Pergamon

Tetrahedron 55 (1999) 5965–5976

TETRAHEDRON

Synthesis of Oligosaccharide Fragments of Mannosylated Lipoarabinomannan from *Mycobacterium tuberculosis*

Vinodhkumar Subramaniam and Todd L. Lowary*

Department of Chemistry, The Ohio State University, 100 West 18th Avenue, Columbus, Ohio 43210

Received 7 January 1999; revised 10 March 1999; accepted 19 March 1999

Abstract. The synthesis of three mannosyl-arabinosides **4–6** found as terminal epitopes of mannosylated lipoarabinomannan (ManLAM) from *Mycobacterium tuberculosis* is reported. A key step in the synthesis of the required protected octyl β-D-arabinofuranoside derivative **7** involved glycosylation of octanol by 5-*O*-acetyl-2,3-di-*O*-benzyl-α-D-arabinofuranosyl chloride (**12**) in the absence of a promoter. The sequential addition of the mannopyranose residues to **7**, using thioglycosides **8** and **9**, provided the protected oligosaccharides **17**, **19**, and **21**. Deprotection by deacylation and then hydrogenation afforded the targets.

Keywords: Carbohydrates, Glycosidation, Thioglycosides

Introduction

Mycobacterium tuberculosis, the causative agent of tuberculosis, claims nearly three million lives each year and a third of the world's population has been estimated to be infected by this pathogen.[1,2] Mycobacterial infections, including those leading to tuberculosis, have recently attracted renewed attention. This is due not only to their increasing incidence in both the industrialized and developing world,[3] but also to the emergence of strains of these organisms resistant to commonly used anti-mycobacterial drugs.[4] Historically, the treatment of these diseases has been difficult, requiring strict adherence to a regimen of three or more antibiotics that must be taken for several months.[5] Such treatments are required because the cell wall of this organism provides a formidable barrier to the passage of antibiotics into the organism.[6]

All mycobacteria possess a protective cell wall complex composed of polysaccharides, proteins, and lipids.[7] The major structural component of this cell wall is an arabinogalactan which is covalently bound to mycolic acids, lipids characteristic to mycobacteria. Interspersed throughout this glycolipid framework is another polysaccharide, lipoarabinomannan (LAM), the major antigenic component of the cell wall. LAM has been implicated in many immunomodulatory events occurring during progression of tuberculosis, including the inhibition of macrophage activation,[8] the induction of cytokines,[9-11] the neutralization of potentially cytotoxic oxygen free radicals,[12] the inhibition of protein kinase activities,[13] and inducing the expression of collegenases that destroy the extracellular matrix of the lung.[14]

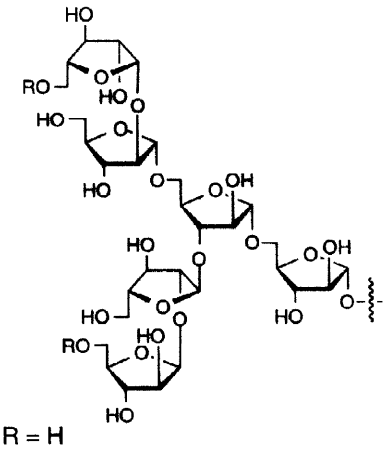
The structure of LAM includes a phosphatidyl inositol moiety which is believed to be noncovalently associated with the plasma membrane of the organism through its lipid portion.[15,16] To this inositol residue is attached a polysaccharide composed of mannopyranose and arabinofuranose, the terminal ends of which are capped with the arabinofuranosyl hexasaccharide **1**. Although in many mycobacterial strains this motif is found

Email: lowary.2@osu.edu

0040-4020/99/$ - see front matter
PII: S0040-4020(99)00260-4

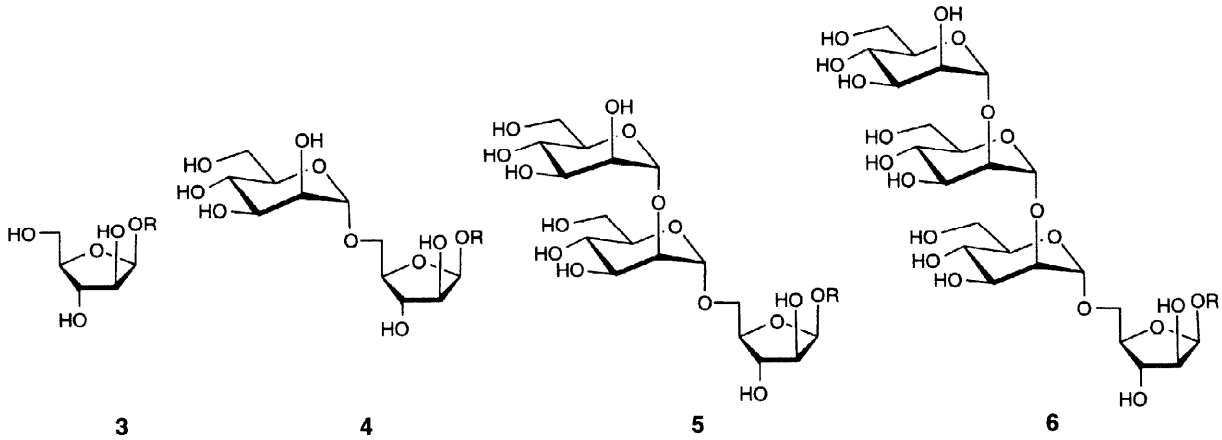
unsubstituted, in others **1** is further substituted with short mannopyranosyl oligosaccharides to provide a polymer known as ManLAM **2**.[7,15,16]

The terminal mannopyranosyl residues of ManLAM have been suggested to be involved in the initial stages of infection by adhering to human cells through their interaction with mannose binding proteins.[17,18] Consequently, drugs that act by inhibiting the enzymes that assemble these oligosaccharides are of possible use in the prevention and treatment of mycobacterial disease. The design of such inhibitors would be greatly facilitated by the isolation and characterization of these mannosyltransferases. This goal is in turn dependent upon the ready availability of potential oligosaccharide substrates which can be used for purification of these enzymes and in the development of assays for their activities. Although biosynthetic studies of LAM have been carried out,[16] the assembly of the mannose caps has not been investigated. We describe here the first syntheses of a series of glycan fragments of ManLAM (**3**–**6**, Figure 2), which are potential substrates for the mannosyltransferases involved in its biosynthesis.



1 R = H
2 R = α-D-Man*p* **or** α-D-Man*p*-α-(1→2)-D-Man*p* **or** α-D-Man*p*- α-(1→2)-D-Man*p* -α-(1→2)-D-Man*p*

Figure 1

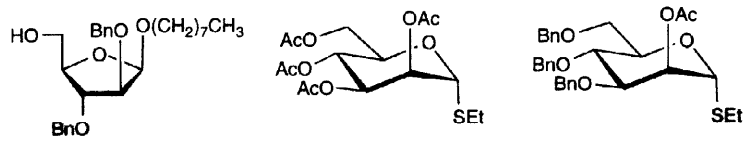


3 **4** **5** **6**

Figure 2. Synthetic Targets, R = $(CH_2)_7CH_3$

Results and Discussion

We envisioned that oligosaccharides **4**–**6** could be prepared straightforwardly from monosaccharide building blocks **7**, **8**, and **9**, through a route involving the sequential addition of mannose residues to arabinofuranosyl-containing acceptors. Thioglycosides were chosen as the glycosyl donors due to their hydrolytic stability as well as the number of methods available for their activation.[19] We chose to prepare octyl glycosides because this hydrophobic group simplifies glycosyltransferase assays by allowing the use of reverse phase (C_{18}) cartridges to separate and quantitate reaction products.[20] A critical issue to be addressed early in the synthetic sequence was the formation of the 1,2-*cis*-glycosidic bond on the arabinofuranose residue. This linkage is directly analogous to that of a β-mannopyranoside, which is notoriously difficult to prepare.[21]



7 8 9

Figure 3. Monosaccharide Building Blocks

Synthesis of monosaccharides. Thioglycosides **8**[22] and **9**[23] are known and were readily prepared from D-mannose as previously described. In contrast, there is considerably less precedence for the synthesis of **7**. Relatively little work has been done on the synthesis of furanosidic oligosaccharides and, since Fletcher and Glaudemans reported the preparation of methyl β-D-arabinofuranoside over 30 years ago,[24] the stereoselective synthesis of linkages of this type has not been investigated. An intermolecular aglycone delivery route for the stereoselective synthesis of β-D-fructofuranosides has been recently reported[25] and, given the close structural similarities between these glycosides and **7**, a similar approach is likely to succeed. However, the simplicity of the approach reported by Fletcher[24] prompted us to investigate this approach first as illustrated in Figure 4.

Commercially available 2,3,5-tri-*O*-benzyl-D-arabinofuranose (**10**) was treated under acetolysis conditions which resulted in the simultaneous cleavage of the O-5 benzyl ether and acetylation of the anomeric hydroxyl group. The product, **11**, was obtained in 86% yield. Reaction of this diacetate with HCl in dichloroethane provided the labile chloride **12** which was not isolated. Instead, immediately after its formation, **12** was reacted with octanol in the absence of a promoter to give the β-glycoside **13**. The product was contaminated with traces of the α-isomer. However, separation could easily be achieved by first removing the acetyl group and then chromatography. The conversion of **11** to **7** proceeded in 35% overall yield. Despite this modest yield, the speed and ease with which **7** can be prepared by this route makes it a viable alternative to more elaborate methods (*e.g.*, the previously mentioned intramolecular aglycone delivery route[25]) that would possibly give the higher yields in the glycosylation step. Chloride **15**, which was prepared in 2 steps from **10**, was reacted with octanol in a manner identical to **12** to provide a 58% yield of the glycoside **16**. In this case, the formation of the α-anomer was not detected.[26] Hydrogenation of **16** gave arabinofuranoside **3** in 82% yield.

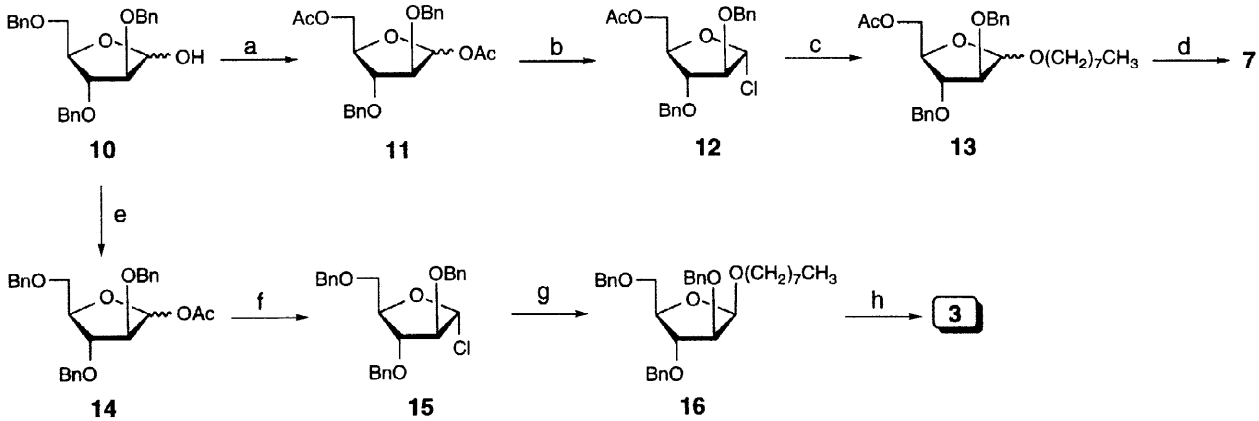


Figure 4. (a) Ac_2O, H_2SO_4, 86%; (b) HCl, $ClCH_2CH_2Cl$; (c) $CH_3(CH_2)_7OH$, CH_2Cl_2; (d) $NaOCH_3$, CH_3OH, 3 steps, 35%; (e) Ac_2O, pyridine, reflux, 100%; (f) HCl, $ClCH_2CH_2Cl$; (g) $CH_3(CH_2)_7OH$, CH_2Cl_2, 2 steps, 58%; (h) Pd/C, H_2, CH_3OH, 82%.

Determining the stereochemical outcome of these glycosylation reactions could be easily done by standard one-dimensional ^{13}C and ^{1}H NMR experiments.[27] The anomeric carbon of α-D-arabinofuranosides resonate in the range of 107–110 ppm whereas the β-anomers appear between 97 and 104 ppm. Furthermore, $^{3}J_{H1,H2}$ is small (0–2 Hz) for the α-anomers and larger (3–5 Hz) for the β-anomers. It should also be mentioned that although $^{1}J_{C1,H1}$ values are unambiguous determinants of anomeric stereochemistry in mannopyranosides,[28] they cannot be used here to make these assignments. In arabinofuranosides the magnitudes of these couplings have been shown to be insensitive to anomeric configuration.[27]

It is tempting to speculate that the formation of β-glycosides from **12** and **15** proceeds through a direct S_N2 displacement reaction. However, the methanolysis of 2-benzylated arabinofuranosyl chlorides has been previously studied[24,29] and it was shown that these reactions proceed through an ion-pair S_N1 mechanism. It should be noted that although the unpromoted glycosylation of these furanosyl halides proceeds efficiently with simple primary alcohols, attempts to react **15** with secondary carbohydrate alcohols produced no glycosides, only the hydrolysis product, **10**.[30] Therefore, while this method is useful for the preparation of simple glycosides it appears to be of limited utility in oligosaccharide synthesis. Furthermore, it should also be mentioned that earlier work on the glycosylation of **12** in the presence of a promoter (Ag_2O, $AgCO_3$) results in the formation of the α-glycoside.[31] This suggests that in the presence of a promoter, a naked carbocation forms and that the attack of the alcohol proceeds through the less sterically hindered face of the ring.

Synthesis of oligosaccharides. With the building blocks **7–9** in hand, the assembly of the oligosaccharides proceeded without serious difficulty. However, careful control of the temperature during the glycosylations was required in order to obtain good stereoselectivities. Despite the presence of a participating protecting group at C-2 in the donor, initiating the glycosylations at 0 °C or -10 °C resulted in the formation of significant amounts of the undesired β-anomers. For example, in the case of glycosylation of **7** with **9** to give **17** (Figure 5), at 0 °C a 3:2 α:β ratio was obtained. When the same reaction was carried out at -10 °C the α:β ratio was 3:1.[32] In addition to lowering the overall yield, the formation of the β-glycosides greatly complicated purification as the two glycosides had very similar chromatographic mobilities in all solvent systems investigated. Best results were obtained by starting the reaction at -40 °C and allowing it to react as it warmed to 0 °C. Under these conditions, only trace amounts of the β-mannoside were formed, which could be separated from the desired compound by chromatography to give the product which was pure by ^{1}H NMR spectroscopy.

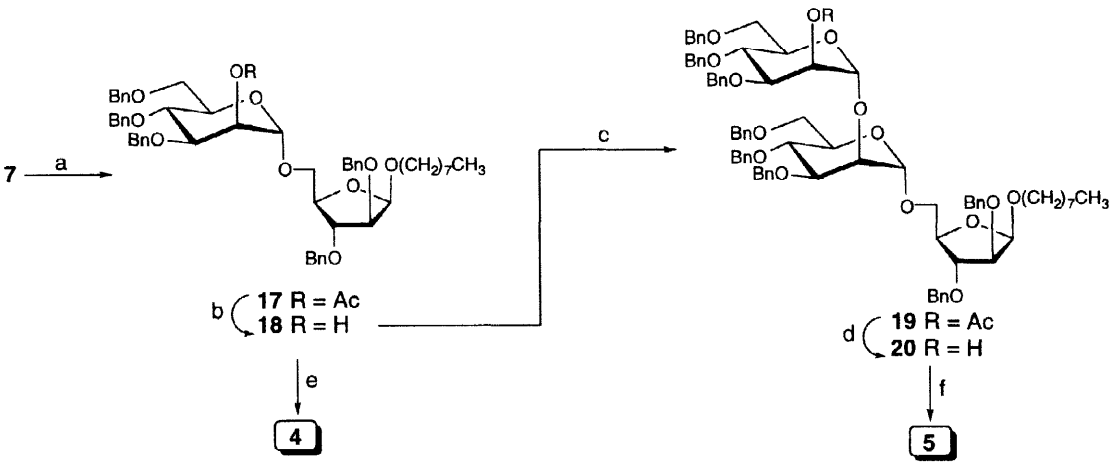


Figure 5. (a) **9**, NIS, AgOTf, CH_2Cl_2, -40 °C → rt, 82%; (b) $NaOCH_3$, CH_3OH, 95%; (c) **9**, NIS, AgOTf, CH_2Cl_2, -40 °C → rt, 87%; (d) $NaOCH_3$, CH_3OH, 96%; (e) Pd/C, H_2, CH_3OH, 90%; (f) Pd/C, H_2, CH_3OH, 93%.

The syntheses of disaccharide **4** and trisaccharide **5** are illustrated in Figure 5. Reaction of alcohol **7** with thioglycoside **9** promoted by *N*-iodosuccinimide and silver triflate afforded the protected disaccharide **17** in 82% yield. Deacetylation proceeded in 95% yield to give alcohol **18**. A portion of this product was completely deprotected by hydrogenation providing **4** in 90% yield. The remainder of **18** was glycosylated, again with **9** under the same reaction conditions used for the synthesis of the disaccharide, to afford trisaccharide **19** in 87% yield. Treatment with sodium methoxide afforded a 96% yield of alcohol **20** which could then be converted, upon hydrogenation, to **5** (93%). Tetrasaccharide **6** was synthesized as shown in Figure 6. Its preparation involved first the reaction of trisaccharide **20** with the fully acylated mannose thioglycoside **8** to give **21** in 76% yield. Treatment with sodium methoxide followed by hydrogenation afforded **6** in 78% yield. Compouds **3–6** were all shown to be pure by 800 MHz ^{1}H NMR spectroscopy.

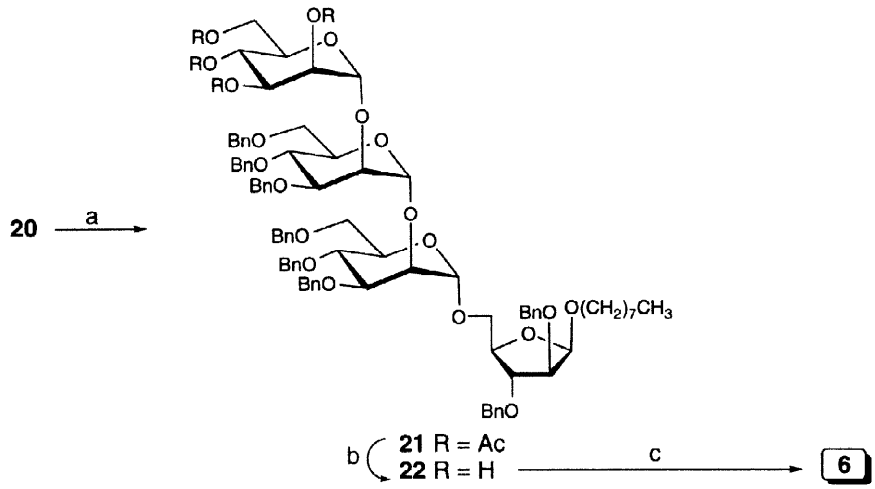


Figure 6. (a) **8**, NIS, AgOTf, -40 °C → rt, 76%; (b) $NaOCH_3$, CH_3OH; (c) Pd/C, H_2, CH_3OH; 78%, two steps.

In conclusion, we have carried out the first syntheses of a series of mannosyl-arabinosides found as constituent parts of ManLAM from *M. tuberculosis*. The availability of these glycans will be important in elucidating the pathways by which this polysaccharide is assembled by the organism. The results of these biosynthetic investigations will be reported in due course.

Experimental

Optical rotations were measured at 22 ± 2 °C. Analytical TLC was perform ed on silica gel 60-F_{254} (0.25 mm, Merck). Spots were detected under UV light or by charring with 10% H_2SO_4 in ethanol. Unless otherwise indicated, all reactions were carried out at room temperature and under positive pressure of argon. Solvents were evaporated under reduced pressure and below 40 °C. Column chromatography was performed on silica gel or Iatrobeads. The ratio between silica gel and compound ranged from 100 to 50:1 (w/w). ^{1}H NMR spectra were recorded at 250 or 300 or 500 MHz, and first order proton chemical shifts δ_H are referenced to either to TMS (δ_H 0.0, $CDCl_3$) or HOD (δ_H 4.78, D_2O). ^{13}C NMR spectra were recorded at 75 or 125 MHz and ^{13}C chemical shifts δ_C are referenced to either to TMS (δ_C 0.0, $CDCl_3$) or dioxane (δ_C 67.4, D_2O). The assignment of resonances in **3–6** were made by two-dimensional homonuclear and heteronuclear shift correlation experiments. One-bond

carbon-hydrogen coupling constants involving the anomeric carbon of the mannose residues in **4–6** were measured to prove glycoside stereochemistry.[28] Elemental analyses were performed by Atlantic Microlab Inc., GA and the samples submitted for analyses were dried overnight under vacuum with phosphorus pentoxide at 56 °C (refluxing acetone). Fast atom bombardment mass spectra were recorded on samples suspended in thioglyceride matrix with a cesium gun. MALDI mass spectra were recorded on samples in an α-cyano-4-hydroxycinnamic acid matrix.

1,5-Di-O-acetyl-2,3-di-O-benzyl-D-arabinofuranose (**11**). To a solution of **10**[33] (2.5 g, 5.95 mmol) in acetic anhydride (15 mL) was added a solution containing 6 drops of conc. H_2SO_4 in acetic anhydride (5 mL). The reaction was stirred for 2 h, and was then poured into ice (100 g) and stirred for 2 h, before being extracted with CH_2Cl_2 (3 x 30 mL). The organic layer was washed with water (30 mL) followed by a saturated $NaHCO_3$ solution (2 x 30 mL) and water (30 mL). The organic extract was dried (Na_2SO_4), filtered and concentrated to a light yellow syrup, which was purified by chromatography (4:1 toluene:EtOAc) to give **11** (2.1 g, 86%) as a colorless syrup in a 9:1 α:β ratio of anomers. R_f 0.27 (4:1 toluene:EtOAc); $[\alpha]_D$ -21.6° (*c* 0.8, $CHCl_3$); ^1H NMR (250 MHz, $CDCl_3$) δ_H 7.25–7.40 (m, Ph), 6.30 (d, $J_{1,2}$ = 4.2 Hz, H-1β), 6.23 (br. s, H-1α), 6.67 (d, J = 11.8 Hz, CH_2Ph) 4.50–4.69 (m, CH_2Ph), 4.37 (ddd, $J_{4,5a}$ = 3.8, $J_{3,4}$ = 5.8, $J_{4,5b}$ = 6.0 Hz, H-4α), 4.27 (dd, $J_{4,5a}$ = 3.8, $J_{5a,5b}$ = 11.9 Hz, H-5aα), 4.13 (dd, $J_{4,5b}$ = 6.0, $J_{5a,5b}$ = 11.9 Hz, H-5bα), 4.08 (d, $J_{2,3}$ = 2.3 Hz, H-2α), 3.89 (dd, $J_{2,3}$ = 2.3, $J_{3,4}$ = 5.8 Hz, H-3α), 2.08 (s, α-OCOCH_3), 2.06 (s, β-OCOCH_3), 2.04 (s, β-OCOCH_3), 2.01 (s, α-OCOCH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 170.7, 169.9 (C=O), 137.7, 137.3, 137.1, 128.6, 128.2, 128.1, 128.0, 128.0 (Ph), 100.5 (C-1α), 94.5 (C-1β), 86.8, 84.1, 83.6, 81.8, 81.2, 72.7, 72.3, 72.2, 65.2, 63.7 (ring and benzylic C), 21.3, 21.2, 20.8 (OCO*C*H$_3$). Anal. Calcd for $C_{23}H_{28}O_8$ (414.22): C, 66.64; H, 6.33. Found: C, 66.40; H, 6.30.

Octyl 2,3-di-O-benzyl-β-D-arabinofuranoside (**7**). HCl gas was bubbled through a solution of **11** (1.4 g, 3.38 mmol) in dry dichloroethane (10 mL) for 30 min. The reaction mixture was stirred for an additional 10 min and the solvent was concentrated to provide 5-*O*-acetyl-2,3-di-*O*-benzyl-α-D-arabinofuranosyl chloride **12** as a light yellow syrup. Without further purification, the crude product was dissolved in dry CH_2Cl_2 (20 mL) and octanol (0.37 mL, 2.35 mmol) was added. After stirring for 90 min, the reaction mixture was diluted with CH_2Cl_2 (40 mL) and then washed with water (30 mL) followed by a saturated $NaHCO_3$ solution (2 x 30 mL) and water (25 mL). The organic layer was dried (Na_2SO_4), filtered and concentrated to give crude **13** as light yellow syrup. The product was dissolved in methanol (15 mL) and a few drops of 0.1 M $NaOCH_3$ was added and the reaction was stirred for 4 h. The reaction mixture was concentrated and the unreacted octanol was removed by azeotropic distillation with water (3 x 40 mL). The light yellow oil obtained was purified by chromatography (85:15 hexane: EtOAc) to obtain **7** (0.5 g, 35%) as a colorless oil. R_f 0.31 (4:1 hexane:EtOAc); $[\alpha]_D$ +14.5° (*c* 0.8, $CHCl_3$); ^1H NMR (200 MHz, $CDCl_3$) δ_H 7.25–7.39 (m, 15 H, Ph), 4.85 (d, 1 H, $J_{1,2}$= 4.4 Hz, H-1), 4.73 (d, 1 H, J = 11.7 Hz, CH_2Ph), 4.60 (s, 2 H, CH_2Ph), 4.58 (d, 1 H, J = 10.7 Hz, CH_2Ph), 4.31 (dd, 1 H, $J_{2,3}$ = 5.8, $J_{3,4}$ = 7.0 Hz, H-3), 4.00–4.10 (m, 2 H, H-2, H-4), 3.50–3.80 (m, 3 H, H-5a, H-5b, octyl OCH_2), 3.39 (dt, 1 H, J = 6.8, 9.6 Hz, octyl OCH_2), 2.37 (dd, 1 H, $J_{5a,5\text{-}OH}$ = 4.2, $J_{5b,5\text{-}OH}$ = 7.7 Hz, 5-OH), 1.58–1.65 (m, 2 H, octyl CH_2), 1.23–1.29 (m, 10 H, octyl CH_2), 0.87 (t, 3 H, J = 6.1 Hz, octyl CH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 138.1, 137.7, 128.4, 128.1, 128.0, 127.8, 127.7 (Ph), 100.8 (C-1), 84.5, 82.2, 81.2, 72.5, 72.4, 69.2, 63.8 (ring and benzylic C, octyl OCH_2), 31.8, 29.6, 29.5, 29.4, 26.1, 22.6

(octyl CH_2), 14.1 (octyl CH_3). Anal. Calcd for $C_{27}H_{38}O_5$ (414.22): C, 73.27; H, 8.65. Found: C, 73.37; H, 8.59.

1-O-Acetyl-2,3,5-tri-O-benzyl-D-arabinofuranose (**14**). This compound was prepared as previously described.[34]

Octyl 2,3,5-tri-O-benzyl-β-D-arabinofuranoside (**16**). HCl gas was bubbled through a solution of **14** (850 mg, 1.87 mmol) in dry dichloroethane (10 mL) for 30 min. After stirring for an additional 10 min the reaction mixture was concentrated to provide 2,3,5-tri-*O*-benzyl-α-D-arabinofuranosyl chloride **15** as a light yellow syrup. Without further purification, the crude product was dissolved in dry CH_2Cl_2 (15 mL) and octanol (0.35 mL, 2.24 mmol) was added. The reaction mixture was stirred for 60 min and then diluted with CH_2Cl_2 (40 mL), before being washed with water (30 mL), a saturated $NaHCO_3$ solution (2 x 30 mL) and water (25 mL). The organic layer was dried (Na_2SO_4), filtered, and concentrated under vacuum to a light yellow syrup, which was purified by column chromatography (9:1 hexane:EtOAc) to give **16** (440 mg, 58%) as a colorless syrup. R_f 0.66 (4:1 hexane:EtOAc); $[\alpha]_D$ -49.6° (*c* 1.4, $CHCl_3$); ^1H NMR (300 MHz, $CDCl_3$) δ_H 7.26–7.40 (m, 15 H, Ph), 4.90 (d, 1 H, $J_{1,2}$ = 4.0 Hz, H-1), 4.70 (d, 1 H, *J* = 11.7 Hz, CH_2Ph), 4.60–4.66 (m, 3 H, CH_2Ph), 4.70 (s, 2 H, CH_2Ph), 4.05–4.22 (m, 3 H, H-2, H-3, H-4), 3.68 (dt, 1 H, *J* = 6.8, 9.2 Hz, octyl OCH_2), 3.50–3.58 (m, 2 H, H-5a, H-5b), 3.34 (dt, 1 H, *J* = 6.8, 9.2 Hz, octyl OCH_2), 1.55–1.62 (m, 2 H, octyl CH_2), 1.29–1.35 (m, 10 H, octyl CH_2), 0.91 (t, 3 H, *J* = 6.4 Hz, octyl CH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 138.2, 138.1, 137.7, 128.3, 128.2, 128.2, 128.0, 127.8, 127.7, 127.6, 127.5, 127.5 (Ph), 100.5 (C-1), 84.2, 83.5, 80.1, 73.2, 72.7, 72.3, 72.2, 67.9 (ring and benzylic C, octyl OCH_2), 31.8, 29.4, 29.3, 29.2, 26.2, 22.6 (octyl CH_2), 14.0 (octyl CH_3). Anal. Calcd for $C_{34}H_{44}O_5$ (532.69): C, 76.66; H, 8.33. Found: C, 76.62; H, 8.36.

Octyl β-D-arabinofuranoside (**3**). To a solution of **16** (250 mg, 0.48 mmol) in methanol (10 mL) was added 10% Pd/C (70 mg). The reaction mixture was stirred under H_2 atmosphere for 6 h at room temperature, filtered, washed with methanol (2 x 5 mL) and CH_2Cl_2 (5 mL) and then concentrated under vacuum to obtain a **3** as colorless syrup (100 mg, 82%). R_f 0.38 (1:4 toluene:EtOAc); $[\alpha]_D$ -29.3° (*c* 1.1, $CHCl_3$); ^1H NMR (800 MHz, D_2O) δ_H 4.90 (d, 1 H, $J_{1,2}$ = 4.7 Hz, H-1), 4.02 (dd, 1 H, $J_{1,2}$ = 4.7, $J_{2,3}$ = 7.8 Hz, H-2), 3.93 (dd, 1 H, $J_{2,3}$ = 7.8, $J_{3,4}$ = 7.3 Hz, H-3), 3.79 (ddd, 1 H, $J_{3,4}$ = 7.3, $J_{4,5a}$ = 3.9, $J_{4,5b}$ = 7.0 Hz, H-4), 3.65–3.70 (m, 2 H, H-5a, octyl OCH_2), 3.55 (dd, 1 H, $J_{4,5b}$ = 7.0, $J_{5a,5b}$ = 11.9 Hz, H-5b), 3.42 (dt, 1 H, *J* = 6.6, 9.5 Hz, octyl OCH_2), 1.50–1.52 (m, 2 H, octyl CH_2), 1.20–1.26 (m, 10 H, octyl CH_2), 0.79 (t, 3 H, *J* = 6.6 Hz, octyl CH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 101.1 (C-1), 82.3 (C-4), 78.2 (C-2), 75.8 (C-3), 69.2 (C-5), 63.0 (octyl OCH_2), 31.7, 29.5, 29.2, 29.1, 25.8, 22.5, (octyl CH_2), 14.0 (octyl CH_3). HR-FAB-MS calcd for $C_{13}H_{27}O_5$ $[M + H]^+$ 263.1859, found 263.1874.

Octyl 2-O-acetyl-3,4,6-tri-O-benzyl-α-D-mannopyranosyl-(1→5)-2,3-di-O-benzyl-β-D-arabinofuranoside (**17**). Thioglycoside **9** (230 mg, 0.43 mmol), alcohol **7** (170 mg, 0.39 mmol), and powdered 4 Å molecular sieves (300 mg) were placed in a round-bottom flask and dried overnight in vacuo. Dry CH_2Cl_2 (20 mL) was added and the mixture was cooled to -40 °C and stirred for 10 min. *N*-iodosuccinimide (115 mg, 0.51 mmol) was added and the reaction was stirred for 20 min before silver triflate (31 mg, 0.12 mmol) was added. The reaction mixture was allowed to gradually warm to 0 °C and was then quenched by addition of triethylamine

(0.5 mL). The yellow solution was filtered, diluted with CH_2Cl_2 (20 mL), washed with saturated $Na_2S_2O_3$ solution (2 x 20 mL) followed by brine (20 mL) and water (20 mL). After drying (Na_2SO_4), the organic layer was concentrated to a light yellow syrup, which was chromatographed (85:15 hexane:EtOAc) to give **17** (323 mg, 82%) as a colorless syrup. R_f 0.33 (4:1 hexane:EtOAc); $[\alpha]_D$ +3.4° (*c* 1.2, $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 7.16–7.36 (m, 25 H, Ph), 5.38 (dd, $J_{1',2'}$ = 1.8, $J_{2',3'}$ = 3.0 Hz, H-2′), 4.41–4.90 (m, 12 H, H-1, H-1′, 10 x C*H*$_2$Ph), 3.60–4.10 (m, 10 H, H-2, H-3, H-4, H-5a, H-5b, H-3′, H-4′, H-5′, H-6′a, octyl OCH_2), 3.51 (dd, 1 H, $J_{5',6'a}$ = 5.6, $J_{6'a,6'b}$ = 10.1 Hz, H-6′b), 3.30 (dt, 1 H, *J* = 6.9, 9.7 Hz, octyl OCH_2), 2.13 (s, 3 H, OCO*C*H$_3$), 1.56–1.64 (m, 2 H, octyl CH_2), 1.21–1.29 (m, 10 H, octyl CH_2), 0.87 (t, 3 H, *J* = 6.7 Hz, octyl CH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 170.3 (C=O), 138.5, 138.2, 138.1, 138.0, 137.9, 137.7, 128.4, 128.3, 128.3, 128.2, 128.1, 128.0, 128.0, 127.9, 127.8, 127.8, 127.8, 127.7, 127.5, 127.5 (Ph), 100.5 (C-1), 98.1 (C-1′), 84.2, 83.3, 79.5, 78.3, 75.1, 74.2, 73.5, 72.4, 71.8, 71.6, 71.5, 70.1, 68.6, 68.5, 68.0 (ring and benzylic C, octyl OCH_2), 31.8, 29.5, 29.4, 29.3, 26.2, 22.6 (octyl CH_2), 21.2 (OCO*C*H$_3$), 14.1 (octyl CH_3). Anal. Calcd for $C_{56}H_{68}O_{11}$ (917.15): C, 73.34; H, 7.47. Found: C, 73.62; H, 7.40.

Octyl α-D-mannopyranosyl-(1→5)-β-D-arabinofuranoside (**4**). A solution of **17** (210 mg, 0.23 mmol) was dissolved in methanol (20 mL) and then 2 drops of 1M NaOMe was added. After stirring overnight, the solution was neutralized with a minimum amount of Amberlite118 H^+ resin and concentrated to a syrup, which was purified by chromatography (8:1 hexane:EtOAc) to give **18** (190 mg, 95%) as a colorless syrup. ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 7.18–7.44 (m, 25 H, Ph), 4.98 (d, $J_{1',2'}$ = 1.4 Hz, H-1′), 4.91 (d, 1 H, $J_{1,2}$ = 4.2 Hz, H-1), 4.87 (d, 1 H, *J* = 10.9 Hz, C*H*$_2$Ph), 4.47–4.77 (m, 9 H, C*H*$_2$Ph), 3.64–4.24 (m, 11 H, H-2, H-3, H-4, H-5a, H-5b, H-2′, H-3′, H-4′, H-5′, H-6′a, octyl OCH_2), 3.59 (dd, 1 H, $J_{5',6'b}$ = 5.7, $J_{6'a,6'b}$ = 10.2 Hz, H-6′b), 3.36 (dt, 1 H, *J* = 6.9, 9.7 Hz, octyl OCH_2), 2.53 (br. s, 1 H, OH), 1.56–1.64 (m, 2 H, octyl CH_2), 1.26–1.33 (m, 10 H, octyl CH_2), 0.94 (t, 3 H, *J* = 6.8 Hz, octyl CH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 138.5, 138.2, 138.2, 138.0, 137.8, 128.6, 128.5, 128.4, 128.2, 128.0, 127.8, 127.7, 127.6 (Ph), 100.5 (C-1), 99.7 (C-1′), 84.3, 83.4, 80.3, 79.6, 75.2, 74.2, 73.5, 72.5, 72.5, 72.1, 71.3, 69.7, 68.8, 68.3, 68.2 (ring and benzylic C, octyl OCH_2), 31.9, 29.6, 29.5, 29.4, 26.3, 22.8 (octyl CH_2), 14.2 (octyl CH_3).

To a solution of **18** (100 mg, 0.11 mmol) in methanol (8 mL), was added 10% Pd/C (25 mg). The solution was stirred overnight under H_2 atmosphere and then the catalyst was separated by filtration and washed with CH_3OH (10 mL). After concentrating, the product was purified by chromatography (4:1 CH_2Cl_2:CH_3OH) on silica gel to give **4** (41 mg, 90%) as a foam. R_f 0.53 (4:1 CH_2Cl_2:CH_3OH); $[\alpha]_D$ +2.9° (*c* 1.2, H_2O); ^{1}H NMR (800 MHz, D_2O) δ_H 4.91 (m, 1 H, H-1), 4.84 (d, 1 H, $J_{1',2'}$ = 1.6 Hz, H-1′), 4.02–4.06 (m, 2 H, H-2, H-3), 3.91–3.94 (m, 1 H, H-4), 3.90 (dd, 1 H, $J_{1',2'}$ = 1.6, $J_{2',3'}$ = 3.3 Hz, H-2′), 3.70–3.81 (m, 4 H, H-5a, H-3′, H-6′a, H-6′b), 3.61–3.68 (m, 3 H, H-4′, H-5b, octyl OCH_2), 3.57 (ddd, 1 H, $J_{4',5'}$ = 9.7, $J_{5',6'a}$ = 2.1, $J_{5',6'b}$ = 5.0 Hz, H-5′), 3.42 (dt, 1 H, *J* = 6.8, 9.6 Hz, octyl OCH_2), 1.58–1.65 (m, 2 H, octyl CH_2), 1.23–1.29 (m, 10 H, octyl CH_2), 0.87 (t, 3 H, *J* = 6.1 Hz, octyl CH_3); ^{13}C NMR (125.8 MHz, D_2O) δ_C 101.4 (C-1, $^1J_{C,H}$ =171.8 Hz), 100.3 (C-1′,$^1J_{C,H}$ = 171.1 Hz), 80.0 (C-4), 76.8 (C-2), 75.3 (C-3), 73.3 (C-5′), 71.0 (C-3′), 70.1 (C-2′), 68.9 (octyl OCH_2), 68.8 (C-5), 67.0 (C-4′), 61.2 (C-6′), 31.7, 29.5, 29.2, 29.1, 25.8, 22.5 (octyl CH_2), 14.0 (octyl CH_3). HR-FAB-MS calcd for $C_{19}H_{37}O_{10}$ $[M + H]^+$ 425.2387, found 425.2386.

Octyl 2-O-acetyl-3,4,6-tri-O-benzyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-O-benzyl-α-D-mannopyranosyl-(1→5)-2,3-di-O-benzyl-β-D-arabinofuranoside (**19**). Thioglycoside **9** (120 mg, 0.22 mmol), disaccharide

18 (150 mg, 0.17 mmol), and powdered 4 Å molecular sieves (200 mg) were placed in a round-bottom flask and dried overnight in vacuo. Dry CH_2Cl_2 (15 mL) was added and the mixture was cooled to -40 °C and stirred for 10 min. *N*-iodosuccinimide (52 mg, 0.23 mmol) was added and the reaction was stirred for 20 min before the addition of silver triflate (13 mg, 0.05 mmol). The reaction mixture was allowed to gradually warm to 0 °C and was then quenched by the addition of triethylamine (0.3 mL). The yellow solution was filtered, diluted with CH_2Cl_2 (20 mL), washed with an saturated $Na_2S_2O_3$ solution (2 x 20 mL) followed by brine (20 mL) and water (20 mL). After drying (Na_2SO_4), it was filtered and concentrated under vacuum to a light yellow syrup, which was chromatographed (85:15 hexane:EtOAc) to give **19** (130 mg, 87%) as a colorless syrup. R_f 0.32 (4:1 hexane:EtOAc); $[\alpha]_D$ +4.7° (*c* 1.3, $CHCl_3$); ^1H NMR (250 MHz, $CDCl_3$) δ_H 7.13–7.37 (m, 40 H, Ph), 5.54 (dd, 1 H, $J_{1'',2''}$ = 1.7, $J_{2'',3''}$ = 3.0 Hz, H-2''), 5.11 (d, 1 H, $J_{1'',2''}$ = 1.7 Hz, H-1''), 4.79–4.92 (m, 5 H, H-1, H-1', 3 x C*H*$_2$Ph), 4.41–4.71 (m, 12 H, C*H*$_2$Ph), 4.38 (d, 1 H, *J* = 10.9 Hz, C*H*$_2$Ph), 3.64–4.13 (m, 16 H, H-2, H-3, H-4, H-5a, H-5b, H-2', H-3', H-4', H-5', H-6'a, H-3'', H-4'', H-5'', H-6''a, H-6''b, octyl OCH_2), 3.47 (dd, 1 H, $J_{5',6'b}$ = 5.6, $J_{6'a,6'b}$ = 10.0 Hz, H-6'b), 3.30 (dt, 1 H, *J* = 6.9, 9.8 Hz, octyl OCH_2), 2.13 (s, 3 H, OCO*C*H$_3$) 1.56–1.64 (m, 2 H, octyl CH_2), 1.21–1.29 (m, 10 H, octyl CH_2), 0.88 (t, 3 H, *J* = 6.9 Hz, octyl CH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 170.1 (C=O), 138.5, 138.4, 138.4, 138.3, 138.2, 138.1, 138.0, 137.7, 128.4, 128.3, 128.2, 128.2, 128.2, 128.1, 128.0, 127.9, 127.9, 127.8, 127.7, 127.7, 127.6, 127.6, 127.6, 127.5 127.4, 127.4, 127.4, 127.3, 127.3 (Ph), 100.4 (C-1), 99.5 (C-1'), 98.9 (C-1''), 84.3, 83.6, 79.9, 79.5, 78.2, 75.1, 74.5, 74.4, 74.2, 73.4, 72.4, 72.1, 72.0, 71.9, 71.8, 69.9, 69.0, 68.8, 68.7, 68.0 (ring and benzylic C, octyl OCH_2), 31.8, 29.5, 29.4, 29.3, 26.2, 22.7 (octyl CH_2), 21.2 (OCO*C*H$_3$), 14.1 (octyl CH_3). Anal. Calcd for $C_{83}H_{96}O_{16}$ (1349.66): C, 73.86; H, 7.17. Found: C, 73.73; H, 7.04. HR-MALDI-MS calcd for $C_{83}H_{96}O_{16}$ $[M + Na]^+$ 1371.6626, found 1371.6596.

Octyl α-D-mannopyranosyl-(1→2)-α-D-mannopyranosyl-(1→5)-β-D-arabinofuranoside (**5**). A solution of **19** (110 mg, 0.23 mmol) was dissolved in methanol (20 mL) and then 2 drops of 1M NaOMe was added. The mixture was allowed to stir overnight and then concentrated to a syrup, which was purified by chromatography (4:1 hexane:EtOAc) to give **20** (100 mg, 96%) as a syrup. ^1H NMR (300 MHz, $CDCl_3$) δ_H 7.19–7.33 (m, 40 H, Ph), 5.14 (d, 1 H, $J_{1'',2''}$ = 1.6 Hz, H-1''), 4.94 (d, 1 H, $J_{1',2'}$ = 1.6 Hz, H-1'), 4.41–4.86 (m 17 H, H-1, 16 x C*H*$_2$Ph), 3.98–4.10 (m, 4 H), 3.60–4.15 (m, 17 H, H-2, H-3, H-4, H-5a, H-5b, H-2', H-3', H-4', H-5', H-6'a, H-2'', H-3'', H-4'', H-5'', H-6''a, H-6''b, octyl OCH_2), 3.47 (dd, 1 H, $J_{5',6'b}$ = 5.6, $J_{6'a,6'b}$ = 10.0 Hz, H-6'b), 3.29 (dt, 1 H, *J* = 6.9, 9.8 Hz, octyl OCH_2), 1.56–1.64 (m, 2 H, octyl CH_2), 1.21–1.29 (m, 10 H, octyl CH_2), 0.88 (t, 3 H, *J* = 6.9 Hz, octyl CH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 170.1 (C=O), 138.5, 138.4, 138.2, 138.1, 138.0, 137.9, 137.7, 137.6, 128.3, 128.3, 128.2, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7, 127.7, 127.6, 127.6, 127.5, 127.4, 127.3, 127.2, 127.2, 127.1 (Ph), 101.0 (C-1), 100.3 (C-1'), 98.9 (C-1''), 84.2, 83.5, 79.9, 79.8, 79.4, 74.9, 74.8, 74.7, 74.6, 74.5, 74.3, 74.2, 73.3, 72.2, 72.0, 71.5, 71.8, 69.9, 69.0, 68.4, 67.9 (ring and benzylic C, octyl OCH_2), 31.7, 29.4, 29.3, 29.2, 26.0, 22.6 (octyl CH_2), 14.0 (octyl CH_3).

To a solution of **20** (100 mg, 0.11 mmol) in methanol (8 mL) was added 10% Pd/C (25 mg). The mixture was stirred overnight under an H_2 atmosphere and the catalyst was separated by filtration and washed with CH_2Cl_2 (10 mL). After concentrating, the product was purified by chromatography (2:1 CH_2Cl_2:CH_3OH) on Iatrobeads to obtain **5** (38 mg, 93%) as a foam. R_f 0.29 (2:1 CH_2Cl_2:CH_3OH); $[\alpha]_D$ +5.2° (*c* 0.9, H_2O); ^1H

NMR (800 MHz, D_2O) δ_H 5.08 (s, 1 H, H-1′), 4.96 (s, 1 H, H-1″), 4.93 (d, 1 H, $J_{1,2}$ = 4.5 Hz, H-1), 4.05 (dd, 1 H, $J_{2,3}$ = 4.6, $J_{3,4}$ = 8.0 Hz, H-3), 4.00–4.02 (m, 2 H, H-2, H-2″), 3.93 (ddd, 1 H, $J_{3,4}$ = 8.0, $J_{4,5a}$ = 3.3, $J_{4,5b}$ = 6.9 Hz, H-4), 3.88–3.91 (m, 2 H, H-2′, H-3′), 3.80–3.83 (m, 2 H, H-6′a, H-6″a), 3.79 (dd, 1 H, $J_{2'',3''}$ = 3.3, $J_{3'',4''}$ = 9.6 Hz, H-3″), 3.63–3.75 (m, 5 H, H-5a, H-4′, H-5″, H-6′b, H-6″b), 3.54–3.61 (m, 4 H, H-5b, H-5′, H-4″, octyl OCH_2), 3.44 (dt, 1 H, J = 7.0, 9.7 Hz, octyl OCH_2), 1.58–1.65 (m, 2 H, octyl CH_2), 1.23–1.29 (m, 10 H, octyl CH_2), 0.87 (t, 3 H, J = 6.1 Hz, octyl CH_3); ^{13}C NMR (125.8 MHz, D_2O) δ_C 102.8 (C-1′, $^1J_{C,H}$ = 170.7 Hz), 101.4 (C-1, $^1J_{C,H}$ = 174.4 Hz), 98.7 (C-1″, $^1J_{C,H}$ = 171.8 Hz), 80.0 (C-4), 79.2 (C-2′), 76.6 (C-3), 75.1 (C-2), 73.5 (C-5″), 73.3 (C-5′), 70.8 (C-3″), 70.6 (C-3′), 70.4 (C-2″), 69.1 (C-5), 69.0 (octyl OCH_2), 67.3 (C-4′), 67.2 (C-4″), 61.6, 61.2 (C-6′, C-6″), 31.6, 29.2, 29.0, 29.0, 25.8, 22.5 (octyl CH_2), 13.9 (octyl CH_3). HR-FAB-MS calcd for $C_{25}H_{46}O_{15}$ $[M + Na]^+$ 609.2734, found 609.2717.

Octyl 2,3,4,6-Tetra-O-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-O-benzyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-O-benzyl-α-D-mannopyranosyl-(1→5)-2,3-di-O-benzyl-β-D-arabinofuranoside (**21**). Thioglycoside **8** (35 mg, 0.09 mmol), trisaccharide **20** (80 mg, 0.06 mmol), and powdered 4 Å molecular sieves (120 mg) were placed in a round-bottom flask and dried overnight in vacuo. The above mixture was dissolved in dry CH_2Cl_2 (10 mL) and the mixture was cooled to -40 °C and stirred for 10 min. *N*-iodosuccinimide (20 mg, 0.09 mmol) was added and the reaction was stirred for 20 min before the addition of silver triflate (5 mg, 0.02 mmol). The reaction mixture was allowed to gradually warm to 0 °C and was then quenched by the addition of triethylamine (0.3 mL). The yellow solution was filtered, diluted with CH_2Cl_2 (15 mL), washed with a saturated $Na_2S_2O_3$ solution (2 x 15 mL) followed by brine (20 mL) and water (20 mL). After drying (Na_2SO_4), it was filtered and concentrated under vacuum to a light yellow syrup, which was purified by chromatography (85:15 hexane:EtOAc) to give **21** (75 mg, 76%) as a syrup. R_f 0.37 (2:1 hexane:EtOAc); $[\alpha]_D$ +11.4° (*c* 0.9, $CHCl_3$); ^1H NMR (300 MHz, $CDCl_3$) δ_H 7.19–7.34 (m, 40 H, Ph), 5.45–5.15 (m, 4 H, H-1‴, H-2‴, H-3‴, H-4‴), 4.95–4.40 (m, 19 H, H-1, H-1′, H-1″, 16 x C*H*$_2$Ph), 3.60–4.15 (m, 20 H, H-2, H-3, H-4, H-5a, H-5b, H-2′, H-3′, H-4′, H-5′, H-6′a, H-2″, H-3″, H-4″, H-5″, H-6″a, H-6″b, H-5‴, H-6‴a, H-6‴b, octyl OCH_2), 3.47 (dd, 1 H, $J_{5',6'b}$ = 5.6, $J_{6'a,6'b}$ = 10.0 Hz, H-6′b), 3.31 (dt, 1 H, J = 6.9, 9.8 Hz, octyl OCH_2), 2.11 (s, 3 H, OCO*C*H$_3$), 1.99 (s, 3 H, OCO*C*H$_3$), 1.98 (s, 3 H, OCO*C*H$_3$), 1.95 (s, 3 H, OCO*C*H$_3$), 1.50–1.57 (m, 2 H, octyl CH_2), 1.23–1.33 (m, 10 H, octyl CH_2), 0.87 (t, 3 H, J = 6.3 Hz, octyl CH_3); ^{13}C NMR (75.5 MHz, $CDCl_3$) δ_C 170.1, 169.8, 169.7, 169.6 (C=O), 138.5, 138.4, 138.4, 138.3, 138.1, 137.8, 128.9, 128.4, 128.3, 128.2, 128.1, 128.1, 128.0, 128.0, 127.9, 127.9, 127.7, 127.6, 127.5, (Ph), 100.5 (C-1‴), 100.3 (C-1″), 99.2 (C-1′), 98.9 (C-1), 84.3, 83.6, 79.8, 79.5, 79.3, 76.8, 75.2, 75.1, 75.0, 74.8, 74.7, 73.4, 73.3, 73.1, 72.5, 72.4, 72.3, 72.2, 72.1, 70.2, 69.6, 69.2, 69.1, 68.9, 68.0, 66.1 (ring and benzylic C, octyl OCH_2), 31.9, 29.5, 29.5, 29.3, 26.2, 22.7 (octyl CH_2), 20.9, 20.7, 20.6 (OCO*C*H$_3$), 14.1 (octyl CH_3). HR-MALDI-MS calcd for $C_{95}H_{112}O_{24}[M + Na]^+$ 1659.7441, found 1659.7504.

Octyl α-D-mannopyranosyl-(1→2)-α-D-mannopyranosyl-(1→2)-α-D-mannopyranosyl-(1→5)-β-D-arabinofuranoside (**6**). A solution of **21** (75 mg, 0.05 mmol) was dissolved in methanol (10 mL) and then 3 drops of 1M NaOMe was added. The mixture was allowed to stir overnight and then concentrated to a syrup, which was purified by chromatography (19:1 CH_2Cl_2:CH_3OH) to obtain **22** (66 mg, 90%). To a solution of **22** (66 mg, 0.045 mmol) in methanol (8 mL) was added 10% Pd/C (15 mg). The mixture was stirred overnight under an H_2 atmosphere and the catalyst was separated by filtration and washed with CH_2Cl_2 (10 mL). After

concentrating, the product was purified by chromatography (2:1 CH_2Cl_2:CH_3OH) on Iatrobeads to obtain **6** (29 mg, 78% from **21**) as a foam. R_f 0.25 (1:1 CH_2Cl_2:CH_3OH); $[\alpha]_D$ +15.4 ° (*c* 1.4, H_2O); ^1H NMR (800 MHz, D_2O) δ_H 5.22 (d, 1 H, $J_{1'',2''}$ = 1.8 Hz, H-1''), 5.06 (d, 1 H, $J_{1',2'}$ = 1.2 Hz, H-1'), 4.98 (d, 1 H, $J_{1''',2'''}$ = 1.8 Hz, H-1'''), 4.93 (d, 1 H, $J_{1,2}$ = 4.6 Hz, H-1), 4.03–4.07 (m, 2 H, H-2, H-2''), 3.99–4.02 (m, 2 H, H-3, H-2'''), 3.93 (ddd, 1 H, $J_{3,4}$ = 7.1, $J_{4,5a}$ = 3.0, $J_{4,5b}$ = 6.9 Hz, H-4), 3.87–3.90 (m, 3 H, H-2', H-3', H-3''), 3.81–3.84 (m, 3 H, H-6a', H-6a'', H-6a'''), 3.79 (dd, 1 H, $J_{2''',3'''}$ = 3.3, $J_{3''',4'''}$ = 9.6 Hz, H-3'''), 3.75-3.63 (m, 8 H, H-5a, H-4', H6b', H-4'', H-6b'', H-4''', H-6b''', octyl OCH_2), 3.53-3.61 (m, 4 H, H-5b, H-5', H-5'', H-5''') 3.42 (dt, 1 H, *J* = 6.6, 9.5 Hz, octyl OCH_2), 1.58–1.64 (m, 2 H, octyl CH_2), 1.22–1.29 (m, 10 H, octyl CH_2), 0.86 (t, 3 H, *J* = 6.3 Hz, octyl CH_3); ^{13}C NMR (125.8 MHz, D_2O) δ_C 102.7 (C-1''', $^1J_{C,H}$ = 169.3 Hz), 101.3 (C-1, $^1J_{C,H}$ = 174.5 Hz), 101.1 (C-1'', $^1J_{C,H}$ = 167.8 Hz), 98.7 (C-1', $^1J_{C,H}$ = 170.0 Hz), 80.0 (C-4), 79.3 (C-2'), 79.3 (C-2''), 76.5 (C-2), 75.0 (C-3), 73.6 (C-5''), 73.5 (C-5'''), 73.3 (C-5'), 70.8 (C-3'''), 70.6 (C-3''), 70.5 (C-2'''), 70.4 (C-3'), 69.1 (C-5), 69.0 (octyl OCH_2), 67.5 (C-4'''), 67.2 (C-4'), 67.2 (C-4''), 61.6 (C-6'''), 61.5 (C-6''), 61.2 (C-6'), 31.5, 29.1, 28.9, 28.8, 25.7, 22.4 (octyl CH_2), 13.8 (octyl CH_3). HR-FAB-MS calcd for $C_{31}H_{57}O_{20}$ $[M + Na]^+$ 749.3443, found 749.3470.

Acknowledgments

This work was supported by The Ohio State University. The authors thank Dr. Charles E. Cottrell for measuring the NMR spectra of **3–6**.

References

1. Dolin, P. J.; Raviglione, M. C.; Kochi, A. *Bull. World Health Org.* **1994**, *72*, 213.
2. Bloom, B. R.; Murray, C. J. L. *Science* **1992**, *257*, 1055.
3. Spinney, L. *New Scientist* March 30, 1996, 8.
4. Gilbert, G. L. *Med. J. Australia* **1996**, *154*, 121.
5. Blanchard, J. S. *Annu. Rev. Biochem.* **1996**, *65*, 215.
6. Connell, N. D.; Nikaido, H. In: Tuberculosis: Pathogenesis, Protection and Control; Bloom, B. R., Ed.; American Society for Microbiology, Washington DC, **1994**, 333.
7. Brennan, P. J.; Nikaido, H. *Annu. Rev. Biochem.* **1995**, *64*, 29.
8. Sibley, L. D.; Hunter, S. W.; Brennan, P. J.; Krahenbuhl, J. L. *Infect. Immun.* **1988**, *56*, 1232.
9. Roach, T. I. A.; Barton, C. H.; Chatterjee, D.; Blackwell, J. M. *J. Immun.* **1993**, *150*, 1886.
10. Yoshida, A.; Koide, Y. *Infect. Immun.* **1997**, *65*, 1953.
11. Zhang, Y.; Broser, M.; Cohen, H.; Bodkin, M.; Law, K.; Reibman, J.; Rom, W. M. *J. Clin. Invest.* **1995**, *95*, 586.
12. Chan, J.; Fan, X.; Hunter, S. W.; Brennan, P. J.; Bloom, B. R. *Infect. Immun.* **1991**, *59*, 1755.
13. Knutson, K. L.; Hmama, Z.; Herrera-Velit, P.; Rochford, R.; Reiner, N. E. *J. Biol. Chem.* **1998**, *273*, 645.
14. Chang, J. C.; Wysocki, A.; Tchou-Wong, K. M.; Moskowitz, N.; Zhang, Y.; Rom, W. N. *Thorax* **1996**, *51*, 306.
15. Chatterjee, D.; Khoo, K. H. *Glycobiology* **1998**, *8*, 113.

16. Besra, G. S.; Morehouse, C. B.; Rittner, C. M.; Waechter, C. J.; Brennan, P. J. *J. Biol. Chem.* **1997**, *272*, 18460.
17. Schlesinger, L. S.; Hull, S. R.; Kaufman, T. M. *J. Immunol.* **1994**, *52*, 4070.
18. Schlesinger, L. S.; Kaufman, T. M.; Iyer, S.; Hull, S. R.; Marchiando, L. K. *J. Immunol.* **1996**, *157*, 4568.
19. Garegg, P. J. *Adv. Carbohydr. Chem. Biochem.* **1997**, *52*, 179.
20. Palcic, M. M.; Heerze, L. D.; Pierce, M.; Hindsgaul, O. *Glycoconjugate J.* **1988**, *5*, 49.
21. Barresi, F.; Hindsgaul, O. *Front. Nat. Prod. Res.* **1996**, *1*, 251.
22. Dasgupta, F.; Garegg, P. J. *Acta Chem. Scand.* **1989**, *43*, 471.
23. Zhang, Y. M.; Mallet, J. M.; Sinaÿ, P. *Carbohydr Res.* **1992**, *236*, 73.
24. Glaudemans, C. P. J.; Fletcher Jr., H. G. *J. Am. Chem. Soc.* **1965**, *87*, 4636.
25. Krog-Jensen, C.; Oscarson, S. *J. Org. Chem.* **1998**, *63*, 1780.
26. Octyl α-D-arabinofuranoside (**16**) and octyl β-D-arabinofuranoside have identical chromatographic mobilities (Subramaniam, V.; Lowary, T. L. Unpublished results), and thus the formation of the undesired isomer would be readily identified by ^{1}H NMR analysis of the reaction products. None was detected.
27. Mizutani, K.; Kasai, R.; Nakamura, M.; Tanaka, O.; Matsuura, H. *Carbohydr. Res.* **1989**, *185*, 27.
28. Bock, K.; Pedersen, C. *J. Chem. Soc. Perkin Trans. 2* **1974**, 293.
29. Glaudemans, C. P. J.; Fletcher Jr., H. G. *J. Am. Chem. Soc.* **1965**, *87*, 2456.
30. Subramaniam, V.; Lowary, T. L. Unpublished results
31. Hasegawa, A.; Kiso, M.; Unger, F. M. Preparation of phosphorylated acyloligosaccharides as pharmaceuticals. German Patent DE 3730905, September 15, 1987.
32. Ratios were determined by integration of signals in the ^{1}H NMR spectra of the products; either the anomeric hydrogen resonances or those arising from the acetate methyl group could be used for this purpose. The samples used for these experiments were prepared by chromatographing the crude reaction mixtures and then pooling and concentrating all fractions containing either glycoside.
33. Commercially available from Pfanstiehl Laboratories.
34. Maryanoff, B.E.; Reitz, A. B.; Tutwiler, G. F.; Benkovic, S. J.; Benkovic, P. A.; Pilkis, S. J. *J. Am. Chem. Soc.* **1984**, *106*, 7851.