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Synthetic 3-*O*-Methylmannose-Containing Polysaccharides (*s*MMPs): Design and Synthesis

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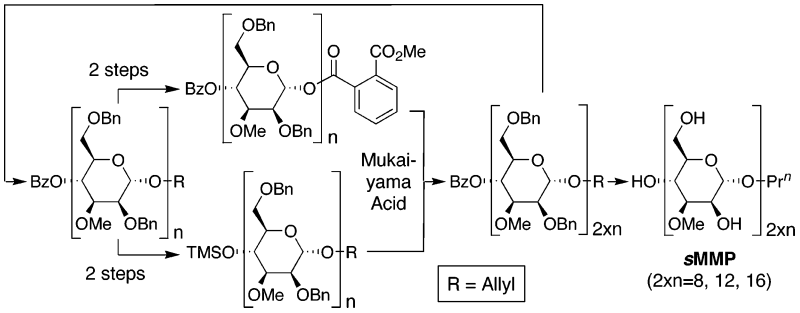

With the hope of mimicking the chemical and biological properties of *natural* 3-*O*-methylmannose-containing polysaccharides (MMPs), *synthetic* 3-*O*-methylmannose-containing polysaccharides (*s*MMPs) were designed and synthesized in a convergent manner. With little modification of the Mukaiyama glycosidation, high α-selectivity (>50:1∼>20:1) and yields (79∼74%) were achieved for the key glycosidation steps. The exceptionally high α-selectivity observed was shown to be consequent to the selective anomerization of β- to α-anomer under the glycosidation conditions. This glycosidation is well suited for a highly convergent oligosaccharide synthesis, particularly because of excellent chemical yields even when using approximately equal-sized donors and acceptors in an approximately 1:1 molar ratio. An iterative reaction sequence allowed the growing oligosaccharide to double in size after each cycle and led to an efficient synthesis of *s*MMP 8-, 12-, and 16-mers **18**−**20**.

Introduction

Extracts of *Mycobacterium smegmatis* cells provide two series of polysaccharides: the 3-*O*-methyl-D-mannose-containing polysaccharides (MMPs) and the 6-*O*-methyl-D-glucose-containing lipopolysaccharides (MGLPs) (Figure 1).[1] MMPs consist of 11−14 α-(1→4)-linked mannosides with most rings bearing a 3-*O*-methyl ether functionality.[2] The reducing end of the oligosaccharides is capped by a methyl aglycon. The four members of the MGLP family share the polysaccharide backbone whose reducing end is capped with a glyceric acid, but they differ in the number of lipophilic side chains attached to each molecule.[3] The side chains include acetate, propionate, isobutyrate, octanoate, and succinates, but the precise distribution of the acyl groups remains unknown. Saponification of each MGLP is known to yield the same 6-*O*-methyl-D-glucose-containing polysaccharide (MGP) that contains 11 internal, contiguous 6-*O*-methyl-D-glucose residues and a 3-*O*-methyl-D-glucose residue at the nonreducing terminus.[4]

(1) For reviews on MMP and MGLP/MGP, see: (a) Bloch, K. *Adv. Enzymol.* **1977**, *45*, 1. (b) Ballou, C. E. *Acc. Chem. Res.* **1968**, *1*, 366. (c) Ballou, C. E. *Pure Appl. Chem.* **1981**, *53*, 107.

(2) For isolation and structural characterization of MMP, see: (a) Gray, G. R.; Ballou, C. E. *J. Biol. Chem.* **1971**, *246*, 6835. (b) Maitra, S. K.; Ballou, C. E. *J. Biol. Chem.* **1977**, *252*, 2459. (c) Weisman, L. S.; Ballou, C. E. *J. Biol. Chem.* **1984**, *259*, 3457. (d) Weisman, L. S.; Ballou, C. E. *J. Biol. Chem.* **1984**, *259*, 3464. (e) Ilton, M.; Jevans, A. W.; McCarthy, E. D.; Vance, D.; White, H. B., III; Bloch, K. *Proc. Natl. Acad. Sci. U.S.A.* **1971**, *68*, 87.

(3) For isolation and structural characterization of MGLP/MGP, see: (a) Lee, Y. C.; Ballou, C. E. *J. Biol. Chem.* **1964**, *239*, PC3602. (b) Saier, M. H., Jr.; Ballou, C. E. *J. Biol. Chem.* **1968**, *243*, 4332. (c) Smith, W. L.; Ballou, C. E. *J. Biol. Chem.* **1973**, *248*, 7118. (d) Forsberg, L. S.; Dell, A.; Walton, D. J.; Ballou, C. E. *J. Biol. Chem.* **1982**, *257*, 3555. Regarding the structural heterogeneity of MGP, Ballou commented that at least two forms of MGP containing 21 hexoses exist: Kamisango, K.; Dell, A.; Ballou, C. E. *J. Biol. Chem.* **1987**, *262*, 4580. Also see: Tuffal, G.; Albigot, R.; Monsarrat, B.; Ponthus, C.; Picard, C.; Rivière, M.; Puzo, G. *J. Carbohydr. Chem.* **1995**, *14*, 631.

(4) The structure of MG(L)P shown in Figure 1 is the revised structure suggested by Rivière on the basis of the structure of polysaccharide isolated from *Mycobacterium bovis* BCG. See: Tuffal, G.; Albigot, R.; Rivière, M; Puzo, G. *Glycobiology* **1998**, *8*, 675−684.

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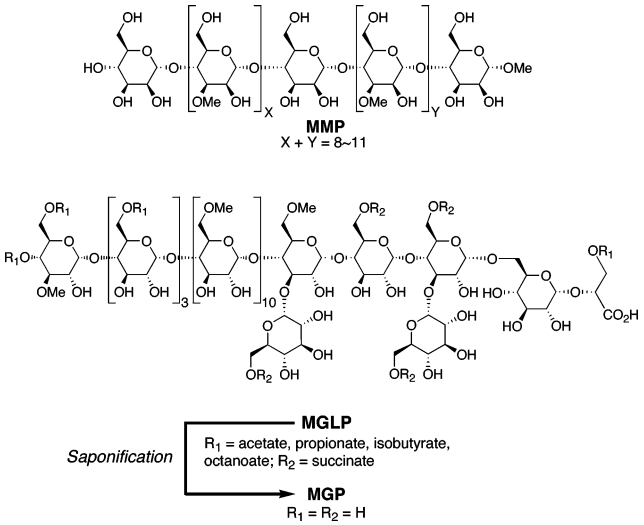


FIGURE 1. Structures of mycobacterial polysaccharides 3-*O*-methyl-D-mannose-containing polysaccharides (MMPs) and 6-*O*-methyl-D-glucose-containing lipopolysaccharides (MGLPs)/6-*O*-methyl-D-glucose-containing polysaccharides (MGPs).

Both MMP and MGLP/MGP are known to affect profoundly the fatty acid biosynthesis in *Mycobacterium smegmatis*.[1a] They increase the overall rate of fatty acid biosynthesis, change the product distribution, and prevent product inhibition of the fatty acid synthetase.[2e,5,6] They form stoichiometric complexes with C_{16}- or longer acyl-CoA.[7] Bloch proposed a structural model for complex formation between palmitoyl-CoA and MMP.[1a,7] In this model, the conformationally flexible glycosidic bonds of MMP rotate to achieve a helical conformation with appropriate dimensions for accommodating the aliphatic side chain of acyl-CoA. In this conformation, the 3-*O*-methyl ether groups are directed toward the core of the helical carbohydrate, thereby increasing the hydrophobic nature of the host and stabilizing the proposed complex via hydrophobic interactions between MMP and the aliphatic side chain of acyl-CoA. Coincidentally, the remaining hydroxyl groups on the 2- and 6-positions of the sugar rings are located on the outside of the helix and can interact with either water molecules or the polar functionalities of acyl-CoA via hydrogen bonding.

We were interested in gaining mechanistic insights for the intriguing biological role(s) of MMP and MGLP/MGP. However, we felt that naturally occurring MMP and MGLP/MGP are not necessarily ideal substrates for our study, as they were isolated as complex mixtures of closely related polysaccharides.[2,3] For this reason, we chose to use synthetic polysaccharides structurally related to natural MMP and MGLP/MGP for two major reasons: (1) synthetic polysaccharides should be available as chemically well-defined and homogeneous materials and (2) synthetic polysaccharides should be structurally tunable for the needs of our investigation. Obviously, the most unique structural feature of natural MMP and MGLP/MGP is the polymeric form of O-methylated mannose and glucose. Thus, we decided to incorporate this structural feature in the synthetic polysaccharides and selected the polymers composed of 3-*O*-methyl-D-mannose and 6-*O*-methyl-D-glucose (Figure 2).

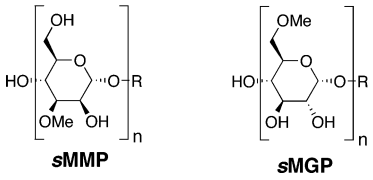


FIGURE 2. Structures of synthetic analogues of mycobacterial polysaccharides, *s*MMPs (synthetic polysaccharides composed of 3-*O*-methyl-D-mannose), and *s*MGPs (synthetic polysaccharides composed of 6-*O*-methyl-D-glucose).

Because *s*MMP and *s*MGP are α-(1→4)-linked oligomers of monomethylated mannose and glucose, respectively, we envisioned that their oligosaccharide backbones could be built by iterative glycosidations. For the overall efficiency of synthesis, we desired to incorporate the highest degree of convergence in the synthetic design and planned to synthesize *s*MMP and *s*MGP from an appropriate combination of a glycosyl donor and a glycosyl acceptor. To execute this plan with high overall efficiency, it is critically important to identify a glycosylation reaction that is not only highly stereoselective but also achievable with an approximately 1:1 mixture of a glycosyl donor and a glycosyl acceptor of approximately the same size. In this paper, using *s*MMP (R = *n*-Pr) with $n = 8$, 12, and 16 as examples, we demonstrate the feasibility of this synthetic plan.

We should note that this project was initiated in 1992,[8] when the chemical synthesis of oligosaccharides was still in the relatively underdeveloped stage. However, for the past one and a half decades, we have witnessed a remarkable development in this general area. A number of glycosyl donors, including halo sugars, pentenyl glycosides, thioglycosides, isopropenyl glycosides, orthoesters, 1-*O*-acyl sugars, 1-*O*-pentenoyl sugars, trichloroacetimidates, glycosyl sulfoxides, glycosyl sulfones, glycosyl thiocyanates, glycosyl dialkylphosphites, glycosyl phosphorodithioates, glycosyl tetramethylphosphorodiamidates, glycals and 1,2-anhydrosugars, seleno glycosides, and glycosyl diazirines, are now known to effect the glycosidation in a stereoselective manner in solution- and/or solid-phase syntheses.[9,10] Many of these synthetic methods have successfully been

(5) Flick, P. K.; Bloch, K. *J. Biol. Chem.* **1974**, *249*, 1031.

(6) Knoche, H.; Esders, T. W.; Koths, K.; Bloch, K. *J. Biol. Chem.* **1973**, *248*, 2317.

(7) (a) Machida, Y.; Bloch, K. *Proc. Natl. Acad. Sci. U.S.A.* **1973**, *70*, 1146. (b) Bergeron, M.; Machida, Y.; Bloch, K. *J. Biol. Chem.* **1975**, *250*, 1223.

(8) M.C.H. started the *s*MMP synthesis as her thesis work in 1992. This paper is largely based on her Dissertation: Hsu, M. C. Synthetic 3-*O*-Methylmannose Polysaccharides (*s*MMP): Complex Formation and Molecular Recognition with Fatty Acids. Dissertation, Harvard University, 1997.

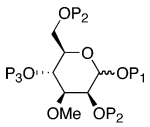
(9) A number of comprehensive monographs on "Chemical Synthesis of Glycosides" are available. For example, see: (a) *Carbohydrates in Chemistry and Biology*; Ernst, B., Hart, G. W., Sinaÿ, P., Eds.; Wiley-VCH: New York, 2000; Vol. 1. The book contains 18 relevant reviews by the leading practitioners. (b) *Carbohydrate-based Drug Discovery*; Wong, C.-H., Ed.; Wiley-VCH: Weinheim, Germany, 2003; Vol. 1. This contains three relevant reviews. (c) *Glycoscience: Chemistry and Chemical Biology*; Fraser-Reid, B. O., Tatsuta, K., Thiem, J., Eds.; Springer-Verlag: Berlin, Germany; Vols. 1 and 2. These contain five relevant reviews. In addition, the December issue of Chemical Reviews (**2000**, *100*) was dedicated to Carbohydrate Chemistry, which contains four relevant reviews.

(10) For recent reviews on "Chemical Synthesis of Glycosides and Oligosaccharides" by the experts in the field, see: (a) Schmidt, R. R. (i) page 5 of the monograph quoted in ref 9a; (ii) *Chem. Rev.* **2000**, *100*, 4423; (iii) *Chem. Rev.* **2006**, *106*, 160. (b) Danishefsky, S. J. (i) page 61 of the monograph quoted in ref 9a; (ii) *Acc. Chem. Res.* **1998**, *31*, 685; (iii) *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 1380. (c) Fraser-Reid, B. (i) page 135 of the monograph quoted in ref 9a. (d) Wong, C.-H. (i) page 117 of the monograph quoted in ref 9a; (ii) *Chem. Rev.* **2000**, *100*, 4465; (iii) *Science*, **2001**, *291*, 2344. (e) Seeberger, P. H. (i) page 103 of the monograph quoted in ref 9b; (ii) *Chem. Rev.* **2000**, *100*, 4349; (iii) *Eur. J. Org. Chem.* **2002**, 595. (f) Hanessian, S. *Chem. Rev.* **2000**, *100*, 4443. (g) Ley, S. V. (i) page 427 of the monograph quoted in ref 9a; (ii) *Chem. Rev.* **2001**, *101*, 53.

applied for the synthesis of a broad range of complex oligosaccharides, most notably mannose-containing oligosaccharides.[11]

Results and Discussion

The synthetic analysis briefly given above calls for appropriately substituted monosaccharides as the starting material. To execute effectively the designed synthesis, we needed to select a minimum of three different protecting groups to differentiate the hydroxyl groups present in **A**. Taking into

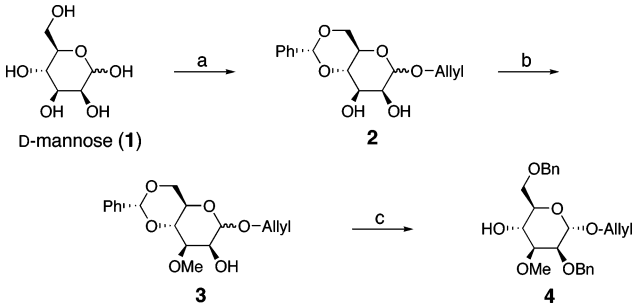


A

account the anticipated reactions for execution of the plan, we were decisive in choosing an allyl group as P_1 and a benzyl group as P_2. However, we postponed the decision on the protecting group P_3 until the first glycosidation was studied.

Using glucose-derived glycosyl donors and acceptors, we then conducted preliminary studies to develop the key glycosidation reaction.[12] However, as discussed in a separate paper,[13] we later realized that the profile of the glycosidation in the mannose series was pronouncedly different from that in the glucose series, and therefore some of the conclusions derived from the preliminary experiments on the glucose series should not be extrapolated to the mannose series. Nonetheless, on the basis of these preliminary studies, it became evident that, among several methods evaluated, the Mukaiyama glycosidation[14] best met our needs. Donors with various aliphatic and aromatic esters at C1 were tested for the Mukaiyama glycosidation, and the monomethyl phthalate ester (cf. **6** in Scheme 2) was found to give the most satisfactory results (chemical yield and α/β-selectivity).[15] With these preliminary results as our guide, we began the synthesis of suitably protected mannoses.

SCHEME 1[a]



[a] Reagents and conditions: (a) (1) $Sc(OTf)_3$, allyl−OH, 80%; (2) $PhCH(OMe)_2$, HBF_4, 83%; (b) (1) *n*-Bu_2SnO, MeOH, 83%; (2) MeI, 74%; (c) (1) NaH, BnBr, imidazole, 66%; (2) $TiCl_4$, $NaBH_3CN$, 73%.

Treatment of D-mannose with $Sc(OTf)_3$ (3 mol %) in allyl alcohol at 100 °C furnished the desired allyl glycoside in 80% isolated yield, with ca. 10:1 stereoselectivity (Scheme 1).[16] Addition of fluoroboric acid (catalytic) by a syringe pump to a solution of allyl mannoside and benzaldehyde dimethyl acetal in DMF gave the known acetal **2**.[17] The competing side reaction was the formation of the five-membered acetal between the C2 and C3 hydroxyl groups. A selective methylation of the C3 hydroxyl group of **2** was achieved via a cyclic stannyl ether,[18] to give **3** in 74% isolated yield. After protecting the C2 hydroxyl as a benzyl ether, the minor amount of the β-anomer was removed at this stage. The cyclic acetal was regioselectively opened by $NaBH_3CN/TiCl_4$,[19] to provide a chromatographically inseparable ca. 4:1 mixture of the desired product **4** and its C6 regioisomer. However, selective silylation of the primary alcohol present in the undesired regioisomer with *t*-butyldiphenylchlorosilane (TBDPSCl) facilitated separation/isolation, to furnish allyl α-mannoside **4** in 73% isolated yield.

The properly protected mannoside **4** gave an easy access to both the glycosyl acceptor **5** and donor **6**. To identify the optimal C4 protecting group, the glycosyl donors **6a**−**d**, with four types of protecting groups, triisopropylsilyl (TIPS), 4-methoxyphenylmethyl (MPM), phenylcarbonate, or benzoyl, were prepared (Scheme 2). Their behaviors under the Mukaiyama conditions (**5** + **6**, $SnCl_4$−$AgClO_4$ (10 mol %), 0 °C, 36 h, MeCN or Et_2O) were tested, thereby demonstrating the following: (1) either the TIPS group or the MPM group was not compatible with the glycosidation conditions; (2) a good α/β-selectivity (10:1) but a poor chemical yield (28%) was observed for **6c**; and (3) excellent α/β-selectivity and chemical yield were observed for **6d** (α/β-selectivity = 16:1 or 17:1, isolated yield = 79% or 76% in MeCN or Et_2O, 20 h, 0 °C).[20] From these preliminary

(11) For recent examples, see the following papers and references cited therein: (a) Merritt, J. R.; Naisang, E.; Fraser-Reid, B. *J. Org. Chem.* **1994**, *59*, 4443. (b) Grice, P.; Ley, S. V.; Pietruszka, J.; Osborn, H. M. I.; Priepke, H. W. M.; Warriner, S. L. *Chem.*−*Eur. J.* **1997**, *3*, 431. (c) Lee, H.-K.; Scanlan, C. N.; Huang, C.-Y.; Chang, A. Y.; Calarese, D. A.; Dwek, R. A.; Rudd, P. M.; Burton, D. R.; Wilson, I. A.; Wong, C.-H. *Angew. Chem., Int. Ed.* **2004**, *43*, 1000. (d) Danishefsky and co-workers synthesized several biologically important oligosaccharides containing mannosides. For the most recent work from the Danishefsky laboratory, see: Geng, X.; Dudkin, V. Y.; Mandal, M.; Danishefsky, S. J. *Angew. Chem., Int. Ed.* **2004**, *43*, 2562. (e) Jiang, L.; Chan, T. H. *Can. J. Chem.* **2005**, *83*, 693. (f) Watt, J. A.; Williams, S. J. *Org. Biomol. Chem.* **2005**, *3*, 1982. (g) Seeberger and co-workers synthesized several biologically important oligosaccharides containing mannosides. For the most recent work from the Seeberger laboratory, see: Liu, X.; Stocker, B. L.; Seeberger, P. H. *J. Am. Chem. Soc.* **2006**, *128*, 3638.

(12) Among the representative glycosidation methods known in 1992, the Schmidt glycosidation gave excellent results in terms of chemical yield and stereoselectivity, but its potential drawback for the current work was that, probably due to the instability of trichloroacetimidates, we had to use an excess of the donor to ensure a high yield for the glycosidation.

(13) Wang, Y.; Cheon, H.-S.; Lee, J.; Kishi, Y., in preparation.

(14) (a) Mukaiyama, T.; Takashima, T.; Katsurada, M.; Aizawa, H. *Chem. Lett.* **1991**, 533. (b) Mukaiyama, T.; Katsurada, M.; Takashima, T. *Chem. Lett.* **1991**, 985.

(15) This screening had been conducted before the second generation of the Mukaiyama ester, 2-(2-methoxyethoxy)acetate, was reported: Matsubara, K.; Sasaki, T.; Mukaiyama, T. *Chem. Lett.* **1993**, 1373. In the mannose series, the monomethyl phthalate and the second generation of the Mukaiyama ester gave comparable results (chemical yield and stereoselectivity), except for the rate of glycosidation, as the second generation of the Mukaiyama ester gave a much faster rate than with the monomethyl phthalate.

(16) The similar stereoselectivity (~10:1 α/β) was observed for formation of allyl mannoside with Dowex-H^+ resin: Lee, R. T.; Lee, Y. C. *Carbohydr. Res.* **1974**, *37*, 193. However, the $Sc(OTf)_3$ conditions furnished allyl mannoside in a higher yield with less side products and shorter reaction times.

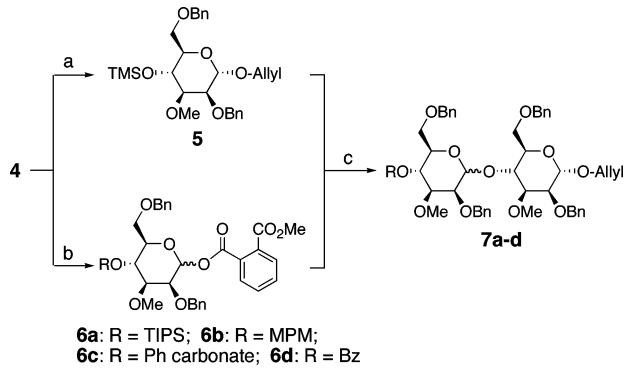
(17) (a) Albert, R.; Dax, K.; Pleschko, R.; Stutz, A. E. *Carbohydr. Res.* **1985**, *137*, 282. (b) Vasella, A.; Witzig, C.; Waldraff, C.; Uhlmann, P.; Briner, K.; Bernet, B.; Panza, L.; Husi, R. *Helv. Chim. Acta* **1993**, *76*, 2847. (c) Winnik, F. M.; Carver, J. P.; Krepinski, J. J. *J. Org. Chem.* **1982**, *47*, 2701.

(18) Nashed, M. A. *Carbohydr. Res.* **1978**, *60*, 200.

(19) Adam, G.; Seebach, D. *Synthesis* **1988**, 373.

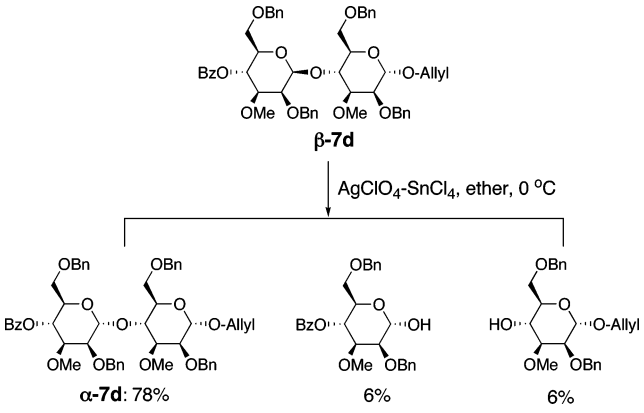
(20) The α- and β-anomers were distinguishable by ^1H NMR; i.e., the anomeric proton of α- and β-anomers resonated at 5.35 and 4.73 ppm, respectively. The stereoselectivity of glycosidation was estimated from the relative intensity of these signals.

SCHEME 2 [a]



[a] Reagents and conditions: (a) TMSOTf, Et_3N, 98%; (b) (1) protection with R; (2) (i) $(Ph_3P)_3RhCl$, DABCO; (ii) HCl; (3) monomethyl phthalate, EDCI, DMAP; (c) $SnCl_4-AgClO_4$ (10 mol %), 0 °C.

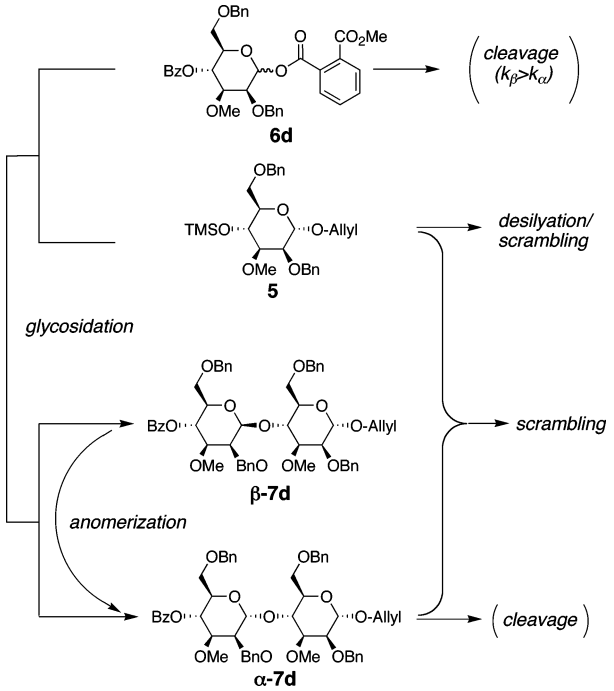
SCHEME 3. Products Formed from β-7d under the Mukaiyama Glycosidation Conditions



experiments, it became evident that the benzoate-protected donor (**6d** as ca. a 2:1 mixture of α- and β-anomers)[21] was the best choice.

Having identified the best combination of glycosyl donor and acceptor, we conducted a time course study to gain insight into the glycosidation. This study showed that the α/β-selectivity was only 3 in 1 h but improved to 5, 9, and 20 in 8, 22, and 30 h, respectively, thereby suggesting that the β-disaccharide β-**7d** anomerized into the α-disaccharide α-**7d** under the glycosidation conditions. Indeed, under the condition of 10 mol % of $SnCl_4-AgClO_4$ for 36 h, the β-disaccharide β-**7d** was converted to a mixture of α-glycoside α-**7d** (78%) and two monosaccharides (Scheme 3). Interestingly, however, the corresponding α-disaccharide α-**7d** was found not to anomerize to the β-disaccharide β-**7d** under the same conditions. Thus, the observed high stereoselectivity is attributed to the selective anomerization from the β- to α-anomer.[22] Interestingly, this

SCHEME 4. Reaction Pathways of the Mannosyl Donor (6d) and Acceptor (5) under the Mukaiyama Glycosidation Conditions



anomerization was not observed in the gluco series, and more extensive mechanistic studies will be reported in a separate paper.[13]

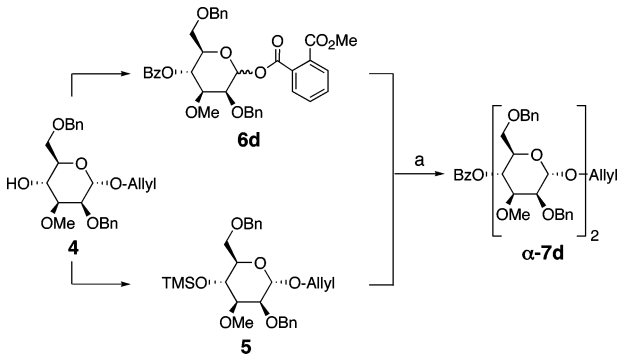
The selective anomerization suggests a possibility that only the desired α-disaccharide α-**7d** would be obtained, if a sufficiently long reaction time was given for the glycosidation. However, this was not a practical option because a significant amount of "scrambled" products were formed when a longer reaction time was employed. To gain insight into this scrambling, the stability of **5**, **6d**, and α-**7d** under the glycosidation conditions was then tested, thereby showing: (1) the acceptor **5** yielded the homocoupling product along with the desilylated starting material; (2) the disaccharide α-**7d** gave a small amount of the cleaved monosaccharides; and (3) the donor **6d** was susceptible to decomposition. Subsequently, a 1:1 mixture of α-**7d** and **5** was found to yield a mixture of scrambled products composed of trisaccharide (17%), tetrasaccharide (2%), and the homocoupling product (7%) under the standard glycosidation conditions. These results are summarized in Scheme 4, in which the reaction pathways on the left side show productive paths to the desired disaccharide α-**7d**, whereas the reaction pathways on the right side indicate paths to the scrambled products. Overall, as the glycosidation of **5** with **6d** is significantly faster than the glycosidation of **5** with **7d** or **5** itself, the byproduct formation via the latter couplings is negligible in a short reaction time but becomes considerable in a prolonged reaction time. Thus, a fine tuning of the reaction time was required. In passing, we noticed that the acceptor reactivity between the alcohol **4** and trimethylsilyl ether **5** showed no appreciable difference in glycosidation, whereas there was considerable difference in reactivity between α-**6d** and β-**6d** ($k_\beta \gg k_\alpha$).

With information on the profile of glycosidation, the optimal conditions were identified to obtain the desired α-**7d**: coupling of **5** (1.0 equiv) with **6d** (1.0 equiv) in the presence of $SnCl_4-AgClO_4$ (10 mol %) in diethyl ether at 0 °C for 36 h gave the

(21) Although a considerable difference was observed in their reactivity, both anomers β-**6d** and α-**6d** were found equally effective for the glycosidation.

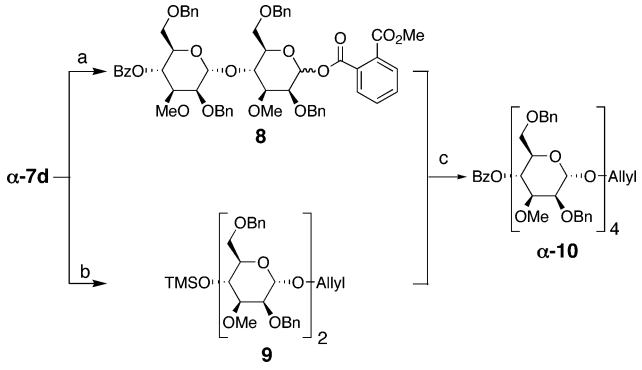
(22) It is generally recognized that β-mannosides are prone to facile anomerization. However, it is worth noting that the Mukaiyama catalyst ($AgClO_4-SnCl_4$) was found to promote the anomerization *only* from the β-mannosides to the corresponding α-mannosides. To the best of our knowledge, the anomerization with the observed level of selectivity is rare. Perhaps, the example reported by Lee and co-workers (Lee, C. K.; Kim, E. J.; Lee, I.-S. H. *Carbohydr. Res.* **1993**, *240*, 197) is most relevant to this case.

SCHEME 5 [a]



[a] Reagents and conditions: (a) $SnCl_4-AgClO_4$, Et_2O, 0 °C, 76%.

SCHEME 6 [a]



[a] Reagents and conditions: (a) (1) (i) $(Ph_3P)_3RhCl$, DABCO; (ii) HCl, 81%; (2) monomethyl phthalate, EDCI, DMAP, 98%; (b) (1) NaOH, MeOH, 92%; (2) TMSOTf, Et_3N, 99%; (c) $SnCl_4-AgClO_4$, Et_2O, −20 °C→0 °C, 76%.

disaccharide as a 20:1 mixture of α/β-anomers, from which the desired α-disaccharide α-**7d** was isolated in 76% yield in a pure form (Scheme 5).

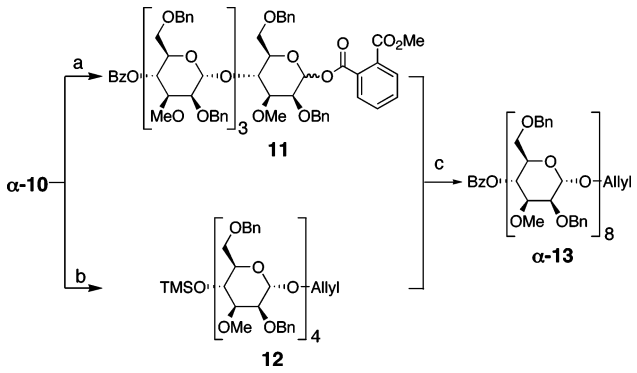
For the synthesis of the tetrasaccharide α-**10**, α-**7d** became the common source of the disaccharide donor and acceptor (Scheme 6). Rh-assisted deallylation[23] of α-**7d**, followed by esterification with monomethyl phthalate, provided the disaccharide donor **8**. The glycosyl acceptor **9** was prepared via saponification of the benzoate and TMS ether formation. The glycosidation proceeded in the same manner as the previous case. An 8:1 mixture of α/β-tetrasaccharides was formed in 3 h at 0 °C, whereas a >50:1 mixture was obtained in 26 h at 0 °C. After the minor β-tetrasaccharide was removed from the crude reaction mixture by HPLC (μ-porasil), purification by JAI (Japan Analytical Industry Co., Ltd., vide infra) gave the α-tetrasaccharide α-**10** in 76% yield with ≥99% purity.

Tetrasaccharide α-**10** was converted to both tetrasaccharide donor **11** and tetrasaccharide acceptor **12** under conditions similar to those described for the smaller fragments (Scheme 7).

Glycosidation of **11** with **12** met with complications when the reaction was left unquenched for over 10 h. The product obtained under such conditions exhibited a clean ^1H NMR spectrum, except the MeO signals (rxn A in Figure 3). Mass spectroscopy analysis (FAB-MS) showed the product contained a broad range of oligomers. Assuming that the oligomers have

(23) (a) Corey, E. J.; Suggs, W. J. *J. Org. Chem.* **1973**, *38*, 3224. (b) Gent, P. A.; Gigg, R. *J. Chem. Soc., Chem. Commun*. **1974**, 277.

SCHEME 7 [a]



[a] Reagents and conditions: (a) (1) (i) $(Ph_3P)_3RhCl$, DABCO; (ii) HCl, 81%; (2) monomethyl phthalate, EDCI, DMAP, 100%; (b) (1) NaOH, MeOH, 92%; (2) TMSOTf, Et_3N, 85%; (c) $SnCl_4-AgClO_4$, Et_2O, −20 °C→0 °C, 79%.

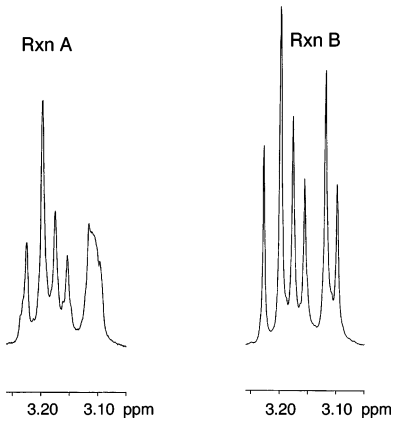
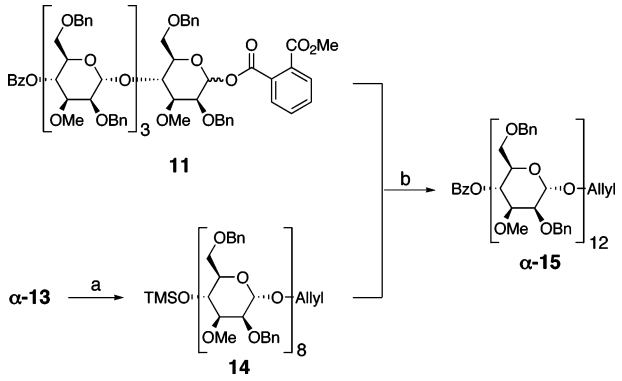


FIGURE 3. ^1H NMR spectra of the MeO signals of **13** (400 MHz, $CDCl_3$). Reaction A was quenched after 21.5 h. Reaction B was quenched after 9.5 h.

similar ionization potentials, the major components in the mixture included the hexamer (ca. 35%), heptamer (ca. 22%), octamer (desired; ca. 24%), nanomer (ca. 9%), and decamer (ca. 6%). Knowing the profile of glycosidation and considering the total number of the glycosidic bonds present in α-**13**, this result was not totally surprising. Quenching the reaction within 10 h provides high chemical conversion with significantly less contamination of scrambled products (FAB-MS: relative contribution of the octamer was ≫70%). The ^1H NMR spectrum showed six sharp signals corresponding to eight methyoxyls. Subsequent purification by JAI recycling HPLC provided the octasaccharide α-**13** in at least 90% purity (rxn B in Figure 3).

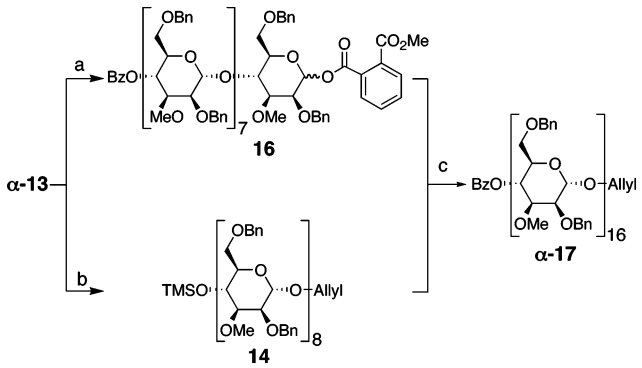
For preparative purposes, the glycosidation of **11** and **12** was quenched after 9.5 h. The ^1H NMR spectrum of the crude product showed that the α/β-stereoselectivity was >50:1 and also that a small amount of unreacted donor **11** and desilylated acceptor remained. Although the three components had similar R_f values by TLC, α-**13** could be isolated in 79% yield by silica gel chromatography. The more preparatively acceptable method of separation involved two chemical derivatization steps: (1) treatment of the crude product with $TMSOTf/NEt_3$ regenerated **12** from the unreacted acceptor, which was readily recovered by chromatography, and (2) base hydrolysis facilitated column-chromatographic separation of α-**13** from the remaining deprotected form of **11**.

SCHEME 8 [a]



[a] Reagents and conditions: (a) (1) NaOH, MeOH, 83%; (2) TMSOTf, Et_3N, 100%; (b) $SnCl_4-AgClO_4$, Et_2O, 0 °C, 75%.

SCHEME 9 [a]



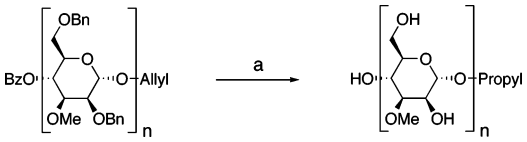
[a] Reagents and conditions: (a) (1) (i) $(Ph_3P)_3RhCl$, DABCO; (ii) HCl, 77%; (2) monomethyl phthalate, EDCI, DMAP, 94%; (b) (1) NaOH, MeOH, 83%; (2) TMSOTf, Et_3N, 100%; (c) $SnCl_4-AgClO_4$, Et_2O, 0 °C, 74%.

Octasaccharide α-**13** was converted to the octasaccharide acceptor **14** in two steps (Scheme 8), and coupling of **14** with tetrasaccharide donor **11** was accomplished under established glycosidation conditions. Minimizing formation of scrambled products, this reaction was quenched after 8.5 h. Crude NMR again showed >50:1 (α/*β*) selectivity. The protected dodecasaccharide α-**15** was isolated in 75% yield.

Following procedures similar to those mentioned for the preparation of other donors, octasaccharide α-**13** was converted to the octasaccharide donor **16** (Scheme 9). Reaction of donor **16** with acceptor **14** allowed the formation of α-hexadecasaccharide α-**17** (>50:1 α/*β*-selectivity) in 9.5 h under the standard conditions (74% yield).

The purification method of oligo- and polysaccharides by JAI (Japan Analytical Industry Co., Ltd.) HPLC deserves a comment. The glucosidation products were purified by a JAI LC-908 preparative liquid chromatographic unit using two polystyrene-based gel permeation columns in tandem. The polystyrene column utilizes the principle of size exclusion, and this chromatographic technique was indispensable for preparative separation/purification of the polysaccharides. As one might speculate from the principle used for this separation technique, the capacity of separating a polysaccharide from the next lower or higher polysaccharide decreases with an increase of its molecular size. Nonetheless, this chromatographic technique allowed us to isolate α-**7d** (disaccharide), α-**10** (tetrasaccharide), α-**13** (octasaccharide), α-**15** (dodecasaccharide), and α-**17** (hexadecasaccharide) with the purity of >98%, >98%, >90%, >85%, and >75%, respectively.[24]

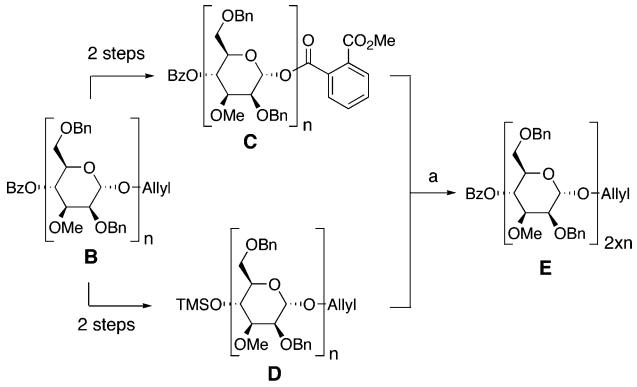
SCHEME 10 [a]



substrate	n	step 1	step 2	*s*MMP
α-**13**	8	83%	100%	**18**
α-**15**	12	78%	83%	**19**
α-**17**	16	87%	55%	**20**

[a] Reagents and conditions: (a) (1) NaOH, MeOH; (2) $Pd(OH)_2$ on C, H_2.

SCHEME 11 [a]



2xn (**E**, product)	selectivity (α:β)	isolated yield (α only)
2	>20:1	76%
4	>50:1	76%
8	>50:1	79%
12	>50:1	75%
16	>50:1	74%

[a] Reagents and conditions: (a) $SnCl_4-AgClO_4$, Et_2O. Note: 12-mer was synthesized via a combination of donor 4-mer and acceptor 8-mer.

Complete deprotection of the oligosaccharides was accomplished in two steps to furnish *s*MMP 8-, 12-, and 16-mers **18**−**20**, respectively (Scheme 10). Hydrolysis of the benzoate proceeded smoothly with methanolic NaOH. The final step was the hydrogenolysis of all benzyl ethers accompanied by the reduction of the allyl aglycon using Pearlman's catalyst and hydrogen. The products were isolated by reverse-phase chromatography on C_{18} silica gel. As discussed, the oligosaccharides α-**13**, α-**15**, and α-**17** were contaminated with scrambled products even after JAI chromatographic purification. However, the purity of *s*MMP 8-, 12-, and 16-mers **18**−**20** was at least 95% by FAB-MS and ^1H NMR analyses, implying that contaminated scrambled products were removed during the deprotection and purification. This may be one of the reasons the overall yield of the deprotection and purification declined from the *s*MMP 8-mer to the *s*MMP 16-mer.[25]

Conclusion

A highly convergent synthesis of *s*MMP has been achieved as summarized in Scheme 11. Beginning with D-mannose, the monosaccharide with the appropriate protecting groups installed (**B** with $n = 1$) was secured in six steps. **B** served as the source of glycosyl donor **C** and glycosyl acceptor **D**, each obtainable

in two steps. Glycosidation under the modified Mukaiyama protocol gave **E**, which is in fact identical to **B** except for *n*. The key steps of this synthesis are the five glycosidations, which proceeded with high α-selectivities and yields. This glycosidation was well suited for a highly convergent oligosaccharide synthesis, particularly because of excellent chemical yields even when using approximately equal-sized donors and acceptors in a molar ratio of approximately 1:1. An iterative reaction sequence allowed the growing oligosaccharide to double in size after each cycle and led to an efficient synthesis of *s*MMP 8-, 12-, and 16-mers **18**−**20**.

*s*MMP 8-, 12-, and 16-mers **18**−**20** thus obtained provided us, for the first time, with an opportunity to study the chemical and biological properties of *synthetic* MMP. To our delight, the preliminary experiments demonstrated that *synthetic* and *natural* MMPs exhibit identical, or at least very similar, properties.[26] With this encouraging result, we felt it to be critically important to address the scalability of *s*MMP synthesis to ensure the supply of the materials. Recognizing a potential problem with (1) the scalability of glycosidation and (2) the isolation of a glycosidation product free from scrambled products, we initiated a study on the second generation of *s*MMP synthesis, resulting in an improved, scalable synthesis of this class of polysaccharides.[27]

Experimental Section

Allyl 4,6-*O*-Benzylidene-α-D-mannopyranoside (2). To a solution of D-mannose (50.1 g, 278 mmol) in allyl alcohol (185 mL) was added $Sc(OTf)_3$ (412 mg, 3 mol %). The reaction mixture was heated to reflux for 6 h. The reaction mixture was concentrated to dryness and flushed through a silica gel column under reduced pressure (9:1 CH_2Cl_2−MeOH), to yield allyl mannopyranoside (49.2 g, 80%, >10:1 α/β mix).[28]

A solution of allyl mannopyranoside (46.3 g, 210 mmol) and benzaldehyde dimethyl acetal (38 mL, 0.25 mol) in DMF (1.0 L) was stirred vigorously. Fluoroboric acid (HBF_4, 54% in Et_2O, 19 mL, 0.11 mmol) was added over 120 min via a syringe pump. After stirring for an additional hour, Et_3N (88 mL, 0.63 mol) was added and the reaction mixture was concentrated to dryness. Silica gel chromatography (1:1 EtOAc/hexanes, EtOAc) gave the product **2** (53.5 g, 83%), a known compound in the literature.[17] ^{1}H NMR of the major anomer ($CDCl_3$, 400 MHz): 7.54−7.45 ppm (m, 2 H), 7.43−7.34 (m, 3 H), 5.91 (m, 1 H), 5.58 (s, 1 H), 5.32 (m, 1 H), 5.23 (m, 1 H), 4.93 (d, J = 1.0 Hz, 1 H), 4.28 (dd, J = 3.4, 9.0 Hz, 1 H), 4.22 (m, 1 H), 4.13 (dd, J = 3.5, 9.2 Hz, 1 H), 4.09 (dd, J = 1.4, 3.5 Hz, 1 H), 4.02 (m, 1 H), 3.94 (dd, J = 9.1, 9.2 Hz, 1 H), 3.84 (m, 2 H), 2.60 (bs, 2 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 137.2 ppm, 133.4, 129.3, 128.4, 126.2, 117.8, 102.3, 99.3, 77.9, 71.0, 68.8, 68.3, 63.1. HRMS (FAB) calcd for $(C_{16}H_{20}O_6Na)^+$ 331.1158, found 331.1158. IR (neat): 3341 cm^{-1}, 3230, 2923, 2903, 2866, 1454, 1397, 1378, 1285, 1219, 1133, 1112, 1098, 1069, 1029. $[\alpha]^{25}_D$ +60.7 (*c* 0.145, CH_2Cl_2).

Methyl Ether 3. A solution of **2** (39.7 g, 129 mmol) and *n*-Bu_2SnO (33.7 g, 135 mmol) in MeOH (650 mL) was heated to reflux. The reaction mixture was concentrated to dryness. The crude stannyl ether and MeI (40 mL, 0.64 mol) were dissolved in DMF (650 mL) and heated overnight at 50 °C. The crude product was flushed through a silica gel column under reduced pressure (9:1 hexanes/EtOAc, 4:1 hexanes/EtOAc, 1:1 hexanes/EtOAc), to yield **3** (30.6 g, 74%). ^{1}H NMR of the major anomer ($CDCl_3$, 400 MHz): 7.53−7.47 ppm (m, 2 H), 7.40−7.31 (m, 3 H), 5.92 (m, 1 H), 5.60 (s, 1 H), 5.32 (m, 1 H), 5.24 (m, 1 H), 4.95 (d, J = 1.0 Hz, 1 H), 4.27 (dd, J = 3.5, 8.8 Hz, 1 H), 4.21 (m, 1 H), 4.14 (m, 1 H), 4.05−3.98 (m, 2 H), 3.91−3.81 (m, 2 H), 3.72 (dd, J = 3.5, 9.6 Hz, 1 H), 3.56 (s, 3 H), 2.58 (d, J = 1.2 Hz, 1 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 137.5 ppm, 133.5, 129.0, 128.2, 126.1, 117.9, 101.8, 99.2, 78.8, 69.3, 68.9, 68.3, 63.3, 58.7. HRMS (FAB) calcd for $(C_{17}H_{22}O_6Na)^+$ 345.1314, found 345.1304. Select IR peaks (neat): 3463 cm^{-1}, 2912, 1456, 1379, 1218, 1172, 1125, 1096, 1049, 1036, 1009. $[\alpha]^{25}_D$ +77.1 (*c* 0.72, CH_2Cl_2).

Secondary Alcohol 4. To a solution of **3** (30.6 g, 95.0 mmol) in THF−DMF (4:1, 500 mL) were slowly added NaH (60% dispersion in mineral oil, 7.6 g, 0.19 mol), imidazole (0.65 g, 9.5 mmol), and benzyl bromide (BnBr, 23 mL, 0.19 mol). After 1 h, the reaction was quenched by addition of excess MeOH and water, followed by removal of solvents in vacuo. The crude mixture was dissolved in a CH_2Cl_2−water mixture, and the two layers were separated. The aqueous layer was extracted with CH_2Cl_2 (3×), and the organic extracts were dried over $MgSO_4$ and concentrated to dryness. The α-anomer (25.8 g, 66%) was isolated by silica gel chromatography under reduced pressure (9:1 hexanes/Et_2O).[28]

To a solution of benzyl ether (25.8 g, 62.5 mmol) and $NaBH_3$-CN (4.3 g, 69 mmol) in CH_3CN (160 mL) at 0 °C was added $TiCl_4$ (7.6 mL, 69 mmol) dropwise. The reaction mixture was stirred at 0 °C for an additional 3 h, then concentrated in vacuo. A solution of NaOH (2 N, ca. 200 mL) was added to the crude mixture, followed by a saturated $NaHCO_3$ solution. The aqueous layer was extracted with CH_2Cl_2 (3×), and the organic extracts were dried over $MgSO_4$ and concentrated to dryness.

The crude product was dissolved in DMF (125 mL) and stirred overnight with *t*-butyldiphenylchlorosilane (TBDPSCl, 16 mL, 63 mmol) and imidazole (2.1 g, 31 mmol). After removal of solvent, **4** (18.9 g, 73%) was isolated by silica gel chromatography (4:1 hexanes/Et_2O, 13:7 hexanes/Et_2O). ^{1}H NMR ($CDCl_3$, 400 MHz): 7.41−7.24 ppm (m, 10 H), 5.88 (m, 1 H), 5.25 (m, 1 H), 5.18 (m, 1 H), 4.95 (d, J = 1.8 Hz, 1 H), 4.69 (dd, J = 12.3, 16.9 Hz, 2 H), 4.61 (dd, J = 12.1, 19.1 Hz, 2 H), 4.19 (m, 1 H), 4.03−3.93 (m, 2 H), 3.84−3.72 (m, 4 H), 3.49 (dd, J = 3.2, 9.5 Hz, 1 H), 3.35 (s, 3 H), 2.58 (br s, 1 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 138.4 ppm, 138.3, 133.9, 128.3, 127.9, 127.7, 127.6, 127.5, 117.4, 97.3, 81.3, 73.6, 73.2, 72.7, 71.6, 70.6, 68.0, 57.2. HRMS (FAB) calcd for $(C_{24}H_{30}O_6Na)^+$ 437.1940, found 437.1940. Select IR peaks (neat): 3456 cm^{-1}, 2913, 1497, 1454, 1367, 1088, 1043. $[\alpha]^{25}_D$ +10.0 (*c* 0.61, CH_2Cl_2).

Monosaccharide Acceptor 5. To a solution of **4** (9.15 g, 22.1 mmol) and Et_3N (12 mL, 88 mmol) in CH_2Cl_2 (55 mL) was slowly added TMSOTf (8.5 mL, 44 mmol). After 30 min, saturated $NaHCO_3$ was added, and the organic components were extracted with CH_2Cl_2 (3×). The organic extracts were dried over $MgSO_4$ and concentrated to dryness, and **5** (10.5 g, 98%) was isolated by silica gel chromatography (9:1 hexanes/Et_2O, 3:1 hexanes/Et_2O). ^{1}H NMR ($CDCl_3$, 400 MHz): 7.47−7.28 ppm (m, 10 H), 5.94 (m, 1 H), 5.31 (m, 1 H), 5.23 (m, 1 H), 4.99 (d, J = 1.5 Hz, 1 H), 4.79 (dd, J = 12.4, 28.3 Hz, 2 H), 4.68 (s, 2 H), 4.25 (m, 1 H), 4.05−3.97 (m, 2 H), 3.89 (dd J = 1.9, 2.7 Hz, 1 H), 3.82−3.63 (m, 3 H), 3.46 (dd, J = 3.1, 9.2 Hz, 1 H), 3.40 (s, 3 H), 0.14 (s, 9 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 138.7 ppm, 134.1, 128.2, 127.7, 127.6, 127.5, 127.3, 117.2, 97.5, 82.1, 73.9, 73.4, 73.3, 72.8, 69.9, 68.3, 67.9, 57.2, 0.5. HRMS (FAB) calcd for $(C_{27}H_{38}O_6SiNa)^+$ 509.2335, found 509.2344. Select IR peaks (neat): 2953 cm^{-1}, 2912, 1454, 1248, 1104, 1060, 1028. $[\alpha]^{25}_D$ +29.6 (*c* 0.75, CH_2Cl_2).

Monosaccharide Donor 6d. A solution of **4** (9.72 g, 23.5 mmol), benzoyl chloride (BzCl, 5.4 mL, 47 mmol), and DMAP (575 mg, 4.71 mmol) in pyr (50 mL) was heated to 65 °C for 1 h. Pyr was removed in vacuo, and the residue was taken up in CH_2Cl_2, washed

(24) The purity was estimated from FAB-MS (see the text).

(25) The second step, i.e., $H_2/Pd(OH)_2$, in Scheme 11 effected deprotection of the benzyl group as well as reduction of the allyl group. However, it should be noted that α-**13**, α-**15**, and α-**17** contain 16, 24, and 32 benzyl groups, respectively, and the increasing number of benzyl groups may also contribute to the observed sharp decline in its efficiency.

(26) (a) Cheon, H. S.; Wang, Y.; Ma, J.; Kishi, Y. *ChemBioChem.*, in press. (b) Papaioannou, N.; Cheon, H.-S.; Kishi, Y., in preparation.

(27) Cheon, H.-S.; Kishi, Y., in preparation.

(28) Complete spectroscopic data of this synthetic intermediate are given in Supporting Information.

with saturated NH_4Cl, and dried over $MgSO_4$. The benzoate was isolated by silica gel chromatography (3:1 hexanes/Et_2O, 1:1 hexanes/Et_2O).[28]

A solution of the benzoate (12.2 g, 23.4 mmol), $(Ph_3P)_3RhCl$ (1.09 g, 5 mol %), and DABCO (400 mg, 15 mol %) in toluene−EtOH−H_2O (6:3:1, 150 mL) was heated overnight in a 100 °C oil bath. Removal of the solvent yielded the enol-ether, which was stirred with a solution of acetone/1 N HCl (9:1, 100 mL) for 2 h at 80 °C. Most of the solvent was removed in vacuo before adding saturated $NaHCO_3$ and extracting with CH_2Cl_2 (3×). The organic extracts were dried over $MgSO_4$. The lactol (9.64 g, 86% over two steps) was isolated by silica gel chromatography (3:1 hexanes/Et_2O, 3:2 hexanes/Et_2O, 1:1 hexanes/Et_2O, 1:3 hexanes-/Et_2O).[28]

A solution of the lactol (9.64 g, 20.2 mmol), monomethyl phthalate (7.26 g, 40.3 mmol), and DMAP (1.23 g, 10.1 mmol) in CH_2Cl_2 (100 mL) was cooled to 0 °C. 1-(3-Dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (EDCI, 5.8 g, 30 mmol) was added in portions to the reaction mixture. The ice-bath was removed, and the reaction mixture was stirred overnight. The solvent was removed in vacuo, and the crude mixture dissolved in EtOAc and H_2O. The organic layer was washed with H_2O, saturated $NaHCO_3$, H_2O, and saturated NaCl, then dried over $MgSO_4$, and concentrated to dryness. **6d** (12.9 g, 100%, ca. 2.4:1 α/β mixture of anomers) was isolated by silica gel chromatography (2:3 Et_2O/hexanes, 3:2 Et_2O/hexanes, 3:1 Et_2O/hexanes, 4:1 Et_2O/hexanes). ^1H NMR ($CDCl_3$, 400 MHz): 8.04−7.11 ppm (m, 19 H), 6.48 (d, J = 1.9 Hz, 1 H, major), 6.01 (s, 1 H, minor), 5.67 (dd, J = 9.9, 9.9 Hz, 1 H, major), 5.62 (dd, J = 9.2, 9.3 Hz, 1 H, minor), 4.86 (s, 2 H, major), 4.83 (s, 2 H, minor), 4.50 (s, 2 H, major), 4.49 (s, 2 H, minor), 4.18−4.09 (m, 2 H), 3.94 (m, 1 H, minor), 3.84 (s, 3 H), 3.77−3.55 (m, 3 H), 3.34 (s, 3 H, minor), 3.32 (s, 3 H, major). ^{13}C NMR ($CDCl_3$, 100 MHz): 167.8 ppm, 167.3, 166.6, 166.1, 165.6, 165.5, 137.8, 133.1, 132.3, 131.6, 131.2, 131.1, 131.0, 129.9, 129.8, 129.4, 129.3, 129.0, 128.8, 128.4, 128.3, 128.2, 128.0, 127.8, 127.6, 127.4, 93.9, 93.7, 81.6, 78.7, 77.3, 77.0, 76.7, 75.4, 74.1, 73.6, 73.4, 73.0, 72.7, 72.5, 69.8, 69.0, 58.2, 58.0, 52.6. HRMS (FAB) calcd for $(C_{37}H_{36}O_{10}Na)^+$ 663.2206, found 663.2191. Select IR peaks (neat): 2915 cm^{-1}, 1727, 1452, 1435, 1356, 1314, 1267, 1176, 1110, 1068, 1039, 1028. $[\alpha]^{25}_D$ +2.9 (*c* 0.65, CH_2Cl_2).

Disaccharide α-7d. Precautions: for handling of anhydrous $AgClO_4$, see refs 29a,b.[b] Solid $AgClO_4$ (310 mg, 1.50 mmol) in a round-bottomed flask was azeotroped with toluene. Under an atmosphere of nitrogen, $AgClO_4$ was dissolved in ether (60 mL), followed by the addition of $SnCl_4$ solution (0.3 M, 5.0 mL of solution, 1.50 mmol). The catalyst was generated by stirring this mixture in the dark. After 1 h, it was cooled to 0 °C with an ice bath. **6d** (9.57 g, 14.9 mmol) and **5** (7.24 g, 14.9 mmol) were premixed in a pear-shaped flask that had been dried overnight with a P_2O_5 trap. When the catalyst was ready, the starting materials were transferred to the reaction mixture with Et_2O (240 mL). The final concentration was 0.05 M relative to the glycosyl donor. The reaction mixture was then placed in a 0 °C refrigerator, stirred for 36 h, and quenched by addition of saturated $NaHCO_3$. The two layers were separated. The aqueous layer was extracted with CH_2Cl_2 (3×), and the combined organic extracts were dried over $MgSO_4$ and concentrated to dryness. Crude NMR showed an α/β ratio of disaccharide to be ca. 17:1. Silica gel chromatography (7:3 hexanes/Et_2O, 1:1 hexanes/Et_2O, 7:3 Et_2O/hexanes) yielded **α-7d** (9.99 g, 76%). ^1H NMR ($CDCl_3$, 400 MHz): 8.02 ppm (m, 2 H), 7.56 (m, 1 H), 7.47−7.10 (m, 22 H), 5.91 (m, 1 H), 5.57 (dd, J = 9.8, 9.8 Hz, 1 H), 5.34 (d, J = 1.9 Hz, 1 H), 5.27 (m, 1 H), 5.19 (m, 1 H), 4.94 (d, J = 1.7 Hz, 1 H), 4.75 (dd, J = 12.6, 16.9 Hz, 2 H), 4.69 (s, 2 H), 4.58−4.40 (m, 4 H), 4.21 (m, 1 H), 4.05−3.88 (m, 5 H), 3.83−3.71 (m, 3 H), 3.68 (dd, J = 2.9, 9.6 Hz, 1 H), 3.60−3.48 (m, 3 H), 3.31 (s, 3 H), 3.20 (s, 3 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 165.6 ppm, 138.8, 138.6, 138.3, 134.0, 132.9, 130.4, 129.8, 128.3, 128.2, 128.1, 127.7, 127.4, 127.2, 117.4, 100.3, 97.0, 81.9, 79.3, 75.1, 74.4, 73.6, 73.3, 72.6, 72.4, 71.7, 71.6, 70.6, 70.3, 69.8, 68.2, 57.9, 56.7. HRMS (FAB) calcd for $(C_{52}H_{58}O_{12}Na)^+$ 897.3826, found 897.3862. Select IR peaks (neat): 2924 cm^{-1}, 1726, 1453, 1268, 1111, 1070, 1050, 1028. $[\alpha]^{25}_D$ +29.2 (*c* 0.67, CH_2Cl_2).

Disaccharide Donor 8. Following the deallylation procedure given for the benzoate derived from **4**, **α-7d** (4.05 g, 4.62 mmol) was converted to the disaccharide lactol (3.12 g, 81%, solvent used for silica gel chromatography: 1:1 Et_2O/hexanes, 6:4 Et_2O/hexanes).[28]

Then, following the procedure described for formation of **6d**, disaccharide lactol (3.11 g, 3.72 mmol) was esterified to give **8** (3.64 g, 98%, solvent used for silica gel chromatography: 1:1 Et_2O/hexanes, 3:1 Et_2O/hexanes). ^1H NMR ($CDCl_3$, 400 MHz): 8.05−7.97 ppm (m, 2 H), 7.84−7.09 (m, 27 H), 6.47 (d, J = 2.0 Hz, 1 H, major), 5.90 (br s, 1 H, minor), 5.59 (dd, J = 9.5, 9.5 Hz, 1 H, minor), 5.57 (dd, J = 9.7, 9.8 Hz, 1 H, major), 5.36 (d, J = 2.0 Hz, 1 H, major), 4.87−4.69 (m, 4 H), 4.58−4.40 (m, 4 H), 4.14−3.95 (m, 3 H), 3.95−3.86 (m, 2 H), 3.86−3.82 (m, 3 H), 3.82−3.75 (m, 1 H), 3.75−3.47 (m, 5 H), 3.37−3.32 (m, 4 H, minor), 3.29 (s, 3 H, major), 3.19 (br s, 3 H, major), 3.18 (br s, 3 H, minor). ^{13}C NMR ($CDCl_3$, 100 MHz): 167.3 ppm, 166.2, 165.5, 138.4, 138.1, 137.8, 132.9, 132.3, 131.6, 131.5, 131.1, 131.0, 130.1, 129.7, 129.3, 129.2, 129.0, 128.8, 128.3, 128.2, 128.1, 127.9, 127.8, 127.6, 127.5, 127.4, 127.3, 100.2, 93.9, 93.7, 84.2, 80.8, 79.2, 79.1, 76.5, 74.4, 74.2, 74.1, 73.5, 73.3, 72.6, 72.4, 72.3, 72.1, 71.5, 70.1, 70.0, 69.5, 58.0, 57.9, 56.8, 56.5, 52.7, 52.6. HRMS (FAB) calcd for $(C_{58}H_{60}O_{15}Na)^+$ 1019.3830, found 1019.3845. Select IR peaks (neat): 2925 cm^{-1}, 1727, 1453, 1313, 1269, 1169, 1112, 1062, 1028. $[\alpha]^{25}_D$ +15.4 (*c* 0.70, CH_2Cl_2).

Disaccharide Acceptor 9. Disaccharide **α-7d** (4.47 g, 5.11 mmol) was stirred with methanolic NaOH (0.2 M, 50 mL) for 3.5 h. The reaction mixture was concentrated to dryness, and the residue was dissolved in EtOAc and washed with water, followed by saturated NaCl. The organic layers were dried over $MgSO_4$ and concentrated to dryness. Purification of the crude product by silica gel column chromatography (1:1 Et_2O/hexanes, 3:2 Et_2O/hexanes) yielded the secondary alcohol (3.63 g, 92%).[28]

Following the procedure given for formation of **5**, the secondary alcohol (3.63 g, 4.70 mmol) was silylated to yield **9** (3.94 g, 99%, solvent used for silica gel chromatography: 7:3 hexanes/Et_2O, 1:1 hexanes/Et_2O). ^1H NMR ($CDCl_3$, 400 MHz): 7.41−7.20 (m, 20 H), 5.90 (m, 1 H), 5.29−5.22 (m, 2 H), 5.18 (m, 1 H), 4.93 (d, J = 1.9 Hz, 1 H), 4.74−4.45 (m, 8 H), 4.19 (m, 1 H), 4.00−3.82 (m, 5 H), 3.79 (dd, J = 2.0, 3.1 Hz, 1 H), 3.76−3.67 (m, 3 H), 3.61−3.53 (m, 2 H), 3.47 (dd, J = 3.1, 9.1 Hz, 1 H), 3.31 (s, 3 H), 3.30 (dd, J = 3.0, 9.2 Hz, 1 H), 3.21 (s, 3 H), 0.08 (s, 9 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 138.9 ppm, 138.8, 138.7, 138.3, 133.9, 128.3, 128.2, 127.7, 127.6, 127.5, 127.2, 117.5, 100.3, 96.9, 81.9, 81.6, 77.2, 75.0, 74.3, 74.0, 73.5, 73.2, 73.1, 72.5, 72.2, 71.6, 70.4, 69.8, 68.1, 68.0, 57.0, 56.8, 0.5. HRMS (FAB) calcd for $(C_{48}H_{62}O_{11}SiNa)^+$ 865.3959, found 865.3978. Select IR peaks (neat): 2924 cm^{-1}, 1454, 1248, 1111, 1047, 1028. $[\alpha]^{25}_D$ +32.3 (*c* 0.39, CH_2Cl_2).

Tetrasaccharide α-10. Following the procedure described for formation of **α-7d**, **8** (4.07 g, 4.08 mmol) and **9** (3.44 g, 4.08 mmol) were glycosidated at −20 °C for 36 h. The desired tetrasaccharide **α-10** (4.92 g, 76% with ≥98% purity) was isolated by HPLC (1:1 hexanes−MeOBu-*t* on an m-porasil column) and then size exclusion HPLC ($CHCl_3$). ^1H NMR ($CDCl_3$, 400 MHz): 8.05−7.93 ppm (m, 2 H), 7.62−7.51 (m, 1 H), 7.47−7.05 (m, 42 H), 5.89 (m, 1 H), 5.61 (dd, J = 9.9, 9.9 Hz, 1 H), 5.37 (d, J = 1.7 Hz, 1 H), 5.29 (d, J = 1.6 Hz, 1 H), 5.28 (d, J = 1.8 Hz, 1 H), 5.24 (m, 1 H), 5.18 (m, 1 H), 4.93 (d, J = 1.7 Hz, 1 H), 4.80−4.36 (m, 16 H), 4.18 (m, 1 H), 4.07−3.60 (m, 16 H), 3.58−3.43 (m, 4 H), 3.40 (dd, J = 3.1, 9.1 Hz, 1 H), 3.32−3.25 (m, 4 H), 3.25−3.13 (m, 11 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 165.5 ppm, 138.7, 138.6, 138.5, 138.4,

(29) (a) Perrin, D. D.; Armarego, W. L. F. *Purification of Laboratory Chemicals*, 3rd ed.; Pergamon: New York, NY, 1988. (b) *Encyclopedia of Reagents for Organic Synthesis*; Paquette, L. A., Ed.; John Wiley & Son: New York, NY, 1995.

138.1, 138.0, 133.7, 132.8, 130.1, 129.7, 128.3, 128.2, 128.1, 127.6, 127.5, 127.4, 127.3, 127.2, 127.1, 117.5, 100.1, 99.7, 96.7, 81.6, 81.3, 79.1, 77.2, 75.3, 75.0, 74.8, 74.2, 74.0, 73.5, 73.4, 73.3, 73.2, 72.9, 72.7, 72.4, 72.3, 72.2, 71.8, 71.7, 71.4, 70.5, 70.0, 69.6, 68.1, 57.8, 56.6, 56.3. MS (FAB) calcd for $(C_{94}H_{106}O_{22}Na)^+$ 1609, found 1609. Select IR peaks (neat): 2921 cm^{-1}, 1726, 1453, 1268, 1112, 1072, 1046, 1028. $[\alpha]^{25}_D$ +34.4 (*c* 0.34, CH_2Cl_2).

Tetrasaccharide Donor 11. Following the deallylation procedure given for the benzoate derived from **4**, α-**10** (1.15 g, 721 mmol) was converted to the tetrasaccharide lactol (905 mg, 81%, solvent used for silica gel chromatography: 2:1 Et_2O/hexanes, 4:1 Et_2O/hexanes).[28]

Then, following the procedure given for the formation of **6d**, the tetrasaccharide lactol (905 mg, 585 mmol) was esterified to yield **11** (997 mg, 100%, solvent used for silica gel chromatography: 7:3 Et_2O/hexanes). ^1H NMR ($CDCl_3$, 400 MHz): 8.05−7.96 ppm (m, 2 H), 7.81−7.06 (m, 47 H), 6.46 (d, J = 1.6 Hz, 1 H, minor), 5.87 (s, 1 H), 5.63 (dd, J = 9.9, 9.9 Hz, 1 H), 5.39 (s, 1 H), 5.32 (s, 1 H), 5.30 (s, 1 H), 4.87−4.67 (m, 6 H), 4.67−4.36 (m, 10 H), 4.10−3.90 (m, 7 H), 3.90−3.60 (m, 14 H), 3.60−3.23 (m, 8 H), 3.21 (s, 3 H), 3.18 (s, 3 H), 3.16 (s, 3 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 167.7 ppm, 165.6, 165.5, 138.7, 138.5, 138.4, 138.3, 138.1, 132.8, 132.2, 131.6, 131.5, 131.1, 130.9, 130.2, 129.7, 129.3, 129.1, 129.0, 128.7, 128.3, 128.2, 128.1, 128.0, 127.9, 127.8, 127.6, 127.5, 127.3, 127.2, 127.1, 100.2, 99.7, 93.8, 93.6, 84.1, 81.3, 80.7, 79.1, 74.9, 74.8, 74.5, 74.3, 74.1, 73.5, 73.4, 73.2, 73.1, 73.0, 72.9, 72.5, 72.4, 72.2, 72.0, 71.9, 71.4, 70.4, 70.1, 70.0, 69.6, 57.9, 56.8, 56.5, 56.3, 52.6, 52.5. MS (FAB) calcd for $(C_{100}H_{108}O_{25}Na)^+$ 1731, found 1731. Select IR peaks (neat): 2925 cm^{-1}, 1727, 1453, 1270, 1113, 1047, 1028. $[\alpha]^{25}_D$ +21.0 (*c* 0.40, CH_2Cl_2).

Tetrasaccharide Acceptor 12. Following the procedure given for hydrolysis of the benzoate of α-**7d**, α-**10** (1.04 g, 657 mmol) was converted to the corresponding secondary alcohol (1.00 g).[28] The crude product was used for the next step without purification.

Then, following the procedure given for formation of **5**, the crude alcohol (974 mg, 657 mmol) was silylated to yield **12** (864 mg, 85% for two steps, solvent used for silica gel chromatography: 6:4 hexanes/Et_2O, 1:1 hexanes/Et_2O, 7:3 Et_2O/hexanes). ^1H NMR ($CDCl_3$, 400 MHz): 7.42−7.15 ppm (m, 40 H), 5.95 (m, 1 H), 5.37 (s, 1 H), 5.34 (s, 1 H), 5.33 (s, 1 H), 5.30 (m, 1 H), 5.24 (m, 1 H), 4.99 (s, 1 H), 4.82−4.69 (m, 6 H), 4.69−4.42 (m, 10 H), 4.25 (m, 1 H), 4.09−3.90 (m, 7 H), 3.90−3.58 (m, 15 H), 3.55−3.48 (m, 1 H), 3.48−3.38 (m, 2 H), 3.33 (s, 3 H), 3.27 (s, 3 H), 3.26 (s, 3 H), 3.23 (s, 3 H), 0.09 (s, 9 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 138.6 ppm, 138.1, 128.3, 128.2, 127.7, 127.6, 127.5, 127.4, 127.3, 127.2, 127.1, 117.6, 100.2, 99.8, 99.7, 96.8, 81.7, 81.5, 81.3, 74.9, 74.6, 74.1, 73.8, 73.4, 73.3, 73.0, 72.7, 72.5, 72.4, 72.0, 71.8, 71.7, 70.4, 69.6, 68.1, 67.9, 57.0, 56.7, 56.5, 0.5. MS (FAB) calcd for $(C_{90}H_{110}O_{21}SiNa)^+$ 1577, found 1577. Select IR peaks (neat): 2924 cm^{-1}, 1454, 1112, 1074, 1046, 1028. $[\alpha]^{25}_D$ +35.8 (*c* 1.20, CH_2Cl_2).

Octasaccharide α-13. Following the procedure given for synthesis of α-**7d**, **11** (844 mg, 493 mmol) and **12** (766 mg, 493 mmol) were glycosidated at 0 °C for 9.5 h. The crude reaction mixture was redissolved in CH_2Cl_2 (5.0 mL), followed by the addition of Et_3N (160 mL, 1.13 mmol) and TMSOTf (110 mL, 569 mmol). After aqueous workup (CH_2Cl_2 extractions (3×) from $NaHCO_3$ solution), silica gel chromatography (1:1 Et_2O/hexanes, 3:2 Et_2O/hexanes, 7:3 Et_2O/hexanes) allowed recovery of unreacted **12** and isolation of desired octasaccharide α-**13** (1.17 g, 79% with 90% estimated purity[24]). Further purification was done by JAI recycling HPLC. ^1H NMR ($CDCl_3$, 400 MHz): 8.03−7.96 ppm (m, 2 H), 7.59−7.52 (m, 1 H), 7.45−7.07 (m, 82m), 5.89 (m, 1 H), 5.62 (dd, J = 9.9, 9.9 Hz, 1 H), 5.38 (s, 1 H), 5.37−5.32 (m, 4 H), 5.31 (s, 1 H), 5.29 (s, 1 H), 5.24 (m, 1 H), 5.18 (m, 1 H), 4.93 (d, J = 1.5 Hz, 1 H), 4.81−4.65 (m, 10 H), 4.65−4.56 (m, 6 H), 4.56−4.33 (m, 16 H), 4.18 (m, 1 H), 4.08−3.88 (m, 11 H), 3.88−3.55 (m, 28 H), 3.55−3.36 (m, 9 H), 3.23 (s, 3 H), 3.20 (s, 6 H), 3.18 (s, 3 H), 3.16 (s, 3 H), 3.12 (s, 6 H), 3.10 (s, 3 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 165.6 ppm, 138.8, 138.5, 138.2, 138.1, 133.8, 132.8, 129.8, 128.3, 128.2, 128.1, 127.6, 127.5, 127.4, 127.3, 127.2, 117.5, 100.3, 99.8, 99.6, 96.8, 81.7, 81.4, 79.2, 75.2, 74.8, 74.1, 73.5, 73.4, 73.3, 73.1, 72.8, 72.5, 72.2, 71.8, 71.5, 70.6, 70.5, 70.1, 69.7, 68.1, 57.9, 56.6, 56.3. MS (FAB) calcd for $(C_{178}H_{202}O_{42}Na)^+$ 3037, found 3036. Select IR peaks (neat): 2923 cm^{-1}, 1727, 1453, 1269, 1114, 1074, 1047, 1028. $[\alpha]^{25}_D$ +38.8 (*c* 0.40, CH_2Cl_2).

Octasaccharide Acceptor 14. Following the procedure given for hydrolysis of the benzoate of α-**7d**, α-**13** (362 mg, 120 mmol) was converted to the corresponding secondary alcohol (290 mg, 83%, solvent used for silica gel chromatography: 2:1 hexanes/EtOAc, 3:2 hexanes/EtOAc, 1:1 hexanes/EtOAc).[28] However, to dissolve the starting material, an equal volume of THF and methanolic NaOH solution was used for this case.

Then, following the procedure described for formation of **5**, the octasaccharide secondary alcohol (286 mg, 98.1 mmol) was silylated to furnish **14** (292 mg, 100%, solvent used for silica gel chromatography: 2:1 hexanes/EtOAc, 3:2 hexanes/EtOAc). ^1H NMR ($CDCl_3$, 500 MHz): 7.48−7.03 ppm (m, 80 H), 5.89 (m, 1 H), 5.37−5.32 (m, 2 H), 5.32−5.29 (m, 5 H), 5.28 (s, 1 H), 5.24 (m, 1 H), 5.18 (m, 1 H), 4.93 (d, J = 1.5 Hz, 1 H), 4.75−4.63 (m, 9 H), 4.63−5.52 (m, 8 H), 4.52−4.44 (m, 8 H), 4.44−4.33 (m, 7 H), 4.18 (m, 1 H), 4.02−3.86 (m, 9 H), 3.86−3.68 (m, 21 H), 3.68−3.52 (m, 9 H), 3.50−3.42 (m, 7 H), 3.39 (dd, J = 2.7, 9.3 Hz, 1 H), 3.33 (dd, J = 2.8, 9.3 Hz, 1 H), 3.23 (s, 3 H), 3.21 (s, 3 H), 3.20 (s, 3 H), 3.18 (s, 3 H), 3.16 (s, 3 H), 3.13 (s, 3 H), 3.12 (s, 6 H), 0.09 (s, 9 H). ^{13}C NMR ($CDCl_3$, 100 MHz): 139.0 ppm, 138.9, 138.6, 138.2, 133.9, 128.3, 128.2, 128.1, 127.6, 127.5, 127.4, 127.1, 117.3, 100.3, 99.8, 99.7, 97.0, 81.8, 81.7, 81.5, 75.2, 74.8, 74.6, 74.4, 74.2, 73.4, 72.7, 72.1, 71.9, 70.6, 70.5, 69.8, 68.2, 68.0, 56.9, 56.6, 56.4, 56.3, 0.5. MS (FAB) calcd for $(C_{174}H_{206}O_{41}SiNa)^+$ 3005, found 3004. Select IR peaks (neat): 2925 cm^{-1}, 1454, 1113, 1074, 1046, 1028. $[\alpha]^{25}_D$ +33.8 (*c* 0.13, CH_2Cl_2).

Dodecasaccharide α-15. Following the procedure given for the synthesis of α-**13**, **11** (54.8 mg, 32.0 mmol) and **14** (95.4 mg, 32.0 mmol) were glycosidated at 0 °C for 8.5 h. The dodecasaccharide α-**15** (107 mg, 75% with 84% estimated purity[23]) was isolated by JAI recycling HPLC. ^1H NMR ($CDCl_3$, 500 MHz): 8.03−7.97 ppm (m, 2 H), 7.58−7.50 (m, 1 H), 7.47−7.03 (m, 122 H), 5.89 (m, 1 H), 5.62 (dd, J = 9.9, 9.9 Hz, 1 H), 5.37 (d, J = 1.6 Hz, 1 H), 5.36−5.33 (m, 8 H), 5.31 (s, 1 H), 5.30 (s, 1 H), 5.24 (m, 1 H), 5.18 (m, 1 H), 4.93 (d, J = 1.6 Hz, 1 H), 4.78−4.65 (m, 14 H), 4.63−4.55 (m, 10 H), 4.55−4.44 (m, 12 H), 4.44−4.32 (m, 12 H), 4.18 (m, 1 H), 4.07−3.88 (m, 15 H), 3.88−3.71 (m, 31 H), 3.71−3.57 (m, 12 H), 3.57−3.37 (m, 14 H), 3.23 (s, 3 H), 3.20 (s, 6 H), 3.18 (s, 3 H), 3.16 (s, 3 H), 3.13−3.04 (m, 21 H). MS (FAB) calcd for $(C_{262}H_{298}O_{62}Na)^+$ 4462, found 4464.

Octasaccharide Donor 16. Following the deallylation procedure given for the benzoate derived from **4**, α-**13** (221 mg, 73.2 mmol) was converted to the octasaccharide lactol (167 mg, 77%, solvent used for silica gel chromatography: 3:1 Et_2O/hexanes, Et_2O).[28]

Then, following the procedure given for preparation of **6d**, octasaccharide lactol (166 mg, 55.8 mmol) was esterified to yield **16** (165 mg, 94%, solvent used for silica gel chromatography: 98:2 CH_2Cl_2/MeOH). ^1H NMR ($CDCl_3$, 400 MHz): 8.05−7.95 ppm (m, 2 H), 7.82−7.50 (m, 5 H), 7.49−7.04 (m, 82 H), 6.46 (s, 1 H, major), 5.87 (s, 1 H, minor), 5.63 (dd, J = 9.9, 9.9 Hz, 1 H), 5.38 (s, 1 H), 5.35 (s, 4 H), 5.32 (s, 1 H), 5.31 (s, 1 H), 4.87−4.67 (m, 10 H), 4.67−4.55 (m, 7 H), 4.55−4.33 (m, 15 H), 4.12−3.90 (m, 11 H), 3.90−3.58 (m, 30 H), 3.58−3.32 (m, 8 H), 3.32−3.23 (m, 4 H, major), 3.21 (s, 3 H, minor), 3.20 (s, 3 H, minor), 3.19 (s, 3 H, major), 3.18 (s, 3 H, major), 3.17 (s, 3 H, minor), 3.16 (s, 3 H, minor), 3.15 (s, 6 H, major), 3.12 (s, 6 H, major), 3.10 (s, 3 H, major). ^{13}C NMR ($CDCl_3$, 100 MHz): 167.6 ppm, 167.2, 166.0, 165.5, 138.7, 138.4, 138.2, 138.1, 137.6, 132.8, 131.8, 131.6, 131.4, 131.1, 131.0, 130.1, 129.7, 129.2, 129.1, 129.0, 128.7, 128.3, 128.2, 128.0, 127.9, 127.8, 127.5, 127.4, 127.3, 127.1, 100.2, 99.8, 99.6, 93.8, 93.5, 84.1, 81.3, 80.8, 79.1, 74.9, 74.8, 74.7, 74.6, 74.5, 74.1,

74.0, 73.4, 73.3, 73.2, 73.1, 72.8, 72.5, 72.4, 72.1, 71.8, 71.4, 70.4, 70.2, 70.0, 69.6, 57.8, 56.7, 56.4, 56.3, 53.7, 52.5. MS (FAB) calcd for $(C_{184}H_{204}O_{45}Na)^+$ 3159, found 3157. Select IR peaks (neat): 2924 cm^{-1}, 1727, 1453, 1270, 1113, 1046, 1028. $[\alpha]^{25}_D$ +35.6 (*c* 0.23, CH_2Cl_2).

Hexadecasaccharide α-17. Following the glycosidation procedure given for α-**13**, **16** (251 mg, 80.0 mmol) and **14** (238 mg, 80.0 mmol) were coupled at 0 °C for 9.5 h. The hexadecasaccharide α-**17** (345 mg, 74% with 76% estimated purity[24]) was isolated by JAI recycling chromatography. ^{1}H NMR ($CDCl_3$, 500 MHz): 8.03−7.95 ppm (m, 2 H), 7.57−7.49 (m, 1 H), 7.48−7.03 (m, 162 H), 5.88 (m, 1 H), 5.61 (dd, J = 9.8, 9.9 Hz, 1 H), 5.37 (s, 1 H), 5.36−5.32 (m, 12 H), 5.31 (s, 1 H), 5.29 (s, 1 H), 5.24 (m, 1 H), 5.18 (m, 1 H), 4.93 (s, 1 H), 4.79−4.64 (m, 17 H), 4.64−4.53 (m, 15 H), 4.53−4.32 (m, 32 H), 4.18 (m, 1 H), 4.07−3.88 (m, 18 H), 3.88−3.64 (m, 46 H), 3.64−3.55 (m, 14 H), 3.55−3.36 (m, 18 H), 3.23 (s, 3 H), 3.20 (s, 6 H), 3.18 (s, 3 H), 3.16 (s, 3 H), 3.14−3.05 (m, 33 H). ^{13}C NMR ($CDCl_3$, 125 MHz): 165.6 ppm, 138.9, 138.5, 129.8, 128.4, 128.2, 128.1, 127.7, 127.6, 127.5, 127.3, 127.1, 117.5, 100.3, 99.7, 81.8, 81.5, 79.2, 75.2, 74.7, 73.5, 73.4, 73.3, 72.6, 72.3, 71.9, 71.5, 70.6, 70.5, 68.2, 57.9, 56.3. MS (FAB) calcd for $(C_{346}H_{394}O_{82}Na)^+$ 5888, found 5887. Select IR peaks (neat): 3366 cm^{-1}, 2923, 1723, 1453, 1273, 1115, 1047, 1028. $[\alpha]^{25}_D$ +45.9 (*c* 0.14, CH_2Cl_2).

***s*MMP 8-mer (18).** A solution of the octasaccharide secondary alcohol (35.7 mg, 11.8 mmol, obtained in 83% by hydrolysis of α-**13** under the conditions given for α-**15**)[28] in EtOH (1.0 mL) was stirred overnight with a portion of 10% $Pd(OH)_2$ on carbon (ca. 3 mg) under an H_2 balloon. The reaction mixture was passed through Celite to remove the catalyst, followed by removal of solvents in vacuo. The deprotected compound was purified by reverse-phase silica gel chromatography (4:1 H_2O−MeOH) to give **18** (17.3 mg, 100%). ^{1}H NMR (D_2O, 500 MHz): 5.23−5.20 (m, 7 H), 4.89 (d, J = 1.5 Hz, 1 H), 4.22−4.20 (m, 1 H), 4.20−4.17 (m, 6 H), 4.17−4.14 (m, 1 H), 3.89−3.61 (m, 40 H), 3.54−3.44 (m, 2 H), 3.45 (br s, 6 × 3 H), 3.43 (br s, 2 × 3 H), 1.64−1.57 (m, 2 H), 0.91 (t, J = 7.6 Hz, 3 H). ^{13}C NMR (D_2O, 100 MHz, 310 K): 101.4 ppm, 101.2, 99.6, 81.1, 80.8, 79.9, 73.9, 72.9, 72.5, 72.4, 71.3, 69.8, 66.4, 66.3, 66.1, 65.6, 61.1, 56.4, 56.3, 22.1, 10.0. MS (FAB) calcd for $(C_{59}H_{103}O_{41})^-$ 1467, found 1467. MS (FAB) calcd for $(C_{59}H_{104}O_{41}Na)^+$ 1492, found 1491. $[\alpha]^{25}_D$ +88.2 (*c* 0.24, H_2O).

***s*MMP 12-mer (19).** Methanolic NaOH solution (0.5 M, 1.0 mL) was added to α-**15** (63.7 mg, 14.3 mmol) in a round-bottomed flask. A small amount of CH_2Cl_2 was added dropwise to dissolve the starting material. The reaction was worked-up as described for hydrolysis of the benzoate of α-**7d**, to yield the corresponding secondary alcohol (48.5 mg, 78%, solvent for silica gel chromatography: 2:1 hexanes/EtOAc, 55:45 hexanes/EtOAc).[28]

Then, following the procedure given for *s*MMP 8-mer (**18**), the secondary alcohol (48.5 mg, 11.2 mmol) was hydrogenated to furnish *s*MMP 12-mer (**19**) (20.0 mg, 83%, solvent used for reverse-phase silica gel chromatography: 4:1 H_2O/MeOH, 3:2 H_2O/MeOH). Note that use of Celite should be avoided because it absorbed the product too tightly. ^{1}H NMR (D_2O, 500 MHz): 5.23−5.20 (m, 11 H), 4.89 (d, J = 1.5 Hz, 1 H), 4.22−4.20 (m, 1 H), 4.20−4.17 (m, 10 H), 4.16−4.14 (m, 1 H), 3.89−3.61 (m, 60 H), 3.54−3.44 (m, 2 H), 3.45 (br s, 10 × 3 H), 3.44 (br s, 2 × 3 H), 1.65−1.57 (m, 2 H), 0.91 (t, J = 7.6 Hz, 3 H). ^{13}C NMR (D_2O, 100 MHz, 310 K): 101.6 ppm, 101.4, 99.6, 80.8, 79.9, 73.8, 73.4, 72.9, 72.4, 71.4, 69.8, 66.3, 65.6, 61.0, 56.3, 22.0, 9.9. MS (FAB) calcd for $(C_{87}H_{151}O_{61})^-$ 2173, found 2173. $[a]^{25}_D$ +92.6 (*c* 0.47, H_2O).

***s*MMP 16-mer (20).** α-**17** (122 mg, 20.9 mmol) was converted to the corresponding secondary alcohol (105 mg, 87%), following the procedure described for hydrolysis of the benzoate of α-**7d**, except that an equal volume of THF and methanolic NaOH was added to enhance the solubility of the starting material in the reaction mixture. Solvent used for silica gel chromatography: 3:2 hexanes/EtOAc, 1:1 hexanes/EtOAc.[28]

Then, following the procedure, except for use of Celite, given for the case of *s*MMP 8-mer (**18**), the secondary alcohol (28.0 mg, 4.86 mmol) was subjected to hydrogenolysis/hydrogenation to provide *s*MMP 16-mer (**20**) (7.7 mg, 55%, solvent used for reverse-phase silica gel chromatography: 4:1 H_2O/MeOH, 3:2 H_2O/MeOH). ^{1}H NMR (D_2O, 500 MHz): 5.23−5.20 (m, 15 H), 4.89 (br s, 1 H), 4.22−4.20 (m, 1 H), 4.20−4.17 (m, 14 H), 4.16−4.14 (m, 1 H), 3.90−3.61 (m, 80 H), 3.54−3.44 (m, 2 H), 3.45 (br s, 14 × 3 H), 3.44 (br s, 2 × 3 H), 1.65−1.57 (m, 2 H), 0.92 (t, J = 7.6 Hz, 3 H). ^{13}C NMR (D_2O, 100 MHz, 310 K): 101.2 ppm, 99.3, 80.8, 80.6, 79.6, 73.6, 72.1, 71.0, 69.6, 66.0, 65.7, 65.3, 60.8, 56.0, 29.5, 21.8, 9.7. MS (FAB) calcd for $(C_{115}H_{199}O_{81})^-$ 2878, found 2878. $[\alpha]^{25}_D$ +95.7 (*c* 0.42, H_2O).

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Supporting Information Available: Includes the General Method Section and spectroscopic data. This material is available free of charge via the Internet at http://pubs.acs.org.

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