Electronic Supplementary Material (ESI) for Chemical Communications.

Automated glycan assembly of branched β-(1,3)-glucans to identify antibody epitopes

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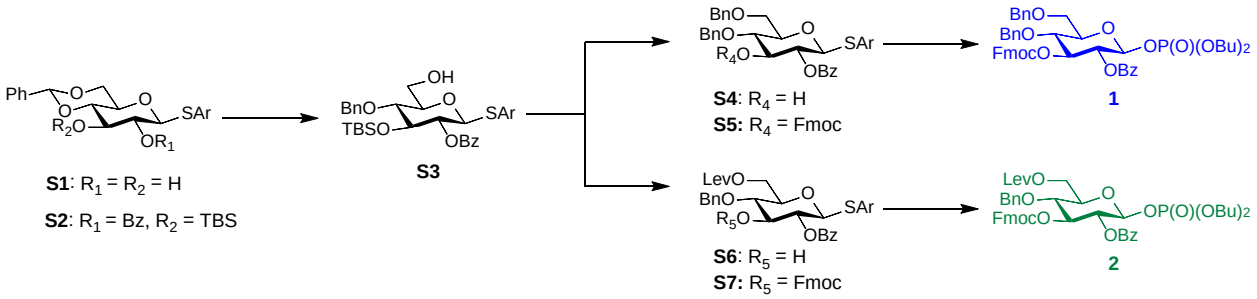
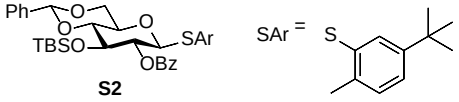
1. General Information

All chemicals used were reagent grade and used as supplied, except where noted. Prior to use, molecular sieves were activated by heating under high vacuum. All reactions were performed in oven-dried glassware under an argon atmosphere, unless noted otherwise. *N*,*N*-Dimethylformamide, (DMF) dichloromethane (DCM), toluene and tetrahydrofuran (THF) were dispensed from a Cycle-Tainer Solvent Delivery System, unless noted otherwise. Analytical thin layer chromatography (TLC) was performed on Merck silica gel 60 F254 plates (0.25 mm). Compounds were visualized by UV-irradiation, or by dipping the plate either in a cerium sulfate ammonium molybdate (CAM) solution or a 1:1 mixture of H_2SO_4 (2 N) and resorcine monomethylether (0.2%) in ethanol. Flash column chromatography was carried out using forced flow of the indicated solvent on Fluka Kieselgel 60 (230-400 mesh). All automated glycosylations were performed on a prototype automated oligosaccharide synthesizer using either anhydrous solvents of the Cycle-Tainer Solvent Delivery System. LCMS chromatograms were recorded on an Agilent 1100 Series spectrometer. Preparative HPLC purifications were performed on an Agilent 1200 Series system. Loading determination of functionalized resins was obtained using a Shimadzu UV-MINI-1240 UV spectrometer. ^{1}H and ^{13}C NMR spectra were recorded using a Varian Mercury 400 (400 MHz), 600 (600MHz) or Bruker DRX700 (700 MHz) spectrometer in $CDCl_3$ or D_2O with chemical shifts referenced to internal standards ($CDCl_3$: 7.26 ppm ^{1}H, 77.16 ppm ^{13}C; CD_3OD: 4.87 or 3.13 ppm ^{1}H, 49.0 ppm ^{13}C) unless stated otherwise. Splitting patterns are indicated as s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; brs, broad singlet for 1H-NMR data. NMR chemical shifts (δ) are reported in ppm and coupling constants (*J*) are reported in Hz. High resolution mass spectrometry (HRMS) was performed by the MS-service in the Department of Organic Chemistry at Freie Universität Berlin using an Agilent 6210 ESI-TOF instrument (Agilent Technologies, Santa Clara, CA, USA). IR spectra were recorded on a Perkin-Elmer 1600 FTIR spectrometer. Optical rotations were measured with a UniPol L 1000 polarimeter (Schmidt & Haensch, Berlin, Germany), with concentrations expressed in g per 100 mL.

2. Building Block Synthesis



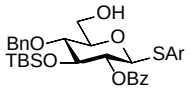
(2-Methyl-5-*tert*-butylphenyl) 2-*O*-benzoyl-4,6-*O*-benzylidene-3-*O*-*tert*-butyldimethylsilyl-1-thio-β-D-glucopyranoside S2



To a solution of compound **S1**[1] (10 g, 23.2 mmol) in anhydrous DCM (46 ml, 0.5 M) were added TBSCl (4.20 g, 27.9 mmol) and imidazole (1.21 g, 32.5 mmol) at 0 ˚C, and the mixture was stirred for 12 h at room temperature. After the reaction mixture was quenched with MeOH and saturated aqueous $NaHCO_3$ it was diluted with DCM and dried over $MgSO_4$, and the combined organic layer was evaporated *in vacuo*. To a solution of this crude product in anhydrous DCM (116 ml, 0.2 M) were added benzoic anhydride (Bz_2O) (10.51 g, 46.4 mmol), triethylamine (Et_3N) (9.70 ml, 69.7 mmol), and a catalytic amount of DMAP (0.57 g, 4.64 mmol) at 0 ˚C and the mixture was stirred at room temperature until completion. After the reaction mixture was quenched by saturated aqueous $NaHCO_3$ it was diluted with DCM and dried over $MgSO_4$, and the solvent was evaporated *in vacuo*. The crude product was purified by column chromatography on silica gel (hexane/EtOAc/DCM = 9:0.5:0.5 to 9.0:1.0:0.5) to afford **S2** (12.1 g, 18.65 mmol, 89% over two steps). $[\alpha]_D^{25}$ = 13.68 (C= 1.00, $CHCl_3$). IR (thin film): υ = 3006, 2959, 1733, 1266, 1097 cm^{-1}; ^1H NMR (400 MHz, Chloroform-*d*) δ 8.10 – 8.05 (m, 2H), 7.61 – 7.54 (m, 2H), 7.52 – 7.43 (m, 4H), 7.41 – 7.33 (m, 3H), 7.20 (dd, *J* = 7.9, 2.1 Hz, 1H), 7.07 (d, *J* = 8.0 Hz, 1H), 5.57 (s, 1H, C*H*Ph), 5.38 – 5.29 (m, 1H, H-2), 4.83 (d, *J* = 10.2 Hz, 1H, H-1), 4.37 (dd, *J* = 10.5, 5.0 Hz, 1H, H-6), 4.06 (t, *J* = 8.8 Hz, 1H, H-3), 3.87 (t, *J* = 10.3 Hz, 1H, H-6), 3.69 (t, *J* = 9.2 Hz, 1H, H-4), 3.55 (td, *J* = 9.7, 5.0 Hz, 1H, H-5), 2.17 (s, 3H, Me), 1.28 (s, 9H, *t*-Bu), 0.69 (s, 9H, *t*-Bu), -0.05 (s, 3H, Me), -0.14 (s, 3H, Me). ^{13}C NMR (150 MHz, $CDCl_3$) *δ* 165.38 (Bz), 149.70, 137.16, 136.81, 133.26, 132.94, 130.18, 130.03, 129.99, 129.59, 129.21, 128.49, 128.29, 126.40, 125.22(Ar), 102.04

(*C*HPh), 88.28 (C-1), 81.51 (C-4), 74.50 (C-3), 73.75 (C-2), 70.77 (C-5), 68.83 (C-6), 34.59 (Cq, *t*-Bu), 31.42 (Me, *t*-Bu), 25.68 (*t*-Bu of TBS), 20.33(Me), 18.08(Cq, TBS), -4.06(Me, TBS), -4.78(Me, TBS). MS ESI-HRMS m/z $[M+Na]^+$ calcd for $C_{37}H_{48}O_6SSiNa$ 671.2833, found 671.2831.

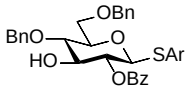
(2-Methyl-5-*tert*-butylphenyl) 2-*O*-benzoyl-4-*O*-benzyl-3-*O*-*tert*-butyldimethylsilyl-1-thio-β-D-glucopyranoside S3



S3

Compound **S2** (11.3 g, 17.41 mmol) was co-evaporated with toluene and dissolved under an Ar atmosphere in DCM (78 mL, 0.2 M). To this solution were added a 1M solution of BH_3 in THF (87ml, 87 mmol), and TMSOTf (1.94 ml, 8.71 mmol) successively at 0 °C. The mixture was stirred for 5 h at 0 °C. After the mixture was quenched with saturated aqueous $NaHCO_3$ it was diluted with DCM and dried over $MgSO_4$. The combined organic layer was evaporated *in vacuo*. The crude compound was purified by column chromatography on silica gel (hexane/EtOAc/DCM = 9:0.5:0.5 to 9:1:0.5) to afford **S3** (10.7g, 16.5 mmol, 95 %). R_f : 0.27 (Hexane/EtOAc/DCM : 8/2/0.5). $[\alpha]_D^{25}$ = 42.23 (C= 2.00, $CHCl_3$). IR (thin film): υ = 3476, 2958, 1732, 1263, 1090 cm^{-1}; ^1H NMR (600 MHz, $CDCl_3$) δ 8.11 – 8.06 (m, 2H), 7.62 – 7.56 (m, 1H), 7.52 (d, *J* = 2.0 Hz, 1H), 7.46 (t, *J* = 7.8 Hz, 2H), 7.39 – 7.33 (m, 4H), 7.32 – 7.28 (m, 1H), 7.20 (dd, *J* = 7.9, 2.1 Hz, 1H), 7.07 (d, *J* = 8.0 Hz, 1H), 5.30 (t, *J* = 9.5 Hz, 1H, H-3), 4.89 (d, *J* = 11.5 Hz, 1H, CH*H*Ph), 4.78 (d, *J* = 10.2 Hz, 1H, H-1), 4.68 (d, *J* = 11.5 Hz, 1H, CH*H*Ph), 4.00 (t, *J* = 8.8 Hz, 1H, H-3), 3.89 (ddd, *J* = 11.9, 5.5, 2.3 Hz, 1H, H-6), 3.72 (ddd, *J* = 12.1, 7.4, 4.7 Hz, 1H, H-6), 3.65 (t, *J* = 9.2 Hz, 1H, H-4), 3.48 (ddd, *J* = 9.7, 4.6, 2.6 Hz, 1H, H-5), 2.17 (s, 3H, Me), 1.29 (s, 9H, *t*-Bu), 0.81 (s, 9H, *t*-Bu), 0.04 (s, 3H, Me), -0.13 (s, 3H, Me). ^{13}C NMR (150 MHz, $CDCl_3$) *δ* 165.58 (Bz), 149.76, 137.99, 136.53, 133.24, 133.21, 130.28, 130.07, 130.03, 129.02, 128.54, 128.46, 127.90, 127.75, 125.07 (Ar), 87.66 (C-1), 79.69 (C-5), 78.53 (C-4), 76.81 (C-3), 75.25 (*C*H_2Ph), 73.43 (C-2), 62.24 (C-6), 34.56 (Cq, *t*-Bu), 31.42 (Me, *t*-Bu), 25.80 (*t*-Bu of TBS), 20.25 (Me), 17.93 (Cq, TBS), -3.88 (Me, TBS), -4.14 (Me, TBS). MS ESI-HRMS m/z $[M+Na]^+$ calcd for $C_{37}H_{50}O_6SSiNa$ 673.2990, found 673.2988.

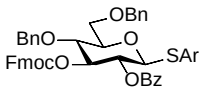
(2-Methyl-5-*tert*-butylphenyl) 2-*O*-benzoyl-4,6-di-*O*-benzyl-1-thio-β-D-glucopyranoside S4



S4

To a solution of compound **S3** (3 g, 4.61 mmol) in THF:DMF (v/v, 9:1) were added benzyl bromide (1.64 ml, 13.83 mmol) and sodium hydride (0.265 mg, 11.06 mmol) at 0 °C, and the mixture was stirred for 2 h at 0 °C. After the reaction was quenched with saturated aqueous NH_4Cl it was diluted with DCM, extracted with DCM, and the combned organic layer was dried over $MgSO_4$ and evaporated *in vacuo*. To a solution of this crude product in anhydrous acetonitrile (57 ml, 0.08 M) was added boron trifluoride etherate ($BF_3{\cdot}OEt_2$) (0.70 mL, 5.54 mmol) at 0 °C, and the mixture was stirred for 20 min. at 0 °C. After the mixture was quenched with saturated aqueous $NaHCO_3$ it was diluted with DCM, and the combined organic layer was dried over $MgSO_4$ and evaporated *in vacuo*. The crude product was purified by column chromatography on silica gel (hexane/EtOAc/DCM = 9:1:1 to 7:3:1) to afford **S4** (2.89g, 4.32 mmol, 94 % over two steps). R_f : 0.11 (Hexane/EtOAc/DCM : 8/2/0.5). $[\alpha]_D^{25}$ = -2.85 (C= 2.67, $CHCl_3$). IR (thin film): υ = 3456, 2961, 1727, 1263 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 8.12 (d, *J* = 8.2 Hz, 2H), 7.65 (d, *J* = 1.6 Hz, 1H), 7.63 – 7.57 (m, 1H), 7.47 (t, *J* = 7.7 Hz, 2H), 7.43 – 7.25 (m, 10H), 7.23 (dd, *J* = 8.0, 1.9 Hz, 1H), 7.11 (d, *J* = 8.0 Hz, 1H), 5.17 (t, *J* = 9.5 Hz, 1H, H-2), 4.86 (d, *J* = 11.2 Hz, 1H, CH*H*Ph), 4.81 (d, *J* = 10.1 Hz, 1H, H-1), 4.64 (dd, *J* = 27.6, 12.0 Hz, 3H, , CH*H*Ph, C*H*$_2$Ph), 3.97 (dd, *J* = 12.4, 5.2 Hz, 1H, H-3), 3.86 – 3.76 (m, 2H, H-6), 3.71 (t, *J* = 9.2 Hz, 1H, H-4), 3.63 – 3.56 (m, 1H, H-5), 2.82 (br, 1H, OH), 2.28 (s, 3H, Me), 1.29 (s, 9H, *t*-Bu). ^{13}C NMR (100 MHz, $CDCl_3$) *δ* 166.45 (Bz), 149.68, 138.14, 138.10, 136.95, 133.47, 132.57, 130.09, 129.93, 129.75, 129.66, 128.59, 128.50, 128.47, 128.17, 128.02, 127.75, 125.07 (Ar), 86.68 (C-1), 79.16 (C-5), 78.07 (C-4), 77.57 (C-3), 74.97 (*C*H_2Ph), 73.70 (C-2), 73.62 (*C*H_2Ph), 68.95 (C-6), 34.50 (Cq, *t*-Bu), 31.33 (Me, *t*-Bu), 20.40 (Me). MS ESI-HRMS m/z $[M+Na]^+$ calcd for $C_{38}H_{42}O_6SNa$ 649.2594, found 649.2560.

(2-Methyl-5-*tert*-butylphenyl) 2-*O*-benzoyl-4,6-di-*O*-benzyl-3-*O*-fluorenylmethoxycarbonyl 1-thio-β-D-glucopyranoside S5

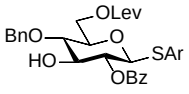


S5

To a solution of compound **S4** (2.74 g, 4.37 mmol) were added 9-fluorenylmethyl chloroformate (2.26 g, 8.74 mmol) and pyridine (2.14 mL, 26.4 mmol) successively at 0 °C, and the reaction

mixture was stirred overnight at room temperature. After the mixture was quenched with 1M aqueous HCl it was diluted with DCM, extracted with DCM, and the combined organic layer was dried over $MgSO_4$ and evaporated *in vacuo*. The crude product was purified by column chromatography on silica gel (hexane/ethyl acetate/DCM = 9:0.5:0.5 to 8.5:1.5:0.5) to afford **S5** (3.55 g, 4.18 mmol, 96 %). R_f : 0.18 (Hexane/EtOAc/DCM : 9/1/0.5). $[\alpha]_D^{25}$ = 55.96 (C= 2.79, $CHCl_3$). IR (thin film): υ = 2960, 1752, 1732, 1451, 1269 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 8.02 (dd, *J* = 8.3, 1.3 Hz, 2H), 7.70 (dd, *J* = 7.6, 3.5 Hz, 2H), 7.58 (d, *J* = 2.1 Hz, 1H), 7.52 – 7.45 (m, 2H), 7.42 (d, *J* = 7.4 Hz, 1H), 7.38 – 7.28 (m, 9H), 7.23 – 7.12 (m, 8H), 7.08 (d, *J* = 8.0 Hz, 1H), 5.39 (t, *J* = 9.7 Hz, 1H, H-2), 5.29 (t, *J* = 9.3 Hz, 1H, H-3), 4.82 (d, *J* = 10.0 Hz, 1H, H-1), 4.67 (d, *J* = 11.2 Hz, 1H, CH*H*Ph), 4.65 (d, *J* = 12.1 Hz, 1H, CH*H*Ph), 4.57 (d, *J* = 11.2 Hz, 1H, CH*H*Ph), 4.56 (d, *J* = 12.2 Hz, 1H, CH*H*Ph), 4.25 (dd, *J* = 10.4, 7.0 Hz, 1H, CH*H*Ph of Fmoc), 4.08 (dd, *J* = 10.4, 8.0 Hz, 1H, CH*H*Ph of Fmoc), 4.01 – 3.90 (m, 2H, H-4, C*H*Ph of Fmoc), 3.77 (d, *J* = 3.0 Hz, 2H, H-6), 3.63 (dt, *J* = 9.8, 3.0 Hz, 1H, H-5), 2.23 (s, 3H, Me), 1.25 (s, 9H, *t*-Bu). ^{13}C NMR (100 MHz, $CDCl_3$) δ 165.22 (Bz), 154.63 (Fmoc), 149.62, 143.38, 142.95, 141.14, 141.04, 137.92, 137.50, 137.02, 133.25, 132.33, 129.95, 129.87, 129.32, 128.40, 128.33, 127.91, 127.82, 127.75, 127.71, 127.11, 125.24, 125.16, 124.95, 119.86 (Ar), 87.10 (C-1), 80.88 (C-3), 79.20 (C-5), 75.70 (C-4), 74.92 (*C*H_2Ph), 73.58 (*C*H_2Ph), 71.02 (C-2), 70.26 (*C*H_2 of Fmoc), 68.50 (C-6), 46.46 (*C*H of Fmoc), 34.41 (Cq, *t*-Bu), 31.24 (Me, *t*-Bu), 20.30 (Me). MS ESI-HRMS m/z $[M+Na]^+$ calcd for $C_{53}H_{52}O_8SNa$ 871.3275, found 871.3276.

(2-Methyl-5-*tert*-butylphenyl) 2-*O*-benzoyl-4-*O*-benzyl-6-*O*-levulinyl-1-thio-β-D-glucopyranoside S6

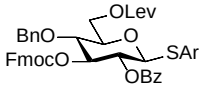


S6

To a solution of compound **S3** (1.7 g, 2.61 mmol) in anhydrous DCM (13 ml, 0.2 M) were added levulinic acid (0.910 g, 7.83 mmol) and DIC (1.22 ml, 7.83 mmol), and a catalytic amount of DMAP (0.096 g, 0.78 mmol) at 0 °C, and the mixture was stirred for 1 h at 0 °C. After the mixture was quenched with saturated aqueous $NaHCO_3$ it was diluted with DCM, extracted with DCM, and the combined organic layer was dried over $MgSO_4$ and evaporated *in vacuo*. To a solution of this crude product in anhydrous acetonitrile (57 ml, 0.08 M) was added boron trifluoride etherate ($BF_3 \cdot OEt_2$) (0.41 mL, 2.72 mmol) at 0 °C, and the mixture was stirred for 20 min. at 0 °C. After

the mixture was quenched with saturated aqueous $NaHCO_3$ it was diluted with DCM, and the combined organic layer was dried over $MgSO_4$ and evaporated *in vacuo*. The crude product was purified by column chromatography on silica gel (hexane/EtOAc/DCM = 9:1:1 to 7:3:1) to afford **S4** (2.89g, 4.32 mmol, 94 % over two steps). R_f : 0.21 (Hexane/EtOAc/DCM : 8/2/0.5). $[\alpha]_D^{25}$ = 56.91 (C= 2.85, $CHCl_3$). IR (thin film): υ = 3482, 2961, 1721, 1263 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 8.10 (dt, *J* = 8.4, 1.5 Hz, 2H), 7.64 – 7.57 (m, 1H), 7.51 (d, *J* = 2.1 Hz, 1H), 7.49 – 7.42 (m, 2H), 7.38 – 7.26 (m, 5H), 7.21 (dd, *J* = 7.9, 2.1 Hz, 1H), 7.09 (d, *J* = 8.0 Hz, 1H), 5.08 (dd, *J* = 10.0, 9.1 Hz, 1H, H-2), 4.88 (d, *J* = 11.1 Hz, 1H, CH*H*Ph), 4.76 (d, *J* = 10.1 Hz, 1H, H-1), 4.72 (d, *J* = 11.1 Hz, 1H, CH*H*Ph), 4.45 – 4.37 (m, 1H, H-6), 4.36 – 4.25 (m, 1H, H-6), 3.97 (m, 1H, H-3), 3.66 – 3.54 (m, 2H, H-4, H-5), 2.78 – 2.69 (m, 2H, Lev), 2.60 (dd, *J* = 10.0, 3.6 Hz, 2H, Lev), 2.24 (s, 3H, Lev), 2.20 (s, 3H, Me), 1.27 (s, 9H, *t*-Bu). ^{13}C NMR (100 MHz, $CDCl_3$) *δ* 206.50 (CO, Lev), 172.64 (Lev), 166.63 (Bz), 149.74, 137.89, 137.48, 133.67, 132.04, 130.18, 130.15, 130.11, 129.55, 128.74, 128.62, 128.44, 128.23, 125.49 (Ar), 86.82 (C-1), 77.87 (C-3), 77.75 (C-4), 77.01 (C-5), 75.16 (*C*H_2Ph), 73.68 (C-2), 63.51 (C-6), 38.04 (Lev), 34.56 (Cq, *t*-Bu), 31.41 (Me, *t*-Bu), 30.04 (Me, Lev), 28.04 (Lev), 20.46 (Me). MS ESI-HRMS m/z $[M+Na]^+$ calcd for $C_{36}H_{42}O_8SNa$ 657.2493, found 657.2489.

(2-Methyl-5-*tert*-butylphenyl) 2-*O*-benzoyl-4-*O*-benzyl-3-*O*-fluorenylmethoxycarbonyl-6-*O*-levulinyl-1-thio-β-D-glucopyranoside S7

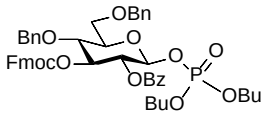


S7

To a solution of compound **S6** (1.61 g, 2.55 mmol) in DCM (12.5 ml, 0.2 M) were added 9-fluorenylmethyl chloroformate (1.32 g, 5.11 mmol) and pyridine (0.62 mL, 7.66 mmol) successively at 0 °C, and the mixture was stirred overnight at room temperature. After the mixture was quenched with saturated aqueous $NaHCO_3$ it was diluted with DCM and extracted with DCM, then the combined organic layer was washed with 1M aqueous HCl, dried over $MgSO_4$ and evaporated *in vacuo*. The crude product was purified by column chromatography on silica gel (hexane/ EtOAc /DCM = 9:0.5:0.5 to 8.5:1.5:0.5) to afford **S7** (2.411 g, 2.46 mmol, 96 %). R_f : 0.35 (hexane/EtOAc/DCM : 9/1/0.5). $[\alpha]_D^{25}$ = 59.08 (C= 2.80, $CHCl_3$). IR (thin film): υ = 2961, 1733, 1268 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 8.04 – 7.97 (m, 2H), 7.69 (dd, J = 7.6, 4.8 Hz, 2H), 7.52 – 7.46 (m, 3H), 7.42 (dd, J = 7.5, 0.8 Hz, 1H), 7.38 – 7.30 (m, 4H), 7.27 – 7.15 (m, 9H), 7.09

(d, J = 8.0 Hz, 1H), 5.36 (t, J = 9.6 Hz, 1H, H-3), 5.30 (t, J = 9.1 Hz, 1H, H-2), 4.81 (d, J = 9.7 Hz, 1H, H-1), 4.65 (m, 2H, C*H*$_2$Ph), 4.39 (dd, J = 12.1, 2.1 Hz, 1H, H-6), 4.31 (dd, J = 12.1, 4.5 Hz, 1H, H-6), 4.26 (dd, J = 10.4, 7.0 Hz, 1H, CH*H* of Fmoc), 4.11 (dd, J = 10.4, 7.9 Hz, 1H, CH*H* of Fmoc), 3.96 (t, J = 7.4 Hz, 1H, C*H* of Fmoc), 3.88 – 3.79 (m, 1H, H-4), 3.68 (ddd, J = 9.8, 4.4, 2.2 Hz, 1H, H-5), 2.80 – 2.70 (m, 2H, Lev), 2.60 (dd, J = 9.9, 3.5 Hz, 2H, Lev), 2.23 (s, 3H, Me), 2.21 (s, 3H, Lev), 1.27 (s, 9H, *t*-Bu). ^{13}C NMR (100 MHz, $CDCl_3$) δ 206.40 (CO, Lev), 172.53 (Lev), 165.35 (Bz), 154.68 (Fmoc), 149.79, 143.47, 143.01, 141.30, 141.19, 137.55, 137.23, 133.48, 132.01, 130.33, 130.12, 130.09, 129.33, 128.59, 128.50, 128.20, 128.16, 127.91, 127.26, 125.63, 125.34, 125.06, 120.03 (Ar), 87.42 (C-1), 80.95 (C-2), 77.12 (C-5), 75.44 (C-5), 75.08 (*C*H_2Ph), 71.00 (C-3), 70.44 (*C*H_2 of Fmoc), 63.13 (C-6), 46.59 (*C*H of Fmoc), 38.02 (*C*H_2 of Lev), 34.55 (Cq, *t*-Bu), 31.41 (*t*-Bu), 30.03 (Me of Lev), 28.00 (*C*H_2 of Lev), 20.43 (Me). MS ESI-HRMS m/z $[M+Na]^+$ calcd for $C_{51}H_{52}O_{10}SNa$ 879.3173, found 879.3179.

Dibutyl 2-*O*-benzoyl-4,6-di-*O*-benzyl-3-*O*-fluorenylmethoxycarbonyl-1-thio-β-D-glucopyranoside 1



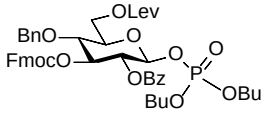
1

Thioglycoside **S5** (6.40 g, 7.55 mmol) was co-evaporated twice with toluene. The residue and NIS (1.87 g, 8.30 mmol) were dissolved in DCM (37.7 mL, 0.2 M) under an Ar atmosphere and the solution was cooled to 0 °C. Dibutyl hydrogen phosphate (2.99 mL, 15.1 mmol) and triflic acid (67 µL, 0.755 mmol) were added and the reaction was stirred at 0 °C for 1 h. After complete conversion of the starting material the reaction mixture was diluted with DCM and extracted with 10% aqueous $Na_2S_2O_3$ followed by saturated aqueous $NaHCO_3$. The organic phase was dried over $MgSO_4$ and the solvent was removed *in vacuo*. The crude product was purified by silica gel flash column chromatography (hexane/ EtOAc, 3:1 to 1:1) to afford building block **1** (6.21 g, 94%).

R_f : 0.35 (Hexane/EtOAc/DCM : 1/1/0.5). $[\alpha]_D^{25}$ = 150.12 (C= 2.10, $CHCl_3$). IR (thin film): υ = 2962, 1751, 1733, 1449 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.00 (dd, *J* = 8.3, 1.2 Hz, 2H), 7.73 (dd, *J* = 7.6, 5.0 Hz, 2H), 7.60 – 7.49 (m, 3H), 7.46 – 7.33 (m, 4H), 7.32 – 7.17 (m, 8H), 7.12 – 6.99 (m, 4H), 5.42 – 5.35 (m, 2H), 5.11 – 5.05 (m, 1H), 4.58 (d, *J* = 11.6 Hz, 1H), 4.54 – 4.46 (m,

3H), 4.33 (qd, *J* = 10.5, 7.1 Hz, 2H), 4.11 (t, *J* = 7.1 Hz, 1H), 4.06 – 3.96 (m, 2H), 3.92 – 3.80 (m, 2H), 3.76 – 3.60 (m, 4H), 1.61 – 1.49 (m, 2H), 1.37 – 1.20 (m, 4H), 1.05 – 0.95 (m, 2H), 0.85 (t, *J* = 7.4 Hz, 3H), 0.66 (t, *J* = 7.4 Hz, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 164.96, 154.24, 143.37, 143.19, 141.43, 141.40, 137.81, 137.24, 133.54, 130.02, 129.40, 128.57, 128.44, 128.42, 128.31, 128.07, 128.05, 128.04, 127.83, 127.78, 127.72, 127.31, 125.19, 125.11, 120.20, 96.66, 96.61, 79.25, 79.23, 77.48, 77.16, 76.84, 75.12, 74.36, 73.78, 73.72, 73.01, 72.93, 70.21, 69.18, 68.17, 68.11, 68.04, 67.98, 46.82, 32.14, 32.07, 31.90, 31.83, 18.67, 18.34, 13.67, 13.48. MS ESI-HRMS m/z $[M+Na]^+$ calcd for $C_{50}H_{55}O_{12}PNa$ 901.3323, found 901.3330.

Dibutyl 2-*O*-benzoyl-4-*O*-benzyl-3-*O*-fluorenylmethoxycarbonyl-6-*O*-levulinyl-1-thio-β-D-glucopyranoside 2



2

Thioglycoside **S7** (3.42 g, 4.00 mmol) was co-evaporated twice with toluene. The residue and NIS (0.99 g, 4.40 mmol) were dissolved in DCM (20.0 mL, 0.2 M) under an Ar atmosphere and the solution was cooled to 0 °C. Dibutyl hydrogen phosphate (1.58 mL, 7.99 mmol) and triflic acid (35 μL, 0.400 mmol) were added and the reaction was stirred at 0 °C for 1 h. After complete conversion of the starting material it was diluted with DCM and extracted with 10% aqueous $Na_2S_2O_3$ and then saturated aqueous $NaHCO_3$. The organic phase was dried over $MgSO_4$ and the solvent was removed *in vacuo*. The crude product was purified by silica gel flash column chromatography (hexane/ethyl acetate, 3:1 to 1:1) to afford building block **2** (2.95 g, 83%).

R_f : 0.28 (Hexane/EtOAc/DCM : 1/1/0.5). $[\alpha]_D^{25}$ = 33.02 (C= 2.40, $CHCl_3$). IR (thin film): υ = 2963, 1736, 1262 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.12 – 8.03 (m, 2H), 7.72 – 7.65 (m, 2H), 7.61 – 7.24 (m, 12H), 7.11 (dtd, *J* = 15.2, 7.5, 1.1 Hz, 2H), 5.80 (dd, *J* = 10.5, 8.0 Hz, 1H), 5.42 (dd, *J* = 8.0, 7.4 Hz, 1H), 5.03 (dd, *J* = 10.5, 3.0 Hz, 1H), 4.80 (d, *J* = 11.2 Hz, 1H), 4.52 (d, *J* = 11.2 Hz, 1H), 4.36 (dd, *J* = 10.4, 7.1 Hz, 1H), 4.32 – 4.19 (m, 3H), 4.11 (t, *J* = 7.0 Hz, 1H), 4.08 – 3.98 (m, 3H), 3.91 (td, *J* = 6.3, 1.2 Hz, 1H), 3.79 – 3.63 (m, 2H), 2.77 – 2.72 (m, 3H), 2.52 (dd, *J* = 6.9, 6.1 Hz, 2H), 2.19 (s, 3H), 1.65 – 1.56 (m, 2H), 1.42 – 1.32 (m, 2H), 1.32 – 1.24 (m, 2H), 1.07 – 0.97 (m, 2H), 0.90 (t, *J* = 7.4 Hz, 3H), 0.68 (t, *J* = 7.4 Hz, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 206.35, 172.19, 165.00, 154.35, 143.13, 142.70, 141.21, 141.10, 137.16, 133.45, 129.93, 129.11,

128.52, 128.48, 128.10, 127.85, 127.11, 127.07, 125.09, 124.90, 120.00 (Ar), 96.73, 96.68, 77.39, 77.37, 77.33, 77.22, 77.02, 76.70, 75.29, 73.02, 72.88, 70.25, 69.77, 69.68, 68.03, 67.97, 67.92, 67.85, 62.09, 46.45, 37.82, 32.02, 31.95, 31.77, 31.70, 29.82, 29.57, 27.69, 18.56, 18.21, 13.56, 13.36. MS ESI-HRMS m/z $[M+Na]^+$ calcd for $C_{48}H_{55}O_{14}PNa$ 909.3222, found 909.3219.

3. Automated Glycan Assembly

Automated glycan assembly of β-(1,3)-glucans:

Solution A1, building block **1** in CH_2Cl_2 (3 equiv. **1** per mL CH_2Cl_2)

Solution A2, building block **1** in CH_2Cl_2 (5 equiv. **1** per mL CH_2Cl_2)

Solution B1, building block **2** in CH_2Cl_2 (3 equiv. **2** per mL CH_2Cl_2)

Solution B2, building block **2** in CH_2Cl_2 (5 equiv. **2** per mL CH_2Cl_2)

Solution C, activator: stoichiometric amounts of TMSOTf in CH_2Cl_2

Solution D, Fmoc deprotection: Et_3N (10% v/v) in DMF

Solution E, Lev deprotection: $N_2H_4 \cdot H_2O$, 0.56 M in pyridine/acetic acid (3:2 v/v)

Module A1: Glycosylation. **Solution A1** (1 mL) is added to the reaction vessel, and the suspension is cooled to -30 °C while the resin is being agitated by an argon flow. Upon reaching the set temperature, **solution C** (1 mL) is added to the reaction vessel. The temperature is maintained for 10 min and then raised to -15 °C for 25 min. The reaction vessel is then drained by argon pressure and the resin is washed with CH_2Cl_2 (6 x).

Module A2: Glycosylation. **Solution A2** (1 mL) is added to the reaction vessel, and the suspension is cooled to -30 °C while the resin is being agitated by an argon flow. Upon reaching the set temperature, **solution C** (1 mL) is added to the reaction vessel. The temperature is maintained for 10 min and then raised to -15 °C for 25 min. The reaction vessel is then drained by argon pressure and the resin is washed with CH_2Cl_2 (6 x).

Module B1: Glycosylation. **Solution B1** (1 mL) is added to the reaction vessel, and the suspension is cooled to -30 °C while the resin is being agitated by an argon flow. Upon reaching the set temperature, **solution C** (1 mL) is added to the reaction vessel. The temperature is maintained for 10 min and then raised to -10 °C for 25 min. The reaction vessel is then drained by argon pressure and the resin is washed with CH_2Cl_2 (6 x).

Module B2: Glycosylation. **Solution B2** (1 mL) is added to the reaction vessel, and the suspension is cooled to -30 °C while the resin is being agitated by an argon flow. Upon reaching the set temperature, **solution C** (1 mL) is added to the reaction vessel. The temperature is maintained for 10 min and then raised to -10 °C for 25 min. The reaction vessel is then drained by argon pressure and the resin is washed with CH_2Cl_2 (6 x).

Module B1*: Glycosylation. **Solution A1** (1 mL) is added to the reaction vessel, and the suspension is cooled to -30 °C while the resin is being agitated by an argon flow. Upon reaching

the set temperature, **solution C** (1 mL) is added to the reaction vessel. The temperature is maintained for 10 min and then raised to -10 °C for 25 min. The reaction vessel is then drained by argon pressure and the resin is washed with CH_2Cl_2 (6 x).

Module B2*: Glycosylation. **Solution A2** (1 mL) is added to the reaction vessel, and the suspension is cooled to -30 °C while the resin is being agitated by an argon flow. Upon reaching the set temperature, **solution C** (1 mL) is added to the reaction vessel. The temperature is maintained for 10 min and then raised to -10 °C for 25 min. The reaction vessel is then drained by argon pressure and the resin is washed with CH_2Cl_2 (6 x).

Module C: Swelling of resin prior to synthesis. The reaction vessel is charged with functionalized resin, and CH_2Cl_2 (2 mL) is added. The resin is swelled in CH_2Cl_2 for 2 h. At the beginning of the synthesis, the reaction vessel is drained by argon pressure.

Module D: Washing before glycosylation. The resin is washed with THF (6 x) and CH_2Cl_2 (6 x) at room temperature.

Module E: Activator wash. **Solution C** is added at -30 °C for 1 min while the suspension is agitated by an argon flow. The reaction vessel is then drained by argon pressure, and the resin is washed with CH_2Cl_2 (6 x).

Module F: Washing after glycosylation cycle. The temperature is raised to 25 °C, and the resin is washed with THF (6 x) and CH_2Cl_2 (6 x).

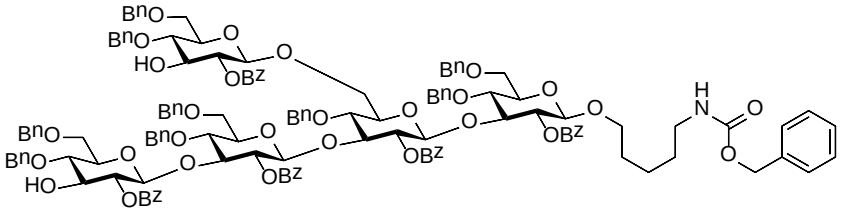
Module G: Fmoc deprotection. The resin is washed with DMF (3 x). **Solution D** (2 mL) is added to the reaction vessel, and the suspension is agitated by an argon flow for 15 min at 25 °C. The reaction vessel is then drained and the solvents are transferred to a fraction collector *via* argon pressure.

Module H: Washing after Fmoc deprotection. The temperature is set to 25 °C, and the resin is washed with DMF (3 x), CH_2Cl_2 (6 x), AcOH in CH_2Cl_2 (6 x), THF (6 x), and CH_2Cl_2 (6 x).

Module I: Lev deprotection. The resin is washed with DMF (3 x). CH_2Cl_2 (1.3 mL) is added to the reaction vessel, followed by the addition of **Solution E** (0.8 mL). The suspension is agigated by pulsed argon bubbling for 30 min at 25 °C., then the reaction vessel is drained by argon pressure.

Module J: Washing after Lev deprotection. The temperature is set to 25 °C, and the resin is washed with CH_2Cl_2 (3 x), THF (3 x), acetone in CH_2Cl_2 (20 % v/v, 6 x), CH_2Cl_2 (6 x), AcOH in CH_2Cl_2 (6 x), THF (6 x), and CH_2Cl_2 (6 x).

***N*-Benzylcarbamoyl aminopentyl 4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-[4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→6)]-4-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranoside (5).**



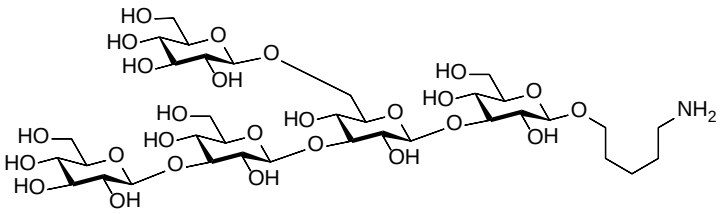
Glucose Unit	Modules (Iterations)
1	**D**(1), **A1** (3), **F** (1), **G** (3), **H** (1), **E** (1)
2	**D**(1), **B1** (3), **F** (1), **G** (3), **H** (1), **E** (1)
3	**D**(1), **A1*** (3), **F** (1), **G** (3), **H** (1), **E** (1)
4	**D**(1), **A1** (3), **F** (1), **I** (3), **J** (1), **E** (1)
5	**D**(1), **A1** (3), **F** (1), **G** (3), **H** (1)

Table 1. Program modules for the synthesis of pentasaccharide **5**.

The reaction vessel of the synthesizer was charged with functionalized Merrifield resin **4** (30 mg, loading: 0.39 mmol/g). Program module **C** was executed once. Then the program described in Table 1 was executed by the synthesizer. The resin was removed from the reaction vessel and swelled in CH_2Cl_2. The suspension was irradiated with UV light by delivering the suspension *via* syringe pump through a FEP tubing (inner diameter: 0.03 inch; volume: 12 mL) wrapped around a UV light source (medium pressure Hg lamp with arc lengths of 27.9 cm and power of 450 W, surrounded by a Pyrex UV filter with 50% transmittance at 305 nm). The resin was slowly injected from a disposable syringe (2 mL) and pushed through the tubing with 15 mL CH_2Cl_2 (flow rate: 300 µL·min^{-1}). The tubing was washed with 15 mL CH_2Cl_2/MeOH (1:1 v/v, flow rate: 300 µL·min^{-1} for 8 mL and 4 mL·min^{-1} for 7 mL), and finally with 15 mL MeOH (flow rate: 4 mL·min^{-1}). The suspension leaving the reactor was directed into a filter where the resin was filtered off and washed with CH_2Cl_2/MeOH (1:1 v/v), MeOH and CH_2Cl_2, while the filtrate was collected. The tubing was re-equilibrated with 15 mL CH_2Cl_2 using a flow rate of 4 mL·min^{-1}. The entire cleavage procedure was repeated three times. The resulting solution was concentrated *in vacuo* to afford compound **5** as a white solid (22.0 mg, 9.2 µmol). $[\alpha]_D^{20}$ = -6.51 (c = 0.3, $CHCl_3$); IR (thin film, cm^{-1}): 3423,

2921, 2867, 1732, 1267, 1094, 1070, 709; ^{1}H NMR (700 MHz, $CDCl_3$) δ 8.00 (d, *J* = 7.1 Hz, 2H), 7.76 (d, *J* = 7.1 Hz, 2H), 7.69 (d, *J* = 7.4 Hz, 2H), 7.61 (d, *J* = 7.4 Hz, 2H), 7.56 (t, *J* = 7.4 Hz, 1H), 7.51 (dd, *J* = 11.8, 4.4 Hz, 3H), 7.48 – 7.41 (m, 2H), 7.39 – 7.13 (m, 61H), 5.08 – 5.01 (m, 3H), 4.98 (t, *J* = 8.6 Hz, 1H), 4.94 – 4.83 (m, 7H), 4.80 (d, *J* = 11.7 Hz, 1H), 4.75 (d, *J* = 11.3 Hz, 1H), 4.71 (d, *J* = 11.2 Hz, 1H), 4.63 – 4.52 (m, 7H), 4.50 – 4.22 (m, 13H), 4.17 (d, *J* = 7.7 Hz, 1H), 4.09 – 3.99 (m, 2H), 3.91 – 3.77 (m, 3H), 3.73 – 3.31 (m, 20H), 3.30 – 3.12 (m, 6H), 2.90 – 2.78 (m, 2H), 1.41 – 1.16 (m, 4H), 1.13 – 0.96 (m, 2H); ^{13}C NMR (151 MHz, $CDCl_3$) δ 166.8, 166.6, 164.6, 164.3, 164.2, 138.6, 138.4, 138.3, 138.3, 138.1, 133.4, 133.3, 133.3, 133.0, 130.2, 129.9, 129.9, 129.8, 129.7, 129.5, 129.4, 128.7, 128.6, 128.6, 128.5, 128.5, 128.4, 128.3, 128.3, 128.2, 128.0, 128.0, 127.9, 127.8, 127.7, 127.7, 127.6, 127.6, 107.5, 100.9, 100.6, 100.4, 99.9, 79.9, 79.8, 79.2, 78.5, 78.5, 76.5, 76.4, 76.3, 76.1, 76.0, 75.5, 75.2, 75.2, 75.1, 75.0, 75.0, 74.9, 74.8, 74.6, 74.6, 74.2, 74.0, 73.7, 73.5, 73.5, 73.4, 69.4, 69.1, 68.9, 68.6, 67.4, 66.6, 40.9, 29.4, 23.2; HSQC (600 MHz, $CDCl_3$) δ 100.9 ($J_{C1,H1}$ = 161.4 Hz, C-1), 100.6 ($J_{C1,H1}$ = 161.4 Hz, C-1), 100.4 ($J_{C1,H1}$ = 161.1 Hz, C-1), 99.9 ($J_{C1,H1}$ = 162.5 Hz, C-1), 99.9 ($J_{C1,H1}$ = 161.1 Hz, C-1); MALDI-TOF MS calcd for $C_{141}H_{143}NO_{33}Na$ $[M + Na]^+$, 2401.9469; found, 2401.810.

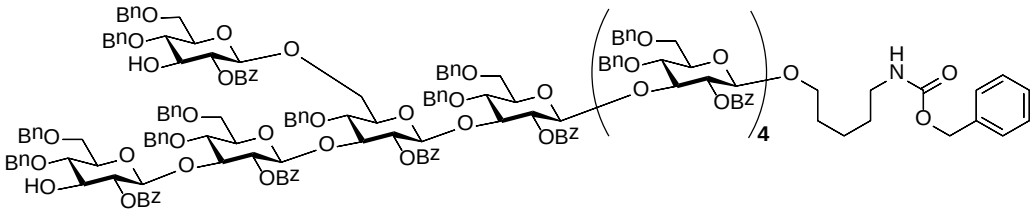
5-Aminopentyl β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-[β-D-glucopyranosyl-(1→6)]-β-D-glucopyranosyl-(1→3)-β-D-glucopyranoside (9).



Compound **5** (22 mg, 9.2 μmol) was dissolved in CH_2Cl_2, a 0.5 M solution of NaOMe in MeOH (184 μL, 92 μmol) was added, and the reaction was stirred overnight. The solution was neutralized with Amberlite® IR-120 (H^+ form) and filtered. The filtrate was concentrated *in vacuo* to afford 13 mg of the deacylated intermediate which was directly used in the next reaction step without further purification. *t*BuOH (10 μL) was added to a round bottom flask and cooled to -78 °C. NH_3 was condensed into the flask at -78 °C, and sodium was added with vigorous stirring until the solution acquired a deep blue color. The deacylated compound was dissolved in THF and added dropwise to the solution. After 30 minutes, the reaction was quenched by the dropwise addition of MeOH, and the ammonia was evaporated. The remaining liquid was neutralized with AcOH and concentrated *in vacuo*. The residue was taken up in H_2O and purified by size exclusion

chromatography over Sephadex® G-10 to afford compound **9** (1.7 mg, 1.9 µmol) as a white solid after lyophilization. ^{1}H NMR (600 MHz, D_2O) δ 4.71 – 4.59 (m, 3H), 4.54 (d, *J* = 7.9 Hz, 1H), 4.51 (d, *J* = 8.1 Hz, 1H), 4.24 (d, *J* = 11.2 Hz, 1H), 4.00 – 3.86 (m, 6H), 3.85 – 3.26 (m, 25H), 3.02 (t, *J* = 7.4 Hz, 2H), 1.75 – 1.61 (m, 4H), 1.52 – 1.41 (m, 2H); ^{13}C NMR (151 MHz, D_2O) δ 105.4, 105.4, 105.3, 105.1, 104.5, 87.8, 86.8, 86.4, 78.6, 78.5, 78.2, 78.2, 78.1, 77.1, 76.0, 75.8, 75.3, 72.7, 72.2, 71.3, 70.9, 70.7, 70.6, 63.3, 42.0, 30.8, 29.0, 24.7; HSQC (600 MHz, $CDCl_3$) δ 105.4 ($J_{C1,H1}$ = 163 Hz, C-1), 105.4 ($J_{C1,H1}$ = 163 Hz, C-1), 105.3 ($J_{C1,H1}$ = 162 Hz, C-1), 105.1 ($J_{C1,H1}$ = 164 Hz, C-1), 104.5 ($J_{C1,H1}$ = 161 Hz, C-1); HRMS calcd for $C_{35}H_{63}NO_{26}Na$ $[M + Na]^+$, 936.3531; found, 936.3558.

***N*-Benzylcarbamoyl aminopentyl 4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-[4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→6)]-4-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranoside (6).**



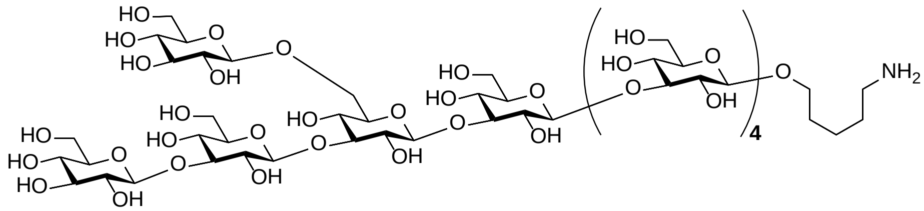
Glucose Unit	Modules (Iterations)
1-4	**D**(1), **A2** (2), **F** (1), **G** (3), **H** (1), **E** (1)
5	**D**(1), **A2** (2), **F** (1), **G** (3), **H** (1), **E** (1)
6	**D**(1), **B2** (2), **F** (1), **G** (3), **H** (1), **E** (1)
7	**D**(1), **A2*** (2), **F** (1), **G** (3), **H** (1), **E** (1)
8	**D**(1), **A2** (2), **F** (1), **I** (3), **J** (1), **E** (1)
9	**D**(1), **A2** (2), **F** (1), **G** (3), **H** (1)

Table 2. Program modules for the synthesis of nonasaccharide **6**.

The reaction vessel of the synthesizer was charged with functionalized Merrifield resin **4** (30 mg, loading: 0.39 mmol/g). Program module **C** was executed once. Then the program described in

Table 2 was executed by the synthesizer. The resin was removed from the reaction vessel and swelled in CH_2Cl_2. The suspension was irradiated with UV light by delivering the suspension *via* syringe pump through a FEP tubing (inner diameter: 0.03 inch; volume: 12 mL) wrapped around a UV light source (medium pressure Hg lamp with arc lengths of 27.9 cm and power of 450 W, surrounded by a Pyrex UV filter with 50% transmittance at 305 nm). The resin was slowly injected from a disposable syringe (2 mL) and pushed through the tubing with 15 mL CH_2Cl_2 (flow rate: 300 μL·min^{-1}). The tubing was washed with 15 mL CH_2Cl_2/MeOH (1:1 v/v, flow rate: 300 μL·min^{-1} for 8 mL and 4 mL·min^{-1} for 7 mL), and finally with 15 mL MeOH (flow rate: 4 mL·min^{-1}). The suspension leaving the reactor was directed into a filter where the resin was filtered off and washed with CH_2Cl_2/MeOH (1:1 v/v), MeOH and CH_2Cl_2, while the filtrate was collected. The tubing was re-equilibrated with 15 mL CH_2Cl_2 using a flow rate of 4 mL·min^{-1}. The entire cleavage procedure was repeated three times. The resulting solution was concentrated *in vacuo* to afford compound **6** (25 mg, 6.0 μmol) as a white solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.86 (d, *J* = 7.2 Hz, 2H), 7.78 – 7.69 (m, 3H), 7.54 – 7.04 (m, 96H), 5.04 (s, 2H), 4.99 – 4.13 (m, 52H), 4.07 – 3.94 (m, 2H), 3.85 – 2.99 (m, 45H), 2.84 – 2.74 (m, 2H), 1.23 – 1.10 (m, 4H), 1.06 – 0.94 (m, 2H); ^{13}C NMR (151 MHz, $CDCl_3$) δ 166.7, 166.4, 164.5, 164.5, 164.4, 164.3, 164.3, 164.2, 138.7, 138.6, 138.6, 138.6, 138.5, 138.4, 138.3, 138.0, 133.3, 133.3, 133.2, 133.1, 133.1, 133.0, 130.2, 130.0, 129.8, 129.8, 129.7, 129.6, 129.3, 128.7, 128.6, 128.6, 128.6, 128.5, 128.4, 128.4, 128.4, 128.4, 128.3, 128.3, 128.2, 128.2, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7, 127.7, 127.6, 127.6, 127.6, 127.5, 127.5, 127.4, 127.4, 100.7, 100.6, 100.5, 100.4, 100.3, 99.9, 80.5, 80.3, 80.0, 79.8, 79.1, 78.5, 78.4, 76.4, 76.3, 76.2, 76.1, 76.0, 75.5, 75.5, 75.4, 75.3, 75.2, 75.2, 75.1, 75.1, 75.0, 74.9, 74.8, 74.7, 74.6, 74.5, 74.1, 74.0, 73.9, 73.9, 73.6, 73.5, 73.5, 73.4, 73.4, 73.3, 69.7, 69.5, 69.4, 69.3, 69.2, 68.9, 67.6, 66.5, 40.9, 29.4, 23.1; MALDI-TOF MS calcd for $C_{249}H_{247}NO_{57}Na$ $[M + Na]^+$, 4187.6420; found, 4187.365.

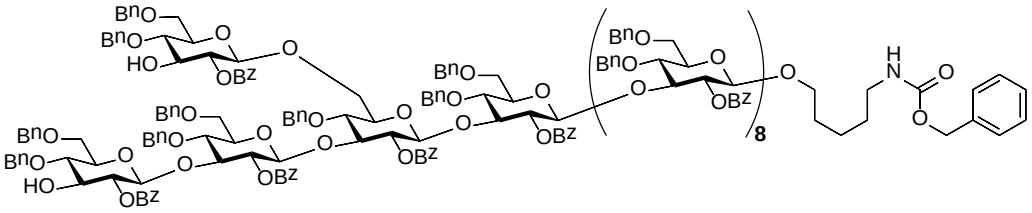
5-Aminopentyl β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-[β-D-glucopyranosyl-(1→6)]-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranoside (10).



Compound **6** (25 mg, 6.0 µmol) was dissolved in CH_2Cl_2, a 0.5 M solution of NaOMe in MeOH (120 µL, 60 µmol) was added, and the reaction was stirred overnight. The solution was neutralized with Amberlite® IR-120 (H^+ form) and filtered. The filtrate was concentrated *in vacuo* to afford the deacylated intermediate (19 mg) which was directly used in the next reaction step without further purification. *t*BuOH (10 µL) was added to a round bottom flask and cooled to -78 °C. NH_3 was condensed into the flask at -78 °C, and sodium was added with vigorous stirring until the solution acquired a deep blue color. The deacylated intermediate (19 mg, 6.0 µmol) was dissolved in THF and added dropwise to the solution. After 30 minutes, the reaction was quenched by the dropwise addition of MeOH, and the ammonia was evaporated. The remaining liquid was neutralized with AcOH and concentrated *in vacuo*. The residue was dissolved in H_2O and purified by size exclusion chromatography over Sephadex® G-10 to afford compound **10** (1.3 mg, 0.8 µmol) as a white solid after lyophilization. ^1H NMR (600 MHz, D_2O) δ 4.79 – 4.68 (m, 7H), 4.55 (d, J = 7.9 Hz, 1H), 4.50 (d, J = 7.9 Hz, 1H), 4.23 (d, J = 10.7 Hz, 1H), 4.00 – 3.88 (m, 10H), 3.84 – 3.29 (m, 45H), 3.00 (t, J = 7.1 Hz, 2H), 1.74 – 1.62 (m, 4H), 1.52 – 1.42 (m, 2H); ^{13}C NMR (151 MHz, D_2O) δ 105.4, 105.4, 105.2, 105.1, 105.1, 104.5, 87.3, 87.0, 86.8, 86.7, 86.4, 78.6, 78.5, 78.2, 78.2, 78.2, 77.1, 76.1, 75.9, 75.9, 75.8, 75.8, 75.7, 75.5, 72.7, 72.2, 71.3, 70.8, 70.7, 70.7, 70.7, 63.3, 42.0, 30.8, 29.2, 24.7; HSQC (600 MHz, $CDCl_3$) δ 105.4 ($J_{C1,H1}$ = 164 Hz, C-1), 105.1 ($J_{C1,H1}$ = 162 Hz, C-1), 104.5 ($J_{C1,H1}$ = 162 Hz, C-1). HRMS calcd for $C_{59}H_{103}NO_{46}HNa$ $[M + H + Na]^{2+}$, 792.7858; found, 792.7835.

***N*-Benzylcarbamoyl aminopentyl 4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-[4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→6)]-4-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-**

benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranoside (7).



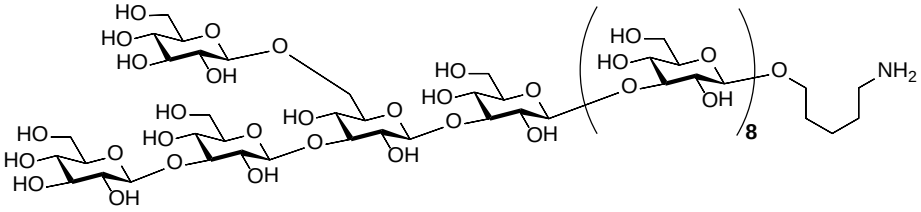
Glucose Unit	Modules (Iterations)
1-8	**D**(1), **A2** (2), **F** (1), **G** (3), **H** (1), **E** (1)
9	**D**(1), **A2** (2), **F** (1), **G** (3), **H** (1), **E** (1)
10	**D**(1), **B2** (2), **F** (1), **G** (3), **H** (1), **E** (1)
11	**D**(1), **A2*** (2), **F** (1), **G** (3), **H** (1), **E** (1)
12	**D**(1), **A2** (2), **F** (1), **I** (3), **J** (1), **E** (1)
13	**D**(1), **A2** (2), **F** (1), **G** (3), **H** (1)

Table 3. Program modules for the synthesis of tridecasaccharide **7**.

The reaction vessel of the synthesizer was charged with functionalized Merrifield resin **4** (65 mg, loading: 0.39 mmol/g). Program module **C** was executed once. Then the program described in Table 3 was executed by the synthesizer. The resin was removed from the reaction vessel and swelled in CH_2Cl_2. The suspension was irradiated with UV light by delivering the suspension *via* syringe pump through a FEP tubing (inner diameter: 0.03 inch; volume: 12 mL) wrapped around a UV light source (medium pressure Hg lamp with arc lengths of 27.9 cm and power of 450 W, surrounded by a Pyrex UV filter with 50% transmittance at 305 nm). The resin was slowly injected from a disposable syringe (2 mL) and pushed through the tubing with 15 mL CH_2Cl_2 (flow rate: 300 µL·min^{-1}). The tubing was washed with 15 mL CH_2Cl_2/MeOH (1:1 v/v, flow rate: 300 µL·min^{-1} for 8 mL and 4 mL·min^{-1} for 7 mL), and finally with 15 mL MeOH (flow rate: 4 mL·min^{-1}). The suspension leaving the reactor was directed into a filter where the resin was filtered off and washed with CH_2Cl_2/MeOH (1:1 v/v), MeOH and CH_2Cl_2, while the filtrate was collected. The tubing was re-equilibrated with 15 mL CH_2Cl_2 using a flow rate of 4 mL·min^{-1}. The entire cleavage procedure was repeated three times. The resulting solution was concentrated *in vacuo* to afford compound **7** as a white solid (118 mg, 19.8 µmol). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.85 (d, *J* = 7.4 Hz, 2H), 7.79 – 7.68 (m, 3H), 7.55 – 7.03 (m, 139H), 5.04 (s, 2H), 5.01 – 4.13 (m, 76H), 4.08 – 2.96 (m, 67H), 2.85 – 2.75 (m, 2H), 1.20 – 1.13 (m, 4H), 1.06 – 0.94 (m, 2H); ^{13}C NMR (101 MHz, $CDCl_3$) δ

166.7, 166.4, 164.4, 164.4, 164.3, 164.2, 164.1, 138.6, 138.4, 138.3, 138.0, 133.3, 133.1, 130.2, 130.0, 129.8, 129.7, 129.6, 129.4, 129.3, 128.7, 128.6, 128.6, 128.5, 128.4, 128.3, 128.3, 128.2, 128.2, 128.0, 127.9, 127.7, 127.6, 127.5, 127.4, 100.8, 100.6, 100.5, 100.4, 100.3, 99.9, 80.4, 80.2, 80.1, 79.9, 79.8, 79.1, 78.5, 78.4, 77.4, 76.4, 76.3, 76.2, 76.1, 75.5, 75.2, 75.0, 74.9, 74.7, 74.5, 74.1, 73.9, 73.8, 73.6, 73.5, 73.4, 73.3, 69.4, 69.3, 69.2, 68.9, 67.6, 66.5, 40.9, 29.3, 23.1; HSQC (400 MHz, $CDCl_3$) δ 100.8 ($J_{C1,H1}$ = 162 Hz, C-1), 100.6 ($J_{C1,H1}$ = 163 Hz, C-1), 100.5 ($J_{C1,H1}$ = 162 Hz, C-1), 100.4 ($J_{C1,H1}$ = 161 Hz, C-1), 100.3 ($J_{C1,H1}$ = 162 Hz, C-1), 99.9 ($J_{C1,H1}$ = 161 Hz, C-1).

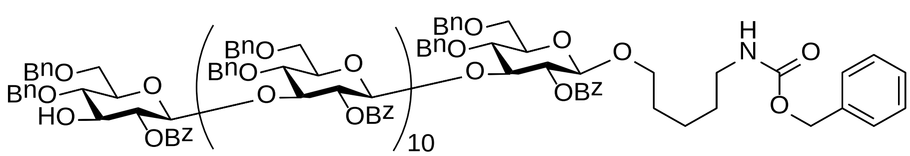
5-Aminopentyl β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-[β-D-glucopyranosyl-(1→6)]-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranoside (11).



Compound **7** (18 mg, 3.0 µmol) was dissolved in CH_2Cl_2, a 0.5 M solution of NaOMe in MeOH (90 µL, 45 µmol) was added, and the reaction was stirred overnight. The solution was neutralized with Amberlite® IR-120 (H^+ form) and filtered. The filtrate was concentrated *in vacuo* to afford the deacylated intermediate (14 mg) which was directly used in the next reaction step without further purification. *t*BuOH (10 µL) was added to a round bottom flask and cooled to -78 °C. NH_3 was condensed into the flask at -78 °C, and sodium was added with vigorous stirring until the solution acquired a deep blue color. The deacylated intermediate (14 mg, 3.0 µmol) was dissolved in THF and added dropwise to the solution. After 30 minutes, the reaction was quenched by the dropwise addition of MeOH, and the ammonia was evaporated. The remaining liquid was neutralized with AcOH and concentrated *in vacuo*. The residue was dissolved in H_2O and purified by size exclusion chromatography over Sephadex® G-10 to afford compound **11** (4.0 mg, 1.8 µmol) as a white solid after lyophilization. ^{1}H NMR (600 MHz, D_2O) δ 4.85 – 4.75 (m, 11H), 4.54 (d, *J* = 7.7 Hz, 1H), 4.50 (d, *J* = 7.6 Hz, 1H), 4.23 (d, *J* = 11.0 Hz, 1H), 4.02 – 3.87 (m, 14H), 3.85 –

3.30 (m, 65H), 3.02 (t, J = 7.1 Hz, 2H), 1.76 – 1.62 (m, 4H), 1.52 – 1.43 (m, 2H); ^{13}C NMR (151 MHz, D_2O) δ 105.4, 105.2, 105.1, 104.5, 87.3, 87.0, 86.8, 86.7, 86.4, 78.6, 78.5, 78.2, 78.2, 77.1, 76.1, 75.9, 75.9, 75.8, 75.8, 75.7, 75.5, 72.7, 72.2, 71.3, 70.8, 70.7, 63.3, 42.0, 30.8, 29.0, 24.7; HSQC (600 MHz, $CDCl_3$) δ 105.4 ($J_{C1,H1}$ = 161 Hz, C-1), 105.2 ($J_{C1,H1}$ = 162 Hz, C-1), 105.1 ($J_{C1,H1}$ = 163 Hz, C-1), 104.5 ($J_{C1,H1}$ = 161 Hz, C-1); HRMS calcd for $C_{83}H_{143}NO_{66}HNa$ [M + H + Na]$^{2+}$, 1116.8915; found,1116.8907.

***N*-Benzylcarbamoyl aminopentyl 4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranosyl-(1→3)-4,6-di-*O*-benzyl-2-*O*-benzoyl-β-D-glucopyranoside (8).**



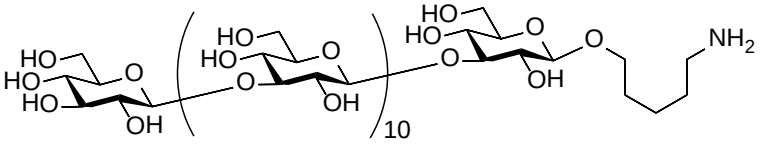
Glucose Unit	Modules (Iterations)
1-12	**D**(1), **A2** (2), **F** (1), **G** (3), **H** (1), **E** (1)

Table 4. Program modules for the synthesis of dodecasaccharide **8**.

The reaction vessel of the synthesizer was charged with functionalized Merrifield resin **4** (65 mg, loading: 0.39 mmol/g). Program module **C** was executed once. Then the program described in Table 4 was executed by the synthesizer. The resin was removed from the reaction vessel and swelled in CH_2Cl_2. The suspension was irradiated with UV light by delivering the suspension *via* syringe pump through a FEP tubing (inner diameter: 0.03 inch; volume: 12 mL) wrapped around a UV light source (medium pressure Hg lamp with arc lengths of 27.9 cm and power of 450 W, surrounded by a Pyrex UV filter with 50% transmittance at 305 nm). The resin was slowly injected

from a disposable syringe (2 mL) and pushed through the tubing with 15 mL CH_2Cl_2 (flow rate: 300 µL·min^{-1}). The tubing was washed with 15 mL CH_2Cl_2/MeOH (1:1 v/v, flow rate: 300 µL·min^{-1} for 8 mL and 4 mL·min^{-1} for 7 mL), and finally with 15 mL MeOH (flow rate: 4 mL·min^{-1}). The suspension leaving the reactor was directed into a filter where the resin was filtered off and washed with CH_2Cl_2/MeOH (1:1 v/v), MeOH and CH_2Cl_2, while the filtrate was collected. The tubing was re-equilibrated with 15 mL CH_2Cl_2 using a flow rate of 4 mL·min^{-1}. The entire cleavage procedure was repeated three times. The resulting solution was concentrated *in vacuo* to afford compound **8** as a white solid (121 mg, 21.6 µmol). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.74 (d, *J* = 7.1 Hz, 2H), 7.69 (d, *J* = 8.1 Hz, 2H), 7.55 – 7.01 (m, 181H), 5.04 (s, 2H), 5.02 – 3.99 (m, 73H), 3.80 – 3.02 (m, 61H), 2.86 – 2.71 (m, 2H), 1.21 – 1.10 (m, 4H), 1.07 – 0.93 (m, 2H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 166.7, 164.5, 164.2, 164.2, 164.2, 164.1, 138.7, 138.5, 138.5, 138.5, 138.4, 138.4, 138.3, 138.0, 133.3, 133.1, 130.1, 130.0, 129.8, 129.7, 129.5, 129.4, 129.3, 129.3, 129.2, 128.7, 128.6, 128.5, 128.5, 128.4, 128.3, 128.3, 128.2, 128.2, 128.2, 128.1, 127.9, 127.7, 127.7, 127.6, 127.6, 127.5, 127.5, 100.7, 100.7, 100.6, 100.4, 99.9, 80.5, 80.3, 80.1, 79.1, 78.4, 76.4, 76.2, 76.1, 75.4, 75.1, 75.1, 75.0, 74.8, 74.0, 74.0, 73.9, 73.8, 73.7, 73.7, 73.5, 73.4, 69.7, 69.4, 69.3, 69.2, 68.8, 66.5, 40.8, 29.3, 23.1; HSQC (400 MHz, $CDCl_3$) 100.7 ($J_{C1,H1}$ = 163 Hz, C-1), 100.7 ($J_{C1,H1}$ = 163 Hz, C-1), 100.6 ($J_{C1,H1}$ = 163 Hz, C-1), 100.4 ($J_{C1,H1}$ = 163 Hz, C-1), 99.9 ($J_{C1,H1}$ = 163 Hz, C-1).

5-Aminopentyl β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranoside (12).

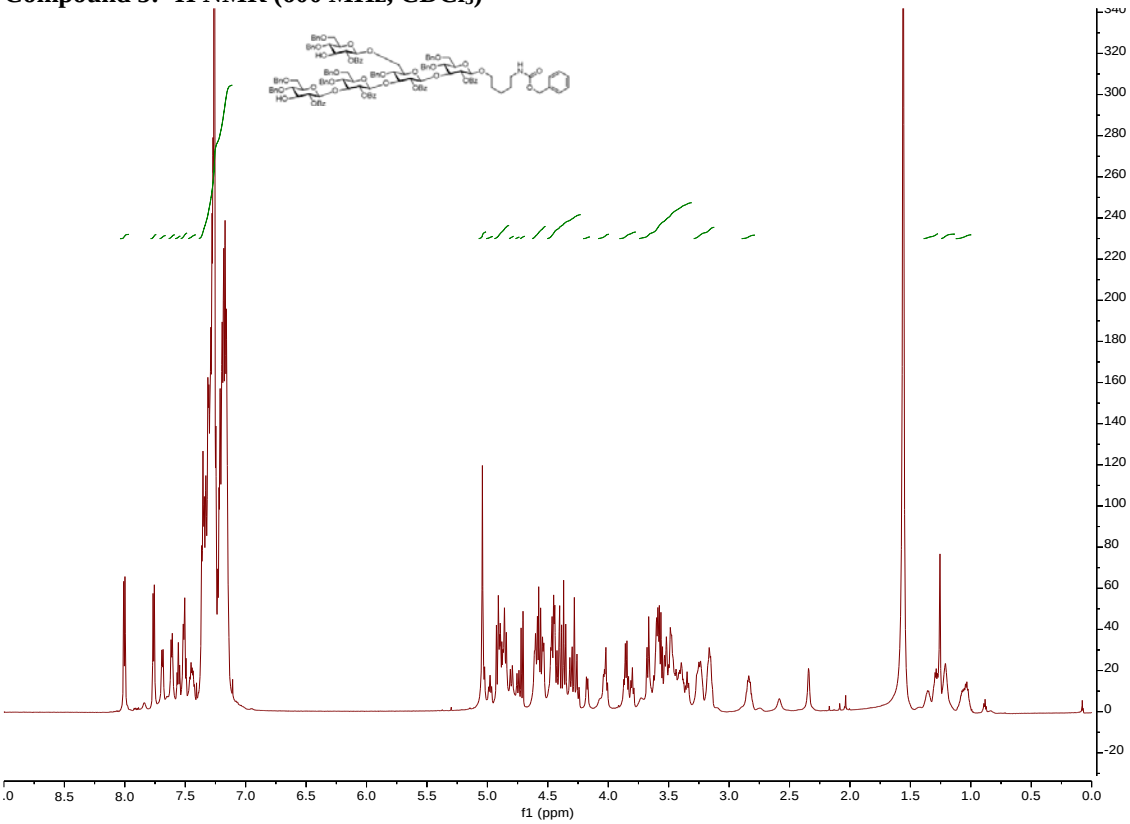


Compound **8** (20 mg, 3.6 µmol) was dissolved in CH_2Cl_2, a 0.5 M solution of NaOMe in MeOH (108 µL, 54 µmol) was added, and the reaction was stirred overnight. The solution was neutralized with Amberlite® IR-120 (H^+ form) and filtered. The filtrate was concentrated *in vacuo* to afford the deacylated intermediate (16 mg, 3.6 µmol), which was used in the next reaction step without further purification. *t*BuOH (10 µL) was added to a round bottom flask and cooled to -78 °C. NH_3 was condensed into the flask at -78 °C, and sodium was added with vigorous stirring until the

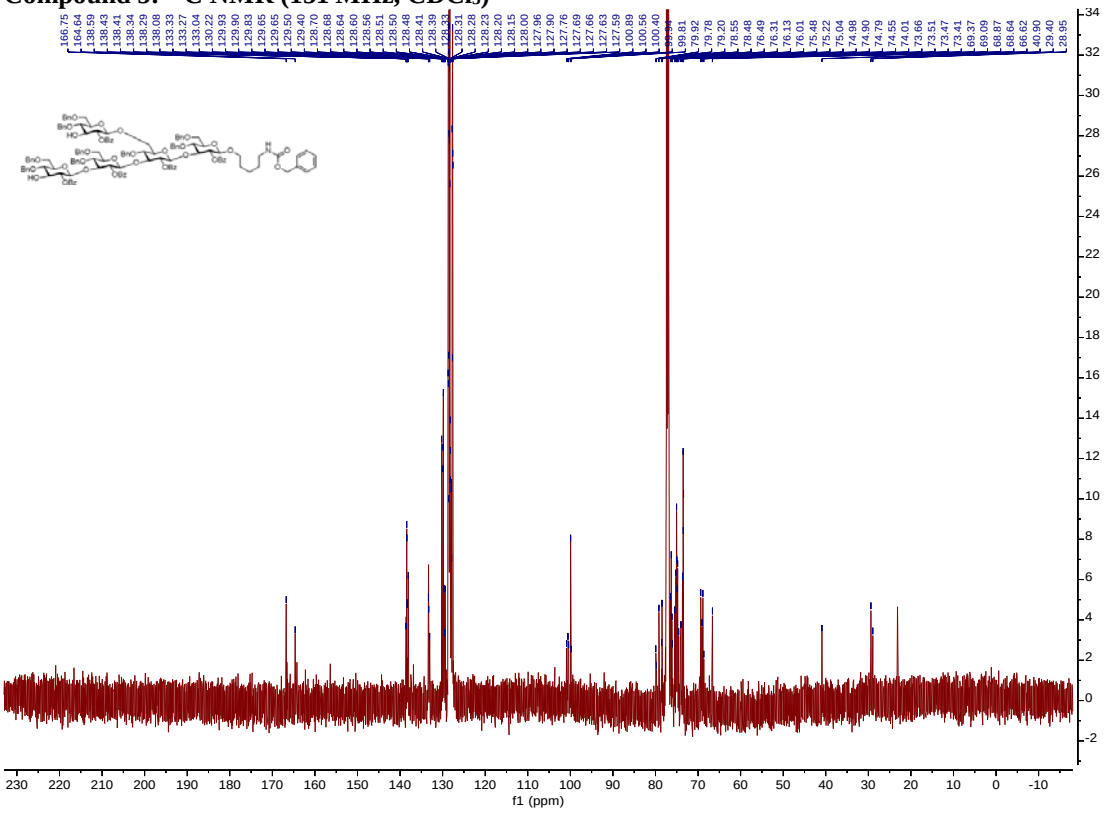
solution acquired a deep blue color. The deacylated intermediate (16 mg, 3.6 µmol) was dissolved in THF and added dropwise to the solution. After 30 minutes, the reaction was quenched by the dropwise addition of MeOH, and the ammonia was evaporated. The remaining liquid was neutralized with AcOH and concentrated *in vacuo*. The residue was dissolved in H_2O and purified by size exclusion chromatography over Sephadex® G-10 to afford compound **12** (5.9 mg, 2.9 µmol) as a white solid after lyophilization. ^{1}H NMR (600 MHz, D_2O) δ 4.84 – 4.76 (m, 11H), 4.50 (d, *J* = 8.0 Hz, 1H), 3.98 – 3.90 (m, 13H), 3.83 – 3.68 (m, 25H), 3.61 – 3.35 (m, 36H), 3.01 (t, *J* = 7.4 Hz, 2H), 1.75 – 1.64 (m, 4H), 1.53 – 1.42 (m, 2H); ^{13}C NMR (151 MHz, D_2O) δ 105.4, 105.1, 104.5, 87.0, 86.9, 86.7, 78.6, 78.2, 78.2, 78.2, 76.1, 75.9, 75.8, 75.5, 72.7, 72.18, 70.8, 70.7, 70.7, 63.3, 42.0, 30.8, 29.1, 24.7; HRMS calcd for $C_{77}H_{133}NO_{61}HNa$ $[M + H + Na]^{2+}$, 1035.8651; found, 1035.8721.

4. NMR Spectra

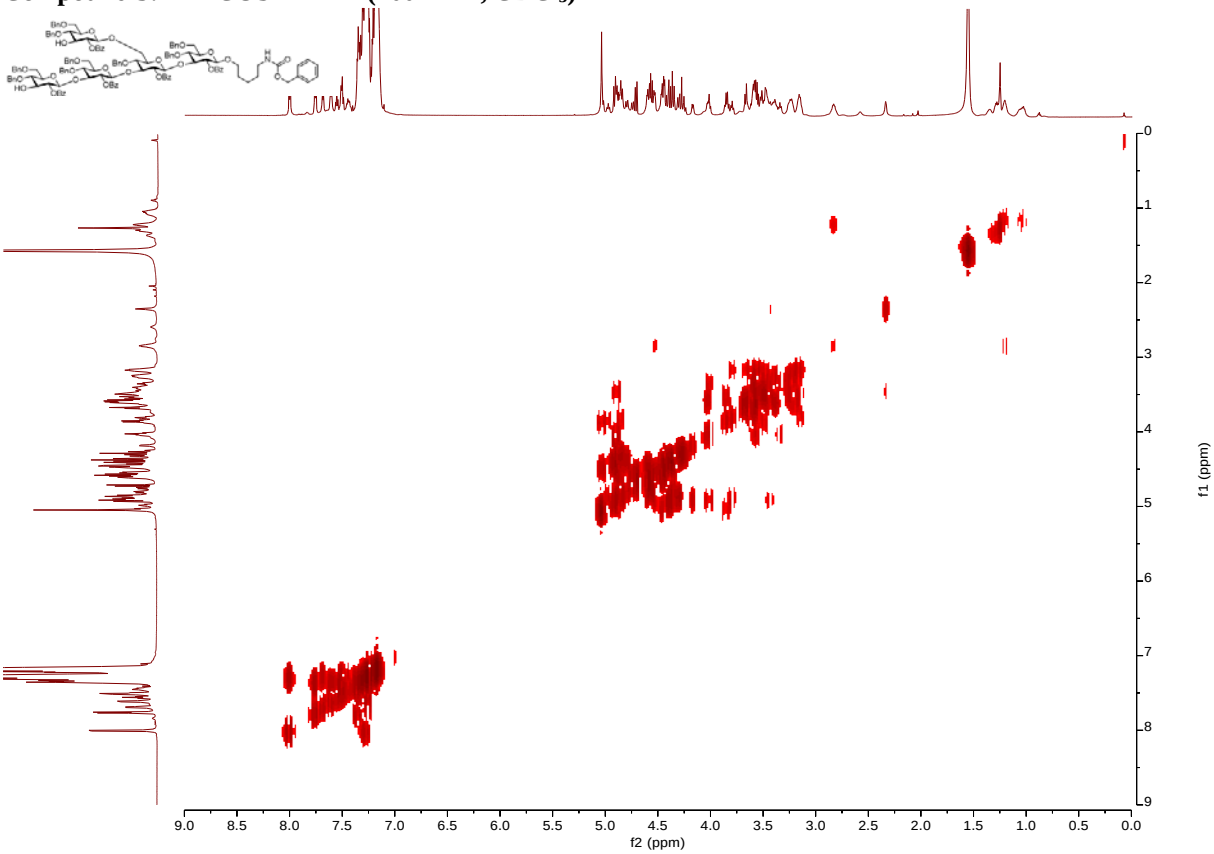
Compound 5: ^{1}H NMR (600 MHz, $CDCl_3$)



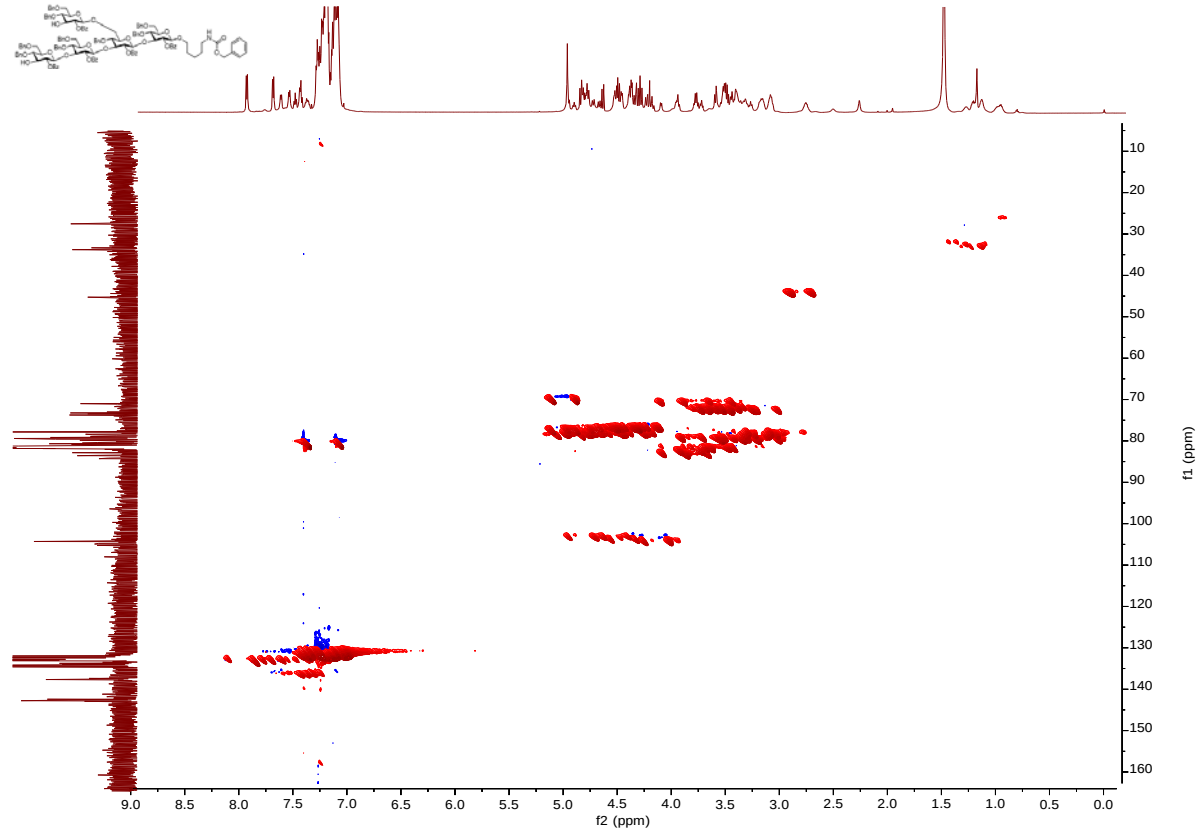
Compound 5: ^{13}C NMR (151 MHz, $CDCl_3$)



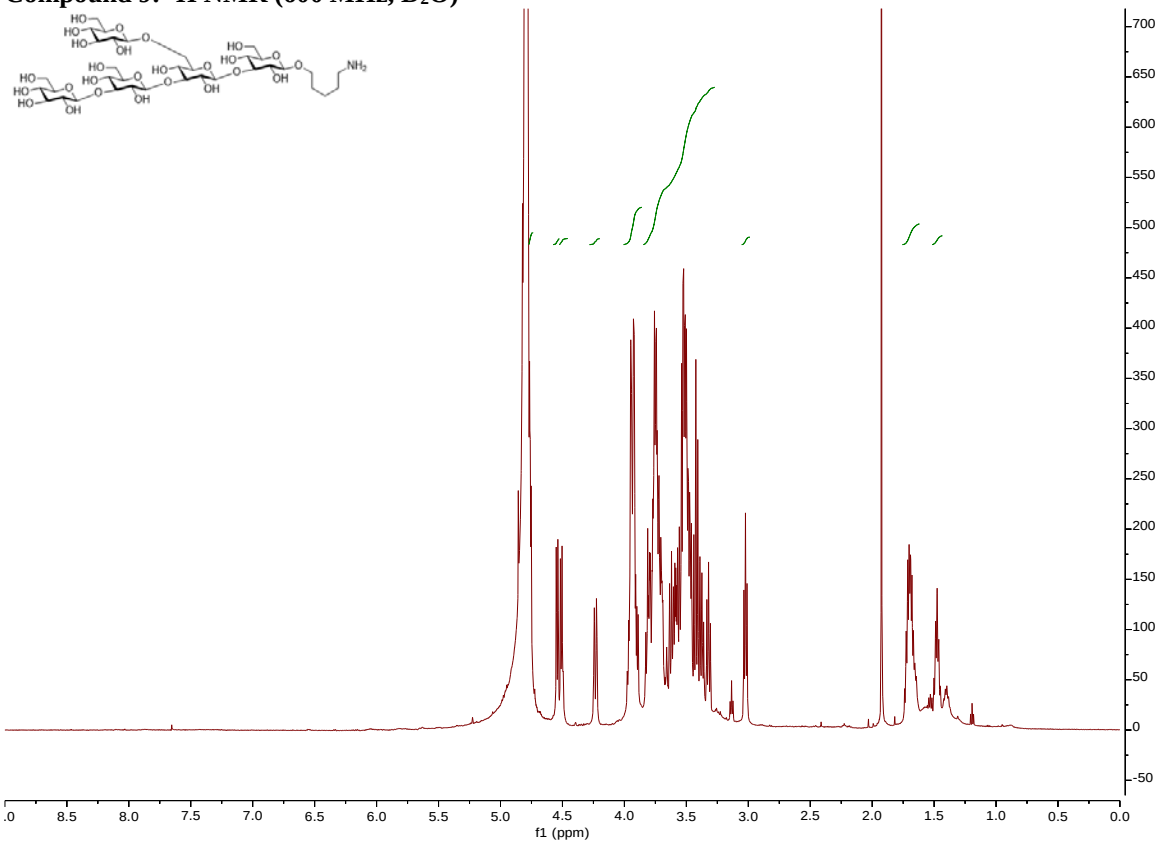
Compound 5: HH-COSY NMR (400 MHz, $CDCl_3$)



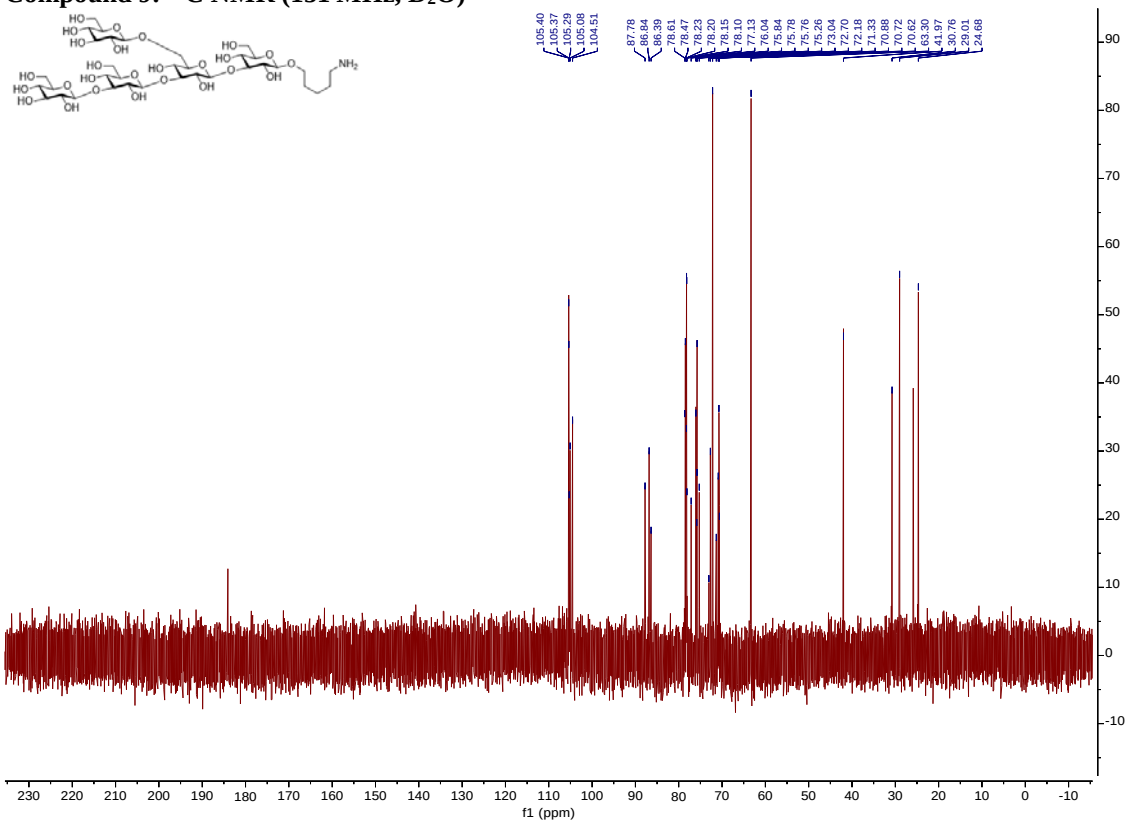
Compound 5: Coupled HSQC NMR (600 MHz, $CDCl_3$)



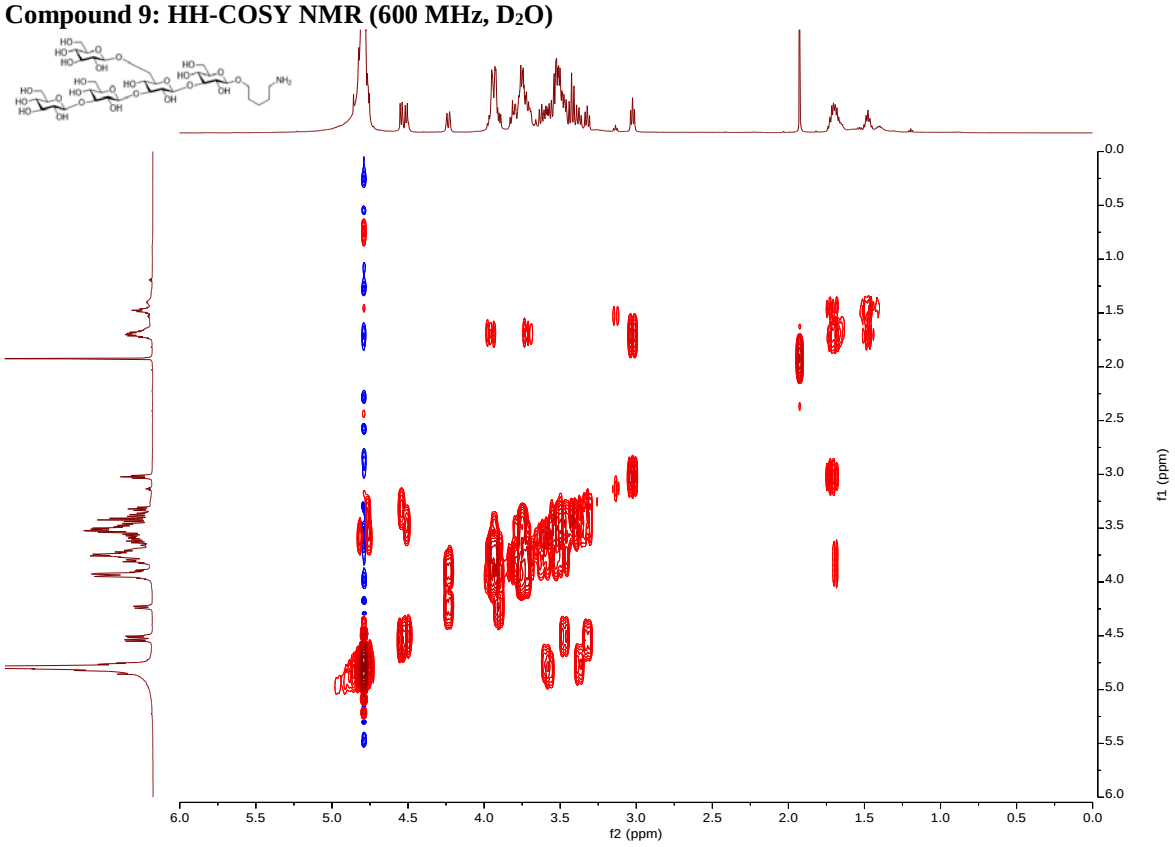
Compound 9: ^{1}H NMR (600 MHz, D_2O)



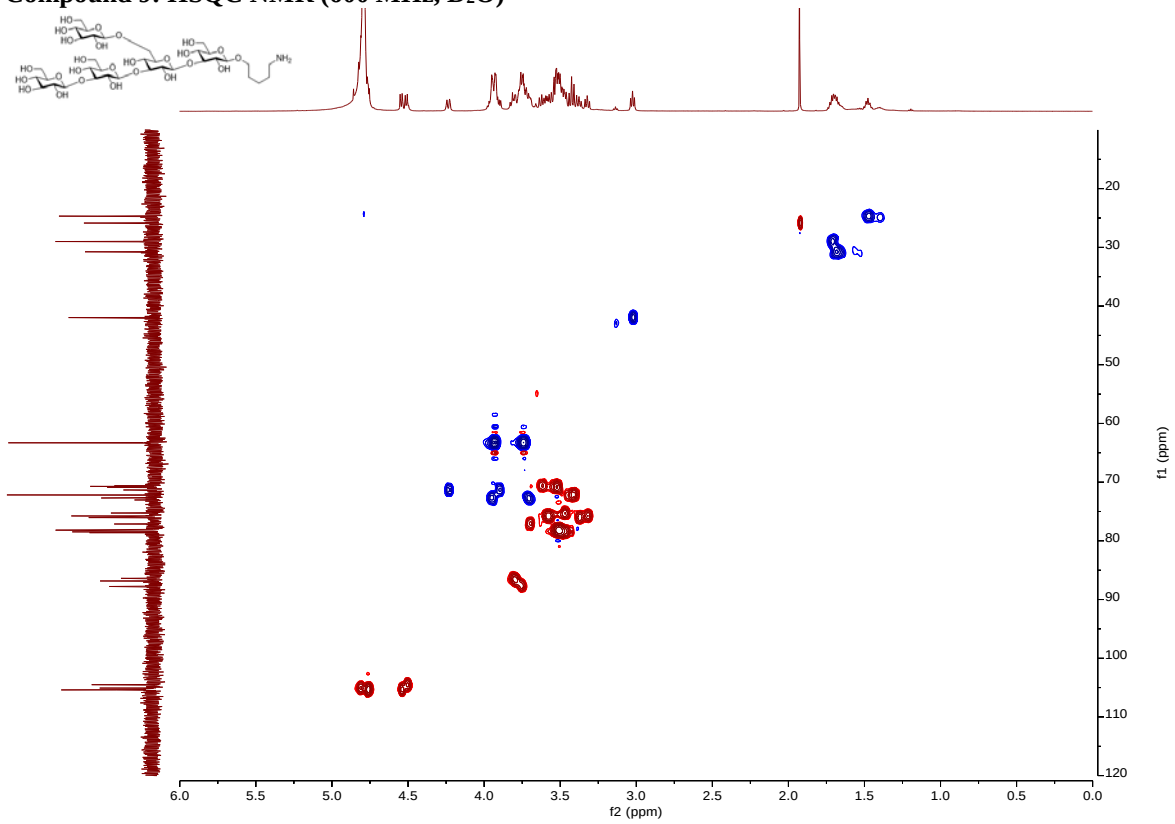
Compound 9: ^{13}C NMR (151 MHz, D_2O)



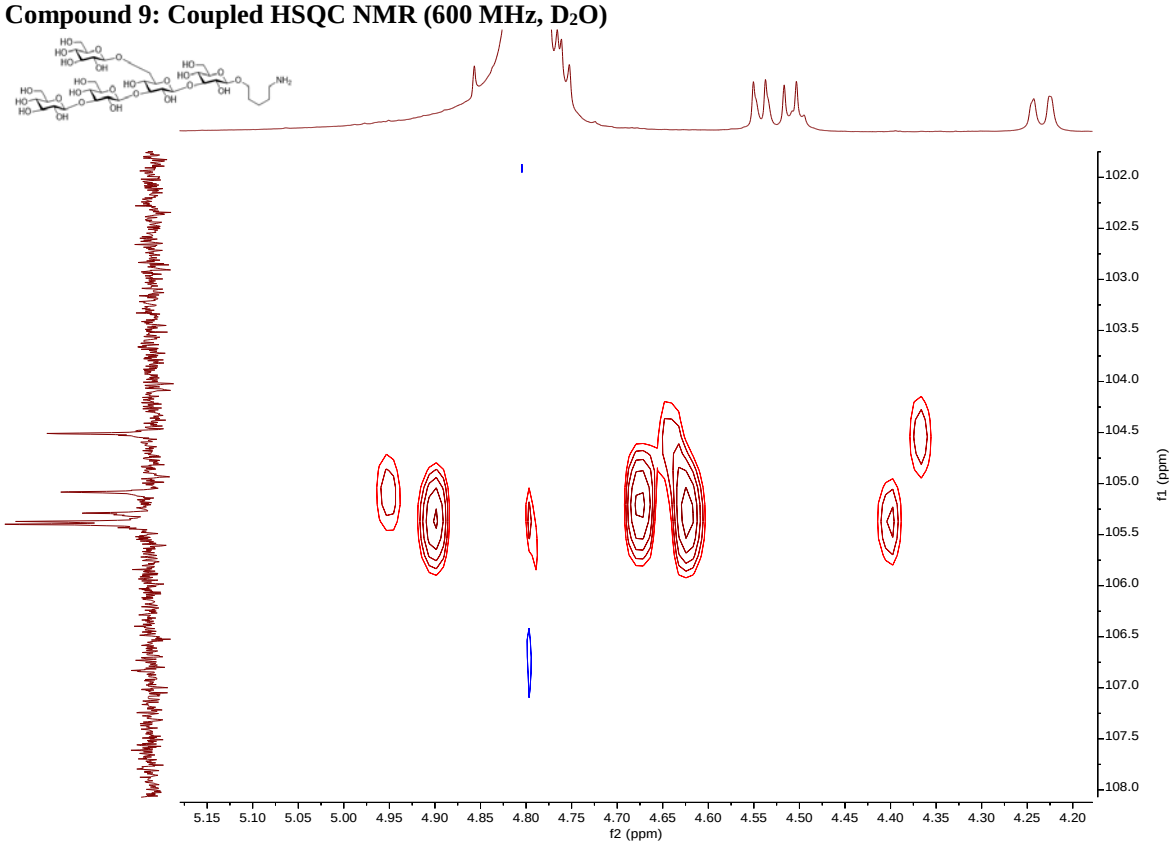
Compound 9: HH-COSY NMR (600 MHz, D_2O)



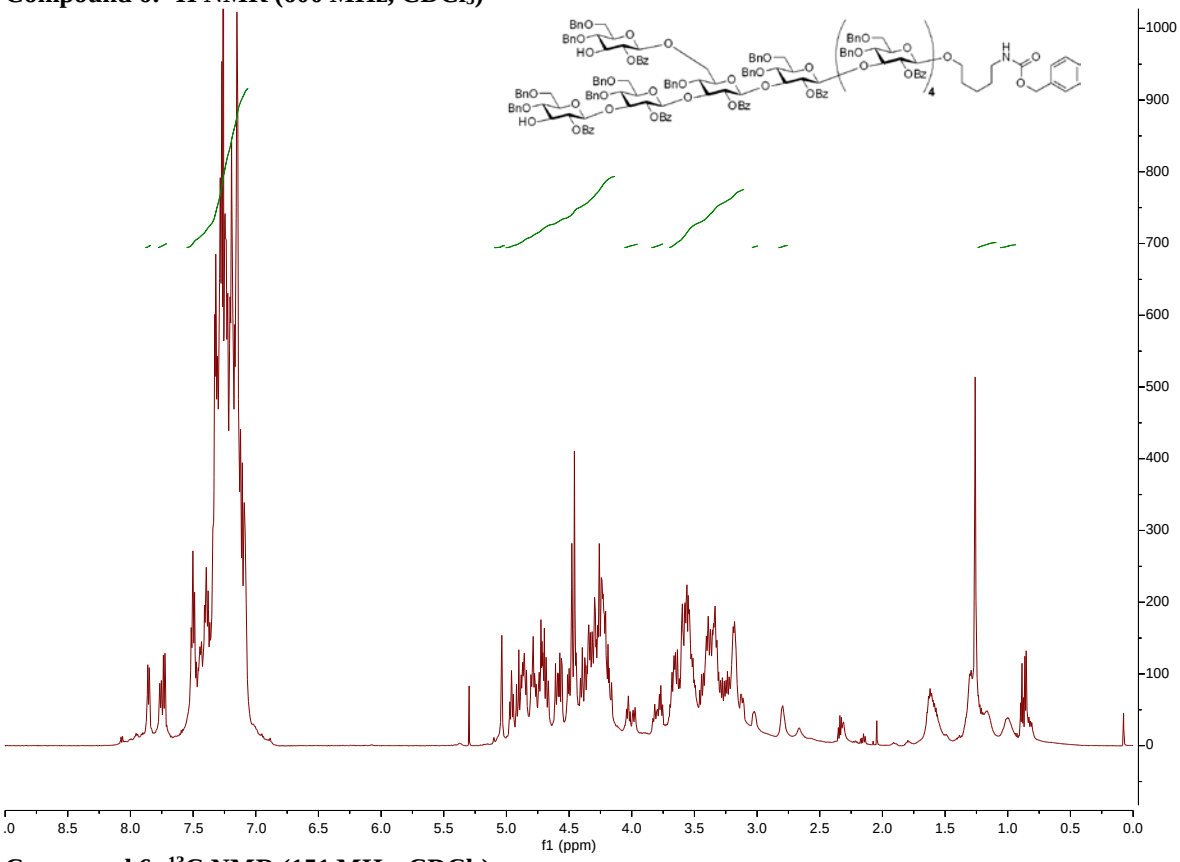
Compound 9: HSQC NMR (600 MHz, D_2O)



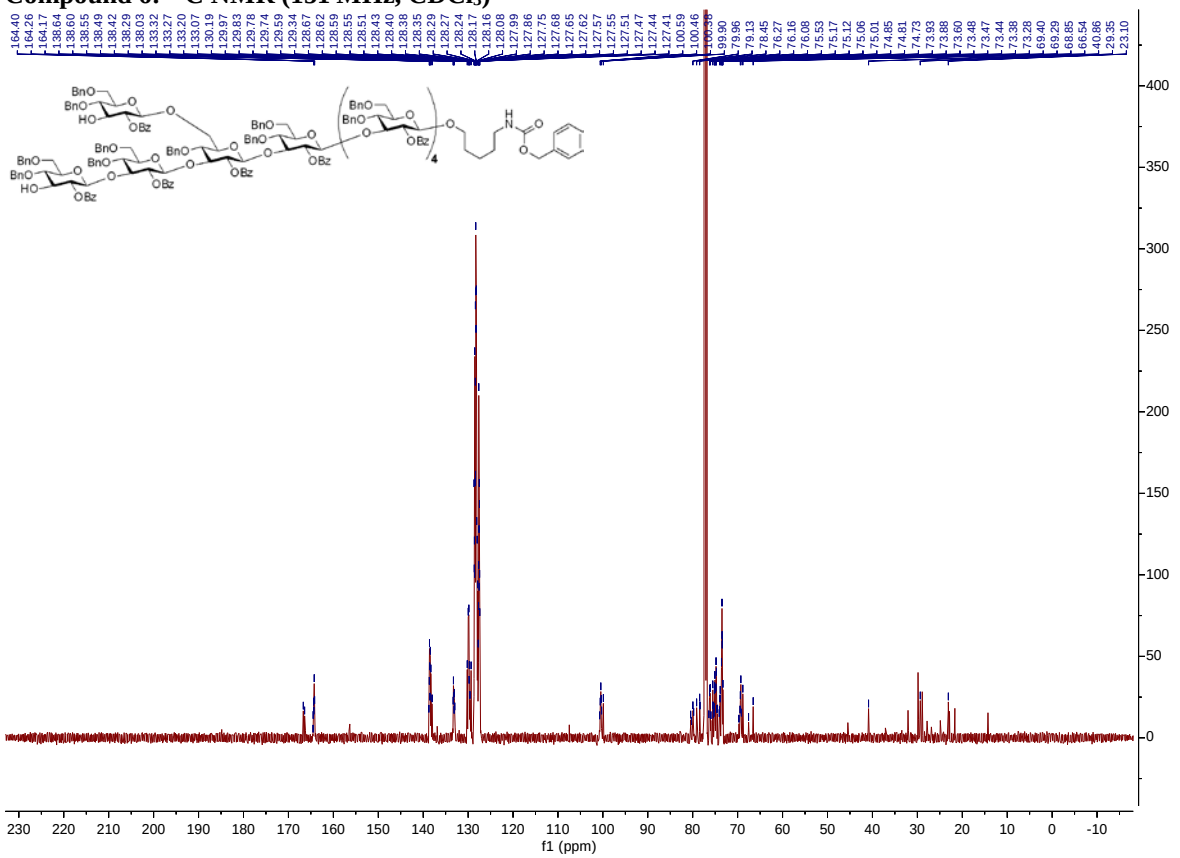
Compound 9: Coupled HSQC NMR (600 MHz, D_2O)



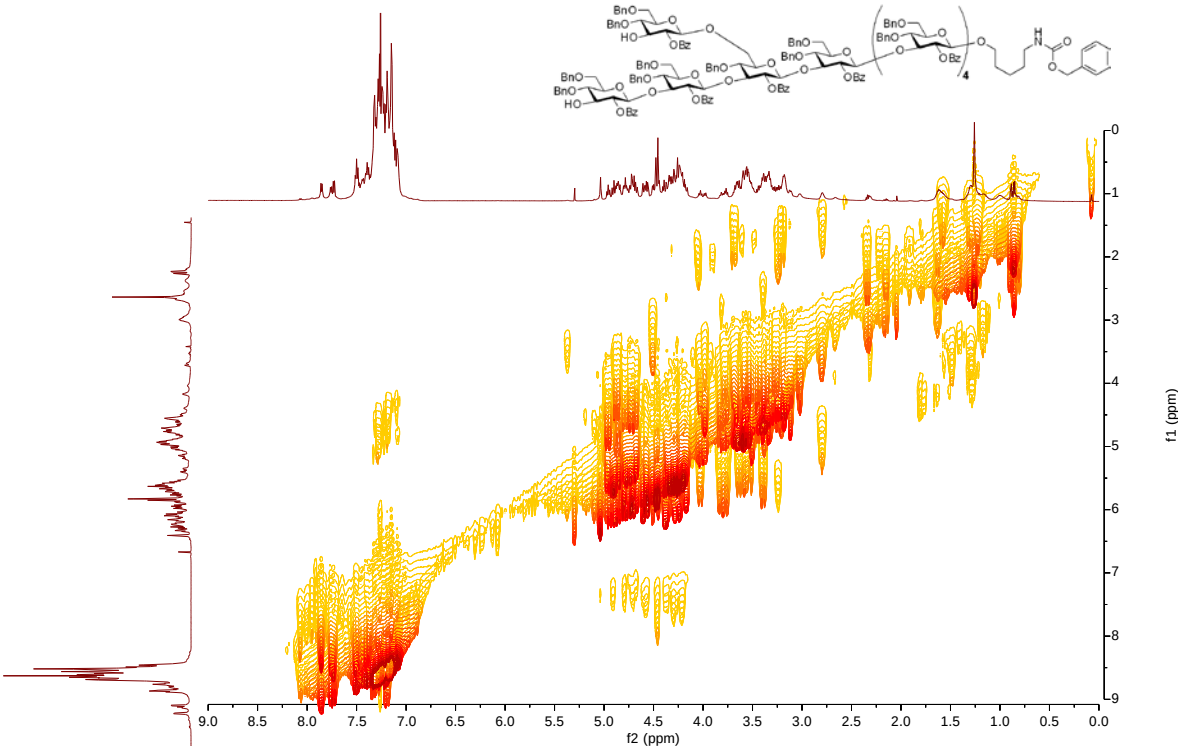
Compound 6: ^{1}H NMR (600 MHz, $CDCl_3$)



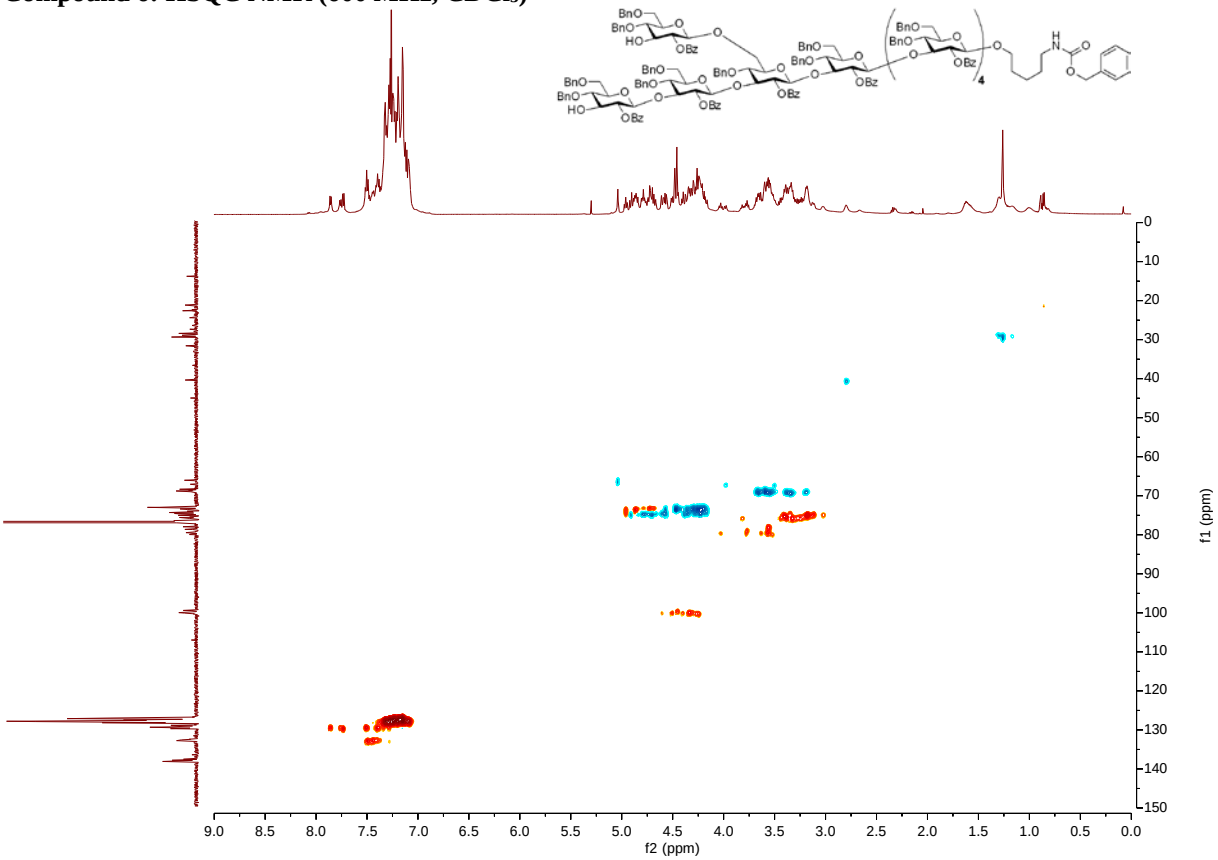
Compound 6: ^{13}C NMR (151 MHz, $CDCl_3$)



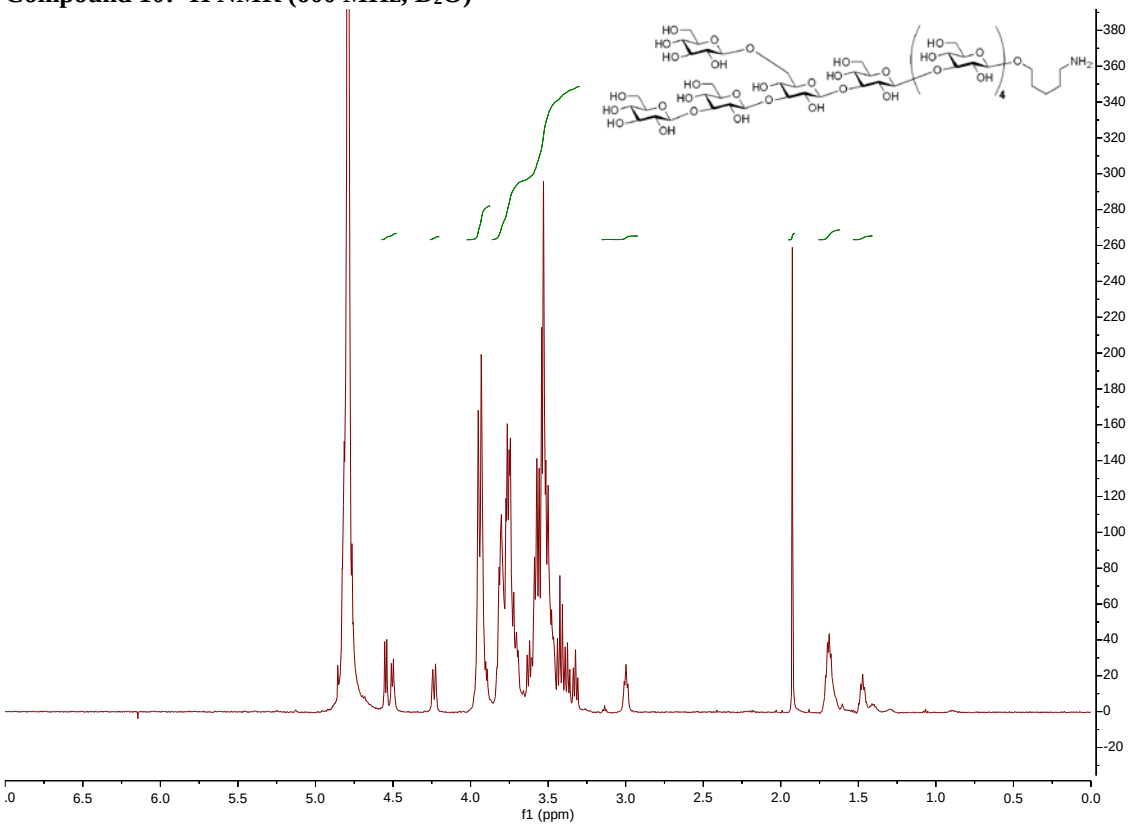
Compound 6: HH-COSY NMR (600 MHz, $CDCl_3$)



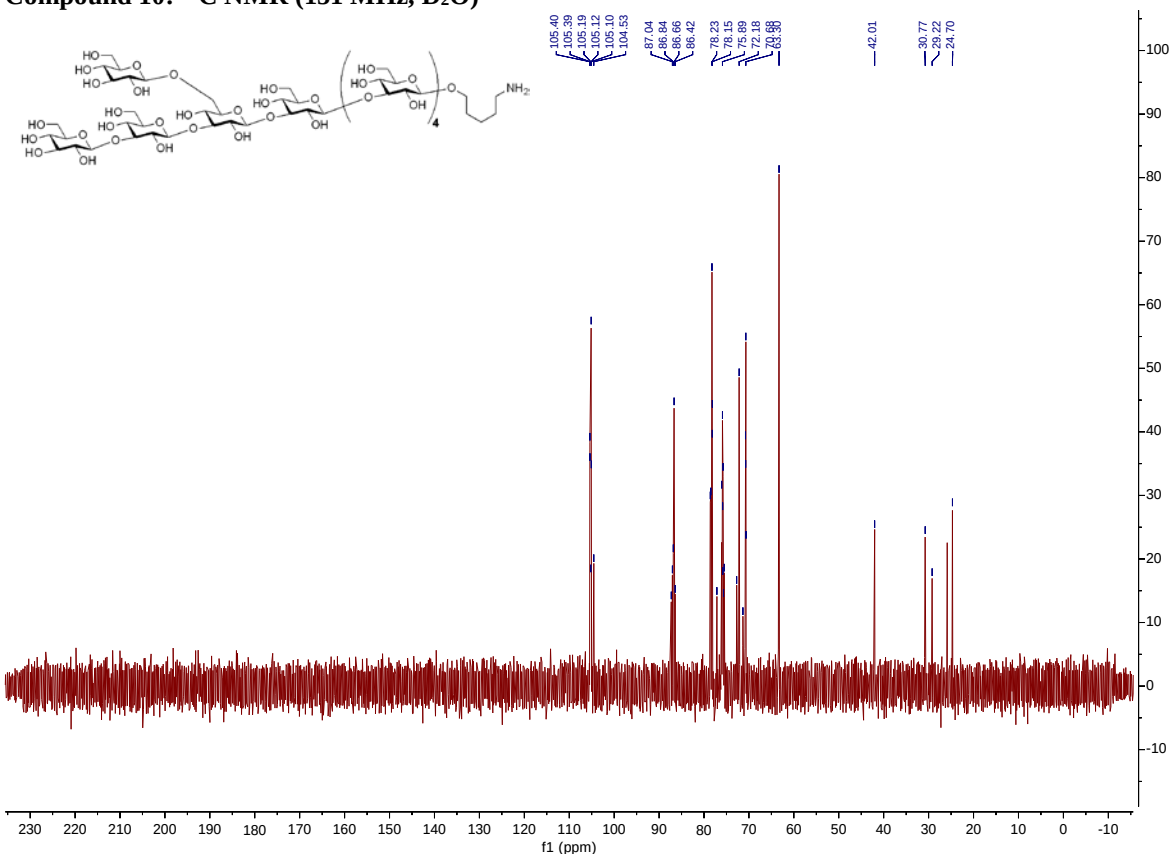
Compound 6: HSQC NMR (600 MHz, $CDCl_3$)



Compound 10: ^{1}H NMR (600 MHz, D_2O)



Compound 10: ^{13}C NMR (151 MHz, D_2O)



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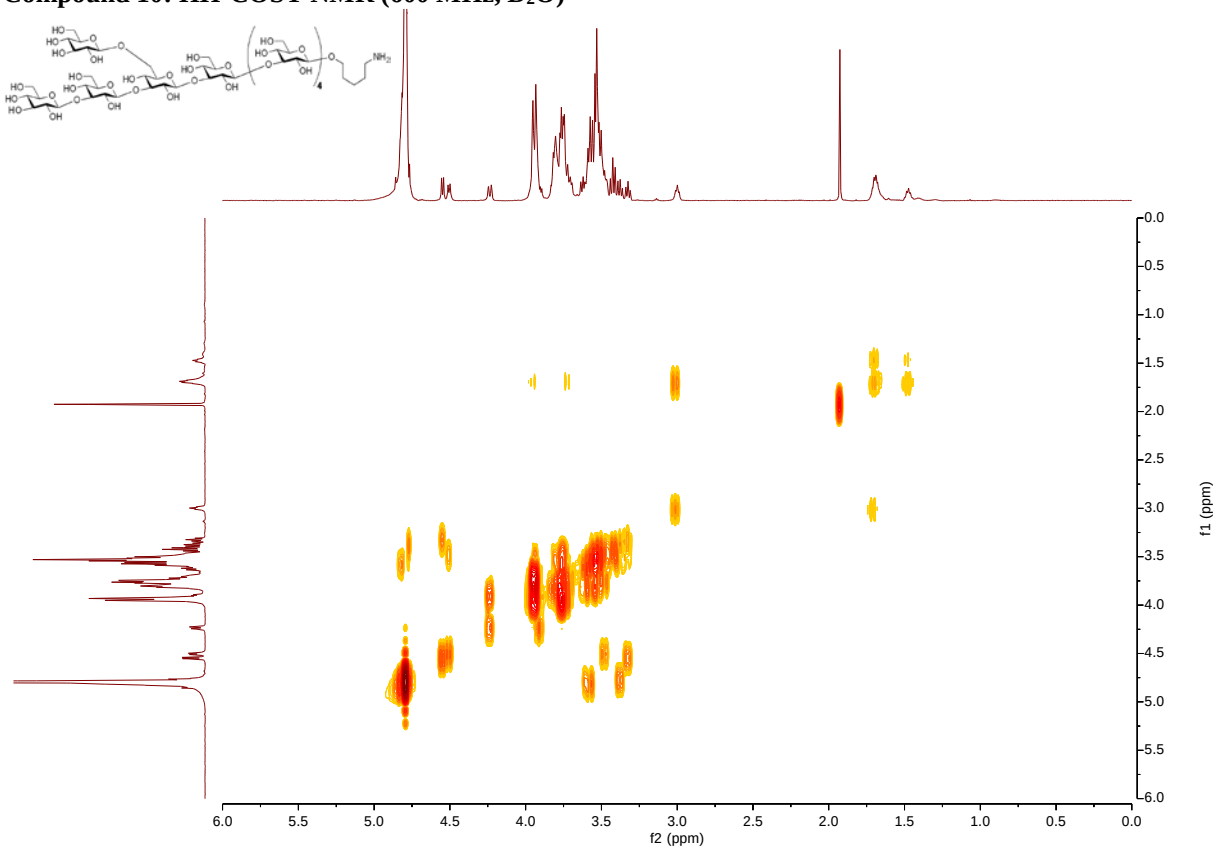

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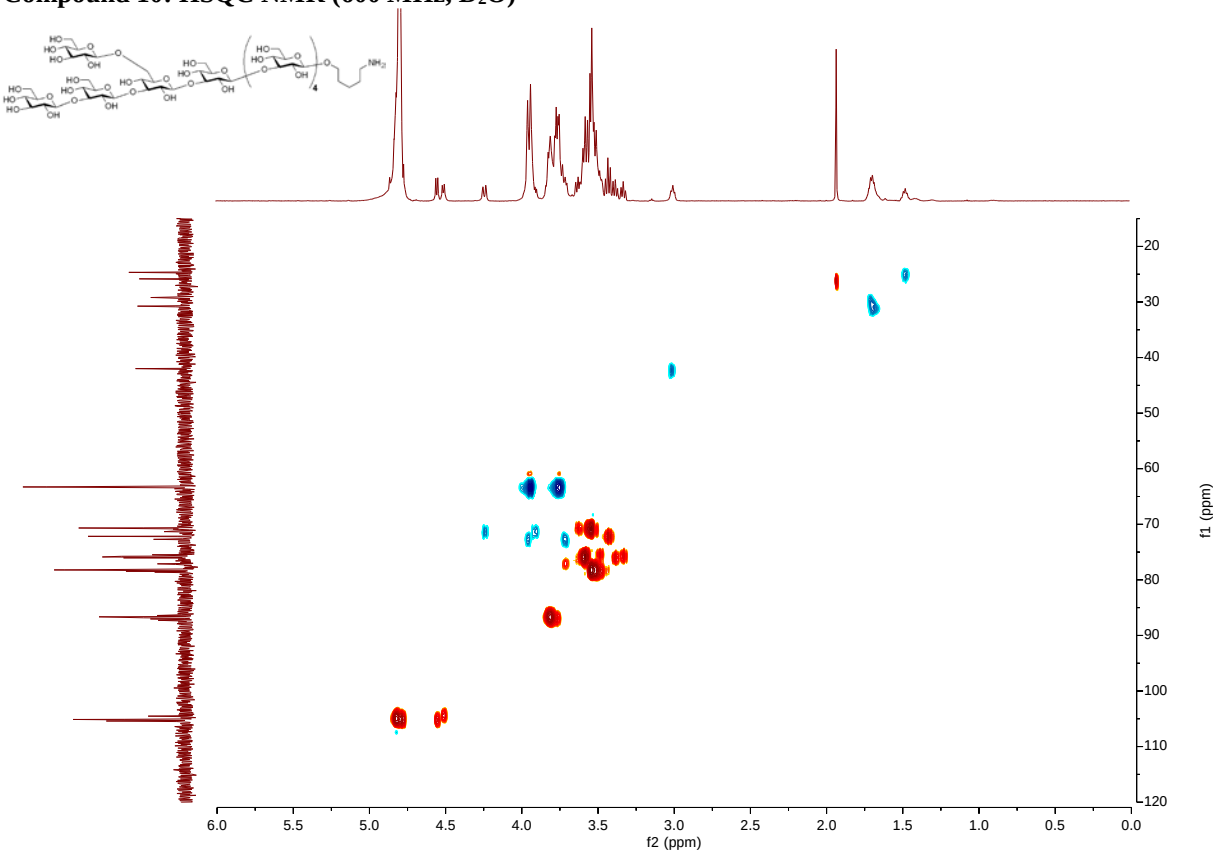
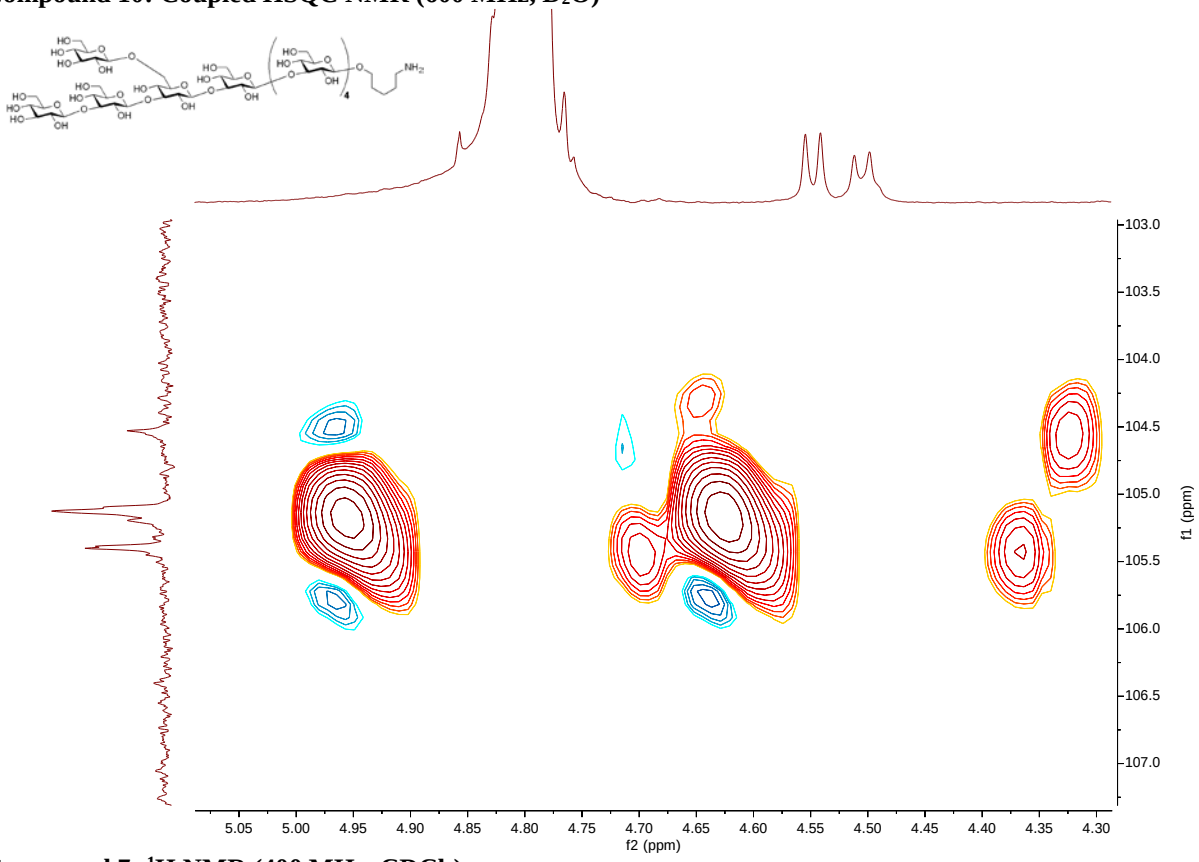
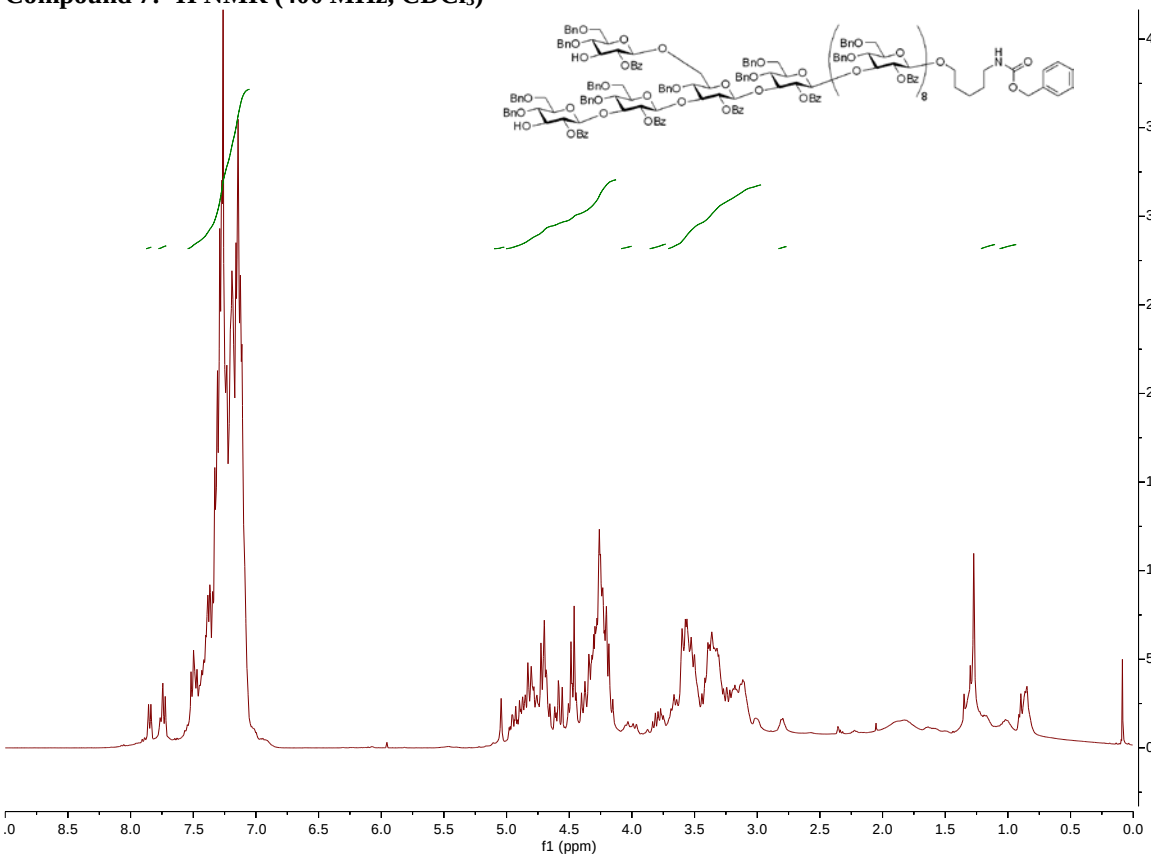


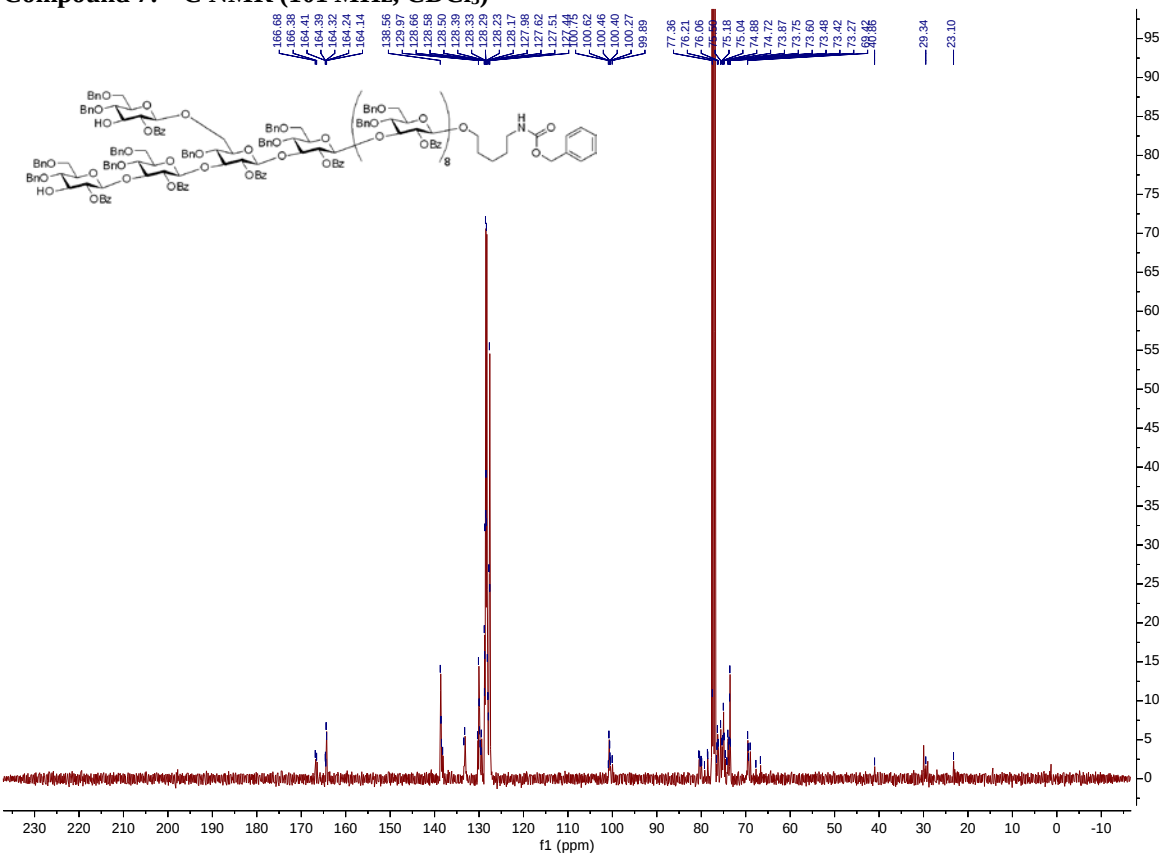
Compound 10: Coupled HSQC NMR (600 MHz, D_2O)



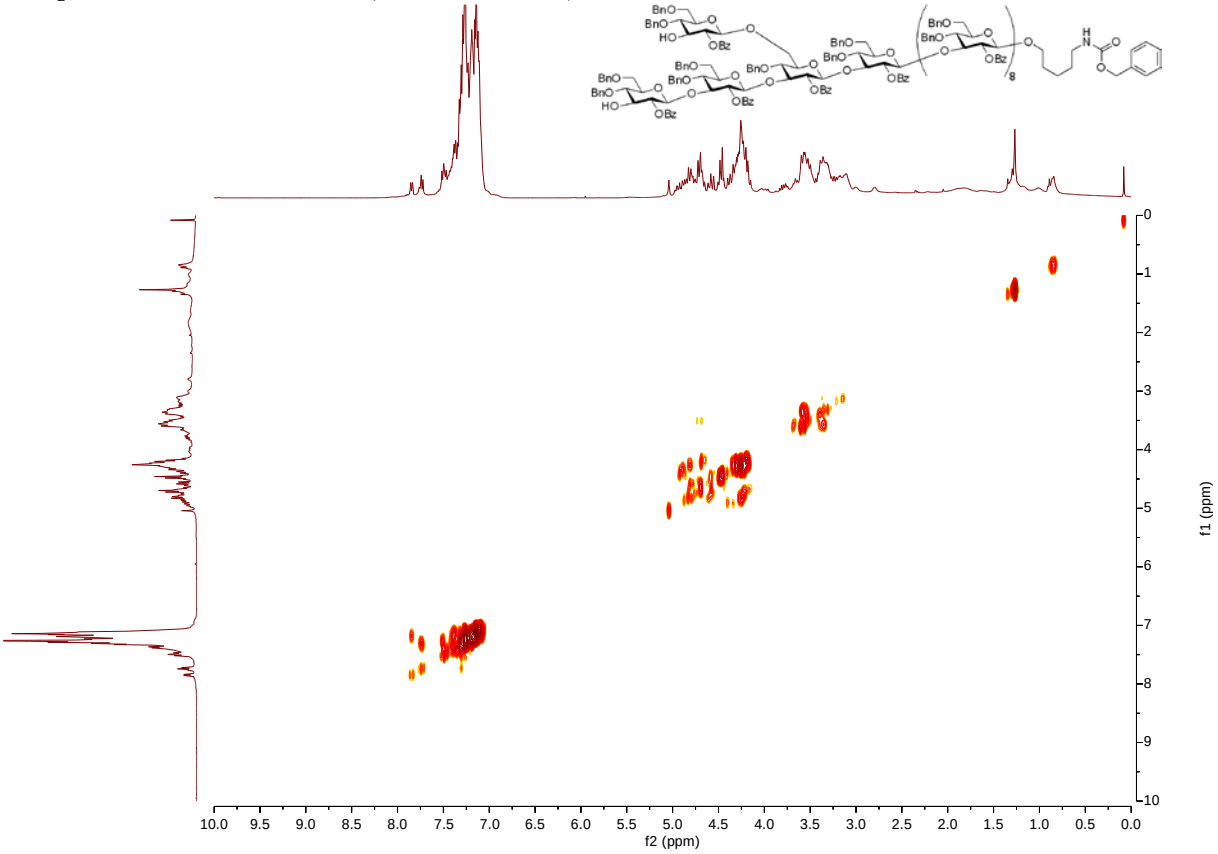
Compound 7: ^1H NMR (400 MHz, $CDCl_3$)



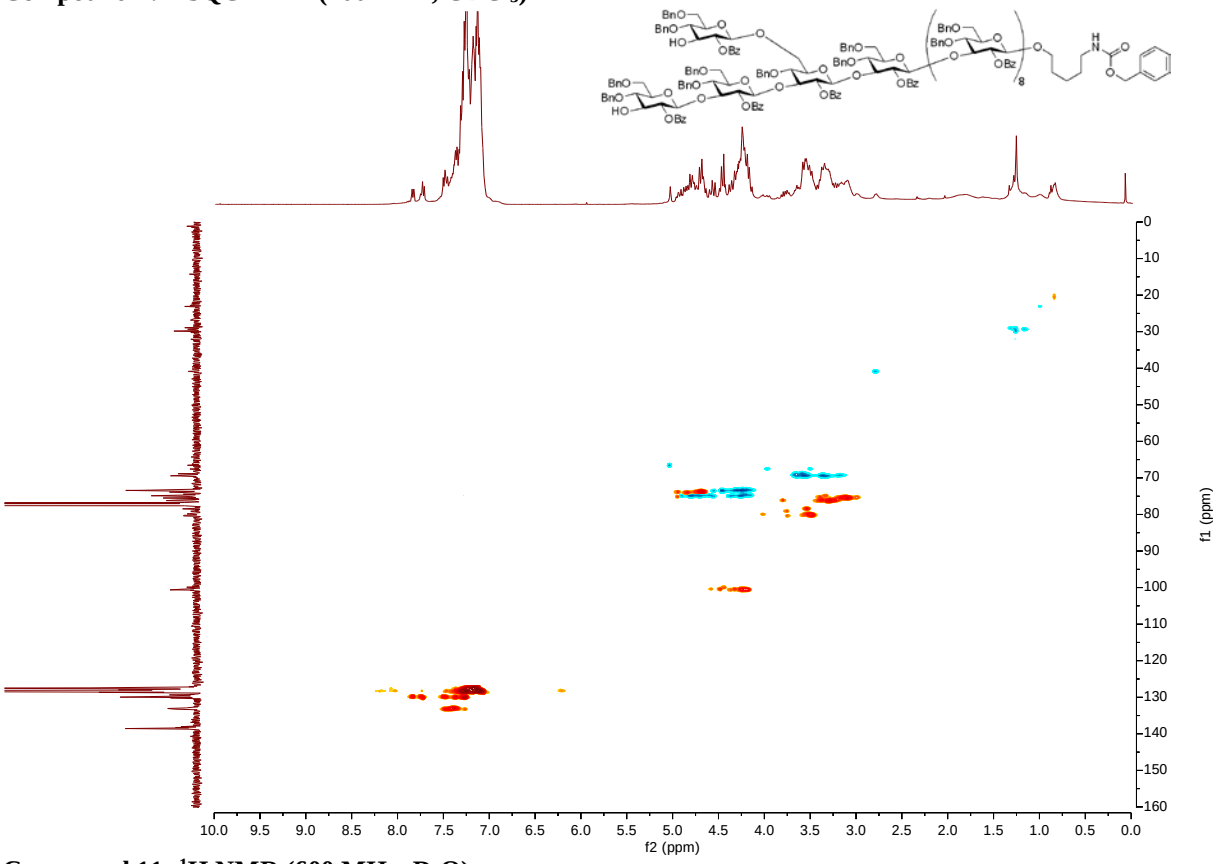
Compound 7: ^{13}C NMR (101 MHz, $CDCl_3$)



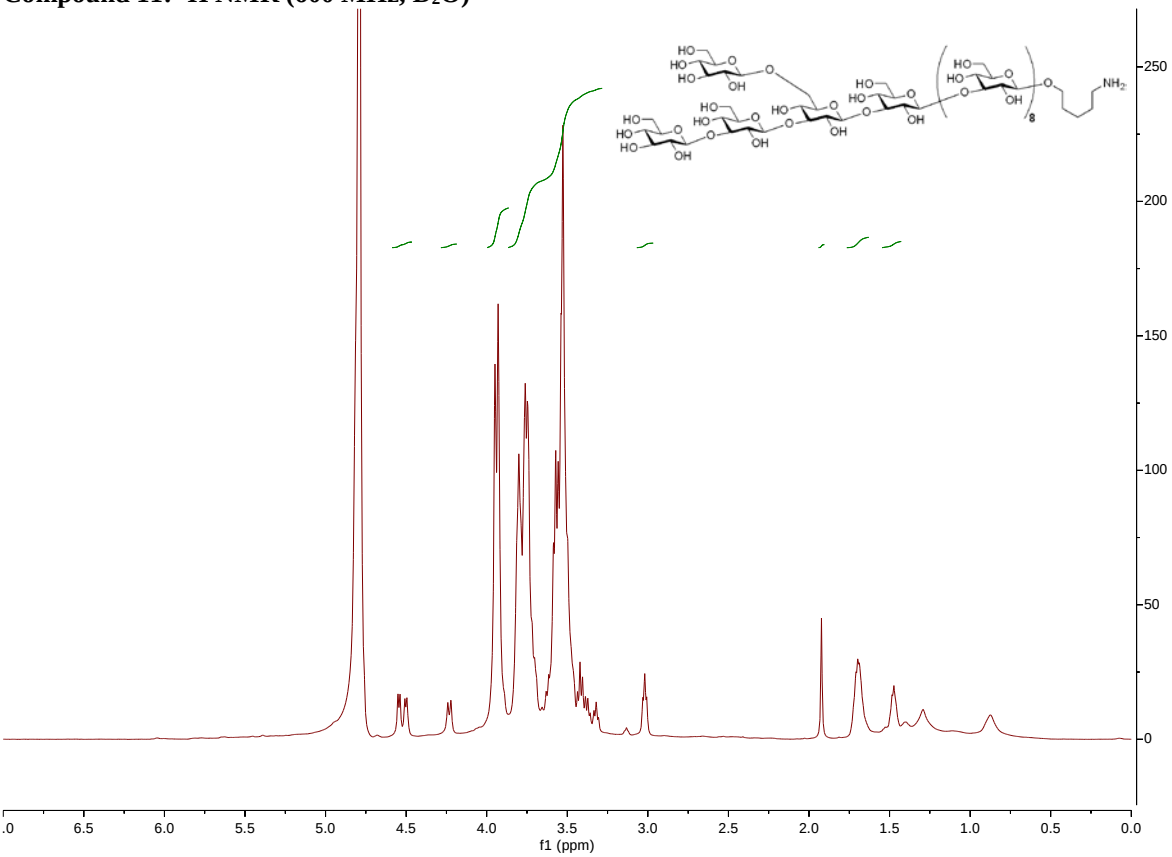
Compound 7: HH-COSY NMR (400 MHz, $CDCl_3$)



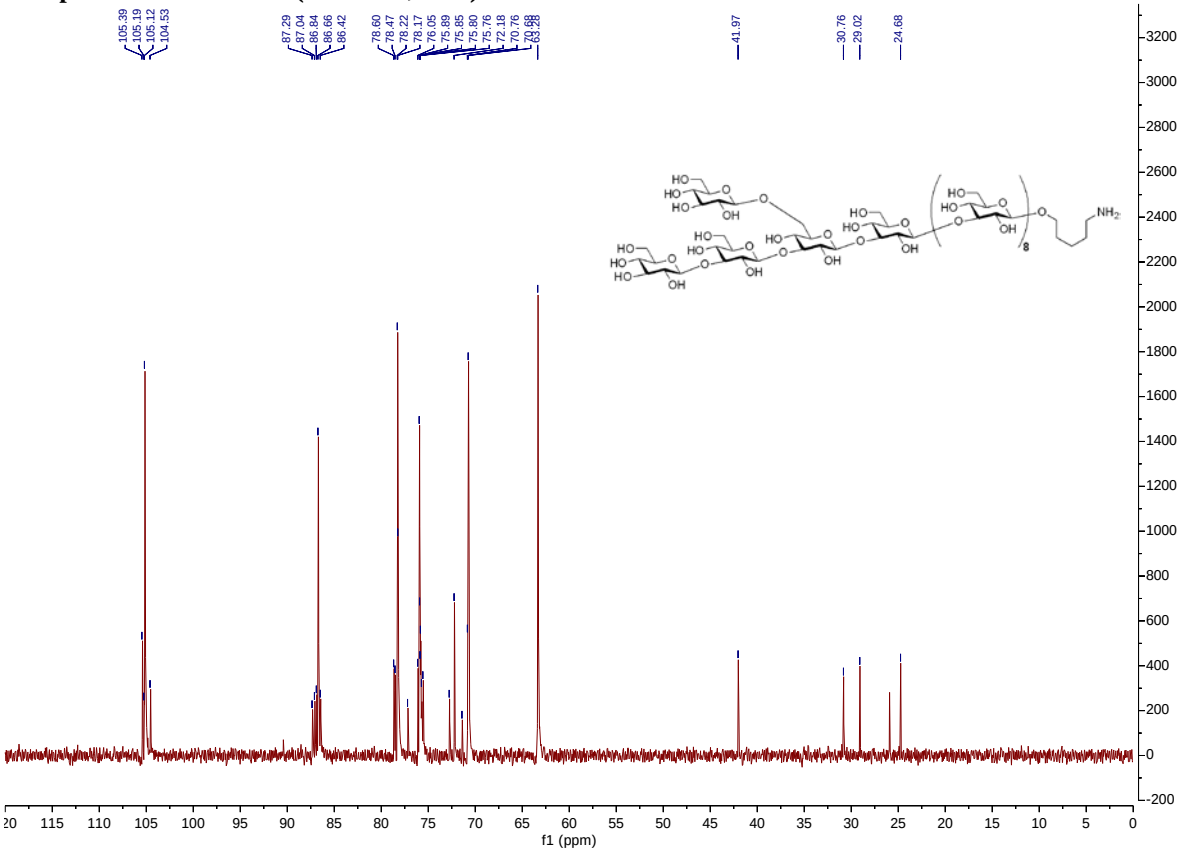
Compound 7: HSQC NMR (400 MHz, $CDCl_3$)



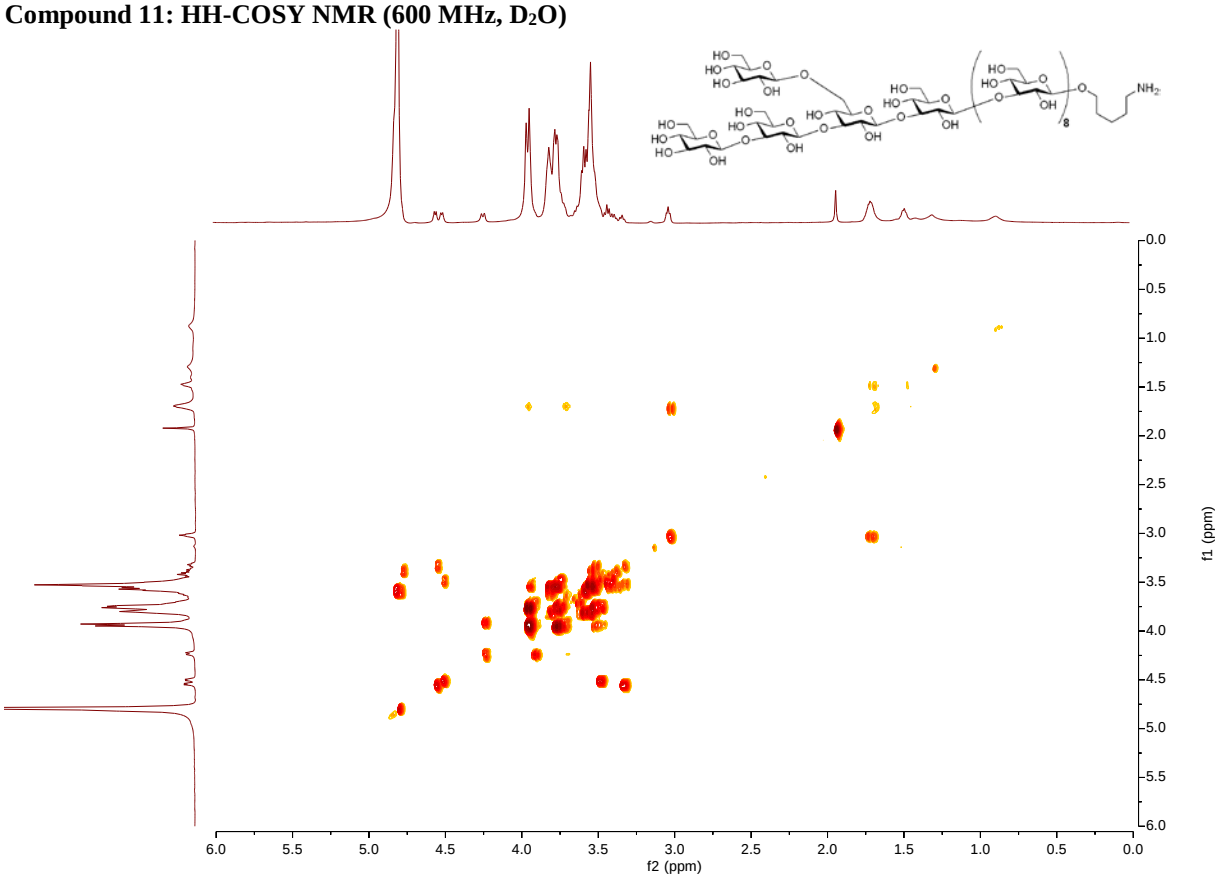
Compound 11: ^1H NMR (600 MHz, D_2O)



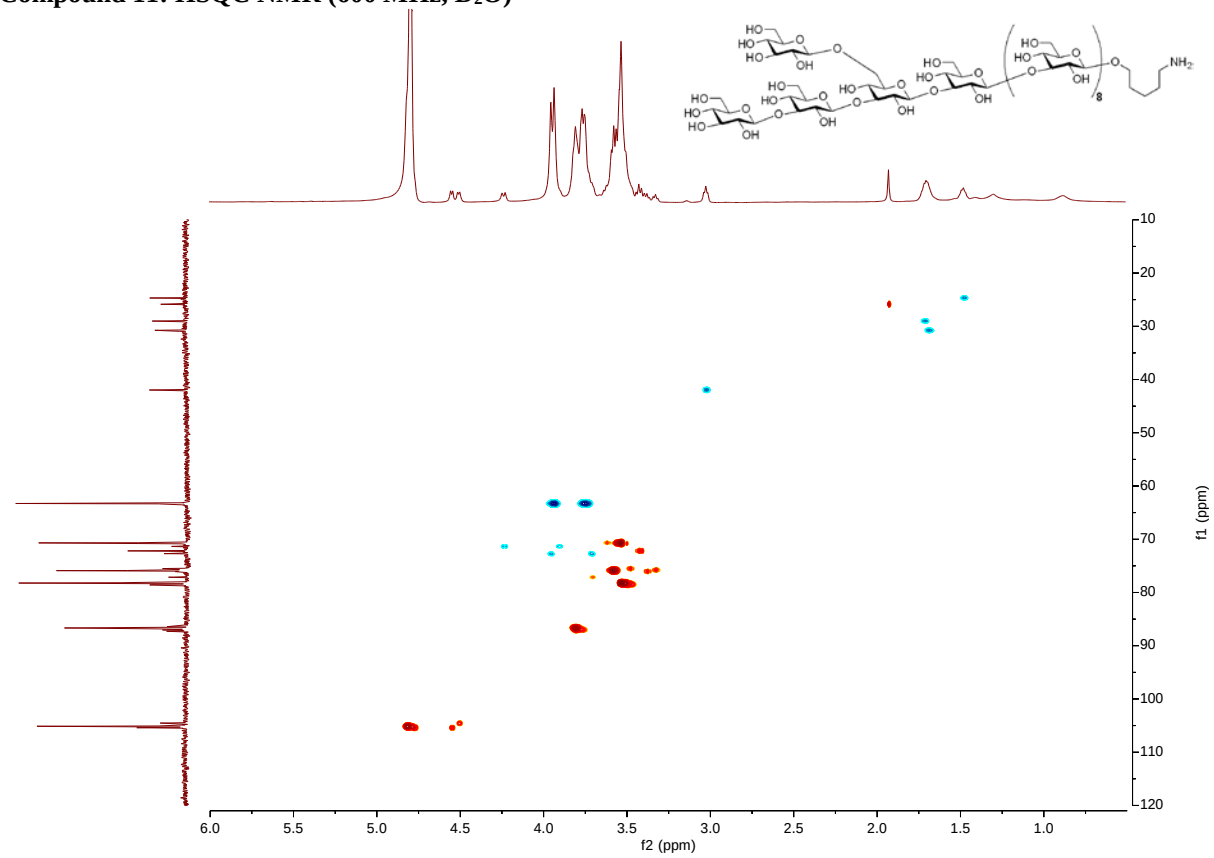
Compound 11: ^{13}C NMR (151 MHz, D_2O)



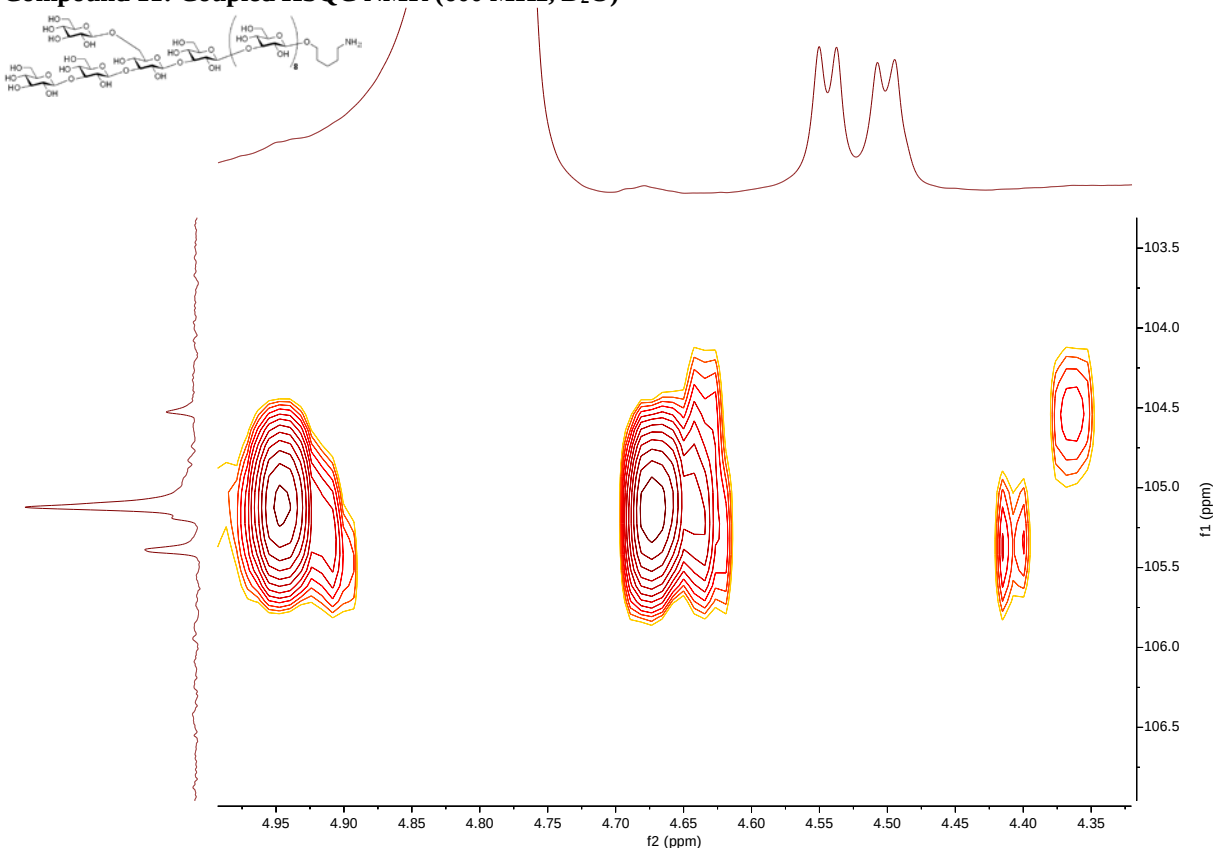
Compound 11: HH-COSY NMR (600 MHz, D_2O)



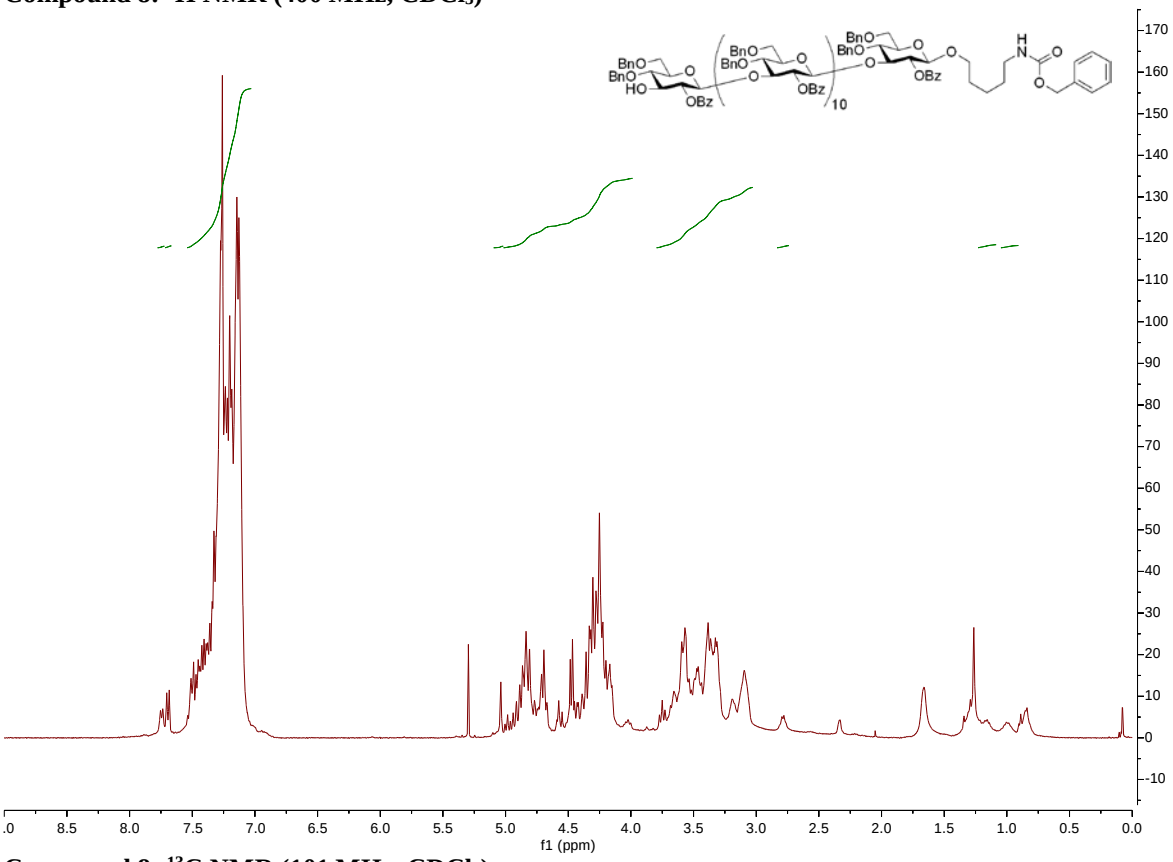
Compound 11: HSQC NMR (600 MHz, D_2O)



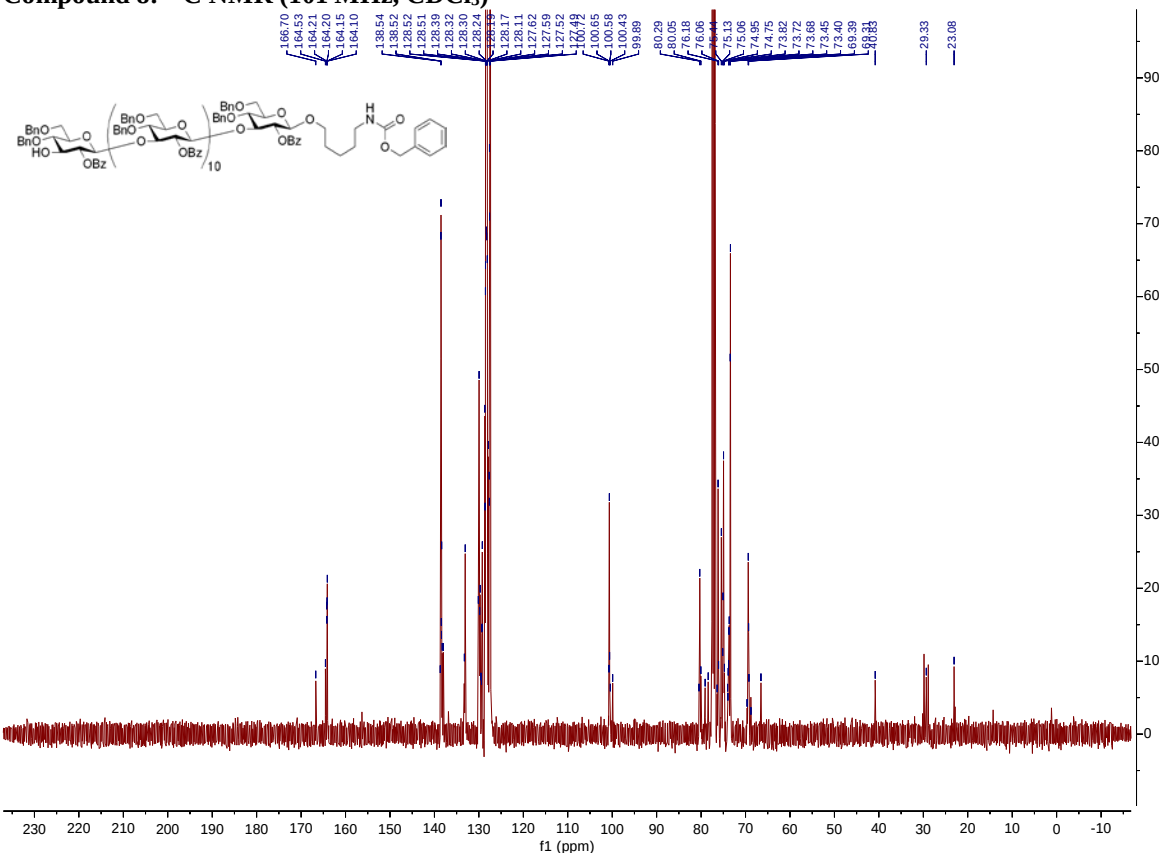
Compound 11: Coupled HSQC NMR (600 MHz, D_2O)



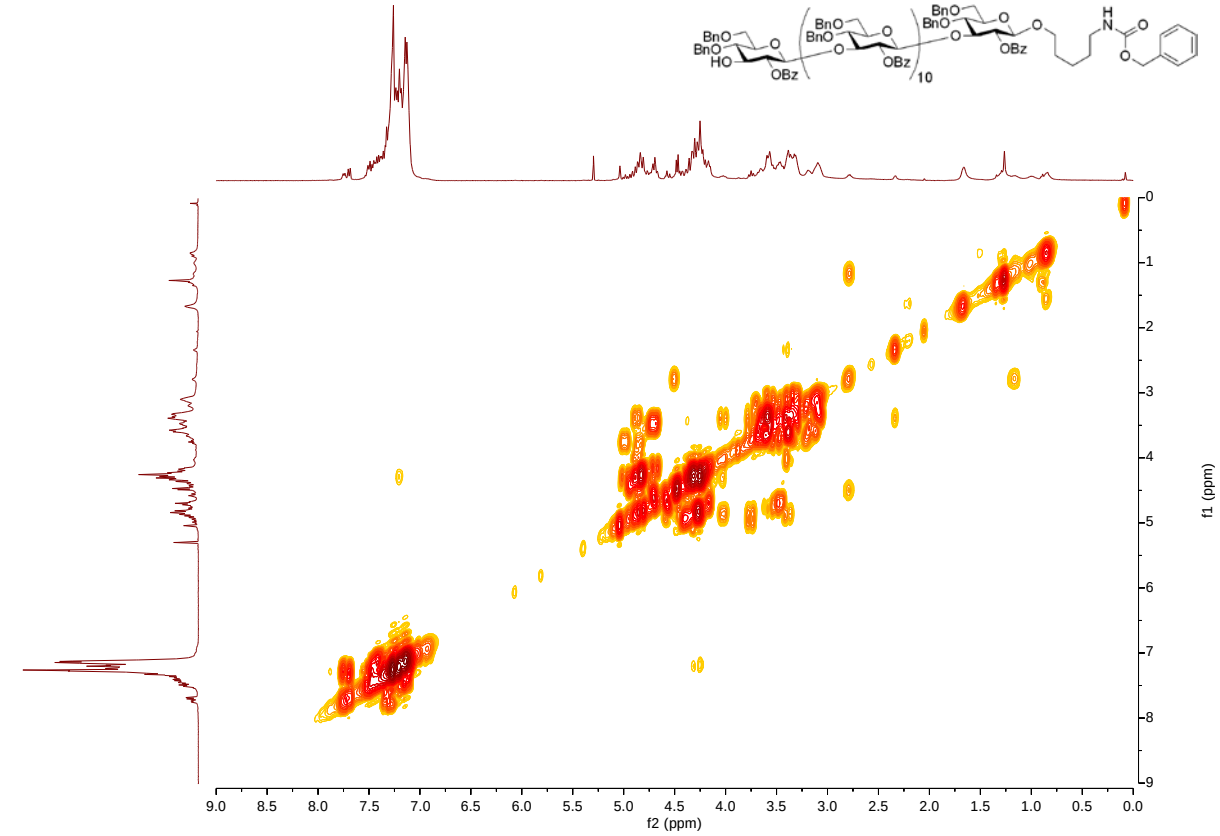
Compound 8: ^{1}H NMR (400 MHz, $CDCl_3$)



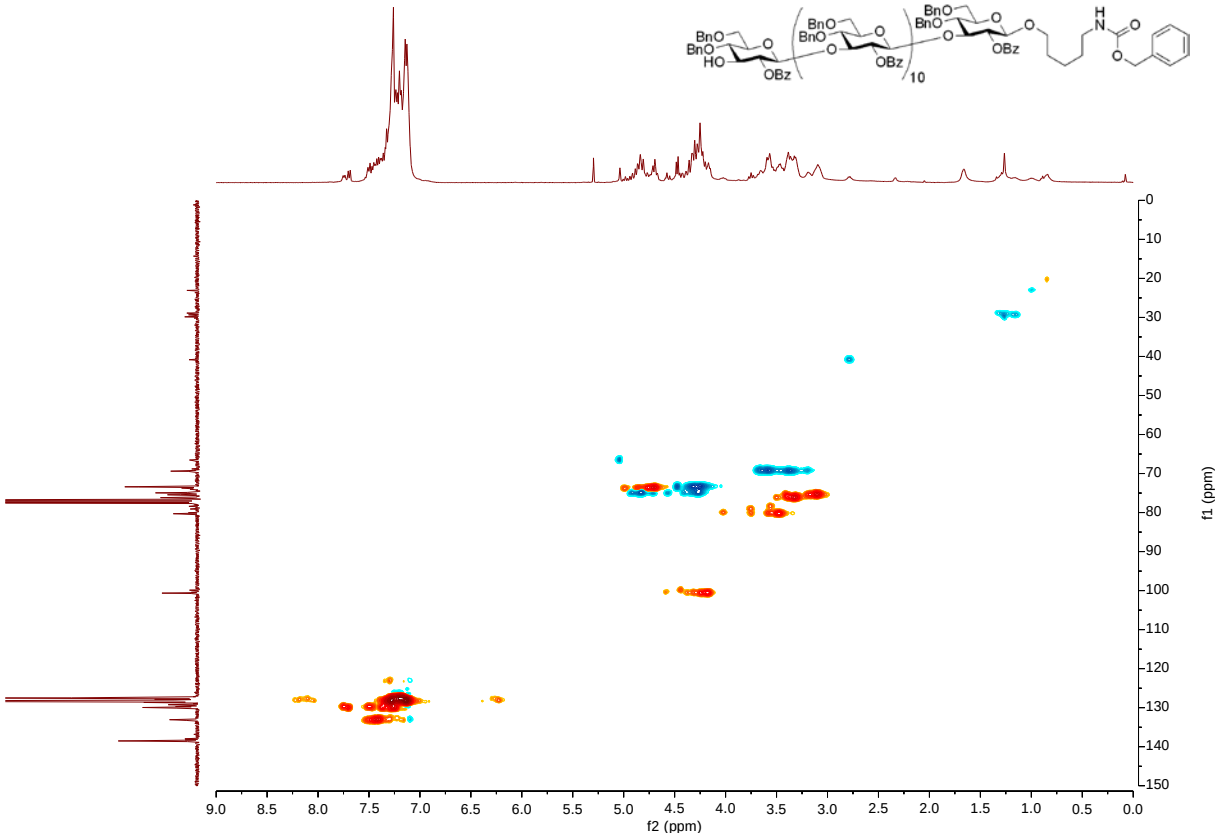
Compound 8: ^{13}C NMR (101 MHz, $CDCl_3$)



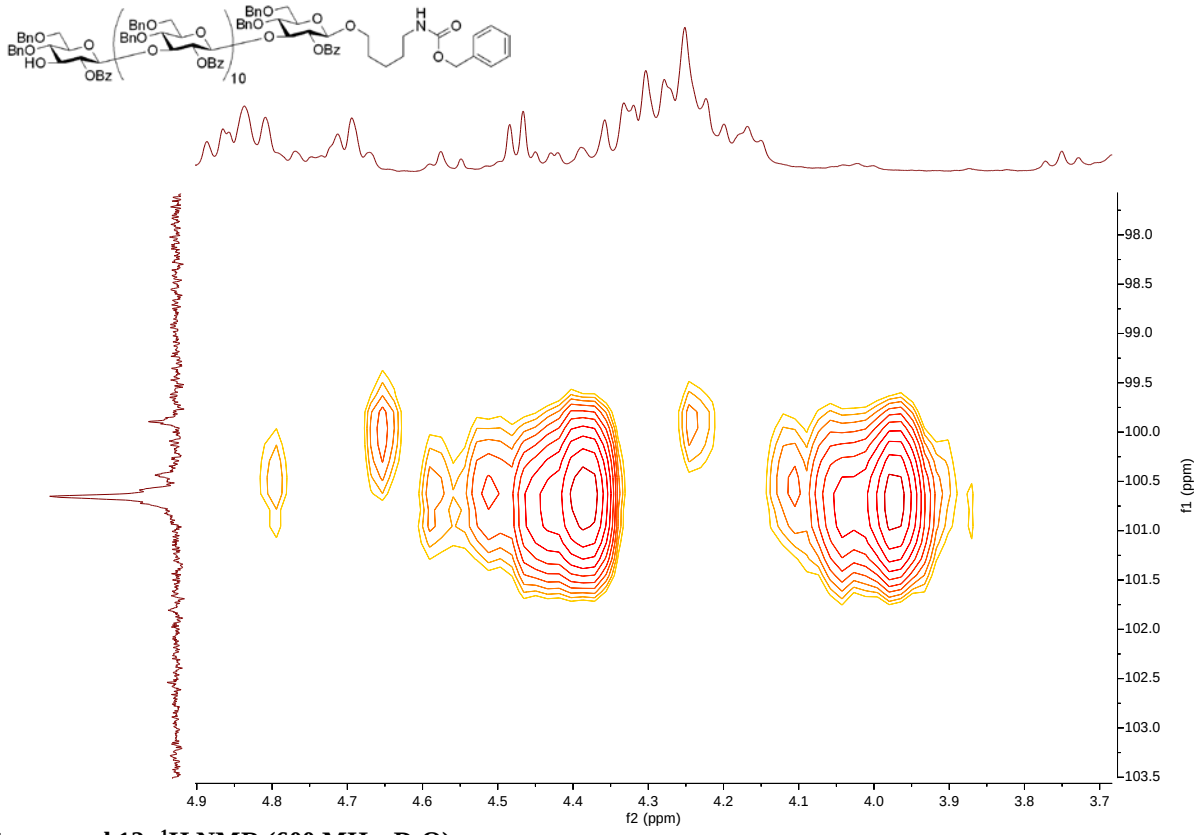
Compound 8: HH-COSY NMR (400 MHz, $CDCl_3$)



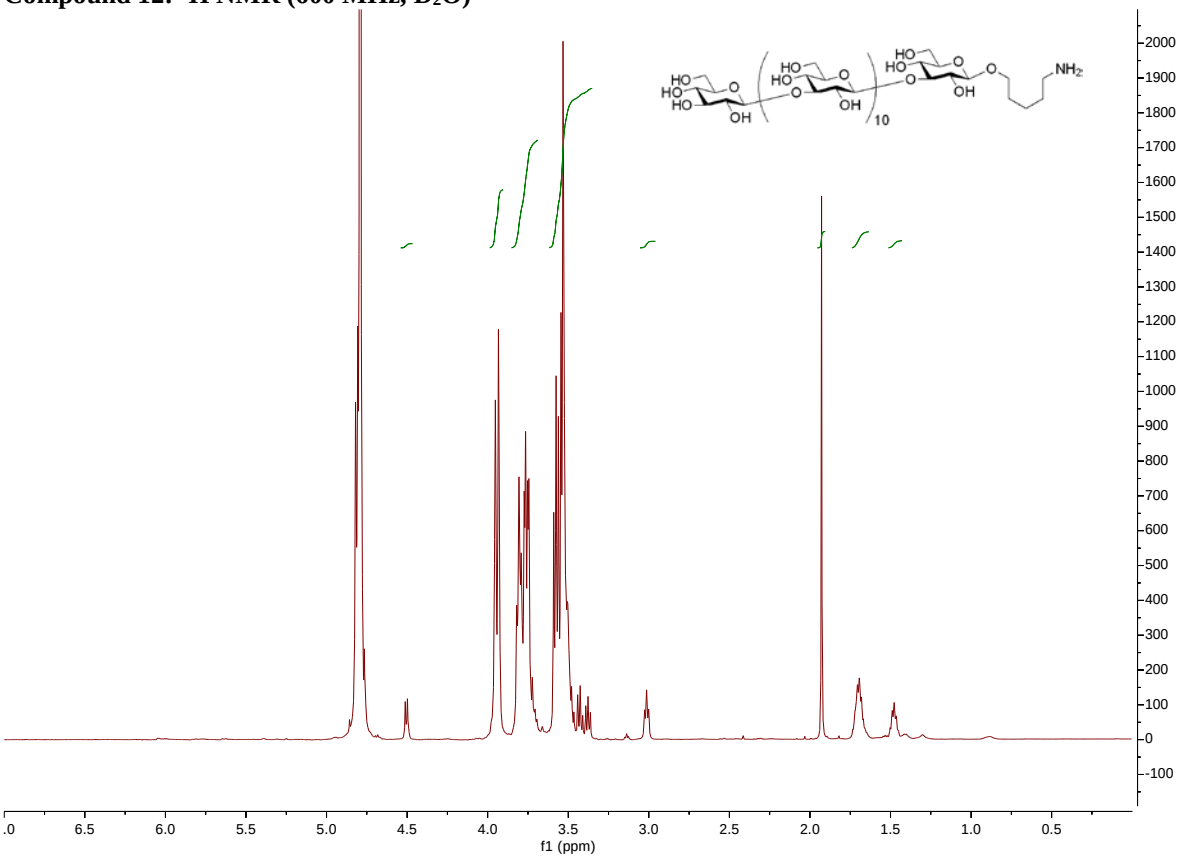
Compound 8: HSQC NMR (400 MHz, $CDCl_3$)



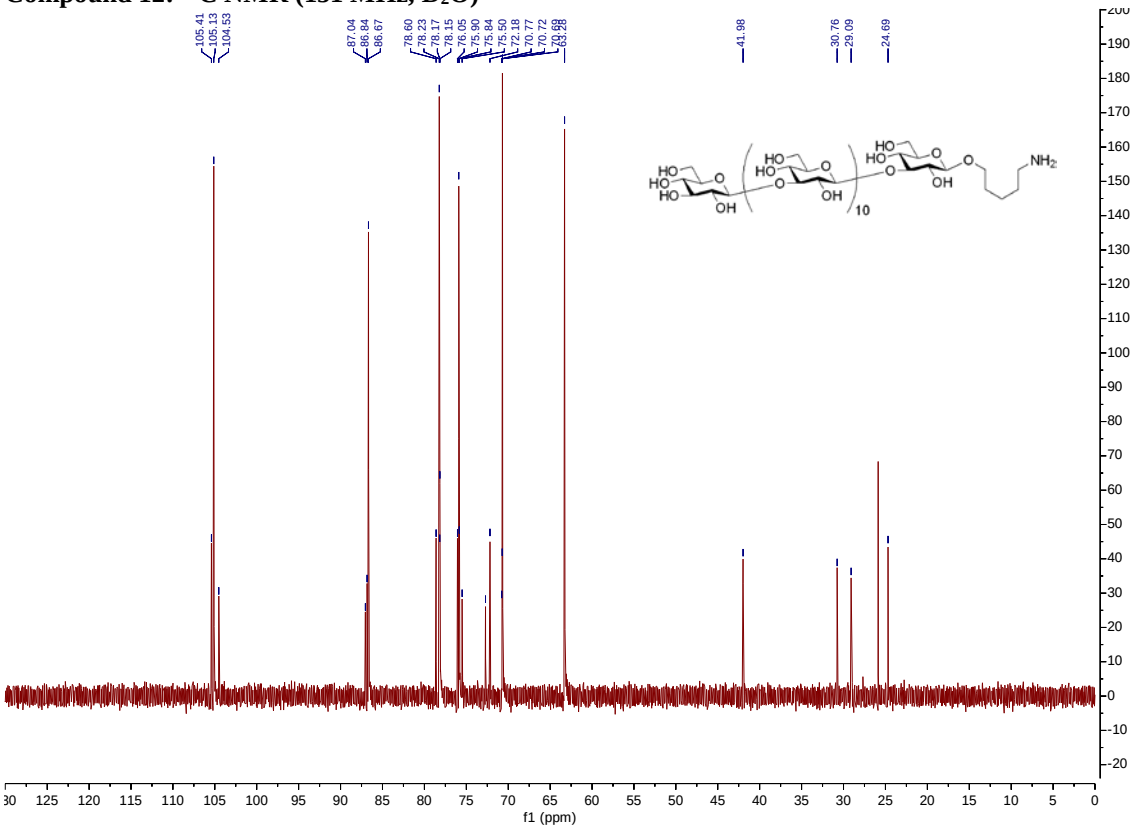
Compound 8: Coupled HSQC NMR (400 MHz, $CDCl_3$)



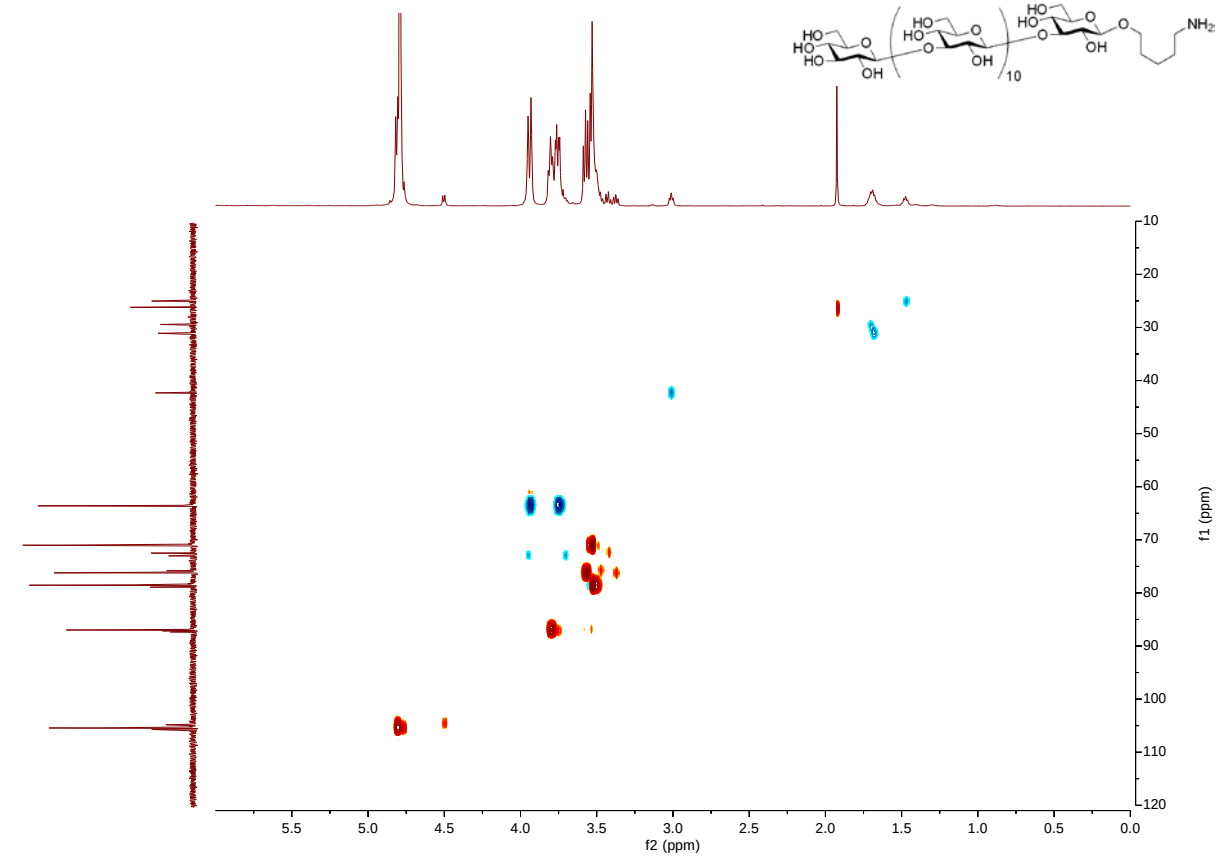
Compound 12: ^1H NMR (600 MHz, D_2O)



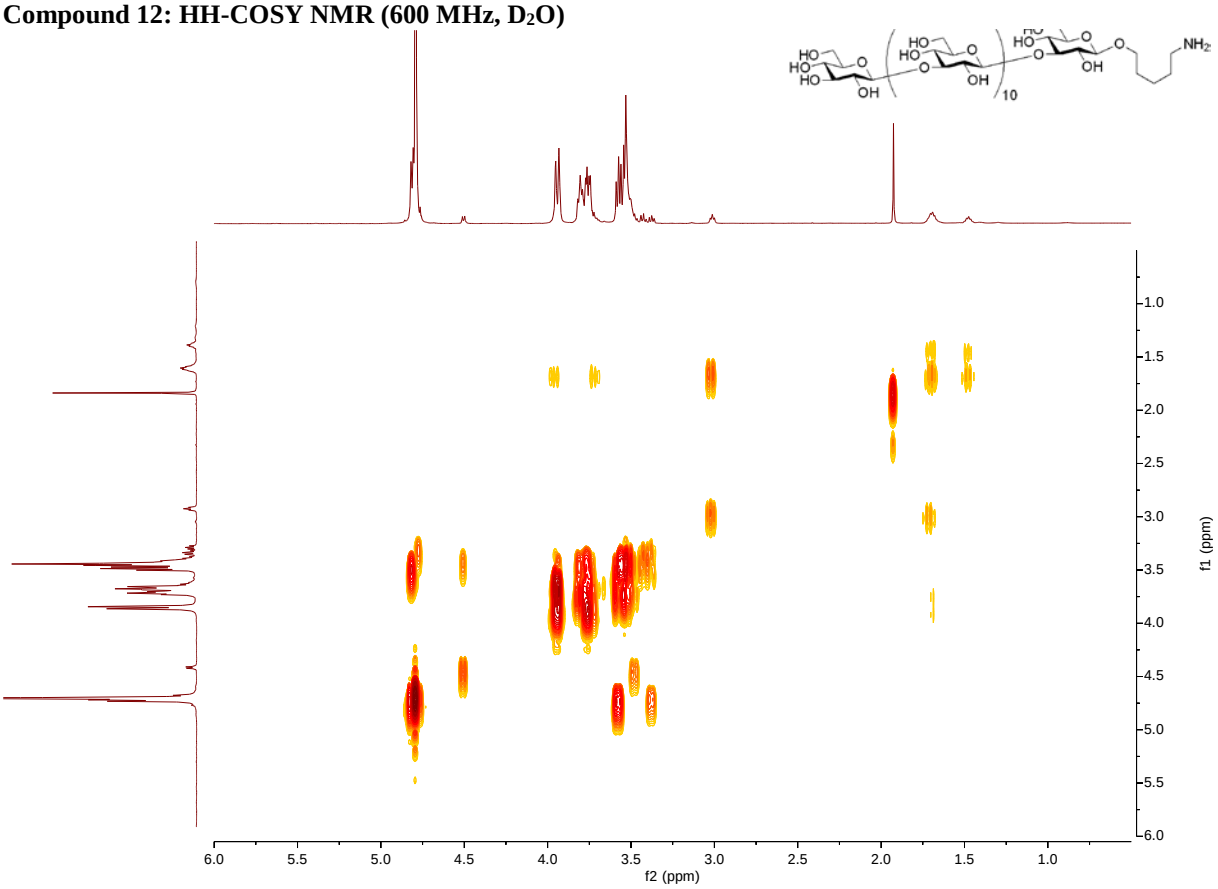
Compound 12: ^{13}C NMR (151 MHz, D_2O)



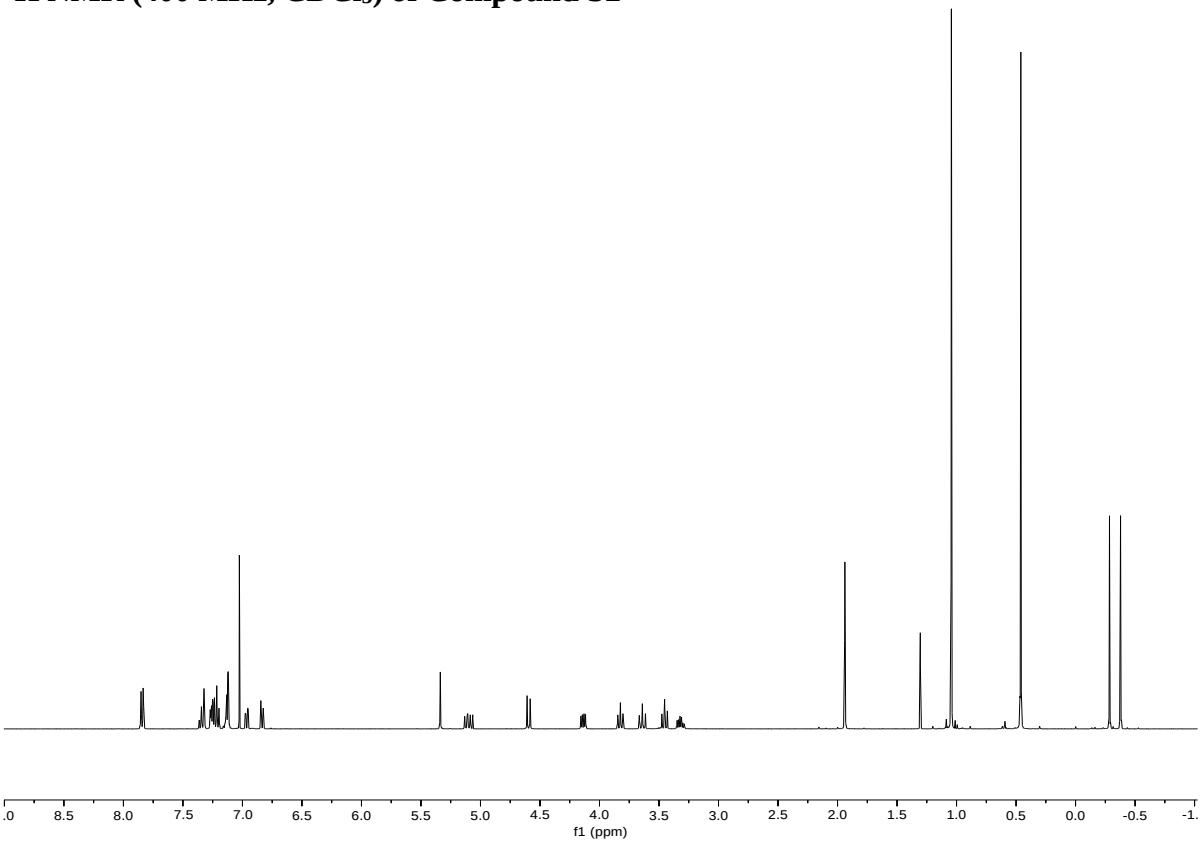
Compound 12: HSQC NMR (600 MHz, D_2O)



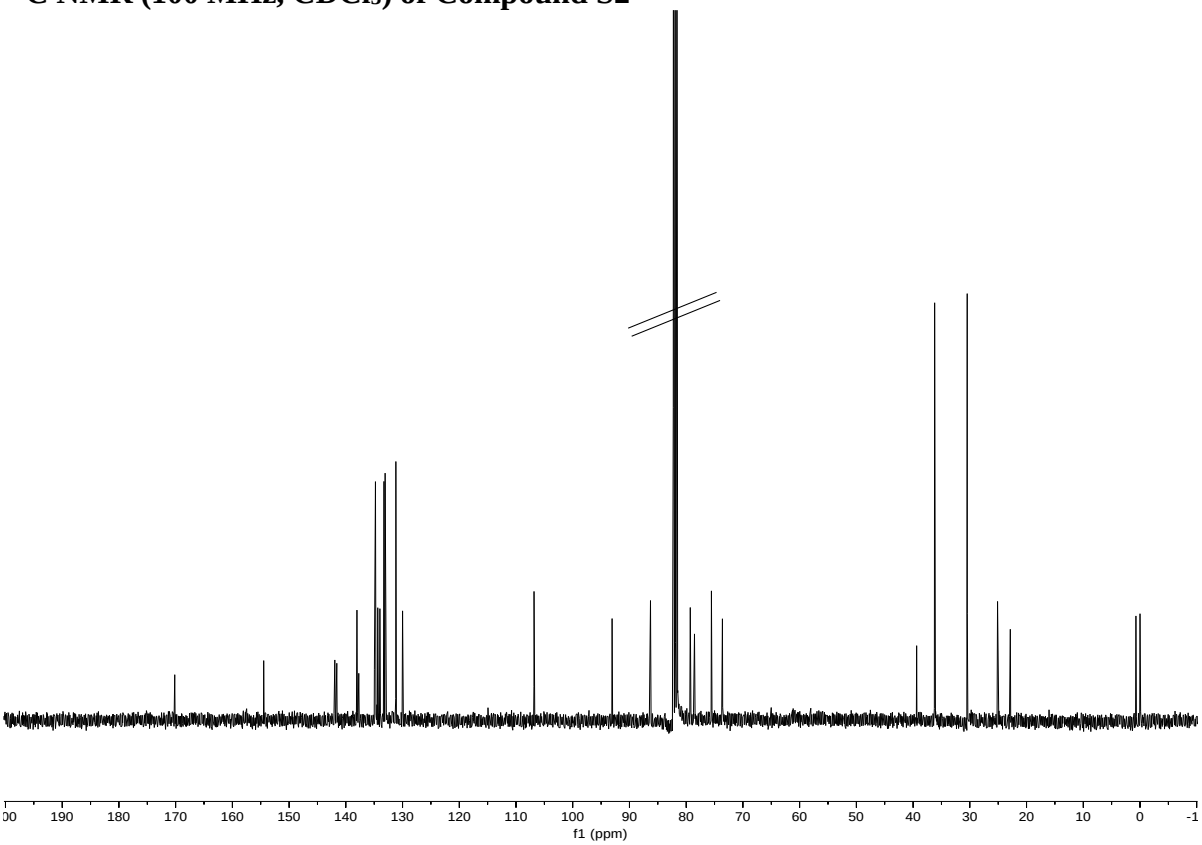
Compound 12: HH-COSY NMR (600 MHz, D_2O)



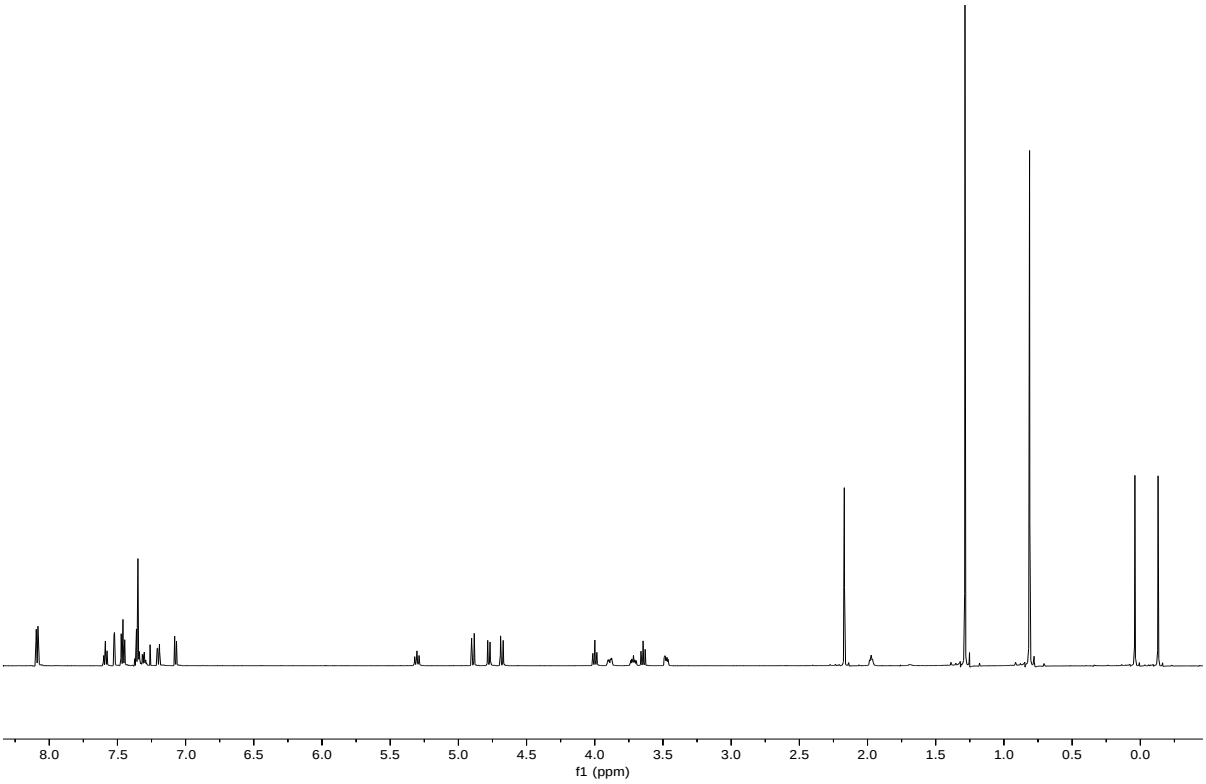
^{1}H NMR (400 MHz, $CDCl_3$) of Compound S2



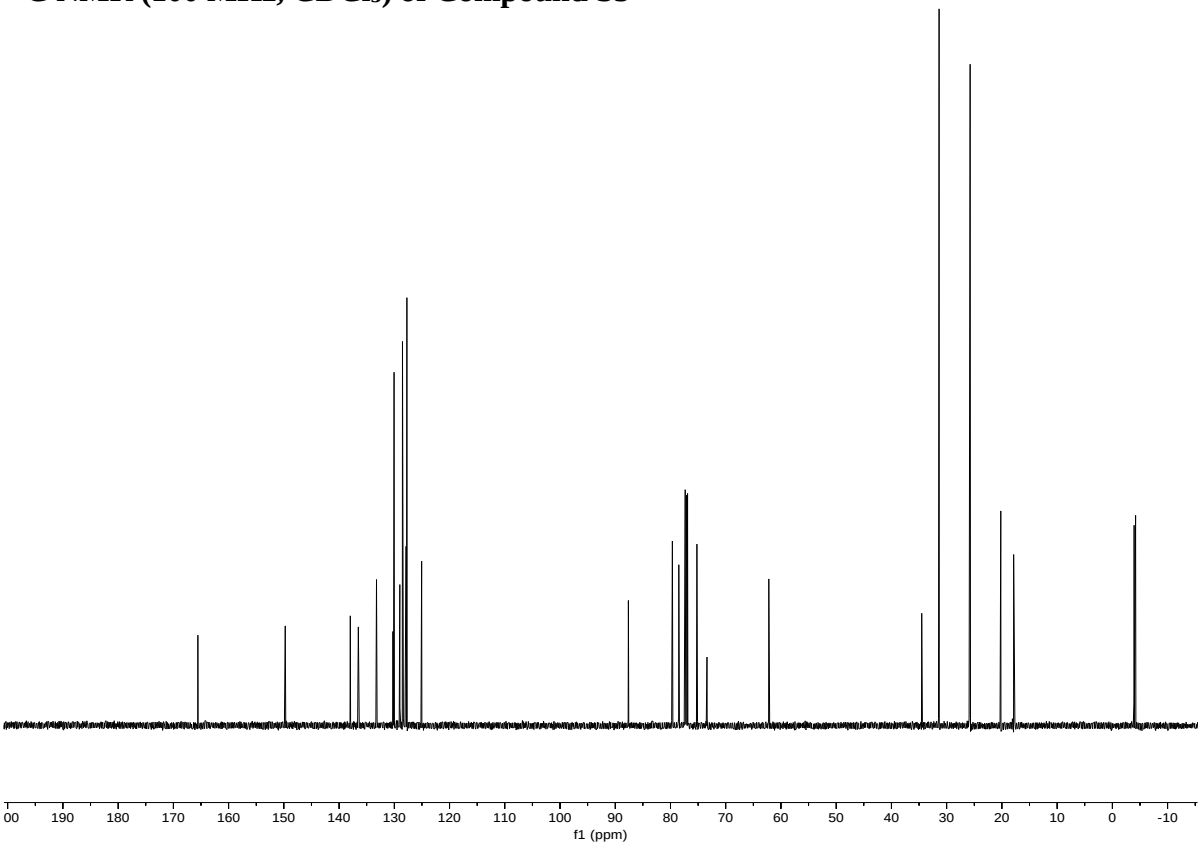
^{13}C NMR (100 MHz, $CDCl_3$) of Compound S2



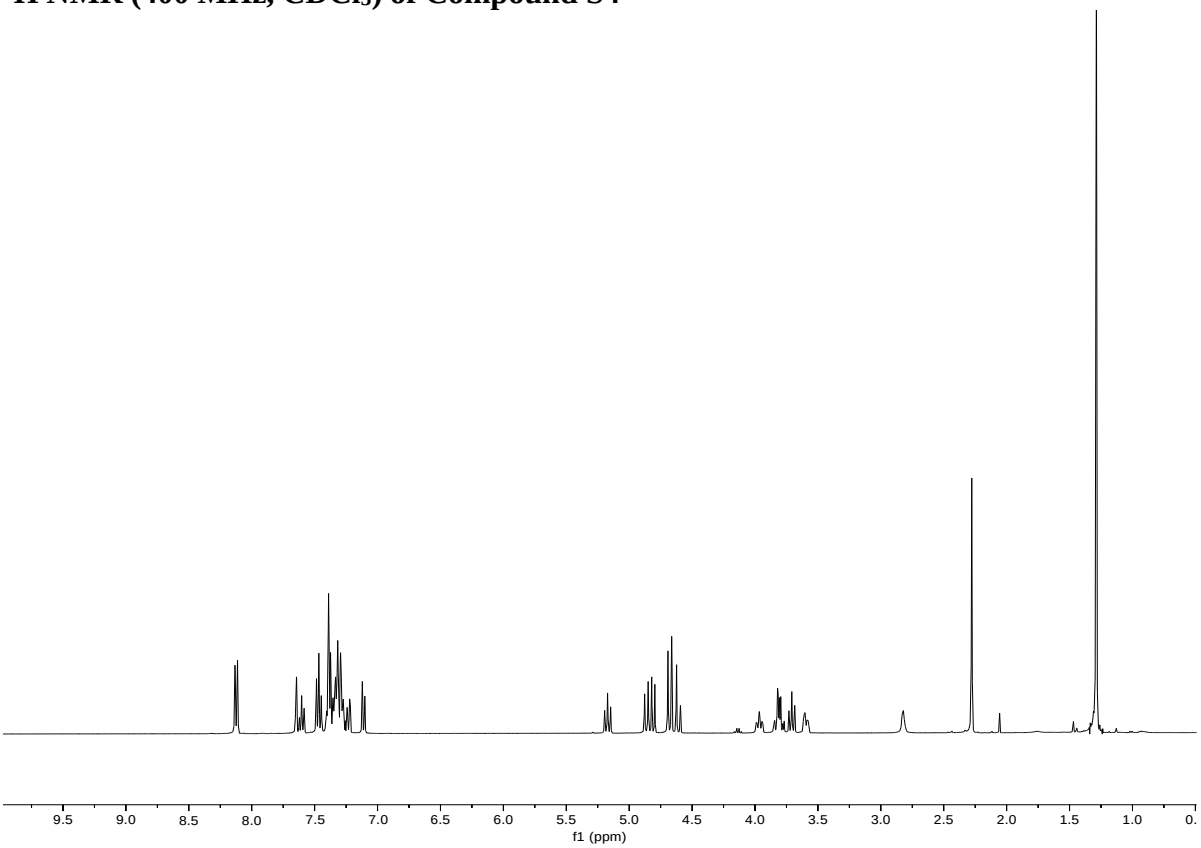
^{1}H NMR (400 MHz, $CDCl_3$) of Compound S3



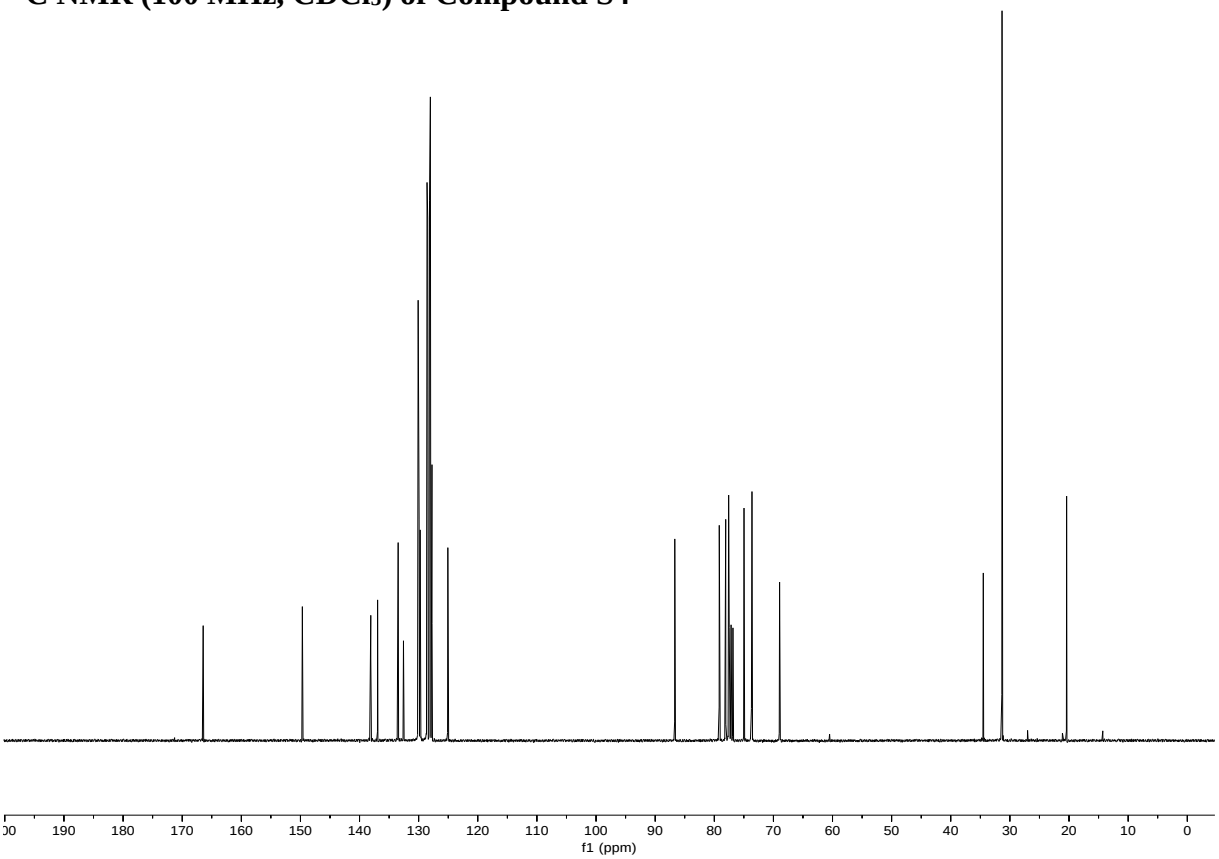
^{13}C NMR (100 MHz, $CDCl_3$) of Compound S3



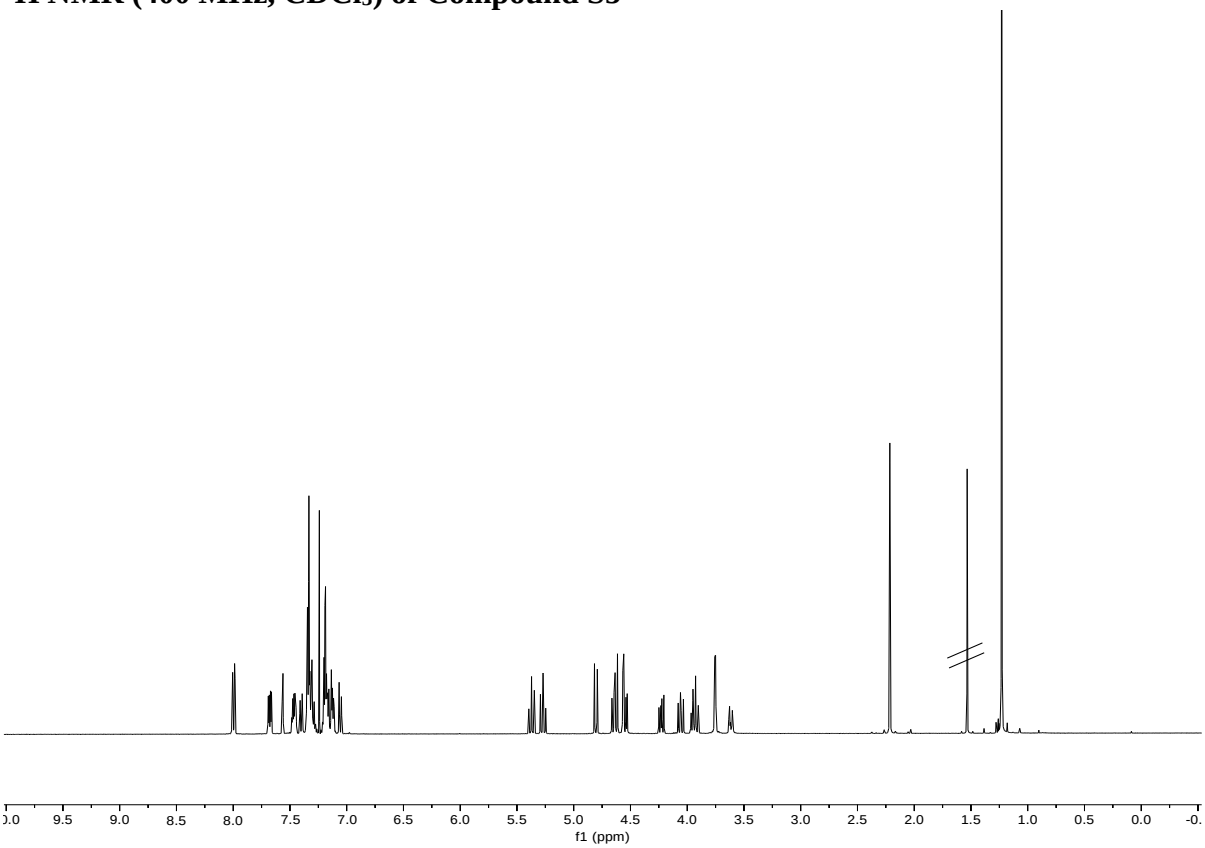
^{1}H NMR (400 MHz, $CDCl_3$) of Compound S4



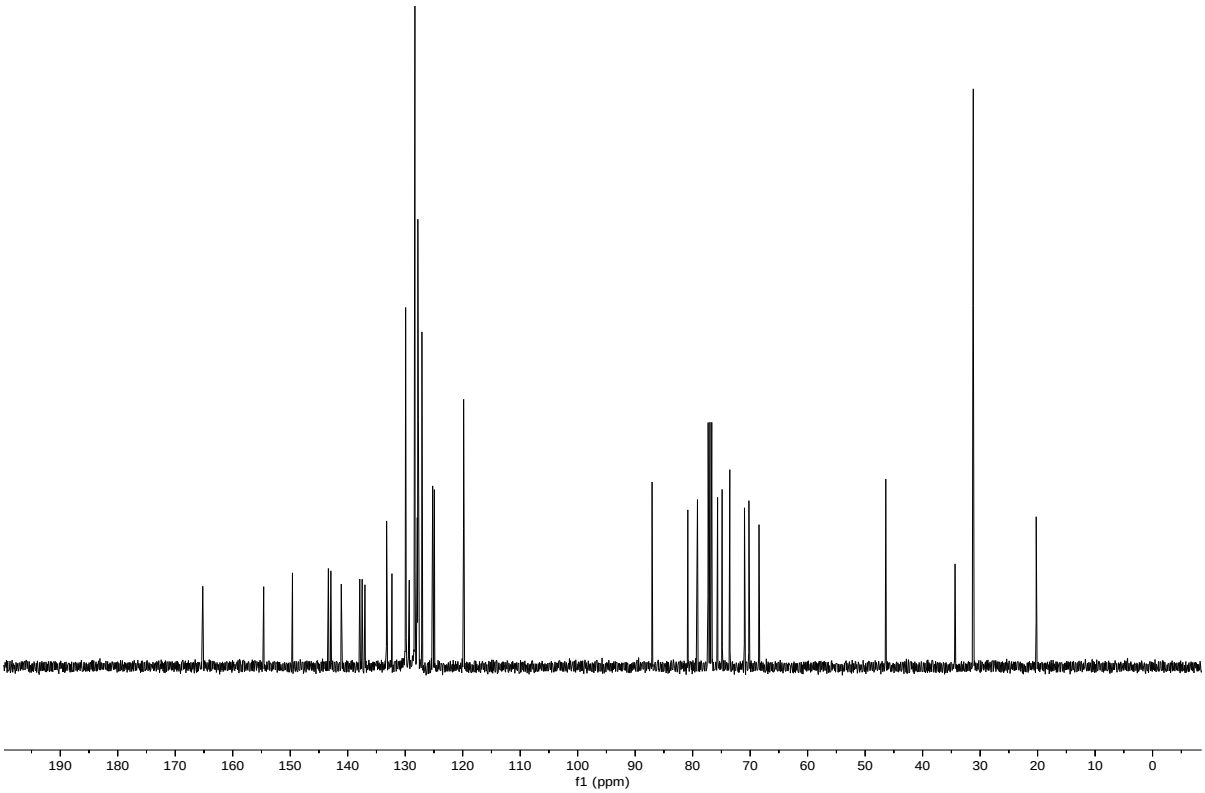
^{13}C NMR (100 MHz, $CDCl_3$) of Compound S4



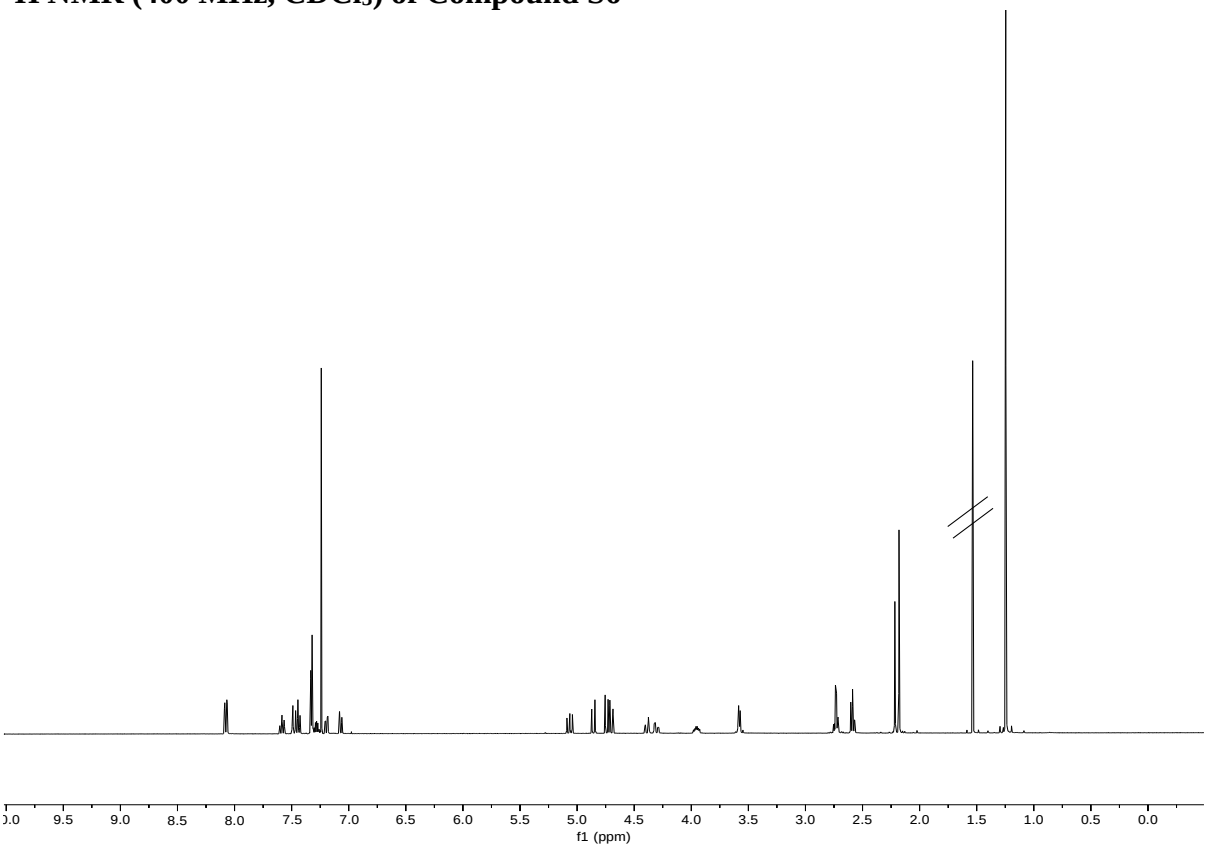
^{1}H NMR (400 MHz, $CDCl_3$) of Compound S5



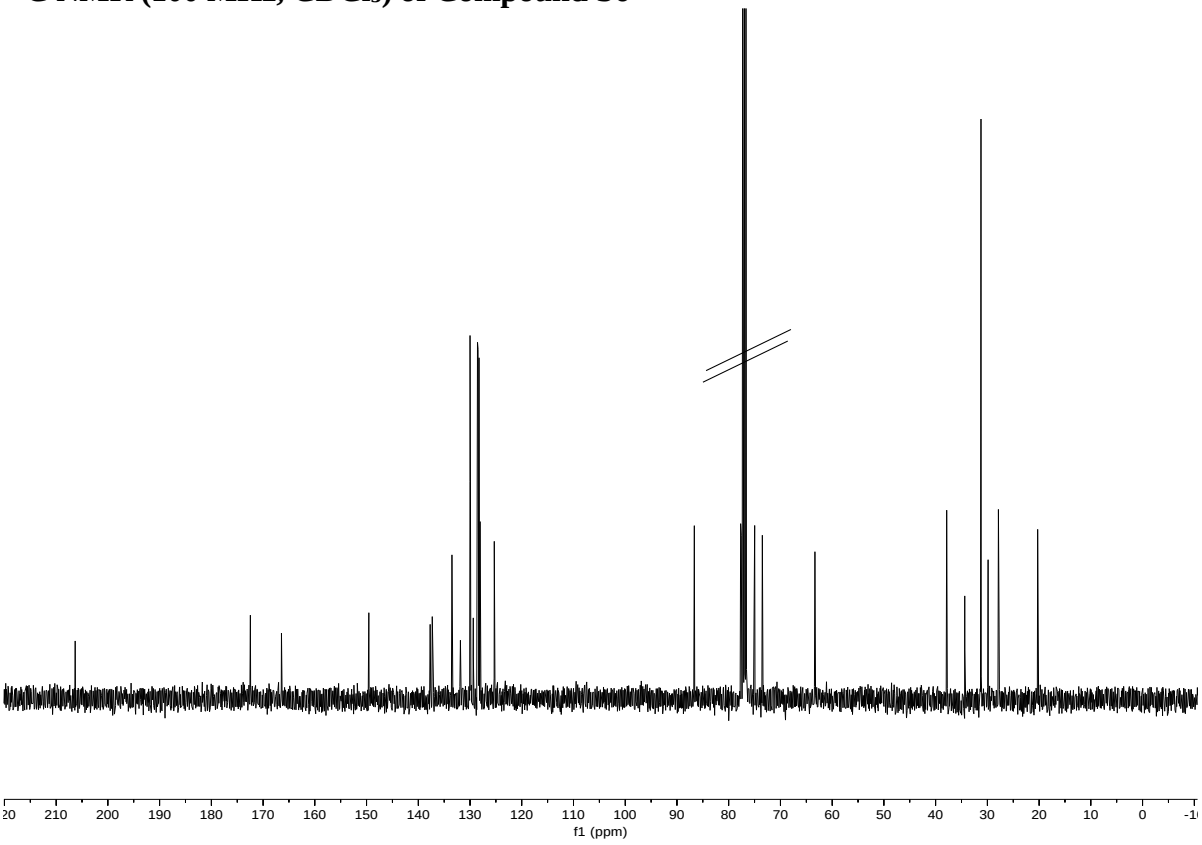
^{13}C NMR (100 MHz, $CDCl_3$) of Compound S5



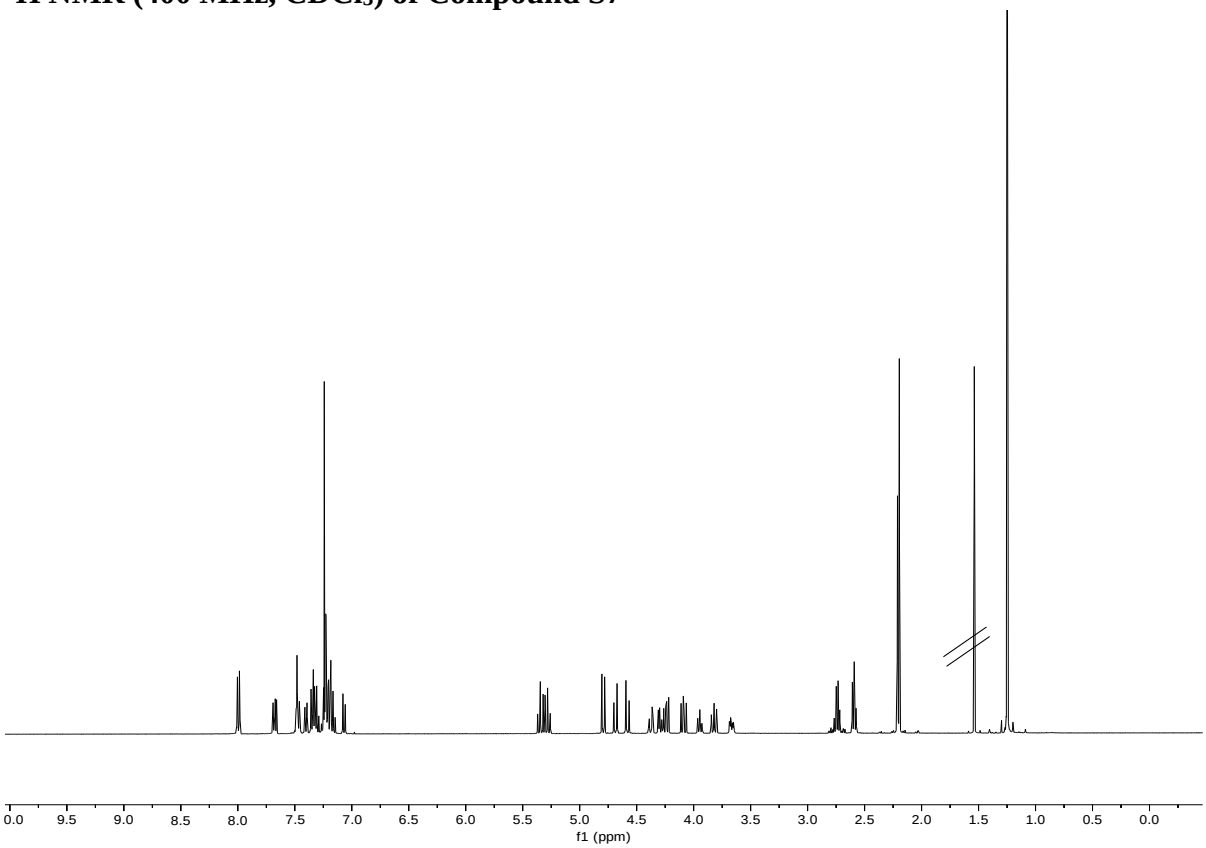
^{1}H NMR (400 MHz, $CDCl_3$) of Compound S6



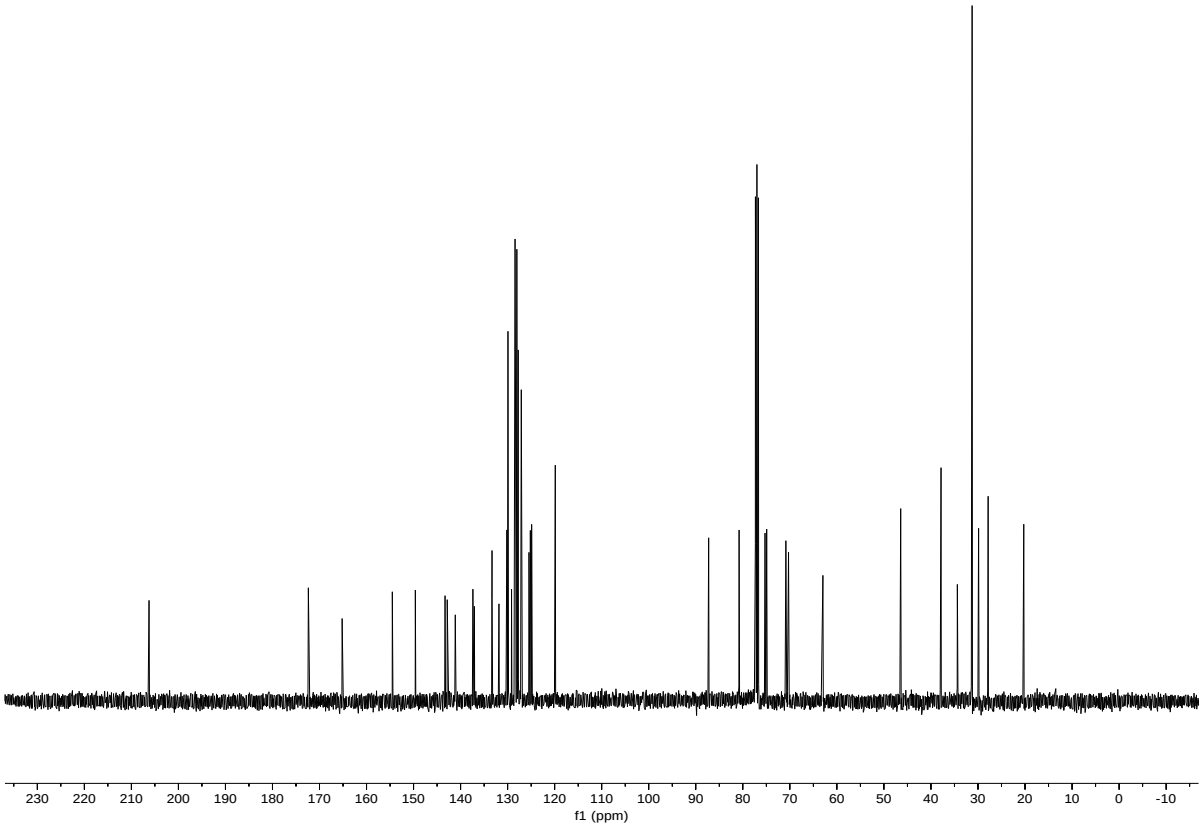
^{13}C NMR (100 MHz, $CDCl_3$) of Compound S6



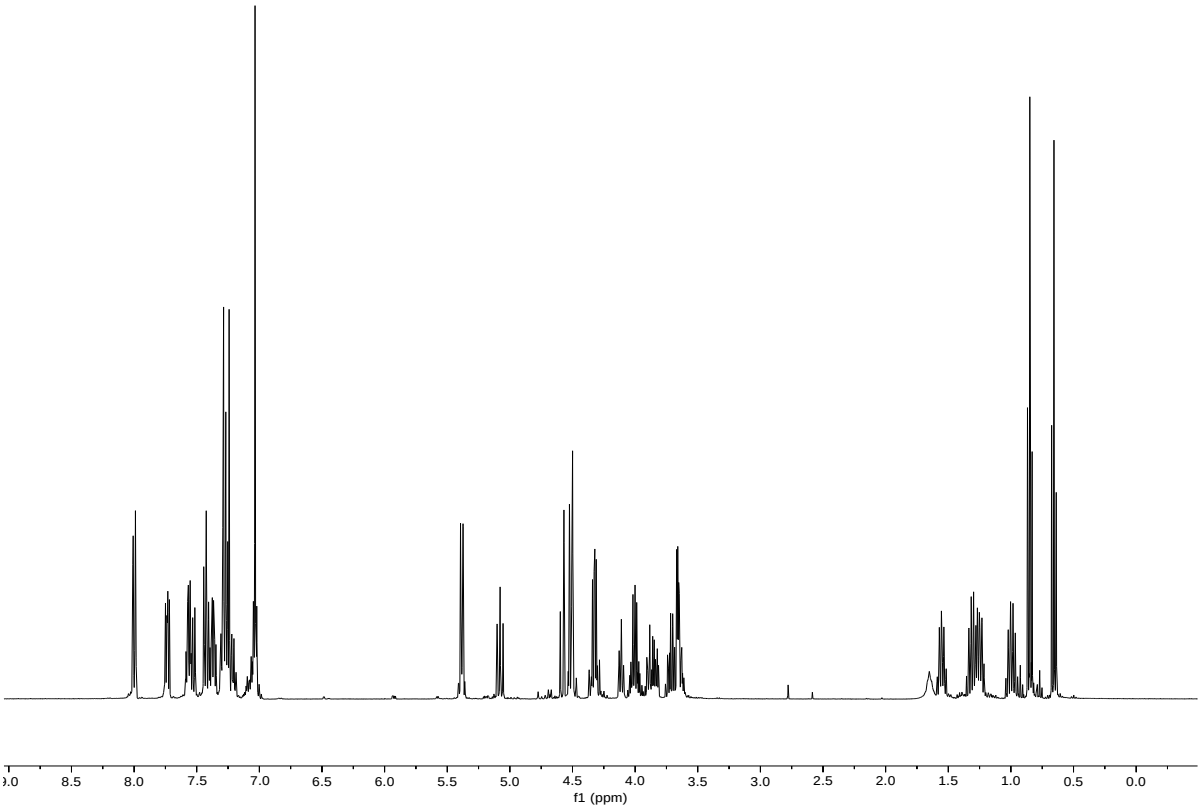
^{1}H NMR (400 MHz, $CDCl_3$) of Compound S7



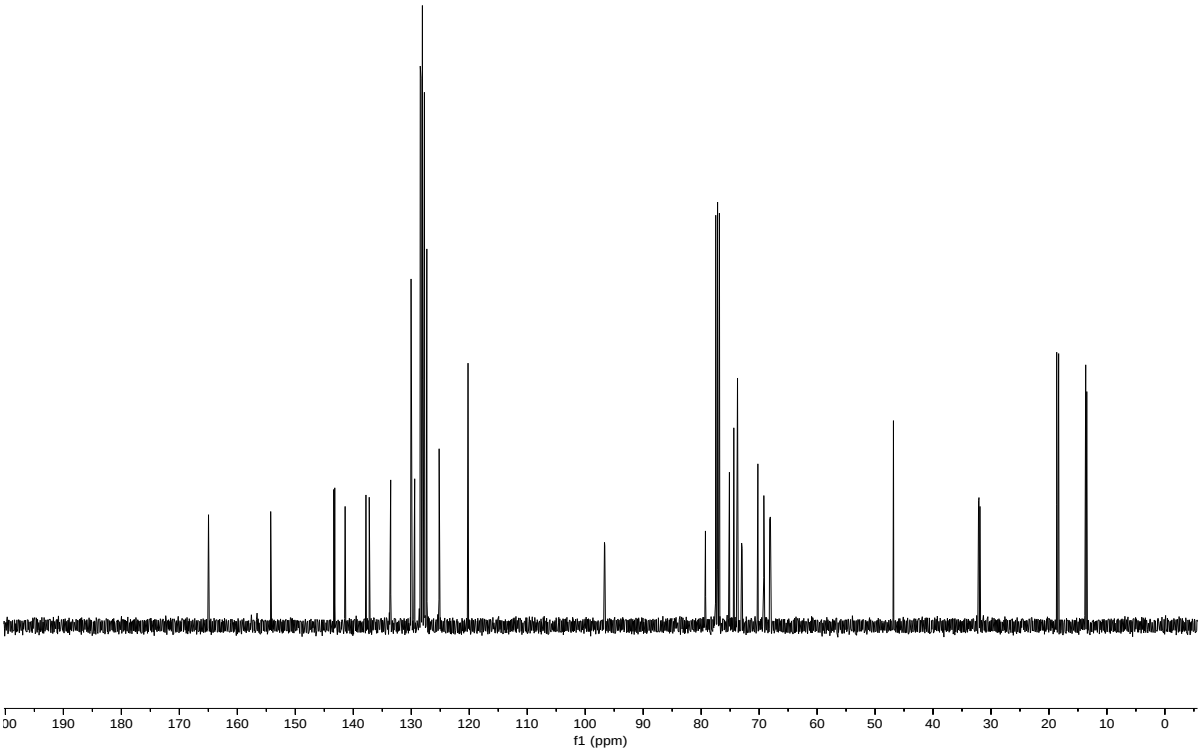
^{13}C NMR (100 MHz, $CDCl_3$) of Compound S7



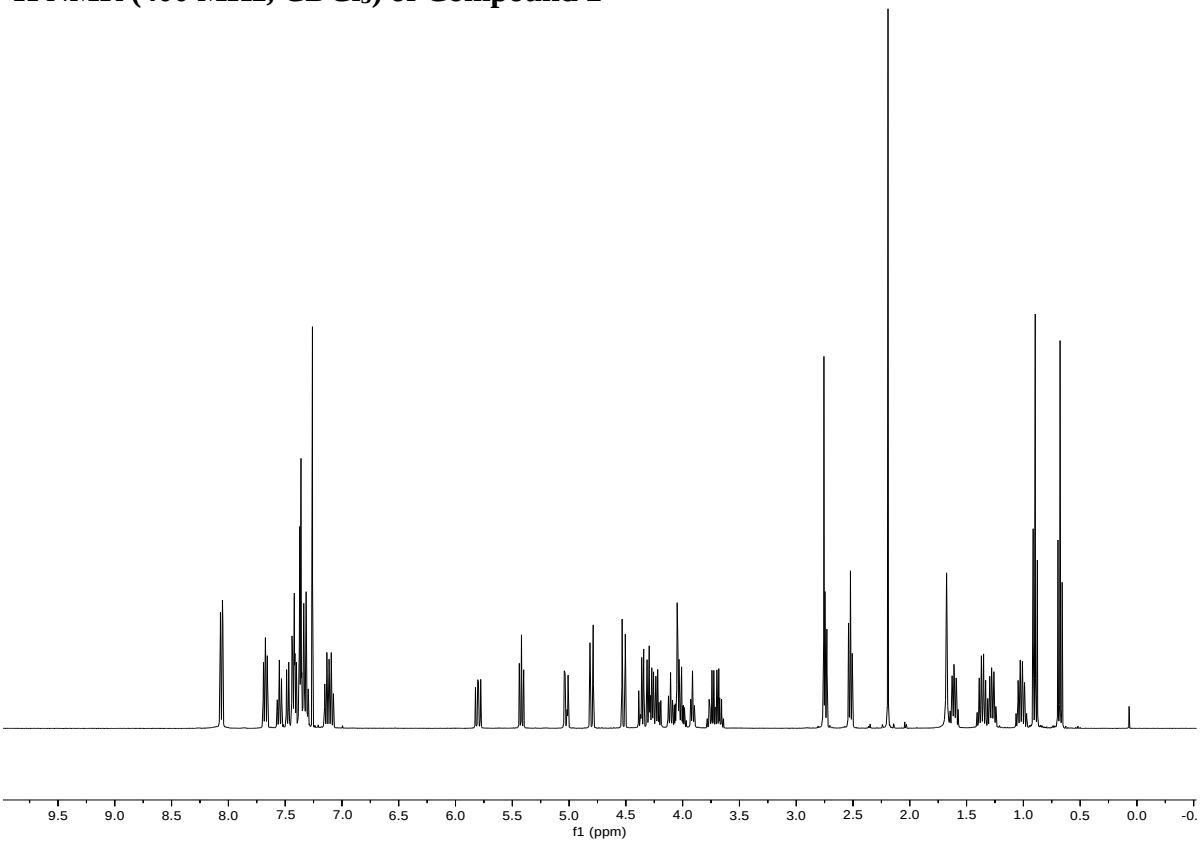
^{1}H NMR (400 MHz, $CDCl_3$) of Compound 1



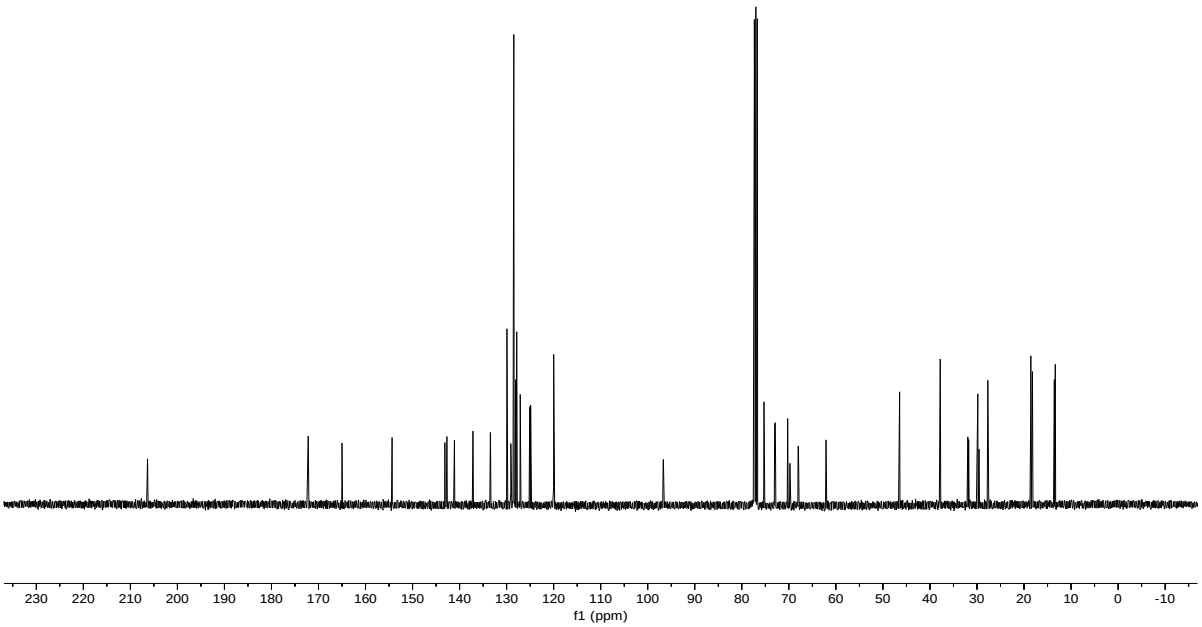
^{13}C NMR (100 MHz, $CDCl_3$) of Compound 1



^{1}H NMR (400 MHz, $CDCl_3$) of Compound 2



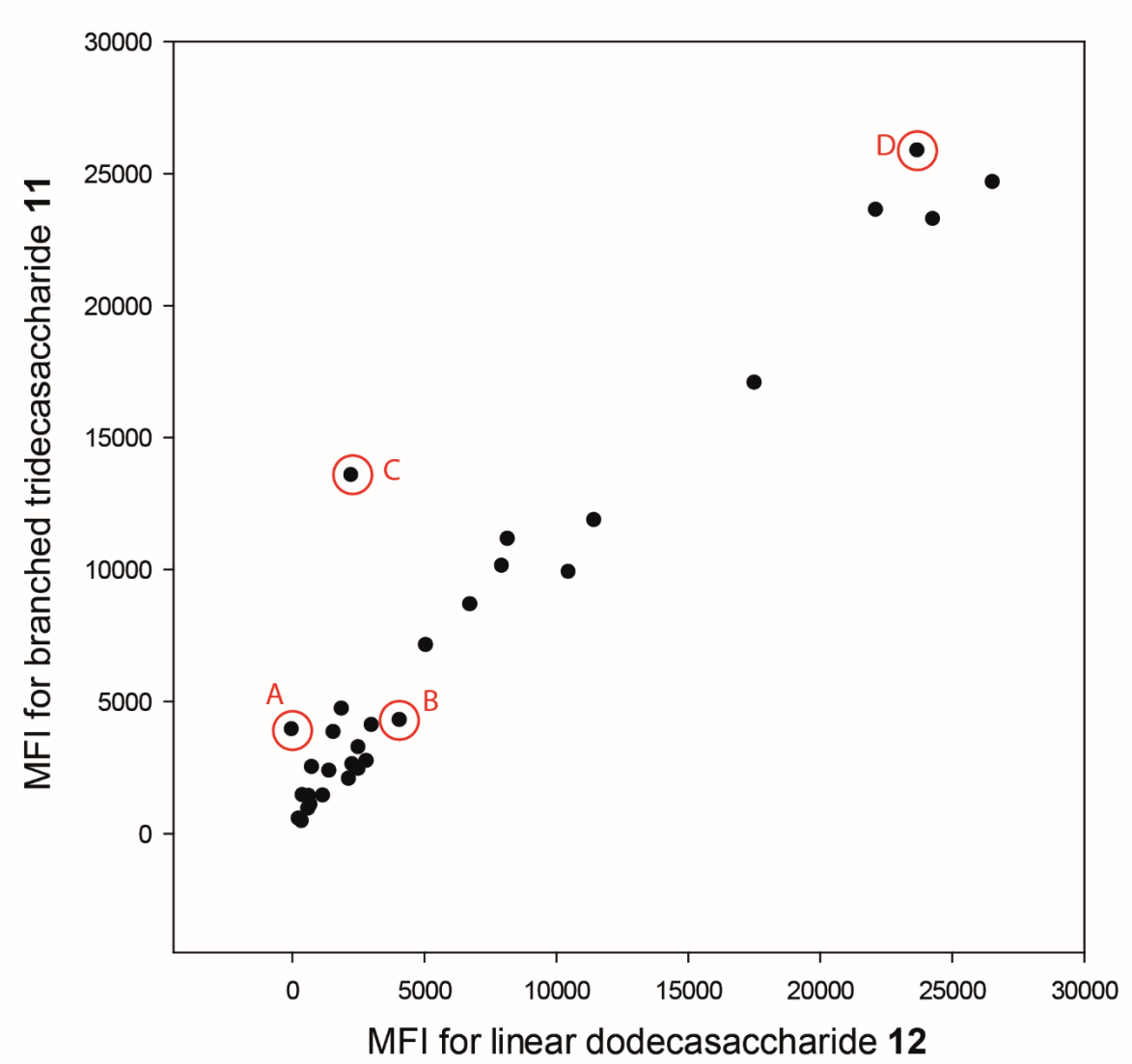
^{13}C NMR (100 MHz, $CDCl_3$) of Compound 2



5. Glycan Array Experiments and Supplementary Figure 1

Glycan Array Experiments

Glycan microarrays were prepared as described previously[2]. Briefly, glycans were dissolved at 0.1 mM and 0.2 mM in 50 mM sodium phosphate buffer pH 8.5 and printed in 64 identical fields to NHS activated hydrogel glass slides (CodeLink slides, Surmodics) using a non-contact S3 microarray spotter (Scienion, Berlin, Germany) according to the pattern shown in Figure 3 (main text). Slides were incubated overnight at high humidity followed by quenching of remaining NHS groups with ethanolamine, drying, and storage at 4 °C. Directly before serum incubation, slides were blocked with 1% (w/v) bovine serum albumin (BSA) in phosphate buffered saline (PBS). A 64 well incubation gasket (FlexWell Grid, Grace Bio Labs) was attached. Human sera (kind gift of Dr. Andreas Bergmann, Sphingotec GmbH, Hennigsdorf, Germany) were diluted 1:100 in 1% BSA-PBS with 0.1% Tween-20, incubated for 10 min at 37 °C, then applied to the slide and incubated overnight at 4°C. Slides were washed thrice with PBS containing 0.1% (v/v) Tween-20 (PBS-T). The gasket was removed; slides were dried and subsequently incubated for 30 min with anti-human IgG-Fc AlexaFluor 488 (Dianova, Cat. 109-545-098) diluted 1:400 in 1% BSA-PBS under a cover slip. After washing twice with PBS-T, once with PBS and rinsing with water, slides were dried and scanned with a GenePix 4300A microarray scanner (Molecular Devices). Intensities were evaluated with GenePix 7.2 (Molecular Devices). Spot diameters were kept identical for the evaluations of all samples. Branched glycans were evaluated with a spot diameter of 240 µm while 210 µm was used for the linear dodecasaccharide 12 for which the spots appeared smaller. Each reported intensity value is the mean of eight spots from two replicate wells with the local background subtracted.



Supporting Figure 1. Glycan microarray comparison of antibody levels in humans to linear dodecasaccharide **12** to represent linear epitopes and branched tridecasaccharide **11**. The positions of the same sera as in Figure 3 (main text) are highlighted.

6. References

1. Martin, C. E.; Weishaupt, M. W.; Seeberger, P. H. *Chem. Commun.* **2011**, *47*, 10260-10262.
2. Pereira, C. L., Geissner, A., Anish, C. and Seeberger, P. H. *Angew. Chem. Int. Ed.* **2015**, 54, 10016–10019; Reinhardt, A, Yang, Y., Claus, H., Pereira, C. L., Cox, A. D., Vogel, U., Anish, C., Seeberger, P. H. *Chem. Biol.* **2015**, 22, Issue 1, 22, 38–49.