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Synthesis of Analogues of the Antitumor (1→6)-Branched (1→3)-Glucohexaose

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β-D-Glcp-(1→3)-[*β-D*-Glcp-(1→6)-]*α-D*-Manp-(1→3)-*β-D*-Glcp-(1→3)-[*β-D*-Glcp-(1→6)-]*D*-Glcp (**18**) and *β-D*-Glcp-(1→3)-[*β-D*-Glcp-(1→6)-]*α-D*-Manp-(1→3)-*β-D*-Glcp-(1→3)-[*β-D*-Glcp-(1→6)-]*β-D*-Glcp-*D*-(1→3)-Glcp-1→OMe (**29**) were synthesized as the analogues of the immunomodulator *β-D*-Glcp-(1→3)-[*β-D*-Glcp-(1→6)-]*α-D*-Glcp-(1→3)-*β-D*-Glcp-(1→3)-[*β-D*-Glcp-(1→6)-]*D*-Glcp through coupling of trisaccharide donors **9** with trisaccharide acceptor **16** and tetrasaccharide acceptor **27** followed by deprotection, respectively.

Keywords oligosaccharide, trichloroacetimidate, regio- and stereoselective synthesis

Introduction

Polysaccharides with antitumor activity separated from fungi such as *Ganoderma lucidum*, *Schizophyllum commune* and *Lentinus edodes* have a *β*-(1→3)-linked glucosyl backbone with *β*-(1→6)-branched glucosyl side chains.[1] Recent studies revealed that *α*-(1→3)-linked glucans also exist in some medically important fungi such as *Cryphonectrini parasitica* and *Ganoderma lucidum*.[2] It was also reported that only higher molecular-weight fractions (M_W ＞16,000) obtained from partial hydrolysis of lentinan with formic acid showed antitumor activity.[3] However, an interesting result in our research revealed[4] that a synthetic hexasaccharide **I**, *β-D*-Glc*p*-(1→3)-[*β-D*-Glc*p*-(1→6)-]*α-D*-Glc*p*-(1→3)-*β-D*-Glc*p*-(1→3)-[*β-D*-Glc*p*-(1→6)-]*D*-Glc*p*, in combination with the chemotherapeutic agent cyclophosphamide (CPA), at a dose of 0.5 to 1 mg/kg substantially increased the inhibition of S_{180} for CPA, but decreased the toxicity caused by CPA. This inspired us to carry out more research regarding the structure function relationships of oligosaccharides. It was reported that 3,6-mannosylated glucans have antitumor activity.[5] We present herein the synthesis of two analogues of **I** containing mannose residue in the (1→3)-linked backbone.

Results and discussion

As outlined in the Scheme 1, replacement of the 2nd glucose residue of the upstream end of the (1→3)-linked backbone of **I** with mannose was carried out to obtain a hexsaose **18** and methyl heptaoside **29** respectively. A co-used trisaccharide donor **9** was synthesized in a concise way. Thus 4,6-*O*-benzylidene-1,2-*O*-ethylidene-*β-D*-mannopyranose (**2**)[6] was used as the starting material. Condensation of **2** with perbenzoylated glucosyl trichloroacetimidate[7] **1** afforded the disaccharide **3** in satisfactory yield (80%). Selective removal of the 4,6-*O*-benzylidene group of **3** with 1∶1000 AcCl-MeOH smoothly offered the disaccharide acceptor **4** (88%), subsequent coupling of **4** with **1** selectively gave the (1→6)-linked trisaccharide **5** (87%). Trisaccharide trichloroacetimidate **9** was obtained by deethylidenation of **5** with 90% CF_3COOH-H_2O, acetylation, selective 1-*O*-deacetylation, and subsequent trichloroacetimidation (65% for four steps). The ^{1}H NMR spectrum of **9** showed a characteristic signal at δ 5.13 with $J_{3,}=J_{4,5}=$ 9.7 Hz for H-4, confirming the *C*-6-glycosylation of **4**. The trisaccharide acceptor **16** was prepared as follows. Coupling of 3-*O*-allyl-2,4,6-tri-*O*-benzoyl-*α-D*-glucopyranosyl trichloroacetimidate (**10**)[8] with 1,2∶5,6-di-*O*-isopropylidene-*α-D*-glucofuranose (**11**) furnished disaccharide **12** (82%). Removal of 5,6-*O*-isopropylidene group of **12** with 90% HOAc-H_2O (90%), followed by selective 6-*O*-glucosylation with **1** (83%), acetylaton, and then deallylation (91%) yielded **16**. For preparation of the tetrasaccharide acceptor **27**, trisaccharide **14** was hydrolyzed to remove the 1,2-*O*-isopropylidene group giving the hemiacetal **19**. Subsequent acetylation, selective 1-*O*-deacetylation, and trichloroacrtimidation yielded the trisaccharide donor **22** (62%, for four steps from **14**). Coupling of **22** with methyl 4,6-*O*-benzylidene-α-*D*-glucopyranoside (**23**)[9] produced the tetrasaccharide **24** (70%). Debenzylidenation of **24**, followed by acetylation and deallylation gave tetrasaccharide acceptor **27** (72%, for three steps). Compared to **24,** the ^{1}H NMR spectrum of **27** clearly showed a new signal at δ 4.76 with $J_{1,2}=3.6$, $J_{2,3}=9.6$ Hz for H-2, confirming the 3-*O*-selective glycosylation of **23**.

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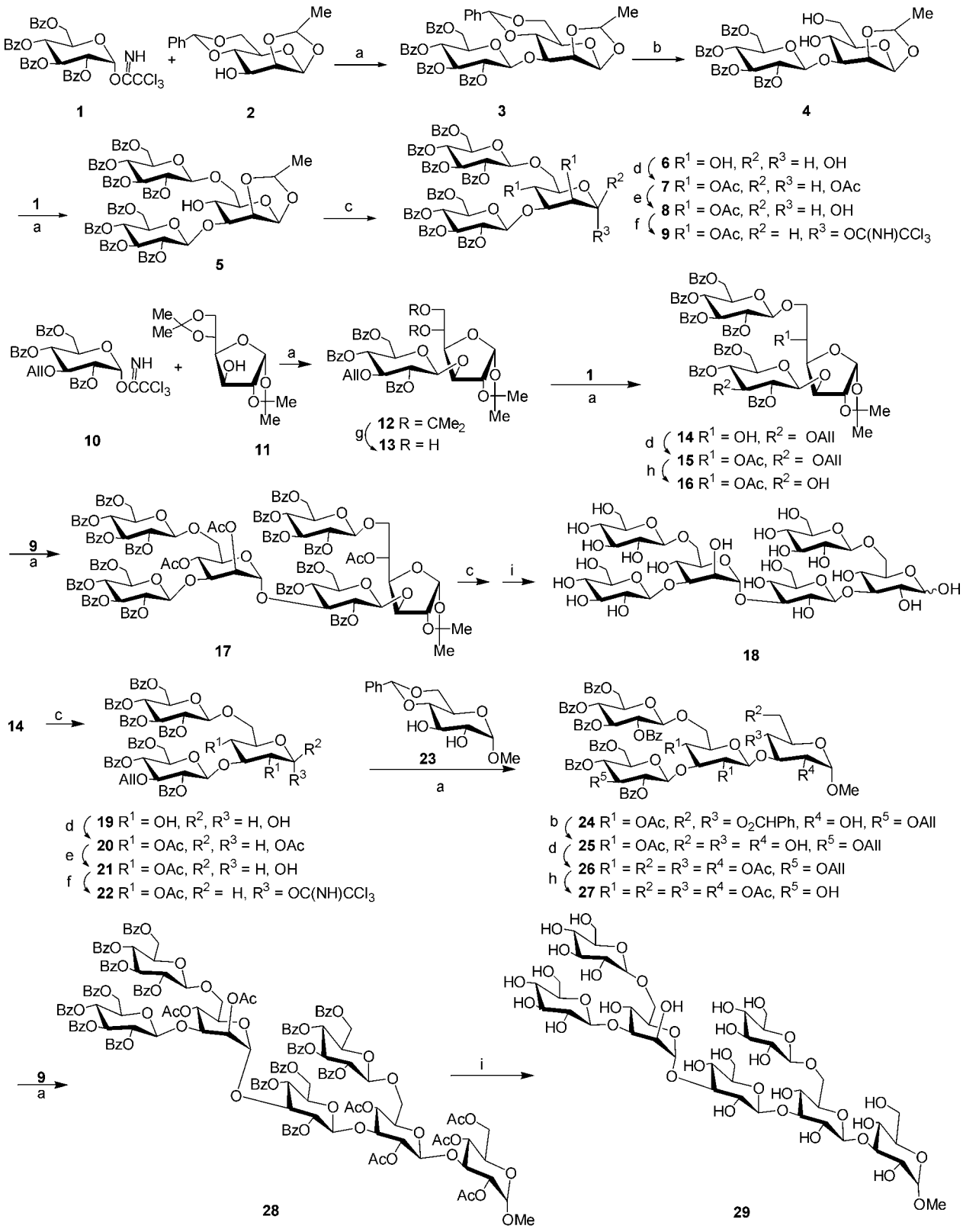
Scheme 1



Conditions and reagents: a: TMSOTf, CH_2Cl_2, −15 ℃ to r.t.; b: MeOH, CH_3COCl, r.t.; c: 90% CF_3COOH-H_2O, 30 ℃, 2 h; d: Ac_2O, pyridine, r.t.,12 h; e: THF, MeOH, NH_3, r.t.; f: CCl_3CN, CH_2Cl_2, DBU, r.t.; g: 90% HOAc-H_2O, 40 ℃, 8 h; h: $PdCl_2$, CH_2Cl_2, MeOH, r.t.; i: MeOH, NH_3, r.t., two weeks.

With the trisaccharide donor **9**, trisaccharide acceptor **16**, and tetrasaccharide acceptor **27** at hand, the target hexaose and heptaoside were readily pepared. Thus, condensation of **16** with **9** followed by deprotection gave the hexaose **18**, while coupling of **27** with **9** followed by deprotection afforded the heptaoside **29**.

The bioassay of **18** and **29** is in progress and the results will be reported in due course.

Experimental

General methods

Optical rotations were determined at 25 ℃ with a Perkin-Elmer Model 241-Mc automatic polarimeter. ^{1}H NMR and ^{13}C NMR spectra were recorded with a Bruker ARX 400 spectrometer (400 MHz for ^{1}H, 100 MHz for ^{13}C) at 25 ℃ for solutions in $CDCl_3$ or D_2O as indicated. Mass spectra were recorded with a VG PLATFORM mass spectrometer using the ESI mode. Thin-layer chromatography (TLC) was performed on silica gel HF_{254} with detection by charring with 30% (*V* ∶ *V*) H_2SO_4 in MeOH or in some cases by a UV lamp. Column chromatography was conducted by elution of a column (16 mm×240 mm, 18 mm×300 mm, 35 mm×400 mm) of silica gel (100—200 mesh) with EtOAc-petroleum ether (60—90 ℃) as the eluent. Solutions were concentrated at ＜60 ℃ under reduced pressure.

2,3,4,6-Tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-1,2-*O*-ethylidene-4,6-*O*-benzylidene-*β*-*D*-mannopyranose (3)

To a cooled solution (0 ℃) of **1** (3.75 g, 5.1 mmol) and **2** (1.36 g, 4.6 mmol) in anhydrous CH_2Cl_2 (50 mL) was added TMSOTf (20 μL, 0.12 mmol). The mixture was stirred for 2 h, during which time the temperature was gradually raised to ambient temperature. The mixture was quenched with Et_3N (4 drops) and then evaporated to give a residue, which was purified by silica gel column chromatography with 2 ∶ 1 petroleum ether-EtOAc as the eluent to give diasaccharide **3** (3.21 g, 80%) as a foamy solid. $[\alpha]_D$ −39.7 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) *δ*: 7.98—7.25 (m, 25H, 4 Bz-H, Ph-H), 5.88 (dd, 1H, $J_{3,4}=J_{4,5}=9.5$ Hz, H-4′), 5.73 (dd, 1H, $J_{2,3}=J_{3,4}=9.5$ Hz, H-3′), 5. 60 (dd, 1H, $J_{1,2}=8.0$ Hz, $J_{2,3}=9.5$ Hz, H-2′), 5.58 (s, 1H, PhCH), 5.13 (d, 1H, $J_{1,2}=8.0$ Hz, H-1′), 5.12 (d, 1 H, $J_{1,2}=2.0$ Hz, H-1), 4.73 (q, 1H, $J=3.7$ Hz, $CH_3C\mathbf{H}$), 4.56 (dd, 1H, $J_{5,6}=3.5$ Hz, $J_{6'e,6'a}=12.1$ Hz, H-6′e), 4.42 (dd, 1H, $J_{5,6}=4.2$ Hz, $J_{6'a,6'e}=12.1$ Hz, H-6′a), 4.09-4.00 (m, 3H, H-2, H-4, H-5′), 4.25 (dd, 1H, $J_{5,6}=5.1$ Hz, $J_{6e,6a}=10.6$ Hz, H-6e), 4.21 (dd, 1H, $J_{5,6}=4.2$ Hz, $J_{6a,6e}=10.6$ Hz, H-6a), 3.74 (dd, 1H, $J_{2,3}=J_{3,4}=10.3$ Hz, H-3), 3.31 (ddd, 1H, $J_{4,5}=10.3$ Hz, $J_{5,6}=5.1$ Hz, $J_{5,6}=4.2$ Hz, H-5), 1.25 (d, 3H, $J=4.7$ Hz, $C\mathbf{H}_3CH$). Anal. calcd for $C_{49}H_{44}O_{15}$: C 67.43, H 5.08; found C 67.38, H 5.09.

2,3,4,6-Tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-1,2-*O*-ethylidene-*β*-*D*-mannopyranose (4)

Acetyl chloride (0.1 mL) was added to a solution of **3** (8.73 g, 10.0 mmol) in anhydrous MeOH (100 mL). The solution was stoppered in a flask and stirred at room temperature until TLC (petroleum ether-EtOAc, 2 ∶ 1, *V* ∶ *V*) showed that the starting material disappeared. The solution was neutralized with Et_3N, then concentrated to dryness. The residue was purified by chromatography with petroleum ether-EtOAc (1 ∶ 2, *V* ∶ *V*) as the eluent to give **4** (6.90 g, 88 %) as a foamy solid. $[\alpha]_D$ −49.7 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) *δ*: 8.07—7.26 (m, 20H, 4×Bz-H), 5.95 (dd, $J_{3,4}=J_{4,5}=9.7$ Hz, 1H, H-4′), 5.64 (dd, $J_{2,3}=J_{3,4}=9.7$ Hz, 1H, H-3′), 5.58 (dd, $J_{1,2}=7.9$ Hz, $J_{2,3}=9.7$ Hz, 1H, H-2′), 5.12 (d, $J_{1,2}=1.5$ Hz, 1H, H-1), 5.01 (d, $J_{1,2}=7.9$ Hz, 1H, H-1′), 4.77 (dd, $J_{2,3}=2.2$ Hz, $J_{3,4}=9.7$ Hz, 1H, H-3), 4.72 (q, $J=4.8$ Hz, 1H, $CH_3C\mathbf{H}$), 4.40 (dd, $J_{5,6}=6.3$ Hz, $J_{6'e,6'a}=12.2$ Hz, 1H, H-6′e), 4.23 (ddd, $J_{4,5}=9.7$ Hz, $J_{5,6}=6.3$ Hz, $J_{5,6}=5.6$ Hz, 1H, H-5′), 3.92 (dd, $J_{3,4}=J_{4,5}=9.7$ Hz, 1H, H-4) , 3.88—3.80 (m, 3H, H-2, 2 H-6), 3.72 (dd, $J_{5,6}=5.6$ Hz, $J_{6'a,6'e}=12.2$ Hz, 1H, H-6′a), 3.29 (ddd, $J_{4,5}=9.7$ Hz, $J_{5,6}=5.1$ Hz, $J_{5,6}=3.8$ Hz, 1H, H-5), 1.26 (d, $J=4.8$ Hz, 3H, $C\mathbf{H}_3CH$). Anal. calcd for $C_{42}H_{40}O_{15}$: C 64.28, H 5.14; found C 64.20, H 5.23.

2,3,4,6-Tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]1,2-*O*-ethylidene-*β*-*D*-mannopyranose (5)

Acceptor **4** (3.93 g, 5 mmol) and donor **1** (3.7 g, 5 mmol) were coupled in anhydrous CH_2Cl_2 (50 mL) in the presence of TMSOTf (50 μL, 0.28 mmol) under the same condition as described for the synthesis of **3** by coupling of **2** with **1**. Purification by chromatography with petroleum ether-EtOAc (2 ∶ 1, *V* ∶ *V*) as the eluent gave trisaccharide **5** (5.93 g, 87%). $[\alpha]_D$ +10.3 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) *δ*: 7.94—7.25 (m, 40H, 8 Bz-H), 5.91 (dd, $J_{3,4}=J_{4,5}=9.7$ Hz, 1H, H-4), 5.86 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.64 (dd, $J_{2,3}=J_{3,4}=9.7$ Hz, 1H, H-3), 5.60 (dd, $J_{2,3}=J_{3,4}=9.6$ Hz, 1H, H-3), 5.52 (dd, $J_{1,2}=7.6$ Hz, $J_{2,3}=9.7$ Hz, 1H, H-2), 5.48 (dd, $J_{1,2}=8.0$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 4.93 (d, $J_{1,2}=7.6$ Hz, 1H, H-1), 4.88 (d, $J_{1,2}=8.0$ Hz, 1H, H-1), 4.84 (d, $J_{1,2}=2.0$ Hz, 1H, H-1), 4.71 (dd, $J_{5,6}=2.6$ Hz, $J_{6,6a}=12.2$ Hz, 1H, H-6e), 4.66 (q, $J=4.8$ Hz, 1H, $CH_3C\mathbf{H}$), 4.61 (dd, $J_{5,6}=3.1$ Hz, $J_{6,6e}=12.2$ Hz, 1H, H-6a), 4.46 (dd, $J_{5,6}=5.1$ Hz, $J_{6,6e}=12.1$ Hz, 1H, H-6a), 4.36 (dd, $J_{5,6}=6.4$ Hz, $J_{6,6a}=12.1$ Hz, 1H, H-6e), 4.19—4.05 (m, 3H, 2×H-5, H-4), 3.77—3.58 (m, 4H, H-2, H-3, 2×H-6), 3.30 (ddd, $J_{4,5}=9.6$ Hz, $J_{5,6}=6.8$ Hz, $J_{5,6}=3.7$ Hz, 1H, H-5), 1.24 (d, $J=4.8$ Hz, 3H, $C\mathbf{H}_3CH$). Anal. calcd for $C_{76}H_{66}O_{24}$: C 66.96, H 4.88; found C 66.75, H, 4.99.

2,3,4,6-Tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]2,4-di-*O*-acetyl-*α*-*D*-mannopyranosyl trichloroacetimidate (9)

Compound **5** (5.45 g, 4.0 mmol) was dissolved in 70 mL of 90% TFA and stirred for 2 h, at the end of which time the reaction mixture was poured directly to 250 mL

of toluene and concentrated. Drying the residue under high vacuum gave a white foamy solid. The foamy solid was dissolved in pyridine (20 mL), and then Ac_2O (10 mL) was added. The reaction mixture was stirred at room temperature for 12 h, and TLC (petroleum ether-EtOAc, 1∶1, *V*∶*V*) indicated that the reaction was complete. The reaction mixture was concentrated to dryness. The resultant crude product **7** was dissolved in 1 mol•L^{-1} solution of ammonia-methanol (200 mL) and stirred at room temperature for 3 h, at the end of which time TLC (petroleum ether-EtOAc, 1∶1, *V*∶*V*) indicated that the reaction was complete. The solution was concentrated to give compound **8** as a syrup. A mixture of **8**, trichloroacetonitrile (4.2 mL, 20 mmol), and 1,8-diazabicyclo[5.4.0]undecene (DBU) (0.50 mL, 4.04 mmol) in dry dichloromethane (50 mL) was stirred under nitrogen protection for 3 h and then concentrated. The residue was purified by flash chromatography with petroleum ether-EtOAc (2∶1, *V*∶*V*) as the eluent to give **9** (4.07 g, 65% for four steps) as a foamy solid. $[\alpha]_D$ +14.3 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 8.41 (s, 1H, $CNHCCl_3$), 8.00—7.26 (m, 40H, 8× Bz-H), 6.02 (d, $J_{1,2}$=2.8 Hz, 1H, H-1), 5.87—5.82 (m, 2H, 2×H-4), 5.66 (dd, $J_{2,3}$=$J_{3,4}$=9.7 Hz, 1H, H-3), 5.64 (dd, $J_{2,3}$=$J_{3,4}$= 9.7 Hz, 1H, H-3), 5.48 (dd, $J_{1,2}$= 7.7 Hz, $J_{2,3}$=9.7 Hz, 1H, H-2), 5.64 (dd, $J_{1,2}$=7.7 Hz, $J_{2,3}$=9.7 Hz, 1H, H-2), 5.24 (dd, $J_{1,2}$=2.8 Hz, $J_{2,3}$=3.4 Hz, 1H, H-2), 5.13 (dd, $J_{3,4}$=$J_{4,5}$=9.7 Hz, 1H, H-4), 4.93 (d, $J_{1,2}$=7.7 Hz, 1H, H-1), 4.90 (d, $J_{1,2}$=7.7 Hz, 1H, H-1), 4.64—4.42 (m, 4H, 2×H-6e, 2×H-6a), 4.26 (dd, $J_{2,3}$=3.4 Hz, $J_{3,4}$=9.7 Hz, 1H, H-3), 4.16—4.89 (m, 4H, 2×H-5, 2×H-6), 3.68 (ddd, $J_{4,5}$=9.7 Hz, $J_{5,6}$=5.8 Hz, $J_{5,6}$=5.7 Hz, 1H, H-5), 1.96 (s, 3H, CH_3CO), 1.30 (s, 3H, CH_3CO). Anal. calcd for $C_{80}H_{68}Cl_3NO_{26}$: C 61.37, H 4.38; found C 61.18, H, 4.35.

3-*O*-Allyl-2,4,6-tri-*O*-benzoyl-β-*D*-glucopyranosyl-(1→3)-1,2∶5,6-di-*O*-isopropylidene-α-*D*-glucofuranose (12)

Compounds **10** (3.38 g, 5 mmol) and **11** (1.18 g, 4.55 mmol) were coupled in anhydrous CH_2Cl_2 (80 mL) in the presence of TMSOTf (50 μL, 0.28 mmol) under the same condition as described for the synthesis of **3** by coupling of **2** with **1**. Purification by chromatography with petroleum ether-EtOAc (3∶1, *V*∶*V*) as the eluent gave disaccharide **12** (2.89 g, 82%). $[\alpha]_D$ +32.2 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 8.09—7.26 (m, 15H, 3×Bz-H), 5.57—5.53 (m, 1H, $CH_2C\mathbf{H}$=CHH), 5.54 (dd, $J_{2,3}$=$J_{3,4}$=9.6 Hz, 1 H, H-4′), 5.46 (d, $J_{1,2}$= 3.8 Hz, 1H, H-1), 5.30 (dd, $J_{1,2}$=7.8 Hz, $J_{2,3}$=9.6 Hz, 1H, H-2'), 5.06 (dd, *J*=1.6, 7.2 Hz, 1H, CH_2CH=C**H**H), 4.95 (dd, *J*=1.4, 10.6 Hz, 1H, CH_2CH=CH**H**), 4.80 (d, $J_{1,2}$=7.8 Hz, 1H, H-1′), 4.71 (ddd, $J_{4,5}$=9.6 Hz, $J_{5,6e}$=6.9 Hz, $J_{5,6a}$=3.7 Hz, 1H, H-5), 4.44 (dd, $J_{3,4}$= $J_{4,5}$=6.6 Hz, 1H, H-4), 4.38 (dd, $J_{2,3}$=$J_{3,4}$=6.6 Hz, 1H, H-3), 4.36—4.21 (m, 7H, C**H**$_2$CH=CHH, H-2, H-3′, H-5′, H-6, H-6′), 4.13—3.92 (m, 2 H, H-6, H-6′), 1.41 (s, 3H, CH_3), 1.36 (s, 3H, CH_3), 1.24 (s, 3H, CH_3), 1.06 (s, 3H, CH_3). Anal. calcd for $C_{42}H_{46}O_{14}$: C 65.11, H 5.98; found C 65.07, H 5.91.

3-*O*-Allyl-2,4,6-tri-*O*-benzoyl-β-*D*-glucopyranosyl-(1→3)-1,2-*O*-isopropylidene-α-*D*-glucofurannose (13)

Compound **12** (3.87 g, 5 mmol) was added to 90% HOAc (50 mL), and the mixture was stirred at 40 ℃ for 8 h, at the end of which time TLC (petroleum ether-EtOAc, 2∶3, *V*∶*V*) indicated that the reaction was complete. The solvents were evaporated to give a residue, which was purified by silica gel column chromatography with 2:3 petroleum ether-EtOAc as the eluent to give diasaccharide **13** as a syrup (3.30 g, 90%). $[\alpha]_D$ +0.00 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 8.20—7.31 (m, 15H, 3×Bz-H), 5.60—5.57 (m, 1H, $CH_2C\mathbf{H}$=CHH), 5.52 (dd, $J_{3,4}$=$J_{4,5}$=9.8 Hz, 1H, H-4′), 5.46 (d, $J_{1,2}$=1.5 Hz, 1H, H-1), 5.30 (dd, $J_{1,2}$=9.1 Hz, $J_{2,3}$=9.2 Hz, 1H, H-2′), 5.05 (dd, *J*=1.4, 17.2 Hz, 1H, CH_2CH=C**H**H), 4.95 (dd, *J*=1.4, 10.8 Hz, 1H, CH_2CH =CH**H**), 4.83 (d, $J_{1,2}$=9.1Hz, 1H, H-1′), 4.76 (ddd, $J_{4,5}$ =9.8 Hz, $J_{5,6}$=6.8 Hz, $J_{5,6}$=3.0 Hz, 1H, H-5), 4.35—4.25 (m , 2 H, H-4, H-3), 4.17 (dd, $J_{1,2}$=1.5 Hz, $J_{2,3}$=9.5 Hz, 1H, H-2), 4.12 (dd, $J_{5,6}$=6.8 Hz, $J_{6e,6a}$= 3.0 Hz, 1H, H-6e), 4.10—3.86 (m, 5H, C**H**$_2$-CH=CHH, H-3′, H-5′, H-6′e), 3.80 (dd, $J_{5,6}$=6.8 Hz, $J_{6a,6e}$=3.0 Hz, 1H, H-6a), 3.63 (dd, $J_{5,6}$=6.1 Hz, $J_{6,6e'}$= 11.4 Hz, 1H, H-6′a), 1.26 (s, 3H, CH_3), 1.18 (s, 3H, CH_3). Anal. calcd for $C_{39}H_{42}O_{14}$: C 63.75, H 5.76; found C63.70, H, 5.71.

2,4,6-Tri-*O*-benzoyl-β-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-β-*D*-glucopyranosyl-(1→6)]-5-*O*-acetyl-1,2-*O*-isopropylidene-α-*D*-glucofuranose (16)

Coupling of **13** (2.56 g, 3.5 mmol) with **1** (2.85 g, 3.85 mmol) in the presence of catalytic TMSOTf (35 μL, 0.20 mmol) was carried out under the same conditions as described for the synthesis of **3** by coupling of **2** with **1**. Purification by a flash chromatography with petroleum ether-EtOAc (2∶1, *V*∶*V*) as the eluent gave compound **14** (3.82 g, 83%). Compound **14** was dissolved in pyridine (20 mL), and then Ac_2O (10 mL) was added. The reaction mixture was stirred at room temperature for 12 h, and TLC (petroleum ether-EtOAc, 4∶1, *V*∶*V*) indicated that the reaction was complete. The reaction mixture was concentrated to dryness, which was purified by chromatography with petroleum ether-EtOAc (2∶1, *V*∶*V*) as the eluent to give triasaccharide **15.** To a solution of **15** (3.79 g, 2.8 mmol) in methanol (100 mL) was added $PdCl_2$ (60 mg, 0.34 mmol) and the mixture was stirred at room temperature for 2 h, at the end of which time TLC (petroleum ether-EtOAc, 2∶1, *V*∶*V*) indicated that the reaction was complete. The mixture was filtered, the filter cake was washed with dichloromethane, and the combined filtrate and washings were concentrated. Purification by chromatography with 2∶1 petroleum ether-EtOAc as the eluent afforded compound **16** (3.3 g, 91%) as a foamy solid. $[\alpha]_D$ −8.2 (*c*, 1.0, $CHCl_3$); ^{1}H NMR (400

MHz, $CDCl_3$), δ: 8.10—7.21 (m, 35H, 7×Bz-H), 5.85 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.70 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.50 (dd, $J_{1,2}=8.0$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 5.40 (dd, $J_{2,3}=J_{3,4}=9.6$ Hz, 1H, H-3), 5.30 (d, $J_{1,2}=3.6$ Hz, 1H, H-1), 5.21—5.12 (m, 2 H, H-2, H-5), 4.87 (d, $J_{1,2}=8.0$ Hz, 1H, H-1), 4.82—4.75 (m, 2H, H-1, H-4), 4.56 (dd, $J_{5,6}=2.9$ Hz, $J_{6,6a}=12.3$ Hz, 1H, H-6e), 4.44—4.35 (m, 4H, H-2, H-3, 2×H-6), 4.27 (dd, $J_{5,6}=3.2$ Hz, $J_{6,6a}=11.2$ Hz, 2H, 2×H-6e), 4.17—4.02 (m, 2H, H-3, H-5), 3.99 (ddd, $J_{4,5}=9.6$ Hz, $J_{5,6}=5.3$ Hz, $J_{5,6}=3.2$ Hz, 1H, H-5), 3.78 (dd, $J_{5,6}=5.3$ Hz, $J_{6,6e}=11.2$ Hz, 1H, H-6a), 1.67 (s, 3H, CH_3CO), 1.30 (s, 3H, CH_3), 1.15 (s, 3H, CH_3). Anal. calcd for $C_{72}H_{66}O_{24}$: C 65.75, H 5.06; found C 65.55, H 5.12.

2,3,4,6-Tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]2,4-di-*O*-acetyl-α-*D*-mannopyranosyl–(1→3)-2,4,6-tri-*O*-bezoyl-*β*-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]5-*O*-acetyl-1,2-*O*-isopropylidene-α-*D*-glucofuranose (17)

Coupling of **16** (158 mg, 0.12 mmol) with **9** (203 mg, 0.13 mmol) in the presence of catalytic TMSOTf (5 μL, 0.028 mmol) was carried out under the same conditions as described for the synthesis of **3** by coupling of **2** with **1**. Purification by a chromatography with petroleum ether-EtOAc (1∶1, *V*∶*V*) as the eluent gave hexasaccharide **17** (261 mg, 80%). $[\alpha]_D$ +12.6 (*c*, 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 8.21—7.22 (m, 75 H, 15×Bz-H), 5.85 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.84 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.73 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.68 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.63 (dd, $J_{2,3}=J_{3,4}=9.6$ Hz, 1H, H-3), 5.60 (dd, $J_{1,2}=7.8$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 5.55 (dd, $J_{1,2}=7.8$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 5.50 (dd, $J_{1,2}=7.8$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 5.35 (dd, $J_{2,3}=J_{3,4}=9.6$ Hz, 1H, H-3), 5.30 (dd, $J_{2,3}=J_{3,4}=9.6$ Hz, 1H, H-3), 5.19 (dd, $J_{1,2}=7.8$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 5.19 (ddd, $J_{4,5}=9.6$ Hz, $J_{5,6}=6.8$ Hz, $J_{5,6}=3.3$ Hz, 1H, H-5), 5.05 (d, $J_{1,2}=3.5$ Hz, 1H, H-1), 4.88 (d, $J_{1,2}=7.8$ Hz, 1H, H-1), 4.78—4.67 (m, 6H, 2×H-1, 2×H-4, 2×H-6), 4.64—4.53 (m, 4H, 2×H-1, 2×H-6), 4.50—4.41 (m, 5H, 2×H-2, H-3, 2×H-6), 4.29—4.16 (m, 4H, 2×H-5, 2×H-6), 4.14—4.00 (m, 3H, 2×H-3, H-5), 3.94 (dd, $J_{5,6}=6.6$ Hz, $J_{6,6a}=9.7$ Hz, 1H, H-6e), 3.85—3.76 (m, 2H, H-5, H-6), 3.63—3.55 (m, 2H, H-5, H-6), 3.26 (dd, $J_{5,6}=6.4$ Hz, $J_{6,6e}=10.5$ Hz, 1H, H-6a), 1.70 (s, 3H, CH_3CO), 1.47 (s, 3H, CH_3CO), 1.29 (s, 3H, CH_3CO) , 1.20 (s, 3H, CCH_3) , 0.78 (s, 3H, CCH_3); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 169.3 (1C, CH_3**C**O), 168.7 (2C, 2×CH_3**C**O), 165.7, 165.6, 165.3, 165.2, 165.16, 165.13, 164.7, 164.6, 164.5, 164.4, 163.9 (17C, 14×Ph**C**O, some signals overlapped), 133.0—132.5, 129.6—128.8, 128.6—127.7 (**Ph**CO), 111.7 (Me_2**C**), 104.4, 100.6, 100.6, 98.6, 97.6, 97.0, (6C, 6×C-1), 72.9, 72.6, 72.5, 72.3, 71.8, 71.7, 71.6, 71.5, 71.4, 71.1, 70.8, 70.7, 69.3, 69.2, 68.8, 68.5, 67.6, 67.4, 65.1, 62.7, 62.6, 62.4, 62.2, 59.9 (C-2—C-6), 26.2, 25.7 (2C, 2×C**C**H_3), 20.0, 19.8, 18.6 (3C, 3×CO**C**H_3). Anal. calcd for $C_{150}H_{132}O_{49}$: C 66.27, H 4.89; found C 66.02, H, 4.77.

β-*D*-Glucopyranosyl-(1→3)-[β-*D*-glucopyranosyl-(1→6)-]α-*D*-mannopyranosyl-(1→3)-β-*D*-glucopyranosyl-(1→3)-[β-*D*-glucopyranosyl-(1→6)-]β-*D*-glucopyranose (18)

The solution of **17** (272 mg, 0.1 mmol) in 10 mL of 90% TFA was stirred for 2 h, at the end of which time the reaction mixture was poured directly to 250 mL of toluene and concentrated. Drying the residue under high vacuum gave a white foamy solid. The foamy solid was dissolved in a saturated solution of ammonia in MeOH (10 mL). After two weeks at room temperature, the reaction solution was concentrated, and the residue was purified on a Biogel P2 column with MeOH-water as the eluent to afford **18** (79 mg, 80% for two steps) as a foamy solid. $[\alpha]_D$ −12.2 (*c* 1.0, H_2O); ^{1}H NMR (D_2O, 400 MHz) δ: 5.16 (d, $J_{1,2}=1.6$ Hz, 1H, H-1), 4.73 (d, $J_{1,2}=8.1$ Hz , 1H, H-1), 4.70 (d, $J_{1,2}=8.7$ Hz , 1H, H-1), 4.53 (d, $J_{1,2}=9.4$ Hz, 1H, H-1), 4.41 (d, $J_{1,2}=8.0$ Hz, 1H, H-1), 4.39 (d, $J_{1,2}=8.0$ Hz, 1H, H-1), 4.18—3.20 (m, 36 H, H-2—H-6); ^{13}C NMR (D_2O, 100 MHz) δ: 105.2, 105.2, 105.1, 103.0, 102.8, 102.8 ($J_{C\text{-}1,H\text{-}1}=160.0$, 160.0, 160.0, 162.4, 162.4, and 176.0 Hz respectively, 6×C-1), 82.6, 79.0, 78.4, 78.3, 78.0, 77.2, 75.5, 75.3, 75.2, 72.4, 72.3, 72.2, 70.2, 67.3, 63.1, 62.9 (C-2—C-6). Anal. calcd for $C_{36}H_{62}O_{31}$: C 43.64, H 6.31; found C 43.43, H, 6.18.

3-*O*-Allyl-2,4,6-tri-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]2,4-di-*O*-acetyl-α-*D*-glucopyranosyl trichloroacetimidate (22)

Compound **14** (341 mg, 0.26 mmol) was dissolved in 35 mL of 90% TFA and stirred for 2 h, at the end of which time the reaction mixture was poured directly to 100 mL of toluene and concentrated. Drying the residue under high vacuum gave **19** as a white foamy solid. This foamy solid was dissolved in pyridine (10 mL), and then Ac_2O (5 mL) was added. The reaction mixture was stirred at room temperature for 12 h, and TLC (petroleum ether-EtOAc, 2∶1, *V*∶*V*) indicated that the reaction was complete. The reaction mixture was concentrated to dryness. The resultant crude product **20** was dissolved in 1 mol/L solution of ammonia-methanol (100 mL) and stirred at room temperature for 3 h, at the end of which time TLC (petroleum ether-EtOAc, 1∶1, *V*∶*V*) indicated that the reaction was complete. The solution was concentrated to give compound **21** as a syrup. A mixture of **21**, trichloroacetonitrile (0.4 mL, 2 mmol), and 1,8- diazabicyclo [5.4.0] undecene (DBU) (0.05 mL, 0.4 mmol) in dry dichloromethane (10 mL) was stirred for 3 h and then concentrated. The residue was purified by flash chromatography with 2:1 petroleum ether-EtOAc as the eluent to give **22** (242 mg, 62 % for four steps) as a foamy solid. $[\alpha]_D$ +15.6 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 8.30 (s, 1H, $NHCCl_3$), 8.15—7.13 (m, 35H, 7×Bz-H), 6.18 (d, $J_{1,2}=3.5$ Hz, 1H, H-1), 5.84 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H,

H-4), 5.61 (dd, $J_{3,4}=J_{4,5}=9.7$ Hz, 1H, H-4), 5.56—5.51 (m, 1H, $CH_2C\mathbf{H}=CHH$), 5.46 (dd, $J_{1,2}=8.0$ Hz, $J_{2,3}=9.7$ Hz, 1H, H-2), 5.42 (dd, $J_{2,3}=J_{3,4}=9.7$ Hz, 1H, H-3), 5.17 (dd, $J_{3,4}=J_{4,5}=8.4$ Hz, 1H, H-4), 4.99 (dd, $J=1.4$, 17.2 Hz, 1H, $CH_2CH=C\mathbf{H}H$), 4.95 (s, $J_{1,2}=7.8$ Hz, 1H, H-1), 4.89 (dd, $J=1.4$, 0.5 Hz, 1H, $CH_2CH=CH\mathbf{H}$), 4.85 (s, $J_{1,2}=8.0$ Hz, 1H, H-1), 4.83 (dd, $J_{1,2}=7.8$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 4.63 (dd, $J_{1,2}=3.5$ Hz, $J_{2,3}=9.7$ Hz, 1H, H-2), 4.65— 4.57 (m, 2H, 2×H-6), 4.47 (dd, $J_{5,6}=5.7$ Hz, $J_{6,6a}=12.2$ Hz, 1H, H-6e), 4.32 (dd, $J_{5,6}=6.2$ Hz, $J_{6,6e}=12.2$ Hz, 1H, H-6a), 4.16—3.88 (m, 8H, 2×H-3, 2×H-5, 2×H-6, $C\mathbf{H}_2CH=CHH$), 3.67 (ddd, $J_{4,5}=9.6$ Hz, $J_{5,6}=6.2$ Hz, $J_{5,6}=5.7$ Hz, 1H, H-5), 2.00 (s, 3H, CH_3CO), 1.81 (s, 3H, CH_3CO). Anal. calcd for $C_{76}H_{68}$- Cl_3NO_{25}: C 60.79, H 4.56; found C 60.53, H 4.64.

Methyl 3-*O*-allyl-2,4,6-tri-*O*-benzoyl-*β*-*D*-glucopyransyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]2,4-di-*O*-acetyl-*β*-*D*-glucopyranosyl-(1→3)-4,6-*O*-benzylidene-*α*-*D*-glucopyranoside (24)

Coupling of **22** (346 mg, 0.23 mmol) with **23** (59 mg, 0.21 mmol) in the presence of catalytic TMSOTf (10 μL, 0.056 mmol) was carried out under the same conditions as described for the synthesis of **3** by coupling of **2** with **1**. Purification by a chromatography with 3:2 petroleum ether-EtOAc as the eluent gave tetrasaccharide **24** (238 mg, 70%). $[\alpha]_D+14.6$ (*c*, 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) *δ*: 8.11—7.22 (m, 40H, 7×Bz-H, Ph-H), 5.87 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.64 (dd, $J_{3,4}=J_{4,5}=9.7$ Hz, 1H, H-4), 5.59—5.44 (m, 5H, PhC**H**, $CH_2C\mathbf{H}=CHH$, 2×H-2, H-3), 5.11 (dd, $J_{3,4}=J_{4,5}=9.7$ Hz, 1H, H-4), 4.98 (dd, $J=1.5$, 17.2 Hz, 1H, $CH_2CH=C\mathbf{H}H$), 4.93 (d, $J_{1,2}=7.9$ Hz, 1H, H-1), 4.89 (dd, $J=1.5$, 10.4 Hz, 1H, $CH_2CH=CH\mathbf{H}$), 4.86 (d, $J_{1,2}=8.0$ Hz, 1H, H-1), 4.84 (d, $J_{1,2}=8.2$ Hz, 1H, H-1), 4.62 (d, $J_{1,2}=3.5$ Hz, 1H, H-1), 4.72—4.63 (m, 3H, H-2, 2×H-6), 4.56 (dd, $J_{5,6}=5.9$ Hz, $J_{6e,6a}=12.7$ Hz, 2H, 2×H-6a), 4.46—4.38 (m, 4H, 4×H-6), 4.19—3.42 (m, 11H, H-2, 3×H-3, H-4, 4×H-5, $C\mathbf{H}_2CH=CHH$), 3.19 (s, 3H, CH_3O), 1.93 (s, 3H, CH_3CO), 1.86 (s, 3H, CH_3CO). Anal. calcd for $C_{88}H_{84}O_{30}$: C 65.18, H 5.22; found C 65.44, H, 5.10.

Methyl 2,4,6-tri-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]2,4-di-*O*-acetyl-*β*-*D*-glucopyranosyl-(1→3)-2,4,6-tri-*O*-acetyl-α-*D*-glucopyranoside (27)

To a solution of **24** (178 mg, 0.11 mmol) in anhydrous MeOH (100 mL) was added acetyl chloride (0.1 mL). The flask was stoppered and the solution was stirred at room temperature until TLC (EtOAc) showed that the starting material disappeared. The solution was neutralized with Et_3N, then concentrated to dryness. The residue was purified by chromatography with petroleum ether-EtOAc (1∶2, *V*∶*V*) as the eluent to give **25** as a white solid. This white solid was dissolved in pyridine (20 mL), and then Ac_2O (10 mL) was added. The reaction mixture was stirred at r.t. for 12 h, and TLC (1∶1 petroleum ether-EtOAc) indicated that the reaction was complete. The reaction mixture was concentrated to dryness, which was purified by chromatography with petroleum ether-EtOAc (1∶1, *V*∶*V*) as the eluent to give tetrasaccharide **26**. To a solution of compound **26** in methanol (100 mL) was added $PdCl_2$ (30 mg, 0.17 mmol) and the mixture was stirred at room temperature for 2 h, at the end of which time TLC (petroleum ether-EtOAc, 1∶1, *V*∶*V*) indicated that the reaction was complete. The mixture was filtered, the filter cake was washed with dichloromethane, and the combined filtrate and washings were concentrated. Purification by column chromatography with petroleum ether-EtOAc (1∶1, *V*∶*V*) as the eluent afforded compound **27** (128 mg, 72% for three steps) as a foamy solid. $[\alpha]_D+24.5$ (*c*, 1.0, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) *δ*: 8.06—7.22 (m, 35H, 7×Bz-**H**), 5.88 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.65 (dd, $J_{3,4}=J_{4,5}=9.7$ Hz, 1H, H-4), 5.48 (dd, $J_{1,2}=7.9$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 5.40 (dd, $J_{2,3}=J_{3,4}=9.7$ Hz, 1H, H-3), 5.28 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.05 (dd, $J_{1,2}=7.9$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 4.95 (dd, $J_{3,4}=J_{4,5}=9.7$ Hz, 1H, H-4), 4.89 (d, $J_{1,2}=7.9$ Hz, 1H, H-1), 4.77 (dd, $J_{1,2}=7.9$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 4.76 (dd, $J_{1,2}=3.6$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 4.73 (d, $J_{1,2}=7.9$ Hz, 1H, H-1), 4.68 (d, $J_{1,2}=3.6$ Hz, 1H, H-1), 4.65 (dd, $J_{5,6}=3.4$ Hz, $J_{6e,6a}=12.6$ Hz, 1H, H-6e), 4.60 (dd, $J_{5,6}=3.4$ Hz, $J_{6a,6e}=12.6$ Hz, 1H, H-6a), 4.49—4.38 (m, 2H, 2×H-6), 4.34 (d, $J_{1,2}=7.9$ Hz, 1H, H-1), 4.20 (dd, $J_{5,6}=5.9$ Hz, $J_{6,6a}=12.3$ Hz, 1H, H-6e), 4.17 —3.53 (m, 10H, 3×H-3, 4×H-5, 3×H-6), 3.20 (s, 3H, CH_3O), 2.02 (s, 3H, CH_3CO), 1.99 (s, 3H, CH_3CO), 1.97 (s, 3H, CH_3CO), 1.93 (s, 3H, CH_3CO), 1.87 (s, 3H, CH_3CO). Anal. calcd for $C_{84}H_{82}O_{33}$: C 62.30, H 5.10; found C 62.01, H, 5.23.

Methyl 2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]2,4-di-*O*-acetyl-*α*-*D*-mannopyranosyl-(1→3)-2,4,6-tri-O-benzoyl-*β*-*D*-glucopyranosyl-(1→3)-[2,3,4,6-tetra-*O*-benzoyl-*β*-*D*-glucopyranosyl-(1→6)-]2,4-di-*O*-acetyl-*β*-*D*-glucopyranosyl-(1→3)-2,4,6-tri-*O*-acetyl-*α*-*D*-glucopyranoside (28)

Coupling of **27** (110 mg, 0.068 mmol) with **9** (125 mg, 0.08 mmol) in the presence of catalytic TMSOTf (5 μL, 0.028 mmol) was carried out under the same conditions as described for the synthesis of **3** by coupling of **2** with **1**. Purification by chromatography with petroleum ether-EtOAc (1∶1, *V*∶*V*) as the eluent gave heptasaccharide **28** (166 mg, 81%). $[\alpha]_D+25.5$ (*c*, 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz) *δ*: 8.16—7.17 (m, 75H, 15×Bz-H), 5.88 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 5.82 (dd, $J_{3,4}=J_{4,5}=9.5$ Hz, 1H, H-4), 5.73—5.57 (m, 4H, 2×H-3, 2×H-4), 5.49 (dd, $J_{1,2}=7.9$ Hz, $J_{2,3}=9.6$ Hz, 1H, H-2), 5.45 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-3), 5.40 (dd, $J_{1,2}=7.9$ Hz, $J_{2,3}=9.5$ Hz, 1H, H-2), 5.29—5.23 (m, 2H, 2×H-2), 4.95 (dd, $J_{3,4}=J_{4,5}=9.6$ Hz, 1H, H-4), 4.90 (d, $J_{1,2}=7.9$ Hz, 1H, H-1), 4.84—4.75 (m, 4H, H-1, 2×H-4, H-6), 4.73—4.38 (m, 14H, 5×H-1, 9×H-6), 4.34—3.52 (m, 18H, 3×H-2, 4×H-3, 7×H-5, 4×H-6),

3.20 (s, 3H, CH_3O), 2.02, 2.00, 1.99, 1.98 (s, 12H, 4× CH_3CO), 1.93 (s, 3H, CH_3CO), 1.88 (s, 3H, CH_3CO), 1.53 (s, 3H, CH_3CO); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 170.7, 169.8, 169.7, 169.5, 169.4, 169.3, 167.7 (7C, 7× CH_3**C**O), 166.1, 166. 0, 165.9, 165.5, 165.4, 165.1, 165.0, 164.8, 164.1 (15C, 15×Ph**C**O, some signals overlapped), 133.7—132.2, 130.8, 130.1—129.0, 128.8—127.9 (**Ph**C), 101.2, 101.0, 100.8, 100.2, 98.7, 98.0, 97.8 (7C, 7×C-1), 73.5, 73.2, 72.8, 72.3, 72.2, 71.8, 71.7, 71.4, 71.0, 69.5, 69.2, 69.0, 68.8, 68.3, 68.1, 67.4, 66.6, 65.4, 63.2, 62.9, 62.8, 62.5, 62.1 (C-2—C-6), 55.4 (1C, **C**H_3O), 20.6, 20.6, 20.6, 20.5, 20.2, 18.9 (7C, 7×**C**H_3CO, some signals overlapped). Anal. calcd for $C_{162}H_{148}O_{58}$: C 64.37, H 4.93; found: C 64.03, H, 4.80.

Methyl *β*-*D*-glucopyranosyl-(1→3)-[*β*-*D*-glucopyranosyl-(1→6)-] *α*-*D*-mannopyranosyl-(1→3)-*β*-*D*-glucopyranosyl-(1→3)-[*β*-*D*-glucopyranosyl-(1→6)-]*β*-*D*-glucopyranosyl-(1→3)-*α*-*D*-glucopyranoside (29)

Compound **28** (272 mg, 0.09 mmol) was dissolved in a saturated solution of ammonia in MeOH (10 mL). After two weeks at room temperature, the reaction solution was concentrated, and the residue was purified on a Biogel P2 column with MeOH-water as the eluent to afford **29** (100 mg, 95%) as a foamy solid. $[\alpha]_D$ +16.5 (*c* 1.0, H_2O); ^1H NMR (D_2O, 400 MHz) δ: 5.23 (d, $J_{1,2}$=2.5 Hz, 1H, H-1), 4.98 (d, $J_{1,2}$=3.6 Hz, 1H, H-1), 4.70 (d, $J_{1,2}$=7.9 Hz, 1H, H-1), 4.56 (d, $J_{1,2}$=8.0 Hz, 1H, H-1), 4.51 (d, $J_{1,2}$=8.0 Hz, 1H, H-1), 4.43 (d, $J_{1,2}$= 7.9 Hz, 1H, H-1), 4.40 (d, $J_{1,2}$=8.0 Hz, 1H, H-1), 4.18—3.15 (m, 45H, H-2—H-6, CH_3); ^{13}C NMR (D_2O, 100 MHz) δ: 103.7, 102.5, 102.4, 102.3, 100.4, 100.0, 98.4 ($J_{C\text{-}1,H\text{-}1}$=165.1, 165.1, 165.1, 165.1, 165.1, 176.4, and 176.4 Hz respectively, 7C-1), 83.6, 82.2, 80.0, 77.8, 75.5, 75.4, 75.3, 75.1, 75.0, 74.8, 74.0, 72.7, 72.6, 72.5, 71.8, 71.6, 71.5, 70.8, 69.4, 69.2, 69.1, 69.0, 68.4, 68.1, 67.5, 67.4, 57.0 (C-2—C-6, O**C**H_3). Anal. calcd for $C_{43}H_{74}O_{36}$: C 44.25, H 6.39; found C 44.03, H, 6.28.

References

1 Sasaki, T.; Takasuka, N. *Carbohydr. Res.* **1976**, *47*, 99.

2 Bao, X.; Liu, C.; Fang, J.; Li, X. *Carbohydr. Res.* **2001**, *332*, 67.

3 Saito, H.; Ohki, T.; Takasuka, N.; Sasaki, T. *Carbohydr. Res.* **1977**, *58*, 293.

4 Ning, J.; Zhang, W.; Yi, Y.; Yang, G.; Wu, Z.; Yi, J.; Kong, F. *Bioorg. Biomed. Chem.* **2003**, *11*, 2193.

5 (a) Inoue, K.; Kawamoto, K.; Kadoya, S. *Carbohydr. Res.* **1983**, *114*, 245.
(b) Inoue, K.; Kohno, M.; Kadoya, S. *Carbohydr. Res.* **1983**, *123*, 305.

6 Zhang, J.; Kong, F. *Tetrahedron: Asymmetry* **2002**, *13*, 243.

7 Schmidt, R. R.; Kinzy, W. *Adv. Carbohydr. Chem. Biochem.* **1994**, *50*, 21.

8 Zeng, Y.; Ning, J.; Kong, F. *Tetrahedron Lett*. **2002**, *20*, 3729.

9 Söderberg, E.; Westman, J.; Oscarson, S. *J. Carbonhydr. Chem.* **2001**, *20*, 397.

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