Supporting information

An Efficient Synthetic Route to Glyco-amino acid building blocks for Glycopeptide Synthesis

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General considerations

The reducing sugars (GlcNAc, cellobiose, maltose, maltotriose), DIPEA, anhydrous DMSO and DMF were purchased from Aldrich, and lactose from Lancaster. Chitobiose octaacetate was purchased from Dextra labs and ammonium carbonate from BDH labs. Hydroxybenzotriazole (HOBt) was purchased from Acros chemicals and HBTU, Wang resin and all N-α-Fmoc-protected L-amino acids from Novabiochem. TLC plates (Silica gel 60F254) were purchased from Merck and silica-gel (35-70 micron) from Fisher Scientific. Developed TLC plates were visualised under UV lamp and stained with p-anisaldehyde solution. Fmoc-SPPS was monitored by ninhydrin test. All microwave reactions were carried out in CEM Discover automated microwave instrument. The coupling and hydrolysis reactions were carried out under nitrogen atmosphere.

$^1$H NMR, $^{13}$C NMR, COSY and HSQC were recorded on Bruker arx250, dpx360, ava 600 instruments. Chemical shifts were relative to the deuterated solvent peak and were in parts per million (ppm). The mass spectra were recorded on HP1050 using electrospray ionisation and FAB technique. Melting points were determined on Gallenkamp melting point apparatus. LC-MS were run on Alliance HT WATERS
Experimental procedures:

General procedure for microwave-assisted Kochetkov amination reaction (2a-f):
The automated microwave tube was charged with reducing sugar (0.25g), ammonium carbonate (1.25g, 5fold excess) and anhydrous DMSO (0.8ml). The tube was sealed and placed under automated microwave at 40°C, 250psi pressure and 10 watts power for 90 minutes under cooling the vessel. The reaction tube was taken off and the reaction mixture was transferred into a round bottom flask. The reaction mixture was freeze dried for over night to remove excess of ammonia and DMSO to afford β-glycosyl amines (2a-f) as colourless hygroscopic solids in excellent yields. These amines were used without further purification.

Preparation of N-α-Cbz-L-Asn[GlcNAc]-O'Bu (5)

To a stirred suspension of 2a (0.17g, 0.77mmol) in anhydrous DMF (3ml) and NaHCO₃ (0.13g, 1.53mmol) was added N-α-Cbz-Asp(F)-O'Bu (0.34g, 1.15mmol) in anhydrous DMF (3ml). The reaction mixture was stirred at room temperature for 2.5hrs. The solvent was removed under reduced pressure. The crude product was purified by flash chromatography using 12% MeOH in CHCl₃ as an eluent to afford 5 as a colourless solid (0.25g, 62%, melting point 196º-198ºC). IR (KBr): 3295, 3091, 1747, 1703, 1654, 1296 cm⁻¹. ¹H NMR (600MHz, CD₃OD-d₄) δ 7.35 (m, 5H), 5.14 (m, 2H), 4.97 (d, 1H, J= 9.7Hz), 4.61(m, 1H), 3.93 (d, 2H, J= 10.7Hz), 3.84 (dd, 1H, J= 1.6Hz, 10.6Hz), 3.76 (dd, 1H, J= 1.6, 10.6Hz), 3.67 (dd, 1H, J= 1.6Hz, 10.5Hz),
3.45 (m, 1H), 3.32 (m, 5H), 2.76 (dd, 1H, J= 5.7Hz, 17.0Hz), 2.70 (dd, 1H, J= 7.2Hz, 16.9Hz), 1.95 (m, 1H), 1.92 (s, 3H), 0.92 (d, 6H, J= 6.7Hz) 13CNMR (600MHz, CD3OD-d4) δ 175.8, 174.2, 173.8, 159.6, 139.3, 130.7, 130.3, 130.2, 81.6, 81.0, 77.5, 73.8, 73.1, 69.9, 57.5, 57.3, 53.4, 39.7, 30.2, 24.1, 20.6. HRMS MS (FAB) calculated mass for C24H35N3O10, 526.2401 [M + H]+ and was found at 526.2405 [M + H]+

Preparation of N-α-Cbz-L-Asn[GlcNAc]-OH (6):

![Structure of 6](image)

To a stirred solution of 5 (0.15g, 0.3mmol) in DMF (2ml) and phosphate buffer (pH 7.0, 2ml) was added 2M HCl to maintain the pH at 7.0, followed by L-cysteine (9mg) and papain (43mg). The reaction mixture was stirred at 35ºC for 8hrs and the pH was maintained at 7.0 by adding 0.2M NaOH solution. The stirring was continued for further 7hrs. The reaction mixture was filtered through a celite pad and was washed with water (3ml) and the filtrate pH was adjusted to 4.0 by adding 2M HCl and the reaction mixture was concentrated under reduced pressure. The crude product was purified by flash chromatography using 20% MeOH in CHCl3 as an eluent to afford 6 as a white solid (85mg, 63%, isolated yield). IR (KBr): 3294, 1704, 1683, 1650, 1272 cm⁻¹. 1H NMR (360MHz, DMSO-d6) δ 8.22 (d, 1H (NH), J= 9.0Hz), 7.83 (d, 1H(NH), J= 8.9Hz), 7.38 (m, 6H (ArHx5, NH), 4.80 (t, 1H, J= 9.3Hz), 4.32 (dd, 1H, J= 7.6Hz, 13.4Hz), 3.70 (dd, 1H, J= 7.6Hz, 13.5Hz), 3.55 (m, 1H), 3.23 (m, 4H), 2.64 (dd, 1H, J= 5.3Hz, 14.3Hz), 2.48 (dd, 1H, J= 5.3Hz, 14.3Hz), 1.77 (s, 3H). 13C NMR (250MHz, D2O) δ 175.2, 172.9, 172.3, 157.9, 136.2, 129.1, 129.0, 128.5, 79.3, 78.3, 74.4, 70.4, 68.2, 61.2, 55.3, 51.2, 38.4, 31.3, 22.7. MS (ESI) calculated mass for C20H27N3O10, m/z 469.31 and was found at 470.13 [M+H]+, 492.14 [M+Na]+
Preparation of N-α-Fmoc-L-Asn[GlcNAc]-O^tBu (3a)

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\text{3a}
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To a 25ml round-bottom flask was added N-α-Fmoc-L-aspartic acid α-tert butyl ester (0.49g, 1.2mmol), anhydrous DMF (3ml), HOBt (0.15g, 1.1mmol) and HBTU (1.13g, 3.0mmol). After stirring for 15 min, a solution of 2a (0.22g, 1.0mmol) was added in anhydrous DMF (3ml). The reaction mixture was stirred at room temperature under nitrogen atmosphere for 24hrs. The solvent was removed under reduced pressure. The crude product was purified by flash chromatography using 13% MeOH in CHCl\textsubscript{3} as an eluent to afford 3a as a white solid (0.54g, 88%, melting point 182º-184ºC). IR (KBr): 3304, 2924, 1702, 1655, 1547, 1084 cm\textsuperscript{-1}. \textsuperscript{1}H NMR (250MHz, CD\textsubscript{3}OD-d\textsubscript{4}) \(\delta\) 7.63 (d, 2H, J= 7.3Hz), 7.54 (d, 2H, J= 7.3Hz), 7.25 (t, 2H, J= 7.4Hz), 7.15 (t,2H, J= 7.4Hz), 4.82 (d, 1H, J= 9.7Hz), 4.31(m, 2H), 4.09 (m, 2H) , 3.65 (m, 3H), 3.23 (m, 3H), 2.70 (m, 2H), 1.82 (s, 3H), 1.28 (s, 9H). \textsuperscript{13}C NMR (360 MHz, CD\textsubscript{3}OD-d\textsubscript{4}) \(\delta\) 172.6, 170.8, 170.1, 162.9, 156.9, 143.3, 140.7, 126.9, 126.3, 124.3, 119.0, 81.1, 78.4, 77.8, 74.3, 69.8, 66.1, 54.2, 50.8, 36.6, 35.0, 29.7, 26.3, 21.0. HRMS (FAB) calculated mass for C\textsubscript{31}H\textsubscript{39}N\textsubscript{3}O\textsubscript{10}, 614.2714[M + H]\textsuperscript{+} and was found 614.2712[M+H]\textsuperscript{+}.

Preparation of N-α-Fmoc-L-Asn[GlcNAc]-OH (4a)

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To a 50ml round-bottom flask was added 3a (0.49g, 1.2mmol), anhydrous DCM (9ml), TFA (9ml) and anisole (0.9ml). The reaction mixture was stirred at room
temperature under nitrogen atmosphere for 4hrs. The reaction mixture was concentrated under reduced pressure and azeotroped with toluene (2 x 20ml) and DCM (2 x 20ml). The residue was triturated with ether to afford a white coloured solid. The crude product was purified by flash chromatography using 20% MeOH in CHCl3 as an eluent to afford 4a as a white solid (0.155g, 86%, melting point 240ºC dec). IR (KBr): 3306, 3072, 1697, 1654, 1540, 1283, 1081 cm⁻¹. ¹H NMR (360MHz, D₂O + DMSO-d⁶) δ 7.78 (d, 2H, J= 7.3Hz), 7.60 (d, 2H, J= 7.3Hz), 7.37 (t, 2H, J= 7.5Hz), 7.35 (t, 2H, J= 7.5Hz), 4.98 (d, 1H, J= 9.7Hz), 4.42 (t, 1H, J= 6.1Hz), 4.28 (t, 2H, J= 6.1Hz), 4.19 (m, 1H), 3.65 (m, 3H), 3.35 (m, 2H), 3.31 (d, 1H, J= 6.2Hz), 2.42 (m, 2H), 1.87 (s, 3H). ¹³C NMR (360MHz, D₂O + DMSO-d⁶) δ 178.2, 175.6, 174.4, 158.1, 144.9, 141.9, 129.2, 128.6, 126.3, 121.3, 79.2, 78.6, 75.2, 70.5, 67.5, 61.5, 55.3, 53.8, 47.9, 23.1. HRMS (FAB) calculated mass for C₂₇H₃₁N₃O₁₀, 558.2088 [M + H]⁺ and was found at 558.2089 [M+H]⁺

**Preparation of N-α-Fmoc-L-Asn [Glc(β1-4)Glc]-O'Bu (3b)**

![Chemical Structure](3b)

To a 25ml round-bottom flask was added N-α-Fmoc-L-aspartic acid α-tert butyl ester (0.25g, 0.62mmol), anhydrous DMF (3ml), HOBt (83 mg, 0.61mmol) and HBTU (0.58g, 1.53mmol). After stirring for 15 min, was added a solution of 2b (0.175g, 0.51mmol) in anhydrous DMF (3ml). The reaction mixture was stirred at room temperature under nitrogen atmosphere for 24hrs. The solvent was removed under reduced pressure. The crude product was purified by flash chromatography using 20% MeOH in CHCl₃ as an eluent to afford 3b as a white solid (0.34g, 92%, melting point 176º-178ºC). IR (KBr): 3398, 1716, 1539, 1429 cm⁻¹. ¹H NMR (250MHz, CD₃OD-d⁴) δ 7.7 (d, 2H, J= 7.3Hz), 7.60(d, 2H, J= 7.3Hz), 7.34 (t, 2H, J= 7.0Hz), 7.26 (t, 2H, J=...
7.0Hz), 4.96 (d, 1H, J= 12.1Hz), 4.35 (t, 1H, J= 6.0Hz), 4.26 (m, 2H), 4.17 (t, 1H, J= 6.7Hz), 3.77 (ddd, 4H, J= 1.86Hz, 11.9Hz, 22.7Hz), 3.57 (dd, 1H, J= 4.5Hz, 11.2Hz), 3.45 (dd, 1H, J= 4.5Hz, 11.0Hz), 3.38 (m, 2H), 3.21(m, 5H), 2.76 (m, 2H), 1.35 (s, 9H) 13C NMR (360MHz, CD3OD-d4) δ 171.3, 170.2, 156.5 , 143.3, 140.6, 126.9, 126.3, 124.4, 119.0, 102.7, 81.3, 78.9, 78.4, 76.2, 75.8, 75.4, 72.9, 71.7, 69.4, 66.2, 60.5, 59.7, 50.7, 36.9, 26.3. HRMS MS (FAB) calculated mass for C35H46N2O15, 735.2976 [M + H]+ and was found at 735.2976 [M + H]+

Preparation of N-α-Fmoc-L-Asn[Glc(β1-4)Glc]-OH (4b)

To a 50ml round-bottom flask was added 3b (0.15g, 0.2mmol), anhydrous DCM (6ml), TFA (6ml) and anisole (0.6ml). The reaction mixture was stirred under nitrogen atmosphere at room temperature for 4hrs. The reaction mixture was concentrated under reduced pressure and azeotroped with toluene (2 × 20ml) and DCM (2 × 20ml). Finally the residue was triturated with ether to afford 4b as a white coloured solid (0.11g, 80%, melting point 180º-182ºC). IR (KBr): 3367, 2919, 1699, 1541, 1163, 1078 cm⁻¹. 1H NMR (360MHz, D2O + DMSO-d6) δ 7.71 (d, 2H, J= 7.5Hz), 7.51 (d, 2H, J= 7.5Hz), 7.31 (t, 2H, J= 7.3Hz), 7.23 (t, 2H, J= 7.3Hz), 4.75 (d, 1H, J= 9.2Hz), 4.35 (m, 3H), 4.11 (t, 2H, J= 6.2Hz), 3.72 (m, 2H), 3.63 (m, 2H), 3.55 (m, 4H), 3.35 (m, 5H) 2.85 (m, 2H). 13C NMR (360MHz, D2O + DMSO-d6) δ 175.7, 174.1, 158.9, 145.2, 142.4, 129.8, 129.2, 126.8, 122.7, 104.2, 80.7, 79.8, 77.8, 77.6, 77.0, 76.5, 74.7, 73.0, 72.7, 71.0, 68.2, 62.1, 61.3, 52.3, 48.2, 42.9, 42.4, 39.4, 38.5. HRMS (FAB) calculated mass for C31H38N2O15, 679.2350 [M + H]+ and was found at 679.2352 [M + H]+.
Preparation of N-α-Fmoc-L-Asn[Gal(β1-4)Glc]-O′Bu (3c)

To a 25ml round-bottom flask was added N-α-Fmoc-L-aspartic acid α-tert butyl ester (0.23g, 0.56mmol), anhydrous DMF (3ml), HOBt (76mg, 0.56mmol) and HBTU (0.53g, 1.41mmol). After stirring for 15 min, was added a solution of 2c (0.16g, 0.47mmol) in anhydrous DMF (3ml). The reaction mixture was stirred at room temperature under nitrogen atmosphere for 24hrs. The solvent was removed under reduced pressure. The crude product was purified by flash chromatography using 20% MeOH in CHCl₃ as an eluent to afford 3c as a white solid (0.24g, 70 %, melting point 170ºC dec). IR (KBr): 3397, 1698, 1539, 1428 cm⁻¹. ¹H NMR (360MHz, CD₃OD-d⁴) δ 7.87 (d, 2H, J= 7.5Hz), 7.71 (d, 2H, J= 7.5Hz), 7.43 (t, 2H, J= 7.4Hz), 7.38 (t, 2H, J= 7.4Hz), 4.93 (d, 1H, J= 9.2Hz), 4.51 (t, 1H, J= 6.7Hz), 4.40 (m, 2H), 4.28 (t, 1H, J= 6.7Hz), 3.84 (m, 5H), 3.55 (m, 6H), 3.34 (m, 2H), 2.79 (m, 2H), 1.49 (s, 9H). ¹³C NMR (360MHz, CD₃OD) δ 170.9, 170.1, 156.2, 143.3, 140.6, 127.0, 126.4, 124.5, 117.2, 103.2, 81.1, 78.9, 78.7, 76.2, 75.3, 75.2, 72.8, 71.6, 70.5, 68.3, 66.0 60.5, 59.8, 50.6, 37.9, 36.6, 26.3. HRMS MS (FAB) calculated mass for C₃₅H₄₆N₂O₁₅, 735.2976 [M + H]+ and was found at 735.2977 [M+H]+.

Preparation of N-α-Fmoc-L-Asn[Gal(β1-4)Glc]-OH (4c)

To a 50ml round-bottom flask was added 3c (0.15g, 0.2mmol), anhydrous DCM (6ml), TFA (6ml) and anisole (0.6ml). The reaction mixture was stirred at room
temperature under nitrogen atmosphere for 4hrs. The reaction mixture was concentrated under reduced pressure and azeotroped with toluene (2 × 20ml) and DCM (2 × 20ml). Finally the residue was triturated with ether to afford 4c as a white coloured solid (0.12g, 87%, 192°-194°C). IR (KBr): 3389, 2931, 1715, 1654, 1540, 1080 cm⁻¹. ¹H NMR (250MHz, DMSO-d⁶) δ 8.16 (d, 2H, J= 7.3Hz), 7.98 (d, 2H, J= 7.3Hz), 7.67 (t, 2H, J= 7.4Hz), 7.61 (t, 2H, J= 7.4Hz), 5.10 (t, 1H, J= 9.1Hz), 4.63 (m, 1H), 4.47 (m, 3H), 3.95 (m, 6H), 3.35 (m, 7H), 2.85 (m, 2H). HRMS (FAB) calculated mass for C₃₁H₃₈N₂O₁₅, 679.2350 [M + H]⁺ and was found at 679.2355 [M + H]⁺

**Preparation of N-α-Fmoc-L-Asn[GlcNAc(β1-4)GlcNAc]-O^Bu (3d)**

![3d Diagram]

To a 25ml round-bottom flask was added N-α-Fmoc-L-aspartic acid α-tert butyl ester (0.15g, 0.38mmol), anhydrous DMF (3ml), HOBt (50mg, 0.38mmol) and HBTU (0.21g, 0.57mmol. After stirring for 15 min, was added a solution of 2d (0.16g, 0.47mmol) in anhydrous DMF (3ml) and DIPEA (67µl, 0.38mmol). The reaction mixture was stirred at room temperature under nitrogen atmosphere for 24hrs. The solvent was removed under reduced pressure. The crude product was purified by flash chromatography using 23% MeOH in chloroform as an eluent to afford 3d as a white solid (0.106g, 70%). ¹H NMR (360MHz, DMSO-d⁶) δ 8.30 (d, 1H, J= 8.9Hz), 7.91 (d, 2H, J= 7.4Hz), 7.83 (d, 1H, J= 8.9Hz), 7.73 (d, 2H, J= 7.4Hz), 7.34 (t, 2H, J= 7.4Hz), 5.15 (d, 1H, J= 5.4Hz), 5.05 (d, 1H, J= 5.3Hz), 4.85 (t, 1H, J= 9.4Hz), 4.81 (d, 1H, J= 4.7Hz), 4.78 (t, 1H, J= 4.7Hz), 4.63 (t, 1H, J= 4.8Hz), 4.38 (m, 3H), 3.75 (m, 2H), 3.64 (M, 2H), 3.20 (d, 2H, J= 10.1Hz), 3.05 (m, 2H), 2.75 (m, 2H), 1.87 (s, 3H), 1.82 (s, 3H), 1.41 (s, 9H). ¹³C NMR (360MHz, DMSO-d⁶) δ 172.0, 171.1, 170.9,
170.6, 157.3, 145.2, 142.1, 129.1, 128.8, 128.5, 121.6, 104.3, 82.7, 80.6, 80.1, 78.4, 78.1, 75.4, 74.3, 72.1, 67.1, 62.4, 61.1, 58.9, 56.8, 52.2, 48.0, 38.4, 29.8, 24.5, 24.2.

MS (ESI) calculated mass for C_{39}H_{52}N_{4}O_{15}, m/z 816.3 and was found at 817.1 [M+H]^+.

**Preparation of N-α-Fmoc-L-Asn[GlcNAc(β1-4)GlcNAc]-OH (4d)**

To a 25ml round-bottom flask was added 3d (32mg, 0.04mmol), anhydrous DCM (2ml), TFA (2ml) and anisole (0.2ml). The reaction mixture was stirred at room temperature under nitrogen atmosphere for 4hrs. The reaction mixture was concentrated under reduced pressure and azeotroped with toluene (2 × 10ml) and DCM (2 × 10ml). Finally the residue was triturated with ether to afford 4d as a white coloured solid (26mg, 89%). ^1H NMR (360MHz, DMSO-d^6) δ 8.57 (d, 1H, J= 9.4Hz), 8.24 (d, 1H, J= 9.4Hz), 8.16 (bs, 1H, 7.88 (d, 2H, J= 7.3Hz), 7.74 (d, 1H, J= 9.2Hz), 7.71 (d, 2H, J= 7.3Hz), 7.46 (d, 1H, J= 8.32Hz), 7.41 (t, 2H, J= 7.42Hz), 7.32 (t, 2H, J= 7.10Hz), 4.83 (t, 1H, J= 9.3Hz), 4.25 (m, 4H), 3.76 (m, 2H), 3.52 (m, 6H), 3.22 (m, 2H), 3.13 (m, 3H), 2.83 (m, 2H), 1.85 (s, 3H), 1.81 (s, 3H). MS (ESI) calculated mass for C_{35}H_{44}N_{4}O_{15}, m/z 760.6 and was found at 783.6 [M+Na]^+.

**Preparation of N-α-Fmoc-L-Asn[Glc(α1-4)Glc(α1-4)Glc]-O'Bu (3e)**
To a 25ml round-bottom flask was added N-α-Fmoc-L-aspartic acid α-tert butyl ester (0.2g, 0.48mmol), anhydrous DMF (3ml), HOBt (64mg, 0.48mmol) and HBTU (0.27g, 0.71mmol). After stirring for 15 min, was added a solution of 2e (0.12g, 0.24mmol) in anhydrous DMF (3ml). The reaction mixture was stirred at room temperature under nitrogen atmosphere for 24hrs. The solvent was removed under reduced pressure. The crude product was purified by flash chromatography using 25% MeOH in CHCl₃ as an eluent to afford 3e as a white coloured crystalline solid (0.15g, 71%, melting point 126º-130ºC). ¹H NMR (360MHz, CD₃OD-d⁴) δ 7.68 (d, 2H, J= 7.4Hz), 7.55 (d, 2H, J= 7.4Hz), 7.28 (t, 2H, J= 7.3Hz), 7.20 (t, 2H, J= 7.3Hz), 5.08 (m, 2H), 4.80 (d, 1H, J= 13.6Hz), 4.39 (t, 1H, J= 6.0Hz), 4.26 (m, 2H), 4.12 (t, 1H, J= 12.9 Hz), 3.74 (m, 5H), 3.59 (m, 5H), 3.45 (m, 5H), 3.22 (m, 3H), 2.75 (m, 2H), 1.35 (s, 9H). ¹³C NMR (360MHz, CD₃OD-d⁴) δ 171.2, 170.14, 156.45, 143.2, 140.5, 126.8, 126.2, 124.3, 118.9, 100.9, 100.7, 81.2, 79.3, 79.0, 78.9, 76.7, 76.1, 73.1, 72.9, 72.8, 72.2, 71.7, 71.4, 71.3, 69.5, 66.2, 60.7, 60.1, 50.6, 47.3, 36.9, 26.2. MS (ESI) calculated mass for C₄₁H₅₆N₂O₂₀, m/z 896.8 and was found at 897.4 [M+H]⁺, 919.3 [M+Na]⁺.

**Preparation of N-α-Fmoc-L-Asn[Glc(α1-4) Glc(α1-4) Glc]-OH (4e)**

![Image of 4e](image)

To a 25ml round-bottom flask was added 3e (95mg, 0.11mmol), anhydrous DCM (4ml), TFA (4ml) and anisole (0.4ml). The reaction mixture was stirred at room temperature under nitrogen atmosphere for 4hrs. The reaction mixture was concentrated under reduced pressure and azeotroped with toluene (2 × 15ml) and...
DCM (2 × 15ml). Finally the residue was triturated with ether to afford 4e as a white coloured solid (79mg, 92%). IR (KBr): 3387, 2932, 1675, 1540, 1151 cm⁻¹. ¹H NMR (360MHz, CD₃OD-d₄) δ 7.81 (d, 2H, J= 7.4Hz), 7.68 (d, 2H, J= 7.4Hz), 7.41 (t, 2H, J= 7.3Hz), 7.33 (t, 2H, J= 7.3Hz), 5.20 (m, 2H), 4.83 (d, 1H, J= 10.6Hz), 4.62 (m, 4H), 4.32 (m, 3H), 3.84 (m, 5H), 3.71 (m, 4H), 3.56 (m, 5H), 3.35 (m, 4H), 2.87 (m, 2H).

¹³C NMR (360MHz, CD₃OD-d₄) δ 172.9, 171.4, 156.5, 143.2, 140.6, 126.8, 126.3, 124.4, 119.0, 100.9, 100.6, 79.1, 77.0, 76.7, 76.2, 73.1, 73.0, 72.8, 72.3, 71.8, 71.5, 71.4, 69.5, 66.3, 65.0, 60.7, 60.1, 49.9, 47.1, 36.6. HRMS (FAB) calculated mass for C₃₇H₄₈N₂O₂₀, 841.2879 [M + H]⁺ and was found at 841.2866 [M + H]⁺.

**Fmoc-SPPS of glycopeptide 7 and 8**

Glycopeptides 7 and 8 were synthesised using standard Fmoc-SPPS protocol. Peptides were assembled on Wang resin (0.82mmol/gram), and the first amino acid was coupled using symmetrical anhydride method. The loading was calculated using UV detection of the Fmoc deprotection (0.56mmol/gram).

The remaining SPPS including coupling of glycosylated asparagine was performed using standard coupling conditions (Fmoc-amino acid (4eq), TBTU (4eq), HOBT (4eq), DIPEA (8eq), and DMF (2ml) for 4hrs followed Fmoc deprotection (20% piperidine in DMF for 1hr). The glyco-amino acids were incorporated into peptide sequence, using double coupling. Finally the resin and side chain protecting groups were removed by using a mixture (3ml) of TFA (94%), H₂O (2.5%), EDT (2.5%) and TIS (1%) for 4hrs, and the resin was filtered off and the filtrate was concentrated under reduced pressure followed by precipitation with cold ether. The glycopeptides 7 and 8 were dissolved separately in two round bottom flasks using (MeCN: H₂O: TFA (1:1:0.1%) and were lyophilised to afford white solids, glycopeptides 7 (19mg, 67%) and 8 (17mg, 54%). These glyco peptides were analysed by LC-MS, using electrospray ionisation technique and HRMS (FAB) Calculated mass for
C\textsubscript{35}H\textsubscript{57}N\textsubscript{13}O\textsubscript{16}S (glycopeptide 7), 948.3845 \text{[M + H]}^+ and was found at 948.3849 \text{[M + H]}^+. Calculated mass for C\textsubscript{30}H\textsubscript{64}N\textsubscript{12}O\textsubscript{21}S (glycopeptide 8), 1069.4108 \text{[M + H]}^+ and was found at 1069.4106 \text{[M + H]}^+. 
Glycopeptide 7

Calculated m/z 947.9 amu
Glycopeptide 7

Structural formula:

-CO-NH-Asp-Arg-His-Thr-Asp-COOH

Calculated mass 907.9 amu
BM-CELLOBIOSE-NEW 115 (4,213) Cm (105:125)

H₂N-Cys-Asp-Arg-NH→CO-His-Thr-COOH

Calculated mass 1050.6 amu

Glycopeptide 8