SUPPORTING INFORMATION

Glycan based detection and drug susceptibility of influenza virus

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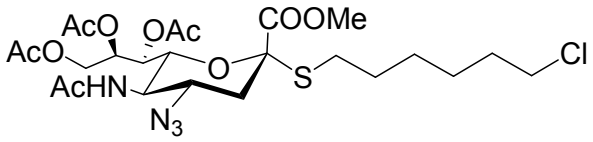
General information

All reagents and solvents were reagent grade or were purified by standard methods before use. Column chromatography was carried out on flash silica gel (Sorbent 230–400 mesh). TLC analysis was conducted on silica gel plates (Sorbent Silica G UV254). NMR spectra were recorded at 1H (400 MHz) and 13C (100 MHz) on a Bruker instrument. Chemical shifts and coupling constants (*J* values) are given in ppm and hertz, respectively, using solvents (1H NMR, 13C NMR) as the internal standard. All reactions were performed under argon atmosphere using degassed solvents.

Abbreviations: N,N Dimethyl formamide, DMF; Ethyl acetate, EtOAc; Trifluroacetic acid, TFA; Acetonitrile, CH_3CN; Azidotrimethylsilane, $TMSN_3$; Diethylamine, DEA; Trimethylsilyltrifluoromethanesulfonate, TMSOTf; p-Toluene sulfonyl, Tos; Di-tert-butyl dicarbonate, $(Boc)_2O$; tert-Butyl alcohol, t-BuOH; Methanesulfonyl chloride, MsCl; Methanol, MeOH; Tetrahydrofuran, THF; Dichloromethane, DCM; Hydrochloric acid, HCl; Triphenylphosphine, PPh_3; Sodium sulfate, Na_2SO_4; Sodium azide, NaN_3; N-methyl morpholine, NMM; 2-Chloro-4,6-DiMethoxy-1,3,5-Triazine, CDMT; Sodium methoxide, NaOMe; Cupric sulfate, $CuSO_4$; Triethylamine, Et_3N; De-ionized water, DI water; Hydrogen gas, H_2; water, H_2O; Triethylamine, Et_3N; Mercury(II) chloride, $HgCl_2$; Dimethylamino pyridine, DMAP; Sodium bicarbonate, $NaHCO_3$; Acetonitrile, CH_3CN.

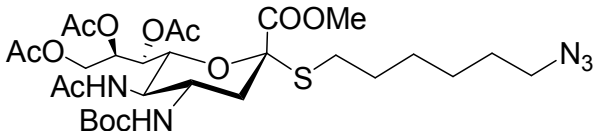
Compound 2: 6-*S*-[Methyl 5-acetamido-7, 8, 9-tri-O- acetyl-4-azido-3, 4, 5-trideoxy -D-glycero-α-D-galacto-non-2-ulopyranosyl) onate]-1-chloro-hexane.



To a stirring solution of compound **1**[1] (0.50 g, 0.94 mmol) in DMF (1.0 ml), DEA (0.95 ml, 9.4 mmol) was added. The reaction mixture was stirred for 10 min at rt. 6-chlorohexyl 4-methylbenzenesulfonate (0.35 g, 1.1 mmol) was added and stirred for 12 h. The reaction mixture was quenched using brine solution (0.20 L, 1x) and extracted with ethyl acetate

(0.050 L, 3x). Organic layers were combined and washed with HCl solution (0.05 ml, 1M, 1x), dried over Na_2SO_4 and concentrated *in vacuo*. The title compound was purified using column chromatography with hexane:acetone (3:1) as eluent to yield a yellow oil (0.46 g, 80% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.57 (d, *J* = 8.0 Hz, 1H), 5.39-5.36 (m, 1H), 5.31 (d, *J* = 7.6 Hz, 1H), 4.34-4.30 (m, 1H), 4.25-4.19 (m, 1H), 4.11 (d, *J* = 10.8 Hz, 1H), 4.06-4.02 (m, 1H), 3.83 (s, 3H), 3.56 (vt, *J* = 13.2, 6.4 Hz, 2H), 3.33-3.25 (m, 1H), 2.82-2.73 (m, 2H), 2.59-2.53 (m, 1H), 2.18 (d, *J* = 3.2 Hz, 6H), 2.06 (s, 3H), 2.01 (s, 3H), 1.81-1.73 (m, 3H), 1.55-1.39 (m, 6H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 170.8, 170.7, 170.6, 170.0, 168.3, 82.9, 72.8, 68.3, 68.0, 62.0, 58.0, 52.3, 45.0, 38.2, 32.4, 29.1, 28.7, 28.0, 26.3, 23.4, 21.1, 21.0, 20.7. HRMS (ESI): Calculated for: $C_{24}H_{37}ClN_4O_{10}SH$, 608.1919 ; found, 609.1975 (M+H).

Compound 3a: 6-*S*-[Methyl 5-acetamido-7, 8, 9-tri-O- acetyl-3, 4, 5-trideoxy -4-(N-tert-butyloxycarbonyl)-amino-D-glycero-α-D-galacto-non-2-ulopyranosyl)onate]-1-azido-hexane

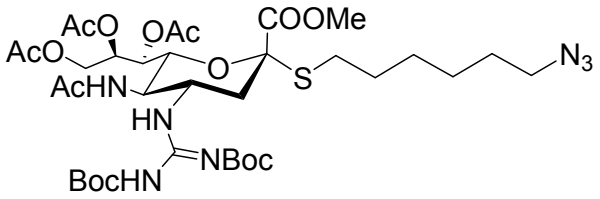


To a stirring solution of compound **2** (0.20 g, 0.33 mmol) in a mixture of THF: H_2O (5.0 ml, 1:1), PPh_3 (0.10 g, 0.39 mmol) was added and the reaction was heated at 40 ^{0}C for 12 h. The solvent was removed *in vacuo* and the residue purified by column chromatography to obtain a clear oil. This compound was directly taken to the next step without further purification. To a stirring solution of the crude product (0.18 g, 0.31 mmol) in THF (0.010 L) and TEA (31 mg, 0.31 mmol), di-*t*-butyl carbonate anhydride (0.10 g, 0.47 mmol) was added. Reaction mixture was stirred for 12h at rt. The white solid was filtered and the residue was concentrated, dissolved in DCM (25 ml) and washed with HCl (25 ml, 1M, 1x). The organic layer was dried over Na_2SO_4 and concentrated *in vacuo*. Product was purified via column chromatography with hexane: acetone (5:1) to yield a clear yellow oil (0.13 g, 60% yield over two steps). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.46 (s, 1H), 5.33 (s, 2H), 4.75 (d, *J* = 8.8 Hz, 1H), 4.28 (d, *J* = 12 Hz, 1H), 4.09 (d, *J* = 8.8 Hz, 1H), 3.88 (d, *J* = 10.4 Hz, 1H), 3.79-3.73 (m, 5H), 3.52 (t, *J* = 6.4, 3H), 2.72 (d, *J* = 11.6 Hz, 2H), 2.54-2.48 (m, 1H), 2.15 (s, 3H), 2.11 (s, 3H),

2.03 (s, 3H), 1.88 (s, 4H), 1.77-1.71 (m, 4H), 1.38 (s, 19H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.0, 170.7, 170.0 169.9, 168.7, 156.0, 83.4, 80.0, 68.5, 67.5, 62.2, 52.9, 50.2, 50.0, 45.0, 39.2, 32.4, 29.1, 28.7, 28.3, 26.4, 23.2, 21.2, 20.8, 20.7. HRMS (ESI): Calculated for $C_{29}H_{47}ClN_2O_{12}S$: 682.2538; Found: 683.2613 (M+H).

To a stirring solution of the chloro derivative (71 mg, 0.10 mmol) in DMF (1.0 ml) NaN_3 (65 mg, 1.04 mmol) was added and stirred at 60 ^{0}C. After 12 h, the reaction mixture was quenched with DI water (25 ml) and extracted with DCM (25 ml, 3x). The organic layers were collected and washed with brine (25 ml, 1x), dried over Na_2SO_4 and concentrated *in vacuo*, to yield a clear oil (64 mg, 90% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.47 (d, *J* = 12 Hz, 1H), 5.33 (s, 2H), 4.75 (d, *J* = 8 Hz, 1H), 4.29 (d, *J* = 12 Hz, 1H), 4.09 (d, *J* = 8 Hz, 1H), 3.87 (m, 1H), 3.79 (s, 3H), 3.60 (m, 1H), 3.52 (vt, *J* = 12, 8.0 Hz, 2H), 2.73 (d, *J* = 12 Hz, 2H), 2.54-2.48 (m, 1H), 2.15 (s, 3H), 2.11 (s, 3H), 2.03 (s, 3H), 1.88 (s, 3H), 1.77-1.74 (m, 3H), 1.5-1.40 (m, 2H), 1.38 (s, 14H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.0, 170.7, 170.1, 170.0, 168.7, 156.0, 83.4, 80.0, 68.5, 67.4, 62.2, 52.9, 50.2, 50.0, 45.0 39.2, 32.4, 28.3, 26.4, 23.2, 21.2, 20.84, 20.80. HRMS (ESI): Calculated for $C_{29}H_{47}N_5O_{12}S$: 689.2942; Found: 690.3004 (M+H).

Compound 3b: 6-*S*-[Methyl 5-acetamido-7, 8, 9-tri-O- acetyl-3, 4, 5-trideoxy-4-(bis-N, N'-tert-butyloxycarbonyl)-guanidino-D-glycero-α-D-galacto-non-2-ulopyranosyl)onate]-hexane

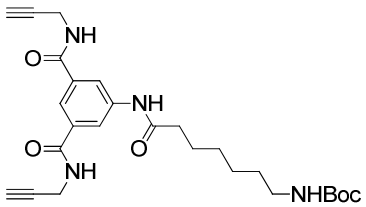


To a stirring solution of compound **2** (0.11 g, 0.20 mmol) in a mixture of THF and H_2O (2.0 ml, 1:1), PPh_3 (62 mg, 0.24 mmol) was added and heated at 40 ^{0}C for 12 h. The solvent was removed *in vacuo* and purified by column chromatography using DCM:MeOH (10:1) and used in the next step without purification. This compound (0.10 g, 0.16 mmol) in DCM (0.01 L), TEA (0.3ml, 1.6 mmol) was added and stirred for 10 min at rt. 1,3-bis(tert-butoxy-carbonyl)-2-methyl-2-thiopseudourea (55 mg, 0.19 mmol) and $HgCl_2$ (51

mg, 0.19 mmol) was added and stirred for 12 h. Next, the reaction mixture was washed with H_2O (25 ml, 1x) and brine (25 ml, 1x). The organic layers were separated, dried over Na_2SO_4 and concentrated *in vacuo*. Product was purified using column chromatography with hexane:acetone (3:1 ratio) to yield a clear oil (0.12 g, 80% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 11.30 (s, 1H), 8.35 (d, *J* = 8.0 Hz, 1H), 6.05 (d, *J* = 8.0 Hz, 1H), 5.37-5.29 (m, 2H), 4.36-4.33 (m, 1H), 4.06 (s, 3H), 3.54 (t, *J* = 12, 4.0 Hz, 2H), 2.82-2.74 (m, 2H), 2.56-2.53 (m, 1H), 2.17 (s, 3H), 2.14 (s, 3H), 2.04 (s, 3H), 1.83 (s, 4H), 1.48 (s, 24H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.8, 170.7, 170.2, 170.1, 168.8, 163.0, 156.8, 152.7, 83.8, 83.3, 79.5, 75.4, 69.0, 67.8, 62.4, 52.9, 50.5, 49.7, 45.0, 38.8, 32.4, 29.1, 28.7, 28.3, 28.0, 27.9, 26.4, 23.0, 21.2, 21.0, 20.8. HRMS (ESI): Calculated for $C_{35}H_{57}ClN_4O_{14}S$: 824.3281; Found: 825.3348 (M+H).

To a stirring solution of the chloro compound (47 mg, 0.06 mmol) in DMF (1.0 ml), NaN_3 (39 mg, 0.60 mmol) was added and stirred at 60 ^{0}C. After 12 h, the reaction mixture was quenched with H_2O and extracted with DCM (25 ml, 3x). The organic layers were collected and washed with brine (25 ml, 1x), dried over Na_2SO_4 and concentrated *in vacuo*, to yield a clear oil (40 mg, 85% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 11.31 (s, 1H), 8.40 (d, *J* = 7.0 Hz, 1H), 6.03 (d, *J* = 8.7 Hz, 1H), 5.33 (t, *J* = 15.6 Hz, 4H), 4.35 (d, *J* = 12.4 Hz, 2H), 4.14 – 3.94 (m, 4H), 3.91 – 3.69 (m, 7H), 3.27 (t, *J* = 6.8 Hz, 4H), 2.99 – 2.47 (m, 9H), 2.17 (s, 5H), 2.15 (s, 5H), 2.05 (s, 6H), 1.84 (s, 5H), 1.60 (d, *J* = 6.3 Hz, 6H), 1.49 (s, 22H), 1.39 (s, 10H), 1.26 (s, 8H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.8, 170.6, 170.2, 170.1, 168.8, 162.9, 156.8, 152.7, 83.8, 83.3, 79.5, 77.3, 77.0, 76.7, 75.4, 69.0, 67.8, 62.4, 52.9, 51.3, 50.4, 49.7, 38.8, 29.7, 29.1, 28.7, 28.7, 28.3, 28.0, 26.2, 23.0, 21.2, 21.0, 20.8. HRMS (ESI): Calculated for $C_{35}H_{57}N_7O_{14}S$: 831.3684; Found: 832.3695 (M+H).

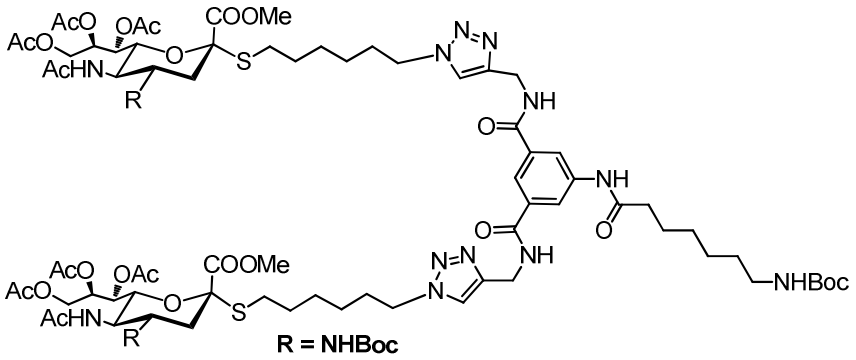
Compound 4: Tert-butyl(7-((3,5-bis(prop-2-yn-1-ylcarbamoyl)phenyl)amino)-7-oxoheptyl) carbamate.



To a stirring solution of the known spacer (64 mg, 0.26 mmol) in THF (5.0 ml), CDMT (91

mg, 0.52 mmol) and NMM (52 mg, 0.52 mmol) was added at 0 ^{0}C for 12 h. In a separate flask, the known dimeric scaffold[2] was dissolved in THF (5 ml) with NMM (52 mg, 0.52 mmol) at 0 ^{o}C and added to the activated acid and stirred for 12 h. The reaction mixture was quenched with H_2O, extracted with EtOAc (10 ml, 3x), dried over Na_2SO_4 and concentrated *in vacuo*. The compound was purified using flask chromatography with hexane: acetone (3:1) to yield a white solid (96 mg, 80% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.39 (s, 1H), 8.27 (s, 2H), 8.04 (s, 1H), 7.75 (s, 2H), 4.80 (s, 1H), 4.17 (s, 4H), 3.00 (d, *J* = 6.1 Hz, 2H), 2.25 (m, 5H), 1.42 (s,15H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.0, 166.7, 159.4, 139.4, 134.6, 121.8, 79.4, 77.4, 77.0, 76.7, 71.7, 56.0, 37.0, 31.6, 30.9, 29.8, 28.6, 28.4, 26.3, 25.2, 22.6, 14.1. HRMS (ESI) Calculated for $C_{26}H_{34}N_4O_5$: 482.2529; Found: 505.2404 (M+Na).

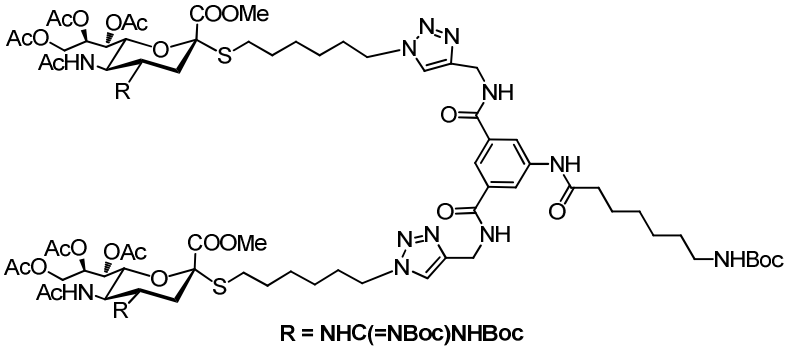
Compound 5a:



To a stirring solution of **3a** (40 mg, 0.06 mmol) in THF/ H_2O (1.0 ml, 1:1), **4** (9.6 mg, 0.02 mmol) was added. $CuSO_4$ (10 mg, 0.04 mmol) was added along with sodium L-ascorbate (7.9 mg, 0.04 mmol) and the reaction was stirred at rt for 12 h. Solvent was removed *in vacuo* and product was purified using flash column chromatography with DCM:MeOH (10:1) to yield **5a** (33 mg, 60% yield). ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.21 (s, 1H), 8.35 (s, 3H), 8.07 (s, 3H), 6.08 (s, 2H), 5.53 (d, *J* = 8.0 Hz, 2H), 5.36-5.31 (m, 5H), 4.70 (s, 5H), 4.32-4.29 (m, 6H), 4.07 (dd, *J* = 12, 4.0 Hz, 2H), 3.99-3.92 (m, 2H), 3.78-3.75 (m, 3H), 3.71 (s, 6H), 3.55 (d, *J* = 8.0 Hz, 2H), 3.10 (d, *J* = 4.0 Hz, 2H), 2.72-2.65 (m, 6H), 2.55-2.51 (m, 2H), 2.38 (s, 2H), 2.19 (m, 3H), 2.13 (s, 6H), 2.02 (s, 6H), 1.98 (s, 6H), 1.93 (s, 6H), 1.45 (s, 12H), 1.36 (s, 27H), 1.27 (s, 12H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 210.8, 172.3, 171.3, 170.7, 170.1, 170.0, 168.8, 166.6, 156.3, 156.1, 139.7, 135.0, 134.9, 123.1, 121.5, 121.4, 121.3, 83.5, 79.7, 79.13, 79.11, 77.9,

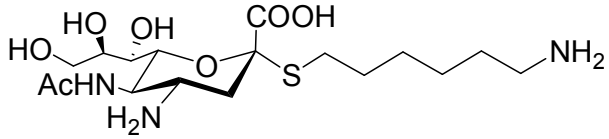
77.6, 77.5, 77.2, 76.6, 76.5, 74.4, 69.5, 68.4, 68.2, 68.0, 62.4, 53.8, 52.9, 51.0, 50.3, 50.2, 50.15, 50.1, 40.5, 40.4, 38.8, 37.2, 37.1, 35.5, 35.4, 31.9, 31.8, 29.9, 29.8, 29.7, 29.6, 29.5, 29.4, 29.3, 28.8, 28.7, 28.7, 28.6, 28.5, 28.4, 28.3, 28.2, 27,3, 27.2, 26.4, 25.9, 25.6, 25.5, 25.4, 25.3, 25.2, 23.3, 23.2, 22.7, 22.6, 21.1, 21.0, 20.8, 20.7, 14.1. HRMS (ESI) Calculated for $C_{84}H_{128}N_{14}O_{29}S_2$: 1860.8413; Found: 1923.6098 (M+ 3Na).

Compound 5b:



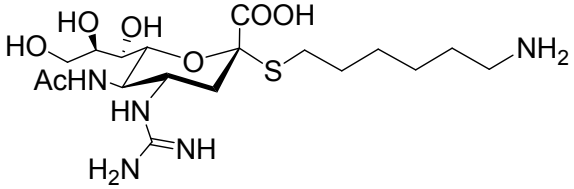
To a stirring solution of compound **3b** (26 mg, 0.04 mmol) in THF: H_2O (1.0 ml, 1:1), **4** (8.6 mg, 0.017 mmol) was added. $CuSO_4$ (7.5 mg, 0.38 mmol) was added with sodium L-ascorbate (6.7 mg, 0.28) and the reaction was stirred for 12 h at rt. Solvent was removed *in vacuo* and the product was purified using flash column chromatography with DCM:MeOH (10:1) to yield compound **5b** (84 mg, 65% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 11.30 (s, 2H), 8.50 (m, 3H), 8.38 – 8.19 (m, 2H), 7.86 – 7.59 (m, 6H), 6.25 (s, 2H), 5.33 (m, 5H), 4.72 (m, 6H), 4.34 (m, 7H), 4.21 – 4.01 (m, 9H), 3.88 – 3.69 (m, 10H), 3.11 (d, *J* = 6.1 Hz, 1H), 2.87 – 2.61 (m, 5H), 2.58 – 2.44 (m, 2H), 2.38-1.87 (m, J = 7.3 Hz, 22H), 1.57 – 1.14 (m, 67H) . ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.9, 170.7, 170.2, 168.7, 166.3, 162.9, 156.8, 152.5, 144.6, 134.9, 122.9, 121.3, 114.1, 83.5, 77.3, 77.2, 77.0, 76.7, 75.0, 68.8, 67.8, 62.5, 56.1, 53.8, 52.9, 50.2, 35.5, 31.9, 29.8, 29.7, 29.4, 29.3, 28.6, 28.4, 28.2, 27.9, 27.5, 25.8, 23.0, 22.7, 21.2, 20.9, 20.8, 14.1. HRMS (ESI) Calculated for $C_{96}H_{148}N_{18}O_{33}S_2$: 2144.9898; Found: 2145.9985 (M+H).

Compound SC1.



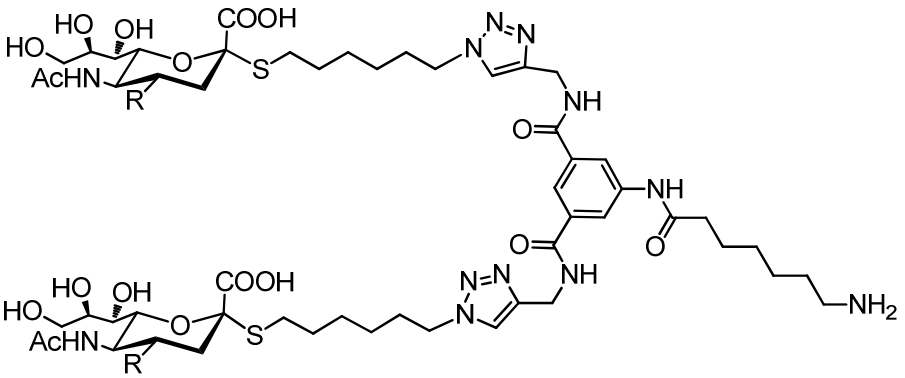
To a stirring solution of compound **3a** (10 mg, 16 µmol) in MeOH(1.0 ml), NaOMe (5.4 M, 0.25 ml) was added. The reaction was stirred for 1 h, neutralized with Dowex H^+ resin until the pH was 7. The resin was filtered, solvent was removed *in vacuo* and the residue was re-dissolved in DCM:TFA (1.0 ml, 1:1) and stirred for 1 h and solvent removed *in vacuo*. The residue was dissolved in EtOH with calaytic amount of $Pd(OH)_2$ and stirred for 8 h. Reaction mixture was filtered and solvent removed, re-dissolved in MeOH (1.0 ml), NaOH (10 mM, 1.0 ml) was added and stirred for 1 h. Dowex H^+ resin was used to neutralize to pH 7 and solvent was removed and compounds were purified with Bio-Gel P-2 Gel with DI water as solvent (5.0 mg, 75% yield). ^{1}H NMR (400 MHz, D_2O) δ 3.72 (t, *J* = 11.4 Hz, 2H), 3.62 – 3.32 (m, 5H), 3.22 (s, 1H), 2.87 (t, *J* = 7.6 Hz, 1H), 2.75 – 2.41 (m, 4H), 1.91 (d, *J* = 13.9 Hz, 3H), 1.52 (m, 4H), 1.28 (s, 4H). HRMS (ESI) Calculated for $C_{17}H_{33}N_3O_7S$: 423.2039; Found: 422.1962 (M-1, negative ion).

Compound SC2.



SC2 was synthesized in a manner from **3b** (5.0 mg, 16 µmol) similar to that of **Sc1** and the product was purified using Bio-Gel P-2 Gel with DI water as solvent to give pure **SC2** (1.9 mg, 70 %). 1H NMR (400 MHz, D_2O) δ 4.25-4.08 (m, 3H), 3.99 (m, 3H), 3.58-3.55 (m , 1H), 3.36-3.34 (m, 1H), 3.12-3.07 (m, *J* = 8.2 Hz, 2H), 2.95-2.78 (m, 1H), 2.44-2.35 (m, 3H), 2.15 (s, 1H), 2.03 (s, 1H), 1.87 (s, 5H), 1.69-1.54 (m, 3H). HRMS (ESI) Calculated for $C_{18}H_{35}N_5O_7S$: 465.2257; Found: 464.1987 (M-1, negative ion mode).

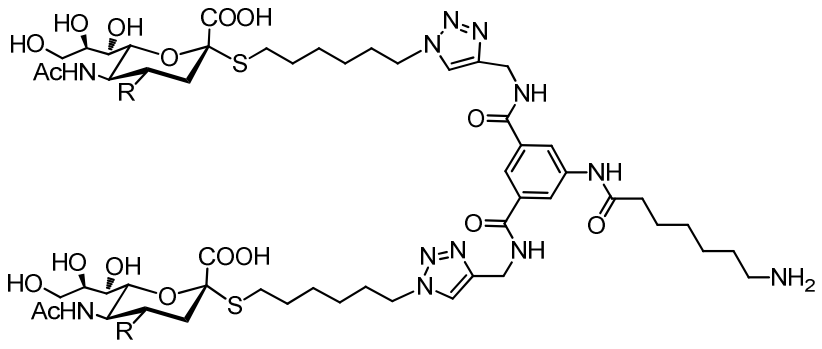
Compound SC3.



R = NH_2

To a stirring solution of compound **5a** (4.0 mg, 2.4 μmol) in MeOH (1.0 ml), NaOMe (5.4 M, 0.25 ml) was added. The reaction was stirred for 1 h, neutralized with Dowex H^+ resin. The resin was filtered, solvent was removed *in vacuo* and residue was re-dissolved in DCM/TFA (1.0 ml, 1:1) and stirred for 1 h. Solvent was removed *in vacuo*, re-dissolved in MeOH (1.0 ml) and NaOH (10 mM, 1.0 ml) was added and stirred for 1 h. Dowex H^+ resin was used to neutralized to pH 7 and solvent was removed. The product was purified with Bio-Gel P-2 Gel with DI water as solvent (2.2 mg, yield 80%). ^{1}H NMR (400 MHz, D_2O) δ 7.88 – 7.69 (m, 5H), 4.21 (q, *J* = 6.6 Hz, 2H), 4.06 – 3.90 (m, 1H), 3.76 – 3.44 (m, 11H), 3.30 – 3.14 (m, 2H), 2.90 – 2.83 (m, 2H), 2.80 – 2.70 (m, 2H), 2.62 – 2.47 (m, 3H), 2.45 – 2.33 (m, 2H), 2.32 – 2.21 (m, 2H), 1.92 (s, 6H), 1.77 (q, *J* = 12.9 Hz, 2H), 1.72 – 1.61 (m, 4H), 1.57 – 1.45 (m, 4H), 1.43 – 0.91 (m, 20H). HRMS (ESI) Calculated for $C_{55}H_{88}N_{14}O_{17}S_2$: 1280.5893; Found: 641.3013 (2M+H).

Compound SC4

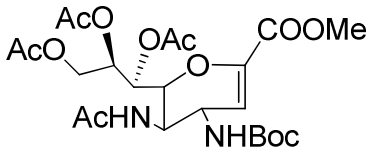


R = NHC(=NH)NH_2

SC4 was synthesized in a manner similar to **SC3** using **5b** (5.1 mg, 2.4 μmol) to yield a white solid (2.3 mg, 70% yield). ^{1}H NMR (400 MHz, D_2O) δ 7.99 – 7.73 (m, 5H), 4.28 (d, *J* =

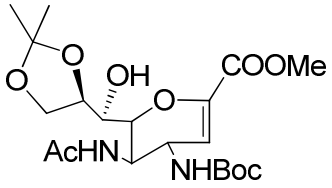
6.2 Hz, 1H), 4.05 (s, 1H), 3.83 (m, 3H), 3.65 (m, 10H), 3.48 (m,10H), 2.68 – 2.43 (m, 5H), 2.46 – 2.23 (m, 4H), 1.87 (s, 6H), 1.81 – 1.46 (m, 10H), 1.42 – 0.98 (m, 18H). HRMS (ESI) Calculated for $C_{57}H_{92}N_{18}O_{17}S_2$: 1364.6329; Found 1365.6420 (M+H).

Compound 7: 5-Acetylamino-4- N-tert-butyloxycarbonyl-6- (1,2,3-triacetoxy-propyl) -5,6-dihydro-4H-pyran-2-carboxylic acid methyl ester.



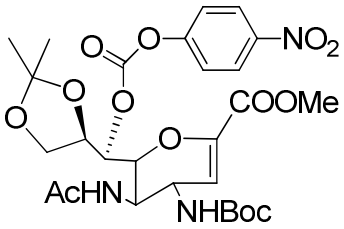
To a solution of compound **6** [3,4](1.5 g, 3.3 mmol) in EtOH (25 ml), Lindlar catalyst (0.15 g, 0.10 equivalent) was added. H_2 gas was bubbled to the solution and stirred at rt for 12 h. After filtering using celite, the filtrate was collected and solvent removed *in vacuo* to give a white product (1.4 g, quantitative). To this compound (0.53 g, 1.2 mmol) in THF (20 ml), Et_3N (1.6 ml, 1.5 mmol) was added. The solution was stirred rt for 30 min and Boc_2O (0.54 g, 2.5 mmol) was added and reaction stirred for 12 h at rt. Upon completion, THF was removed *in vacuo*. The residue was washed using HCl (1M, 25 ml) and extracted by DCM (30 ml, 3x), the organic phases were combined and dried over Na_2SO_4. DCM was removed *in vacuo* and the reaction mixture was purified using column chromatography with hexane: acetone (3:1) as eluent to give a white product. (0.56 g, 86%). ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.46 (d, *J* = 9.0 Hz, 1H), 5.94 (s, 1H), 5.43 (s, 1H), 5.27 (s, 1H), 4.63 (d, *J* = 12.4 Hz, 1H), 4.44 (d, *J* = 10.0 Hz, 1H), 4.35 (d, *J* = 8.9 Hz, 1H), 4.15 (dd, *J* = 12.3, 7.1 Hz, 1H), 3.95 (d, *J* = 9.2 Hz, 1H), 3.77 (s, 3H), 2.14 (s, 9H), 2.10 (s, 3H), 2.03 (s, 3H), 2.02 (s, 3H), 1.95 (s, 3H). 13C NMR (100 MHz, $CDCl_3$) δ 170.9, 170.6, 170.3, 170.0, 161.8, 156.2, 144.6, 111.0, 80.2, 71.4, 67.8, 62.2, 60.4, 52.4, 50.1, 47.3, 28.2, 23.1, 21.0, 20.9, 20.8. HRMS. Calculated for $C_{23}H_{34}N_2O_{12}$: 530.2112; Found: 531.2181 (M+H)

Compound 8: 5-Acetylamino-4- N-tert-butyloxycarbonyl-6-[(2,2-dimethyl-[1,3]dioxolan-4-yl] 5,6-dihydro-4H-pyran-2-carboxylic acid methyl ester.



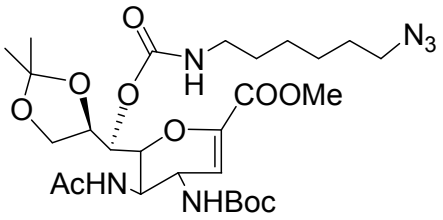
To a solution of compound **7** (0.56 g, 1.1 mmol) in MeOH (0.010 L), NaOMe (0.05 eq) was added. The solution was stirred at rt for 5 h. The progress of the reaction was monitored by TLC. Upon completion, the reaction mixture was neutralized by H^+ resin, the suspension was filtered. The liquid phase was collected and dried *in vacuo* to give a colorless compound. To a solution of this compound (0.42 g, 1.1 mmol) in dry acetone (0.010 L), H^+ resin was added to adjust the pH to 4. The solution was stirred at rt for 12 h. The suspension was filtered, acetone was removed *in vacuo*, the residue was washed by saturated $NaHCO_3$, extracted by DCM (3 × 0.020 mL), the organic phases were combined and dried over Na_2SO_4, DCM was removed *in vacuo* and the reaction mixture was purified by flash column chromatography using hexane : acetone (4:1) to give **8**. (0.41 g, 88%). 1H NMR (400 MHz, $CDCl_3$) δ 6.65 (d, *J* = 6.4 Hz, 1H), 5.79 (s, 1H), 5.08 (d, *J* = 4.2 Hz, 1H), 4.82 (d, *J* = 9.0 Hz, 1H), 4.59 (t, *J* = 9.4 Hz, 1H), 4.38 (dd, *J* = 13.5, 5.3 Hz, 1H), 4.26 – 4.06 (m, 2H), 4.01 (d, *J* = 10.6 Hz, 1H), 3.91 (td, *J* = 10.1, 6.7 Hz, 1H), 3.76 (s, 3H), 3.50 (dd, *J* = 8.3, 4.3 Hz, 1H), 2.03 (s, 3H), 1.44 (s, 9H), 1.40 (s, 3H), 1.36 (s, 3H). ^{13}C NMR (400 MHz, $CDCl_3$) δ 173.9, 162.0, 157.2, 146.3, 109.2, 107.8, 81.0, 78.3, 74.0, 69.7, 67.3, 52.4, 52.1, 48.7, 28.2, 27.1, 25.3, 23.0. HRMS(ESI) Calculated for $C_{20}H_{32}N_2O_9$: 444.2108; Found: 445.2180 (M+H).

Compound 9: 5-Acetylamino-4-N-tert-butyloxycarbonyl-6- [(2,2-dimethyl-[1,3]dioxolan-4-yl)-(4-nitro-phenoxycarbonyloxy)-methyl]-5,6-dihydro-4H-pyran-2-carboxylic acid methyl ester.



To a solution of compound **3** (35 mg, 0.079 mmol) in pyridine (0.010 L), DMAP (19 mg, 0.16 mmol) was added. The solution was stirred at rt for 30 min and 4-nitrophenylchloroformate (32 mg, 0.16 mmol) was added. The reaction was stirred at rt for 16 h. The reaction mixture was washed by HCl (1 M, 0.025 L) and extracted with DCM (3 × 0.020 L), the organic phases were combined and dried over Na_2SO_4. DCM was removed *in vacuo* and the product was purified by column chromatography using hexane: acetone (3:1) to give **9**. (39 mg, 80%). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.27 (d, *J* = 8.9 Hz, 2H), 7.50 (d, *J* = 8.9 Hz, 2H), 5.97 (d, *J* = 9.6 Hz, 1H), 5.90 (s, 1H), 5.31 (t, *J* = 5.7 Hz, 1H), 4.81 (d, *J* = 9.6 Hz, 1H), 4.54 (t, *J* = 9.7 Hz, 1H), 4.47 – 4.29 (m, 2H), 4.24 (dd, *J* = 8.9, 5.7 Hz, 2H), 4.17 – 4.05 (m, 1H), 3.79 (s, 3H), 1.95 (s, 3H), 1.42 (s, 9H), 1.41 (s, 3H), 1.38 (s, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.3, 161.6, 156.4, 155.7, 152.5, 145.6, 144.8, 125.2, 122.3, 110.5, 108.9, 80.6, 75.1, 74.2, 65.5, 52.5, 49.5, 48.1, 28.2, 26.4, 25.5, 23.2. HRMS(ESI) Calculated for $C_{27}H_{35}N_3O_{13}$: 609.2170; Found: 632.2057 (M+Na).

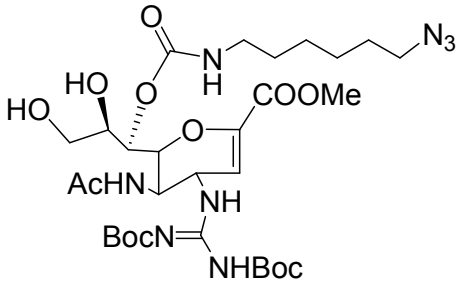
Compound 10a: 5-Acetylamino-4-N-tert-butyloxycarbonyl-6-[(6-azido-hexylcarbamoyloxy) -(2,2-dimethyl-[1,3]dioxolan-4-yl)-methyl]-5,6-dihydro-4H-pyran-2-carboxylic acid methyl ester.



To a solution of 6-azidohex-1-amine (19 mg, 0.13 mmol) in CH_3CN (8.0 ml), Et_3N (20 mg, 0.19 mmol) was added. The solution was stirred at rt for 30 min. **9** (39 mg, 0.064 mmol) in CH_3CN (2.0 ml) was added. The reaction was stirred at rt for 3 h. The progress of the reaction was monitored by TLC. Upon completion, CH_3CN was removed *in vacuo* and the reaction mixture was washed by HCl (1 M, 0.025 L), extracted by DCM (3 × 0.020 L), the organic phases were combined and dried over Na_2SO_4. DCM was removed *in vacuo* and the product was purified by column chromatography using hexane:EtOAc (1:1) to give **10a**. (35 mg, 89%).

^{1}H NMR (400 MHz, $CDCl_3$) δ 6.15 (d, *J* = 9.4 Hz, 1H), 5.90 (s, 1H), 5.24 (d, *J* = 3.7 Hz, 1H), 5.01 – 4.83 (m, 2H), 4.51 (t, *J* = 8.9 Hz, 1H), 4.33 (dd, *J* = 13.6, 7.8 Hz, 2H), 4.19 – 4.04 (m, 2H), 4.04 – 3.92 (m, 2H), 3.76 (s, 3H), 3.73(s, 1H) 3.24 (t, *J* = 6.9 Hz, 2H), 3.11 (m, 2H), 1.93 (s, 3H), 1.66 – 1.53 (m, 2H), 1.48 (m, 2H), 1.41 (m, 2H), 1.39 (s, 9H), 1.34(s, 3H), 1.23(s, 3H). ^{13}C NMR (1001 MHz, $CDCl_3$) δ 170.8, 162.1, 156.2, 155.5, 144.4, 111.4, 108.9, 80.1, 74.9, 69.7, 65.9, 52.4, 51.3, 50.0, 47.6, 41.1, 29.6, 28.7, 28.3, 26.4, 26.3, 25.4, 23.2. HRMS(ESI) Calculated for $C_{27}H_{44}N_6O_{10}$: 612.3119;Found: 635.3006 (M+Na).

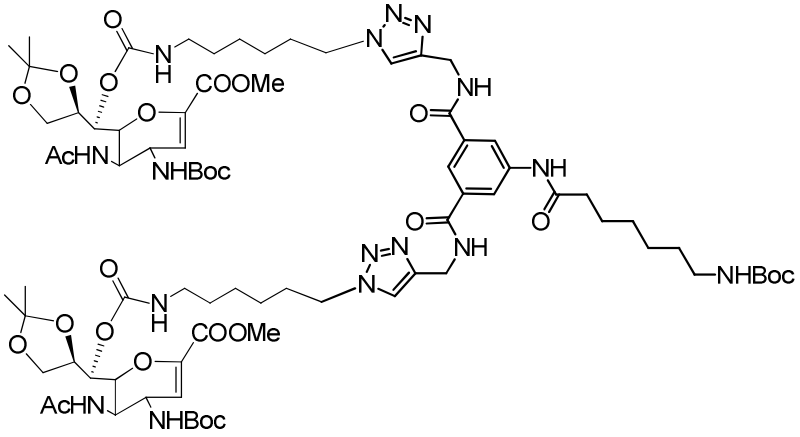
Compound 10b: 5-Acetylamino-4-[2,3-bis(tert-butoxycarbonyl)guanidine]-6-[(6-azido-hexyl carbamoyloxy)-2,3 dihydroxy propyl]-5,6-dihydro-4H-pyran-2-carboxylic acid methyl ester.



To a solution of compound **10a** (40 mg, 0.084 mmol) in THF (3.0 ml),TFA (3.0 ml) was added, the reaction was stirred at rt for 1 h. THF was removed *in vacuo*, Et_3N (26 µl, 0.25 mmol) was added. The solution was stirred at rt for 30 min. $HgCl_2$ (27 mg, 0.10 mmol) and 1,3-Bis(tert-butoxycarbonyl)-2-methyl-2-thiopseudourea (29 mg, 1.2 mmol) was added. The reaction was stirred at rt for 12 h. The reaction mixture was washed with HCl (1M, 0.025 L) , extracted with DCM (3 x 0.010 L), DCM was removed *in vacuo* and the product was purified by column chromatography using DCM : MeOH (25 :1) to give the product **10b**. (50 mg, 84%). ^{1}H NMR (400 MHz, $CDCl_3$) δ 11.40 (s, 1H), 8.54 (d, *J* = 8.7 Hz, 1H), 7.39 (s, 1H), 7.28 (s, 1H), 6.85 (s, 1H), 5.89 (s, 1H), 5.32 (s, 1H), 5.20 (t, *J* = 9.5 Hz, 1H), 5.03 (t, *J* = 5.7 Hz, 1H), 4.82 (d, *J* = 9.4 Hz, 1H), 4.49 (d, *J* = 10.5 Hz, 1H), 4.37 (m, 1H), 4.14 (dd, *J* = 14.2, 7.1 Hz, 1H), 4.05 (d, *J* = 9.2 Hz, 1H), 3.80 (s, 3H), 3.68 (m, 1H), 3.27 (t, *J* = 6.8 Hz, 2H), 3.17 (m, 2H), 1.95 (d, *J* = 10.0 Hz, 3H), 1.60 (dd, *J* = 13.8, 6.8 Hz, 2H), 1.50 (s, 18H), 1.38 (m, 4H), 1.27 (dd, *J* = 7.7, 6.5 Hz, 2H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.0, 162.8, 162.4, 162.2, 162.1, 157.1, 152.5, 144.9, 110.5, 83.7, 79.7, 69.4, 68.7, 62.4, 53.5, 52.31, 51.26, 49.4, 47.1, 45.7, 41.2, 29.4,

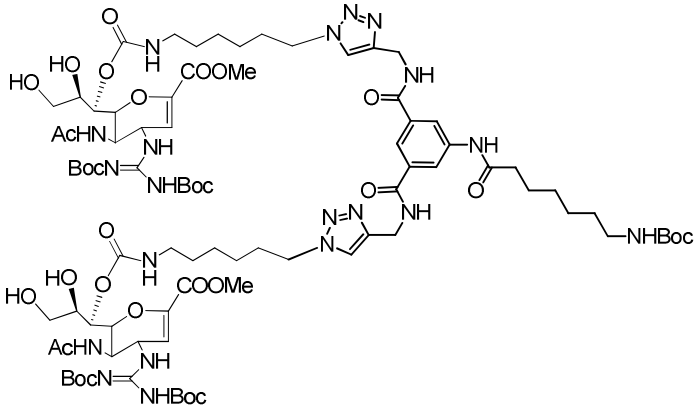
28.7, 28.3, 28.2, 28.1, 27.9, 26.3, 26.2, 22.7. HRMS(ESI) Calculated for $C_{30}H_{50}N_8O_{12}$: 714.3548; Found: 715.3627 (M+H).

Compound 11a:



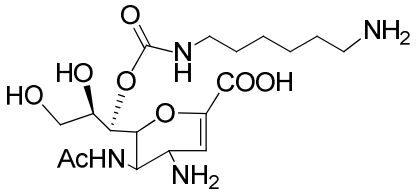
To a solution of **10a** (54 mg, 0.088 mmol) in THF/H_2O (1.0 ml, 1:1), **4** (18 mg, 0.036 mmol) was added. $CuSO_4$ (15 mg, 0.10 mmol) was added along with sodium L-ascorbate (.020 g, 0.10 mmol). The reaction was stirred at rt for 12 h. Upon completion by TLC, solvent was removed *in vacuo* and the product was purified by column chromatography using EtOAc: MeOH (30:1) to give **11a**. (48 mg, 78%). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.23(s, 3H), 7.83 (s, 2H), 6.67 (s, 2H), 5.95 (s, 2H), 5.64 (s, 1H), 5.24(s,3H), 4.60(m, 6H), 4.33(m, 8H), 4.12 (m, 8H), 3.98 (s, 2H), 3.71 (s, 6H), 3.04 (m, 4H), 2.40 (s, 1H), 2.19 (s, 2H), 2.09 (s, 2H), 2.06 (s, 2H), 1.90 (s, 10H), 1.68 (s, 2H), 1.44 (m, 14H), 1.26– 1.36 (m, 42H). HRMS (ESI) Calculated for $C_{80}H_{122}N_{16}O_{25}$: 1706.8767; Found: 1707.8838(M+H).

Compound 11b:



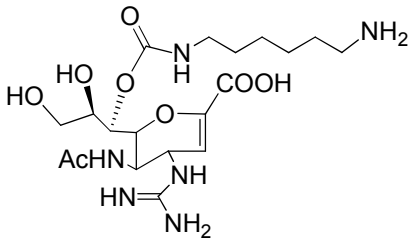
To a solution of **10b** (40 mg, 0.056 mmol) in THF/H_2O (1.0 ml, 1:1), **4** (13 mg, 0.025 mmol) was added. $CuSO_4$ (14 mg, 0.088 mmol) was added with sodium L-ascorbate (17 mg, 0.088 mmol). The reaction was stirred at rt for 12 h. Upon completion, solvent was removed *in vacuo* and the product was purified by column chromatography using EtOAc : MeOH (20:1) to give **11b**. (38 mg, 80%). ^{1}H NMR (400 ^{1}H NMR (400 MHz, $CDCl_3$) δ 11.38 (s, 1H), 9.74 (s, 1H), 8.19 (m, 5H), 7.28 (s, 2H), 5.89 (s, 2H), 5.31 (s, 6H), 5.21 – 5.05 (m, 2H), 4.78 (s, 2H), 4.52 (s, 5H), 4.17 – 3.90 (m, 4H),4.09-3.85 (m, 4H), 3.80-3.74 (m, 8H), 3.54 (s, 2H), 3.36 (s, 1H), 3.30 – 2.72 (m, 6H), 2.35 (d, *J* = 13.9 Hz, 2H), 2.05 (s, 2H), 1.81 (s, 7H), 1.54 (m, 22H), 1.45 – 1.33 (m, 12H), 1.33 – 0.99 (m, 28H), 0.90-0.84 (m, 10H). HRMS(ESI) Calculated for $C_{86}H_{134}N_{20}O_{29}$: 1910.9626; Found: 1912.0572(M+H)

Compound SC5:



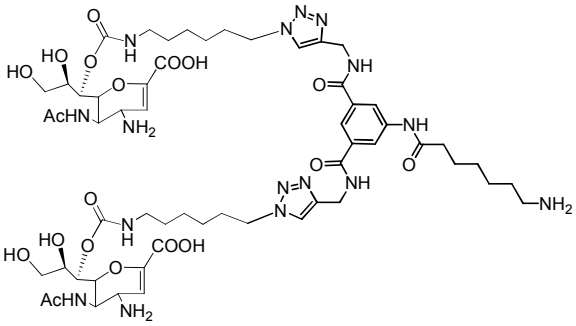
To a solution of compound **10a** (1.8 mg, 0.29 μmol) in MeOH (5.0 ml), NaOH (0.50 M, 1.0 ml) was added. The solution was stirred at rt for 2 h. The progress of the reaction was monitored by TLC. Upon completion, the reaction mixture was neutralized by H^+ resin, the suspension was filtered. The filtrate was collected and dried *in vacuo*. The residue was added to DCM/TFA (5.0 ml, 1:1), the mixture was stirred at rt for 1 h. DCM was removed *in vacuo*, the product was dissolved in EtOH (5.0 ml), Lindlar catalyst (10%) was added. H_2 gas was bubbled to the solution and stirred at rt for 12 h. The suspension was filtered and the filtrate, which contained product **SC5**, was collected and crude product was purified by Biogel P2 column to give pure **SC5** (1.0 mg, 76%). ^{1}H NMR (400 MHz, D_2O) δ 5.73 (s, 1H), 4.34 – 4.23 (m, 2H), 4.16 – 4.04 (m, 2H), 3.71 – 3.57 (m, 1H), 3.39 (dt, *J* = 13.3, 6.8 Hz, 2H), 3.02 (s, 4H), 1.98 (s, 3H), 1.55 – 1.30 (m, 4H), 1.23-1.18 (m, 4H). HRMS(ESI) Calculated for $C_{18}H_{32}N_4O_8$, 432.2220. Found:433.2292 (M+H).

Compound SC6:



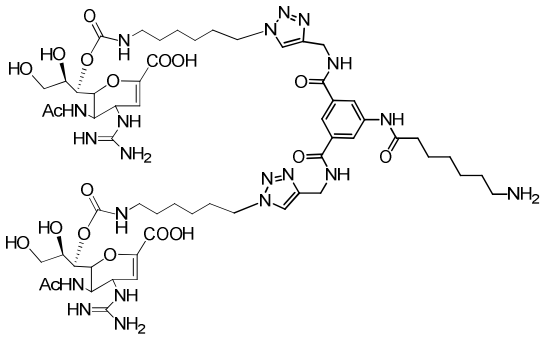
SC6 was synthesized in a manner similar to that of **SC5** using **10b** (1.6 mg, .22 μmol) and the crude product was purified by P2 column to give the **SC6** (0.75 mg, 72%). ^{1}H NMR (400 MHz, D_2O) δ 5.63 (s, 1H), 4.38 – 4.18 (m, 2H), 4.10-3.87 (m, 2H), 3.74-3.61(m, 2H), 3.38 (t, *J* = 7.2 Hz, 1H), 3.02 (s, 4H), 1.93 (s, 3H), 1.52 – 0.99 (m, 8H). HRMS(ESI) Calculated for $C_{19}H_{34}N_6O_8$: 474.2438; Found:475.2516 (M+H).

Compound SC7:



To a solution of compound **11a** (4.5 mg, 0.26 μmol) in MeOH (5.0 ml), NaOH (0.50 M, 1.0 ml) was added. The solution was stirred at rt for 2 h. The progress of the reaction was monitored by TLC. Upon completion, the reaction mixture was neutralized by H^+ resin and the suspension was filtered. The filtrate was dried *in vacuo* and DCM/TFA (1:1, 5.0 ml) was added to the residue and was stirred at rt for 1 h. After removal of solvent, the crude product was purified by P2 column to give **SC7**. (2.4 mg, 70%). ^{1}H NMR (400 MHz, D_2O) δ 7.89 (s, 2H), 7.80 (s, 3H), 5.91 (s, 2H), 4.84 (d, *J* = 9.4 Hz, 2H), 4.54 (s, 2H), 4.47 (d, *J* = 10.5 Hz, 2H), 4.29 (s, 3H), 4.25 – 4.08 (m, 3H), 3.96 (s, 4H), 3.53 (d, *J* = 9.8 Hz, 2H), 3.37 (dd, *J* = 11.9, 6.4 Hz, 2H), 2.88 (d, *J* = 7.4 Hz, 4H), 1.90 (s, 6H), 1.76 (s, 2H), 1.57 (s, 2H), 1.30 (s, 6H), 1.13-1.05 (m, 14H), 0.70-0.63 (m, 6H). HRMS(ESI) Calculated for $C_{57}H_{86}N_{16}O_{19}$: 1298.6255; Found: 1299.6329(M+H).

Compound SC8:



SC8 was synthesized in a manner similar to **SC7** using **11b** (3.5 mg, 0.18 μmol) and the crude product was purified by P2 column to give **SC8**. (2.0 mg, 79%). ^{1}H NMR (400 MHz, D_2O) δ 7.89 (s, 5H), 5.80 (d, *J* = 17.5 Hz, 2H), 4.44 (d, *J* = 10.1 Hz, 2H), 4.38 (d, *J* = 8.5 Hz, 2H), 4.34 – 4.20 (m, 4H), 4.16 (dd, *J* = 26.4, 10.1 Hz, 2H), 4.01 (dt, *J* = 13.9, 6.9 Hz, 4H), 3.62 (d, *J* = 8.8 Hz, 2H), 3.51 (d, *J* = 11.6 Hz, 1H), 2.99 – 2.79 (m, 5H), 2.29 (d, *J* = 7.5 Hz, 2H), 1.96 (s, 2H), 1.95 – 1.85 (m, 5H), 1.82 (d, *J* = 14.9 Hz, 2H), 1.71 (s, 3H), 1.54 (s, 4H), 1.27 (s, 8H), 1.20 – 1.11 (m, 6H), 1.06 (s, 6H). HRMS(ESI) Calculated for $C_{57}H_{86}N_{16}O_{19}$:1382.6691; Found: 1383.6761(M+H).

References:

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