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Synthesis of the Tetrasaccharide Repeating Unit from *Acinetobacter baumannii* Serogroup O18 Capitalizing on Phosphorus-Containing Leaving Groups

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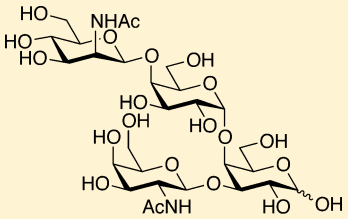
Supporting Information

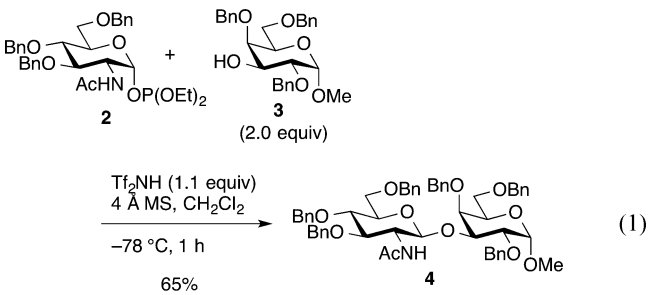
ABSTRACT: The first convergent synthesis of the tetrasaccharide repeating unit of the polymeric O antigen isolated from *Acinetobacter baumannii* serogroup O18 has been achieved. The ManNAcβ1→4Gal and GalNAcβ1→3Gal units were successfully obtained through β-selective glycosylation with 2-azido-4,6-*O*-benzylidene-2-deoxymannosyl diphenyl phosphate and Tf_2NH-promoted glycosylation with 2-acetamido-2-deoxygalactosyl diethyl phosphite, respectively. The disaccharide units could be coupled with the aid of $TMSClO_4$ as an activator of the diphenyl phosphate leaving group, and global deprotection completed the synthesis of the tetrasaccharide.



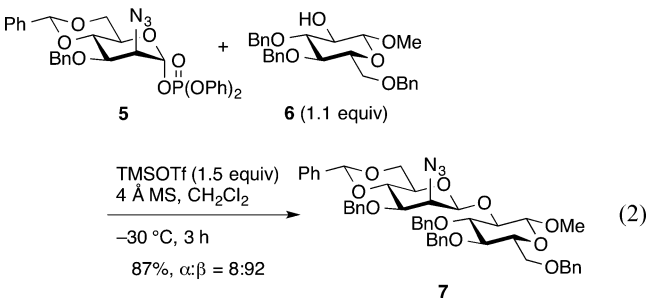
INTRODUCTION

Lipopolysaccharides (LPS) are expressed at the outer membrane of Gram-negative bacteria, and they play an important role in bacterial virulence and resistance to innate immunity.[1] Structurally, LPS is characterized by a highly variable polysaccharide attached through a core oligosaccharide to endotoxic lipid A, which functions as an anchor to the bacterial cell. Since the polysaccharide, comprising iterations of a repeating unit, defines the serotype and is responsible for the O-antigenic properties, fragments of various polysaccharides have been synthesized for potential vaccine development and pathogen detection.[2]

As mentioned in the preceding paper,[9b] we have developed novel glycosylation reactions capitalizing on phosphorus-containing leaving groups.[3] By use of donors with these leaving groups, various types of glycosidic linkages could be constructed in a stereoselective manner by appropriate choice of reaction conditions.[4−9] To demonstrate the synthetic utility of our method, we then undertook synthesis of the tetrasaccharide repeating unit of the polymeric O-antigen obtained from LPS extracted from isolated, defatted cell walls of the reference strain for *Acinetobacter baumannii* serogroup O18 (Figure 1).[10,11] The tetrasaccharide [ManNAcβ1→4Galα1→4(GalNAcβ1→3)Gal] (**1**) contains three types of glycosidic linkages, all of which required us to devise strategies for stereoselective construction. As described in the preceding paper,[9b] we have demonstrated that 2-acetamido-2-deoxyglycosyl diethyl phosphites could be employed for the glycosylation of reactive alcohols, such as 3-O-unprotected glycoside alcohol, at −78 °C (eq 1). With regard to the synthesis of 2-acetamido-2-deoxymannosides, van der Marel and co-workers,[12] inspired by Crich's β-mannosylation,[13] developed an efficient approach using 2-azido-4,6-*O*-benzylidene-2-deoxy-1-thiomannoside. In experiments patterned after their reports, we explored the glycosylation with 2-azido-2-deoxymannosyl donors carrying phosphorus-containing leaving groups and finally achieved a high-yield coupling with diphenyl phosphate as a leaving group (eq 2).[8] We surmised



(1)



(2)

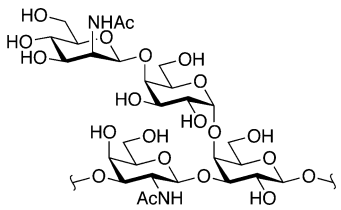


Figure 1. Structure of O-antigen from *Acinetobacter baumannii* serogroup O18.

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that the targeted tetrasaccharide **1** would be synthesized by taking advantage of these reactions. In this paper, we describe a convergent and stereocontrolled approach allowing access to the tetrasaccharide **1** by capitalizing on diethyl phosphite and diphenyl phosphate as leaving groups.

RESULTS AND DISCUSSION

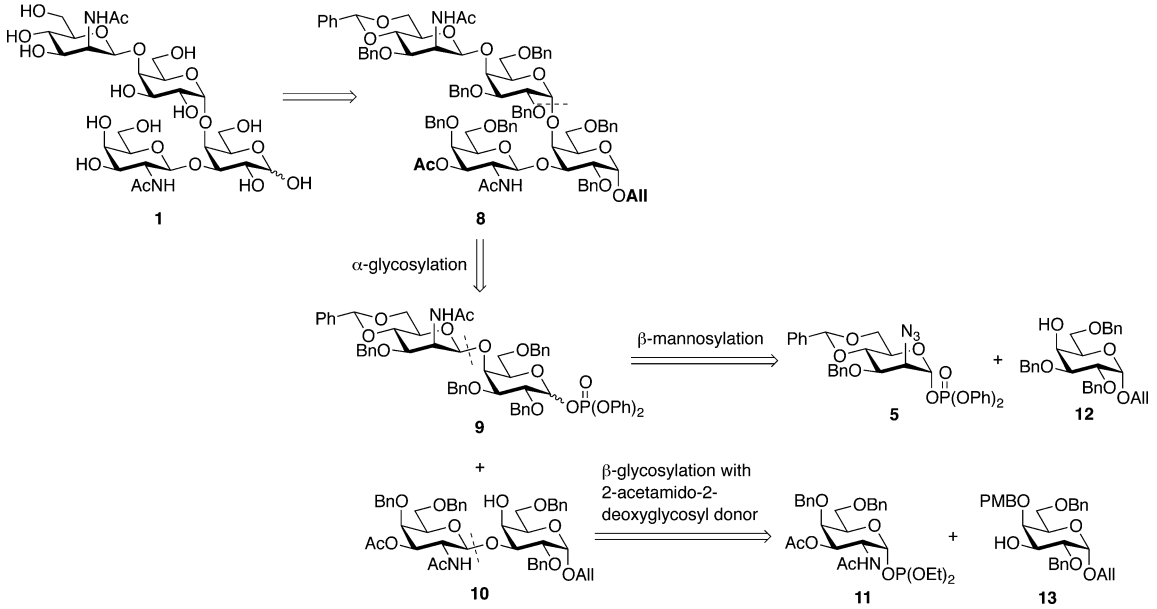
Our retrosynthetic analysis of tetrasaccharide **1** is depicted in Scheme 1. We considered that selectively removable masking groups should be employed for protection of the hydroxyl groups at the 3-position of GalNAc and the reducing terminus, so that the deprotected products could be used as building blocks for synthesis of polymeric saccharides. From the standpoint of convergency, coupling between disaccharides **9**[14] and **10** seemed to be attractive for the construction of tetrasaccharide **8**. The acetamido group of disaccharide **9** needs to be masked as an azide to ensure β-selective mannosylation[8] of 4-O-unprotected galactoside **12**.[15] On the other hand, disaccharide **10** would be available via glycosylation[9] of 3-O-unprotected galactoside **13** with 2-acetamido-2-deoxygalactosyl diethyl phosphite **11**.

Preparation of GalNAc donor **11** began with protection of known alcohol **14**[16] with BnBr, affording benzyl ether **15** in 96% yield (Scheme 2). After deprotection of the allyl glycoside by a two-step sequence involving olefin isomerization with *t*-BuOK and bromination in aqueous tetrahydrofuran (THF),[17] the *p*-methoxybenzyl (PMB) group was oxidatively removed with 2,3-dichloro-5,6-dicyanobenzoquinone (DDQ)[18] to give diol **17** in 74% overall yield for the three steps. Protection of the C3 hydroxyl group was achieved by the diacetylation−selective deacetylation sequence: the acetate at C1 could be hydrolyzed, with the acetate at C3 remaining intact, by employing Hauser's conditions (H_2O_2, $NaHCO_3$)[19] to furnish hemiacetal **19** with an α:β ratio of 95:5 in 92% yield. Phosphitylation of hemiacetal **19** with diethyl chlorophosphite in the presence of Et_3N gave α-linked diethyl phosphite **11** in 82% yield.

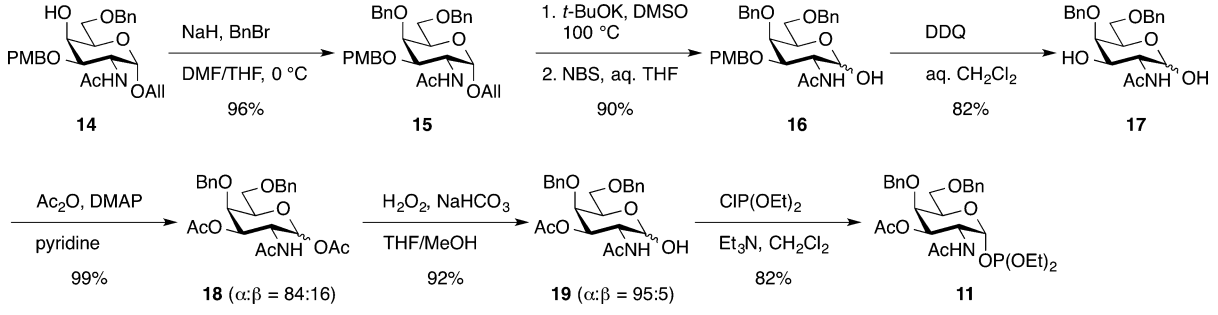
Both galactoside acceptors **12** and **13** were prepared from the common diol **20**[20] (Scheme 3). Monobenzylation of diol **20** via the corresponding stannylene acetal under modified conditions[21] provided 4-O-unprotected galactoside **12**[15] in almost quantitative yield. On the other hand, 3-O-unprotected galactoside **13** was obtained as a single isomer in 96% yield by the reduction of acetal **22**, prepared upon treatment of diol **20** with anisaldehyde dimethyl acetal, with $NaBH_3CN$ with the aid of trifluoroacetic acid (TFA) in the presence of 4 Å molecular sieves (MS). It is interesting that formation of undesired 4-O-unprotected galactoside was observed by the reduction of less polar isomer **21** under identical conditions. Stereochemical assignments for isomers **21** and **22** were determined by nuclear Overhauser enhancement (NOE) experiments.

With monosaccharide units **11**−**13** in hand, efforts were next focused on glycosylation reactions (Scheme 4). As expected,

Scheme 1. Retrosynthetic Analysis of Tetrasaccharide 1



Scheme 2. Preparation of 2-Acetamido-2-deoxygalactosyl Donor 11



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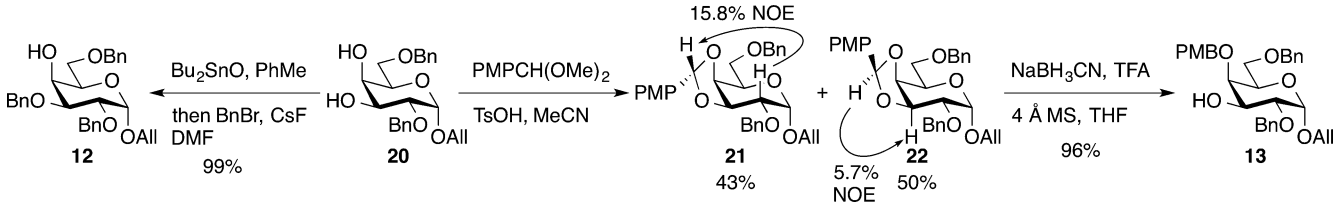
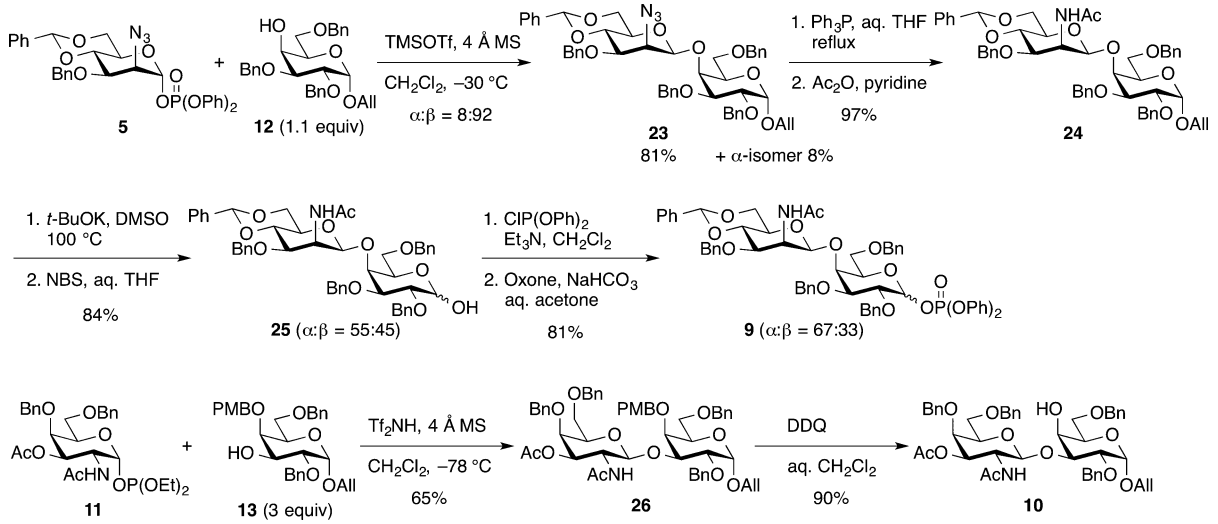
Scheme 3. Preparation of Galactosyl Acceptors 12 and 13



Scheme 4. Synthesis of Disaccharide Units 9 and 10 through β-Selective Glycosylations



glycosylation of 4-O-unprotected galactoside **12** with 2-azido-2-deoxymannosyl diphenyl phosphate **5** under optimized conditions[8] [trimethylsilyl trifluoromethanesulfonate (TMSOTf), 4 Å molecular sieves, CH_2Cl_2, −30 °C] proceeded to completion within 3 h to give disaccharides in 89% combined yield with exceptionally high β-selectivity (8:92). After separation of the anomers, azide **23** was transformed to acetamide **24** in 97% yield by treatment with Ph_3P in aqueous THF, followed by acetylation with Ac_2O in pyridine. Hemiacetal **25** (α:β = 55:45), obtained in 84% yield from allyl glycoside **24** by a two-step procedure that entailed treatment with *t*-BuOK in dimethyl sulfoxide (DMSO) at 100 °C and bromination with *N*-bromosuccinimide (NBS) in aqueous THF,[17] was successfully converted to diphenyl phosphate **9** (α:β = 67:33) in 81% yield via oxidation[22] of the corresponding phosphite with potassium peroxymonosulfate (Oxone)[23] in aqueous acetone.[24]

With regard to glycosylation with 2-acetamido-2-deoxygalactosyl diethyl phosphite **11**, 3 equiv of 3-O-unprotected galactoside **13** was employed as an acceptor since galactoside **13** could be easily prepared from inexpensive galactose on a large scale and the unreacted alcohol could be quantitatively recovered after column chromatography. The Tf_2NH-promoted reaction proceeded at −78 °C to give β-linked disaccharide **26** as a single isomer in 65% yield. Oxidative removal of the PMB group with DDQ in aqueous CH_2Cl_2[18] furnished alcohol **10** in 90% yield.

Having established access to disaccharide units **9** and **10**, the stage was now set for the crucial coupling reaction. During the course of our synthesis of globotriaosylceramide (Gb_3),[27] it was found that alcohols with poor nucleophilicity favored axial attack on the oxocarbenium ion generated from the galactosyl phosphorodiamidate with the aid of TMSOTf in CH_2Cl_2, leading to predominant formation of the corresponding α-glycosides. With this precedent in mind, we first selected TMSOTf as promoter. As expected, tetrasaccharide **8** was obtained as a sole product by the TMSOTf-promoted reaction of diphenyl phosphate **9** with alcohol **10** in the presence of 5 Å molecular sieves in CH_2Cl_2 at 0 °C; however, the yield was only 35% even with 2 equiv of alcohol **10** (Table 1, entry 1).[28] Gratifyingly, the product yield was improved to 75% by use of $TMSClO_4$ instead of TMSOTf (entry 2).[29] A solvent survey revealed that slower reaction and reduced yield (32%)[28] were observed in THF (entry 3)[30] and that the use of toluene as a cosolvent had no discernible benefit (entry 4). Examination of the temperature profile of the reaction demonstrated that a decrease in reaction temperature resulted in a decrease in product yield (entries 2 vs 5 and 6). Stereochemical assignment of the newly formed stereocenter in tetrasaccharide **8** was established by a significant ^{1}H NOE interaction and a coupling constant (J = 2.9 Hz) between H-1′ and H-2′.

With monosaccharide units **5** and **11**−**13** successfully assembled, the remaining operations necessary for synthesis of tetrasaccharide repeating unit **1** involved removal of all protecting groups. Exposure of tetrasaccharide **8** to NaOMe in MeOH effected methanolysis of the acetate at the 3-position of GalNAc to provide alcohol **27**, which can be employed as an acceptor for synthesis of the dimeric repeating unit (Scheme 5). The allyl protecting group at the reducing terminus in **27** could be safely removed by a two-step sequence involving $(Ph_3P)_3RhCl$-catalyzed olefin isomerization and bromination in aqueous THF,[17] to provide hemiacetal **28** in 82% overall yield. Finally, global deprotection employing 10% Pd/C in MeOH under a hydrogen atmosphere afforded the target tetrasaccharide **1** in 94% yield.

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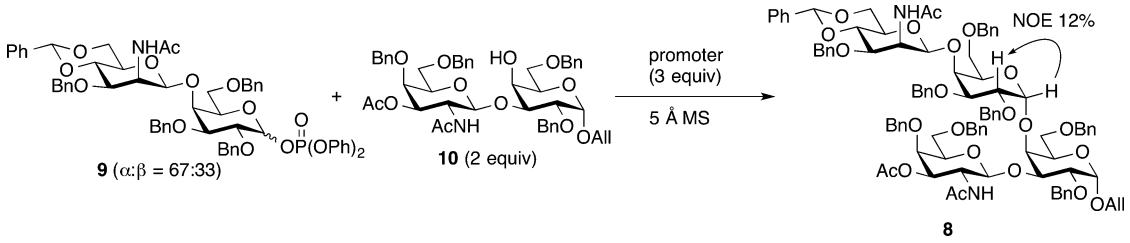
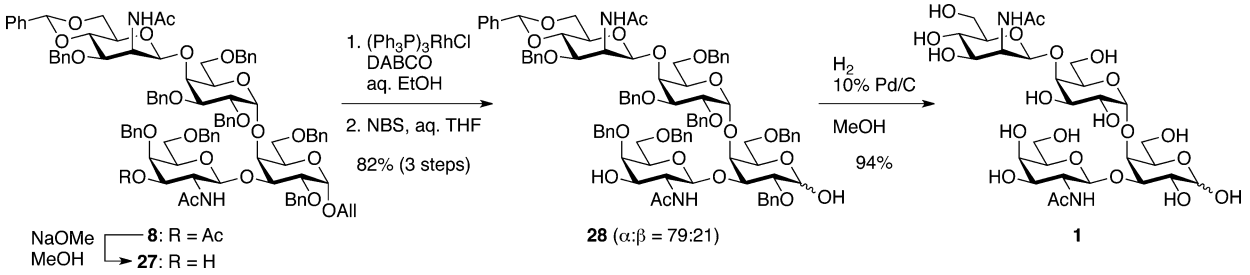
Table 1. Coupling of Disaccharide Units 9 and 10



entry	promoter	solvent	temp (°C)	time (h)	yield (%)
1	TMSOTf	CH_2Cl_2	0	1	35
2	$TMSClO_4$	CH_2Cl_2	0	0.5	75
3	$TMSClO_4$	THF	0	2	32
4	$TMSClO_4$	1:1 PhMe/CH_2Cl_2	0	0.5	77
5	$TMSClO_4$	CH_2Cl_2	−23	24	65
6	$TMSClO_4$	CH_2Cl_2	−45	72	58

Scheme 5. Completion of Synthesis of Tetrasaccharide Repeating Unit



CONCLUSION

We have completed the first total synthesis of the tetrasaccharide repeating unit of the polymeric O antigen isolated from *Acinetobacter baumannii* serogroup O18 (**1**), wherein all of the anomeric configurations could be controlled by proper choice of both phosphorus-containing leaving group and reaction conditions. The use of selectively removable protective groups for protection of the hydroxyl groups at the 3-position of GalNAc and the reducing terminus would allow access to polymeric saccharides. The synthesis provides a good illustration of the utility of phosphorus-containing leaving groups in oligosaccharide synthesis.

EXPERIMENTAL SECTION

2-Azido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-D-manno-pyranosyl Diphenyl Phosphate (5). NaOMe in MeOH (1.0 M, 0.6 mL, 0.6 mmol) was added to a solution of *tert*-butyldimethylsilyl 3,4,6-tri-*O*-acetyl-2-azido-2-deoxy-*β*-D-mannopyranoside[31] (1.65 g, 3.70 mmol) in MeOH (30 mL). After 1 h of stirring, the reaction mixture was neutralized with Amberlite IR-120B acidic resin. Filtration and evaporation in vacuo furnished the crude product (1.19 g), which was used without further purification.

Anhydrous *p*-toluenesulfonic acid (33 mg, 0.19 mmol) was added to a stirred solution of the crude triol and benzaldehyde dimethyl acetal (0.67 mL, 4.48 mmol) in MeCN (10 mL). After 30 min of stirring, the reaction was quenched with Et_3N (0.2 mL), and the volatile elements were removed in vacuo. Purification of the pale yellow residue by column chromatography (silica gel 30 g, 9:1 → 4:1 *n*-hexane/AcOEt) afforded *tert*-butyldimethylsilyl 2-azido-4,6-*O*-benzylidene-2-deoxy-*β*-D-mannopyranoside (1.21 g, 80%) as a white amorphous solid.

A 60% dispersion of NaH in mineral oil (143 mg, 3.58 mmol) was added to an ice-cooled (0 °C) mixture of the benzylidene acetal (1.12 g, 2.75 mmol), benzyl bromide (0.43 mL, 3.58 mmol), and Bu_4NI (102 mg, 0.275 mmol) in 9:1 THF/*N*,*N*-dimethylformamide (DMF) (20 mL). After 30 min of stirring, the reaction was quenched with MeOH (0.5 mL), and the resulting mixture was partitioned between AcOEt (70 mL) and saturated aqueous NH_4Cl (10 mL). The organic layer was successively washed with H_2O (10 mL) and brine (2 × 10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the pale yellow oil, which was purified by column chromatography (silica gel 30 g, 10:1 *n*-hexane/AcOEt) to give *tert*-butyldimethylsilyl 2-azido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-*β*-D-mannopyranoside (1.23 g, 90%) as a colorless syrup. R_f 0.71 (2:1 *n*-hexane/AcOEt); $[\alpha]_D^{23}$ −62.1 (*c* 2.03, $CHCl_3$); IR (neat) 3034, 2930, 2858, 2106, 1454, 1379, 1255, 1199, 1099, 1005 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) *δ* 0.12 (s, 3H), 0.16 (s, 3H), 0.92 (s, 9H), 3.32 (ddd, *J* = 5.0, 9.3, 10.2 Hz, 1H), 3.70 (dd, *J* = 3.8, 9.5 Hz, 1H), 3.85−3.89 (m, 2H), 3.98 (dd, *J* = 9.3, 9.5 Hz, 1H), 4.26 (dd, *J* = 5.0, 10.5 Hz, 1H), 4.75 (d, *J* = 12.4 Hz, 1H), 4.87 (d, *J* = 12.4 Hz, 1H), 4.89 (d, *J* = 1.0 Hz, 1H), 5.58 (s, 1H), 7.28−7.40 (m, 8H), 7.48 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) *δ* −5.6, −4.3, 17.7, 25.4, 65.0, 67.1, 68.1, 72.5, 75.7, 78.1, 95.7, 101.2, 125.8, 127.4, 127.5, 127.9, 128.2, 128.7, 137.2, 137.7; HRMS (FAB) *m*/*z* $[M + H]^+$ calcd for $C_{26}H_{36}N_3O_5Si$ 498.2424, found 498.2416. Anal. Calcd for $C_{26}H_{35}N_3O_5Si$: C, 62.75; H, 7.09; N, 8.44. Found: C, 62.57; H, 7.16; N, 8.39.

Bu_4NF in THF (1.0 M, 2.5 mL, 2.50 mmol) was added to an ice-cooled (0 °C) solution of the *tert*-butyldimethylsilyl (TBS) ether (1.05 g, 2.11 mmol) and AcOH (0.24 mL, 4.19 mmol) in THF (10 mL). After 20 min of stirring, saturated aqueous $NaHCO_3$ (10 mL) was added, and the resulting mixture was extracted with AcOEt (70 mL). The organic extract was successively washed with saturated aqueous $NaHCO_3$ (10 mL) and brine (2 × 10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (1.28 g), which was purified by column chromatography (silica gel 30 g, 2:1 *n*-hexane/AcOEt) to give 2-azido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-D-mannopyranose (798 mg, 99%, *α*:*β* = 56:44) as a white amorphous solid. The anomeric *α*:*β* ratio of the product was determined by 500 MHz ^1H NMR.

Diphenylphosphoryl chloride (0.52 mL, 2.50 mmol) was added to an ice-cooled (0 °C) solution of the hemiacetal (798 mg, 2.08 mmol) and 4-dimethylaminopyridine (DMAP; 508 mg, 4.16 mmol) in

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CH_2Cl_2 (8 mL). After 20 min, the reaction was quenched by addition of a piece of crushed ice, followed by stirring at room temperature for 15 min. The mixture was poured into a two-layer mixture of Et_2O (5 mL) and saturated aqueous $NaHCO_3$ (10 mL), and the resulting mixture was extracted with AcOEt (50 mL). The organic extract was washed with brine (2 × 10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (1.97 g, pale yellow oil), which was purified by column chromatography (silica gel 40 g, 4:1 → 2:1 *n*-hexane/AcOEt with 0.5% Et_3N) to give diphenyl phosphate **5** (917 mg, 72%) and its *β*-isomer (347 mg, 27%) as pale yellow syrups.

Data for *α*-phosphate **5**: R_f 0.46 (2:1 *n*-hexane/AcOEt); $[\alpha]_D^{23}$ +37.2 (*c* 1.51, $CHCl_3$); IR (neat) 3065, 2870, 2112, 1591, 1489, 1454, 1377, 1298, 1103 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) *δ* 3.71 (t, *J* = 10.2 Hz, 1H), 3.86 (ddd, *J* = 4.8, 9.8, 10.2 Hz, 1H), 3.93 (dd, *J* = 1.5, 3.5 Hz, 1H), 3.98 (dd, *J* = 4.8, 10.2 Hz, 1H), 4.07 (dd, *J* = 3.5, 9.8 Hz, 1H), 4.13 (t, *J* = 9.8 Hz, 1H), 4.67 (d, *J* = 12.1 Hz, 1H), 4.85 (d, *J* = 12.1 Hz, 1H), 5.57 (s, 1H), 5.79 (dd, *J* = 1.5, 6.3 Hz, 1H), 7.17−7.23 (m, 6H), 7.29−7.38 (m, 12H), 7.46 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) *δ* 62.3 (d, J_{C-P} = 10.3 Hz), 65.7, 67.9, 73.4, 74.6, 78.1, 97.7 (d, J_{C-P} = 5.0 Hz), 101.6, 119.9 (d, J_{C-P} = 5.0 Hz), 120.0 (d, J_{C-P} = 5.0 Hz), 125.7, 125.8, 125.9, 127.5, 127.8, 128.1, 128.4, 129.0, 129.85, 129.89, 137.0, 137.6, 150.0 (d, J_{C-P} = 7.6 Hz), 150.1 (d, J_{C-P} = 7.6 Hz); ^{31}P NMR (202 MHz, $CDCl_3$) *δ* −13.62; HRMS (FAB) *m*/*z* $[M + H]^+$ calcd for $C_{32}H_{31}N_3O_8P$ 616.1849, found 616.1860.

Data for *β*-phosphate: R_f 0.39 (2:1 *n*-hexane/AcOEt); $[\alpha]_D^{23}$ −25.7 (*c* 1.52, $CHCl_3$); IR (neat) 3065, 2876, 2112, 1591, 1489, 1454, 1386, 1286, 1188, 1064 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) *δ* 3.25 (ddd, *J* = 4.9, 9.5, 10.0 Hz, 1H), 3.65 (dd, *J* = 3.7, 10.0 Hz, 1H), 3.66 (t, *J* = 10.0 Hz, 1H), 3.79 (dd, *J* = 1.2, 3.7 Hz, 1H), 3.92 (t, *J* = 9.5 Hz, 1H), 4.07 (dd, *J* = 4.9, 9.5 Hz, 1H), 4.58 (d, *J* = 12.2 Hz, 1H), 4.73 (d, *J* = 12.2 Hz, 1H), 5.36 (dd, *J* = 1.2, 6.9 Hz, 1H), 5.45 (s, 1H), 7.09−7.14 (m, 6H), 7.21−7.27 (m, 12H), 7.36 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) *δ* 63.3 (d, J_{C-P} = 7.9 Hz), 67.7, 67.8, 73.1, 76.1, 77.7, 96.1 (d, J_{C-P} = 3.8 Hz), 101.5, 120.1 (d, J_{C-P} = 3.8 Hz), 120.2 (d, J_{C-P} = 3.8 Hz), 125.6, 125.8, 125.9, 127.6, 127.9, 128.1, 128.4, 129.0, 129.4, 129.6, 129.9, 136.9, 137.3, 149.9 (d, J_{C-P} = 7.6 Hz), 150.2 (d, J_{C-P} = 7.6 Hz); ^{31}P NMR (202 MHz, $CDCl_3$) *δ* −13.11; HRMS (FAB) *m*/*z* $[M + H]^+$ calcd for $C_{32}H_{31}N_3O_8P$ 616.1849, found 616.1851.

Allyl 2-Acetamido-4,6-di-*O*-benzyl-2-deoxy-3-*O*-(4-methoxybenzyl)-*α*-D-galactopyranoside (15). A solution of glycoside alcohol **14** (1.94 g, 4.11 mmol) in THF (15 mL) was added to an ice-cooled (0 °C) suspension of NaH (65% in oil, 197 mg, 5.34 mmol) in DMF (25 mL). After 10 min of stirring at 0 °C, BnBr (0.60 mL, 4.93 mmol) was added, and the mixture was stirred at this temperature for 1 h. The reaction was quenched with MeOH (2 mL), and the resulting mixture was partitioned between AcOEt (150 mL) and saturated aqueous NH_4Cl (30 mL). The organic layer was successively washed with saturated aqueous NH_4Cl (30 mL) and brine (2 × 40 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (2.66 g, white solid), which was purified by column chromatography (silica gel 50 g, 20:1 → 10:1 CH_2Cl_2/acetone) to give benzyl ether **15** (2.21 g, 96%) as a white solid. R_f 0.40 (3:1 toluene/acetone); mp 126.5−128 °C (colorless needles from AcOEt/*n*-hexane); $[\alpha]_D^{20}$ +90.6 (*c* 1.03, $CHCl_3$); IR (KBr) 3324, 3065, 3030, 1649, 1549, 1252, 1119, 1044, 739 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) *δ* 1.92 (s, 3H), 3.55 (dd, *J* = 6.5, 9.2 Hz, 1H), 3.60 (dd, *J* = 2.3, 11.0 Hz, 1H), 3.62 (dd, *J* = 6.5, 9.2 Hz, 1H), 3.81 (s, 3H), 3.89 (t, *J* = 6.5 Hz, 1H), 3.94 (ddd, *J* = 1.1, 6.2, 13.2 Hz, 1H), 3.98 (br s, 1H), 4.11 (ddd, *J* = 1.5, 6.5, 13.2 Hz, 1H), 4.39 (d, *J* = 11.9 Hz, 1H), 4.42 (d, *J* = 12.0 Hz, 1H), 4.48 (d, *J* = 12.0 Hz, 1H), 4.57 (d, *J* = 11.5 Hz, 1H), 4.65 (d, *J* = 11.9 Hz, 1H), 4.66 (ddd, *J* = 3.8, 8.9, 11.0 Hz, 1H), 4.92 (d, *J* = 3.8 Hz, 1H), 4.96 (d, *J* = 11.5 Hz, 1H), 5.15 (ddd, *J* = 1.1, 1.4, 10.2 Hz, 1H), 5.19 (ddd, *J* = 1.4, 1.5, 17.4 Hz, 1H), 5.25 (br d, *J* = 8.9 Hz, 1H), 5.85 (m, 1H), 6.89 (m, 2H), 7.22−7.35 (m, 12H); ^{13}C NMR (126 MHz, $CDCl_3$) *δ* 23.4, 48.9, 55.2, 68.1, 68.9, 69.6, 70.8, 72.5, 73.4, 74.3, 76.5, 96.9, 113.7, 117.1, 127.4, 127.7, 127.8, 128.0, 128.1, 128.3, 129.2, 130.1, 133.8, 137.8, 138.5, 159.2, 169.6; HRMS (FAB) *m*/*z* $[M + H]^+$ calcd for $C_{33}H_{40}NO_7$ 562.2805, found 562.2794. Anal. Calcd for $C_{33}H_{39}NO_7$: C, 70.57; H, 7.00; N, 2.49. Found: C, 70.57; H, 6.92; N, 2.41.

2-Acetamido-4,6-di-*O*-benzyl-2-deoxy-3-*O*-(4-methoxybenzyl)-D-galactopyranose (16). Potassium *tert*-butoxide (571 mg, 5.09 mmol) was added to a stirred solution of allyl glycoside **15** (2.12 g, 3.77 mmol) in DMSO (10 mL). After 30 min of stirring at 100 °C, the reaction mixture was cooled with a water bath, and the reaction was quenched with crushed ice (ca. 1 g). Addition of cold water (10 mL) resulted in the precipitation of a white solid, which was filtered and washed with cold water.

NBS (739 mg, 4.15 mmol) was added to a stirred solution of the crude prop-1-enyl glycoside in 50:3 THF/H_2O (31.8 mL). After 5 min of stirring, the volatile elements were removed in vacuo, and the residue was partitioned between AcOEt (100 mL) and 10% aqueous $Na_2S_2O_3$ (30 mL). The organic layer was successively washed with H_2O (2 × 20 mL) and brine (2 × 30 mL), and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (2.19 g, slightly yellow solid), which was recrystallized from *i*-PrOH/*i*-Pr_2O to give hemiacetal **16** (1.78 g, 90%) as colorless needles. R_f 0.46 (1:1 toluene/acetone); mp 190.0−190.5 °C; $[\alpha]_D^{22}$ +60.7 (*c* 1.03, $CHCl_3$); IR (KBr) 3369, 3311, 3031, 2928, 1648, 1554, 1515, 1251, 1097, 1057, 819 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$, data for *α*-anomer) *δ* 1.92 (s, 3H), 3.49 (dd, *J* = 6.4, 9.1 Hz, 1H), 3.56 (dd, *J* = 6.4, 9.1 Hz, 1H), 3.66 (br d, *J* = 3.2 Hz, 1H), 3.74 (dd, *J* = 2.6, 10.8 Hz, 1H), 3.81 (s, 3H), 3.95 (br s, 1H), 4.11 (br t, *J* = 6.4 Hz, 1H), 4.37 (d, *J* = 11.9 Hz, 1H), 4.41 (d, *J* = 11.8 Hz, 1H), 4.46 (ddd, *J* = 3.2, 8.2, 10.8 Hz, 1H), 4.49 (d, *J* = 11.8 Hz, 1H), 4.57 (d, *J* = 11.7 Hz, 1H), 4.64 (d, *J* = 11.9 Hz, 1H), 4.93 (d, *J* = 11.7 Hz, 1H), 5.30 (t, *J* = 3.2 Hz, 1H), 5.42 (d, *J* = 8.2 Hz, 1H), 6.87−6.90 (m, 2H), 7.20−7.34 (m, 12H); ^{13}C NMR (100 MHz, $CDCl_3$, data for *α*-anomer) *δ* 23.4, 50.0, 55.3, 69.3, 71.1, 72.8, 73.4, 74.4, 75.8, 92.2, 113.8, 127.5, 127.7, 127.9, 128.12, 128.15, 128.3, 129.3, 130.0, 137.6, 138.3, 159.2, 170.6; HRMS (FAB) *m*/*z* $[M + H]^+$ calcd for $C_{30}H_{36}NO_7$ 522.2499, found 522.2484.

2-Acetamido-4,6-di-*O*-benzyl-2-deoxy-D-galactopyranose (17). DDQ (847 mg, 3.82 mmol) was added to an ice-cooled (0 °C), biphasic mixture of PMB ether **16** (1.66 g, 3.18 mmol) in 20:1 CH_2Cl_2/pH 7 phosphate buffer (31.5 mL). After 1 h of stirring at room temperature, the reaction was quenched with saturated aqueous $NaHCO_3$ (5 mL), and the resulting mixture was partitioned between AcOEt/*n*-hexane (9:1, 100 mL) and saturated aqueous $NaHCO_3$ (10 mL). The organic layer was successively washed with saturated aqueous $NaHCO_3$ (5 × 15 mL) and brine (2 × 30 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (1.60 g, slightly yellow solid), which was purified by column chromatography (silica gel 40 g, 2:1 CH_2Cl_2/acetone) to give diol **17** (1.05 g, 82%) as a white solid. R_f 0.25 (*α*-anomer), 0.18 (*β*-anomer) (3:2 CH_2Cl_2/acetone); mp 188.0−188.5 °C (colorless needles from *i*-PrOH/$CHCl_3$); $[\alpha]_D^{21}$ +53.9 (*c* 0.10, EtOH); IR (KBr) 3384, 3321, 3033, 2929, 1640, 1552, 1451, 827 cm^{-1}; ^1H NMR (400 MHz, CD_3OD, data for *α*-anomer) *δ* 1.98 (s, 3H), 3.48 (dd, *J* = 6.5, 9.4 Hz, 1H), 3.52 (dd, *J* = 6.5, 9.4 Hz, 1H), 3.84 (br d, *J* = 3.2 Hz, 1H), 3.95 (dd, *J* = 3.2, 11.1 Hz, 1H), 4.19 (t, *J* = 6.5 Hz, 1H), 4.29 (dd, *J* = 3.5, 11.1 Hz, 1H), 4.39 (d, *J* = 11.7 Hz, 1H), 4.46 (d, *J* = 11.7 Hz, 1H), 4.54 (d, *J* = 11.1 Hz, 1H), 4.91 (d, *J* = 11.1 Hz, 1H), 5.11 (d, *J* = 3.5 Hz, 1H), 7.24−7.34 (m, 10H); ^{13}C NMR (100 MHz, CD_3OD, data for *α*-anomer) *δ* 22.7, 52.5, 70.1, 70.3, 74.3, 76.4, 78.6, 92.8, 128.5, 128.6, 128.9, 129.10, 129.12, 129.3, 139.2, 140.1, 173.8; HRMS (FAB) *m*/*z* $[M + H]^+$ calcd for $C_{22}H_{28}NO_6$ 402.1917, found 402.1910. Anal. Calcd for $C_{22}H_{27}NO_6$: C, 65.82; H, 6.78; N, 3.49. Found: C, 65.56; H, 6.69; N, 3.48.

2-Acetamido-3-*O*-acetyl-4,6-di-*O*-benzyl-2-deoxy-D-galactopyranosyl Acetate (18). Acetic anhydride (0.65 mL, 6.93 mmol) was added to a stirred solution of diol **17** (927 mg, 2.31 mmol) and DMAP (28 mg, 0.23 mmol) in pyridine (20 mL). After 1.5 h of stirring, the reaction was quenched with 10% aqueous HCl (60 mL), and the resulting mixture was extracted with AcOEt (140 mL). The organic extract was successively washed with brine (20 mL), saturated aqueous $NaHCO_3$ (2 × 50 mL), and brine (2 × 50 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (1.19 g), which was purified by column chromatography (silica gel 20 g, 4:1 CH_2Cl_2/acetone) to give diacetate **18** (1.11 g, 99%, *α*:*β* = 84:16) as a colorless oil. The *α*- and *β*-anomers

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were separated by flash column chromatography with 5:1 CH_2Cl_2/acetone.

Data for α-anomer: R_f 0.42 (4:1 CH_2Cl_2/acetone); $[\alpha]_D^{22}$ +85.4 (c 1.05, $CHCl_3$); IR (neat) 3282, 1741, 1659, 1548, 1226 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 1.93 (s, 3H), 2.04 (s, 3H), 2.14 (s, 3H), 3.53 (dd, J = 5.3, 9.1 Hz, 1H), 3.64 (dd, J = 8.3, 9.1 Hz, 1H), 4.00 (br d, J = 2.9 Hz, 1H), 4.04 (dd, J = 5.3, 8.3 Hz, 1H), 4.40 (d, J = 11.5 Hz, 1H), 4.46 (d, J = 11.5 Hz, 1H), 4.56 (d, J = 11.4 Hz, 1H), 4.79 (d, J = 11.4 Hz, 1H), 4.87 (ddd, J = 3.5, 9.4, 11.4 Hz, 1H), 5.20 (dd, J = 2.9, 11.4 Hz, 1H), 5.50 (d, J = 9.4 Hz, 1H), 6.17 (d, J = 3.5 Hz, 1H), 7.26–7.36 (m, 10H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 20.9, 21.0, 23.1, 47.5, 67.6, 70.7, 71.2, 73.4, 73.8, 75.0, 91.5, 127.6, 127.7, 127.8, 128.2, 128.3, 137.4, 137.7, 169.0, 169.9, 171.3; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{26}H_{32}NO_8$ 486.2128, found 486.2113. Anal. Calcd for $C_{26}H_{31}NO_8$: C, 64.32; H, 6.44; N, 2.88. Found: C, 64.04; H, 6.37; N, 2.94.

Data for β-anomer: R_f 0.37 (4:1 CH_2Cl_2/acetone); mp 152.0–152.5 °C (colorless needles from acetone/n-hexane); $[\alpha]_D^{21}$ +9.49 (c 1.00, $CHCl_3$); IR (KBr) 3336, 1750, 1727, 1658, 1543, 1235 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 1.92 (s, 3H), 2.01 (s, 3H), 2.08 (s, 3H), 3.57–3.65 (m, 2H), 3.80 (br t, J = 6.8 Hz, 1H), 3.94 (br d, J = 2.9 Hz, 1H), 4.41 (d, J = 11.7 Hz, 1H), 4.47 (d, J = 11.7 Hz, 1H), 4.57 (d, J = 11.7 Hz, 1H), 4.58 (ddd, J = 8.8, 9.6, 11.4 Hz, 1H), 4.76 (d, J = 11.7 Hz, 1H), 5.00 (dd, J = 2.9, 11.4 Hz, 1H), 5.39 (d, J = 9.6 Hz, 1H), 5.62 (d, J = 8.8 Hz, 1H), 7.26–7.33 (m, 10H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 20.8, 20.9, 50.2, 67.5, 73.1, 73.2, 74.0, 74.9, 93.1, 127.6, 127.7, 128.0, 128.1, 128.3, 137.4, 137.7, 169.6, 170.0, 170.8; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{26}H_{32}NO_8$ 486.2128, found 486.2139. Anal. Calcd for $C_{26}H_{31}NO_8$: C, 64.32; H, 6.44; N, 2.88. Found: C, 64.06; H, 6.30; N, 2.93.

2-Acetamido-3-*O*-acetyl-4,6-di-*O*-benzyl-2-deoxy-D-galactopyranose (19). $NaHCO_3$ (281 mg, 3.34 mmol) and 35% aqueous H_2O_2 (1.0 mL) were added to a stirred solution of diacetate **18** (813 mg, 1.67 mmol) in 5:1 THF/MeOH (24 mL). After 24 h of stirring, the reaction was quenched with 10% aqueous $Na_2S_2O_3$ (20 mL), and the resulting mixture was extracted with AcOEt (100 mL). The organic extract was successively washed with half-saturated brine (2 × 20 mL) and brine (30 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (753 mg, white amorphous solid), which was purified by column chromatography (silica gel 15 g, 4:1 → 2:1 CH_2Cl_2/acetone) to give hemiacetal **19** (679 mg, 92%, α:β = 95:5) as a white solid. The anomeric α:β ratio of the product was determined by ^1H NMR. R_f 0.53 (α-anomer), 0.44 (β-anomer) (2:3 CH_2Cl_2/acetone); mp 131.5–132.5 °C (colorless needles from 8:1 acetone/n-hexane); $[\alpha]_D^{23}$ +49.0 (c 1.02, $CHCl_3$); IR (KBr) 3391, 3311, 1731, 1656, 1547, 1250 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$, data for α-anomer) δ 1.94 (s, 3H), 2.04 (s, 3H), 3.46 (dd, J = 6.8, 9.7 Hz, 1H), 3.56 (dd, J = 6.8, 9.7 Hz, 1H), 3.58 (br d, J = 3.5 Hz, 1H), 3.87 (br d, J = 2.9 Hz, 1H), 4.22 (br t, J = 6.8 Hz, 1H), 4.41 (d, J = 12.0 Hz, 1H), 4.49 (d, J = 12.0 Hz, 1H), 4.50 (d, J = 12.0 Hz, 1H), 4.71 (ddd, J = 3.5, 9.7, 11.4 Hz, 1H), 4.80 (d, J = 12.0 Hz, 1H), 5.21 (dd, J = 2.9, 11.4 Hz, 1H), 5.26 (t, J = 3.5 Hz, 1H), 5.73 (d, J = 9.7 Hz, 1H), 7.26–7.35 (m, 10H); ^{13}C NMR (100 MHz, $CDCl_3$, data for α-anomer) δ 21.0, 23.2, 48.6, 68.87, 68.94, 71.3, 73.2, 74.5, 74.8, 91.9, 127.66, 127.69, 127.74, 128.1, 128.2, 128.3, 137.4, 137.8, 170.3, 171.1; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{24}H_{30}NO_7$ 444.2022, found 444.2020. Anal. Calcd for $C_{24}H_{29}NO_7$: C, 65.00; H, 6.59; N, 3.16. Found: C, 64.86; H, 6.54; N, 3.18.

2-Acetamido-3-*O*-acetyl-4,6-di-*O*-benzyl-2-deoxy-α-D-galactopyranosyl Diethyl Phosphite (11). Diethyl chlorophosphite (0.23 mL, 1.60 mmol) was added to an ice-cooled (0 °C) solution of hemiacetal **19** (591 mg, 1.33 mmol) and Et_3N (0.56 mL, 4.00 mmol) in CH_2Cl_2 (13 mL). After 30 min of stirring at 0 °C, the reaction was quenched with crushed ice, followed by stirring at room temperature for 10 min. The resulting mixture was partitioned between AcOEt (45 mL) and saturated aqueous $NaHCO_3$ (10 mL), and the organic layer was washed with brine (2 × 10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (935.2 mg, white solid), which was purified by column chromatography (Wako gel 15 g, 3:1 n-hexane/acetone with 3% Et_3N) to give diethyl phosphite **11** (611 mg, 82%) as a white solid. R_f 0.35 (3:1 n-hexane/acetone, with Et_3N-doped silica gel plate); $[\alpha]_D^{23}$ +89.2 (c 1.04, $CHCl_3$); mp 70.5–71.5 °C; IR (neat) 3290, 3064, 3031, 2978, 2928, 1741, 1651, 1237, 1027 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 1.23 (t, J = 6.9 Hz, 3H), 1.26 (t, J = 6.9 Hz, 3H), 1.94 (s, 3H), 2.03 (s, 3H), 3.53 (dd, J = 5.7, 9.1 Hz, 1H), 3.61 (dd, J = 7.5, 9.1 Hz, 1H), 3.86–3.94 (m, 4H), 3.93 (br d, J = 2.9 Hz, 1H), 4.21 (dd, J = 5.7, 7.5 Hz, 1H), 4.42 (d, J = 12.0 Hz, 1H), 4.48 (d, J = 12.0 Hz, 1H), 4.55 (d, J = 12.0 Hz, 1H), 4.78 (ddd, J = 3.5, 9.7, 10.9 Hz, 1H), 4.79 (d, J = 12.0 Hz, 1H), 5.21 (dd, J = 2.9, 10.9 Hz, 1H), 5.53 (dd, J = 3.5, 8.0 Hz, 1H), 5.65 (d, J = 9.7 Hz, 1H), 7.26–7.35 (m, 10H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 16.9 (d, J_{C-P} = 4.7 Hz), 17.1 (d, J_{C-P} = 4.8 Hz), 21.1, 23.4, 48.8, 58.5 (d, J_{C-P} = 10.7 Hz), 58.7 (d, J_{C-P} = 11.9 Hz), 68.1, 70.2, 71.2, 73.3, 74.2, 74.9, 92.7 (d, J_{C-P} = 14.3 Hz), 127.41, 127.45, 127.8, 128.0, 128.1, 137.5, 137.7, 169.4, 170.9; ^{31}P NMR (202.5 MHz, $CDCl_3$) δ 139.9; HRMS (ESI) m/z $[M + Na]^+$ calcd for $C_{28}H_{38}NO_9PNa$ 586.2182, found 586.2183. Anal. Calcd for $C_{28}H_{38}NO_9P$: C, 59.67; H, 6.80; N, 2.49. Found: C, 59.43; H, 6.69; N, 2.50.

Allyl 2,6-Di-*O*-benzyl-3,4-*O*-[(*R*)-4-methoxybenzylidene]-α-D-galactopyranoside (21) and Allyl 2,6-Di-*O*-benzyl-3,4-*O*-[(*S*)-4-methoxybenzylidene]-α-D-galactopyranoside (22). p-Toluenesulfonic acid (76.9 mg, 0.41 mmol) was added to an ice-cooled (0 °C) solution of diol **20** (1.62 g, 4.05 mmol) and p-methoxybenzaldehyde dimethyl acetal (0.86 mL, 4.85 mmol) in MeCN (10 mL). After 30 min of stirring at room temperature, the reaction was quenched with Et_3N (0.1 mL), and the volatile elements were removed in vacuo. Purification of the residue (2.59 g) by flash column chromatography (silica gel 60 g, 10:1 n-hexane/AcOEt) afforded (*R*)-isomer **21** (898 mg, 43%) and (*S*)-isomer **22** (1.05 g, 50%) as colorless oils.

Data for (*R*)-isomer **21**: R_f 0.62 (2:1 n-hexane/AcOEt); $[\alpha]_D^{21}$ +47.1 (c 1.01, $CHCl_3$); IR (neat) 3063, 3030, 2911, 2870, 1613, 1514, 1454, 1249, 1095, 1031 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 3.66 (dd, J = 3.5, 7.9 Hz, 1H), 3.71–3.74 (m, 2H), 3.81 (s, 3H), 4.03 (dd, J = 6.2, 12.9 Hz, 1H), 4.18–4.24 (m, 3H), 4.50 (d, J = 12.3 Hz, 1H), 4.61 (d, J = 12.3 Hz, 1H), 4.68 (dd, J = 5.0, 7.9 Hz, 1H), 4.77 (d, J = 12.6 Hz, 1H), 4.84 (d, J = 12.6 Hz, 1H), 4.93 (d, J = 3.5 Hz, 1H), 5.23 (d, J = 10.3 Hz, 1H), 5.35 (d, J = 17.0 Hz, 1H), 5.90 (s, 1H), 5.94 (m, 1H), 6.87–6.90 (m, 2H), 7.24–7.41 (m, 12H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 55.3, 66.8, 68.3, 69.6, 72.2, 73.1, 73.3, 73.6, 77.1, 95.7, 102.4, 113.5, 117.7, 127.2, 127.26, 127.34, 127.5, 127.6, 127.7, 128.0, 128.1, 128.2, 131.0, 133.3, 137.7, 137.8, 159.8; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{31}H_{35}O_7$ 519.2383, found 519.2375. Anal. Calcd for $C_{31}H_{34}O_7$: C, 71.80; H, 6.61. Found: C, 71.73; H, 6.65.

Data for (*S*)-isomer **22**: R_f 0.59 (2:1 n-hexane/AcOEt); $[\alpha]_D^{20}$ +23.4 (c 1.00, $CHCl_3$); IR (neat) 3063, 3029, 2911, 2870, 1614, 1518, 1454, 1250, 1092, 1032 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 3.58 (dd, J = 3.5, 7.7 Hz, 1H), 3.75 (dd, J = 7.1, 10.3 Hz, 1H), 3.82 (dd, J = 5.0, 10.3 Hz, 1H), 3.83 (s, 3H), 4.03 (dd, J = 6.4, 12.9 Hz, 1H), 4.21 (dd, J = 5.0, 12.9 Hz, 1H), 4.25–4.32 (m, 2H), 4.49 (dd, J = 6.2, 7.7 Hz, 1H), 4.54 (d, J = 12.0 Hz, 1H), 4.60 (d, J = 12.6 Hz, 1H), 4.63 (d, J = 12.0 Hz, 1H), 4.73 (d, J = 12.6 Hz, 1H), 4.85 (d, J = 3.5 Hz, 1H), 5.22 (dd, J = 1.6, 10.3 Hz, 1H), 5.34 (dd, J = 1.6, 17.3 Hz, 1H), 5.84 (s, 1H), 5.94 (m, 1H), 6.84–6.87 (m, 2H), 7.20–7.35 (m, 12H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 55.2, 66.5, 68.3, 69.3, 72.1, 73.3, 75.5, 76.1, 76.7, 95.9, 103.9, 113.5, 117.8, 127.38, 127.44, 127.8, 128.0, 128.1, 128.2, 129.5, 133.5, 137.9, 138.0, 160.1; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{31}H_{35}O_7$ 519.2383, found 519.2396. Anal. Calcd for $C_{31}H_{34}O_7$: C, 71.80; H, 6.61. Found: C, 71.71; H, 6.65.

Allyl 2,6-Di-*O*-benzyl-4-*O*-(4-methoxybenzyl)-α-D-galactopyranoside (13). TFA (0.26 mL, 3.38 mmol) was added to a cooled (10 °C) mixture of acetal **22** (877 mg, 1.69 mmol), $NaBH_3CN$ (164 mg, 2.54 mmol) and pulverized 4 Å molecular sieves (800 mg) in THF (12 mL). After 10 min of stirring at this temperature, the reaction was quenched with Et_3N (0.5 mL). The resulting mixture was diluted with AcOEt (15 mL) and passed through a Celite pad. The filtrate was partitioned between AcOEt (15 mL) and saturated aqueous $NaHCO_3$ (10 mL). The organic layer was successively washed with saturated aqueous $NaHCO_3$ (10 mL) and brine (2 × 10 mL), and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the

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crude product (1.45 g, slightly yellow oil), which was purified by column chromatography (silica gel 30 g, 2:1 *n*-hexane/AcOEt) to give alcohol **13** (847 mg, 96%) as a white solid. R_f 0.50 (15:1 CH_2Cl_2/acetone); mp 68.0−69.0 °C; $[\alpha]_D^{20}$ +54.8 (*c* 1.00, $CHCl_3$); IR (neat) 3437, 2927, 2872, 1613, 1516, 1249, 1101 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 2.27 (d, *J* = 4.7 Hz, 1H), 3.54 (d, *J* = 6.4 Hz, 2H), 3.78 (s, 3H), 3.79 (dd, *J* = 3.5, 9.1 Hz, 1H), 3.89−3.94 (m, 2H), 3.99 (t, *J* = 6.4 Hz, 1H), 4.07 (ddd, *J* = 4.1, 4.7, 9.1 Hz, 1H), 4.13 (dd, *J* = 5.0, 12.9 Hz, 1H), 4.43 (d, *J* = 11.7 Hz, 1H), 4.52 (d, *J* = 11.7 Hz, 1H), 4.57 (d, *J* = 11.5 Hz, 1H), 4.64 (d, *J* = 11.8 Hz, 1H), 4.68 (d, *J* = 11.8 Hz, 1H), 4.73 (d, *J* = 11.5 Hz, 1H), 4.88 (d, *J* = 3.5 Hz, 1H), 5.18 (d, *J* = 10.3 Hz, 1H), 5.29 (d, *J* = 17.0 Hz, 1H), 5.89 (m, 1H), 6.84 (m, 2H), 7.20−7.37 (m, 12H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 55.3, 68.4, 68.9, 69.2, 70.1, 72.7, 73.3, 74.6, 76.0, 77.3, 95.6, 113.5, 117.5, 127.38, 127.42, 127.6, 127.8, 128.1, 128.2, 129.6, 130.3, 133.6, 137.7, 137.8, 158.9; HRMS (FAB) *m/z* $[M + H]^+$ calcd for $C_{31}H_{37}O_7$ 521.2540, found 521.2545. Anal. Calcd for $C_{31}H_{36}O_7$: C, 71.52; H, 6.97. Found: C, 71.33; H, 6.94.

Allyl 4-*O*-(2-Azido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-*β*-D-mannopyranosyl)-2,3,6-tri-*O*-benzyl-*α*-D-galactopyranoside (23). A 1.0 M solution of TMSOTf in CH_2Cl_2 (2.25 mL, 2.25 mmol) was added to a cooled (−30 °C) mixture of diphenyl phosphate **5** (923 mg, 1.50 mmol), 4-O-unprotected galactoside **12** (810 mg, 1.65 mmol), and pulverized 4 Å molecular sieves (920 mg) in CH_2Cl_2 (15 mL). After 3 h of stirring at −30 °C, the reaction was quenched with Et_3N (3 mL). The mixture was diluted with AcOEt (25 mL) and passed through a Celite pad. The filtrate was partitioned between AcOEt (35 mL) and saturated aqueous $NaHCO_3$ (10 mL). The organic layer was successively washed with saturated aqueous $NaHCO_3$ (10 mL) and brine (2 × 10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (1.65 g, slightly yellow oil), which was purified by column chromatography (silica gel 35 g, 9:1 → 6:1 *n*-hexane/AcOEt) to give a mixture of *β*-linked disaccharide **23** and its *α*-isomer (1.192 g, 93%) as a slightly yellow oil. The anomeric ratio of the disaccharides was determined to be 8:92 by HPLC analysis [eluent, 6:1 *n*-hexane/AcOEt; flow rate, 1.0 mL/min; t_R (*α*-anomer) = 16.6 min, t_R (*β*-anomer **23**) = 26.0 min]. Separation of disaccharides by flash column chromatography (silica gel 40 g, 9:1 → 6:1 *n*-hexane/AcOEt) afforded *β*-linked disaccharide **23** (1.042 g, 81%) as a colorless oil, along with its *α*-isomer (102 mg, 8%) as a colorless oil. R_f 0.41 (4:1 *n*-hexane/AcOEt); $[\alpha]_D^{21}$ +5.88 (*c* 1.00, EtOH); IR (neat) 3030, 2868, 2106, 1497, 1454, 1379, 1098, 1057 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 3.12 (ddd, *J* = 4.9, 9.6, 10.4 Hz, 1H), 3.44 (dd, *J* = 3.6, 9.6 Hz, 1H), 3.62 (dd, *J* = 5.8, 9.5 Hz, 1H), 3.74 (dd, *J* = 6.9, 9.5 Hz, 1H), 3.77 (t, *J* = 10.4 Hz, 1H), 3.91 (t, *J* = 9.6 Hz, 1H), 3.93 (dd, *J* = 3.2, 10.1 Hz, 1H), 3.95 (dd, *J* = 2.6, 10.1 Hz, 1H), 3.96−4.04 (m, 3H), 4.11 (dd, *J* = 4.9, 10.4 Hz, 1H), 4.14−4.18 (m, 2H), 4.53 (s, 2H), 4.58 (d, *J* = 12.2 Hz, 1H), 4.59 (d, *J* = 11.3 Hz, 1H), 4.67 (d, *J* = 12.2 Hz, 2H), 4.76 (d, *J* = 12.2 Hz, 1H), 4.77 (d, *J* = 3.6 Hz, 1H), 4.89 (d, *J* = 3.2 Hz, 1H), 4.92 (d, *J* = 11.3 Hz, 1H), 5.21 (dd, *J* = 1.4, 10.1 Hz, 1H), 5.30 (dd, *J* = 1.4, 17.2 Hz, 1H), 5.54 (s, 1H), 5.92 (m, 1H), 7.25−7.36 (m, 23H), 7.45 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 63.1, 67.3, 68.2, 68.3, 68.7, 68.8, 72.5, 73.1, 73.3, 73.9, 76.2, 76.5, 77.9, 78.0, 95.7, 101.2, 101.5, 117.9, 125.7, 127.1, 127.27, 127.34, 127.49, 127.53, 127.6, 127.8, 127.9, 128.09, 128.13, 128.3, 128.7, 133.5, 136.9, 137.5, 137.9, 138.0, 138.2; HRMS (FAB) *m/z* $[M + H]^+$ calcd for $C_{50}H_{54}N_3O_{10}$ 856.3809, found 856.3815. Anal. Calcd for $C_{50}H_{53}N_3O_{10}$: C, 70.16; H, 6.24; N, 4.91. Found: C, 69.94; H, 6.25; N, 4.89.

Data for *α*-isomer: R_f 0.45 (4:1 *n*-hexane/AcOEt); $[\alpha]_D^{23}$ +55.7 (*c* 1.02, CH_2Cl_2); IR (neat) 3032, 2106, 1735, 1454, 1371 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 3.38 (t, *J* = 9.1 Hz, 1H), 3.46 (dd, *J* = 5.4, 9.1 Hz, 1H), 3.58 (t, *J* = 10.0 Hz, 1H), 3.70 (dd, *J* = 5.0, 10.0 Hz, 1H), 3.82 (dd, *J* = 3.7, 10.0 Hz, 1H), 3.84 (dd, *J* = 2.3, 3.6 Hz, 1H), 3.85 (dd, *J* = 2.7, 10.0 Hz, 1H), 3.88 (ddd, *J* = 3.6, 5.4, 9.1 Hz, 1H), 3.96 (dd, *J* = 3.6, 10.0 Hz, 1H), 3.97 (m, 1H), 4.03 (t, *J* = 10.0 Hz, 1H), 4.08 (m, 1H), 4.11 (dd, *J* = 2.7, 3.6 Hz, 1H), 4.22 (dt, *J* = 5.0, 10.0 Hz, 1H), 4.47 (d, *J* = 11.3 Hz, 1H), 4.53 (d, *J* = 11.3 Hz, 1H), 4.64 (d, *J* = 11.8 Hz, 1H), 4.73 (d, *J* = 12.2 Hz, 1H), 4.74 (d, *J* = 12.2 Hz, 2H), 4.79 (d, *J* = 3.7 Hz, 1H), 4.80 (d, *J* = 2.3 Hz, 1H), 4.84 (d, *J* = 12.2 Hz, 1H), 4.90 (d, *J* = 11.8 Hz, 1H), 5.22 (dd, *J* = 1.8, 10.5 Hz, 1H), 5.29 (dd, *J* = 1.8, 17.4 Hz, 1H), 5.56 (s, 1H), 5.92 (m, 1H), 7.18−7.41 (m, 23H), 7.48 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 63.0, 64.1, 67.8, 68.40, 68.44, 72.9, 73.2, 73.6, 73.9, 74.4, 76.1, 76.5, 76.9, 79.2, 96.4, 100.4, 101.2, 118.0, 126.0, 127.3, 127.4, 127.6, 127.97, 128.03, 128.18, 128.24, 128.28, 128.32, 128.5, 128.6, 133.6, 136.9, 137.6, 138.1, 138.2; HRMS (FAB) *m/z* $[M + H]^+$ calcd for $C_{50}H_{54}N_3O_{10}$ 856.3809, found 856.3805. Anal. Calcd for $C_{50}H_{53}N_3O_{10}$: C, 70.16; H, 6.24; N, 4.91. Found: C, 70.03; H, 6.30; N, 4.85.

Allyl 4-*O*-(2-Acetamido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-*β*-D-mannopyranosyl)-2,3,6-tri-*O*-benzyl-*α*-D-galactopyranoside (24). Triphenylphosphine (309 mg, 1.18 mmol) was added to a stirred solution of azide **23** (916 mg, 1.07 mmol) in THF (10 mL). After 30 min of stirring, H_2O (0.2 mL) was added, and the mixture was refluxed for 10 h. The reaction mixture was partitioned between AcOEt (30 mL) and brine (15 mL), and the organic layer was dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the colorless oil, which was used without further purification.

Acetic anhydride (0.30 mL, 3.18 mmol) was added to a stirred solution of the crude amine in pyridine (4 mL). After 3 h of stirring, the reaction mixture was partitioned between AcOEt (30 mL) and 10% aqueous HCl (15 mL). The organic layer was successively washed with H_2O (5 mL), saturated aqueous $NaHCO_3$ (10 mL), and brine (10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (1.59 g, slightly yellow amorphous solid), which was purified by column chromatography (silica gel 30 g, 2:1 → 2:3 *n*-hexane/AcOEt) to give acetamide **24** (908 mg, 97%) as a white amorphous solid. R_f 0.31 (5:1 toluene/acetone); $[\alpha]_D^{21}$ +7.62 (*c* 1.00, $CHCl_3$); IR (KBr) 3437, 3335, 2922, 2870, 1679, 1454, 1372, 1099, 1027 cm^{-1}; ^1H NMR (400 MHz, acetone-d_6) δ 1.90 (br s, 3H), 3.32 (ddd, *J* = 4.7, 9.7, 10.0 Hz, 1H), 3.53 (dd, *J* = 4.4, 9.7 Hz, 1H), 3.58 (dd, *J* = 6.1, 10.0 Hz, 1H), 3.68 (t, *J* = 10.0 Hz, 1H), 3.72 (dd, *J* = 5.6, 10.0 Hz, 1H), 3.85 (t, *J* = 9.7 Hz, 1H), 3.95−4.01 (m, 4H), 4.07 (dd, *J* = 4.7, 10.0 Hz, 1H), 4.15 (ddd, *J* = 1.5, 5.0, 13.2 Hz, 1H), 4.33 (d, *J* = 12.0 Hz, 1H), 4.37 (br s, 1H), 4.51 (d, *J* = 11.7 Hz, 1H), 4.56 (d, *J* = 11.7 Hz, 1H), 4.69 (d, *J* = 11.5 Hz, 1H), 4.70 (d, *J* = 12.0 Hz, 1H), 4.78 (d, *J* = 11.7 Hz, 1H), 4.80 (d, *J* = 11.5 Hz, 1H), 4.82 (d, *J* = 2.7 Hz, 1H), 4.85 (d, *J* = 11.7 Hz, 1H), 5.03 (d, *J* = 1.8 Hz, 1H), 5.08 (ddd, *J* = 1.8, 4.4, 9.7 Hz, 1H), 5.12 (ddd, *J* = 1.5, 1.8, 11.7 Hz, 1H), 5.32 (dd, *J* = 1.8, 17.3 Hz, 1H), 5.89 (s, 1H), 5.94 (m, 1H), 6.99 (d, *J* = 9.7 Hz, 1H), 7.20−7.36 (m, 19H), 7.44−7.48 (m, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 23.3, 50.2, 66.5, 68.2, 68.7, 68.8, 69.3, 71.7, 73.2, 73.6, 73.9, 75.7, 76.2, 76.7, 78.2, 79.0, 96.0, 100.8, 101.2, 117.8, 125.7, 127.1, 127.2, 127.4, 127.5, 127.6, 127.8, 127.9, 127.96, 127.98, 128.1, 128.3, 128.7, 133.6, 136.9, 137.8, 137.9, 138.0, 138.2, 170.0; HRMS (FAB) *m/z* $[M + H]^+$ calcd for $C_{52}H_{58}NO_{11}$ 872.4010, found 872.4024.

4-*O*-(2-Acetamido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-*β*-D-mannopyranosyl)-2,3,6-tri-*O*-benzyl-D-galactopyranose (25). Potassium *tert*-butoxide (140 mg, 1.25 mmol) was added to a stirred solution of allyl glycoside **24** (906 mg, 1.04 mmol) in DMSO (5 mL), and the mixture was heated at 100 °C for 20 min. After cooling to room temperature, the reaction was quenched with H_2O (1 mL), and the mixture was partitioned between AcOEt (25 mL) and saturated aqueous NH_4Cl (5 mL). The organic layer was washed with brine (2 × 10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the slightly yellow oil, which was used without further purification.

NBS (203 mg, 1.14 mmol) was added to a stirred solution of the crude prop-1-enyl glycoside in 20:1 THF/H_2O (10.5 mL). After 5 min of stirring, the volatile elements were removed in vacuo, and the residue was partitioned between AcOEt (30 mL) and 10% aqueous $Na_2S_2O_3$ (10 mL). The organic layer was successively washed with H_2O (2 × 10 mL) and brine (10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (938 mg, slightly yellow oil), which was purified by column chromatography (silica gel 35 g, 1:1 *n*-hexane/AcOEt with 0.5% Et_3N) to give hemiacetal **25** (726 mg, 84%, *α*:*β* = 55:45) as a slightly yellow amorphous solid. R_f 0.23 (3:1 toluene/acetone); $[\alpha]_D^{22}$ −21.4 (*c* 1.01, MeCN); IR (Nujol) 3328, 1659, 1496, 1212 cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 1.90 (s, 3H), 3.30−3.35 (m, 1H),

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3.52−3.57 (m, 1.55H), 3.62 (dd, J = 5.2, 8.6 Hz, 0.45H), 3.66−3.76 (m, 3.35H), 3.84−3.89 (m, 1H), 3.95 (dd, J = 3.5, 10.3 Hz, 0.55H), 4.02 (dd, J = 2.9, 10.3 Hz, 0.55H), 4.05−4.20 (m, 1H), 4.19 (br dd, J = 5.7, 6.3 Hz, 0.55H), 4.31 (br s, 0.45H), 4.33−4.38 (m, 1.55H), 4.49−4.57 (m, 2.1H), 4.63 (br dd, J = 6.3, 6.9 Hz, 0.45H), 4.70−4.86 (m, 4.45H), 4.98 (d, J = 11.5 Hz, 0.45H), 5.01−5.10 (m, 1H), 5.04 (d, J = 1.7 Hz, 0.55H), 5.07 (d, J = 1.7 Hz, 0.45H), 5.20 (dd, J = 3.5, 4.0 Hz, 0.55H), 5.34 (d, J = 4.0 Hz, 0.55H), 5.50 (s, 0.55H), 5.51 (s, 0.45H), 5.91 (d, J = 6.9 Hz, 0.45H), 6.94 (d, J = 9.8 Hz, 0.45H), 6.98 (d, J = 9.7 Hz, 0.55H), 7.20−7.48 (m, 25H); ^{13}C NMR (100 MHz, acetone-d_6) δ 22.4, 24.2, 51.75, 51.79, 68.8, 70.0, 70.3, 71.3, 72.2, 74.2, 74.3, 74.37, 74.44, 74.5, 75.6, 76.2, 77.4, 78.4, 78.7, 79.7, 79.9, 82.5, 83.6, 93.1, 99.4, 102.4, 102.8, 102.9, 126.7, 127.7, 128.46, 128.52, 128.6, 128.7, 128.77, 128.79, 128.85, 128.96, 128.99, 129.1, 129.2, 129.3, 129.4, 129.5, 129.6, 129.8, 130.1, 130.3, 139.6, 140.4, 140.6, 141.0, 141.1, 171.0, 171.1; HRMS (FAB) m/z [M + H]$^+$ calcd for $C_{49}H_{54}NO_{11}$ 832.3697, found 832.3685. Anal. Calcd for $C_{49}H_{53}NO_{11}$: C, 70.74; H, 6.42; N, 1.62. Found: C, 71.04; H, 6.59; N, 1.65.

4-*O*-(2-Acetamido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-β-D-mannopyranosyl)-2,3,6-tri-*O*-benzyl-D-galactopyranosyl Diphenyl Phosphate (9). Diphenyl chlorophosphite (50 mg, 0.20 mmol) was added to an ice-cooled (0 °C) solution of hemiacetal **25** (83 mg, 0.10 mmol) and Et_3N (55 μL, 0.39 mmol) in CH_2Cl_2 (2 mL). After 10 min of stirring at 0 °C, the reaction was quenched by addition of a piece of crushed ice, followed by stirring at room temperature for 10 min. The mixture was partitioned between AcOEt (15 mL) and saturated aqueous $NaHCO_3$ (5 mL), and the organic layer was washed with brine (2 × 10 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (164 mg, slightly yellow oil), which was purified by column chromatography (Wako gel 4 g, 2:1 → 1:1 n-hexane/AcOEt) to give the corresponding diphenyl phosphite (92 mg, 88%) as a colorless oil.

Potassium peroxymonosulfate (Oxone, 162 mg, 0.26 mmol) was added to an ice-cooled (0 °C) solution of diphenyl phosphite (92 mg, 0.088 mmol) and $NaHCO_3$ (36.9 mg, 0.44 mmol) in 2:1 acetone/H_2O (3 mL). After 30 min of stirring at 0 °C, the reaction mixture was diluted with Et_2O (2 mL) and partitioned between AcOEt (20 mL) and saturated aqueous $NaHCO_3$ (5 mL). The organic layer was successively washed with saturated aqueous $NaHCO_3$ (5 mL) and brine (2 × 5 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (95 mg), which was purified by column chromatography (Wako gel 2.5 g, 1:1 n-hexane/AcOEt with 2% Et_3N) to give diphenyl phosphate **9** (86.2 mg, 92%, α:β = 67:33) as a colorless foam. R_f 0.39 (α-anomer), 0.59 (β-anomer) (3:1 toluene/acetone); $[\alpha]_D^{22}$ +7.67 (c 1.05, MeCN); IR (KBr) 3331, 3063, 3032, 2871, 1676, 1591, 1490, 1190 cm^{-1}; ^{1}H NMR (500 MHz, acetone-d_6) δ 1.89 (br s, 1H), 1.90 (br s, 2H), 3.32−3.36 (m, 1H), 3.44 (dd, J = 5.7, 9.2 Hz, 0.67H), 3.55−3.60 (m, 1H), 3.63 (dd, J = 6.5, 10.1 Hz, 0.33H), 3.67−3.73 (m, 1.67H), 3.77 (dd, J = 5.8, 10.1 Hz, 0.33H), 3.85−3.91 (m, 1.33H), 3.95−4.00 (m, 1.33H), 4.06−4.14 (m, 1.67H), 4.18 (dt, J = 9.7, 3.4 Hz, 0.67H), 4.34 (d, J = 12.0 Hz, 0.67H), 4.38 (d, J = 12.0 Hz, 0.33H), 4.40 (br d, J = 1.7 Hz, 0.33H), 4.45 (br d, J = 0.9 Hz, 0.67H), 4.48 (d, J = 12.1 Hz, 0.67H), 4.52 (d, J = 12.1 Hz, 0.67H), 4.57 (s, 0.67H), 4.68−4.72 (m, 1.33H), 4.76−4.85 (m, 3.67H), 5.03 (d, J = 1.7 Hz, 0.67H), 5.04 (d, J = 1.7 Hz, 0.33H), 5.06−5.14 (m, 1H), 5.42 (dd, J = 6.9, 8.0 Hz, 0.33H), 5.51 (s, 0.67H), 5.52 (s, 0.33H), 5.99 (dd, J = 3.4, 6.3 Hz, 0.67H), 7.03 (br d, J = 10.3 Hz, 0.33H), 7.04 (br d, J = 10.3 Hz, 0.67H), 7.15−7.48 (m, 35H); ^{13}C NMR (100 MHz, acetone-d_6) δ 22.5, 26.1, 51.2, 68.3, 69.5, 69.9, 70.0, 70.3, 70.9, 71.2, 71.7, 72.8, 73.8, 73.9, 74.7, 75.0, 75.3, 76.1, 76.2, 76.7 (d, J_{C-P} = 6.7 Hz), 77.8, 78.8, 79.4, 80.0 (d, J_{C-P} = 9.3 Hz), 82.7, 99.5 (d, J_{C-P} = 5.8 Hz), 101.2 (d, J_{C-P} = 5.7 Hz), 102.2, 102.5, 116.4, 120.4, 121.18, 121.21, 121.31, 121.35, 121.55, 121.59, 121.67, 121.70, 121.85, 121.89, 126.0, 126.37, 126.40, 126.47, 126.51, 127.4, 128.2, 128.27, 128.33, 128.38, 128.44, 128.50, 128.53, 128.6, 128.7, 128.8, 128.9, 129.0, 129.1, 129.2, 129.25, 129.33, 129.38, 129.42, 129.5, 129.6, 129.8, 130.5, 130.7, 130.75, 130.82, 130.9, 131.0, 139.37, 139.40, 139.8, 139.9, 140.0, 140.07, 140.10, 140.16, 140.19, 140.25, 151.80, 151.85, 151.91, 151.95, 151.97, 152.01, 152.05, 152.36, 152.42, 158.6, 171.1; ^{31}P NMR (202.5 MHz, acetone-d_6) δ −11.3 (α-anomer), −11.6 (β-anomer); HRMS (ESI) m/z [M + Na]$^+$ calcd for $C_{61}H_{62}NO_{14}PNa$ 1086.3806, found 1086.3804. Anal. Calcd for $C_{61}H_{62}NO_{14}P$: C, 68.85; H, 5.87; N, 1.32. Found: C, 68.68; H, 6.13; N, 1.33.

Allyl 3-*O*-(2-Acetamido-3-*O*-acetyl-4,6-di-*O*-benzyl-2-deoxy-β-D-galactopyranosyl)-2,6-di-*O*-benzyl-4-*O*-(4-methoxybenzyl)-α-D-galactopyranoside (26). A 1.0 M solution of Tf_2NH in EtCN (0.55 mL, 0.55 mmol) was added to a cooled (−78 °C) mixture of diethyl phosphite **11** (280 mg, 0.50 mmol), 3-O-unprotected galactoside **13** (776 mg, 1.49 mmol), and pulverized 4 Å molecular sieves (500 mg) in CH_2Cl_2 (5 mL). After 2 h of stirring at this temperature, the reaction was quenched with Et_3N (0.3 mL). The mixture was diluted with AcOEt (5 mL) and passed through a Celite pad. The filtrate was partitioned between AcOEt (50 mL) and saturated aqueous $NaHCO_3$ (10 mL). The organic layer was successively washed with saturated aqueous $NaHCO_3$ (10 mL) and brine (2 × 10 mL), and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (1.25 g, slightly yellow oil), which was purified by flash column chromatography (silica gel 40 g, 8:1 → 6:1 toluene/acetone) to give β-linked disaccharide **26** (305 mg, 65%) as a white solid, along with recovered alcohol **13** (594 mg). R_f 0.25 (4:1 toluene/acetone); mp 117.5−118.5 °C (colorless plates from AcOEt/n-hexane); $[\alpha]_D^{21}$ −8.8 (c 1.02, $CHCl_3$); IR (KBr) 3259, 2932, 2862, 1743, 1646, 1510, 1241, 735, 697 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 1.71 (s, 3H), 2.00 (s, 3H), 3.34 (dd, J = 5.7, 9.8 Hz, 1H), 3.48 (dd, J = 6.3, 9.8 Hz, 1H), 3.56 (dd, J = 5.2, 8.6 Hz, 1H), 3.65−3.72 (m, 2H), 3.73 (s, 3H), 3.89 (br dd, J = 5.7, 6.3 Hz, 1H), 3.91−3.98 (m, 4H), 4.08 (dd, J = 2.9, 10.3 Hz, 1H), 4.10 (ddd, J = 1.1, 5.1, 13.1 Hz, 1H), 4.34 (d, J = 12.0 Hz, 1H), 4.38 (d, J = 11.7 Hz, 1H), 4.43 (d, J = 11.7 Hz, 1H), 4.45 (d, J = 12.0 Hz, 1H), 4.50 (ddd, J = 8.5, 9.3, 11.1 Hz, 1H), 4.54 (d, J = 11.7 Hz, 1H), 4.60 (d, J = 12.0 Hz, 1H), 4.61 (d, J = 11.5 Hz, 1H), 4.65 (d, J = 12.0 Hz, 1H), 4.72 (d, J = 8.5 Hz, 1H), 4.78 (d, J = 3.6 Hz, 1H), 4.82 (d, J = 11.7 Hz, 1H), 4.86 (d, J = 11.5 Hz, 1H), 4.93 (dd, J = 2.9, 11.1 Hz, 1H), 5.16 (dd, J = 1.7, 10.3 Hz, 1H), 5.17 (d, J = 9.3 Hz, 1H), 5.26 (ddd, J = 1.1, 1.7, 17.2 Hz, 1H), 5.88 (m, 1H), 6.71 (d, J = 6.9 Hz, 2H), 7.18−7.38 (m, 22H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 21.0, 23.4, 51.6, 55.2, 68.1, 68.2, 69.5, 72.7, 73.1, 73.2, 73.4, 73.7, 73.9, 74.9, 75.5, 76.2, 78.5, 95.8, 102.8, 113.2, 117.8, 127.19, 127.22, 127.25, 127.3, 127.4, 127.5, 127.6, 128.0, 128.2, 128.3, 130.4, 130.6, 133.6, 137.5, 137.86, 137.94, 138.1, 158.6, 169.3, 170.5; HRMS (FAB) m/z [M + H]$^+$ calcd for $C_{55}H_{64}NO_{13}$ 946.4378, found 946.4385. Anal. Calcd for $C_{55}H_{63}NO_{13}$: C, 69.82; H, 6.71; N, 1.48. Found: C, 69.65; H, 6.78; N, 1.42.

Allyl 3-*O*-(2-Acetamido-3-*O*-acetyl-4,6-di-*O*-benzyl-2-deoxy-β-D-galactopyranosyl)-2,6-di-*O*-benzyl-α-D-galactopyranoside (10). DDQ (83 mg, 0.37 mmol) was added in three portions to a stirred biphasic mixture of PMB ether **26** (295 mg, 0.31 mmol) in 20:1 CH_2Cl_2/pH 7 phosphate buffer (3.15 mL). After 1 h of stirring, the reaction mixture was diluted with AcOEt (5 mL) and passed through a Celite pad. The filtrate was partitioned between AcOEt (10 mL) and saturated aqueous $NaHCO_3$ (5 mL). The organic layer was successively washed with saturated aqueous $NaHCO_3$ (3 × 5 mL) and brine (5 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (362 mg), which was purified by column chromatography (silica gel 12 g, 5:1 → 3:1 toluene/acetone) to give alcohol **10** (232 mg, 90%) as a white amorphous solid. R_f 0.25 (4:1 toluene/acetone); $[\alpha]_D^{23}$ +22.0 (c 1.02, $CHCl_3$); IR (KBr) 3472, 3290, 2929, 2865, 1725, 1660, 1496, 1314, 697 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 1.73 (s, 3H), 1.99 (s, 3H), 2.78 (s, 1H), 3.53 (dd, J = 6.3, 9.2 Hz, 1H), 3.57 (t, J = 9.2 Hz, 1H), 3.669 (dd, J = 6.3, 9.2 Hz, 1H), 3.671 (dd, J = 5.2, 9.2 Hz, 1H), 3.68 (dd, J = 6.3, 9.2 Hz, 1H), 3.84 (dd, J = 3.4, 10.3 Hz, 1H), 3.89 (d, J = 2.9 Hz, 1H), 3.97 (br dd, J = 5.2, 6.3 Hz, 1H), 4.00 (m, 1H), 4.01 (dd, J = 1.2, 10.3 Hz, 1H), 4.09 (dd, J = 1.2, 1.7 Hz, 1H), 4.15 (dd, J = 5.2, 13.2 Hz, 1H), 4.38 (ddd, J = 8.6, 9.2, 11.2 Hz, 1H), 4.39 (d, J = 12.0 Hz, 1H), 4.43 (d, J = 12.0 Hz, 1H), 4.50 (d, J = 12.0 Hz, 1H), 4.53 (d, J = 11.5 Hz, 1H), 4.56 (d, J = 12.0 Hz, 1H), 4.61 (d, J = 12.1 Hz, 1H), 4.64 (d, J = 12.1 Hz, 1H), 4.70 (d, J = 8.6 Hz, 1H), 4.71 (d, J = 11.5 Hz, 1H), 4.82 (d, J = 3.4 Hz, 1H), 4.95 (dd, J = 2.9, 11.2 Hz, 1H), 5.15 (d, J = 9.2 Hz, 1H), 5.19 (dd, J = 1.7, 11.3 Hz, 1H), 5.29 (dd, J = 1.7, 17.1 Hz, 1H), 5.92 (m, 1H), 7.23−7.47 (m, 20H); ^{13}C NMR (100 MHz,

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$CDCl_3$) δ 20.6, 23.1, 51.0, 67.9, 68.4, 68.9, 69.4, 72.7, 73.0, 73.09, 73.13, 73.2, 74.7, 75.0, 78.6, 95.7, 102.2, 117.8, 127.2, 127.4, 127.48, 127.53, 127.96, 128.00, 128.1, 128.16, 128.22, 137.3, 137.4, 138.0, 138.3, 170.0, 170.7; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{47}H_{56}NO_{12}$ 826.3803, found 826.3797. Anal. Calcd for $C_{47}H_{55}NO_{12}$: C, 68.35; H, 6.71; N, 1.70. Found: C, 68.25; H, 6.70; N, 1.67.

Allyl (2-Acetamido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-β-D-mannopyranosyl)-(1→4)-(2,3,6-tri-*O*-benzyl-α-D-galactopyranosyl)-(1→4)-[2-acetamido-3-*O*-acetyl-4,6-di-*O*-benzyl-2-deoxy-β-D-galactopyranosyl-(1→3)]-2,6-di-*O*-benzyl-α-D-galactopyranoside (8). A 1.0 M solution of $TMSClO_4$ in dioxane (0.12 mL, 0.12 mmol) was added to an ice-cooled (0 °C) mixture of diphenyl phosphate **9** (42.6 mg, 0.04 mmol), alcohol **10** (66.1 mg, 0.08 mmol), and pulverized 5 Å molecular sieves (80 mg) in CH_2Cl_2 (0.6 mL). After 30 min of stirring, the reaction was quenched with Et_3N (0.2 mL). The mixture was diluted with AcOEt (4 mL) and passed through a Celite pad. The filtrate was partitioned between AcOEt (20 mL) and saturated aqueous $NaHCO_3$ (5 mL). The organic layer was successively washed with saturated aqueous $NaHCO_3$ (5 mL) and brine (2 × 5 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (104.5 mg, white solid), which was purified by flash column chromatography (silica gel 12 g, 5:1 toluene/acetone) to give tetrasaccharide **8** (49.5 mg, 75%) as a colorless oil. R_f 0.50 (2:1 toluene/acetone); $[\alpha]_D^{22}$ +29.0 (c 1.03, $CHCl_3$); IR (KBr) 3329, 3031, 2925, 2870, 1746, 1681, 1454, 1368, 1239 cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 1.56 (s, 3H), 1.90 (br s, 3H), 1.91 (s, 3H), 3.33 (ddd, J = 4.6, 9.2, 9.7 Hz, 1H), 3.50 (dd, J = 6.9, 9.7 Hz, 1H), 3.53 (dd, J = 4.9, 10.2 Hz, 1H), 3.48−3.70 (m, 3H), 3.79−4.05 (m, 14H), 4.09 (ddd, J = 1.7, 5.2, 13.2 Hz, 1H), 4.21 (dd, J = 2.9, 10.3 Hz, 1H), 4.25 (br d, J = 2.9 Hz, 1H), 4.29−4.35 (m, 3H), 4.45 (d, J = 12.0 Hz, 1H), 4.51 (d, J = 12.0 Hz, 1H), 4.51−4.60 (m, 5H), 4.64−4.83 (m, 8H), 5.05−5.16 (m, 4H), 5.19 (d, J = 8.6 Hz, 1H), 5.28 (dt, J = 17.2, 1.7 Hz, 1H), 5.38 (dd, J = 3.5, 10.8 Hz, 1H), 5.50 (s, 1H), 5.90 (m, 1H), 6.64 (br d, J = 8.0 Hz, 1H), 7.00 (br d, J = 9.8 Hz, 1H), 7.21−7.51 (m, 45H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 20.7, 23.09, 23.11, 50.4, 52.4, 66.5, 67.9, 68.2, 68.6, 68.8, 69.7, 71.7, 72.4, 72.7, 72.8, 72.9, 73.1, 73.2, 74.0, 74.1, 75.0, 75.1, 75.4, 75.9, 76.6, 76.8, 78.6, 95.4, 97.9, 101.1, 101.3, 101.6, 118.1, 125.2, 126.0, 127.3, 127.38, 127.42, 127.5, 127.55, 127.64, 127.7, 127.8, 128.0, 128.13, 128.15, 128.2, 128.26, 128.34, 128.4, 128.5, 128.9, 129.0, 133.8, 137.2, 137.4, 137.8, 138.0, 138.3, 138.4, 138.5, 138.6, 138.7, 169.5, 170.3, 170.5; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{96}H_{107}N_2O_{22}$ 1639.7316, found 1639.7300. Anal. Calcd for $C_{96}H_{106}N_2O_{22}$: C, 70.31; H, 6.52; N, 1.71. Found: C, 70.08; H, 6.56; N, 1.70.

Allyl (2-Acetamido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-β-D-mannopyranosyl)-(1→4)-(2,3,6-tri-*O*-benzyl-α-D-galactopyranosyl)-(1→4)-[2-acetamido-4,6-di-*O*-benzyl-2-deoxy-β-D-galactopyranosyl-(1→3)]-2,6-di-*O*-benzyl-α-D-galactopyranoside (27). A 1.0 M solution of NaOMe in MeOH (35 μL, 0.035 mmol) was added to an ice-cooled (0 °C) solution of tetrasaccharide **8** (56.5 mg, 0.034 mmol) in MeOH (0.5 mL). After 1.5 h of stirring at this temperature, the reaction mixture was neutralized with Amberlite IR-120 acidic resin. Filtration and evaporation in vacuo furnished alcohol **27** (56.7 mg), which was used without further purification. An analytical sample was obtained by silica gel column chromatography with 4:1 → 3:1 toluene/acetone. R_f 0.44 (2:1 toluene/acetone); $[\alpha]_D^{22}$ +28.0 (c 0.97, $CHCl_3$); IR (neat) 3333, 3063, 3031, 2870, 1667, 1539, 1496, 1099 cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 1.54 (s, 3H), 1.89 (br s, 3H), 3.32 (t, J = 11.8 Hz, 1H), 3.49 (dd, J = 6.3, 9.5 Hz, 1H), 3.51 (m, 1H), 3.63−3.75 (m, 4H), 3.77−4.05 (m, 14H), 4.13 (dd, J = 5.0, 13.1 Hz, 1H), 4.18 (dd, J = 2.7, 10.4 Hz, 1H), 4.20 (br d, J = 2.7 Hz, 1H), 4.28 (s, 2H), 4.31 (d, J = 11.8 Hz, 1H), 4.32 (d, J = 11.3 Hz, 1H), 4.45−4.52 (m, 3H), 4.53 (d, J = 11.3 Hz, 1H), 4.64 (d, J = 11.8 Hz, 1H), 4.70 (d, J = 11.8 Hz, 1H), 4.71−4.75 (m, 3H), 4.72 (d, J = 11.3 Hz, 1H), 4.77 (d, J = 11.3 Hz, 1H), 4.84 (d, J = 7.5 Hz, 1H), 4.86 (d, J = 11.3 Hz, 1H), 4.96 (d, J = 3.2 Hz, 1H), 5.04−5.07 (m, 4H), 5.09 (d, J = 10.4 Hz, 1H), 5.29 (dd, J = 1.4, 17.2 Hz, 1H), 5.39 (br s, 1H), 5.52 (s, 1H), 5.92 (m, 1H), 6.74 (br d, J = 3.2 Hz, 1H), 7.00 (br d, J = 9.9 Hz, 1H), 7.21−7.50 (m, 45H); ^{13}C NMR (100 MHz, acetone-d_6) δ 23.4, 23.6, 51.3, 57.3, 68.3, 69.2, 69.48, 69.54, 69.9, 70.3, 71.2, 71.8, 72.2, 73.6, 73.7, 73.8, 73.9, 75.0, 75.1, 75.7, 76.3, 76.4, 77.1, 77.7, 78.0, 78.2, 78.4, 78.6, 79.4, 80.0, 97.1, 99.8, 102.5, 103.0, 104.1, 117.6, 127.4, 128.2, 128.3, 128.35, 128.40, 128.5, 128.58, 128.64, 128.67, 128.71, 129.0, 129.07, 129.13, 129.2, 129.28, 129.30, 129.32, 129.4, 129.51, 129.53, 129.8, 130.0, 135.9, 139.4, 139.9, 140.03, 140.05, 140.3, 140.5, 140.6, 140.8, 170.8, 173.1; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{94}H_{105}N_2O_{21}$ 1597.7210, found 1597.7201.

(2-Acetamido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-β-D-mannopyranosyl)-(1→4)-(2,3,6-tri-*O*-benzyl-α-D-galactopyranosyl)-(1→4)-[2-acetamido-4,6-di-*O*-benzyl-2-deoxy-β-D-galactopyranosyl-(1→3)]-2,6-di-*O*-benzyl-D-galactopyranose (28). Tris(triphenylphosphine)rhodium(I) chloride (4.8 mg, 5.2 μmol) was added to a stirred solution of allyl glycoside **27** (56.7 mg, 0.034 mmol) and 1,4-diazabicyclo[2.2.2]octane (DABCO; 1.2 mg, 0.011 mmol) in 9:1 EtOH/H_2O (1 mL), and the mixture was heated at 100 °C for 2 h. After cooling to room temperature, the reaction mixture was diluted with AcOEt (2 mL) and passed through a Celite pad. The filtrate was partitioned between AcOEt (15 mL) and saturated aqueous NH_4Cl (3 mL). The organic layer was washed with brine (2 × 3 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the colorless oil, which was used without further purification.

NBS (7.6 mg, 0.041 mmol) was added to a stirred solution of the crude prop-1-enyl glycoside in 20:1 THF/H_2O (1.26 mL). After 10 min of stirring, the volatile elements were removed in vacuo, and the residue was partitioned between AcOEt (15 mL) and 10% aqueous $Na_2S_2O_3$ (3 mL). The organic layer was successively washed with 10% aqueous $Na_2S_2O_3$ (3 mL) and brine (2 × 3 mL) and dried over anhydrous Na_2SO_4. Filtration and evaporation in vacuo furnished the crude product (67.2 mg, slightly yellow oil), which was purified by column chromatography (silica gel 6 g, 4:1 CH_2Cl_2/acetone with 1% Et_3N) to give hemiacetal **28** (44.2 mg, 82% for three steps, α:β = 79:21) as a colorless viscous oil. R_f 0.41 (2:1 toluene/acetone); $[\alpha]_D^{20}$ +21.2 (c 1.01, $CHCl_3$); IR (Nujol) 3324, 2922, 2853, 1657, 1455, 1376, 1076, 734, 696 cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 1.50 (s, 2.4H), 1.52 (s, 0.6H), 1.88 (br s, 3H), 3.32 (m, 1H), 3.42 (dd, J = 6.3, 9.7 Hz, 0.8H), 3.52 (m, 1H), 3.58 (m, 0.2H), 3.64−3.73 (m, 4.4H), 3.76−3.91 (m, 7.8H), 3.95−4.03 (m, 3H), 4.17 (t, J = 6.3 Hz, 1H), 4.20 (t, J = 6.3 Hz, 0.2H), 4.24−4.33 (m, 5.4H), 4.39−4.55 (m, 5.4H), 4.60−4.80 (m, 7.2H), 4.84−4.86 (m, 1.6H), 4.91 (d, J = 8.0 Hz, 0.2H), 5.01−5.11 (m, 4H), 5.18 (d, J = 3.4 Hz, 0.2H), 5.34 (t, J = 3.7 Hz, 0.8H), 5.42 (br s, 0.8H), 5.48 (d, J = 3.4 Hz, 0.8H), 5.51 (s, 1H), 6.00 (d, J = 6.3 Hz, 0.2H), 6.63 (d, J = 4.6 Hz, 0.2H), 6.74 (d, J = 4.0 Hz, 0.8H), 6.98 (d, J = 9.7 Hz, 1H), 7.21−7.49 (m, 45H); ^{13}C NMR (100 MHz, acetone-d_6) δ 23.38, 23.44, 23.6, 51.3, 55.8, 57.2, 57.4, 68.3, 68.6, 69.5, 69.8, 69.9, 70.0, 70.06, 70.10, 70.3, 70.6, 71.8, 73.58, 73.64, 73.7, 73.8, 73.9, 74.0, 74.2, 74.6, 74.9, 75.0, 75.1, 75.2, 75.7, 76.3, 76.4, 76.5, 77.1, 77.3, 77.66, 77.72, 78.0, 78.2, 78.3, 78.4, 79.4, 79.6, 80.1, 80.3, 80.9, 91.7, 99.2, 99.7, 99.8, 102.5, 102.9, 103.0, 103.1, 104.1, 127.4, 128.2, 128.25, 128.34, 128.4, 128.5, 128.6, 128.66, 128.69, 129.0, 129.09, 129.14, 129.2, 129.29, 129.33, 129.4, 129.50, 129.54, 129.8, 139.4, 139.85, 139.87, 139.96, 140.00, 140.04, 140.25, 140.29, 140.32, 140.55, 140.61, 140.76, 140.81, 170.8, 173.18, 173.22; HRMS (ESI) m/z $[M + Na]^+$ calcd for $C_{91}H_{100}N_2O_{21}Na$ 1579.6716, found 1579.6704.

(2-Acetamido-2-deoxy-β-D-mannopyranosyl)-(1→4)-(α-D-galactopyranosyl)-(1→4)-[2-acetamido-2-deoxy-β-D-galactopyranosyl-(1→3)]-D-galactopyranose (1). Palladium on carbon (10%, 10 mg) was added to a stirred solution of hemiacetal **28** (21.5 mg, 0.014 mmol) in MeOH (1 mL), and the mixture was stirred under 1 atm of hydrogen for 14 h. The reaction mixture was diluted with MeOH (2 mL) and passed through a Celite pad. The filtrate was evaporated in vacuo to furnish the crude product (9.8 mg), which was purified by column chromatography (Sephadex 5 g, eluting with MeOH) to give tetrasaccharide **1** (9.7 mg, 94%, α:β = 1:1) as a colorless amorphous solid. R_f 0.13 (9:1 EtOH/H_2O); $[\alpha]_D^{21}$ +35.7 (c 0.62, MeOH); IR (KBr) 3376, 1642, 1562, 1376, 1065 cm^{-1}; ^1H NMR (500 MHz, CD_3OD) δ 1.98 (s, 1.5H), 1.99 (s, 1.5H), 2.02 (s, 3H), 3.20−3.25 (m, 1H), 3.43 (t, J = 9.7 Hz, 1H), 3.44−3.88 (m, 16H), 3.89−3.98 (m, 1.5H), 4.07 (t, J = 7.4 Hz, 0.5H), 4.12−4.16 (m, 1H), 4.20 (d, J = 2.9 Hz, 0.5H), 4.26 (d, J = 2.9 Hz, 0.5H),

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4.48 (d, J = 7.4 Hz, 0.5H), 4.51−4.55 (br q, J = 7.4 Hz, 1H), 4.58−4.64 (m, 2H), 4.82−4.84 (d, J = 3.4 Hz, 1H), 4.93 (d, J = 4.0 Hz, 0.5H), 4.97 (d, J = 3.4 Hz, 0.5H), 5.14 (d, J = 4.0 Hz, 0.5H); ^{13}C NMR (100 MHz, CD_3OD) δ 22.72, 22.73, 23.09, 23.11, 54.7, 54.8, 60.8, 61.0, 61.3, 62.6, 62.7, 68.8, 69.80, 69.83, 70.4, 70.7, 70.9, 71.1, 71.2, 71.4, 71.5, 71.9, 73.4, 73.7, 73.8, 74.2, 74.8, 75.6, 78.2, 78.3, 78.4, 79.3, 82.1, 94.3, 98.9, 100.3, 100.5, 102.0, 102.1, 105.1, 174.5, 174.7, 175.0, 175.1; HRMS (FAB) m/z $[M + H]^+$ calcd for $C_{28}H_{49}N_2O_{21}$ 749.2828, found 749.2835.

■ ASSOCIATED CONTENT

Supporting Information

Additional text with general information and ^{1}H and ^{13}C NMR spectra for all new compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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■ REFERENCES

(1) For reviews, see (a) Raetz, C. R.; Whitfield, C. *Annu. Rev. Biochem.* **2002**, *71*, 635−700. (b) Caroff, M.; Karibian, D. *Carbohydr. Res.* **2003**, *338*, 2431−2447. (c) Weintraub, A. *Carbohydr. Res.* **2003**, *338*, 2539−2547. (d) Kabanov, D. S.; Prokhorenko, I. R. *Biochemistry (Moscow)* **2010**, *75*, 469−491.

(2) For reviews, see (a) Astronomo, R. D.; Burton, D. R. *Nat. Rev. Drug Discovery* **2010**, *9*, 308−324. (b) Morelli, L.; Poletti, L.; Lay, L. *Eur. J. Org. Chem.* **2011**, 5723−5777.

(3) For reviews, see (a) Zhang, Z.; Wong, C.-H. In *Carbohydrates in Chemistry and Biology*; Ernst, B., Hart, G. W., Sinaÿ, P., Eds.; Wiley−VCH: Weinheim, Germany, 2000; Part I, pp 117−134. (b) Vankayalapati, H.; Jiang, S.; Singh, G. *Synlett* **2002**, 16−25. (c) Palmacci, E. R.; Plante, O. J.; Seeberger, P. H. *Eur. J. Org. Chem.* **2002**, 595−606. (d) Nakamura, S.; Nambu, H.; Hashimoto, S. In *Handbook of Chemical Glycosylation*; Demchenko, A. V., Ed.; Wiley−VCH: Weinheim, Germany, 2008; pp 223−259.

(4) For use of diphenyl phosphate as a leaving group, see (a) Hashimoto, S.; Honda, T.; Ikegami, S. *J. Chem. Soc., Chem. Commun.* **1989**, 685−687. (b) Koshiba, M.; Suzuki, N.; Arihara, R.; Tsuda, T.; Nambu, H.; Nakamura, S.; Hashimoto, S. *Chem.—Asian J.* **2008**, *3*, 1664−1677. (c) Kakita, K.; Tsuda, T.; Suzuki, N.; Nakamura, S.; Nambu, H.; Hashimoto, S. *Tetrahedron* **2012**, *68*, 5005−5017.

(5) For use of *P*,*P*-diphenyl-*N*-(*p*-toluenesulfonyl)phosphinimidate as a leaving group, see Hashimoto, S.; Honda, T.; Ikegami, S. *Tetrahedron Lett.* **1991**, *32*, 1653−1654.

(6) For use of *N*,*N*,*N*′,*N*′-tetramethylphosphorodiamidate as a leaving group, see Hashimoto, S.; Yanagiya, Y.; Honda, T.; Harada, H.; Ikegami, S. *Tetrahedron Lett.* **1992**, *33*, 3523−3526.

(7) For use of diethyl phosphite as a leaving group, see Hashimoto, S.; Umeo, K.; Sano, A.; Watanabe, N.; Nakajima, M.; Ikegami, S. *Tetrahedron Lett.* **1995**, *36*, 2251−2254.

(8) For β-selective glycosylation with 2-azido-2-deoxymannosyl diphenyl phosphate, see Nakamura, S.; Tsuda, T.; Suzuki, N.; Hashimoto, S. *Heterocycles* **2009**, *77*, 843−848.

(9) For glycosylation with 2-acetamido-2-deoxyglycosyl diethyl phosphites, see (a) Arihara, R.; Nakamura, S.; Hashimoto, S. *Angew. Chem., Int. Ed.* **2005**, *44*, 2245−2249. (b) Arihara, R.; Kakita, K.; Suzuki, N.; Nakamura, S.; Hashimoto, S. *J. Org. Chem.* **2015**, DOI: 10.1021/acs.joc.5b00138.

(10) Haseley, S.; Wilkinson, S. G. *Carbohydr. Res.* **1997**, *301*, 187−192. While the chemical structure of the polymeric antigen was reported, its biological repeating unit has not been defined.

(11) Synthesis of the trisaccharide ManNAcβ1→4Galα1→4Gal involved in the tetrasaccharide repeating unit of the O18 antigen has already been reported: Koto, S.; Shinoda, Y.; Hirooka, M.; Sekino, A.; Ishizumi, S.; Koma, M.; Matuura, C.; Sakata, N. *Bull. Chem. Soc. Jpn.* **2003**, *76*, 1603−1615.

(12) (a) Litjens, R. E. J. N.; Leeuwenburgh, M. A.; van der Marel, G. A.; van Boom, J. H. *Tetrahedron Lett.* **2001**, *42*, 8693−8696. (b) Codée, J. D. C.; Litjens, R. E. J. N.; den Heeten, R.; Overkleeft, H. S.; van Boom, J. H.; van der Marel, G. A. *Org. Lett.* **2003**, *5*, 1519−1522. (c) Codée, J. D. C.; van den Bos, L. J.; Litjens, R. E. J. N.; Overkleeft, H. S.; van Boeckel, C. A. A.; van Boom, J. H.; van der Marel, G. A. *Tetrahedron* **2004**, *60*, 1057−1064. (d) Litjens, R. E. J. N.; den Heeten, R.; Timmer, M. S. M.; Overkleeft, H. S.; van der Marel, G. A. *Chem.—Eur. J.* **2005**, *11*, 1010−1016. (e) Litjens, R. E. J. N.; van den Bos, L. J.; Codée, J. D. C.; van den Berg, R. J. B. H. N.; Overkleeft, H. S.; van der Marel, G. A. *Eur. J. Org. Chem.* **2005**, 918−924. (f) van den Bos, L. J.; Duivenvoorden, B. A.; de Koning, M. C.; Filippov, D. V.; Overkleeft, H. S.; van der Marel, G. A. *Eur. J. Org. Chem.* **2007**, 116−124.

(13) For reviews, see (a) Crich, D. *Acc. Chem. Res.* **2010**, *43*, 1144−1153. (b) Crich, D. *J. Org. Chem.* **2011**, *76*, 9193−9209.

(14) We selected diphenyl phosphate as the leaving group of disaccharide **9** with the expectation that higher α-selectivity could be obtained as compared with diethyl phosphite leaving group when treated with a Lewis acid. See ref 4b.

(15) Nashed, M. A.; Anderson, L. *Tetrahedron Lett.* **1976**, 3503−3506.

(16) Ueda, T.; Feng, F.; Sadamoto, R.; Niikura, K.; Monde, K.; Nishimura, S.-I. *Org. Lett.* **2004**, *6*, 1753−1756.

(17) Hébert, N.; Just, G. *J. Chem. Soc., Chem. Commun.* **1990**, 1497−1498. In general, either NIS or iodine has been employed to cleave prop-1-enyl glycosides, but the use of iodine in aqueous THF in the presence of pyridine led to partial loss of a benzylidene protecting group if present. We therefore used NBS for oxidative hydrolysis of prop-1-enyl groups throughout the synthesis to avoid the side reaction.

(18) (a) Oikawa, Y.; Yoshioka, T.; Yonemitsu, O. *Tetrahedron Lett.* **1982**, *23*, 885−888. (b) Horita, K.; Yoshioka, T.; Tanaka, T.; Oikawa, Y.; Yonemitsu, O. *Tetrahedron* **1986**, *42*, 3021−3028.

(19) Zheng, Q. Y.; Darbie, L. G.; Cheng, X.; Murray, C. K. *Tetrahedron Lett.* **1995**, *36*, 2001−2004.

(20) (a) Nashed, M. A.; Anderson, L. *Carbohydr. Res.* **1976**, *51*, 65−72. (b) He, D.-Y.; Li, Z.-J.; Cai, M.-S. *Carbohydr. Res.* **1993**, *239*, 267−271.

(21) (a) Nagashima, N.; Ohno, M. *Chem. Lett.* **1987**, 141−144. (b) Nagashima, N.; Ohno, M. *Chem. Pharm. Bull.* **1991**, *39*, 1972−1982.

(22) Westerduin, P.; Veeneman, G. H.; Marugg, J. E.; van der Marel, G. A.; van Boom, J. H. *Tetrahedron Lett.* **1986**, *27*, 1211−1214.

(23) For use of Oxone as an oxidizing agent of organophosphorus compounds, see Woźniak, L. A.; Stec, W. J. *Tetrahedron Lett.* **1999**, *40*, 2637−2640.

(24) All attempts to introduce diphenyl phosphate leaving group directly at C1 met with little success; for example, (a) $ClP(O)(OPh)_2$, DMAP, CH_2Cl_2, 0 to 50 °C, complex mixture[25] and (b) BuLi, THF, −78 °C, then $ClP(O)(OPh)_2$, −78 to 0 °C, ca. 60−70%.[26]

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J. Org. Chem. 2015, 80, 4278−4288

(25) Sabesan, S.; Neira, S. *Carbohydr. Res.* **1992**, *223*, 169−185.
(26) Inage, M.; Chaki, H.; Kusumoto, S.; Shiba, T. *Chem. Lett.* **1982**, 1281−1284.
(27) Hashimoto, S.; Sakamoto, H.; Honda, T.; Abe, H.; Nakamura, S.; Ikegami, S. *Tetrahedron Lett.* **1997**, *38*, 8969−8972.
(28) Hemiacetal **25** was obtained as a major product.
(29) For use of $TMSClO_4$ as an activator of glycosyl phosphates, see ref 4b.
(30) The beneficial effect of THF in glycosylation with glycosyl diphenyl phosphates has been reported; see ref 4c.
(31) Kinzy, W.; Schmidt, R. R. *Liebigs Ann. Chem.* **1985**, 1537−1545.

DOI: 10.1021/acs.joc.5b00139
J. Org. Chem. 2015, 80, 4278−4288