz, 2H, Ar*H*), 7.52 (br s, 1H, Ar*H*), 7.44 (t, *J* = 7.2 Hz, 1H, Ar*H*), 7.37 (br s, 1H, Ar*H*), 7.22 (t, *J* = 7.8 Hz, 2H, Ar*H*), 7.05 (br s, 1H, Ar*H*), 5.69 (dd, *J* = 10.8, 9.6 Hz, 1H, H-3$^{GlcN-B)}$), 5.45 (d, *J* = 8.4 Hz, 1H, H-1^{GlcN-B}), 5.39 (dd, *J* = 10.2, 3.6 Hz, 1H, H-3^{Rha-C}), 5.33 (dd, *J* = 3.6, 1.8 Hz, 1H, H-2^{Rha-C}), 5.26 (d, *J* = 1.2 Hz, 1H, H-1^{Rha-C}), 5.15 (t, *J* = 9.6, 1H, H-4^{GlcN-B}), 5.06 (t, *J* = 9.6 Hz, 1H, H-4^{Rha-A}), 5.05 (t, *J* = 10.2 Hz, 1H, H-4^{Rha-C}), 4.66 (d, *J* = 1.8 Hz, 1H, H-1^{Rha-A}), 4.53 (dd, *J* = 10.8, 8.4 Hz, 1H, H-2^{GlcN-B}), 4.22 (dd, *J* = 12.0, 2.4 Hz, 1H, H-6a^{GlcN-B}), 4.16 (dd, *J* = 3.0, 1.8 Hz, 1H, H-2^{Rha-A}), 4.14–4.08 (m, 2H, H-3^{Rha-A}, H-6b^{GlcN-B}), 3.97–3.89 (m, 1H, H-5^{Rha-C}), 3.84 (ddd, *J* = 9.6, 4.8, 2.4 Hz, 1H, H-5^{GlcN-B}), 3.79–3.70 (m, 2H, H-5^{Rha-A}, -OC*H*$_2$CH$_2$-), 3.52–3.46 (m, 1H, -OC*H*$_2$CH$_2$-), 3.46–3.37 (m, 2H, -C*H*$_2$N$_3$), 2.25, 2.10, 2.03, 2.02, 1.98, 1.75 (6 s, 6×3H, 6×-C*H*$_3$CO), 1.95–1.85 (m, 2H, -OCH$_2$C*H*$_2$CH$_2$N$_3$), 1.19 (d, *J* = 6.6 Hz, 3H, H-6^{Rha-C}), 1.06 (d, *J* = 6.6 Hz, 3H, H-6^{Rha-A}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.67, 170.17, 170.06, 169.70, 169.64, 169.40, 164.53, 133.65, 132.70, 130.88, 129.44, 129.15, 128.14, 99.08 (C-1^{GlcN-B}), 98.96 (H-1^{Rha-C}), 98.93 (H-1^{Rha-A}), 77.34 (C-3^{Rha-A}), 77.06 (C-2^{Rha-A}), 72.51 (C-4^{Rha-C}), 71.84 (C-4^{Rha-A}), 71.32 (C-5^{GlcN-B}), 70.11 (C-2^{Rha-C}), 70.08 (C-3^{GlcN-B}), 68.86 (C-4^{GlcN-B}), 68.58 (C-3^{Rha-C}), 66.66 (C-5^{Rha-C}), 66.55 (H-5^{Rha-A}), 64.45 (-O*C*H$_2$CH$_2$-), 62.03 (C-6^{GlcN-B}), 54.17 (C-2^{GlcN-B}), 48.40 (-*C*H$_2$N$_3$), 28.74 (-OCH$_2$*C*H$_2$CH$_2$N$_3$), 21.09, 20.83, 20.63, 20.38 (6C, 6×-C*H*$_3$CO), 17.45 (C-6^{Rha-C}), 17.42 (C-6^{Rha-A}); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{48}H_{60}O_{22}N_5$ $[M+NH_4]^+$: 1058.3724; found m/z: 1058.3733.

3-Azidopropyl α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranoside (5a). Ammonia was bubbled into a mixture of **18** (200 mg, 0.192 mmol) in MeOH (30 mL) at 0 °C until saturation. The reaction mixture was kept at rt for 7 days and then evaporated to dryness. The product was dissolved in dry methanol (10 mL) containing acetic anhydride (1.0 mL). The reaction mixture was stirred at rt overnight and then concentrated. The resulting residue was purified by size exclusion chromatography on a Bio-Gel P-2 column with distilled water as the eluent and then lyophilized to give **5a** (98 mg, 86%) as a white foamy solid. $[\alpha]_D^{25} = -21°$ (c 0.1, H_2O); ^{1}H NMR (600 MHz, D_2O): δ 4.98 (d, J = 1.2 Hz, 1H, H-1$^{Rha\text{-}C}$), 4.65 (1H, H-1$^{Rha\text{-}A}$ overlapped in H_2O), 4.50 (d, J = 9.0 Hz, 1H, H-1$^{GlcN\text{-}B}$), 4.00 (dd, J = 3.0, 1.2 Hz, 1H, H-2$^{Rha\text{-}A}$), 3.86 (dd, J = 3.6, 1.8 Hz, 1H, H-2$^{Rha\text{-}C}$), 3.76–3.72 (m, 1H, H-6a$^{GlcN\text{-}B}$), 3.69 (dd, J = 9.6, 3.0 Hz, 1H, H-3$^{Rha\text{-}A}$), 3.66–3.62 (m, 1H, -OCH_2CH$_2$-), 3.62–3.50 (m, 5H, H-6b$^{GlcN\text{-}B}$, H-2$^{GlcN\text{-}B}$, H-3$^{Rha\text{-}C}$, H-5$^{Rha\text{-}A}$, H-5$^{Rha\text{-}C}$), 3.44–3.38 (m, 1H, -OCH_2CH$_2$-), 3.38–3.31 (m, 2H, H-3$^{GlcN\text{-}B}$, H-4$^{Rha\text{-}A}$), 3.30–3.21 (m, 5H, H-4$^{GlcN\text{-}B}$, H-5$^{GlcN\text{-}B}$, H-4$^{Rha\text{-}C}$, -CH_2N$_3$), 1.84 (s, 3H, -CH_3CO), 1.78–1.67 (m, 2H, -OCH$_2$CH_2CH$_2$N$_3$), 1.11 (d, J = 6.6 Hz, 3H, H-6$^{Rha\text{-}A}$), 1.09 (d, J = 6.6 Hz, 3H, H-6$^{Rha\text{-}C}$); ^{13}C NMR (150 MHz, D_2O): δ 174.57, 102.50 (C-1$^{GlcN\text{-}B}$), 101.45 (C-1$^{Rha\text{-}C}$), 98.48 (C-1$^{Rha\text{-}A}$), 79.83 (C-3$^{Rha\text{-}A}$), 76.58 (C-2$^{Rha\text{-}A}$), 75.59 (C-4$^{GlcN\text{-}B}$), 73.60 (C-3$^{GlcN\text{-}B}$), 71.87 (C-4$^{Rha\text{-}C}$), 70.87 (C-4$^{Rha\text{-}A}$), 69.83 (C-3$^{Rha\text{-}C}$), 69.79 (C-2$^{Rha\text{-}C}$), 69.63 (C-5$^{GlcN\text{-}B}$), 68.97 (C-5$^{Rha\text{-}C}$), 68.88 (C-5$^{Rha\text{-}A}$), 64.94 (-OCH$_2$CH$_2$-), 60.66 (C-6$^{GlcN\text{-}B}$), 55.70 (C-2$^{GlcN\text{-}B}$), 48.20 (-CH$_2$NH$_2$), 27.70 (-OCH$_2$$CH_2CH_2N_3$), 22.05 (-C$H_3$CO), 16.48 (C-6$^{Rha\text{-}A}$), 16.45 (C-6$^{Rha\text{-}C}$); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{23}H_{41}O_{14}N_4$ $[M+H]^+$: 597.2614; found m/z: 597.2610.

3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-2-chloroacetyl-α-L-rhamnopyranosyl trichloroacetimidate (19). To a solution of **16** (1.60 g, 1.95 mmol) in CH_2Cl_2 (25 mL) containing CH_3CN (0.6 mL) and H_2O (0.2 mL) was added NIS (655 mg, 2.93 mmol) and AgOTf (150 mg, 0.585 mmol) at rt. The reaction mixture was stirred at same temperature for 2 h, and then neutralized with triethylamine and concentrated, and the residue was briefly purified by flash column chromatography (petroleum ether–ethyl acetate 1:1). The resultant hemiacetal was then dissolved in CH_2Cl_2 (5 mL) at 0 ºC, and trichloroacetonitrile (0.38 mL, 3.90 mmol) and DBU (38 uL) were added. The reaction mixture was stirred at 0 ºC for 1 h, and then concentrated. The residue was subjected to flash column chromatography (petroleum ether–ethyl acetate 2:1) to give **19** (1.24 g, 71% for 2 steps) as a white foamy solid. $[\alpha]_D^{25}$ = +23° (c 0.2, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.78 (s, 1H, N*H*), 7.63–7.36 (d, J = 8.4 Hz, 6H, Ar*H*), 7.25 (t, J = 7.8 Hz, 2H, Ar*H*), 7.16 (br s, 1H, Ar*H*), 6.25 (d, J = 2.4 Hz, 1H, H-1^{Rha}), 5.66 (dd, J = 10.8, 9.0 Hz, 1H, H-3^{GlcN}), 5.57 (dd, J = 3.6, 2.4 Hz, 1H, H-2^{Rha}), 5.49 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 5.22 (t, J = 10.2 Hz, 1H, H-4^{Rha}), 5.13 (dd, J = 10.2, 9.0 Hz, 1H, H-4^{GlcN}), 4.31 (d, J = 15.6 Hz, 1H, -ClC*H*$_2$CO), 4.26 (dd, J = 10.2, 3.6 Hz, 1H, H-3^{Rha}), 4.22 (d, J = 15.6 Hz, 1H, -ClC*H*$_2$CO), 4.21 (dd, J = 10.8, 7.8 Hz, 1H, H-2^{GlcN}), 4.19–4.16 (m, 2H, H-6a^{GlcN}, H-6b^{GlcN}), 4.05-3.98 (m, 1H, H-5^{Rha}), 3.83 (dt, J = 10.2, 3.0 Hz, 1H, H-5^{GlcN}), 2.11, 1.98, 1.75 (3 s, 3×3H, 3×-C*H*$_3$CO), 1.10 (d, J = 6.6 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.71, 169.97, 169.30, 166.40, 164.71, 159.61, 133.87, 133.14, 130.74, 129.52, 128.58, 128.26, 123.17, 98.58, 94.06, 90.71, 75.02, 71.80, 71.55, 71.39, 70.11, 69.08, 68.48, 61.38, 54.45, 40.79, 20.74, 20.58, 20.32, 17.36.

Isopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-2-chloroacetyl-α-L-rhamnopyranosyl-(1→3)-2,4-di-*O*-benzoyl-1-thio-α-L-rhamnopyranoside (7). After a mixture of **10** (343 mg, 0.799 mmol), 19 (795 mg, 0.879 mmol), and 4 Å molecular sieves in dry CH_2Cl_2 (8 mL) were stirred at rt for 20 min under a N_2 atmosphere, TMSOTf (16 μL, 88 μmol) were added at 0 ºC. The reaction mixture was stirred under these conditions for 1 h, at which time TLC (petroleum ether-ethyl acetate 2:1) indicated the complete disappearance of **10**, and then neutralized with triethylamine, filtered through a Celite pad, and concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 2:1) to yield **7** (705 mg, 68%) as a white foamy solid. $[\alpha]_D^{25}$ = +33° (c 1, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.21 (d, J = 8.4 Hz, 2H, Ar*H*), 8.18 (d, J = 8.4 Hz, 2H, Ar*H*), 7.65–7.59 (m, 2H, Ar*H*), 7.58–7.51 (m, 4H, Ar*H*), 7.42 (br s, 2H, Ar*H*), 7.39–7.30 (m, 5H, Ar*H*), 7.09 (t, J = 7.8 Hz, 2H, Ar*H*), 5.54 (t, J = 9.6 Hz, 1H, H-4$^{Rha\text{-}C}$), 5.52 (dd, J = 3.0, 1.8 Hz, 1H, H-2$^{Rha\text{-}C}$), 5.50 (dd, J = 10.2, 9.6 Hz, 1H, H-3$^{GlcN\text{-}E}$), 5.47 (d, J = 1.8 Hz, 1H, H-1$^{Rha\text{-}C}$), 5.06 (d, J = 8.4 Hz, 1H, H-1$^{GlcN\text{-}E}$), 4.96 (t, J = 9.6 Hz, 1H, H-4$^{Rha\text{-}D}$), 4.95–4.89 (m, 3H, H-1$^{Rha\text{-}D}$, H-2$^{Rha\text{-}D}$, H-4$^{GlcN\text{-}E}$), 4.44–4.37 (m, 1H, H-5$^{Rha\text{-}C}$), 4.26 (dd, J = 9.6, 3.0 Hz, 1H, H-3$^{Rha\text{-}C}$), 4.10 (d, J = 15.6 Hz, 1H, -ClC*H*$_2$CO), 4.09 (dd, J = 10.2, 8.4 Hz, 1H, H-2$^{GlcN\text{-}E}$), 4.01 (d, J = 15.6 Hz, 1H, -ClC*H*$_2$CO), 4.00 (dd, J = 12.6, 6.6 Hz, 1H, H-6a$^{GlcN\text{-}E}$), 3.93 (dd, J = 9.6, 3.6 Hz, 1H, H-3$^{Rha\text{-}D}$), 3.80–3.73 (m, 1H, H-5$^{Rha\text{-}D}$), 3.38 (dd, J = 12.6, 1.8 Hz, 1H, H-6b$^{GlcN\text{-}E}$), 3.15–3.09 (m, 1H, -C*H*(CH$_3$)$_2$), 2.90 (dt, J = 10.2, 1.8 Hz, 1H, H-5$^{GlcN\text{-}E}$), 2.03, 1.97, 1.71 (3 s, 3×3H, 3×-C*H*$_3$CO), 1.35 (d, J = 7.2 Hz, 6H, -CH(C*H*$_3$)$_2$), 1.32 (d, J = 6.0 Hz, 1H, H-6$^{Rha\text{-}D}$), 0.79 (d, J = 6.0 Hz, 1H, H-6$^{Rha\text{-}C}$); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.61, 169.96, 169.14, 166.44, 165.79, 165.68, 164.49, 133.72, 133.56, 133.49, 132.72, 129.98, 129.91, 129.28, 128.77, 128.69, 128.04, 123.09, 98.37 (C-1$^{GlcN\text{-}E}$), 98.27 (C-1$^{Rha\text{-}D}$), 81.41 (C-1$^{Rha\text{-}C}$), 77.14 (C-3$^{Rha\text{-}C}$), 75.72 (C-3$^{Rha\text{-}D}$), 74.21 (C-2$^{Rha\text{-}C}$), 73.11 (C-4$^{Rha\text{-}C}$), 72.42 (C-2$^{Rha\text{-}D}$), 71.49 (C-4$^{Rha\text{-}}$

^{D}), 71.15 (C-5^{GlcN-E}), 70.19 (C-3^{GlcN-E}), 67.77 (C-4^{GlcN-E}), 67.24 (C-5^{Rha-D}), 67.15 (C-5^{Rha-C}), 60.49 (C-6^{GlcN-E}), 54.13 (C-2^{GlcN-E}), 40.81 (-Cl*C*H$_2$CO), 36.68 (-*C*H(CH$_3$)$_2$), 23.75 (-CH(*C*H$_3$)$_2$), 23.58 (-CH(*C*H$_3$)$_2$), 20.68, 20.56, 20.30 (3C, 3×-*C*H$_3$CO), 17.57 (C-6^{Rha-C}), 16.94 (C-6^{Rha-D}); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{58}H_{64}O_{21}N_2ClS$ $[M+NH_4]^+$: 1191.3405; found m/z: 1191.3409.

Isopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-α-L-rhamnopyranosyl-(1→3)-2,4-di-*O*-benzoyl-1-α-L-rhamnopyranoside (20). To a solution of **7** (300 mg, 0.256 mmol) in methanol (20 mL) and CH_2Cl_2 (5 mL) was added thiourea (98 mg, 1.28 mmol) and 2,6-lutidine (30 μL, 0.256 mmol) at rt. The reaction mixture was stirred at 70 ºC for overnight, at which time TLC showed the disappearance of **7** (petroleum ether-ethyl acetate 2:1). After cooling down to rt, the mixture was diluted with ethyl acetate (50 mL), and then washed with 1 M HCl and water. The organic phase was dried over Na_2SO_4 and concentrated, and the residue was purified by column chromatography to give **20** (255 mg, 91 %) as a white foamy solid. $[\alpha]_D^{25}$ = +20° (c 0.5, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.19 (d, J = 8.4 Hz, 2H, Ar*H*), 8.14 (d, J = 8.4 Hz, 2H, Ar*H*), 7.67–7.59 (m, 2H, Ar*H*), 7.57 (t, J = 7.8 Hz, 2H, Ar*H*), 7.52 (t, J = 7.8 Hz, 2H, Ar*H*), 7.44–7.39 (m, 2H, Ar*H*), 7.37–7.27 (m, 5H, Ar*H*), 7.08 (t, J = 7.8 Hz, 2H, ArH), 5.56 (dd, J = 10.8, 9.0 Hz, 1H, H-3^{GlcN-E}), 5.55 (d, J = 3.6, 1.2 Hz, 1H, H-2^{Rha-C}), 5.52 (t, J = 9.6 Hz, 1H, H-4^{Rha-C}), 5.48 (d, J = 1.2 Hz, 1H, H-1^{Rha-C}), 5.15 (d, J = 8.4 Hz, 1H, H-1^{GlcN-E}), 5.03 (t, J = 9.6 Hz, 1H, H-4^{Rha-D}), 4.96 (d, J = 1.2 Hz, 1H, H-1^{Rha-D}), 4.95 (t, J = 9.6 Hz, 1H, H-4^{GlcN-E}), 4.45–4.36 (m, 1H, H-5^{Rha-C}), 4.28 (dd, J = 9.6, 3.6 Hz, 1H, H-3^{Rha-C}), 4.17 (dd, J =10.8, 8.4 Hz, 1H, H-2^{GlcN-E}), 4.03 (dd, J =12.6, 4.2 Hz, 1H, H-6a^{GlcN-E}), 3.83–3.74 (m, 3H, H-2^{Rha-D}, H-3^{Rha-D}, H-5^{Rha-D}), 3.62 (dd, J = 12.6, 1.8 Hz, 1H, H-6b^{GlcN-E}), 3.24–3.19 (m, 1H, H-

$5^{GlcN\text{-}E}$), 3.17–3.08 (m, 1H, -C*H*(CH_3)$_2$), 1.99, 1.95, 1.72 (3 s, 3×3H, 3×-C*H*$_3$CO), 1.35 (d, *J* = 6.6 Hz, 6H, -CH(C*H*$_3$)$_2$), 1.30 (d, *J* = 6.0 Hz, 3H, H-$6^{Rha\text{-}C}$), 0.87 (d, *J* = 6.0 Hz, 3H, H-$6^{Rha\text{-}D}$); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.51, 169.92, 169.20, 165.83, 165.51, 164.57, 133.72, 133.57, 133.44, 132.52, 129.94, 129.76, 129.68, 129.47, 129.29, 129.00, 128.79, 128.69, 127.99, 100.79 (C-$1^{Rha\text{-}D}$), 98.51 (C-$1^{GlcN\text{-}E}$), 81.58 (C-$1^{Rha\text{-}C}$), 79.44 (C-$3^{Rha\text{-}D}$), 75.89 (C-$3^{Rha\text{-}C}$), 74.47 (C-$2^{Rha\text{-}C}$), 73.65 (C-$4^{Rha\text{-}C}$), 71.54 (C-$4^{Rha\text{-}D}$), 71.46 (C-$5^{GlcN\text{-}E}$), 70.20 (C-$3^{GlcN\text{-}E}$), 69.96 (C-$2^{Rha\text{-}D}$), 68.26 (C-$4^{GlcN\text{-}E}$), 67.25 (C-$5^{Rha\text{-}C}$), 67.05 (C-$5^{Rha\text{-}D}$), 61.02 (C-$6^{GlcN\text{-}E}$), 54.09 (C-$2^{GlcN\text{-}E}$), 36.60 (-*C*H(CH_3)$_2$), 23.77 (-CH(*C*H$_3$)$_2$), 23.58 (-CH(*C*H$_3$)$_2$), 20.58 (2C, 2×-*C*H$_3$CO) 20.28 (*C*H$_3$CO), 17.54 (C-$6^{Rha\text{-}C}$), 17.06 (C-$6^{Rha\text{-}D}$); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{56}H_{63}O_{20}N_2S$ $[M+NH_4]^+$: 1115.3689; found m/z: 1115.3688.

Isopropyl 2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→2)-[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranosyl-(1→3)-2,4-di-*O*-benzoyl-1-thio-α-L-rhamnopyranoside (21). After a mixture of **8** (103 mg, 0.238 mmol), **20** (200 mg, 0.182 mmol), and 4 Å molecular sieves in dry CH_2Cl_2 (4 mL) were stirred at rt for 30 min under a N_2 atmosphere, TMSOTf (5 μL, 23.8 μmol) were added at 0 °C. The reaction mixture was stirred under these conditions for 2 h, and then neutralized with triethylamine, filtered through a Celite pad, and concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 1:1) to yield **21** (215 mg, 86%) as a white foamy solid. $[\alpha]_D^{25}$ = –4° (c 0.1, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.21 (d, *J* = 8.4 Hz, 2H, Ar*H*), 8.08 (d, *J* = 8.4 Hz, 2H, Ar*H*), 7.69 (t, *J* = 7.8 Hz, 1H, Ar*H*), 7.64–7.31 (m, 11H, Ar*H*), 7.16 (t, *J* = 7.8 Hz, 2H, Ar*H*), 7.09 (d, *J* = 6.6 Hz, 1H, Ar*H*), 5.60 (dd, *J* = 10.8, 9.6 Hz, 1H, H-$3^{GlcN\text{-}E}$), 5.55 (dd, *J* = 3.6, 1.2 Hz, 1H, H-$2^{Rha\text{-}C}$), 5.50 (t, *J* = 9.6 Hz, 1H, H-$4^{Rha\text{-}C}$),

5.47 (d, J = 1.2 Hz, 1H, H-1$^{Rha\text{-}C}$), 5.25 (dd, J = 9.6, 3.6 Hz, 1H, H-3$^{Rha\text{-}F}$), 5.20 (dd, J = 3.6, 1.8 Hz, 1H, H-2$^{Rha\text{-}F}$), 5.11 (d, J = 8.4 Hz, 1H, H-1$^{GlcN\text{-}E}$), 5.04 (d, J = 1.8 Hz, 1H, H-1$^{Rha\text{-}F}$), 4.98 (t, J = 10.2 Hz, H-4$^{GlcN\text{-}E}$), 4.95 (t, J = 9.6 Hz, 1H, H-4$^{Rha\text{-}D}$), 4.88 (t, J = 9.6 Hz, 1H, H-4$^{Rha\text{-}F}$), 4.83 (d, J = 1.8 Hz, 1H, H-1$^{Rha\text{-}D}$), 4.41–4.36 (m, 1H, H-5$^{Rha\text{-}C}$), 4.35 (dd, J = 10.8, 8.4 Hz, 1H, H-2$^{GlcN\text{-}E}$), 4.28 (dd, J = 9.6, 3.6 Hz, 1H, H-3$^{Rha\text{-}C}$), 4.04 (dd, J = 12.0, 4.2 Hz, 1H, H-6a$^{GlcN\text{-}E}$), 3.93 (dd, J = 3.0, 1.8 Hz, 1H, H-2$^{Rha\text{-}D}$), 3.91 (dd, J = 9.6, 3.0 Hz, 1H, H-3$^{Rha\text{-}D}$), 3.81–3.72 (m, 1H, H-5$^{Rha\text{-}D}$), 3.62–3.55 (m, 2H, H-6b$^{GlcN\text{-}E}$, H-5$^{Rha\text{-}F}$), 3.28 (ddd, J = 10.2, 4.2, 2.4 Hz, 1H, H-5$^{GlcN\text{-}E}$), 3.16–3.07 (m, 1H, -C*H*$(CH_3)_2$), 2.17, 2.00, 1.99, 1.98, 1.97, 1.72 (6 s, 6×3H, 6×-C*H*$_3$CO), 1.35 (d, J = 6.6 Hz, 6H, -CH(C*H*$_3$)$_2$), 1.28 (d, J = 6.6 Hz, 3H, H-6$^{Rha\text{-}C}$), 0.92 (d, J = 6.6 Hz, 3H, H-6$^{Rha\text{-}D}$), 0.71 (d, J = 6.0 Hz, 3H, H-6$^{Rha\text{-}F}$); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.49, 170.08, 169.99, 169.50 (2C), 169.27, 165.82, 165.40, 164.31, 133.64, 133.57, 133.40, 132.53, 129.94, 129.73, 129.61, 129.47, 129.37, 129.12, 128.77, 128.75, 128.02, 100.33 (C-1$^{Rha\text{-}D}$), 99.14 (C-1$^{GlcN\text{-}E}$), 98.41 (C-1$^{Rha\text{-}F}$), 81.57 (C-1$^{Rha\text{-}C}$), 77.12 (C-3$^{Rha\text{-}D}$), 76.42 (C-2$^{Rha\text{-}D}$), 74.97 (C-3$^{Rha\text{-}C}$), 74.33 (C-2$^{Rha\text{-}C}$), 73.69 (C-4$^{Rha\text{-}C}$), 72.04 (C-4$^{Rha\text{-}D}$), 71.68 (C-4$^{Rha\text{-}F}$), 71.19 (C-5$^{GlcN\text{-}E}$), 69.97 (C-3$^{GlcN\text{-}E}$), 69.83 (C-2$^{Rha\text{-}F}$), 68.51 (C-3$^{Rha\text{-}F}$), 68.39 (C-4$^{GlcN\text{-}E}$), 67.63 (C-5$^{Rha\text{-}D}$), 67.34 (C-5$^{Rha\text{-}C}$), 66.50 (C-5$^{Rha\text{-}F}$), 61.27 (C-6$^{GlcN\text{-}E}$), 54.01 (C-2$^{GlcN\text{-}E}$), 36.60 (-*C*H$(CH_3)_2$), 23.76 (-CH(*C*H$_3$)$_2$), 23.57 (-CH(*C*H$_3$)$_2$), 21.00, 20.82, 20.75, 20.62, 20.59, 20.36 (6C, 6×-*C*H$_3$CO), 17.58 (C-6$^{Rha\text{-}C}$), 17.18 (C-6$^{Rha\text{-}D}$), 16.94 (C-6$^{Rha\text{-}F}$); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{68}H_{79}O_{27}N_2S$ $[M+NH_4]^+$: 1387.4585; found m/z: 1387.4580.

3-Azidopropyl 2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→2)-[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranosyl-(1→3)-2,4-di-*O*-benzoyl-α-L-rhamnopyranosyl-(1→2)-

[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranoside (22). A solution of **6** (37 mg, 47.6 μmol), **21** (82 mg, 59.6 μmol), and 4 Å molecular sieves in dry CH_2Cl_2 (3 mL) were stirred at rt for 30 min under a N_2 atmosphere. The mixture was then cooled down to 0 ºC, and NIS (16 mg, 71.2 μmol) and TMSOTf (1.3 μL, 7.12 μmol) were added sequentially. After stirring at 0 ºC for 1 h, the reaction mixture was neutralized with triethylamine, filtered through a Celite pad, and concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 2:3) to yield **22** (76 mg, 78%) as a white foamy solid. $[\alpha]_D^{25}$ = +13° (c 0.2, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.36 (d, J = 7.2 Hz, 2H, Ar*H*), 8.11 (d, J = 7.2 Hz, 2H, Ar*H*), 7.72–7.32 (m, 16H, Ar*H*), 7.28–7.08 (m, 7H, Ar*H*), 7.90 (d, J = 7.2 Hz, 1H, Ar*H*), 5.70 (dd, J = 10.2, 9.0 Hz, 1H, H-3^{GlcN-B}), 5.64 (dd, J = 10.2, 9.0 Hz, 1H, H-3^{GlcN-E}), 5.60 (dd, J = 3.0, 1.8 Hz, 1H, H-2^{Rha-C}), 5.56 (t, J = 9.6 Hz, 1H, H-4^{Rha-C}), 5.52 (d, J = 8.4 Hz, 1H, H-1^{GlcN-B}), 5.51 (d, J = 1.8 Hz, 1H, H-1^{Rha-C}), 5.34 (t, J = 10.2 Hz, 1H, H-4^{GlcN-B}), 5.27 (dd, J = 9.6, 3.6 Hz, 1H, H-3^{Rha-F}), 5.23 (dd, J = 3.6, 1.8 Hz, 1H, H-2^{Rha-F}), 5.17 (d, J = 8.4 Hz, 1H, H-1^{GlcN-E}), 5.13–5.08 (m, 2H, H-4^{Rha-A}, H-1^{Rha-F}), 5.07 (t, J = 10.2 Hz, 1H, H-4^{Rha-D}), 5.02 (t, J = 9.6 Hz, 1H, H-4^{GlcN-E}), 5.00 (d, J = 1.8 Hz, 1H, H-1^{Rha-D}), 4.88 (t, J = 9.6 Hz, 1H, H-4^{Rha-F}), 4.85 (dd, J = 10.8, 8.4Hz, 1H, H-2^{GlcN-B}), 4.72 (d, J = 1.2 Hz, 1H, H-1^{Rha-A}), 4.52 (dd, J = 9.6, 3.0 Hz, 1H, H-3^{Rha-C}), 4.37 (dd, J =10.8, 8.4 Hz, 1H, H-2^{GlcN-E}), 4.23 (dd, J = 3.0, 1.2 Hz, 1H, H-2^{Rha-A}), 4.19 (dd, J =12.6, 2.4 Hz, 1H, H-6a^{GlcN-B}), 4.14 (dd, J = 12.6, 4.2 Hz, 1H, H-6a^{GlcN-E}), 4.12–4.04 (m, 4H, H-5^{Rha-C}, H-3^{Rha-A}, H-6b^{GlcN-B}, H-3^{Rha-D}), 4.04–4.02 (m, 1H, H-2^{Rha-D}), 4.02–3.95 (m, 1H, H-5^{Rha-D}), 3.85 (ddd, J = 10.2, 4.2, 2.4 Hz, 1H, H-5^{GlcN-B}), 3.80–3.73 (m, 1H, H-5^{Rha-A}, -OC*H*$_2$CH$_2$-), 3.68 (dd, J = 12.6, 2.4 Hz, 1H, H-6b^{GlcN-E}), 3.65–3.58 (m, 1H, H-5^{Rha-F}), 3.52–3.47 (m, 1H, -OC*H*$_2$CH$_2$-), 3.45–3.39 (m, 3H, H-5^{GlcN-E}, -C*H*$_2$N$_3$), 2.19, 2.04, 2.00, 1.99, 1.98, 1.97 (6 s, 6×3H, 6×-C*H*$_3$CO), 1.94–1.85 (m, 2H, -OCH$_2$C*H*$_2$CH$_2$N$_3$), 1.77, 1.73, 1.72 (3 s, 3×3H, 3×-C*H*$_3$CO), 1.21 (d, J = 6.6

Hz, 3H, H-6$^{Rha\text{-}C}$), 1.12 (d, J = 6.0 Hz, 3H, H-6$^{Rha\text{-}D}$), 1.09 (d, J = 6.0 Hz, 3H, H-6$^{Rha\text{-}A}$), 0.69 (d, J = 6.0 Hz, 3H, H-6$^{Rha\text{-}F}$); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.77, 170.64, 170.20, 170.01 (2C), 169.61, 169.43, 169.35, 169.21, 165.58, 165.22, 164.64, 164.28, 133.56, 133.45, 133.32, 132.54, 132.34, 130.89, 130.19, 129.95, 129.86, 129.53, 129.41, 129.30, 128.95, 128.75, 128.71, 128.09, 127.90, 100.01 (C-1$^{Rha\text{-}D}$), 99.44 (C-1$^{GlcN\text{-}B}$), 99.33 (C-1$^{Rha\text{-}C}$), 99.22 (C-1$^{Rha\text{-}A}$), 99.02 (C-1$^{GlcN\text{-}E}$), 98.31 (C-1$^{Rha\text{-}F}$), 78.67 (C-2$^{Rha\text{-}A}$), 77.71 (C-3$^{Rha\text{-}A}$), 77.22 (C-3$^{Rha\text{-}D}$), 76.47 (C-2$^{Rha\text{-}D}$), 73.91 (C-4$^{Rha\text{-}C}$), 73.06 (C-3$^{Rha\text{-}C}$), 72.49 (2C, C-2$^{Rha\text{-}C}$, C-4$^{Rha\text{-}D}$), 72.09 (C-4$^{Rha\text{-}A}$), 71.72 (C-4$^{Rha\text{-}F}$), 71.26 (2C, C-5$^{GlcN\text{-}B}$, C-5$^{GlcN\text{-}E}$) 70.44 (C-3$^{GlcN\text{-}B}$), 69.99 (C-3$^{GlcN\text{-}E}$), 69.82 (C-2$^{Rha\text{-}F}$), 68.67 (C-3$^{Rha\text{-}F}$), 68.61 (C-4$^{GlcN\text{-}B}$), 68.47 (C-4$^{GlcN\text{-}E}$), 67.89 (C-5$^{Rha\text{-}D}$), 66.88 (C-5$^{Rha\text{-}C}$), 66.50 (C-5$^{Rha\text{-}F}$), 66.41 (C-5$^{Rha\text{-}A}$), 64.50 (-O*C*H$_2$CH$_2$-), 61.88 (C-6$^{GlcN\text{-}B}$), 61.32 (C-6$^{GlcN\text{-}E}$), 54.11 (C-2$^{GlcN\text{-}E}$), 53.91 (C-2$^{GlcN\text{-}B}$), 48.39 (-*C*H$_2$N$_3$), 28.74 (-OCH$_2$*C*H$_2$CH$_2$N$_3$), 21.10, 20.88, 20.79, 20.72, 20.67, 20.62, 20.44, 20.40, 20.24 (9C, 9×-*C*H$_3$CO), 17.65 (C-6$^{Rha\text{-}C}$), 17.52 (C-6$^{Rha\text{-}A}$), 17.14 (C-6$^{Rha\text{-}D}$), 16.94 (C-6$^{Rha\text{-}F}$); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{101}H_{115}O_{42}N_7$ $[M+2NH_4]^{2+}$: 1048.8534; found m/z: 1048.8538.

3-Azidopropyl α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranoside (5b). Ammonia was bubbled into a mixture of **22** (59 mg, 28.8 μmol) in MeOH (10 mL) at 0 °C until saturation. The reaction mixture was kept at rt for 10 days and then evaporated to dryness. The product was dissolved in dry methanol (5 mL) containing acetic anhydride (0.5 mL). The reaction mixture was stirred at rt overnight and then concentrated. The resulting residue was purified by size exclusion chromatography on a Bio-Gel P-2 column with distilled water as the

eluent and then lyophilized to give **5b** (26 mg, 83%) as a white foamy solid. $[\alpha]_D^{25} = -11.0°$ (c 0.1, H_2O); ^1H NMR (600 MHz, D_2O): δ 5.02 (s, 1H, H-1^{Rha}), 5.00 (s, 1H, H-1^{Rha}), 4.94 (s, 1H, H-1^{Rha}), 4.64 (1H, H-1^{Rha} overlapped by H_2O), 4.56 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 4.49 (d, J = 7.8 Hz, 1H, H-1^{GlcN}), 4.11 (s, 1H, H-2^{Rha}), 4.02 (s, 1H, H-2^{Rha}), 3.90 (s, 1H, H-2^{Rha}), 3.86 (s, 1H, H-2^{Rha}), 3.81 (dd, J = 10.2, 3.0 Hz, 1H, H-3^{Rha}), 3.76 (d, J = 11.4 Hz, 1H, H-6a^{GlcN}), 3.74 (d, J = 11.4 Hz, 1H, H-6a^{GlcN}), 3.71–3.46 (m, 12H), 3.45–3.23 (m, 13H), 1.85 (s, 3H, -CH_3CO), 1.84 (s, 3H, -CH_3CO), 1.79–1.68 (m, 2H, -OCH$_2$CH_2CH$_2$N$_3$), 1.13 (d, J = 6.6 Hz, 3H, H-6^{Rha}), 1.12 (d, J = 6.6 Hz, 3H, H-6^{Rha}), 1.10 (d, J = 6.6 Hz, 3H, H-6^{Rha}), 1.08 (d, J = 6.0 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, D_2O): δ 174.56, 174.35, 102.58 (C-1^{GlcN}), 102.41 (C-1^{GlcN}), 101.48 (C-1^{Rha}), 101.12 (C-1^{Rha}), 100.76 (C-1^{Rha}), 98.50 (C-1^{Rha}), 79.96, 79.60, 76.43, 76.41, 76.09, 75.58 (2C), 73.74, 73.65, 71.89, 71.65, 71.11, 70.93, 69.87, 69.77, 69.66 (3C), 69.17 (2C), 69.00, 68.94, 64.91, 60.70, 60.64, 55.74, 55.70, 48.21, 27.70, 22.07, 16.67, 16.56, 16.45, 16.40; ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{43}H_{74}O_{27}N_5$ $[M+H]^+$: 1092.4566; found m/z: 1092.4573.

3-Azidopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-2-chloroacetyl-α-L-rhamnopyranosyl-(1→3)-2,4-di-*O*-benzoyl-α-L-rhamnopyranosyl-(1→2)-[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranoside (23). After a solution of **6** (97.2 mg, 0.127 mmol), 7 (161 mg, 0.137 mmol), and 4 Å molecular sieves in dry CH_2Cl_2 (5 mL) were stirred at rt for 30 min under a N_2 atmosphere, NIS (42 mg, 0.187 mmol) and TMSOTf (4 μL, 18.7 μmol) were sequentially added 0 °C. The reaction mixture was stirred at these conditions for 2 h, and then neutralized with triethylamine, filtered through a Celite pad, and concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 1:1) to yield **23** (188 mg, 80%) as a white foamy

solid. $[\alpha]_D^{25}$ = +29° (c 0.5, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.33 (d, J = 7.8 Hz, 2H, Ar*H*), 8.21 (d, J = 7.8 Hz, 2H, Ar*H*), 7.65–7.17 (m, 19H, Ar*H*), 7.15 (t, J = 7.8 Hz, 2H, Ar*H*), 7.08 (t, J = 7.8 Hz, 2H, Ar*H*), 6.89 (d, J = 7.2 Hz, 1H, Ar*H*), 5.71 (dd, J = 10.2, 9.6 Hz, 1H, H-3$^{GlcN\text{-}B}$), 5.65–5.59 (m, 2H, H-4$^{Rha\text{-}C}$, H-2$^{Rha\text{-}C}$), 5.58–5.52 (m, 2H, H-3$^{GlcN\text{-}E}$, H-1$^{GlcN\text{-}B}$), 5.47 (s, 1H, H-1$^{Rha\text{-}C}$), 5.40 (t, J = 9.6 Hz, 1H, H-4$^{GlcN\text{-}B}$), 5.17 (d, J = 8.4 Hz, 1H, H-1$^{GlcN\text{-}E}$), 5.13 (s, 1H, H-1$^{Rha\text{-}D}$), 5.11–5.04 (m, 3H, H-4$^{Rha\text{-}A}$, H-2$^{Rha\text{-}D}$, H-4$^{Rha\text{-}D}$), 4.97 (t, J = 9.6 Hz, 1H, H-4$^{GlcN\text{-}E}$), 4.90 (dd, J = 10.2, 8.4 Hz, 1H, H-2$^{GlcN\text{-}B}$), 4.74 (s, 1H, H-1$^{Rha\text{-}A}$), 4.50 (dd, J = 10.2, 3.6 Hz, 1H, H-3$^{Rha\text{-}C}$), 4.24 (s, 1H, H-2$^{Rha\text{-}A}$), 4.20 (dd, J = 12.0, 2.4 Hz, 1H, H-6a$^{GlcN\text{-}B}$), 4.18–4.02 (m, 8H, H-5$^{Rha\text{-}C}$, H-2$^{GlcN\text{-}E}$, H-6b$^{GlcN\text{-}B}$, H-6a$^{GlcN\text{-}E}$, -ClC*H*$_2$CO, H-3$^{Rha\text{-}A}$, H-3$^{Rha\text{-}D}$), 4.02–3.94 (m, 1H, H-5$^{Rha\text{-}D}$), 3.88–3.83 (m, 1H, H-5$^{GlcN\text{-}B}$), 3.81–3.73 (m, 2H, H-5$^{Rha\text{-}A}$, -OC*H*$_2$CH$_2$-), 3.55–3.46 (m, 2H, H-6b$^{GlcN\text{-}E}$, -OC*H*$_2$CH$_2$-), 3.46–3.39 (m, 2H, -C*H*$_2$N$_3$), 3.16–3.10 (m, 1H, H-5$^{GlcN\text{-}E}$), 2.06, 1.99, 1.97, 1.77, 1.75, 1.72 (6 s, 6×3H, 6×-C*H*$_3$CO), 1.95–1.86 (m, 2H, -OCH$_2$C*H*$_2$CH$_2$N$_3$), 1.26 (d, J = 6.0 Hz, 3H, H-6$^{Rha\text{-}C}$), 1.11 (d, J = 6.0 Hz, 3H, H-6$^{Rha\text{-}A}$), 1.00 (d, J = 6.0 Hz, 3H, H-6$^{Rha\text{-}D}$); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.76 (2C), 170.20, 169.98, 169.24, 169.21, 166.00, 165.84, 165.23, 164.78, 164.41, 133.67, 133.44, 133.28, 132.59, 130.21, 130.02, 129.84, 129.54, 129.51, 129.30, 129.02, 128.93, 128.66, 128.12, 127.98, 123.10, 99.46 (C-1$^{Rha\text{-}C}$), 99.44 (C-1$^{GlcN\text{-}B}$), 99.17 (C-1$^{Rha\text{-}A}$), 98.27 (C-1$^{GlcN\text{-}E}$), 98.23 (C-1$^{Rha\text{-}D}$), 79.29 (C-2$^{Rha\text{-}A}$), 77.71 (C-3$^{Rha\text{-}A}$), 75.70 (C-3$^{Rha\text{-}D}$), 74.96 (C-3$^{Rha\text{-}C}$), 73.29 (C-4$^{Rha\text{-}C}$), 72.47 (C-2$^{Rha\text{-}C}$), 72.43 (C-2$^{Rha\text{-}D}$), 72.21 (C-4$^{Rha\text{-}D}$), 72.00 (C-4$^{Rha\text{-}A}$), 71.28 (C-5$^{GlcN\text{-}B}$), 71.22 (C-5$^{GlcN\text{-}E}$), 70.52 (C-3$^{GlcN\text{-}B}$), 70.27 (C-3$^{GlcN\text{-}E}$), 68.65 (C-4$^{GlcN\text{-}B}$), 67.93 (C-4$^{GlcN\text{-}E}$), 67.20 (C-5$^{Rha\text{-}D}$), 66.96 (C-5$^{Rha\text{-}C}$), 66.33 (C-5$^{Rha\text{-}A}$), 64.53 (-O*C*H$_2$CH$_2$-), 61.89 (C-6$^{GlcN\text{-}B}$), 60.64 (C-6$^{GlcN\text{-}E}$), 54.27 (C-2$^{GlcN\text{-}B}$), 53.97 (C-2$^{GlcN\text{-}E}$), 48.41 (-*C*H$_2$N$_3$), 40.87 (-Cl*C*H$_2$CO), 28.76 (-OCH$_2$*C*H$_2$CH$_2$N$_3$), 20.73, 20.67, 20.58, 20.44, 20.32, 20.23 (6C, 6×-*C*H$_3$CO),

17.64 (C-$6^{Rha\text{-}C}$), 17.54 (C-$6^{Rha\text{-}A}$), 16.97 (C-$6^{Rha\text{-}D}$); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{91}H_{100}O_{36}N_7Cl$ $[M+2NH_4]^{2+}$: 950.7944; found m/z: 950.7956.

3-Azidopropyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)-4-*O*-benzoyl-α-L-rhamnopyranosyl-(1→3)-2,4-di-*O*-benzoyl-α-L-rhamnopyranosyl-(1→2)-[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranoside (24). To a solution of **23** (136 mg, 73.5 μmol) in 15 mL of MeOH and CH_2Cl_2 (v/v 4:1) was added thiourea (28 mg, 0.363 mmol) and 2,6-lutidine (9 μL, 73.5μmol) at rt. The reaction was stirred at 70 °C for 26 h, and then cooled down to rt. The mixture was diluted with CH_2Cl_2 (50 mL), and washed with 1 M HCl and brine. The organic phase was dried over Na_2SO_4, filtered and concentrated. The residue was purified by column chromatography (petroleum ether-ethyl acetate 2:3) to give **24** (112 mg, 86 %) as a white foamy solid. $[\alpha]_D^{25}$ = +40° (c 0.2, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.33 (d, J = 7.2 Hz, 2H, Ar*H*), 8.16 (d, J = 7.2 Hz, 2H, Ar*H*), 7.66–7.27 (m, 16H, Ar*H*), 7.26–7.12 (m, 5H, Ar*H*), 7.06 (t, J = 7.8 Hz, 2H, Ar*H*), 6.87 (d, J = 6.6 Hz, 1H, Ar*H*), 5.73 (dd, J = 10.8, 9.6 Hz, 1H, H-$3^{GlcN\text{-}B}$), 5.64–5.56 (m, 3H, H-$4^{Rha\text{-}C}$, H-$2^{Rha\text{-}C}$, H-$3^{GlcN\text{-}E}$), 5.51 (d, J = 8.4 Hz, 1H, H-$1^{GlcN\text{-}B}$), 5.47 (d, J = 1.2 Hz, 1H, H-$1^{Rha\text{-}C}$), 5.41 (dd, J = 10.2, 9.6 Hz, 1H, H-$4^{GlcN\text{-}B}$), 5.27 (d, J = 8.4 Hz, 1H, H-$1^{GlcN\text{-}E}$), 5.18–5.13 (m, 2H, H-$1^{Rha\text{-}D}$, H-$4^{Rha\text{-}D}$), 5.08 (t, J = 9.6 Hz, 1H, H-$4^{Rha\text{-}A}$), 4.98 (dd, J = 10.2, 9.0 Hz, 1H, H-$4^{GlcN\text{-}E}$), 4.90 (dd, J = 10.8, 8.4 Hz, 1H, H-$2^{GlcN\text{-}B}$), 4.74 (d, J = 1.8 Hz, 1H, H-$1^{Rha\text{-}A}$), 4.55 (dd, J = 10.2, 3.6 Hz, 1H, H-$3^{Rha\text{-}C}$), 4.24–4.18 (m, 3H, H-$2^{GlcN\text{-}E}$, H-$2^{Rha\text{-}A}$, H-6a$^{GlcN\text{-}B}$), 4.17–4.07 (m, 4H, H-$5^{Rha\text{-}C}$, H-$3^{Rha\text{-}A}$, H-6b$^{GlcN\text{-}B}$, H-6a$^{GlcN\text{-}E}$), 4.04–3.98 (m, 1H, H-$5^{Rha\text{-}D}$), 3.91 (br s, 1H, H-$2^{Rha\text{-}D}$), 3.89 (dd, J = 9.6, 3.0 Hz, 1H, H-$3^{Rha\text{-}D}$), 3.86 (ddd, J = 10.2, 4.2, 2.4 Hz, 1H, H-$5^{GlcN\text{-}B}$), 3.80–3.73 (m, 2H, H-$5^{Rha\text{-}A}$, -OC$H_2$$CH_2$-), 3.69

(dd, J = 12.6, 2.4 Hz, 1H, H-6b^{GlcN-E}), 3.53–3.47 (m, 1H, -OCH_2CH$_2$-), 3.45–3.38 (m, 3H, H-5$^{GlcN-E,}$ -CH_2N$_3$), 2.00, 1.99, 1.96, 1.77, 1.75, 1.72(6 s, 6×3H, 6×-CH_3CO), 1.95-1.86 (m, 2H, -OCH$_2$CH_2CH$_2$N$_3$), 1.24 (d, J = 6.0 Hz, 3H, H-6^{Rha-C}), 1.09 (d, J = 6.0 Hz, 3H, H-6^{Rha-A}), 1.05 (d, J = 6.6 Hz, 3H, H-6^{Rha-D}); ^{13}C NMR (150 MHz, CDCl$_3$): δ 170.80, 170.61, 170.21, 169.95, 169.28, 169.26, 165.72, 165.29, 164.88, 164.33, 133.68, 133.53, 133.25, 132.56, 132.37, 130.23, 129.99, 129.85, 129.61, 129.47, 129.32, 128.97, 128.76, 128.65, 128.10, 127.92, 123.13, 100.45 (C-1^{Rha-D}), 99.44 (C-1^{Rha-C}), 99.40 (C-1^{GlcN-B}), 99.18 (C-1^{Rha-A}), 98.36 (C-1^{GlcN-E}), 79.52 (C-3^{Rha-D}), 79.12 (C-2^{Rha-A}), 77.47 (C-3^{Rha-A}), 73.85 (C-4^{Rha-C}), 73.65 (C-3^{Rha-C}), 72.69 (C-2^{Rha-C}), 72.22 (C-4^{Rha-A}), 71.96 (C-4^{Rha-D}), 71.46 (C-5^{GlcN-E}), 71.30 (C-5^{GlcN-B}), 70.40 (C-3^{GlcN-B}), 70.28 (C-3^{GlcN-E}), 70.11 (C-2^{Rha-D}), 68.69 (C-4^{GlcN-B}), 68.39 (C-4^{GlcN-E}), 67.15 (C-5^{Rha-D}), 66.99 (C-5^{Rha-C}), 66.37 (C-5^{Rha-A}), 64.52 (-O*C*H$_2$CH$_2$-), 61.85 (C-6^{GlcN-B}), 61.25 (C-6^{GlcN-E}), 54.20 (C-2^{GlcN-E}), 53.90 (C-2^{GlcN-B}), 48.39 (-*C*H$_2$N$_3$), 28.75 (-OCH$_2$*C*H$_2$CH$_2$N$_3$), 20.69, 20.60 (2C), 20.47, 20.31, 20.25 (6C, 6×-*C*H$_3$CO), 17.62 (C-6^{Rha-C}), 17.55 (C-6^{Rha-A}), 17.07 (C-6^{Rha-D}); ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{89}H_{99}O_{35}N_7$ $[M+2NH_4]^{2+}$: 912.8086; found m/z: 912.8096.

3-Azidopropyl 2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→2)-[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranosyl-(1→3)-2,4-di-*O*-benzoyl-α-L-rhamnopyranosyl-(1→2)-[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranosyl-(1→3)-2,4-di-*O*-benzoyl-α-L-rhamnopyranosyl-(1→2)-[3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl-(1→3)]-4-*O*-benzoyl-α-L-rhamnopyranoside (25). A solution of **21** (60 mg, 43.4 μmol), 24 (68 mg, 37.7 μmol), and 4 Å molecular sieves in dry CH_2Cl_2 (4 mL) were stirred at rt for 40 min under a N_2 atmosphere. Then, the solution was cooled down to 0 ºC,

and NIS (12 mg, 56.6 μmol) and TMSOTf (1.0 μL, 6 μmol) were added sequentially. The reaction mixture was stirred at 0 °C for 2 h, and then neutralized with triethylamine, filtered, and concentrated. The residue was purified by flash column chromatography (petroleum ether-ethyl acetate 1:2) to yield **25** (78 mg, 67%) as a white foamy solid. $[\alpha]_D^{25}$ = +17° (c 0.5, CHCl3); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.35 (d, *J* = 7.8 Hz, 2H, Ar*H*), 8.30 (d, *J* = 7.8 Hz, 2H, Ar*H*), 8.13 (d, *J* = 7.8 Hz, 2H, Ar*H*), 8.04 (d, *J* = 7.8 Hz, 2H, Ar*H*), 7.73–7.08 (m, 35H, Ar*H*), 7.04 (t, *J* = 7.8 Hz, 2H, Ar*H*), 6.97–6.90 (m, 2H, Ar*H*), 5.68 (dd, *J* = 10.8, 9.0 Hz, 1H, H-3^{GlcN}), 5.63 (dd, *J* = 10.8, 9.6 Hz, 2H, 2×H-3^{GlcN}), 5.60 (dd, *J* = 3.0, 1.8 Hz, 1H, H-2^{Rha}), 5.57–5.51 (m, 3H, H-1^{GlcN}, H-1^{Rha}, H-4^{Rha}), 5.50 (dd, *J* = 3.0, 1.8 Hz, 1H, H-2^{Rha}), 5.41 (t, *J* = 9.6 Hz, 1H, H-4^{Rha}), 5.31 (t, *J* = 9.6 Hz, 1H, H-4^{GlcN}), 5.29–5.23 (m, 3H, H-1^{GlcN}, H-1^{Rha}, H-3^{Rha}), 5.22 (dd, *J* = 3.6, 1.8 Hz, 1H, H-2^{Rha}), 5.16 (t, *J* = 9.6 Hz, 1H, H-4^{GlcN}), 5.15 (d, *J* = 8.4 Hz, 1H, H-1^{GlcN}), 5.12 (t, *J* = 9.6 Hz, 1H, H-4^{Rha}), 5.08 (t, *J* = 9.6 Hz, 1H, H-3^{GlcN}), 5.07–5.02 (m, 4H, H-1^{Rha}, H-4^{Rha}), 5.00 (t, *J* = 9.6 Hz, 1H, H-4^{GlcN}), 4.96 (d, *J* = 1.2 Hz, 1H, H-1^{Rha}), 4.87 (t, *J* = 9.6 Hz, 1H, H-4^{Rha}), 4.83 (dd, *J* = 10.8, 8.4 Hz, 1H, H-2^{GlcN}), 4.72 (s, 1H, H-1^{Rha}), 4.71 (dd, *J* = 10.2, 8.4 Hz, 1H, H-2^{GlcN}), 4.47–4.41 (m, 2H, 2×H-3^{Rha}), 4.36 (dd, *J* = 10.8, 8.4 Hz, 1H, H-2^{GlcN}), 4.24 (dd, *J* = 3.6, 1.2 Hz, 1H, H-2^{Rha}), 4.21–3.89 (m, 13H, 2×H-2^{Rha}, 3×H-3^{Rha}, 4×H-5^{Rha}, 4×H-6^{GlcN}), 3.88–3.82 (m, 1H, H-5^{GlcN}), 3.80–3.71 (m, 2H, H-5^{Rha}, -OC*H*$_2$CH$_2$-), 3.69–3.59 (m, 3H, 2×H-6^{GlcN}, H-5^{Rha}), 3.53–3.46 (m, 1H, -OC*H*$_2$CH$_2$-), 3.46–3.38 (m, 3H, H-5^{GlcN}, -C*H*$_2$N$_3$), 3.38–3.32 (m, 1H, H-5^{GlcN}), 2.18, 2.03, 2.00, 1.99, 1.98, 1.97, 1.78, 1.73, 1.71, 1.67 (10 s, 12×3H, 12×-C*H*$_3$CO), 1.21 (d, *J* = 6.6 Hz, 3H, H-6^{Rha}), 1.13 (d, *J* = 6.0 Hz, 3H, H-6^{Rha}), 1.09 (d, *J* = 6.6 Hz, 3H, H-6^{Rha}), 1.08 (d, *J* = 6.6 Hz, 3H, H-6^{Rha}), 0.80 (d, *J* = 6.0 Hz, 3H, H-6^{Rha}), 0.70 (d, *J* = 6.6 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.76, 170.68, 170.63, 170.28, 170.20, 170.17, 170.02, 169.59, 169.41, 169.35, 169.18, 169.13, 165.68, 165.63, 165.23, 164.96, 164.65, 164.36, 164.33, 133.55, 133.44, 133.33, 133.30, 133.15, 132.58,

132.29, 132.15, 130.89, 130.21, 130.13, 130.10, 129.99, 129.91, 129.73, 129.69, 129.46, 129.41, 129.28, 129.19, 128.89, 128.81, 128.76, 128.68, 128.64, 128.11, 127.91, 127.88, 100.24 (C-1^{Rha}), 99.71 (C-1^{Rha}), 99.52 (C-1^{GlcN}), 99.46 (C-1^{GlcN}), 99.34 (C-1^{Rha}), 99.24 (C-1^{Rha}), 99.05 (C-1^{GlcN}), 98.82 (C-1^{Rha}), 98.34 (C-1^{Rha}), 78.62, 78.49, 77.82, 77.69, 77.17, 76.80, 74.03, 73.84, 73.81, 72.94, 72.51, 72.41, 72.38, 72.05 (2C), 71.77, 71.24, 71.19, 71.14, 70.49, 70.43, 70.04, 69.89, 68.72, 68.59, 68.52, 68.14, 67.77, 67.66, 66.82, 66.66, 66.44 (2C), 64.56, 61.82, 61.37, 61.03, 54.10, 53.96, 53.86, 48.38, 28.73, 21.08, 20.91, 20.82, 20.71, 20.65 (2C), 20.62, 20.44, 20.41 (2C), 20.24 (2C), 17.64, 17.51, 17.29, 17.26, 17.16, 16.97; ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{154}H_{166}O_{62}N_8$ $[M+2NH_4]^{2+}$: 1559.5036; found m/z: 1559.5048.

3-Azidopropyl α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranoside (5c). Ammonia was bubbled into a mixture of **23** (55 mg, 17.9 μmol) in MeOH (10 mL) at 0 °C until saturation. The reaction mixture was kept at rt for 14 days and then evaporated to dryness. The product was dissolved in dry methanol (4 mL) containing acetic anhydride (0.4 mL). The reaction mixture was stirred at rt overnight and then concentrated. The resulting residue was purified by size exclusion chromatography on a Bio-Gel P-2 column with distilled water as the eluent and then lyophilized to give **5c** (23.3 mg, 82%) as a white foamy solid. $[\alpha]_D^{25}$ = –22° (c 0.1, H_2O); ^1H NMR (600 MHz, D_2O): δ 5.01 (s, 2H, 2×H-1^{Rha}), 4.99 (s, 1H, H-1^{Rha}), 4.93 (s, 1H, H-1^{Rha}), 4.92 (s, 1H, H-1^{Rha}), 4.63 (1H, H-1^{Rha} overlapped by H_2O), 4.55 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 4.54 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 4.49 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 4.11 (s, 2H,

2×H-2^{Rha}), 4.02 (s, 1H, H-2^{Rha}), 3.89 (s, 2H, 2×H-2^{Rha}), 3.86 (dd, *J* = 3.0, 1.8 Hz, 1H, H-2^{Rha}), 3.81 (dd, *J* = 9.6, 3.0 Hz, 2H, H-3^{Rha}), 3.75 (d, *J* = 12.0 Hz, 2H, 3×H-6a^{GlcN}), 3.73 (d, *J* = 10.8 Hz, 1H, 3×H-6a^{GlcN}), 3.70–3.21 (m, 35H), 1.85, 1.84, 1.84 (3 s, 3×3H, 3×-C*H*$_3$CO), 1.78–1.67 (m, 2H, -OCH$_2$C*H*$_2$CH$_2$N$_3$), 1.15-1.05 (m, 18H, 6×H-6^{Rha}); ^{13}C NMR (150 MHz, D$_2$O): *δ* 174.56, 174.34, 174.33, 102.57 (C-1^{GlcN}), 102.46 (C-1^{GlcN}), 102.40 (C-1^{GlcN}), 101.48 (C-1^{Rha}), 101.16 (C-1^{Rha}), 101.10 (C-1^{Rha}), 100.79 (2C, 2×C-1^{Rha}), 98.50 (C-1^{Rha}), 79.94, 79.59 (2C), 76.66, 76.49, 76.40, 76.12, 75.96, 75.59, 75.57 (3C), 73.76, 73.74, 73.65, 71.89, 71.64, 71.60, 71.16, 71.11, 70.92, 69.87, 69.78, 69.67 (3C), 69.65, 69.23, 69.21, 69.19, 69.18, 68.98, 68.96, 64.90, 60.69, 60.65, 60.64, 55.74, 55.72, 55.69, 48.20, 27.69, 22.09, 22.08, 22.06, 16.75, 16.64, 16.55, 16.46, 16.39, 16.37; ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{63}H_{106}O_{40}N_6Na_2$ $[M+2Na]^{2+}$: 816.3115; found m/z: 816.3113.

3-Aminopropyl α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranoside (26a). To a stirred mixture of **5a** (61 mg, 0.103 mmol) in H$_2$O (3 mL) was added Pd/C (10%, 5 mg), then the atmosphere was placed with H$_2$ and the reaction was stirred at rt for 2 h. After detection with MALDI-TOF mass spectra indicated the completion of reaction, the mixture was filtered and the filtrate was lyophilized to give the product **26a** (57 mg, 98%) as white solid flocs without further purification. $[\alpha]_D^{25}$ = –39° (c 0.1, H$_2$O); ^{1}H NMR (600 MHz, D$_2$O): *δ* 4.96 (d, *J* = 1.8 Hz, 1H, H-1^{Rha}), 4.63 (1H, H-1^{Rha} overlapped in H$_2$O), 4.53 (d, *J* = 8.4 Hz, 1H, H-1^{GlcN}), 3.98 (dd, *J* = 3.0, 1.8 Hz, 1H, H-2^{Rha}), 3.85 (dd, *J* = 3.6, 1.8 Hz, 1H, H-2^{Rha}), 3.74 (d, *J* = 12.0 Hz, 1H, H-6a^{GlcN}), 3.68 (dd, *J* = 9.6, 3.0 Hz, 1H, H-3^{Rha}), 3.66–3.62 (m, 1H, -OC*H*$_2$CH$_2$-), 3.61–3.43 (m, 5H, H-6b^{GlcN}, H-2^{GlcN}, H-3^{Rha}, H-5^{Rha}, H-5^{Rha}), 3.41–3.29 (m, 3H, -OC*H*$_2$CH$_2$-, H-4^{GlcN}, H-4^{Rha}), 3.29–3.22 (m, 3H, H-3^{GlcN}, H-5^{GlcN}, H-4^{Rha}), 2.96–2.86 (m, 2H, -C*H*$_2$NH$_2$), 1.84 (s, 3H, -C*H*$_3$CO), 1.83–1.76

(m, 2H, -$OCH_2CH_2CH_2NH_2$), 1.10 (d, J = 6.0 Hz, 3H, H-6^{Rha}), 1.08 (d, *J* = 6.6 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, D_2O): δ 174.54, 102.29(C-1^{GlcN}), 101.51(C-1^{Rha}), 98.47(C-1^{Rha}), 79.36, 76.69, 75.63, 73.54, 71.84, 70.95, 69.80, 69.77, 69.68, 68.96, 68.84, 64.82, 60.72, 55.70, 37.20, 26.73, 22.05, 16.50, 16.46; ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{23}H_{43}O_{14}N_2$ $[M+H]^+$: 571.2709; found m/z: 571.2707.

3-Aminopropyl α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranoside (26b). To the mixture of **5b** (20 mg, 18.1 μmol) in H_2O (3 ml) was added Pd/C (10%, 4 mg), then the atmosphere was placed with H2 and the reaction was stirred at rt for 4 h. After detection with MALDI-TOF mass spectra indicated the completion of reaction, the mixture was filtered and the filtrate was lyophilized to give the product **26b** (18 mg, 95%) as white solid flocs without further purification. $[\alpha]_D^{25}$ = –27° (c 0.1, H_2O); ^{1}H NMR (600 MHz, D_2O): δ 5.03 (d, *J* = 1.8 Hz, 1H, H-1^{Rha}), 5.02 (d, *J* = 1.8 Hz, 1H, H-1^{Rha}), 4.96 (d, *J* = 1.2 Hz, 1H, H-1^{Rha}), 4.63 (1H, H-1^{Rha} overlapped in H_2O), 4.58 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 4.54 (d, *J* = 8.4 Hz, 1H, H-1^{GlcN}), 4.13 (dd, *J* = 3.0, 1.8 Hz, 1H, H-2^{Rha}), 4.03 (dd, J = 3.0, 1.8 Hz, 1H, H-2^{Rha}), 3.91 (dd, *J* = 3.0, 1.8 Hz, 1H, H-2^{Rha}), 3.88 (dd, *J* = 3.6, 1.8 Hz, 1H, H-2^{Rha}), 3.83 (dd, *J* = 9.6, 3.0 Hz, 1H, H-3^{Rha}), 3.80–3.74 (m, 2H), 3.72–3.46 (m, 12H), 3.45–3.25 (m, 11H), 2.93–2.85 (m, 2H, -CH_2NH_2), 1.87 (s, 3H, -CH_3CO), 1.87 (s, 3H, -CH_3CO), 1.83-1.73 (m, 2H, -$OCH_2CH_2CH_2NH_2$), 1.15 (d, *J* = 6.0 Hz, 3H, H-6^{Rha}), 1.14 (d, *J* = 6.0 Hz, 3H, H-6^{Rha}), 1.12 (d, *J* = 6.0 Hz, 3H, H-6^{Rha}), 1.10 (d, *J* = 6.6 Hz, 3H, H-6^{Rha}); ^{13}C NMR (150 MHz, D_2O): *δ* 174.55, 174.30, 102.41 (2C, 2×C-1^{GlcN}), 101.48 (C-1^{Rha}), 101.15 (C-1^{Rha}), 100.76 (C-1^{Rha}), 98.51 (C-1^{Rha}), 79.55, 76.42, 76.35, 76.08, 75.65, 75.58,

73.68, 73.63, 71.86, 71.62, 71.18, 70.92, 69.86, 69.75, 69.71, 69.66 (2C), 69.16, 68.99, 68.91, 64.79, 60.71 (2C), 55.75, 55.68, 37.20, 26.84, 22.08, 22.06, 16.68, 16.56, 16.44, 16.41; ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{43}H_{76}O_{27}N_3$ $[M+H]^+$: 1066.4661; found m/z: 1066.4660.

3-Aminopropyl α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-[2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→3)]-α-L-rhamnopyranoside (26c). To the mixture of **5c** (19 mg, 11.9 μmol) in H_2O was added Pd/C (10%, 3 mg), then the atmosphere was placed with H_2 and the reaction was stirred in rt for 6 h. After detection with MALDI-TOF mass spectra indicated the completion of reaction, the mixture was filtered and the filtrate was lyophilized to give the product **26c** (17 mg, 96%) as white solid flocs without further purification. $[\alpha]_D^{25}$ = –45° (c 0.1, H_2O); ^{1}H NMR (600 MHz, D_2O): δ 5.02 (s, 2H, 2×H-1^{Rha}), 5.00 (s, 1H, H-1^{Rha}), 4.94 (s, 1H, H-1^{Rha}), 4.92 (s, 1H, H-1^{Rha}), 4.63 (s, 1H, H-1^{Rha}), 4.56 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 4.55 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 4.52 (d, J = 8.4 Hz, 1H, H-1^{GlcN}), 4.12 (s, 2H, 2×H-2^{Rha}), 4.01 (s, 1H, H-2^{Rha}), 3.90 (s, 2H, 2×H-2^{Rha}), 3.86 (s, 1H, H-2^{Rha}), 3.81 (br d, J = 10.2 Hz, 2H, 2×H-3^{Rha}), 3.78–3.22 (m, 36H), 3.05–2.82 (m, 2H, -CH_2NH$_2$), 1.853, 1.849, 1.844 (3 s, 3×3H, 3×-CH_3CO), 1.84–1.75 (m, 2H, -OCH$_2$CH_2CH$_2$NH$_2$), 1.18–1.04 (m, 18H, 6×H-6^{Rha}); ^{13}C NMR (150 MHz, D_2O): δ 174.57, 174.34 (2C), 102.46 (C-1^{GlcN}), 102.41 (C-1^{GlcN}), 102.37 (C-1^{GlcN}), 101.48 (C-1^{Rha}), 101.44 (C-1^{Rha}), 101.15 (C-1^{Rha}), 100.79 (2C, 2×C-1^{Rha}), 98.52 (C-1^{Rha}), 79.56 (2C), 76.65, 76.41, 76.14, 76.09, 75.97, 75.65, 75.57 (2C), 73.75, 73.68, 73.64, 71.88, 71.61, 71.59, 71.19, 71.15, 71.11, 70.92, 69.77, 69.67 (5C), 69.19 (4C), 68.98, 68.92, 64.77, 60.69 (3C), 55.75,

55.72, 55.68, 37.19, 26.69, 22.09 (3C), 16.76, 16.63, 16.57, 16.46, 16.40, 16.37; ESI-LTQ-Orbitrap HRMS (positive ion): Calcd for $C_{63}H_{109}O_{40}N_4$ $[M+H]^+$: 1561.6613; found m/z: 1561.6608.

Preparation of Activated Esters 28a-c. A mixture of oligosaccharide haptens **26a**, **26b** or **26c** (12 mg) and glutarate **27** (15 equiv.) in DMF and PBS buffer (0.1 M, pH 8.0) (v/v 4:1, 2 mL) was gently stirred at rt for 4 h, and the solvents were removed under reduced pressure. The residue was then washed with nine volumes of ethyl acetate (10 times) to remove excess DSG **27**, and dried under high vacuum to furnish the monoester products **28a-c** that were used directly for protein conjugation.

Preparation of Glycoproteins 2a-c, 3a-c, 4a-c and 29a-c. Each activated oligosaccharide and ScpA193, CRM197, TT or BSA in a mass ratio of 1:2 (oligosaccharide/carrier protein) were dissolved in PBS buffer (0.2 M, 0.5 mL, pH 8.0), and the solution was stirred at rt for 5 days. The reaction was monitored with MALDI-TOF MS, and after the observed increase in molecular mass had stopped, the reaction mixture was applied to a Biogel A0.5 column to remove any unreacted oligosaccharides through gel-filtration chromatography (0.1 M PBS buffer, pH 8.0). Fractions that contained glycoproteins, confirmed by MALDI-TOF MS analysis, were combined and dialyzed against distilled water (3×5 mL). The residual solution was lyophilized to give glycoconjugates **2a-c**, **3a-c**, **4a-c** and **29a-c** as white powders. The carbohydrate loadings were analyzed by a previously reported protocol (Liao, G. et al, *Bioconjugate chem.*, **2015**, *26*, 466–476).

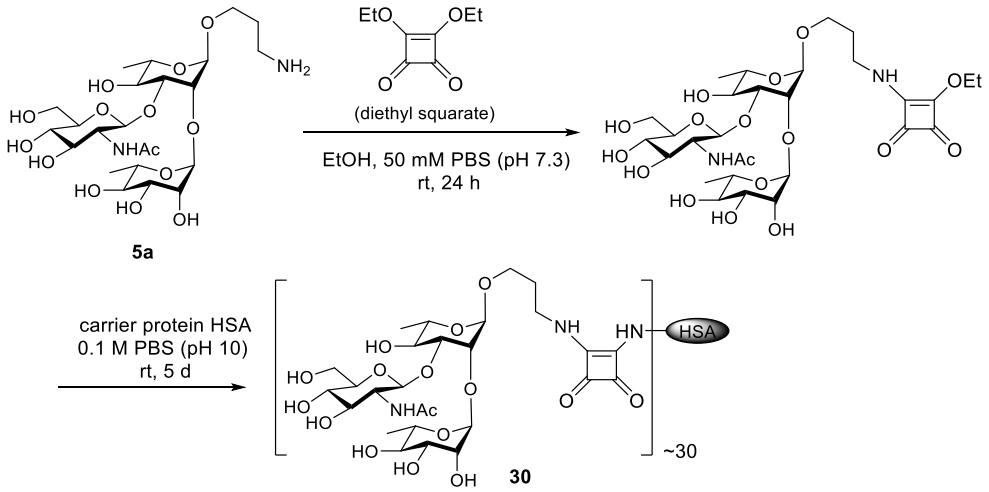
Preparation of the trisaccharide-HSA Glycoprotein 30. A mixture of **5a** (2 mg) and diethyl squarate (2.0 equiv) in ethanol and PBS buffer (50 mM, pH 7.3) (v/v 1:1, 1.5 mL) was stirred at rt for 24 h. After the solvents were removed under reduced pressure, the residue was purified by Bio-Gel P2 with distilled water as the eluent and lyophilized to give the monoethyl squarate activated esters. The activated oligosaccharide and HSA in a mass ratio of 1:2 were dissolved in PBS buffer (0.1 M, 0.5 mL, pH 10), and the solution was stirred at rt for 5 days. The excessive oligosaccharide was removed by using Dialysis cup, and the PBS buffer was replaced with ultra-pure water at the same time. The residual solution was lyophilized to give the product **30** (3.6 mg) as white solid flocs. The carbohydrate loading (22.7%) was analyzed by its MALDI-TOF mass spectra.

Immunization of Mice. Female C57BL/6 mice (6-8 weeks old) were purchased from Shandong University Laboratory Animal Center. A solution of glycoconjugates **2-4a-c** (0.45~3.0 mg, containing 90 μg oligosaccharide), oligosaccharide **5a-c** (90 μg) dissolved in 1.5 mL of 2×PBS buffer was thoroughly mixed with 1.5 mL of CFA or IFA according to the manufacturer's instructions to generate an emulsion. Mice (6 each/group) were inoculated first via s.c. injection of 0.1 mL of the CFA emulsion of a specific conjugate (3 μg carbohydrate antigen/mouse/injection) on day 1. Following the initial immunization, the mice were boosted 4 times on day 14, day 21, day 28, and day 42 via s.c. injection of the IFC emulsion (0.1 mL) of the same vaccine. Mouse blood samples were collected on day 0 prior to the initial immunization and on day 56 after boost immunizations and were clotted to obtain the antisera by the standard protocols that were stored at -80 ^{o}C before use. The animal protocols involved in this research were performed in strict accordance with the National Institute for Health Guide for the Care and

Use of Laboratory Animals (National Research Council, 8th Ed. National Academies Press (US); Washington DC: 2011), and approved by the Institutional Animal Care and Use Committee (IACUC) of Shandong University.

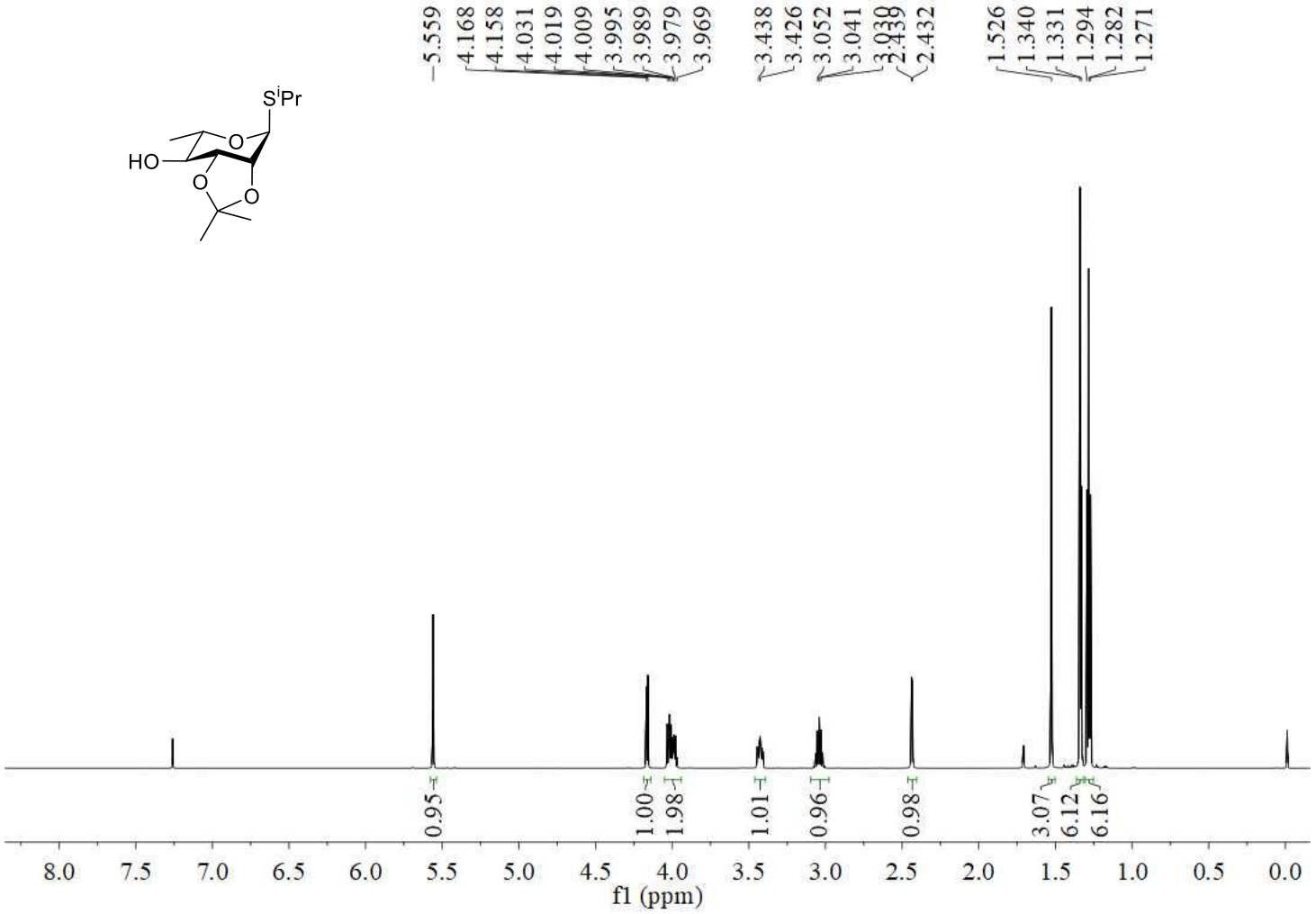
ELISA. ELISA plates were treated with a solution of BSA conjugate **29a-c**, HSA conjugate **30**, BSA, HSA, ScpA193 or ScpA (2 μg/mL, 100 μL/well) dissolved in the coating buffer (0.1 M aqueous bicarbonate, pH 9.6) at 4 °C overnight and then at 37 °C for 1 h. It was followed by washing with PBS buffer containing 0.05% Tween-20 (PBST) three times. The plates were incubated with the blocking buffer (1% BSA in PBST, 100 μL/well) at rt for 1 h and washed with PBST three times. Each mouse serum with serial dilutions from 1:300 to 1:937500 in PBS (100 μL/well) was added to the coated plates, and the plates were incubated at 37 °C for 2 h. After being washed with PBST, the plates were incubated with a 1:1000 diluted solution of an alkaline phosphatase-linked goat anti-mouse kappa antibody (Abcam), respectively, at rt for 1 h. The plates were again washed with PBST three times and developed with a *p*-nitrophenylphosphate (PNPP) solution (1.67 mg/mL in buffer, 100 μL) at rt for 30 min. The reaction was quenched by adding 25 μL of the quenching solution (3 M NaOH) to each well. Finally, the plates were examined with a microplate reader at 405 nm wavelength. The OD values after deducting the background OD values obtained with the day 0 sera were plotted against dilution numbers, and the best-fit equation was obtained for each set of data and used to calculate the antibody titer, defined as the dilution number giving an adjusted OD value of 0.20.

II. Conjugation of trisaccharide 5a with HAS protein via bifunctional squarate linker

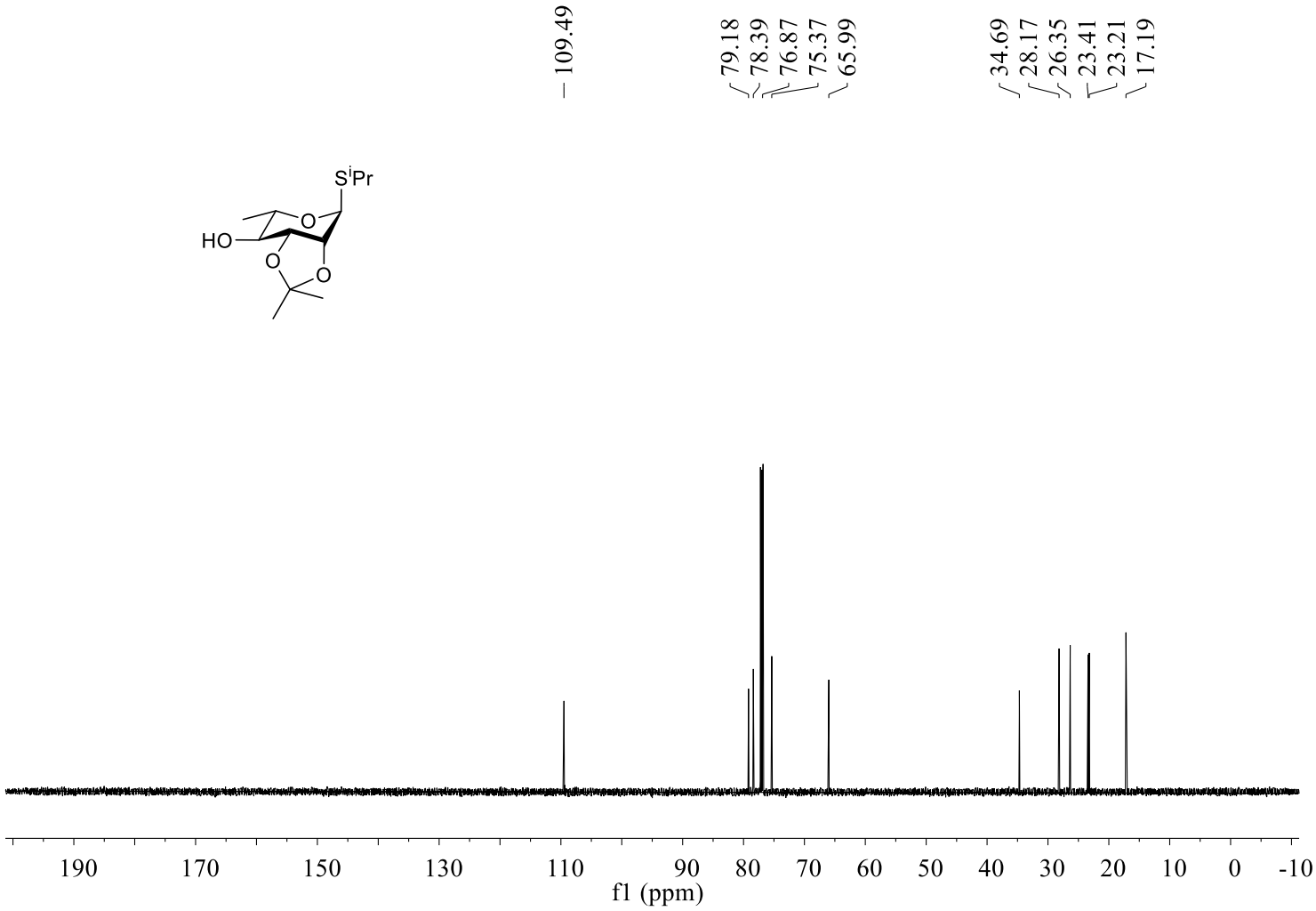


Scheme S1.

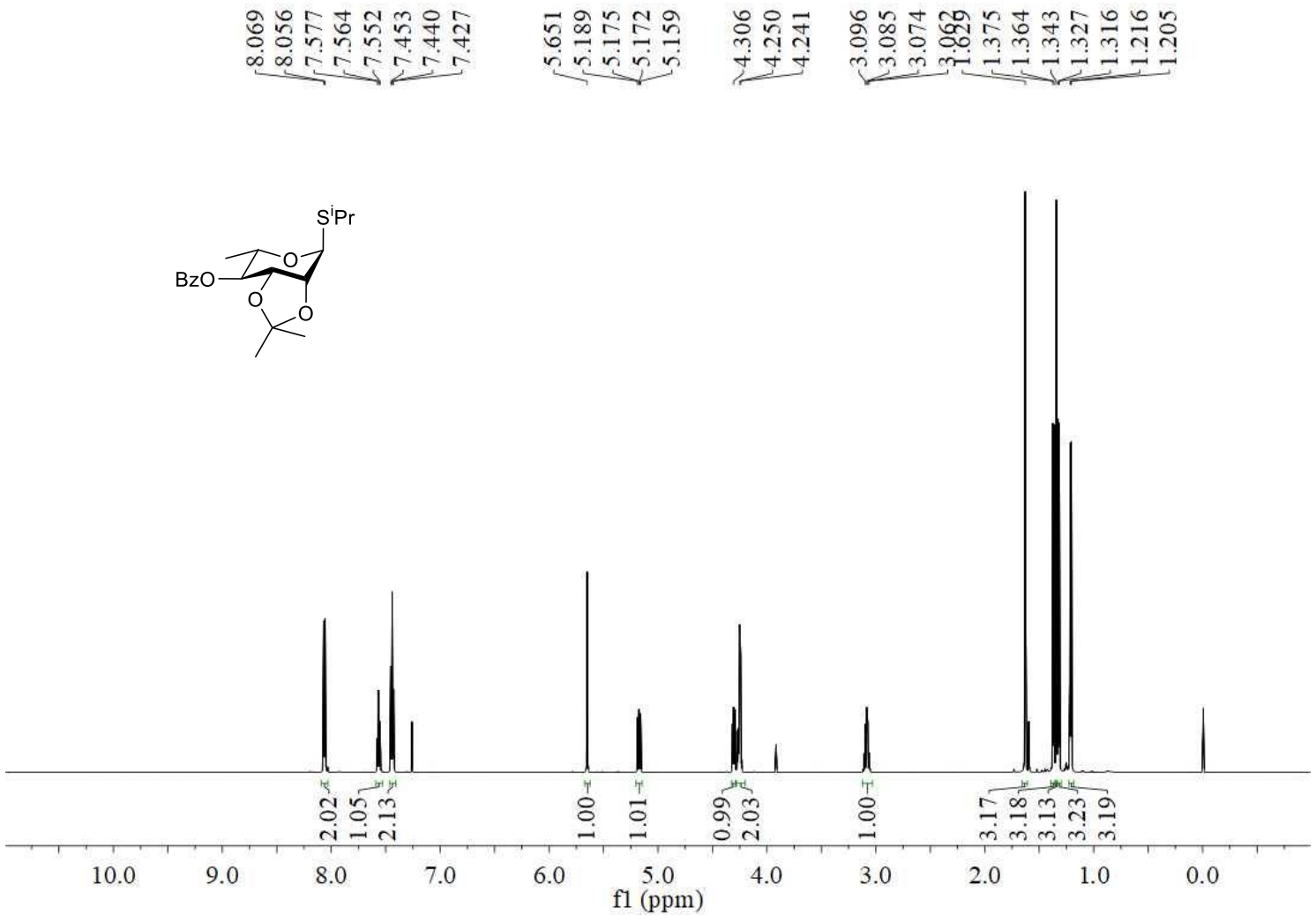
III. NMR and MS data of synthetic compounds



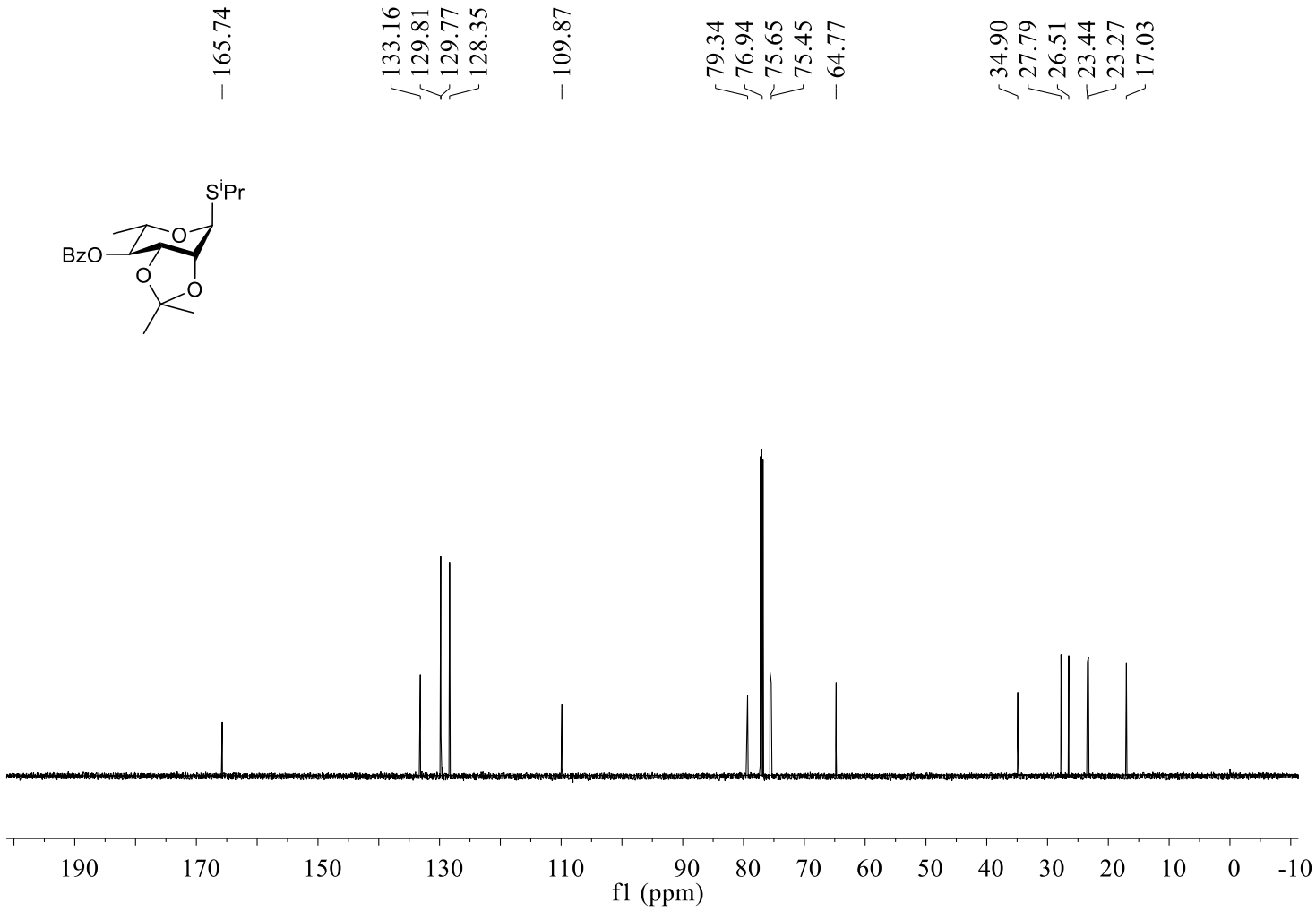
^{1}H NMR spectrum of compound **13** (600 MHz, $CDCl_3$)



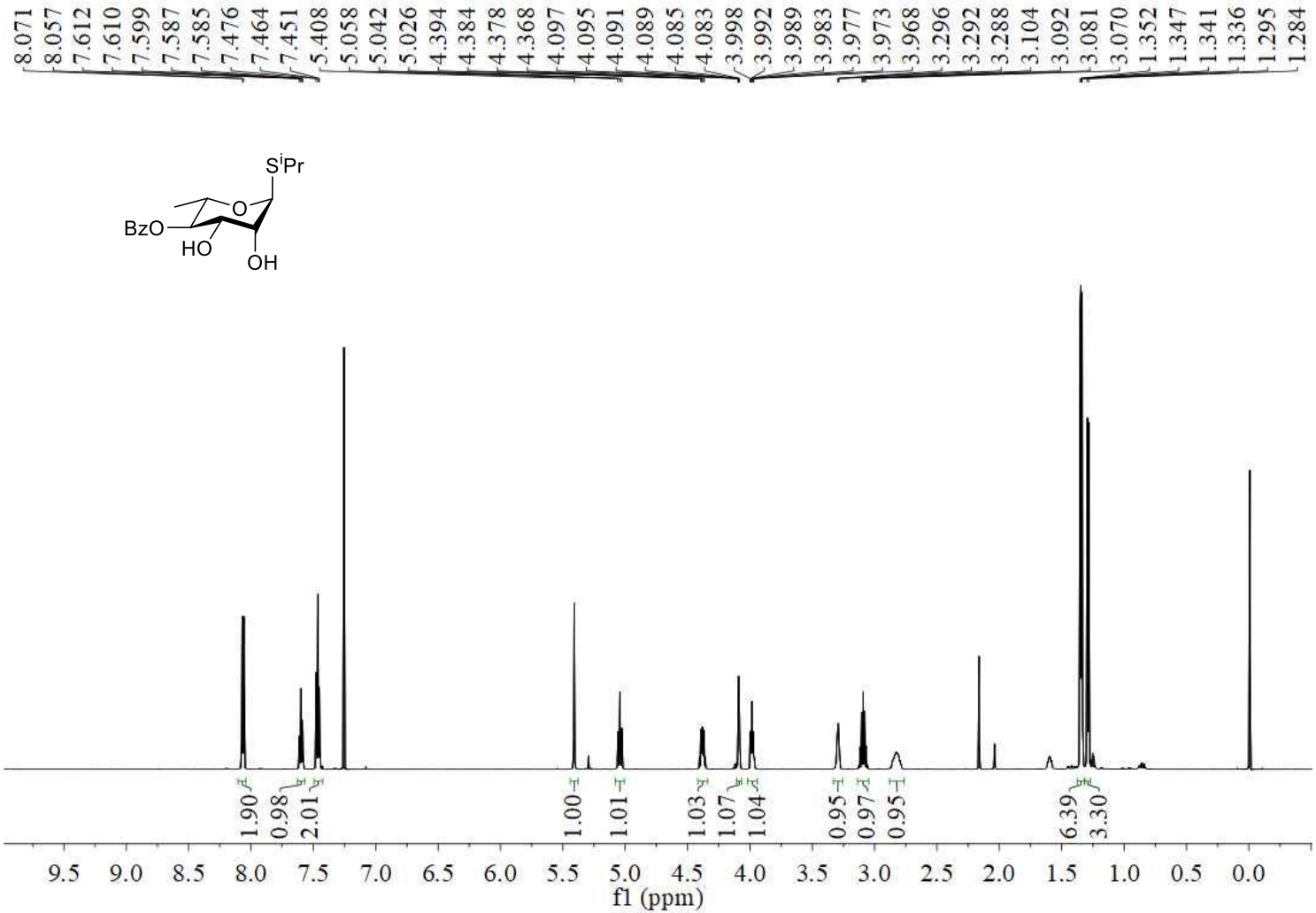
^{13}C NMR spectrum of compound **13** (150 MHz, $CDCl_3$)



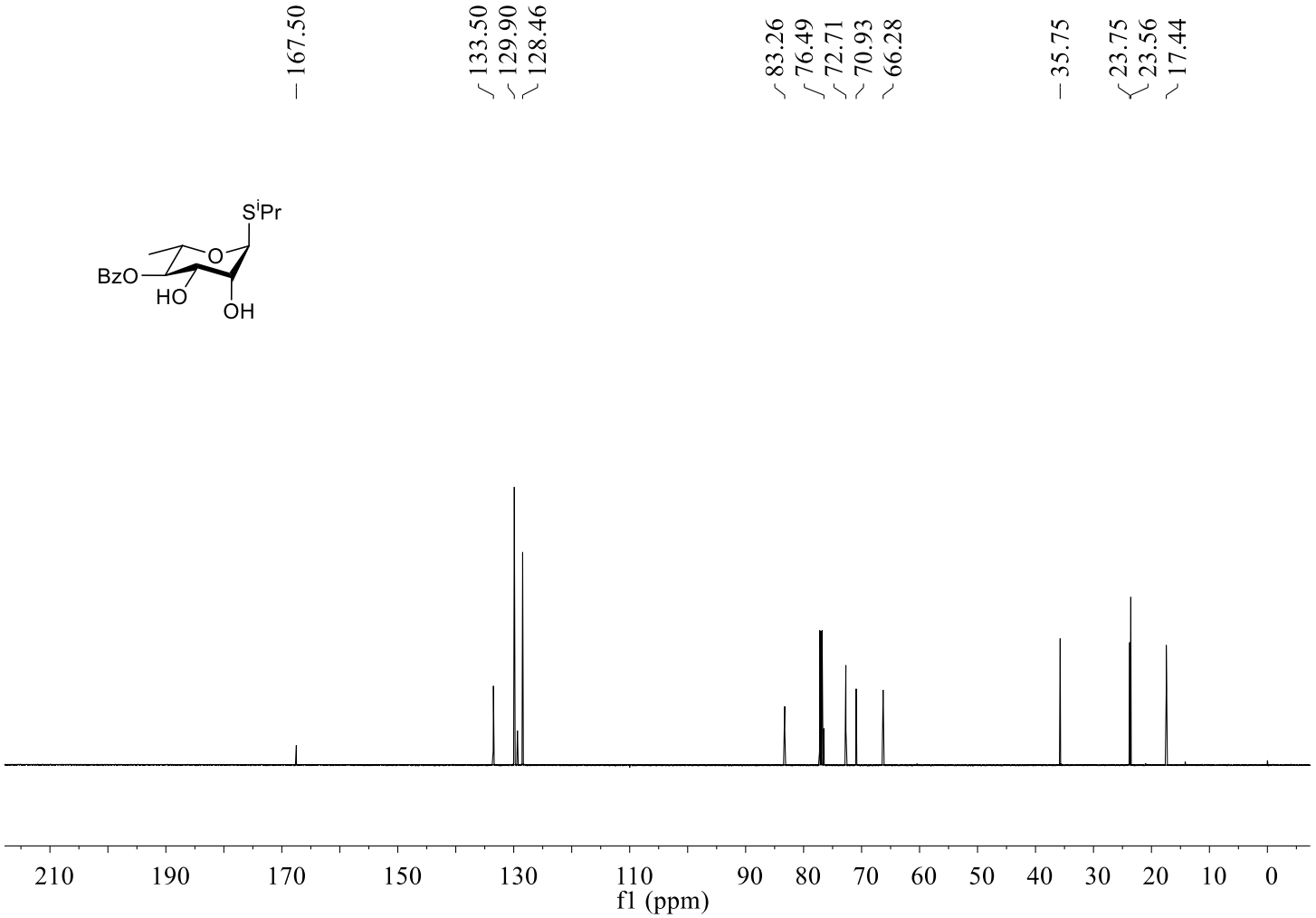
^{1}H NMR spectrum of compound **14** (600 MHz, $CDCl_3$)



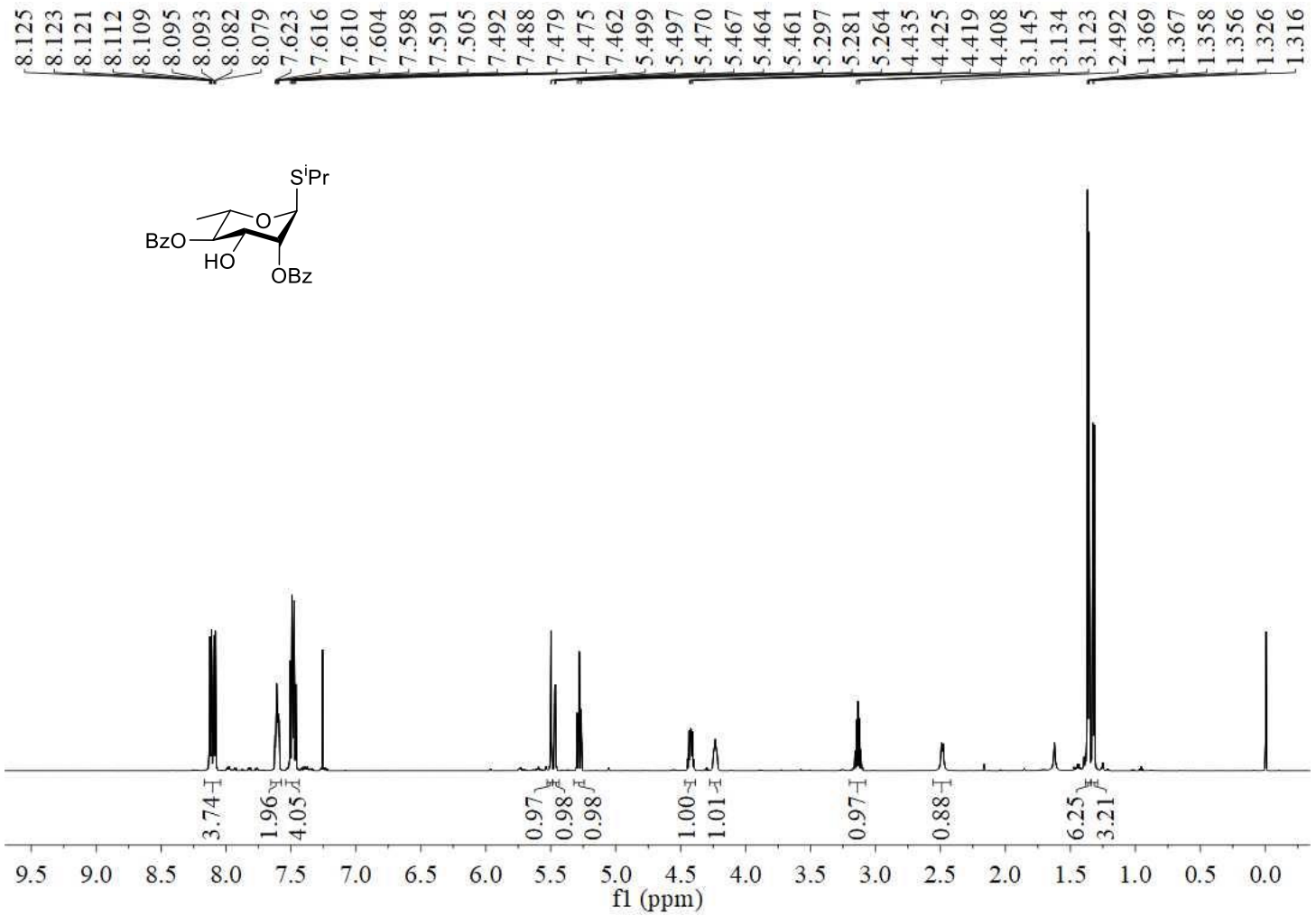
^{13}C NMR spectrum of compound **14** (150 MHz, $CDCl_3$)



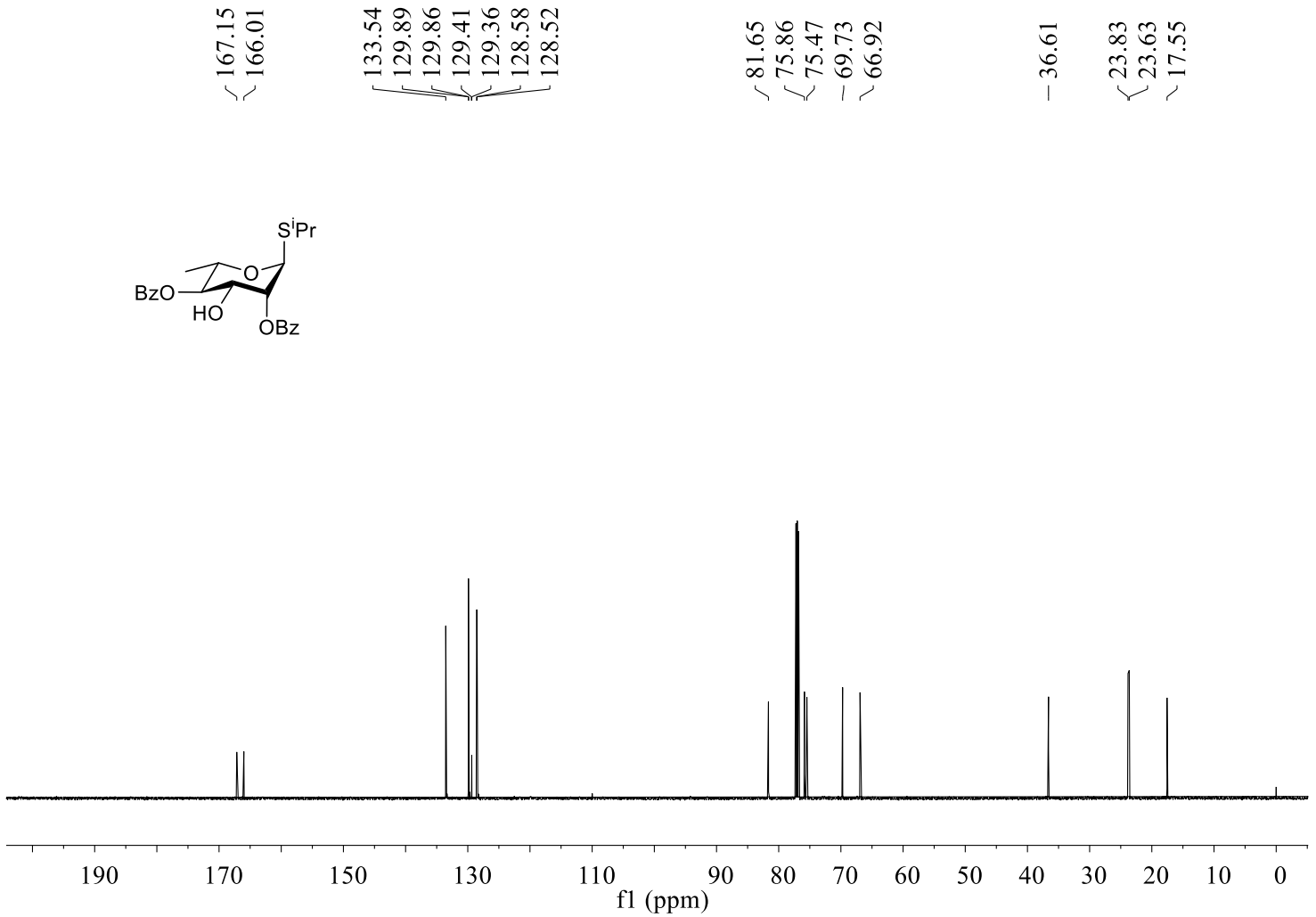
^{1}H NMR spectrum of compound **9** (600 MHz, $CDCl_3$)



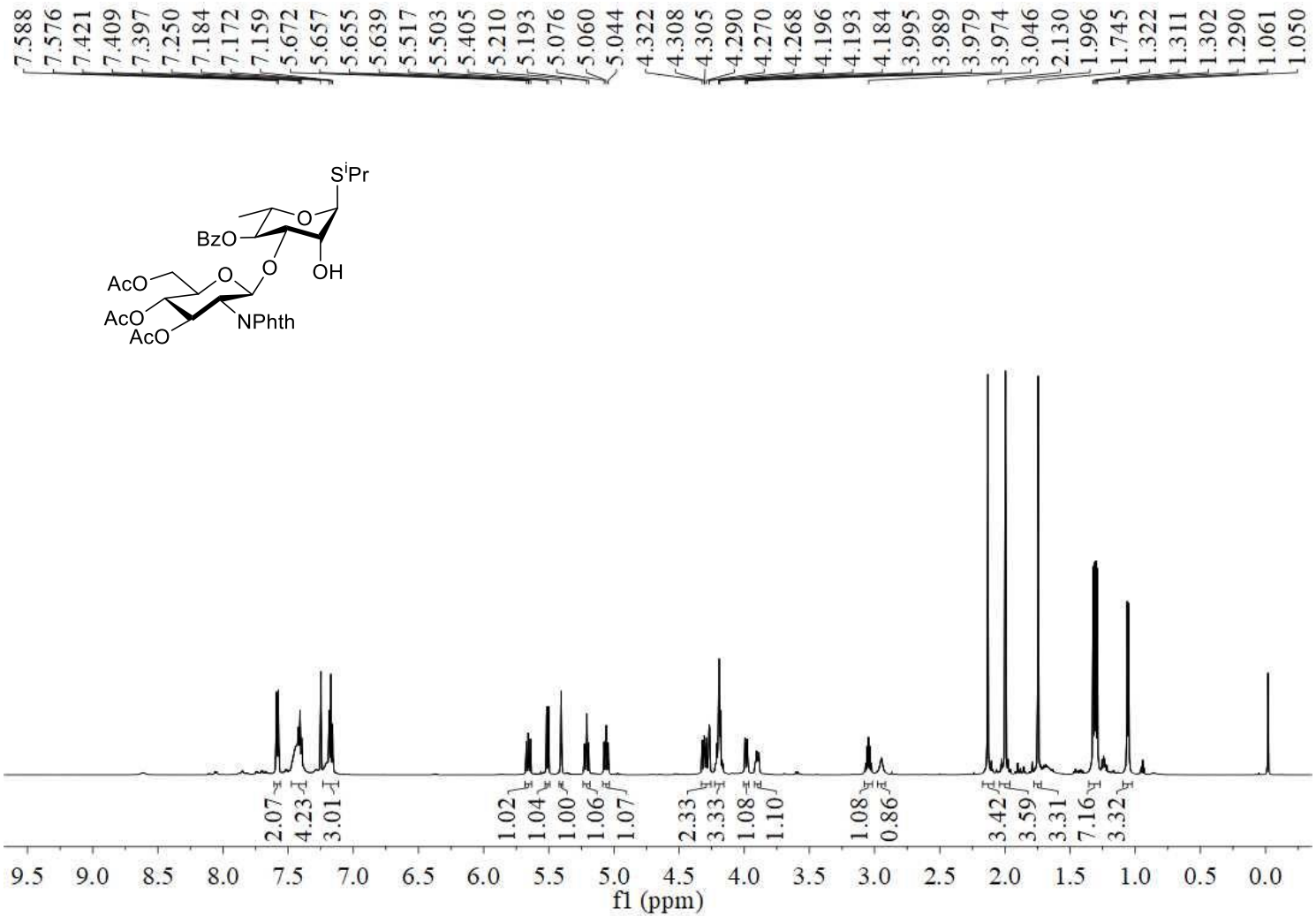
^{13}C NMR spectrum of compound **9** (150 MHz, $CDCl_3$)



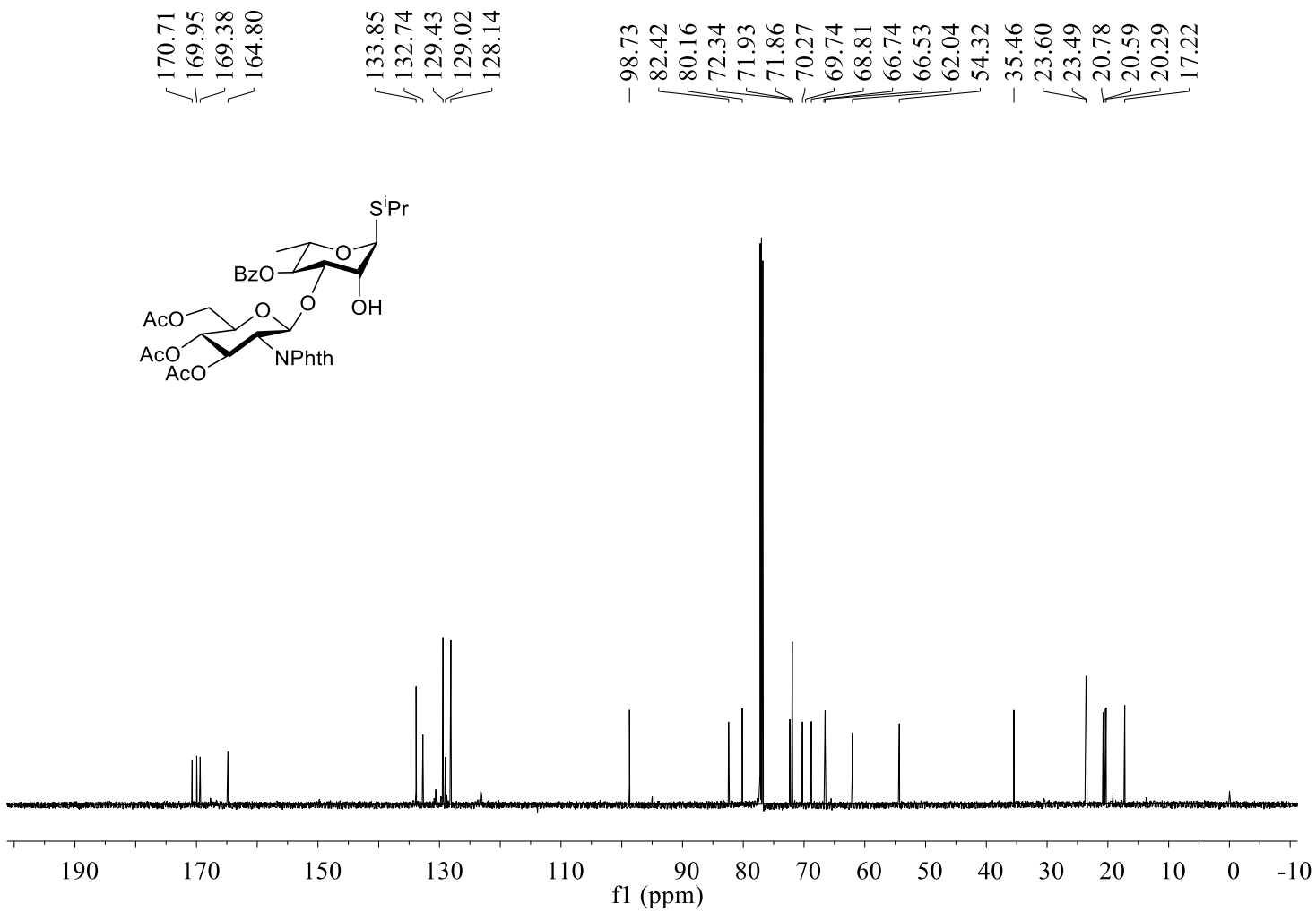
^{1}H NMR spectrum of compound **10** (600 MHz, $CDCl_3$)



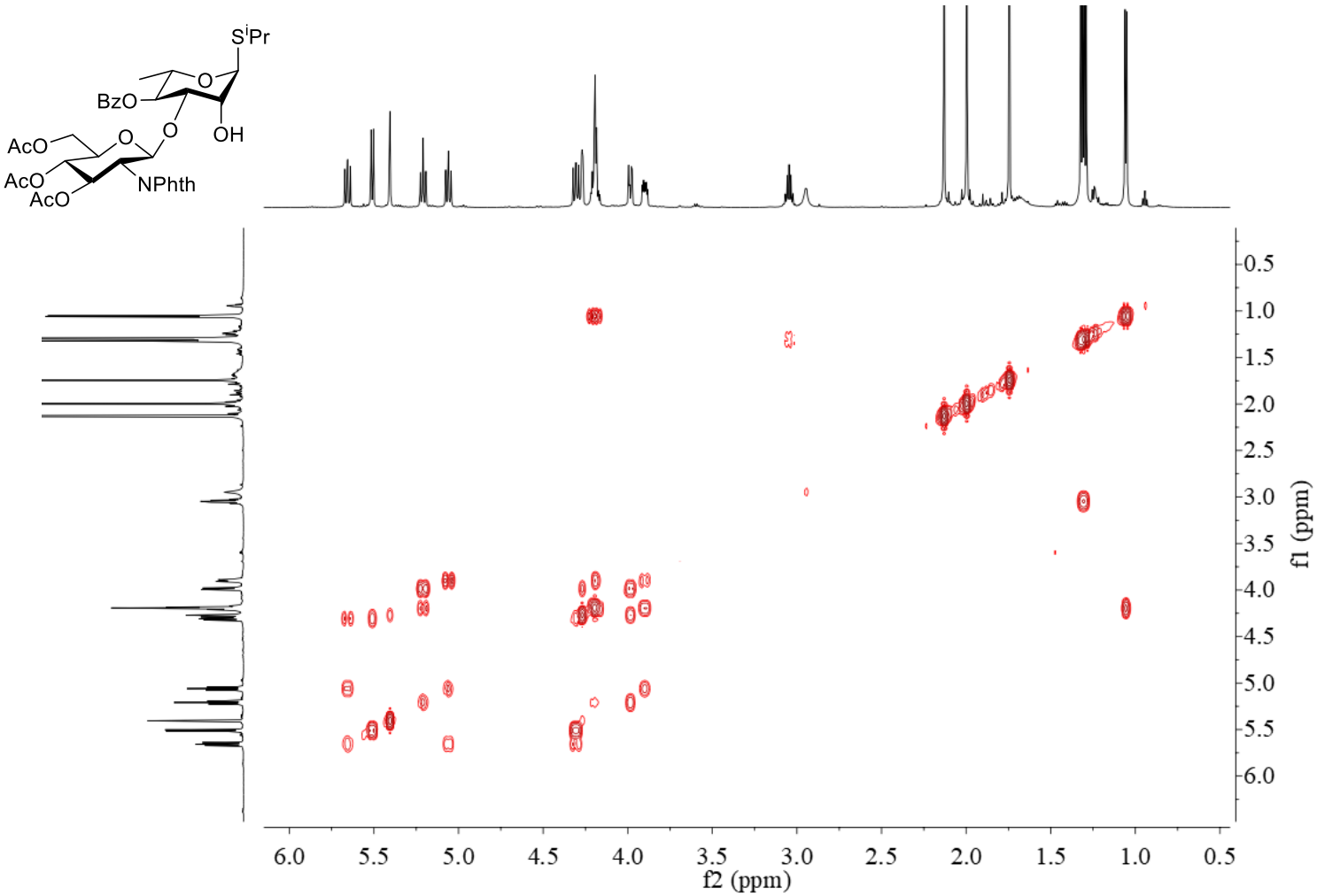
^{13}C NMR spectrum of compound **10** (150 MHz, $CDCl_3$)



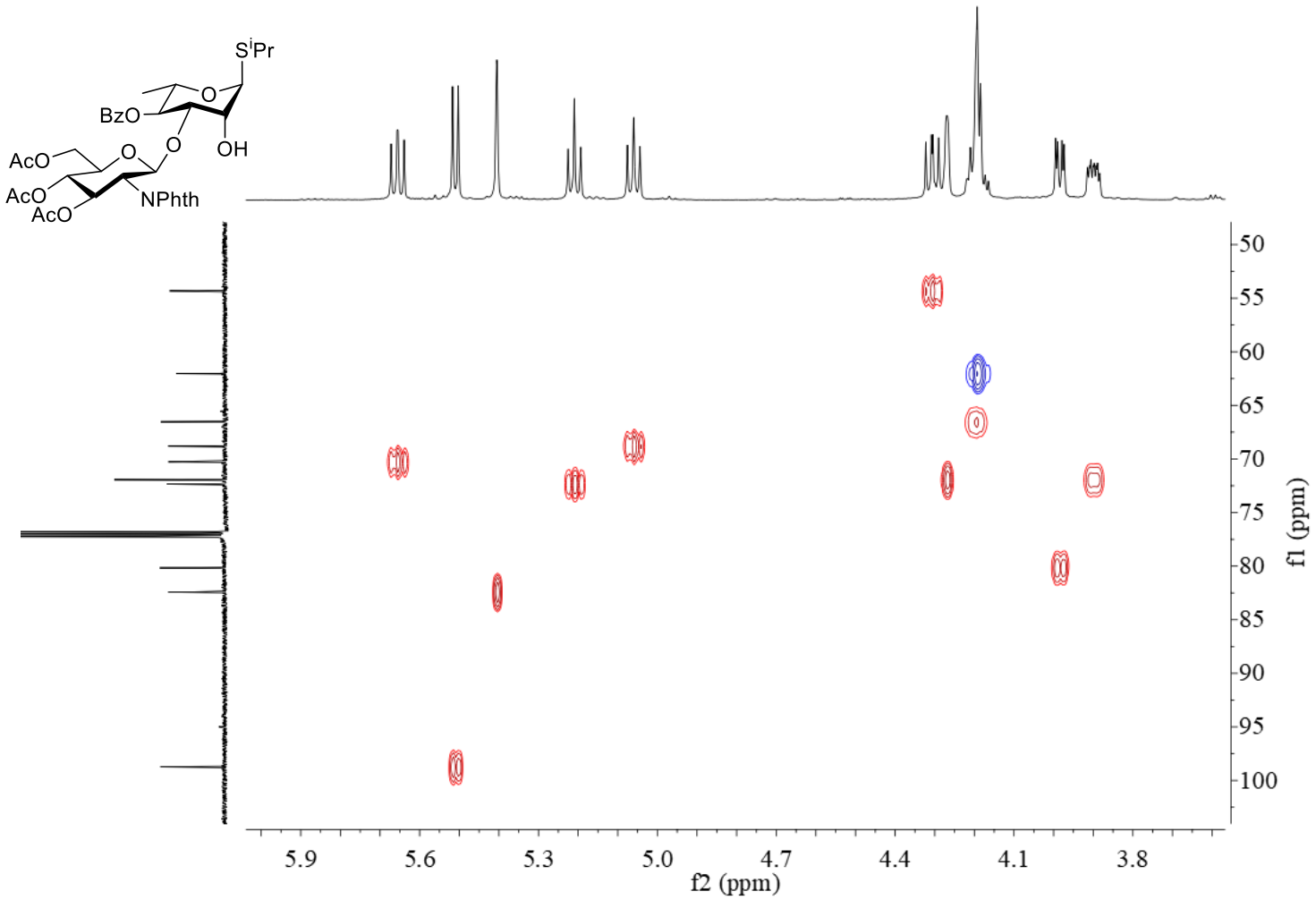
^{1}H NMR spectrum of compound **15** (600 MHz, $CDCl_3$)



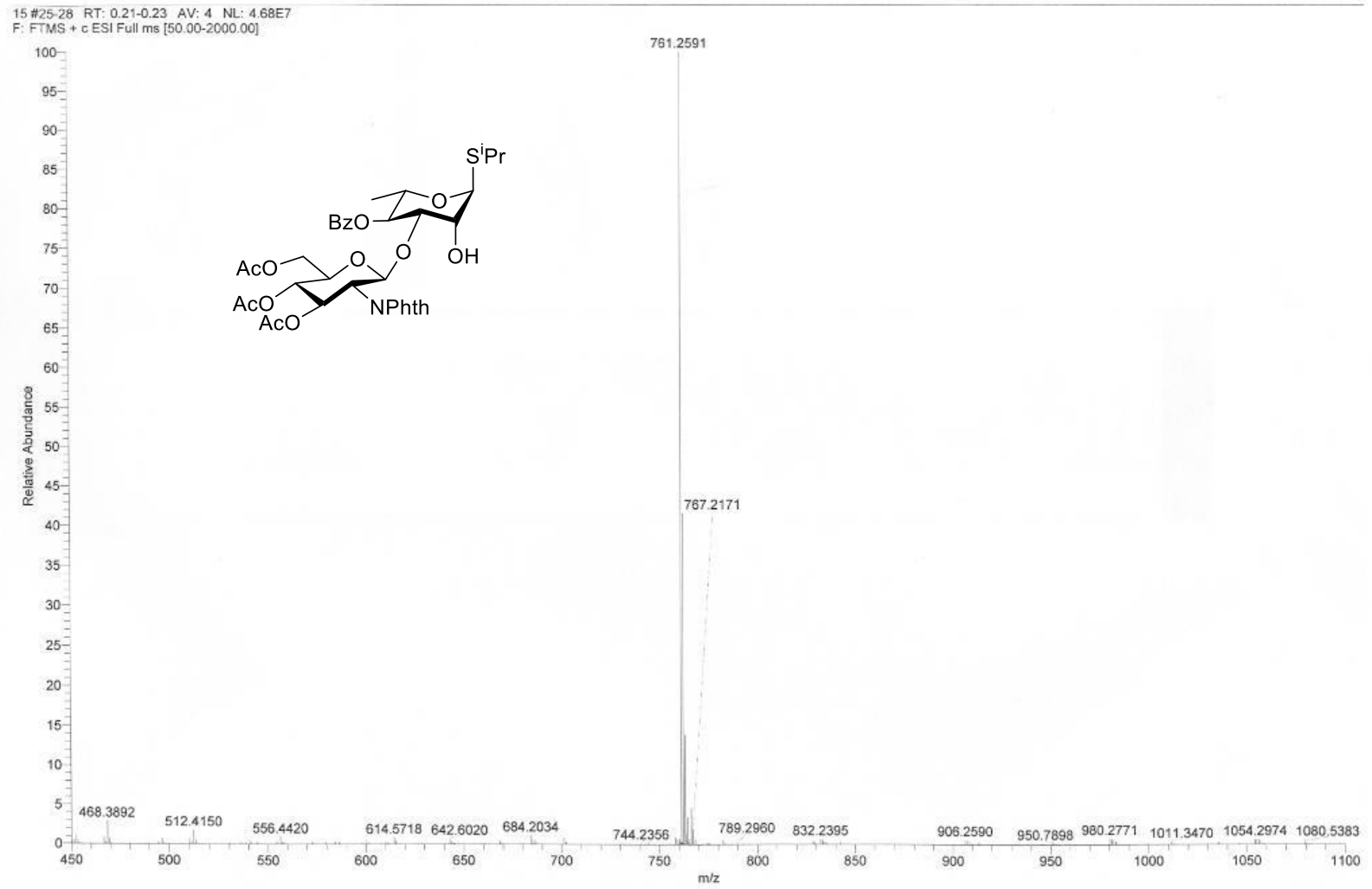
^{13}C NMR spectrum of compound **15** (150 MHz, $CDCl_3$)



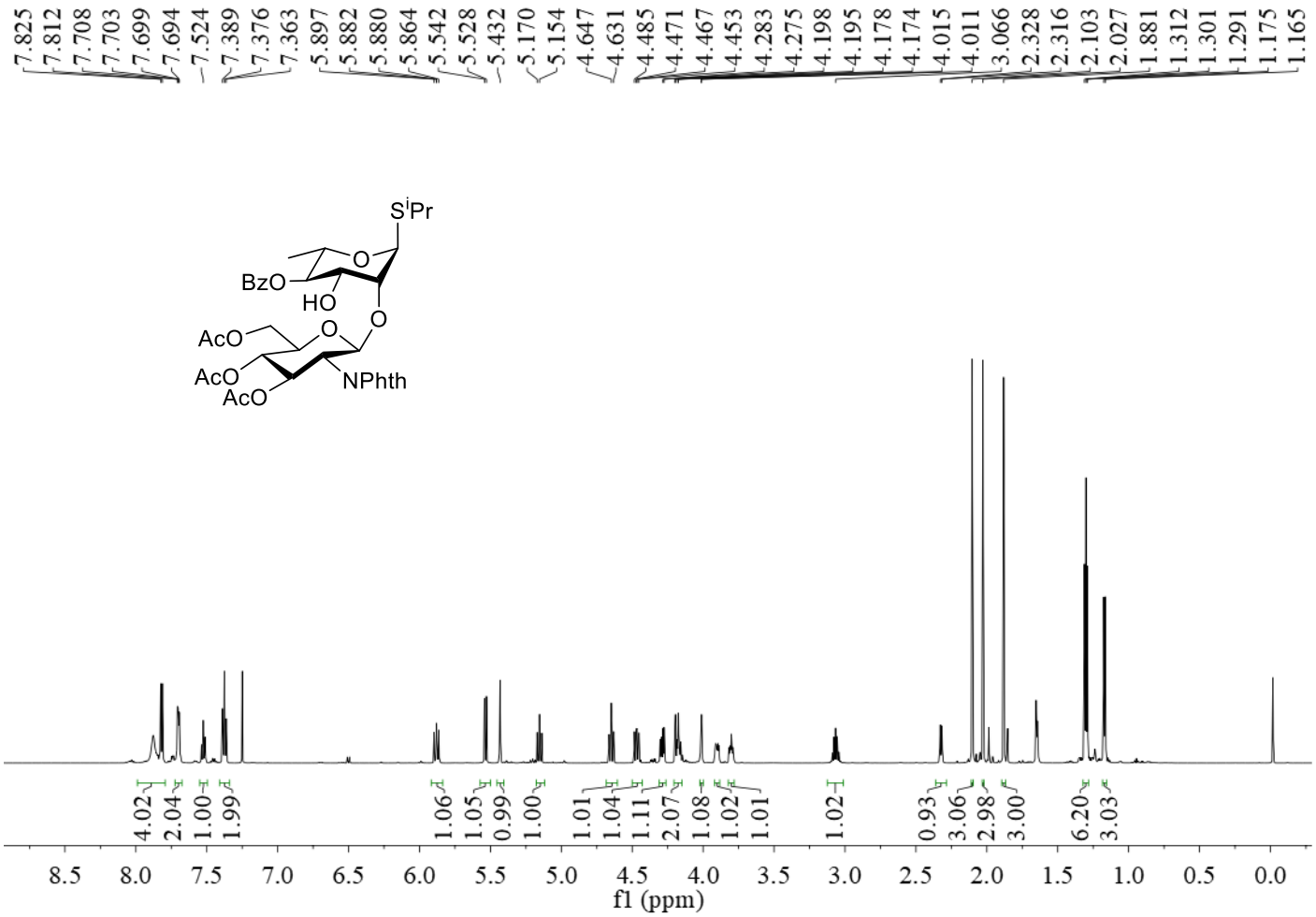
^{1}H-^{1}H COSY spectrum of compound **15** (600 MHz, $CDCl_3$)



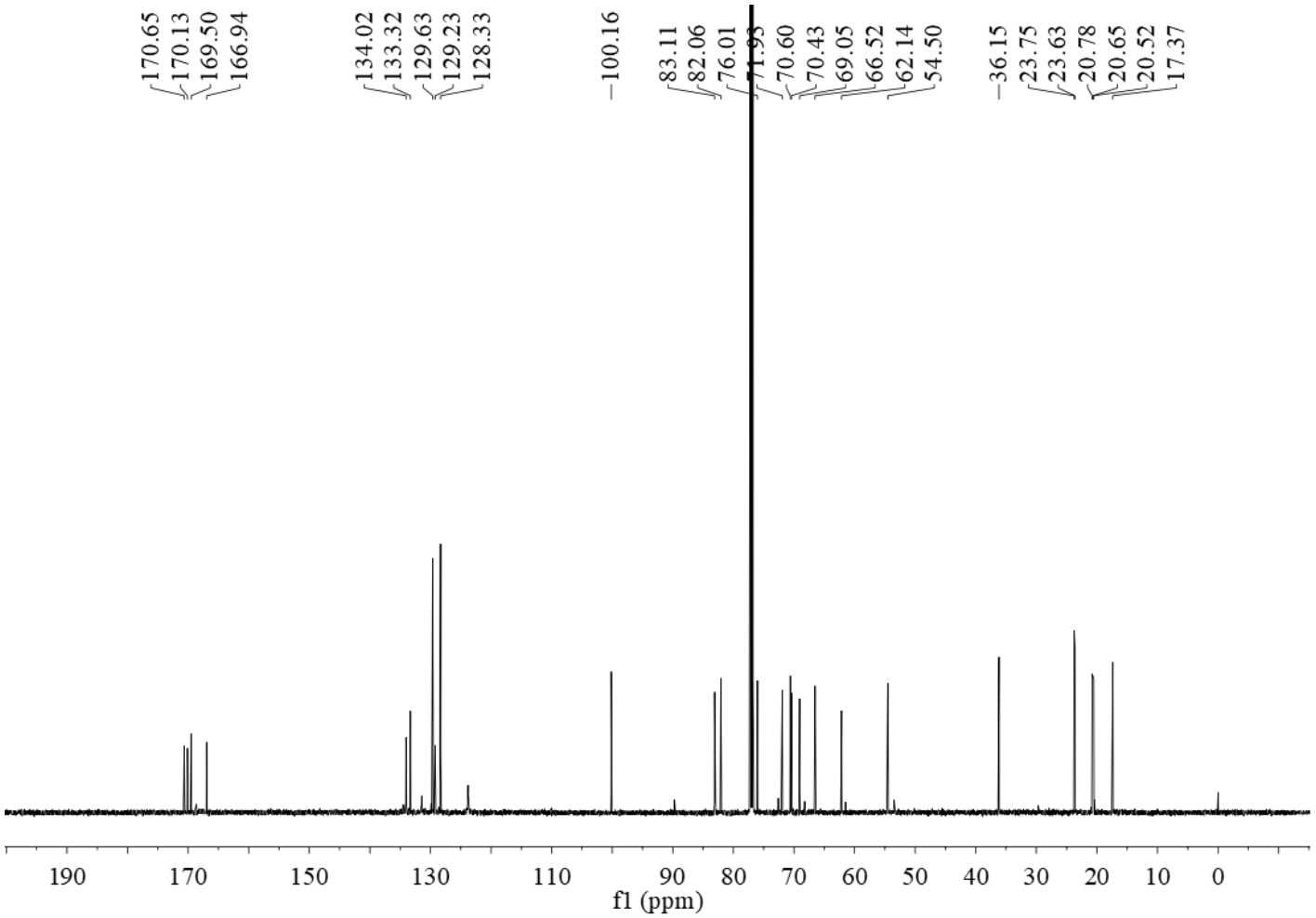
^{1}H-^{13}C HSQC spectrum of compound **15** (600/150 MHz, $CDCl_3$)



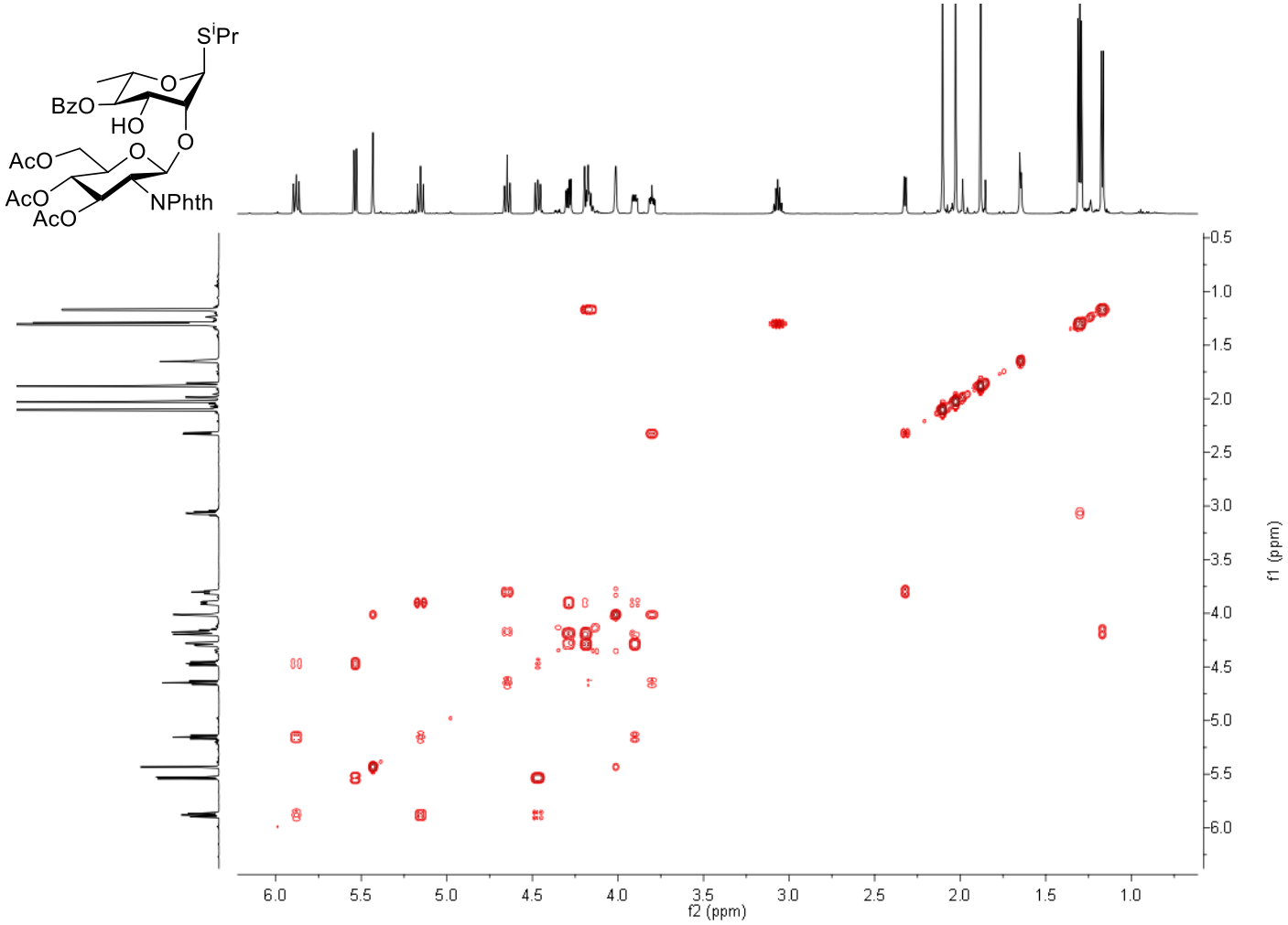
HR-ESI-(+) mass spectrum of compound **15**



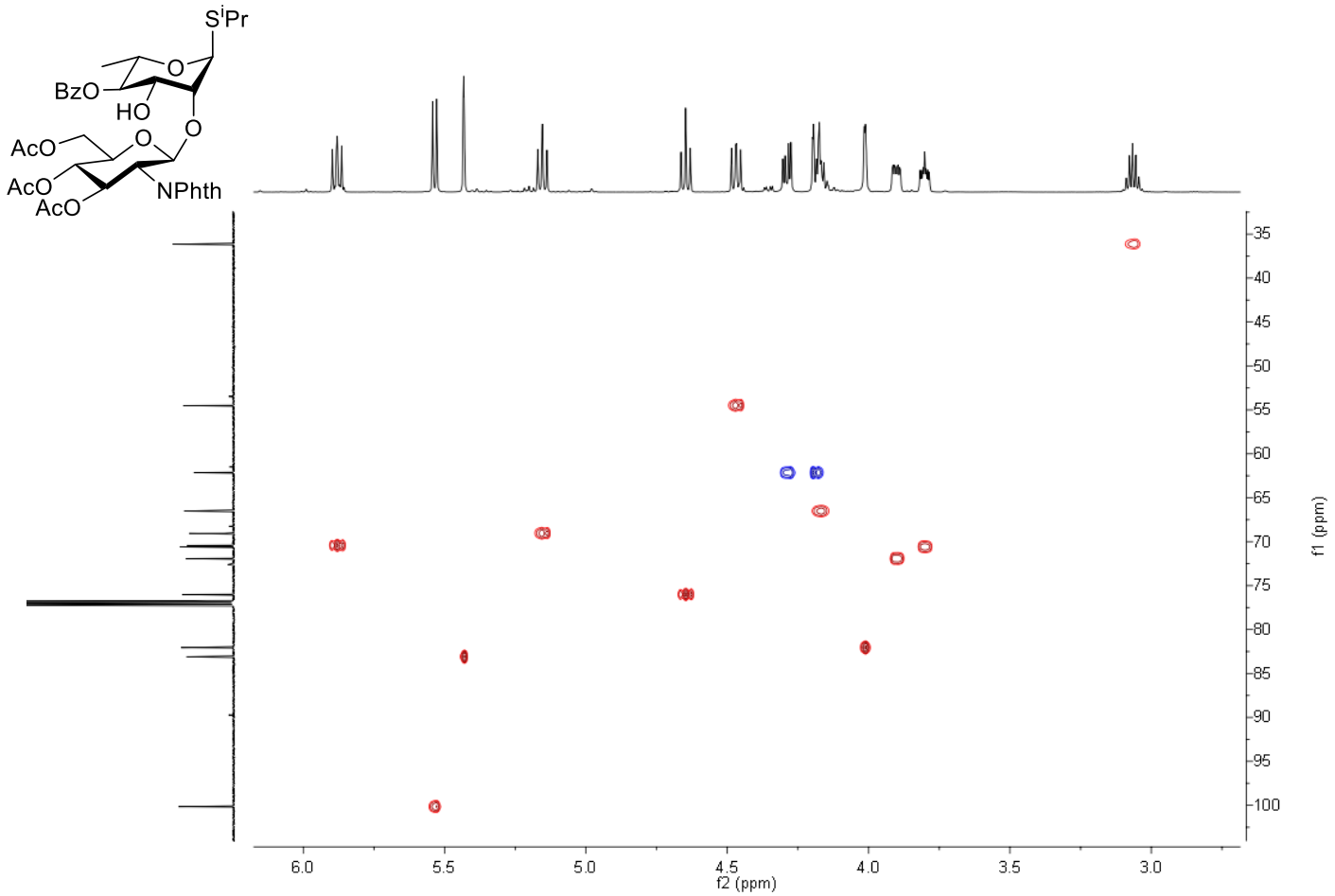
^{1}H NMR spectrum of compound **15a** (600 MHz, $CDCl_3$)



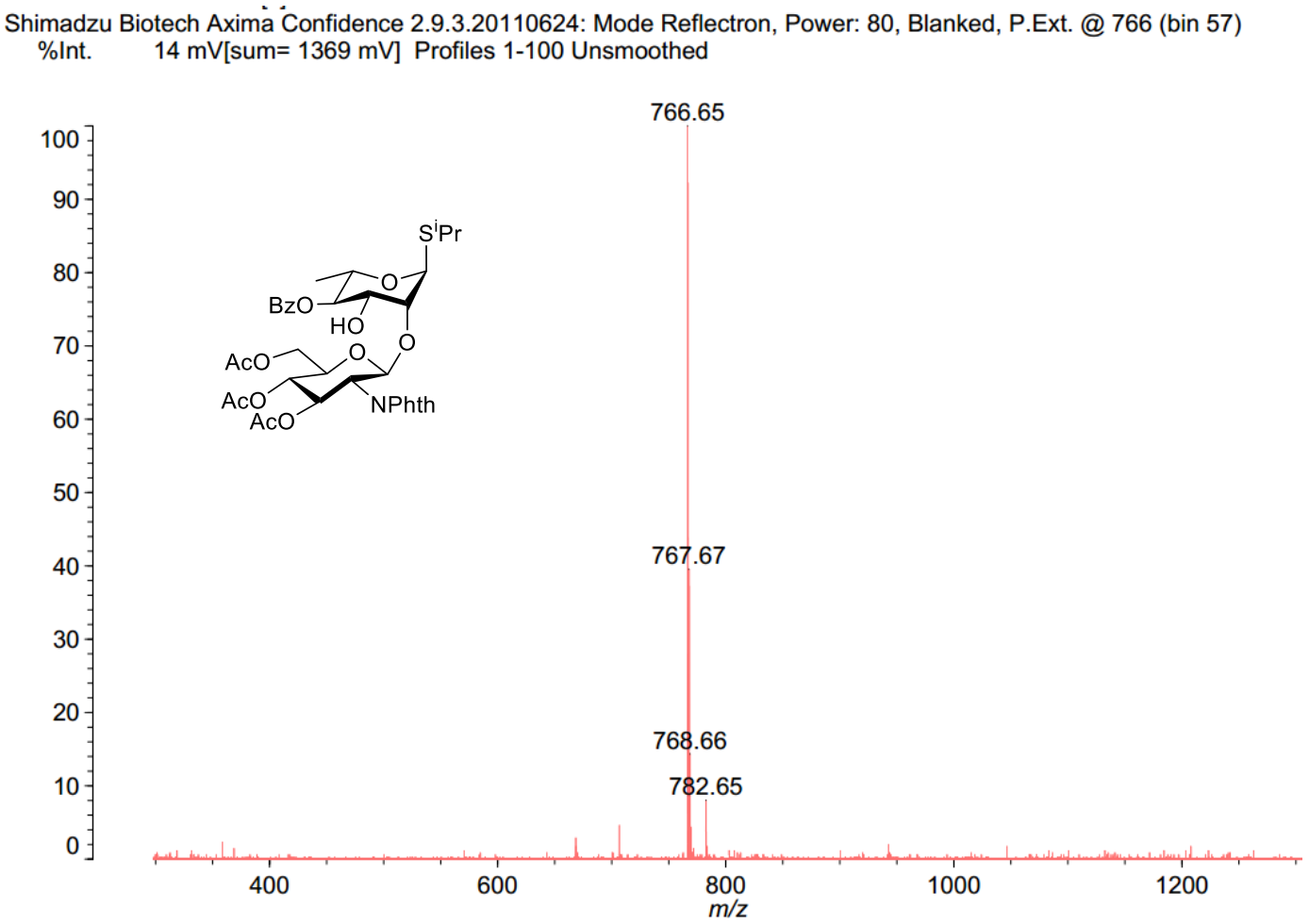
^{13}C NMR spectrum of compound **15a** (150 MHz, $CDCl_3$)



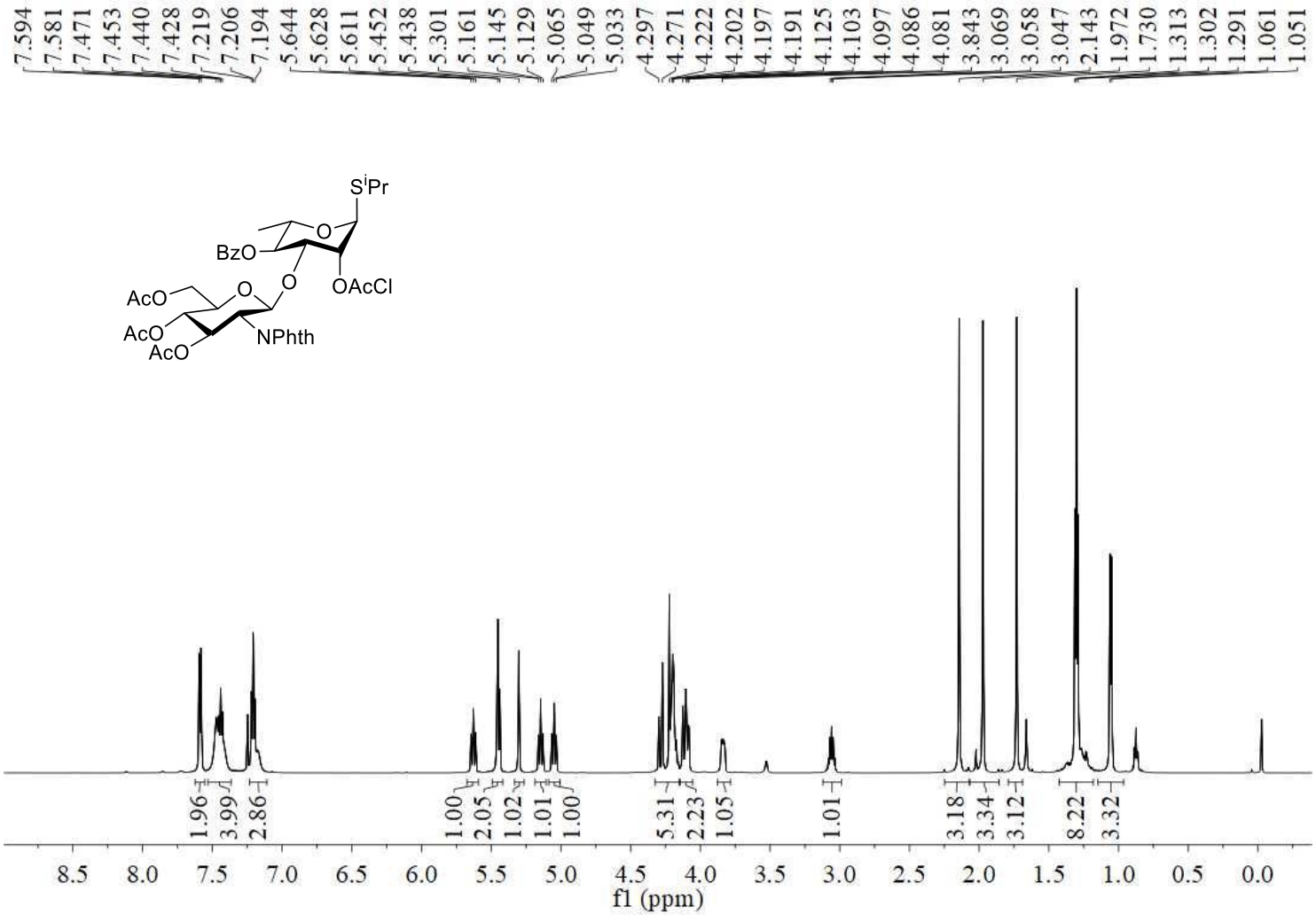
^{1}H-^{1}H COSY spectrum of compound **15a** (600 MHz, $CDCl_3$)



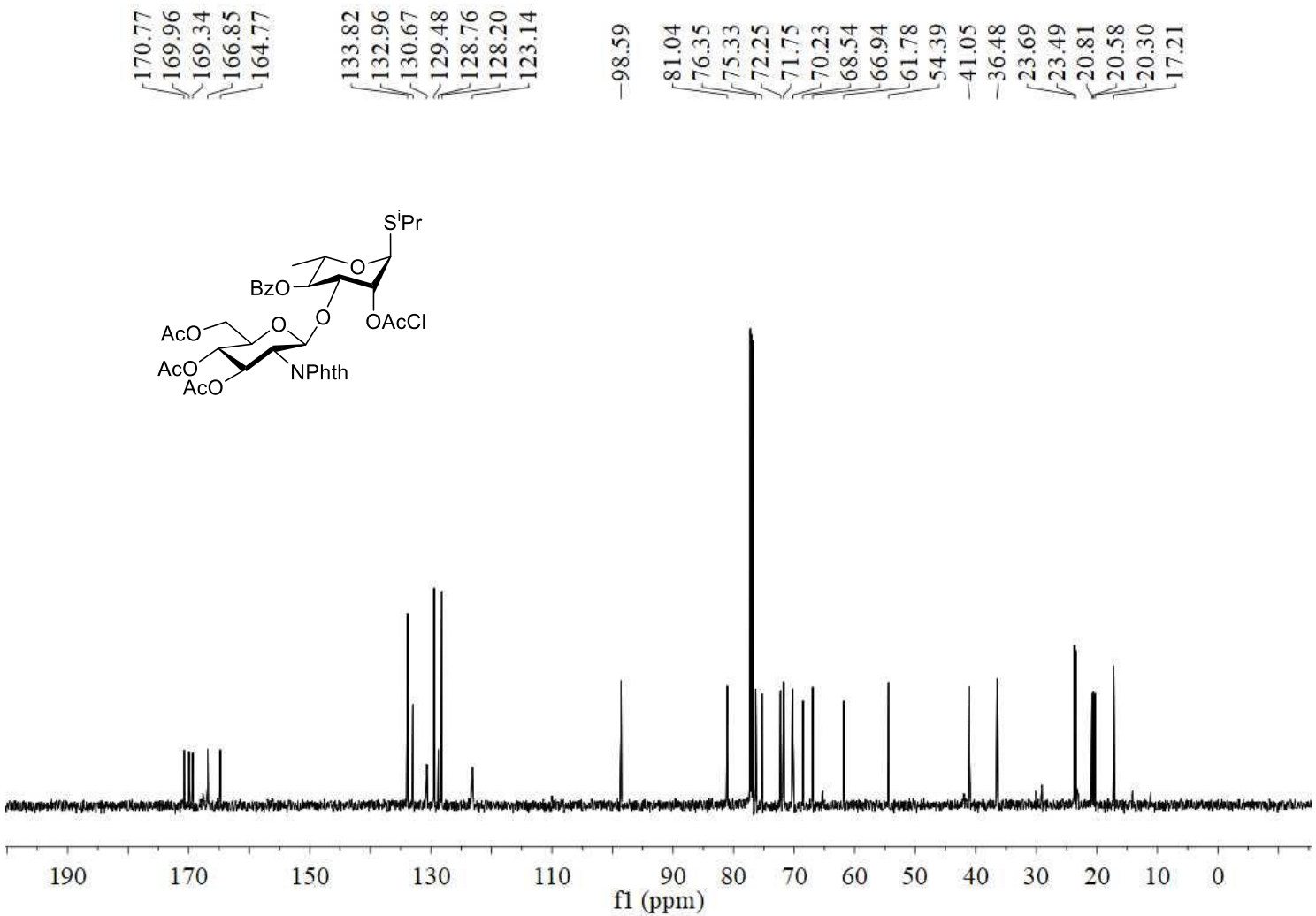
^{1}H-^{13}C HSQC spectrum of compound **15a** (600/150 MHz, $CDCl_3$)



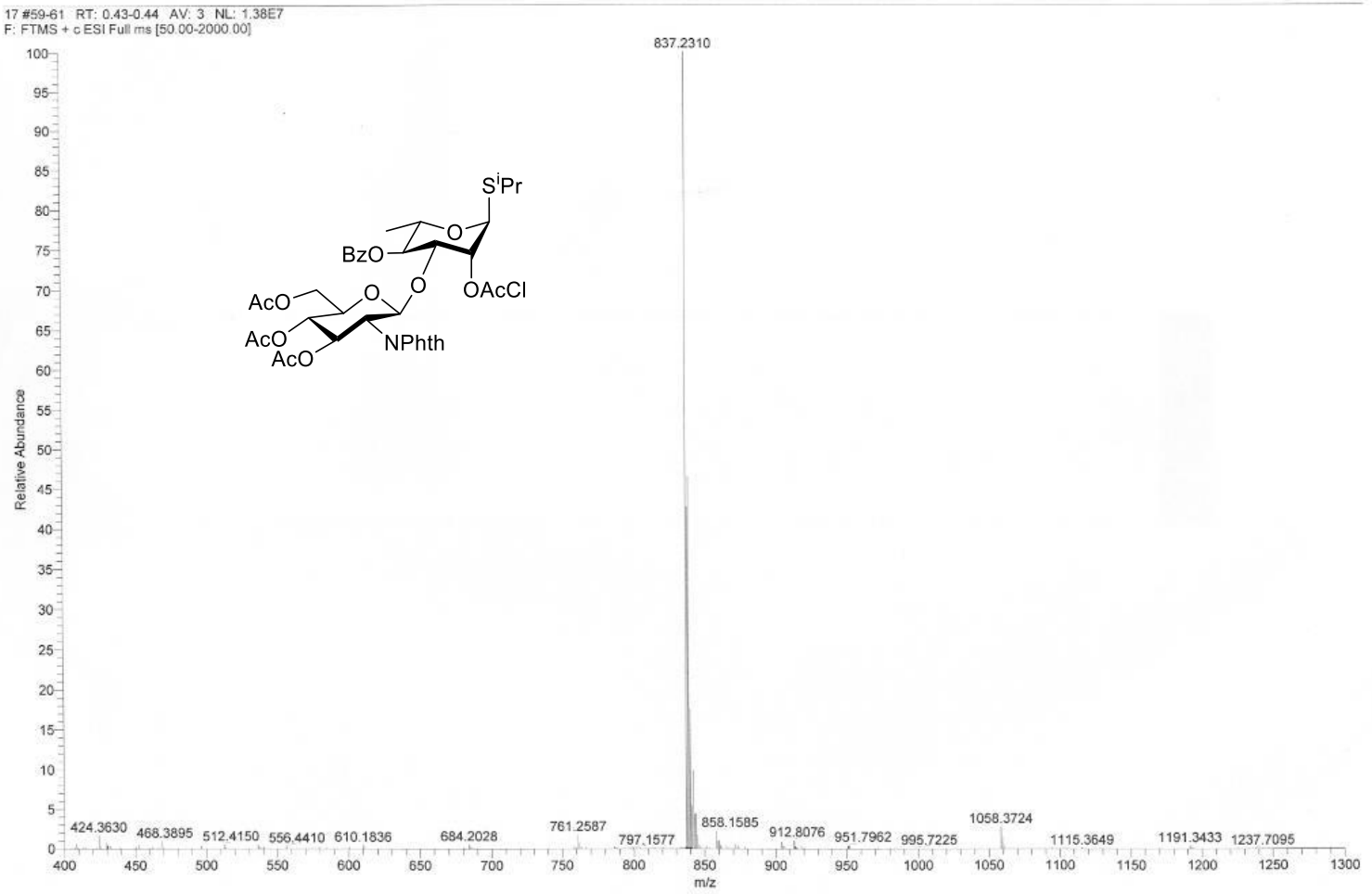
MALDI-TOF-(+) mass spectrum of compound **15a**



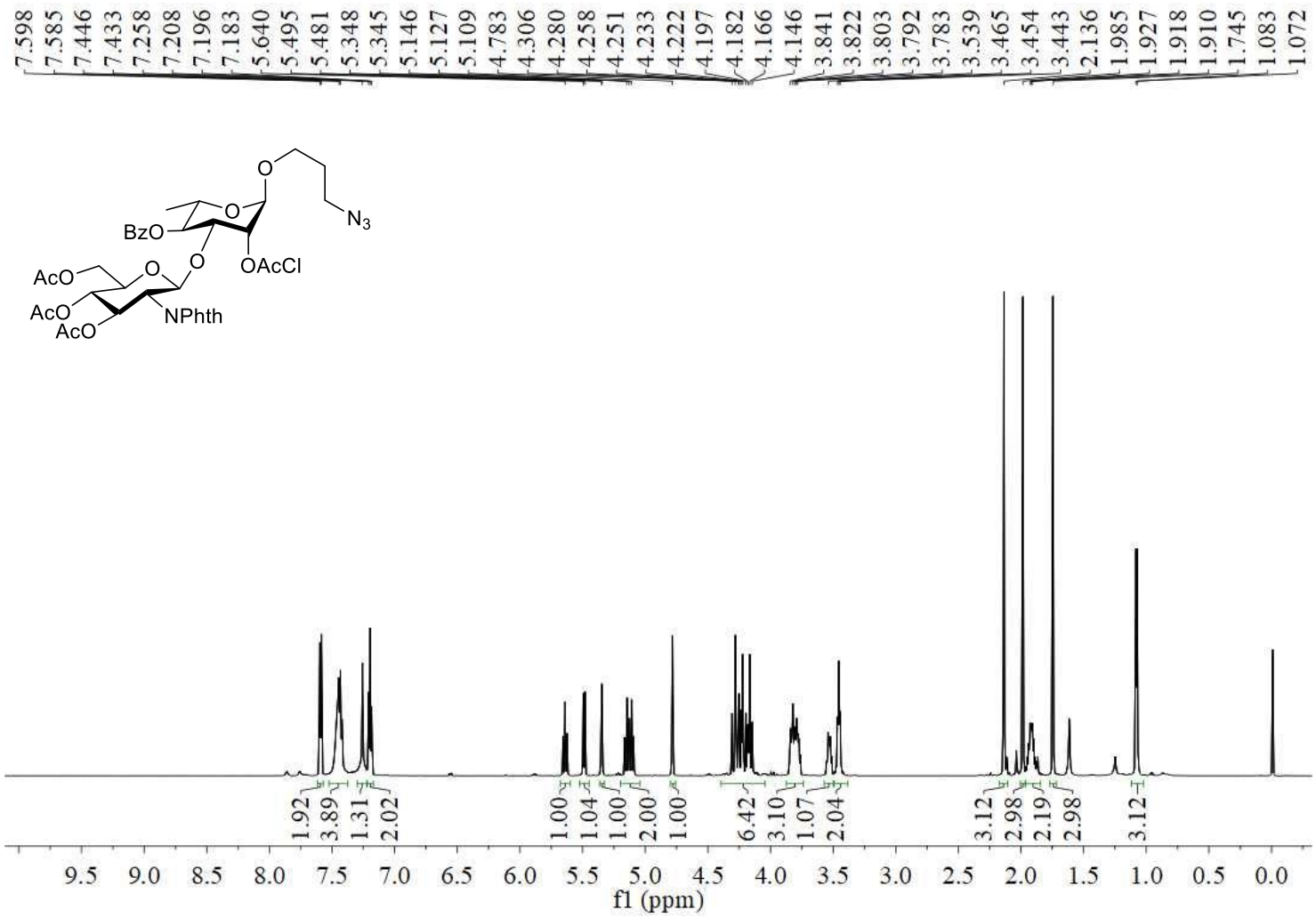
^{1}H NMR spectrum of compound **16** (600 MHz, $CDCl_3$)



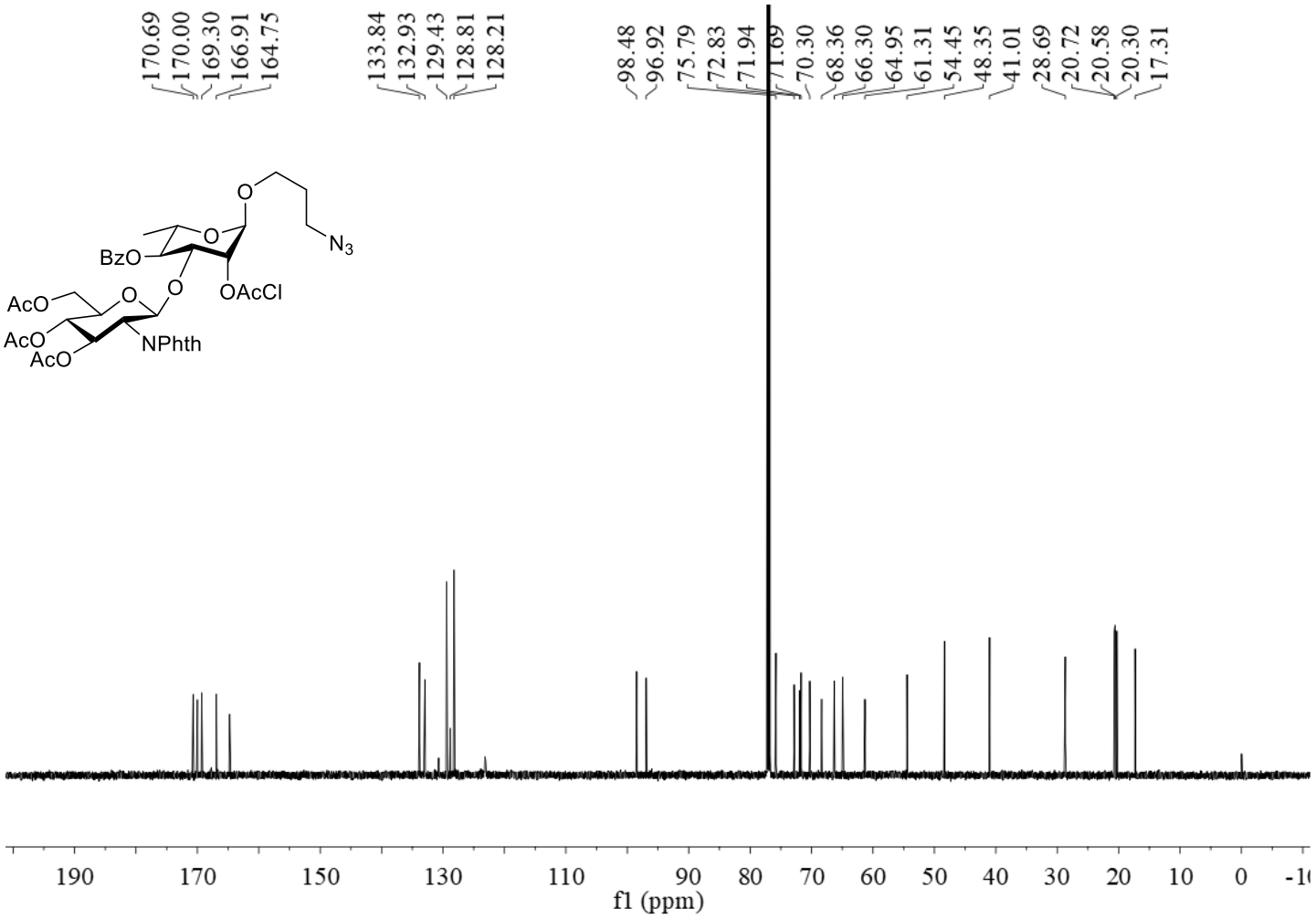
^{13}C NMR spectrum of compound **16** (150 MHz, $CDCl_3$)



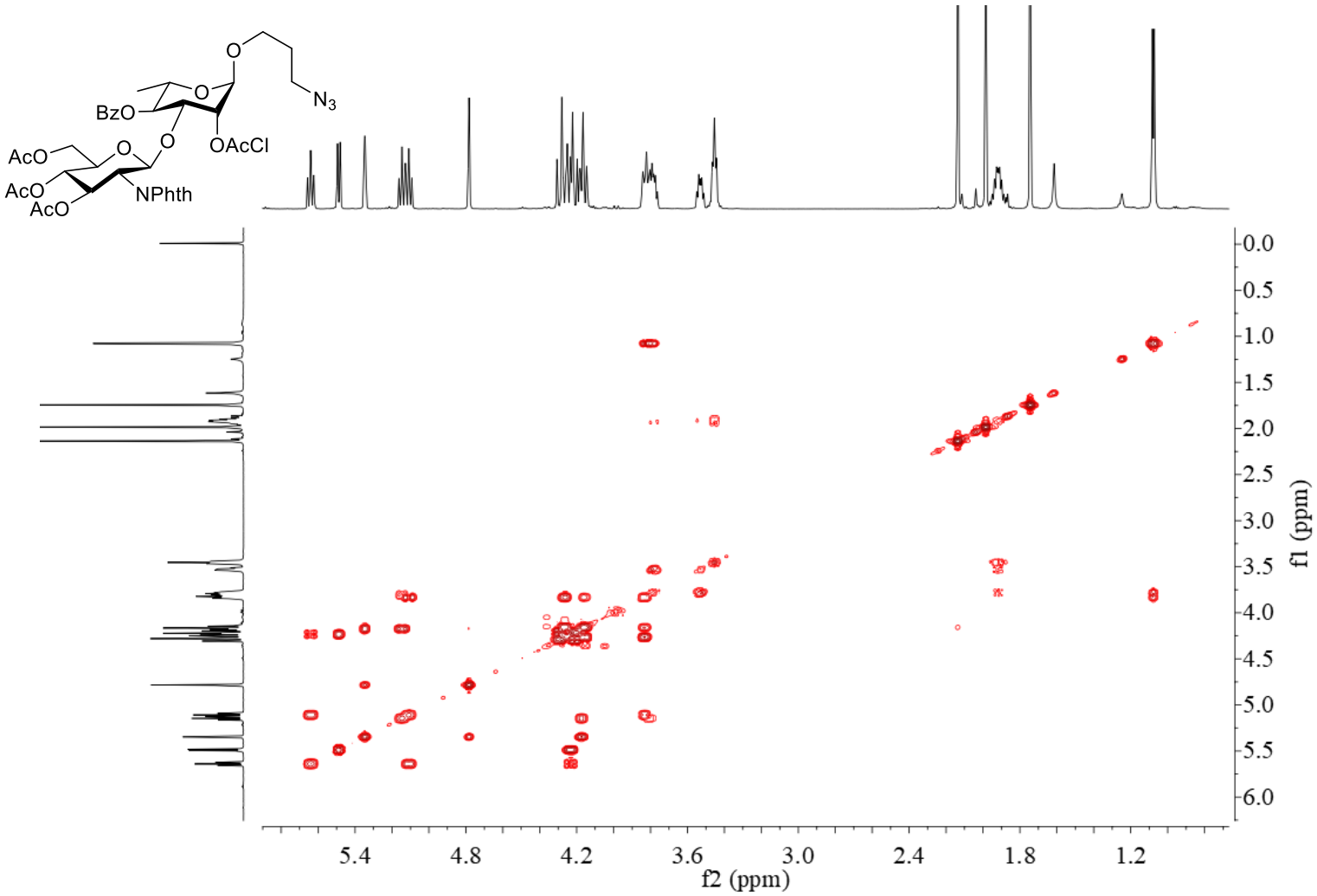
HR-ESI-(+) mass spectrum of compound **16**



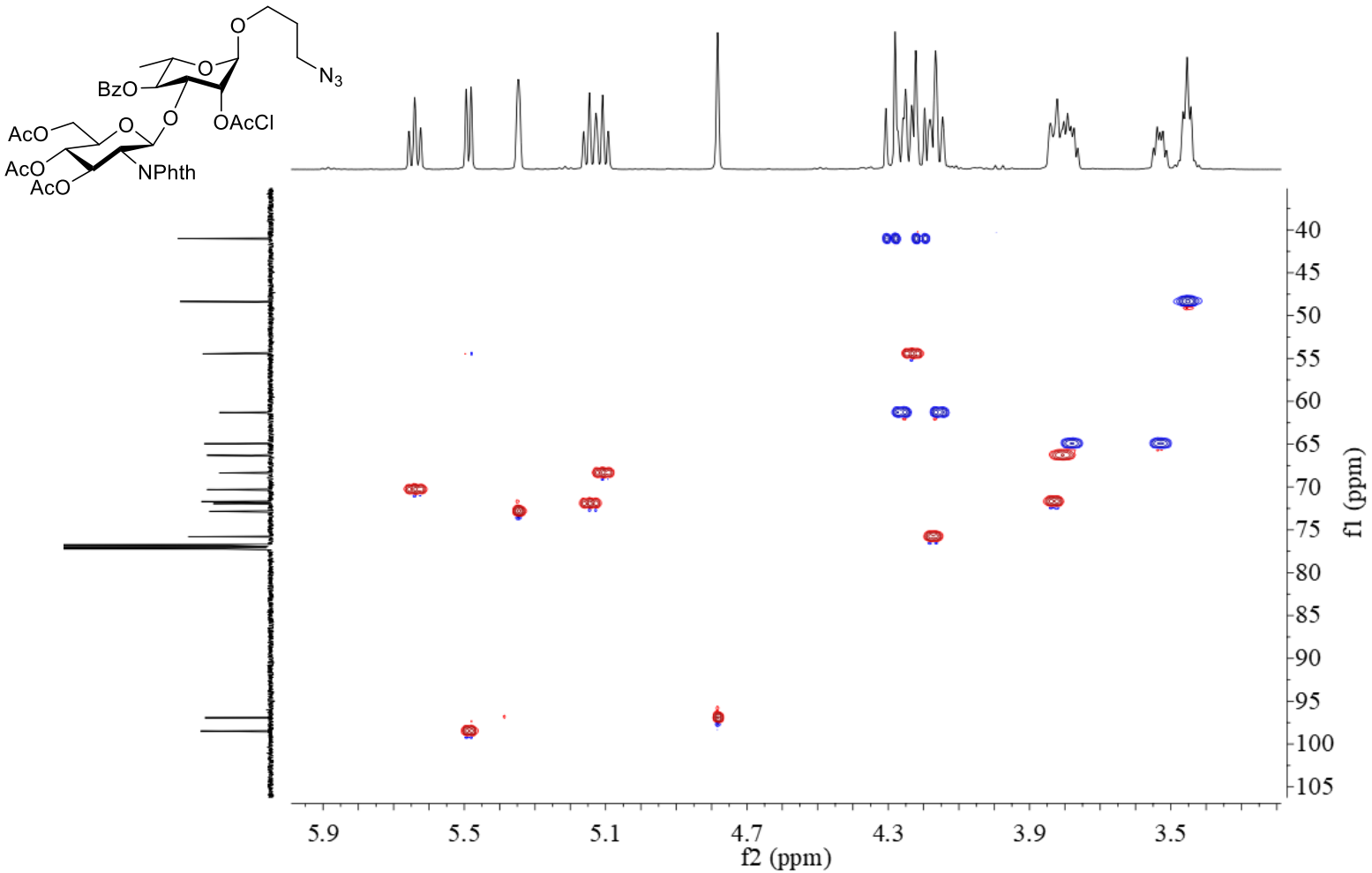
^{1}H NMR spectrum of compound **17** (600 MHz, $CDCl_3$)



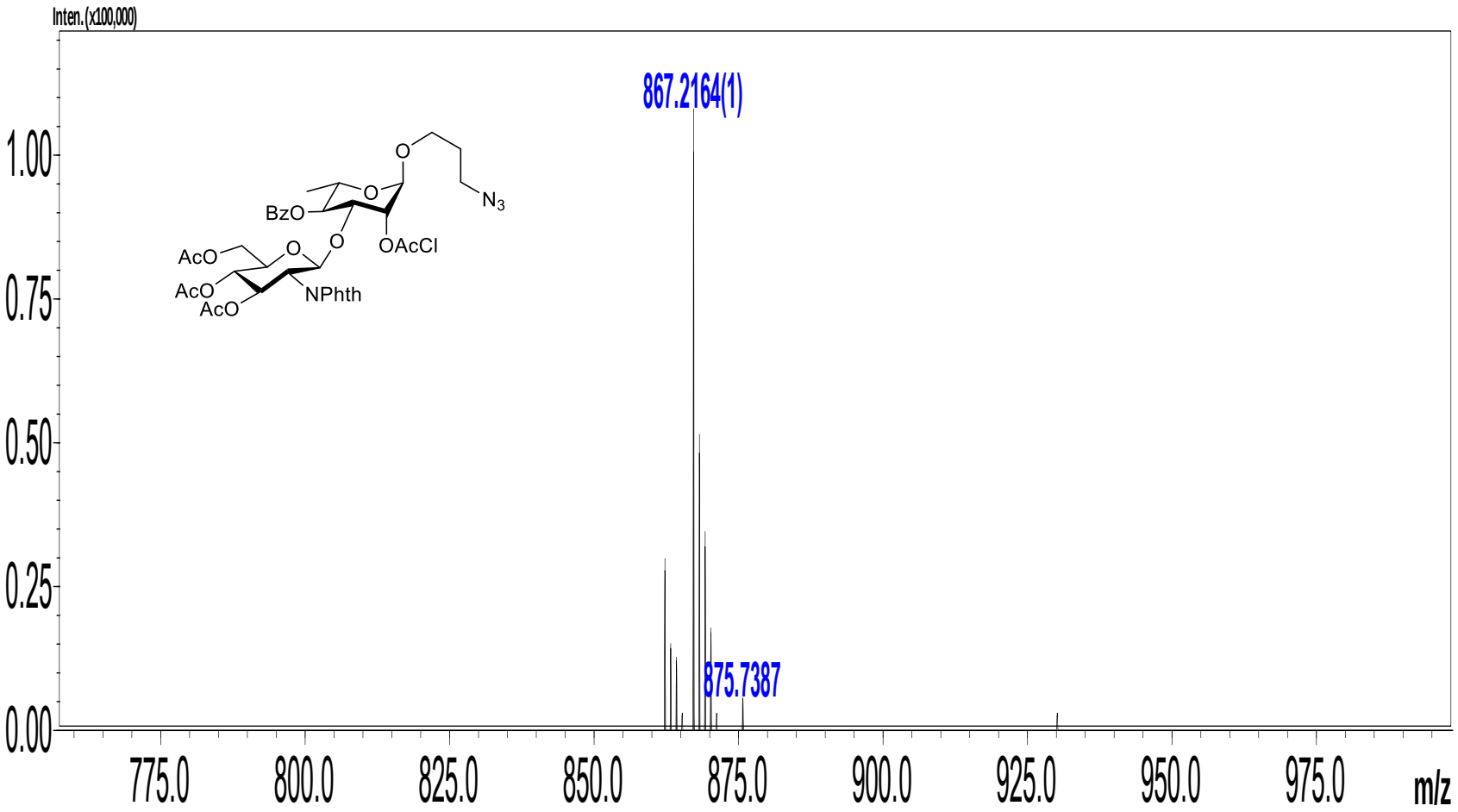
^{13}C NMR spectrum of compound **17** (150 MHz, $CDCl_3$)



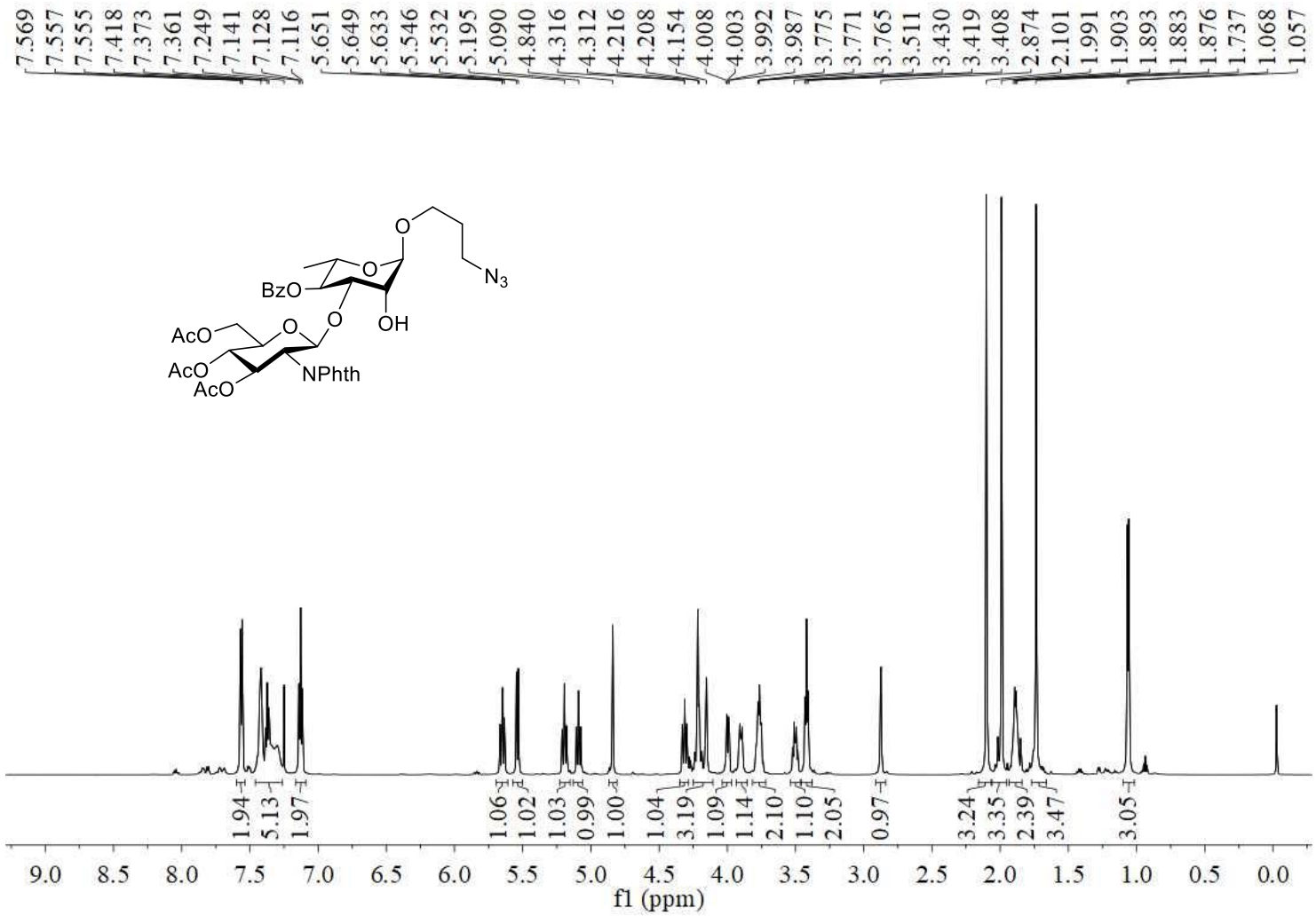
^{1}H-^{1}H COSY spectrum of compound **17** (600 MHz, $CDCl_3$)



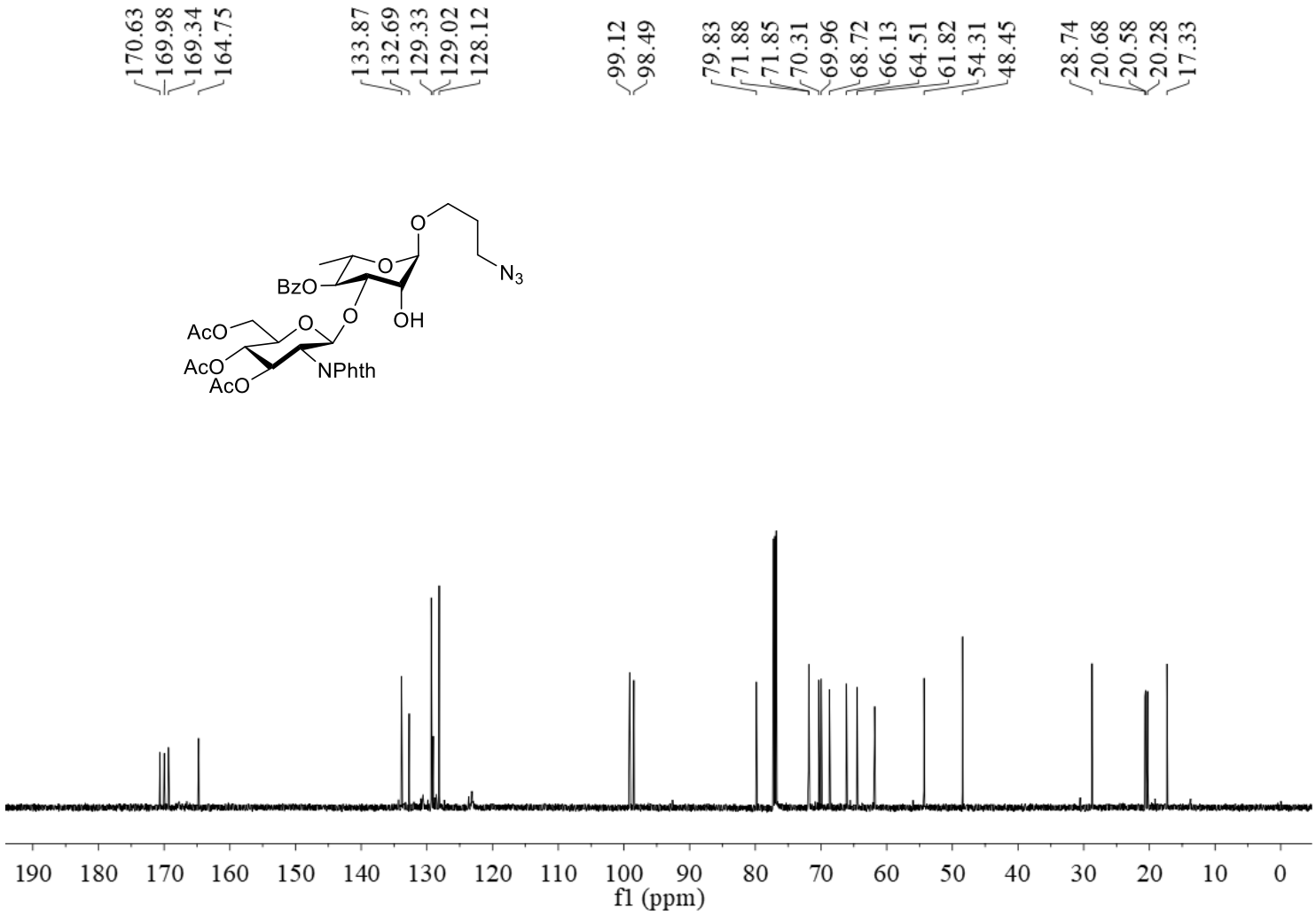
^{1}H-^{13}C HSQC spectrum of compound **17** (600/150 MHz, $CDCl_3$)



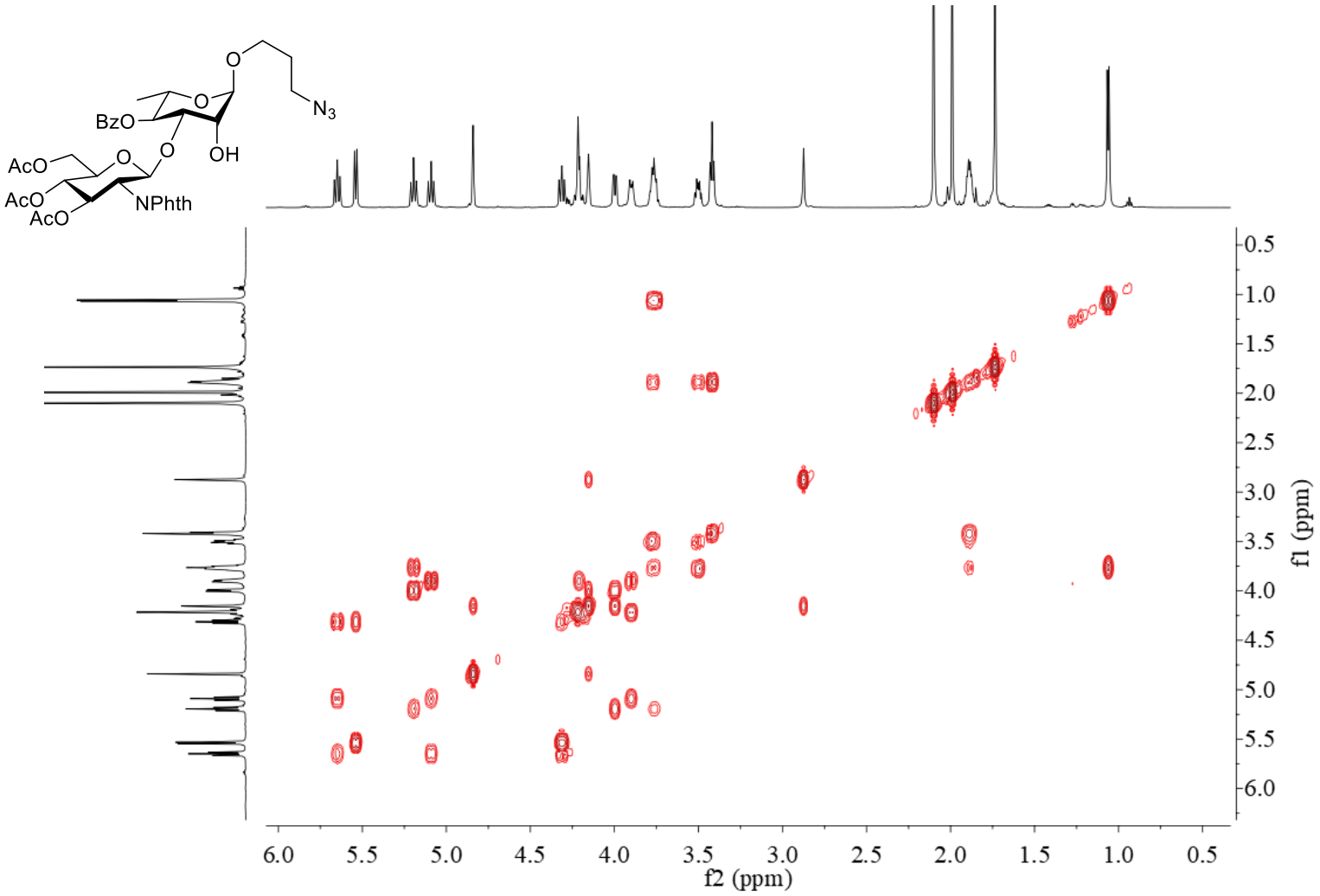
HR-ESI-(+) mass spectrum of compound **17**



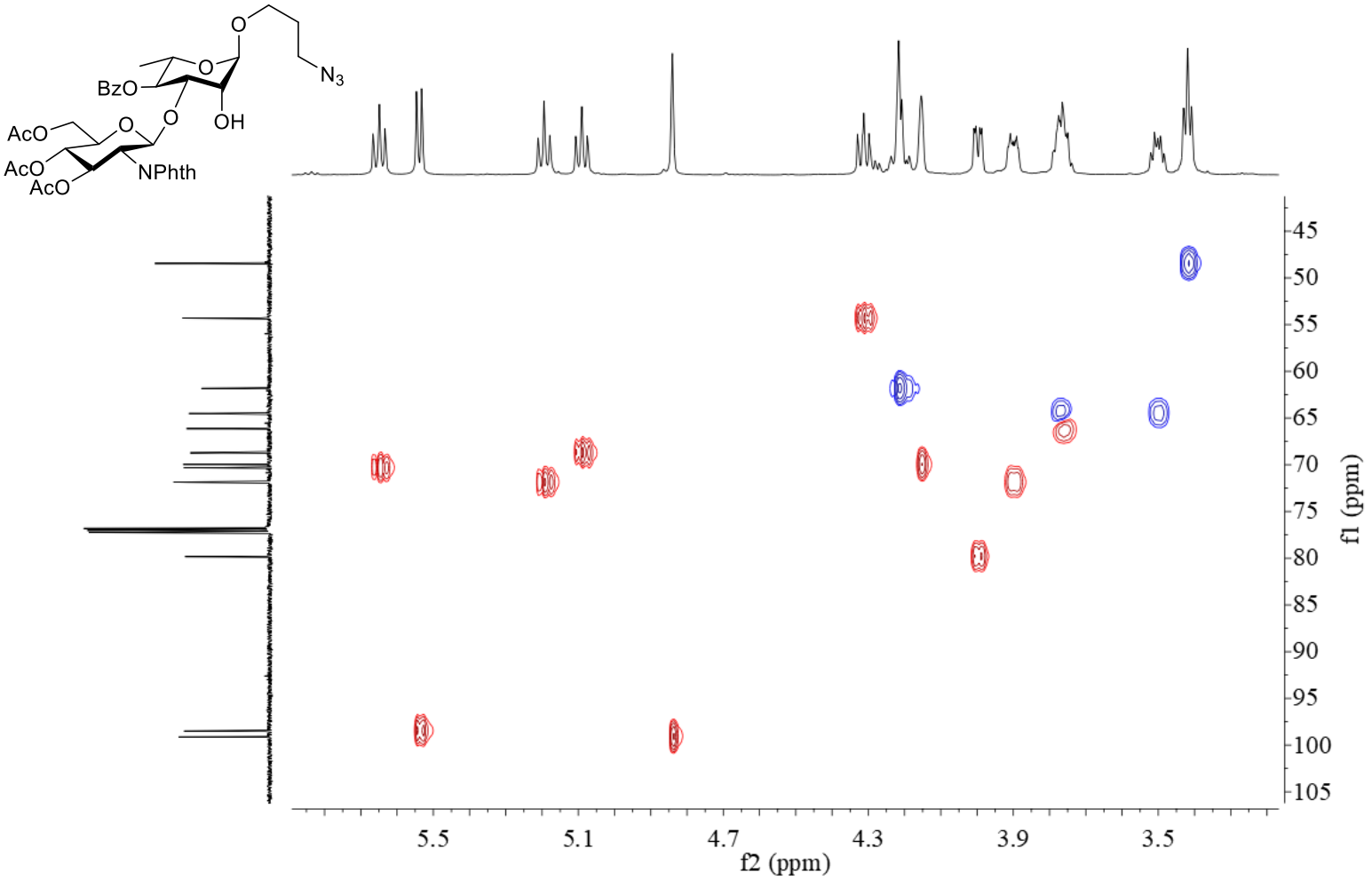
^{1}H NMR spectrum of compound **6** (600 MHz, $CDCl_3$)



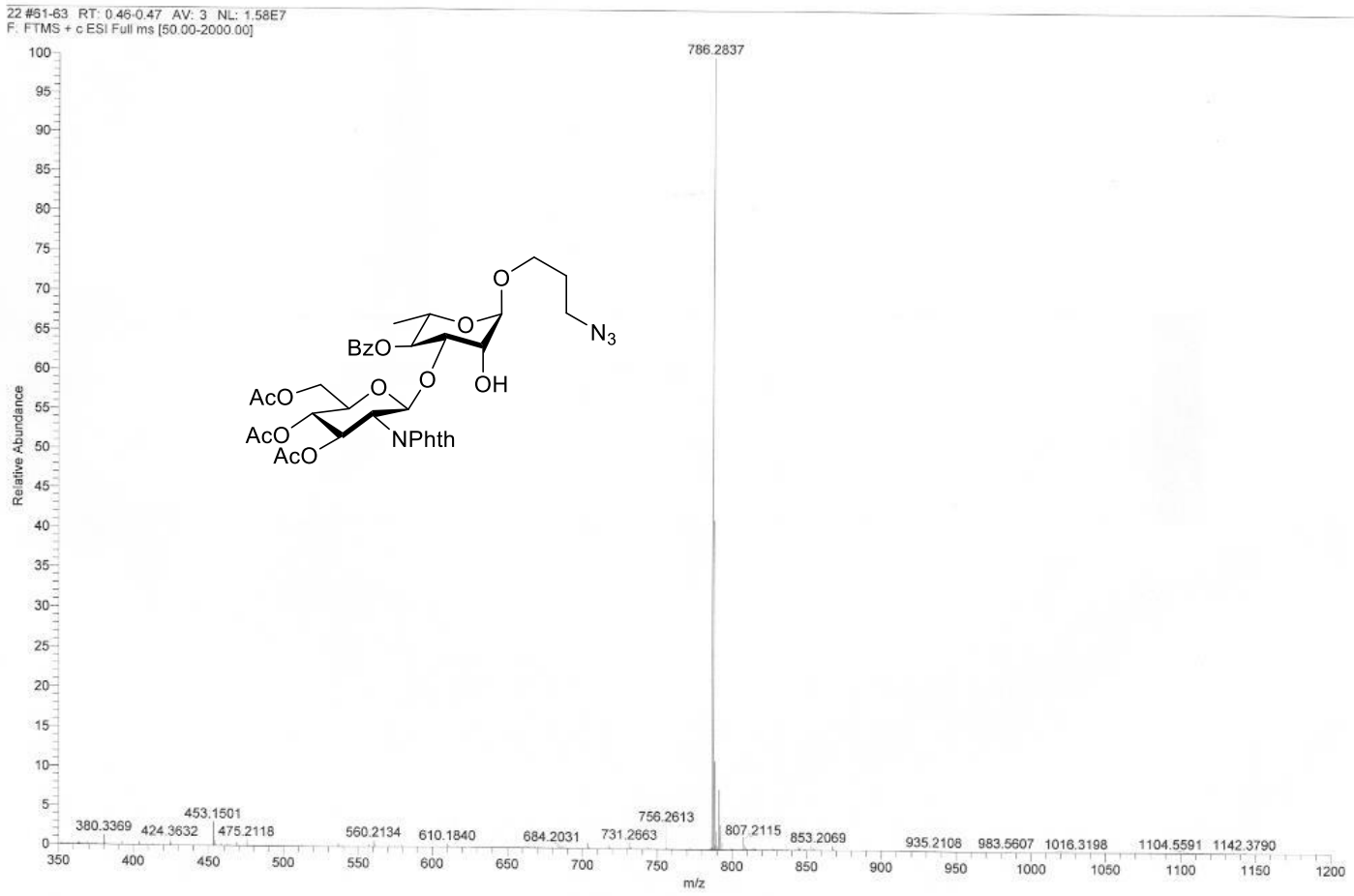
^{13}C NMR spectrum of compound **6** (150 MHz, $CDCl_3$)



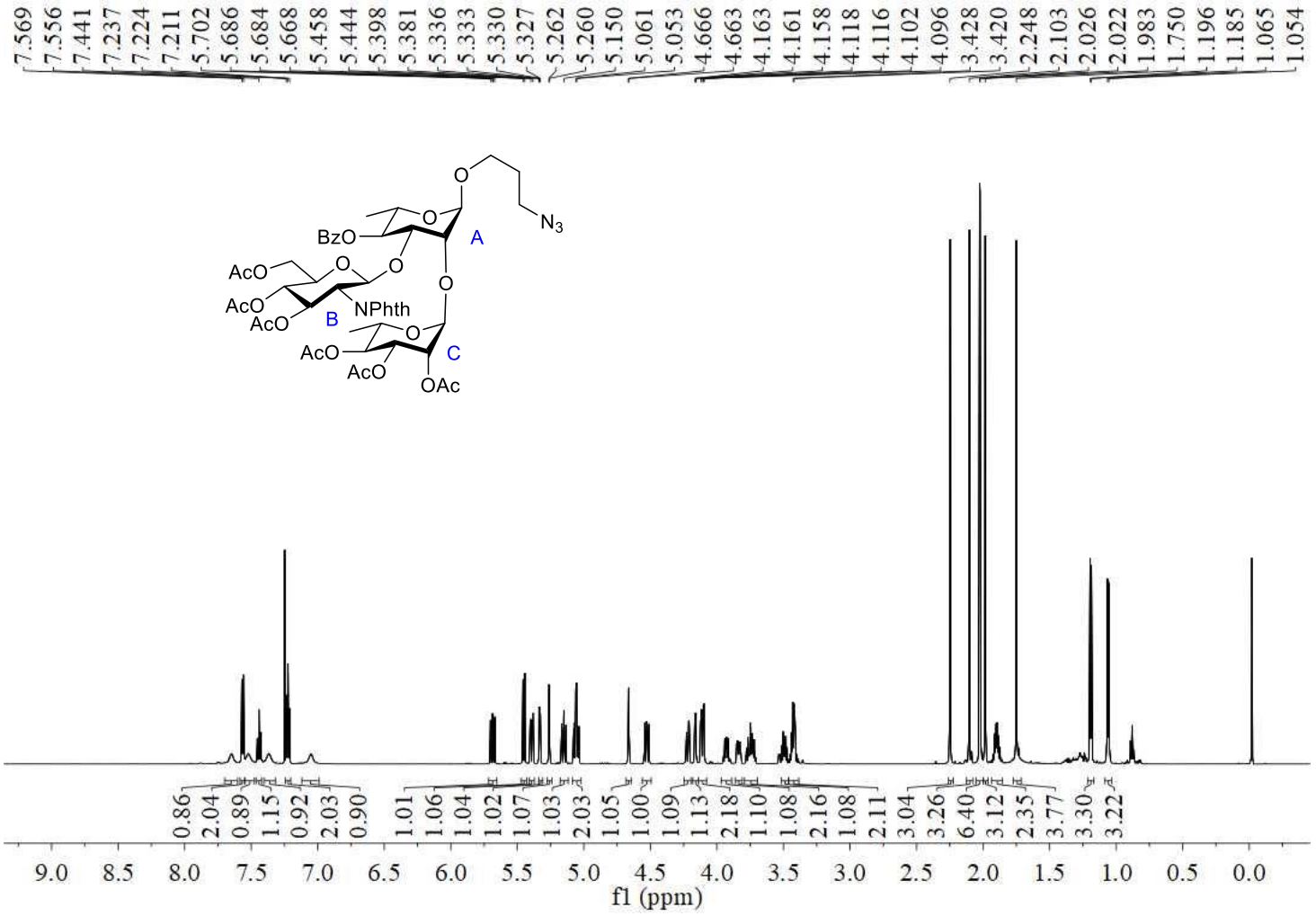
^{1}H-^{1}H COSY spectrum of compound **6** (600 MHz, $CDCl_3$)



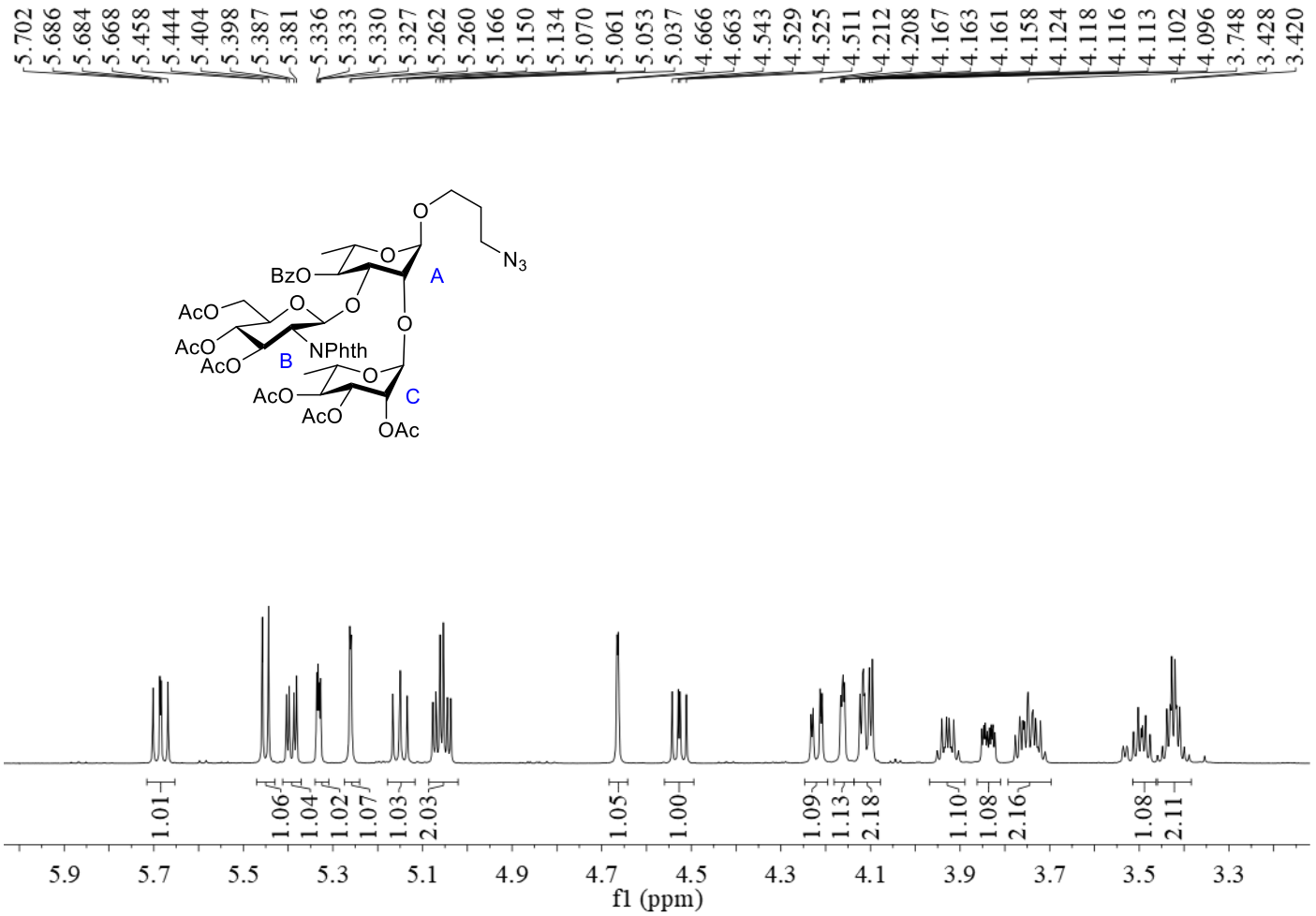
^{1}H-^{13}C HSQC spectrum of compound **6** (600/150 MHz, $CDCl_3$)



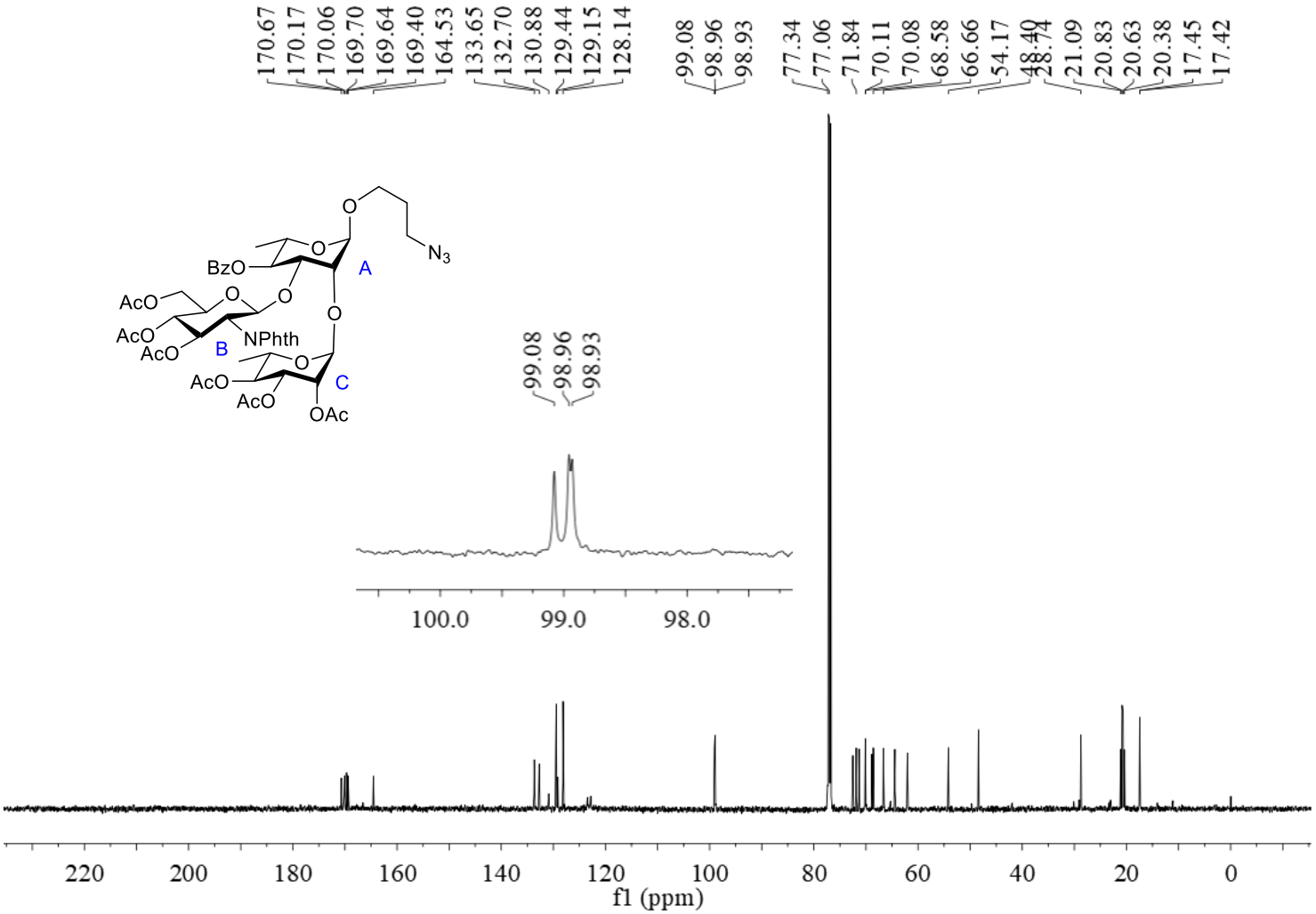
HR-ESI-(+) mass spectrum of compound **6**



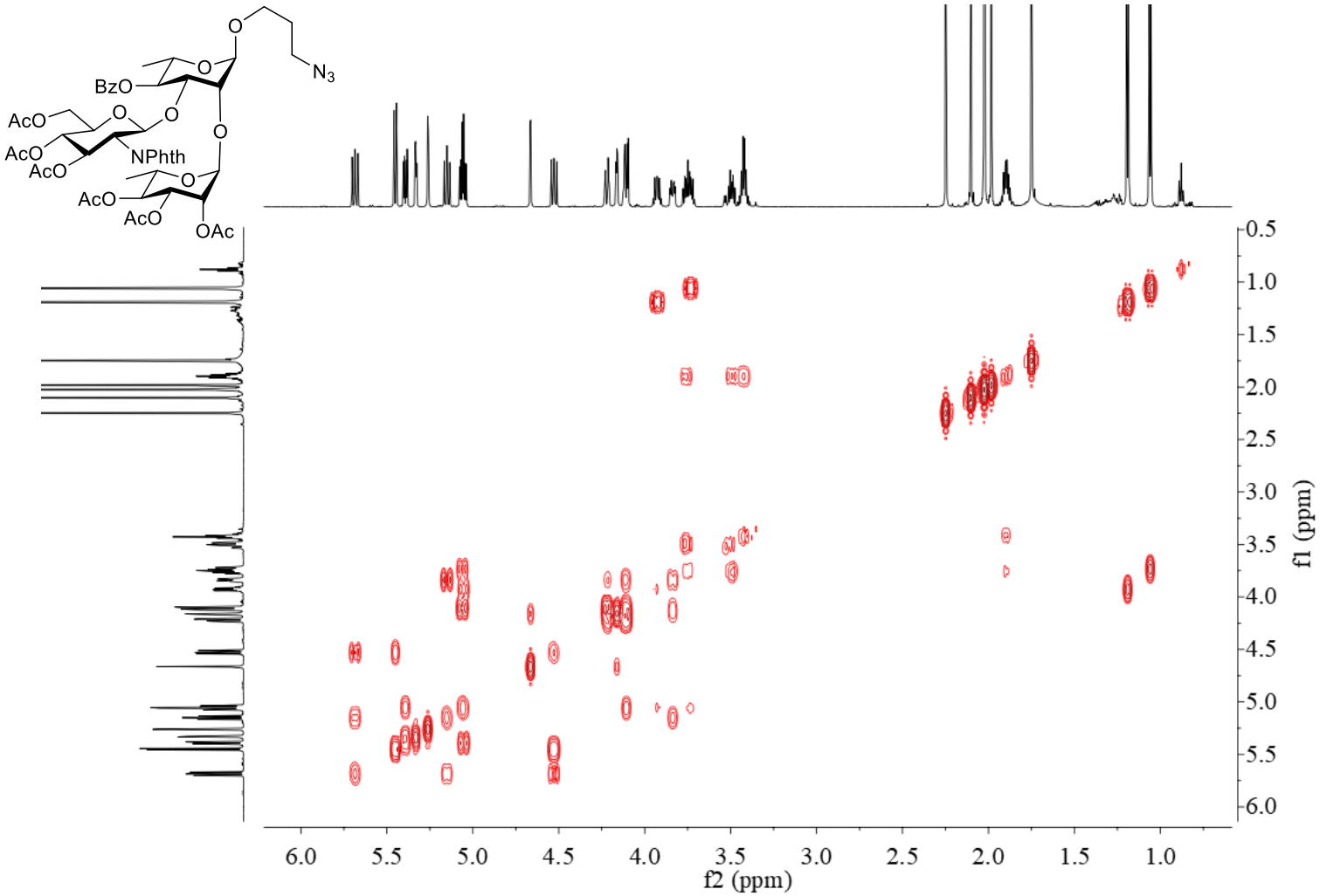
^{1}H NMR spectrum of compound **18** (600 MHz, $CDCl_3$)



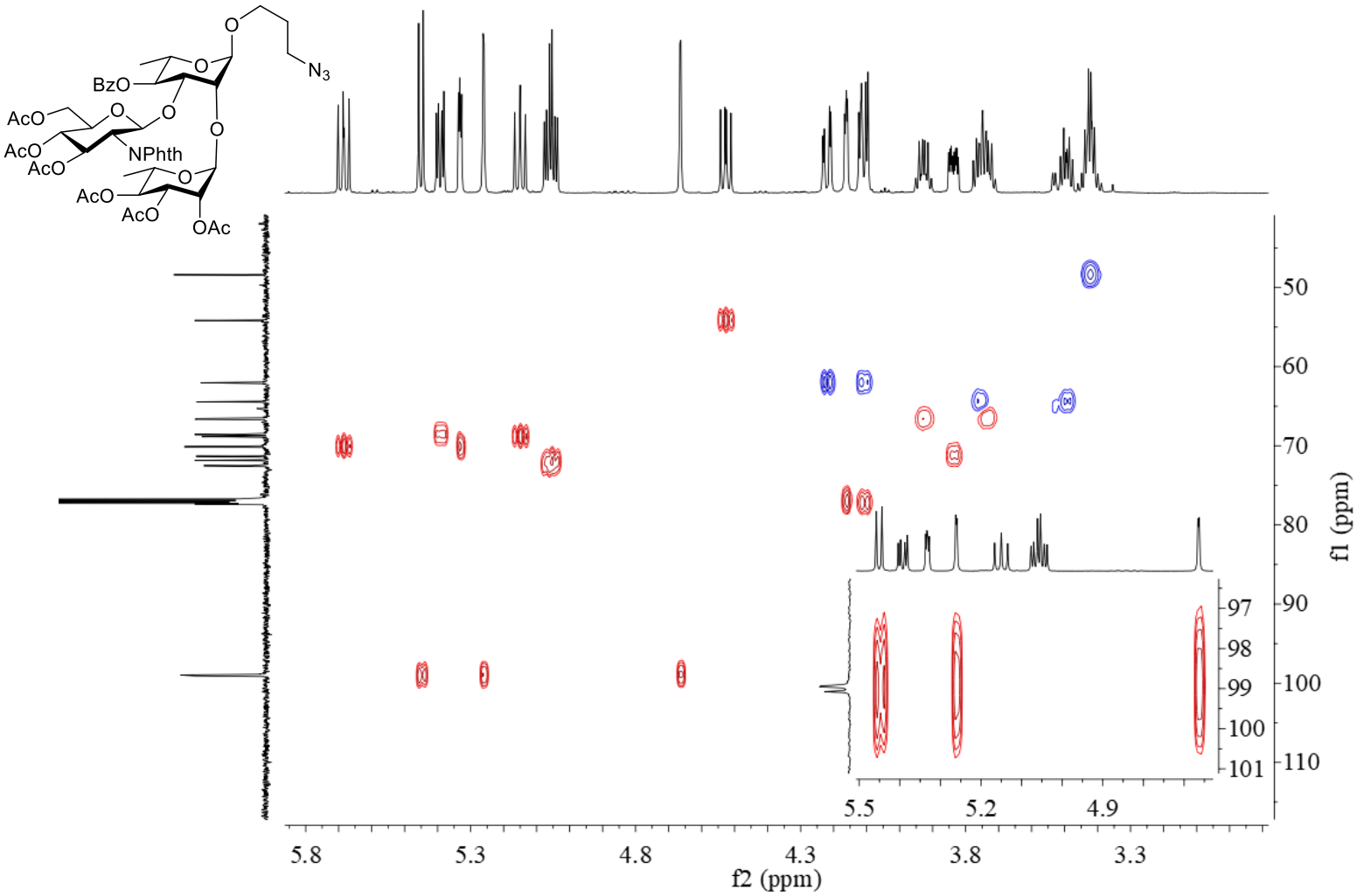
^{1}H NMR spectrum of compound **18** (expanded sugar region, 600 MHz, $CDCl_3$)



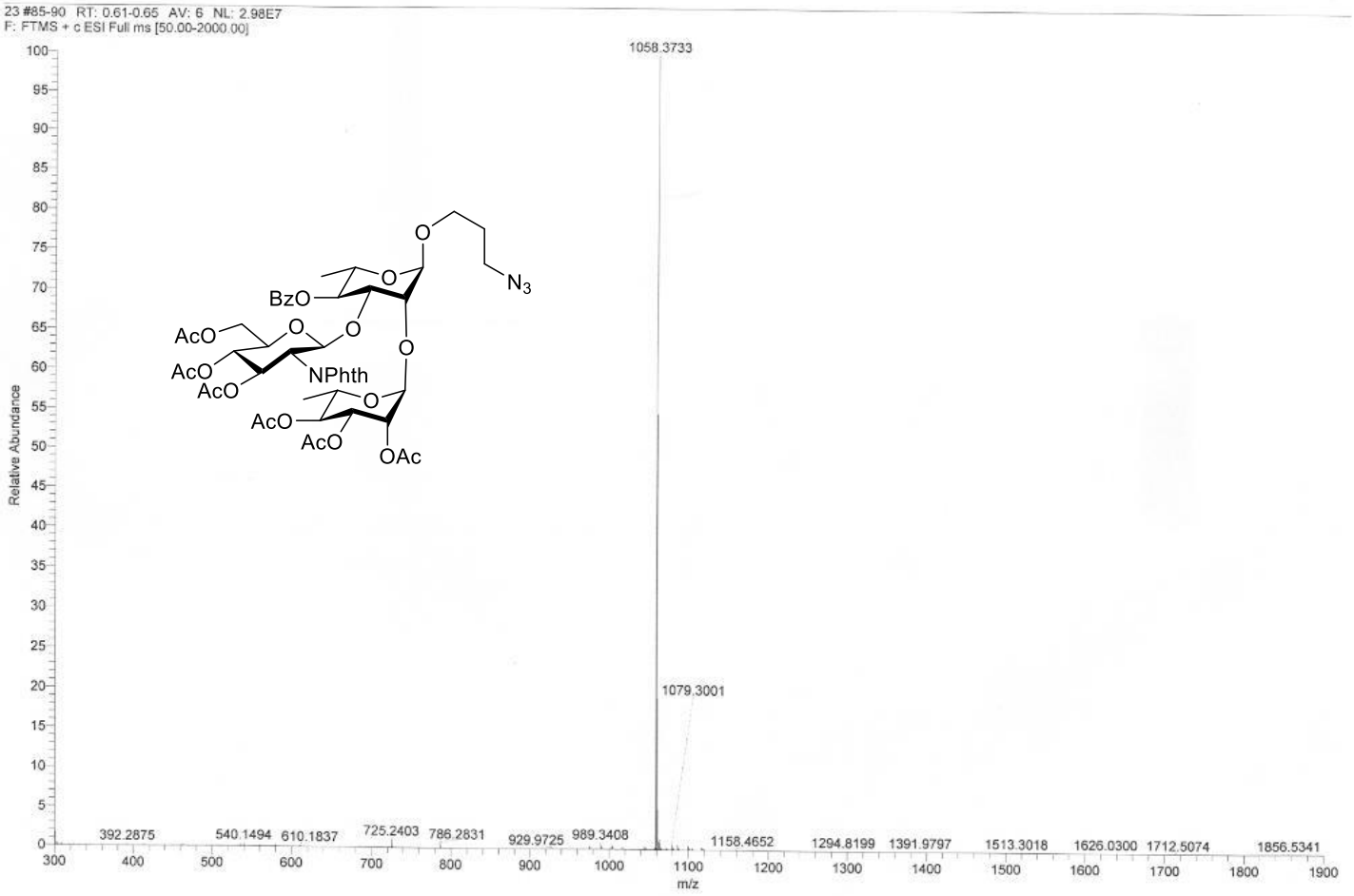
^{13}C NMR spectrum of compound **18** (150 MHz, $CDCl_3$)



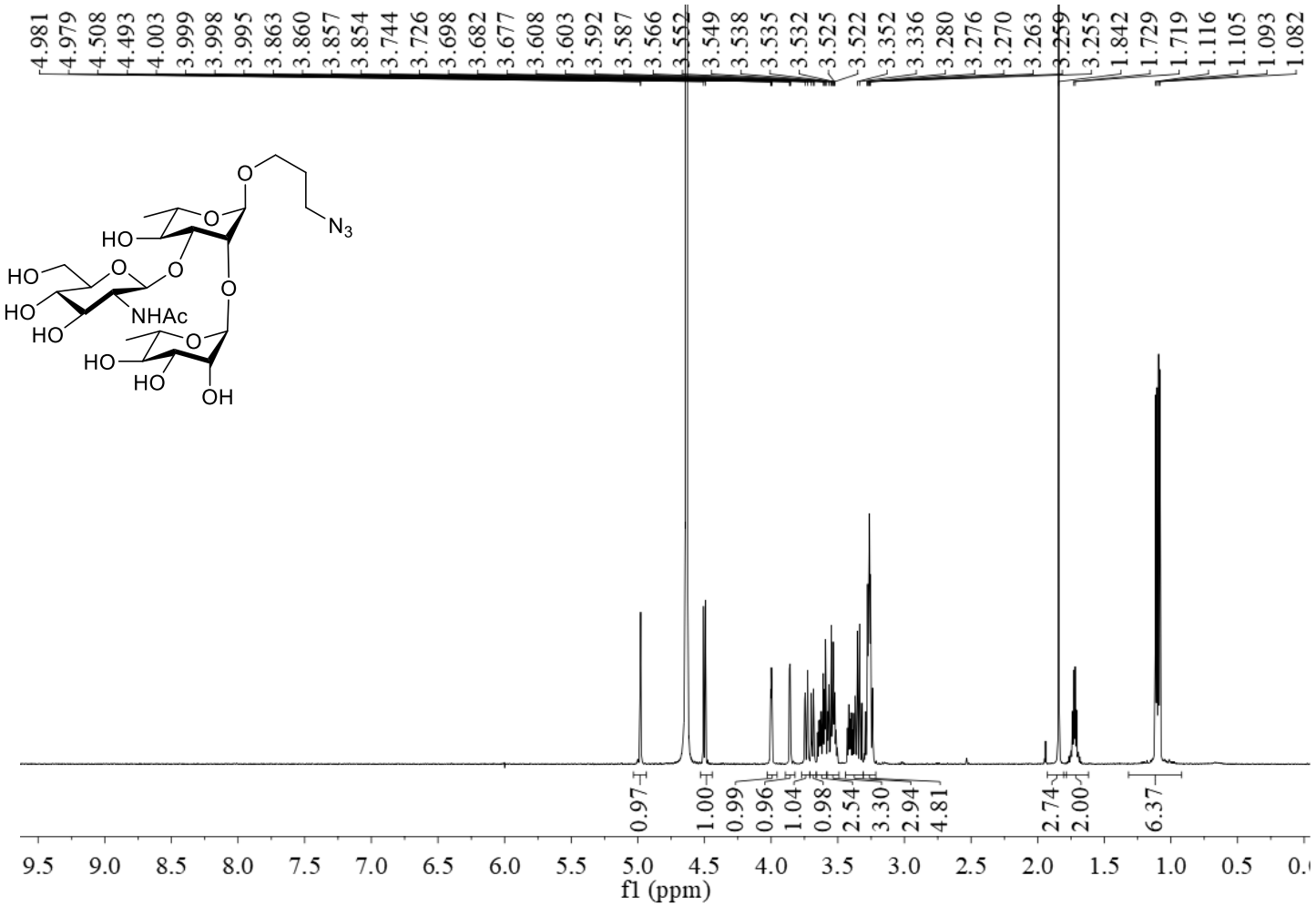
^{1}H-^{1}H COSY spectrum of compound **18** (600 MHz, $CDCl_3$)



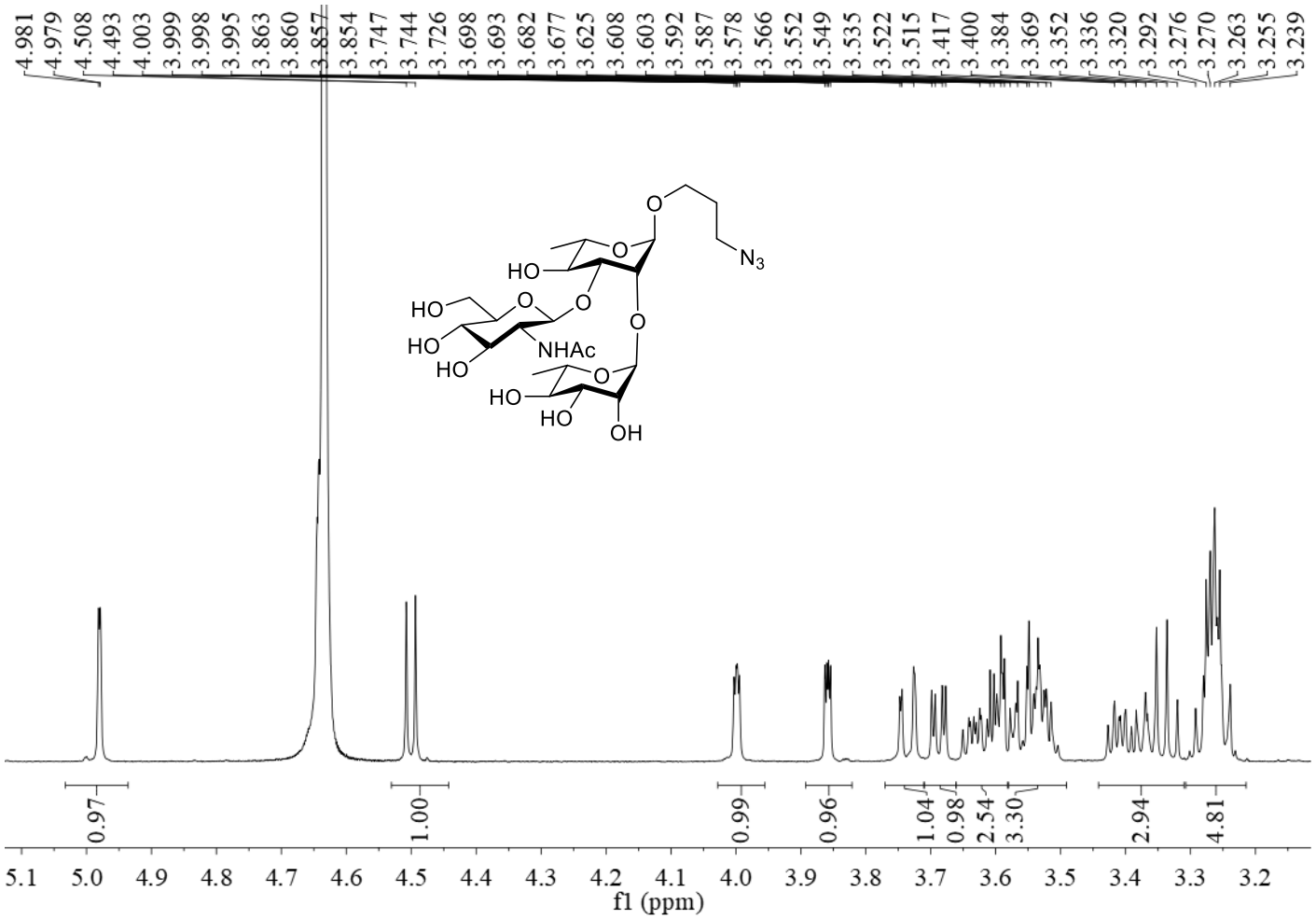
^{1}H-^{13}C HSQC spectrum of compound **18** (600/150 MHz, $CDCl_3$)



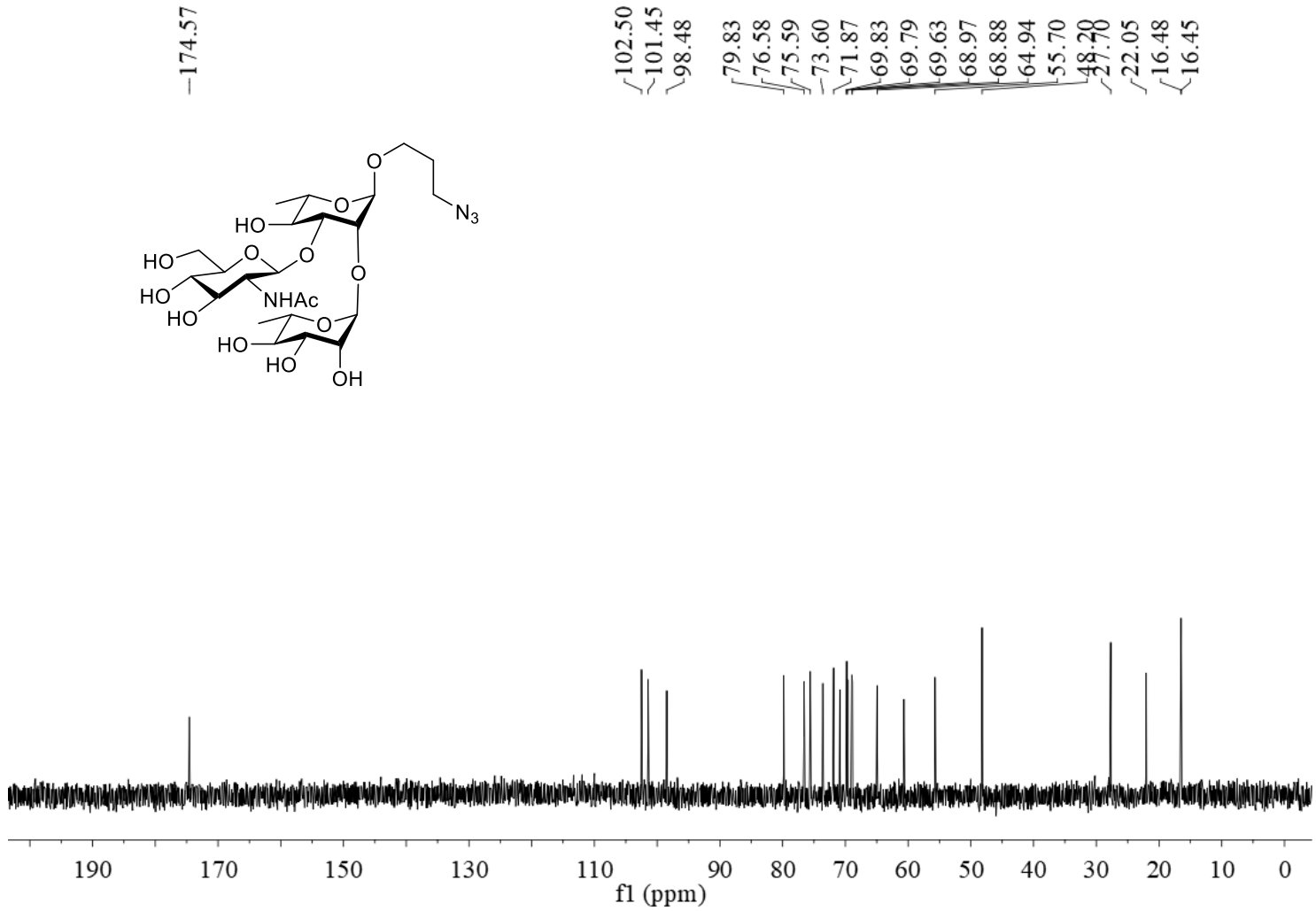
HR-ESI-(+) mass spectrum of compound **18**



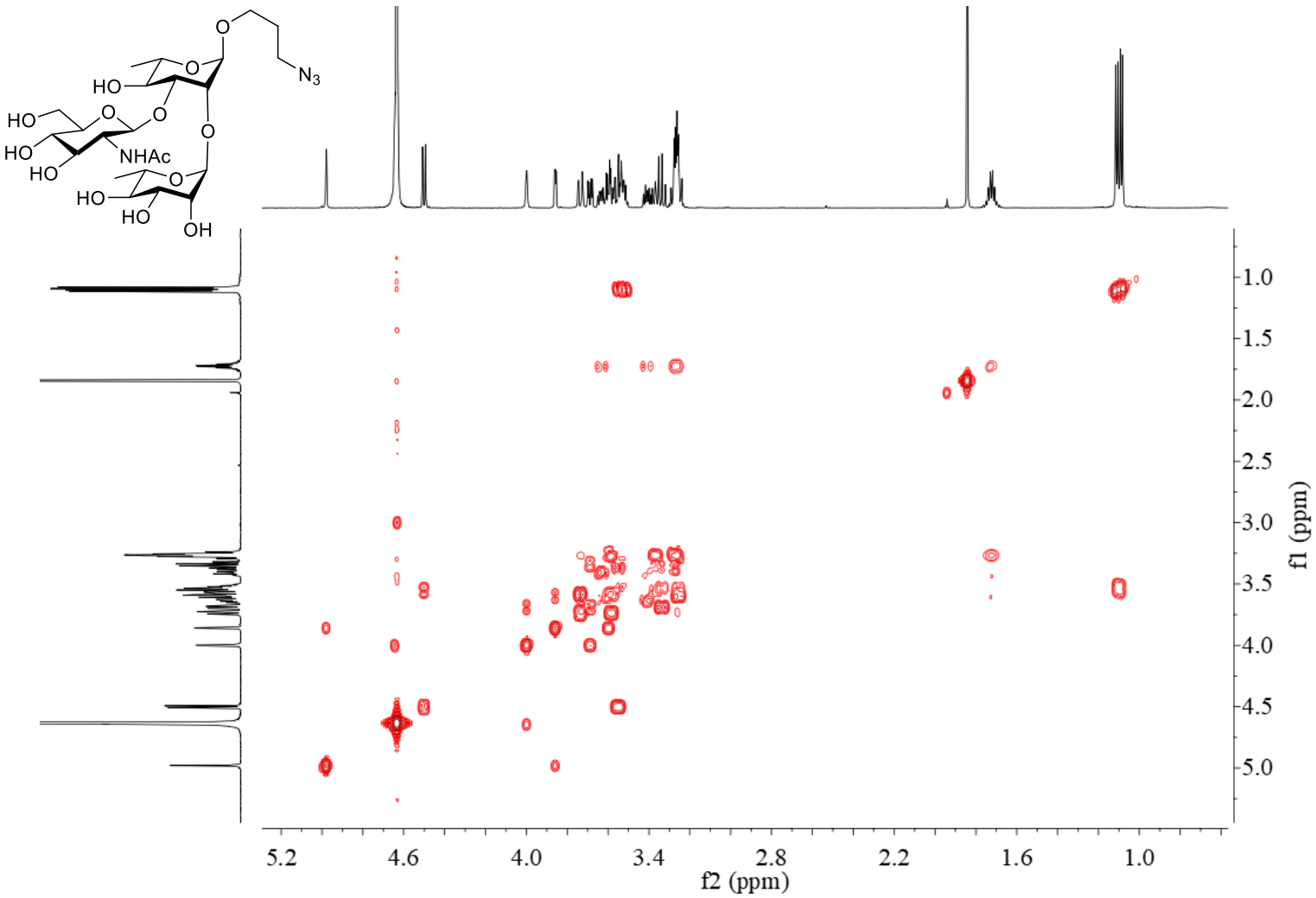
^{1}H NMR spectrum of compound **5a** (600 MHz, D_2O)



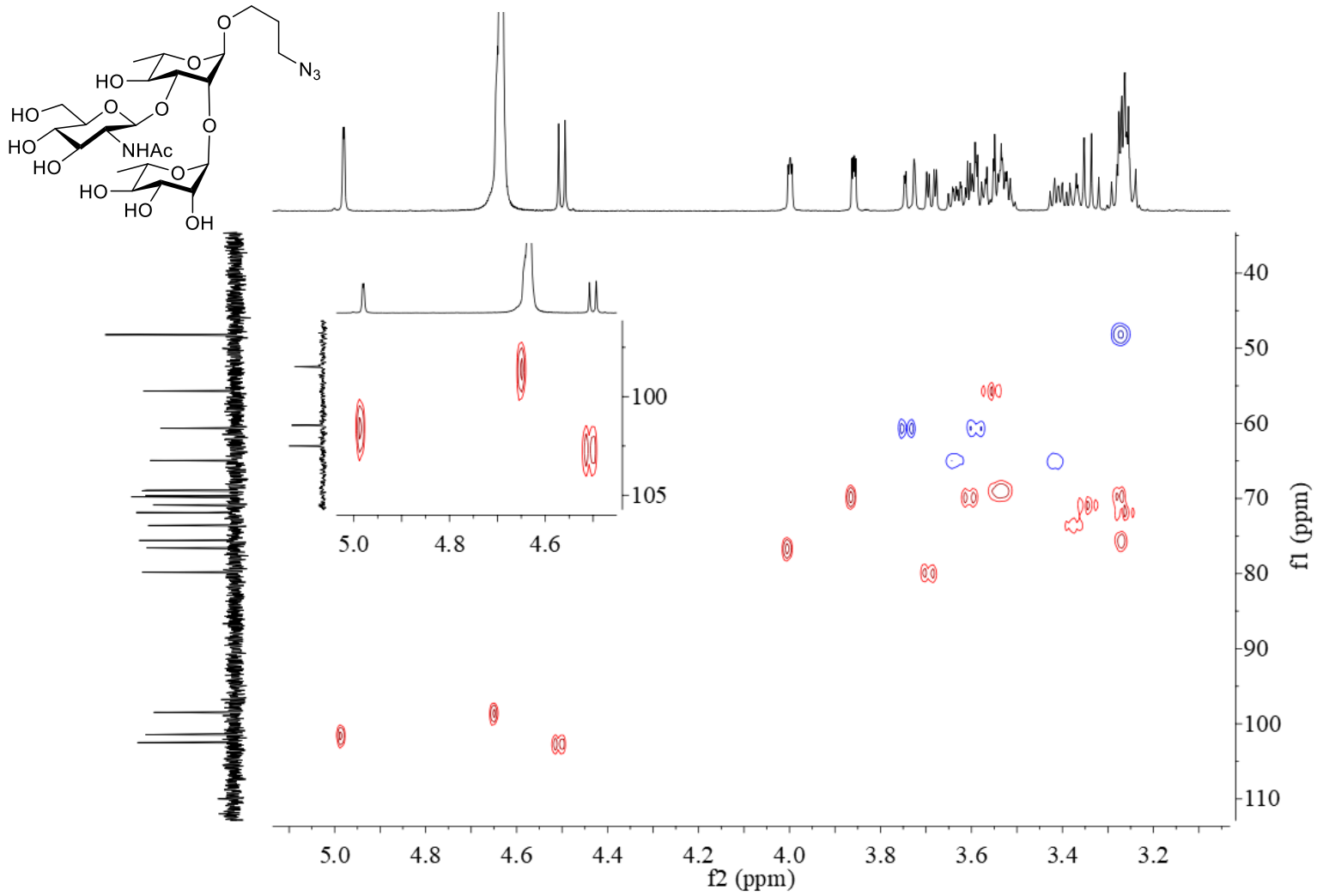
^{1}H NMR spectrum of compound **5a** (expanded sugar region, 600 MHz, D_2O)



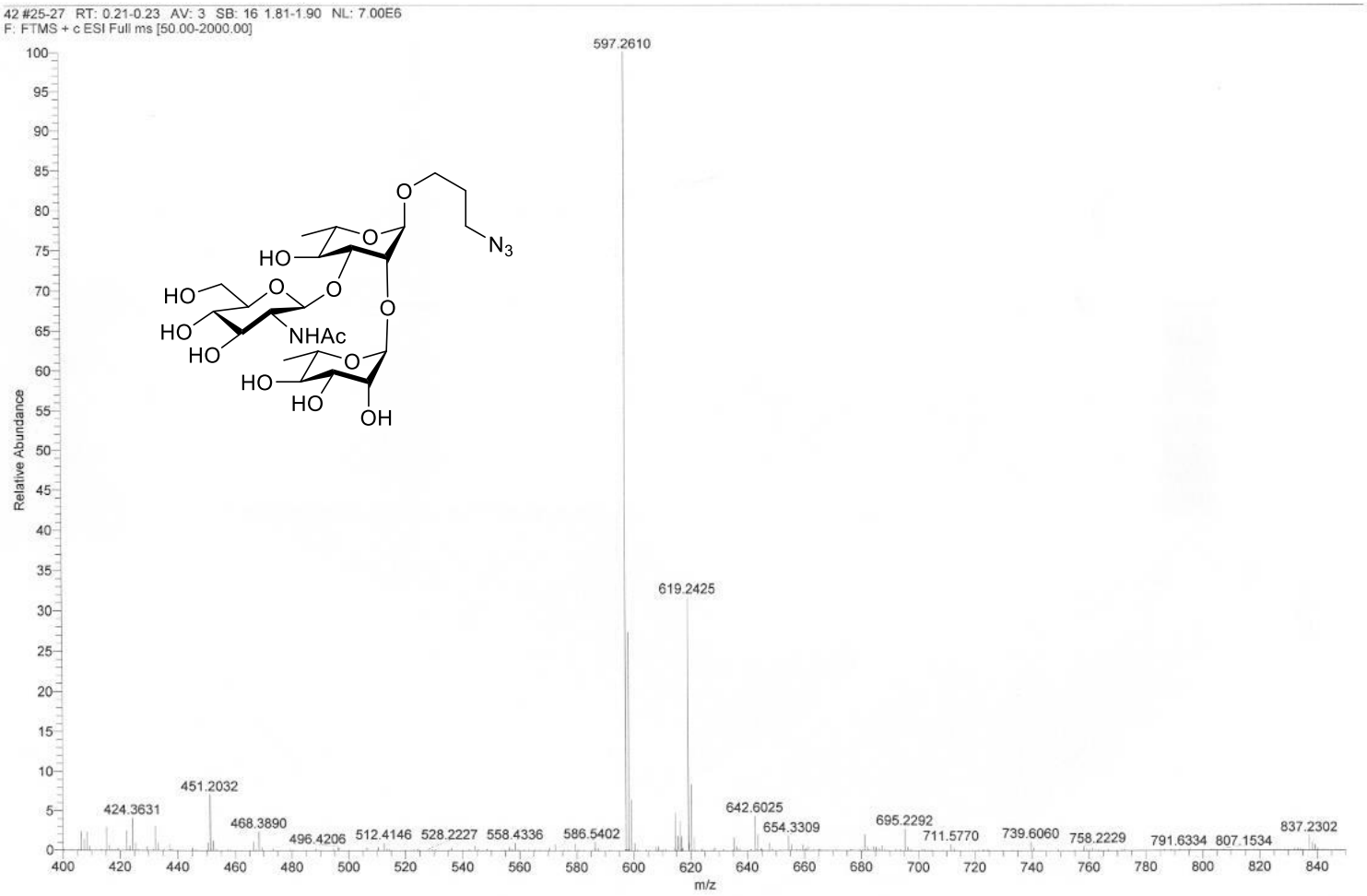
^{13}C NMR spectrum of compound **5a** (150 MHz, D_2O)



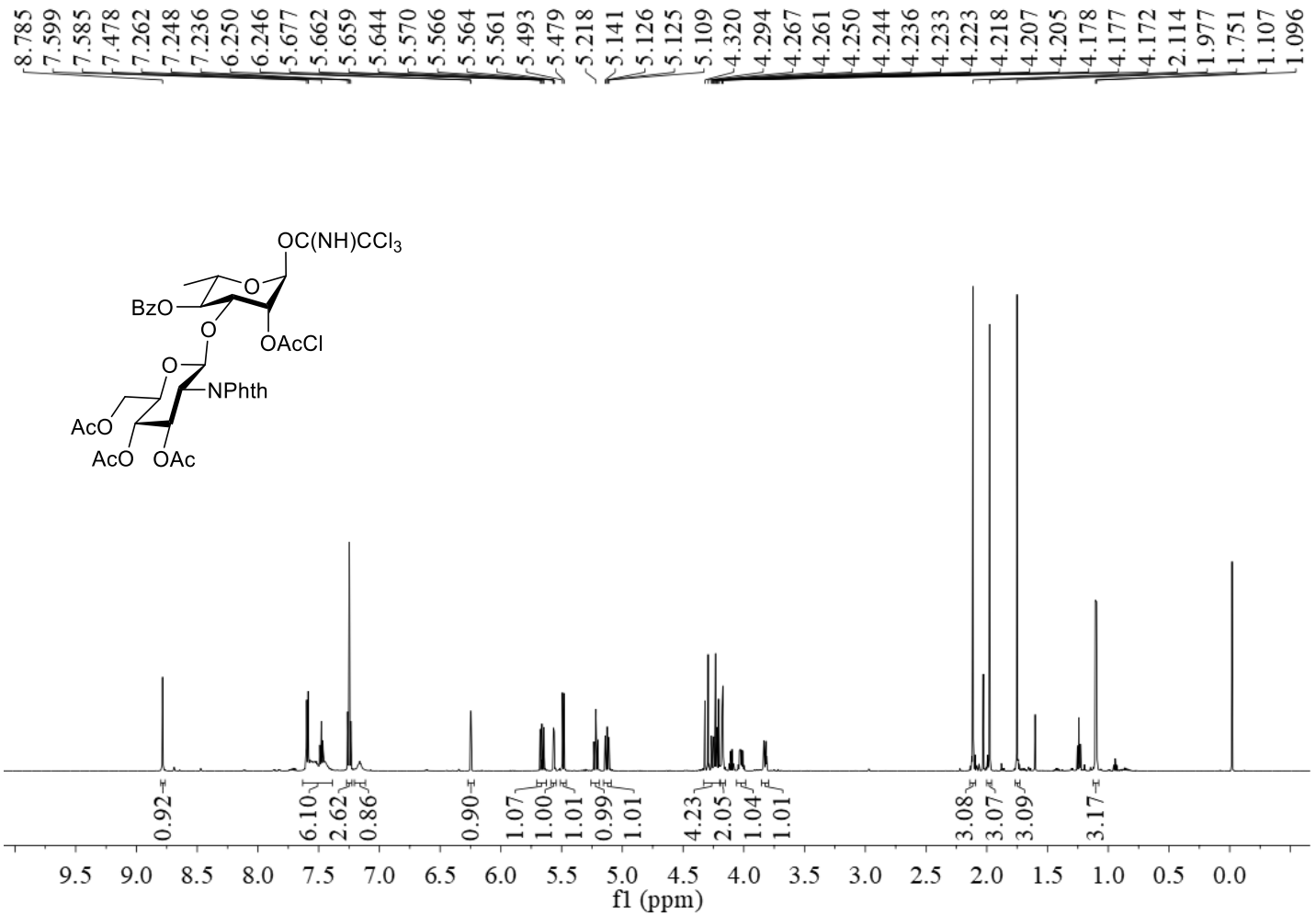
^{1}H-^{1}H COSY spectrum of compound **5a** (600 MHz, D_2O)



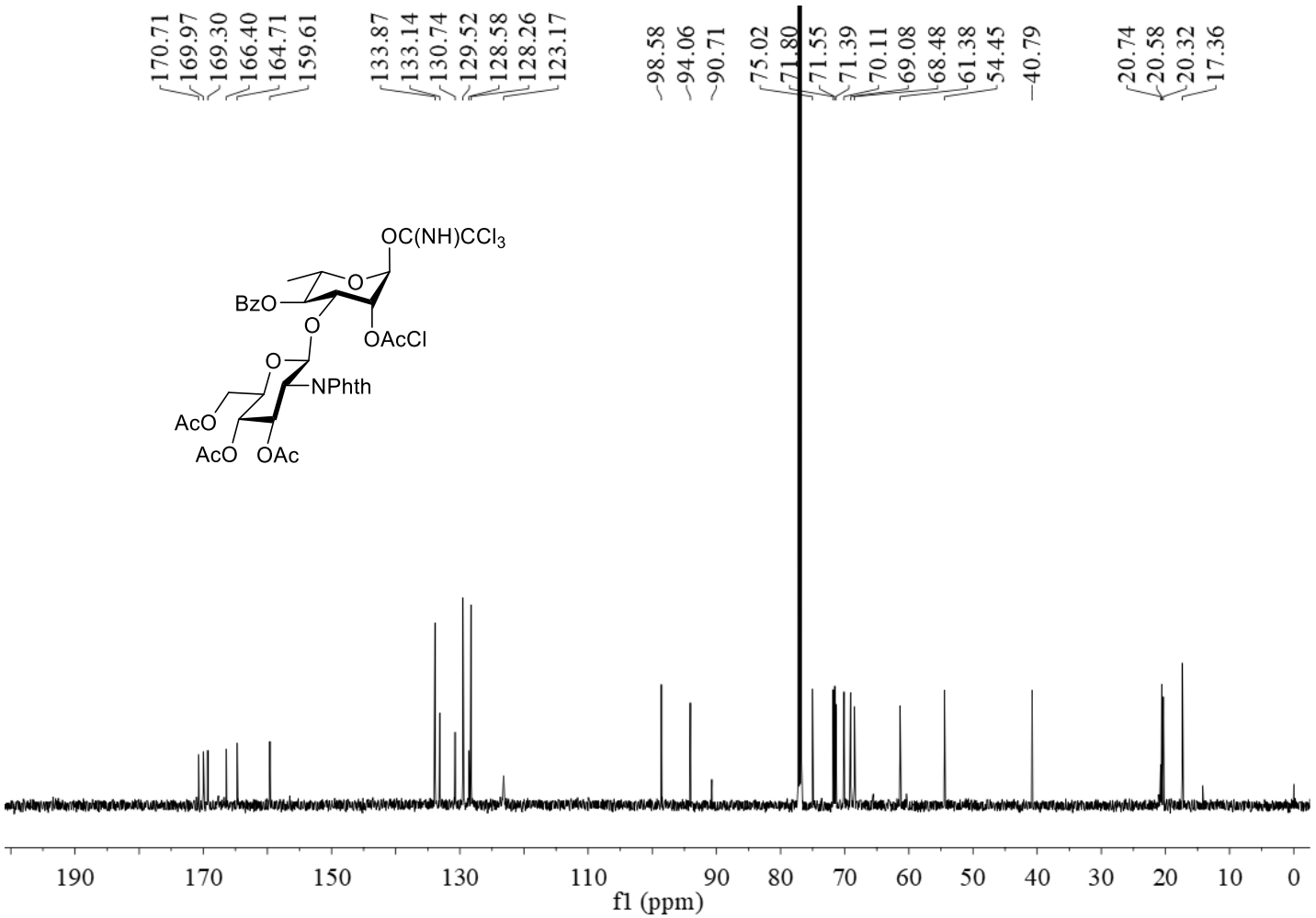
^{1}H-^{13}C HSQC spectrum of compound **5a** (600/150 MHz, D_2O)



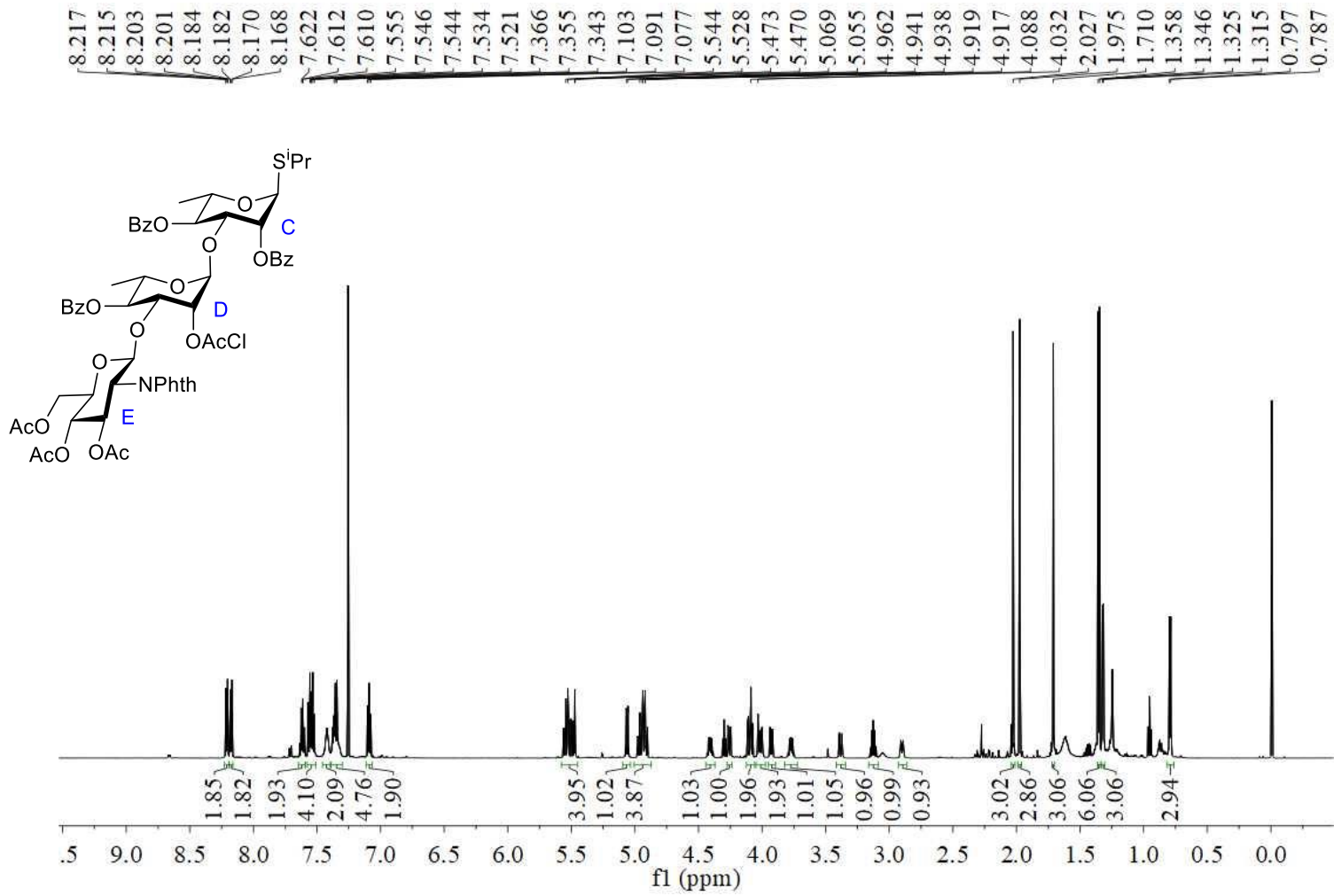
HR-ESI-(+) mass spectrum of compound **5a**



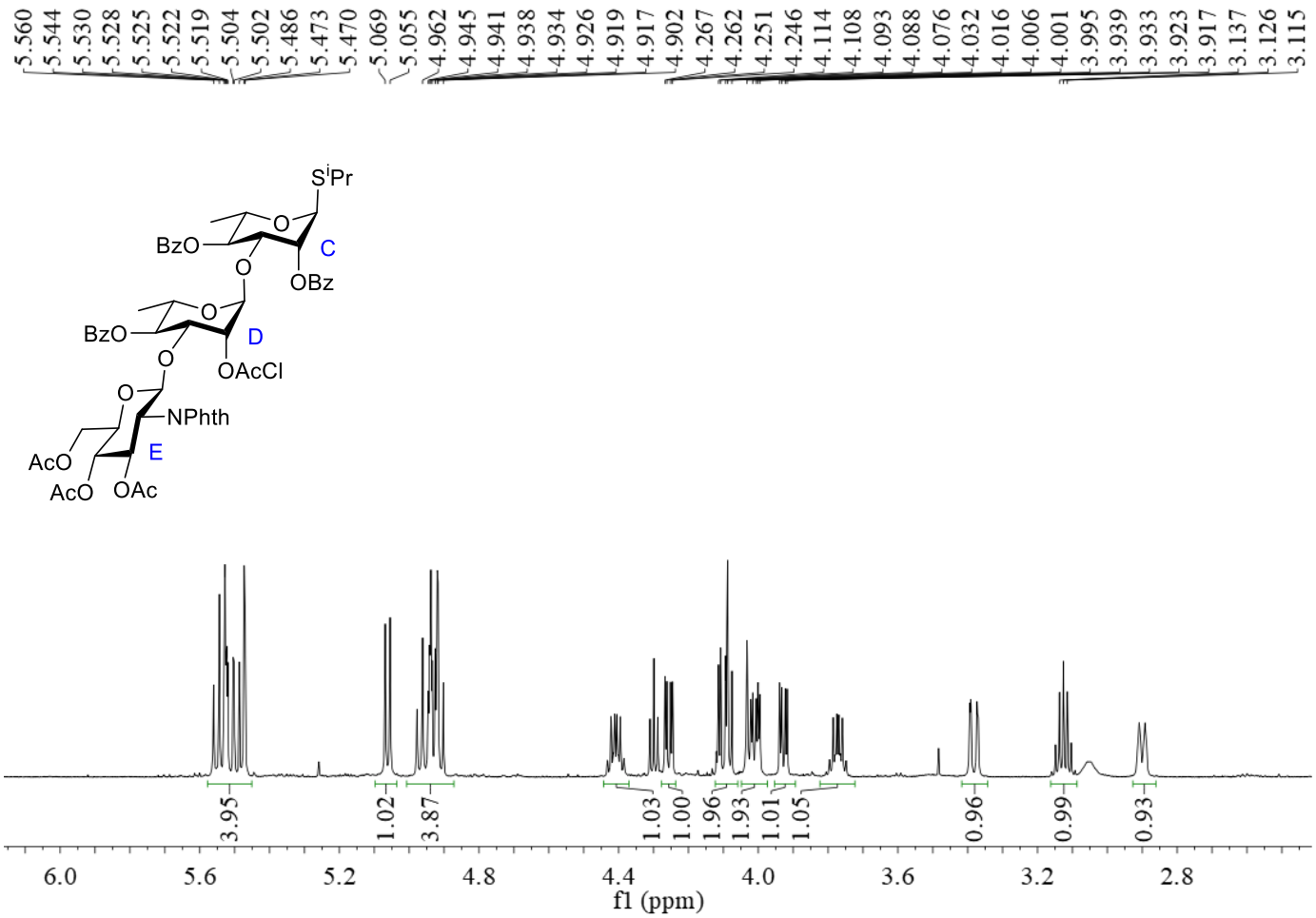
^{1}H NMR spectrum of compound **19** (600 MHz, $CDCl_3$)



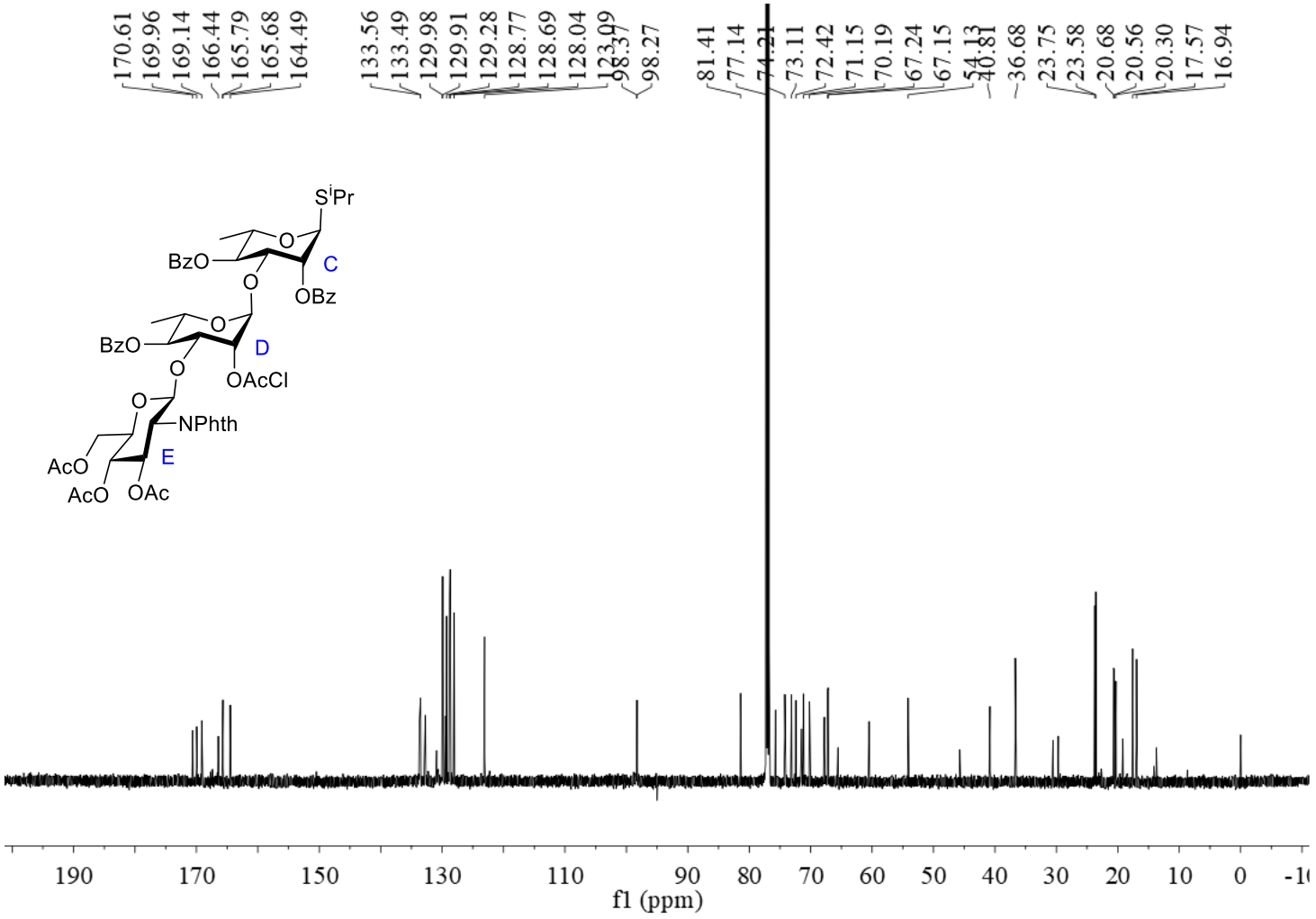
^{13}C NMR spectrum of compound **19** (150 MHz, $CDCl_3$)



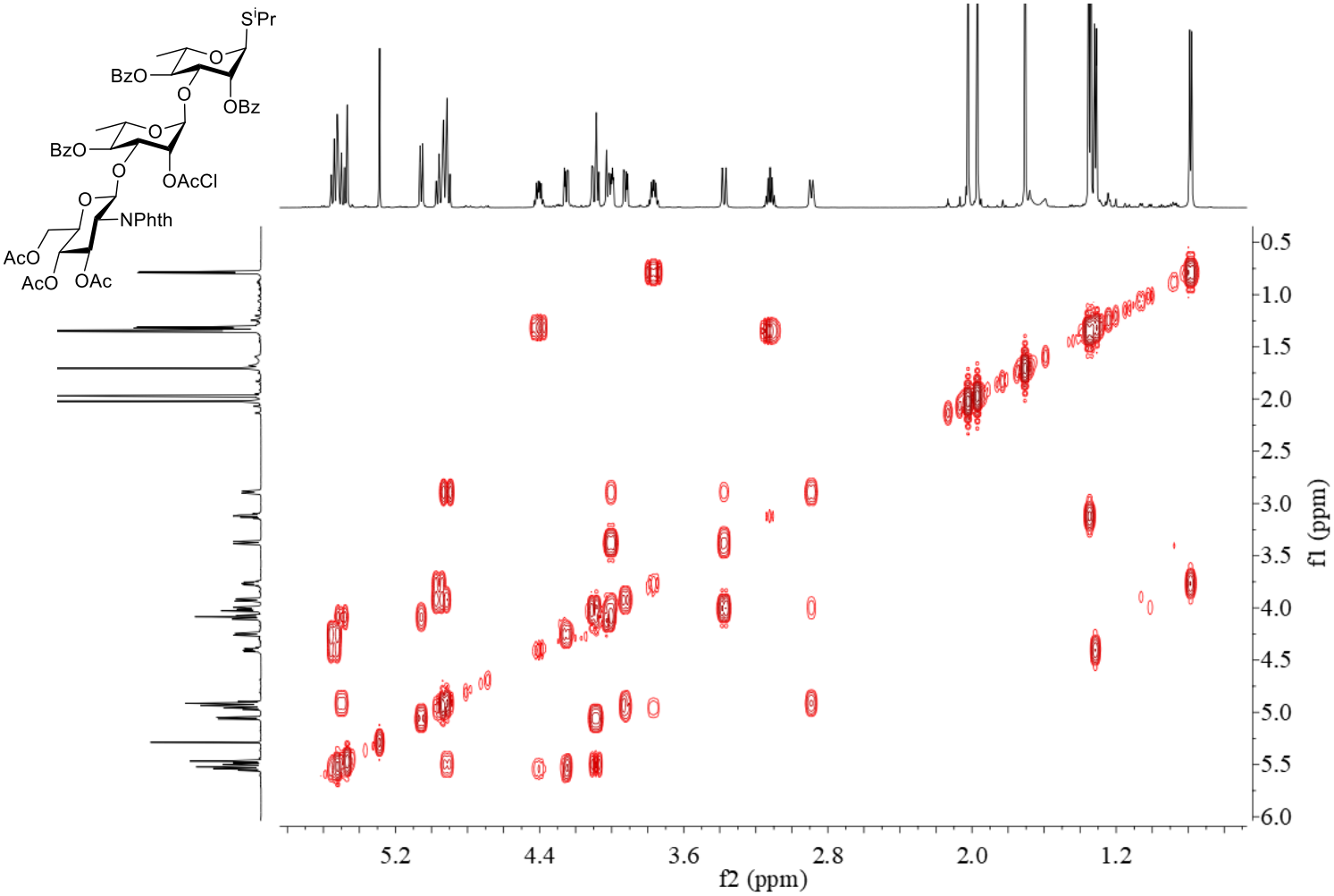
^{1}H NMR spectrum of compound **7** (600 MHz, $CDCl_3$)



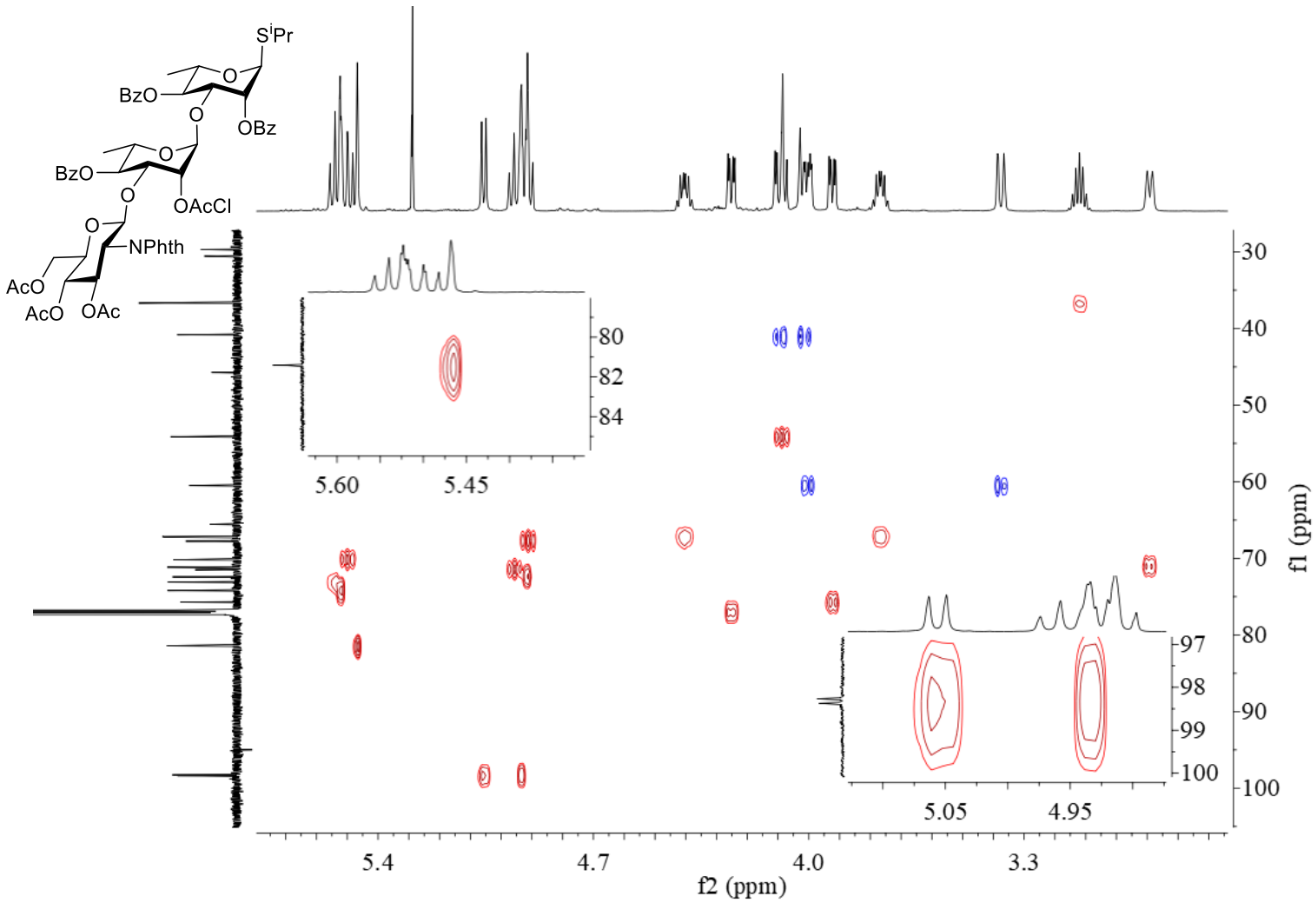
^{1}H NMR spectrum of compound **7** (expanded sugar region, 600 MHz, $CDCl_3$)



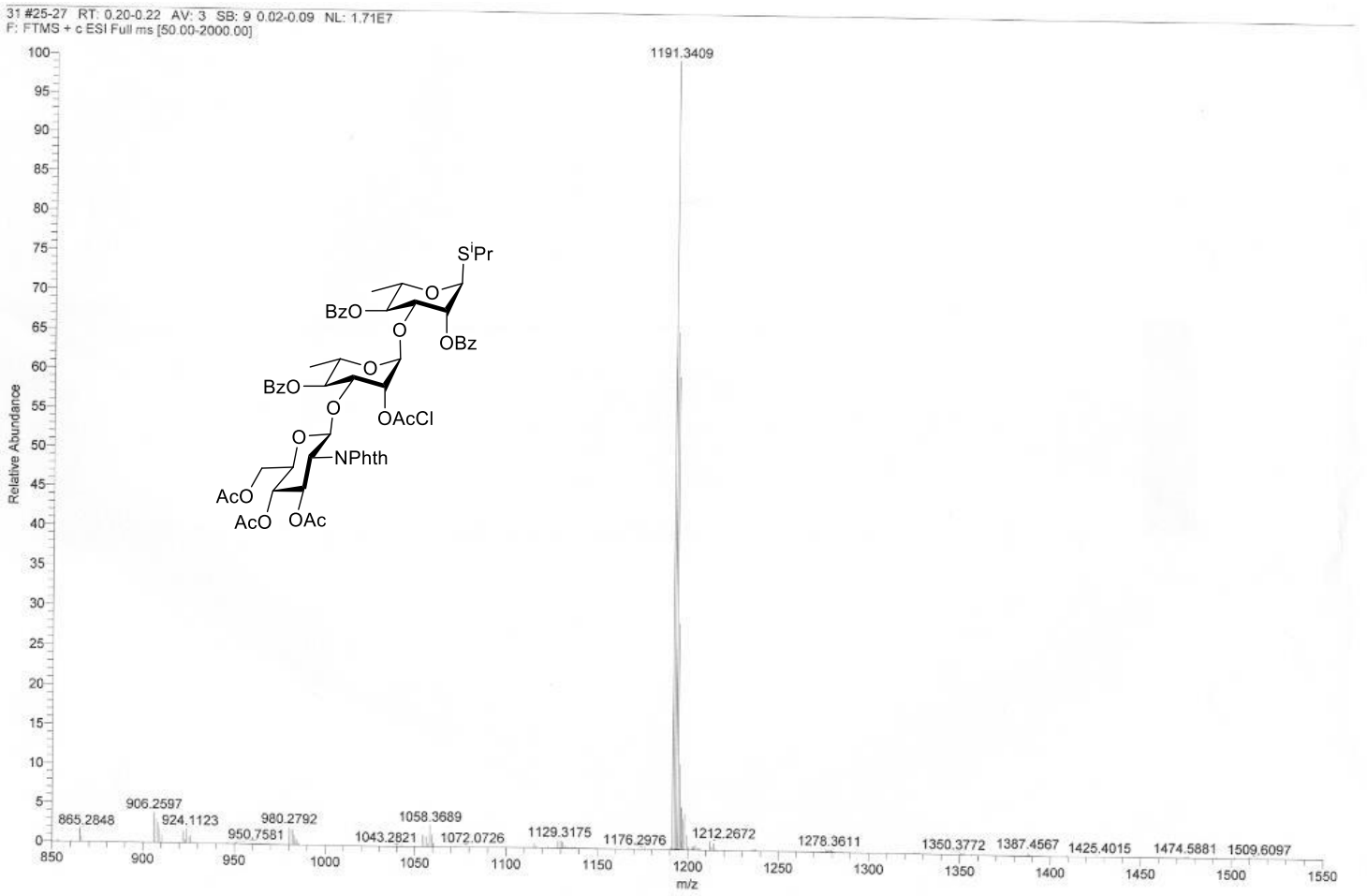
^{13}C NMR spectrum of compound **7** (150 MHz, $CDCl_3$)



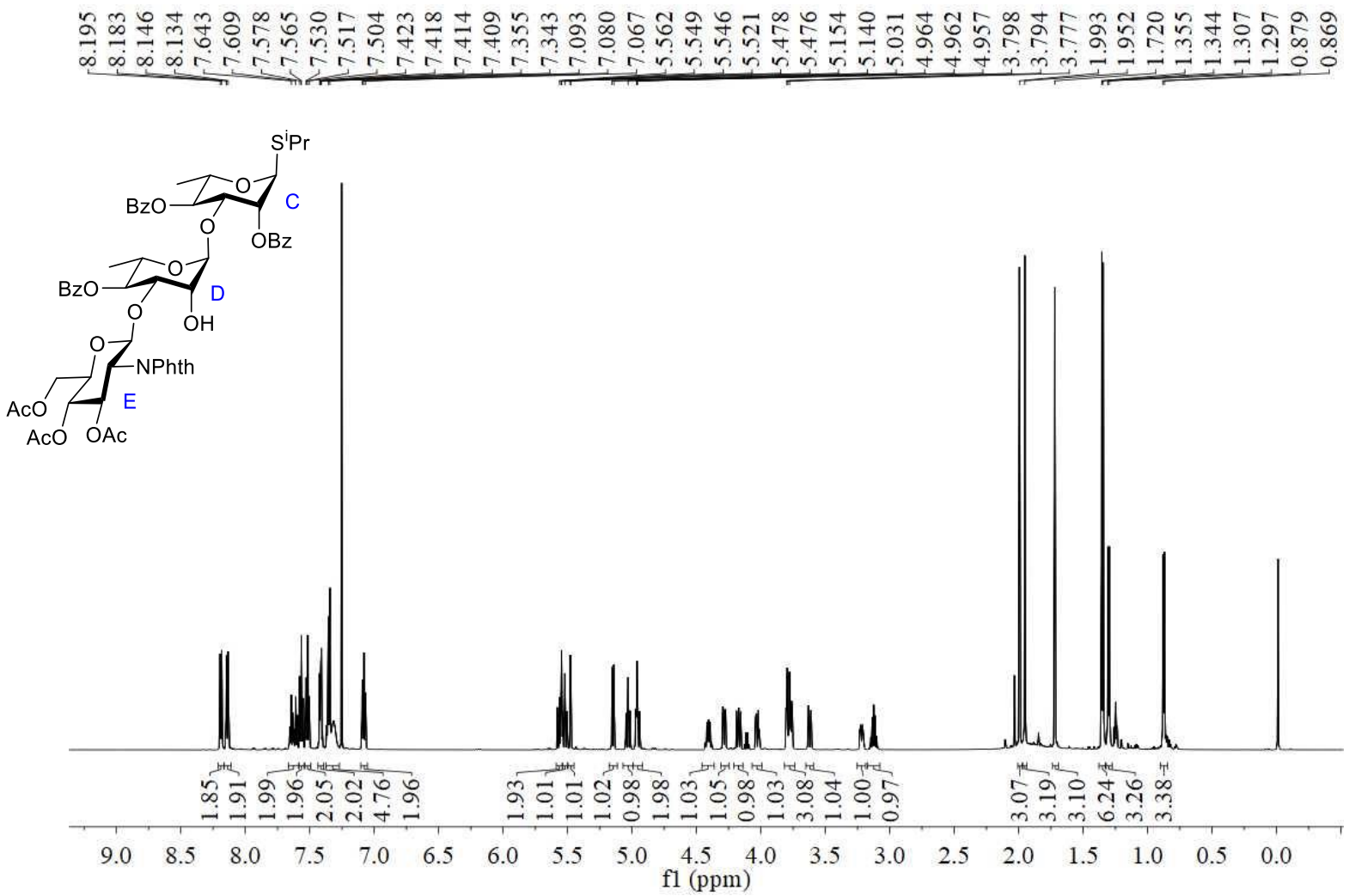
^{1}H-^{1}H COSY spectrum of compound **7** (600 MHz, $CDCl_3$)



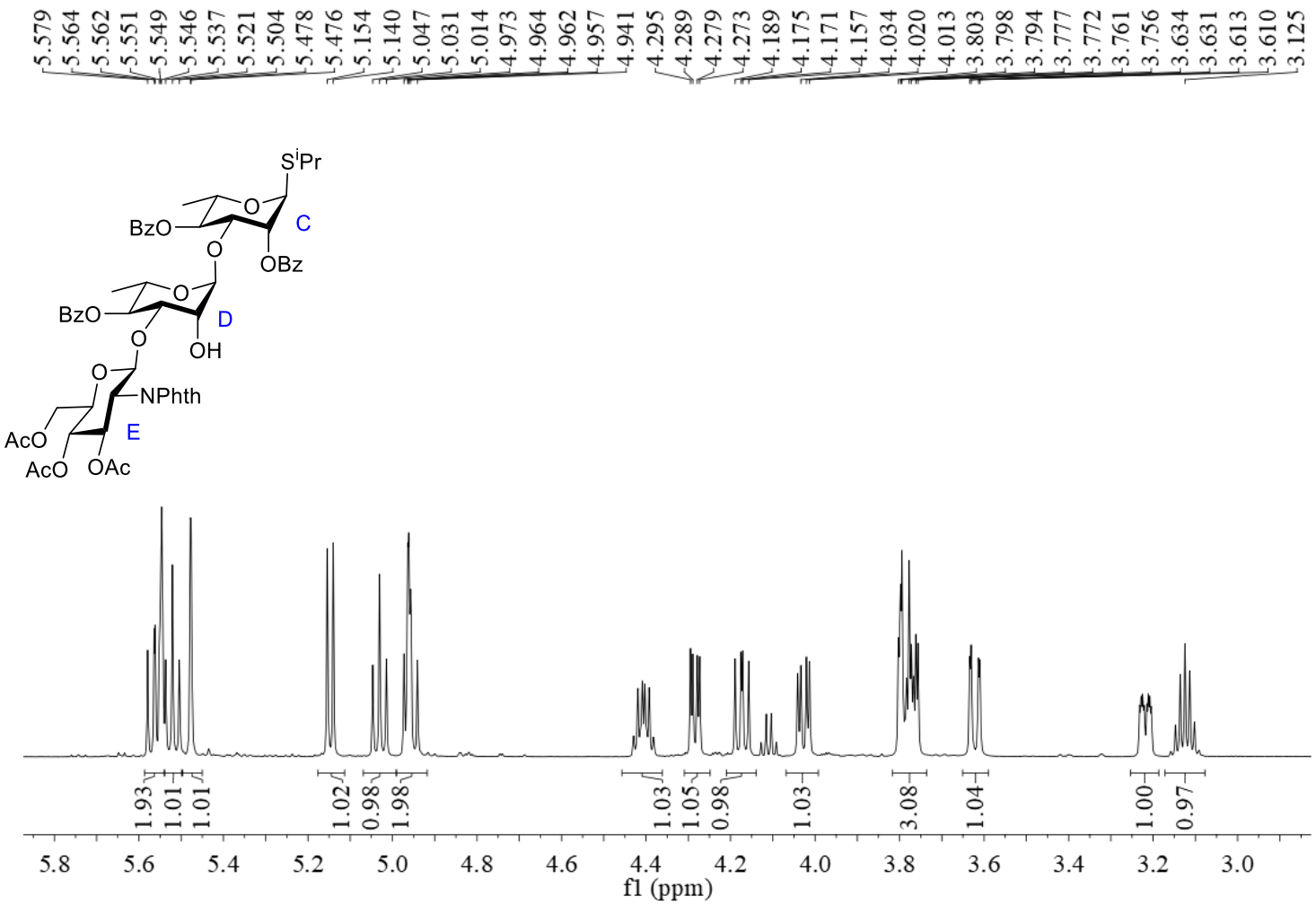
^{1}H-^{13}C HSQC spectrum of compound **7** (600/150 MHz, $CDCl_3$)



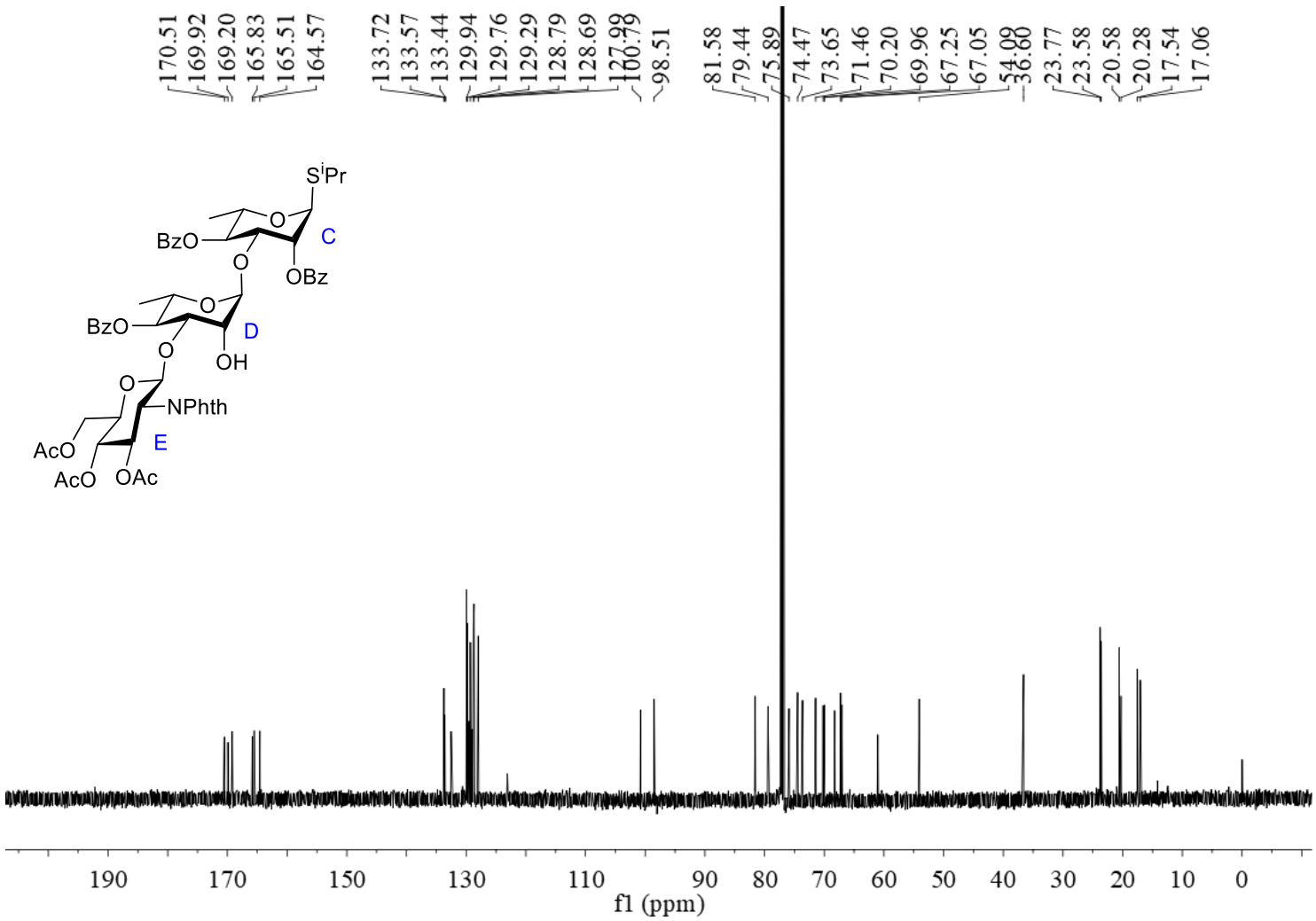
HR-ESI-(+) mass spectrum of compound **7**



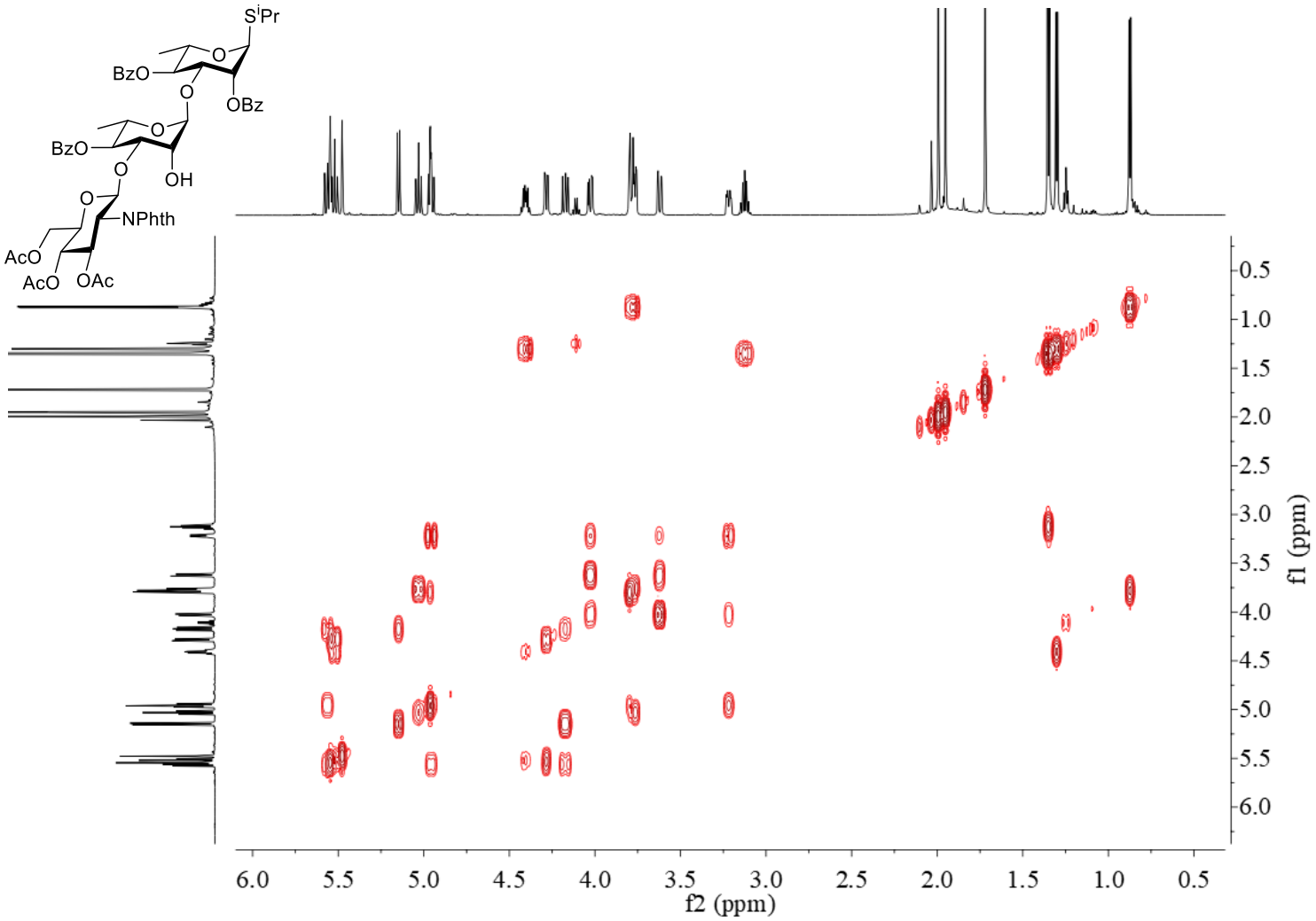
^{1}H NMR spectrum of compound **20** (600 MHz, $CDCl_3$)



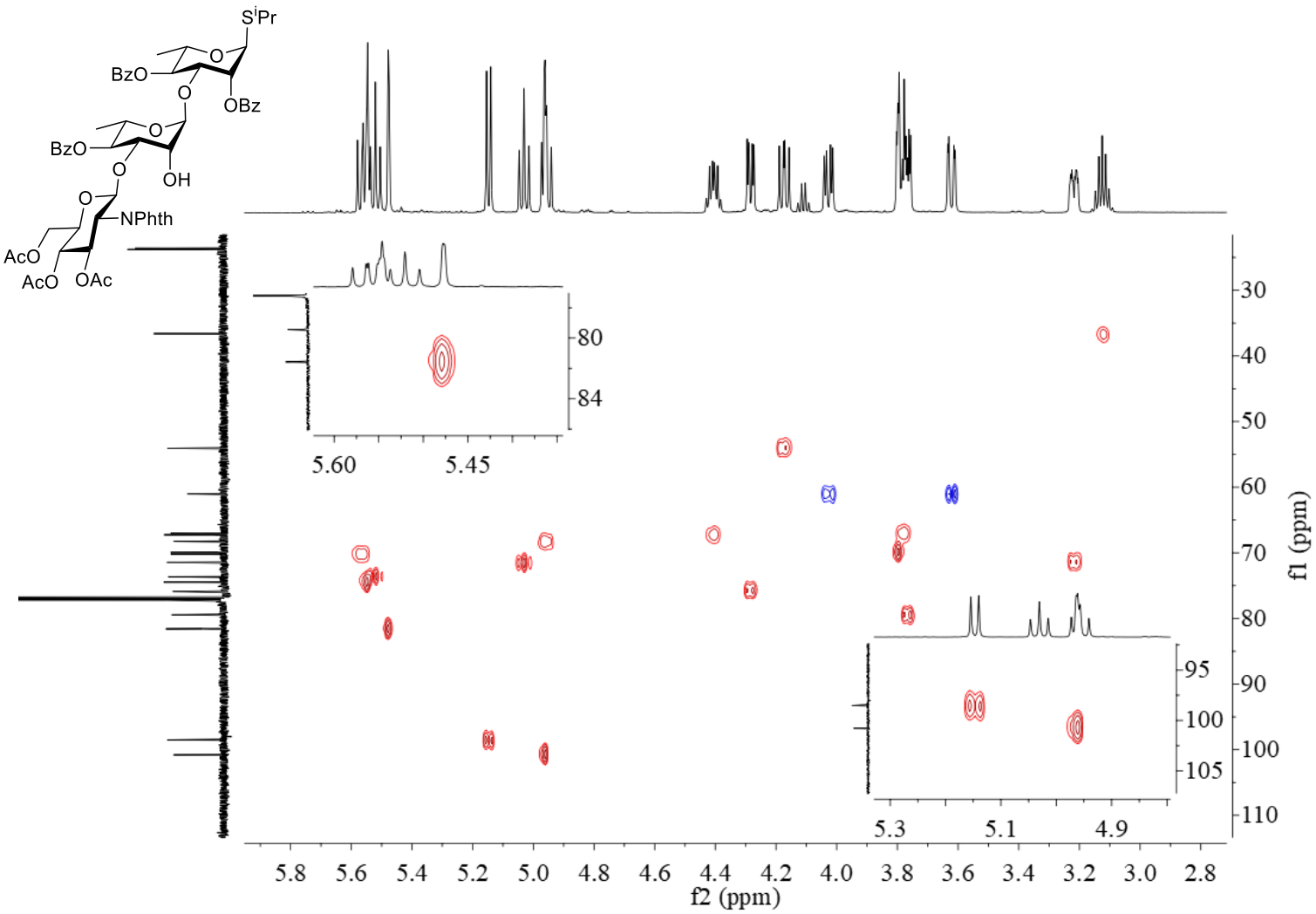
^{1}H NMR spectrum of compound **20** (expanded sugar region, 600 MHz, $CDCl_3$)



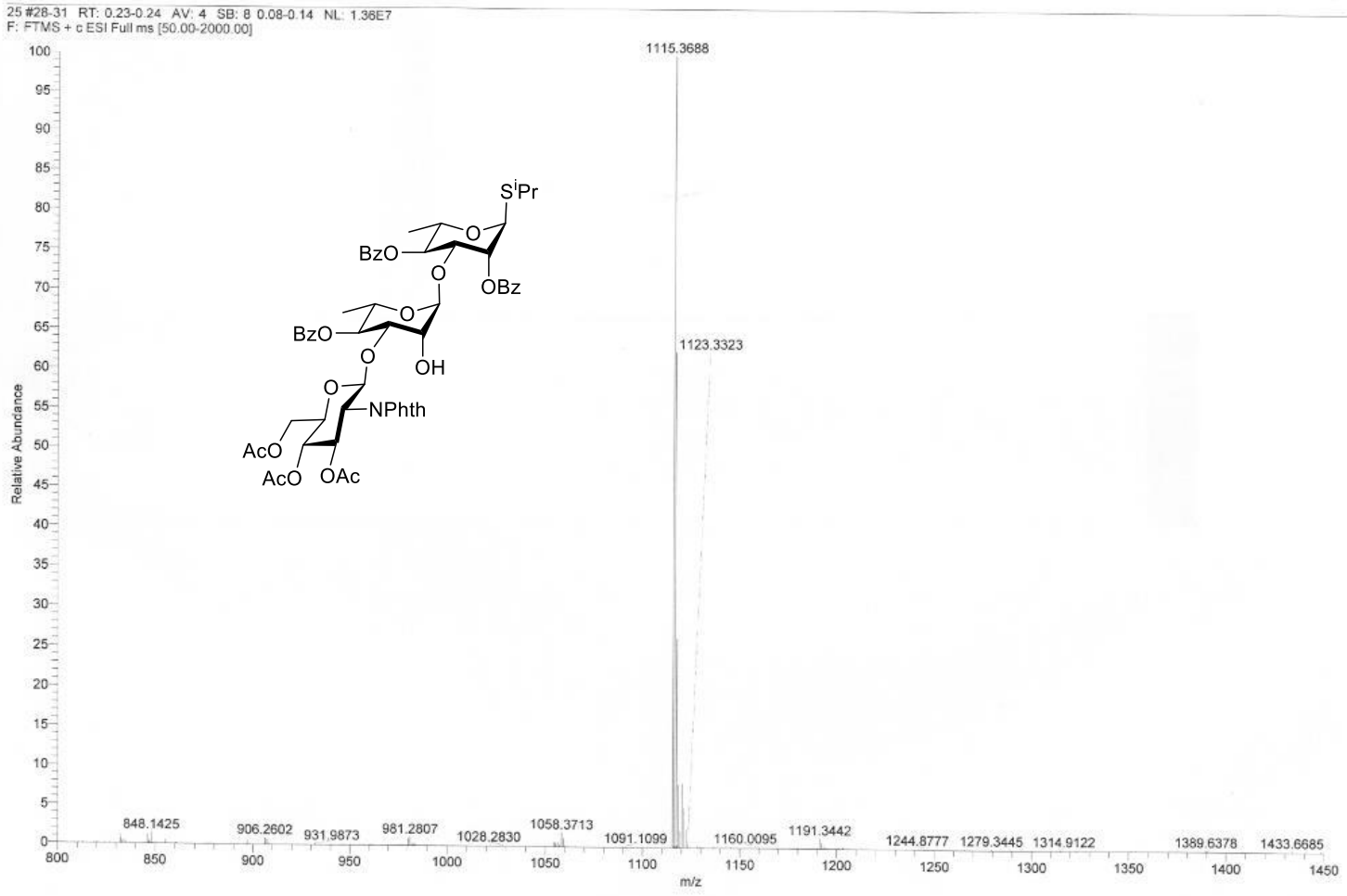
^{13}C NMR spectrum of compound **20** (150 MHz, $CDCl_3$)



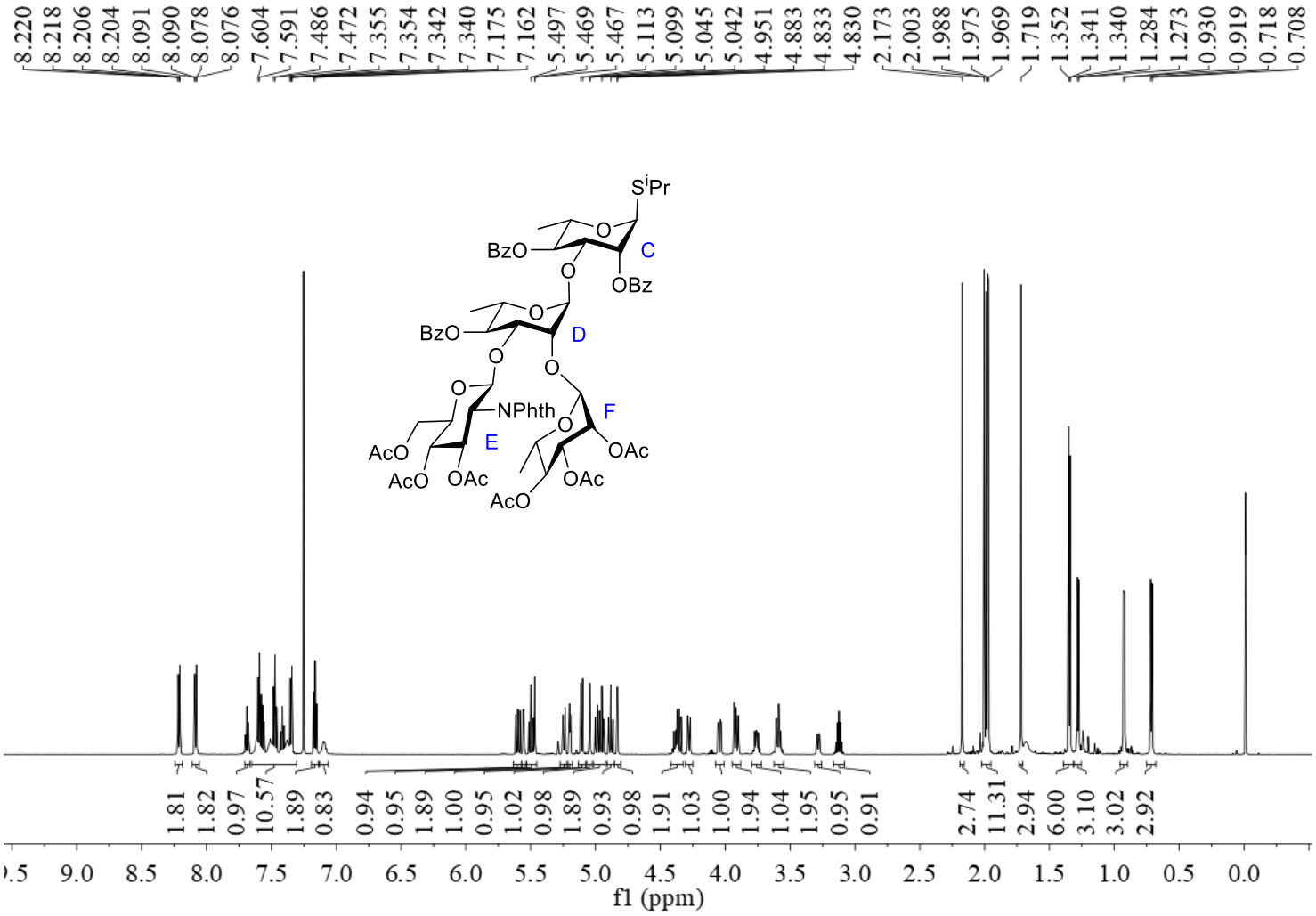
^{1}H-^{1}H COSY spectrum of compound **20** (600 MHz, $CDCl_3$)



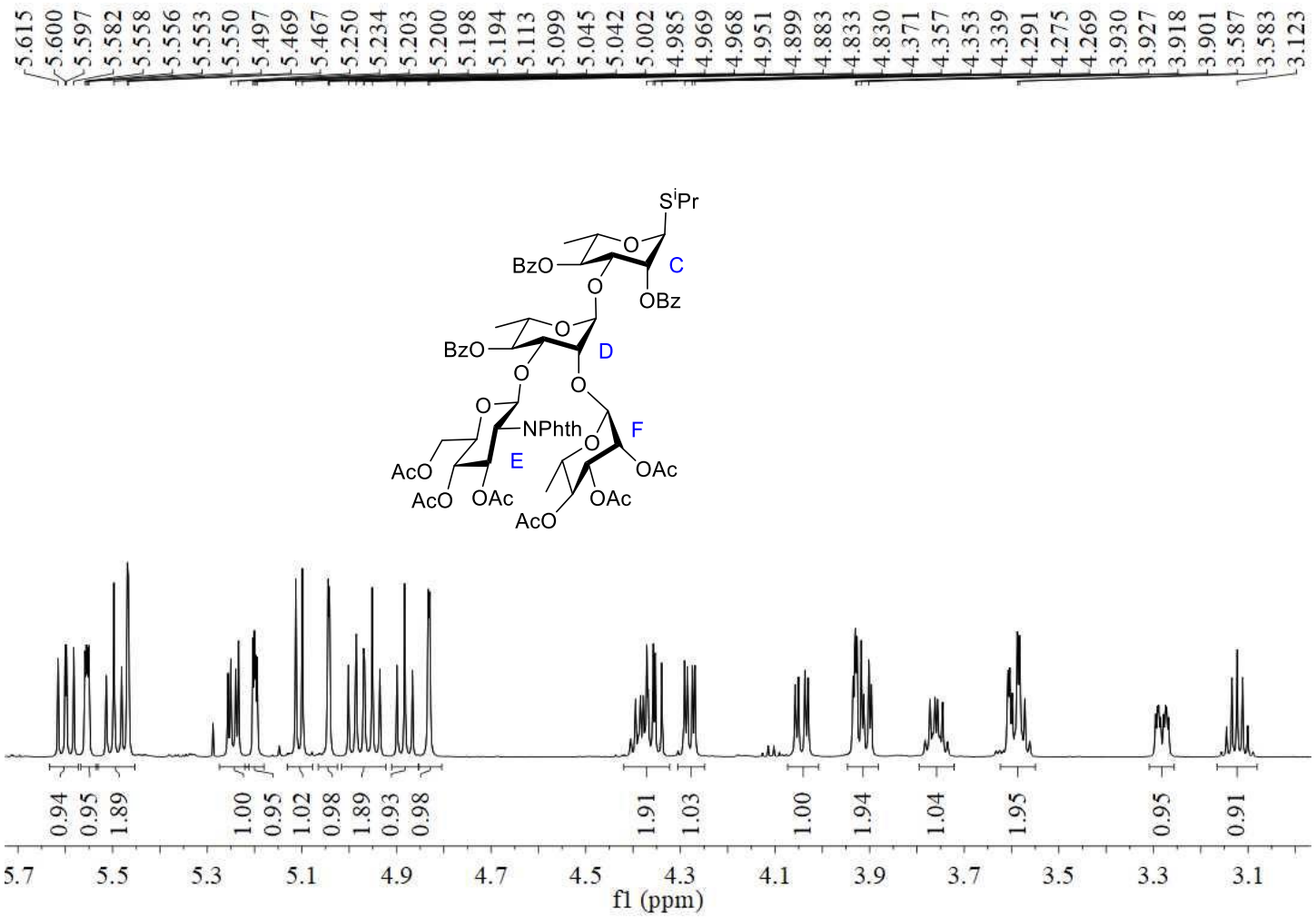
^{1}H-^{13}C HSQC spectrum of compound **20** (600/150 MHz, $CDCl_3$)



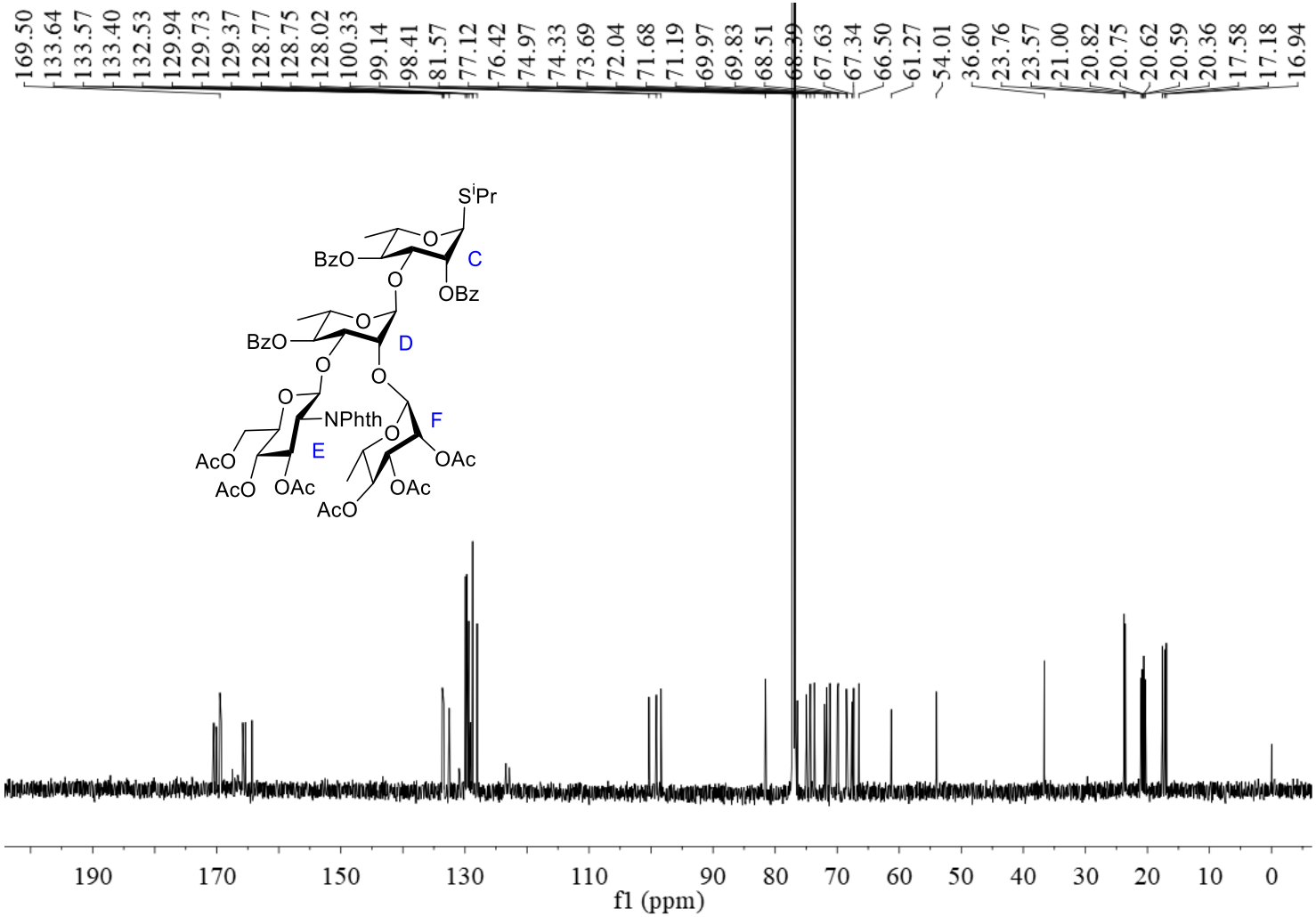
HR-ESI-(+) mass spectrum of compound **20**



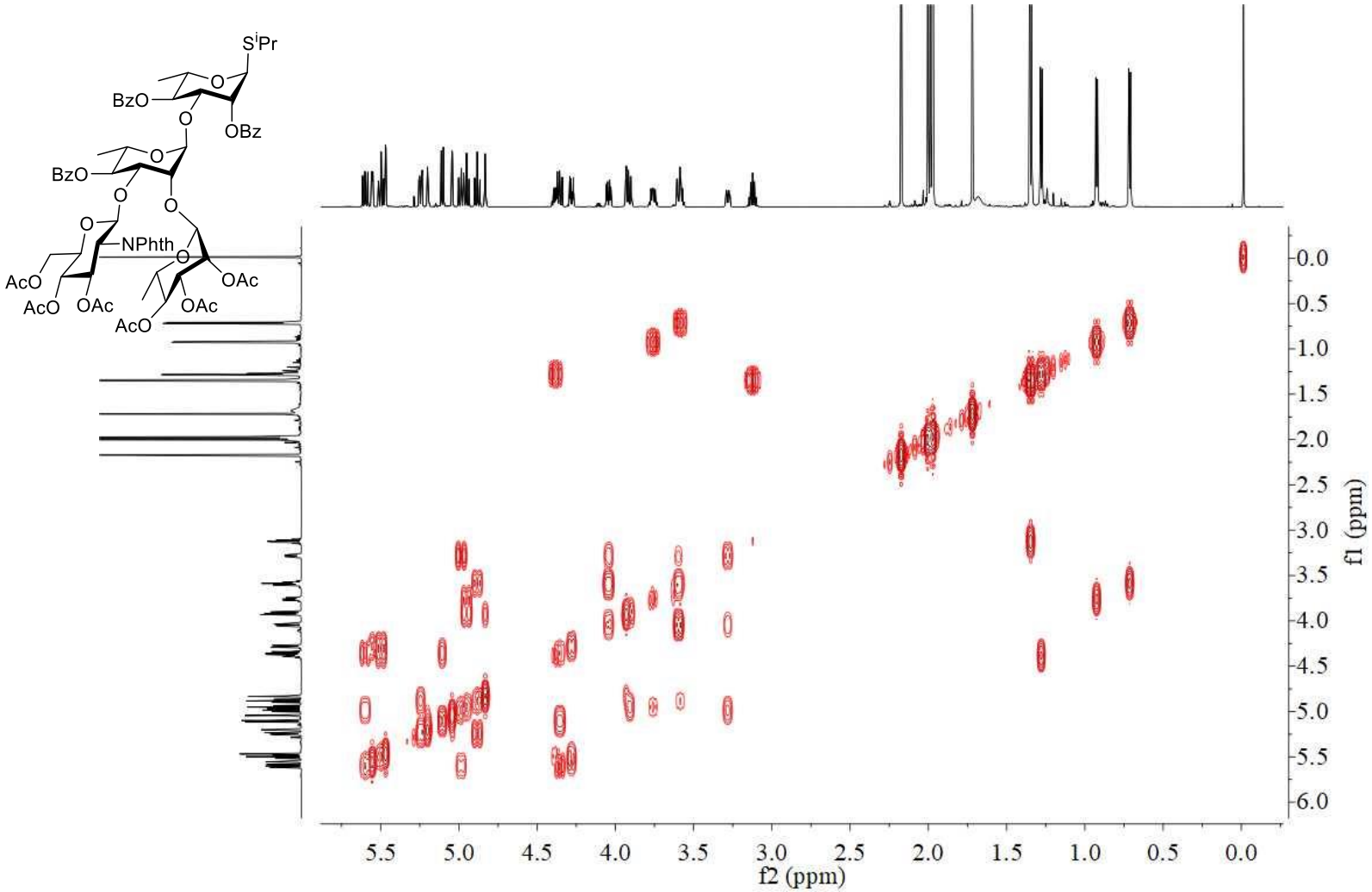
^{1}H NMR spectrum of compound **21** (600 MHz, $CDCl_3$)



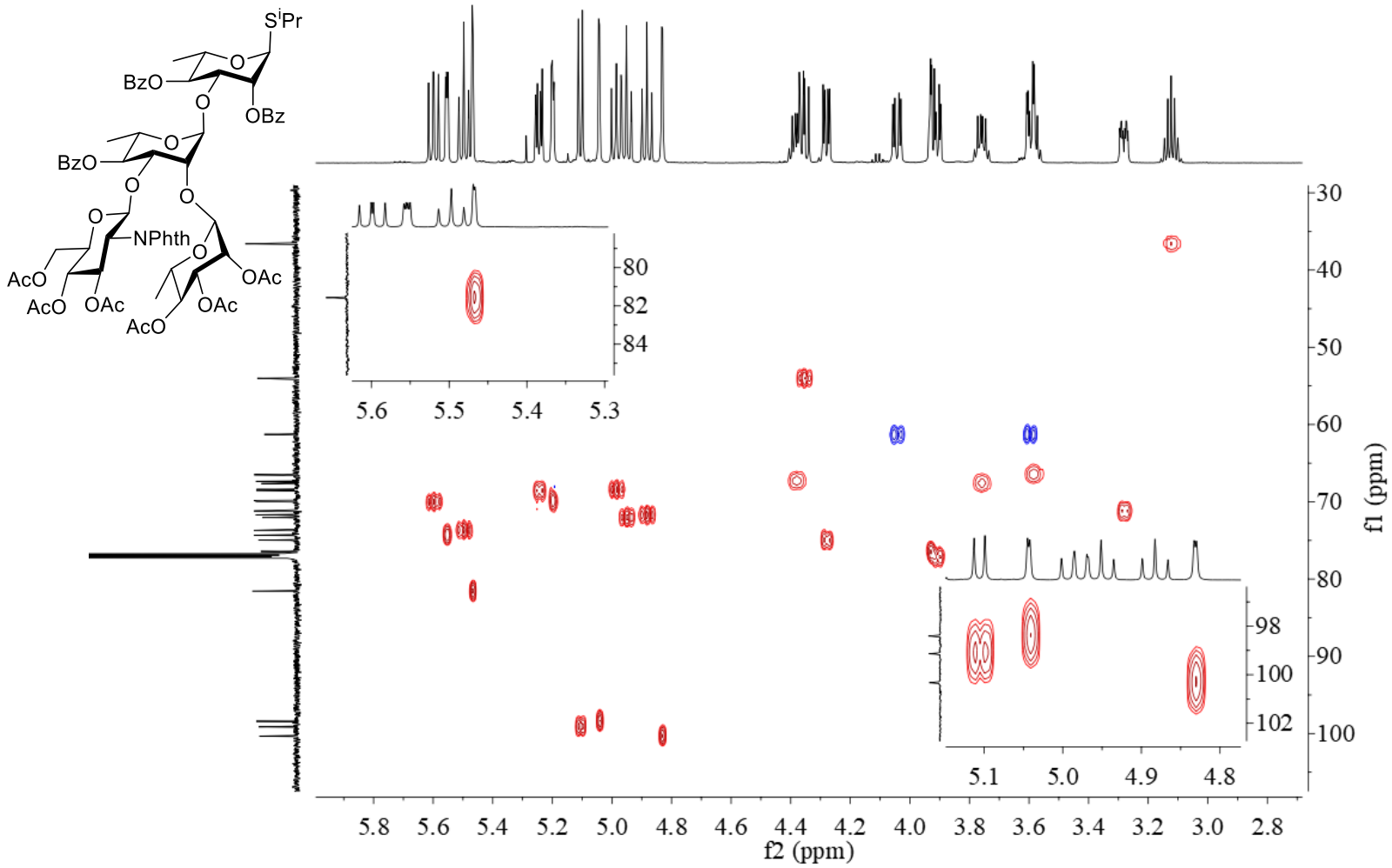
^{1}H NMR spectrum of compound **21** (expanded sugar region, 600 MHz, $CDCl_3$)



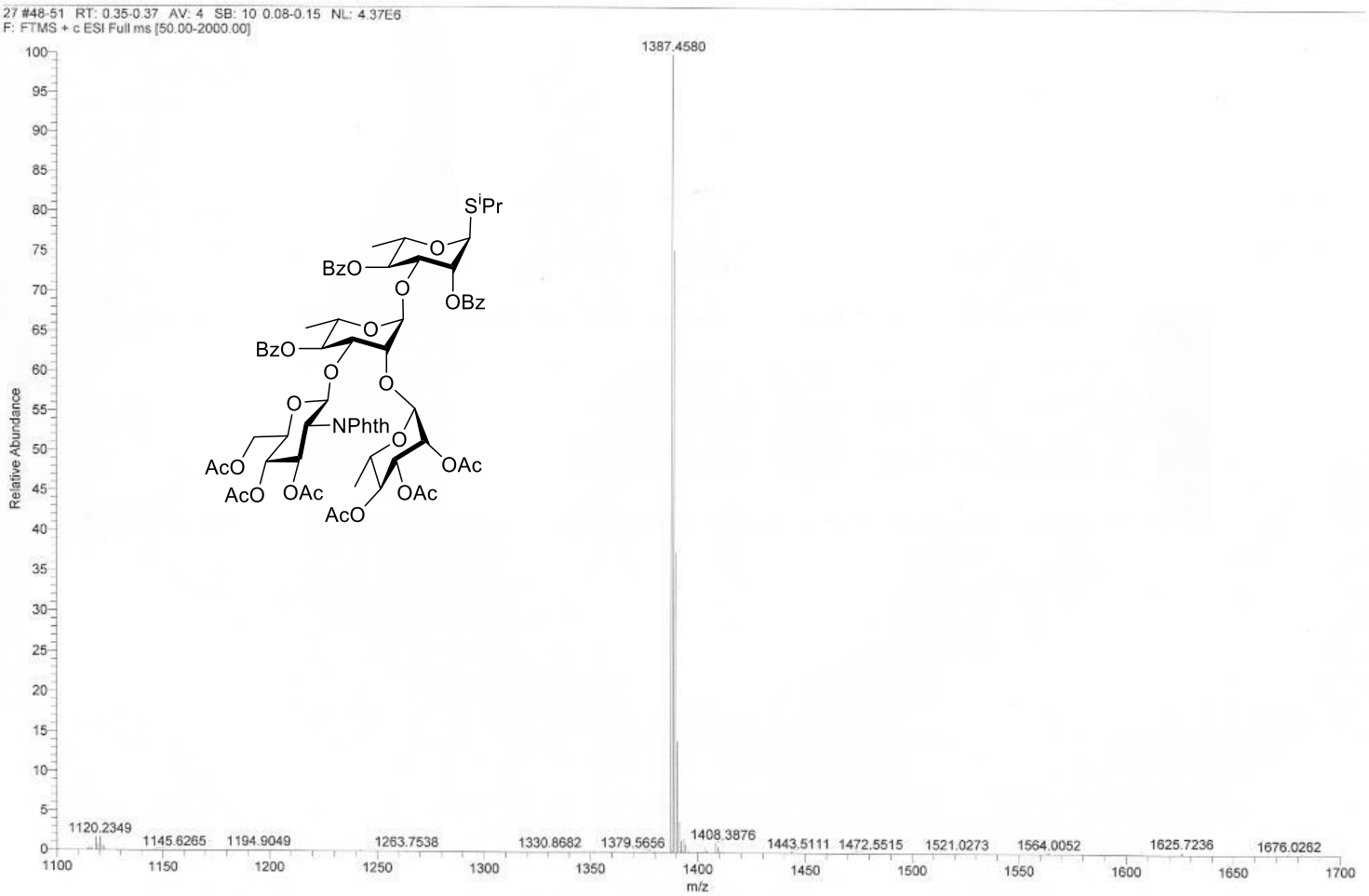
^{13}C NMR spectrum of compound **21** (150 MHz, $CDCl_3$)



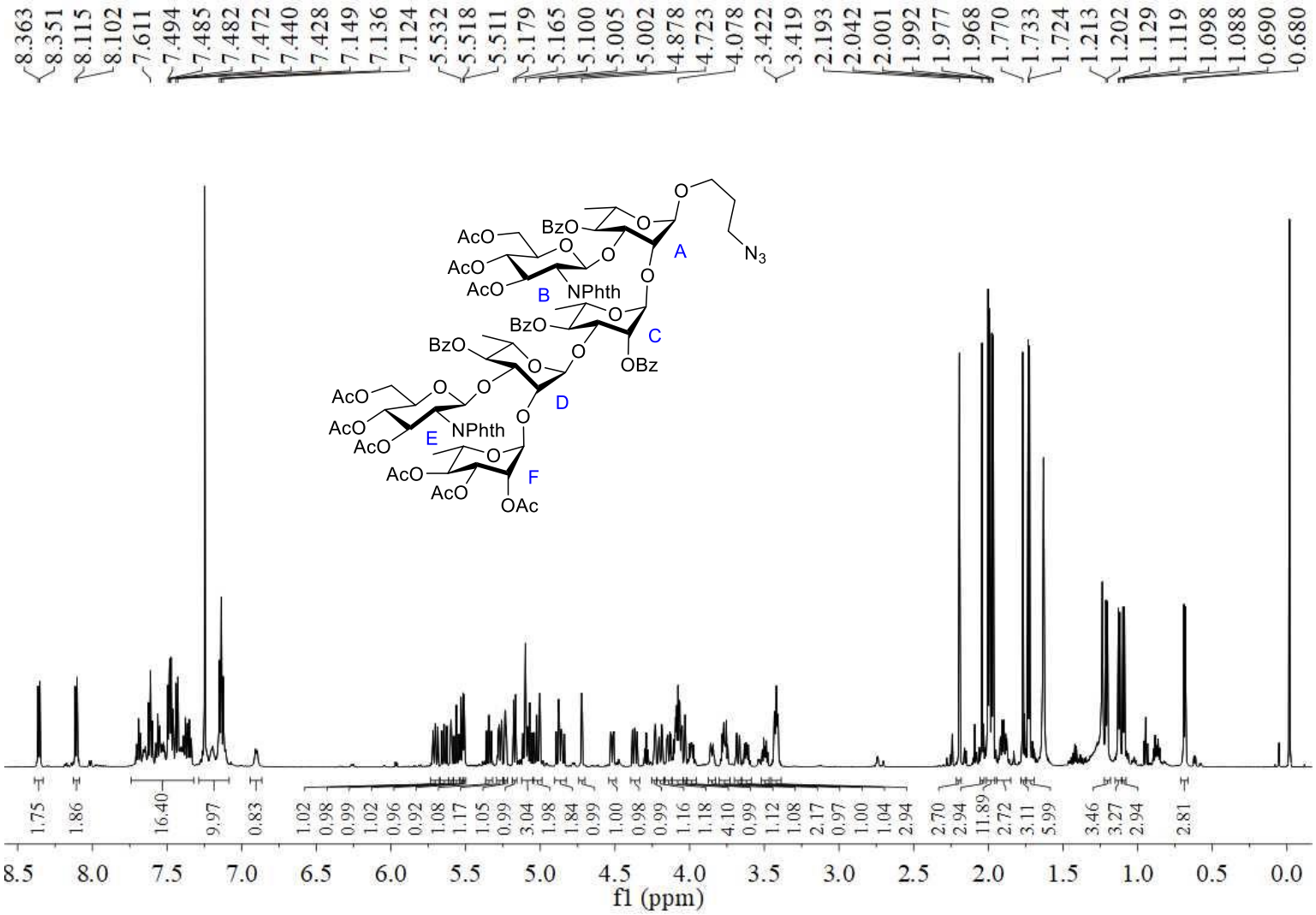
^{1}H-^{1}H COSY spectrum of compound **21** (600 MHz, $CDCl_3$)



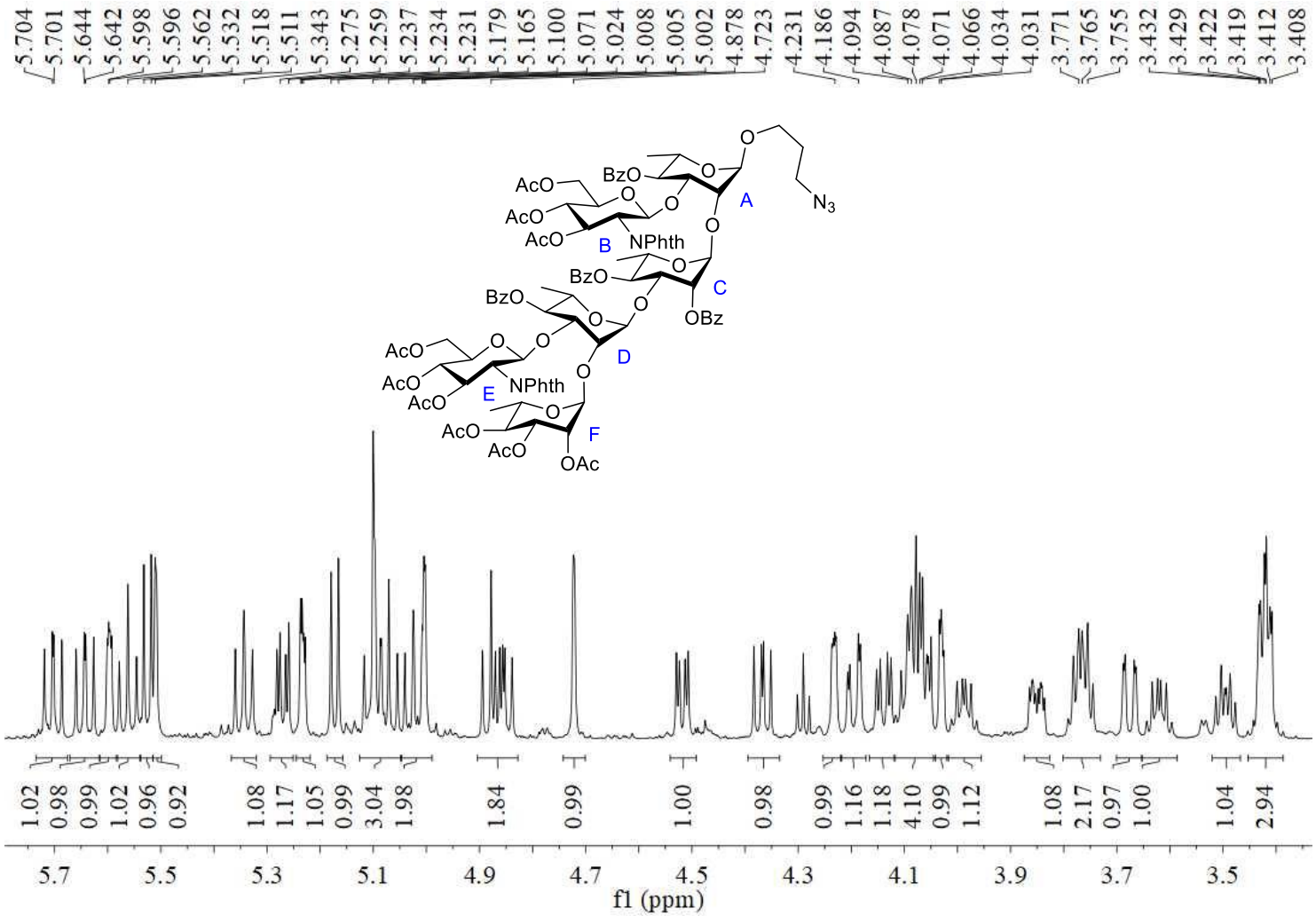
^{1}H-^{13}C HSQC spectrum of compound **21** (600/150 MHz, $CDCl_3$)



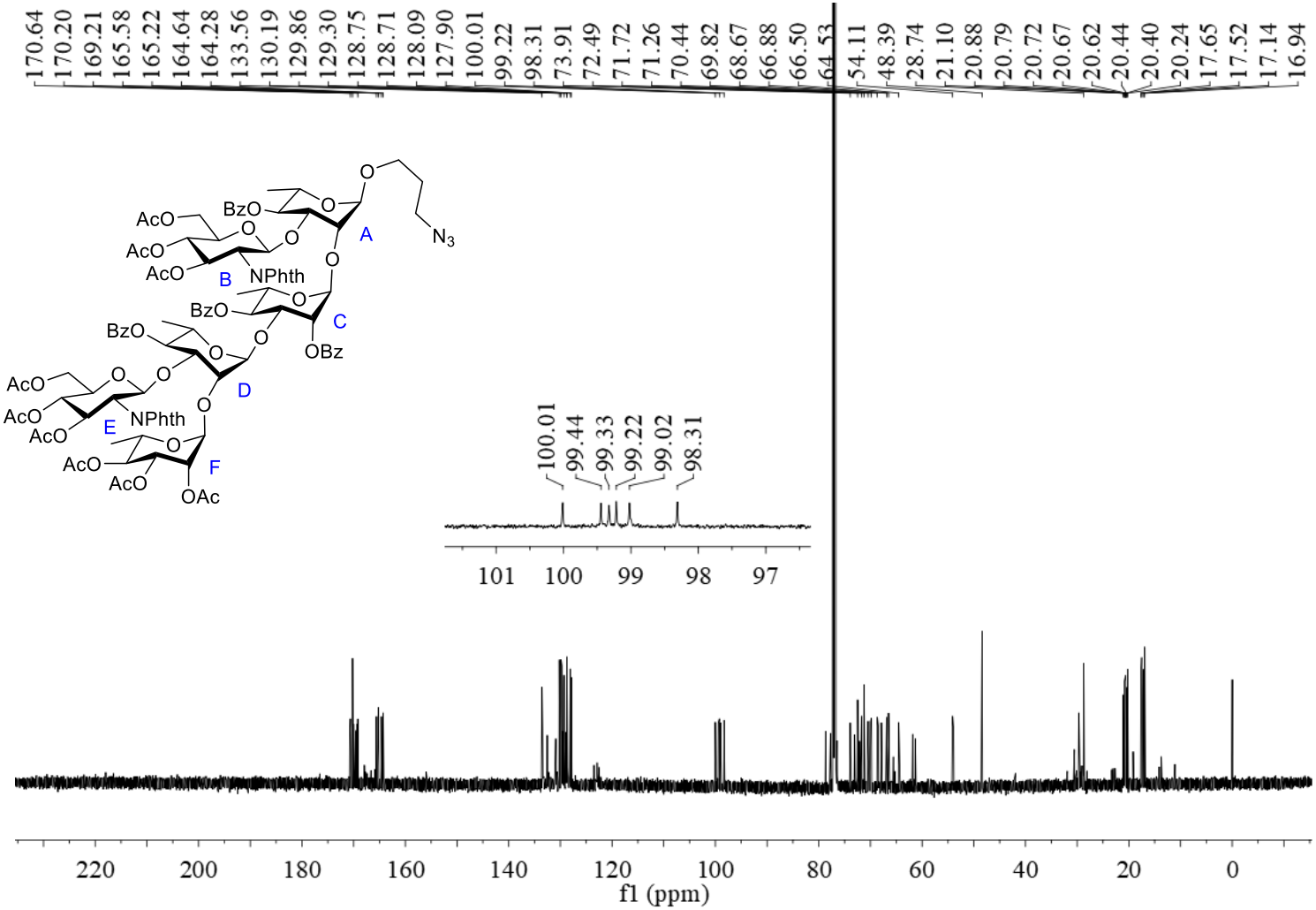
HR-ESI-(+) mass spectrum of compound **21**



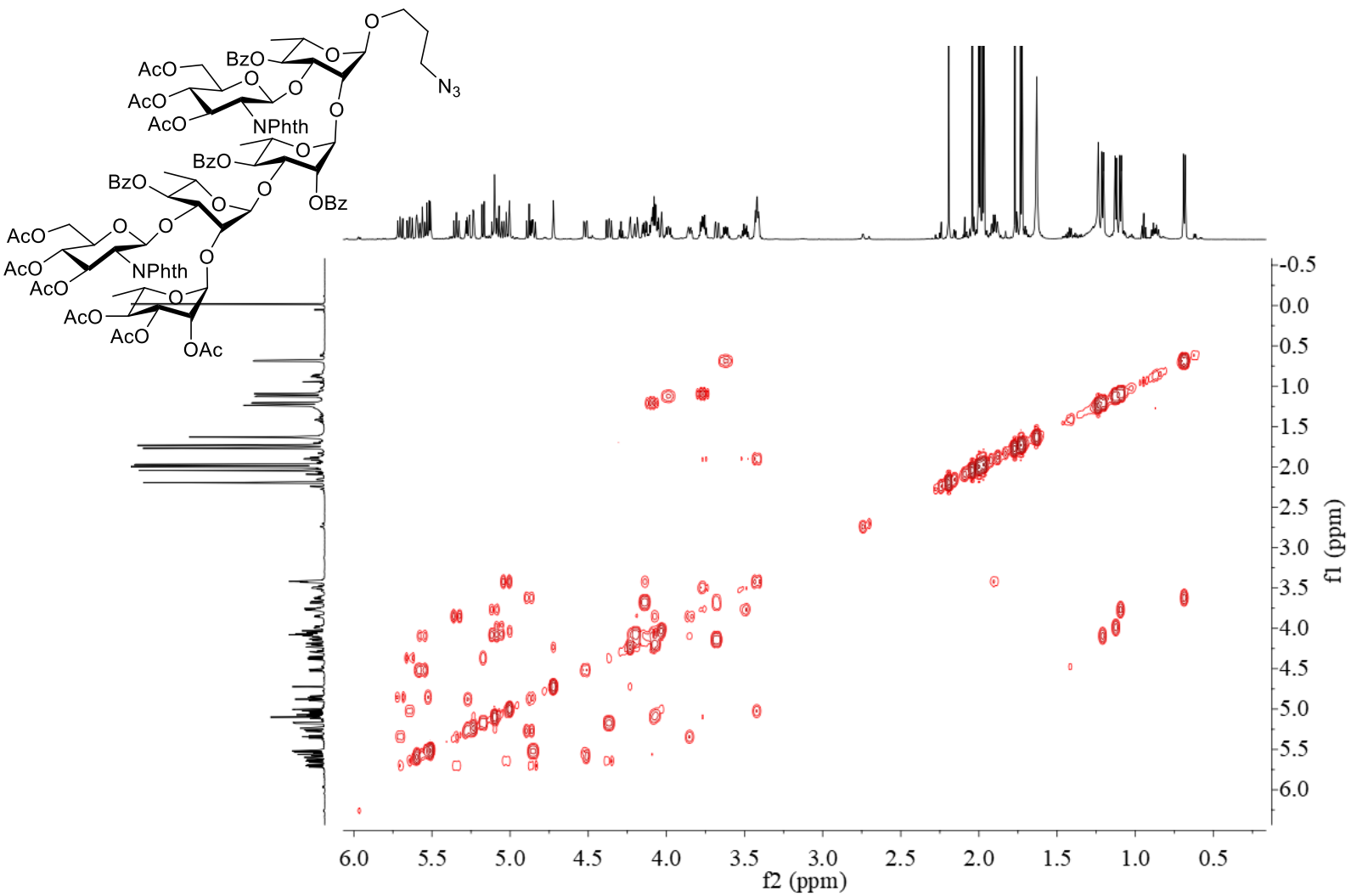
^{1}H NMR spectrum of compound **22** (600 MHz, $CDCl_3$)



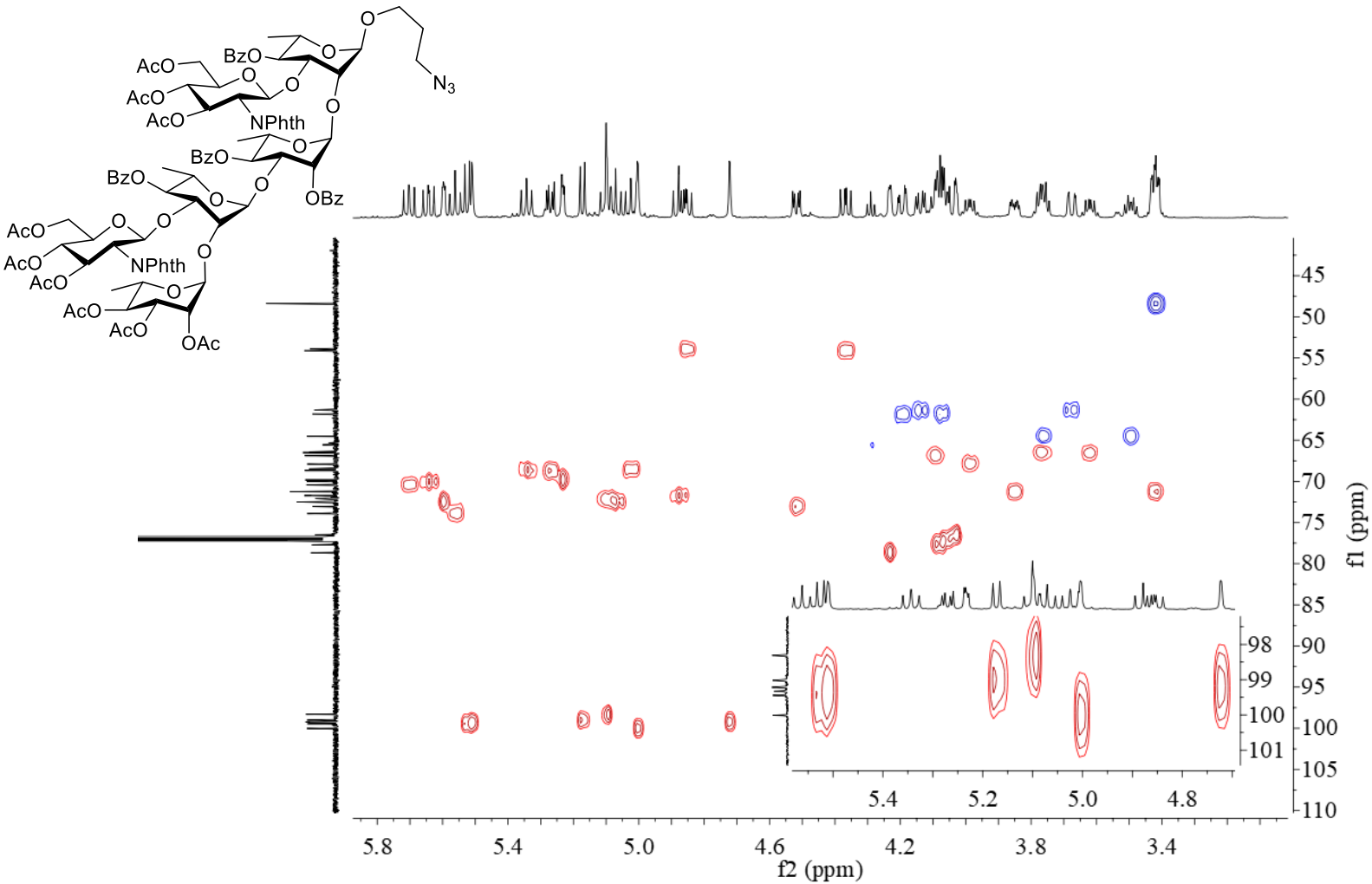
^{1}H NMR spectrum of compound **22** (expanded sugar region, 600 MHz, $CDCl_3$)



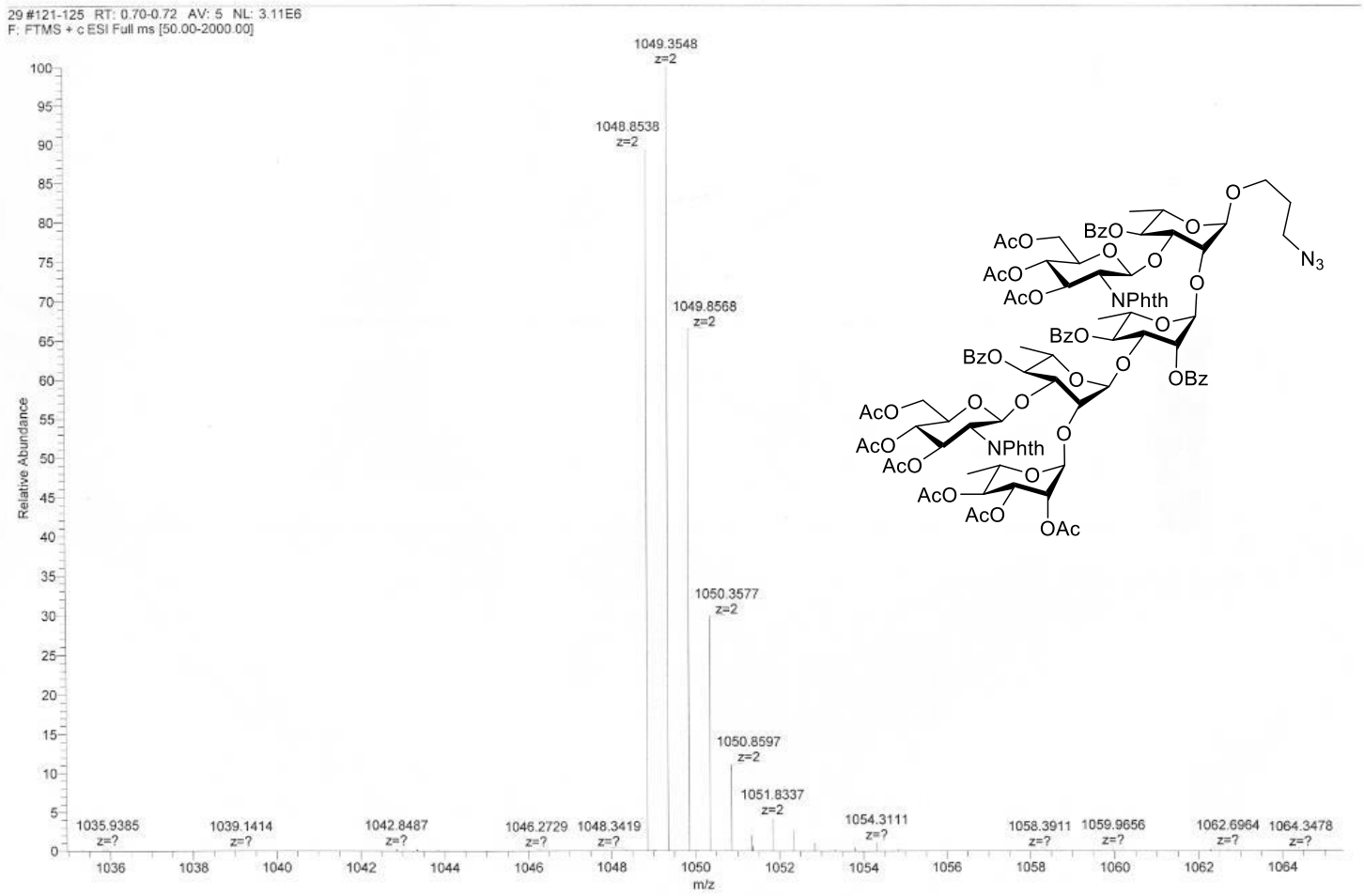
^{13}C NMR spectrum of compound **22** (150 MHz, $CDCl_3$)



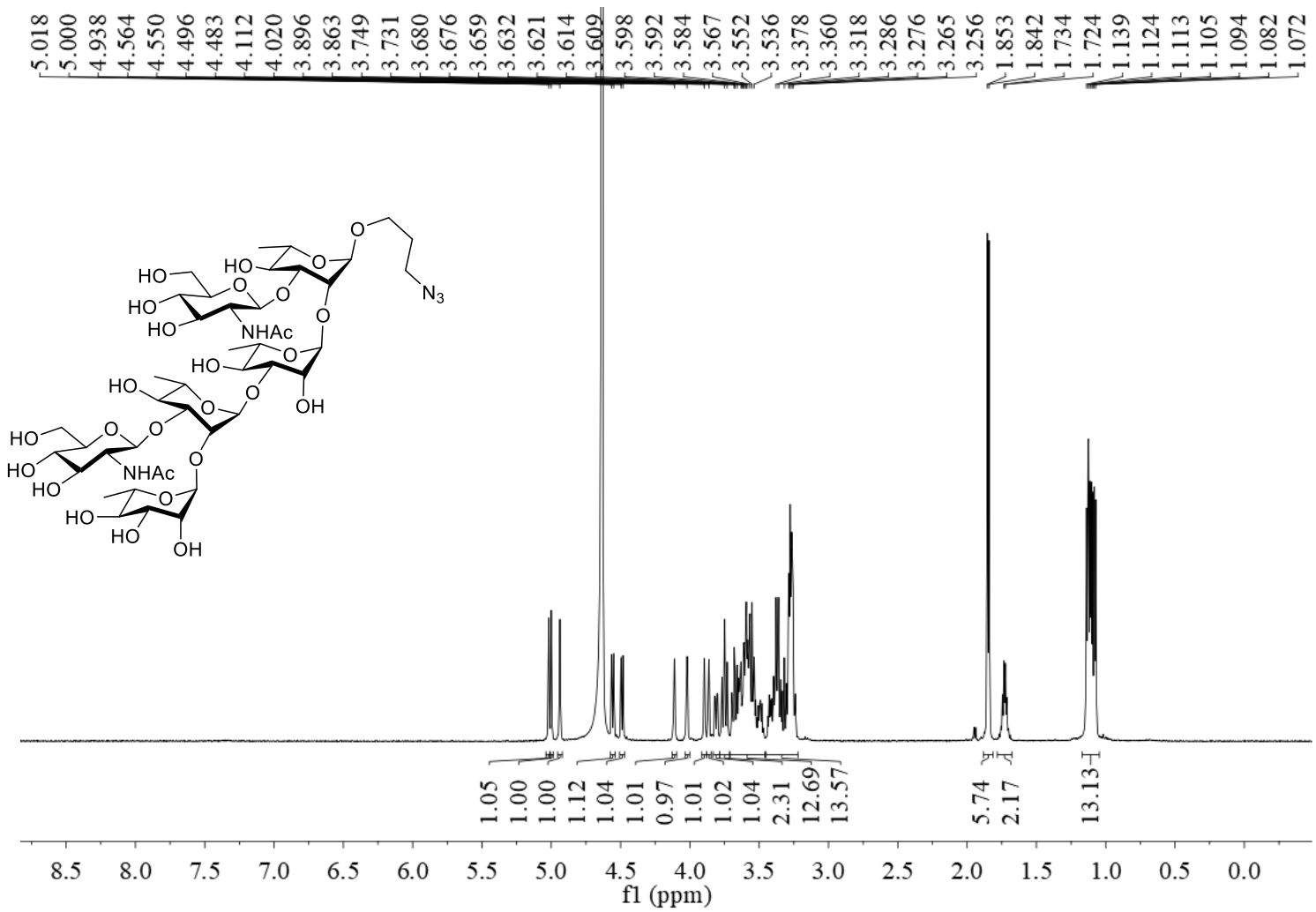
^{1}H-^{1}H COSY spectrum of compound **22** (600 MHz, $CDCl_3$)



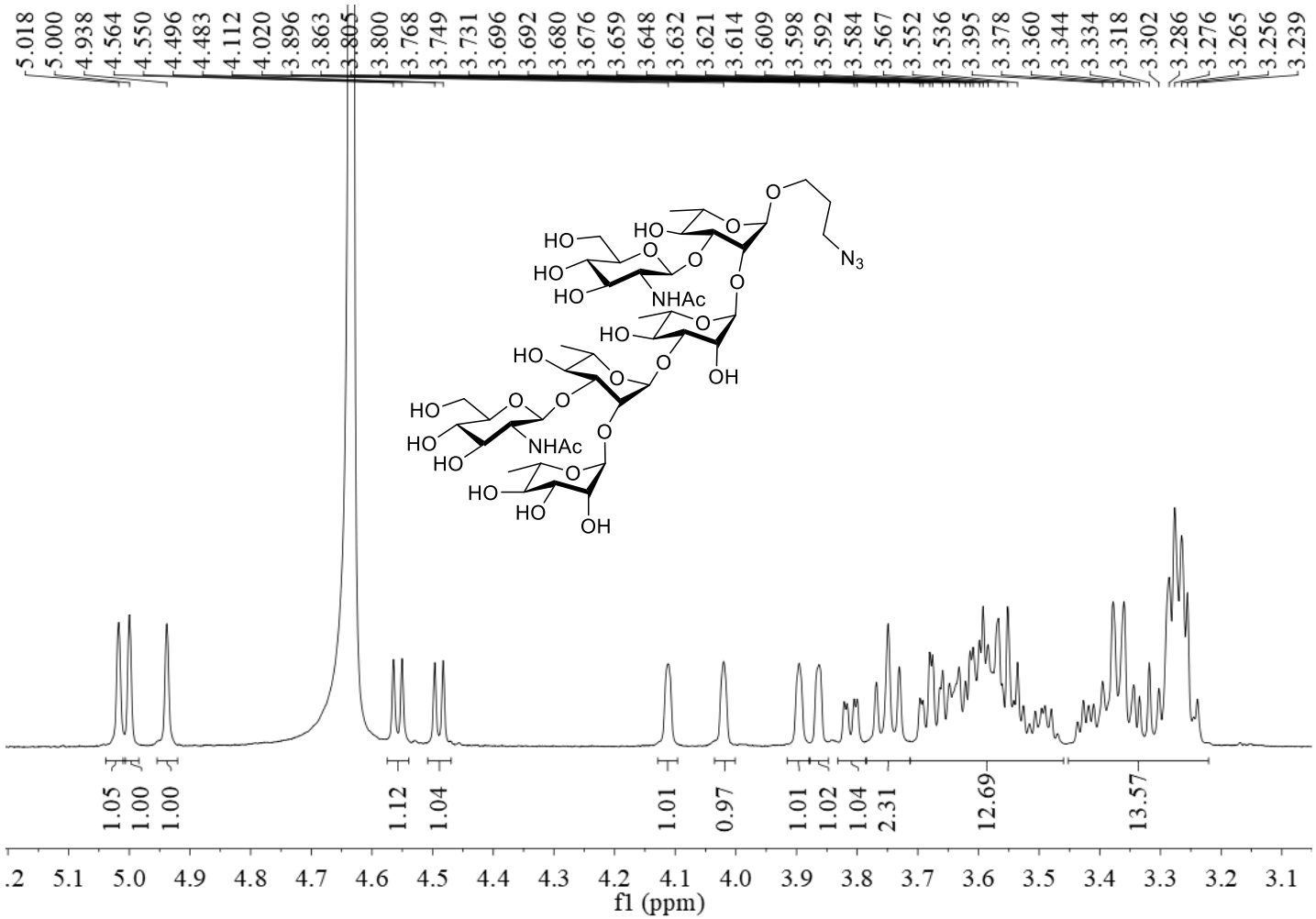
^{1}H-^{13}C HSQC spectrum of compound **22** (600/150 MHz, $CDCl_3$)



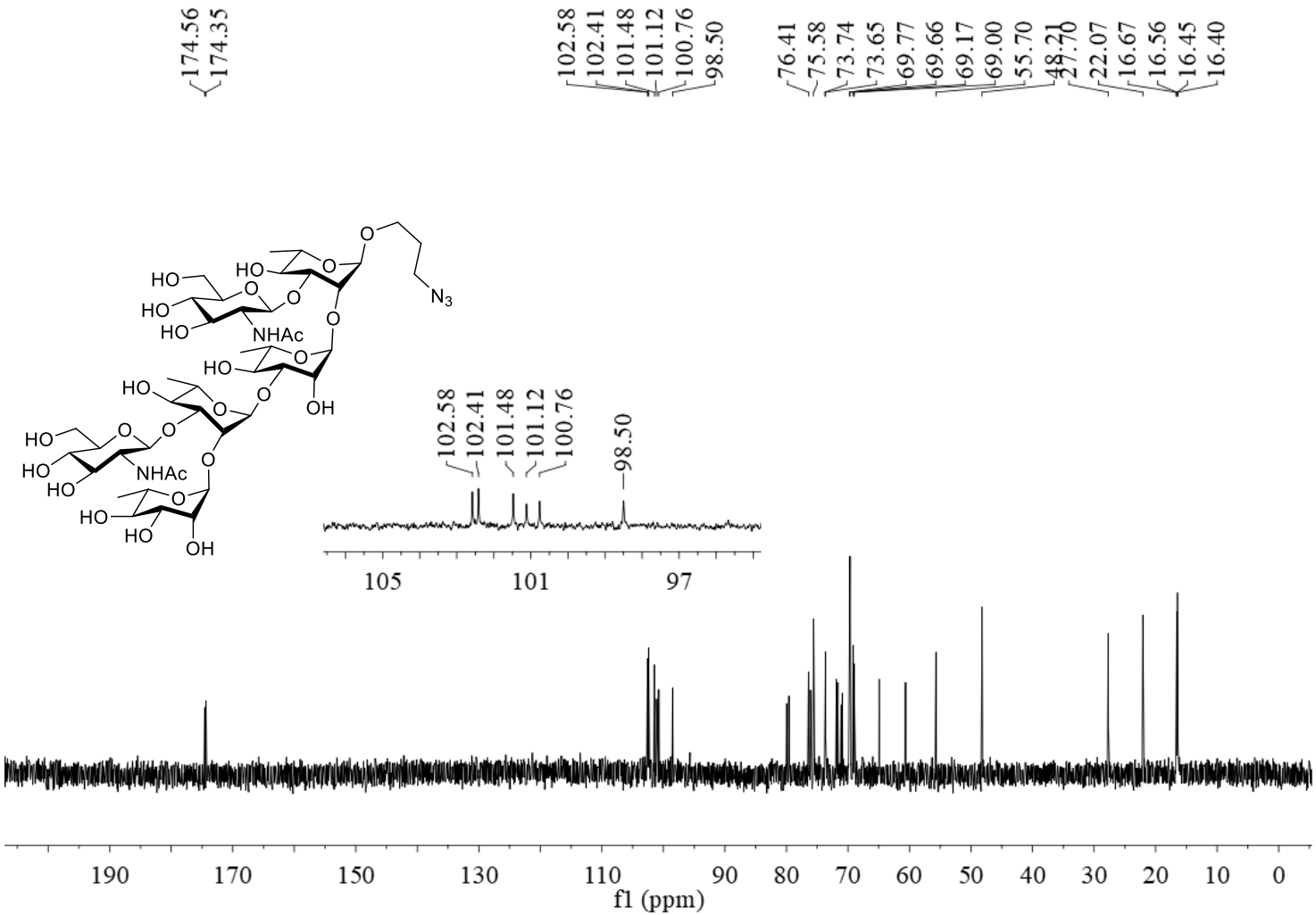
HR-ESI-(+) mass spectrum of compound **22**



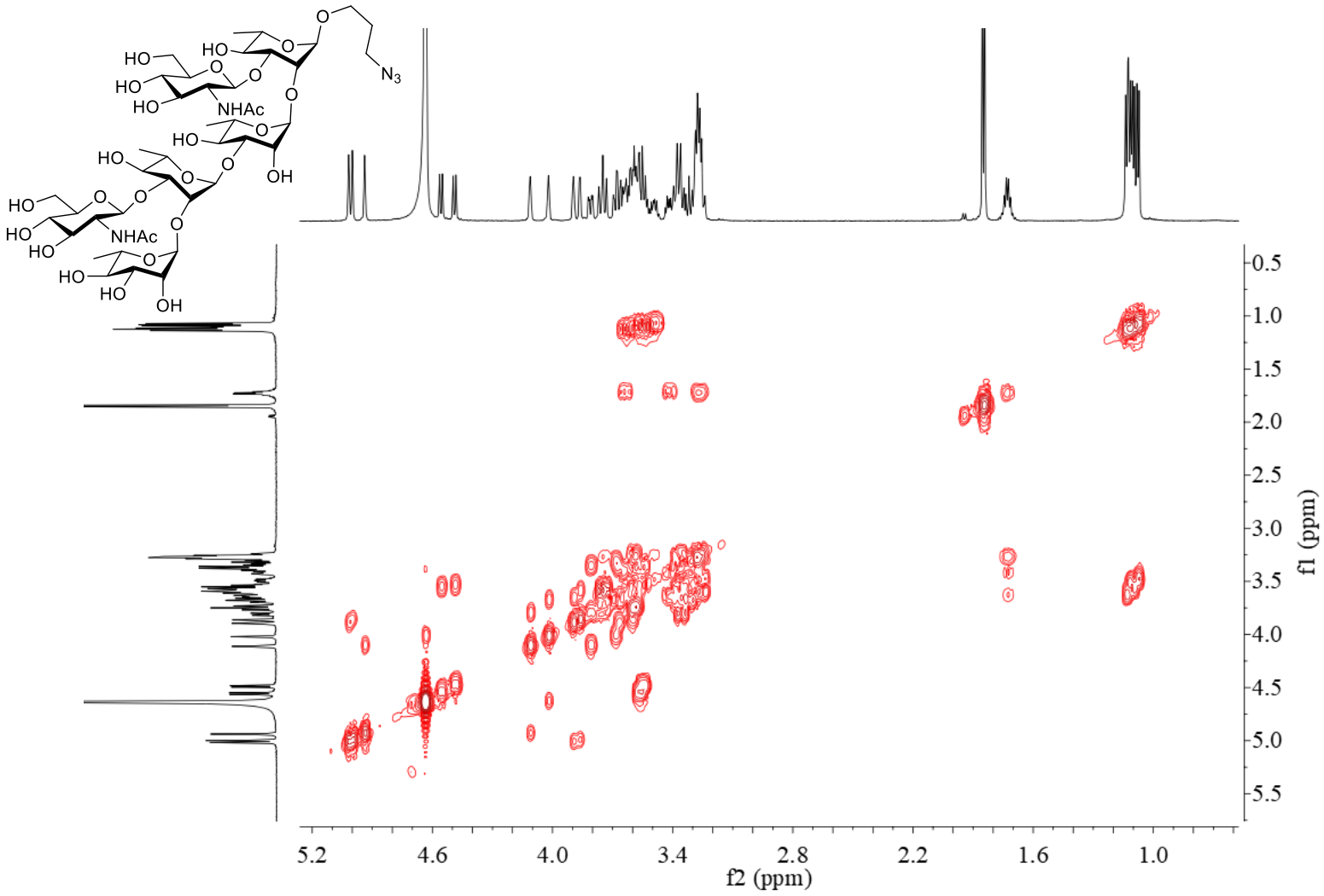
^{1}H NMR spectrum of compound **5b** (600 MHz, D_2O)



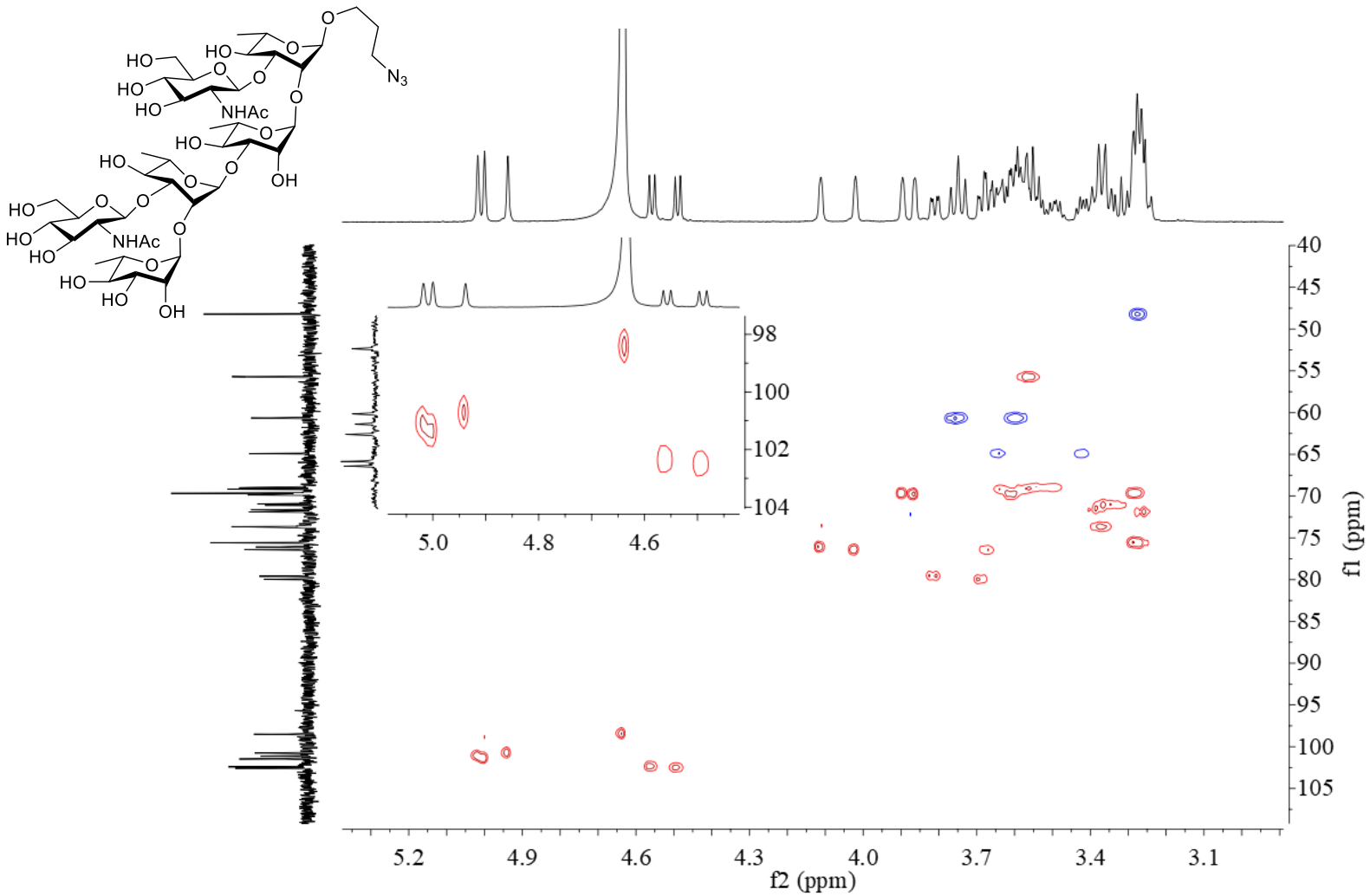
^{1}H NMR spectrum of compound **5b** (expanded sugar region, 600 MHz, D_2O)



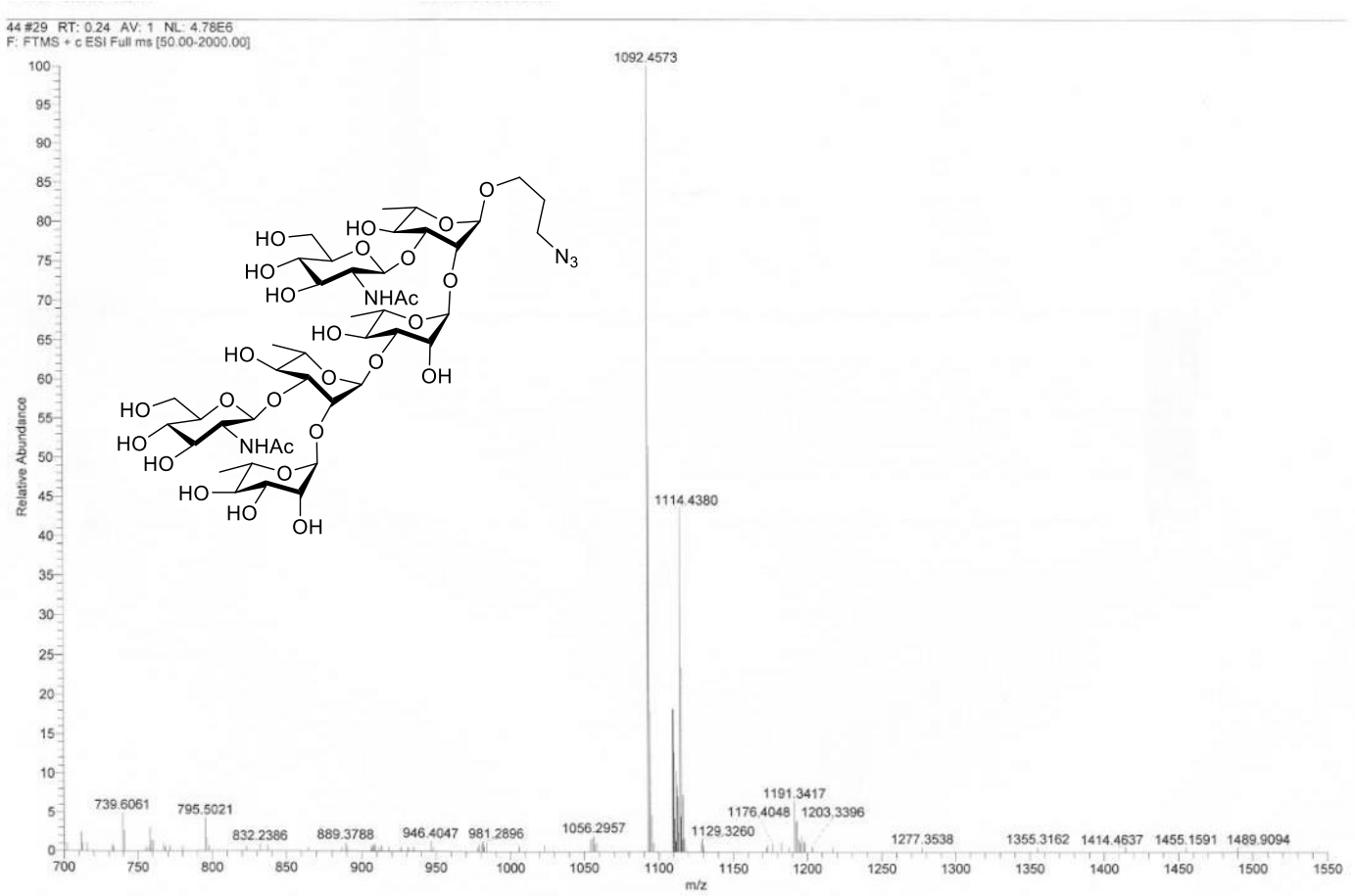
^{13}C NMR spectrum of compound **5b** (150 MHz, D_2O)



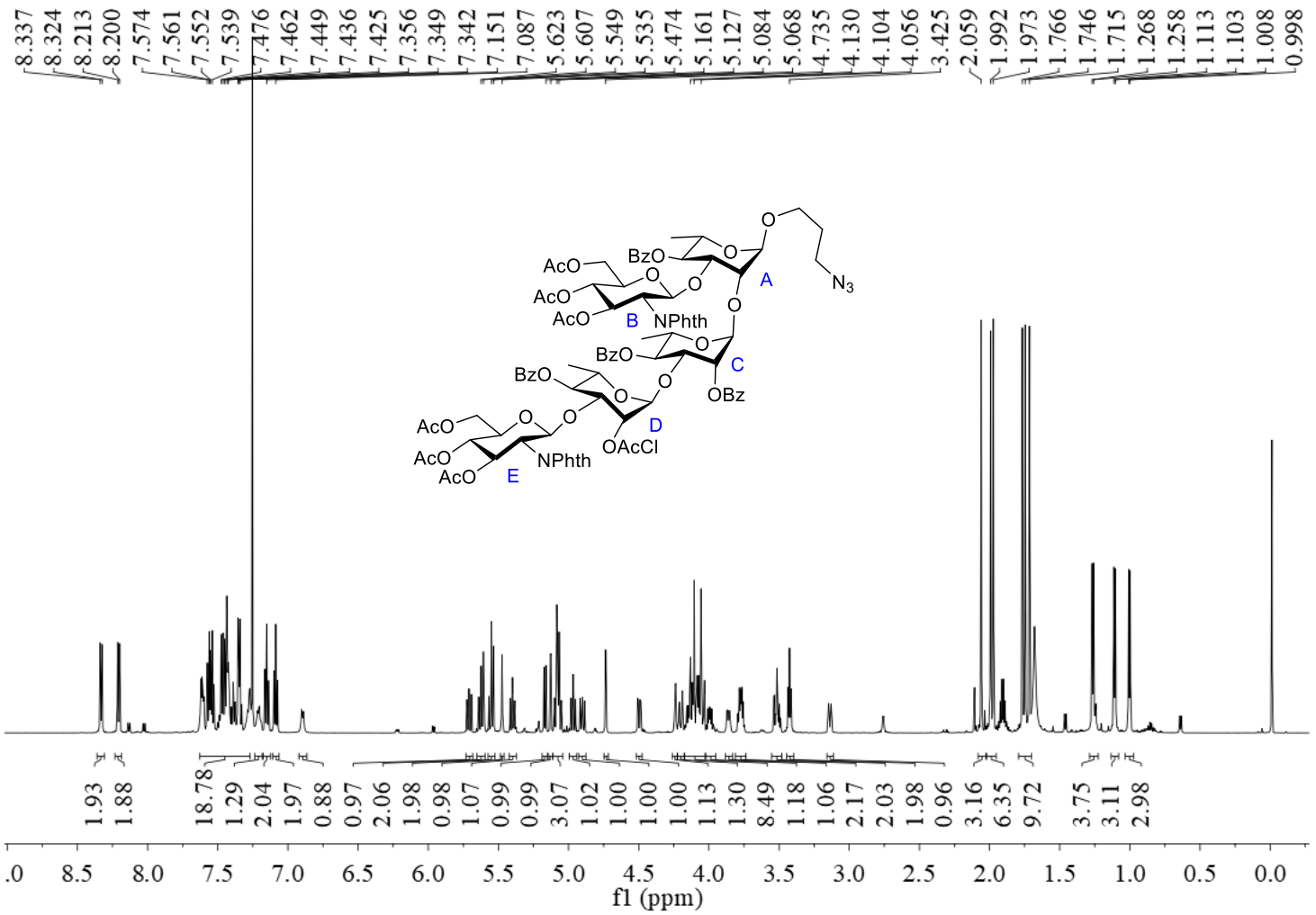
^{1}H-^{1}H COSY spectrum of compound **5b** (600 MHz, D_2O)



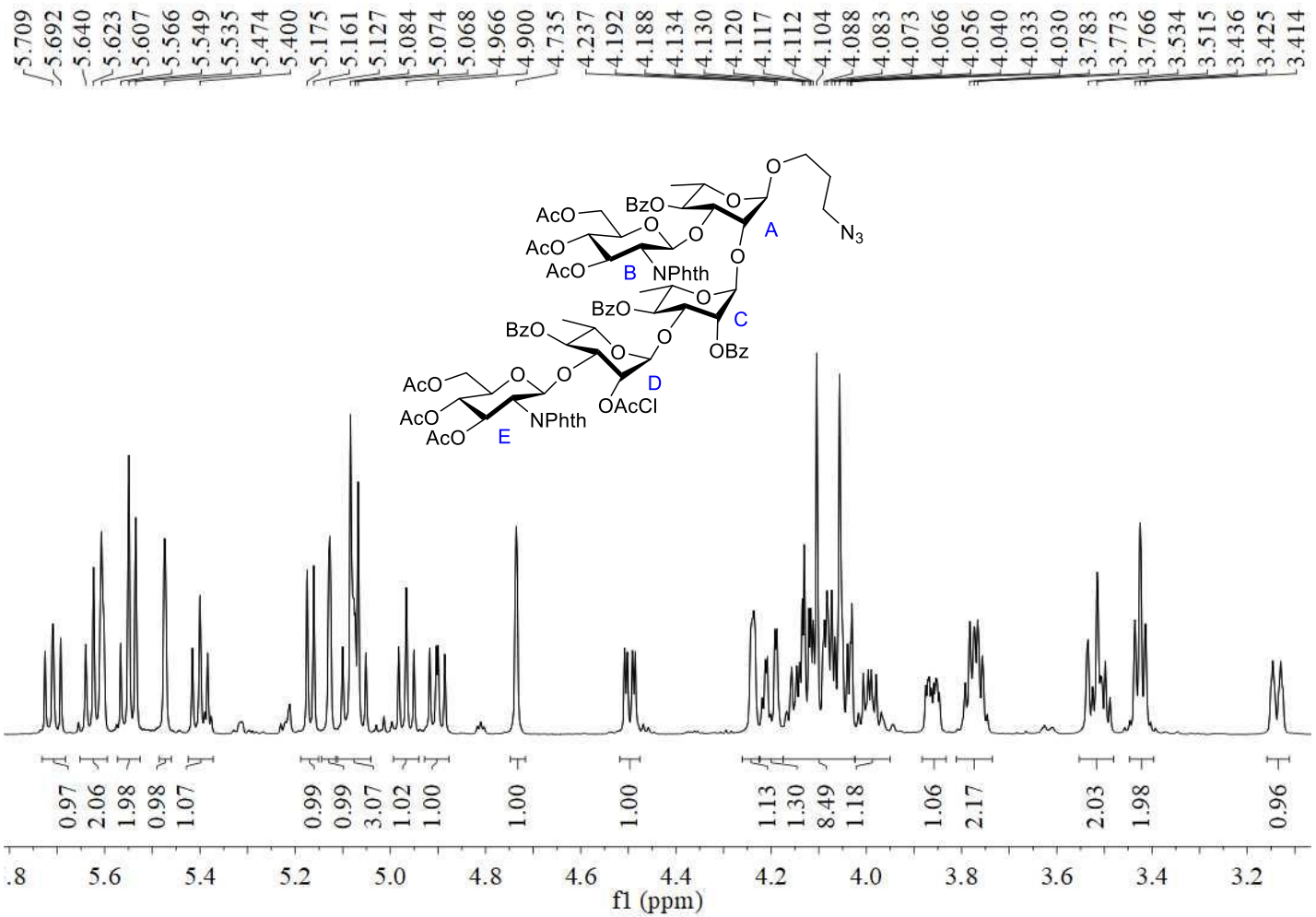
^{1}H-^{13}C HSQC spectrum of compound **5b** (600/150 MHz, D_2O)



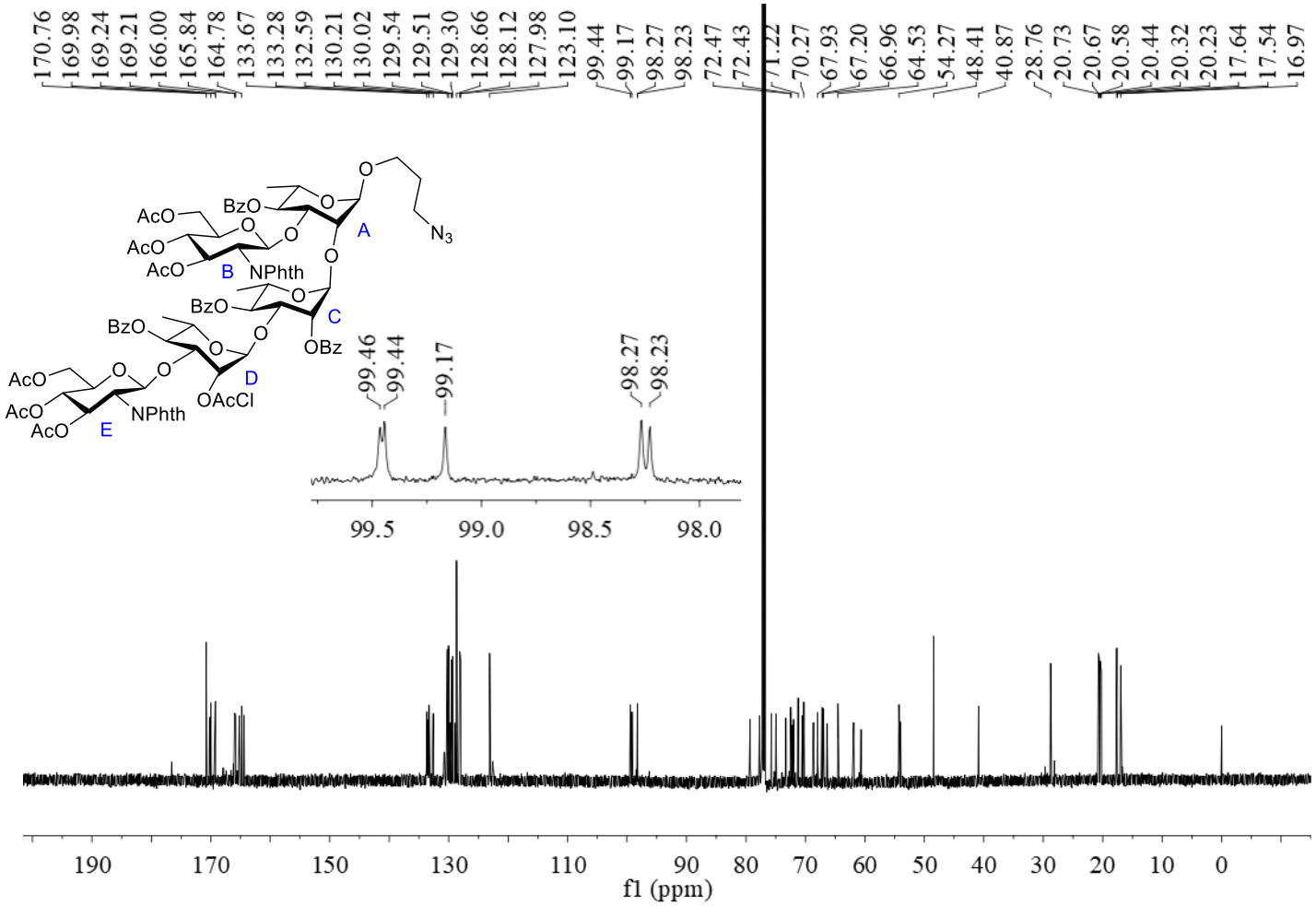
HR-ESI-(+) mass spectrum of compound **5b**



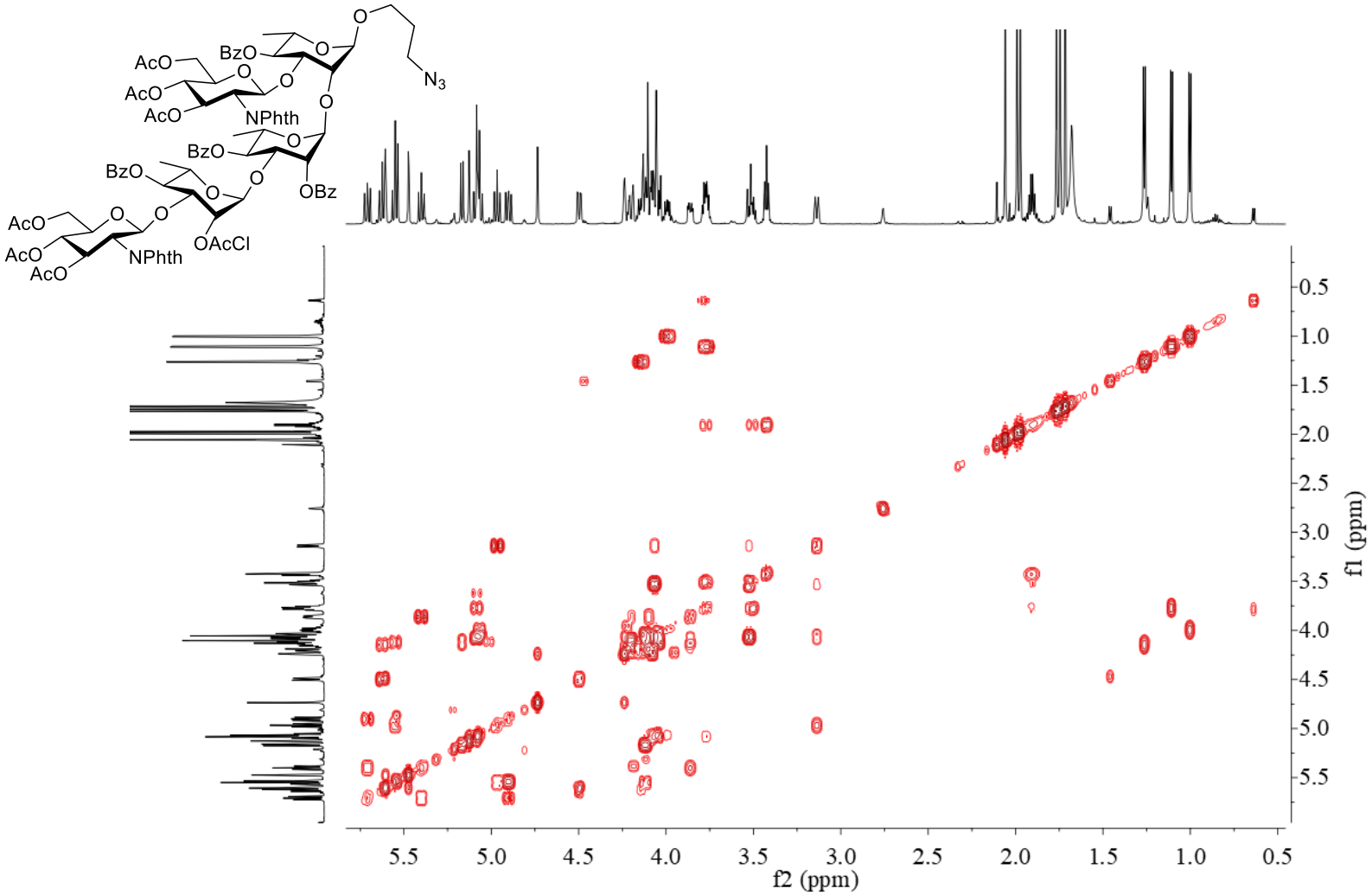
^{1}H NMR spectrum of compound **23** (600 MHz, $CDCl_3$)



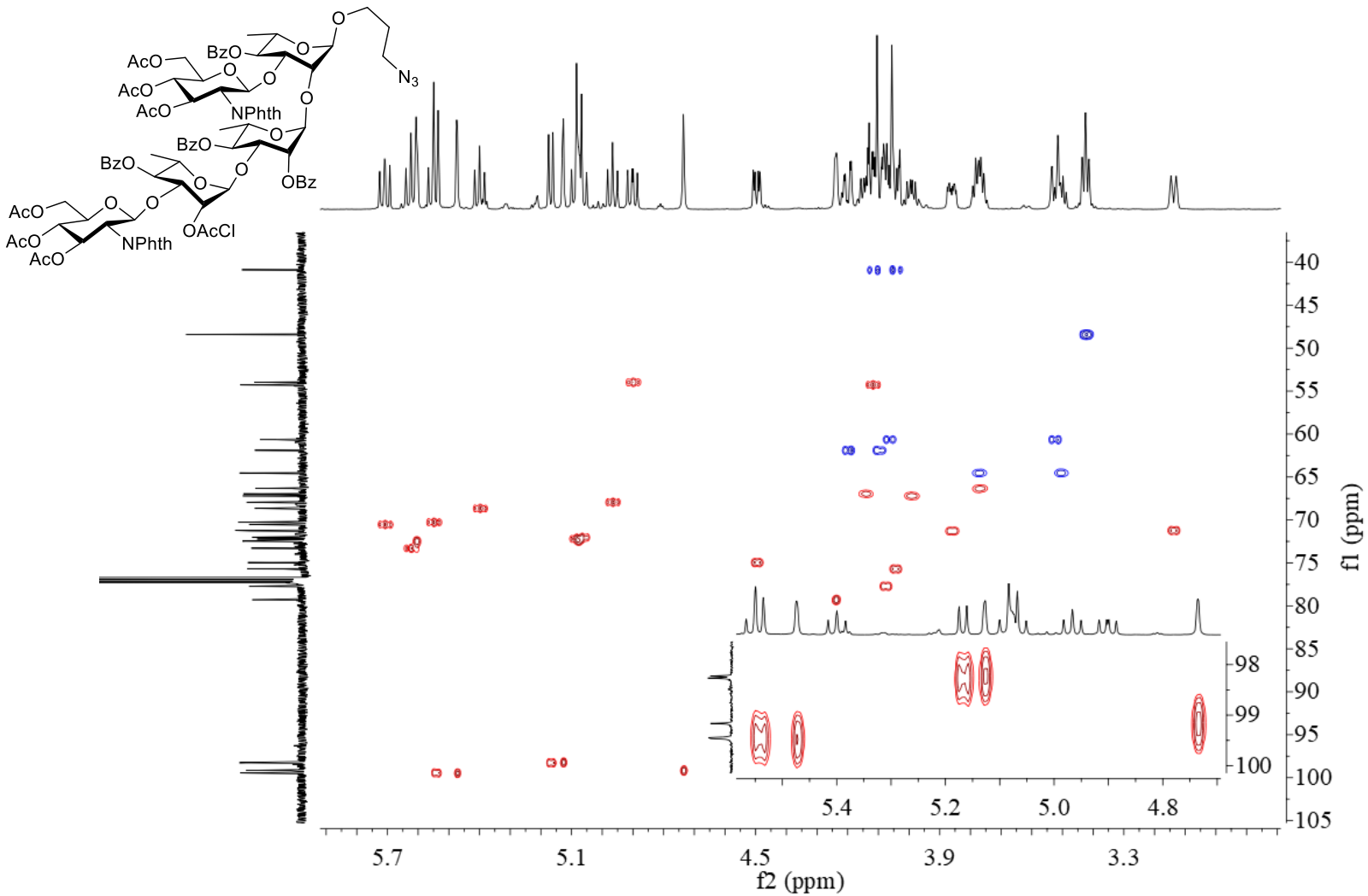
^{1}H NMR spectrum of compound **23** (expanded sugar region, 600 MHz, $CDCl_3$)



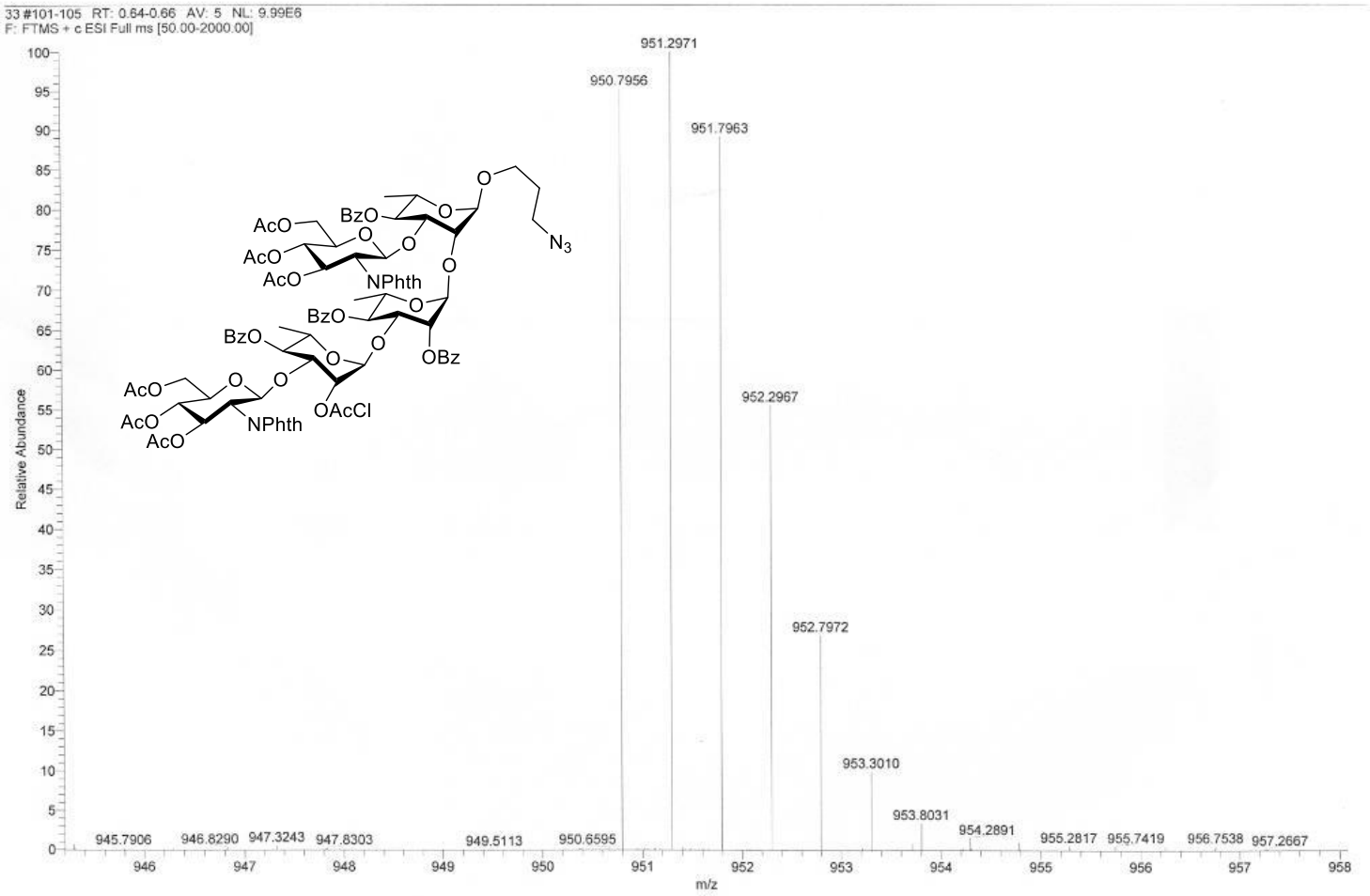
^{13}C NMR spectrum of compound **23** (150 MHz, $CDCl_3$)



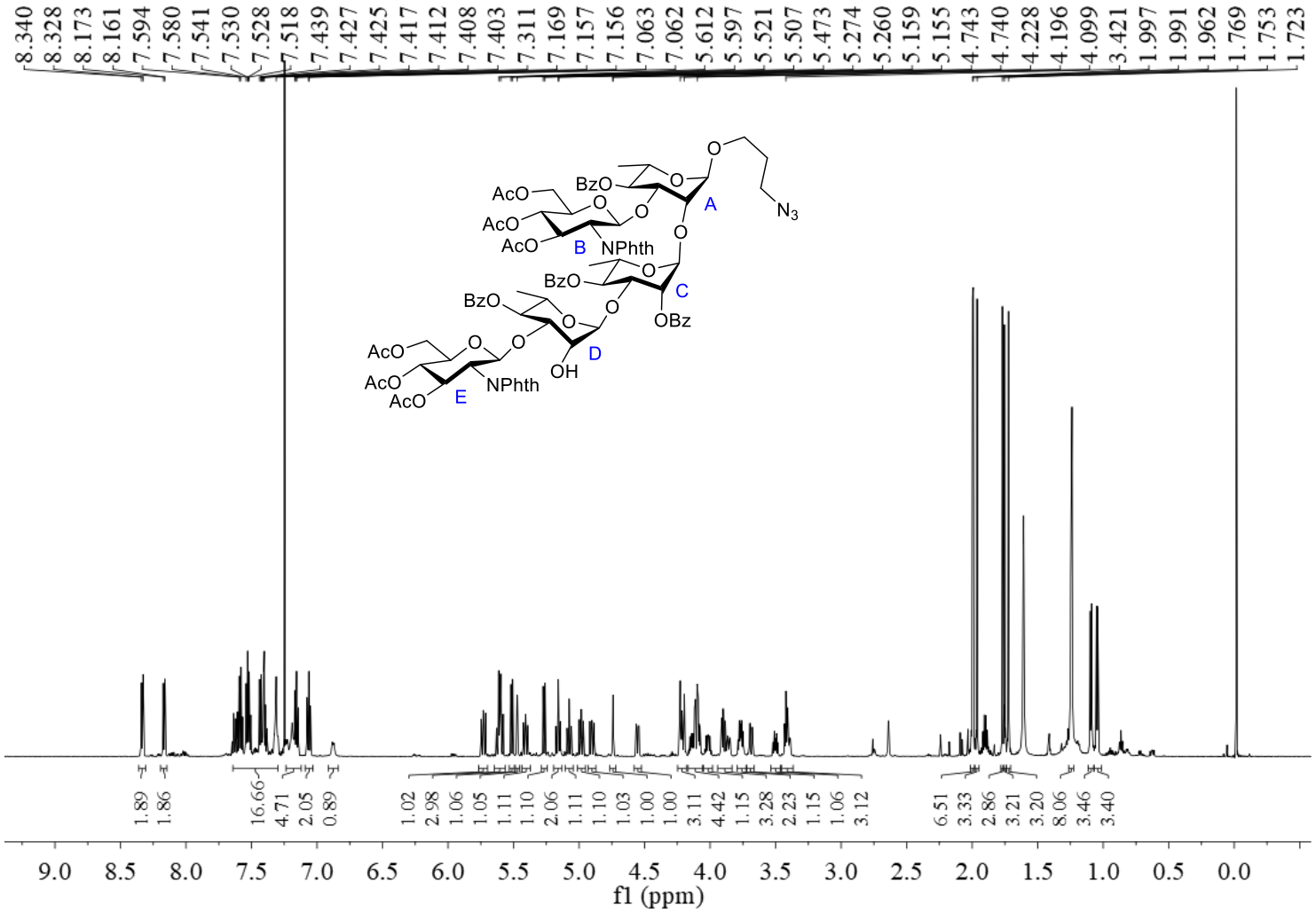
^{1}H-^{1}H COSY spectrum of compound **23** (600 MHz, $CDCl_3$)



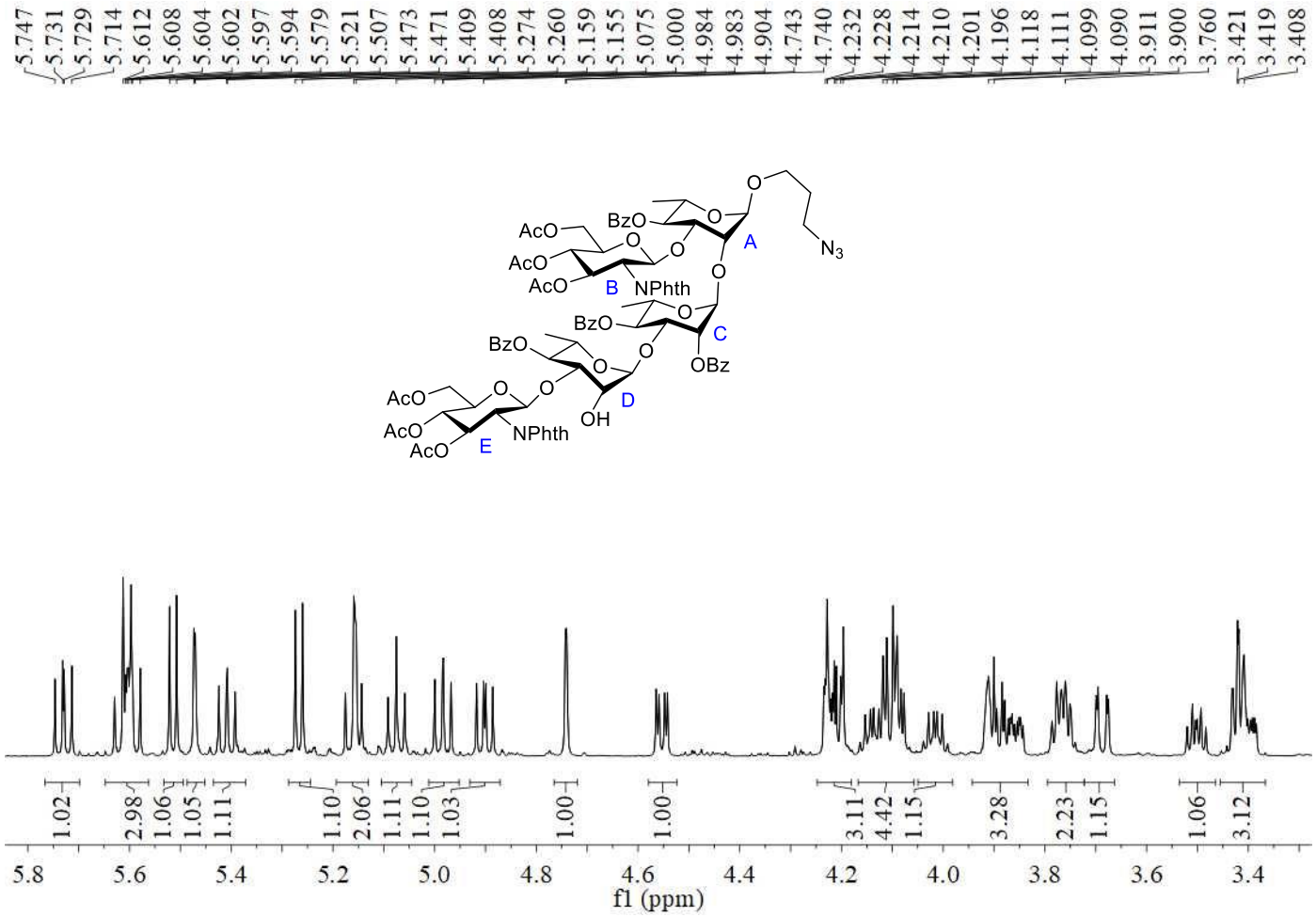
^{1}H-^{13}C HSQC spectrum of compound **23** (600/150 MHz, $CDCl_3$)



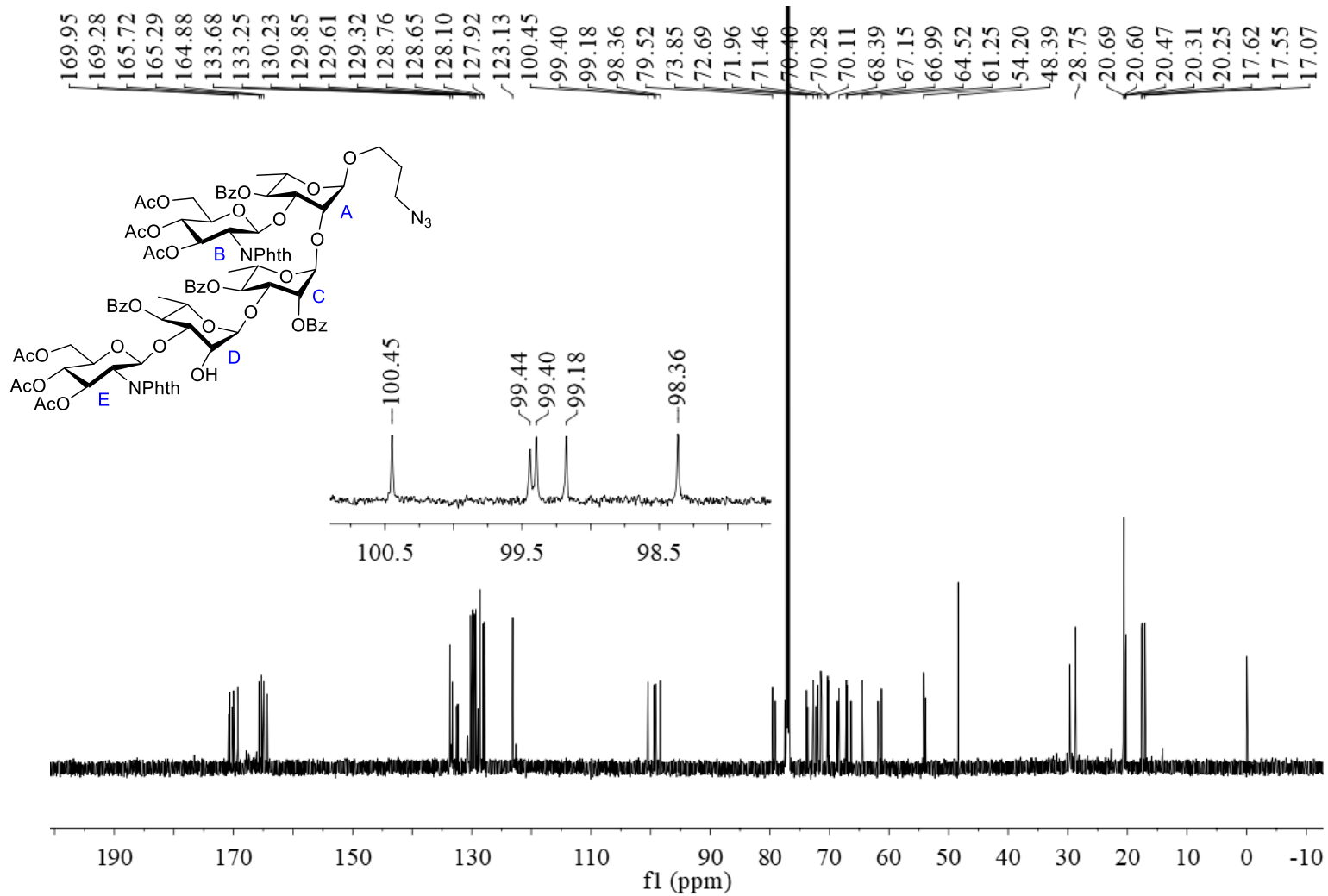
HR-ESI-(+) mass spectrum of compound **23**



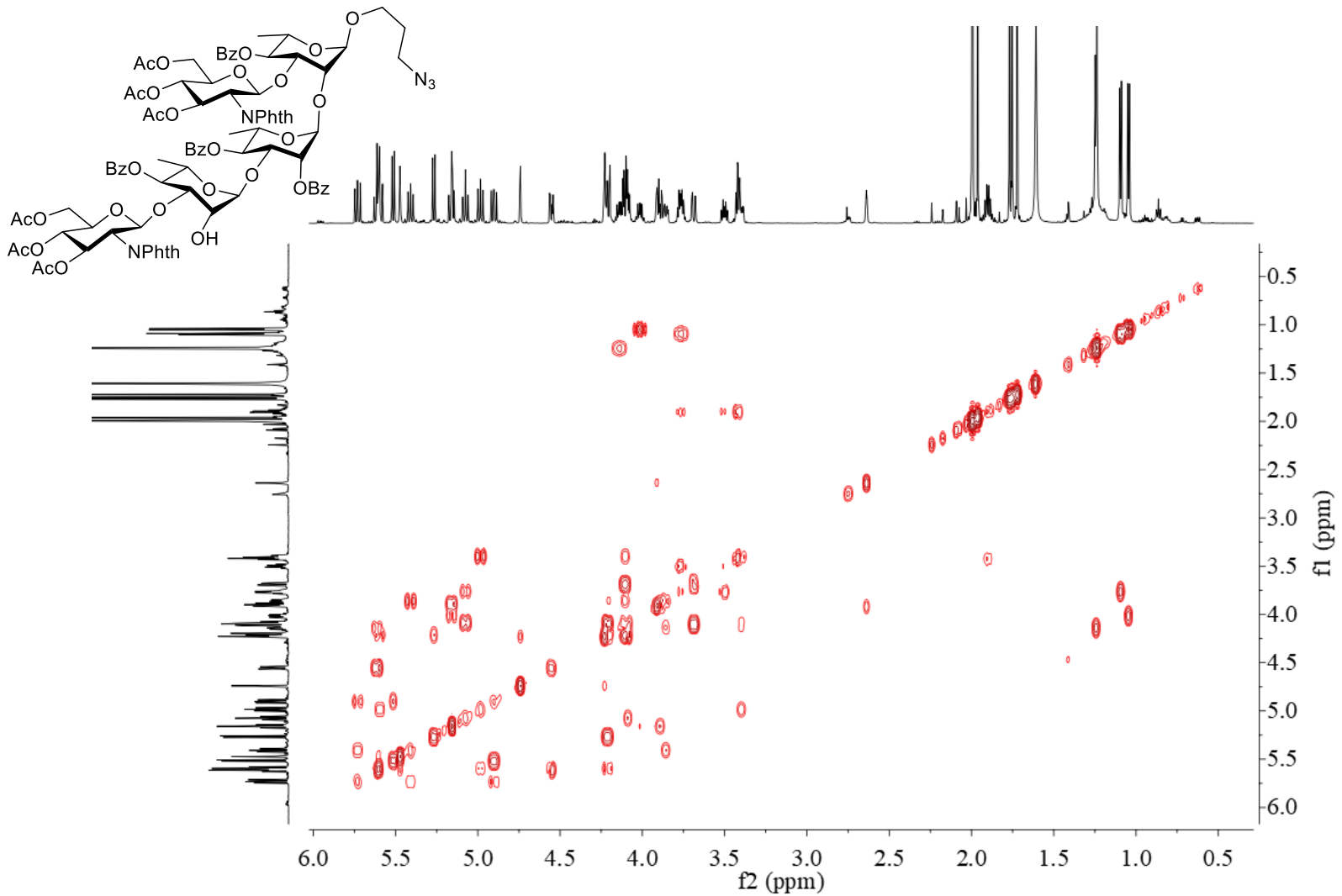
^{1}H NMR spectrum of compound **24** (600 MHz, $CDCl_3$)



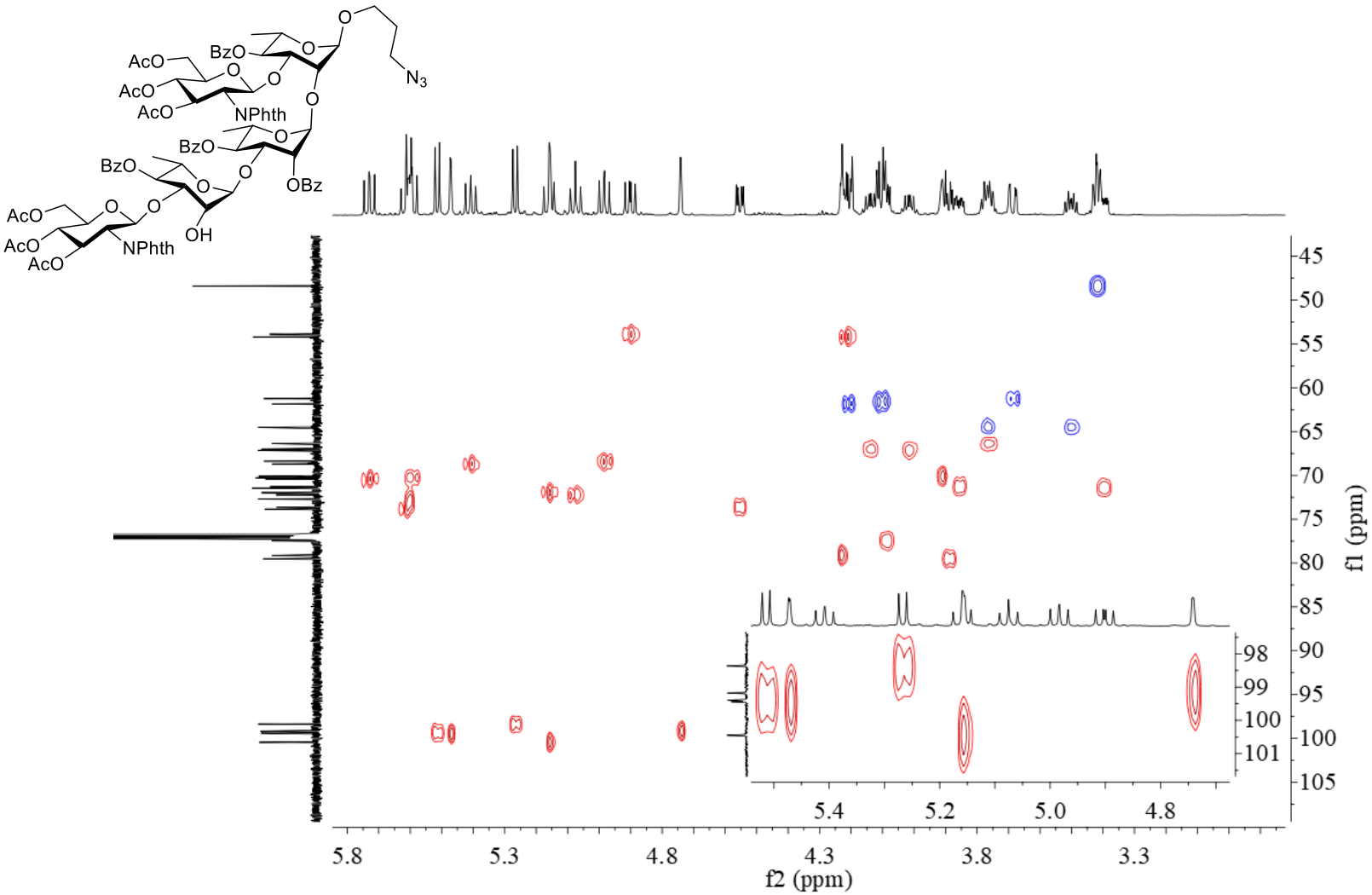
^{1}H NMR spectrum of compound **24** (expanded sugar region, 600 MHz, $CDCl_3$)



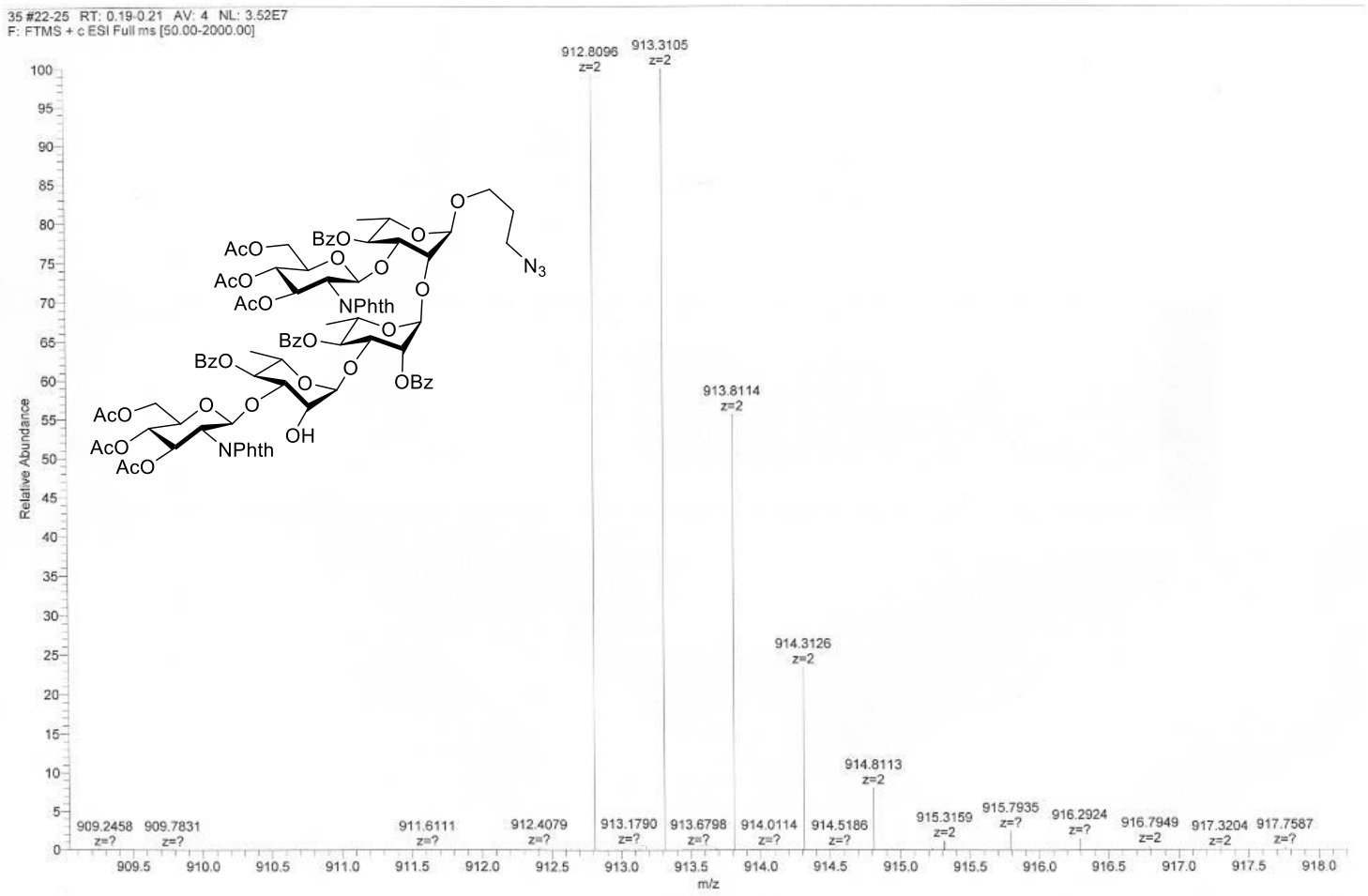
^{13}C NMR spectrum of compound **24** (150 MHz, $CDCl_3$)



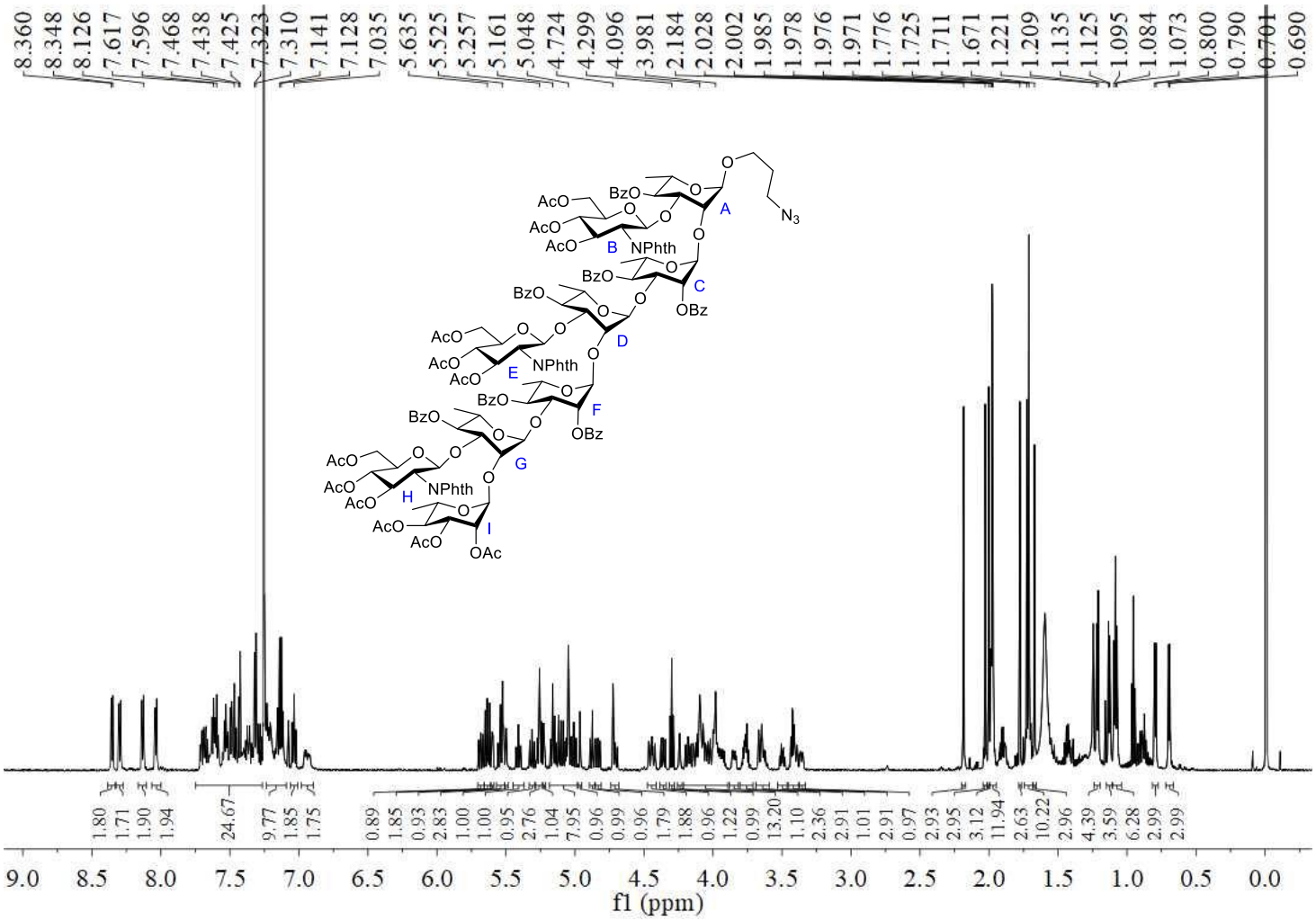
^{1}H-^{1}H COSY spectrum of compound **24** (600 MHz, $CDCl_3$)



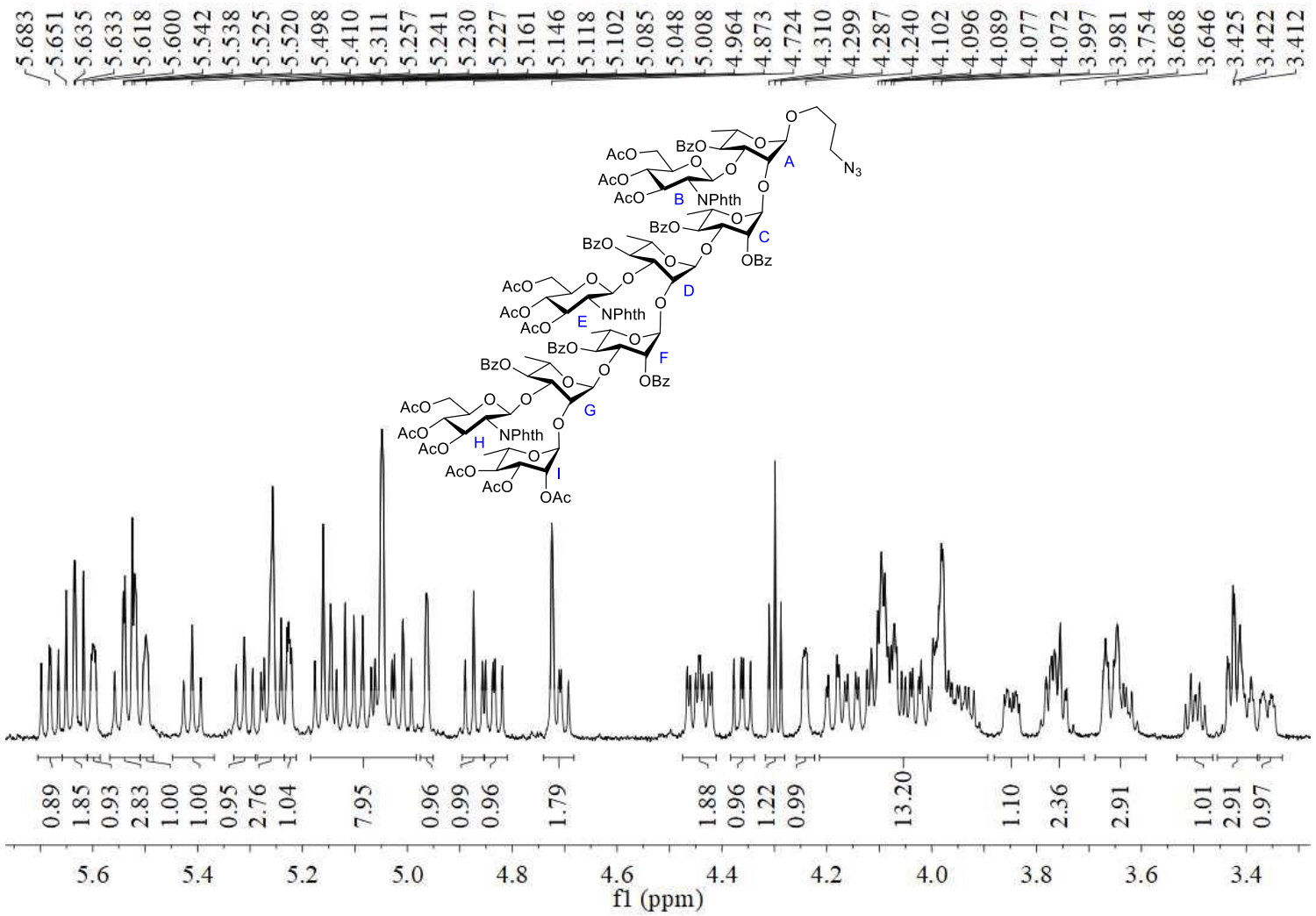
^{1}H-^{13}C HSQC spectrum of compound **24** (600/150 MHz, $CDCl_3$)



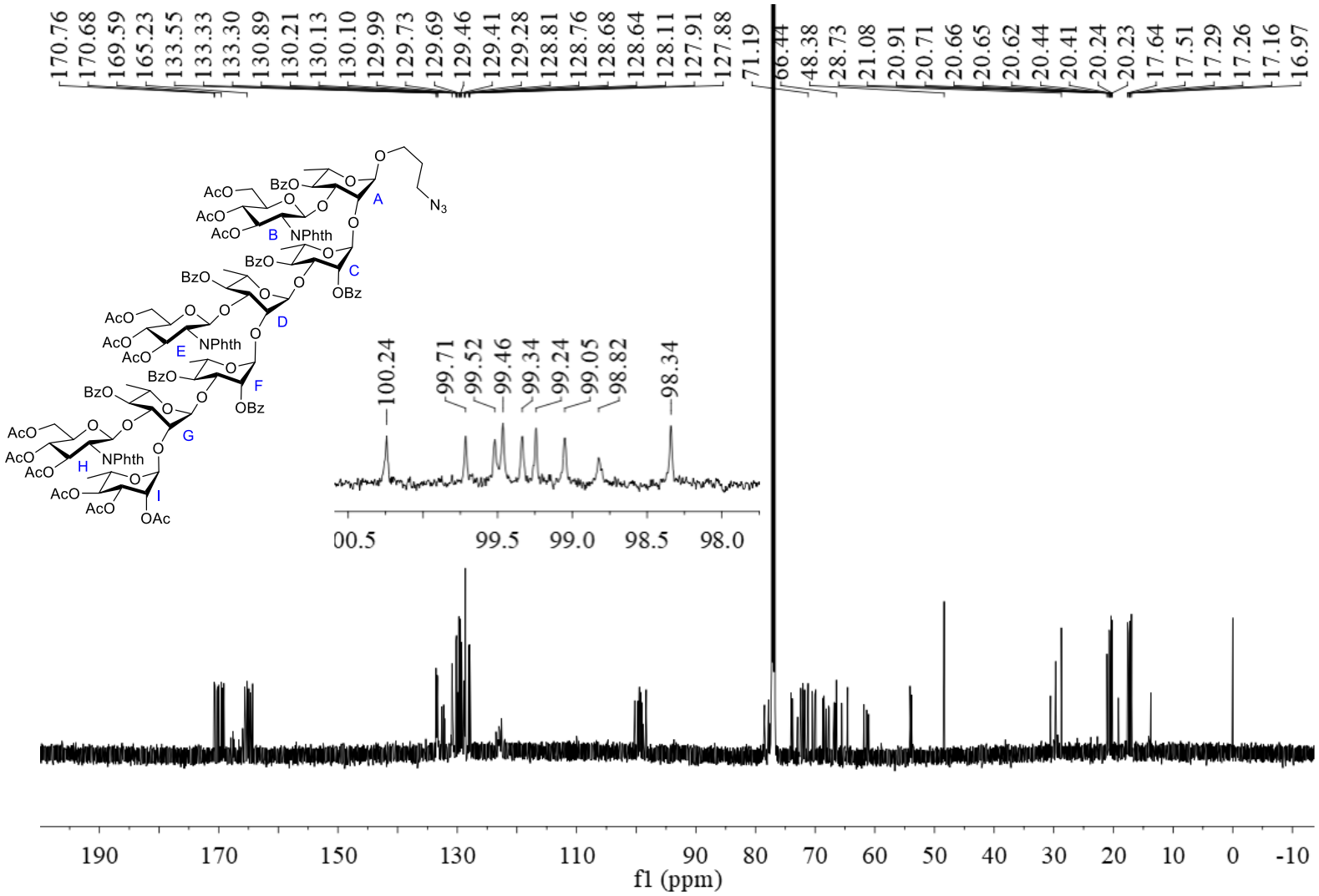
HR-ESI-(+) mass spectrum of compound **24**



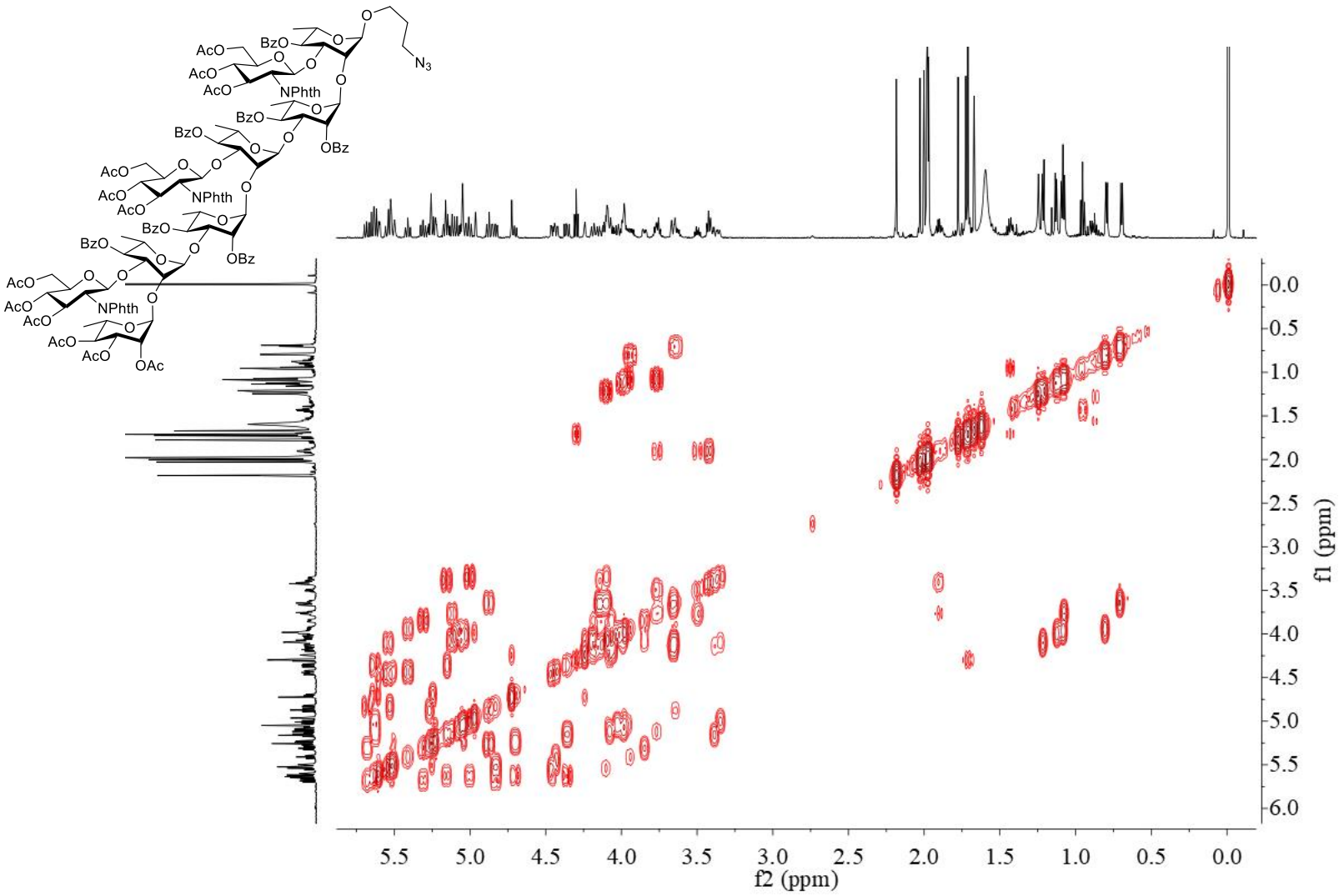
^{1}H NMR spectrum of compound **25** (600 MHz, $CDCl_3$)



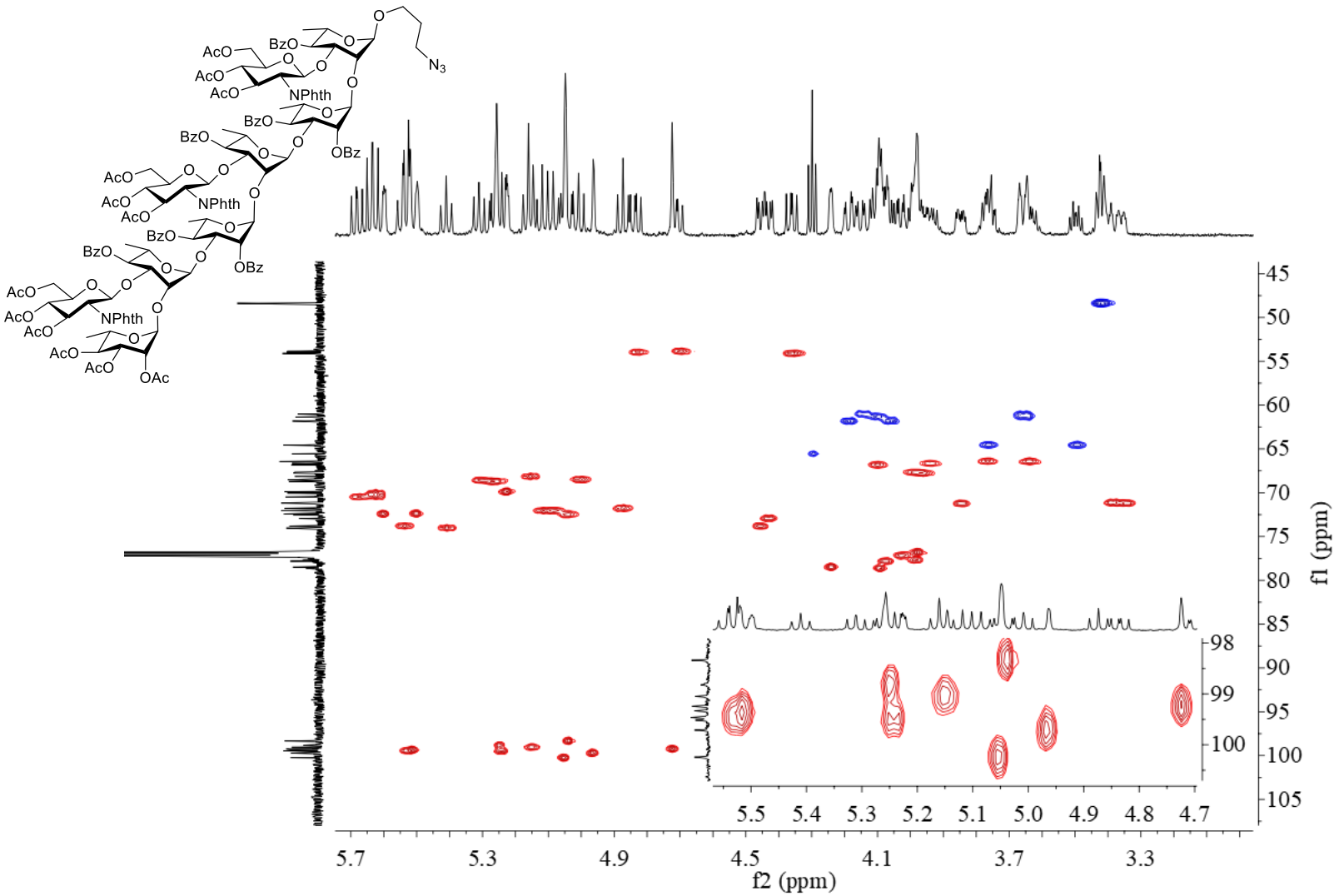
^{1}H NMR spectrum of compound **25** (expanded sugar region, 600 MHz, $CDCl_3$)



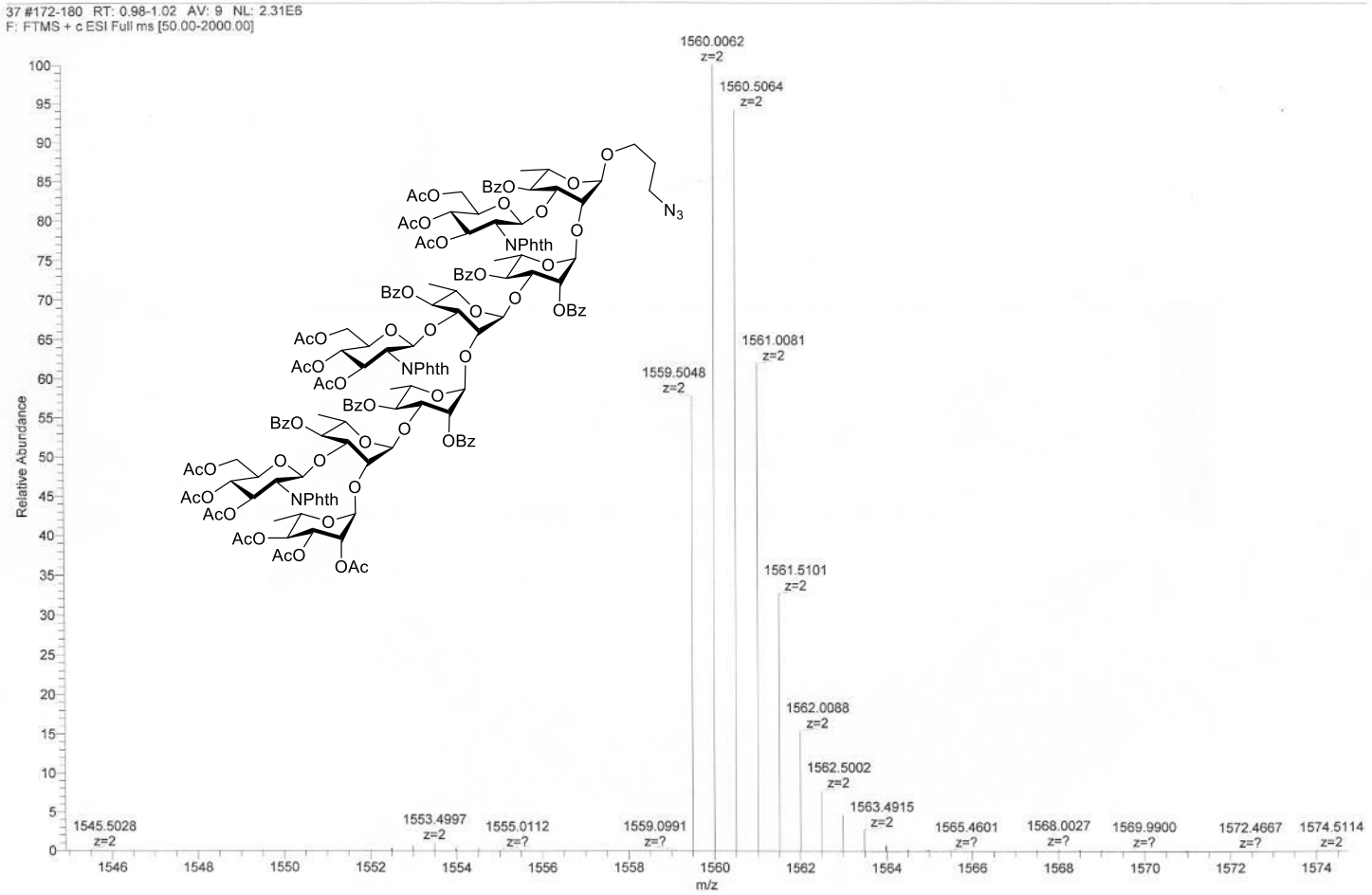
^{13}C NMR spectrum of compound **25** (150 MHz, $CDCl_3$)



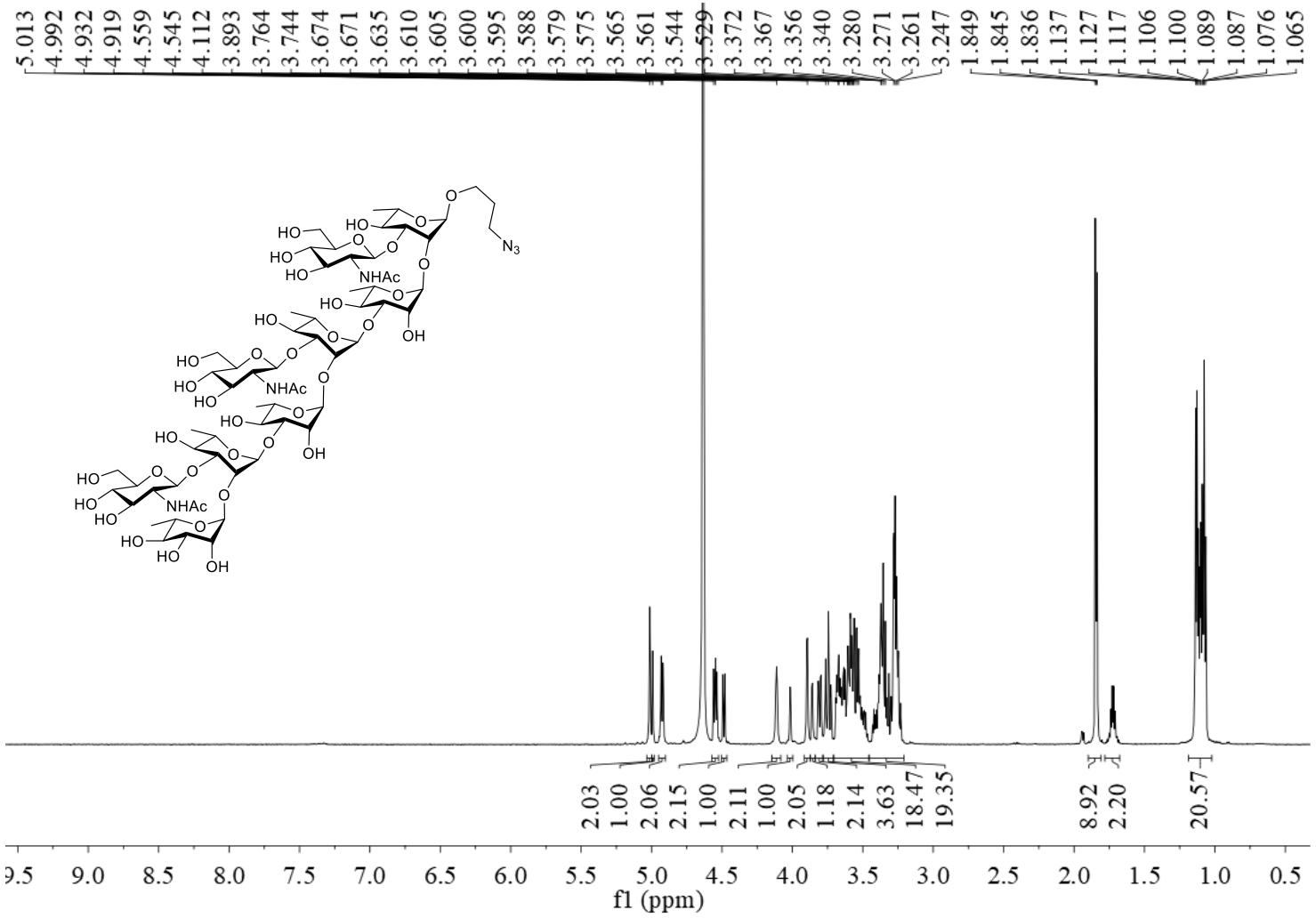
^{1}H-^{1}H COSY spectrum of compound **25** (600 MHz, $CDCl_3$)



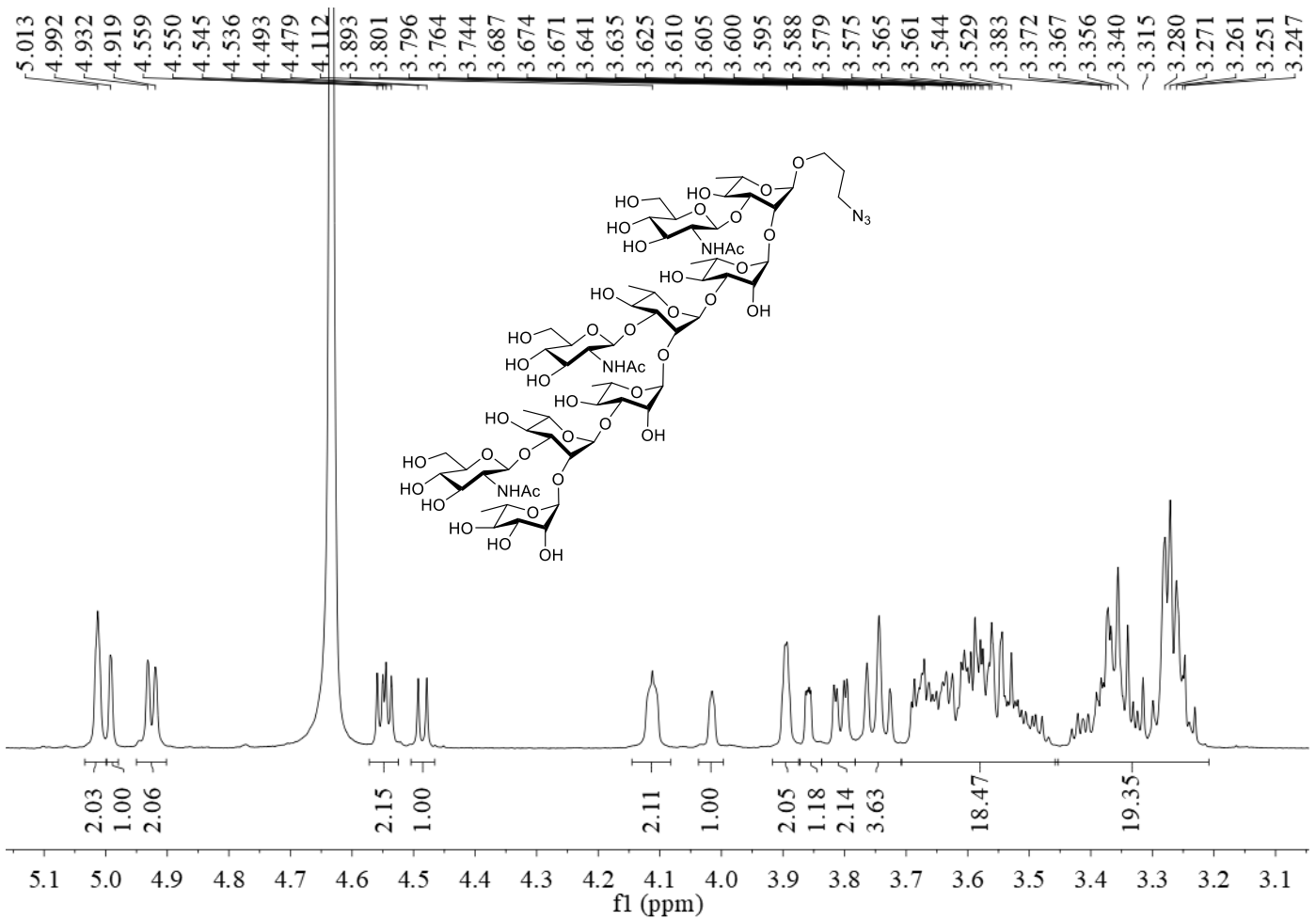
^{1}H-^{13}C HSQC spectrum of compound **25** (600/150 MHz, $CDCl_3$)



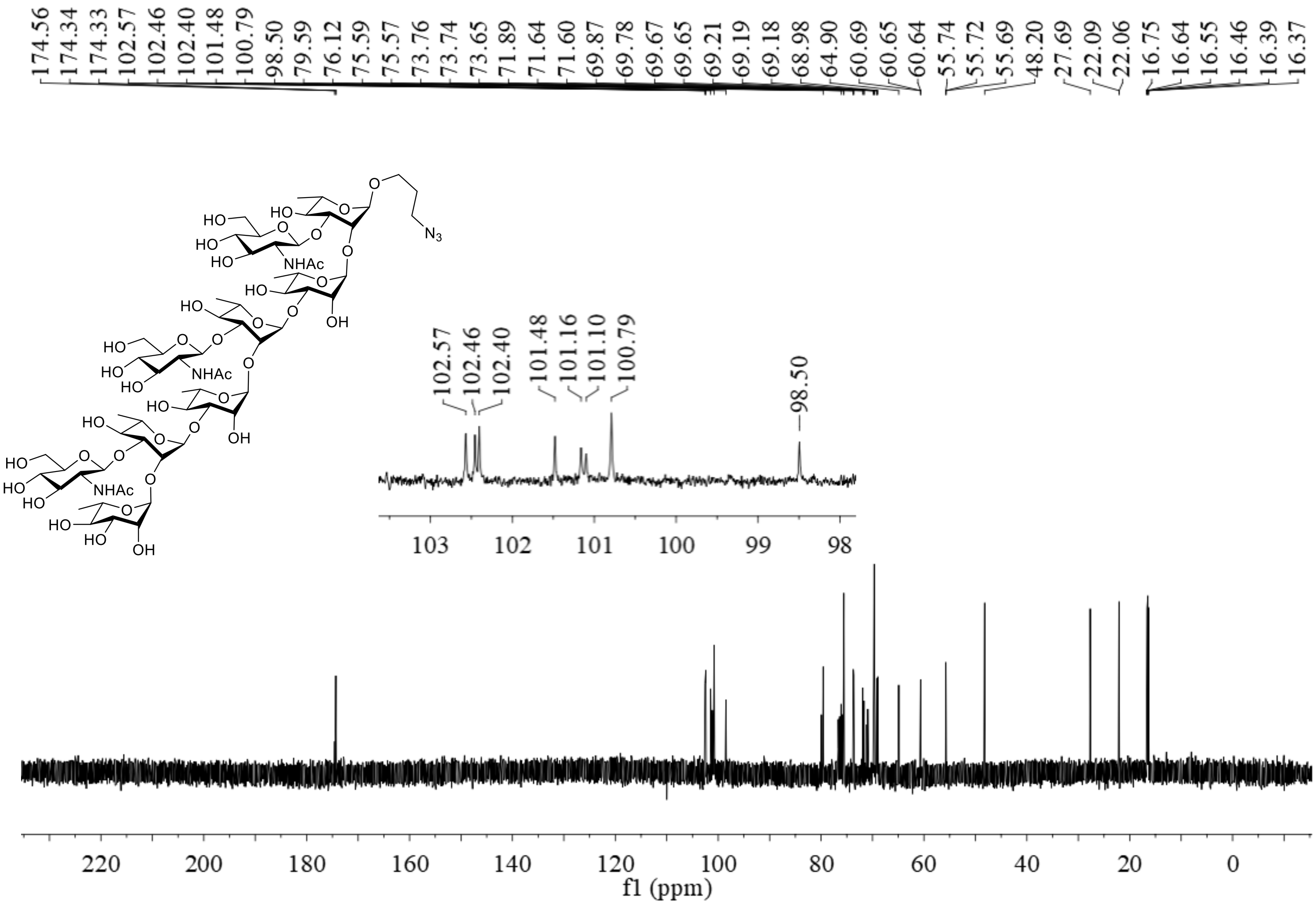
HR-ESI-(+) mass spectrum of compound **25**



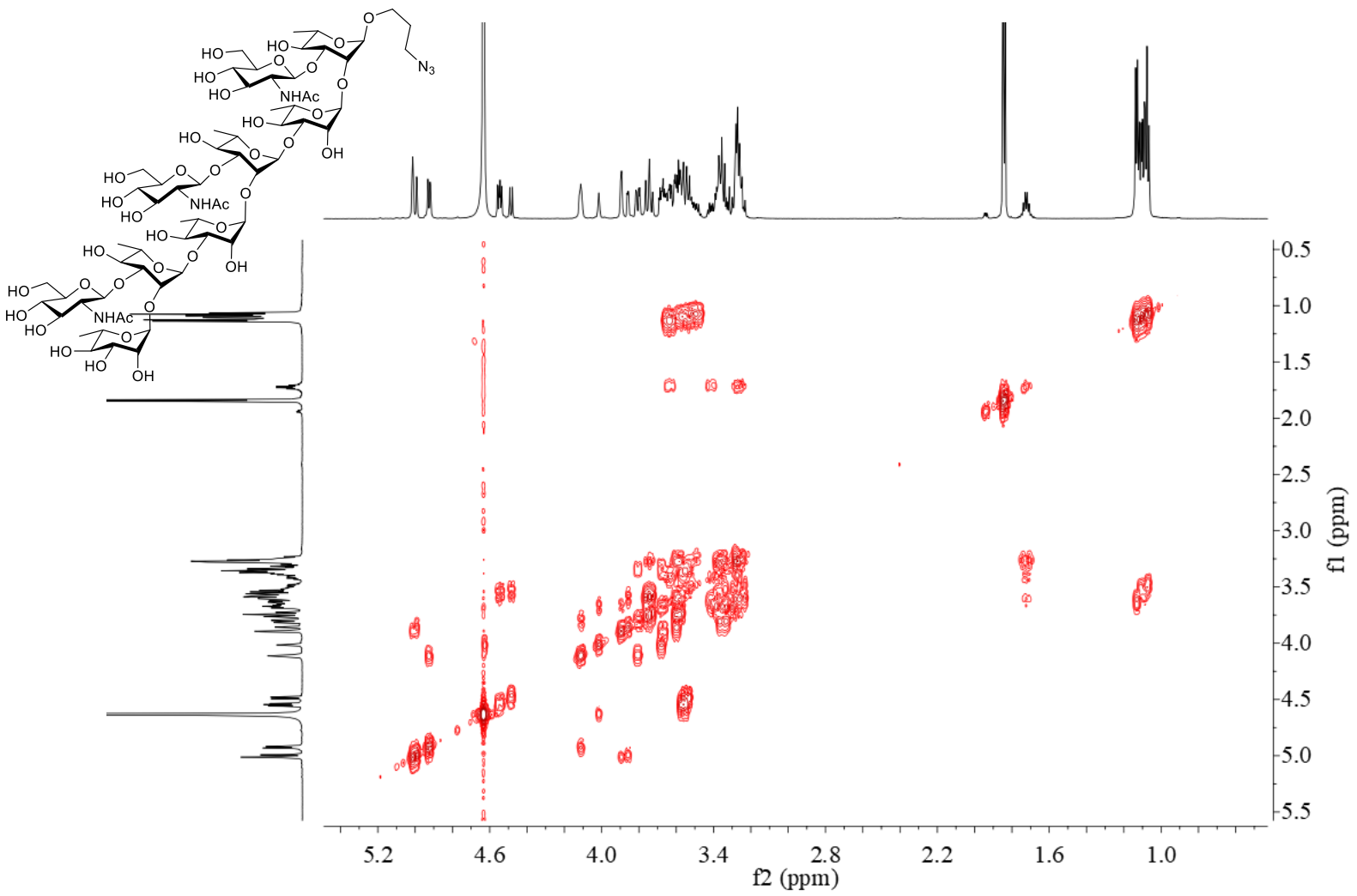
^{1}H NMR spectrum of compound **5c** (600 MHz, D_2O)



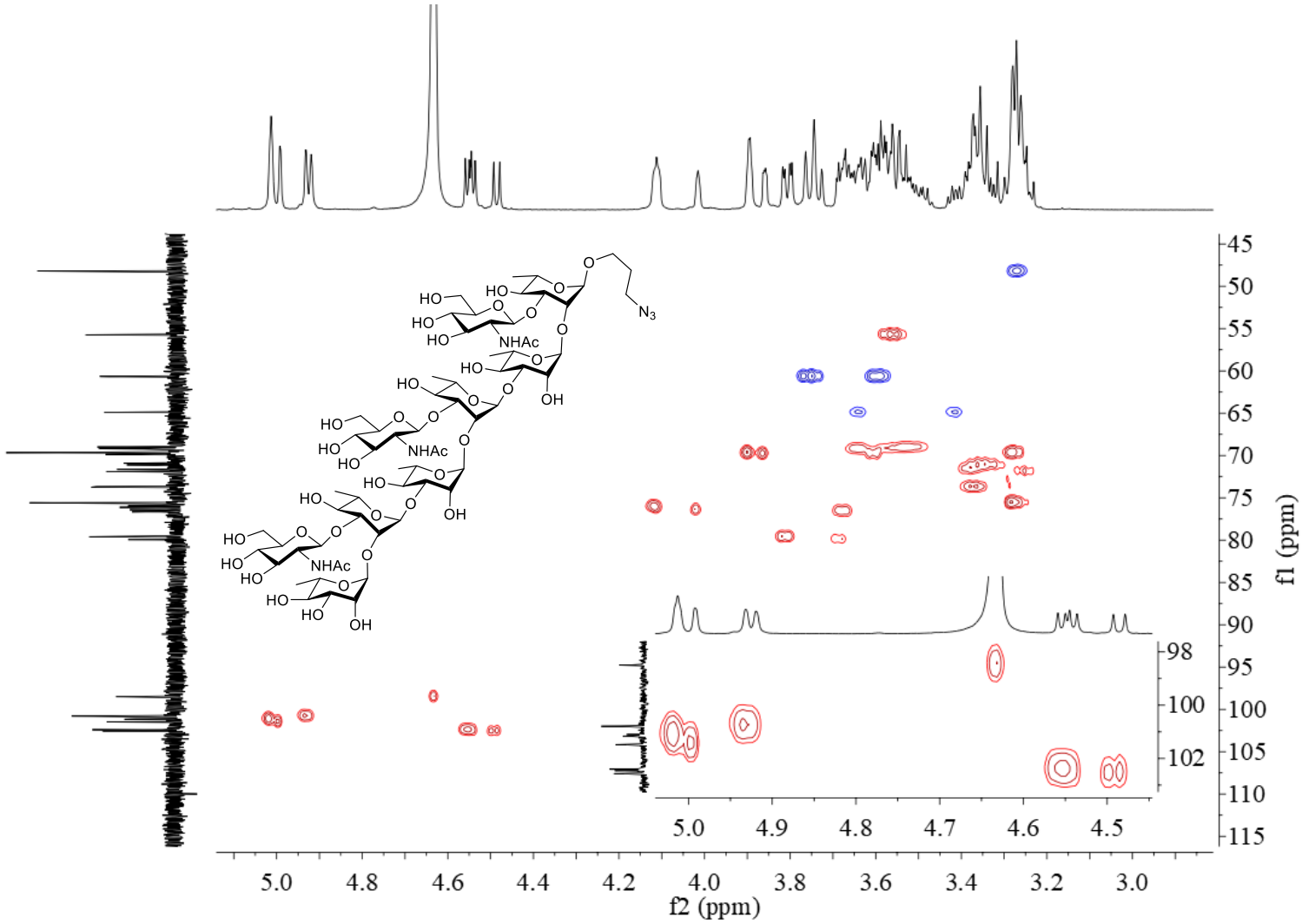
^{1}H NMR spectrum of compound **5c** (expanded sugar region, 600 MHz, D_2O)



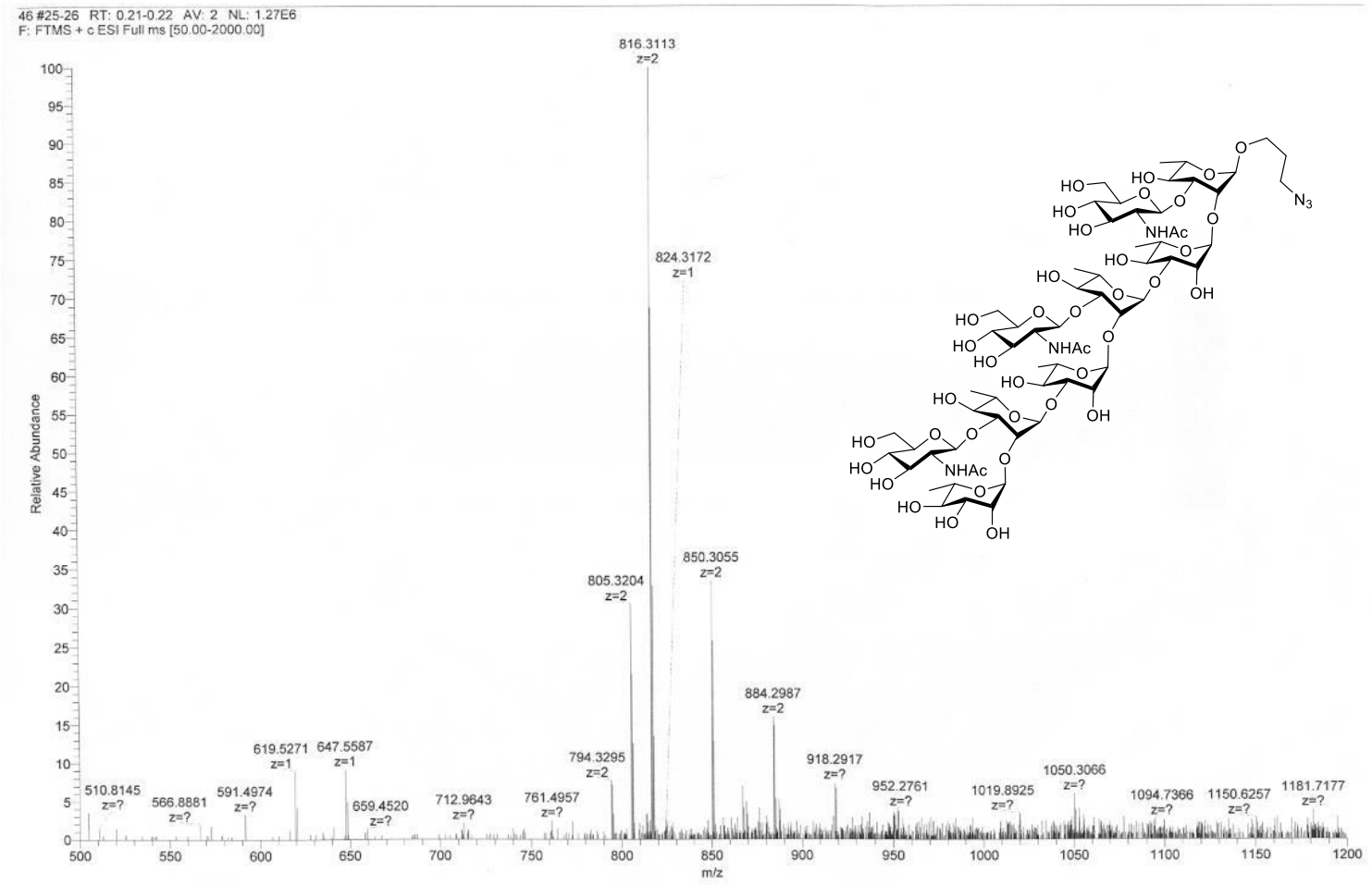
^{13}C NMR spectrum of compound **5c** (150 MHz, D_2O)



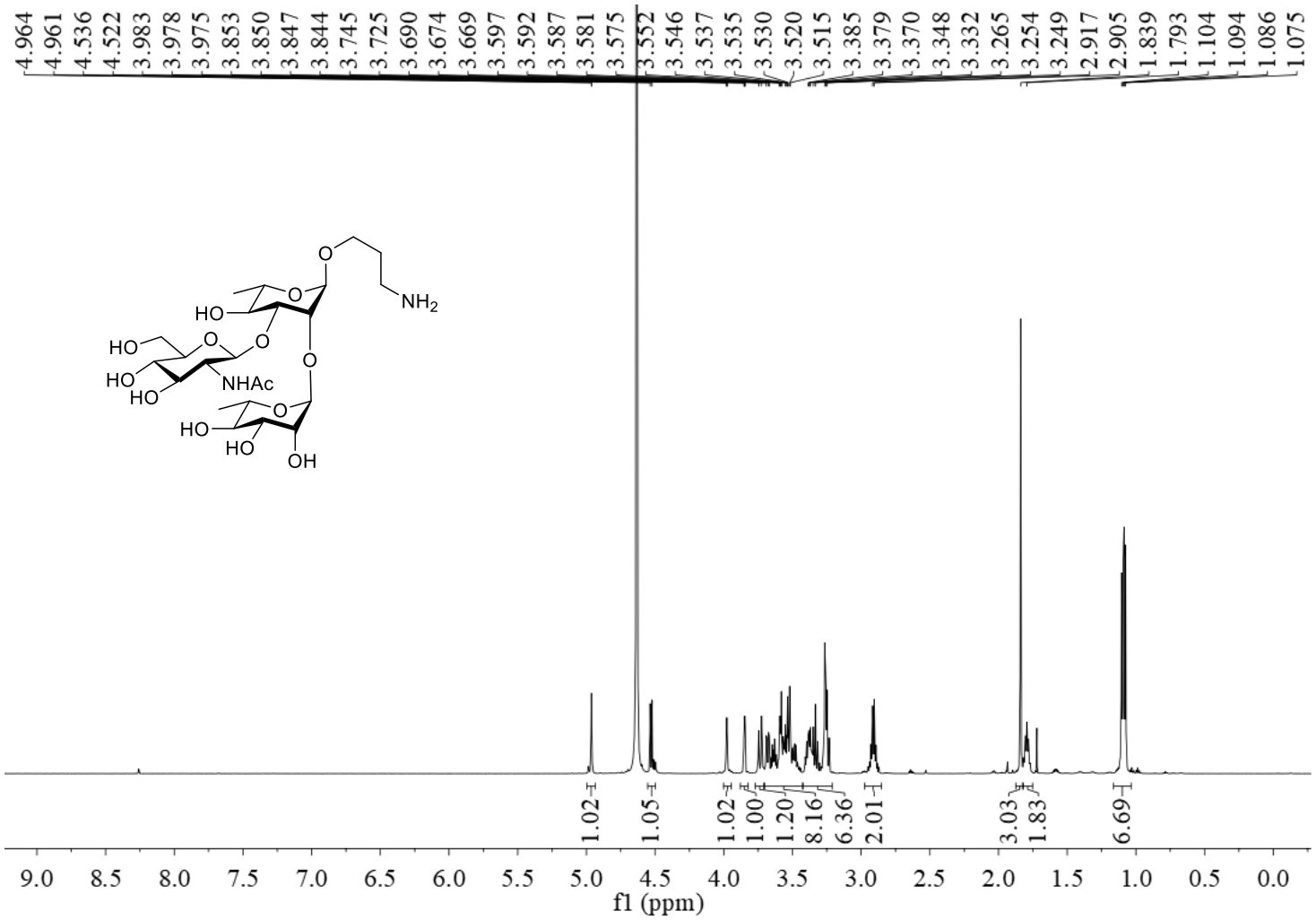
^{1}H-^{1}H COSY spectrum of compound **5c** (600 MHz, D_2O)



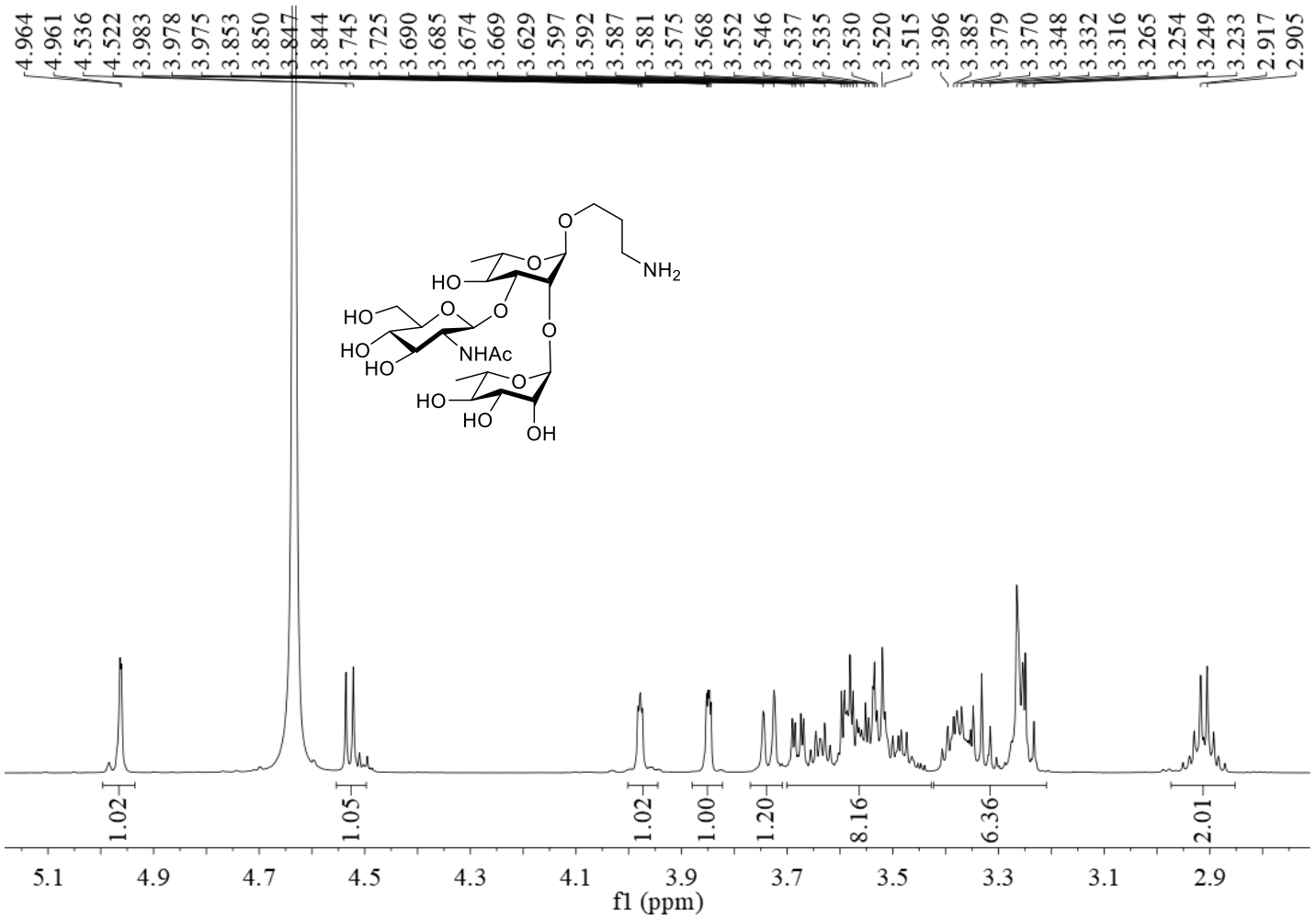
^{1}H-^{13}C HSQC spectrum of compound **5c** (600/150 MHz, D_2O)



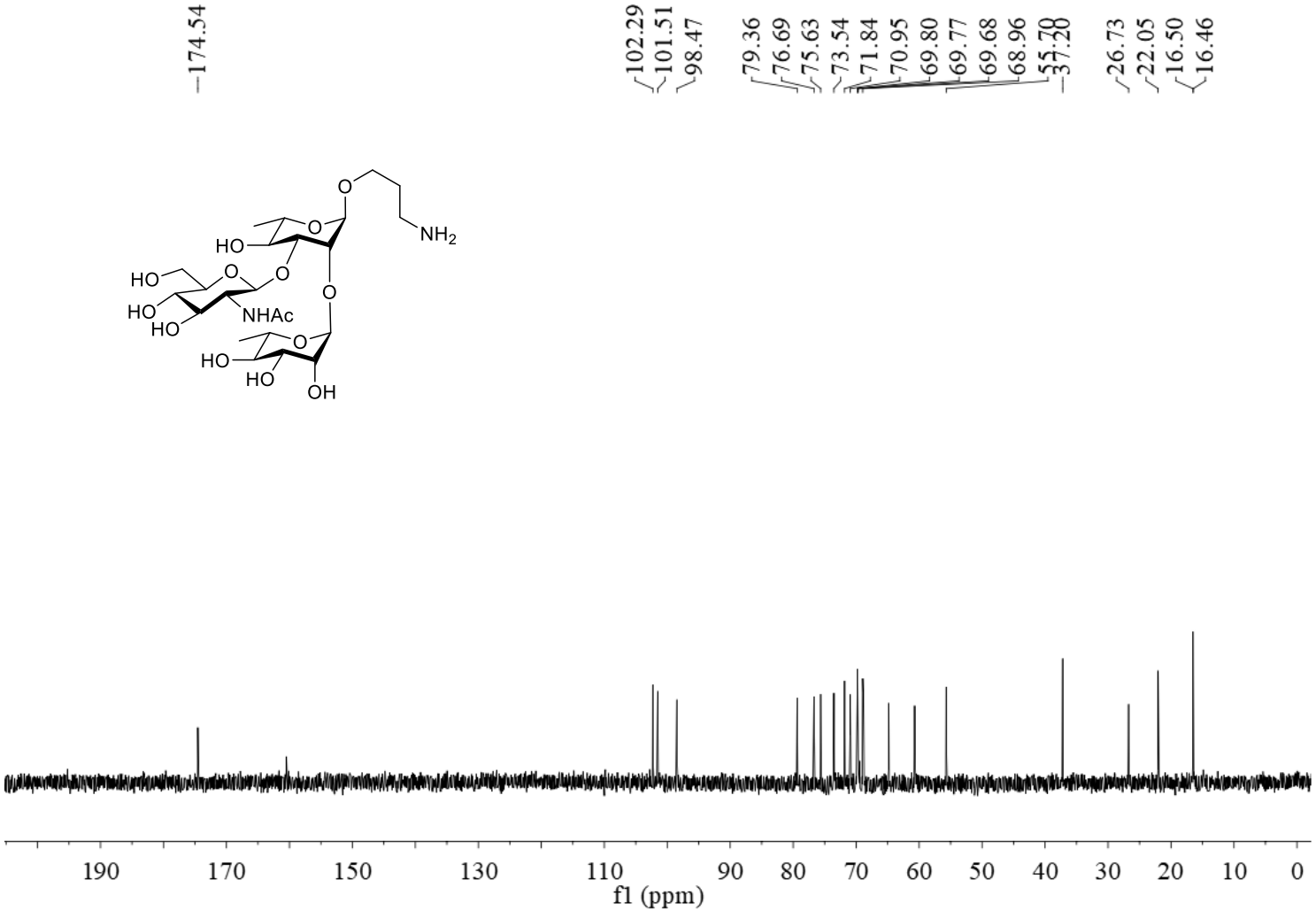
HR-ESI-(+) mass spectrum of compound **5c**



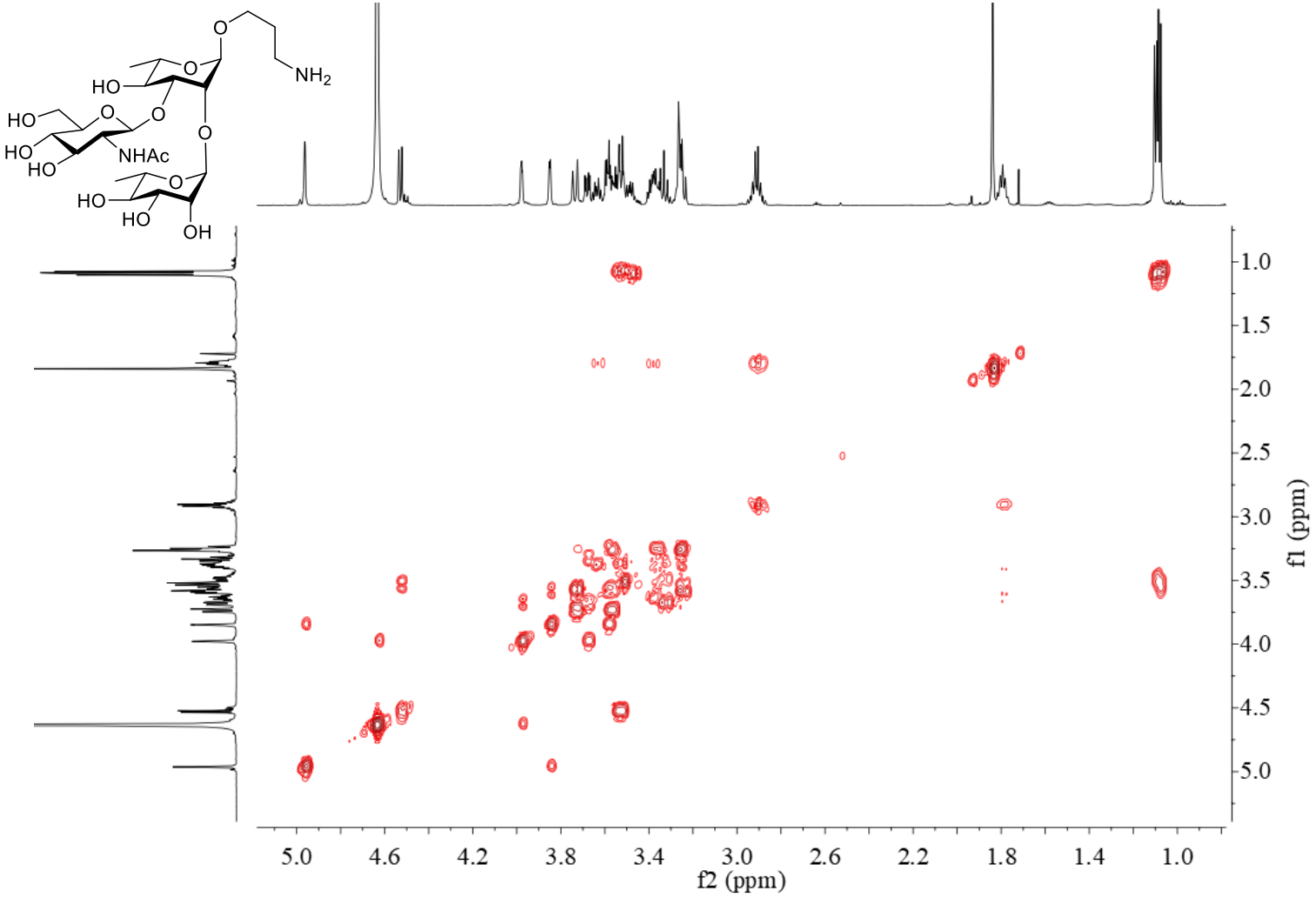
^{1}H NMR spectrum of compound **26a** (600 MHz, D_2O)



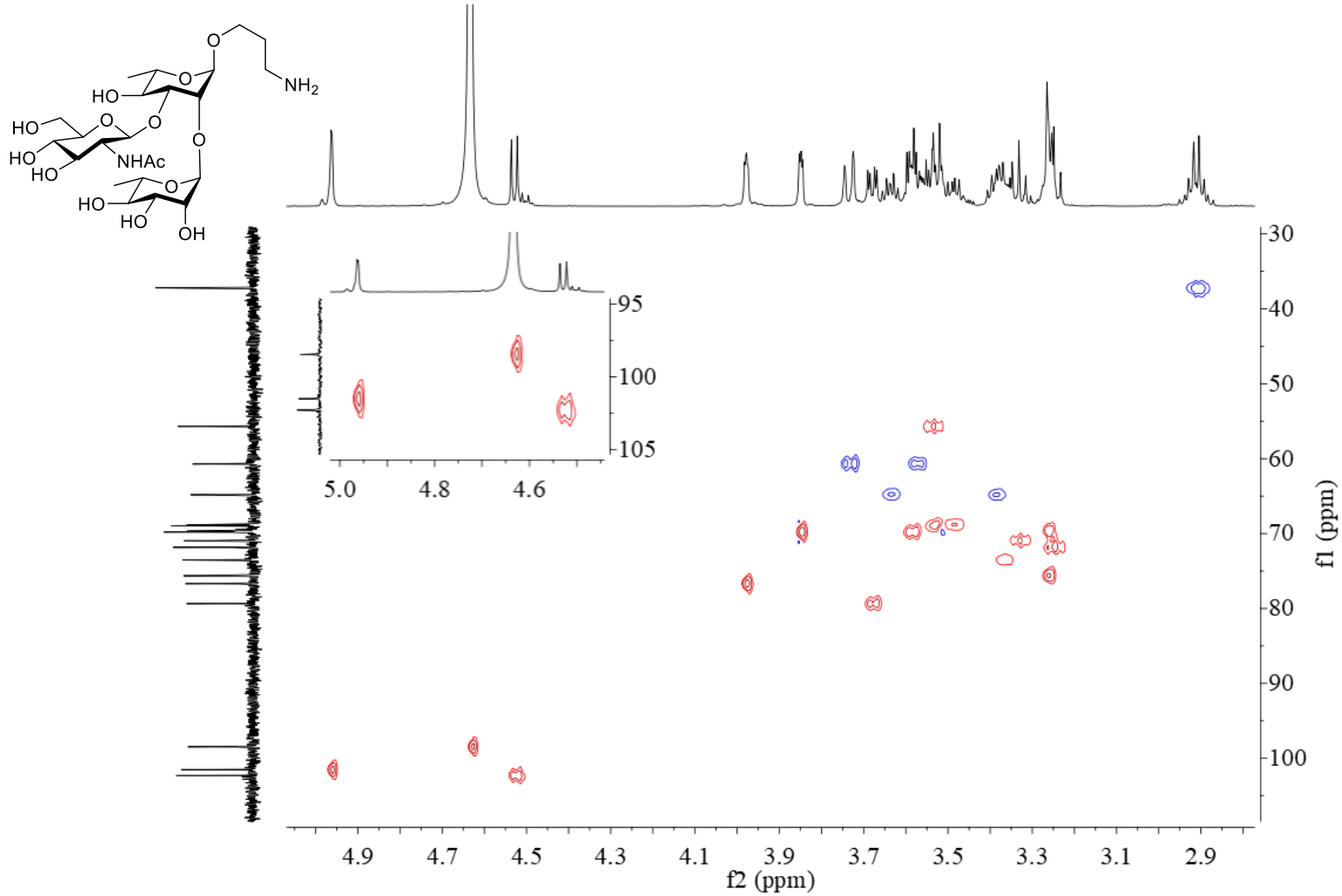
^{1}H NMR spectrum of compound **26a** (expanded sugar region, 600 MHz, D_2O)



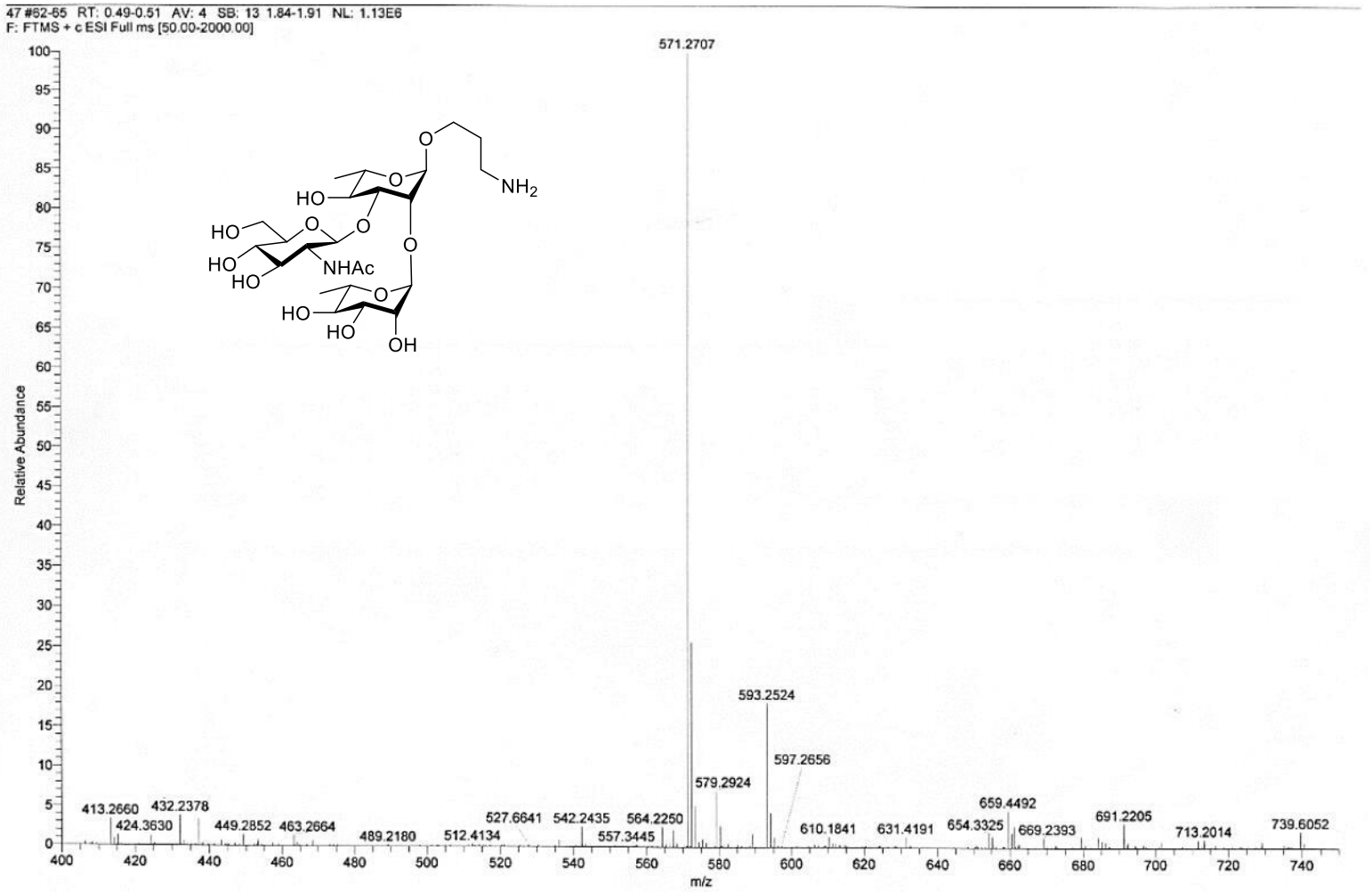
^{13}C NMR spectrum of compound **26a** (150 MHz, D_2O)



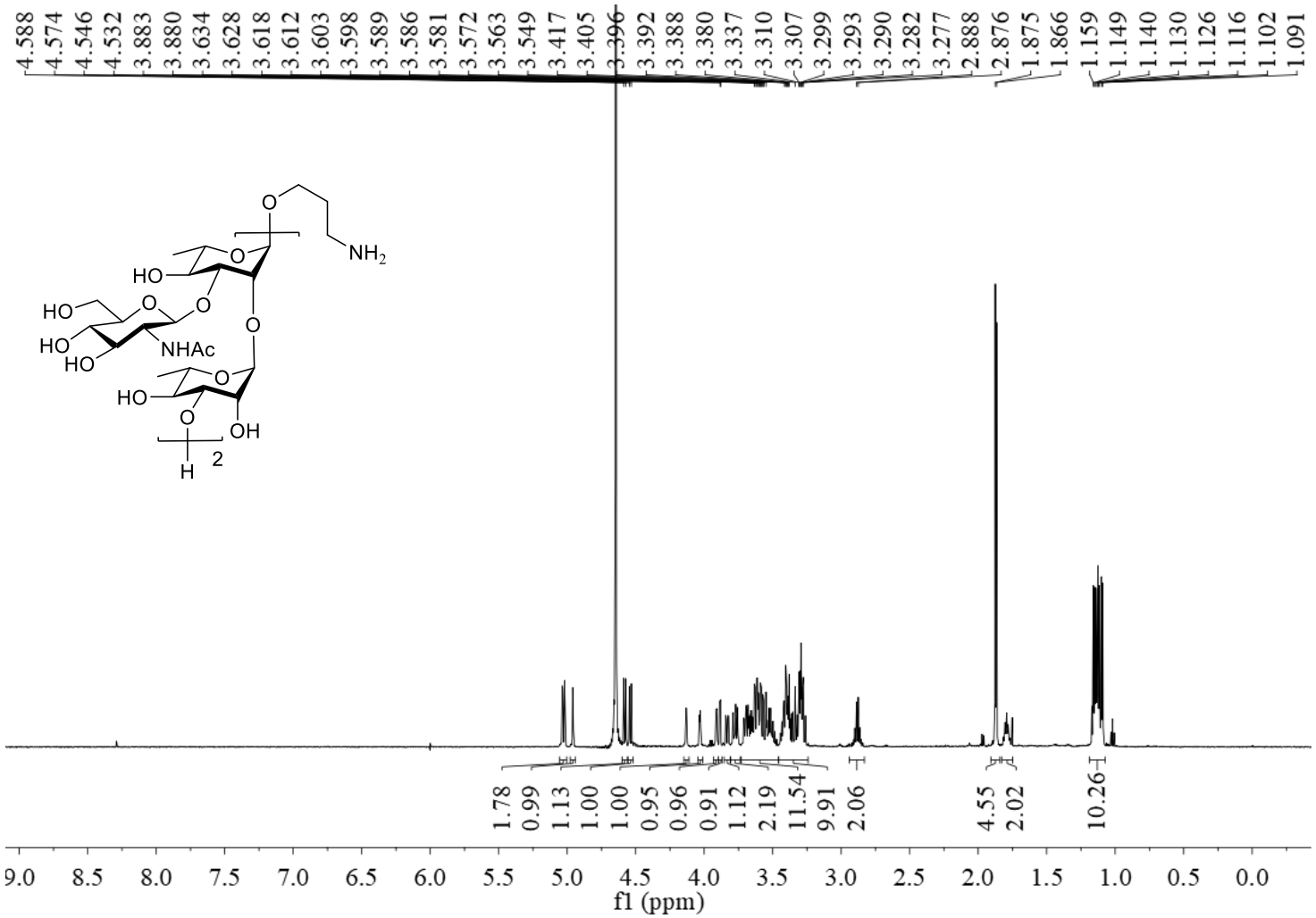
^{1}H-^{1}H COSY spectrum of compound **26a** (600 MHz, D_2O)



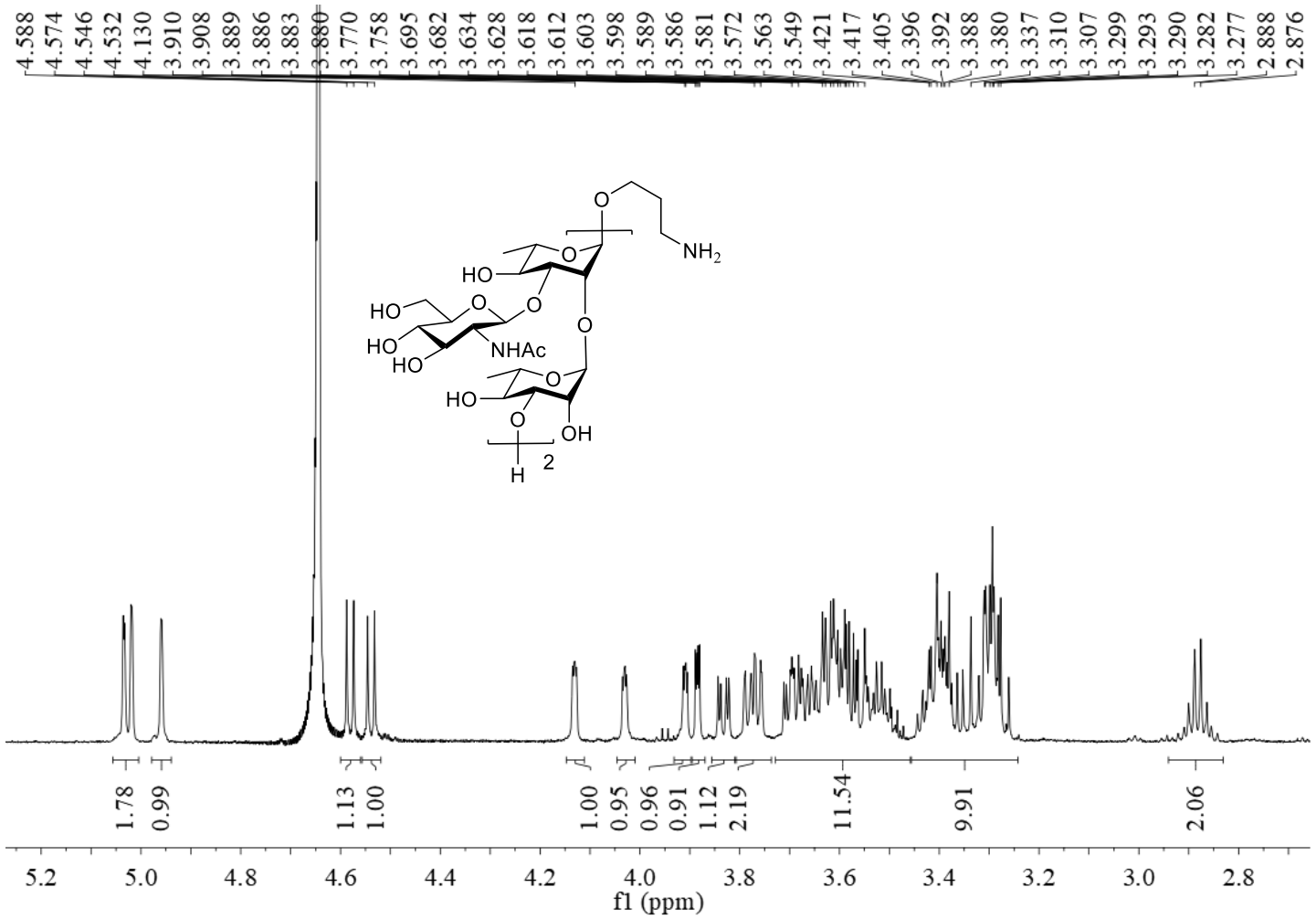
^{1}H-^{13}C HSQC spectrum of compound **26a** (600/150 MHz, D_2O)



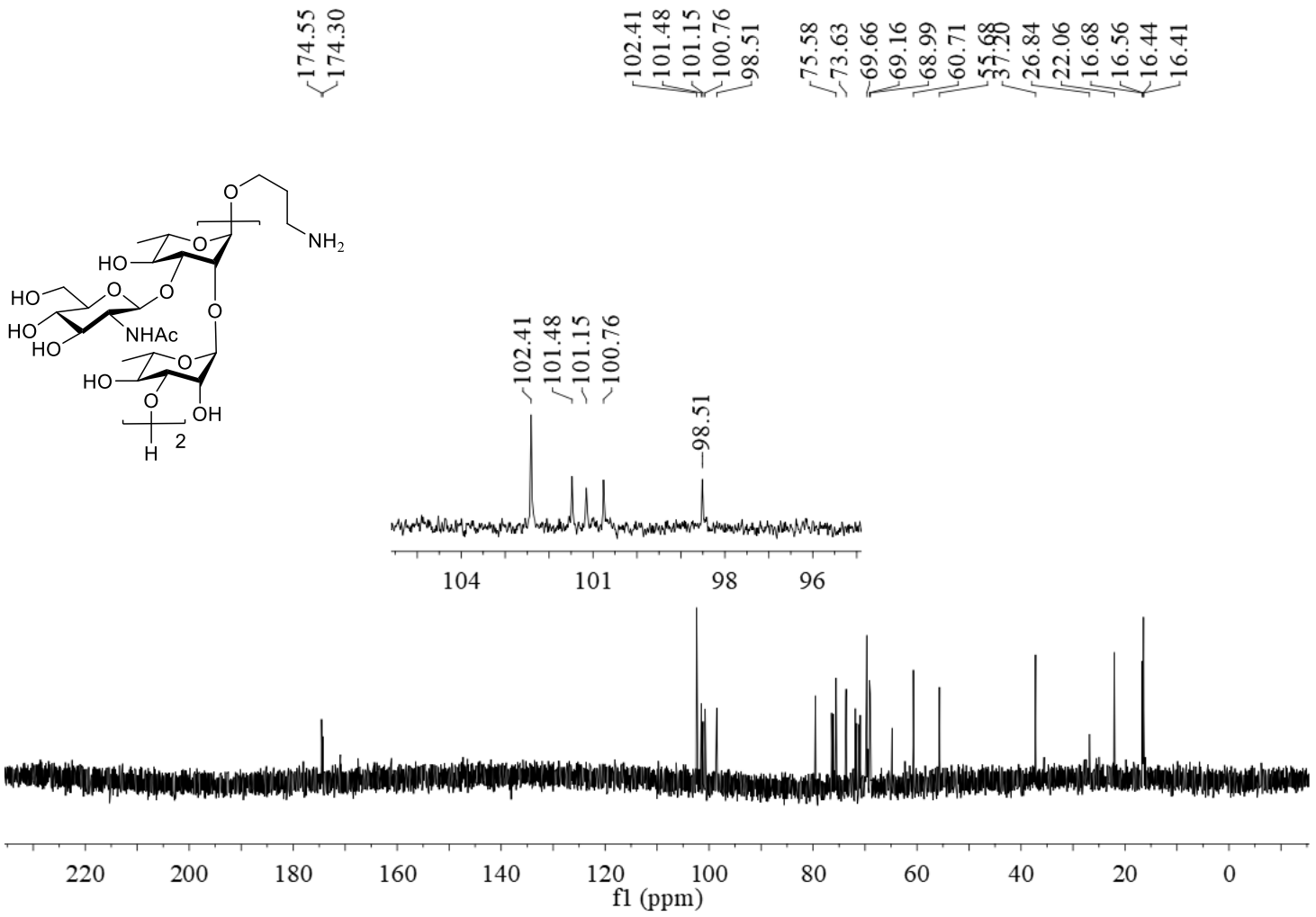
HR-ESI-(+) mass spectrum of compound **26a**



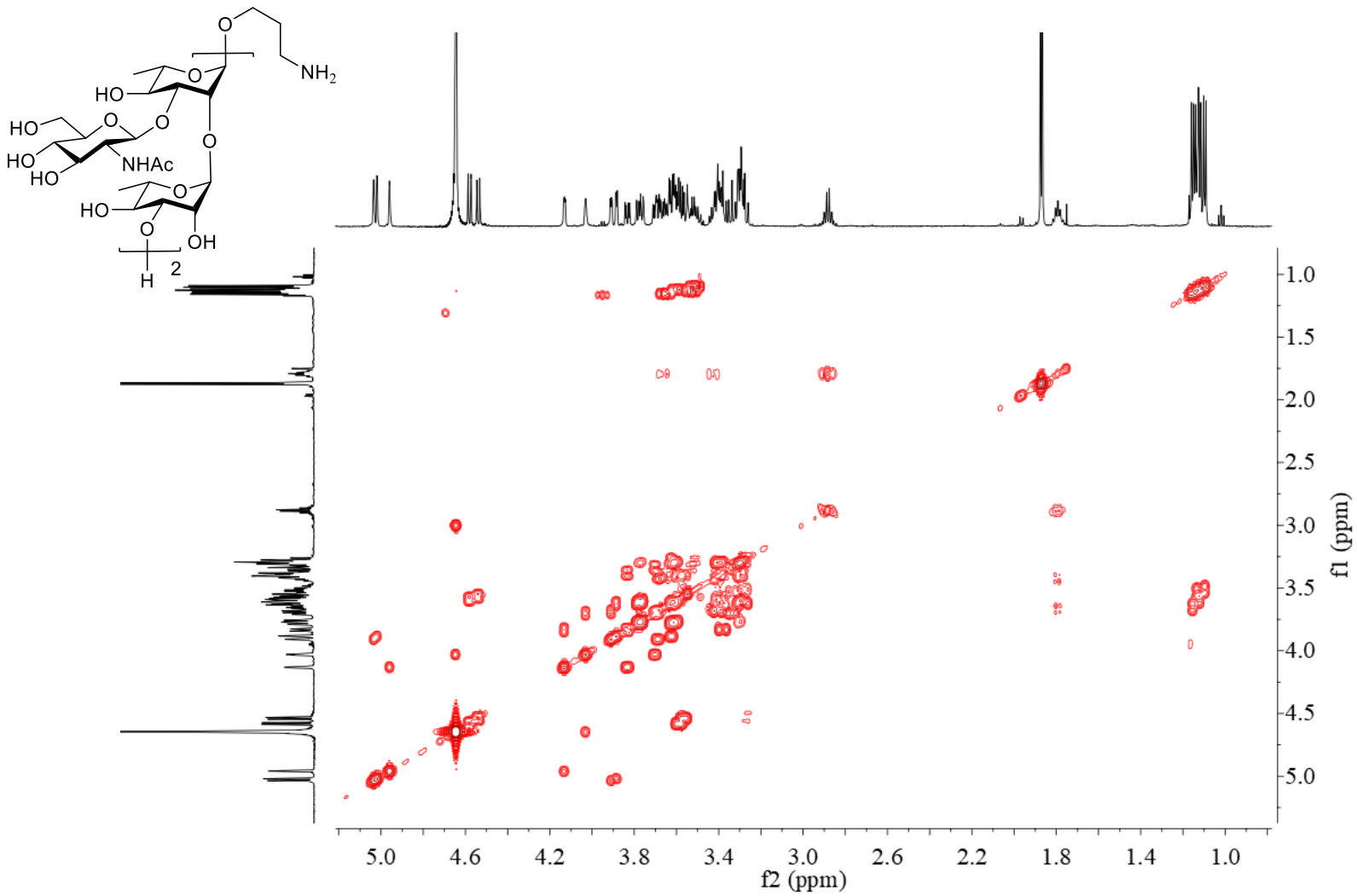
^{1}H NMR spectrum of compound **26b** (600 MHz, D_2O)



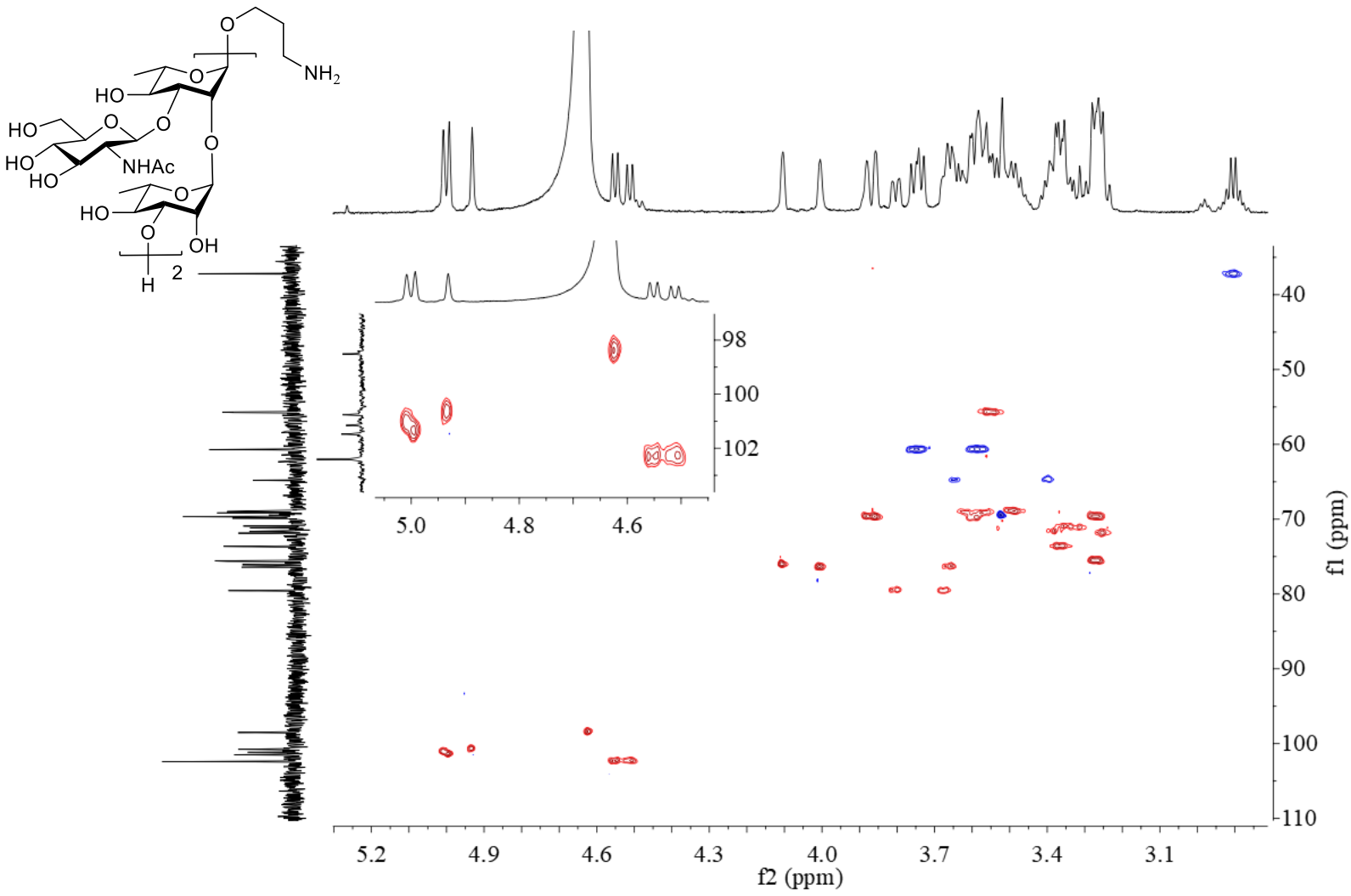
^{1}H NMR spectrum of compound **26b** (expanded sugar region, 600 MHz, D_2O)



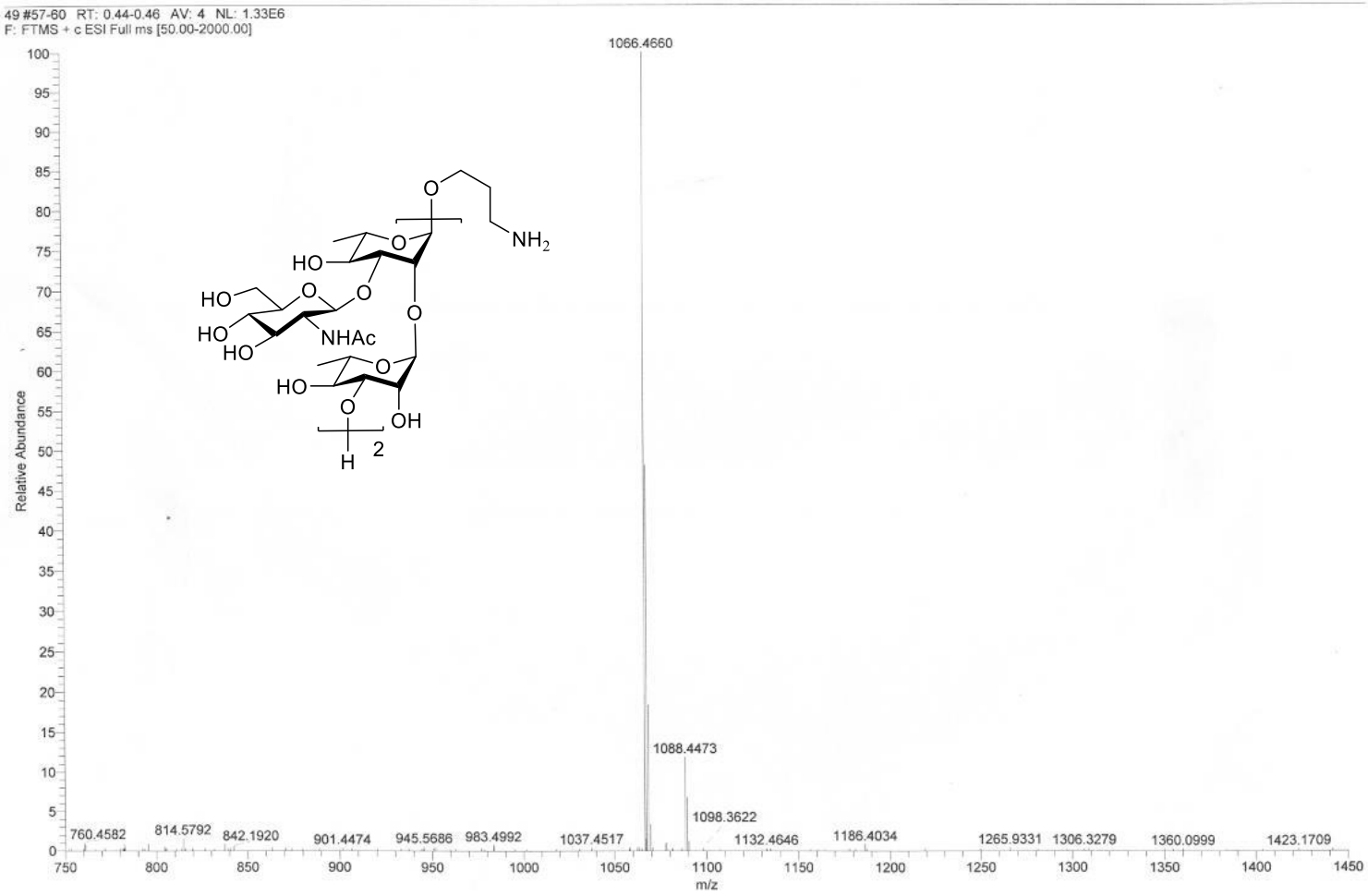
^{13}C NMR spectrum of compound **26b** (150 MHz, D_2O)



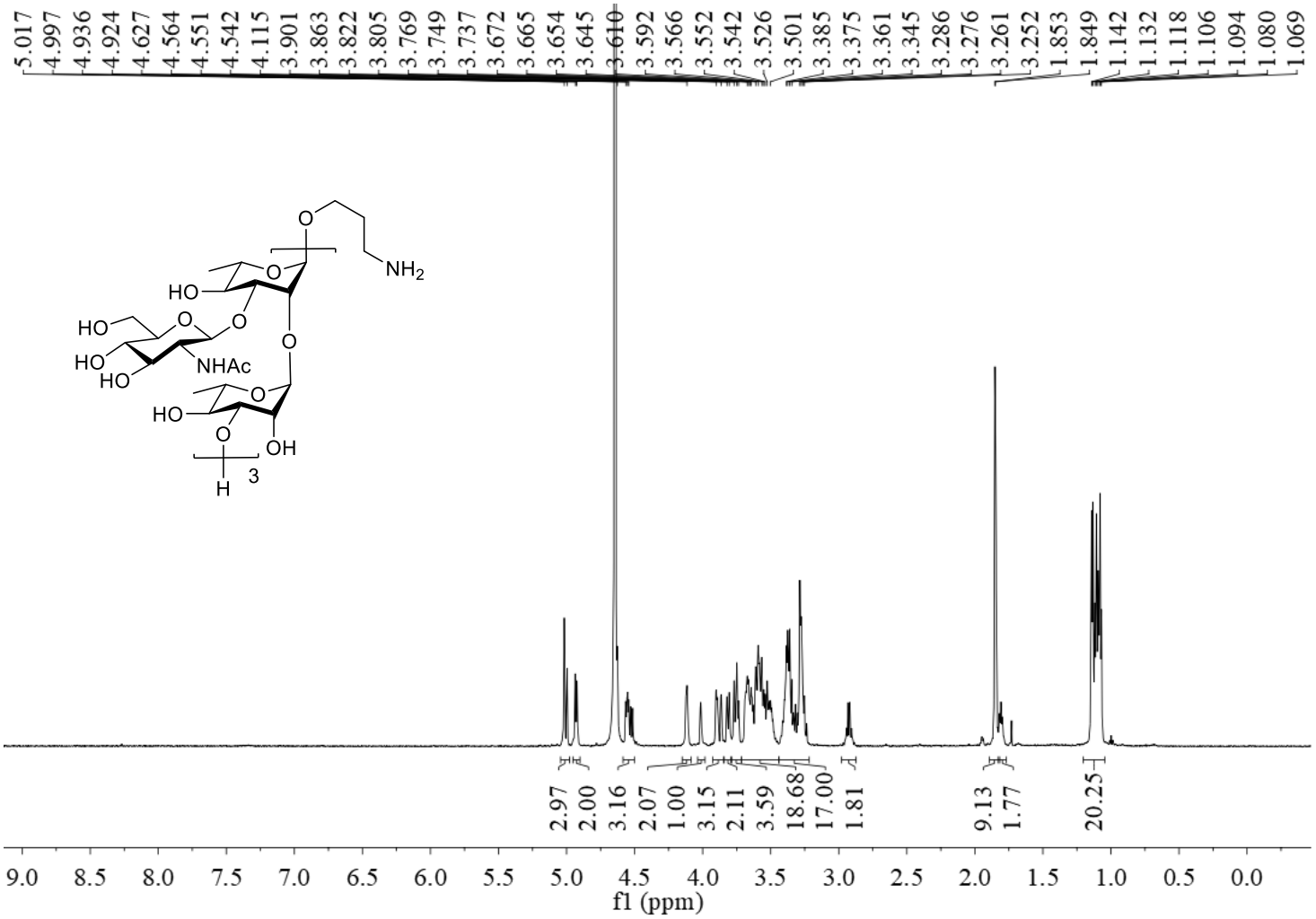
^{1}H-^{1}H COSY spectrum of compound **26b** (600 MHz, D_2O)



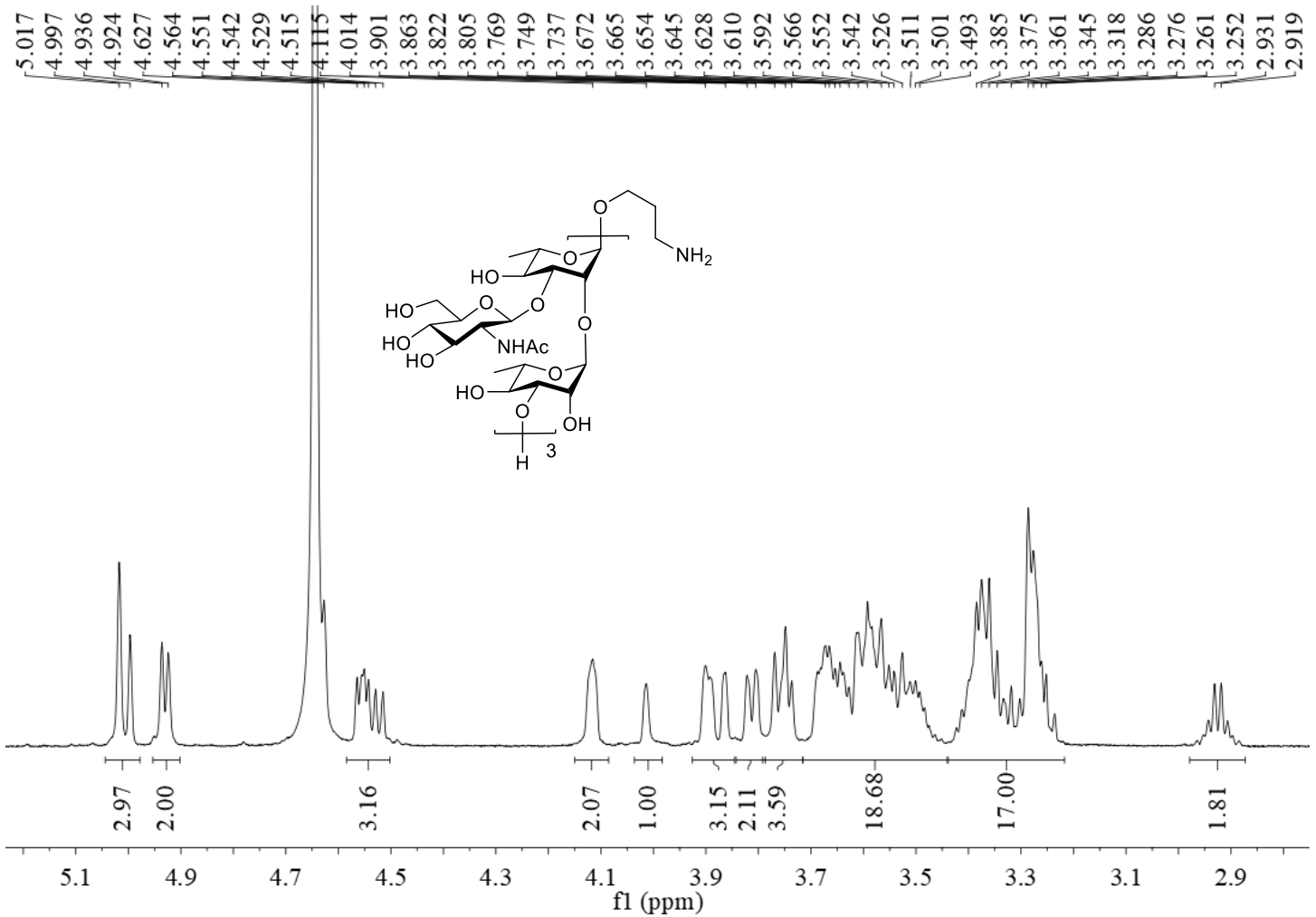
^{1}H-^{13}C HSQC spectrum of compound **26b** (600/150 MHz, D_2O)



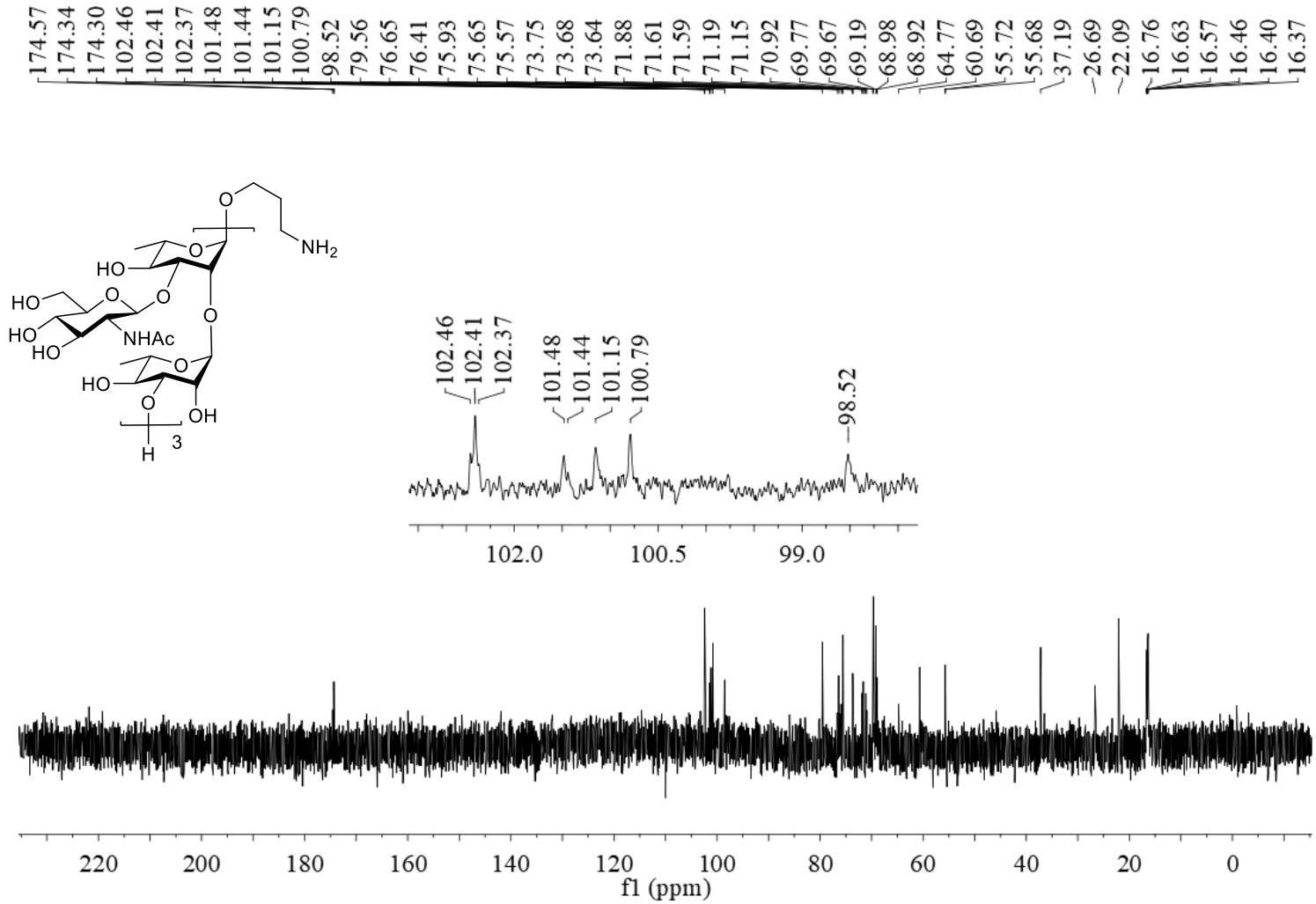
HR-ESI-(+) mass spectrum of compound **26b**



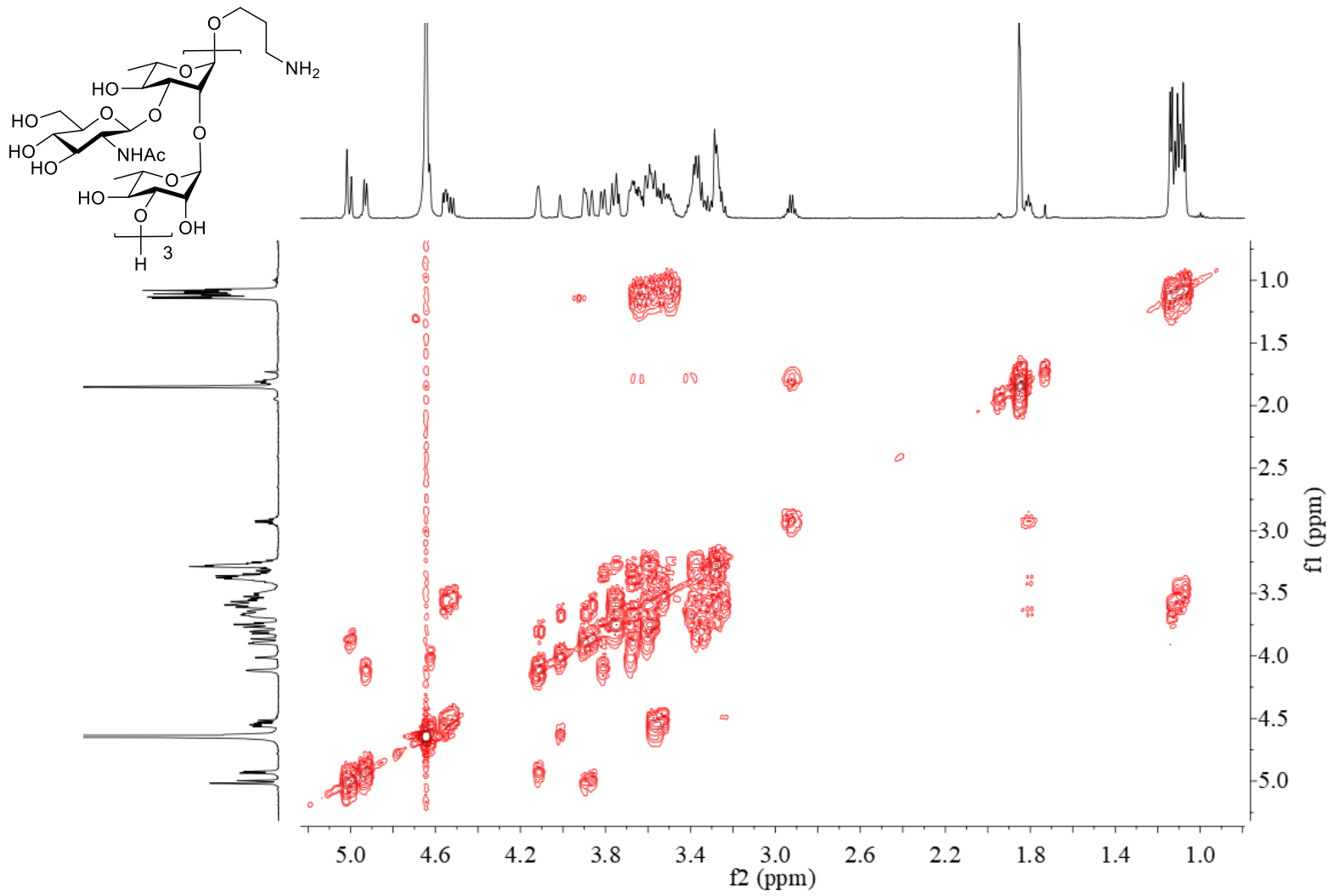
^{1}H NMR spectrum of compound **26c** (600 MHz, D_2O)



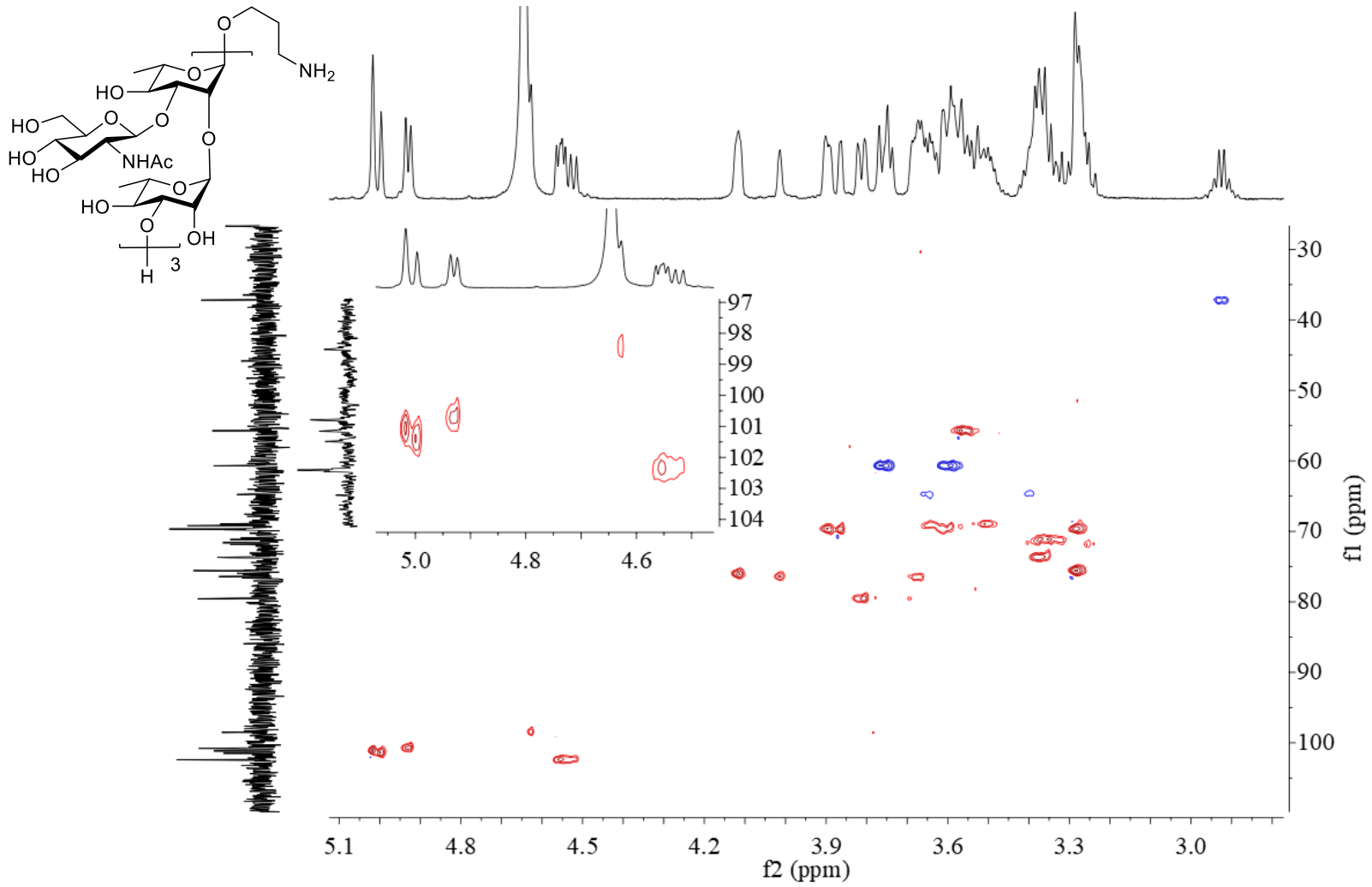
^{1}H NMR spectrum of compound **26c** (expanded sugar region, 600 MHz, D_2O)



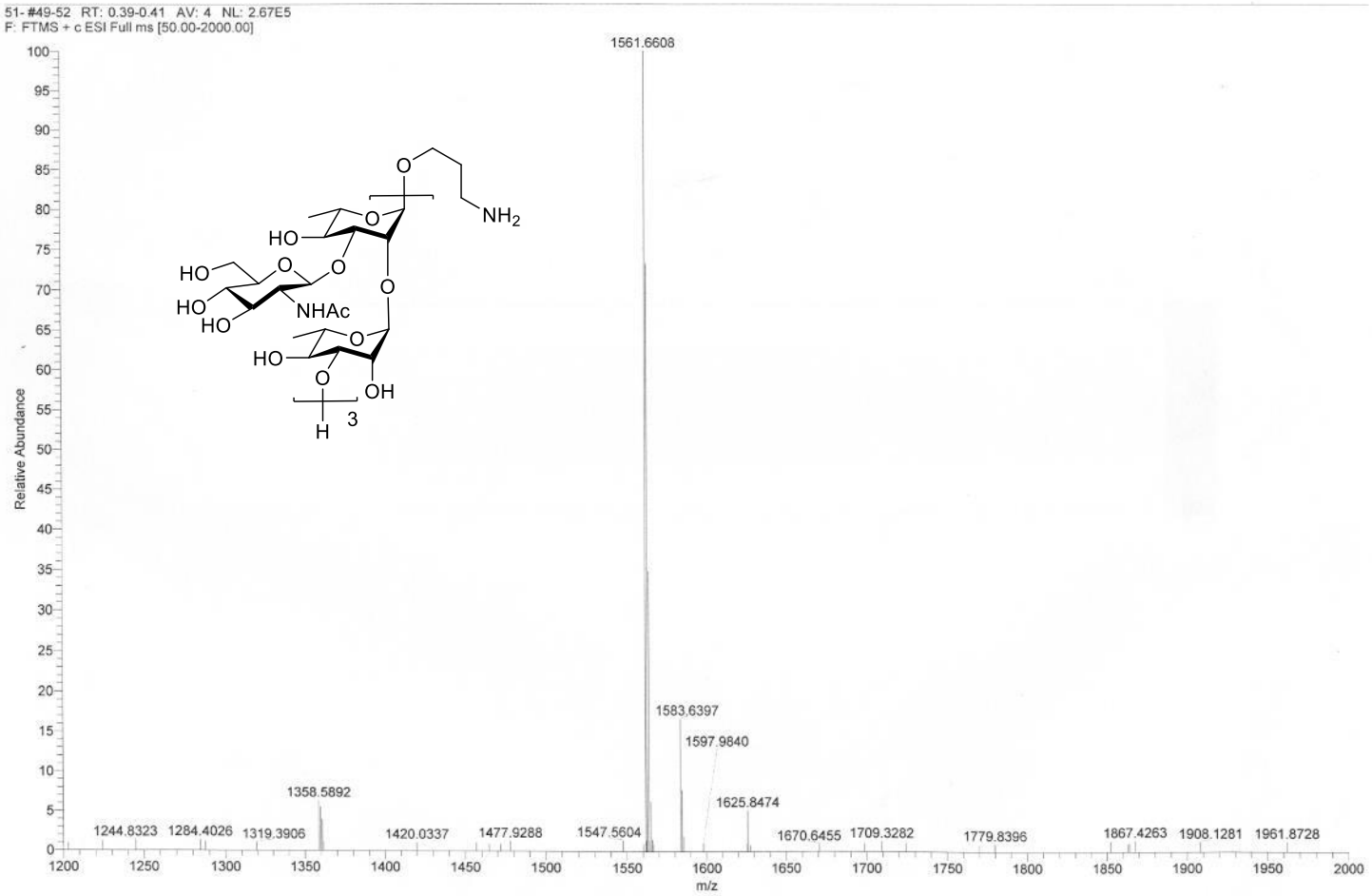
^{13}C NMR spectrum of compound **26c** (150 MHz, D_2O)



^{1}H-^{1}H COSY spectrum of compound **26c** (600 MHz, D_2O)

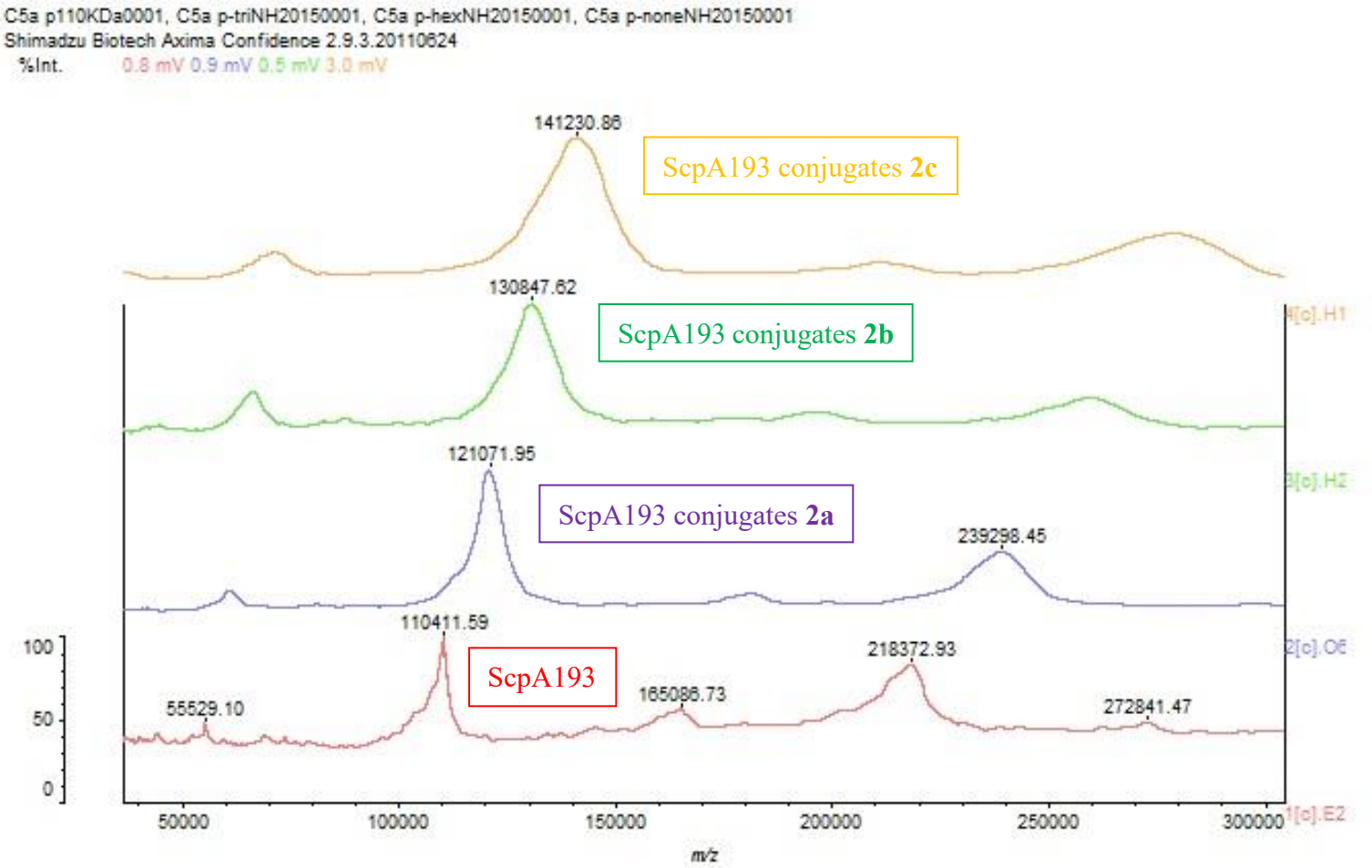


^{1}H-^{13}C HSQC spectrum of compound **26c** (600/150 MHz, D_2O)

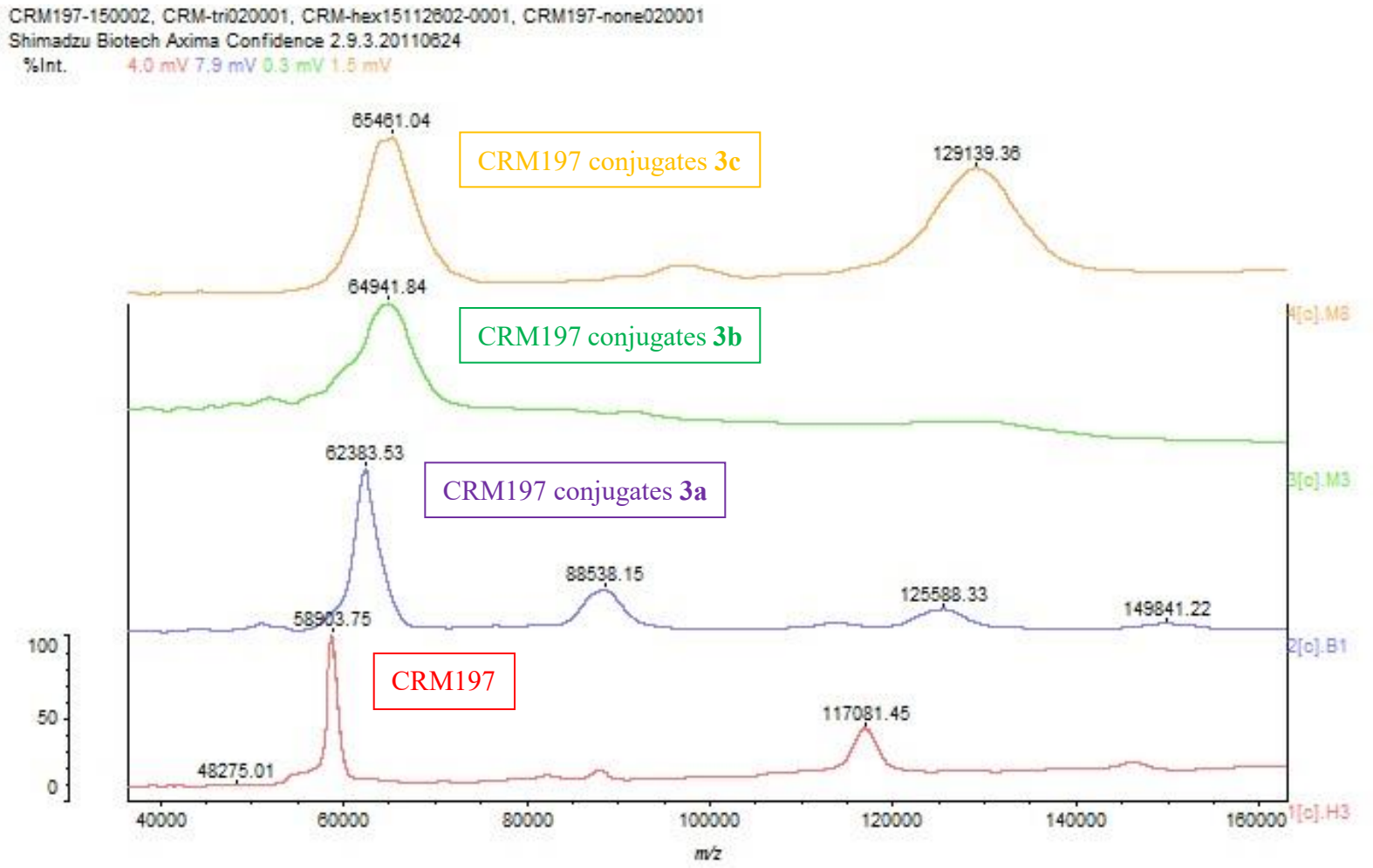


HR-ESI-(+) mass spectrum of compound **26c**

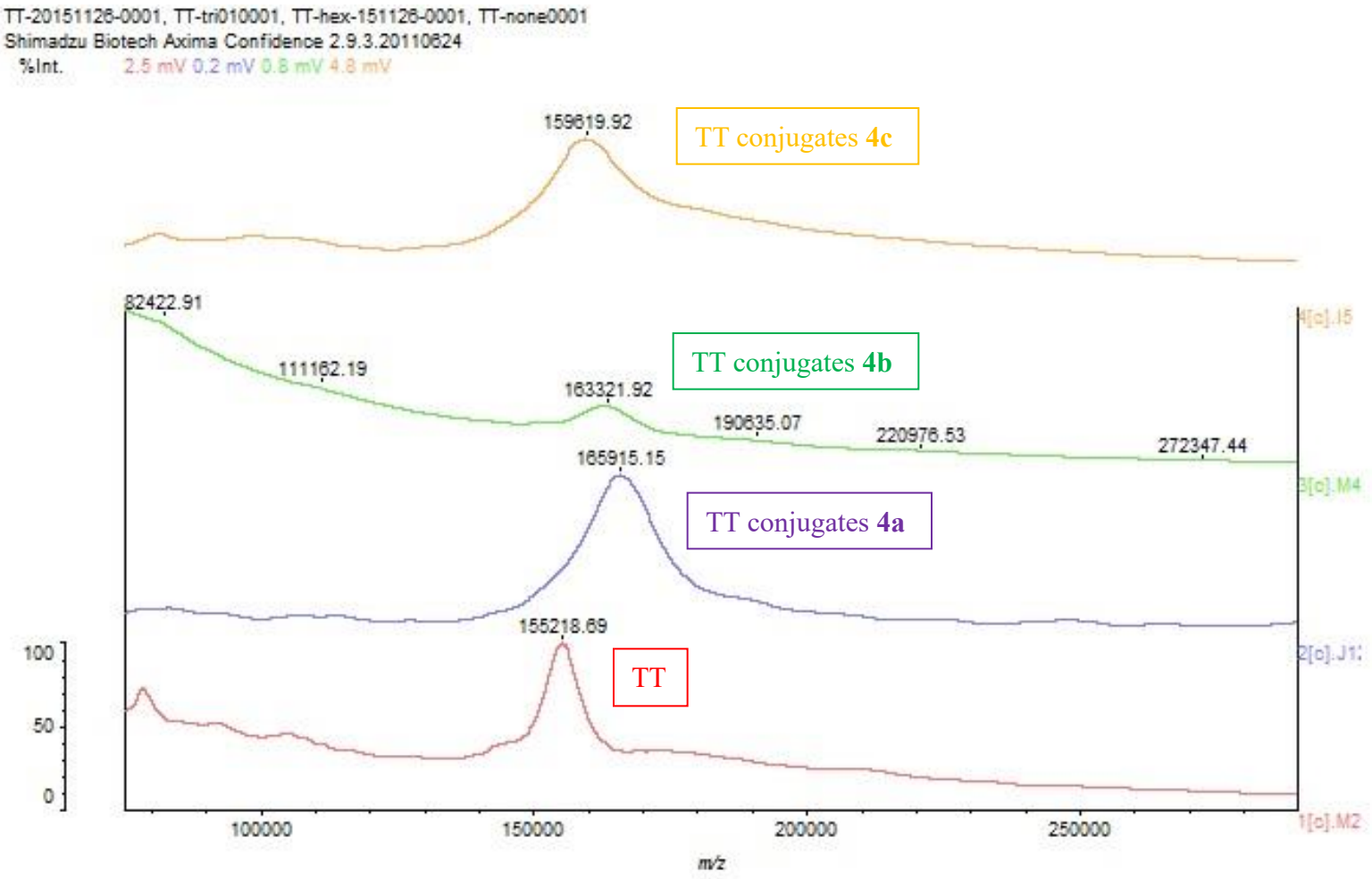
IV. MS characterization of GAS oligosaccharide-protein conjugates



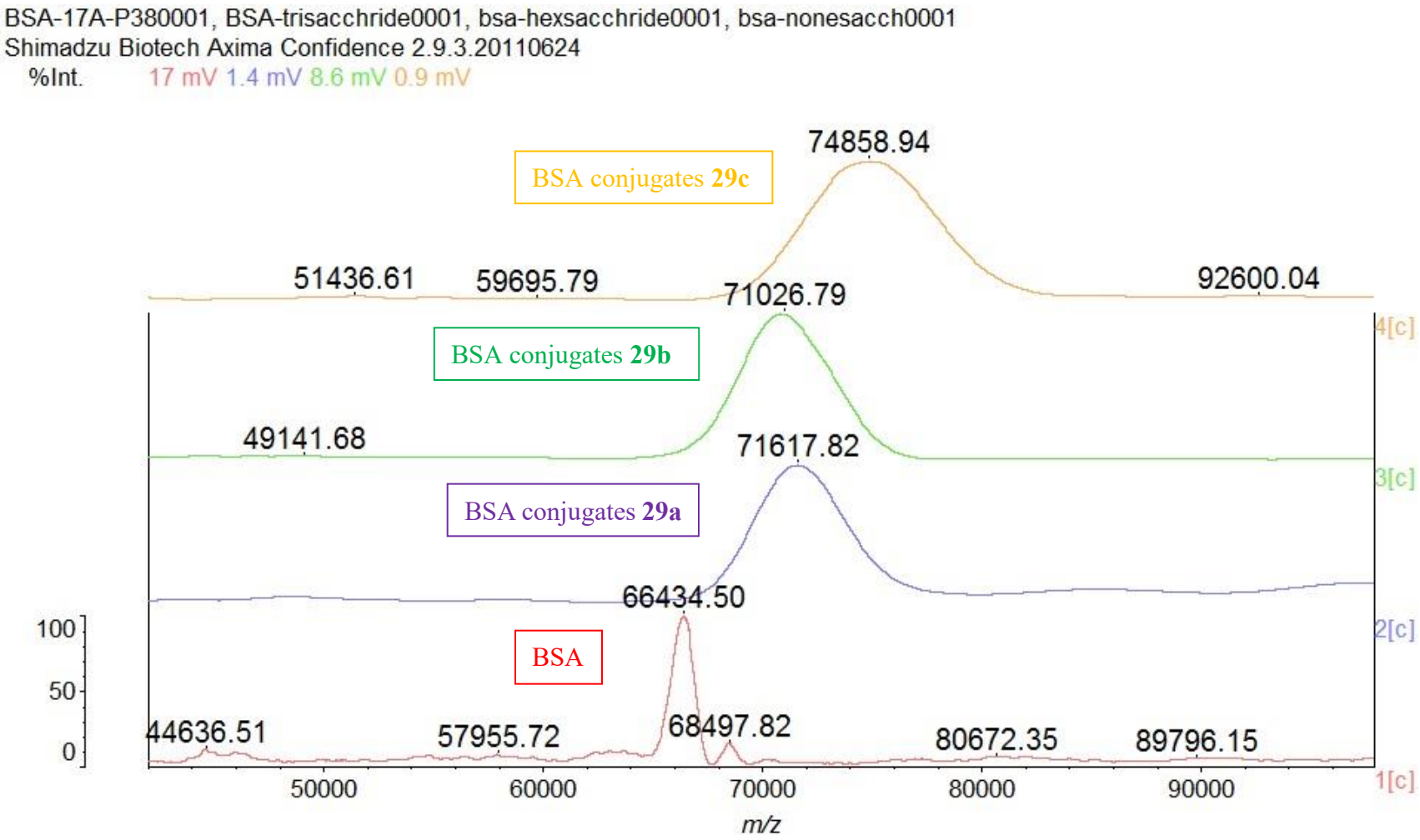
MALDI-TOF MS of ScpA193 and ScpA193 conjugates **2a-c**



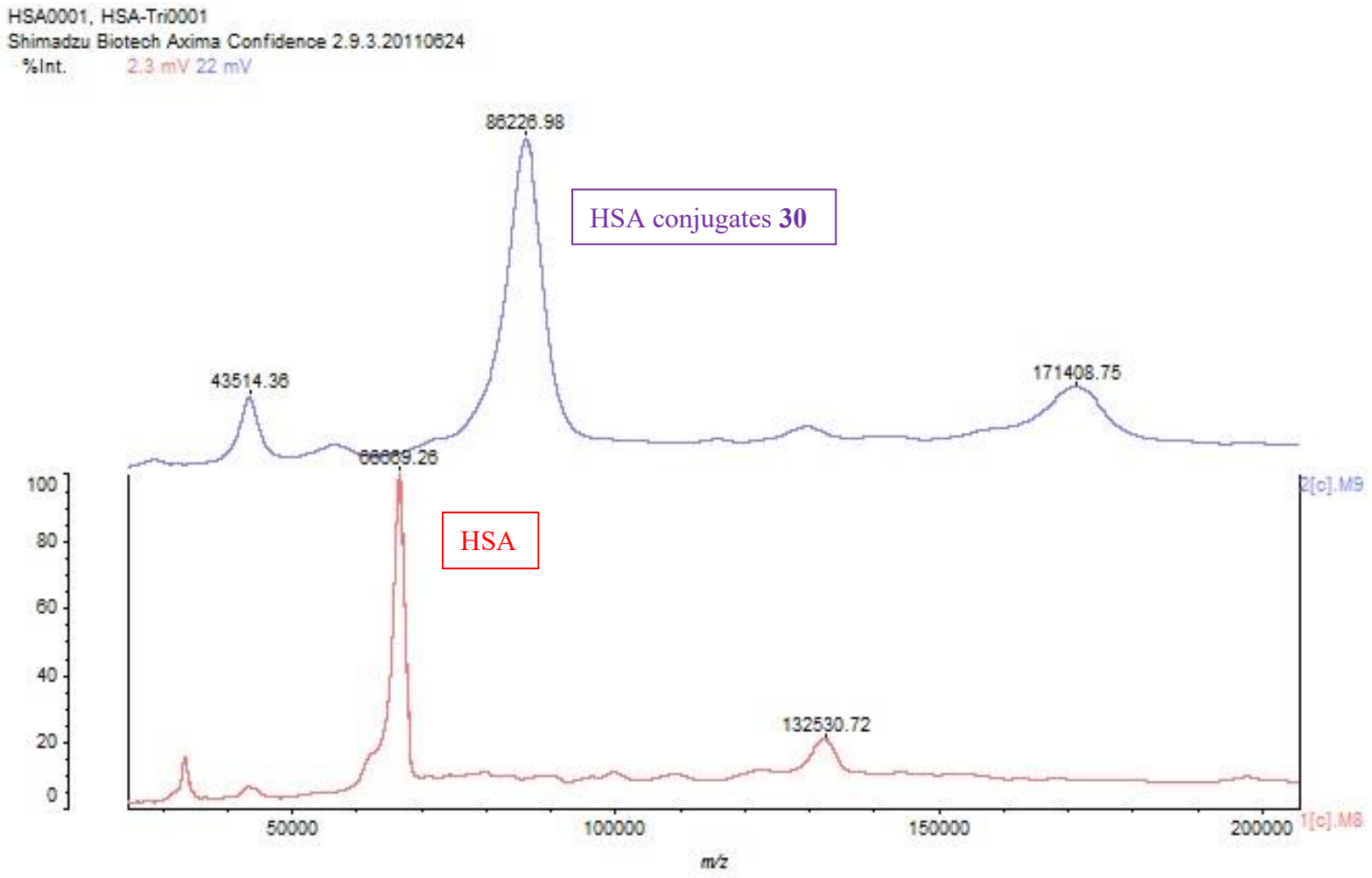
MALDI-TOF MS of CRM197 and CRM197 conjugates **3a-c**



MALDI-TOF MS of TT and TT conjugates **4a-c**



MALDI-TOF MS of BSA and BSA conjugates **29a-c**



MALDI-TOF MS of HSA and HSA conjugates **30**