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Synthesis of a Tristearoyl Lipomannan via Preactivation-Based Iterative One-Pot Glycosylation

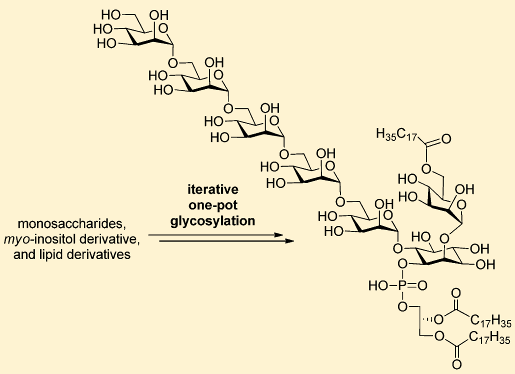
Jian Gao[†] and Zhongwu Guo*[,†,‡]

[†]Department of Chemistry, Wayne State University, Detroit, Michigan 48202, United Stated

[‡]National Glycoengineering Research Center, Shandong University, Jinan 250100, China

Supporting Information

ABSTRACT: A convergent and efficient strategy was developed for the synthesis of lipomannan (LM), useful for vaccine development. Thioglycosides were employed as glycosyl donors to construct two key pseudotrisaccharide and tetramannose intermediates through preactivation-based glycosylation strategy. These building blocks were then successfully coupled to form the LM core, which was lapidated, phospholipidated, and finally globally deprotected to afford the target molecule. The intermediate LM core involved in this synthesis contained orthogonal protections, which would facilitate its variable modifications for the preparation of other complex LM derivatives and for the synthesis of LM conjugates as LM-based vaccines.



■ INTRODUCTION

Mycobacterium tuberculosis (*Mtb*) is the causative pathogen of tuberculosis (TB), one of the most detrimental diseases worldwide, which causes more than two million deaths each year. A major virulent factor of *Mtb* is its cell envelope glycolipids, especially the phosphatidylinositol-anchored lipoglycans, such as lipoarabinomannans (LAMs) and lipomannans (LMs).[1,2] LMs have exhibited a variety of bioactivities, such as stimulating proinflammatory cytokine secretion through the toll-like receptor 2/CD14-dependent pathway and inducing macrophage apoptosis and IL-12 expression.[3−6] Due to the structural complexity and the intriguing immunoregulatory activities, LMs and related molecules have been the targets of a number of chemical total syntheses.[7−18]

In an effort to explore LM-derived vaccines, we have developed a highly convergent and efficient strategy for LM synthesis via preactivation-based iterative one-pot glycosylation using thioglycosides as glycosyl donors.[19,20] Many syntheses have proved that this strategy can save time and improve efficiency by abolishing multiple experimental preparation and intermediate separation steps. Our synthetic plan (Scheme 1) was to assemble the target molecule **1** from pseudotrisaccharide **2**, tetramannose **3**, and phosphoglycerolipid **4**. The key intermediate **3** was constructed from two monosaccharide building blocks **6** and **8** via iterative one-pot glycosylation, whereas the orthogonal allyl (All), *tert*-butyldimethylsilyl (TBS), and *p*-methoxybenzyl (PMB) protecting groups in **2** allowed for regioselective glycosylation, lipidation, and phospholipidation. Furthermore, the 2-*O*-positions of **6**, **7**, **8**, and **9** were protected as acetyl esters to safeguard α-selective glycosylation resulting from neighboring group participation.

■ RESULTS AND DISCUSSION

Mannosyl donors and acceptors **6**−**9** were prepared from **10**[21] and **12**[22] according to the procedures shown in Scheme 2. Stannylene acetal-directed benzylation of **10** was selective for the 3-*O*-position, which was followed by 2-*O*-acetylation and then regioselective benzylidene ring-opening in the presence of borane tetrahydrofuran complex (BH_3·THF) and trimethylsilyl trifluoromethanesulfonate (TMSOTf) to afford **6**. Thereafter, the free hydroxyl group in **6** was silylated with TBSCl under the influence of imidazole to obtain **7**. Compound **8** was prepared from orthoester **12** following perbenzylation and subsequent thioglycosylation using $SnCl_4$ as the promoter. En route to **9**, the 6-*O*-position of **12** was regioselectively protected with TBS first, followed by benzylation of the remaining free hydroxyl groups to produce **14**. Consecutively, the 6-*O*-TBS group was swapped for an allyl group to distinguish from **7**, via sequential desilylation mediated by tetrabutylammonium fluoride (TBAF) and reaction with allyl bromide and sodium hydride. Finally, reaction of **15** with *p*-thiocresol and $SnCl_4$ gave **9** which was derived from **12** in five steps and a 47% overall yield, involving only two column purification operations.

The assembly of pseudotrisaccharide **2** (Scheme 3), which had two mannose residues linked to the inositol 2-*O*- and 6-*O*-positions while the 1-*O*-position was uniquely protected to facilitate subsequently selective deprotection and phospholipidation, commenced from an optically pure 1,2,6-*O*-differentiated *myo*-inositol derivative **5** obtained from methyl α-D-glucopyranoside according to a reported procedure.[23] Glyco-

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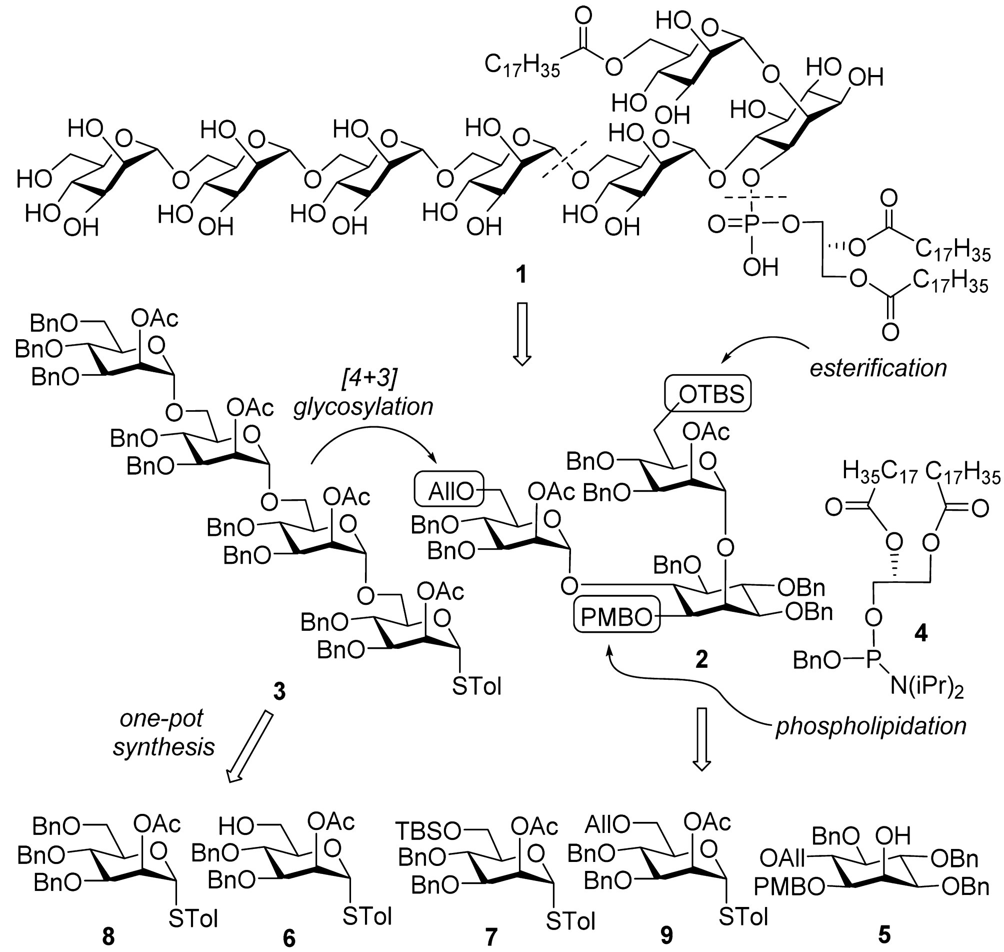
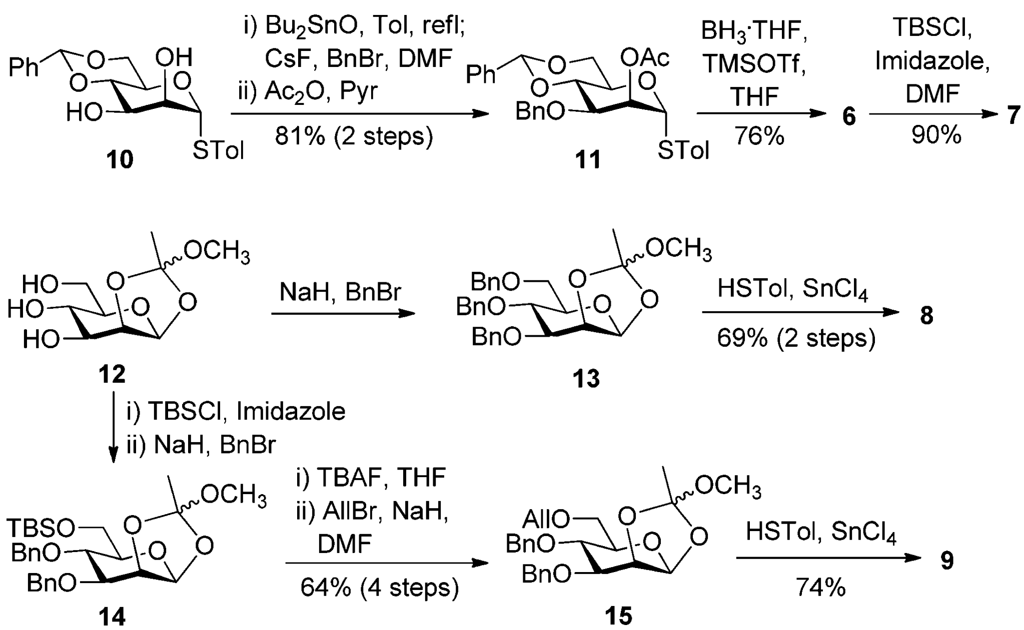
Scheme 1. Retrosynthesis of the Target Molecule 1

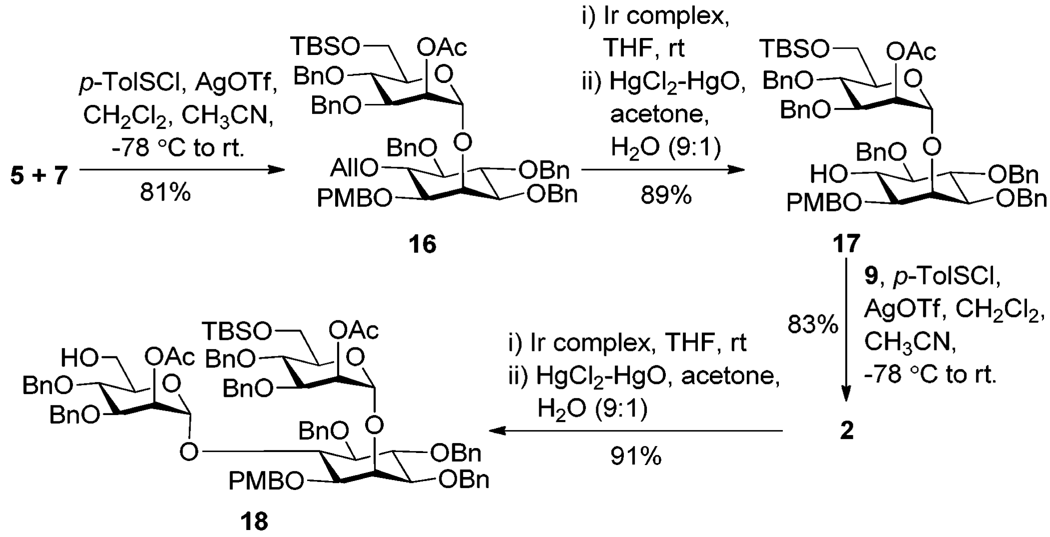


Scheme 2. Synthesis of Mannose Building Blocks 6−9

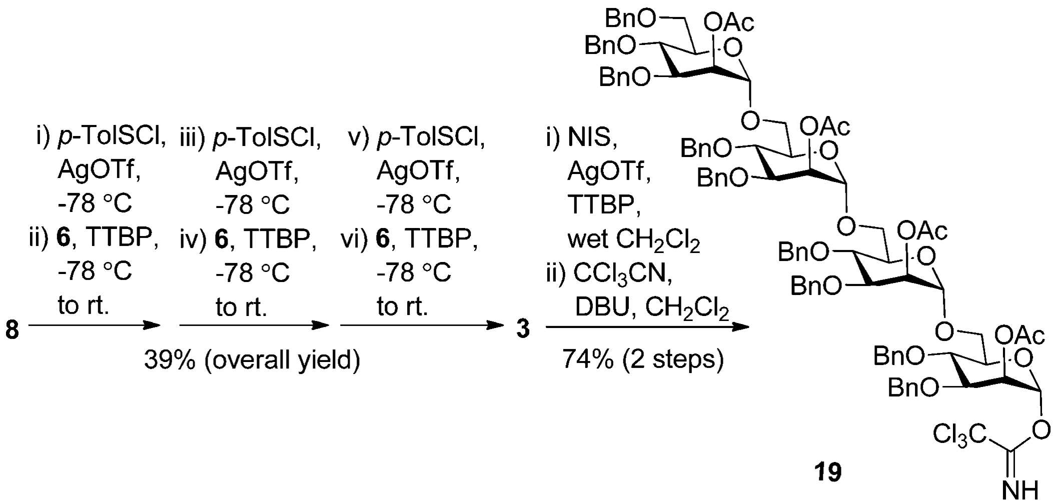


sylation of **5** with **7** under the influence of *p*-toluenesulfenyl triflate (*p*-TolSOTf) generated in situ from the reaction of *p*-toluenesulfenyl chloride (*p*-TolSCl) and silver triflate (AgOTf)[19,20] gave α-linked pseudodisaccharide **16** (81%) stereospecifically, as a result of neighboring acetyl group participation in the reaction. The allyl group on the inositol 6-*O*-position was thereafter removed by Iridium complex-catalyzed olefin rearrangement and then Hg(II)-catalyzed

Scheme 3. Synthesis of the Key Pseudotrisaccharide 2



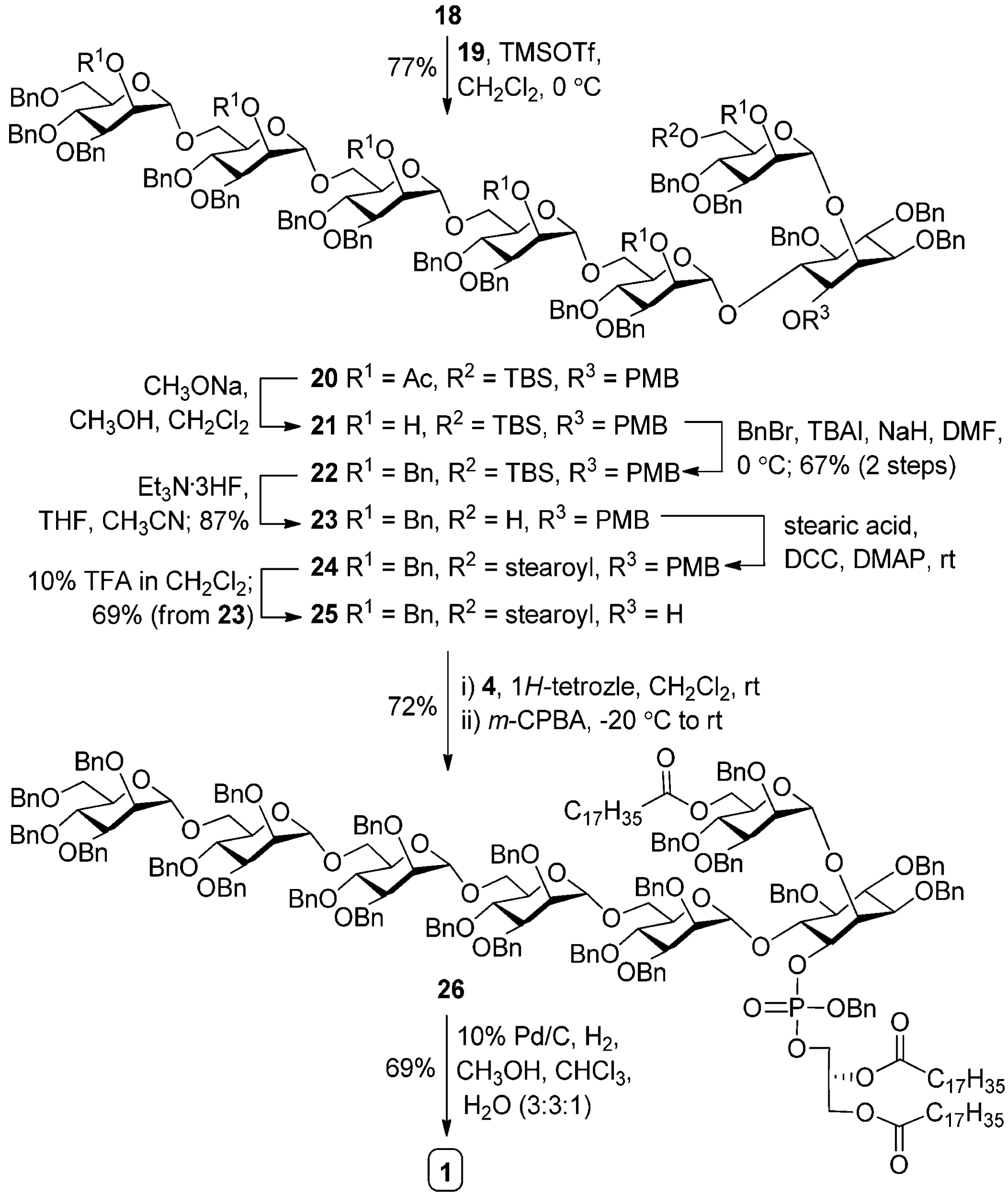
Scheme 4. Preactivation-Based Iterative One-Pot Synthesis of 3 and 19



hydrolysis[24] to form **17**. Mannosylation of **17** with **9** was again promoted by *p*-TolSOTf to give the desired **2** in an 83% yield. The anomeric C−H coupling constants ($^1J_{C,H}$ = 175 and 177 Hz) of **2** confirmed the *α*-stereochemistry of its glycosidic linkages.[25] Cleavage of the allyl protection by the Ir complex/Hg(II) method mentioned above finally afforded **18**, which was ready for further elongation of the glycan to get the target molecule or other LM derivatives.

The synthesis of tetramannose **3** via preactivation-based iterative one-pot glycosylation is outlined in Scheme 4. Preactivation of the thioglycosyl donors was achieved at −78 °C with *p*-TolSOTf as the promoter. Glycosylation reactions were furnished using a sterically hindered base, 2,4,6-tri-*tert*-butylpyrimidine (TTBP), as a scavenger for trifluoromethanesulfonic acid generated from the reactions. Each glycosylation was kept at room temperature for ca. 20 min to endorse complete reaction as shown by TLC. It is noteworthy that 1.0 equiv of *p*-TolSCl and 0.9 equiv of **6** (relative to the donors) were used in the reactions to further ensure complete consumption of the glycosyl acceptor in each step so as to minimize any potential interference with the reactions followed. Clearly, iterative one-pot glycosylation for oligosaccharide synthesis could improve the synthetic efficiency by obviating some time-consuming purification processes.[26−28] Thus, after three sequential glycosylation steps, **3** was obtained in 6 h and a 39% overall yield, giving an average of more than 73% yield for each glycosylation step. All of the reactions were proved *α*-specific (anomeric $^1J_{C,H}$ values of **3** were between 169 and 176 Hz). Although **3** could be directly utilized as a glycosyl donor for the glycosylation of **18**, the reactions employing *p*-TolSCl/AgOTf/TTBP or NIS/AgOTf/TTBP as the promoters gave relatively low yields of the desired product with an orthoester as the main byproduct. Due to neighboring group participation, glycosylation reactions using 2-*O*-acylated donors, such as **3**, usually involve orthoesters as the reaction intermediates, which can be transformed into the desired glycosides in the presence of strong Lewis acids. However, **3** and **18** were both complex and sterically hindered, and the reaction condition was almost neutral, thus the reaction might have stopped at the orthoester stage. To avoid this problem, **3** was converted to trichloroacetimidate **19** via hydrolysis of the thioglycoside in the presence of NIS/AgOTf/TTBP and then reaction of the resultant hemiacetal with trichloroacetonitrile and 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU).

Scheme 5. Final Assembly of the Synthetic Target 1



Glycosylation of **18** with **19** in the presence of TMSOTf was α-specific to afford **20** in a 77% yield (Scheme 5). The ^{13}C NMR spectrum of **20** displayed six discrete carbon signals at δ 98.6, 98.3, 98.2, 98.1, 98.0, and 97.9 with $^{1}J_{C,H}$ values between 172 and 176 Hz. Upon *O*-deacetylation and benzylation, the acetyl groups in **20** were replaced with benzyl groups to provide **22**, which was ready for the installation of lipids. The purpose for the protecting group exchange was to avoid using bases for global deprotection later on, as the target molecule contained ester linkages that were rather sensitive to basic conditions. Treatment of **22** with $Et_3N{\cdot}3HF$ to remove TBS was followed by acylation of the exposed hydroxyl group with stearic acid under the influence of *N,N*′-dicyclohexylcarbodiimide (DCC) and 4-dimethylaminopyridine (DMAP) and, thereafter, cleavage of the PMB ether with 10% TFA in CH_2Cl_2 to generate **25**. Compound **25** was smoothly phospholipidated using a two-step one-pot protocol, including reaction with freshly prepared phosphoramidite **4** in the presence of 1*H*-tetrazole and oxidation of resultant phosphite intermediate with *m*-chloroperoxybenzoic acid (*m*-CPBA) to afford **26** (72%) as a 1:3 diastereomeric mixture, originating from the stereogenic phosphorus atom. Global debenzylation of **26** was achieved in a mixture of chloroform, methanol, and water (3:3:1) under a H_2 atmosphere with 10% Pd/C as the catalyst to eventually yield the synthetic target **1**, which was characterized with ^{1}H and ^{31}P NMR spectroscopy and MALDI-TOF MS.

■ CONCLUSIONS

In summary, an efficient and convergent strategy was developed for the synthesis of LMs. It is highlighted by the construction of tetramannose **3** through preactivation-based iterative one-pot glycosylation, which has markedly reduced the number of synthetic and purification steps as compared to the reported syntheses[7−18] and thus improved the synthetic efficiency. Furthermore, taking advantage of neighboring group participation, all of the glycosylation reactions involved in the synthesis were stereoselective to form α-glycosidic linkages. The synthetic strategy reported here can be generally applicable to the preparation of various LM derivatives and conjugates. For example, intermediate **22**, which contained orthogonal TBS and PMB protecting groups, can be selectively deprotected to facilitate regioselective introduction of linkers for the conjugation with proteins or other carrier molecules to formulate LM-based vaccines or introduction of various lipids or phospholipids at these positions to obtain different LM derivatives. Furthermore, using differently protected monosaccharides from **8** as starting materials for Scheme 4, intermediates suitable for further elongation of the carbohydrate chain of the LM skeleton can be obtained for the synthesis of more complex LM molecules.

■ EXPERIMENTAL SECTION

General Experimental Methods. Chemicals and materials were obtained from commercial sources and were used as received without further purification unless otherwise noted. MS 4 Å was flame-dried under high vacuum and used immediately after cooling under a N_2 atmosphere. Analytical TLC was carried out on silica gel 60 Å F_{254} plates with detection by a UV detector and/or by charring with 15% (v/v) H_2SO_4 in EtOH. NMR spectra were recorded on a 400, 500, or 600 MHz machine with chemical shifts reported in ppm (δ) downfield from tetramethylsilane (TMS), which was used as an internal reference.

*p-Tolyl 2-O-Acetyl-3-O-benzyl-4,6-O-benzylidene-1-thio-α-D-mannopyranoside (**11**).*[29] The mixture of diol **10** (2.0 g, 5.35 mmol) and Bu_2SnO (1.6 g, 6.42 mmol) in toluene (80 mL) was refluxed under a N_2 atmosphere for 6 h. After the reaction was cooled to room temperature, toluene was removed under vacuum. The residue was dissolved in 20 mL of anhydrous DMF and mixed with CsF (2.4 g, 16.05 mmol) and BnBr (960 μL, 8.03 mmol). The mixture was stirred at room temperature for 24 h, at the end of which time TLC indicated the complete reaction. The solution was diluted with EtOAc (300 mL) and washed with saturated aq NaCl. The organic layer was dried with Na_2SO_4 and evaporated. The residue was dried under high vacuum for 1 h and then dissolved in 15 mL of pyridine and 2 mL of acetic anhydride. After 1 h of reaction, the mixture was coevaporated with toluene, and the residue was finally purified by silica gel column chromatography with a 1:7 mixture of EtOAc and hexane as eluent to give **11** (2.2 g, 81% for two steps) as a syrup. The following NMR data agreed well with that of the reported.[29] ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.52−7.49 (m, 2H, Ph), 7.40−7.24 (m, 10H, Ph), 7.12 (d, J = 8.4 Hz, 2H, Ph), 5.63 (s, 1H, PhCH), 5.61 (dd, J = 3.3, 1.0 Hz, 1H, H-2), 5.37 (d, J = 1.0 Hz, 1H, H-1), 4.70 (m, 2H, Bn), 4.36 (dt, J = 10.2, 4.8 Hz, 1H, H-5), 4.22 (dd, J = 10.2, 4.8 Hz, 1H, H-6), 4.12 (t, J = 10.2, 1H, H-4), 4.01 (dd, J = 10.2, 3.3 Hz, 1H, H-3), 3.85 (t, J = 10.2 Hz, 1H, H-6′), 2.32 (s, 3H, Tol), 2.14 (s, 3H, Ac). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 170.0, 138.3, 137.7, 137.4, 132.7, 129.9, 128.9, 128.3, 128.1, 127.7, 126.1, 101.6, 87.5 (C-1), 78.5, 74.1, 72.3, 71.3, 68.4, 65.1, 21.1, 21.0. MS (ESI-TOF) m/z: calcd for $C_{29}H_{30}O_6SNa$ [M + Na]$^+$ 529.1, found 528.8.

*p-Tolyl 2-O-Acetyl-3,4-di-O-benzyl-1-thio-α-D-mannopyranoside (**6**).* To a solution of **11** (2.0 g, 3.95 mmol) and MS 4 Å in 30 mL of anhydrous THF at −40 °C was added BH_3·THF (19.75 mL, 19.75 mmol) under a N_2 atmosphere. Fifteen minutes later, TMSOTf (924 μL, 5.14 mmol) was added dropwise to this solution. The mixture was stirred under these conditions for 1 h and then stirred at room temperature for another 24 h. The reaction was quenched with saturated aq $NaHCO_3$ at 0 °C. The solution was diluted with CH_2Cl_2 (400 mL) and washed with saturated aq $NaHCO_3$ and NaCl solutions. The organic layer was dried with Na_2SO_4 and concentrated; the residue was purified by silica gel column chromatography with a mixture of EtOAc and hexane (1:4) as the eluent to give **6** (76%, 1.53 g) as syrup. ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.40−7.29 (m, 12H, Ph), 7.13 (d, J = 8.0 Hz, 2H, Ph), 5.62 (dd, J = 3.2, 1.6 Hz, 1H, H-2), 5.40 (d, J = 1.6 Hz, 1H, H-1), 4.95 (d, J = 11.2 Hz, 1H, Bn), 4.75 (d, J = 11.7 Hz, 1H, Bn), 4.66 (d, J = 10.7 Hz, 1H, Bn), 4.59 (d, J = 11.2 Hz, 1H, Bn), 4.22 (dt, J = 9.6, 3.4 Hz, 1H, H-5), 3.98 (dd, J = 9.4, 3.2 Hz, 1H, H-3), 3.90 (t, J = 9.2 Hz, 1H, H-4), 3.80−3.85 (m, 2H, H-6,6′), 2.33 (s, 3H, Tol), 2.14 (s, 3H, Ac). ^{13}C NMR (100 MHz, $CDCl_3$) δ: 170.2, 138.3, 138.2, 137.6, 132.8, 129.9, 129.4, 128.5, 128.2, 128.0, 127.9, 127.8, 86.6 (C-1), 78.3, 75.3, 74.3, 72.9, 71.9, 70.3, 70.2, 62.0, 21.1. HR MS (ESI-TOF) m/z: calcd for $C_{29}H_{32}O_6SNa$ [M + Na]$^+$ 531.1817, found 531.1824. MS (ESI-TOF): found 530.9.

*p-Tolyl 2-O-Acetyl-3,4-di-O-benzyl-6-O-tert-butydimethylsilyl-1-thio-α-D-mannopyranoside (**7**).* To a solution of **6** (388 mg, 0.764 mmol) in anhydrous DMF (10 mL) were added imidazole (104 mg, 1.53 mmol) and TBSCl (192 mg, 1.15 mmol) at 0 °C. The mixture was stirred at room temperature for 2 h and then diluted with EtOAc (150 mL). The organic phase, after being washed with saturated aq NaCl solution, was dried over Na_2SO_4 and concentrated in vacuo, and the residue was purified by silica gel column chromatography with EtOAc and hexane (1:14) as the eluent to give **7** (427 mg, 90%) as a white solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.34−7.27 (m, 12H, Ph), 7.08 (d, J = 7.8 Hz, 2H, Ph), 5.54 (s, 1H, H-2), 5.37 (s, 1H, H-1), 4.90 (d, J = 10.8 Hz, 1H, Bn), 4.71 (d, J = 11.2 Hz, 1H, Bn), 4.64 (d, J = 10.8 Hz, 1H, Bn), 4.56 (d, J = 11.2 Hz, 1H, Bn), 4.10−4.12 (m, 1H, H-5), 3.95−3.91 (m, 3H, H-3,4,6), 3.81 (d, J = 11.4 Hz, 1H, H-6′), 2.30 (s, 3H, Tol), 2.09 (s, 3H, Ac), 0.89 (s, 9H, *t*Bu), 0.03 (s, 3H, SiMe), 0.04 (s, 3H, SiMe). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 170.2, 138.6, 137.7, 137.6, 132.1, 130.3, 129.7, 128.4, 128.3, 128.1, 127.9, 127.8, 127.6, 86.5 (C-1), 78.3, 75.2, 74.4, 73.8, 71.9, 70.6, 62.2, 60.0, 25.9, 21.0, 20.9, 18.3, −5.2, −5.4. HR MS (ESI-TOF) m/z: calcd for $C_{35}H_{46}O_6SSiNa$ [M + Na]$^+$ 645.2682, found 645.2689. MS (ESI-TOF): found, 644.9.

*p-Tolyl 2-O-Acetyl-3,4,6-tri-O-benzyl-1-thio-α-D-mannopyranoside (**8**).*[30] To a solution of **12** (410 mg, 1.74 mmol) in anhydrous DMF were added NaH (208 mg, 8.69 mmol) and BnBr (1.04 mL, 8.69 mmol) at 0 °C under N_2. Half an hour later, the reaction was quenched with saturated aq NaCl solution, and the mixture was diluted with EtOAc (150 mL). The organic phase, after being washed with saturated aq NaCl, was dried over Na_2SO_4, concentrated in vacuo and the residue was purified by silica gel column with EtOAc and hexane (1:8) as the eluent to give **13** as a pale yellow solid. Compound **13**, *p*-thiocresol (248 mg, 2.0 mmol), and activated 4 Å MS were mixed in 20 mL of anhydrous CH_2Cl_2. Then, a catalytic amount of $SnCl_4$ (300 μL, 0.3 mmol, 1 M in CH_2Cl_2) was added at 0 °C under N_2. The mixture was stirred at room temperature for 30 min, and TLC indicated the completion of the reaction. After the reaction was quenched with triethylamine, the solvent was evaporated in vacuum to give the crude oil, which was purified by silica gel column chromatography with EtOAc and hexane (1:10) as the eluent to give **8** (717 mg, 69% for two steps) as a white solid. The following NMR data agreed well with that of the reported.[30] ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.38−7.25 (m, 15H, Ph), 7.20 (d, J = 7.2 Hz, 2H, Ph), 7.05 (d, J = 7.2 Hz, 2H, Ph), 5.60 (d, J = 1.8 Hz, 1H, H-2), 5.46 (s, 1H, H-1), 4.89 (d, J = 10.8 Hz, 1H, Bn), 4.73 (d, J = 10.8 Hz, 1H, Bn), 4.66 (d, J = 12.0 Hz, 1H, Bn), 4.57 (d, J = 11.4 Hz, 1H, Bn), 4.51 (d, J = 10.8 Hz, 1H, Bn), 4.46 (d, J = 12.0 Hz, 1H, Bn), 4.34 (m, 1H, H-5), 3.96−3.93 (m, 2H, H-3,4), 3.85 (ddd, J = 10.8, 4.8, 1.8 Hz, 1H, H-6), 3.73 (d, J = 10.8 Hz, 1H, H-6′), 2.30 (s, 3H), 2.13 (s, 3H). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 170.3, 138.3, 138.2, 137.8, 137.6, 132.3, 129.9, 129.8, 128.4, 128.3, 128.2, 128.1, 127.8, 127.7, 127.6, 127.5, 86.5 (C-1), 78.5, 75.2, 74.6, 73.3, 72.4, 71.9, 70.3, 68.9, 21.07, 21.06. MS (ESI-TOF) m/z: calcd for $C_{36}H_{38}O_6SNa$ [M + Na]$^+$ 621.2, found 620.9.

*3,4-Di-O-benzyl-6-O-allyl-1,2-O-methoxyethylidene-β-D-mannopyranose (**15**).* To a solution of **12** (330 mg, 1.40 mmol) in anhydrous DMF (20 mL) was added imidazole (190 mg, 2.80 mmol) and TBSCl (252 mg, 1.68 mmol) at 0 °C. The mixture was stirred at room temperature for 2 h and then diluted with EtOAc (200 mL). The organic phase, after being washed with saturated aq NaCl solution, was dried over Na_2SO_4 and concentrated in vacuo to afford a residue that was dissolved in anhydrous DMF (10 mL) and then mixed with NaH (101 mg, 4.2 mmol) and BnBr (670 μL, 5.6 mmol) at 0 °C under N_2.

Half an hour later, the reaction was quenched with methanol, diluted with EtOAc (200 mL), and washed with saturated aq NaCl solution. The organic phase was dried over Na_2SO_4 and concentrated in vacuo to give the crude **14**. To a solution of crude **14** in THF (8 mL) was added TBAF (4.2 mL, 4.2 mmol, 1 M in THF). The mixture was stirred at room temperature for 3 h, at which time TLC indicated the completion of the reaction, and then diluted with EtOAc (200 mL). The organic phase, after being washed with saturated aq NaCl solution, was dried over Na_2SO_4, and concentrated in vacuo to afford a residue, which was dissolved in anhydrous DMF (10 mL) and mixed with NaH (67 mg, 2.8 mmol) and allyl bromide (242 μL, 2.8 mmol) at 0 °C under N_2. An hour later, the reaction was quenched with methanol, diluted with EtOAc (100 mL), and then washed with saturated aq NaCl solution. The organic phase was dried over Na_2SO_4 and concentrated in vacuo, and the product was purified by silica gel column chromatography with EtOAc and hexane (1:8) as the eluent to give **15** (408 mg, 64% for four steps) as a white solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.39−7.28 (m, 10H, Ph), 5.87 (m, 1H, All), 5.33 (d, *J* = 2.4 Hz, 1H, H-1), 5.26 (dd, *J* = 17.2, 1.8 Hz, 1H, All), 5.14 (dd, *J* = 10.2, 1.2 Hz, 1H, All), 4.92 (d, *J* = 10.8 Hz, 1H, Bn), 4.80−4.75 (m, 2H, Bn), 4.66 (d, *J* = 10.8 Hz, 1H, Bn), 4.38 (dd, *J* = 3.6, 2.4 Hz, 1H, H-2), 4.04 (dd, *J* = 13.2, 5.4 Hz, 1H, All), 3.99 (dd, *J* = 13.2, 5.4 Hz, 1H, All), 3.89 (t, *J* = 9.6 Hz, 1H, H-4), 3.72−3.63 (m, 3H, H-3,4,6′), 3.39−3.36 (m, 1H, H-5), 3.27 (s, 3H, OMe), 1.73 (s, 3H, Me). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 138.3, 137.8, 134.6, 128.5, 128.4, 128.0, 127.9, 127.7, 123.9, 116.6, 97.5 (C-1), 79.0, 77.1, 75.2, 74.2, 74.1, 72.4, 72.3, 68.9, 49.7, 24.3. HR MS (ESI-TOF) *m/z*: calcd for $C_{26}H_{32}O_7Na$ $[M + Na]^+$ 479.2046, found 479.2052. MS (ESI-TOF): found 478.9.

*p-Tolyl 2-O-Acetyl-3,4-di-O-benzyl-6-O-allyl-1-thio-α-D-mannopyranoside (**9**).* To a solution of **15** (400 mg, 0.877 mmol), *p*-thiocresol (131 mg, 1.05 mmol), and activated MS 4 Å in 15 mL of anhydrous CH_2Cl_2 was added $SnCl_4$ (176 μL, 0.176 mmol, 1 M in CH_2Cl_2) at 0 °C under N_2. The reaction mixture was stirred at room temperature for 30 min, at which time TLC indicated the completion of the reaction. The reaction was quenched with triethylamine, and the solvent was evaporated in vacuo. The residue was purified by silica gel column chromatography EtOAc and hexane (1:8) as the eluent to give **9** (344 mg, 74%) as a colorless syrup. ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.38−7.30 (m, 12H, Ph), 7.11 (d, *J* = 8.0 Hz, 2H, Ph), 5.98−5.88 (m, 1H, All), 5.61 (s, 1H, H-2), 5.47 (s, 1H, H-1), 5.30 (dd, *J* = 17.2, 1.2 Hz, 1H, All), 5.18 (d, *J* = 10.4 Hz, 1H, All), 4.95 (d, *J* = 10.8 Hz, 1H, Bn), 4.75 (d, *J* = 11.2 Hz, 1H, Bn), 4.63 (d, *J* = 10.8 Hz, 1H, Bn), 4.58 (d, *J* = 11.2 Hz, 1H, Bn), 4.32 (m, 1H, H-5), 4.11 (dd, *J* = 12.8, 5.2 Hz, 1H, All), 4.01−3.94 (m, 3H, H-3,4,All), 3.82 (dd, *J* = 10.8, 4.4 Hz, 1H, H-6), 3.69 (d, *J* = 10.8 Hz, 1H, H-6′), 2.33 (s, 3H, Tol), 2.16 (s, 3H, Ac). ^{13}C NMR (100 MHz, $CDCl_3$) δ: 170.3, 138.4, 137.8, 137.7, 134.7, 132.2, 129.9, 129.8, 128.5, 128.4, 128.2, 127.9, 127.7, 116.9, 86.6 (C-1), 78.5, 75.3, 74.6, 72.36, 72.34, 71.9, 70.4, 68.9, 21.1. HR MS (ESI-TOF) *m/z*: calcd for $C_{32}H_{36}O_6SNa$ $[M + Na]^+$ 571.2130, found 571.2137. MS (ESI-TOF): found 570.9.

Preactivation-Based Glycosylation (General Procedure A). After a solution of glycosyl donor (1.1 equiv) and acceptor (1.0 equiv) in anhydrous CH_2Cl_2 mixed with TTBP (1.0 equiv) and MS 4 Å was stirred at room temperature for 40 min, it was cooled to −78 °C. Then, a solution of AgOTf (3.3 equiv) in acetonitrile was added. Fifteen minutes later, *p*-TolSCl (1.1 equiv) was added dropwise. The reaction mixture was warmed to room temperature slowly within 1 h and stirred for another 20 min. The reaction was quenched with Et_3N, and the mixture was diluted with CH_2Cl_2 and filtered. The filtrate was concentrated in vacuum, and the resultant residue was purified by flash silica gel column chromatography with a mixture of EtOAc and toluene as eluent to afford the desired product.

Ir-Complex/Hg(II)-Catalyzed Removal of the Allyl Group (General Procedure B). The solution of $[Ir(COD)(PMePh_2)_2]PF_6$ (0.15 equiv) in anhydrous THF was stirred under a H_2 atmosphere at room temperature until the red color turned to pale yellow (in ca. 15 min). Then, H_2 was exchanged with argon three times before a solution of the allyl-protected intermediate (1.0 equiv) in anhydrous THF was added slowly. The reaction mixture was stirred at room temperature for 40 min, at which point TLC showed the complete reaction. The reaction mixture was concentrated in vacuum, and after the residue was dissolved in acetone and water (9:1, v/v), the solution was treated with $HgCl_2$ (5.0 equiv) and HgO (0.15 equiv). Ten minutes later, the solution was concentrated and the residue was purified by silica gel column chromatography to give the desired product.

*2-O-(2-O-Acetyl-3,4-di-O-benzyl-6-O-tert-butydimethylsilyl-α-D-mannopyranosyl)-1-O-(p-methoxybenzyl)-3,4,5-tri-O-benzyl-6-O-allyl-D-myo-inositol (**16**).* General procedure A was used to prepare **16** (279 mg, 81%) from **7** (213 mg, 0.342 mmol) and **5** (190 mg, 0.311 mmol). ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.35−7.19 (m, 27H), 6.86 (d, *J* = 8.4 Hz, 2H), 5.99−5.93 (m, 1H, All), 5.42 (s, 1H), 5.28 (dd, *J* = 17.4, 1.8 Hz, 1H, All), 5.20 (d, *J* = 1.2 Hz, 1H, Man H-1), 5.17 (dd, *J* = 10.8, 1.2 Hz, 1H, All), 4.86−4.53 (m, 12H), 4.38 (dd, *J* = 12.0, 6.0 Hz, 1H), 4.32 (dd, *J* = 12.0, 6.0 Hz, 1H), 4.27 (s, 1H), 3.98−3.92 (m, 3H), 3.81−3.76 (m, 5H), 3.67 (dd, *J* = 12.0, 2.4 Hz, 1H), 3.44 (d, *J* = 10.8 Hz, 1H), 3.37 (t, *J* = 9.6 Hz, 1H), 3.27 (m, 2H), 2.05 (s, 3H), 0.89 (s, 9H), 0.04 (s, 3H), 0.007 (s, 3H). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 169.9, 159.1, 139.1, 138.7, 138.6, 138.1, 138.0, 135.4, 130.3, 129.0, 128.3, 128.2, 128.1, 128.0, 127.9, 127.6, 127.4, 127.3, 127.1, 116.7, 113.7, 98.5 (Man C-1, J_{CH} = 177 Hz), 83.4, 81.1, 81.0, 80.3, 78.9, 77.6, 76.1, 75.7, 75.0, 74.6, 73.9, 72.37, 72.35, 72.0, 71.8, 68.8, 61.8, 55.2, 25.9, 20.9, 18.3, −5.1, −5.4. HR MS (ESI-TOF) *m/z*: calcd for $C_{66}H_{80}O_{13}SiNa$ $[M + Na]^+$ 1131.5266, found 1131.5243. MS (MALDI-TOF): found 1132.4.

*2-O-(2-O-Acetyl-3,4-di-O-benzyl-6-O-tert-butydimethylsilyl-α-D-mannopyranosyl)-1-O-(p-methoxybenzyl)-3,4,5-tri-O-benzyl-D-myo-inositol (**17**).* General procedure B was used to prepare **17** (197 mg, 89%) from **16** (230 mg, 0.208 mmol). ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.38−7.20 (m, 27H), 6.87 (d, *J* = 8.4 Hz, 2H), 5.40 (t, *J* = 1.6 Hz, 1H), 5.16 (d, *J* = 1.6 Hz, 1H, Man H-1), 4.89−4.51 (m, 12H), 4.32 (t, *J* = 2.0 Hz, 1H), 4.02−3.94 (m, 4H), 3.84−3.79 (m, 4H), 3.69 (dd, *J* = 11.6, 2.4 Hz, 1H), 3.46 (d, *J* = 10.8 Hz, 1H), 3.36−3.29 (m, 2H), 3.17 (dd, *J* = 9.6, 2.0 Hz, 1H), 2.07 (s, 3H), 0.89 (s, 9H, *t*Bu), 0.04 (s, 3H, SiMe), 0.01 (s, 3H, SiMe). ^{13}C NMR (100 MHz, $CDCl_3$) δ: 170.0, 159.4, 139.0, 138.7, 138.6, 138.1, 138.0, 129.7, 129.4, 128.5, 128.4, 128.3, 128.1, 128.0, 127.9, 127.7, 127.6, 127.5, 127.2, 113.9, 98.5 (Man C-1), 83.2, 80.9, 79.7, 79.1, 77.6, 75.7, 75.6, 75.1, 73.9, 72.9, 72.48, 72.44, 72.0, 71.9, 71.3, 68.9, 61.8, 55.3, 25.9, 21.0, 18.3, −5.0, −5.3. HR MS (ESI-TOF) *m/z*: calcd for $C_{63}H_{76}O_{13}SiNa$ $[M + Na]^+$ 1091.4953, found 1091.4956. MS (MALDI-TOF): found 1092.1.

*6-O-(2-O-Acetyl-3,4-di-O-benzyl-6-O-allyl-α-D-mannopyranosyl)-3,4,5-tri-O-benzyl-2-O-(2-O-acetyl-3,4-di-O-benzyl-6-O-tert-butydimethylsilyl-α-D-mannopyranosyl)-1-O-(p-methoxybenzyl)-D-myo-inositol (**2**).* General procedure A was used to parepare **2** (173 mg, 83%) from **9** (84 mg, 0.154 mmol) and **17** (150 mg, 0.140 mmol). ^{1}H NMR (500 MHz, $CDCl_3$) δ: 7.42−7.17 (m, 35H), 7.11 (t, *J* = 7.5 Hz, 2H), 6.88 (d, *J* = 8.5 Hz, 2H), 5.80 (m, 1H, H-All), 5.49 (m, 2H), 5.46 (s, 1H, Man^A H-1), 5.16 (m, 2H, Man^B H-1, H-All), 5.05 (d, *J* = 10.0 Hz, 1H, H-All), 4.95−4.48 (m, 16H), 4.32 (s, 1H), 4.08 (t, *J* = 9.5 Hz, 1H), 4.02−3.94 (m, 7H), 3.88 (t, *J* = 9.5 Hz, 1H), 3.82 (s, 3H), 3.75−3.70 (m 2H), 3.52 (d, *J* = 11.5 Hz, 1H), 3.36−3.24 (m, 5H), 2.10 (s, 6H), 0.92 (s, 9H), 0.06 (s, 3H), 0.02 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$) δ: 170.3, 169.7, 159.4, 139.2, 139.0, 138.4, 138.2, 138.1, 137.9, 134.9, 129.9, 129.2, 128.6, 128.3, 128.2, 128.1, 128.0, 127.9, 127.6, 127.5, 127.3, 127.2, 116.6, 113.8, 98.6 (Man^B C-1), 98.3 (Man^A C-1), 81.4, 80.9, 78.9, 78.2, 77.4, 76.0, 75.7, 75.5, 75.06, 75.00, 74.1, 73.8, 72.6, 72.4, 72.3, 71.7, 71.6, 71.3, 70.6, 68.6, 68.5, 68.1, 61.9, 55.2, 25.9, 21.2, 21.0, 18.3, −5.1, −5.3. HR MS (ESI-TOF) *m/z*: calcd for $C_{88}H_{104}O_{19}SiNa$ $[M + Na]^+$ 1515.6839, found 1515.6865. MS (MALDI-TOF): found 1516.6.

*6-O-(2-O-Acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)-3,4,5-tri-O-benzyl-2-O-(2-O-acetyl-3,4-di-O-benzyl-6-O-tert-butydimethylsilyl-α-D-mannopyranosyl)-1-O-(p-methoxybenzyl)-D-myo-inositol (**18**).* General procedure B was used to prepare **18** (124 mg, 91%) from **2** (140 mg, 0.094 mmol). ^{1}H NMR (500 MHz, $CDCl_3$) δ: 7.40−7.19 (m, 35H), 7.10 (t, *J* = 7.5 Hz, 2H), 6.89 (d, *J* = 8.5 Hz, 2H), 5.49 (dd, *J* = 2.5, 1.0 Hz, 1H), 5.46−5.43 (m, 2H, Man^A H-1, Man^B H-2), 5.19 (d, *J* = 1.0 Hz, 1H, Man^B H-1), 4.92−4.47 (m, 16H), 4.35 (s, 1H), 4.10 (t, *J* = 9.5 Hz, 1H), 4.02−3.91 (m, 5H), 3.87 (t, *J* = 9.5 Hz, 1H), 3.83−3.78 (m, 4H), 3.76 (dd, *J* = 11.0, 2.0 Hz, 1H), 3.55 (d, *J* =

11.5 Hz, 1H), 3.49 (dd, J = 12.0, 2.0 Hz, 1H), 3.40 (dd, J = 12.0, 3.0 Hz, 1H), 3.35−3.27 (m, 3H), 2.11 (s, 3H), 2.09 (s, 3H), 0.92 (s, 9H), 0.08 (s, 3H), 0.04 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$) δ: 170.2, 170.1, 159.4, 138.9, 138.8, 138.4, 138.1, 138.0, 137.8, 130.0, 129.1, 128.4, 128.3, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.5, 127.4, 127.3, 113.8, 98.2 (Man^B C-1), 98.0 (Man^A C-1), 81.5, 81.4, 80.9, 78.8, 77.9, 76.2, 75.7, 75.1, 74.0, 73.9, 72.6, 72.5, 71.6, 71.5, 70.0, 68.7, 68.6, 61.9, 61.4, 55.2, 25.9, 21.1, 21.0, 18.3, −5.1, −5.3. HR MS (ESI-TOF) m/z: calcd for $C_{85}H_{100}O_{19}SiNa$ [M + Na]$^+$ 1475.6526, found 1475.6366. MS (MALDI-TOF): found 1476.4.

*p-Tolyl (2-O-Acetyl-3,4,6-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2-O-acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2-O-acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)-(1→6)-3,4-di-O-benzyl-2-O-acetyl-α-D-mannopyranoside (**3**).* A mixture of **8** (370 mg, 0.619 mmol) and activated MS 4 Å in anhydrous CH_2Cl_2 (8 mL) was stirred at room temperature for 40 min and then cooled to −78 °C. A solution of AgOTf (477 mg, 1.856 mmol) in acetonitrile (1.5 mL) was added. After 10 min of stirring, p-TolSCl (89 μL, 0.619 mmol) was added dropwise. Fifteen minutes later, a solution of **6** (286 mg, 0.563 mmol) and TTBP (140 mg, 0.563 mmol) in anhydrous CH_2Cl_2 (1.5 mL) was added. The reaction mixture was warmed to room temperature slowly over 1 h, stirred for another 20 min, and then cooled to −78 °C, which was followed by the same sequence of addition of AgOTf (434 mg, 1.689 mmol) in acetonitrile (1 mL), p-TolSCl (81 μL, 0.563 mmol), a solution of **6** (260 mg, 0.512 mmol) and TTBP (127 mg, 0.512 mmol) in anhydrous CH_2Cl_2 (1.5 mL). The reaction mixture was warmed to room temperature slowly in 1 h, stirred for another 20 min, and then cooled to −78 °C, and again was followed by the same sequential addition of AgOTf (395 mg, 1.536 mmol) in acetonitrile (1 mL), p-TolSCl (74 μL, 0.512 mmol), a solution of **6** (236 mg, 0.465 mmol) and TTBP (115 mg, 0.465 mmol) in anhydrous CH_2Cl_2 (1.5 mL). The reaction mixture was warmed to room temperature slowly in 1 h, stirred for another 20 min, and then quenched with Et_3N, diluted with CH_2Cl_2, and finally filtered. The filtrate was concentrated in vacuum, and the residue purified by silica gel column chromatography with EtOAc and toluene (1:12) as the eluent to give **3** (317 mg, 39% for three glycosylation steps) as a foamy solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.35−7.08 (m, 49H), 5.61 (dd, J = 2.5, 1.2 Hz, 1H), 5.47 (m, 2H), 5.46 (dd, J = 2.5, 1.2 Hz, 1H), 5.38 (s, 1H, Man^D C-1), 4.98 (s, 1H, Man^C H-1), 4.93−4.82 (m, 6H, Man^A H-1, Man^B H-1, 4 × Bn-CH_2-), 4.74−4.39 (m, 14H), 4.31 (dd, J = 10.2, 4.8 Hz, 1H), 3.98 (dd, J = 9.6, 3.0 Hz, 1H), 3.96−3.80 (m, 8H), 3.77 (dd, J = 11.4, 3.0 Hz, 1H), 3.74−3.68 (m, 2H), 3.68−3.58 (m, 4H), 3.57−3.50 (m, 3H), 2.19 (s, 3H), 2.15 (s, 3H), 2.14 (s, 6H), 2.13 (s, 3H). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 170.3, 170.2, 170.19, 170.18, 138.5, 138.4, 138.2, 137.9, 137.7, 137.6, 137.5, 132.0, 129.9, 128.5, 128.4, 128.3, 128.2, 127.9, 127.8, 127.7, 127.6, 127.5, 127.4, 127.3, 98.0 (Man^C C-1), 97.98 (Man^A C-1), 97.97 (Man^B C-1), 86.6 (Man^D C-1), 78.5, 77.9, 77.7, 77.6, 75.2, 75.1, 75.0, 74.9, 74.3, 74.1, 73.8, 73.7, 73.4, 72.1, 71.8, 71.6, 71.4, 71.3, 71.15, 71.11, 70.2, 68.6, 68.2, 68.1, 68.0, 66.3, 65.5, 65.3, 29.7, 21.1, 21.04, 21.03, 21.00. HR MS (ESI-TOF) m/z: calcd for $C_{102}H_{110}O_{24}SNa$ [M + Na]$^+$ 1773.7005, found 1773.7000. MS (MALDI-TOF): found 1774.6.

*6-O-[(2-O-Acetyl-3,4,6-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2-O-acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2-O-acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)]-3,4-di-O-benzyl-2-O-acetyl-α-D-mannopyranosyl Trichloroacetimidate (**19**).* To a solution of **3** (310 mg, 0.177 mmol) and TTBP (132 mg, 0.532 mmol) in wet CH_2Cl_2 were added N-iodosuccinimide (80 mg, 0.354 mmol) and silver triflate (91 mg, 0.354 mmol) at 0 °C. The reaction mixture was stirred at room temperature for 2 h, quenched with saturated aq $Na_2S_2O_3$ solution, and diluted with CH_2Cl_2 (150 mL). The organic phase, after being washed with saturated aq NaCl solution and H_2O, was dried over Na_2SO_4 and concentrated in vacuo to afford a residue that was purified with silica gel column chromatography with EtOAc and toluene (1:5) as the eluent to give a white solid. It was dissolved in 10 mL of anhydrous CH_2Cl_2, and then DBU (14 μL, 0.089 mmol) and trichloroacetonitrile (91 μL, 0.89 mmol) were added at 0 °C. After 1.5 h of stirring, the mixture was concentrated in vacuum, and the residue was purified on a Et_3N-neutralized silica gel column with EtOAc and toluene (1:8) as the eluent to give **19** (234 mg, 74% for two steps) as a solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ: 8.67 (s, 1H), 7.32−7.07 (m, 45H), 6.19 (d, J = 1.8 Hz, 1H), 5.48 (dd, J = 3.0, 1.8 Hz, 1H), 5.46 (dd, J = 3.0, 1.8 Hz, 1H), 5.45 (dd, J = 3.0, 1.8 Hz, 1H), 5.41 (dd, J = 3.0, 1.8 Hz, 1H), 4.98 (d, J = 1.2 Hz, 1H, Man H-1), 4.91−4.38 (m, 20H, 3 × Man H-1, 17 × Bn-CH_2-), 4.02 (dd, J = 9.0, 3.0 Hz, 1H), 3.94−3.87 (m, 4H), 3.86−3.86 (m, 9H), 3.67 − 3.59 (m, 3H), 3.56 (d, J = 11.4 Hz, 2H), 3.51 (d, J = 10.8, 1.2 Hz, 1H), 2.19 (s, 3H), 2.15 (s, 3H), 2.13 (s, 3H), 2.12 (s, 3H). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 170.3, 170.2, 170.17, 170.15, 159.6, 138.4, 138.2, 137.9, 137.7, 137.6, 137.4, 128.4, 128.3, 128.2, 127.9, 127.8, 127.5, 127.4, 98.0 (Man C-1), 97.9 (Man C-1), 97.6 (Man C-1), 94.9 (Man C-1), 90.7, 77.8, 77.7, 77.6, 77.5, 75.3, 75.1, 74.97, 74.95, 74.1, 73.9, 73.76, 73.72, 73.4, 73.3, 72.0, 71.5, 71.45, 71.41, 71.13, 71.11, 68.6, 68.2, 68.1, 68.0, 67.2, 65.42, 65.40, 65.3, 21.1, 21.04, 21.03, 20.9.

*6-O-[(2-O-Acetyl-3,4,6-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2-O-acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2-O-acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2-O-acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2-O-acetyl-3,4-di-O-benzyl-α-D-mannopyranosyl)]-3,4,5-tri-O-benzyl-2-O-(2-O-acetyl-3,4-di-O-benzyl-6-O-tert-butyldimethylsilyl-α-D-mannopyranosyl)-1-O-(p-methoxybenzyl)-D-myo-inositol (**20**).* To a stirred mixture of **18** (60 mg, 0.041 mmol), **19** (96 mg, 0.054 mmol), and MS 4 Å in anhydrous CH_2Cl_2 (4 mL) was added TMSOTf (1 μL, 5.4 μmol) under N_2 protection at 0 °C. After the reaction mixture was stirred for another 30 min, it was neutralized with Et_3N, filtered, and concentrated. The residue was subjected to silica column chromatography with EtOAc and toluene (1:10) as the eluent to afford **20** (97 mg, 77%) as a white solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.38 (d, J = 7.8 Hz, 2H), 7.35−7.01 (m, 78H), 6.99 (d, J = 7.2 Hz, 2H), 6.84 (d, J = 7.2 Hz, 2H), 5.52 (s, 1H), 5.49 (m, 4H), 5.41 (s, 1H), 5.38 (s, 1H, Man^A H-1), 5.14 (s, 1H, Man^B H-1), 5.01 (d, J = 10.8 Hz, 1H), 4.97 (s, 1H, Man^C H-1), 4.93−4.20 (m, 36H, 3 × Man H-1, 33 × Bn-CH_2-), 4.01−3.69 (m, 21H), 3.64−3.58 (m, 3H), 3.51−3.37 (m, 7H), 3.31−3.22 (m, 6H), 3.15 (d, J = 12.0 Hz, 1H), 3.12 (d, J = 12.0 Hz, 1H), 2.14 (s, 3H), 2.11 (s, 3H), 2.10 (s, 9H), 2.05 (s, 3H), 0.88 (s, 9H), 0.03 (s, 3H), −0.003 (s, 3H). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 170.18, 170.13, 170.0, 169.9, 169.7, 159.4, 139.0, 138.9, 138.6, 138.5, 138.3, 138.2, 138.1, 138.0, 137.9, 137.8, 137.7, 137.5, 130.0, 129.0, 128.7, 128.5, 128.4, 128.3, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.5, 127.3, 127.2, 127.1, 126.9, 126.6, 113.8, 98.6 (Man^A C-1), 98.3 (Man^B C-1), 98.2 (Man C-1), 98.1 (Man^C C-1), 98.0 (Man C-1), 97.9 (Man C-1), 81.4, 81.3, 80.6, 78.8, 77.8, 77.7, 77.67, 77.61, 76.4, 75.7, 75.1, 74.9, 74.8, 74.6, 74.4, 74.0, 73.8, 73.6, 73.5, 73.4, 73.3, 72.6, 71.68, 71.65, 71.5, 71.44, 71.40, 71.3, 71.2, 72.1, 71.0, 70.8, 70.5, 69.9, 68.6, 68.5, 68.2, 67.9, 67.8, 67.7, 65.3, 65.2, 61.9, 55.2, 25.9, 21.1, 21.09, 21.03, 21.00, 18.3, −5.1, −5.4. HR MS (ESI-TOF) m/z: calcd for $C_{180}H_{202}O_{43}SiNa_2$ [M + 2Na]$^{2+}$ 1562.6593; found, 1562.6454. MS (MALDI-TOF) m/z: calcd for $C_{180}H_{202}O_{43}SiNa$ [M + Na]$^+$ 3102.3, found, 3103.2.

*6-O-[(2,3,4,6-Tetra-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)]-3,4,5-tri-O-benzyl-2-O-(2,3,4-tri-O-benzyl-6-O-tert-butyldimethylsilyl-α-D-mannopyranosyl)-1-O-(p-methoxybenzyl)-D-myo-inositol (**22**).* To a solution of **20** (90 mg, 0.029 mmol) in CH_2Cl_2 and CH_3OH (1:2, 9 mL) was added CH_3ONa solution (0.5 M in CH_3OH) until the pH reached 11. The solution was stirred at 35 °C for 24 h before the solvent was removed in vacuo. The residue was dissolved in a mixture of CH_2Cl_2 and CH_3OH (1:10) and filtered, and the filtrate was concentrated under vacuum to give crude **21**. To a solution of crude **21** in dry DMF were added BnBr (42 μL, 0.35 mmol) and TBAI (4.0 mg, 10.8 μmol). After the mixture was stirred for 10 min, NaH was added (5.6 mg, 0.232 mmol) at 0 °C and the resulting mixture stirred for 1.5 h. Then, MeOH was added to quench the reaction before water was added. The aqueous phase was extracted with CH_2Cl_2 (3 × 50 mL), and the organic layer was dried over Na_2SO_4 and concentrated. The residue was purified by silica gel column chromatography with EtOAc and toluene (1:16) as the eluent to give **22** (66 mg, 67% for two steps) as a syrup. ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.40−6.98 (m, 112H), 6.58 (d, J = 8.4 Hz, 2H), 5.39 (s, 1H, Man^A H-1), 5.29 (s, 1H, Man^F H-1), 5.09

(s, 1H, Man^E H-1), 5.02 (d, J = 11.4 Hz, 1H), 5.00 (s, 1H, Man^C H-1), 4.94 (s, 1H, Man^B H-1), 4.93−4.27 (m, 46H, Man^D H-1, 45 × Bn-CH_2-), 4.05 (t, J = 9.6 Hz, 1H), 4.03−3.78 (m, 21H), 3.73 (s, 1H), 3.68 (d, J = 9.6 Hz, 2H), 3.62−3.54 (m, 6H), 3.53−3.48 (m, 3H), 3.46−3.43 (m, 2H), 3.38 (d, J = 9.0 Hz, 1H), 3.35−3.25 (m, 6H), 3.17 (d, J = 11.4 Hz, 1H), 3.10 (d, J = 10.8 Hz, 1H), 0.85 (s, 9H), −0.002 (s, 3H), −0.01 (s, 3H). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 159.4, 139.1, 138.9, 138.8, 138.7, 138.6, 138.5, 138.3, 138.2, 138.1, 137.9, 129.5, 129.0, 128.5, 128.3, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.5, 127.4, 127.3, 127.2, 127.1, 126.9, 126.8, 113.9, 99.1 (Man^A C-1), 98.6 (Man^B C-1), 98.4 (Man^C C-1), 98.2 (Man^D C-1), 98.15 (Man^E C-1), 98.10 (Man^F C-1), 82.0, 81.4, 80.6, 79.6, 79.4, 79.3, 79.2, 79.1, 78.9, 76.0, 75.9, 75.7, 75.6, 75.0, 74.92, 74.90, 74.8, 74.7, 74.67, 74.62, 74.52, 74.50, 74.4, 74.2, 73.9, 73.8, 73.6, 73.2, 72.8, 72.63, 72.61, 72.5, 72.4, 72.3, 72.2, 72.1, 71.9, 71.8, 71.5, 71.4, 71.26, 71.22, 71.20, 71.1, 71.0, 69.0, 65.8, 65.7, 65.6, 62.2, 55.0, 53.4, 25.9, 18.3, −5.1, −5.4. HR MS (ESI-TOF) m/z: calcd for $C_{210}H_{226}O_{37}SiNa_2$ $[M + 2Na]^{2+}$ 1706.7684, found 1706.7743. MS (MALDI-TOF) m/z: calcd for $C_{210}H_{226}O_{37}SiNa$ $[M + Na]^+$ 3390.5, found 3291.2.

*6-O-[(2,3,4,6-Tetra-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)]-3,4,5-tri-O-benzyl-2-O-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-1-O-(p-methoxybenzyl)-D-myo-inositol (**23**).* A solution of **22** (60.0 mg, 0.018 mmol) in THF and CH_3CN (2:13, mL) and triethylamine trihydrofluoride (1.0 mL) was stirred at room temperature overnight under Ar. The solution was quenched with dropwise addition of saturated aq $NaHCO_3$ solution. The aq phase was extracted with CH_2Cl_2 (3 × 40 mL), and the organic layer was dried over Na_2SO_4, concentrated. The residue was purified by silica gel column chromatography with EtOAc and toluene (1:10) as the eluent to give **23** (50.4 mg, 87%) as syrup. ^{1}H NMR (600 MHz, $CDCl_3$) δ: 7.40−7.04 (m, 112H), 6.65 (dd, J = 7.8 Hz, 2H), 5.44 (d, J = 3.6 Hz, 1H, Man^A H-1), 5.21 (d, J = 4.2 Hz, 1H, Man^B H-1), 5.12 (d, J = 4.2 Hz, 1H, Man^E H-1), 5.07−5.04 (d, J = 10.8 Hz, 1H), 5.02 (d, J = 3.6 Hz, 1H, Man^D H-1), 4.96−4.85 (m, 8H, Man^C H-1, Man^F H-1, 6 × Bn-CH_2-), 4.77 (d, J = 10.8 Hz, 1H), 4.72−4.29 (m, 38H), 4.07−3.42 (m, 36H), 3.40 (d, J = 8.4 Hz, 1H), 3.36−3.25 (m, 6H), 3.18 (d, J = 10.8 Hz, 1H), 3.12 (d, J = 10.8 Hz, 1H), 2.11 (s, 1H). ^{13}C NMR (150 MHz,$CDCl_3$) δ: 159.4, 138.9, 138.8, 138.7, 138.6, 138.5, 138.4, 138.3, 138.2, 138.1, 138.0, 137.7, 129.2, 128.9, 128.5, 128.4, 128.3, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.5, 127.4, 127.3, 127.2, 127.1, 127.0, 126.8, 113.9, 99.1 (Man^A C-1), 99.0 (Man^B C-1), 98.7 (Man^C C-1), 98.4 (Man^D C-1), 98.3 (Man^E C-1), 98.2 (Man^F C-1), 81.8, 81.3, 80.7, 79.6, 79.4, 79.3, 79.2, 79.0, 78.7, 75.9, 75.8, 75.7, 75.1, 74.9, 74.8, 74.7, 74.6, 74.5, 74.2, 74.0, 73.9, 73.7, 73.2, 72.8, 72.6, 72.5, 72.4, 72.26, 72.21, 72.1, 72.0, 71.8, 71.6, 71.5, 71.3, 71.2, 71.1, 70.9, 69.1, 65.8, 65.7, 65.6, 62.1, 55.1. HR MS (ESI-TOF) m/z: calcd for $C_{204}H_{212}O_{37}Na_2$ $[M + 2Na]^{2+}$ 1649.7252; found, 1649.7317. MS (MALDI-TOF) m/z: calcd for $C_{204}H_{212}O_{37}Na$ $[M + Na]^+$ 3276.4, found 3277.1.

*6-O-[(2,3,4,6-Tetra-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)]-3,4,5-tri-O-benzyl-2-O-(2,3,4-tri-O-benzyl-6-O-stearoyl-α-D-mannopyranosyl)-D-myo-inositol (**25**).* To a solution of **23** (49.0 mg, 0.015 mmol) in anhydrous CH_2Cl_2 (2 mL) were added stearic acid (21.3 mg, 0.075 mmol), DCC (15.5 mg, 0.075 mmol), and DMAP (9.2 mg, 0.075 mmol) at room temperature. After having been stirred overnight, the mixture was filtered off through a Celite pad, and the filtrate was concentrated to get a residue that was purified by silica gel column chromatography with toluene and EtOAc (15:1) as the eluent to give **24** as a white solid. To a solution of **24** in CH_2Cl_2 (1.5 mL) was added 20% TFA/CH_2Cl_2, giving a final concentration of about 10% of TFA. The mixture was stirred for 3 h, at which time TLC indicated the completion of the reaction. The solution was coevaporated with toluene 3 times to remove TFA completely. Purification of the residue by silica gel column chromatography with EtOAc and toluene (1:12) as the eluent gave **25** (35.3 mg, 69% from **23**) as a white solid. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.31−7.01 (m, 110H), 5.36 (s, 1H, Man^A H-1), 5.09 (s, 1H, Man^B H-1), 5.04 (s, 1H, Man^F H-1), 4.96 (s, 2H, Man^C H-1, Man^D H-1), 4.85−4.73 (m, 8H, Man^E H-1, 7 × Bn-CH_2-), 4.65 (d, J = 10.8 Hz, 1H), 4.60−4.27 (m, 36H), 4.16 (s, 1H), 4.08 (d, J = 9.6 Hz, 1H), 4.05 (dd, J = 12.0, 3.6 Hz, 1H), 3.96−3.64 (m, 24H), 3.60 (d, J = 12.0, 2.4 Hz, 1H), 3.58−3.53 (m, 2H), 3.50−3.35 (m, 9H), 3.30−3.27 (m, 1H), 3.22 (dd, J = 9.6, 1.8 Hz, 1H), 3.12 (t, J = 9.6 Hz, 1H), 2.10 (t, J = 7.8 Hz, 2H), 1.48−1.41 (m, 2H), 1.24−1.12 (m, 28H), 0.80 (t, J = 7.2 Hz, 3H). ^{13}C NMR (150 MHz, $CDCl_3$) δ: 173.6, 138.8, 138.7, 138.6, 138.5, 138.4, 138.3, 138.2, 138.1, 138.0, 137.9, 137.6, 128.5, 128.4, 128.3, 128.2, 128.1, 127.9, 127.8, 127.7, 127.6, 127.5, 127.4, 127.3, 127.2, 98.6 (Man^A C-1), 98.5 (Man^B C-1, Man^C C-1, Man^D C-1), 98.4 (Man^E C-1), 98.2 (Man^F C-1), 81.2, 80.4, 80.3, 79.4, 79.3, 79.2, 78.8, 78.3, 75.5, 75.3, 75.2, 75.1, 75.0, 74.9, 74.87, 74.81, 74.7, 74.6, 74.3, 74.1, 74.0, 73.9, 73.2, 73.0, 72.67, 72.61, 72.5, 72.2, 72.0, 71.9, 71.8, 71.7, 71.67, 71,61, 71.3, 71.28, 71.26, 71.20, 70.1, 69.1, 66.5, 65.8, 65.7, 65.6, 63.1, 34.1, 31.9, 29.7, 29.65, 29.60, 29.5, 29.3, 29.25, 29.20,, 24.8, 22.7, 14.1. HR MS (ESI-TOF) m/z: calcd for $C_{214}H_{238}O_{37}Na_2$ $[M + 2Na]^{2+}$ 1722.8269; found, 1722.8318. MS (MALDI-TOF) m/z: calcd for $C_{214}H_{238}O_{37}Na$ $[M + Na]^+$ 3422.6, found 3423.0

*6-O-[(2,3,4,6-Tetra-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-O-benzyl-α-D-mannopyranosyl)]-3,4,5-tri-O-benzyl-2-O-(2,3,4-tri-O-benzyl-6-O-stearoyl-α-D-mannopyranosyl)-1-O-(1,2-di-O-stearoyl-sn-glycero-3-benzylphosphoryl)-D-myo-inositol (**26**).* To a mixture of **25** (16.0 mg, 4.7 μmol), freshly prepared glycerylphosphoramidite **4** (20.2 mg, 0.024 mmol), and MS 4 Å in CH_2Cl_2 and CH_3CN (2:1, 3 mL) was added 1*H*-tetrazole (0.45 M in CH_3CN, 105 μL, 0.047 mmol). After being stirred at room temperature under Ar for 40 min, the reaction mixture was cooled to −20 °C, and *m*-CPBA (4.1 mg, 0.024 mmol) was added. The reaction mixture was slowly warmed to room temperature in 1 h, and then quenched with saturated aq NaS_2O_3 solution. The aqueous layer was extracted with CH_2Cl_2 (3 × 30 mL), and the organic phase, after being dried over Na_2SO_4, was concentrated with the residue purified by silica gel column chromatography with EtOAc and toluene (1:13) as the eluent to give **26** (14.1 mg, 72%, mixture of two isomers in about 1:3 ratio) as a white solid. ^{1}H and ^{13}C NMR spectroscopic data for the major stereoisomer. ^{1}H NMR (600 MHz, $CDCl_3$) δ: 5.33 (s, 1H, Man^A H-1), 5.19 (s, 1H, Man^B H-1), 5.03 (s, 1H, Man^C H-1), 4.93 (s, 1H, Man^D H-1), 4.86 (s, 1H, Man^E H-1), 4.76 (s, 1H, Man^F H-1); ^{13}C NMR (150 MHz, $CDCl_3$) δ: 99.2 (Man^B C-1), 99.1 (Man^A C-1), 99.0 (Man^E C-1), 98.8 (Man^D C-1), 98.5 (Man^F C-1), 98.4 (Man^C C-1). ^{31}P NMR (162 MHz, $CDCl_3$) δ: −0.42 (minor isomer), −0.19 (major isomer). HR MS (ESI-TOF) m/z: calcd for $C_{260}H_{319}O_{44}PNa_2$ $[M + 2Na]^{2+}$ 2111.1129, found 2111.1270.

*6-O-[(α-D-Mannopyranosyl)-(1→6)-(α-D-mannopyranosyl)-(1→6)-(α-D-mannopyranosyl)-(1→6)-(α-D-mannopyranosyl)-(1→6)-(α-D-mannopyranosyl)]-2-O-[(6-O-stearoyl-α-D-mannopyranosyl)]-1-O-(1,2-di-O-stearoyl-sn-glycero-3-phosphoryl)-D-myo-inositol (**1**).* The mixture of **26** (10 mg, 2.4 μmol) and 10% Pd/C (15 mg) in $CHCl_3$, MeOH, and H_2O (3:3:1, 3 mL) was stirred under a 50 psi H_2 atmosphere for 3 days. The reaction solution was filtered off through a pad of Celite, and the filtrate was concentrated in vacuum to give the target compound **1** (3.5 mg, 69%) as a pale yellow solid. ^{1}H NMR (600 MHz, CD_3OD, $CDCl_3$, and D_2O 3:3:1) δ: 5.33 (s, 1H, Man^A H-1), 5.19 (s, 1H, Man^B H-1), 5.12 (s, 1H, Man^C H-1), 4.90 (s, 1H, Man^D H-1), 4.84 (s, 1H, Man^E H-1), 4.83 (s, 1H, Man^F H-1), 4.60−2.98 (m, 47H), 2.38−2.16 (m, 6H), 1.65−1.55 (m, 6H), 1.40−1.15 (m, 84H), 0.90−0.80 (m, 9H). ^{31}P NMR (162 MHz, $CD_3OD/CDCl_3/D_2O$ 3:3:1) δ: 0.54. MS (MALDI-TOF) m/z: calcd for $C_{99}H_{180}O_{44}PK_3$ $[M + 3K - H]^{2+}$ 1110.5, found 1109.2.

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

^{1}H, ^{13}C, ^{31}P, and ^{1}H−^{1}H gCOSY and ^{1}H−^{13}C gHMQC NMR spectra of all new compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

AUTHOR INFORMATION

Corresponding Author

*E-mail: zwguo@chem.wayne.edu.

Notes

The authors declare no competing financial interest.

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