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Total synthesis of the heptasaccharide repeating unit of the iron-binding exopolysaccharide secreted by *Klebsiella oxytoca* BAS-10

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ABSTRACT

The first total synthesis of a heptasaccharide found in the iron-binding exopolysaccharide produced by *Klebsiella oxytoca* BAS-10 has been achieved in excellent yield using a block synthetic strategy. A trisaccharide glycosyl donor was stereoselectively coupled with a tetrasaccharide glycosyl acceptor using the trichloroacetimidate activation procedure. The yields and stereo outcome were excellent in each step of glycosylation. A late stage oxidation protocol was adopted for the oxidation of the primary hydroxyl group to the carboxylic functionality while keeping a secondary hydroxyl group unaffected.

1. Introduction

Klebsiella bacteria are gram-negative opportunistic pathogens which cause several infections such as pneumonia, septicemia, and urinary tract infections.[1] Among several species of *Klebsiella*, *Klebsiella oxytoca* infections are the second most frequently found in several patients after *Klebsiella pneumonia* infections.[2] *K. Oxytoca* produces exopolysaccharides with several pharmaceutical and environmental importances.[3] Very recently, the structure of an exopolysaccharide secreted by *Klebsiella oxycota* BAS-10 containing two glucuronic acids at the non-reducing terminus has been reported by De Castro et al.[4] This particular exopolysaccharide has shown strong iron binding affinity[5] useful in the preparation of bacterial biopolymers. It is believed that due to the presence of the glucuronic acid moieties it could form stable complexes with the metal ions through strong electrostatic attractions between metal ion and the negatively charged uronic acid moieties.[6] In addition to the pharmaceutical properties of biopolymers, they can also be successfully applied to the selective removal of heavy metals and radioactive metals from water and soils.[7]

The preparation of carbohydrate-derived molecules for their use in therapeutics is a thrust area in medicinal chemistry research. Although the oligosaccharides can be isolated from natural sources, their bio- and pharmaceutical evaluation require reasonably higher quantities of materials, which cannot be managed by the isolation from natural sources. Therefore, concise chemical synthetic strategies are always useful in gaining access to large quantities of oligosaccharides in pure form.[8] In this report, we describe a concise total synthesis of the iron-binding heptasaccharide repeating unit found in the exopolysaccharide secreted by *K. oxytoca* BAS-10 (Fig. 1). The 4-methoxyphenyl group has been chosen as the anomeric protection because of its easy removal whenever we are required to modify the synthesized heptasaccharide moiety to prepare the glycoconjugate molecule.

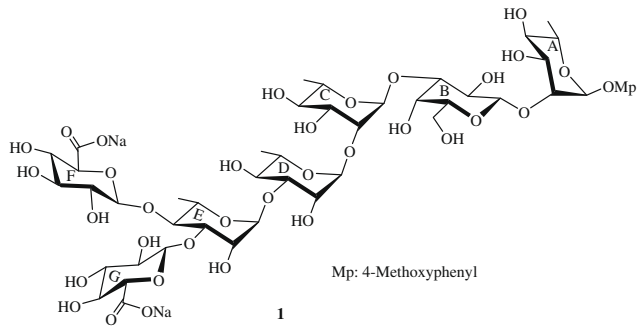

Figure 1. Structure of the synthesized heptasaccharide antigen from *Klebsiella oxytoca* BAS-10 as its 4-methoxyphenyl glycoside **1**.

2. Results and discussion

In order to synthesize the target heptasaccharide **1** as its 4-methoxyphenyl glycoside, a block synthetic strategy has been adopted. A trisaccharide trichloroacetimidate derivative **18** and a tetrasaccharide derivative **15** were prepared separately starting from a series of suitably protected monosaccharide intermediates (Schemes 1 and 2). Stereoselective glycosylation of compound **15** with compound **18** furnished a heptasaccharide derivative **19**, which was further transformed into the target heptasaccharide **1** after selective oxidation of the primary hydroxyl groups and removal of protecting groups (Scheme 2). For this purpose, a

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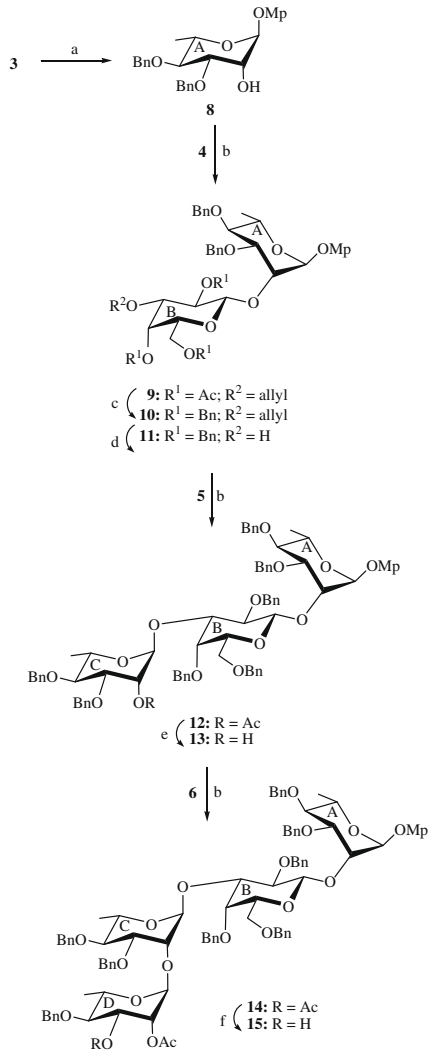
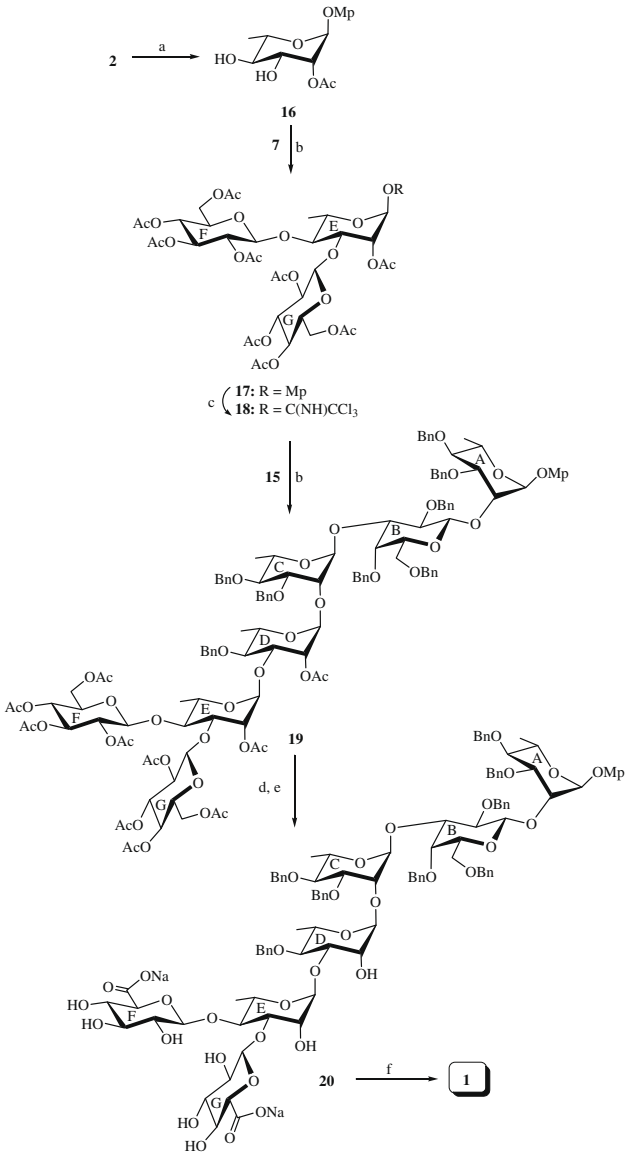


Scheme 1. Reagents and conditions: (a) (i) dibutyltin oxide, CH_3OH, 80 °C, 2 h; (ii) benzyl bromide, tetrabutylammonium bromide, DMF, room temperature, 12 h, 76%; (b) NIS, TMSOTf, MS 4 Å, CH_2Cl_2, −45 °C, 1 h, 90% for compound **9**, 88% for compound **12**, 85% for compound **14**; (c) benzyl bromide, NaOH, TBAB, THF, room temperature, 6 h, 91%; (d) $PdCl_2$, CH_3OH, room temperature, 6 h, 76%; (e) CH_3ONa, CH_3OH, room temperature, 3 h, 96%; (f) (i) CH_3ONa, CH_3OH, room temperature, 3 h; (ii) $CH_3CH(OCH_3)_2$, *p*-TsOH, DMF, 2 h; (iii) 80% aq AcOH, room temperature, 30 min, 91%.



Scheme 2. Reagents and conditions: (a) (i) $CH_3CH(OCH_3)_2$, *p*-TsOH, DMF, room temperature, 2 h; (ii) 80% aq AcOH, room temperature, 30 min, 84%; (b) TMSOTf, MS 4 Å, CH_2Cl_2, −10 °C, 3 h, 64% for compound **17**, 68% for compound **19**; (c) (i) ceric ammonium nitrate, CH_3CN–H_2O (4:1), room temperature, 3 h; (ii) CCl_3CN, DBU, CH_2Cl_2, −10 °C, 4 h, 70%; (d) CH_3ONa, CH_3OH, room temperature, 5 h; (e) (i) NaBr, CH_2Cl_2, H_2O, TBAB, TEMPO, $NaHCO_3$, NaOCl, 0–5 °C, 3 h; (ii) *tert*-butanol, 2-methyl-but-2-ene, $NaClO_2$, NaH_2PO_4, room temperature, 3 h; (f) H_2, 20% $Pd(OH)_2$–C, CH_3OH, room temperature, 24 h, overall yield 59%.

number of differentially protected monosaccharide intermediates **2**,[9] **3**,[10] **4**,[11] **5**,[12] **6**,[13] and **7**[14] were prepared from commercially available reducing sugars using earlier reported reaction conditions (Fig. 2).

Selective benzylation via the formation of stannylidene acetal **3**[10] furnished 4-methoxyphenyl 3,4-di-*O*-benzyl-α-L-rhamnopyranoside **8** in 76% yield. Stereoselective glycosylation of compound **8** with thioglycoside derivative **4**,[11] in the presence of *N*-iodosuccinimide (NIS) and trimethylsilyl trifluoromethanesulfonate (TMSOTf)[15] furnished disaccharide derivative **9** in 90% yield. The exclusive formation of the 1,2-*trans*-glycosidic bond was achieved due to the presence of a neighboring group *O*-acetoxy at the C-2 position of compound **4**. The presence of signals in the ^1H NMR [δ 5.50 (br s, H-1_A), 4.51 (d, J = 8.0 Hz, H-1_B)] and in the ^{13}C NMR [δ 103.4 (C-1_B), 97.9 (C-1_A)] confirmed the formation of compound **9**. Compound **9** was transformed to compound **10** using one-pot deacetylation–benzylation reaction conditions[16] in 91% yield. Removal of the allyl ether protection from compound **10** was carried out using palladium

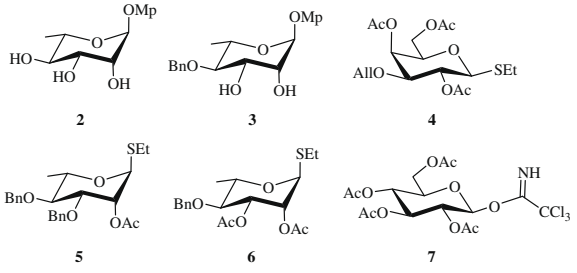


Figure 2. Suitably protected monosaccharide intermediates used in the construction of heptasaccharide **1**.

chloride[17] to give compound **11** in 76% yield. The stereoselective glycosylation of the disaccharide acceptor **11** with thioglycoside

derivative **5**[12] in the presence of NIS-TMSOTf[15] as the glycosylation promoter afforded trisaccharide derivative **12** in 88% yield, which on deacetylation furnished trisaccharide acceptor **13** in 96% yield. The formation of compound **12** was confirmed from spectroscopic analysis [appearance of a new signal at δ 5.31 (br s, 1H, H-1_C) in the ^{1}H NMR and signals at δ 104.9 (C-1_B), 100.5 (C-1_C), and 98.2 (C-1_A) in the ^{13}C NMR spectra]. Glycosylation of trisaccharide derivative **13** with thioglycoside derivative **6**[13] was carried out using NIS-TMSOTf[15] as the thiophilic activator to furnish the tetrasaccharide derivative **14** in 85% yield. The presence of an *O*-acetoxy group at the C-2 position of compound **6** directed the exclusive formation of compound **14**, which was supported by its spectroscopic analysis [appearance of a signal at δ 4.76 (br s, H-1_D) in ^{1}H NMR and signals at δ 104.9 (C-1_B), 100.1 (C-1_C), 98.9 (C-1_D), and 98.2 (C-1_A) in the ^{13}C NMR spectra]. Removal of the *O*-acetyl groups in compound **14** using saponification followed by selective acetylation of the C-2 hydroxy group of the terminal L-rhamnosyl moiety via formation of an orthoester[18] using triethyl orthoacetate resulted in tetrasaccharide acceptor **15** in 91% overall yield (Scheme 1).

In another experiment, compound **2**[9] was selectively 2-O-acetylated via orthoester formation[18] using triethyl orthoacetate in the presence of *p*-toluene sulfonic acid to give diol derivative **16** in 84% yield. Two consecutive stereoselective 1,2-*trans*-glycosylations of the diol derivative **16** with trichloroacetimidate derivative **7**[14] using Schmidt's glycosylation condition[19] furnished trisaccharide derivative **17** in 64% yield, which was confirmed from its spectroscopic analysis [signals at δ 5.24 (br s, H-1_E), 4.74 (d, J = 7.7 Hz, H-1_F), and 4.69 (d, J = 8.0 Hz, H-1_G) in the ^{1}H NMR and at δ 101.1 (C-1_F), 99.2 (C-1_G), and 96.4 (C-1_E) in the ^{13}C NMR spectra]. Compound **17** was treated with ceric ammonium nitrate (CAN)[10] in moist CH_3CN to give the trisaccharide hemiacetal, which was treated with trichloroacetonitrile in the presence of DBU[20] to give trisaccharide trichloroacetimidate derivative **18** in 70% overall yield. The final stereoselective glycosylation of the trisaccharide trichloroacetimidate derivative **18** with the tetrasaccharide derivative **15** under Schmidt's glycosylation conditions[19] furnished heptasaccharide derivative **19** in 68% yield, which was confirmed from its spectroscopic analysis [presence of signals at δ 104.9 (C-1_B), 100.6 (C-1_F), 100.0 (C-1_C), 98.8 (C-1_G), 98.6 (C-1_D), 98.4 (C-1_E), and 98.1 (C-1_A) in the ^{13}C NMR spectrum]. Saponification of the heptasaccharide derivative **19** using sodium methoxide followed by TEMPO-mediated selective oxidation[21] of the polyhydroxylated product in phase transfer reaction conditions furnished two D-glucuronic acids containing the heptasaccharide derivative, which was hydrogenolized over Pearlman's catalyst[22] to give target heptasaccharide **1** as its 4-methoxyphenyl glycoside in 59% overall yield. It is worth mentioning that late stage phase transfer TEMPO-mediated oxidation conditions have been adopted for the selective oxidation of two primary hydroxyl groups in the presence of secondary hydroxyl groups, which furnished D-glucuronic acid containing heptasaccharide derivative **20** quite efficiently. The oxidized product **20** was directly subjected to hydrogenation to give target heptasaccharide **1**. The stereochemical assignments of the glycosyl linkages in compound **1** were confirmed from its 1D and 2D NMR spectra [signals at δ 5.58 (br s, H-1_A), 5.14 (br s, H-1_C), 4.95 (br s, H-1_E), 4.85 (br s, H-1_D), 4.67 (d, J = 7.9 Hz, H-1_G), 4.52 (d, J = 7.5 Hz, H-1_F), and 4.43 (d, J = 7.8 Hz, H-1_B) in the ^{1}H NMR and at δ 107.5 (C-1_B), 105.1 (C-1_F), 104.3 (C-1_G), 103.7 (C-1_D), 103.4 (C-1_E), 102.3 (C-1_C), and 99.7 (C-1_A) in the ^{13}C NMR spectra] and mass spectroscopic analysis (Scheme 2). The 4-methoxyphenyl group used as the anomeric-protecting group remained unaffected under the oxidation and hydrogenation conditions, which was confirmed from the appearance of signals in the NMR spectra of compound **1** [δ 6.88 (d, J = 9.0 Hz, 2H), 6.74 (d, J = 9.0 Hz, 2H) in ^{1}H NMR and at δ 156.5, 151.7, 118.9 (2C), and 115.6 (2C)].

3. Conclusion

In conclusion, the total synthesis of a heptasaccharide containing two D-glucuronic acid moieties found in the iron-binding exopolysaccharide produced by *K. oxytoca* BAS-10 was achieved in excellent yield using a block synthetic strategy. Most of the glycosylation steps are highly stereoselective and reproducible for scale-up. A late stage selective TEMPO-mediated oxidation of the primary hydroxyl groups was achieved using a two-step, one-pot phase transfer oxidation protocol without affecting the secondary hydroxyl groups present in the molecule. The 4-methoxybenzyl group was chosen as the temporary protecting group at the reducing end for its easy removal whenever necessary.

4. Experimental

4.1. General methods

All reactions were monitored by thin layer chromatography over silica gel-coated TLC plates. The spots on TLC were visualized by warming ceric sulfate (2% $Ce(SO_4)_2$ in 2 N H_2SO_4) sprayed plates on a hot plate. Silica gel 230–400 mesh was used for column chromatography. ^{1}H and ^{13}C NMR, 2D COSY, and HMQC spectra were recorded on a Brucker 500 MHz NMR spectrometer (DRX-500) using $CDCl_3$ and D_2O as solvents and TMS as internal reference unless stated otherwise. Chemical shift value is expressed in δ ppm. ESI-MS were recorded on a MICROMASS QUTTRO II triple quadrupole mass spectrometer. Elementary analysis was carried out on Carlo ERBA-1108 analyzer. Optical rotations were measured at 25 °C on a Rudolf Autopol III polarimeter. Commercially available grades of organic solvents of adequate purity were used in many reactions.

4.1.1. 4-Methoxyphenyl 3,4-di-*O*-benzyl-α-L-rhamnopyranoside 8

To a solution of compound **3** (6 g, 16.65 mmol) in anhydrous CH_3OH (120 mL) was added dibutyltin(IV) oxide (4.9 g, 19.68 mmol) and the reaction mixture was allowed to stir at 80 °C for 2 h. The solvents were removed under reduced pressure and to a solution of the crude stannylidene acetal derivative in anhydrous DMF (50 mL) were added benzyl bromide (4 mL, 33.63 mmol) and tetrabutylammonium bromide (3.3 g, 1 mmol). The reaction mixture was allowed to stir at room temperature for 12 h and the solvents were removed under reduced pressure. The crude product was dissolved in EtOAc (150 mL) and the organic layer was washed with 1 M HCl, satd $NaHCO_3$, and water successively. The organic layer was dried (Na_2SO_4) and concentrated to give the crude product, which was purified over SiO_2 using hexane–EtOAc (4:1) as eluant to give pure compound **8** (5.7 g, 76%); Oil; $[\alpha]_D^{25} = -43$ (*c* 1.5, $CHCl_3$); IR (neat): 2932, 2368, 1654, 1513, 1457, 1389, 1227, 1100, 1043, 774, 699 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$):δ 7.58–7.19 (m, 10H, Ar-H), 6.98 (d, J = 9.0 Hz, 2H, Ar-H), 6.83 (d, J = 9.0 Hz, 2H, Ar-H), 5.44 (br s, 1H, H-1), 4.94 (d, J = 11.0 Hz, 1H, PhCH_2), 4.78 (br s, 2H, PhCH_2), 4.70 (d, J = 11.0 Hz, 1H, PhCH_2), 4.20 (br s, 1H, H-2), 4.05 (dd, J = 9.4, 2.9 Hz, 1H, H-3), 3.92–3.83 (m, 1H, H-5), 3.80 (s, 3H, OCH_3), 3.53 (t, J = 9.4 Hz, 1H, H-4), 1.30 (d, J = 6.0 Hz, 3H, CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 154.9–114.6 (Ar-C), 96.2 (C-1), 79.9 (2C, 2PhCH_2), 75.4 (C-4), 72.2 (C-2), 68.6 (C-5), 68.0 (C-3), 55.5 (OCH_3), 17.9 (CCH_3); ESI-MS: 473.2 $[M+Na]^+$; Anal. Calcd for $C_{27}H_{30}O_6$ (450.20): C, 71.98; H, 6.71. Found: C, 71.80; H, 6.95.

4.1.2. 4-Methoxyphenyl (2,4,6-tri-*O*-acetyl-3-*O*-allyl-β-D-galactopyranosyl)-(1→2)-3,4-di-*O*-benzyl-α-L-rhamnopyranoside 9

To a solution of compound **8** (5 g, 11.1 mmol) and compound **4** (5.2 g, 13.32 mmol) in anhydrous CH_2Cl_2 (70 mL) was added MS

4 Å (10 g) and the reaction mixture was allowed to stir at room temperature for 1 h under argon. The reaction mixture was cooled to −45 °C and *N*-iodosuccinimide (NIS; 3.5 g, 15.55 mmol) followed by trimethylsilyltrifluoromethane sulfonate (TMSOTf; 30 μL) were added to it and the reaction mixture was allowed to stir at same temperature for 1 h. The reaction mixture was quenched with Et_3N (0.1 mL), filtered through a Celite® bed and washed with CH_2Cl_2 (200 mL). The organic layer was washed with 5% aq $Na_2S_2O_3$, satd aq $NaHCO_3$, and water, dried (Na_2SO_4) and concentrated to dryness. The crude product was purified over SiO_2 using hexane–EtOAc (5:1) as eluant to give pure **9** (7.8 g, 90%); Oil; $[\alpha]_D^{25} = +12.6$ (*c* 1.5, $CHCl_3$); IR (neat): 3020, 2361, 1746, 1506, 1371, 1217, 1042, 761 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 7.41–7.27 (m, 10H, Ar-H), 6.94 (d, *J* = 9.0 Hz, 2H, Ar-H), 6.75 (d, *J* = 9.0 Hz, 2H, Ar-H), 5.85–5.75 (m, 1H, C*H*=CH_2), 5.50 (br s, 1H, H-1_A), 5.39 (d, *J* = 2.9 Hz, 1H, H-4_B), 5.32–5.19 (m, 2H, CH_2=CH), 5.14 (dd, *J* = 8.0 Hz each, 1H, H-2_B), 4.87 (d, *J* = 10.9 Hz, 1H, PhCH_2), 4.79–4.69 (m, ABq, *J* = 11.5 Hz, 2H, PhCH_2), 4.63 (d, *J* = 10.9 Hz, 1H, PhCH_2), 4.51 (d, *J* = 8.0 Hz, 1H, H-1_B), 4.17–4.09 (m, 3H, H-6_{abB}, OCH_{2a}), 4.03–3.92 (m, 3H, H-2_A, H-5_A, OCH_{2b}), 3.76 (s, 3H, OCH_3), 3.80–3.73 (m, 2H, H-4_A, H-5_B), 3.53 (dd, *J* = 8.2, 2.0 Hz, 1H, H-3_B), 3.51–3.45 (m, 1H, H-3_A), 2.18, 1.92, 1.90 (3 s, 9H, 3COCH_3), 1.28 (d, *J* = 6.1 Hz, 3H, CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 170.2, 170.1, 169.2 (3 *C*OCH_3), 154.8–138.5 (Ar-C), 134.2 (CH_2=*C*H), 128.4–114.6 (Ar-C), 117.2 (*C*H_2=CH), 103.4 (C-1_B), 97.9 (C-1_A), 80.3 (C-3_A), 79.5 (C-5_A), 77.5 (C-2_A), 76.3 (C-3_A), 75.4 (PhCH_2), 72.8 (PhCH_2), 70.8 (C-4_A), 70.6 (OCH_2), 70.5 (C-2_B), 68.7 (C-5_B), 65.9 (C-4_B), 61.9 (C-6_B), 55.5 (OCH_3), 20.8, 20.7, 20.4 (3COCH_3), 17.9 (CCH_3); ESI-MS: 801.3 $[M+Na]^+$; Anal. Calcd for $C_{42}H_{50}O_{14}$ (778.32): C, 64.77; H, 6.47. Found: C, 64.60; H, 6.72.

4.1.3. 4-Methoxyphenyl (3-*O*-allyl-2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(1→2)-3,4-di-*O*-benzyl-α-L-rhamnopyranoside 10

To a solution of compound **9** (6 g, 7.70 mmol) in anhydrous THF (80 mL) were added powdered NaOH (2 g, 50 mmol), benzyl bromide (4.5 mL, 37.84 mmol), and tetrabutylammonium bromide (500 mg, 1.55 mmol) and the reaction mixture was allowed to stir briskly at room temperature for 6 h. The reaction mixture was quenched with CH_3OH (5 mL) and concentrated under reduced pressure. The crude mass was dissolved in CH_2Cl_2 (200 mL) and the organic layer was washed with water, dried (Na_2SO_4), and concentrated. The crude product was purified over SiO_2 using hexane–EtOAc (8:1) as eluant to give pure compound **10** (6.5 g, 91%); Oil; $[\alpha]_D^{25} = -38.6$ (*c* 1.5, $CHCl_3$); IR (neat): 3020, 2360, 1598, 1507, 1216, 1074, 928, 760 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 7.43–7.15 (m, 25H, Ar-H), 6.90 (d, *J* = 9.0 Hz, 2H, Ar-H), 6.74 (d, *J* = 9.0 Hz, 2H, Ar-H), 5.97–5.85 (m, 1H, C*H*=CH_2), 5.54 (br s, 1H, H-1_A), 5.33–5.14 (m, 3H, CH_2=CH, PhCH_2), 4.90 (d, *J* = 11.6 Hz, 1H, PhCH_2), 4.80–4.65 (m, 5H, H-1_B, PhCH_2), 4.58 (d, *J* = 11.6 Hz, 1H, PhCH_2), 4.44 (d, *J* = 10.9 Hz, 1H, PhCH_2), 4.39–4.30 (ABq, *J* = 11.9 Hz, 2H, PhCH_2), 4.28–4.12 (m, 3H, H-2_A, OCH_2), 4.05 (dd, *J* = 9.4, 2.8 Hz, 1H, H-3_A), 3.82–3.75 (m, 3H, H-2_B, H-4_B, H-5_A), 3.73 (s, 3H, OCH_3), 3.55 (t, *J* = 9.4 Hz each, 1H, H-4_A), 3.51–3.42 (m, 3H, H-4_A, H-5_B, H-6_{abB}), 3.35 (dd, *J* = 9.7, 2.8 Hz, 1H, H-3_B), 1.23 (d, *J* = 6.1 Hz, 3H, CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 154.9–137.9 (Ar-C), 135.2 (*C*H=CH_2), 128.4–114.5 (Ar-C), 116.4 (CH=*C*H_2), 104.7 (C-1_B), 98.2 (C-1_A), 81.7 (C-3_B), 80.5 (C-4_A), 79.9 (C-3_A), 79.5 (C-2_B), 75.3 (PhCH_2), 74.8 (PhCH_2), 74.7 (C-2_A), 74.6 (PhCH_2), 73.8 (C-5_A), 73.6 (C-5_B), 73.5 (PhCH_2), 72.3 (PhCH_2), 72.2 (OCH_2), 69.1 (C-6_B), 68.6 (C-4_B), 55.5 (OCH_3), 18.0 (CCH_3); ESI-MS: 945.4 $[M+Na]^+$; Anal. Calcd for $C_{57}H_{62}O_{11}$ (922.43): C, 74.16; H, 6.77. Found: C, 74.0; H, 7.98.

4.1.4. 4-Methoxyphenyl (2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(12)-3,4-di-*O*-benzyl-α-L-rhamnopyranoside 11

To a solution of compound **10** (6 g, 6.5 mmol) in anhydrous CH_3OH (80 mL) was added $PdCl_2$ (700 mg, 3.95 mmol) and the reaction mixture was allowed to stir at room temperature for 6 h. The reaction mixture was concentrated to dryness under reduced pressure and the crude product was purified over SiO_2 using hexane–EtOAc (5:1) as eluant to give pure compound **11** (4.4 g, 76%); Oil; $[\alpha]_D^{25} = -5.9$ (*c* 1.5, $CHCl_3$); IR (neat): 3460, 3020, 2926, 2359, 1595, 1505, 1216, 1074, 927, 761 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 7.41–7.16 (m, 25H, Ar-H), 6.90 (d, *J* = 9.0 Hz, 2H, Ar-H), 6.74 (d, *J* = 9.0 Hz, 2H, Ar-H), 5.56 (br s, 1H, H-1_A), 5.25 (d, *J* = 11.2 Hz, 1H, PhCH_2), 4.91–4.46 (m, 8H, H-1_B, PhCH_2), 4.42–4.32 (m, ABq, *J* = 11.9 Hz, 2H, PhCH_2), 4.24–4.22 (m, 1H, H-2_A), 4.07 (dd, *J* = 9.5, 2.3 Hz, 1H, H-3_A), 3.85–3.77 (m, 2H, H-4_B, H-5_A), 3.74 (s, 3H, OCH_3), 3.68–3.59 (m, 2H, H-2_B, H-4_A), 3.56–3.50 (m, 4H, H-3_B, H-5_B, H-6_{abB}), 1.24 (d, *J* = 6.1 Hz, 3H, CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 154.7–114.5 (Ar-C), 104.8 (C-1_B), 98.2 (C-1_A), 80.4 (C-3_B), 79.9 (C-3_A), 79.3 (C-2_B), 75.4 (C-2_A), 75.3 (C-5_A), 75.2 (PhCH_2), 74.9 (PhCH_2), 74.4 (PhCH_2), 74.0 (C-4_A), 73.8 (C-5_B), 73.5 (PhCH_2), 72.5 (PhCH_2), 69.0 (C-6_B), 68.5 (C-4_B), 55.5 (OCH_3), 18.0 (CCH_3); ESI-MS: 905.4 $[M+Na]^+$; Anal. Calcd for $C_{54}H_{58}O_{11}$ (882.40): C, 73.45; H, 6.62. Found: C, 73.20; H, 6.90.

4.1.5. 4-Methoxyphenyl (2-*O*-acetyl-3,4-di-*O*-benzyl-α-L-rhamnopyranosyl)-(13)-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(12)-3,4-di-*O*-benzyl-α-L-rhamnopyranoside 12

To a solution of compound **11** (4 g, 4.53 mmol) and compound **5** (2.3 g, 5.34 mmol) in anhydrous CH_2Cl_2 (30 mL) was added MS 4 Å (3 g) and the reaction mixture was allowed to stir at room temperature for 1 h under argon. The reaction mixture was cooled to −45 °C and *N*-iodosuccinimide (NIS; 1.4 g, 6.22 mmol) followed by trimethylsilyltrifluoromethane sulfonate (TMSOTf; 20 μL) were added to it and the reaction mixture was allowed to stir at the same temperature for 1 h. The reaction mixture was quenched with Et_3N (0.1 mL), filtered through a Celite® bed, and washed with CH_2Cl_2 (150 mL). The organic layer was washed with 5% aq $Na_2S_2O_3$, satd aq $NaHCO_3$, and water, dried (Na_2SO_4) and concentrated to dryness. The crude product was purified over SiO_2 using hexane–EtOAc (5:1) as eluant to give pure **12** (5 g, 88%); Oil; $[\alpha]_D^{25} = +2.6$ (*c* 1.5, $CHCl_3$); IR (neat): 3019, 2925, 2360, 1739, 1594, 1369, 1216, 1073, 926, 761 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 7.46–7.15 (m, 35H, Ar-H), 6.94 (d, *J* = 9.0 Hz, 2H, Ar-H), 6.78 (d, *J* = 9.0 Hz, 2H, Ar-H), 5.59 (br s, 1H, H-1_A), 5.58–5.57 (m, 1H, H-2_C), 5.31 (br s, 1H, H-1_C), 5.26 (d, *J* = 10.8 Hz, 1H, PhCH_2), 4.92 (d, *J* = 11.1 Hz, 1H, PhCH_2), 4.84–4.44 (m, 10H, H-1_B, PhCH_2), 4.39–4.26 (m, 3H, PhCH_2), 4.22–4.20 (m, 1H, H-2_A), 4.10 (dd, *J* = 9.4, 2.8 Hz, 1H, H-3_A), 3.91 (dd, *J* = 8.0 Hz each, 1H, H-2_B), 3.87–3.79 (m, 4H, H-3_B, H-3_C, H-5_A, H-5_C), 3.77 (s, 3H, OCH_3), 3.71 (br s, 1H, H-4_B), 3.57 (dd, *J* = 9.5 Hz each, 1H, H-4_A), 3.56–3.50 (m, 3H, H-5_B, H-6_{abB}), 3.39 (t, *J* = 9.4 Hz each, 1H, H-4_C), 2.12 (s, 3H, COCH_3), 1.33, 1.25 (2 d, *J* = 6.1 Hz, 6H, 2CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 169.6 (*C*OCH_3), 154.7–114.5 (Ar-C), 104.9 (C-1_B), 98.8 (C-1_C), 98.3 (C-1_A), 80.4 (C-4_A), 79.9 (C-3_A), 79.7 (C-4_C), 79.2 (C-2_B), 77.8 (C-3_B), 76.9 (C-3_C), 75.9 (C-4_B), 75.4 (PhCH_2), 75.3 (C-2_A), 75.2 (PhCH_2), 75.0 (PhCH_2), 74.4 (PhCH_2), 73.7 (C-5_B), 73.5 (PhCH_2), 72.5 (PhCH_2), 71.4 (PhCH_2), 68.9 (C-6_B), 68.6 (C-5_A), 68.5 (C-5_C), 68.1 (C-2_C), 55.4 (OCH_3), 20.9 (COCH_3), 18.2, 18.0 (2CCH_3); ESI-MS: 1273.6 $[M+Na]^+$; Anal. Calcd for $C_{76}H_{82}O_{16}$ (1250.56): C, 72.94; H, 6.60. Found:C, 72.75; H, 6.84.

4.1.6. 4-Methoxyphenyl (3,4-di-*O*-benzyl-α-L-rhamnopyranosyl)-(13)-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(12)-3,4-di-*O*-benzyl-α-L-rhamnopyranoside 13

A solution of compound **12** (4.5 g, 3.6 mmol) in 0.1 M CH_3ONa in CH_3OH (100 mL) was allowed to stir at room temperature for 3 h and neutralized with Amberlite IR-120 (H^+) resin. The reaction mixture was filtered and concentrated under reduced pressure to give a crude product, which was passed through a short pad of SiO_2 column using hexane–EtOAc (3:1) to give pure compound **13** (4.2 g, 96%); Oil; $[\alpha]_D^{25} = -1.7$ (*c* 1.5, $CHCl_3$); IR (neat): 3397, 3021, 2926, 2358,

1595, 1216, 1044, 928, 762 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 7.37–7.15 (m, 35H, Ar-H), 6.90 (d, J = 9.0 Hz, 2H, Ar-H), 6.73 (d, J = 9.0 Hz, 2H, Ar-H), 5.55 (br s, 1H, H-1_A), 5.24 (br s, 1H, H-1_C), 5.16 (d, J = 11.4 Hz, 1H, PhCH_2), 4.86–4.33 (m, 14H, H-1_B, PhCH_2), 4.22–4.21 (m, 1H, H-2_A), 4.05 (dd, J = 9.4, 2.8 Hz, 1H, H-3_A), 3.85–3.74 (m, 5H, H-2_B, H-2_C, H-3_C, H-5_A, H-5_C), 3.73 (s, 3H, OCH_3), 3.69 (br s, 1H, H-4_B), 3.64 (dd, J = 9.1, 3.1 Hz, 1H, H-3_B), 3.55–3.48 (m, 4H, H-4_A, H-5_B, H-6_{abB}), 3.41 (t, J = 9.4 Hz each, 1H, H-4_C), 1.26, 1.23 (2 d, J = 6.1 Hz each, 6H, 2CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 154.7–114.5 (Ar-C), 104.9 (C-1_B), 100.5 (C-1_C), 98.2 (C-1_A), 80.3 (C-4_A), 79.8 (C-3_A), 79.7 (C-3_B), 79.5 (C-2_B), 79.3 (C-4_C), 77.7 (C-3_C), 76.1 (C-4_B), 75.3 (C-2_A), 75.2 (PhCH_2), 75.1 (PhCH_2), 74.9 (PhCH_2), 74.5 (PhCH_2), 73.6 (C-5_B), 73.5 (PhCH_2), 72.3 (PhCH_2), 71.7 (PhCH_2), 68.9 (C-6_B), 68.5 (C-5_A), 68.3 (C-5_C), 68.1 (C-2_C), 55.5 (OCH_3), 18.1, 18.0 (2CCH_3); ESI-MS: 1231.5 $[M+Na]^+$; Anal. Calcd for $C_{74}H_{80}O_{15}$ (1208.55): C, 73.49; H, 6.67. Found: C, 73.32; H, 6.90.

4.1.7. 4-Methoxyphenyl (2,3-di-*O*-acetyl-4-*O*-benzyl-α-L-rhamnopyranosyl)-(12)-(3,4-di-*O*-benzyl-α-L-rhamnopyranosyl)-(13)-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(12)-3,4-di-*O*-benzyl-α-L-rhamnopyranoside 14

To a solution of compound **13** (3.5 g, 2.9 mmol) and compound **6** (1.3 g, 3.4 mmol) in anhydrous CH_2Cl_2 (25 mL) was added MS 4 Å (2 g) and the reaction mixture was allowed to stir at room temperature for 1 h under argon. The reaction mixture was cooled to −45 °C and NIS (0.9 g, 4.0 mmol) followed by TMSOTf (10 μL) were added to it and the reaction mixture was allowed to stir at same temperature for 1 h. The reaction mixture was quenched with Et_3N (0.1 mL), filtered through a Celite® bed, and washed with CH_2Cl_2 (150 mL). The organic layer was washed with 5% aq $Na_2S_2O_3$, satd aq $NaHCO_3$, and water, dried (Na_2SO_4) and concentrated to dryness. The crude product was purified over SiO_2 using hexane–EtOAc (5:1) as eluant to give pure **14** (3.8 g, 85%); Oil; $[\alpha]_D^{25} = -7.5$ (c 1.5, $CHCl_3$); IR (neat): 3020, 2925, 2360, 1749, 1593, 1429, 1216, 1081, 929, 762 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 7.40–7.16 (m, 40H, Ar-H), 6.88 (d, J = 9.0 Hz, 2H, Ar-H), 6.72 (d, J = 9.0 Hz, 2H, Ar-H), 5.52 (br s, 1H, H-1_A), 5.42–5.40 (m, 1H, H-2_D), 5.31 (dd, J = 9.6, 3.2 Hz, 1H, H-3_D), 5.21 (br s, 1H, H-1_C), 5.0 (d, J = 12.1 Hz, 1H, PhCH_2), 4.87–4.83 (m, 2H, PhCH_2), 4.76 (br s, 1H, H-1_D), 4.74–4.30 (m, 14H, H-1_B, PhCH_2), 4.23–4.21 (m, 1H, H-2_A), 4.01 (dd, J = 9.4, 3.0 Hz, 1H, H-3_A), 3.88–3.81 (m, 2H, H-2_C, H-5_D), 3.80–3.60 (m, 6H, H-2_B, H-3_B, H-4_B, H-4_C, H-5_A, H-5_C), 3.72 (s, 3H, OCH_3), 3.58–3.45 (m, 5H, H-3_C, H-4_A, H-5_B, H-6_{abB}), 3.40 (t, J = 9.4 Hz, 1H, H-4_D), 2.06, 1.96 (2 s, 6H, 2COCH_3), 1.27, 1.17, 1.12 (3 d, J = 6.1 Hz, 9H, 3CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 169.6 (COCH_3), 154.7–114.5 (Ar-C), 104.9 (C-1_B), 100.1 (C-1_C), 98.9 (C-1_D), 98.2 (C-1_A), 80.4 (C-4_A), 79.9 (C-3_B), 79.8 (C-3_A), 79.5 (C-2_B), 78.9 (C-4_D), 77.7 (C-4_C), 77.6 (C-5_D), 76.4 (C-3_C), 75.5 (C-4_B), 75.3 (PhCH_2), 75.2 (PhCH_2), 75.0 (PhCH_2), 74.9 (C-2_A), 74.8 (PhCH_2), 73.7 (C-5_B), 73.5 (2C, PhCH_2), 72.2 (PhCH_2), 71.8 (PhCH_2), 71.6 (C-3_D), 70.4 (C-2_D), 69.2 (C-5_A), 68.9 (C-6_B), 68.5 (C-5_C), 68.2 (C-2_C), 55.5 (OCH_3), 20.9 (2C, 2COCH_3), 18.2, 18.0, 17.8 (3 CCH_3); ESI-MS: 1551.6 $[M+Na]^+$; Anal. Calcd for $C_{91}H_{100}O_{21}$ (1528.68): C, 71.45; H, 6.59. Found: C, 71.20; H, 6.86.

4.1.8. 4-Methoxyphenyl (2-*O*-acetyl-4-*O*-benzyl-α-L-rhamnopyranosyl)-(12)-(3,4-di-*O*-benzyl-α-L-rhamnopyranosyl)-(13)-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(12)-3,4-di-*O*-benzyl-α-L-rhamnopyranoside 15

A solution of compound **14** (1.8 g, 1.17 mmol) in 0.1 M CH_3ONa in CH_3OH (50 mL) was allowed to stir at room temperature for 3 h and neutralized with Amberlite IR-120 (H^+) resin. The reaction mixture was filtered and evaporated to dryness. To a solution of the crude product in anhydrous DMF (10 mL) were added $CH_3C(OC_2H_5)_3$ (1 mL, 5.45 mmol) followed by p-TsOH (100 mg) and the reaction mixture was allowed to stir at room temperature for 2 h. The solvents were removed under vacuum and the resulting *ortho*-ester derivative was dissolved in 80% aq AcOH (50 mL) and the reaction mixture was allowed to stir at room temperature for 30 min. The solvents were removed under reduced pressure and co-evaporated with toluene to give the crude product, which was purified over SiO_2 using hexane–EtOAc (5:1) as eluant to give pure compound **15** (1.6 g, 91%); Oil; $[\alpha]_D^{25} = +6.9$ (c 1.5, $CHCl_3$); IR (neat): 3018, 2926, 2361, 1737, 1594, 1503, 1368, 1216, 1074, 924, 760 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 7.41–7.14 (m, 40H, Ar-H), 6.88 (d, J = 9.0 Hz, 2H, Ar-H), 6.74 (d, J = 9.0 Hz, 2H, Ar-H), 5.52 (br s, 1H, H-1_A), 5.20 (br s, 2H, H-1_C, H-2_D), 5.0 (d, J = 12.1 Hz, 1H, PhCH_2), 4.88–4.56 (m, 11H, H-1_B, H-1_D, PhCH_2), 4.48–4.32 (m, 6H, PhCH_2), 4.22 (br s, 1H, H-2_A), 4.08 (dd, J = 9.4, 3.0 Hz, 1H, H-3_D), 4.01 (dd, J = 9.4, 3.0 Hz, 1H, H-3_A), 3.87 (br s, 1H, H-2_C), 3.83–3.66 (m, 10H, H-2_B, H-3_B, H-4_B, H-4_C, H-5_A, H-5_C, H-5_D, OCH_3), 3.50–3.40 (m, 5 H, H-3_C, H-4_A, H-5_B, H-6_{abB}), 3.25 (t, J = 9.4 Hz each, 1H, H-4_D), 2.12 (s, 3H, COCH_3), 1.25, 1.19, 1.11 (3 d, J = 6.1 Hz each, 9H, 3 CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 170.4 (COCH_3), 154.7–114.5 (Ar-C), 104.9 (C-1_B), 100.2 (C-1_C), 98.8 (C-1_D), 98.2 (C-1_A), 81.7 (C-4_A), 80.4 (C-3_B), 79.9 (2C, C-2_B, C-3_A), 79.1 (C-4_D), 77.9 (C-4_C), 77.7 (C-5_D), 76.3 (C-3_C), 75.2 (PhCH_2), 75.1 (PhCH_2), 75.0 (PhCH_2), 74.9 (C-4_B), 74.8 (PhCH_2), 73.7 (C-2_A), 73.5 (2C, 2 PhCH_2), 72.7 (C-5_B), 72.3 (PhCH_2), 71.7 (PhCH_2), 70.1 (C-3_D), 69.0 (C-6_B), 68.9 (C-2_D), 68.5 (C-5_A), 67.9 (2C, C-2_C, C-5_C), 55.5 (OCH_3), 21.0 (COCH_3), 18.3, 18.0, 17.9 (3CCH_3); ESI-MS: 1509.6 $[M+Na]^+$; Anal. Calcd for $C_{89}H_{98}O_{20}$ (1486.67): C, 71.85; H, 6.64. Found: C, 71.63; H, 6.90.

4.1.9. 4-Methoxyphenyl 2-*O*-acetyl-α-L-rhamnopyranoside 16

To a solution of compound **2** (2 g, 7.4 mmol) in anhydrous DMF (5 mL) were added $CH_3C(OC_2H_5)_3$ (7.5 mL, 40.9 mmol) followed by p-TsOH (100 mg) and the reaction mixture was allowed to stir at room temperature for 2 h. The solvents were removed under reduced pressure and the crude *ortho*-ester derivative was dissolved in 80% aq AcOH (50 mL). The reaction mixture was allowed to stir at room temperature for 30 min and evaporated and co-evaporated with toluene (3 × 50 mL) to give the crude product, which was purified over SiO_2 using hexane–EtOAc (1:1) as eluant to give pure compound **16** (1.9 g (84%); Oil; $[\alpha]_D^{25} = +10$ (c 1.0, $CHCl_3$);IR (neat): 2362, 1754, 1721, 1620, 1368, 1216, 760 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 6.99 (d, J = 9.0 Hz, 2H, Ar-H), 6.79 (d, J = 9.0 Hz, 2H, Ar-H), 5.35 (br s, 1H, H-1), 5.25–5.24 (m, 1H, H-2), 4.19–4.11 (m, 1H, H-5), 3.86–3.79 (m, 1H, H-3), 3.77 (s, 3H, OCH_3), 3.51 (t, J = 9.3 Hz each, 1H, H-4), 2.16 (s, 3H, COCH_3), 1.29 (d, J = 6.1 Hz, 3H, CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 170.9 (COCH_3), 155.2–114.6 (Ar-C), 96.7 (C-1), 73.2 (C-4), 72.4 (C-2), 70.1 (C-5), 68.8 (C-3), 55.5 (OCH_3), 20.9 (COCH_3), 17.6 (CCH_3); ESI-MS: 335.1 $[M+Na]^+$; Anal. Calcd for $C_{15}H_{20}O_7$ (312.12): C, 57.69; H, 6.45. Found: C, 57.50; H, 6.70.

4.1.10. 4-Methoxyphenyl (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-(13)-[(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-(14)]-2-*O*-acetyl-α-L-rhamnopyranoside 17

To a solution of compound **16** (1 g, 3.2 mmol) in anhydrous CH_2Cl_2 (20 mL) were added compound **7** (6 g, 12.17 mmol) and MS 4 Å (2 g) and the reaction mixture was cooled to −10 °C under argon. To the cooled reaction mixture was added TMSOTf (200 μL, 1.1 mmol) and allowed to stir at same temperature for 3 h. The reaction mixture was quenched with Et_3N (0.5 mL), filtered, and washed with CH_2Cl_2 (150 mL). The organic layer was washed with aq $NaHCO_3$, water in succession, dried (Na_2SO_4), and concentrated. The crude product was purified over SiO_2 using hexane–EtOAc (3:1) as eluant to give pure compound **17** (2 g, 64%); Oil; $[\alpha]_D^{25} = -2.3$ (c 1.0, $CHCl_3$); IR (neat): 3020, 2357, 1759, 1721, 1650, 1368, 1220, 929, 739 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 6.95 (d, J = 9.0 Hz, 2H, Ar-H), 6.77 (d, J = 9.0 Hz, 2H, Ar-H), 5.36–5.33 (m, 1H, H-2_E), 5.24 (br s, 1H, H-1_E), 5.16–4.99 (m, 5H, H-2_F, H-3_F, H-4_F, H-3_G, H-4_G), 4.92 (t, J = 8.0 Hz each, 1H, H-2_G), 4.74

(d, J = 7.7 Hz, 1H, H-1_F), 4.69 (d, J = 8.0 Hz, 1H, H-1_G), 4.28–4.12 (m, 4H, H-3_E, H-6_{abF}, H-6_{aG}), 4.10–4.05 (dd, J = 12.0, 2.9 Hz, 1H, H-6_{bG}), 3.80–3.77 (m, 2H, H-4_E, H-5_E), 3.75 (s, 3H, OCH_3), 3.72–3.60 (m, 2H, H-5_F, H-5_G), 2.21, 2.14, 2.11, 2.09, 2.04, 2.03, 2.02, 2.01, 1.98 (9 s, 27H, 9 COCH_3), 1.23 (d, J = 6.1 Hz, 3H, CCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 170.2, 170.1, 169.8, 169.7 (3C), 169.1, 169.0, 168.7 (9*C*OCH$_3$), 155.7–114.5 (Ar-C), 101.1 (C-1_F), 99.2 (C-1_G), 96.4 (C-1_E), 78.6 (C-3_E), 75.1 (C-4_E), 73.2 (C-3_F), 72.7 (C-3_G), 72.1 (C-2_F), 72.0 (C-2_G), 71.8 (2C, C-2_E, C-5_F), 71.4 (C-5_G), 68.5 (C-4_F), 67.9 (C-4_G), 67.3 (C-5_E), 61.6 (C-6_F), 61.5 (C-6_G), 55.4 (O*C*H$_3$), 21.1, 20.9, 20.7, 20.6, 20.5 (3 C), 20.4 (2C), 17.9 (*C*CH$_3$). ESI-MS: 995.3 [M+Na]$^+$; Anal. Calcd for $C_{43}H_{56}O_{25}$ (972.31): C, 53.09; H, 5.80. Found: C, 52.87; H, 5.68.

4.1.11. 4-Methoxyphenyl (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-(13)-[(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-(14)]-2-*O*-acetyl-α-L-rhamnopyranosyl-(13)-(2-*O*-acetyl-4-*O*-benzyl-α-L-rhamnopyranosyl)-(12)-(3,4-di-*O*-benzyl-α-L-rhamnopyranosyl)-(13)-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(12)-3,4-di-*O*-benzyl-α-L-rhamnopyranoside 19

To a solution of compound **17** (1.8 g, 1.85 mmol) in 80% aq CH_3CN (40 mL) was added ceric ammonium nitrate (CAN; 1.6 g, 2.92 mmol) and the reaction mixture was allowed to stir at room temperature for 3 h. The reaction mixture was diluted with CH_2Cl_2 (100 mL) and the organic layer was washed with satd $NaHCO_3$ and water in succession, dried (Na_2SO_4), and concentrated. To a solution of crude hemiacetal derivative in anhydrous CH_2Cl_2 (20 mL) were added CCl_3CN (1.5 mL, 14.95 mmol) and DBU (120 μL, 0.82 mmol) at 0 °C and the reaction mixture was allowed to stir at 0 °C for 4 h. The solvents were removed under reduced pressure and the crude trichloroacetimidate derivative was purified over SiO_2 using hexane–EtOAc (6:1) as eluant to give pure compound **18** (1.3 g, 70%), which was used immediately in the next step. To a solution of compound **15** (1.5 g, 1.0 mmol) in anhydrous CH_2Cl_2 (15 mL) were added MS 4 Å (1 g) and compound **18** (1.2 g, 1.2 mmol) and the reaction mixture was cooled to −10 °C under argon. To the cooled reaction mixture was injected TMSOTf (50 μL) and it was allowed to stir at the same temperature for 3 h. The reaction mixture was quenched with Et_3N (0.5 mL), filtered, and washed with CH_2Cl_2 (100 mL). The organic layer was washed with satd $NaHCO_3$ and water in succession, dried (Na_2SO_4), and concentrated under reduced pressure. The crude product was purified over SiO_2 using hexane–EtOAc (3:1) to give pure compound **19** (1.6 g (68%); Oil; $[\alpha]_D^{25} = +3.2$ (*c* 1.5, $CHCl_3$); IR (neat): 3021, 2971, 2928, 2360, 1750, 1624, 1428, 1216, 1045, 928, 760 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.35–7.13 (m, 40H, Ar-H), 6.90 (d, J = 9.0 Hz, 2H, Ar-H), 6.74 (d, J = 9.0 Hz, 2H, Ar-H), 5.51 (br s, 1H, H-1_A), 5.19–5.16 (m, 3H, H-1_C, H-2_D, PhC*H*$_2$), 5.15–5.0 (m, 7H, H-2_F, H-3_F, H-3_G, PhC*H*$_2$), 4.98 (br s, 1H, H-1_E), 4.94 (t, J = 8.0 Hz, 1H, H-2_G), 4.85–4.80 (m, 3H, H-4_F, H-4_G, PhC*H*$_2$), 4.79 (br s, 1H, H-1_D), 4.73 (d, J = 7.7 Hz, 1H, H-1_G), 4.71–4.64 (m, 6H, H-1_B, PhC*H*$_2$), 4.59–4.50 (m, 3H, H-1_F, H-2_E, PhC*H*$_2$), 4.46–4.30 (m, 4H, PhC*H*$_2$), 4.28–4.17 (m, 4H, H-2_A, H-6_{abF}, H-6_{aG}), 4.15–4.09 (m, 2H, H-2_C, H-3_E), 4.07–3.90 (m, 3H, H-3_A, H-3_D, H-6_{bG}), 3.83–3.65 (m, 12H, H-2_B, H-3_B, H-4_B, H-5_A, H-5_C, H-5_D, H-5_E, H-6_{abB}, OC*H*$_3$), 3.48–3.30 (m, 8H, H-3_C, H-4_A, H-4_C, H-4_D, H-4_E, H-5_B, H-5_F, H-5_G), 2.18, 2.13, 2.08, 2.07, 2.06, 2.03, 2.0, 1.99 (8 s, 30 H, 10 COC*H*$_3$), 1.25–1.20 (m, 6H, 2CC*H*$_3$), 1.17 (d, J = 6.1 Hz, 3H, CC*H*$_3$), 1.10 (d, J = 6.1 Hz, 3H, CC*H*$_3$); ^{13}C NMR (75 MHz, $CDCl_3$): δ 170.0, 169.9, 169.7, 169.6, 169.5, 169.4, 169.3 (2C), 169.2, 168.8 (10*C*OCH$_3$), 154.6–114.4 (Ar-C), 104.9 (C-1_B), 100.6 (C-1_F), 100.0 (C-1_C), 98.8 (C-1_G), 98.6 (C-1_D), 98.4 (C-1_E), 98.1 (C-1_A), 80.3 (2C, C-3_B, C-4_A), 79.9 (C-3_A), 79.6 (C-2_B), 78.8 (C-4_D), 78.4 (C-3_E), 77.7 (C-4_C), 77.2 (C-5_D), 76.4 (C-3_C), 76.2 (C-4_E), 75.4 (C-4_B), 75.2 (PhC*H*$_2$), 75.0 (3C, C-3_F, 2PhC*H*$_2$), 74.8 (C-3_G), 74.7 (PhC*H*$_2$), 73.6 (PhC*H*$_2$), 73.4 (2C, 2 PhC*H*$_2$), 72.9 (C-2_A), 72.6 (C-2_F), 72.1 (PhC*H*$_2$), 72.0 (2C, C-2_G, C-5_F), 71.9 (C-5_G), 71.5 (3C, C-2_E, C-3_D, PhC*H*$_2$), 71.2 (C-5_C), 68.9 (2C, C-2_D, C-6_B), 68.6 (C-5_A), 68.4 (C-4_F), 68.1 (C-4_G), 67.6 (C-2_C), 67.3 (C-5_E), 61.7 (C-6_G), 61.0 (C-6_F), 55.5 (OC*H*$_3$), 21.0, 20.9, 20.7, 20.5 (3C), 20.4 (4C) (10COC*H*$_3$), 18.1, 17.9, 17.7, 17.6 (4 C*C*H$_3$); MALDI-TOF MS: 2358.0 [M+Na]$^+$; Anal. Calcd for $C_{125}H_{146}O_{43}$ (2334.92): C, 64.26; H, 6.30. Found: C, 64.0; H, 6.50.

4.1.12. 4-Methoxyphenyl (sodium β-D-glucopyranosyl uronate)-(13)-[(sodium β-D-glucopyranosyl uronate)-(14)]-α-L-rhamnopyranosyl-(13)-(α-L-rhamnopyranosyl)-(12)-(α-L-rhamnopyranosyl)-(13)-(β-D-galactopyranosyl)-(12)-α-L-rhamnopyranoside 1

A solution of compound **19** (1.5 g, 0.64 mmol) in 0.1 M CH_3ONa in CH_3OH (100 mL) was allowed to stir at room temperature for 5 h and neutralized with Dowex-50W X8 (H^+) resin. The reaction mixture was filtered and concentrated under reduced pressure. To a solution of the crude product in CH_2Cl_2 (30 mL) and H_2O (7 mL) were added aq solution of NaBr (2 mL; 1 M), aq solution of TBAB (4 mL; 1 M), TEMPO (150 mg, 0.96 mmol), satd aq solution of $NaHCO_3$ (15 mL), and 4% aq NaOCl (20 mL) in succession and the reaction mixture was allowed to stir at 0–5 °C for 3 h. The reaction mixture was neutralized with the addition of 1 M aq HCl solution. To the reaction mixture were added *tert*-butanol (30 mL), 2-methyl-but-2-ene (40 mL; 2 M solution in THF), aq solution of $NaClO_2$ (2 g in 10 mL), and aq solution of NaH_2PO_4 (2 g in 10 mL) and the reaction mixture was allowed to stir at room temperature for 3 h. The reaction mixture was diluted with satd aq NaH_2PO_4 and extracted with CH_2Cl_2 (200 mL). The organic layer was washed with water, dried (Na_2SO_4), and concentrated to dryness. To a solution of the crude product in 90% aq CH_3OH (20 mL) was added 20% $Pd(OH)_2$–C (200 mg) and the reaction mixture was allowed to stir at room temperature under a positive pressure of hydrogen for 24 h. The reaction mixture was filtered through a Celite® bed and washed with CH_3OH–H_2O (60 mL; 5:1 v/v). The combined filtrate was evaporated under reduced pressure to furnish compound **1**, which was purified through a Sephadex LH-20 column using CH_3OH–H_2O (6:1) as eluant to give pure compound **1** (480 mg (59%); white powder; $[\alpha]_D^{25} = -64$ (*c* 1.0, CH_3OH); IR (KBr): 3020, 2362, 1754, 1721, 1216, 929, 760 cm^{-1}; ^{1}H NMR (300 MHz, CH_3OD–D_2O): δ 6.88 (d, J = 9.0 Hz, 2H, Ar-H), 6.74 (d, J = 9.0 Hz, 2H, Ar-H), 5.58 (br s, 1H, H-1_A), 5.14 (br s, 1H, H-1_C), 4.95 (br s, 1H, H-1_E), 4.85 (br s, 1H, H-1_D), 4.67 (d, J = 7.9 Hz, 1H, H-1_G), 4.52 (d, J = 7.5 Hz, 1H, H-1_F), 4.43 (d, J = 7.8Hz, 1H, H-1_B), 4.18–4.17 (m, 1H, H-2_E), 4.02 (dd, J = 9.3, 2.8 Hz, 1H, H-3_E), 4.0–3.99 (m, 1H, H-2_A), 3.97–3.96 (m, 1H, H-2_D), 3.91–3.90 (m, 1H, H-2_C), 3.84–3.74 (m, 5H, H-3_A, H-3_C, H-4_B, H-5_A, H-6_{aB}), 3.70–3.66 (m, 4H, H-3_D, H-3_F, H-5_F, H-5_G), 3.65 (s, 3H, OC*H*$_3$), 3.64–3.56 (m, 6H, H-2_B, H-3_B, H-4_F, H-5_B, H-5_C, H-6_{bB}), 3.52–4.46 (m, 2H,H-4_E, H-5_D), 3.40 (t, J = 9.5 Hz each, 1H, H-4_C), 3.35 (t, J = 9.5 Hz, 1H, H-4_D), 3.30–3.16 (m, 5H, H-2_F, H-3_G, H-4_G, H-4_A, H-5_E), 1.26–1.14 (m, 12H, 4 CC*H*$_3$); ^{13}C NMR (75 MHz, $CDCl_3$): δ 174.4, 174.3 (2*C*OONa), 156.5 (Ar-C), 151.7 (Ar-C), 118.9 (2C, Ar-C), 115.6 (2C, Ar-C), 107.5 (C-1_B), 105.1 (C-1_F), 104.3 (C-1_G), 103.7 (C-1_D), 103.4 (C-1_E), 102.3 (C-1_C), 99.7 (C-1_A), 82.7 (C-3_E), 82.2 (C-2_A), 81.0 (C-4_E), 79.5 (C-2_C), 79.3 (C-3_D), 79.1 (C-5_B), 78.5 (C-4_F), 78.2 (C-5_A), 77.8 (C-5_E), 77.7 (C-3_F), 76.7 (C-5_D), 75.7 (C-5_F), 75.4 (C-5_G), 74.4 (C-4_A), 74.3 (C-4_D), 73.3 (C-4_C), 72.7 (C-3_G), 72.3 (C-2_B), 72.0 (C-2_F), 71.9 (2C, C-2_D, C-4_B), 71.6 (C-2_E), 71.2 (C-4_G), 70.6 (C-2_G), 70.4 (C-3_B), 70.3 (C-5_C), 70.0 (C-3_C), 68.9 (C-3_A), 62.9 (C-6_B), 56.1 (OC*H*$_3$), 18.4, 18.2, 18.0, 17.9 (4 C*C*H$_3$); ESI-MS: 1267.0 [M]$^+$; Anal. Calcd for $C_{49}H_{72}Na_2O_{35}$: C, 46.45; H, 5.73. Found: C, 46.27; H, 5.98.

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References

1. Grimont, F.; Grimont, P. A. D. In *The Prokaryotes*; Balows, A., Ed.; Springer: New York, 1992.
2. (a) Bleich, A.; Kirsch, P.; Sahly, H.; Fahey, J.; Smoczek, A.; Hedrich, H.-J.; Sundberg, J. P. *Lab. Anim.* **2008**, *42*, 369–375; (b) Seliskar, A.; Zdovc, I.; Zorko, B. *Slov. Vet. Res.* **2007**, *44*, 115–122; (c) Watson, J. T.; Jones, R. C.; Siston, A. M.; Fernandez, J. R.; Martin, K.; Beck, E.; Sokalski, S.; Jensen, B. J.; Arduino, M. J.; Srinivasan, A.; Gerber, S. I. *Arch. Int. Med.* **2005**, *165*, 2639–2643.
3. (a) Sugihara, R.; Yoshimura, M.; Mori, M.; Kanayama, N.; Hikida, M.; Ohmori, H. *Immunopharmacology* **2000**, *49*, 325–333; (b) Evans, E.; Brown, M. R. W.; Gilbert, P. *Microbiology* **1994**, *140*, 153–157.
4. Leone, S.; De Castro, C.; Parrilli, M.; Baldi, F.; Lanzetta, R. *Eur. J. Org. Chem.* **2007**, 5183–5189.
5. Baldi, F.; Minacci, A.; Pepi, M.; Scozzafava, A. *FEMS Microbiol. Ecol.* **2001**, *36*, 169–174.
6. Gutnick, D. L.; Bach, H. *Appl. Microbiol. Biotechnol.* **2000**, *54*, 451–460.
7. D'Annibale, A.; Leonardi, V.; Baldi, F.; Zecchini, F.; Petruccioli, M. *Appl. Microbiol. Biotechnol.* **2007**, *74*, 1135–1144.
8. (a) Mandal, P. K.; Misra, A. K. *Tetrahedron* **2008**, *64*, 8685–8691; (b) Mandal, P. K.; Misra, A. K. *Synthesis* **2009**, 1348–1354; (c) Mandal, P. K.; Misra, A. K. *Glycoconjugate J.* **2008**, *25*, 713–722; (d) Agnihotri, G.; Mandal, P. K.; Misra, A. K. *Tetrahedron* **2007**, *63*, 7240–7245; (e) Mukherjee, C.; Misra, A. K. *Tetrahedron: Asymmetry* **2009**, *20*, 473–477; (f) Mukherjee, C.; Misra, A. K. *Tetrahedron: Asymmetry* **2008**, *19*, 2746–2751; (g) Panchadhayee, R.; Misra, A. K. *Glycoconjugate J.* **2008**, *25*, 817–826; (h) Kumar, R.; Maulik, P. R.; Misra, A. K. *Glycoconjugate J.* **2008**, *25*, 511–519; (i) Mukherjee, C.; Misra, A. K. *Glycoconjugate J.* **2008**, *25*, 611–624; (j) Tiwari, P.; Misra, A. K. *Glycoconjugate J.* **2008**, *25*, 85–99; (k) Mukherjee, C.; Misra, A. K. *Glycoconjugate J.* **2008**, *25*, 111–119; (l) Mandal, P. K.; Misra, A. K. *Synthesis* **2007**, 2660–2666; (m) Agnihotri, G.; Misra, A. K. *Tetrahedron Lett.* **2006**, *47*, 8493–8497.
9. Sarkar, K.; Mukherjee, I.; Roy, N. *J. Carbohydr. Chem.* **2003**, *22*, 95–108.
10. Werz, D. B.; Seeberger, P. H. *Angew. Chem., Int. Ed.* **2005**, *44*, 6315–6318.
11. Rabuka, D.; Hindsgaul, O. *Carbohydr. Res.* **2002**, *337*, 2127–2151.
12. Sajtos, F.; Hajkó, J.; Kövér, K. E.; Lipták, A. *Carbohydr. Res.* **2001**, *334*, 253–259.
13. Veeneman, G. H.; Van Leeuwen, S. H.; Zuurmond, H.; van Boom, J. H. *J. Carbohydr. Chem.* **1990**, *9*, 783–796.
14. Schmidt, R. R.; Stumpp, M. *Liebigs Ann. Chem.* **1983**, *7*, 1249–1256.
15. (a) Veeneman, G. H.; van Leeuwen, S. H.; van Boom, J. H. *Tetrahedron Lett.* **1990**, *31*, 1331–1334; (b) Konradsson, P.; Udodong, U. E.; Fraser-Reid, B. *Tetrahedron Lett.* **1990**, *31*, 4313–4316.
16. Madhusudan, S. K.; Agnihotri, G.; Negi, D. S.; Misra, A. K. *Carbohydr. Res.* **2005**, *340*, 1373–1377.
17. Ogawa, T.; Yamamoto, H. *Agric. Biol. Chem.* **1985**, *49*, 475–482.
18. Field, R. A.; Otter, A.; Fu, W.; Hindsgaul, O. *Carbohydr. Res.* **1995**, *276*, 347–363.
19. Schmidt, R. R. *Angew. Chem., Int. Ed. Engl.* **1986**, *25*, 212–236.
20. Schmidt, R. R.; Jung, K.-H. In *Preparative Carbohydrate Chemistry*; Hanessian, S., Ed.; Marcel Dekker Inc.: New York, 1997; pp 283–312.
21. (a) Huang, L.; Teumelsan, N.; Huang, X. *Chem. Eur. J.* **2006**, *12*, 5246–5252; (b) Anelli, P. L.; Biffi, C.; Montanari, F.; Quici, S. *J. Org. Chem.* **1987**, *52*, 2559–2562.
22. Pearlman, W. M. *Tetrahedron Lett.* **1967**, *8*, 1663–1664.