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Bioorganic & Medicinal Chemistry 11 (2003) 4027–4037

BIOORGANIC &  
MEDICINAL  
CHEMISTRY

# Facile Syntheses of the Hexasaccharide Repeating Unit of the Exopolysaccharide from *Cryptococcus Neoformans* Serovar A

Jianjun Zhang and Fanzuo Kong\*

Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, Beijing 100085, China

Received 25 March 2003; accepted 29 May 2003

**Abstract**—Two hexasaccharides,  $\beta$ -D-Xylp-(1 $\rightarrow$ 2)- $\alpha$ -D-Manp-(1 $\rightarrow$ 3)-[ $\beta$ -D-Xylp-(1 $\rightarrow$ 2)-] $\alpha$ -D-Manp-(1 $\rightarrow$ 3)-[ $\beta$ -D-GlcA-(1 $\rightarrow$ 2)-] $\alpha$ -D-Manp and  $\beta$ -D-GlcA-(1 $\rightarrow$ 2)- $\alpha$ -D-Manp-(1 $\rightarrow$ 3)-[ $\beta$ -D-Xylp-(1 $\rightarrow$ 2)-] $\alpha$ -D-Manp-(1 $\rightarrow$ 3)-[ $\beta$ -D-Xylp-(1 $\rightarrow$ 2)-] $\alpha$ -D-Manp, the repeating unit of the exopolysaccharide from *Cryptococcus neoformans* serovar A, were synthesized as their methyl glycosides in a regio- and stereoselective manner.

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## Introduction

*Cryptococcus neoformans* is an opportunistic pathogenic yeast that causes a life-threatening meningoencephalitis in individuals with an impaired immune system.<sup>1</sup> The incidence of cryptococcosis has increased dramatically in recent years as a consequence of the AIDS epidemic, and it is a leading cause of death in patients with AIDS<sup>2–4</sup>. *C. neoformans* is unusual among pathogenic fungi in that it has a polysaccharide capsule, glucuronoxylomannan (GXM).<sup>5</sup> GXM is antipathogenic and poorly immunogenic, and acapsular strains have significantly reduced virulence.<sup>6</sup> In vitro, GXM inhibits leukocyte migration,<sup>7</sup> enhances HIV infection in human lymphocytes<sup>8</sup> and promotes L-selectin shedding from neutrophils.<sup>9</sup> There are four major serotypes<sup>10</sup> for GXM designated A–D (Scheme 1). All the four serotypes are composed of a linear  $\alpha$ -1,3-linked mannosyl backbone with  $\beta$ -glucopyranosyluronic acid,  $\beta$ -xylopyranosyl, and 6-*O*-acetyl substituents.<sup>11</sup> Serotype D is the most heavily *O*-acetylated and serotype C the least *O*-acetylated. However, the *O*-acetyl groups are not essential for binding, though they have a significant contribution.<sup>5</sup>

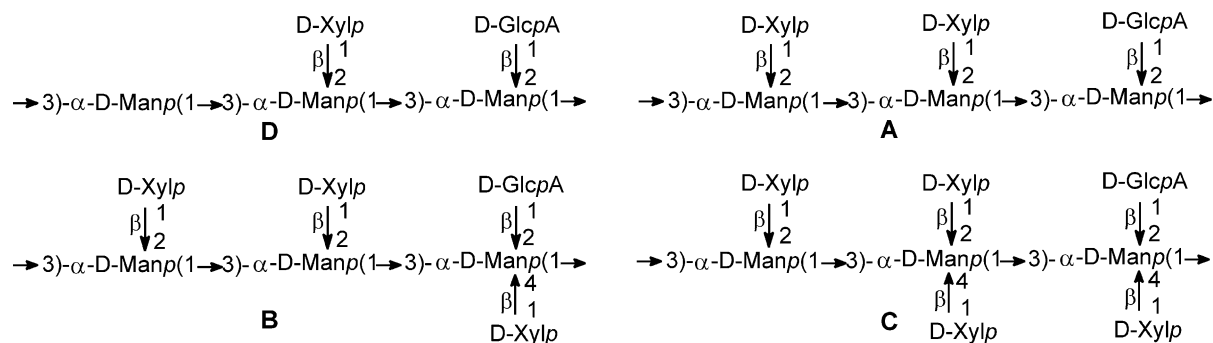
As part of an ongoing project to synthesize oligosaccharides<sup>12</sup> existing in cell-wall polysaccharides, we have reported successful syntheses of 3,6-branched glucans,<sup>12a,b,c</sup> 3- or 2-branched rhamnans,<sup>12d,e</sup> 2,6-<sup>12f,g</sup> and 3,6-branched mannans.<sup>12h,i</sup> A major problem in the

assembly of a repeating unit of the exopolysaccharide of *C. neoformans* entails inter alia the introduction of a  $\beta$ -D-glucuronic acid residue at the axially orientated *O*-2 position of a mannopyranoside moiety.<sup>13</sup> The synthesis of trisaccharide and tetrasaccharide fragments corresponding to structures in capsular polysaccharides of *C. neoformans* has been reported<sup>14,15</sup> and the synthesis of a pentasaccharide—the repeating unit of the polysaccharide in *C. neoformans* serovar D has appeared.<sup>16</sup> In these syntheses, multiple steps and orthogonal masking groups were involved making the procedure rather complex. As a result, it would be difficult to synthesize by the reported methods, the higher oligosaccharides — the repeating units of *C. neoformans* serotypes A, B, and C. Previously, we have reported the regio- and stereoselective synthesis of oligosaccharides with un- or lightly-protected mannose<sup>17</sup> and rhamnose<sup>18</sup> as the glycosyl acceptors and glycosyl trichloroacetimidates as the donors giving satisfactory results. We also reported in a preliminary communication<sup>19</sup> the synthesis of the hexasaccharide repeating unit of *O*-deacetylated GXM of *C. neoformans* serotype A with 4,6-*O*-isopropylidened mannose derivatives as the acceptors. We present herein a new regio- and stereoselective synthesis of the frame-shifted hexasaccharide and the synthesis in the precedent communication in detail.

## Results and Discussion

Scheme 2 outlines the new synthesis of the frame-shifted hexasaccharide,  $\beta$ -D-Xylp-(1 $\rightarrow$ 2)- $\alpha$ -D-Manp-(1 $\rightarrow$ 3)-[ $\beta$ -D-

\*Corresponding author. Tel.: +86-10-629-36613; fax: +86-10-629-23563; e-mail: fzkong@mail.rcees.ac.cn

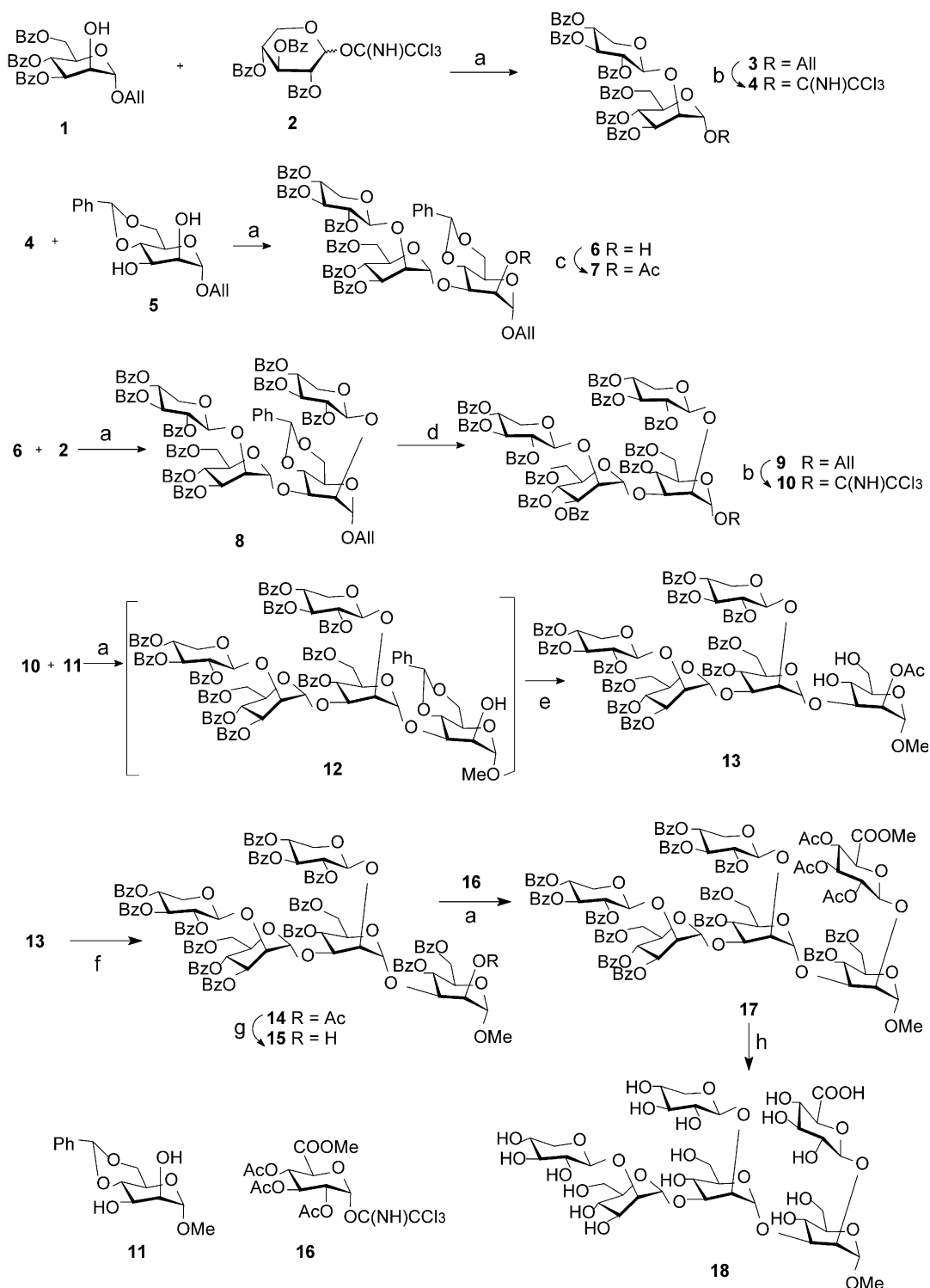


**Scheme 1.** Model structures of GXM of *C. neoformans* serotypes A–D.

Xylp-(1→2)-]α-D-Manp-(1→3)-[β-D-GlcpA-(1→2)-]α-D-Manp. Due to the unstability of methyl ester linkage of glucuronate residue under either basic or acidic conditions, assembly of the glucuronate unit was arranged at the end of the reaction series. So, a synthetic route starting from construction of the nonreducing end blocks and ending with the glucuronosylation was designed and carried out. Therefore, allyl 3,4,6-tri-*O*-benzoyl-α-D-mannopyranoside **1**, obtained from rearrangement of 3,4,6-tri-*O*-benzoyl-1,2-*O*-allyloxyethylidene-α-D-mannopyranose<sup>20</sup> followed by selective deacetylation,<sup>21</sup> was chosen as the starting material. Coupling of **1** with 2,3,4-tri-*O*-benzoyl-α,β-D-xylopyranosyl trichloroacetimidate **2** afforded (1→2)-linked disaccharide **3**. Subsequent 1-*O*-deallylation with PdCl<sub>2</sub> and activation with trichloroacetonitrile<sup>22</sup> in the presence of potassium carbonate gave the disaccharide donor **4**. Condensation of **4** with the acceptor allyl 4,6-*O*-isopropylidene-α-D-mannopyranoside **5** selectively afforded the (1→3)-linked trisaccharide **6** (80.3%). The regioselectivity of the coupling was confirmed by acetylation of **6** to give **7**, and the <sup>1</sup>H NMR spectrum of **7** showed a newly emerged downfield doublet of doublets at δ 5.40 ppm with *J*<sub>1,2</sub>=0.6 and *J*<sub>2,3</sub>=3.0 Hz for H-2, compared to that of **6**. Coupling of **6** with **2** gave the tetrasaccharide **8**. Because of the presence of benzylidene group, an attempt for deallylation of **8** with PdCl<sub>2</sub> was not successful since a complex product was obtained. Thus, debenzylidenation of **8** with 90% TFA followed by benzylation gave fully benzyolated tetrasaccharide **9** (91.9%). Dallylation of **9** followed by trichloroacetimidation afforded the tetrasaccharide donor **10**. Again, 3-*O*-selective glycosylation of methyl 4,6-*O*-isopropylidene-α-D-mannopyranoside **11** with the donor **10** yielded (1→3)-linked pentasaccharide **12**. Purification of **12** was not easy since an unidentified byproduct had very similar behavior to **12** in column chromatography separation. Thus, acetylation of **12** followed by debenzylidenation yielded the pentasaccharide diol **13** (54.3% for three steps) that was easily purified. The <sup>1</sup>H NMR spectrum of **13** showed a characteristic downfield doublet of doublets at δ 5.24 ppm with *J*<sub>1,2</sub>=1.4 and *J*<sub>2,3</sub>=3.2 Hz for H-2, confirming the regioselectivity in the coupling reaction. Benzylation of **13** was difficult at room temperature due to the steric hindrance caused by xylose residue. So, the benzylation was carried out in the presence of catalytic DMAP at 70 °C for 12 h, and **14** was obtained in satisfactory yield (79%). Subsequent selective deacetylation with CH<sub>3</sub>COCl–MeOH–CH<sub>2</sub>Cl<sub>2</sub>

gave the pentasaccharide acceptor **15**. TMSOTf promoted glucuronosylation of **15** with methyl 2,3,4-tri-*O*-acetyl-α-D-glucuronate trichloroacetimidate **16** went smoothly giving the required hexasaccharide **17** in satisfactory yield (72.7%). The <sup>13</sup>C NMR spectrum of **17** showed all of the characteristic signals such as two methyl signals (δ 54.7 and 52.7 ppm, respectively), 13 benzoyl C=O signals (δ 167.3, 167.2, 166.1, 166.0, 165.8, 165.4, 165.3, 165.3, 165.3, 165.2, 165.2, 165.1, 164.6 ppm), four acetyl C=O signals (δ 171.3, 171.0, 169.7, 169.2 ppm), and six anomeric C signals 102.6, 100.6, 100.6, 99.6, 98.9, 98.2. Deprotection of **17** was carried out in methanol with MeONa for 96 h, then water (10 equiv) was added to cleave the methyl ester. After stirring at room temperature for 2 h, the reaction mixture was concentrated and purified on a Bio-Gel P2 column (eluent: water), affording the target hexasaccharide **18** as a foamy solid. Determination of *J*<sub>C1,H1</sub> of **18** confirmed mannosyl linkages (*J*<sub>C1,H1</sub>=176.2, 172.7, and 174.1 Hz), β-xylosyl linkages (*J*<sub>C1,H1</sub>=163.4 and 163.4 Hz), and β-glucopyranosyluronic acid linkage (*J*<sub>C1,H1</sub>=166.8 Hz).

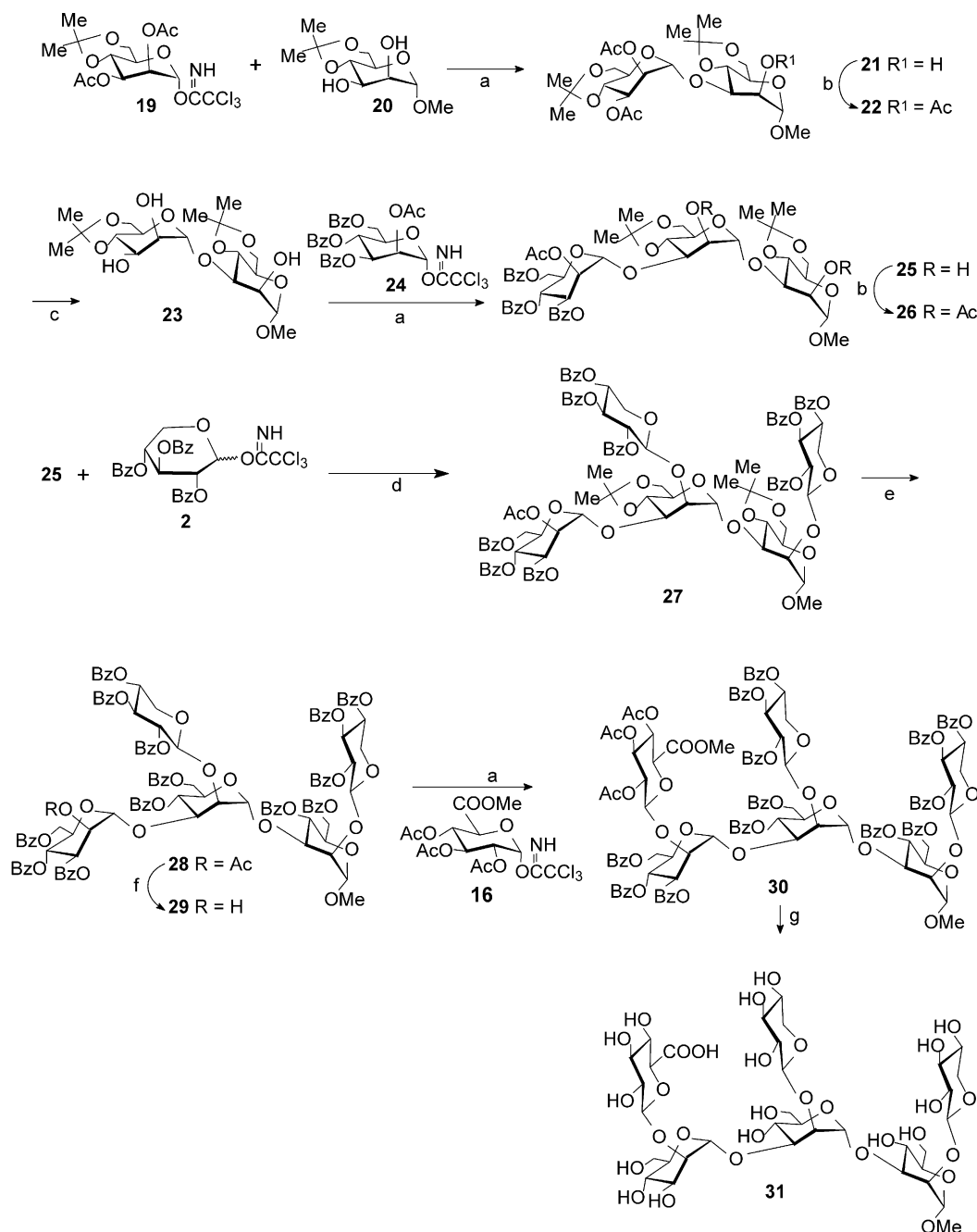
**Scheme 3** shows synthesis of the another hexasaccharide, β-D-GlcpA-(1→2)-α-D-Manp-(1→3)-[β-D-Xylp-(1→2)-]α-D-Manp-(1→3)-[β-D-Xylp-(1→2)-]α-D-Manp, with the glucuronic acid residue at the nonreducing end. For the same reason of the unstability of methyl ester of glucuronate, the synthesis started from construction of the reducing end blocks, then extended to nonreducing end, and finally ended with the glucuronosylation. Thus, 2,3-di-*O*-acetyl-4,6-*O*-isopropylidene-α-D-mannopyranosyl trichloroacetimidate **19** was obtained from selective 4,6-*O*-isopropylidenation of mannose,<sup>23</sup> acetylation, selective 1-*O*-deacetylation, and followed by trichloroacetimidation. Condensation of **19** with the acceptor methyl 4,6-*O*-isopropylidene-α-D-mannopyranoside **20** selectively afforded the (1→3)-linked disaccharide **21** (74%). The regioselectivity of the coupling was confirmed by acetylation of **21** to give **22**, and the <sup>1</sup>H NMR spectrum of **22** showed a newly emerged downfield doublet of doublets at δ 5.34 ppm with *J*<sub>1,2</sub>=1.5 and *J*<sub>2,3</sub>=3.0 Hz for H-2, compared to that of **21**. Deacetylation of **21** or **22** in a solution of ammonia in methanol furnished the disaccharide triol acceptor **23** quantitatively. Again, 3-*O*-selective glycosylation of **23** with the donor 2-*O*-acetyl-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl trichloroacetimidate **24** yielded (1→3)-linked trisaccharide **25** (67%). The trisaccharide **25** was



**Scheme 2.** Reagents and conditions: (a) TMSOTf (0.01–0.25 equiv), 4 Å MS, CH<sub>2</sub>Cl<sub>2</sub>, –20 °C, 2–4 h (86% for **3**, 80% for **6**, 64% for **8**, 54% for **12**, 73% for **17**); (b) (i) PdCl<sub>2</sub>, HOAc–NaOAc, rt, 10–12 h; (ii) CH<sub>2</sub>Cl<sub>2</sub>, CCl<sub>3</sub>CN (2–3.3 equiv), DBU (0.18–0.2 equiv), rt, 2 h, (90% for **4**, 85% for **10**); (c) Ac<sub>2</sub>O–pyridine, 100%; (d) (i) 90% TFA, rt, 2 h; (ii) BzCl–pyridine, rt, 10 h, 92%; (e) (i) Ac<sub>2</sub>O–pyridine; (ii) 90% TFA, rt, 2 h; (f) BzCl–pyridine–DMAP, 70 °C, 12 h, 79%; (g) 2.5% CH<sub>3</sub>COCl in CH<sub>2</sub>Cl<sub>2</sub>–CH<sub>3</sub>OH, rt, 89%; (h) MeONa–MeOH, rt, 96 h, then water was added, 2 h, 64%.

an ideal acceptor since it contained two free hydroxyl groups at the positions where the xylosyl residue should be attached. Thus, TMSOTf promoted dixylosylation of **25** with 2,3,4-tri-*O*-benzoyl-D-xylopyranosyl trichloroacetimidate **2** was achieved with an ‘inverse Schmidt’ strategy, that is after formation of monoxylated tetrasaccharide, additional TMSOTf was added,

and after stirring the reaction mixture for 20 min, further 2 equiv of donor **2** was added. Purification of **27** was not easy since the tetrasaccharide byproducts had very similar behavior to **27**. Thus, deisopropylidenation of **27** in 90% TFA followed by benzoylation with benzoyl chloride in pyridine gave the protected pentasaccharide **28** (42% for three steps). Selective deacetylation of **28**



**Scheme 3.** Model structures of GXM of *C. neoformans* serotypes A–D. Reagents and conditions: (a) TMSOTf (0.01–0.05 equiv), 4 Å MS,  $\text{CH}_2\text{Cl}_2$ ,  $-20^\circ\text{C}$ , 2–4 h (74% for **21**, 67% for **25**, 88% for **30**); (b)  $\text{Ac}_2\text{O}$ –pyridine, 100%; (c)  $\text{CH}_3\text{OH}$  saturated with ammonia, rt, 12 h, 100%; (d) **2** (2 equiv), TMSOTf (0.1 equiv), 4 Å MS,  $\text{CH}_2\text{Cl}_2$ ,  $-0^\circ\text{C}$ , 0.5 h; then TMSOTf (1.0 equiv), 0.5 h, and **2** (2 equiv) was added, 42%; (e) (i) 90% HOAc,  $60^\circ\text{C}$ , 20 h; (ii)  $\text{BzCl}$ –pyridine, rt, 10 h, 91%; (f) 2%  $\text{CH}_3\text{COCl}$  in  $\text{CH}_2\text{Cl}_2$ – $\text{CH}_3\text{OH}$ , rt, 20 h, 67%; (g) methanol saturated with ammonia, rt, 36 h, then water (2 equiv) was added, 5 h, 65%.

with 2% acetyl chloride–methanol gave the penta-saccharide acceptor **29** in 67% yield. Coupling of **29** with methyl 2,3,4-tri-*O*-acetyl- $\alpha$ -D-glucopyranosyluronate trichloroacetimidate **16** went smoothly affording the protected hexasaccharide in good yield (88%). The  $^1\text{H}$  and  $^{13}\text{C}$  NMR spectra of **30** showed two methyl signals ( $\delta$  3.69 and 3.23 ppm, respectively), 13 benzoyl C=O signals ( $\delta$  165.9, 165.9, 165.9, 165.4, 165.4, 165.4, 165.3, 165.2, 165.1, 165.0, 164.9, 164.6, 164.6 ppm), four acetyl C=O signals ( $\delta$  167.0, 168.5, 168.5, 168.3 ppm), six anomeric C signals (100.9,  $J_{\text{C1,H1}} = 175$  Hz, Manp; 100.3,  $J_{\text{C1,H1}} = 163$  Hz, GluAp; 99.9,  $J_{\text{C1,H1}} = 164$  Hz,

Xylp; 99.5,  $J_{\text{C1,H1}} = 163$  Hz, Xylp; 98.5,  $J_{\text{C1,H1}} = 172$  Hz, Manp; 95.2,  $J_{\text{C1,H1}} = 176$  Hz, Manp). Deprotection of **30** was carried out in a saturated solution of ammonia in methanol for 36 h, then water was added to cleave the methyl ester. After standing at room temperature for 5 h, the reaction mixture was concentrated and purified on a Bio-Gel P2 column (eluent: water), affording the target hexasaccharide **31** as a foamy solid.

In summary, efficient syntheses of the hexasaccharide repeat unit of *O*-deacetylated GXM of *C. neoformans* serotype A with lightly protected mannose derivatives

as the acceptors was achieved. Glycosylation of either 4,6-*O*-isopropylidenated or 4,6-*O*-benzylidenated mannopyranosides showed excellent 3-*O*-selectivity. The strategy presented here also provides a route to the synthesis of more complex repeating units of GXM of *C. neoformans* serotype B and C.

### Experimental

Melting points were determined with a 'Mel-Temp' apparatus. Optical rotations were determined at 25 °C with digital polarimeter. The NMR spectra were recorded in CDCl<sub>3</sub> with TMS internal standard or D<sub>2</sub>O with ethanol as standard on ARX 400 MHz. Mass spectra were recorded on an autospec mass spectrometer using ESI technique to introduce the sample. Elemental analyses were done on elemental analyzer model 1108 EA. Thin-layer chromatography (TLC) was performed on silica gel HF<sub>254</sub> with detection by charring with 30% (v/v) H<sub>2</sub>SO<sub>4</sub> in MeOH or in some cases by a UV detector. Column chromatography was conducted by elution of a column (10×240 mm, 18×300 mm, 35×400 mm) of silica gel (100–200 mesh) with EtOAc-petroleum ether (60–90 °C) as the eluent. Solutions were concentrated at <60 °C under diminished pressure. Dry solvents were distilled over CaH<sub>2</sub> and stored over molecular sieves.

**Allyl 2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-D-mannopyranoside (3).** To a cooled solution (0 °C) of **1** (5.32 g, 10 mmol) and **2** (6.67 g, 1.1 mmol) in anhydrous CH<sub>2</sub>Cl<sub>2</sub> (50 mL) was added TMSOTf (18 μL, 0.01 mmol). The mixture was stirred at this temperature for 2 h, and then quenched with Et<sub>3</sub>N (two drops). The solvents were evaporated in vacuo to give a residue, which was purified by silica gel column chromatography (3:1 petroleum ether–EtOAc) to give disaccharide **3** (8.41 g, 86.2%) as a foamy solid. [α]<sub>D</sub> –23.7° (*c* 1.3, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.16–7.32 (m, 30H, 6*Ph*), 5.92 (m, 1H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 5.91 (dd, 1H, *J*<sub>3,4</sub>=*J*<sub>4,5</sub>=10.2 Hz, H-4), 5.65 (dd, 1H, *J*<sub>2,3</sub>=3.3, *J*<sub>3,4</sub>=10.2 Hz, H-3), 5.64 (dd, 1H, *J*<sub>2,3</sub>=*J*<sub>3,4</sub>=4.9 Hz, H-3'), 5.36 (dd, 1H, *J*<sub>1,2</sub>=3.6, *J*<sub>2,3</sub>=4.9 Hz, H-2'), 5.30–5.18 (m, 2H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 5.14 (m, 1H, H-4'), 4.99 (d, 1H, *J*<sub>1,2</sub>=1.6 Hz, H-1), 4.96 (d, 1H, *J*<sub>1,2</sub>=3.6 Hz, H-1'), 4.49 (dd, 1H, *J*<sub>1,2</sub>=1.6, *J*<sub>2,3</sub>=3.3 Hz, H-2), 4.41–4.36 (m, 2H, H-5'), 4.31 (ddd, 1H, *J*<sub>4,5</sub>=10.2, *J*<sub>5,6a</sub>=6.3, *J*<sub>5,6b</sub>=4.6 Hz, H-5), 4.22 (m, 1H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 4.08 (dd, 1H, *J*<sub>5,6a</sub>=6.3, *J*<sub>6a,6b</sub>=12.0 Hz, H-6a), 4.02 (m, 1H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 3.37 (dd, 1H, *J*<sub>5,6b</sub>=4.6, *J*<sub>6a,6b</sub>=12.0 Hz, H-6b); <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>) δ 166.1, 166.0, 165.5, 165.4, 165.3, 165.1 (6C, 6*PhCO*), 133.7–128.1 (*PhCO*, CH<sub>2</sub>=CHCH<sub>2</sub>O), 118.3 (CH<sub>2</sub>=CHCH<sub>2</sub>O), 99.1, 97.1 (2C, 2C-1), 76.3, 71.6, 69.5, 69.0, 68.8, 68.6, 68.0, 67.5, 63.9, 60.0 (10C, C-2–6, C-2'–5', CH<sub>2</sub>=CHCH<sub>2</sub>O). Anal. calcd for C<sub>56</sub>H<sub>48</sub>O<sub>16</sub>: C 68.84; H 4.95. Found: C 68.58; H 5.15.

**2,3,4-Tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl trichloroacetimidate (4).**

To a solution of **3** (4.88 g, 5 mmol) in 90% acetic acid (50 mL) containing sodium acetate (1.46 g, 15 mmol) was added PdCl<sub>2</sub> (270 mg, 2.5 mmol), and the mixture was stirred for 12 h, at the end of which time TLC (2:1 petroleum ether–EtOAc) indicated that the reaction was complete. The mixture was diluted with dichloromethane (150 mL), washed with water and satd aq sodium bicarbonate. The organic layer was concentrated, and the residue was passed through a short silica gel column with 2:1 petroleum ether–EtOAc as the eluent to give crude 2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-D-mannopyranose as a syrup. Dried under high vacuum for 2 h, the solid was dissolved in dichloromethane (30 mL), and CCl<sub>3</sub>CN (1.0 mL, 10 mmol) and DBU (135 μL, 0.9 mmol) were added. The reaction mixture was stirred for 2 h, at the end of which time TLC (2:1 petroleum ether–EtOAc) indicated that the reaction was complete. Concentration of the reaction mixture, followed by purification of the crude product on a silica gel column with 3:1 petroleum ether–EtOAc as the eluent, furnished the disaccharide donor **4** (4.91 g, 90.9%) as a foamy solid. [α]<sub>D</sub> –17.5° (*c* 1.0, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.73 (s, 1H, CNHCCl<sub>3</sub>), 8.19–7.14 (m, 30H, 5*Ph*), 6.43 (s, 1H, H-1), 6.04 (dd, 1H, *J*<sub>3,4</sub>=*J*<sub>4,5</sub>=10.1 Hz, H-4), 5.69 (dd, 1H, *J*<sub>2,3</sub>=3.2, *J*<sub>3,4</sub>=10.1 Hz, H-3), 5.67 (dd, 1H, *J*<sub>2,3</sub>=*J*<sub>3,4</sub>=5.1 Hz, H-3'), 5.39 (dd, 1H, *J*<sub>1,2</sub>=3.8, *J*<sub>2,3</sub>=5.1 Hz, H-2'), 5.12 (m, 1H, H-4'), 5.09 (d, 1H, *J*<sub>1,2</sub>=3.8 Hz, H-1'), 4.75 (dd, 1H, *J*<sub>1,2</sub>=0.6, *J*<sub>2,3</sub>=3.2 Hz, H-2), 4.52 (ddd, 1H, *J*<sub>4,5</sub>=10.2, *J*<sub>5,6a</sub>=6.3, *J*<sub>5,6b</sub>=4.6 Hz, H-5), 4.45 (dd, 1H, *J*<sub>4',5'a</sub>=2.8, *J*<sub>5'a,5'b</sub>=12.1 Hz, H-5'a), 4.33 (dd, 1H, *J*<sub>4',5'b</sub>=3.2, *J*<sub>5'a,5'b</sub>=12.1 Hz, H-5'b), 4.14 (dd, 1H, *J*<sub>5,6a</sub>=5.8, *J*<sub>6a,6b</sub>=12.1 Hz, H-6a), 3.43 (dd, 1H, *J*<sub>5,6b</sub>=5.0, *J*<sub>6a,6b</sub>=12.1 Hz, H-6b). Anal. calcd for C<sub>55</sub>H<sub>44</sub>Cl<sub>3</sub>NO<sub>16</sub>: C 61.09; H 4.10. Found: C 61.18; H 4.33.

**Allyl 2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl-(1→3)-4,6-*O*-benzylidene-α-D-mannopyranoside (6).** To a cooled solution (–20 °C) of **5** (1.54 g, 5 mmol) and **4** (5.94 g, 5.5 mmol) in anhydrous CH<sub>2</sub>Cl<sub>2</sub> (50 mL) was added TMSOTf (18 μL, 0.05 mmol). The mixture was stirred at this temperature for 2 h, and then quenched with Et<sub>3</sub>N (two drops). The solvents were evaporated in vacuo to give a residue, which was purified by silica gel column chromatography (2:1 petroleum ether–EtOAc) to give trisaccharide **6** as a syrup (4.92 g, 80.3%). [α]<sub>D</sub> –70.4° (*c* 0.5, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 7.98–7.25 (m, 35H, 7*Ph*), 5.87 (m, 1H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 5.81 (dd, 1H, *J*<sub>3,4</sub>=*J*<sub>4,5</sub>=9.9 Hz, H-4'), 5.64 (dd, 1H, *J*<sub>2,3</sub>=3.2, *J*<sub>3,4</sub>=9.9 Hz, H-3'), 5.62 (s, 1H, PhCHO<sub>2</sub>), 5.45 (dd, 1H, *J*<sub>2,3</sub>=*J*<sub>3,4</sub>=6.9 Hz, H-3''), 5.32 (dd, 1H, *J*<sub>1,2</sub>=5.2, *J*<sub>2,3</sub>=6.9 Hz, H-2''), 5.30 (d, 1H, *J*<sub>1,2</sub>=1.2 Hz, H-1'), 5.30–5.20 (m, 2H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 5.06 (m, 1H, H-4''), 4.87 (d, 1H, *J*<sub>1,2</sub>=1.1 Hz, H-1), 4.47 (dd, 1H, *J*<sub>1,2</sub>=1.2, *J*<sub>2,3</sub>=3.2 Hz, H-2'), 4.41 (d, 1H, *J*<sub>1,2</sub>=5.2 Hz, H-1''), 4.36–4.28 (m, 3H), 4.18–4.09 (m, 5H), 4.01–3.85 (m, 4H), 2.91 (dd, 1H, *J*<sub>5,6b</sub>=6.9, *J*<sub>6a,6b</sub>=12.1 Hz, H-6b); <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>) δ 166.3, 166.2, 165.5, 165.4, 165.3, 164.8 (6C, 6*PhCO*), 133.7–126.2 (*PhCO*, CH<sub>2</sub>=CHCH<sub>2</sub>O), 118.1 (CH<sub>2</sub>=CHCH<sub>2</sub>O), 102.3 (PhCHO<sub>2</sub>), 99.4, 99.1, 98.8 (3C, 3C-1), 78.8, 75.5, 74.7,

71.4, 70.8, 70.3, 70.2, 69.2, 69.0, 69.0, 68.2, 67.3, 63.7, 63.6, 60.2 (15C, C-2-6, C-2'-6', C-2''-5'', CH<sub>2</sub>=CHCH<sub>2</sub>O). Anal. calcd for C<sub>69</sub>H<sub>62</sub>O<sub>21</sub>: C 67.53; H 5.09. Found: C 67.70; H 5.00.

**Allyl 2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl-(1→3)-2-*O*-acetyl-4,6-*O*-benzylidene-α-D-mannopyranoside (7).** To a solution of **6** (123 mg, 0.1 mmol) in pyridine (5 mL) was added acetic anhydride (2.0 mL, 2 mmol). The reaction mixture was stirred at rt for 12 h, at the end of which time TLC (3:1 petroleum ether–EtOAc) indicated that the reaction was complete. The reaction mixture was concentrated, and then the residue was purified by flash column chromatography on a silica gel column (3:1 petroleum ether–EtOAc) to give compound **7** (115 mg, 90.6%) as a foamy solid. [α]<sub>D</sub> –70.8° (*c* 0.5, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.13–7.24 (m, 35H, 7*Ph*), 5.87 (dd, 1H, *J*<sub>3,4</sub>=*J*<sub>4,5</sub>=9.8 Hz, H-4'), 5.82 (m, 1H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 5.64 (s, 1H, PhCHO<sub>2</sub>), 5.50 (dd, 1H, *J*<sub>2,3</sub>=3.3, *J*<sub>3,4</sub>=9.8 Hz, H-3'), 5.46 (dd, 1H, *J*<sub>2,3</sub>=*J*<sub>3,4</sub>=6.3 Hz, H-3''), 5.40 (dd, 1H, *J*<sub>1,2</sub>=0.6, *J*<sub>2,3</sub>=3.0 Hz, H-2), 5.28 (d, 1H, *J*<sub>1,2</sub>=1.4 Hz, H-1'), 5.26 (dd, 1H, *J*<sub>1,2</sub>=4.6, *J*<sub>2,3</sub>=6.3 Hz, H-2''), 5.31–5.21 (m, 2H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 5.01 (m, 1H, H-4''), 4.82 (d, 1H, *J*<sub>1,2</sub>=0.6 Hz, H-1), 4.47 (d, 1H, *J*<sub>1,2</sub>=4.6 Hz, H-1''), 4.43 (dd, 1H, *J*<sub>1,2</sub>=1.4, *J*<sub>2,3</sub>=3.3 Hz, H-2'), 4.37–4.29 (m, 3H), 4.18–4.11 (m, 2H), 4.09 (dd, 1H, *J*<sub>3,4</sub>=*J*<sub>4,5</sub>=9.4 Hz, H-4), 4.03–3.84 (m, 4H), 2.99 (dd, 1H, *J*<sub>5,6b</sub>=6.2, *J*<sub>6a,6b</sub>=12.0 Hz, H-6b). Anal. calcd for C<sub>71</sub>H<sub>64</sub>O<sub>22</sub>: C 67.18; H 5.08. Found: C 67.37; H 5.25.

**Allyl 2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl-(1→3)-[2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)]-4,6-*O*-benzylidene-α-D-mannopyranoside (8).** To a cooled solution (–10 °C) of **6** (3.68 g, 3.0 mmol) and **2** (2.00 g, 3.3 mmol) in anhydrous CH<sub>2</sub>Cl<sub>2</sub> (50 mL) was added TMSOTf (54 μL, 0.3 mmol). After stirring at this temperature for 30 min, further TMSOTf (360 μL, 2.0 mmol) and 2.2 equiv of donor **2** (4.00 g, 6.6 mmol) was added to the reaction mixture subsequently. The mixture was stirred for 2 h, and then quenched with Et<sub>3</sub>N (four drops). The solvents were evaporated in vacuo to give a residue, which was purified by silica gel column chromatography (2:1 petroleum ether–EtOAc) to give tetrasaccharide **8** (3.21 g, 64.2%) as a foamy solid. [α]<sub>D</sub> –67.5° (*c* 1.3, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.29–7.19 (m, 50H, 10*Ph*), 5.92 (dd, 1H, *J*<sub>2,3</sub>=3.2, *J*<sub>3,4</sub>=9.8 Hz, H-3, Man*p*), 5.87 (dd, 1H, *J*<sub>3,4</sub>=*J*<sub>4,5</sub>=9.8 Hz, H-4, Man*p*), 5.78 (m, 1H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 5.66 (dd, 1H, *J*<sub>1,2</sub>=*J*<sub>2,3</sub>=4.2 Hz, H-2, Xyl*p*), 5.53 (s, 1H, PhCHO<sub>2</sub>), 5.52 (dd, 1H, *J*<sub>2,3</sub>=5.6, *J*<sub>3,4</sub>=6.0 Hz, H-3, Xyl*p*), 5.40–5.38 (m, 2H, H-2 and H-4, Xyl*p*), 5.38 (d, 1H, *J*<sub>1,2</sub>=1.3, H-1, Man*p*), 5.33 (dd, 1H, *J*<sub>2,3</sub>=4.4, *J*<sub>3,4</sub>=6.0 Hz, H-3, Xyl*p*), 5.24–5.12 (m, 3H), 5.08 (m, 1H, H-4, Xyl*p*), 5.04 (d, 1H, *J*<sub>1,2</sub>=1.7 Hz, H-1, Man*p*), 5.00 (d, 1H, *J*<sub>1,2</sub>=4.2 Hz, H-1, Xyl*p*), 4.83 (d, 1H, *J*<sub>1,2</sub>=0.7 Hz, H-1, Man*p*), 4.60 (d, 1H, *J*<sub>1,2</sub>=4.4 Hz, H-1, Xyl*p*), 4.52–4.40 (m, 3H), 4.28 (dd, 1H, *J*<sub>2,3</sub>=3.3, *J*<sub>3,4</sub>=9.8 Hz, H-3, Man*p*), 4.20–4.05 (m, 6H), 3.92–3.86 (m, 2H), 3.76 (m, 1H), 3.43 (dd, 1H, *J*<sub>3,4</sub>=*J*<sub>4,5</sub>=9.8 Hz, H-4, Man*p*), 3.02 (dd, 1H, *J*=6.3, 12.0 Hz, H-6b, Man*p*); <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>) δ

166.2, 165.8, 165.5, 165.5, 165.4, 165.4, 165.3, 164.9, 164.8 (9C, 9PhCO), 118.1 (CH<sub>2</sub>=CHCH<sub>2</sub>O), 101.7 (PhCHO<sub>2</sub>), 98.7, 98.1, 97.1, 97.0 (4C, 4C-1), 78.7, 76.6, 74.2, 74.1, 69.9, 69.9, 69.6, 69.5, 68.7, 68.6, 68.4, 68.4, 67.9, 67.7, 67.6, 64.1, 63.9, 60.0, 59.9. Anal. calcd for C<sub>95</sub>H<sub>82</sub>O<sub>28</sub>: C 68.25; H 4.94. Found: C 68.16; H 5.10.

**Allyl 2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl-(1→3)-[2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)]-4,6-di-*O*-benzoyl-α-D-mannopyranoside (9).** Compound **8** (3.00 g, 1.8 mmol) was dissolved in 50 mL 90% TFA and stirred for 2 h, at the end of which time the reaction mixture was poured into 200 mL toluene and then the mixture was concentrated to give a yellow syrup. Dried under high vacuum for 2 h, the solid was dissolved in pyridine (20 mL), and benzoyl chloride (3.5 mL, 30 mmol) was added dropwise in 30 min. The mixture was stirred overnight at room temperature, and TLC (2:1 petroleum ether–EtOAc) indicated that the reaction was complete. Ice water was added, and the mixture was diluted with dichloromethane, washed with M HCl, water, and satd aq sodium bicarbonate subsequently. The organic layer was combined, dried, and concentrated. Purification by column chromatography (2:1 petroleum ether–EtOAc) gave **9** (2.96 g, 91.9% for two steps) as a foamy solid. [α]<sub>D</sub> –92.9° (*c* 1.2, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.16–7.31 (m, 55H, 11*Ph*), 5.85 (m, 1H, CH<sub>2</sub>=CHCH<sub>2</sub>O), 5.84 (dd, 1H, *J*<sub>3,4</sub>=*J*<sub>4,5</sub>=9.9 Hz, H-4, Man*p*), 5.76–5.62 (m, 3H), 5.49 (m, 1H, H-4, Xyl*p*), 5.45 (dd, 1H, *J*<sub>1,2</sub>=*J*<sub>2,3</sub>=5.7 Hz, H-2, Xyl*p*), 5.35–5.19 (m, 3H), 5.14 (d, 1H, *J*<sub>1,2</sub>=0.7 Hz, H-1, Man*p*), 5.12 (d, 1H, *J*<sub>1,2</sub>=4.2 Hz, H-1, Xyl*p*), 5.06 (m, 1H, H-4, Xyl*p*), 5.01 (dd, 1H, *J*<sub>2,3</sub>=5.4, *J*<sub>3,4</sub>=6.0 Hz, H-3, Xyl*p*), 4.87 (d, 1H, *J*<sub>1,2</sub>=0.9 Hz, H-1, Man*p*), 4.43 (d, 1H, *J*<sub>1,2</sub>=5.1 Hz, H-1, Xyl*p*), 4.36–3.86 (m, 13H), 2.92 (dd, 1H, *J*=6.1, 12.2 Hz, H-6b, Man*p*), 2.81 (dd, 1H, *J*=6.3, 12.1 Hz, H-6b, Man*p*); <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>) 166.2, 166.1, 165.6, 165.6, 165.5, 165.4, 165.3, 165.3, 165.1, 164.8, 164.7 (11C, 11PhCO), 118.3 (CH<sub>2</sub>=CHCH<sub>2</sub>O), 100.1, 99.4, 99.0, 96.8 (4C, 4C-1), 78.2, 76.0, 75.9, 70.3, 69.8, 69.7, 69.3, 68.8, 68.7, 68.6, 68.1, 67.6, 64.0, 63.7, 60.6, 60.5 (some signals overlapped). Anal. calcd for C<sub>102</sub>H<sub>86</sub>O<sub>30</sub>: C 68.37; H 4.84. Found: C 68.52; H 4.99.

**2,3,4-Tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl-(1→3)-[2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)]-4,6-di-*O*-benzoyl-α-D-mannopyranosyl trichloroacetimidate (10).** To a solution of **9** (2.69 g, 1.5 mmol) in 90% acetic acid (15 mL) containing sodium acetate (0.44 g, 4.5 mmol) was added PdCl<sub>2</sub> (81 mg, 0.75 mmol), and the mixture was stirred for 12 h, at the end of which time TLC (2:1 petroleum ether–EtOAc) indicated that the reaction was complete. The mixture was diluted with dichloromethane (150 mL), washed with water and satd aq sodium bicarbonate. The organic layer was concentrated, and the residue was passed through a short silica gel column with 1:1 petroleum ether–EtOAc as the eluent to give crude 2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl-(1→3)-[2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)]-4,6-di-*O*-benzoyl-α-D-manno-

pyranose as a syrup. Dried under high vacuum for 2 h, the solid was dissolved in dichloromethane (10 mL), and  $\text{CCl}_3\text{CN}$  (0.5 mL, 5 mmol) and DBU (40  $\mu\text{L}$ , 0.3 mmol) were added. The reaction mixture was stirred for 2 h, at the end of which time TLC (2:1 petroleum ether–EtOAc) indicated that the reaction was complete. Concentration of the reaction mixture, followed by purification of the crude product on a silica gel column with 2:1 petroleum ether–EtOAc as the eluent, furnished the tetrasaccharide donor **10** (2.44 g, 85.8%) as a foamy solid.  $[\alpha]_{\text{D}} -71.2^\circ$  (*c* 1.3,  $\text{CHCl}_3$ );  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  8.70 (s, 1H,  $\text{CNHCCl}_3$ ), 8.13–7.31 (m, 55H, 11*Ph*), 6.40 (s, 1H, H-1, Manp), 5.82 (dd, 1H,  $J_{3,4}=J_{4,5}=9.7$  Hz, H-4, Manp), 5.78 (dd, 1H,  $J_{2,3}=3.2$ ,  $J_{3,4}=9.8$  Hz, H-3, Manp), 5.74 (dd, 1H,  $J_{1,2}=J_{2,3}=4.1$  Hz, H-2, Xylp), 5.72 (dd, 1H,  $J_{3,4}=J_{4,5}=9.8$  Hz, H-4, Manp), 5.51 (m, 1H, H-4, Xylp), 5.45 (dd, 1H,  $J_{1,2}=J_{2,3}=4.0$  Hz, H-2, Xylp), 5.36 (dd, 1H,  $J_{2,3}=4.0$ ,  $J_{3,4}=6.5$  Hz, H-3, Xylp), 5.16 (d, 1H,  $J_{1,2}=4.1$  Hz, H-1, Xylp), 5.06 (d, 1H,  $J_{1,2}=1.7$  Hz, H-1, Manp), 5.04 (m, 1H, H-4, Xylp), 4.58 (d, 1H,  $J_{1,2}=4.0$  Hz, H-1, Xylp). Anal. calcd for  $\text{C}_{101}\text{H}_{82}\text{Cl}_3\text{NO}_{30}$ : C 63.98; H 4.36. Found: C 63.85; H 4.51.

**Methyl 2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)-3,4,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)-[4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-2-*O*-acetyl- $\alpha$ -D-mannopyranoside (13).** To a cooled solution ( $-20^\circ\text{C}$ ) of **10** (1.90 g, 1.0 mmol) and **11** (0.42 g, 1.5 mmol) in anhydrous  $\text{CH}_2\text{Cl}_2$  (50 mL) was added TMSOTf (18  $\mu\text{L}$ , 0.1 mmol). The mixture was stirred at this temperature for 2 h, and then quenched with  $\text{Et}_3\text{N}$  (two drops). The solvents were evaporated in vacuo to afford the crude pentasaccharide **12**. Compound **12** was dissolved in pyridine (20 mL), and  $\text{Ac}_2\text{O}$  (10 mL) was added. The mixture was stirred at rt for 12 h, and then was evaporated and coevaporated with toluene in vacuo to give a residue. The residue was dissolved in 50 mL 90% TFA and stirred for 2 h, at the end of which time the reaction mixture was poured into 200 mL toluene and then the mixture was concentrated. The residue was purified by silica gel column chromatography (1:1 petroleum ether–EtOAc) to give **13** (1.07 g, 54.3% for three steps) as a syrup.  $[\alpha]_{\text{D}} -41.3^\circ$  (*c* 1.3,  $\text{CHCl}_3$ );  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  8.07–6.86 (m, 55H, 11*Ph*), 5.76 (dd, 1H,  $J_{3,4}=J_{4,5}=9.8$  Hz, H-4, Manp), 5.74 (dd, 1H,  $J_{2,3}=3.1$ ,  $J_{3,4}=9.8$  Hz, H-3, Manp), 5.66 (dd, 1H,  $J_{3,4}=J_{4,5}=9.8$  Hz, H-4, Manp), 5.58 (dd, 1H,  $J_{1,2}=J_{2,3}=4.6$  Hz, H-2, Xylp), 5.39–5.36 (m, 2H), 5.31 (d, 1H,  $J_{1,2}=1.0$  Hz, H-1, Manp), 5.29 (m, 1H, H-4, Xylp), 5.24 (dd, 1H,  $J_{1,2}=1.4$ ,  $J_{2,3}=3.2$  Hz, H-2, Manp), 5.20 (dd, 1H,  $J_{1,2}=J_{2,3}=5.5$  Hz, H-2, Xylp), 5.06 (m, 1H, H-4, Xylp), 5.02 (d, 1H,  $J_{1,2}=0.6$  Hz, H-1, Manp), 4.93 (d, 1H,  $J_{1,2}=0.8$  Hz, H-1, Manp), 4.63 (d, 1H,  $J_{1,2}=4.4$  Hz, H-1, Xylp), 4.57 (d, 1H,  $J_{1,2}=5.5$  Hz, H-1, Xylp), 4.42–4.34 (m, 4H), 4.29–4.27 (m, 2H), 4.17 (dd, 1H,  $J_{2,3}=3.2$ ,  $J_{3,4}=9.9$  Hz, H-3, Manp), 4.10–4.05 (m, 3H), 4.00–3.93 (m, 2H), 3.87–3.81 (m, 3H), 3.69–3.57 (m, 2H), 3.29 (s, 3H,  $\text{OCH}_3$ ), 2.89 (dd, 1H,  $J=6.2$  Hz, 12.2, H-6b, Manp), 2.04 (s, 3H,  $\text{CH}_3\text{CO}$ );  $^{13}\text{C}$  NMR (100 MHz,  $\text{CDCl}_3$ )  $\delta$  170.1 ( $\text{CH}_3\text{CO}$ ), 167.3, 166.0, 165.5, 165.3, 165.2, 165.1, 165.1, 164.9, 164.8, 164.6,

164.5 (11C, 11*PhCO*), 99.8, 99.5, 98.7, 98.4, 98.2 (5C, 5C-1), 75.6, 71.9, 71.3, 70.0, 69.7, 69.4, 69.3, 68.5, 68.2, 67.8, 67.5, 64.7, 63.3, 61.6, 60.2, 59.9 (C-2–6, Manp; C-2–5, Xylp, some signals overlapped), 54.4 ( $\text{OCH}_3$ ), 24.4 ( $\text{CH}_3\text{CO}$ ). Anal. calcd for  $\text{C}_{108}\text{H}_{96}\text{O}_{36}$ : C 65.85; H 4.91. Found: C 65.71; H 5.13.

**Methyl 2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)-3,4,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-*O*-benzoyl- $\alpha$ -D-xylopyranosyl-(1 $\rightarrow$ 2)-[4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-2-*O*-acetyl-4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranoside (14).** To a solution of **13** (1.00 g, 0.51 mmol) in pyridine (10 mL) was added benzoyl chloride (1.2 mL, 10 mmol) and DMAP (12 mg, 0.1 mmol). The reaction mixture was stirred at  $70^\circ\text{C}$  for 12 h, at the end of which time TLC (1:1 petroleum ether–EtOAc) indicated that the reaction was complete, and then quenched with MeOH (2.0 mL). The reaction mixture was concentrated, and purification of the residue by column chromatography on a silica gel column (2:1 petroleum ether–EtOAc) gave compound **14** (877 mg, 79.3%) as a syrup.  $[\alpha]_{\text{D}} -48.8^\circ$  (*c* 0.5,  $\text{CHCl}_3$ );  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  8.02–7.16 (m, 65H, 13*Ph*), 5.90 (dd, 1H,  $J_{3,4}=J_{4,5}=9.9$  Hz, H-4, Manp), 5.71 (dd, 1H,  $J_{2,3}=3.2$ ,  $J_{3,4}=9.8$  Hz, H-3, Manp), 5.69 (dd, 1H,  $J_{3,4}=J_{4,5}=9.8$  Hz, H-4, Manp), 5.64 (dd, 1H,  $J_{3,4}=J_{4,5}=9.6$  Hz, H-4, Manp), 5.38–5.34 (m, 3H), 5.32 (dd, 1H,  $J_{1,2}=1.0$ ,  $J_{2,3}=3.1$  Hz, H-2, Manp), 5.23 (dd, 1H,  $J_{1,2}=J_{2,3}=5.1$  Hz, H-2, Xylp), 5.18 (dd, 1H,  $J_{2,3}=5.1$ ,  $J_{3,4}=6.3$  Hz, H-3, Xylp), 5.06 (d, 1H,  $J_{1,2}=0.7$  Hz, H-1, Manp), 5.01 (d, 1H,  $J_{1,2}=0.8$  Hz, H-1, Manp), 4.98 (m, 1H, H-4, Xylp), 4.70 (d, 1H,  $J_{1,2}=1.0$  Hz, H-1, Manp), 4.55 (d, 1H,  $J_{1,2}=4.9$  Hz, H-1, Xylp), 4.50 (d, 1H,  $J_{1,2}=5.1$  Hz, H-1, Xylp), 4.40–4.28 (m, 10H), 4.07–3.84 (m, 6H), 3.34 (s, 3H,  $\text{OCH}_3$ ), 2.90 (dd, 1H,  $J=6.0$ , 11.9 Hz, H-6, Manp), 2.04 (s, 3H,  $\text{CH}_3\text{CO}$ );  $^{13}\text{C}$  NMR (100 MHz,  $\text{CDCl}_3$ )  $\delta$  170.3 ( $\text{CH}_3\text{CO}$ ), 166.0, 165.9, 165.8, 165.4, 165.3, 165.3, 165.1, 165.1, 164.9, 164.8, 164.6, 164.5, 164.4 (13C, 13*PhCO*), 99.8, 98.6, 98.6, 98.4, 98.4 (5C, 5C-1), 76.4, 74.7, 74.2, 73.6, 70.9, 69.7, 69.6, 69.3, 69.1, 68.8, 68.4, 68.3, 68.2, 67.8, 63.6, 63.3, 63.2, 60.4, 60.3, 60.3 (C-2–6, Manp; C-2–5, Xylp, some signals overlapped), 55.1 ( $\text{OCH}_3$ ), 22.6 ( $\text{CH}_3\text{CO}$ ). Anal. calcd for  $\text{C}_{122}\text{H}_{104}\text{O}_{38}$ : C 67.27; H 4.81. Found: C 67.08; H 4.77.

**Methyl 2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)-3,4,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)-[4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranoside (15).** To a solution of **14** (762 mg, 0.35 mmol) in anhydrous  $\text{CH}_2\text{Cl}_2$  (10 mL) was added anhyd MeOH (50 mL), then acetyl chloride (1.5 mL) was added to the reaction mixture at  $0^\circ\text{C}$ . The solution was stoppered in a flask and stirred at room temperature until TLC (3:1 petroleum ether–EtOAc) showed that the starting material disappeared. The solution was neutralized with  $\text{Et}_3\text{N}$ , then concentrated to dryness. The residue was passed through a short silica gel column to give **15** (664 mg, 88.9%) as a foamy solid.  $[\alpha]_{\text{D}} -27.4^\circ$  (*c* 1.3,  $\text{CHCl}_3$ );  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  8.12–6.99 (m, 65H, 13*Ph*), 5.83 (dd, 1H,  $J_{3,4}=J_{4,5}=10.0$  Hz, H-4, Manp), 5.72 (dd, 1H,  $J_{3,4}=J_{4,5}=9.5$  Hz, H-4,



Manp), 5.65 (dd, 1H,  $J_{2,3}=3.3$ ,  $J_{3,4}=10.0$  Hz, H-3, Manp), 5.57 (dd, 1H,  $J_{3,4}=J_{4,5}=9.8$  Hz, H-4, Manp), 5.51 (dd, 1H,  $J_{1,2}=J_{2,3}=5.1$  Hz, H-2, Xylp), 5.41–5.35 (m, 4H), 5.29 (dd, 1H,  $J_{1,2}=J_{2,3}=4.5$  Hz, H-2, Xylp), 5.20 (dd, 1H,  $J_{2,3}=5.1$ ,  $J_{3,4}=5.3$  Hz, H-3, Xylp), 5.07 (d, 1H,  $J_{1,2}=1.1$  Hz, H-1, Manp), 5.03 (d, 1H,  $J_{1,2}=0.6$  Hz, H-1, Manp), 5.02 (d, 1H,  $J_{1,2}=4.5$  Hz, H-1, Xylp), 5.01 (m, 1H, H-4, Xylp), 4.58 (d, 1H,  $J_{1,2}=0.9$ , H-1, Manp), 4.54 (dd, 1H,  $J_{2,3}=2.9$ ,  $J_{3,4}=9.9$  Hz, H-3, Manp), 4.49 (d, 1H,  $J_{1,2}=5.1$  Hz, H-1, Xylp), 4.42–4.26 (m, 9H), 4.11–3.86 (m, 6H), 3.36 (s, 3H, OCH<sub>3</sub>); <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>) δ 166.1, 166.1, 165.7, 165.4, 165.4, 165.2, 165.1, 165.1, 165.0, 164.9, 164.8, 164.7, 164.7 (13C, 13PhCO), 100.7, 99.9, 99.5, 98.8, 98.7 (5C, 5C-1), 77.7, 75.0, 74.9, 70.2, 69.9, 69.7, 69.6, 69.5, 68.6, 68.3, 68.2, 68.1, 68.0, 67.9, 64.2, 63.4, 63.3, 60.2, 60.1 (C-2–6, Manp; C-2–5, Xylp, some signals overlapped), 55.0 (OCH<sub>3</sub>). Anal. calcd for C<sub>120</sub>H<sub>102</sub>O<sub>37</sub>: C 67.47; H 4.81. Found: C 67.72; H 5.01.

**Methyl 2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-mannopyranosyl-(1→3)-[2,3,4-tri-*O*-benzoyl-β-D-xylopyranosyl-(1→2)-4,6-di-*O*-benzoyl-α-D-mannopyranosyl-(1→3)]-methyl 2,3,4-tri-*O*-acetyl-β-D-glucopyranosyluronate-(1→2)-[4,6-di-*O*-benzoyl-β-D-mannopyranoside (17).** To a cooled solution (0 °C) of **15** (427 mg, 0.2 mmol) and methyl 2,3,4-tri-*O*-acetyl-α-D-glucopyranosyluronate trichloroacetimidate **16** (144 mg, 0.3 mmol) in anhydrous CH<sub>2</sub>Cl<sub>2</sub> (10 mL) was added TMSOTf (8 μL, 0.05 mmol). The mixture was stirred at this temperature for 2 h, and then quenched with Et<sub>3</sub>N (one drop). The solvents were evaporated in vacuo to give a residue, which was purified by silica gel column chromatography (1:1 petroleum ether–EtOAc) to give **17** (356 mg, 72.7%) as a foamy solid.  $[\alpha]_D^{25} -61.7^\circ$  (c 1.3, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.20–6.90 (m, 65H, 13 PhH), 6.55 (dd, 1H,  $J_{1,2}=J_{2,3}=7.7$  Hz, H-2, Xylp), 6.09 (dd, 1H,  $J_{3,4}=J_{4,5}=9.6$  Hz, H-4, Manp), 6.01 (dd, 1H,  $J_{3,4}=J_{4,5}=8.8$  Hz, H-4, Manp), 5.78 (dd, 1H,  $J_{3,4}=J_{4,5}=9.9$  Hz, H-4, Manp), 5.28 (d, 1H,  $J_{1,2}=1.1$  Hz, H-1, Manp), 5.18 (d, 1H,  $J_{1,2}=0.8$  Hz, H-1, Manp), 5.12 (d, 1H,  $J_{1,2}=1.1$  Hz, H-1, Manp), 4.81 (d, 1H,  $J_{1,2}=7.9$  Hz, H-1, Xylp), 4.77 (d, 1H,  $J_{1,2}=7.7$  Hz, H-1, Xylp), 4.39 (dd, 1H,  $J_{1,2}=1.1$ ,  $J_{2,3}=3.2$  Hz, H-2, Manp), 4.17 (dd, 1H,  $J_{1,2}=0.8$ ,  $J_{2,3}=2.8$  Hz, H-2, Manp), 4.07 (d, 1H,  $J_{1,2}=7.2$  Hz, H-1, GluAp), 3.79 (dd, 1H,  $J_{1,2}=1.1$ ,  $J_{2,3}=2.9$  Hz, H-2, Manp), 3.52 (s, 3H, COOCH<sub>3</sub>), 3.18 (s, 3H, OCH<sub>3</sub>), 2.15, 2.13, 2.01 (3s, 9H, 3COCH<sub>3</sub>). <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>): 171.3, 171.0, 169.7, 169.2 (4C, 3COCH<sub>3</sub>, COOMe), 167.3, 167.2, 166.1, 166.0, 165.8, 165.4, 165.3, 165.3, 165.3, 165.2, 165.2, 165.1, 164.6 (13C, 13COPh), 102.6, 100.6, 100.6, 99.6, 98.9, 98.2 (6C, 6C-1), 54.7 (OCH<sub>3</sub>), 52.7 (COOCH<sub>3</sub>), 208, 20.7, 20.5 (3C, 3COCH<sub>3</sub>). Anal. calcd for C<sub>133</sub>H<sub>118</sub>O<sub>46</sub>: C, 65.13; H, 4.85. Found: C, 65.32; H, 4.97.

**Methyl β-D-xylopyranosyl-(1→2)-α-D-mannopyranosyl-(1→3)-[β-D-xylopyranosyl-(1→2)-α-D-mannopyranosyl-(1→3)]-(β-D-glucopyranosyluronic acid)-(1→2)-[α-D-mannopyranoside, sodium salt (18).** Hexasaccharide **17** (245 mg, 0.10 mmol) was dissolved in anhydrous CH<sub>2</sub>Cl<sub>2</sub> (5 mL), then MeOH (30 mL) and 1 mL of

MeONa–MeOH (0.2 N) was added to the solvent subsequently. After 96 h at room temperature, water (18 μL, 1.0 mmol) was added to the mixture to cleave the methyl ester. After stirring at room temperature for 2 h, the reaction mixture was concentrated and purified on a Bio-Gel P2 column (eluent: water), affording the target hexasaccharide **18** (63 mg, 64.5%) as a foamy solid.  $[\alpha]_D^{25} +56.9^\circ$  (c 0.5, H<sub>2</sub>O); <sup>1</sup>H NMR (D<sub>2</sub>O, 400 MHz): δ 5.05 (s, 1H, H-1, Manp), 4.97 (s, 1H, H-1, Manp), 4.65 (s, 1H, H-1, Manp), 4.49 (d, 1H,  $J_{1,2}=8.0$  Hz, H-1, GluAp), 4.28 (d, 1H,  $J_{1,2}=8.8$  Hz, H-1, Xylp), 4.24 (d, 1H,  $J_{1,2}=8.4$  Hz, H-1, Xylp), 3.28 (s, 3H, OCH<sub>3</sub>); <sup>13</sup>C NMR (100 MHz, D<sub>2</sub>O): 173.9 (–COONa), 103.5 (C-1,  $J_{C1,H1}=163.4$  Hz, Xylp), 103.5 (C-1,  $J_{C1,H1}=163.4$  Hz, Xylp), 102.7 (C-1,  $J_{C1,H1}=176.2$  Hz, Manp), 100.6 (C-1,  $J_{C1,H1}=172.7$  Hz, Manp), 99.1 (C-1,  $J_{C1,H1}=174.1$  Hz, Manp), 96.0 (C-1,  $J_{C1,H1}=166.8$  Hz, GluAp), 78.4, 78.2, 76.2, 76.1, 76.0, 75.7, 74.1, 73.5, 73.4, 72.8, 72.2, 70.5, 70.2, 69.4, 66.9, 66.5, 66.2, 65.3, 65.3, 61.5, 60.4, 60.4 (C-2–6, Manp; C-2–5, Xylp; C-2–5, GluAp; some signals overlapped), 55.0 (O–CH<sub>3</sub>). Negative-ESI-MS calcd for C<sub>35</sub>H<sub>57</sub>O<sub>30</sub>Na: [M] 980.8, Found: [M–Na] 957.9.

**2,3-Di-*O*-acetyl-4,6-*O*-isopropylidene-α-D-mannopyranosyl trichloroacetimidate (19).** Compound 1,2,3-*O*-acetyl-4,6-*O*-isopropylidene-α-D-mannopyranoside (6.9 g, 20.0 mmol) was dissolved in 1 M solution of ammonia–methanol (100 mL) and stirred for 4 h, at the end of which time TLC (3:1 petroleum ether–EtOAc) indicated that the reaction was complete. The solution was concentrated and dried under high vacuum giving a white foamy solid. This foamy solid was dissolved in dry dichloromethane (50 mL), then trichloroacetonitrile (6.3 mL, 30 mmol) and 1,8-diazabicyclo[5.4.0]undecene (DBU) (0.50 mL, 4.04 mmol) was added subsequently. The reaction mixture was stirred under nitrogen for 3 h and then concentrated. The residue was purified by chromatography (4:1 petroleum ether–EtOAc) to give **19** (7.40 g, 82.2%) as a syrup.  $[\alpha]_D^{25} +42^\circ$  (c 1.0, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 8.72 (s, 1H, CNHCCl<sub>3</sub>), 6.17 (s, 1H,  $J_{1,2}=1.6$  Hz, H-1), 5.50 (dd, 1H,  $J_{1,2}=1.6$ ,  $J_{2,3}=3.4$  Hz, H-2), 5.30 (dd, 1H,  $J_{2,3}=3.4$ ,  $J_{3,4}=10.3$  Hz, H-3), 4.11 (dd, 1H,  $J_{3,4}=10.3$ ,  $J_{4,5}=10.0$  Hz, H-4), 3.98–3.85 (m, 3H), 2.20 (s, 3H, CH<sub>3</sub>CO), 2.04 (s, 3H, CH<sub>3</sub>CO), 1.55 (s, 3H, isopropylidene), 1.42 (s, 3H, isopropylidene). Anal. calcd for C<sub>15</sub>H<sub>20</sub>Cl<sub>3</sub>NO<sub>8</sub>: C 40.12; H 4.49. Found: C 40.40; H 4.73.

**Methyl 2,3-di-*O*-acetyl-4,6-*O*-isopropylidene-α-D-mannopyranosyl-(1→3)-4,6-*O*-isopropylidene-α-D-mannopyranoside (21).** To a cooled solution (–20 °C) of **20** (1.17 g, 5 mmol) and **19** (2.46 g, 5.5 mmol) in anhyd CH<sub>2</sub>Cl<sub>2</sub> (50 mL) was added TMSOTf (18 μL, 0.05 mmol). The mixture was stirred at this temperature for 2 h, and then quenched with Et<sub>3</sub>N (two drops). The solvents were evaporated in vacuo to give a residue, which was purified by silica gel column chromatography (2:1 petroleum ether–EtOAc) to give disaccharide **21** (1.93 g, 74.2%) as a syrup.  $[\alpha]_D^{25} +43.3^\circ$  (c 1.0, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 5.36 (dd, 1H,  $J_{1,2}=1.0$ ,  $J_{2,3}=3.2$  Hz, H-2'), 5.21 (dd, 1H,  $J_{2,3}=3.2$ ,  $J_{3,4}=9.9$  Hz, H-3'), 5.20 (s, 1H,  $J_{1,2}=1.0$  Hz, H-1'), 4.73 (s, 1H,



$J_{1,2}=1.1$  Hz, H-1), 4.14 (dd, 1H,  $J_{3,4}=J_{4,5}=9.9$  Hz, H-4), 4.06–4.02 (m, 2H), 3.95 (dd, 1H,  $J_{2,3}=3.3$ ,  $J_{3,4}=10.0$  Hz, H-3), 3.87–3.76 (m, 5H), 3.60 (m, 1H, H-5), 3.36 (s, 3H, OCH<sub>3</sub>), 2.17 (s, 3H, CH<sub>3</sub>CO), 2.15 (s, 3H, CH<sub>3</sub>CO), 1.52 (s, 6H, isopropylidene), 1.40 (s, 3H, isopropylidene), 1.29 (s, 3H, isopropylidene); <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>): 170.0, 169.6 (2C, 2COCH<sub>3</sub>), 101.4 (1C, Me<sub>2</sub>CO<sub>2</sub>), 100.1, 100.0 (2C, 2C-1), 99.5 (1C, Me<sub>2</sub>CO<sub>2</sub>), 74.1, 71.6, 71.1, 70.0, 68.9, 68.6, 65.4, 64.2, 62.4, 62.2 (10C, C2–6, C2'–6'), 54.9 (OCH<sub>3</sub>), 29.1, 29.0, 20.8, 20.7 [4C, 2(CH<sub>3</sub>)<sub>2</sub>CO<sub>2</sub>], 19.3, 19.1 (2C, 2COCH<sub>3</sub>). Anal. calcd for C<sub>23</sub>H<sub>36</sub>O<sub>13</sub>: C, 53.07; H, 6.97. Found: C, 53.32; H, 6.70.

**Methyl 2,3-di-*O*-acetyl-4,6-*O*-isopropylidene- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-2-*O*-acetyl-4,6-*O*-isopropylidene- $\alpha$ -D-mannopyranoside (22).** To a solution of 21 (104 mg, 0.2 mmol) in pyridine (5 mL) was added acetic anhydride (2.0 mL, 2 mmol). The reaction mixture was stirred at rt for 12 h, at the end of which time TLC (3:1 petroleum ether–EtOAc) indicated that the reaction was complete. The reaction mixture was concentrated, and then the residue was purified by flash column chromatography on a silica gel column (3:1 petroleum ether–EtOAc) to give compound 22 (100 mg, 89.3%) as a foamy solid.  $[\alpha]_D +38.9^\circ$  (*c* 1.5, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  5.37 (dd, 1H,  $J_{1,2}=1.2$ ,  $J_{2,3}=3.3$  Hz, H-2'), 5.34 (dd, 1H,  $J_{1,2}=1.5$ ,  $J_{2,3}=3.0$  Hz, H-2), 5.26 (dd, 1H,  $J_{2,3}=3.3$ ,  $J_{3,4}=9.8$  Hz, H-3'), 5.12 (s, 1H,  $J_{1,2}=1.2$  Hz, H-1'), 4.70 (s, 1H,  $J_{1,2}=1.5$  Hz, H-1), 4.20 (dd, 1H,  $J_{3,4}=J_{4,5}=9.9$  Hz, H-4), 4.11–3.87 (m, 8H), 3.66 (m, 1H, H-5), 3.34 (s, 3H, OCH<sub>3</sub>), 2.21 (s, 3H, CH<sub>3</sub>CO), 2.19 (s, 3H, CH<sub>3</sub>CO), 2.14 (s, 3H, CH<sub>3</sub>CO), 1.55 (s, 3H, isopropylidene), 1.53 (s, 3H, isopropylidene), 1.38 (s, 3H, isopropylidene), 1.31 (s, 3H, isopropylidene). Anal. calcd for C<sub>25</sub>H<sub>38</sub>O<sub>14</sub>: C, 53.37; H, 6.81. Found: C, 53.41; H, 6.62.

**Methyl 4,6-*O*-isopropylidene- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-4,6-*O*-isopropylidene- $\alpha$ -D-mannopyranoside (23).** Disaccharide 21 (2.60 mg, 5.0 mmol) was dissolved in a satd methanolic ammonia (25 mL). After 2 h at room temperature, the reaction mixture was concentrated, and the residue was purified by flash column chromatography on a silica gel column (EtOAc) to give compound 23 (2.10 g, 96.3%) as a foamy solid.  $[\alpha]_D +90.6^\circ$  (*c* 0.5, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  5.21 (s, 1H, H-1'), 4.69 (s, 1H, H-1), 3.34 (s, 3H, CH<sub>3</sub>CO), 1.52 (s, 3H, isopropylidene), 1.49 (s, 3H, isopropylidene), 1.43 (s, 3H, isopropylidene), 1.37 (s, 3H, isopropylidene). <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>): 101.4, 101.3 (2C, 2Me<sub>2</sub>CO<sub>2</sub>), 100.2, 99.8 (2C, 2C-1), 73.6, 71.4, 71.3, 71.2, 70.9, 68.9, 64.7, 64.3, 62.3, 62.2 (10C, C2–6, C2'–6'), 54.9 (OCH<sub>3</sub>), 29.3, 29.2, 19.4, 19.3 [4C, 2(CH<sub>3</sub>)<sub>2</sub>CO<sub>2</sub>]. Anal. calcd for C<sub>19</sub>H<sub>32</sub>O<sub>11</sub>: C, 52.28; H, 7.39. Found: C, 52.51; H, 7.67.

**Methyl 2-*O*-acetyl-3,4,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-4,6-*O*-isopropylidene- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-4,6-*O*-isopropylidene- $\alpha$ -D-mannopyranoside (25).** To a cooled solution (–20°C) of 23 (2.18 g, 5 mmol) and 24 (3.73 g, 5.5 mmol) in anhyd CH<sub>2</sub>Cl<sub>2</sub> (50 mL) was added TMSOTf (18  $\mu$ L, 0.05 mmol). The mixture was

stirred at this temperature for 2 h, and then quenched with Et<sub>3</sub>N (two drops). The solvents were evaporated in vacuo to give a residue, which was purified by silica gel column chromatography (1:1 petroleum ether–EtOAc) to give trisaccharide 25 (3.20 g, 67.2%) as a syrup.  $[\alpha]_D +91.7^\circ$  (*c* 1.0, CHCl<sub>3</sub>); <sup>1</sup>H NMR (CDCl<sub>3</sub>, 400 MHz):  $\delta$  7.95–7.34 (m, 15H, 3 PhH), 5.94 (dd, 1H,  $J_{3,4}=J_{4,5}=10.0$  Hz, H-4), 5.75 (dd, 1H,  $J_{2,3}=3.3$  Hz,  $J_{3,4}=10.0$  Hz, H-3), 5.53 (dd, 1H,  $J_{1,2}=1.8$  Hz,  $J_{2,3}=3.3$  Hz, H-2), 5.43 (d, 1H,  $J_{1,2}=1.8$  Hz, H-1), 5.31 (d, 1H,  $J_{1,2}=1.0$  Hz, H-1), 4.71 (d, 1H,  $J_{2,1}=1.0$  Hz, H-1), 4.66–4.58 (m, 3H), 4.26–4.18 (m, 4H), 4.04–3.99 (m, 2H), 3.90–3.82 (m, 4H), 3.68–3.63 (m, 2H), 3.36 (s, 3H, OCH<sub>3</sub>), 2.17 (s, 3H, COCH<sub>3</sub>), 1.56, 1.45 (2s, 6H, isopropylidene), 1.37 (s, 6H, isopropylidene); <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>): 169.5 (COCH<sub>3</sub>), 166.3, 165.6, 163.6 (5C, 5 C-Ph), 101.2, 100.8, (2C, 2 Me<sub>2</sub>C) 99.9, 99.4, 98.3 (3C, 3 C-1), 54.8 (OCH<sub>3</sub>), 29.1, 28.9 (2C, CH<sub>3</sub>CCH<sub>3</sub>), 20.6 (COCH<sub>3</sub>), 19.2, 19.0 (2C, CH<sub>3</sub>CCH<sub>3</sub>). Anal. calcd for C<sub>48</sub>H<sub>56</sub>O<sub>20</sub>: C, 60.50; H, 5.92. Found: C, 60.45; H, 5.66.

**Methyl 2-*O*-acetyl-3,4,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-2-*O*-acetyl-4,6-*O*-isopropylidene- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-2-*O*-acetyl-4,6-*O*-isopropylidene- $\alpha$ -D-mannopyranoside (26).** To a solution of 25 (95 mg, 0.1 mmol) in pyridine (5 mL) was added acetic anhydride (2.0 mL, 2 mmol). The reaction mixture was stirred at rt for 12 h, at the end of which time TLC (3:1 petroleum ether–EtOAc) indicated that the reaction was complete. The reaction mixture was concentrated, and then the residue was purified by flash column chromatography on a silica gel column (3:1 petroleum ether–EtOAc) to give compound 26 (83 mg, 80.6%) as a foamy solid.  $[\alpha]_D +82.6^\circ$  (*c* 1.0, CHCl<sub>3</sub>); <sup>1</sup>H NMR (CDCl<sub>3</sub>, 400 MHz):  $\delta$  8.10–7.23 (m, 65H, 13 PhH), 6.04 (dd, 1H,  $J_{3,4}=J_{4,5}=10.0$  Hz, H-4), 5.64 (dd, 1H,  $J_{2,3}=3.2$  Hz,  $J_{3,4}=10.0$  Hz, H-3), 5.45 (dd, 1H,  $J_{1,2}=2.0$  Hz,  $J_{2,3}=2.9$  Hz, H-2), 5.40 (dd, 1H,  $J_{1,2}=1.3$  Hz,  $J_{2,3}=3.4$  Hz, H-2), 5.32 (d, 1H,  $J_{1,2}=1.7$  Hz, H-1), 5.22 (dd, 1H,  $J_{1,2}=1.3$  Hz,  $J_{2,3}=2.9$  Hz, H-2), 5.10 (d, 1H,  $J_{1,2}=1.2$  Hz, H-1), 4.67 (dd, 1H,  $J_{2,3}=2.9$  Hz,  $J_{3,4}=12.1$  Hz, H-3), 4.63 (d, 1H,  $J_{1,2}=1.2$  Hz, H-1), 4.46–4.38 (m, 2H), 4.09–4.05 (m, 4H), 3.87–3.81 (m, 4H), 3.70–3.60 (m, 2H), 3.36 (s, 3H, OCH<sub>3</sub>), 2.31, 2.18, 2.07 (3s, 9H, 3 COCH<sub>3</sub>), 1.56, 1.52 (2s, 6H, isopropylidene), 1.37 (s, 6H, isopropylidene). Anal. calcd for C<sub>52</sub>H<sub>60</sub>O<sub>22</sub>: C, 60.22; H, 5.83. Found: C, 60.47; H, 5.61.

**Methyl 2-*O*-acetyl-3,4,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)]-4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)]-4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranoside (28).** To a cooled solution (–10°C) of 25 (1.90 g, 0.2 mmol) and 2 (303 mg, 0.5 mmol) in anhyd CH<sub>2</sub>Cl<sub>2</sub> (30 mL) was added TMSOTf (18 L, 0.1 mmol). After stirring at this temperature for 30 min, further TMSOTf (72  $\mu$ L, 0.4 mmol) was added to the reaction mixture and stirred for 20 min, then donor 2 (485 mg, 0.8 mmol) was added to the reaction mixture. The reaction mixture was stirred for 2 h, and then quenched with Et<sub>3</sub>N (four drops). The solvents were evaporated in vacuo to give a residue, which was

dissolved in 50 mL 90% TFA and stirred for 2 h, at the end of which time the reaction mixture was poured into 200 mL toluene and then the mixture was concentrated. Dried under high vacuo for 2 h, the residue was dissolved in pyridine (10 mL), and benzoyl chloride (1 mL) was added. The mixture was stirred at rt for 12 h, and then was evaporated and coevaporated with toluene in vacuo to give a residue. The residue was purified by silica gel column chromatography (3:1:1 petroleum ether–toluene–EtOAc) to give **28** (1.84 g, 42.3% for three steps) as a syrup.  $[\alpha]_D -58.6^\circ$  (*c* 1.0, CHCl<sub>3</sub>); <sup>1</sup>H NMR (CDCl<sub>3</sub>, 400 MHz):  $\delta$  8.11–7.34 (m, 65H, 13 PhH), 5.94 (dd, 1H,  $J_{3,4}=J_{4,5}=10.0$  Hz, H-4, Manp), 5.77 (dd, 1H,  $J_{2,3}=3.2$  Hz,  $J_{3,4}=10.0$  Hz, H-3, Manp), 5.71 (dd,  $J_{1,2}=J_{2,3}=6.3$  Hz, H-2, Xylp), 5.59 (dd, 1H,  $J_{3,4}=J_{4,5}=10.0$  Hz, H-4, Manp), 5.54 (dd, 1H,  $J_{3,4}=J_{4,5}=9.9$  Hz, H-4, Manp), 5.47–5.40 (m, 3H), 5.31–5.27 (m, 2H), 5.18 (d, 1H,  $J_{1,2}=0.7$  Hz, H-1, Manp), 5.16 (dd, 1H,  $J_{1,2}=1.0$  Hz,  $J_{2,3}=3.0$  Hz, H-2, Manp), 4.99 (d, 1H,  $J_{1,2}=0.8$  Hz, H-1, Manp), 4.80 (d, 1H,  $J_{1,2}=4.8$  Hz, H-1, Xylp), 4.53 (d, 1H,  $J_{1,2}=1.0$  Hz, H-1, Manp), 4.45 (d, 1H,  $J_{1,2}=6.1$  Hz, H-1, Xylp), 3.19 (s, 3H, OCH<sub>3</sub>), 1.90 (s, 3H, COCH<sub>3</sub>). <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>): 168.9 (COCH<sub>3</sub>), 166.0, 165.9, 165.8, 165.4, 165.3, 165.3, 165.2, 165.1, 165.0, 164.9, 164.7, 164.6 (13C, 13 C-1), 99.7, 99.7, 99.7, 99.0, 98.5 (5C, 5 C-1), 54.8 (OCH<sub>3</sub>), 20.3 (COCH<sub>3</sub>). Anal. calcd for C<sub>122</sub>H<sub>104</sub>O<sub>38</sub>: C, 67.27; H, 4.81. Found: C, 67.45; H, 5.96.

**Methyl 3,4,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)]-4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)]-4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranoside (**29**).** To a solution of **28** (1.09 g, 0.05 mmol) in anhyd CH<sub>2</sub>Cl<sub>2</sub> (10 mL) was added anhyd MeOH (40 mL), then acetyl chloride (1.0 mL) was added to the reaction mixture at 0°C. The solution was stoppered in a flask and stirred at room temperature until TLC (3:1 petroleum ether–EtOAc) showed that the starting material disappeared. The solution was neutralized with Et<sub>3</sub>N, then concentrated to dryness. The residue was passed through a short silica gel column to give **29** (715 mg, 66.8%) as a foamy solid.  $[\alpha]_D -17.5^\circ$  (*c* 0.6, CHCl<sub>3</sub>); <sup>1</sup>H NMR (CDCl<sub>3</sub>, 400 MHz):  $\delta$  8.12–7.34 (m, 65H, 13 PhH), 6.02 (dd, 1H,  $J_{3,4}=J_{4,5}=10.3$  Hz, H-4, Manp), 5.75 (dd,  $J_{1,2}=J_{2,3}=6.4$  Hz, H-2, Xylp), 5.66 (dd, 1H,  $J_{2,3}=3.2$  Hz,  $J_{3,4}=10.2$  Hz, H-3, Manp), 5.57 (dd, 1H,  $J_{3,4}=J_{4,5}=10.0$  Hz, H-4, Manp), 5.56 (dd, 1H,  $J_{3,4}=J_{4,5}=9.7$  Hz, H-4, Manp), 5.54–5.42 (m, 2H), 5.39 (m, 1H, H-4, Xylp), 5.32–5.29 (m, 2H), 5.22 (d, 1H,  $J_{1,2}=1.2$  Hz, H-1, Manp), 4.98 (s, 1H, H-1, Manp), 4.82 (d, 1H,  $J_{1,2}=4.8$  Hz, H-1, Xylp), 4.53 (d, 1H,  $J_{1,2}=1.7$  Hz, H-1, Manp), 4.52 (d, 1H,  $J_{1,2}=5.1$  Hz, H-1, Xylp), 3.18 (s, 3H, OCH<sub>3</sub>). <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>): 166.0, 165.9, 165.9, 165.6, 165.4, 165.4, 165.3, 165.2, 165.1, 165.0, 164.9, 164.6, 164.6 (13C, 13 C-1), 102.4, 99.9, 99.7, 99.3, 98.7 (5C, 5 C-1), 54.8 (OCH<sub>3</sub>). Anal. calcd for C<sub>120</sub>H<sub>102</sub>O<sub>37</sub>: C 67.47; H 4.81. Found: C 67.59; H 4.63.

**Methyl [methyl 2,3,4-tri-*O*-acetyl- $\beta$ -D-glucopyranosyluronate-(1 $\rightarrow$ 2)]-3,4,6-tri-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-*O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)]-4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[2,3,4-tri-**

***O*-benzoyl- $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)]-4,6-di-*O*-benzoyl- $\alpha$ -D-mannopyranoside (**30**).** To a cooled solution (0°C) of **29** (640 mg, 0.3 mmol) and methyl 2,3,4-tri-*O*-acetyl- $\alpha$ -D-glucopyranosyluronate trichloroacetimidate **16** (240 mg, 0.5 mmol) in anhyd CH<sub>2</sub>Cl<sub>2</sub> (10 mL) was added TMSOTf (8  $\mu$ L, 0.05 mmol). The mixture was stirred at this temperature for 2 h, and then quenched with Et<sub>3</sub>N (1 drop). The solvents were evaporated in vacuo to give a residue, which was purified by silica gel column chromatography (1:1 petroleum ether–EtOAc) to give hexasaccharide **30** (644 mg, 87.6%) as a foamy solid.  $[\alpha]_D -22.5^\circ$  (*c* 0.5, CHCl<sub>3</sub>); <sup>1</sup>H NMR (CDCl<sub>3</sub>, 400 MHz):  $\delta$  6.00 (dd, 1H,  $J_{3,4}=J_{4,5}=10.0$  Hz, H-4, Manp), 5.71 (dd, 1H,  $J_{2,3}=3.2$  Hz,  $J_{3,4}=10.0$  Hz, H-3, Manp), 5.15 (d, 1H,  $J_{1,2}=1.0$  Hz, H-1, Manp), 4.96 (d, 1H,  $J_{1,2}=0.9$  Hz, H-1, Manp), 4.75 (d, 1H,  $J_{1,2}=4.9$  Hz, H-1, Xylp), 4.60 (d, 1H,  $J_{1,2}=4.8$  Hz, H-1, Xylp), 4.56 (d, 1H,  $J_{1,2}=1.0$  Hz, H-1, Manp), 4.04 (d, 1H,  $J_{1,2}=6.6$  Hz, H-1, GluAp), 3.69 (s, 3H, COOCH<sub>3</sub>), 3.23 (s, 3H, OCH<sub>3</sub>), 1.96, 1.92, 1.31 (3s, 9H, 3 COCH<sub>3</sub>); <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>): 167.0, 168.5, 168.5, 168.3 (4C, 3 COCH<sub>3</sub>, COOMe), 165.9, 165.9, 165.9, 165.4, 165.4, 165.4, 165.3, 165.2, 165.1, 165.0, 164.9, 164.6, 164.6 (13C, 13 C-1), 100.9 (C-1,  $J_{C1,H1}=175$  Hz, Manp), 100.3 (C-1,  $J_{C1,H1}=163$  Hz, GluAp), 99.9 (C-1,  $J_{C1,H1}=164$  Hz, Xylp), 99.5 (C-1,  $J_{C1,H1}=163$  Hz, Xylp), 98.5 (C-1,  $J_{C1,H1}=172$  Hz, Manp), 95.2 (C-1,  $J_{C1,H1}=176$  Hz, Manp), 54.8 (OCH<sub>3</sub>), 52.3 (COOCH<sub>3</sub>), 20.5, 20.3, 20.2 (COCH<sub>3</sub>). Anal. calcd for C<sub>133</sub>H<sub>118</sub>O<sub>46</sub>: C, 65.13; H, 4.85. Found: C, 65.45; H, 4.56.

**Methyl ( $\beta$ -D-glucopyranosyluronic acid)-(1 $\rightarrow$ 2)- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[ $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)]- $\alpha$ -D-mannopyranosyl-(1 $\rightarrow$ 3)-[ $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 2)]- $\alpha$ -D-mannopyranoside, ammonium salt (**31**).** Hexasaccharide **30** (490 mg, 0.2 mmol) was dissolved in a satd methanolic ammonia (50 mL). After 36 h at room temperature, water (1.0 mL) was added to the mixture to cleave the methyl ester. After stirring at room temperature for 5 h, the reaction mixture was concentrated and purified on a Bio-Gel P2 column (eluent: water), affording the target hexasaccharide **31** (129 mg, 66.2%) as a foamy solid.  $[\alpha]_D +99.6^\circ$  (*c* 0.5, H<sub>2</sub>O); <sup>1</sup>H NMR (D<sub>2</sub>O, 400 MHz):  $\delta$  5.11 (s, 1H, H-1, Manp), 4.75 (s, 1H, H-1, Manp), 4.31 (s, 1H, H-1, Manp), 4.29 (d, 1H,  $J_{1,2}=8.0$  Hz, H-1, GluAp), 4.12 (d, 1H,  $J_{1,2}=8.9$  Hz, H-1, Xylp), 4.10 (d, 1H,  $J_{1,2}=9.2$  Hz, H-1, Xylp), 3.33 (s, 3H, OCH<sub>3</sub>); <sup>13</sup>C NMR (100 MHz, D<sub>2</sub>O): 174.1 (–COONH<sub>4</sub>), 103.4, 103.3, 103.2, 102.5, 100.3, 100.2 (6 C-1), 79.0, 78.5, 78.4, 78.3, 78.3, 76.4, 76.2, 75.9, 75.8, 73.6, 73.4, 73.2, 72.9, 72.9, 70.7, 70.3, 69.5, 69.5, 68.4, 67.0, 66.7, 66.5, 65.4, 65.3, 61.5, 60.6, 60.6, 56.6 (O–CH<sub>3</sub>). MALDI-TOF MS calcd for the ammonium salt of **31**, C<sub>35</sub>H<sub>61</sub>O<sub>30</sub>N: 975.8 [M]. Found: 975.8 (M); 980.9 (M–NH<sub>4</sub><sup>+</sup> + Na<sup>+</sup>).

## Acknowledgements

This work was supported by The Chinese Academy of Sciences (KZCX3-J-08) and by The National Natural Science Foundation of China (Projects 30070185 and 39970864).

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