Scalable Synthesis of Fmoc-Protected GalNAc-Threonine Amino Acid and \( T_N \) Antigen via Nickel Catalysis

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Supporting Information

**Methods and Reagents.** All reactions were performed in oven-dried Schlenk flasks fitted with glass stoppers under a positive pressure of argon. Organic solutions were concentrated by rotary evaporation below 40 °C at 25 torr. Analytical thin-layer chromatography (TLC) was routinely used to monitor the progress of the reactions and performed using pre-coated glass plates with 230-400 mesh silica gel impregnated with a fluorescent indicator (250 nm). Visualization was achieved using UV light, iodine, or ceric ammonium molybdate. Flash chromatography was performed and employed 230-400 mesh silica gel. Dichloromethane was distilled from calcium hydride under an argon atmosphere at 760 torr. All other chemicals were obtained from commercial vendors and used without further purification.

**Instrumentation.** All proton (\(^1\)H) nuclear magnetic resonance spectra were recorded on 300, 400, and 500 MHz spectrometers. All carbon (\(^1\)C) nuclear magnetic resonance spectra were recorded on 75, 100, 125, 150 MHz NMR spectrometer. Chemical shifts are expressed in parts per million (δ scale) downfield from tetramethylsilane and are referenced to the residual proton in the NMR solvent (CDCl₃: δ 7.27 ppm, δ 77.23 ppm; DMSO-d₆: δ 2.50 ppm, δ 39.52 ppm; D₂O: δ 4.79 ppm). Data are presented as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, and bs = broad singlet), integration, and coupling constant in hertz (Hz). Infrared (IR) spectra were reported in cm\(^{-1}\). High resolution (ESI) mass spectrometry was performed to identify the purity of the compounds.

**Preparation of Ni Catalyst – Ni(F-Ph-CN)₄Cl₂**

A 100 mL Schlenk flask was charged with anhydrous NiCl₂ (1.0 g, 7.71 mmol, 1.0 equiv), 4-fluorobenzonitrile (25.0 g, 206.6 mmol, 26.8 equiv), and anhydrous dichloromethane (10 mL). The reaction mixture was stirred vigorously at room temperature 48 h, transferred to a flask containing hexanes (150 mL), and stirred for 30 min. The mixture was then left unperturbed for 10 min, and the colorless supernatant liquid was removed. Hexanes (150 mL) was added to the precipitate and stirred for 5 min. The mixture was then left unperturbed for 10 min and the colorless supernatant liquid was removed. This process was repeated one more time. Finally the yellow precipitate was dried under high vacuum at 100 °C for 24 h (456.7 mg).

**Synthesis of threonine acceptor 13**

A 100 mL oven-dried Schlenk flask was charged with Fmoc-Thr-OH (3 g, 8.788 mmol, 1.0 equiv), allyl bromide (1.50 mL, 17.577 mmol, 2.00 equiv), DIPEA (3.10
mL, 17.577 mmol, 2.00 equiv), and anhydrous DMF (35 mL). The resulting mixture was stirred at room temperature overnight. The reaction mixture was diluted with ethyl acetate (150 mL) and washed with brine (4 x 40 mL). The organic layer was dried with anhydrous sodium sulfate and concentrated to a white solid. The crude product was purified by flash chromatography on silica gel (hexane/ethyl acetate = 2/1) to afford 13 as a white solid (2.70 g, 80%).

^1H NMR (CDCl₃, 300 MHz): δ = 7.79 – 7.30 (m, 8H), 5.99 – 5.86 (m, 2H), 5.35 (d, J = 17.1 Hz, 1H), 5.26 (d, J = 11.7 Hz, 1H), 4.69 (d, J = 5.7 Hz, 2H), 4.46 – 4.40 (m, 4H), 4.28 – 4.24 (m, 1H), 2.74 (bs, 1H), 1.28 (d, J = 6.3 Hz, 3H).

The ^1H NMR Spectrum of 13 is matched with previously published results.¹

Synthesis of galactosamine imidate donor 16 (Large Scale)

The former synthetic route for making galactosamine donor 16 was deemed to inefficient for large scale preparation.² A more streamlined synthetic route is shown below.

C(2)-N-ortho-Trifluoromethylbenzylideneamino-D-Galactosaminepyranoside A. An oven dried 1 L round bottom flask was charged with D-Galactosamine hydrochloride (23.0 g, 1067 mmol, 1 equiv), 2-(trifluoromethyl)-benzaldehyde (42.2 mL, 320.0 mmol, 3.0 equiv), anhydrous pyridine (213 mL), and triethylamine (22.3 mL, 160.05 mmol, 1.5 equiv). The resulting mixture was stirred at 60 °C for 24 h. The reaction mixture was cooled to room temperature, and acetic anhydride (81.3 mL, 853.6 mmol, 8.0 equiv) was added. The reaction mixture was then stirred at room temperature overnight and concentrated to a dark oil. The crude product was purified by flash chromatography on silica gel (hexane/ethyl acetate = 5/1 → 3/1 with 1% Et₃N) to afford A as a white solid (12.55 g, 55%).

^1H NMR (CDCl₃, 300 MHz): δ = 8.67 (d, J = 2.1 Hz, 1H), 8.06 (d, J = 7.2 Hz, 1H), 7.70 – 7.67 (m, 1H), 7.61 – 7.51 (m, 2H), 5.98 (d, J = 8.1 Hz, 1H), 5.46 (d, J = 3.3 Hz, 1H), 5.30 (dd, J = 10.5, 3.6 Hz, 1H), 4.23 – 4.15 (m, 3H), 3.72 (dd, J = 10.5, 8.4 Hz, 1H), 2.19 (s, 3H), 2.05 (s, 3H), 2.04 (s, 3H), 1.91 (s, 3H).

Galactosaminepyranosyl N-Phenyl Trifluoroacetimidate 16. A 500 mL round bottom flask was charged with A (12.55 g, 24.36 mmol, 1 equiv) and anhydrous THF (125 mL). The solution was cooled to 0 °C, ammonia in methanol solution (7 N, 53.4 mL, 373.96 mmol, 15.0 equiv) was added. The resulting mixture was stirred at room temperature for 2.5 h and monitored by TLC. The round bottom flask was unsealed, and the reaction mixture was stirred under positive air flow for 1 h. The reaction mixture was concentrated to a yellow oil, and the crude hemiacetal used in the next step without further purification. A 250 mL round bottom flask was charged with the yellow-oil hemiacetal, 2,2,2-trifluoro-N-phenyl-ethanimidoyl chloride (4.4 mL, 27.3 mmol, 1.1 equiv), diazobicycloundecane (DBU) (2.0 mL, 13.65 mmol, 0.5 equiv) and anhydrous dichloromethane (53 mL). The resulting solution was stirred at room temperature overnight. When the reaction mixture was complete as monitored by TLC (hexane/ethyl acetate = 1/1), the reaction mixture was evaporated and purified by flash chromatography on silica gel (hexane/ethyl acetate = 5/1 → 3/1 with 1% Et3N) to afford 16 as a white solid (6.30 g, 40%; α:β = 1:2).

1H NMR (CDCl3, 300 MHz): δ = 8.73 (d, J = 2.4 Hz, 1H), 8.12 (d, J = 7.5 Hz, 1H), 7.73 (d, J = 6.9 Hz, 1H), 7.67 – 7.56 (m, 2H), 7.31 (t, J = 7.5 Hz, 2H), 7.13 (t, J = 7.2 Hz, 1H), 6.82 (d, J = 7.8 Hz, 2H), 6.73 (d, J = 7.8 Hz, 0.25H), 6.02 (bs, 0.75H), 5.45 (d, J = 3.0 Hz, 1H), 5.28 (bs, 1H), 4.22 (d, J = 6.0 Hz, 2H), 4.06 (bs, 1H), 3.85 (t, J = 9.3 Hz, 1H), 2.24 (s, 3H), 2.06 (s, 3H), 1.94 (s, 3H).


General Glycosylation Procedure Using Ni(4-F-PhCN)₄(OTf)₂ as the Catalyst (Batch A and Batch B)

Batch A:
A 100 mL oven-dried Schlenk flask was charged with Ni(4-F-PhCN)₄Cl₂ (78.6 mg, 0.128 mmol, 10 mol%) and AgOTf (65.8 mg, 0.256 mmol, 20 mol%) in dichloromethane (4 mL). The resulting mixture was stirred at room temperature for 30 min. A solution of D-galactosamine N-phenyl trifluoroacetimidate donor 16 (808 mg, 1.278 mmol, 1.0 equiv), threonine acceptor 13 (585 mg, 1.534 mmol, 1.20 equiv), and CH₂Cl₂ (4.5 mL) was added. The resulting mixture was stirred under argon at 35 °C overnight. The reaction mixture was filtered through Celite and then purified by silica
gel flash chromatography (3/1, hexanes/ethyl acetate + 1% triethylamine) to give the desired glycosyl amino acid 15 (783 mg, 74%, α only).

**1H NMR (CDCl3, 300 MHz):** δ = 8.68 (d, J = 2.1 Hz, 1H), 8.39 (d, J = 2.4 Hz, 1H), 7.79 – 7.60 (m, 9H), 7.49 – 7.30 (m, 10H), 6.21 (d, J = 9.0 Hz, 1H), 6.10 (s, 1H), 5.80 – 5.68 (m, 1H), 5.53 – 5.47 (m, 2H), 5.21 – 5.13 (m, 2H), 5.03 (d, J = 3.3 Hz, 1H), 4.53 – 4.40 (m, 6H), 4.34 – 4.24 (m, 2H), 4.19 (d, J = 6.6 Hz, 2H), 3.86 (dd, J = 10.5, 3.9 Hz, 1H), 2.21 (s, 3H), 2.10 (s, 3H), 1.91 (s, 3H), 1.44 (d, J = 6.3 Hz, 3H).

**13C NMR (CDCl3, 100 MHz):** δ = 170.4, 170.2, 170.2, 161.9, 156.8, 143.9, 143.8, 141.2, 132.2, 131.2, 130.7, 129.1, 128.7, 127.7, 127.1, 125.3 (q, J_{C-F} = 5.4 Hz), 121.0, 119.9, 119.0, 100.0, 77.2, 75.3, 68.2, 68.0, 67.6, 67.2, 67.0, 66.0, 62.1, 58.8, 47.2, 20.8, 20.7, 20.4, 19.3.

**IR (film, cm⁻¹):** ν = 3154, 2982, 2892, 1746, 1471, 1378, 1315, 1245, 1173, 1095, 911.

**HRMS (ESI):** calc. for C₄₂H₄₄N₂O₁₂F₃ (M+H): 825.2846; found: 825.2850.

**Batch B:**

A 100 mL oven-dried Schlenk flask was charged with Ni(4-F-PhCN)₄Cl₂ (92.0 mg, 0.15 mmol, 10 mol%) and AgOTf (77.0 mg, 0.30 mmol, 20 mol%) in dichloromethane (5 mL). The resulting mixture was stirred at room temperature for 30 min. A solution of D-galactosamine \( N \)-phenyl trifluoroacetimidate donor 16 (947 mg, 1.5 mmol, 1.0 equiv), threonine acceptor 13 (685 mg, 1.8 mmol, 1.20 equiv), and CH₂Cl₂ (5 mL) was added. The resulting mixture was stirred under argon at 35 °C overnight. The reaction mixture was filtered through Celite and then purified by silica gel flash chromatography* (3/1, hexanes/ethyl acetate + 1% triethylamine) to give the desired glycosyl amino acid 15 (779 mg, 63%, α only).

* We first tried to separate the crude product 15 from unreacted threonine residue 13 by automated chromatography on a Teledyne Isco CombiFlash Rf system utilizing normal phase pre-column cartridges and gold high performance columns. However, it was not successful. Thus, the product 15 was then purified by manual silica gel flash chromatography.

**General Glycosylation Procedure Using Ni(4-F-PhCN)₄(OTf)₂ as the Catalyst (Batch C)**

![Diagram of the general glycosylation procedure](attachment:diagram.png)
A 100 mL oven-dried Schlenk flask was charged with Ni(4-F-PhCN)₄Cl₂ (434.7 mg, 0.71 mmol, 10 mol%) and AgOTf (363.8 mg, 1.42 mmol, 20 mol%) in dichloromethane (23 mL). The resulting mixture was stirred at room temperature for 30 min. A solution of D-galactosamine N-phenyl trifluoroacetimidate donor 16 (5.73 g, 9.059 mmol, 1.28 equiv), threonine acceptor 13 (2.7 g, 7.078 mmol, 1.0 equiv), and CH₂Cl₂ (24 mL) was added. The resulting mixture was stirred under argon at 35 °C overnight. The reaction mixture was filtered through Celite and then purified by silica gel flash chromatography (3/1, hexanes/ethyl acetate + 1% triethylamine) to give the desired glycosyl amino acid 15 (3.77 g, 66%, α only).

**General Glycosylation Procedure Using Ni(4-F-PhCN)$_4$(OTf)$_2$ as the Catalyst (Small Scale)**

![Chemical Diagram]

A 10 mL oven-dried Schlenk flask was charged with D-galactosamine N-phenyl trifluoroacetimidate donor 16 (93.0 mg, 0.147 mmol, 1.0 equiv), Z-Thr-OBzl (60.6 mg, 0.176 mmol, 1.2 equiv), and CH₂Cl₂ (0.5 mL). A preformed solution of Ni(4-F-PhCN)₄(OTf)₂, which was generated in situ from Ni(4-F-PhCN)₄Cl₂ (9.0 mg, 0.0147 mmol, 10 mol%) and AgOTf (7.6 mg, 0.0294 mmol, 20 mol%) in dichloromethane (0.5 mL) was then added to the solution. The resulting mixture was stirred under argon at 35 °C overnight, and purified by silica gel flash chromatography (3/1, hexanes/ethyl acetate + 1% triethylamine) to give the desired disaccharide 21 (76.0 mg, 67%, α only).

**$^1$H NMR (CDCl₃, 500 MHz):** δ = 8.56 (s, 1H), 8.25 (d, $J = 7.5$ Hz, 1H), 7.63 (d, $J = 8.0$ Hz, 1H), 7.42 – 7.15 (m, 10H), 6.09 (d, $J = 9.0$ Hz, 1H), 5.47 (d, $J = 2.0$ Hz, 1H), 5.38 (dd, $J = 10.5$, 3.0 Hz, 1H), 5.21 – 5.13 (m, 2H), 5.01 – 4.89 (m, 2H), 4.83 (d, $J = 4.0$ Hz, 1H), 4.51 (dd, $J = 12.5$, 6.5 Hz, 1H), 4.40 (d, $J = 7.0$ Hz, 2H), 4.15 (d, $J = 7.0$ Hz, 2H), 3.70 (dd, $J = 15.0$, 3.5 Hz, 1H), 2.20 (s, 3H), 2.07 (s, 3H), 1.87 (s, 3H), 1.39 (d, $J = 6.5$ Hz, 3H).

**$^{13}$C NMR (CDCl₃, 100 MHz):** δ = 170.4, 170.1, 169.9, 161.7, 156.9, 136.3, 135.0, 133.1, 132.2, 130.6, 129.0, 129.0, 128.7, 128.5, 128.3, 128.2, 128.1, 125.5, 125.3 (q, $J_{C,F} = 5.4$ Hz), 122.8, 99.6, 74.9, 68.1, 67.7, 67.7, 67.1, 66.9, 62.1, 58.7, 53.4, 20.7, 20.7, 20.3, 19.4.

**IR (film, cm$^{-1}$):** ν = 3432, 3342, 3064, 3031, 2925, 2251, 1728, 1638, 1495, 1376, 1315, 1229, 1166, 1066, 1017.

**HRMS (ESI):** calc. for C$_{39}$H$_{42}$N$_2$O$_{12}$F$_3$ (M+H): 787.2690; found: 787.2709.
$^1$H NMR (CDCl$_3$, 400 MHz): $\delta = 8.60$ (s, 1 H), 8.37-8.26 (m, 2 H), 7.82-7.18 (m, 15 H), 6.30 (d, $J = 8.8$ Hz, 1 H), 5.51-5.42 (m, 2 H), 5.00 (d, $J = 12.0$ Hz, 1 H), 4.91 (d, $J = 12.$ Hz, 1 H), 4.87 (d, $J = 2.4$ Hz, 1 H), 4.55-4.37 (m, 4 H), 4.30 (t, $J = 7.2$ Hz, 1 H), 4.22 (d, $J = 7.2$ Hz, 1 H), 4.15 (d, $J = 6.4$ Hz, 2 H), 3.74 (dd, $J = 3.2$, 10.4 Hz, 1 H), 2.20 (s, 3 H), 2.06 (s, 3 H), 1.89 (s, 3 H), 1.40 (d, $J = 6.4$ Hz, 3 H).

$^{13}$C NMR (CDCl$_3$, 100 MHz): $\delta = 170.5$, 170.3, 170.2, 170.0, 161.9, 156.9, 155.0, 143.81, 143.75, 141.2, 135.3, 130.7, 129.3, 128.3, 128.4, 128.3, 127.67, 127.66, 127.1, 126.2, 125.24, 125.19, 120.6, 119.9, 99.9, 75.3, 68.1, 67.9, 67.6, 67.2, 67.1, 66.9, 62.1, 58.8, 47.1, 20.7, 20.6, 20.3, 19.2.

IR (film, cm$^{-1}$): $\nu = 3323, 1747, 1641, 1314.$

HRMS (ESI): calc. for C$_{46}$H$_{46}$F$_3$N$_2$O$_{12}$ (M+H): 875.3003; found: 875.3026.

$^1$H NMR (CDCl$_3$, 400 MHz): $\delta = 8.67$ (s, 1 H), 8.36 (d, $J = 7.6$ Hz, 1 H), 7.82-7.27 (m, 11 H), 6.37 (d, $J = 8.8$ Hz, 1 H), 5.5-5.44 (m, 2 H), 5.07 (d, $J = 2.4$ Hz, 1 H), 4.58-4.40 (m, 6 H), 4.328-4.25 (m, 2 H), 4.19 (d, $J = 6.4$ Hz, 2 H), 3.85 (dd, $J = 3.6$, 10.4 Hz, 1 H), 2.40 (s, 1 H), 2.21 (s, 3 H), 2.09 (s, 3 H), 1.90 (s, 3 H), 1.45 (d, $J = 6.4$ Hz, 3 H).

$^{13}$C NMR (CDCl$_3$, 100 MHz): $\delta = 170.4$, 170.1, 169.9, 169.7, 162.1, 156.8, 143.8, 143.7, 141.1, 133.1, 132.2, 130.7, 129.0, 127.63, 127.61, 127.0, 125.5 (q, $J_{C-F} = 5.4$ Hz), 125.22, 125.18, 119.9, 100.0, 75.5, 75.4, 68.0, 67.5, 67.1, 66.9, 62.1, 58.7, 52.6, 47.0, 20.73, 20.65, 20.3, 19.1.

IR (film, cm$^{-1}$): $\nu = 3328, 1738, 1643, 1316.$

HRMS (ESI): calc. for C$_{42}$H$_{42}$F$_3$N$_2$O$_{12}$ (M+H): 823.2690; found: 823.2705.
1H NMR (CDCl₃, 400 MHz): δ = 8.78 (s, 1 H), 8.41 (d, J = 6.8 Hz, 1 H), 7.82-7.22 (m, 21 H), 6.38 (d, J = 8.8 Hz, 1 H), 5.76-5.66 (m, 1 H), 5.18-5.07 (m, 2 H), 4.99 (d, J = 11.6 Hz, 1 H), 4.93 (d, J = 3.2 Hz, 1 H), 4.69-4.55 (m, 3 H), 4.38-4.02 (m, 14 H), 3.96 (s, 1 H), 2.04 (s, 3 H), 1.39 (d, J = 6.4 Hz, 3 H).

13C NMR (CDCl₃, 100 MHz): δ = 170.6, 170.1, 161.3, 156.8, 143.8, 141.2, 138.1, 138.0, 132.0, 131.2, 130.4, 128.7, 128.40, 128.37, 128.2, 127.8, 127.7, 126, 127.4, 127.0, 125.5 (q, J_C-F = 5.4 Hz), 125.2, 119.9, 118.9, 100.6, 75.1, 74.6, 73.2, 72.4, 70.8, 69.4, 67.3, 65.9, 63.9, 58.9, 47.2, 20.8, 19.2.

IR (film, cm⁻¹): ν = 3341, 1727, 1313.

HRMS (ESI): calc. for C₂₅H₂₅F₃N₂O₁₀ (M+H): 921.3574; found: 921.3590.

1H NMR (CDCl₃, 400 MHz): δ = 8.76 (d, J = 1.6 Hz, 1 H), 8.42 (d, J = 6.4 Hz, 1 H), 7.83-7.17 (m, 26 H), 6.39 (d, J = 7.2 Hz, 1 H), 5.05-4.85 (m, 4 H), 4.62-3.92 (m, 14 H), 2.04 (s, 3 H), 1.37 (d, J = 6.0 Hz, 3 H).

13C NMR (CDCl₃, 100 MHz): δ = 170.6, 170.2, 161.3, 156.8, 143.84, 143.77, 141.2, 138.1, 138.0, 134.9, 133.5, 131.9, 130.4, 128.8, 128.7, 128.6, 128.5, 128.40, 128.37, 128.24, 128.19, 127.8, 127.7, 127.6, 127.5, 127.4, 127.0, 125.5 (q, J_C-F = 5.4 Hz), 119.9, 100.7, 75.2, 74.6, 73.1, 72.4, 70.6, 69.4, 67.3, 67.1, 65.9, 63.9, 59.0, 47.2, 20.8, 19.1.

IR (film, cm⁻¹): ν = 3342, 1727, 1313.

HRMS (ESI): calc. for C₂₅H₂₅F₃N₂O₁₀ (M+H): 971.3731; found: 971.3756.
\[^1\text{H} \text{NMR (CDCl}_3, 500 \text{ MHz)}: \delta = 8.13 (d, J = 8.0 \text{ Hz, 1H}), 8.04 (d, J = 8.0 \text{ Hz, 1.5H}), 7.92 - 7.81 (m, 4H), 7.75 - 7.65 (m, 5H), 6.25 (d, J = 8.0 \text{ Hz, 0.5H}), 6.10 (d, J = 3.5 \text{ Hz, 1H}), 5.62 (dd, J = 10, 3.0 \text{ Hz, 1H}), 5.60 - 5.54 (m, 2H), 5.50 (s, 1H), 5.47 (bs, 1H), 5.44 (bs, 1.5H), 5.14 (d, J = 1.0 \text{ Hz, 1H}), 4.71 (t, J = 6.5 \text{ Hz, 1H}), 4.55 - 4.52 (m, 2.5H), 4.31 - 4.08 (m, 9H), 3.77 (t, J = 7.0 \text{ Hz, 2H}), 3.14 (d, J = 6.0 \text{ Hz, 2H}), 2.18 (s, 3H), 2.15 (s, 3H), 2.11 (s, 1.5H), 2.10 (s, 1.5H), 2.09 (s, 3H), 1.94 (s, 1.5H), 1.92 (s, 3H), 1.46 - 1.43 (m, 4.5H).\]

\[^{13}\text{C} \text{NMR (CDCl}_3, 100 \text{ MHz)}: \delta = 189.2, 189.0, 183.6, 169.8, 163.7, 161.8, 161.1, 156.6, 155.4, 134.4, 131.1, 130.3, 129.9, 129.9, 128.9, 128.7, 128.5, 126.1, 125.7, 122.7, 119.1, 100.0, 99.5, 93.8, 90.1, 88.5, 86.1, 85.4, 69.8, 68.4, 67.6, 66.5, 61.3, 57.2, 55.1, 53.4, 51.1, 50.2, 42.6, 30.9, 29.7, 26.4, 22.7, 21.4, 20.7.\]

\[\text{IR (film, cm}^{-1}\rfloor: \nu = 3156, 2992, 2896, 1747, 1474, 1377, 1315, 1243, 1171, 1095, 909.\]

\[\text{HRMS (ESI): calc. for C}_{42}\text{H}_{44}\text{N}_{2}\text{O}_{12}\text{F}_{3} \text{(M+H)}: 825.2846; found: 825.2864.}\]

An oven dried 250 mL round bottom flask was charged with 15 (3.77 g, 4.57 mmol, 1 equiv), anhydrous methanol (57 mL), and acetyl chloride (520 μL, 7.32 mmol, 1.6 equiv). The resulting mixture was stirred at room temperature overnight. Pyridine (40 mL) was added, and the reaction mixture was concentrated to a yellow oil. The yellow oil was then placed under high vacuum for 2 h to remove trace methanol. The yellow oil was dissolved in anhydrous pyridine (46 mL). Acetic anhydride (3.5 mL, 36.56 mmol, 8.0 equiv) was added to the solution. The reaction mixture was stirred at room temperature overnight and then concentrated to afford crude 22. The resulting residue was purified by silica gel flash chromatography (1/1 hexane/ethyl acetate → 1/2 hexane/ethyl acetate) to provide pure 22 (2.25 g, 70%) as a yellow solid.

\[^1\text{H} \text{NMR (CDCl}_3, 400 \text{ MHz)}: \delta = 7.79 (d, J = 7.2 \text{ Hz, 2H}), 7.65 (d, J = 7.6 \text{ Hz, 2H}), 7.44 - 7.33 (m, 4H), 5.94 - 5.84 (m, 1H), 5.77 (d, J = 10.4 \text{ Hz, 1H}), 5.58 (d, J = 9.6 \text{ Hz, 1H}), 5.41 - 5.30 (m, 4H), 5.10 (dd, J = 11.6, 3.2 \text{ Hz, 1H}), 4.90 (d, J = 3.2 \text{ Hz, 1H}), 4.69 (dd, J = 13.2, 7.2 \text{ Hz, 1H}), 4.62 - 4.56 (m, 3H), 4.46 (dd, J = 7.6, 4.0 \text{ Hz, 2H}), 4.31 - 4.22 (m, 3H), 4.16 - 4.07 (m, 3H), 2.18 (s, 3H), 2.06 (s, 3H), 2.02 (s, 6H), 1.35 (d, J = 6.4 \text{ Hz, 3H}).\]

\[^{13}\text{C} \text{NMR (CDCl}_3, 100 \text{ MHz)}: \delta = 170.9, 170.6, 170.3, 170.3, 156.5, 143.8, 143.6, 141.3, 130.8, 127.8, 127.1, 125.1, 125.1, 120.1, 120.0, 100.1, 68.4, 67.5, 67.3, 66.4, 62.1, 58.5, 53.4, 47.5, 47.1, 29.7, 23.2, 20.7, 20.6, 18.2.\]

\[\text{IR (film, cm}^{-1}\rfloor: \nu = 3432, 3145, 2933, 2843, 2251, 1744, 1670, 1499, 1372, 1229, 1037.\]

\[\text{HRMS (ESI): calc. for C}_{36}\text{H}_{44}\text{N}_{2}\text{O}_{12} \text{(M+H)}: 711.2765; found: 711.2771.}\]
A 250 mL oven-dried round bottom flask was charged with glycosyl amino acid 22 (1.12 g, 1.576 mmol, 1 equiv) and anhydrous tetrahydrofuran (44 mL). The solution was cooled to 0 °C, N-methylaniline (838.8 µL, 7.75 mmol, 4.92 equiv) and tetrakis(triphenylphosphine)palladium (179 mg, 0.155 mmol, 0.098 equiv) were then added. The resulting mixture was stirred at room temperature while being monitored by TLC. After 1 h, the reaction mixture was concentrated to a dark oil and purified by silica gel flash chromatography (9/1, dichloromethane/methanol) to provide pure 8 (1.02 g, 97%) as an off-white solid.

$^1\text{H NMR (CDCl}_3, 500 \text{ MHz)}$: $\delta = 7.78 - 7.31 \text{ (m, 10H)}, 5.40 \text{ (s, 1H)}, 5.31 - 5.26 \text{ (m, 1H)}, 5.15 \text{ (d, } J = 7.5 \text{ Hz, 1H)}, 5.02 \text{ (d, } J = 27 \text{ Hz, 1H)}, 4.59 - 4.42 \text{ (m, 4H)}, 4.29 - 4.25 \text{ (m, 3H)}, 4.20 \text{ (d, } J = 5.0 \text{ Hz, 1H)}, 4.14 - 4.09 \text{ (m, 3H)}, 2.18 \text{ (s, 3H)}, 2.05 \text{ (s, 3H)}, 2.01 \text{ (s, 3H)}, 1.99 \text{ (s, 3H)}, 1.29 \text{ (d, } J = 13 \text{ Hz, 3H}).$

The $^1\text{H NMR spectrum of 8 matches the result obtained by Sigma-Aldrich website.}$

An oven dried 250 mL round bottom flask was charged with 15 (1.50 g, 1.82 mmol, 1 equiv), anhydrous methanol (23 mL), and acetyl chloride (207 µL, 2.91 mmol, 1.6 equiv). The reaction mixture was stirred at room temperature overnight. Pyridine (20 mL) was added, and the reaction mixture was concentrated to a yellow oil. The yellow oil was then placed under high vacuum for 2 h to remove trace methanol. The yellow oil was dissolved in anhydrous pyridine (20 mL). Acetic anhydride (1.39 mL, 14.6 mmol, 8.0 equiv) was added to the reaction mixture. The reaction mixture was stirred at room temperature overnight. The reaction mixture was concentrated to afford crude 22. The resulting residue was purified by silica gel flash chromatography (1/1 hexane/ethyl acetate $\rightarrow$ 1/2 hexane/ethyl acetate) to provide pure 22 (1.18 g, 91%) as a yellow solid.
An oven dried 100 mL round bottom flask was charged with 22 (1.18 g, 1.67 mmol, 1 equiv), sodium hydroxide (0.2 M in methanol) (33.4 mL, 6.680 mmol, 4.00 equiv), and triethylamine (1.16 mL, 8.350 mmol, 5.00 equiv). The resulting mixture was stirred at room temperature overnight. The reaction mixture was concentrated to a yellow oil. The yellow oil was washed with dichloromethane (3 x 5 mL). The residue was redissolved in water and brought to neutral pH using Amberlyst 15 hydrogen form. The aqueous solution was decanted and lyophilized to a white solid 1 (0.51 g, 99%).

\(^1\)H NMR (CDCl\textsubscript{3}, 300 MHz): \(\delta = 4.85 \text{ (d, } J = 3.9 \text{ Hz, } 1\text{H}), 4.32 \text{ (dd, } J = 6.9, 2.1 \text{ Hz, } 1\text{H}), 4.02 \text{ (dd, } J = 7.5, 3.6 \text{ Hz, } 1\text{H}), 3.94 \text{ (t, } J = 6.3 \text{ Hz, } 1\text{H}), 3.87 \text{ (d, } J = 2.7 \text{ Hz, } 1\text{H}), 3.76 \text{ (dd, } J = 8.1, 3.0 \text{ Hz, } 1\text{H}), 3.66 – 3.61 \text{ (m, } 3\text{H}) 1.95 \text{ (s, } 3\text{H}), 1.30 \text{ (d, } J = 6.6 \text{ Hz, } 1\text{H}).

The \(^1\)H NMR Spectrum of 1 is matched with previously published results.iii

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† The authors contributed equally to the present work
1.2 1.3 1.4 1.5 1.6 1.7 1.8 1.9 2.0 2.1 2.2 2.3 2.4 2.5 2.6 2.7 2.8 2.9 3.0 3.1 3.2 3.3 3.4 3.5 3.6 3.7 3.8 3.9 4.0 4.1 4.2 4.3 4.4 4.5 4.6 4.7 4.8 4.9 5.0

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