Supplementary Information

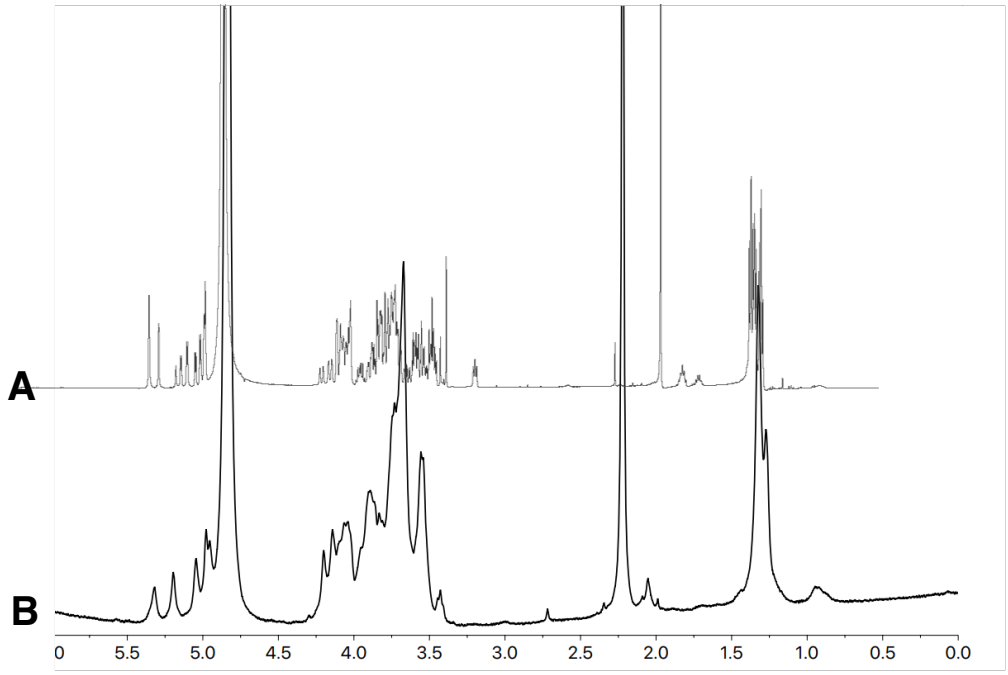
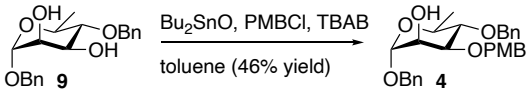


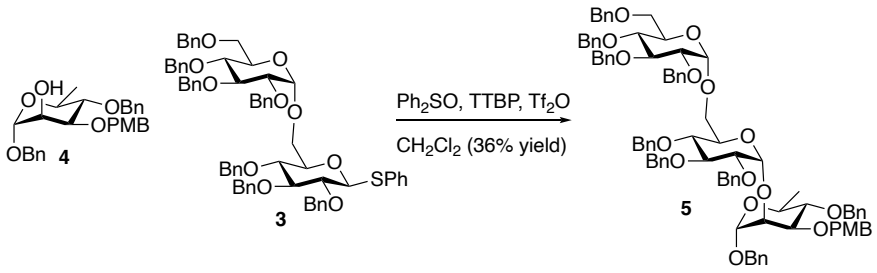
Figure S1. Overlay of ^{1}H NMR spectra of **A:** pentasaccharide **1** and **B:** glycorhamnan from *R. gnavus* from ref. 9.

Synthesis of **1**



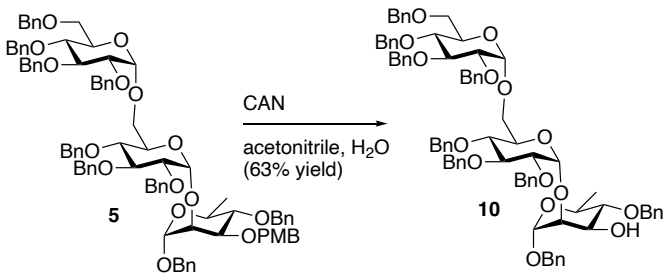
Preparation of 4: Compound **9**[15] (3.28 g, 9.53 mmol) was dissolved in anhydrous toluene (75 mL) before addition of Bu_2SnO (2.37 g, 9.53 mmol). The reaction was heated to 85 °C and stirred until the reaction turned clear (1 h), after which PMBCl (1.28 mL, 9.53 mmol) and TBAB (3.07 g, 9.53 mmol) were added. The reaction was stirred for 4 h at 85 °C and then concentrated. Column chromatography (SiO_2, ethyl acetate/hexanes; 1/1) gave **4** as a clear oil (2.06 g, 46%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.28 – 7.22 (m, 12H), 6.83 (dd, J = 8.6, 2.5 Hz, 2H), 4.86 (d, J = 11.7 Hz, 2H), 4.69 – 4.58 (m, 2H), 4.58 (s, 2H), 4.44 (d, J = 11.9 Hz, 1H), 4.01 (t, J = 2.6 Hz,

1H), 3.86 (dt, J = 9.1, 2.6 Hz, 1H), 3.82 – 3.75 (m, 1H), 3.74 (d, J = 1.9 Hz, 3H), 3.46 (t, J = 9.3 Hz, 1H), 2.7 (s, 1H), 1.30 (s, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 159.49, 138.58, 138.56, 137.47, 130.20, 130.18, 129.66, 128.66, 128.51, 128.49, 128.08, 128.06, 127.91, 127.81, 114.02, 98.49, 98.46, 80.08, 79.91, 76.99, 75.47, 71.82, 69.10, 68.71, 67.71, 67.69, 55.32, 18.04. HRMS (ESI) m/z: $[M+NH_4]^+$ 482.2548; calculated: 482.2537.

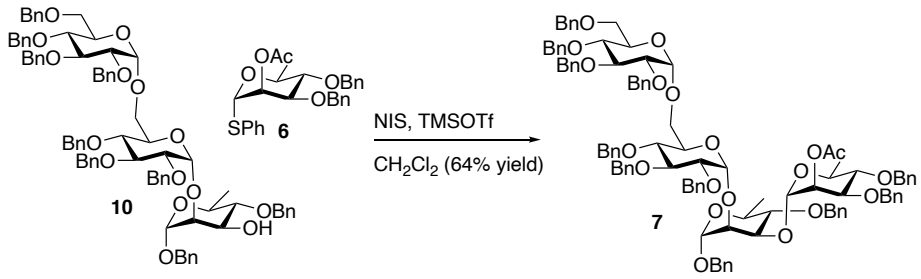


Preparation of 5: Compound **3**[16] (164 mg, 0.15 mmol) was dissolved in dichloromethane (3 mL) before addition of Ph_2SO (78 mg, 0.39 mmol), TTBP (134 mg, 0.54 mmol), and molecular sieves 4Å (400 mg). The reaction was stirred for 1 h at room temperature and then cooled to -60 ^oC before addition of Tf_2O (36 μL, 0.22 mmol). The solution was allowed to warm to -40 ^oC while stirring (20 min). Compound **4** (107 mg, 0.23 mmol) was then dissolved in dichloromethane (2 mL) and added dropwise to the reaction mixture. The cold bath was removed, and the reaction was allowed to stir for 1 h, after which triethylamine was added (4 drops). The solution was concentrated and subjected to SiO_2 chromatography (ethyl acetate/hexanes; 1/1) which yielded **5** as a colorless oil (79 mg, 36%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.03 – 7.95 (m, 4H), 7.55 (dt, J = 30.5, 7.4 Hz, 4H), 7.40 (dd, J = 11.8, 4.0 Hz, 2H), 7.34 (s, 2H), 7.35 – 7.28 (m, 7H), 7.27 (d, J = 10.7 Hz, 12H), 7.27 – 7.22 (m, 8H), 7.22 (d, J = 4.5 Hz, 1H), 7.18 (q, J = 5.3, 2.9 Hz, 4H), 7.09 (dd, J = 6.6, 3.0 Hz, 1H), 6.78 (d, J = 8.3 Hz, 1H), 5.73 (d, J = 3.6 Hz, 1H), 5.05 – 4.99 (m, 2H), 4.96 – 4.89 (m, 3H), 4.88 – 4.75 (m, 2H), 4.73 –

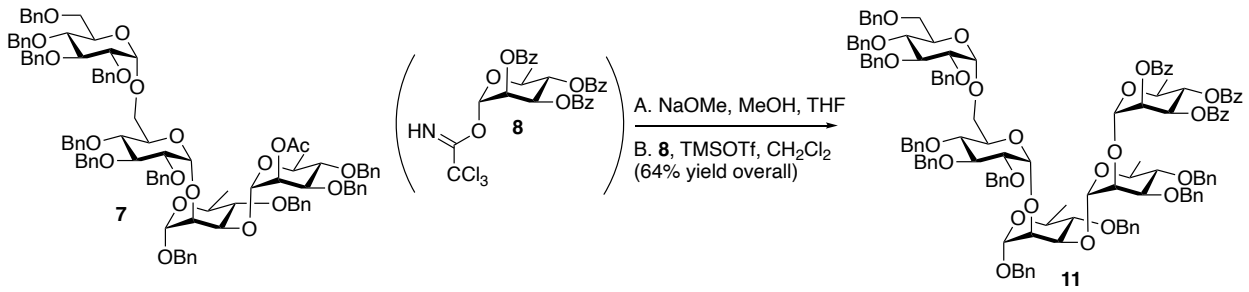
4.65 (m, 2H), 4.67 – 4.62 (m, 2H), 4.64 – 4.48 (m, 5H), 4.45 – 4.38 (m, 2H), 4.23 (dd, J = 17.0, 10.9 Hz, 2H), 4.18 – 4.02 (m, 2H), 3.99 – 3.88 (m, 2H), 3.82 – 3.58 (m, 4H), 3.63 (s, 3H), 3.55 – 3.41 (m, 2H), 3.41 – 3.33 (m, 1H), 1.61 (s, 2H), 1.37 (d, J = 6.2 Hz, 2H), 1.30 (s, 2H), 1.27 (s, 4H), 1.23 (s, 1H), 0.89 (dq, J = 19.2, 7.7, 6.6 Hz, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 158.98, 141.62, 139.16, 138.58, 138.51, 138.32, 137.95, 137.48, 133.19, 129.29, 129.10, 128.63, 128.40, 128.35, 128.33, 128.29, 128.24, 128.18, 128.10, 128.05, 128.03, 127.88, 127.80, 127.74, 127.68, 127.63, 127.61, 127.55, 127.48, 127.43, 127.30, 127.22, 127.00, 126.86, 113.70, 96.91, 96.58, 81.91, 81.58, 80.05, 79.57, 78.53, 77.66, 75.45, 75.14, 74.98, 73.98, 73.43, 73.06, 72.83, 72.70, 72.43, 71.62, 70.85, 70.22, 68.87, 68.72, 68.47, 68.17, 55.09, 29.72, 18.05. HRMS (ESI) m/e: $[M+NH_4]^+$ 1436.6773; calculated: 1436.6880.



Preparation of 10: Compound **5** (79 mg, 56 μmol) and CAN (122 mg, 220 μmol) were dissolved in acetonitrile/H_2O (9:1, 5 mL). The reaction was stirred at rt for 30 min, then diluted with dichloromethane and washed with aqueous saturated sodium sulfite. The organic phase was collected, concentrated, and subjected to SiO_2 chromatography (ethyl acetate/hexanes; 1/1) to afford **10** as a clear, colorless oil (46 mg, 63%). HRMS (ESI) m/e: $[M+NH_4]^+$ 1316.6287; calculated: 1316.6305.

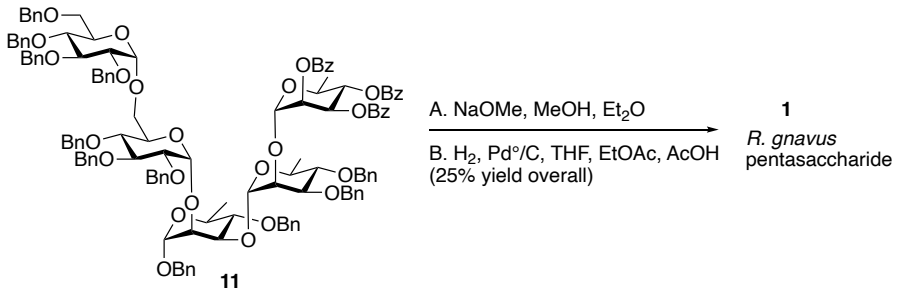


Preparation of 7: Compound **6**[18] (104 mg, 80 μmol) was dissolved in anhydrous dichloromethane and molecular sieves 4Å (200 mg) were added. The solution was stirred at rt for 1 h before being cooled to -30 °C. NIS (27 mg, 120 μmol) and TMSOTf (2 drops, cat.) were added followed by **10** in dichloromethane. The reaction was stirred at -30 °C for 30 min. The reaction was quenched with triethylamine and then concentrated. Column chromatography (SiO_2, ethyl acetate/hexanes; 1/2) afforded **7** as a white solid (85 mg, 64%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.31 – 7.14 (m, 55H), 5.14 (d, J = 4.2 Hz, 2H), 4.97 (d, J = 10.4 Hz, 4H), 4.95 – 4.85 (m, 2H), 4.85 – 4.72 (m, 4H), 4.71 – 4.59 (m, 3H), 4.59 (d, J = 8.9 Hz, 5H), 4.57 – 4.42 (m, 4H), 4.36 (dq, J = 12.1, 6.9, 6.4 Hz, 4H), 4.18 – 4.05 (m, 2H), 4.02 (dd, J = 8.9, 4.9 Hz, 2H), 4.01 – 3.94 (m, 1H), 3.90 – 3.76 (m, 4H), 3.74 – 3.64 (m, 1H), 3.64 – 3.54 (m, 1H), 3.42 (dq, J = 8.1, 4.0 Hz, 2H), 2.06 (s, 3H), 1.38 (dt, J = 10.2, 5.5 Hz, 1H), 1.30 (td, J = 13.6, 12.4, 6.0 Hz, 6H), 1.15 (s, 1H), 0.92 (dt, J = 17.3, 6.3 Hz, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 169.96, 139.04, 138.94, 138.72, 138.70, 138.61, 138.12, 138.07, 137.46, 128.49, 128.44, 128.39, 128.36, 128.35, 128.32, 128.29, 128.27, 128.25, 128.19, 128.11, 128.08, 128.04, 127.97, 127.95, 127.91, 127.88, 127.85, 127.82, 127.78, 127.75, 127.72, 127.69, 127.67, 127.65, 127.64, 127.60, 127.58, 127.51, 127.49, 127.48, 127.46, 127.41, 127.38, 99.43, 97.31, 95.31, 94.69, 81.63, 81.61, 80.93, 80.31, 80.24, 79.87, 78.64, 77.57, 77.02, 75.47, 75.40, 75.20, 75.13, 74.90, 73.37, 72.41, 72.24, 71.88, 71.56, 70.38, 69.05, 68.87, 68.69, 68.46, 68.23, 65.63, 29.75, 20.99, 18.09, 18.04, 18.00. HRMS (ESI) m/e: $[M+NH_4]^+$ 1684.8023; calculated: 1684.7929



Preparation of 11: Compound **7** (85 mg, 51 μmol) was dissolved in methanol/THF (1:2, 3 mL) and NaOMe (0.1 mL, 1 *M*) was added. The reaction was stirred for 24 h. Dichloromethane was added (5 mL) and the mixture was washed with water. The organic phase was collected, concentrated and dissolved in anhydrous dichloromethane (5 mL). Compound **8**[20] (42 mg, 68 μmol) and molecular sieves 4Å (120 mg) were added and stirred at rt for 1 h. The mixture was then cooled to -10 °C before the addition of TMSOTf (1 drop, cat.). The reaction was stirred at -10 °C for 20 min before being quenched with triethylamine (3 drops). The solution was then filtered, concentrated, and subjected to SiO_2 chromatography (ethyl acetate/hexanes; 1/2) to afford **11** as a white solid (68 mg, 64%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.09 (d, J = 7.7 Hz, 2H), 7.97 (d, J = 7.7 Hz, 2H), 7.83 (d, J = 7.7 Hz, 2H), 7.64 (t, J = 7.4 Hz, 1H), 7.52 (t, J = 7.8 Hz, 5H), 7.47 – 7.40 (m, 6H), 7.40 – 7.28 (m, 25H), 7.26 -- 7.037 (m, 27H), 5.86 – 5.78 (m, 2H), 5.56 (m, 1H), 5.15 (s, 1H), 5.10 (s, 1H), 5.00 – 4.92 (m, 2H), 4.93 (s, 2H), 4.87 (dd, J = 14.6, 9.2 Hz, 2H), 4.82 – 4.60 (m, 2H), 4.60 – 4.42 (m, 3H), 4.34 (dd, J = 17.7, 9.2 Hz, 2H), 4.22 – 3.92 (m, 3H), 3.89 – 3.76 (m, 2H), 3.72 (s, 2H), 3.72 – 3.56 (m, 2H), 3.50 – 3.36 (m, 1H), 1.62 (s, 3H), 1.33 – 1.23 (m, 15H), 1.23 –1.14 (m, 5H), 1.14 – 0.97 (m, 4H), 0.96 – 0.78 (m, 5H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 165.83, 165.28 (d, J = 33.8 Hz), 138.99, 138.68, 138.58, 138.05, 137.38, 133.30, 133.23, 132.95, 129.94, 129.80, 129.69, 129.43, 129.35, 128.52, 128.45, 128.41, 128.38, 128.35, 128.33, 128.30, 128.29, 128.27, 128.25, 128.23, 128.21, 128.19, 128.17, 128.10,

128.05, 127.98, 127.83, 127.80, 127.76, 127.73, 127.71, 127.65, 127.63, 127.59, 127.57, 127.47, 127.45, 127.37, 127.33, 101.12, 99.41, 97.27, 94.62, 81.76, 81.58, 80.52, 80.22, 80.11, 79.73, 77.78, 77.58, 75.57, 75.45, 75.43, 75.26, 74.89, 73.39, 73.11, 72.39, 71.91, 71.87, 71.75, 70.60, 70.37, 69.96, 69.13, 68.69, 68.47, 67.00, 65.59, 31.94, 29.72, 22.71, 18.04, 17.90, 17.50, 14.15, 1.04. HRMS (ESI) m/e: $[M+NH_4]^+$ 2100.9231; calculated: 2100.9189.



Preparation of 1: Compound **11** (34 mg, 16 μmol) was dissolved in methanol/ethyl ether (2:1, 3 mL) before the addition of NaOMe (0.1 mL, 1 *M*). The reaction was stirred at rt for 30 min, after which the solvent was removed and the crude mixture was passed through a SiO_2 plug (2:1 EtOAc:Hexanes). The eluent was concentrated and dissolved in THF/EtOAc/AcOH (3:6:3, 5 mL) before addition of activated Pd/C (100 mg, 10%). The solution was placed into a pressure chamber and was pressurized with H_2 to 500 psi. The reaction was stirred for 3 d, before being filtered, concentrated and subjected to SiO_2 chromatography (EtOAc/MeOH/H_2O, 12/5/1) to afford target compound **1** as a white solid (3.2 mg, 25%). ^{1}H NMR (500 MHz, D_2O) δ 5.18 (s, 1H), 5.11 (s, 1H), 4.85 – 4.79 (m, 3H), 4.05 – 3.88 (series of m, 24H), 1.75 (s, 1H), 1.19 – 1.07 (series of m, 9H). ^{13}C NMR (126 MHz, D_2O) δ 102.16, 102.04, 100.23, 99.95, 98.44, 97.95, 97.92, 93.66, 91.38, 82.47, 78.10, 77.91, 77.72, 76.53, 74.39, 73.28, 72.96, 72.94, 72.82, 72.58, 72.52, 72.24, 72.18, 71.98, 71.93, 71.81, 71.79, 71.54, 71.50, 71.27, 71.23, 70.93, 69.95, 69.92,

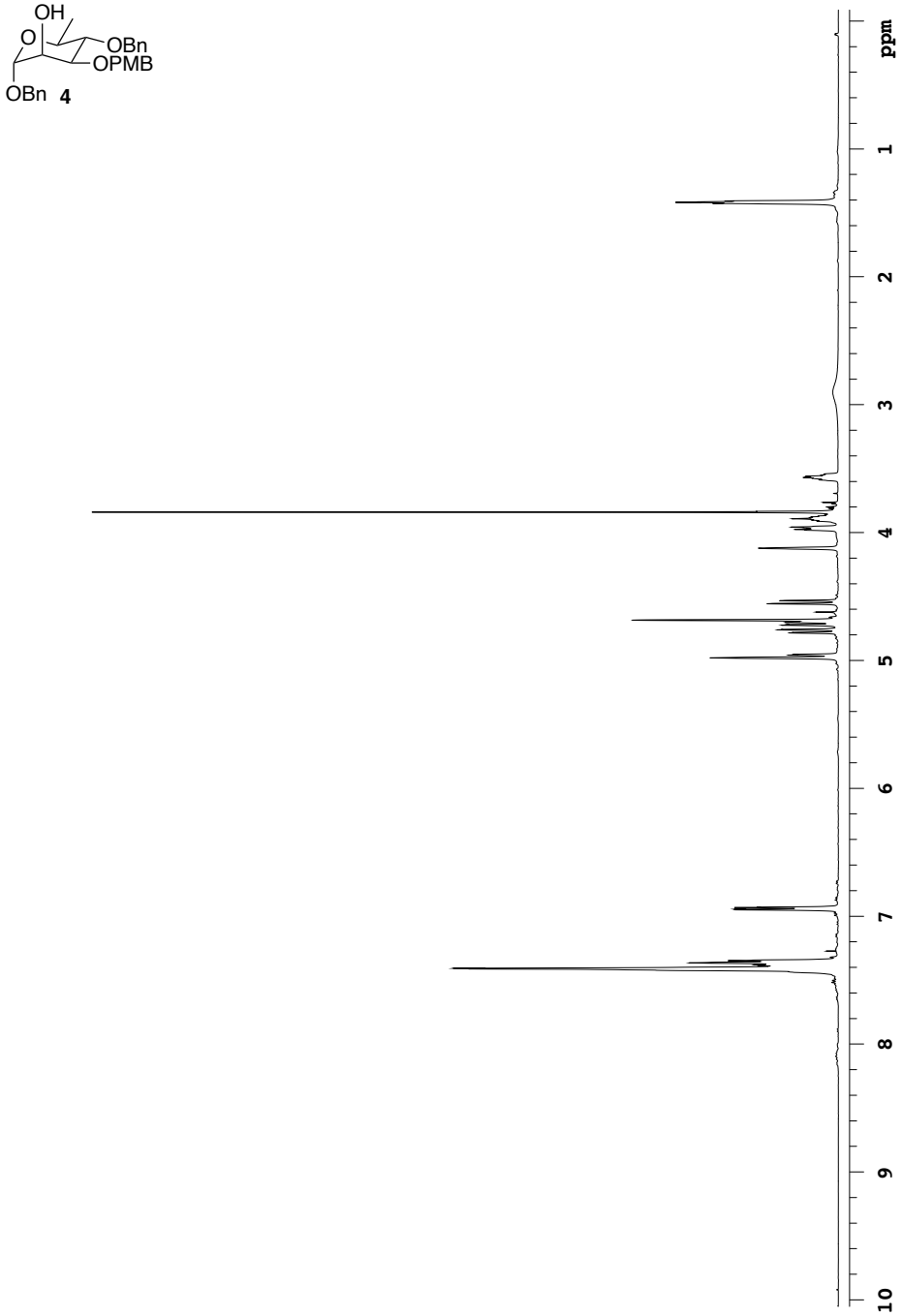
69.40, 69.24, 69.04, 68.90, 68.81, 68.61, 68.41, 64.89, 64.61, 60.36, 23.17, 19.96, 16.60, 16.57, 16.54, 16.52, 16.46, 16.43. HRMS (ESI) m/e: $[M+NH_4]^+$ 798.3189; calculated: 798.3238.

Generation and in vitro stimulation of bone marrow-derived dendritic cells (BMDCs)

Femur and tibia of wild type C57Bl/6J and Tlr4 KO mice (purchased from The Jackson Laboratory) were crushed with a plunger. Bone marrow cells and bone debris was washed with sterile PBS (Corning) and filtered through a 100-µm cell strainer. Cells were pelleted, resuspended in 1 x RBC lysis buffer (Thermo Fisher) and incubated for 5 min on ice. Cells were washed with PBS and resuspended in complete RPMI-1640 medium (HyClone) supplemented with 10% fetal calf serum (Gibco), 1 x MEM Non-Essential Amino Acids (Gibco), Pen Strep Glutamine (Gibco), 10 mM HEPES (Gibco), 1 mM Sodium Pyruvate (Gibco), 50 µM 2-Mercaptoethanol (Gibco) and 20 ng/ml GM-CSF (Biolegend). Cells were plated in a 6 well plate at a density of 2.5 x 10^6 cells per ml. Media was exchanged every 2-3 days and cell purity checked using antibodies against CD11c (N418), CD11b (M1/70), MHCII (M5/114.15.2) and TLR4 (MTS510).

For *in vitro* stimulation cells were harvested at day 8 or day 9 and transferred to a 96 well flat bottom plate (200k cells per well in 200 µl complete culture medium) in the presence of LPS (invivogen), Mannan (Sigma), PBS-281 (*R. gnavus* pentasaccharide) or PBS-195 (*S. pneumoniae* serotype 14 tetrasaccharide) and stimulated for 24 hrs. Interleukin 6 and TNF-alpha was in the supernatant was measured using ELISA-MAX™ kits (Biolegend).

^{1}H NMR Spectra of Compounds 1, 4, 5, 7 and 11.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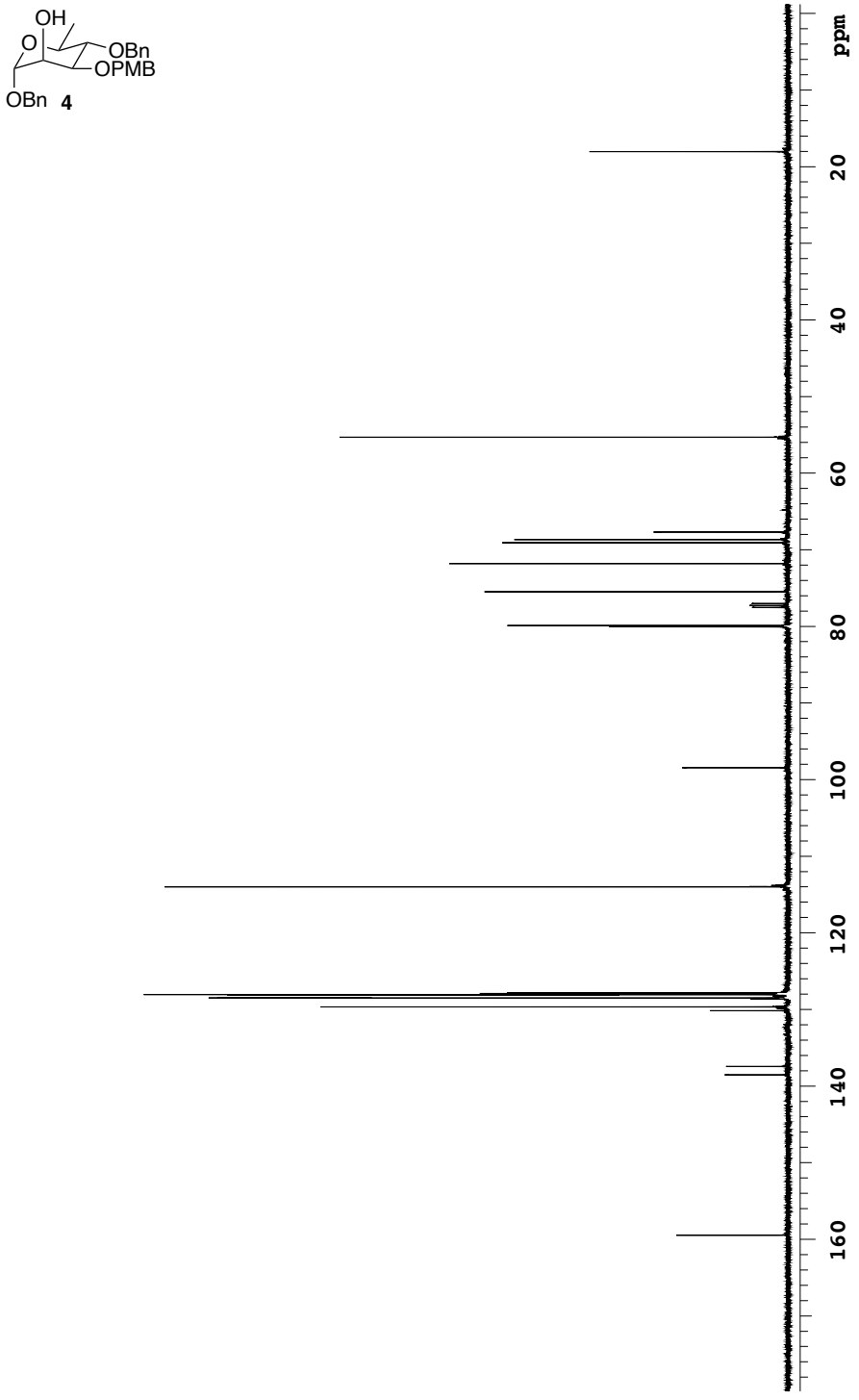
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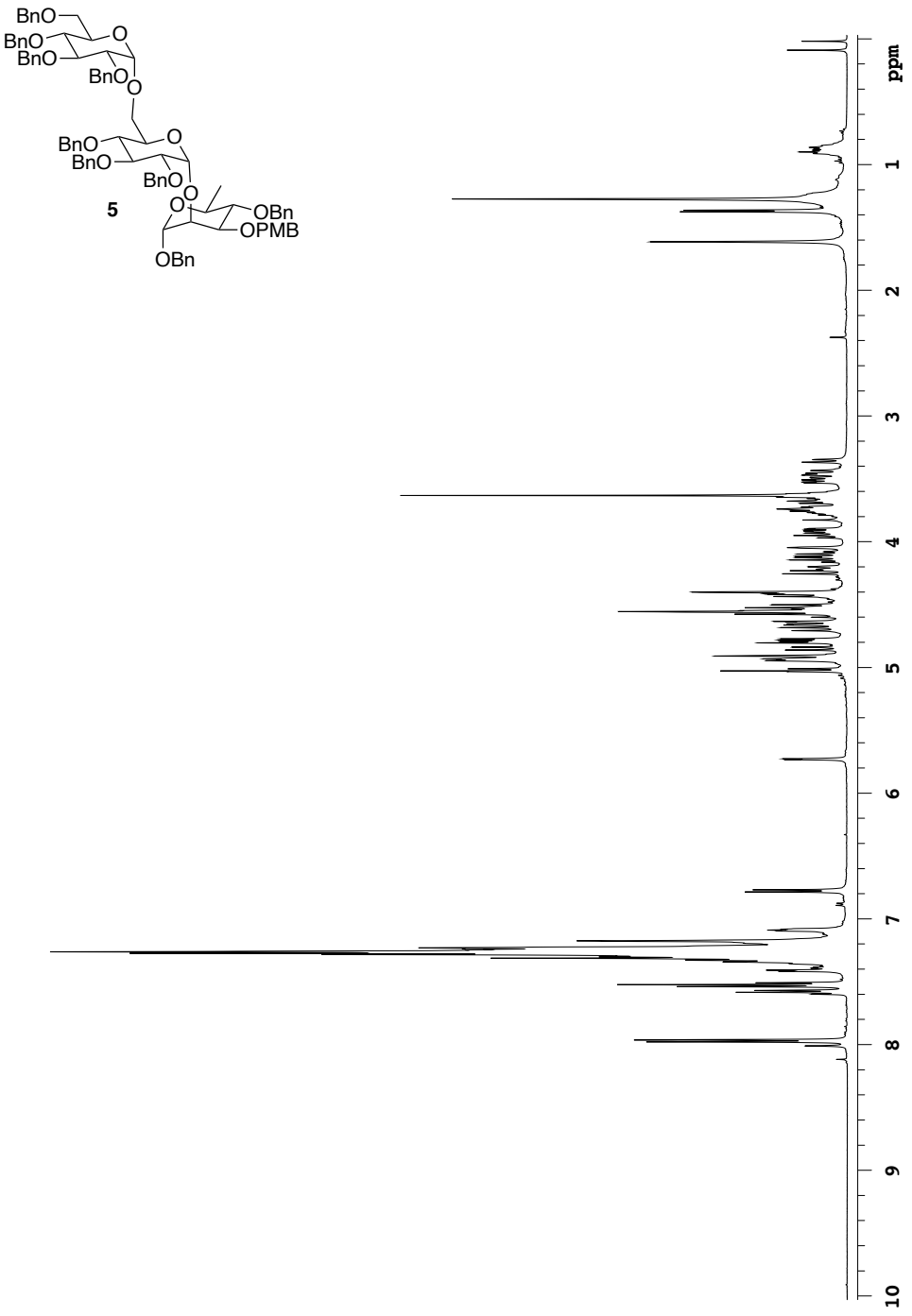
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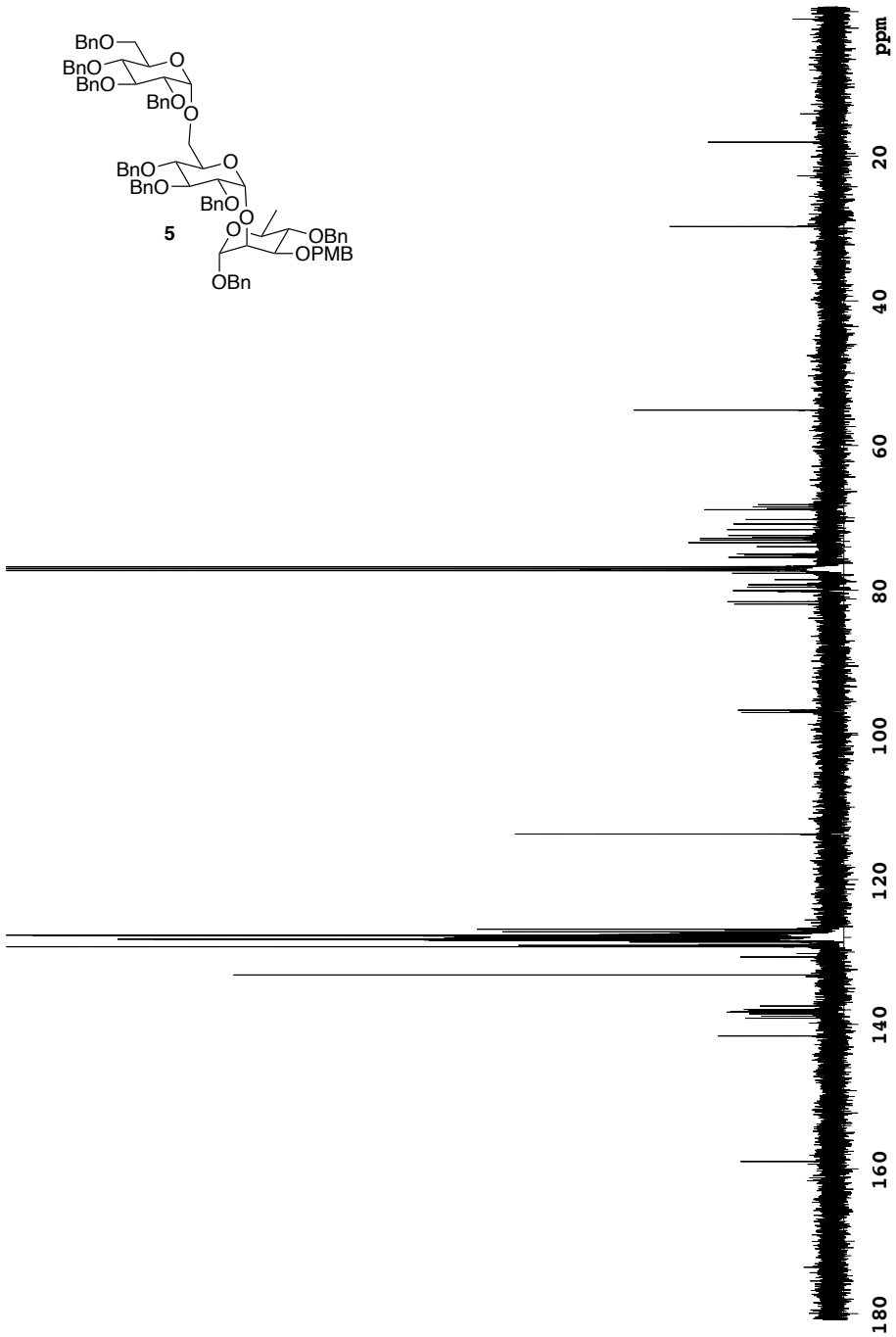
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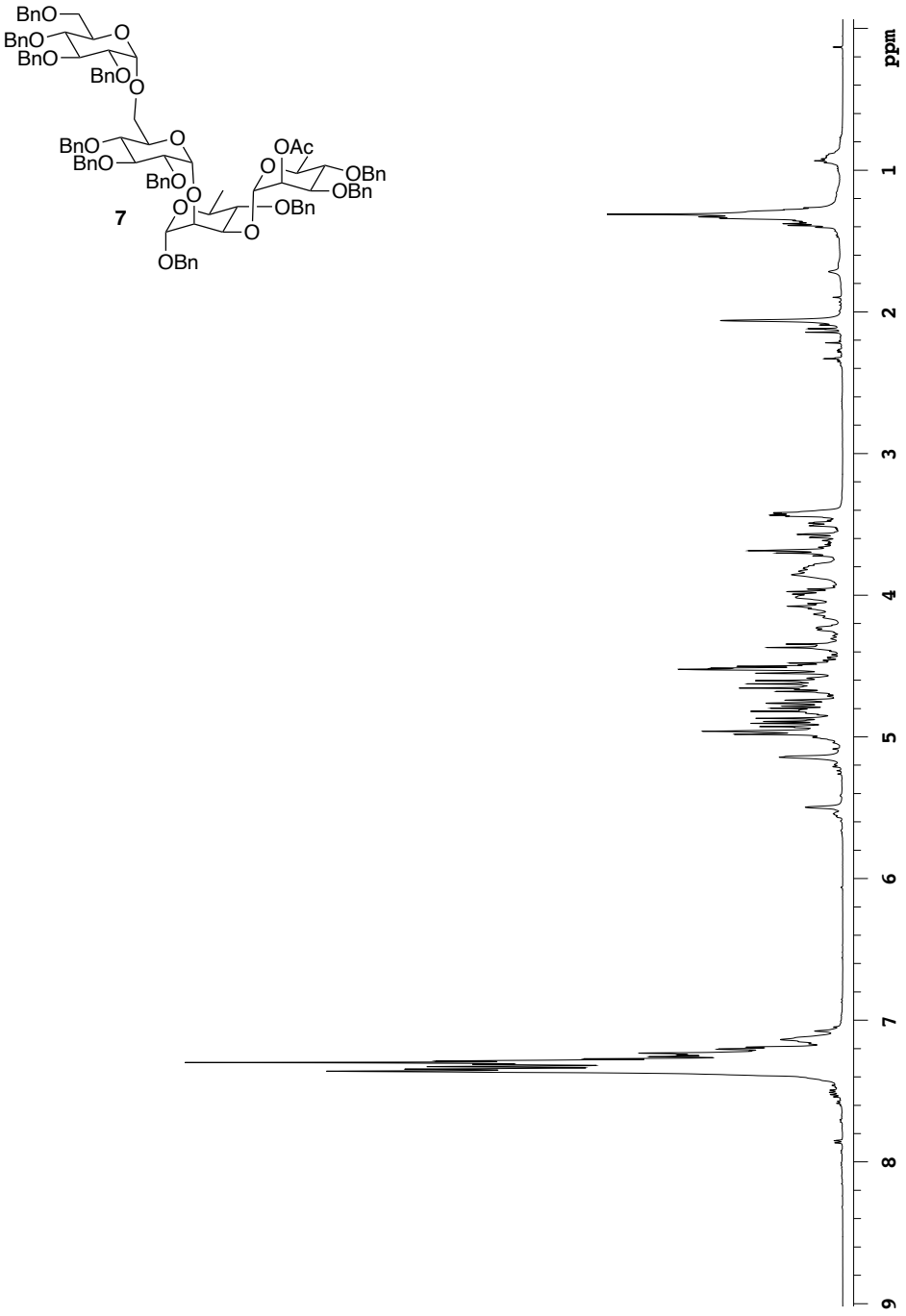
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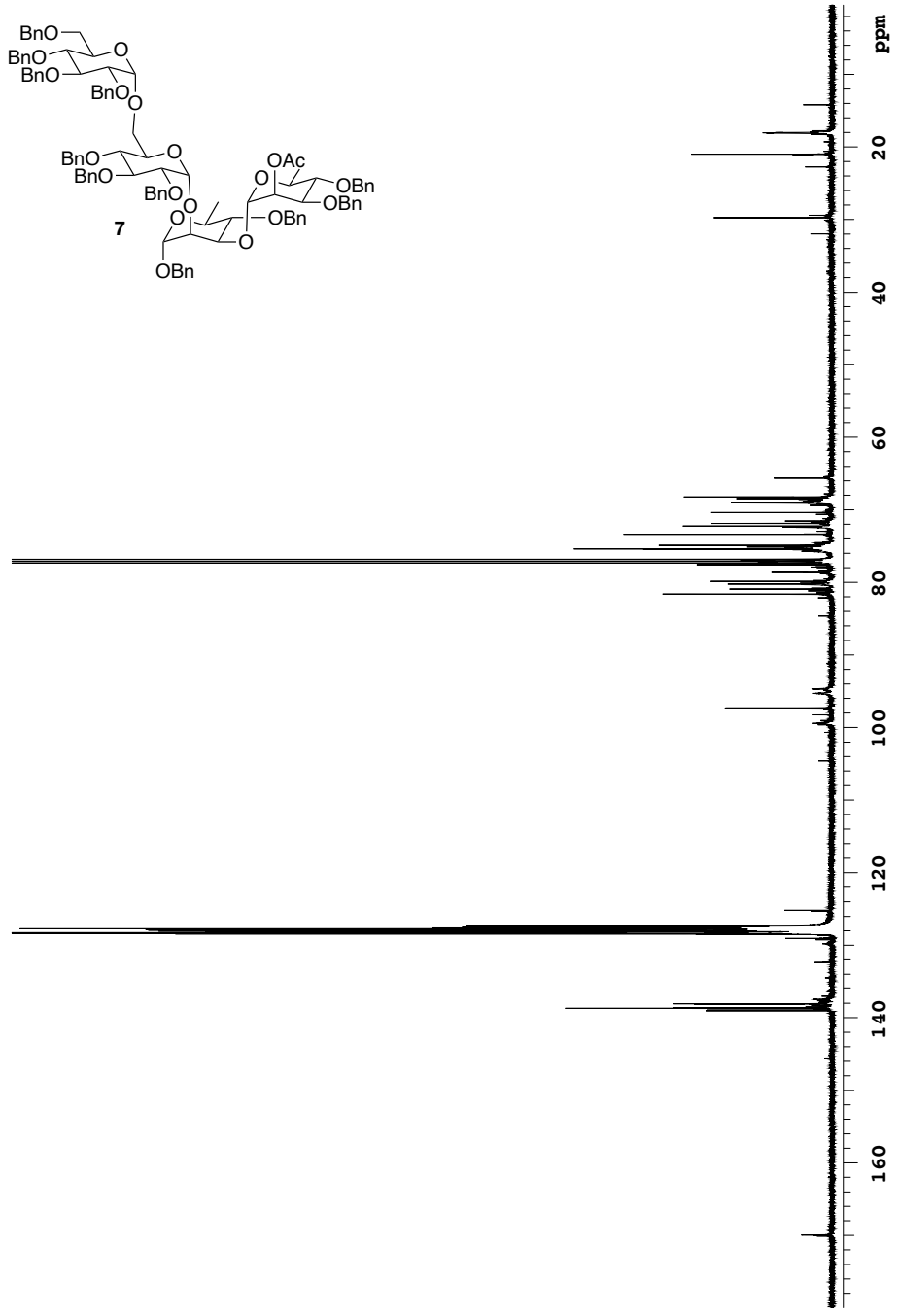
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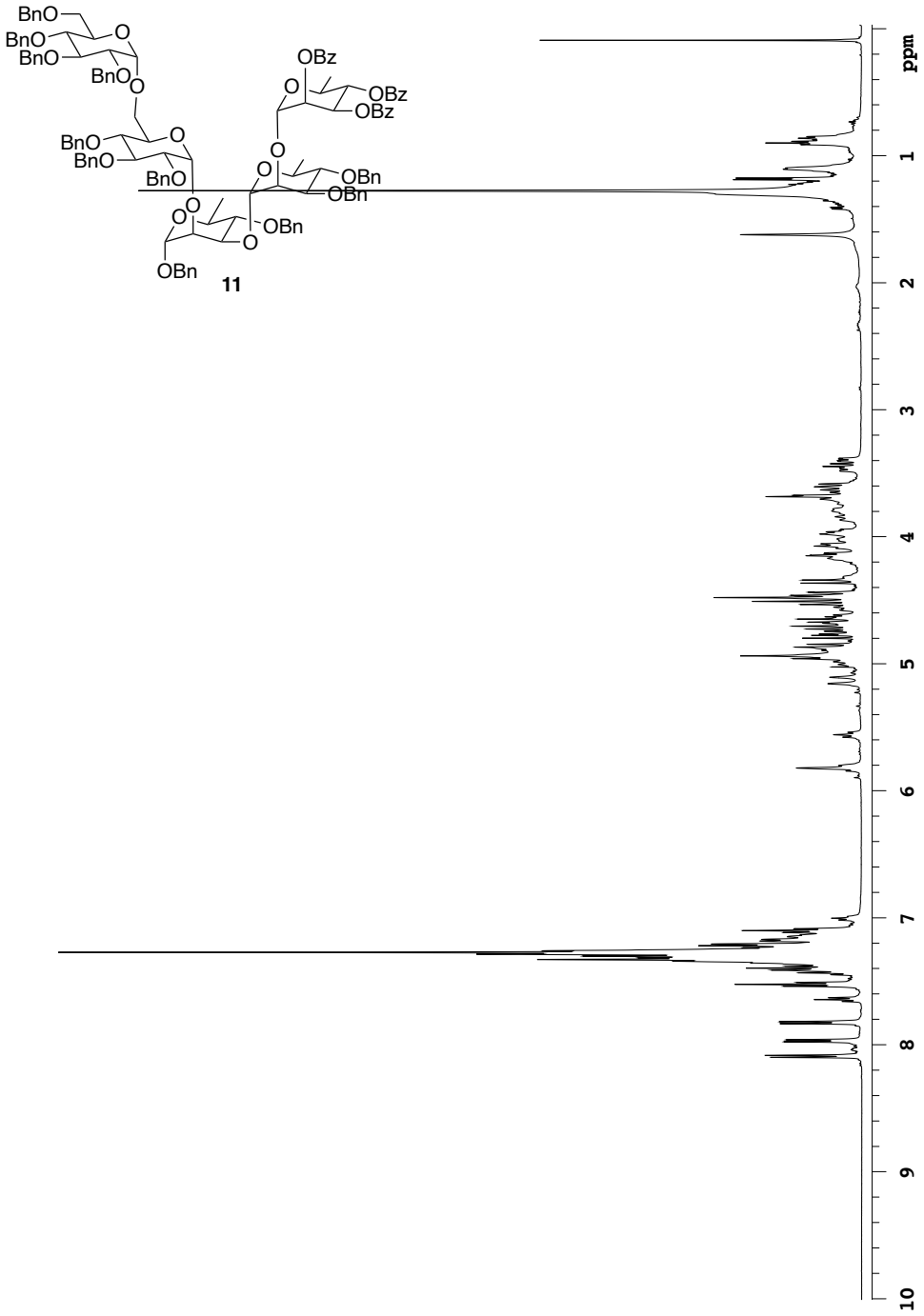
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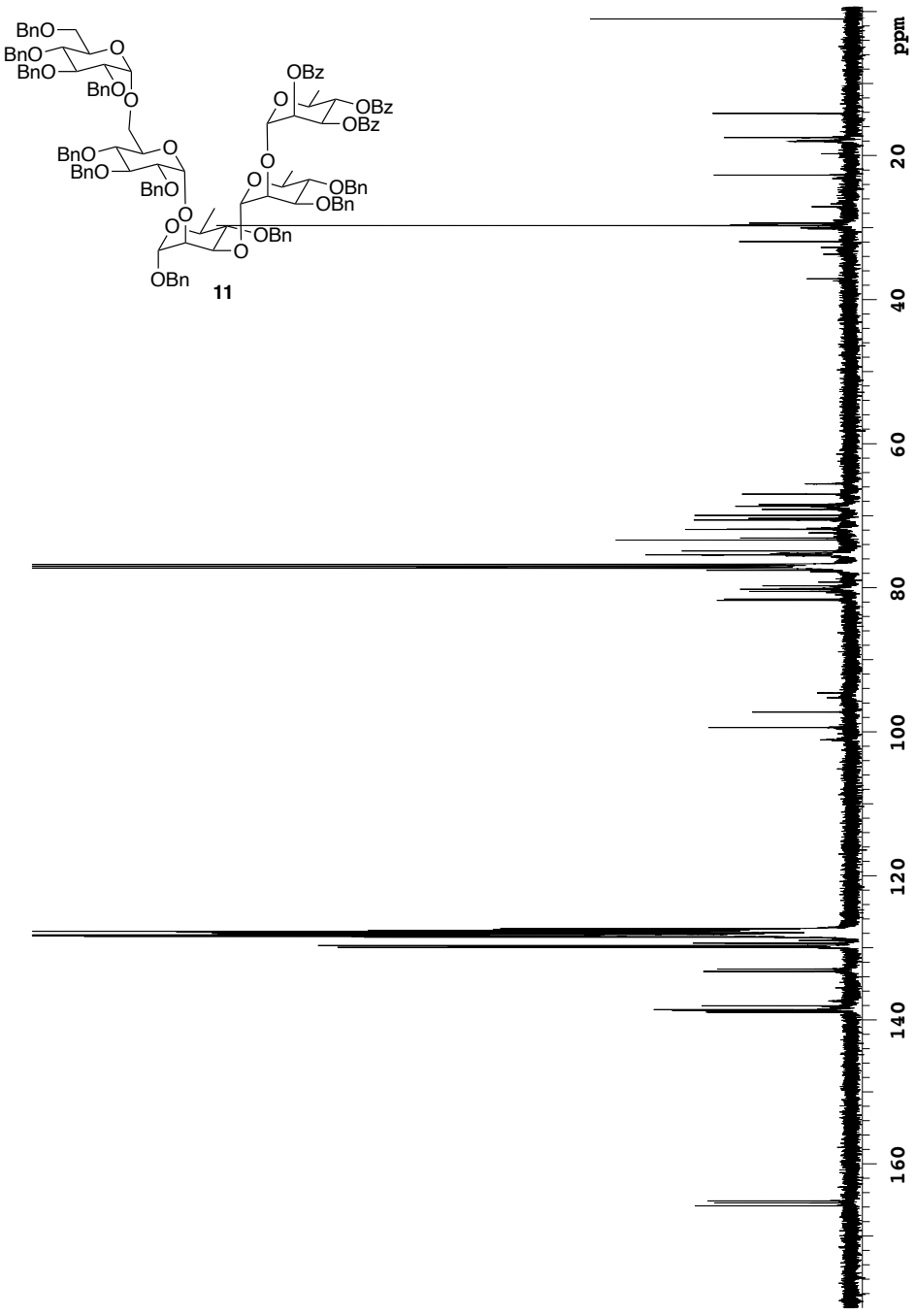
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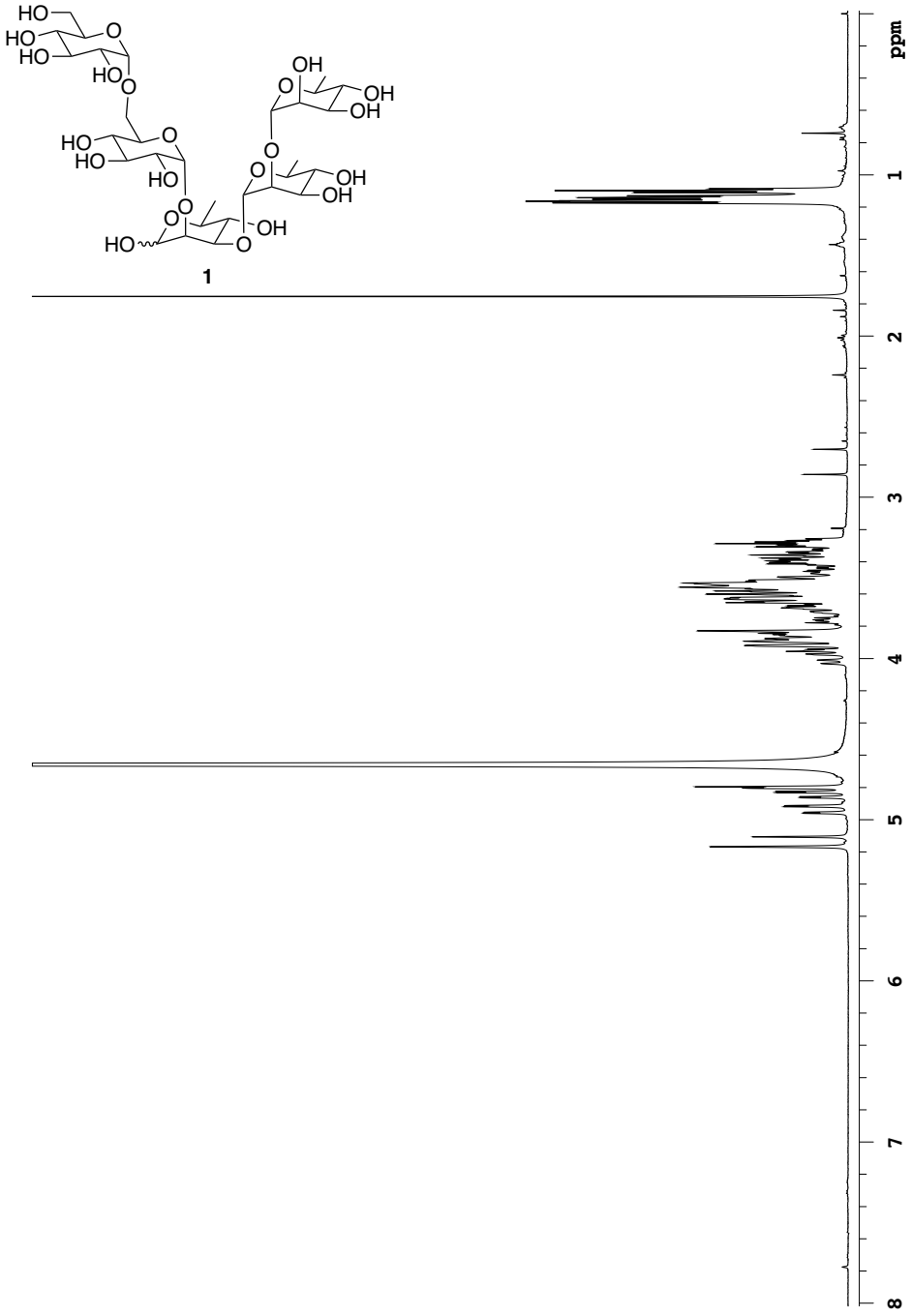
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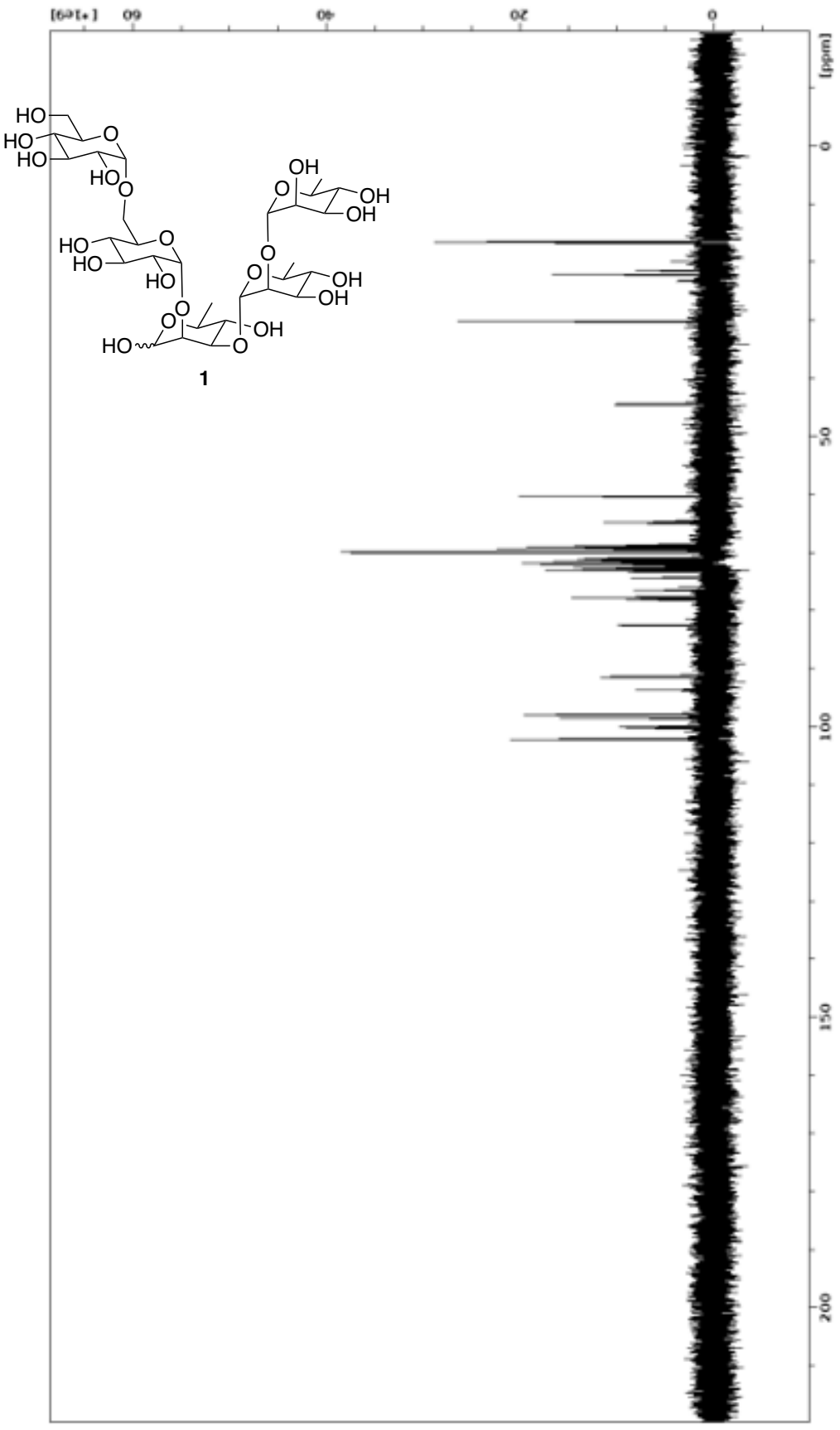
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