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ARTICLE

Total Synthesis of MECA-79

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MECA-79 antigen is a sulfated mucin type core-1 extended *O*-glycan which is a potential anti-inflammatory agent. Herein we report a total synthesis of MECA-79 *via* a convergent [2+2] glycosylation route. The synthesis relies on efficient transformation of D-glucosamine into the orthogonally protected Tn antigen derivative and its elaboration into TF antigen *en route* to MECA-79.

Introduction

Mucin type *O*-glycoproteins are heavily *O*-glycosylated proteins which are ubiquitously distributed on the surface of epithelial tissues. The *O*-glycans although show a great structural diversity, they all contain a common building block, comprising N-acetyl D-galactosamine (GalNAc) residue α-linked to L-serine/L-threonine, called the Tn antigen.[1] The extension of Tn antigen at O3 and O6 with D-galactose, GlcNAc and GalNAc generates various core structures (core 1 to core 8).[2] Some of these *O*-glycans are specifically over-expressed on the surfaces of certain cancer cells and are well-established markers for tumor progression.[3] The tumor associated carbohydrate antigens (TACAs), are explored for the development of anti-cancer vaccines.[4-9] In 2001, Fukuda and co-workers identified a sulfated extended core-1 mucin type *O*-glycan called MECA-79 and proposed its structure as Galβ1→4(sulfo→6)GlcNAcβ1→3Galβ1→3GalNAc **1** (Figure 1).[10] This sulfated tetrasaccharide is of significant biological interest due to its unique expression at the site of chronic inflammation. Biological studies indicate that antibodies with specificities similar to MECA-79 may serve as anti-inflammatory agents.[11,12]

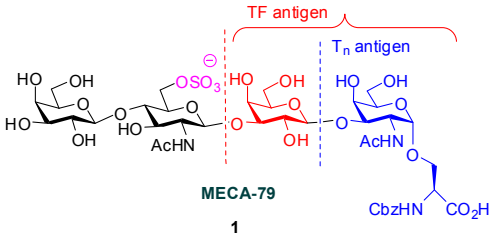
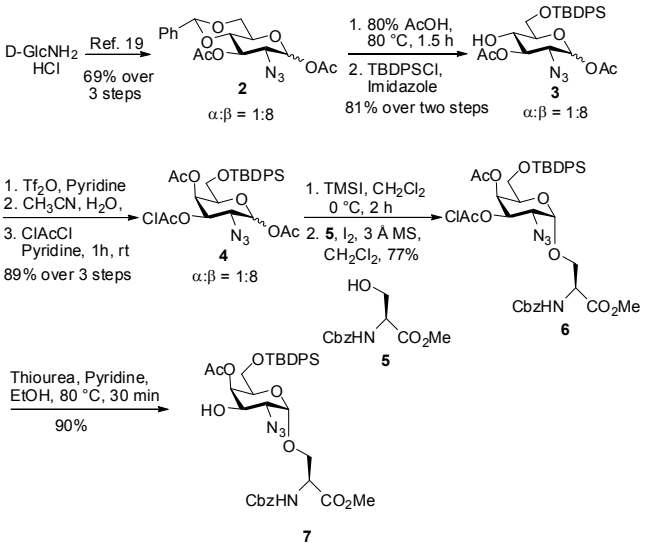


Figure 1 Structure of MECA-79 antigen

Soon after its isolation, Bélot and co-workers reported a synthesis of the *n*-octyl derivative of MECA-79.[13] Subsequently, Bertozzi and co-workers reported a synthesis of a thioether linked derivative of MECA-79 and its incorporation into a *O*-glycopeptide.[14] They also reported the first total synthesis of L-serine linked MECA-79 *en route* to sulfoadhesins *via* regioselective glycosylations.[15] In continuation of our studies directed towards the synthesis of glycosamine containing glycoconjugates,[16-18] we report herein a convenient synthesis of orthogonally protected Tn antigen and its application in the total synthesis of MECA-79 antigen.

Results and discussion

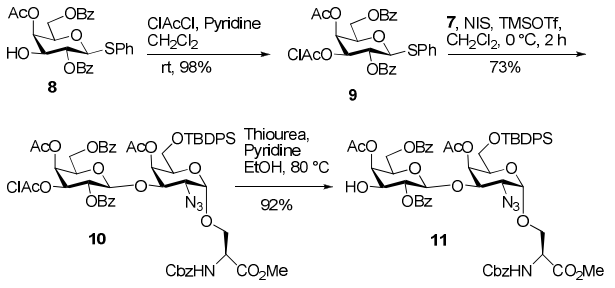
A practical synthesis of an orthogonally protected Tn antigen derivative workable on a 5 g scale is shown in Scheme 1. Synthesis of Tn antigen[1] entails a α-stereoselective coupling of a D-galactosamine donor with L-serine or L-threonine acceptor. Since D-galactosamine (D-GalNH$_2$) is expensive, we selected its C4 epimer D-glucosamine (D-GlcNH$_2$) as a cheap and abundant starting material. First, commercially available D-glucosamine was rapidly converted to fully protected derivative **2** *via* Hung's one-pot protection protocol in very good overall yields.[19] Accordingly, starting from 5 g D-GlcNH$_2$.HCl, the three stage sequence involving diazo transfer, per-*O*-silylation and one-pot protection could be carried out in 3 days to obtain 6 g of **2** after a single column purification (69% overall). Hydrolysis of the 4,6-*O*-benzylidene acetal of **2** followed by selective TBDPS protection of the primary hydroxyl group cleanly afforded the C4-alcohol derivative **3** (81%, over two steps). Triflation of **3** (Tf$_2$O, pyridine) and its subsequent reaction with water in acetonitrile as a solvent under reflux conditions effected the migration of C3-OAc group to C4 position with concomitant S$_N$2 displacement of the C4 triflate, resulting in the inversion of configuration at C4.[17,20] The free C3-hydroxyl group was capped with a chloroacetyl group to furnish the desired D-galactosamine building block **4** in 89% yield over three steps. For the synthesis of Tn antigen, the anomeric acetate **4** was first treated with TMSI to generate the requisite α-glycosyl iodide, which was stereoselectively glycosylated with the known L-serine derived acceptor **5**[21] using I$_2$ as a promoter[22] to obtain the α-linked glycosyl serine derivative **6** (^{1}H NMR H-1 δ 4.91, J = 3.6 Hz, ^{13}C NMR C-1 δ 99.2) in 77% yield over two steps. Selective removal of the chloroacetyl group in **6** by using thiourea[23] in pyridine



Scheme 1 Synthesis of orthogonally protected Tn antigen from D-glucosamine

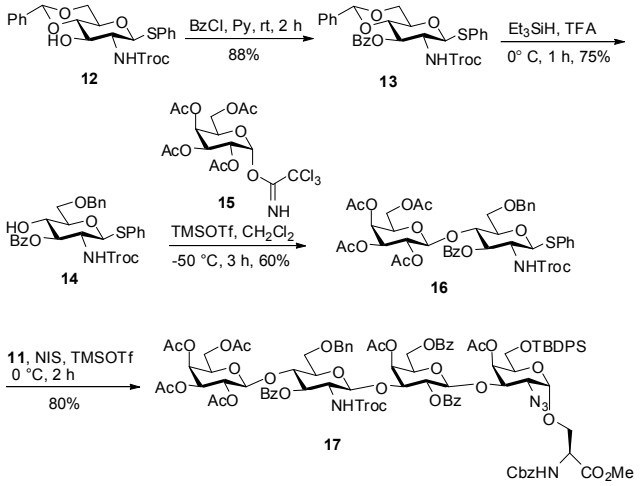
afforded acceptor **7** (90%). The entire sequence from D-$GlcNH_2$.HCl can be carried out in a week.

The appropriately protected Tn antigen derivative **6** was utilized for the synthesis of TF antigen derivatives as shown in Scheme 2. For this purpose, the known C3-alcohol D-galactose derivative **8**[24] was first treated with chloroacetyl chloride in pyridine to obtain the fully protected D-galactose building block **9** (98%). Glycosylation of donor **9** with Tn antigen acceptor **6** under NIS, TMSOTf conditions at 0 °C afforded the corresponding TF antigen derivative **10** in 73% yields. The chloroacetyl group at O3″ was subsequently removed with thiourea to furnish the desired C3″-alcohol disaccharide acceptor **11** in excellent yield.



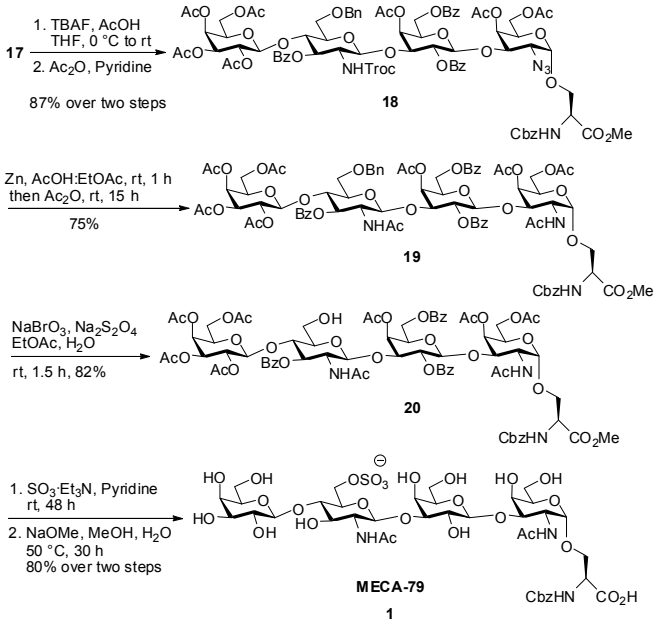
Scheme 2 Synthesis of TF antigen derivative **11**

Scheme 3 outlines the preparation of the left hand disaccharide donor **16** and its assembly with the TF disaccharide acceptor **11** to obtain tetrasaccharide **17**. For the synthesis of disaccharide **16**, we started with the known 4,6-*O*-benzylidene protected derivative **12**[25] which was easily obtained from D-$GlcNH_2$. By using benzoyl chloride and pyridine, the free C3-hydroxyl group in **12** was benzoylated to furnish **13**, which was then subjected to a regioselective reductive ring opening of 4,6-*O*-benzylidene acetal by using Et_3SiH and TFA[26] to afford the C4 alcohol **14**. Coupling of trichloroacetimidate donor **15**[27] with C4-OH acceptor **14** by using TMSOTf as a promoter at -50 °C in CH_2Cl_2 cleanly generated the *β*-linked disaccharide **16** in 60% yield along with a small amount of the corresponding aglycon transferred product (15%). Finally, thioglycoside donor **16** was coupled with acceptor **11** by using NIS, TMSOTf as a promoter to give exclusively *β*-linked tetrasaccharide **17** (80%) corresponding to MECA-79 antigen.



Scheme 3 Assembly of tetrasaccharide **17**

Scheme 4 outlines the global deprotection of fully protected tetrasaccharide **17**. Sequential removal of TBDPS group by treating with TBAF in AcOH followed by acylation by using acetic anhydride in pyridine furnished **18** as a sole product in 87% yield over two steps. One-step reduction of both the azide and NHTroc groups by using Zn in AcOH: EtOAc (2:1),[28] and subsequent acetylation by treating with acetic anhydride afforded the *N*-acetylated derivative **19** in good yields. Oxidative debenzylation using sodium bromate and sodium dithionate[29] in water and EtOAc furnished the 6‴-OH derivative 20, which was



Scheme 4 Global deprotection and completion of the synthesis of MECA-79

sulfated by treating with sulfur trioxide-triethyl amine complex in pyridine as a solvent followed by removal of all the ester functionalities with NaOMe in MeOH/H_2O to afford the target molecule **1** in 80% yield over two steps. The spectral data of **1** matched perfectly with the reported one confirming its identity.[15] Target molecule **1** and all the synthetic intermediates are thoroughly characterized by ^1H, ^{13}C, and 2D NMR (See Supporting Information).

Conclusions

In conclusion, we have successfully carried out the total synthesis of MECA-79 *via* a convergent [2+2] glycosylation route. The tetrasaccharide is equipped with a functional handle for further conjugation to a carrier protein or microarrays. The synthesis involves efficient transformation of D-glucosamine into the orthogonally protected Tn antigen derivative which also allows access to various mucin type oligosaccharides (core 1 to core 8) by selective removal of the protecting groups at O3 and O6 and coupling with respective glycosyl donors. Also the overall protecting group pattern of the fully protected tetrasaccharide **17** will further facilitate its elaboration into extended core 2 structures such as tetra-core 2 and hexa-core 2.

Experimental section

All reactions were conducted under a dry nitrogen atmosphere. Solvents (CH_2Cl_2 >99%, THF 99.5%, acetonitrile 99.8%, DMF 99.5%) were purchased in capped bottles and dried under sodium or CaH_2. All other solvents and reagents were used without further purification. All glassware used was oven dried before use. TLC was performed on pre-coated Aluminium plates of Silica Gel 60 F254 (0.25 mm, E. Merck). Developed TLC plates were visualized under a short-wave UV lamp and by heating plates that were dipped in ammonium molybdate/cerium (IV) sulfate solution. Silica gel column chromatography was performed using Silica Gel (100-200 mesh) and employed a solvent polarity correlated with TLC mobility. NMR experiments were conducted on 500 and 400 MHz instrument using $CDCl_3$ (D, 99.8%) or $(CD_3)_2CO$ (D, 99.9%) as solvents. Chemical shifts are relative to the deuterated solvent peaks and are in parts per million (ppm). ^1H-^1H COSY was used to confirm proton assignments. Mass spectra were acquired in the ESI mode. Melting points were determined by capillary apparatus. Specific rotation experiments were measured at 589 nm (Na) and 20 °C. IR spectra were recorded on an FT-IR spectrometer using CsCl plates.

1,3-Di-*O*-acetyl-2-azido-2-deoxy-6-*O*-*t*-butyldiphenylsilyl-*α,β*-D-glucopyranoside (3)

1,3-Di-*O*-acetyl-2-azido-4,6-*O*-benzylidene-2-deoxy-*α,β*-D-glucopyranoside **2** (3.50 g, 9.27 mmol) was dissolved in 80% AcOH (140 mL) and kept for reflux at 80 °C. After 1.5 h reaction mixture was concentrated *in vacuo* and subsequently co-evaporated with toluene (3 × 60 mL). After evaporation of solvent, the crude product was dissolved in CH_3CN (35 mL) and to this solution, imidazole (1.50 g, 23.17 mmol), followed by TBDPSCl (2.85 mL, 11.12 mmol) were added. After 15 min, the reaction mixture was concentrated *in vacuo* and the desired product was purified by column chromatography on silica gel (10% ethyl acetate: pet ether) to afford the desired product **3** as a yellowish liquid (3.90 g, 82%, *α/β* = 1:8): $[\alpha]^{20}_D$ = +8.9 (*c* 1.0, $CHCl_3$); IR ($CHCl_3$) *ν* 3498, 3018, 2931, 2858, 2112, 1759, 1428, 1372, 1218, 1113, 1082, 759, 704, 506 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) *δ* 7.67-7.64 (m, 4H, ArH), 7.45-7.35 (m, 6H, ArH), 6.24 (d, *J* = 3.6 Hz, H-1α), 5.53 (d, *J* = 8.5 Hz, H-1β), 4.97 (dd, *J* = 10.1, 9.4 Hz, 1H, H-3), 3.94 (dd, *J* = 10.9, 4.2 Hz, 1H, H-6a), 3.86 (dd, *J* = 10.9, 4.6 Hz, 1H, H-6b), 3.81-3.79 (m, 2H, H-4), 3.56-3.48 (m, 2H, H-2, H-5), 2.19 (s, 3H, $COCH_3$), 2.16 (s, 3H, $COCH_3$), 1.05 (s, 9H, $(CH_3)_3CSi$); ^{13}C NMR (100 MHz, $CDCl_3$) *δ* 171.2, 168.8, 135.8, 135.7, 132.7, 130.1, 130.09, 128.0, 127.9, 92.8, 90.4, 75.7, 75.5, 73.4, 70.5, 64.0, 62.7, 26.9, 21.1, 21.0, 19.3; HRMS-ESI $[M + H]^+$ calcd for $C_{26}H_{33}O_7SiN_3$ 528.2166, found 528.2161.

1,4-Di-*O*-acetyl-2-azido-2-deoxy-3-*O*-chloroacetyl-6-*O*-*t*-butyldiphenylsilyl-*α,β*-D-galactopyranoside (4)

Trifluoromethanesulfonic anhydride (1.07 mL, 6.37 mmol) was added dropwise at 0 °C to a stirred solution of **3** (2.8 g, 5.31 mmol), pyridine (2.56 mL, 31.8 mmol) in CH_2Cl_2 (40 mL). After 2 h, the reaction mixture was diluted with CH_2Cl_2 (100 mL) and washed successively with 1M HCl (30 mL), aq. $NaHCO_3$ (30 mL), and water (30 mL). Separated organic layer was dried over Na_2SO_4, concentrated and this crude product was used for the next step without purification.

The crude product was dissolved in acetonitrile (30 mL), H_2O (2.1 mL) and kept for reflux at 65 °C for 90 min. Then reaction mixture was concentrated *in vacuo* and the residue obtained was dissolved in EtOAc and washed with brine (3 × 50 mL). Separated organic layer was dried over Na_2SO_4 and concentrated. The crude product which was obtained after removal of solvent was dissolved in CH_2Cl_2 (30 mL). To this clear solution ClAcCl (1.3 mL, 16.0 mmol) and pyridine (1.3 mL, 16.0 mmol) were added. After 10 min, reaction mixture was concentrated *in vacuo* and chromatographed on silica gel (20% ethyl acetate: pet ether) to obtain the desired product **4** as a foam (2.85 g, 89%, *α/β* = 1:8): $[\alpha]^{20}_D$ = −16.8 (*c* 0.1, $CHCl_3$); IR ($CHCl_3$) *ν* 3685, 3020, 2400, 1520, 1424, 1216, 928, 769, 669, 627 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) *δ* 7.60-7.56 (m, 4H, ArH), 7.46-7.35 (m, 6H, ArH), 6.27 (d, *J* = 3.7 Hz, H-1α), 5.58 (dd, *J* = 3.2, 0.9 Hz, 1H, H-4), 5.53 (d, *J* = 8.5 Hz, H-1β), 4.97 (dd, *J* = 10.8, 3.2 Hz, 1H, H-3), 4.08, 4.06 (ABq, *J* = 15.3 Hz, 2H, -CH_2), 3.91-3.86 (m, 1H, H-6a), 3.83 (dd, *J* = 10.8, 8.5 Hz, 1H, H-2), 3.74 (dd, *J* = 9.8, 5.6 Hz, 1H, H-6b), 3.59 (dd, *J* = 9.8, 8.7 Hz, 1H, H-5), 2.15 (s, 3H, $COCH_3$), 2.01 (s, 3H, $COCH_3$), 1.01 (s, 9H, $(CH_3)_3CSi$); ^{13}C NMR (100 MHz, $CDCl_3$) *δ* 170.2, 168.7, 166.3, 135.8, 135.7, 132.7, 130.1, 128.0, 127.9, 93.0, 90.5, 73.9, 73.4, 71.2, 71.0, 65.8, 60.9, 60.5, 59.9, 57.1, 53.9, 40.6, 26.86, 26.83, 20.9, 20.7, 19.1; HRMS-ESI $[M + H]^+$ calcd for $C_{28}H_{34}O_8SiClN_3$ 604.1882, found 604.1870.

N-(Benzyloxycarbonyl)-*O*-(4-*O*-acetyl-3-*O*-chloroacetyl-2-azido-2-deoxy-6-*O*-*t*-butyldiphenylsilyl-*α*-D-galactopyranosyl)-L-serine methylester (6)

TMSI (0.8 mL, 5.56 mmol) was added to a clear solution of **4** (2.8 g, 4.63 mmol) in CH_2Cl_2 (30 mL) at 0 °C and reaction mixture was stirred at the same temperature for 2 h. After

complete consumption of starting material, benzene (15 × 2 mL) was added and the mixture was azeotroped twice on a rotary evaporator under high vacuum to obtain the crude glycosyl iodide. This brownish oil was dissolved in anhydrous CH_2Cl_2 (30 mL) and cannulated dropwise into a mixture of acceptor (1.0 g, 3.93 mmol) and 3 Å MS (2 g) at room temperature. After 6 h the crude reaction mixture was filtered through Celite. The filtrate was concentrated and chromatographed on silica gel (20% ethyl acetate: pet ether) to obtain **6** as a viscous oily liquid (2.41 g, 77%): $[\alpha]^{20}_D$ = +52.6 (*c* 1.8, $CHCl_3$); IR ($CHCl_3$) *v* 3018, 2929, 2113, 1749, 1216, 1046, 759, 668 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.63-7.58 (m, 5H, ArH), 7.44-7.31 (m, 10H, ArH), 5.66 (d, *J* = 8.4 Hz, 1H, NH), 5.59 (d, *J* = 3.2 Hz, 1H, H-4), 5.36 (dd, *J* = 11.0, 3.2 Hz, 1H, H-3), 5.09, 5.08 (ABq, *J* = 11.3 Hz, 2H, CH_2 of Cbz), 4.91 (d, *J* = 3.6 Hz, 1H, H-1), 4.58-4.55 (m, 1H, -CH), 4.09-4.02 (m, 5H, H-5, $-CH_2$, $-CH_2$), 3.79 (s, 3H), 3.75-3.63 (m, 1H, H-6a), 3.61-3.54 (m, 2H, H-2, H-6b), 1.98 (s, 3H, CH_3), 1.01 (s, 9H, $(CH_3)_3$CSi); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.3, 170.2, 166.4, 156.2, 136.1, 135.72, 135.71, 132.9, 132.8, 130.8, 130.08, 130.05, 128.7, 128.4, 128.3, 128.0, 99.2, 70.3, 69.5, 67.4, 67.2, 61.2, 57.6, 54.4, 53.0, 40.6, 26.8, 20.7, 19.1; HRMS-ESI [M + H]$^+$ calcd for $C_{38}H_{46}O_{11}SiClN_4$ 797.2621, found 797.2600.

N-(Benzyloxycarbonyl)-O-(4-O-acetyl-2-azido-2-deoxy-6-O-t-butyldiphenylsilyl-α-D-galactopyranosyl)-L-serine methylester (7)

Thiourea (0.36 g, 4.8 mmol) was added to a clear solution of **6** (0.54 g, 0.68 mmol) in pyridine (7 mL) and EtOH (7 mL) and the reaction mixture was kept for reflux at 80 °C. After 30 min, solvents were removed and the crude product was chromatographed on silica gel (25% ethyl acetate: pet ether) to afford **7** as a viscous liquid (0.41 g, 84%): $[\alpha]^{20}_D$ = +30.2 (*c* 1.0, $CHCl_3$); IR ($CHCl_3$) *v* 3018, 2975, 2112, 1735, 1427, 1216, 1047, 758, 669 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.63-7.59 (m, 5H, ArH), 7.42-7.31 (m, 10H, ArH), 5.68 (d, *J* = 8.4 Hz, 1H, NH), 5.47 (d, *J* = 3.0 Hz, 1H, H-4), 5.12-5.03 (m, 2H, $-CH_2$), 4.86 (d, *J* = 3.6 Hz, 1H, H-1), 4.57-4.55 (m, 1H, -CH), 4.22 (dd, *J* = 10.6, 3.0 Hz, 1H, H-3), 4.02-3.90 (m, 3H, H-5, $-CH_2$), 3.78 (s, 3H, CH_3), 3.74-3.58 (m, 2H, H-6a, H-6b), 3.40 (dd, *J* = 10.6, 3.6 Hz, 1H, H-2), 2.01 (s, 3H, CH_3), 1.02 (s, 9H, $(CH_3)_3$CSi); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.7, 170.4, 156.1, 135.7, 133.0, 132.9, 130.1, 130.0, 128.7, 128.4, 128.3, 128.0, 99.3, 70.1, 69.7, 69.4, 67.5, 67.4, 61.4, 60.2, 54.4, 53.0, 26.8, 20.9, 19.2; HRMS-ESI [M + Na]$^+$ calcd for $C_{36}H_{44}O_{10}SiN_4$ 743.2719, found 743.2706.

Phenyl 4-acetyl-3-chloroacetyl-2,6-di-O-benzoyl-1-thio-β-D-galactopyranoside (9)

To the clear solution of **8** (2.3 g, 4.401 mmol) in CH_2Cl_2 (24 mL) and pyridine (1.06 mL, 13.20 mmol) was added ClAcCl (1.05 mL, 1.49 mmol) at 0 °C. Then reaction mixture was kept up to 1 h at room temperature. After consumption of the starting material, the crude reaction mixture was concentrated and chromatographed on silica gel (25% ethyl acetate: pet ether) to get **9** as a yellowish solid (2.6 g, 98%): $[\alpha]^{20}_D$ = +21.3 (*c* 1.7, $CHCl_3$); mp 146 °C; IR ($CHCl_3$) *v* 3020, 1729, 1266, 1216, 1112, 758, 711, 668 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 8.02-8.00 (m, 4H, ArH), 7.62-7.44 (m, 8H, ArH), 7.26-7.23 (m, 1H, ArH), 7.19-7.15 (m, 2H, ArH), 5.60-5.54 (m, 2H, H-4, H-2), 5.36 (dd, *J* = 9.9, 3.3 Hz, 1H, H-3), 4.93 (d, *J* = 10.0 Hz, 1H, H-1), 4.55 (dd, *J* = 11.4, 7.2 Hz, 1H, H-6a), 4.39 (dd, *J* = 11.4, 5.7 Hz, 1H, H-6b), 4.19 (t, *J* = 6.4 Hz, 1H, H-5), 3.89-3.90 (m, 2H), 2.19 (s, 3H, $COCH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): δ 170.6, 166.8, 166.1, 165.4, 133.8, 133.6, 132.7, 132.6, 130.0, 129.9, 129.4, 129.2, 129.0, 128.8, 128.7, 128.3, 87.2, 74.7, 73.9, 67.8, 67.6, 62.3, 40.5, 20.8; HRMS-ESI [M + Na]$^+$ calcd for $C_{30}H_{27}ClO_9S$ 621.0957, found 621.0943.

N-(Benzyloxycarbonyl)-O-(4-O-acetyl-2,6-di-O-benzoyl-3-O-chloroacetyl-β-D-galactopyranosyl-(1→3)-2-azido-2-deoxy-4-O-acetyl-6-O-t-butyldimethylsilyl-α-D-galactopyranosyl)-L-serine methylester (10)

NIS (0.18 g, 1.38 mmol) was added to a suspension of thiogalactoside donor **9** (0.28 g, 0.46 mmol), acceptor **7** (0.27 g, 0.37 mmol) and 3Å MS (0.4 g) in CH_2Cl_2 (5 mL). Then the reaction mixture was brought to 0 °C and TMSOTf (25 µL, 0.14 mmol) was added slowly dropwise and kept stirring at rt for 2 h. After completion of the starting material, Et_3N (1 mL) was added and stirred for 10 min. Then the reaction mixture was filtered through Celite, washed with aq. $Na_2S_2O_3$, the separated organic layer was dried over Na_2SO_4 and concentrated. The residue was purified by silica gel chromatography (25% ethyl acetate: pet ether) to obtain the desired product **10** as a colourless foam (0.32 g, 73%): $[\alpha]^{20}_D$ = +37.4 (*c* 0.9, $CHCl_3$); IR ($CHCl_3$) *v* 2931, 2110, 1732, 1268, 1226, 1112, 1071, 757, 710, 614, 504 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.00-7.99 (d, *J* = 7.7 Hz, 4H, ArH), 7.63-7.52 (m, 6H, ArH), 7.45-7.30 (m, 15H, ArH), 5.61 (d, *J* = 8.6 Hz, 1H, NH), 5.53 (m, 2H, H-4, H-4′), 5.47 (dd, *J* = 10.4, 7.8 Hz, 1H, H-2′), 5.30 (dd, *J* = 10.4, 3.3 Hz, 1H, H-3′), 5.12-5.05 (m, 2H, CH_2 of Cbz), 4.92 (d, *J* = 7.8 Hz, 1H, *β*, H-1′), 4.83 (d, *J* = 3.6 Hz, 1H, *α*, H-1), 4.58-4.55 (m, 2H, -CH, H-6a′), 4.33 (dd, *J* = 10.9, 8.1 Hz, 1H, H-6b′), 4.14 (t, *J* = 6.8 Hz, 1H, H-5′), 4.04 (dd, *J* = 10.6, 3.2 Hz, 1H, H-3), 3.95-3.91 (m, 3H, CH_2, H-6a), 3.89-3.85 (m, 2H, $-CH_2$), 3.70 (s, 3H, CO_2Me), 3.63-3.54 (m, 2H, H-5, H-6b), 3.47 (dd, *J* = 10.6, 3.6 Hz, 1H, H-2), 2.20 (s, 3H, $COCH_3$), 1.97 (s, 3H, $COCH_3$), 1.02 (s, 9H, $(CH_3)_3$CSi); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.6, 170.4, 169.3, 166.8, 165.9, 165.2, 156.1, 136.1, 135.71, 135.70, 133.6, 133.5, 133.2, 133.1, 129.9, 129.8, 129.4, 129.2, 128.7, 128.6, 128.58, 128.5, 128.3, 127.9, 127.8, 101.7, 98.8, 74.3, 72.6, 70.8, 70.7, 69.4, 69.3, 68.9, 67.4, 66.8, 62.5, 61.1, 59.5, 54.3, 52.8, 40.5, 26.8, 20.8, 20.7, 19.2; HRMS-ESI [M + Na]$^+$ calcd for $C_{60}H_{65}ClN_4O_{19}Si$ 1231.3593 found 1231.3542.

N-(Benzyloxycarbonyl)-O-(4-O-acetyl-2,6-di-O-benzoyl-3-β-D-galactopyranosyl-(1→3)-2-azido-2-deoxy-4-O-acetyl-6-O-t-butyldimethylsilyl-α-D-galactopyranosyl)-L-serine methylester (11)

Thiourea (0.14 g, 1.90 mmol) was added to a clear solution of **10** (0.32 g, 0.27 mmol) in pyridine (3.5 mL) and EtOH (3.5 mL) and the reaction mixture was kept for reflux at 80 °C. After 30 min, solvents were removed and the crude product was chromatographed on silica gel (25% ethyl acetate: pet ether) to afford **11** as a white foam (0.28 g, 92%): $[\alpha]^{20}_D$ = +29.8 (*c* 2.8, $CHCl_3$); IR ($CHCl_3$) *v* 3436, 3020, 2931, 2110, 1728, 1270, 1230, 1112, 1070, 758, 710, 504 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.10-8.02 (m, 4H, ArH), 7.66-7.63 (m, 5H, ArH), 7.59-7.33 (m, 16H, ArH), 5.66 (d, *J* = 8.0 Hz, 1H, NH), 5.56 (d, *J* = 2.3 Hz, 1H, H-4), 5.50 (d, *J* = 2.9 Hz, 1H, H-4′), 5.27-5.23 (m, 1H, H-2′),

5.14, 5.10 (ABq, J = 12.0 Hz, 2H, CH_2 of Cbz), 4.90 (d, J = 7.7 Hz, 1H, β, H-1′), 4.87 (d, J = 3.5 Hz, 1H, α, H-1), 4.64-4.53 (m, 2H, -CH, H-6a′), 4.34 (dd, J = 11.1, 7.3 Hz, 1H, H-6b′), 4.09-4.05 (m, 2H, H-3, H-5′), 4.02 (dd, J = 9.8, 3.3 Hz, 1H, H-3′), 3.96-3.92 (m, 3H, CH_2, H-6a), 3.74 (s, 3H, CO_2Me), 3.68-3.57 (m, 2H, H-5, H-6b), 3.53 (dd, J = 10.4, 3.5 Hz, 1H, H-2), 2.22 (s, 3H, $COCH_3$), 1.96 (s, 3H, $COCH_3$), 1.05 (s, 9H, $(CH_3)_3$CSi); ^{13}C NMR (125 MHz, $CDCl_3$) δ 171.0, 170.5, 170.3, 169.2, 166.5, 166.1, 165.9, 165.2, 156.0, 136.0, 135.6, 135.5, 133.3, 133.2, 133.0, 132.9, 129.8, 129.7, 129.5, 129.4, 128.6, 128.5, 128.4, 128.3, 128.2, 128.0, 127.8, 127.74, 127.71, 101.5, 98.7, 74.5, 73.0, 72.9, 72.4, 71.1, 70.9, 70.8, 69.9, 69.5, 68.7, 67.3, 67.0, 62.5, 61.7, 59.3, 54.2, 52.7, 26.7, 20.8, 20.6, 19.1, 19.0; HRMS-ESI [M + Na]$^+$ calcd for $C_{58}H_{64}N_4O_{18}Si$ 1155.3877, found 1155.3923.

Phenyl 4,6-(benzylidene)-3-benzoyl-2-deoxy-2-[[(2,2,2-trichloroethoxy)carbonyl]amino]-1-thio-β-D-glucopyranoside (13)

BzCl (0.6 mL, 5.40 mmol), pyridine (0.43 mL, 5.40 mmol) were added to a clear solution of **12** (0.9 g, 1.80 mmol) in CH_2Cl_2 (10 mL) at 0 °C. The reaction mixture was stirred at room temperature for 2 h. After that the reaction mixture was diluted with CH_2Cl_2 and washed with $NaHCO_3$ (3 × 20 mL) and brine (20 mL). Then dried over Na_2SO_4, concentrated and chromatographed on silica gel (12% ethyl acetate: pet ether) to obtain the desired product **13** as a white solid (1.0 g, 87%): $[\alpha]^{20}_D$ = −15.4 (c 3.3, $CHCl_3$); mp 215 °C; IR ($CHCl_3$) ν 3019, 1712, 1541, 1369, 1270, 1216, 1082, 1023, 759 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.02 (d, J = 7.3 Hz, 2H, ArH), 7.56-7.53 (m, 1H, ArH), 7.47-7.46 (m, 2H, ArH), 7.41-7.37 (m, 2H, ArH), 7.34-7.32 (m, 2H, ArH), 7.28-7.19 (m, 4H, ArH), 7.14-7.11 (m, 2H, ArH), 6.35 (d, J = 10.0 Hz, 1H, NH), 5.79 (t, J = 9.8 Hz, 1H, H-3), 5.46 (s, 1H, benzylidene), 4.84 (d, J = 10.4 Hz, 1H, H-1), 4.14 (d, J = 10.4 Hz, 1H, H-2), 4.63 (s, 2H, CH_2 of Troc), 4.06 (dd, J = 10.2, 4.7 Hz, 1H, H-6a), 3.81 (t, J = 9.4 Hz, 1H, H-4), 3.70 (t, J = 10.2 Hz, 1H, H-6b), 3.62-3.56 (m, 1H, H-5); ^{13}C NMR (125 MHz, $CDCl_3$): δ 167.0, 154.6, 136.9, 133.7, 132.7, 132.5, 130.1, 129.0, 128.9, 128.5, 128.14, 128.11, 125.8, 100.9, 95.4, 88.3, 78.7, 74.4, 73.8, 70.4, 68.3, 55.5; HRMS-ESI [M + Na]$^+$ calcd for $C_{29}H_{26}Cl_3NO_7S$ 660.0388, found 660.0382.

Phenyl 3-benzoyl-6-benzyl-2-deoxy-2-[[(2,2,2-trichloroethoxy)carbonyl]amino]-1-thio-β-D-glucopyranoside (14)

Triethyl silane (0.5 mL, 3.0 mmol) and TFA (0.3 mL, 3.0 mmol) were added dropwise at 0 °C to a stirred solution of **13** (0.38 g, 0.60 mmol) in CH_2Cl_2 (4 mL) and kept the reaction mixture for 1 h. After completion of the starting material, the reaction mixture was diluted with CH_2Cl_2 and washed with $NaHCO_3$ (20 mL) and brine (10 mL). Separated organic layer was dried over Na_2SO_4. The desired product was purified by column chromatography on silica gel by using (25% ethyl acetate: pet ether) as eluent to afford the desired product **14** as a white foam (0.29 g, 75%): $[\alpha]^{20}_D$ = +15.6 (c 7.6, $CHCl_3$); IR ($CHCl_3$) ν 3350, 3019, 2868, 1719, 1528, 1279, 1216, 1070, 1026, 820, 759, 712, 668, 570 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.93 (d, J = 7.6 Hz, 2H, ArH), 7.52-7.49 (m, 3H, ArH), 7.33-7.19 (m, 10H, ArH), 5.83-5.79 (m, 1H, NH), 5.40 (t, J = 9.6 Hz, 1H, H-3), 4.83 (d, J = 10.3 Hz, 1H, H-1), 4.67 (d, J = 12.0 Hz, 1H, CH*H*Ph), 4.55-4.49 (m, 3H, CH*H*Ph, CH_2 of NHTroc), 4.00-3.94 (m, 1H, H-2), 3.85-3.66 (m, 4H, H6a, H6b, H-5, H-4); ^{13}C NMR (100 MHz, $CDCl_3$) δ 167.4, 154.5, 137.8, 133.7, 133.5, 133.1, 132.1, 131.9, 130.1, 129.1, 129.0, 128.8, 128.6, 128.3, 127.9, 127.6, 95.4, 86.9, 78.6, 74.3, 73.6, 70.4, 69.9, 55.0, 54.7; HRMS-ESI [M + Na]$^+$ calcd for $C_{29}H_{28}Cl_3NO_7S$ 662.0544, found 662.0540.

Phenyl *O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl-(1→4)-3-*O*-benzoyl-6-*O*-benzyl-2-deoxy-2-(2,2,2-trichloroethoxycarbonylamino)-1-thio-β-D-glucopyranoside (16)

TMSOTf (25 µL, 0.01 mmol) was added dropwise to a suspension of imidate **15** (0.70 g, 1.42 mmol), acceptor **14** (0.91 g, 1.42 mmol) and 3Å MS (0.3 g) at −50 °C and the reaction mixture was stirred at the same temperature for 3 h. Then the mixture was diluted with CH_2Cl_2 filtered through Celite and concentrated. The residue was purified by silica gel column chromatography (25% ethyl acetate: pet ether) to give the desired product **16** as a foam (0.65 g, 60%). Along with the product some amount of aglycon transferred product (phenyl 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-galactopyranoside) was also isolated (15%) which was confirmed by ^{1}H and ^{13}C NMR: $[\alpha]^{20}_D$ = −11.7 (c 2.3, $CHCl_3$); IR ($CHCl_3$) ν 2925, 1750, 1369, 1222, 1060, 760 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.99 (d, J = 7.3 Hz, 2H, ArH), 7.55-7.54 (m, 3H, ArH), 7.40-7.33 (m, 7H, ArH), 7.28-7.26 (m, 3H, ArH), 6.03 (d, J = 9.6 Hz, 1H, NH), 5.47 (t, J = 9.6 Hz, 1H, H-3), 5.09 (bs, 1H, H-4′), 4.97-4.93 (m, 2H, H-1, H-2′), 4.77-4.75 (m, 2H, H-3′), 4.67 (d, J = 12.0 Hz, 1H, CH*H*Ph), 4.57 (d, J = 12.0 Hz, 1H, CH*H*Ph), 4.51-4.45 (m, 3H, CH_2 of NHTroc, H-1′), 4.13-4.03 (m, 2H, H-4, H-2), 3.82-3.73 (m, 2H, H-6a′ & H-6b′), 3.67-3.66 (m, 1H, H-5), 3.42-3.36 (m, 3H, H-6a & H-6b, H-5′), 1.97 (s, 6H, $COCH_3$), 1.95 (s, 3H, $COCH_3$), 1.92 (s, 3H, $COCH_3$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.2, 170.1, 168.9, 166.4, 154.4, 137.8, 133.4, 132.9, 131.7, 129.9, 129.6, 129.0, 128.6, 128.4, 128.2, 128.1, 127.5, 100.2, 95.4, 86.4, 78.8, 76.9, 74.7, 74.6, 74.3, 73.8, 70.9, 70.3, 69.3, 67.6, 66.5, 60.3, 54.9, 20.8, 20.7, 20.6, 20.3; HRMS-ESI [M + Na]$^+$ calcd for $C_{43}H_{46}Cl_3NO_{16}S$ 994.1476, found 994.1419.

***N*-(Benzyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl-(1→4)-3-*O*-benzoyl-6-*O*-benzyl-2-deoxy-2-(2,2,2-trichloroethoxycarbonylamino)-β-D-glucopyranosyl-(1→3)-4-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-galactopyranosyl-(1→3)-2-azido-2-deoxy-4-*O*-acetyl-6-*O*-*t*-butyldimethylsilyl-α-D-galactopyranosyl)-L-serine methylester (17)**

To a suspension of donor **16** (0.13 g, 0.13 mmol), acceptor **8** (0.12 g, 0.10 mmol) and 3Å MS (0.2 g) in CH_2Cl_2 (3 mL), NIS (0.09 g, 0.66 mmol) was added. After that TMSOTf (12 µL, 0.06 mmol) was added slowly dropwise at 0 °C and the reaction mixture was kept for stirring at rt for 2 h. After consumption of the starting material, Et_3N (1.0 mL) was added to quench the reaction. Then the reaction mixture was filtered through Celite, washed with $Na_2S_2O_3$, the separated organic layer was dried over Na_2SO_4 and concentrated. The crude product was purified by column chromatography on silica gel (40% ethyl acetate: pet ether) to afford 17 as a white foam (0.17 g, 80%): $[\alpha]^{20}_D$ = +25.1 (c 1.0, $CHCl_3$); IR ($CHCl_3$) ν 2930, 2110, 1749, 1369, 1228, 1070, 758, 711, 668, 504 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.07-8.05 (m, 4H, ArH), 7.94-7.93 (m, 2H, ArH), 7.64-7.30 (m, 29H, ArH), 5.64-5.61 (m, 2H, NH), 5.53-5.52 (m, 1H), 5.43 (dd,

J = 9.6, 8.3 Hz, 1H), 5.19 (dd, *J* = 10.0, 9.8 Hz, 1H), 5.15-5.06 (m, 3H), 4.94 (dd, *J* = 10.1, 8.1 Hz, 1H), 4.84-4.82 (m, 2H), 4.78-4.74 (m, 2H), 4.70-4.68 (m, 1H), 4.58-4.54 (m, 3H), 4.50-4.42 (m, 3H), 4.39-4.36 (m, 1H), 4.07-4.00 (m, 5H), 3.99-3.91 (m, 3H), 3.75-3.72 (m, 2H), 3.71 (s, 3H, CO_2CH_3), 3.60-3.54 (m, 3H), 3.49-3.38 (m, 4H), 2.23 (s, 3H, $COCH_3$), 1.98 (s, 3H, $COCH_3$), 1.97 (s, 3H, $COCH_3$), 1.92 (s, 6H, $COCH_3$), 1.90 (s, 3H, $COCH_3$), 1.03 (s, 9H, $(CH_3)_3CSi$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.4, 170.3, 170.24, 170.2, 170.1,169.4, 169.1, 166.1, 165.9, 164.9, 156.1, 154.0, 138.1, 136.1, 135.75, 135.7, 133.6, 133.5, 133.3, 133.2, 129.9, 129.8, 129.7, 129.5, 128.7, 128.6, 128.5, 128.47, 128.4, 128.0, 127.9, 127.8, 102.2, 101.8, 100.3, 98.9, 95.4, 75.0, 74.0, 73.9, 72.6, 71.5, 71.2, 71.0, 70.5, 69.6, 69.3, 69.1, 68.8, 67.6, 67.4, 66.6, 62.7, 62.3, 60.5, 59.5, 56.3, 54.3, 52.9, 26.8, 21.0, 20.83, 20.8, 20.7, 20.6, 20.5, 19.2; HRMS-ESI $[M + Na]^+$ calcd for $C_{95}H_{104}Cl_3N_5O_{34}Si$ 2016.5297, found 2016.5254.

***N*-(Benzyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-*β*-D-galactopyranosyl-(1→4)-3-*O*-benzoyl-6-*O*-benzyl-2-deoxy-2-(2,2,2-trichloroethoxycarbonylamino)-*β*-D-glucopyranosyl-(1→3)-4-*O*-acetyl-2,6-di-*O*-benzoyl-*β*-D-galactopyranosyl-(1→3)-2-azido-2-deoxy-4,6-di-*O*-acetyl-*α*-D-galactopyranosyl)-L-serine methylester (18)**

A solution of TBAF (1.3 mL, 4.48 mmol, 1 M solution in THF) and AcOH (0.1 mL, 2.26 mmol) with pH 7 was added at 0 °C to a clear solution of **17** (0.12 g, 0.06 mmol) in THF (6.8 mL) and the reaction mixture was stirred at rt overnight. After complete consumption of starting material, solvents were removed *in vacuo* and the reaction mixture was azoetroped twice with toluene (2 × 5 mL). The crude product obtained after solvent evaporation was dissolved in pyridine (2.0 mL) and Ac_2O (0.7 mL, 7.42 mmol) and kept for stirring. After consumption of the starting material, the solvent was concentrated *in vacuo* and chromatographed on silica gel (60% ethyl acetate: pet ether) to obtain **18** as a viscous liquid (0.09 g, 87%): $[\alpha]^{20}_D$ = +8.4 (*c* 0.1, $CHCl_3$); IR ($CHCl_3$) *v* 3019, 2110, 1518, 1424, 1217, 759, 669, 627 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.10-8.01 (m, 4H, ArH), 7.99-7.92 (m, 2H, ArH), 7.61-7.27 (m, 19H, ArH), 5.73 (d, *J* = 8.1 Hz, 1H, NH), 5.63 (d, *J* = 3.1 Hz, 1H), 5.48-5.42 (m, 2H), 5.21-5.17 (m, 2H), 5.14-5.07 (m, 2H), 4.98-4.91 (m, 2H), 4.82-4.79 (m, 2H), 4.76-4.73 (m, 2H), 4.70-4.67 (m, 1H), 4.62-4.53 (m, 3H), 4.51-4.43 (m, 4H), 4.38-4.35 (m, 1H), 4.05-3.95 (m, 8H), 3.92-3.85 (m, 2H), 3.71 (s, 3H, CO_2CH_3), 3.56-3.53 (m, 2H), 3.47-3.36 (m, 4H), 2.21 (s, 3H, $COCH_3$), 2.09 (s, 3H, $COCH_3$), 2.00 (s, 3H, $COCH_3$), 1.96 (s, 3H, $COCH_3$), 1.90 (s, 6H, $COCH_3$), 1.89 (s, 3H, $COCH_3$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.3, 170.2, 170.15, 170.13, 170.1, 169.7, 169.1, 166.2, 165.8, 164.8, 155.9, 154.0, 138.1, 136.1, 133.6, 133.5, 133.3, 129.83, 129.81, 129.7, 129.6, 129.5, 129.2, 128.7, 128.6, 128.57, 128.5, 128.44, 128.4, 128.3, 128.2, 128.1, 128.04, 128.0, 102.2, 102.0, 100.3, 99.0, 95.4, 75.0, 74.7, 74.3, 74.1, 74.0, 73.9, 72.5, 71.5, 71.0, 70.9, 70.5, 69.5, 69.4, 69.3, 69.1, 68.0, 67.6, 67.3, 66.5, 62.7, 62.5, 60.4, 59.1, 56.3, 54.4, 52.9, 20.2, 20.8, 20.7, 20.6, 20.5; HRMS-ESI $[M + Na]^+$ calcd for $C_{81}H_{88}Cl_3N_5O_{35}$ 1820.4219, found 1820.4216.

***N*-(Benzyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-*β*-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-3-*O*-benzoyl-6-*O*-benzyl-*β*-D-glucopyranosyl-(1→3)-4-*O*-acetyl-2,6-di-*O*-benzoyl-*β*-D-galactopyranosyl-(1→3)-2-acetamido-2-deoxy-4,6-di-*O*-acetyl-*α*-D-galactopyranosyl)-L-serine methylester (19)**

To a solution of compound **18** (0.06 g, 0.03 mmol) in EtOAc (3.4 mL) was added zinc (1.0 g, 16.04 mmol), acetic acid (550 µL, 9.65 mmol) and the solution was stirred at rt for 1 h. The reaction mixture was filtered through Celite and washed with EtOAc. To the filtrate was added Ac_2O (605 µL) and the resulting mixture was stirred at rt for 15 h. The reaction mixture was concentrated and chromatographed on silica gel (90% ethyl acetate: pet ether) to give **19** as a colourless foam (0.04 g, 75%): $[\alpha]^{20}_D$ = +10.4 (*c* 0.1, $CHCl_3$); IR ($CHCl_3$) *v* 3686, 3019, 1745, 1519, 1215, 1027, 928, 761, 669 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 8.06-8.01 (m, 4H, ArH), 7.93 (d, *J* = 7.3 Hz, 2H, ArH), 7.62-7.31 (m, 19H, ArH), 5.60-5.59 (m, 2H), 5.44-5.38 (m, 3H), 5.22-5.05 (m, 4H), 4.99-4.84 (m, 3H), 4.75-4.69 (m, 4H), 4.63 (d, *J* = 8.2 Hz, 1H), 4.45-4.31 (m, 6H), 4.07-3.74 (m, 10H), 3.71-3.68 (m, 2H), 3.65 (s, 3H, $COOCH_3$), 3.57-3.30 (m, 4H), 2.17 (s, 3H, $COCH_3$), 2.03 (s, 3H, $COCH_3$), 2.00 (s, 3H, $COCH_3$), 1.94 (s, 3H, $COCH_3$), 1.90 (s, 3H, $COCH_3$), 1.89 (s, 6H, $COCH_3$), 1.42 (s, 3H, CH_3), 1.35 (s, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.6, 170.5, 170.3, 170.26, 170.20, 170.16, 170.09, 169.2, 166.3, 166.1, 164.8, 155.9, 138.1, 136.0, 134.1, 133.5, 133.3, 129.9, 129.8, 129.6, 129.4, 129.2, 128.8, 128.7, 128.63, 128.6, 128.54, 128.5, 128.1, 128.03, 128.0, 102.0, 100.8, 100.3, 98.5, 71.8, 70.9, 70.5, 69.4, 69.3, 69.2, 68.4, 67.9, 67.8, 67.5, 66.6, 63.0, 62.6, 60.4, 54.6, 54.4, 52.7, 49.0, 22.7, 22.5, 20.9, 20.8, 20.7, 20.6, 20.5; HRMS-ESI $[M + Na]^+$ calcd for $C_{82}H_{93}N_3O_{35}$ 1702.5482, found 1702.5481.

***N*-(Benzyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-*β*-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-3-*O*-benzoyl-*β*-D-glucopyranosyl-(1→3)-4-*O*-acetyl-2,6-di-*O*-benzoyl-*β*-D-galactopyranosyl-(1→3)-2-acetamido-2-deoxy-4,6-di-*O*-acetyl-*α*-D-galactopyranosyl)-L-serine methylester (20)**

A solution of $NaBrO_3$ (0.04 g, 0.32 mmol) in water (1.2 mL) was added to a clear solution of **19** (0.09 g, 0.05 mmol) in EtOAc (3.0 mL). To this biphasic layer a solution of $Na_2S_2O_4$ (0.05 g, 0.32 mmol) in water (1.8 mL) was added drop wise over 5 min. After 45 min, reaction mixture was quenched with aq. $Na_2S_2O_3$ solution and extracted with EtOAc (3 × 30 mL). Combined organic layers dried over Na_2SO_4, concentrated and chromatographed on silica gel (5% methanol: ethyl acetate) to afford the desired product **20** as a foam (70 mg, 82%): $[\alpha]^{20}_D$ = −6.4 (*c* 0.1, $CHCl_3$); IR ($CHCl_3$) *v* 3415, 2931, 1746, 1656, 1218, 1071, 917, 769, 713, 481 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.10-7.97 (m, 6H, ArH), 7.60-7.39 (m, 14H, ArH), 5.8 (bs, 1H), 5.60 (d, *J* = 7.7 Hz, 1H, NH), 5.46-5.35 (m, 4H), 5.16-5.03 (m, 5H), 4.89-4.86 (m, 3H), 4.75-4.74 (m, 1H), 4.61-4.56 (m, 2H), 4.49-4.34 (m, 2H), 4.20-4.02 (m, 5H), 3.96-3.72 (m, 8H), 3.67 (s, 3H, $COOCH_3$), 3.64-3.59 (m, 1H), 3.47-3.36 (m, 3H), 2.25 (s, 3H, $COCH_3$), 2.09 (s, 3H, $COCH_3$), 2.08 (s, 3H, $COCH_3$), 2.03 (s, 3H, $COCH_3$), 1.98 (s, 3H, $COCH_3$), 1.94 (s, 3H, $COCH_3$), 1.94 (s, 3H, $COCH_3$), 1.44 (s, 3H, CH_3), 1.03 (s, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$) δ 171.1, 170.6, 170.3, 170.2, 170.1, 170.0, 169.7, 166.2, 165.9, 164.8, 155.9, 134.0, 133.7, 133.6, 133.5, 130.2, 129.9, 129.8, 129.7, 129.5, 129.4, 129.1, 128.8, 128.7, 128.6, 128.5, 101.8, 101.2, 100.9, 98.5, 71.3, 71.0, 70.6, 69.6, 69.4, 69.3, 68.3, 67.9, 67.5, 66.5, 63.0, 61.5, 60.1, 55.3, 54.4, 52.8, 49.0, 22.5, 22.3,

21.3, 20.9, 20.8, 20.73, 20.7, 20.6; HRMS-ESI $[M + Na]^+$ calcd for $C_{75}H_{87}N_3O_{35}$ 1612.5007, found 1612.5012.

***N*-(Benzyloxycarbonyl)-*O*-(*β*-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-6-sulfo-*β*-D-glucopyranosyl-(1→3)-*β*-D-galactopyranosyl-(1→3)-2-acetamide-2-deoxy-*α*-D-galactopyranosyl)-L-serine (1)**

To a solution of **20** (0.03 mg, 0.02 mmol) in dry pyridine (1 mL) was added SO_3•NEt_3 (40 mg, 0.22 mmol). The reaction was stirred for 48 h at rt. After complete consumption of the starting material, the reaction mixture was concentrated and co-evaporated with toluene (3 × 5 mL). A solution of NaOMe (24 mg) in MeOH (1 mL) was added to the crude product in MeOH (1 mL) and water (1 mL) at 55 °C (pH 10). After stirring at the same temperature for 30 h, the reaction mixture was neutralized by adding AcOH until the pH adjusted to 6. The reaction mixture was then concentrated and purified by silica gel chromatography (7:2:1 ethyl acetate: MeOH: H_2O) to give **1** as a waxy solid (17 mg, 80%): ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.33-7.30 (m, 5H, ArH), 4.60-4.58 (m, 2H), 4.38 (d, J = 7.8 Hz, 1H), 4.32 (d, J = 7.8 Hz, 1H), 4.28-4.25 (m, 1H), 4.20-4.16 (m, 2H), 4.10-4.01 (m, 3H), 3.91 (d, J = 3.0 Hz, 1H), 3.90-3.89 (m, 1H), 3.86-3.79 (m, 3H), 3.76-3.61 (m, 5H), 3.60-3.44 (m, 13H), 3.43-3.38 (m, 3H), 1.89 (s, 3H, CH_3), 1.79 (s, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$) δ 182.5, 181.5, 174.9, 174.6, 157.7, 136.5, 128.8, 128.7, 128.4, 127.8, 127.7, 104.7, 102.7, 98.0, 82.3, 75.4, 74.6, 72.6, 72.5, 72.2, 71.0, 70.8, 69.7, 68.8, 68.7, 68.6, 68.4, 67.1, 66.6, 61.1, 55.2, 48.5, 23.3, 22.3, 22.2, 20.1; HRMS-ESI $[M + Na]^+$ calcd for $C_{39}H_{58}N_3NaO_{28}S$ 1094.2717, found 1094.2714.

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Notes and references

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Table of Contents Graphics

Total Synthesis of MECA-79

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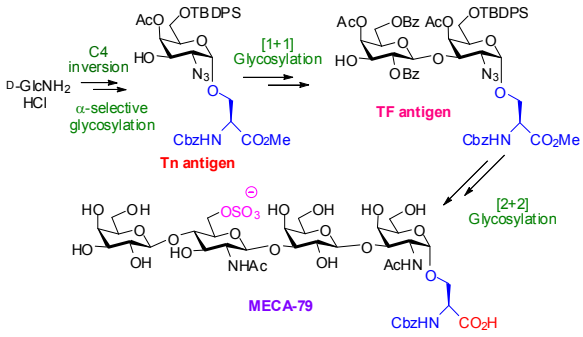

MECA-79 antigen is a sulfated mucin type core-1 extended *O*-glycan which is a potential anti-inflammatory agent. Herein we report a total synthesis of MECA-79 via a convergent [2+2] glycosylation route. The synthesis relies on efficient transformation of D-glucosamine into the orthogonally protected Tn antigen derivative and its elaboration into TF antigen *en route* to MECA-79.