CHEMISTRY
A EUROPEAN JOURNAL

DOI: 10.1002/chem.200902460

Synthesis of Monomeric and Dimeric Repeating Units of the Zwitterionic Type 1 Capsular Polysaccharide from *Streptococcus pneumoniae*

Xiangyang Wu,[b] Lina Cui,[c] Tomasz Lipinski,[a] and David R. Bundle*[a]

Abstract: Zwitterionic polysaccharides (ZPSs) from *Bacteroides fragilis* and *Streptococcus pneumoniae* display unique T-cell activities. The first synthesis of a hexasaccharide representing two repeating units of the zwitterionic capsular polysaccharide from *S. pneumoniae* type 1 (Sp1) is reported. Key elements of the approach are stereoselective construction of 1,4-*cis*-α-galactose linkages based on a reactive trichloroacetimidate donor that incorporates a 6-*O*-acetyl group, which may contribute to the high α selectivity in glycosylation. After assembly of the fully protected hexasaccharide from five monosaccharide synthons **2**–**4**, **24** and **25**, selective deprotection of the primary hydroxyl groups of the four galactose residues followed by oxidation to the corresponding uronic acids provides hexasaccharide **19**. The trisaccharide counterpart **1** was synthesized in similar fashion from three synthons, **2**–**4**. This approach employed both conventional and dehydrative glycosylation methodologies and avoids the use of poorly reactive uronic acid derived glycosyl donors and acceptors.

Keywords: glycosylation · oxidation · polysaccharides · T-cell activation · uronic acid

Introduction

The immune system has evolved the ability for T cells to recognize nearly any biological polymer, including peptides, protein superantigens, and glycolipids through presentation by the major histocompatibility complex (MHC) proteins such as MHC class I (MHCI), MHC class II (MHCII), and CD1. However, polysaccharides are poorly immunogenic, T-cell-independent (TI-2) antigens[1] and are not recognized by MHC proteins. Recent and unexpected additions to the list of biopolymers recognized by T-cells are zwitterionic capsular polysaccharide (ZPS). These bacterial molecules utilize MHCII presentation to activate T cells by αβ T cell receptor (αβTCR) proteins. These zwitterionic polysaccharides (ZPSs) from *Bacteroides fragilis* induce a variety of T-cell specific responses such as cell proliferation, cytokine secretion, and regulation of antibody production.[2] ZPSs activate $CD4^+$ T cells by a recently described mechanism of processing and presentation by antigen-presenting cells that requires nitric oxide-mediated degradation of these polymers in the MHCII pathway.[3]

Active ZPSs share a common structural motif: a high density of positively charged amino and negatively charged carboxyl or phosphate groups. Functional group modification established that the zwitterionic motif was essential for activity since conversion of positively charged amines to neutral acetamido groups eliminated the activity of ZPSs.[4] Fragmentation of native capsular polysaccharides has further established that large oligosaccharides with molecular weight around 8 kDa retain biological activity.[3,5] However, successful synthesis and deprotection of oligosaccharide fragments, or isolation of single repeating units or oligomers containing the minimum structural element for biological activity has yet to be achieved. Efficient syntheses of the repeating units of ZPSs are a prerequisite for establishing their minimal size for activity and an appreciation of precise structure-function relationships.

The structure of the type 1 capsular polysaccharide of *Streptococcus pneumoniae* (Sp1)[6] is less complex than either the polysaccharides A and B of *Bacteroides fragilis*

[a] Dr. T. Lipinski, Prof. Dr. D. R. Bundle
Department of Chemistry, University of Alberta
Alberta Ingenuity Centre for Carbohydrate Science
Edmonton, AB T6G 2G2 (Canada)
E-mail: dave.bundle@ualberta.ca

[b] Dr. X. Wu
Molecular Pharmacology and Chemistry
Memorial Sloan Kettering Cancer Center
1275 York Avenue, New York, NY 10065 (USA)

[c] Dr. L. Cui
719 Latimer Hall, Department of Chemistry
University of California, Berkeley, CA 94720-1460 (USA)

Supporting information for this article is available on the WWW under http://dx.doi.org/10.1002/chem.200902460.

(Figure 1) and exhibits similar T-cell activity,[7] rendering the synthesis of Sp1 repeating units more tractable. Sp1 is a linear polymer of trisaccharide repeating units, containing two galacturonic acids (GalA, residues **a** and **c**) and a 2-acetamido-4-amino-2,4,6-trideoxygalactose (residue **b**), (in Figure 1). All anomeric linkages have the α-configuration. Each repeating unit of Sp1 contains one positively charged amine and two negatively charged carboxyl groups. The inherent challenges confronting the synthesis of Sp1 repeating units are the low reactivity of uronic acid derivatives in glycosylation reactions and the synthesis of α-galactopyranosides with high stereoselectivity. There have been attempts to synthesize the PS-A of *B. fragilis*, however, the complete repeating unit was not reached nor was the oligosaccharide deprotected.[8] In other work a blocked but not deprotected trisaccharide repeating unit of Sp1 has been reported.[9] Here we report the first completed chemical synthesis and deprotection of a zwitterionic polysaccharide trisaccharide repeating unit **1** and the corresponding hexasaccharide **19** containing two repeating units.

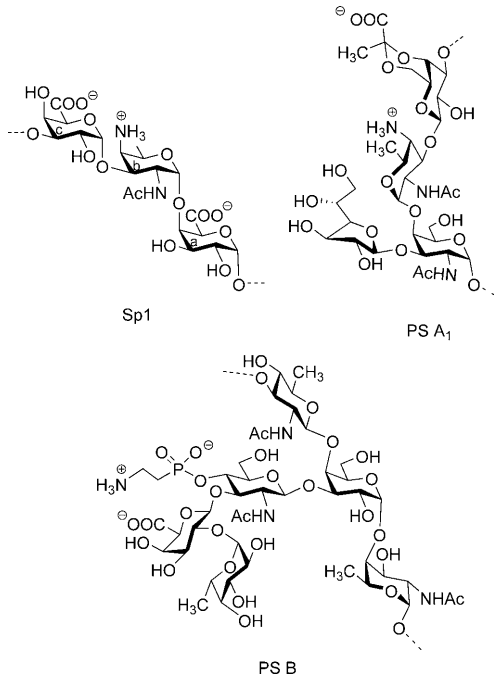


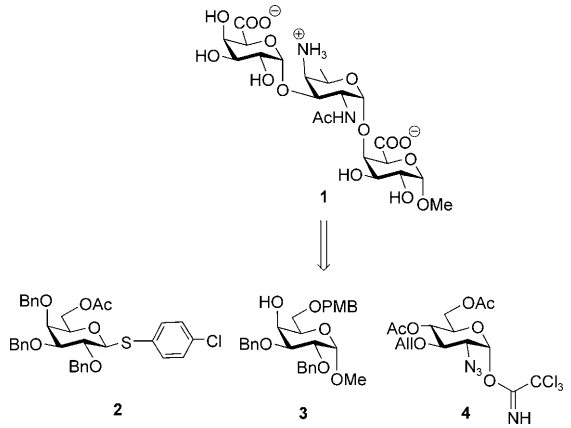
Figure 1. Chemical structures of the repeating units of various zwitterionic polysaccharides. PS A_1 and PS B are capsules from *B. fragilis*. Sp1 is the capsule from *S. pneumoniae* type 1.

Results and Discussion

A key issue for the synthesis of the target molecules is the availability of high-yielding glycosylation strategies, especially in light of the low reactivity of uronic acid derivatives which generally result in poor glycosylation yields.[10–13] The alternative strategy adopted here is to create uronic acid units by oxidation of the hydroxymethyl group of appropriately prepared substrate oligosaccharides.[11–12,34] Implementation of this approach requires the stereoselective construction of α1,3 and α1,4 galactopyranosyl linkages. Typically these require glycosyl donors possessing a non-participating functionality at the C2 position, which may lead to anomeric mixtures. Although there are numerous methods for α-glycoside synthesis,[14–16] direct synthesis of the α-galactopyranosyl linkage with high stereoselectivity remains a challenge. Traditionally, reasonable yields of α-glycosides could be achieved by extensive optimization of reaction conditions, such as solvent, temperature, promoter, as well as leaving group and protecting-group pattern.[17] Conformationally locked *N*-benzyl 2,3-*trans*-oxazolidione derivatives described by Ito and co-workers exhibit good α-selectivity for amino sugar glycoside formation.[18] However, when considering the synthesis of a 2,4-diamino-2,4,6-trideoxy-α-D-hexopyranoside, that approach required more steps than the adoption of 2-azido-2-deoxy-hexose donors for the construction of 1,2-*cis* glycosides.[17] In this work stereoselective formation of the 1,4-*cis*-α-galactose linkage of the diamino-dideoxy-hexose residue could be achieved using an azido moiety at C-2 as a nonparticipating group combined with an *O*-6 acetate. Although Crich et al. have established that there is no evidence for long range neighboring group participation for such esters,[19a] there is empirical evidence that a 6-*O*-acetate may contribute to stereoselective α-glycosylation.[19b,c–20]

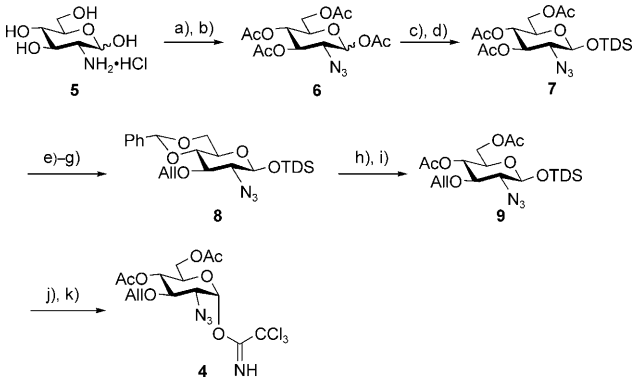
Based on the above principles, retrosynthesis of trisaccharide **1** starting from the reducing terminus led to three different building blocks **2**, **3** and **4** (Scheme 1). Building blocks **2**[21] and **3**[22] were readily synthesized according to published literature. Utilization of the *p*-methoxybenzyl (PMB) group as a temporary protecting group at C-6 in compound **3** was intended to enhance the nucleophilicity of the 4-hydroxyl group, and facilitate its glycosylation by donor **4**. The choice of 4-chlorothiophenol as leaving group for the donor **2** avoids strong thiophenol odors. For the efficient construction of 1,4-*cis* glycoside, we introduced an azido moiety at C-2 as a nonparticipating group.[17]

Synthesis of building block **4** started from commercially available D-glucosamine hydrochloride **5** (Scheme 2). Diazo transfer from triflyl azide to D-glucosamine hydrochloride



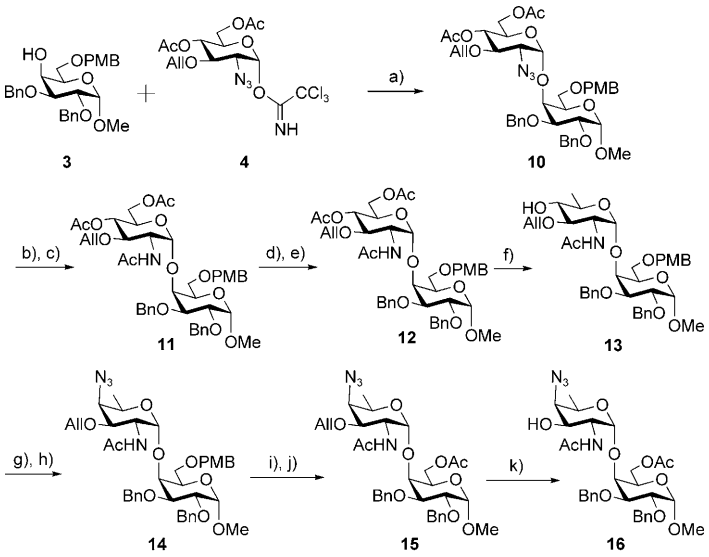
Scheme 1. Retrosynthesis of target trisaccharide **1**.

5[23] with cupric sulfate as catalyst, followed by acetylation furnished **6** in 86% yield in two steps. Selective removal of the anomeric acetate with hydrazinium acetate in DMF gave the desired intermediate which upon treatment with thexyldimethylsilyl chloride (TDSCl) in DMF afforded compound **7** in excellent yield.[24] The utilization of a very bulky TDS protecting group at the anomeric position gave the pure β-isomer and thus simplified the purification. Deacetylation by transesterification, followed by reaction with benzaldehyde dimethyl acetal in the presence of catalytic amount of toluenesulfonic acid (TsOH) in acetonitrile, and subsequent allylation afforded compound **8** in an excellent yield. Selective cleavage of the 4,6-*O*-benzylidene group[25] with ethanethiol in the presence of TsOH as catalyst in dichloromethane, followed by acetylation gave the desired compound **9**. Removal of the *O*-silyl group with tetrabutylammonium fluoride (TBAF)[26] under acidic conditions and then reaction with trichloroacetonitrile in the presence of 1,8-diazabicyclohexylcarbodiimide (DBU)[27] as base furnished the desired building block **4** in good yield.



Scheme 2. a) TfN_3, CH_2Cl_2, K_2CO_3, H_2O, $CuSO_4$, MeOH; b) Ac_2O, pyridine, 86% over two steps; c) $NH_2NH_2{\cdot}HOAc$, DMF, 86%; d) TDSCl, CH_2Cl_2, imidazole, 93%; e) NaOMe, MeOH, 100%; f) $PhCH(OMe)_2$, TsOH, CH_3CN, 80%; g) AllBr, DMF, NaH, 90%; h) EtSH, TsOH, CH_2Cl_2, 85%; i) Ac_2O, pyridine, 100%; j) TBAF, AcOH, THF, 74%; k) CCl_3CN, DBU, CH_2Cl_2, 80%.

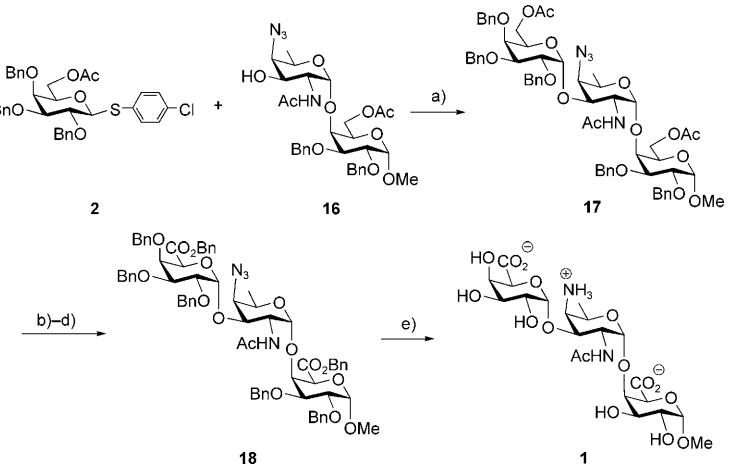
With the required building blocks at our disposal, the multistep synthesis of trisaccharide **1** was initiated (Scheme 3). Activation of donor **4** by trimethylsilyl trifluoromethane sulfonate (TMSOTf) was employed for the glycosylation of acceptor **3** to give disaccharide **10** in 60% yield. Excellent diastereoselectivity was observed in this strategy since ^{1}H NMR detected none of the β-glucopyranosyl anomer. The NMR data and $^3J_{1,2}$ coupling constant values indicated the presence of the 1,2-*cis*-glucopyranosidic linkage ($\delta = 4.91$ ppm, 1b-H, $J_{1,2} = 3.62$ Hz; $\delta = 3.34$ ppm, 2b-H, $J_{2,1} = 3.62$, $J_{2,3} = 10.2$ Hz), which was also confirmed by its $^1J_{C-H}$ value (174 Hz). The high α selectivity suggested a possible remote stereoelectronic effect,[19b,c,20] while the relatively low yield (60%) was ascribed to the low reactivity and steric hindrance at the 4-*O* position of the galactopyranoside acceptor **3**. Reduction of compound **10** with hydrogen sulfide (H_2S) under basic conditions afforded the free amine intermediate followed by acetylation with acetic anhydride in pyridine to give the desired disaccharide **11** in 74% yield over two steps. Transesterification of the acetate group of **11** followed by selective mesylation gave intermediate **12**, and subsequent reduction with sodium borohydride in DMSO at 85°C furnished the 6-deoxy disaccharide **13** in good yield while leaving the *N*-acetyl group intact.[28] Triflation of **13** with triflic anhydride (Tf_2O) and pyridine at −30°C gave the unstable 4-*O*-triflate which was immediately treated with sodium azide (NaN_3) in DMF at room temperature to produce the required 2-acetamido-4-azido-galactopyranose disaccharide **14** in 57% yield.[29] Selective removal of the *p*-methoxybenzyl group with trifluoroacetic acid (TFA), followed by acetylation gave **15**. Subsequent $PdCl_2$-catalyzed deallylation[30] afforded the selectively deprotected building block **16** in moderate yield. Coordination of the azido group with $PdCl_2$ may account for the low yield of the deallylation step.



Scheme 3. a) TMSOTf, CH_2Cl_2, −15°C → RT, 60%; b) H_2S, pyridine, H_2O, NEt_3; c) Ac_2O, pyridine, 74% over two steps; d) NaOMe, MeOH, 100%; e) MsCl, pyridine, −15°C, 71%; f) DMSO, $NaBH_4$, 85°C, 80%; g) Tf_2O, pyridine, CH_2Cl_2, −30°C; h) NaN_3, DMF, RT, 57%; i) TFA (1%) in CH_2Cl_2; j) Ac_2O, pyridine, 80%; k) $PdCl_2$, NaOAc, AcOH, H_2O, 53%.

The glycosylation[31] of disaccharide **16** by thiogalactoside **2** in the presence of *N*-iodosuccinimide (NIS) and trifluoromethane sulfonic acid (TfOH) at −30°C in dichloromethane (CH_2Cl_2) furnished the required trisaccharide **17** in 73% yield (Scheme 4). Only trace amounts of β-linked galactose were detectable by ^{1}H NMR. The heteronuclear one-bond coupling constant ($^1J_{C-H} = 174$ Hz) unambiguously established the α-anomeric configuration of the newly introduced glycosidic bond.[32] Consistent with the findings of others,[19b,c,20] we attribute the α selectivity to remote stereoelectronic effects associated with the 6-*O*-acetyl group. In order

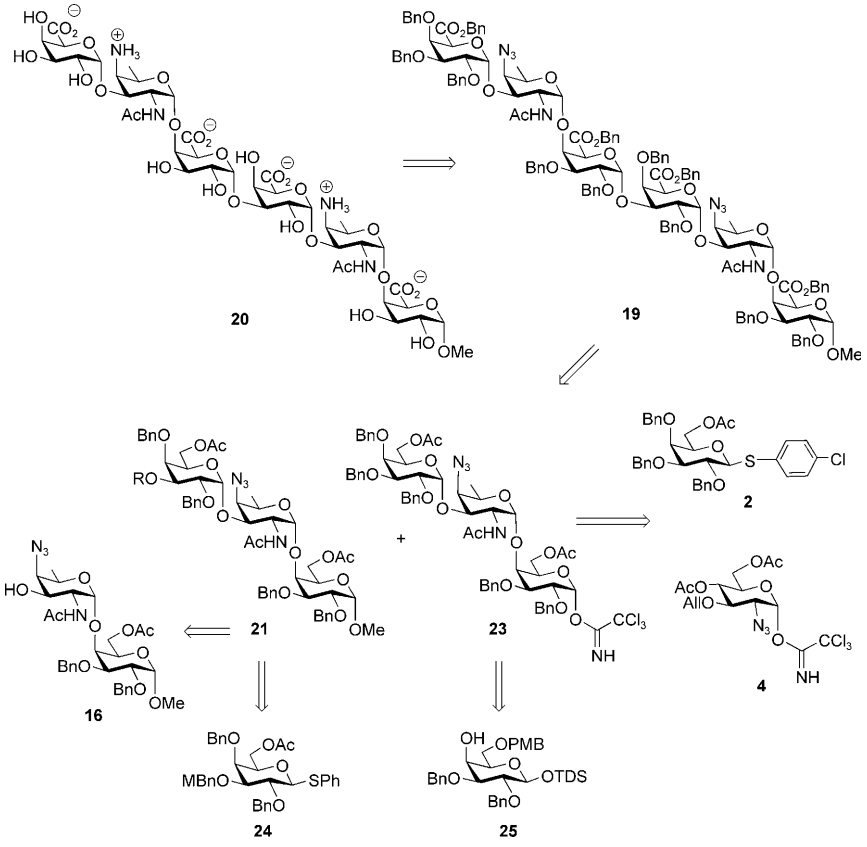
to successfully transform two primary alcohol groups to carboxylic acids simultaneously, an efficient oxidation conditions was highly desirable. Jones oxidation[33] requires strong acidic conditions which are not compatible with sensitive functional groups, such as electron-rich aromatic rings, acid labile isopropylidene ketals and glycosidic linkages, and precludes its application for the synthesis of complex oligosaccharides. Recently, Huang[34] and co-workers developed a novel two-step, one-pot procedure for the conversion of primary alcohols to carboxylic acids under very mild conditions in good yields. Using a similar procedure employing sodium hypochlorite and 2,2,6,6-tetramethylpiperidine-oxyl (TEMPO), the two hydroxymethyl groups after selective deacetylation of trisaccharide **17** were concurrently oxidized to the corresponding uronic acids, which were protected as benzyl ethers[12,35] for ease of purification to give the trisaccharide **18**. Final hydrogenolysis of the fully protected trisaccharide **18** afforded the target molecular **1** in 58% yield.



Scheme 4. a) NIS, TfOH, CH_2Cl_2, −30 → RT, 73%; b) NaOMe, MeOH, 100%; c) TEMPO, KBr, NaOCl; d) BnBr, CsF, DMF, 51% over three steps; e) H_2, $HOAc/H_2O$, Pd/C (10%), 58%.

However, biological evaluation showed that trisaccharide **1** did not activate T-cells, so we turned our attention to the synthesis of hexasaccharide **19** containing two repeating units. Based on the synthetic strategy described above, retrosynthetic analysis of hexasaccharide **19** and its protected form **20** led to two key trisaccharide building blocks, acceptor **21** and trisaccharide donor **23** (Scheme 5). The terminal reducing trisaccharide acceptor **21** could be readily synthesized by glycosylation of disaccharide **16** with monosaccharide **24** using similar conditions to those described above. Trisaccharide donor **23** (shown in Scheme 5) can be assembled from building blocks **2**, **4** and **25**.

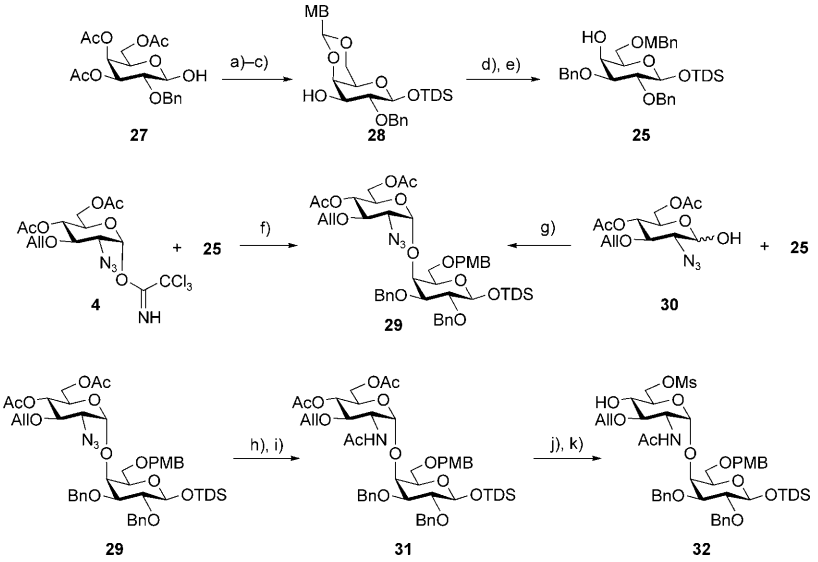
In order to synthesize trisaccharide acceptor **21**, we required a temporary protecting group for the 3″-hydroxyl group, which is compatible with the glycosylation conditions and acetyl group removal. A *p*-methoxybenzyl (PMB) group fulfilled this requirement (Scheme 5). The synthesis of monosaccharide **24** could be achieved starting from the known 4,6-*O*-benzylidene acetal **26** (Scheme 6).[35] *p*-Methoxybenzylation of **26** with *p*-methoxylbenzyl chloride (PMBCl) and sodium hydride (NaH) in DMF followed by selective ring-opening[21] of the 4,6-*O*-benzylidene group with dibutyl boranetriflate (Bu_2BOTf) and BH_3·THF and subsequent acetylation in pyridine provided the desired product **24** in excellent yield. The glycosylation of disaccharide acceptor **16** with donor **24** in the presence of NIS and silver triflate (AgOTf) gave trisaccharide **22** in 79% yield without affecting the *p*-methoxybenzyl group.[31] Cleavage of the *p*-methoxybenzyl group with 2,3-dichloro-5,6-dicyanobenzoquinone (DDQ) in a heterogeneous mixture of dichloromethane (CH_2Cl_2) and



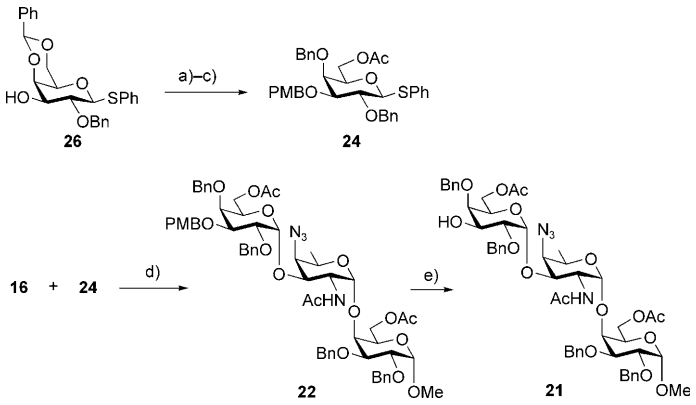
Scheme 5. Retrosynthetic analysis of hexasaccharide **19**.

H_2O gave trisaccharide acceptor **21** in 73% yield (Scheme 6).

In an attempt to improve the coupling efficiency of glycosylation, careful adjustment of donor and acceptor reactivity via protecting group strategy is crucial. In the construction of trisaccharide trichloroacetimidate donor **23**, it is highly desirable that the temporary protecting group at the anomeric position of the terminal reducing galactose residue (Scheme 7) is orthogonal to other protecting groups and the glycosylation conditions without dampening the reactivity of acceptor. After a few attempts, we were delighted to find that the TDS group could satisfy these requirements.[25a] Silyla-



Scheme 7. a) TDSCl, imidazole, CH_2Cl_2, 95%; b) NaOMe, MeOH; c) MeO-C_6H_4-$CH(OMe)_2$, 70%; d) BnBr, NaH, DMF, 95%;[21] e) $NaCNBH_3$, HCl in Et_2O, 65%; f) TMSOTf, CH_2Cl_2, 0°C, 30%; g) Ph_2SO, Tf_2O, TTBP, CH_2Cl_2, −25°C, 73%; h) H_2S, pyridine/H_2O/Et_3N; i) Ac_2O, pyridine, 75% over two steps; j) NaOMe, MeOH; k) MsCl, pyridine, −15 → 0°C, 95%.



Scheme 6. a) PMBnCl, NaH, DMF, 90%; b) Bu_2BOTf, BH_3·THF, 70%; c) Ac_2O, pyridine, 100%; d) NIS/AgOTf, CH_2Cl_2, −20°C, 79%; e) CH_2Cl_2, H_2O, DDQ, 73%.

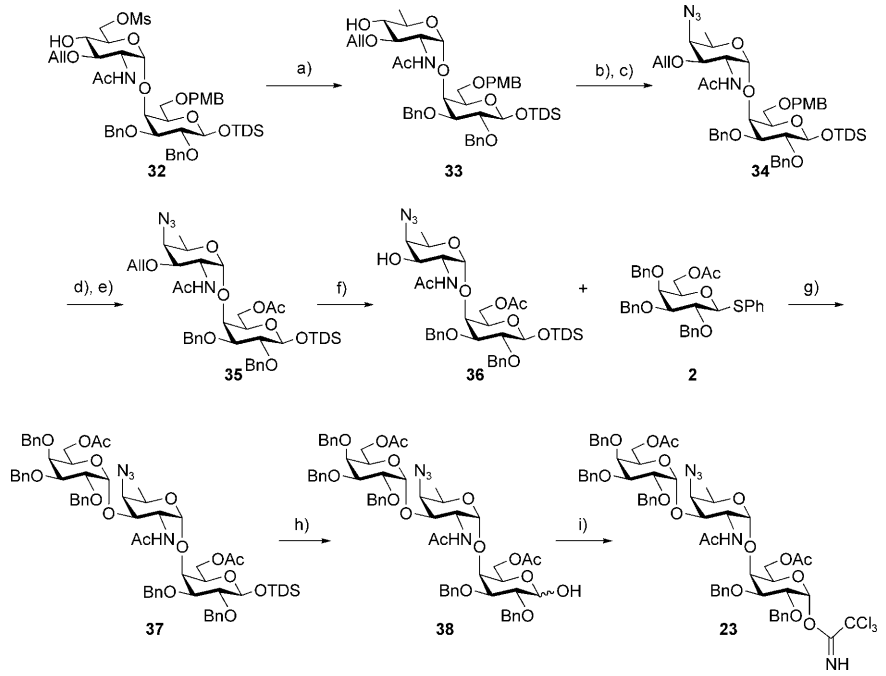
tion of starting material **27**[35b] with TDSCl under basic conditions followed by deacetylation with sodium methoxide and subsequent reaction with anisaldehyde dimethyl acetal under acidic conditions provided **28** in excellent yield. Benzylation of **28** with benzyl bromide (BnBr) followed by sodium cyanoborohydride ($NaCNH_3$) reductive ring-opening[36] of the 4,6-*O*-*p*-methoxybenzylidene acetal group with HCl in Et_2O and subsequent acetylation provided acceptor **25** in good yield (Scheme 7).

In an initial attempt to perform the glycosylation of acceptor **25** with donor **4** using TMSOTf as catalyst, the desired disaccharide **29** was obtained in only 30% yield. The low yield was probably due to steric hindrance at the axial 4-OH of galactopyranoside **25**, which was accentuated by the bulky TDS group. However, using the dehydration strategy developed by Gin and co-workers,[37] glycosylation of acceptor **25** by donor **30** performed in the presence of phenyl sulfoxide, triflic anhydride and tri-*tert*-butylpyrimidine (TTBP) gave disaccharide **29** in 73% yield.[37] Selective reduction of the azide group of disaccharide **29** with hydrogen sulfide (H_2S) under basic conditions was followed by *N*-acetylation to give the fully protected 2-acetamido-2-deoxy disaccharide **31**. Transesterification of the ester groups and subsequent mesylation gave the 2-acetamido-2,6-deoxy disaccharide mesylate **32** in good yield.

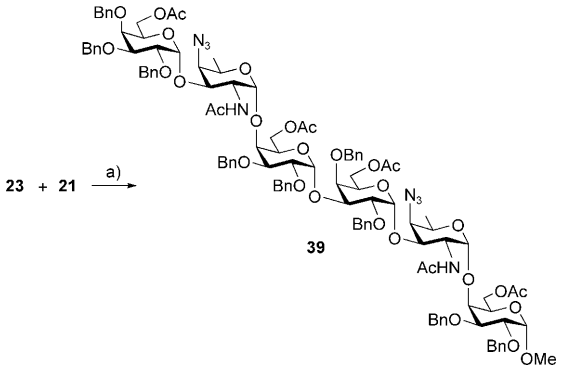
Manipulation of protecting groups utilizing similar procedures to those described for compound **16** provided the desired disaccharide **36** via derivatives **33**–**35** (Scheme 8). Glycosylation of disaccharide **36** with donor **2** using *N*-iodosuccinimide (NIS) and silver triflate (AgOTf) provided trisaccharide **37** in 66% yield. Desilylation[25a] of trisaccharide **37** with tetrabutylammonium fluoride (TBAF) under acidic conditions gave the reducing trisaccharide **38** in 92% yield without cleaving the acetyl groups, and subsequent reaction with trichloroacetonitrile using DBU provided the trisaccharide trichloroacetimidate donor **23** in 71% yield.[27]

The glycosidation of trisaccharide donor **23** with trisaccharide acceptor **21** was performed in the presence of TMSOTf at room temperature to give hexasaccharide **39** in 85% yield (Scheme 9). Careful tuning of the reaction temperature and the equivalents of donor and Lewis acid (TMSOTf) was crucial to achieving this high yield.

Transesterification of hexasaccharide **39** gave tetraol **40** quantitatively (Scheme 10). The TEMPO/NaClO procedure used to prepare trisaccharide **1** failed to oxidize hexasaccharide **40**. However, by using Huang's oxidation procedure,[34] all four hydroxyl groups of the hexasaccharide **40** could be oxidized by vigorous stirring of a heterogeneous mixture of water/organic solvents containing TEMPO/



Scheme 8. a) $NaBH_4$, DMSO, 85 °C, 85 %; b) Tf_2O, pyridine, CH_2Cl_2, −30 °C; c) NaN_3, DMF, 62 % over two steps; d) DDQ, CH_2Cl_2, H_2O; e) Ac_2O, py. 80 % over two steps; f) $PdCl_2$, NaOAc, HOAc, 66 %; g) NIS/AgOTf, CH_2Cl_2, 66 %; h) TBAF, HOAc, THF, 92 %; i) CCl_3CN, DBU, CH_2Cl_2, 71 %.



Scheme 9. a) TMSOTf, CH_2Cl_2, 85 %.

NaClO. Immediate benzylation with benzyl bromide under slightly basic conditions provided the desired hexasaccharide **20** smoothly in 52 % yield over three steps. Hydrogenolysis of hexasaccharide **20** was challenging owing to different polarity of the reactant and its product, therefore a solvent system that could provide good solubility of both was critical to the hydrogenolysis reaction. Here we used a mixture of CH_2Cl_2/MeOH/H_2O 1:3:0.5 for the hydrogenolysis of hexasaccharide **20** using palladium hydroxide [$Pd(OH)_2$] on charcoal to give after gel permeation chromatography pure hexasaccharide **19** in 53 % yield.

Trisaccharide **1** and hexasacharide **19** were purified from lower molecular weight side products by gel-permeation chromatography as their ammonium salts. The NMR spectra were also recorded for these salts and compared (Figure 2) with the published spectrum of the Sp1 polysaccharide.[6] The internal anomeric proton resonances of the hexasaccharide such as a′, c, begin to adopt chemical shifts that are similar to those of the Sp1 polysaccharide. However, it is clear that residues a, b, b′ are quite distinct in their chemical shifts when compared to the corresponding resonances of the polysaccharide suggesting that the conformation of even the hexasaccharide should be distinct from the average conformation for the repeating unit as it occurs in the polysaccharide. Initial biological assays indicate that there is only very low if any activity for even the hexasaccharide, consistent with the inference of solution conformational preferences.

Fragmentation of zwitterionic polysaccharides established that large oligosaccharides of 8 kDa molecular weight retain biological activity.[3,5] This would correspond to an oligosaccharide of approximately 10–15 repeating units. Clearly given the synthetic challenges in preparing two repeating units, compound **19**, the resolution of the unresolved issue of the minimal size oligosaccharide for biological activity is unlikely to be addressed by oligosaccharide synthesis.

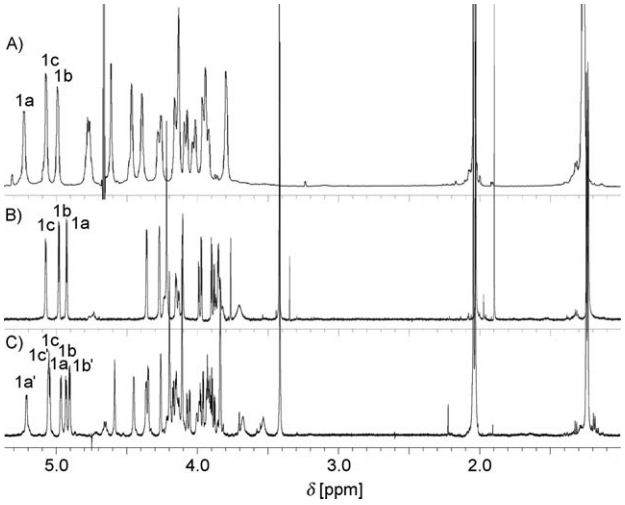
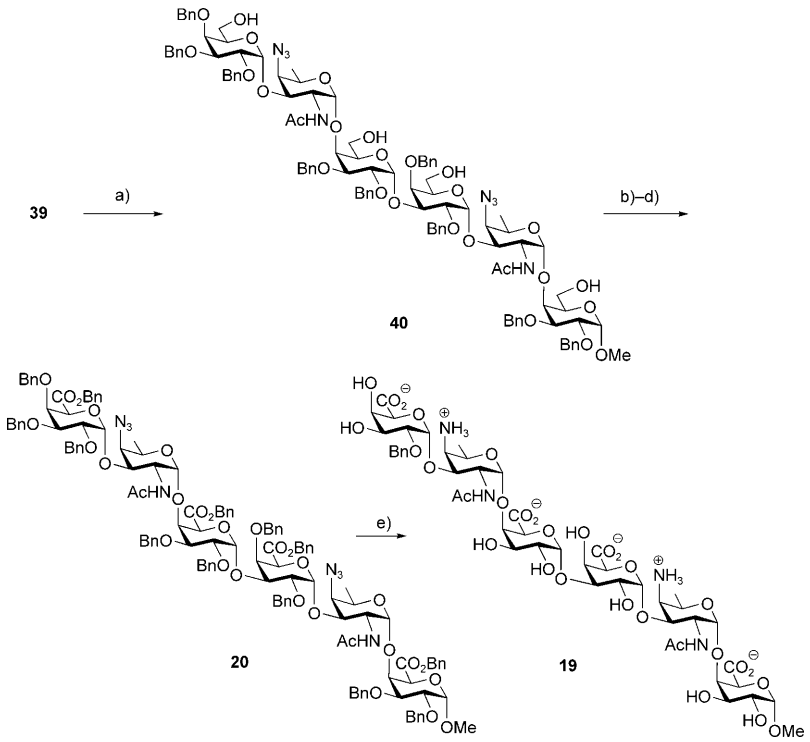


Figure 2. A comparison of the ^1H NMR spectra of A) polysaccharide Sp1, B) trisaccharide **1** and C) hexasaccharide **19**.



Scheme 10. a) NaOMe, MeOH; b) TBABr, $NaHCO_3$, TEMPO, CH_2Cl_2, NaOCl; c) HCl, *t*BuOH, 2-methylbut-2-ene, $NaClO_2$, NaH_2PO_4; d) CsF, BnBr, DMF, 52% over three steps; e) H_2, $Pd(OH)_2$, CH_2Cl_2, MeOH, H_2O, 53%.

Conclusion

A fully synthetic strategy was developed to synthesize the trisaccharide and hexasacharide repeating units of the zwitterionic capsular polysaccharide antigen of *Streptococcus pneumoniae* type 1. The creation of trisaccharide and hexasaccharide target compounds containing, 2- and 4-α-linked galacturonic acids residues, respectively, was accomplished by first creating the corresponding α-galactopyranosides. This avoids the inherently low reactivity of uronic acid glycosyl donors. Careful simultaneous oxidation of two and four primary hydroxymethyl groups gave the trisaccharide **1** and hexasaccharide **19** could be obtained in respectable yields. Excellent stereocontrol of α-galactosylation was accomplished by employing the very reactive trichloroacetimidate anomeric activating group in combination with a donor 6-*O*-acetyl group. The utilization of a 6-*O*-*p*-methoxybenzyl group as a temporary protecting group enhanced the nucleophilicity of the 4-*O* position of the galactose acceptor, thereby improving the glycosylation yield. Initial immunological evaluation of hexasaccharide **19** suggests that structures containing more than two repeating units will be necessary to achieve significant biological activity.

Experimental Section

Analytical thin-layer chromatography (TLC) was performed on silica gel 60-F254 (Merck). TLC detection was achieved by charring with 5% sulfuric acid in ethanol. All commercial reagents were used as supplied. Column chromatography used silica gel (SiliCycle, 230–400 mesh, 60 Å), and solvents were distilled. High-performance liquid chromatography (HPLC) was performed using a Waters HPLC system that consisted of a Waters 600S controller, 626 pump, and 486 tunable absorbance detector. HPLC separations were performed on a Beckmann C18 semi preparative reversed-phase column with a combination of methanol and water as eluents. Photoadditions were carried out using a spectroline model ENF-260C UV lamp and cylindrical quartz vessels. ^{1}H NMR spectra were recorded at either 400, 500, or 600 MHz, and are referenced to the residual protonated solvent peaks; δ_H 7.24 ppm for solutions in $CDCl_3$, and 0.1% external acetone (δ_H 2.225) for solutions in D_2O. Optical rotations were measured with a Perkin–Elmer 241 polarimeter at 22 °C. Mass spectrometric analysis was performed by positive-mode electrospray ionization on a Micromass ZabSpec Hybrid Sector-TOF mass spectrometer. MALDI mass spectrometric analysis was performed on a Voyager-Elite system from Applied Biosystems.

Dimethylthexylsilyl 3-*O*-allyl-2-azido-4,6-*O*-benzylidene-2-deoxy-β-D-glucopyranoside (8): Allyl bromide (0.34 mL, 3.59 mmol) was added to a stirred solution of dimethylthexylsilyl 2-azido-4,6-*O*-benzylidene-2-deoxy-β-D-glucopyranoside[24] (1.2 g, 2.76 mmol) in DMF (15 mL). After 15 min at 0 °C sodium hydride (190 mg, 4.75 mmol) was added to the solution under argon. The resulting mixture was warmed to room temperature and stirred for two hours. TLC indicated the consumption of starting material, and the reaction mixture was diluted with ethyl acetate (30 mL), and excess NaH was neutralized with methanol. The resulting solution was washed with brine and dried over magnesium sulfate and concentrated to yield a yellow oil. The residue was purified by flash chromatography (hexane/ethyl acetate 6:1) to afford **8** (1.18 g, 90%) as a white oil. $[\alpha]_D = -65.5°$ (c = 1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ = 7.5–7.37 (m, 5H, Ar), 5.93 (m, 1H, OCH_2CH=CH_2), 5.56 (s, 1H, CHPh), 5.33–5.19 (m, 2H, OCH_2CH=CH_2), 4.57 (d, $J_{1,2}$ = 7.7 Hz, 1H, 1-*H*), 4.43–4.39 (m, 1H, OCH_2CH=CH_2), 4.31–4.24 (m, 2H, OCH_2CH=CH_2, 6-*H*), 3.78 (t, ^3J = 10.3 Hz, 1H, 6′-*H*), 3.64 (t, ^3J = 9.2 Hz, 1H, 4-*H*), 3.44 (t, ^3J = 9.2, 9.6 Hz, 1H, 3-*H*), 3.38 (m, 1H, 5-*H*), 3.33 (dd, ^3J = 7.7, 9.6 Hz, 1H, 2-*H*), 1.69 (m, 1H, $CH(CH_3)_2$), 0.92 (m, 12H, 4 × CH_3), 0.21 ppm (m, 6H, 2 × CH_3); HRMS (ESI): *m*/*z*: calcd for $C_{24}H_{37}N_3O_5SiNa$: 498.2395; found: 498.2397 [M+Na^+].

Dimethylthexylsilyl 4,6-di-*O*-acetyl-3-*O*-allyl-2-azido-2-deoxy-β-D-glucopyranoside (9): To a solution of **8** (0.57 g, 1.13 mmol) in CH_2Cl_2 (10 mL) at room temperature was added TsOH (30 mg, 0.23 mmol), followed by addition of ethanethiol (494 μL, 6.78 mmol). The reaction mixture was stirred for 30 min, and then evaporated at low pressure, followed by addition of acetic anhydride (5 mL) and pyridine (5 mL). The resulting mixture was stirred overnight and the resulting solution was concentrated to slightly yellow oil. The residue was purified by flash chromatography (hexanes/ethyl acetate 4:1) to afford **9** (228 mg, 85%) as a white solid. $[\alpha]_D = -29.5°$ (c = 1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ = 5.88–5.84 (m, 1H, OCH_2CH=CH_2), 5.27–5.17 (m, 2H, OCH_2CH=CH_2), 4.93 (dd, ^3J = 9.1, 10.1 Hz, 1H, 4-*H*), 4.50 (d, ^3J = 7.6 Hz, 1H, 1-*H*), 4.29–4.26 (m, 1H, OCH_2CH=CH_2), 4.14–4.08 (m, 3H, OCH_2CH=CH_2, 6-*H*, 6′-*H*), 3.53 (m, 1H, 5-*H*), 3.34 (dd, ^3J = 7.6, 9.8 Hz, 1H, 2-*H*), 3.27 (dd, ^3J = 9.1, 9.8 Hz, 1H, 3-*H*), 2.07 (m, 6H, 2 × CH_3CO), 1.66 (m, 1H, $CH(CH_3)_2$),

0.89 (m, 12H, 4×CH_3), 0.19 ppm (m, 6H, 2×CH_3); (HRMS) EMS: *m*/*z*: calcd for $C_{21}H_{37}N_3O_7SiNa$: 494.2293; found: 494.2294 [*M*+Na^+].

4,6-Di-*O*-acetyl-3-*O*-allyl-2-azido-2-deoxy-α-D-glucopyranosyl trichloroacetimidate (4): Acetic acid (0.3 mL) was added to a solution of **9** (360 mg, 0.72 mmol) in THF (10 mL) at room temperature, followed by slow addition of TBAF (1 mL, 1.01 mmol). The reaction was stirred for 2 h at which point TLC check indicated that the starting material had reacted. The solution was diluted with ethyl acetate (30 mL), and neutralized by extraction with saturated sodium bicarbonate solution (10 mL). The aqueous layer was extracted with ethyl acetate (3×20 mL) and the combined organic layers were dried over magnesium sulfate and concentrated in vacuo. The residue was purified by flash chromatography (hexanes/ethyl acetate 3:1) to afford 4,6-di-*O*-acetyl-3-*O*-allyl-2-azido-2-deoxy-D-glucopyranose as a mixture of anomers (190 mg, 74%). This glucopyranose (170 mg, 0.476 mmol) was dissolved in a mixture of CH_2Cl_2 (5 mL) and Cl_3CCN (0.11 mL) under argon, and then one drop of DBU was added at room temperature. After 2 h, the resulting mixture was concentrated at low pressure. The residue was purified by flash chromatography (hexanes/ethyl acetate 4:1) to afford **4** (190 mg, 80%). $[\alpha]_D$ = +60.4° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=8.77 (s, 1H, N*H*), 6.41 (d, ^3J=3.5 Hz, 1H, 1-*H*), 5.92–5.85 (m, 1H, OCH_2CH=CH_2), 5.29–5.18 (m, 2H, OCH_2CH=CH_2), 5.13 (dd, ^3J=9.5, 10.0 Hz, 1H, 4-*H*), 4.30 (m, 1H, OCH_2CH=CH_2), 4.21–4.15 (m, 2H, OCH_2CH=CH_2, 6-*H*), 5.07 (m, 2H, 5-*H*, 6′-*H*), 3.90 (t, ^3J=9.5, 9.9 Hz, 1H, 3-*H*), 3.72 (dd, ^3J= 3.5, 10.1 Hz, 1H, 2-*H*), 2.10–2.03 ppm (m, 6H, 2×CH_3CO); (HRMS) EMS: *m*/*z*: calcd for $C_{15}H_{19}N_4O_7Cl_3Na$: 495.0211; found: 495.0214 [*M*+Na^+].

Methyl 4,6-di-*O*-acetyl-3-*O*-allyl-2-azido-2-deoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-methoxybenzyl-α-D-galactopyranoside (10): Glycosyl donor **4** (50 mg, 0.1 mmol), monosaccharide acceptor **3**[22] (50 mg, 0.1 mmol) and activated 4 Å molecular sieves (20 mg) were dried together under vacuum for one hour in a pear-shaped flask (5 mL). The contents of the flask were then dissolved in CH_2Cl_2 (3 mL). The suspension was stirred for 10 min at room temperature under argon, and then the temperature was reduced to −15°C, and TMSOTf (50 μL, 0.1 M in CH_2Cl_2) was added dropwise and the reaction mixture was slowly warmed to room temperature. After 30 min, the reaction mixture was neutralized with triethylamine and concentrated in vacuum. The residue was purified by flash chromatography (hexanes/ethyl acetate 6:1→3:1) to afford **10** (50 mg, 60%). $[\alpha]_D$ = +78.0° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=7.41–6.89 (m, 14H, Ar), 5.92–5.85 (m, 1H, OCH_2CH=CH_2), 5.28–5.16 (m, 2H, OCH_2CH=CH_2), 5.04 (t, ^3J=9.5, 10.2 Hz, 1H, 4b-*H*), 4.91 (d, ^3J=3.6 Hz, 1H, 1b-*H*), 4.83–4.70 (m, 5H, 1a-*H*, $2CH_2$Ph), 4.51–4.42 (m, 2H, CH_2Ph), 4.38 (m, 1H, 5b-*H*), 4.25 (m, 1H, OCH_2CH=CH_2), 4.16 (m, 1H, 4a-*H*), 4.08 (m, 1H, OCH_2CH=CH_2), 3.91–3.81 (m, 5H, 2a-*H*, 3a-*H*, 5a-*H*, 6a-*H*, 6b-*H*), 3.80 (s, 3H, OCH_3), 3.72 (t, ^3J=9.6, 9.9 Hz, 1H, 3b-*H*), 3.49–3.45 (m, 2H, 6a′-*H*, 6b′-*H*), 3.37 (s, 3H, OCH_3), 3.31 (dd, ^3J=3.5, 10.2 Hz, 1H, 2b-*H*), 2.1–2.0 (m, 6H, 2× $COCH_3$); (HRMS) EMS: *m*/*z*: calcd for $C_{42}H_{51}N_3O_{13}Na$: 828.3314; found: 828.3313 [*M*+Na^+].

Methyl 2-acetamido-4,6-di-*O*-acetyl-3-*O*-allyl-2-deoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-methoxybenzyl-α-D-galactopyranoside (11): H_2S was bubbled into a solution of **10** (54 mg, 0.066 mmol) in a mixture of pyridine/H_2O/NEt_3 (10:1:0.3) (5 mL) for 3 h. The reaction mixture was co-evaporated with toluene to give a residue which was dissolved in pyridine (3 mL) and acetic anhydride (1 mL). The resulting solution was stirred overnight, concentrated and the residue was purified by flash chromatography (*n*-hexane/acetone 2:1) to afford **11** (41 mg, 74%). $[\alpha]_D$ = +66.7° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=7.44–6.84 (m, 14H, Ar), 5.83–5.74 (m, 1H, OCH_2CH=CH_2), 5.71 (d, 1H, NH), 5.21–5.10 (m, 2H, OCH_2CH=CH_2, 4b-*H*), 4.94 (d, ^3J=3.6 Hz, 1H, 1b-*H*), 4.86–4.67 (m, 5H, 1a-*H*, CH_2Ph), 4.43–4.29 (m, 4H, 2b-*H*, 5b-*H*, CH_2Ph), 4.19 (m, 1H, 4a-*H*), 4.06–3.97 (m, 2H, OCH_2CH=CH_2), 3.88–3.77 (m, 7H, 2a-*H*, 3a-*H*, 5a-*H*, 6b-*H*, OCH_3), 3.62–3.55 (m, 2H, 3b-*H*, 6′b-*H*), 3.42 (m, 2H, 6a-*H*, 6′a-*H*), 3.36 (s, 3H, OCH_3), 2.08–1.94 ppm (m, 9H, 3×$COCH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): δ=138.1–126.8, 116.4, 114.1, 98.4, 98.1, 76.6, 76.4, 75.8, 73.2, 70.8, 68.8, 68.2, 66.9, 61.5, 60.4, 55.4, 55.1, 51.8 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{44}H_{55}NO_{14}Na$: 844.3515; found: 844.3515 [*M*+Na^+].

Methyl 2-acetamido-3-*O*-allyl-6-*O*-methanesulfonyl-2-deoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-methoxybenzyl-α-D-galactopyranoside (12): Compound **11** (30 mg, 0.037 mmol) was dissolved in methanol (3 mL), and NaOMe in methanol (0.54 mL, 0.1 M) was added. After one hour, the reaction mixture was neutralized with Amberlite IR120 (H^+ form), and filtered. The resulting solution was evaporated at low pressure to give the crude diol which was used in the next step without purification. The residue in pyridine (3 mL) was treated with methanesulfonyl chloride (MsCl) (4.5 μL, 0.056 mmol) under argon at −15°C and stirred for 30 min. Then the reaction mixture was slowly warmed to 0°C and kept at this temperature for 3 h. TLC indicated that the starting material had completely reacted. The reaction mixture was quenched with MeOH (1 mL), concentrated and the residue was purified by flash chromatography (*n*-hexane/acetone 3:2) to afford the **12** (22 mg, 71%). $[\alpha]_D$ = +65.0° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=7.44–6.85 (m, 14H, Ar), 5.92–5.84 (m, 1H, OCH_2CH=CH_2), 5.73 (d, 1H, NH), 5.26–5.15 (m, 2H, OCH_2CH=CH_2), 4. 87–4.63 (m, 6H, 1a-*H*, 1b-*H*, 2× CH_2Ph), 4.39 (m, 2H, CH_2Ph), 4.12–4.23 (m, 5H, 4a-*H*, OCH_2CH=CH_2, 6b-*H*, 5b-*H*), 3.96–3.82 (m, 5H, 6′b-*H*, 2a-*H*, 2b-*H*, 3a-*H*, 3b-*H*), 3.81 (s, 3H, OCH_3), 3.68–3.61 (m, 2H, 5a-*H*, 4b-*H*), 3.44–3.38 (m, 6H, 6a-*H*, 6a′-*H*, 5a-*H*, OCH_3), 2.88 (s, 3H, CH_3), 2.58 (d, 1H, OH), 1.98 ppm (s, 3H, Ac); ^{13}C NMR (125 MHz, $CDCl_3$): δ=135.6, 129.5–127.8, 116.8, 114.2, 98.6, 98.2, 79.8, 77.5, 75.8, 74.4, 73.8, 73.6, 69.8, 68.8, 68.6, 68.0, 66.8, 60.6 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{41}H_{53}NO_{14}Na$: 838.3079; found: 838.3083 [*M*+Na^+].

Methyl 2-acetamido-3-*O*-allyl-2,6-dideoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl-α-D-galactopyranoside (13): Sodium borohydride (230 mg, 6.13 mmol) was added to a solution of methyl 2-acetamido-3-*O*-allyl-6-*O*-methanesulfonyl-2-deoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-methoxybenzyl-α-D-galactopyranoside (250 mg, 0.306 mmol) in DMSO (6 mL) under argon at 85°C and the solution was stirred overnight. The resulting mixture was quenched with methanol at room temperature and diluted with ethyl acetate (20 mL). This solution was washed with water (10 mL). The aqueous wash was extracted with ethyl acetate (3×20 mL) and the combined organic extracts were dried over $MgSO_4$ and concentrated in vacuum. The residue was purified by flash chromatography (*n*-hexane/acetone 5:2) to afford **13** (184 mg, 80%). $[\alpha]_D$ = +47.0° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=7.41–6.85 (m, 14H, Ar), 5.93–5.85 (m, 1H, OCH_2CH=CH_2), 5.76 (d, 1H, NH), 5.27–5.14 (m, 2H, OCH_2CH=CH_2), 4.88–4.71 (m, 5H, 1b-*H*, 2×CH_2Ph), 4.66 (d, ^3J=2.8 Hz, 1H, 1a-*H*), 4.43–4.36 (m, 2H, CH_2Ph), 4.26–4.08 (m, 5H, OCH_2CH=CH_2, 2b-*H*, 5b-*H*, 3a-*H*), 3.87–3.80 (m, 6H, 2a-*H*, 6a-*H*, 6a′-*H*, OCH_3), 3.46–3.34 (m, 5H, 3b-*H*, 5a-*H*, OCH_3), 3.28 (t, ^3J=9.6 Hz, 1H, 4b-*H*), 2.26 (m, 1H, OH), 1.93 (s, 3H, $COCH_3$), 1.04 ppm (d, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): δ=135.2, 129.6–127.4, 116.8, 114.2, 98.8, 98.2, 80.2, 77.5, 75.7, 75.5, 73.6, 73.4, 73.2, 72.6, 72.1, 68.8, 68.6, 67.2 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{40}H_{51}NO_{11}Na$: 744.3354; found: 744.3353 [*M*+Na^+].

Methyl 2-acetamido-3-*O*-allyl-4-azido-2,4,6-trideoxy-α-D-galactopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-methoxybenzyl-α-D-galactopyranoside (14): Pyridine (25 μL, 0.287 mmol) and Tf_2O (30 μL, 0.191 mmol) were added to a stirred to a solution of **13** (65 mg, 0.096 mmol) in CH_2Cl_2 (6 mL) at −30°C under argon. After 2.0 h, TLC indicated that the starting material had been consumed. The reaction mixture was diluted with CH_2Cl_2 (10 mL), and extracted with a saturated $NaHCO_3$ solution (10 mL). The aqueous layer was extracted with ethyl acetate (3×10 mL) and the combined organic layers were dried over magnesium sulfate and concentrated in vacuum. The crude triflate was dissolved in dry DMF (2 mL) under argon, NaN_3 (123 mg, 1.914 mmol) was added and the reaction mixture was stirred overnight. The reaction mixture was diluted with ethyl acetate (5 mL), and then washed with water (5 mL). The aqueous layer was extracted with ethyl acetate (3×5 mL) and the combined organic extracts were dried over $MgSO_4$ and concentrated in vacuo. The residue was purified by flash chromatography (*n*-hexane/acetone 3:1→ 2:1) to afford **14** (32 mg, 57%). $[\alpha]_D$ = +86.1° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=7.42–6.84 (m, 14H, Ar), 5.94–5.84 (m, 1H,

$OCH_2CH{=}CH_2$), 5.57 (d, 1H, NH), 5.31–5.18 (m, 2H, $OCH_2CH{=}CH_2$), 4.87–4.72 (m, 6H, 1a-*H*, 1b-*H*, 2×CH_2Ph), 4.87 (d, ^3J=3.6 Hz, 1H, 1b-*H*), 4.53 (ddd, ^3J=3.6, 10.7 Hz, 1H, 2b-*H*), 4.45–4.31 (m, 3H, 5b-*H*, CH_2Ph), 4.22–4.15 (m, 2H, 3a-H, $OCH_2CH{=}CH_2$), 3.99–3.92 (m, 1H, $OCH_2CH{=}CH_2$), 3.87–3.76 (m, 6*H*, 2a-*H*, 4a-*H*, 6a-H, OCH_3), 3.71 (m, 1H, 4b-*H*), 3.58–3.53 (dd, ^3J=3.3, 10.8 Hz, 1H, 3b-*H*), 3.42 (m, 2H, 5a-*H*, 6′a-*H*), 3.37 (s, 3H, OCH_3), 1.95 (s, 3H, $COCH_3$), 0.98 ppm (d, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): δ=135.2, 129.6–127.4, 116.8, 114.2, 98.4, 98.2, 77.0, 76.8, 73.4, 73.3, 73.1, 70.5, 68.8, 67.2, 65.3, 63.3 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{40}H_{50}N_4O_{10}Na$: 769.3419; found: 769.3421 [*M*+Na^+].

Methyl 2-acetamido-3-*O*-allyl-4-azido-2,4,6-trideoxy-α-D-galactopyranosyl-(1→4)-6-*O*-acetyl-2,3-di-*O*-benzyl-α-D-galactopyranoside (15): Trifluoroacetic acid (1%) (20 μL) was added to a solution of disaccharide **14** (70 mg, 0.094 mmol) in CH_2Cl_2 (3 mL) and the mixture was stirred for two hours. TLC check indicated that the starting material had reacted completely. The reaction mixture was neutralized with triethylamine (0.1 mL) and concentrated. The residue was dissolved in a mixture of pyridine (4 mL) and Ac_2O (1 mL) and the solution was stirred for 18 h. Concentration gave the crude product and this residue was purified by flash chromatography (*n*-hexane/acetone 3:1→3:2) to afford **15** (50 mg, 80%). $[\alpha]_D$ = +75.0° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ= 7.42–7.25 (m, 10H, Ar), 5.94–5.83 (m, 1H, $OCH_2CH{=}CH_2$), 5.52 (d, 1H, NH), 5.31–5.19 (m, 2H, $OCH_2CH{=}CH_2$), 4.86 (d, ^3J=3.8 Hz, 1H, 1b-*H*), 4.81–4.72 (m, H, 1a-*H*, 2×CH_2Ph), 4.47 (m, 1H, 2b-*H*), 4.39 (t, 1H, 6a-*H*), 4.27 (t, 1H, 5b-*H*), 4.19 (m, 1H, $OCH_2CH{=}CH_2$), 4.01 (m, 1H, 4a-*H*), 3.95 (m, 1H, $OCH_2CH{=}CH_2$), 3.88–3.75 (m, 4H, 3a-*H*, 2a-*H*, 5a-*H*, 6′a-*H*), 3.71 (m, 1H, 4b-*H*), 3.57 (dd, ^3J=3.3, 10.9 Hz, 1H, 3b-*H*), 3.37 (s, 3H, CH_3), 2.05–1.98 (m, 6H, 2$COCH_3$), 1.00 ppm (d, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): δ=138.2, 134.3, 128.4–127.2, 117.3, 98.8, 98.2, 76.4, 76.2, 75.1, 73.4, 73.1, 68.4, 65.8, 63.2 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{34}H_{44}N_4O_{10}Na$: 691.2949; found: 691.2951 [*M*+Na^+].

Methyl 2-acetamido-4-azido-2,4,6-trideoxy-α-D-galactopyranosyl-(1→4)-6-*O*-acetyl-2,3-di-*O*-benzyl-α-D-galactopyranoside (16): A solution of NaOAc (29 mg, 0.036 mmol) in a mixture of HOAc/H_2O 20:1 (3 mL) was degassed under argon for 30 min. Compound **15** (30 mg, 0.045 mmol) was dissolved in this degassed solution under an atmosphere of argon. $PdCl_2$ (30 mg, 0.135 mmol) was added and the reaction mixture was stirred until TLC indicated that starting material had reacted. The reaction mixture was passed through a filter (0.2 mm) to give a yellow solution, which was diluted with ethyl acetate (10 mL), and extracted with a saturated $NaHCO_3$ solution (10 mL). The aqueous layer was extracted with ethyl acetate (3×10 mL) and the combined organic layers were dried over $MgSO_4$ and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 3:1→3:2) to afford **16** (12 mg, 53%). $[\alpha]_D$ = +88.2° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=7.41–7.25 (m, 10H, Ar), 6.01 (d, 1H, NH), 4.84–4.78 (m, 2H, CH_2Ph), 4.77 (d, ^3J= 3.95 Hz, 1H, 1b-*H*), 4.74–4.69 (m, 3H, 1a-*H*, CH_2Ph), 4.41–4.27 (m, 3H, 2b-*H*, 5b-*H*, 6a-*H*), 4.09 (m, 1H, 4a-*H*), 3.93–3.80 (m, 4H, 2a-*H*, 3b-*H*, 5a-*H*, 6′a-*H*), 3.59 (m, 1H, 4b-*H*), 3.37 (s, 3H, CH_3), 2.07 (m, 6H, 2× $COCH_3$), 0.97 ppm (d, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): δ=135.2, 129.6–127.4, 98.2, 97.8, 76.8, 76.1, 73.8, 73.2, 71.5, 68.2, 66.4, 63.8 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{31}H_{40}N_4O_{10}Na$: 651.2637; found: 651.2639 [*M*+Na^+].

Methyl 2,3,4-tri-*O*-benzyl-6-*O*-acetyl-α-D-galactopyranosyl-(1→3)-2-acetamido-4-azido-2,4,6-trideoxy-α-galactopyranosyl-(1→4)-6-*O*-acetyl-2,3-di-*O*-benzyl-α-D-galactopyranoside (17): NIS (10.2 mg, 0.045 mmol) was added to a solution of **2**[21] (37.2 mg, 0.06 mmol) and **16** (19 mg, 0.03 mmol) containing 4 Å molecular sieves (50 mg) in CH_2Cl_2 (3 mL) under argon at −30°C and the mixture was stirred for 15 min. The resulting mixture was treated with TfOH (1 mg, 0.006 mmol) at −30°C and the reaction mixture was slowly warmed to room temperature. After 30 min, TLC indicated that the starting material had been consumed. The reaction mixture was quenched with triethylamine, diluted with CH_2Cl_2 (10 mL), and this solution was washed with saturated $NaHCO_3$ solution, $Na_2S_2O_3$ solution (10%) and brine. The organic phase was dried over $MgSO_4$, concentrated to give the crude product and this residue was purified by flash chromatography (*n*-hexane/acetone 5:2) to afford **17** (24 mg, 73%). $[\alpha]_D$ = +42.4° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ= 7.44–7.21 (m, 15H, Ar), 5.72 (d, 1H, NH), 4.97 (m, 2H, CH_2Ph), 4.86–4.70 (m, 9H, 1a-*H*, 1b-*H*, 1c-*H*, 3×CH_2Ph), 4.69–4.57 (m, 2H, CH_2Ph), 4.56–4.50 (m, 1H, 2b-*H*), 4.32 (m, 1H, 6a-*H*), 4.19–4.03 (m, 5H, 2c-*H*, 4b-*H*, 5b-*H*, 6b-*H*, 6′b-*H*), 4.00 (m, 1H, 4a-*H*), 3.89–3.79 (m, 5H, 2a-*H*, 3a-*H*, 3c-*H*, 5a-*H*, 6′a-H), 3.67–3.62 (m, 2H, 3b-*H*, 4b-*H*), 3.35 (s, 3H, CH_3), 2.05–1.89 (m, 9H, 3×$COCH_3$), 0.89 ppm (d, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): δ=128.4–127.8, 101.2, 98.6, 98.2, 79.5, 79.1, 77.2, 76.8, 76.4, 76.0, 75.4, 74.8, 74.5, 74.3, 73.3, 73.0, 69.4, 68.3, 65.9 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{60}H_{70}N_4O_{16}Na$: 1125.4679; found: 1125.4672 [*M*+Na^+].

Methyl [benzyl 2,3,4-tri-*O*-benzyl-α-D-galactopyranosiduronate]-(1→3)-2-acetamido-4-azido-2,4,6-trideoxy-α-D-galactopyranosyl-(1→4)-[benzyl 2,3-*O*-dibenzyl-α-D-galactopyranosiduronate (18): Trisaccharide **17** (11 mg, 0.01 mmol) was dissolved in dry MeOH (2 mL), followed by addition of 0.5 M NaOMe in MeOH (0.01 mmol, 20 μL), and the reaction mixture was stirred for one hour. The resulting mixture was neutralized with Amberlite IR120 (H^+ form), filtered and concentrated to give the trisaccharide diol. This diol (10.2 mg, 0.01 mmol) was dissolved in an acetone (1 mL) and $NaHCO_3$ solution (5%) (0.5 mL) and KBr (2.5 mg, 0.02 mmol) and TEMPO (3.8 mg, 0.014 mmol) were added at 0°C. After 5 min, NaOCl (13%, 42 μL) was added and the mixture was stirred for 10 min at 0°C. The reaction mixture was diluted with ethyl acetate (15 mL), washed with brine and dried over $MgSO_4$, and then the resulting organic phase was concentrated. The residue was dissolved in DMF (2 mL), and CsF (20 mg) and BnBr (16 μL) were added under argon, and the mixture was stirred overnight. The reaction mixture was diluted with ethyl acetate (15 mL), washed with brine, dried over $MgSO_4$, and then the resulting organic phase was concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 5:2) to afford **18** (6.3 mg, 51%). $[\alpha]_D$ = +61.5° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ= 7.41–7.17 (m, 35H, Ar), 5.70 (d, 1H, NH), 5.17 (m, 2H, CH_2Ph), 5.00–4.87 (m, 6H, 1a-*H*, 1c-*H*, 2×CH_2Ph), 4.82–4.71 (m, 6H, 3×CH_2Ph), 4.63 (m, 2H, 1/2×CH_2Ph, 5c-*H*), 4.56 (d, 1H, ^3J=3.84 Hz, 1H, 1b-*H*), 4.46–4.40 (m, 2H, 1/2×CH_2Ph, 2b-*H*), 4.32 (m, 1H, 4c-*H*), 4.28 (m, 1H, 3a-*H*), 4.25 (m, 1H, 4a-*H*), 4.20 (dd, ^3J=3.84, 10.1 Hz, 1H, 3c-*H*), 4.13 (dd, ^3J= 3.52, 10.2 Hz, 1H, 2c-*H*), 4.04 (m, 1H, 5b-*H*), 3.81 (m, 2H, 2a-*H*, 5a-*H*), 3.60 (m, 1H, 4b-*H*), 3.54 (dd, ^3J=3.2, 10.8 Hz, 1H, 3b-*H*), 3.38 (s, 3H, CH_3), 1.83 (s, 3H, $COCH_3$), 0.84 ppm (d, 3H, CH_3); (HRMS) EMS: *m*/*z*: calcd for $C_{70}H_{74}N_4O_{16}Na$: 1249.4992; found: 1249.4995 [*M*+Na^+].

Methyl α-D-galactopyranosiduronate-(1→3)-2-acetamido-4-amino-2,4,6-trideoxy-α-D-galactopyranosyl-(1→4)-α-D-galactopyranosiduronate (1): A solution of **18** (11 mg, 9 μmol) in aqueous acetic acid (90%) (5 mL) was hydrogenated at 400 kPa over Pd/C (10%) (50 mg) for 18 h. The reaction mixture was passed through a filter (0.2 mm) and a reverse phase column (Sep-PaK) to give a yellow solution. Gel filtration of the yellow solution on a column (2.5×85 cm) of Biogel P-2 eluting with ammonium acetate (100 mM) gave pure product **1** (3 mg, 58%) as a white powder after repeated lyophilization. $[\alpha]_D$ = +35.6° (*c*=0.5, D_2O); ^{1}H NMR (600 MHz, D_2O): δ=5.06 (d, ^3J=3.4 Hz, 1H, 1c-*H*), 4.97 (d, ^3J=3.96 Hz, 1H, 1b-*H*), 4.92 (d, ^3J=3.84 Hz, 1H, 1a-*H*), 4.75 (m, 1H, 5b-*H*), 4.35 (d, ^3J=3.2 Hz, 1H, 4a-*H*), 4.26 (m, 1H, 4c-*H*), 4.23 (m, 2H, 3b-*H*, 5a-*H*), 4.13 (dd, ^3J= 3.95 Hz, 1H, 2b-*H*), 4.10 (m, 1H, 5c-*H*), 3.98 (dd, ^3J=3.2, 10.6 Hz, 1H, 3a-*H*), 3.89 (dd, ^3J=3.84, 10.5 Hz, 1H, 2a-*H*), 3.85–3.82 (m, 2H, 2c-*H*, 3c-*H*), 3.74 (m, 1H, 4b-*H*), 2.05 (s, 3H, $COCH_3$), 1.30 ppm (d, 3H, CH_3); (HRMS) EMS: *m*/*z*: calcd for $C_{21}H_{33}N_2O_{16}^-$: 569.1836; found: 569.1806 [*M*−H^-].

Phenyl 6-*O*-acetyl-2,4-di-*O*-benzyl-3-*O*-*p*-methoxybenzyl-1-thio-β-D-galactopyranoside (24): Compound **26**[35] (427 mg, 0.95 mmol) was dissolved in dry DMF (20 mL) under argon, followed by addition of *p*-methoxybenzyl chloride (157 mg, 1.0 mmol). The resulting mixture was treated with NaH (60%) (57 mg, 1.43 mmol) at 0°C, and slowly warmed to room temperature. After one hour, the reaction mixture was quenched with methanol, and extracted with ethyl acetate (2×20 mL), then washed with brine. The resulting organic phase was evaporated at low pressure to give crude phenyl 2,4-di-*O*-benzyl-4,6-*O*-benzylidene-3-*O*-*p*-methoxybenzyl-1-thio-β-D-galactopyranoside, which was used without purification. This residue was dissolved CH_2Cl_2 (10 mL) and BH_3·THF in THF (1.0 M)

(9.5 mmol, 9.5 mL) was added. The reaction mixture was stirred for 10 min followed by dropwise addition of Bu_2BOTf (0.95 mL, 0.95 mmol) at 0 °C. TLC indicated that the starting material had disappeared after 2 h. The resulting mixture was neutralized with triethylamine and excess borane was quenched with methanol. The solution was concentrated and the residue was dissolved in a mixture of pyridine (10 mL) and acetic anhydride (5 mL) and stirred overnight. Purification by chromatography gave **24** (436 mg, 75 %) as a colorless oil. $[\alpha]_D = +45.6°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 7.59$–6.87 (m, 14H, Ar), 5.02 (d, $^2J =$ 11.2 Hz, 1H, $1/2\times CH_2Ph$), 4.84–4.76 (dd, 2H, CH_2Ph), 4.71 (s, 2H, CH_2Ph), 4.60 (m, 2H, $1/2\times CH_2Ph$, 1-*H*), 4.29–4.26 (m, 1H, 6-*H*), 4.13–4.10 (m, 1H, 6′-*H*), 3.94 (t, $^3J = 9.5$ Hz, 1H, 2-*H*), 3.82 (m, 4H, CH_3, 4-*H*), 3.58 (m, 2H, 3*H*, 5-*H*), 2.01 ppm (s, 3H, CH_3CO); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.3$, 134.1–127.2, 113.9, 87.8, 83.9, 76.8, 76.0, 75.7, 74.3, 73.4, 72.8 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{36}H_{38}O_7SNa$: 637.2231; found: 637.2234 [M+Na$^+$].

Methyl 2,4-di-*O*-benzyl-3-*O*-*p*-methoxybenzyl-6-*O*-acetyl-α-D-galactopyranosyl-(1→3)-2-acetamido-4-azido-2,4,6-trideoxy-α-galactopyranosyl-(1→4)-6-*O*-acetyl-2,3-di-*O*-benzyl-α-D-galactopyranoside (22): NIS (30 mg, 0.134 mmol) was added to a solution of **24** (86 mg, 0.134 mmol) and **16** (42 mg, 0.067 mmol) containing 4 Å molecular sieves (200 mg) in CH_2Cl_2 (5 mL) under argon at −30 °C. The mixture was stirred for 15 min and then AgOTf (2.5 mg) and slowly warmed to room temperature. After one hour, TLC indicated that the starting material had been completely consumed. The reaction mixture was quenched with triethylamine, diluted with CH_2Cl_2 (10 mL), and the solution was washed with saturated $NaHCO_3$ solution, $Na_2S_2O_3$ solution (10 %) and brine. The organic phase was dried over $MgSO_4$, concentrated and the residue was purified by flash chromatography (*n*-hexane/acetone 5:2) to afford **22** (58 mg, 79 %). $[\alpha]_D = +41.5°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.41$–6.85 (m, 24H, Ar), 5.75 (d, 1H, NH), 4.97 (m, 2H, CH_2Ph), 4.83–4.57 (m, 11H, 1a-*H*, 1b-*H*, 1c-*H*, $4\times CH_2Ph$), 4.54 (m, 1H, 2b- *H*), 4.32 (m, 1H, 6a-*H*), 4.16 (m, 1H, 5b-*H*), 4.13–4.03 (m, 5H, 2c-*H*, 3c-*H*, 5c-*H*, 6c-*H*, 6′c-*H*), 4.00 (m, 1H, 4a-*H*), 3.87–3.79 (m, 8H, 2a-*H*, 3a-*H*, 4c-*H*, 5a-*H*, 6′a-*H*, CH_3), 3.68–3.63 (m, 2H, 3b-*H*, 4b-*H*), 3.35 (s, 3H, CH_3), 2.03–1.92 (m, 9H, $3\times COCH_3$), 0.90 ppm (d, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.6$–127.6, 113.8, 101.3, 98.8, 98.3, 79.3, 78.7, 76.6, 76.0, 75.4, 74.8, 74.7, 74.4, 73.2, 73.0, 72.9, 69.4, 68.5, 65.7, 64.0, 62.7, 61.9 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{61}H_{72}N_4O_{17}Na$: 1155.4785; found: 1155.4782 [M+Na$^+$].

Methyl 2,4-di-*O*-benzyl-6-*O*-acetyl-α-D-galactopyranosyl-(1→3)-2-acetamido-4-azido-2,4,6-trideoxy-α-galactopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-acetyl-α-D-galactopyranoside (21): DDQ (26 mg, 0.115 mmol) was added a solution of **22** (26 mg, 0.023 mmol) in a mixture of CH_2Cl_2 and H_2O (10:1, 5 mL) and this mixture was stirred for 2 h. After dilution with CH_2Cl_2 (10 mL) the organic phase was washed with $NaHCO_3$ solution, dried over $MgSO_4$ and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 2:1) to afford **21** (17 mg, 73 %). $[\alpha]_D = +21.5°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.41$–6.85 (m, 20H, Ar), 5.72 (d, 1H, NH), 4.91–4.62 (m, 11H, 1a-*H*, 1b-*H*, 1c-*H*, $4\times CH_2Ph$), 4.54 (m, 1H, 2b-*H*), 4.32 (m, 1H, 6c-*H*), 4.42–4.00 (m, 5H, 4a-*H*, 5a-*H*, 5b-*H*, 6a-*H*, 6′a-*H*), 3.92–3.80 (m, 5H, 2a-*H*, 3a-*H*, 4c-*H*, 5c-*H*, 6′c-*H*), 3.71 (dd, $^3J = 3.2$, 11.0 Hz, 1H, 3b-*H*), 3.53 (m, 1H, 4b-*H*), 3.35 (s, 3H, CH_3), 2.03–1.93 (m, 9H, $3\times CH_3CO$), 0.94 ppm (d, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.3$–127.6, 98.6, 98.5, 98.4, 76.9, 76.8, 76.6, 75.9, 75.7, 75.2, 74.5, 73.9, 73.4, 73.1, 70.0, 69.0, 68.4, 65.8, 63.6, 62.8, 61.7 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{53}H_{64}N_4O_{16}Na$: 1035.4209; found: 1035.4214 [M+Na$^+$].

Dimethylthexylsilyl 2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl-β-D-galactopyranoside (25): A solution of dimethylthexylsilyl 2,3-di-*O*-benzyl-4,6-*O*-*p*-methoxybenzylidene-β-D-galactopyranoside[21] (180 mg, 0.29 mmol) and molecule sieve 4 Å (100 mg) in dry THF (6 mL) was cooled to 0 °C, and $NaCNBH_3$ (183 mg, 2.9 mmol) was added, followed by dropwise addition of hydrogen chloride solution in Et_2O (1 M, 1 mL). The reaction was continued until evolution gas ceased. After 30 min, the reaction mixture was diluted with CH_2Cl_2 (10 mL), and extracted with saturated $NaHCO_3$ solution and brine. The organic phase was dried over $MgSO_4$ and evaporated at low pressure to give a residue, that was purified by flash chromatography (hexanes/ethyl acetate 4:1) to afford **25** (117 mg, 65 %). $[\alpha]_D = +11.2°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.38$–6.38 (m, 14H, Ar), 4.93 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.76 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.71 (m, 2H, CH_2Ph), 4.61 (d, $^3J = 7.4$ Hz, 1H, 1-*H*), 4.52 (s, 2H, CH_2Ph), 4.02 (m, 1H, 4-*H*), 3.82 (s, 3H, CH_3), 3.78 (m, 1H, 6-*H*), 3.68 (m, 1H, 6′-*H*), 3.59 (dd, $^3J = 7.4$, 9.4 Hz, 1H, 2-*H*), 3.54 (t, $^3J =$ 5.9 Hz, 1H, 5-*H*), 3.49 (dd, $^3J = 3.4$, 9.4 Hz, 1H, 3-*H*), 2.51 (br s, 1H, OH), 1.71 (m, 1H, $CH(CH_3)_2$), 0.91 (m, 12H, $4\times CH_3$), 0.21 ppm (m, 6H, $2\times CH_3$); (HRMS) EMS: *m*/*z*: calcd for $C_{36}H_{50}O_7SiNa$: 645.3218; found: 645.3219 [M+Na$^+$].

Dimethylthexylsilyl 4,6-di-*O*-acetyl-3-*O*-allyl-2-azido-2-deoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-methoxybenzyl-α-D-galactopyranoside (29): A mixture of **30** (197 mg, 0.6 mmol), diphenylsulfoxide (242 mg, 1.2 mmol) and tri-*tert*-butylpyrimidine (446 mg, 1.8 mmol) in CH_2Cl_2 (8 mL) were stirred over flame dried 4 Å molecule sieves (200 mg) for 30 min. Then the reaction mixture was cooled to −60 °C, Tf_2O (120 μL, 0.72 mmol) was added and the mixture was first brought to −40 °C and then slowly warmed to −20 °C over a period of approximately 1.5 h. After the reaction was cooled to −25 °C, a solution of the acceptor **25** (302 mg, 0.485 mmol) in CH_2Cl_2 (4 mL) was added dropwise. The resulting mixture was warmed very slowly to room temperature (typically overnight), after which time the reaction was quenched with triethylamine and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 4:1) to afford **29** (330 mg, 73 %). $[\alpha]_D = +53.3°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.67$–6.89 (m, 14H, Ar), 5.88–5.95 (m, 1H, OCH_2CH=CH_2), 5.18–5.31 (m, 2H, OCH_2CH=CH_2), 5.04 (t, $^3J = 10.3$ Hz, 1H, 4b-*H*), 4.94 (m, 2H, 1b-*H*, $1/2\times CH_2Ph$), 4.82 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.69 (m, 2H, CH_2Ph), 4.61 (d, $^3J = 7.2$ Hz, 1H, 1a-*H*), 4.46 (s, 2H, CH_2Ph), 4.34–4.28 (m, 2H, 5b-*H*, OCH_2CH=CH_2), 4.14 (m, 1H, OCH_2CH=CH_2), 4.08 (d, $^3J = 3.1$ Hz, 1H, 4a-*H*), 3.90–3.83 (m, 3H, 3b-*H*, 6b-*H*, 6′b-*H*), 3.81 (s, 3H, CH_3), 3.60–3.43 (m, 4H, 2a-*H*, 5a-*H*, 6a-*H*, 6′a-*H*), 3.38 (dd, $^3J = 3.1$, 10.0 Hz, 1H, 3a-*H*), 3.30 (dd, $^3J = 3.5$, 10.3 Hz, 1H, 2b-*H*), 2.08–2.00 (m, 6H, $2\times CH_3CO$), 1.69 (m, 1H, $CH(CH_3)_2$), 0.91 (m, 12H, $4\times CH_3$), 0.19 ppm (m, 6H, $2\times CH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.6$–124.8, 117.3, 113.9, 113.8, 98.6, 98.5, 80.9, 79.9, 74.7, 74.4, 73.4, 73.1, 73.0, 72.7, 69.7, 67.9, 66.9, 63.3, 61.3 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{49}H_{67}N_3O_{13}SiNa$: 956.4335; found: 956.4330 [M+Na$^+$].

Dimethylthexylsilyl 2-acetamido-4,6-di-*O*-acetyl-3-*O*-allyl-2-deoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl-α-D-galactopyranoside (31): H_2S was bubbled into a solution of **29** (330 mg, 0.355 mmol) in a mixture of pyridine/H_2O/NEt_3 10:1:0.3 (10 mL) for 3 h. TLC indicated that the starting material had disappeared and the reaction mixture was co-evaporated with toluene at to give the amino form of the disaccharide. The residue was dissolved in pyridine (10 mL) and acetic anhydride (4 mL) and resulting solution was stirred overnight and then concentrated to give a residue that was purified by flash chromatography (*n*-hexane/acetone 3:1) to afford **31** (255 mg, 75 %). $[\alpha]_D = +61.2°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.38$–6.87 (m, 14H, Ar), 5.85–5.77 (m, 2H, NH, OCH_2CH=CH_2), 5.24–5.10 (m, 3H, 4b-*H*, OCH_2CH=CH_2), 4.93 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.92 (d, $^3J =$ 3.7 Hz. 1H, 1b-*H*), 4.84 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.74–4.66 (m, 2H, CH_2Ph), 4.63 (d, $^3J = 7.2$ Hz, 1H, 1a-*H*), 4.44–4.30 (m, 4H, 2b-*H*, 5b-*H*, CH_2Ph), 4.20 (d, $^3J = 2.95$ Hz, 1H, 4a-*H*), 4.10–4.00 (m, 2H, OCH_2CH=CH_2), 3.82–3.76 (m, 4H, 6b-*H*, CH_3), 3.71 (t, $^3J = 10.5$, 9.8 Hz, 1H, 3b-*H*), 3.60–3.56 (m, 2H, 2a-*H*, 6′b-*H*), 3.52–3.44 (m, 3H, 5a-*H*, 6a-*H*, 6′a-*H*), 3.41 (dd, $^3J = 9.7$, 3.0 Hz, 1H, 3a-*H*), 2.15–2.03 (m, 9H, $3\times CH_3CO$), 1.70 (m, 1H, $CH(CH_3)_2$), 0.91 (m, 12H, $4\times CH_3$), 0.19 ppm (m, 6H, $2\times CH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.4$–127.2, 116.8, 114.0, 98.4, 97.7, 81.2, 79.9, 76.8, 74.9, 73.2, 72.7, 72.6, 71.6, 70.7, 68.6, 68.2, 66.8, 61.4, 60.3 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{51}H_{71}NO_{14}SiNa$: 972.4536; found: 972.4550 [M+Na$^+$].

Dimethylthexylsilyl 2-acetamido-3-*O*-allyl-6-*O*-methanesulfonyl-2-deoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-methoxybenzyl-α-D-galactopyranoside (32): Compound **31** (512 mg, 0.54 mmol) was dissolved in methanol (20 mL), and sodium methoxide (87 mg, 1.62 mmol) was added. After one hour, the reaction mixture was neutralized with Amberlite IR120 (H$^+$ form) and filtered. The resulting solution was evaporated

at low pressure. The crude disaccharide diol was used directly in the next step. Methansulfonyl chloride (62 μL, 0.81 mmol) in pyridine (30 mL) under argon at −15 °C was added to the diol and the solution was stirred for 30 min, then slowly warmed to 0 °C and kept at this temperature for 3 h. The reaction mixture was quenched with MeOH (5 mL) and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 3:1) to afford **32** (483 mg, 95 %). $[\alpha]_D = +35.1°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.38$–6.87 (m, 14H, Ar), 5.94–5.86 (m, 1H, $OCH_2CH{=}CH_2$), 5.84 (d, 1H, NH), 5.31–5.18 (m, 2H, $OCH_2CH{=}CH_2$), 4.94 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.84–4.81 (m, 2H, 1b-*H*, $1/2\times CH_2Ph$), 4.74 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.65–4.62 (m, 2H, 1a-*H*, $1/2\times CH_2Ph$), 4.39 (m, 2H, CH_2Ph), 4.27–4.14 (m, 5H, $OCH_2CH{=}CH_2$, 2b-*H*, 5b-*H*, 4a-*H*), 3.95 (m, 1H, 6b-*H*), 3.82 (s, 3H, CH_3), 3.66 (m, 2H, 4b-*H*, 6′b-*H*), 3.58–3.45 (m, 5H, 2a-*H*, 3b-*H*, 5a-*H*, 6a-*H*, 6′a-*H*), 3.41 (dd, $^3J = 9.78$, 2.86 Hz, 1H, 3a-*H*), 2.90 (s, 3H, CH_3), 1.96 (s, 3H, CH_3), 1.69 (m, 1H, $CH(CH_3)_2$), 0.92 (m, 12H, $4\times CH_3$), 0.19 ppm (m, 6H, $2\times CH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.4$–127.4, 117.2, 114.0, 98.5, 98.0, 81.5, 80.5, 79.5, 77.4, 77.1, 76.9, 75.1, 73.3, 73.1, 72.7, 72.6, 71.9, 69.8, 68.9, 67.9, 66.8 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{48}H_{69}NO_{14}SiNa$: 966.4100; found: 966.4101 [M+Na$^+$].

Dimethylthexylsilyl 2-acetamido-3-*O*-allyl-2,6-dideoxy-α-D-glucopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl-α-D-galactopyranoside (33): Sodium borohydride (230 mg, 6.13 mmol) was added to a solution of **32** (200 mg, 0.21 mmol) in DMSO (10 mL) under argon at 85 °C, and the solution was stirred overnight. The resulting mixture was quenched with methanol at room temperature, and the mixture was diluted with ethyl acetate (15 mL), and the resultant mixture was washed with water (10 mL). The aqueous layer was extracted with ethyl acetate (3×15 mL) and the combined organic extracts were dried over $MgSO_4$ and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 3:1) to afford **33** (153 mg, 85 %). $[\alpha]_D = +45.2°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.37$–6.87 (m, 14H, Ar), 5.95–5.88 (m, 2H, NH, $OCH_2CH{=}CH_2$), 5.32–5.18 (m, 2H, $OCH_2CH{=}CH_2$), 4.90–4.82 (m, 2H, CH_2Ph), 4.79–4.76 (m, 2H, 1b-*H*, $1/2\times CH_2Ph$), 4.63–4.61 (m, 2H, 1a-*H*, $1/2\times CH_2Ph$), 4.40 (m, 2H, CH_2Ph), 4.27–2.12 (m, 5H, $OCH_2CH{=}CH_2$, 2b-*H*, 4a-*H*, 5b-*H*), 3.82 (s, 3H, CH_3), 3.55 (dd, $^3J = 7.2$, 9.8 Hz, 1H, 2a-*H*), 3.50 (t, $^3J = 9.1$ Hz, 1H, 3b-*H*), 3.48 (m, 3H, 5a-*H*, 6a-*H*, 6′b-*H*), 3.40 (dd, $^3J = 2.9$, 9.7 Hz, 1H, 3a-*H*), 3.28 (t, $^3J = 9.3$ Hz, 1H, 4b-*H*), 1.95 (s, 3H, CH_3CO), 1.69 (m, 1H, $CH(CH_3)_2$), 1.05 (d, 3H, CH_3), 0.91 (m, 12H, $4\times CH_3$), 0.18 ppm (m, 6H, $2\times CH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.6$–127.3, 117.2, 114.0, 98.4, 98.0, 81.2, 80.3, 80.1, 75.2, 75.1, 73.3, 72.9, 72.0, 71.9, 70.9, 68.0, 67.0 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{47}H_{67}NO_{11}SiNa$: 872.4376; found: 872.4375 [M+Na$^+$].

Dimethylthexylsilyl 2-acetamido-3-*O*-allyl-4-azido-2,4,6-trideoxy-α-D-galactopyranosyl-(1→4)-2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl-α-D-galactopyranoside (34): Pyridine (42 μL, 0.494 mmol) and Tf_2O (52 μL, 0.33 mmol) were added to a solution of **33** (140 mg, 0.165 mmol) in CH_2Cl_2 (10 mL) at −30 °C, and the reaction mixture was stirred at this temperature under argon for two hours. The reaction mixture was diluted with CH_2Cl_2 (10 mL), and extracted with saturated $NaHCO_3$ solution (10 mL). The aqueous layer was extracted with ethyl acetate (3×10 mL) and the combined organic layers were dried over $MgSO_4$ and concentrated in vacuum. The crude product was dissolved in dry DMF (4 mL) under argon, and NaN_3 (123 mg, 1.914 mmol) was added. The reaction mixture was stirred overnight. Then the reaction mixture was diluted with ethyl acetate (10 mL), and washed with water (10 mL). The aqueous layer was extracted with ethyl acetate (3×10 mL) and the combined organic extracts were dried over $MgSO_4$ and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 3:1→2:1) to afford **34** (90 mg, 62 %). $[\alpha]_D = +32.4°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.39$–6.86 (m, 14H, Ar), 5.95–5.87 (m, 1H, $OCH_2CH{=}CH_2$), 5.69 (d, 1H, NH), 5.35–5.23 (m, 2H, $OCH_2CH{=}CH_2$), 4.93 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.85–4.79 (m, 2H, 1b-*H*, $1/2\times CH_2Ph$), 4.74 (d, 1H, $1/2\times CH_2Ph$), 4.65–4.62 (m, 2H, 1a-*H*, $1/2\times CH_2Ph$), 4.52 (m, 1H, 2b-*H*), 4.44–4.33 (m, 3H, 5b-*H*, CH_2Ph), 4.25–4.20 (m, 1H, $OCH_2CH{=}CH_2$), 4.18 (m, 1H, 4a-*H*), 4.05–4.00 (m, 1H, $OCH_2CH{=}CH_2$), 3.82 (s, 3H, CH_3), 3.71 (m, 1H, 4b-*H*), 3.66 (dd, $^3J = 3.3$, 10.7 Hz, 1H, 3b-*H*), 3.52–3.45 (m, 4H, 2a-*H*, 5a-*H*, 6a-*H*, 6′a-*H*), 3.39 (dd, $^3J = 2.9$, 9.8 Hz, 1H, 3a-*H*), 1.96 (s, 3H, CH_3CO), 1.71 (m, 1H, $CH(CH_3)_2$), 1.00 (d, 3H, CH_3), 0.92 (m, 12H, $4\times CH_3$), 0.19 ppm (m, 6H, $2\times CH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 134.4$–127.3, 117.4, 113.9, 98.4, 97.9, 81.1, 80.3, 76.4, 74.9, 73.2, 72.9, 72.3, 70.9, 70.5, 66.9, 65.2, 63.4 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{47}H_{66}N_4O_{10}SiNa$: 897.4440; found: 897.4436 [M+Na$^+$].

Dimethylthexylsilyl 2-acetamido-3-*O*-allyl-4-azido-2,4,6-trideoxy-α-D-galactopyranosyl-(1→4)-6-*O*-acetyl-2,3-di-*O*-benzyl-α-D-galactopyranoside (35): Compound **34** (45 mg, 0.051 mmol) was dissolved in a mixture of CH_2Cl_2 and H_2O solvents (10:1, 5 mL), followed by addition of DDQ (47 mg, 0.206 mmol) and the reaction mixture was stirred for 2 h. The reaction mixture was diluted with CH_2Cl_2 (10 mL), and washed with $NaHCO_3$ solution the organic phase was dried over $MgSO_4$ and then concentrated. The crude produce was dissolved in pyridine (4 mL) and acetic anhydrate (2 mL), and stirred overnight. The reaction mixture was concentrated and the residue was purified by flash chromatography (*n*-hexane/acetone 3:1) to afford **35** (32 mg, 80 %). $[\alpha]_D = +43.4°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.41$–7.24 (m, 10H, Ar), 5.98–5.84 (m, 1H, $OCH_2CH{=}CH_2$), 5.77 (d, 1H, NH), 5.38–5.20 (m, 2H, $OCH_2CH{=}CH_2$), 4.94 (d, 1H, $1/2\times CH_2Ph$), 4.83–4.76 (m, 2H, 1b-H, $1/2\times CH_2Ph$), 4.71 (m, 2H, CH_2Ph), 4.61 (d, $^2J = 7.1$ Hz, 1H, 1a-*H*), 4.53 (m, 1H, 2b-*H*), 4.43–4.36 (m, 1H, 6a-*H*), 4.31 (m, 1H, 5b-*H*), 4.26–4.18 (m, 1H, $OCH_2CH{=}CH_2$), 4.06–3.96 (m, 1H, $OCH_2CH{=}CH_2$), 3.90 (m, 1H, 6′a-*H*), 3.71–3.67 (m, 2H, 3b-*H*, 4b-*H*), 3.54–3.48 (m, 2H, 2a-*H*, 5a-*H*), 3.39 (dd, $^3J = 2.8$, 9.8 Hz, 1H, 3a-*H*), 2.02 (m, 6H, $2\times CH_3CO$), 1.69 (m, 1H, $CH(CH_3)_2$), 1.00 (d, 3H, CH_3), 0.92 (m, 12H, $4\times CH_3$), 0.20 ppm (m, 6H, $2\times CH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.3$–127.3, 98.5, 97.3, 80.9, 79.8, 75.2, 72.8, 72.4, 72.1, 71.6, 66.5, 65.7, 61.9 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{41}H_{60}N_4O_{10}SiNa$: 819.3971; found: 819.3973 [M+Na$^+$].

Dimethylthexylsilyl 2-acetamido-4-azido-2,4,6-trideoxy-α-D-galactopyranosyl-(1→4)-6-*O*-acetyl-2,3-di-*O*-benzyl-α-D-galactopyranoside (36): A solution of NaOAc (26 mg, 0.32 mmol) in a mixture of HOAc/H_2O (20:1) (4 mL) was degassed under argon for 30 min. Compound **35** (32 mg, 0.04 mmol) was dissolved in the degassed above solution and $PdCl_2$ (27 mg, 0.12 mmol) was added, and mixture was stirred under argon until the starting material had reacted. The reaction mixture was passed through a filter (0.2 mm) to give a yellow solution, followed by dilution with ethyl acetate (10 mL), and then extracted with saturated $NaHCO_3$ solution (10 mL). The aqueous layer was extracted with ethyl acetate ($3\times$ 10 mL) and the combined organic layers were dried over $MgSO_4$ and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 3:1→2:1) to afford **36** (20 mg, 66 %). $[\alpha]_D = +51.5°$ ($c =$ 1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.37$–7.25 (m, 10H, Ar), 6.22 (d, 1H, NH), 4.92 (d, $^2J = 11.2$ Hz, 1H, $1/2\times CH_2Ph$), 4.79–4.76 (m, 2H, 1b-*H*, $1/2\times CH_2Ph$), 4.71 (s, 2H, CH_2Ph), 4.62 (d, $^2J = 7.25$ Hz, 1H, 1a-*H*), 4.45 (m, 1H, 6a-*H*), 4.36–4.30 (m, 2H, 2b-*H*, 5b-*H*), 4.06 (m, 1H, 4a-*H*), 4.00 (dd, $^3J = 3.5$, 10.5 Hz, 1H, 3b-*H*), 3.90 (m, 1H, 6′a-*H*), 3.60 (m, 1H, 4b-*H*), 3.57–3.53 (m, 2H, 2a-*H*, 5a-*H*), 3.42 (dd, $^3J = 9.67$, 2.96 Hz, 1H, 3a-*H*), 2.07 (m, 6H, $2\times CH_3CO$), 1.69 (m, 1H, $CH(CH_3)_2$), 0.99 (d, 3H, CH_3), 0.91 (m, 12H, $4\times CH_3$), 0.18 ppm (m, 6H, $2\times CH_3$); ^{13}C NMR (125 MHz, $CDCl_3$): $\delta = 138.3$–127.3, 98.5, 97.3, 80.9, 79.8, 75.2, 72.8, 72.4, 72.1, 71.6, 66.5, 65.7, 61.9, 50.7 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{38}H_{56}N_4O_{10}SiNa$: 779.3658; found: 779.3658 [M+Na$^+$].

Dimethylthexylsilyl 6-*O*-acetyl-2,3,4-tri-*O*-benzyl-α-D-galactopyranosyl-(1→3)-2-acetamido-4-azido-2,4,6-trideoxy-α-galactopyranosyl-(1→4)-6-*O*-acetyl-2,3-di-*O*-benzyl-α-D-galactopyranoside (37): NIS (12 mg, 0.053 mmol)was added to a solution of containing acceptor **36** (20 mg, 0.026 mmol), donor **2** (33 mg, 0.053 mmol) and 4 Å molecular sieves (200 mg) in CH_2Cl_2 (5 mL) under argon at −30 °C. The mixture was stirred at that temperature for 15 min, then AgOTf (1 mg) was added at −30 °C and the reaction mixture was slowly warmed to room temperature. After 1 h, the reaction was complete as judged by TLC. The reaction mixture was quenched with triethylamine, diluted with CH_2Cl_2 (5 mL), and this solution was extracted with saturated $NaHCO_3$ solution, $Na_2S_2O_3$ solution (10 %) and brine. The organic phase was dried with $MgSO_4$ and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 7:2) to afford **37** (21 mg, 66 %). $[\alpha]_D = +23.4°$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 7.37$–7.25 (m, 25H, Ar), 5.90 (d, 1H, NH), 4.98–4.93 (m, 3H, $3/2\times CH_2Ph$), 4.88 (d, $^3J =$ 3.1 Hz, 1H, 1c-*H*), 4.85–4.72 (m, 5H, 1b-*H*, $2\times CH_2Ph$), 4.67 (m, 2H,

CH_2Ph), 4.59 (m, 2H, 1a-*H*, 1/2×CH_2Ph), 4.55 (m, 1H, 2b-*H*), 4.35 (m, 1H, 6a-*H*), 4.16 (m, 1H, 5b-*H*), 4.13–4.08 (m, 5H, 2c-*H*, 3c-*H*, 5c-*H*, 6c-*H*, 6′c-*H*), 3.97 (m, 1H, 4a-*H*), 3.92–3.88 (m, 2H, 4c-*H*, 6′a-*H*), 3.73 (dd, ^3J=3.2, 10.8 Hz, 1H, 3b-*H*), 3.67 (m, 1H, 4b-*H*), 3.54–3.48 (m, 2H, 2a-*H*, 5a-*H*), 3.37 (dd, ^3J=10.2, 3.0 Hz, 1H, 3a-*H*), 2.04–1.96 (m, 9H, 3×CH_3CO), 1.68 (m, 1H, $CH(CH_3)_2$), 0.9 (m, 15H, 5×CH_3), 0.18 ppm (m, 6H, 2×CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): δ=138.7–127.4, 102.0, 98.7, 98.5, 80.6, 80.5, 79.8, 79.0, 76.3, 74.9, 74.7, 74.5, 74.3, 73.2, 72.9, 72.8, 72.5, 69.9, 65.6, 64.3, 62.4, 61.8 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{67}H_{86}N_4O_{16}SiNa$: 1253.5700; found: 1253.5698 [*M*+Na^+].

6-*O*-Acetyl-2,3,4-tri-*O*-benzyl-α-D-galactopyranosyl-(1→3)-2-acetamido-4-azido-2,4,6-trideoxy-α-galactopyranosyl-(1→4)-6-*O*-acetyl-2,3-*O*-dibenzyl-α/β-D-galactopyranose (38): HOAc (70 μL) and TBAF (0.7 mL, 0.62 mmol) were added to a solution of **37** (306 mg, 0.249 mmol) in THF (10 mL) and the solution was then stirred for two hours. The resulting mixture was diluted with ethyl acetate (20 mL) and this solution was extracted with saturated $NaHCO_3$ solution. The aqueous layer was extracted with ethyl acetate (3×20 mL) and the combined organic layers were dried over $MgSO_4$ and concentrated. The residue was purified by flash chromatography (*n*-hexane/acetone 2:1→1:1) to afford **38** (250 mg, 92%) directly for next step.

6-*O*-Acetyl-2,3,4-tri-*O*-benzyl-α-D-galactopyranosyl-(1→3)-2-acetamido-4-azido-2,4,6-trideoxy-α-galactopyranosyl-(1→4)-2,3-*O*-dibenzyl-6-*O*-acetyl-α-D-galactopyranosyl trichloroacetimidate (23): Compound **38** (108 mg, 0.1 mmol) was dissolved in a mixture of CH_2Cl_2 (5 mL) and Cl_3CCN (0.11 mL) under argon, and then one drop of DBU was added at room temperature. After 2 h, the resulting mixture was concentrated. The residue was purified by flash chromatography (hexanes/ethyl acetate 2:1) to afford **23** (87 mg, 71%). $[\alpha]_D$ = +35.2° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=8.58 (s, 1H, NH), 7.42–7.23 (m, 25H, Ar), 6.55 (d, ^3J=3.5 Hz, 1H, 1a-*H*), 5.67 (d, 1H, NH), 4.98 (m, 2H, CH_2Ph), 4.84 (m, 2H, 1b-*H*, 1c-*H*), 4.81–4.60 (m, 8H, 4×CH_2Ph), 4.55 (m, 1H, 2b-*H*), 4.28 (m, 1H, 6a-*H*), 4.20 (m, 1H, 5b-*H*), 4.15 (m, 1H, 6c-*H*), 4.11–4.05 (m, 6H, 2c-*H*, 4c-*H*, 5c-*H*, 6′c-*H*, 4a-*H*, 6′a-*H*), 4.02 (dd, ^3J=10.2, 3.5 Hz, 1H, 2a-*H*), 3.92–3.88 (m, 3H, 3a-*H*, 3c-*H*, 5a-*H*), 3.67 (m, 2H, 3b-*H*, 4b-*H*), 2.02 (m, 9H, 3×CH_3CO), 0.95 ppm (d, 1H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): δ=138.6–127.6, 100.9, 98.7, 94.2, 78.9, 78.9, 75.9, 75.6, 74.8, 74.7, 74.4, 74.3, 73.7, 73.3, 73.1, 72.6, 71.4, 69.5, 65.8, 63.9, 62.7, 61.4 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{61}H_{68}Cl_3N_5O_{16}Na$: 1254.3727; found: 1254.3725 [*M*+Na^+].

Glycoside 39: Glycosyl donor **23** (173 mg, 0.1 mmol), monosaccharide acceptor **21** (95 mg, 0.094 mmol) and activated 4 Å molecular sieves (200 mg) were dried together under vacuum for 1 h in a pear-shaped flask (25 mL). The contents of the flask were then dissolved in CH_2Cl_2 (10 mL). The suspension was stirred for 10 min at room temperature under argon. Then the temperature was reduced to −15 °C, and TMSOTf (47 μL, 0.1 M in CH_2Cl_2) was added dropwise and the mixture was slowly warmed to room temperature. After 30 min, the reaction mixture was neutralized with triethylamine and concentrated. The residue was purified by flash chromatography (toluene/acetone 10:1) to afford **39** (166 mg, 85%). $[\alpha]_D$ = +45.2° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=4.70 (1a-H), 3.78 (2a-H), 3.82 (3a-H), 4.00 (4a-H); 4.82 (1b-H), 4.55 (2b-H), 3.66 (3b-H, 4b-H), 4.18 (5b-H); 4.86 (1c-H), 4.12 (2c-H, 3c-H), 3.96 (4c-H); 5.28 (^2J=3.4 Hz, 1d-H), 3.88 (2d-H), 3.81 (3d-H), 3.94 (4d-H), 4.04 (5d-H), 3.72 (6d-H), 4.28 (6′d-H), 4.81 (1e-H), 4.48 (2e-H), 3.62 (3e-H), 3.53 (4e-H), 4.16 (5e-H), 4.73 (1f-H), 3.97 (2f-H), 4.14 (3f-H), 4.09 (4f-H), 3.89 (5f-H), 3.81 (6f-H), 3.83 ppm (6′f-H); ^{13}C NMR (125 MHz, $CDCl_3$): δ=138.6–127.7, 101.2, 99.07, 98.7, 98.3, 97.3, 79.3, 78.9, 76.8, 76.6, 76.3, 76.1, 76.0, 75.7, 75.6, 74.8, 74.7, 74.6, 74.5, 74.4, 73.8, 73.3, 73.2, 73.1, 72.3, 69.5, 69.1, 68.4, 65.7, 64.1, 62.7, 62.6, 61.7, 61.2, 55.3, 48.9, 48.8 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{112}H_{130}N_8O_{31}Na$: 2105.8734; found: 2105.8721.

Glycoside 20: Compound **39** (166 mg, 0.08 mmol) was dissolved in methanol (5 mL), and NaOMe (21.6 mg, 0.4 mmol) was added. After 1 h, the reaction mixture was neutralized with Amberlite IR120 (H^+ form), and filtered. The resulting solution was evaporated at low pressure to give the crude 40. An aqueous solution of NaBr (1 M, 12.5 μL), an aqueous solution of TBABr (1 M, 25 μL), TEMPO (1.5 mg) and a saturated aqueous solution of $NaHCO_3$ (62 μL) were added to a solution of crude **40** (10 mg, 5 μmol) in CH_2Cl_2 (0.5 mL) and H_2O (85 μL) in an ice-water bath. An aqueous solution of NaOCl (available chlorine 10–15%, 75 μL) was added and the reaction mixture was continuously stirred for 80 min. As the temperature increased from 0 °C to room temperature, the reaction was neutralized with HCl (0.5 1 M HCl, about 20 μL) to pH 6–7. After neutralization, *t*BuOH (0.35 mL), 2-methylbut-2-ene in THF (2 M, 0.7 mL) and a solution of $NaClO_2$ (25 mg) and NaH_2PO_4 in water (100 μL) were added. The reaction mixture was kept at room temperature for two hours. The reaction mixture was diluted with saturated aqueous NaH_2PO4 solution (5 mL), and extracted with ethyl acetate (3× 10 mL). The organic layers were combined and dried over $MgSO_4$. The crude product from the oxidation reaction was dissolved in DMF (1 mL), followed by addition of CsF (10 mg) and BnBr (40 μL). The reaction mixture was stirred overnight and the resulting mixture was diluted with ethyl acetate (5 mL) and extracted with saturated $NaHCO_3$ solution. The aqueous layer was extracted with ethyl acetate (3×5 mL) and the combined organic layers were dried over $MgSO_4$ and concentrated in vacuum. The residue was purified by flash chromatography (*n*-hexane/acetone 3:1→12:1) to afford **20** (6 mg, 52%). $[\alpha]_D$ = +25.4° (*c*=1.0, $CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$): δ=4.91 (1a-H), 3.82 (2a-H), 3.82 (3a-H), 4.33 (4a-H), 4.25 (5a-H); 4.61 (1b-H), 4.38 (2b-H), 3.52 (3b-H), 3.35 (4b-H), 4.06 (5b-H); 4.86 (1c-H), 3.97 (2c-H), 4.23 (3c-H), 4.31 (4c-H), 4.62 (5c-H); 5.43 (1d-H), 3.87 (2d-H), 3.76 (3d-H), 4.13 (4d-H), 4.34 (5d-H); 4.51 (1e-H), 4.43 (2e-H), 3.51 (3e-H), 3.60 (4e-H), 4.01 (5e-H); 4.93 (1f-H), 4.12 (2f-H), 4.20 (3f-H), 4.30 (4f-H), 4.53 ppm (5f-H); ^{13}C NMR (125 MHz, $CDCl_3$): δ=138.7–126.9, 102.7, 101.9, 99.5, 98.9, 98.4, 95.7, 82.3, 81.9, 78.6, 76.3, 76.0, 75.9, 75.3, 74.8, 74.6, 74.5, 73.8, 73.5, 73.2, 73.1, 72.4, 72.0, 71.7, 70.6, 69.5, 67.5, 67.2, 66.9, 65.7, 65.4, 64.4, 64.2 ppm; (HRMS) EMS: *m*/*z*: calcd for $C_{132}H_{138}N_8O_{31}Na$: 2353.9360; found: 2353.9347 [*M*+Na^+].

Glycoside 19: Compound **20** (6 mg, 2.57 μmol) was dissolved in a mixture of CH_3OH and CH_2Cl_2 and H_2O (3:1:0.5, 4.5 mL), and $Pd(OH)_2$ (20 mg) were also added to the reaction mixture. The reaction mixture was hydrogenated overnight and then passed through a filter (0.2 mm) and reverse phase column (Sep-Pak) to give a yellow solution. Gel filtration of the yellow solution on a column (2.5×85 cm) of Biogel P-2 eluting with ammonium acetate (100 mM) gave the pure product **19** (1.5 mg, 53%) as a white powder after repeated lyophilization. $[\alpha]_D$ = +83.9° (*c*=0.4, D_2O); ^{1}H NMR (600 MHz, D_2O): δ=4.91 (1a-H), 3.89 (2a-H), 3.98 (3a-H), 4.36 (4a-H), 4.24 (5a-H); 4.97 (1b-H), 4.13 (2b-H), 4.28 (3b-H), 3.84 (4b-H), 4.79 (5b-H); 5.06 (1c-H), 3.94 (2c-H), 4.02 (3c-H), 4.46 (4c-H), 4.13 (5c-H); 5.22 (1d-H), 3.98 (2d-H), 4.08 (3d-H), 4.39 (4d-H), 4.61 (5d-H); 4.99 (1e-H), 4.13 (2e-H), 4.25 (3e-H), 3.79 (4e-H), 4.77 (5e-H); 5.08 (1f-H), 3.88 (2f-H), 3.86 (3f-H), 4.27 (4f-H), 4.13 ppm (5f-H); (HRMS) EMS: *m*/*z*: calcd for $C_{41}H_{64}N_4O_{31}$: 1108.3406 [*M*−2H^-]; found: 553.1703 [*M*−2H^{-2}].

Acknowledgements

We thank Dr. Dennis Kasper Harvard medical school for preliminary assays to test for T-cell activity in compounds **1** and **19**. These studies were supported by a Discovery grant from the Natural Science and Engineering Research Council of Canada (NSERC) and funds from the Alberta Ingenuity Foundation's Centre's program.

[1] a) M. H. Nahm, M. A. Apicella, D. E. Briles, *Immunity to Extracellular Bacteria, in Fundamental Immunology* (Ed.: W. E., Paul) Lippincott-Raven, New York, **1999**, pp. 1373–1386; b) L. G. Rubin, *Pediatr. Clin. North Am.* **2000**, *47*, 269–285; c) M. J. Jedrzejas, *Microbiol. Mol. Biol. Rev.* **2001**, *65*, 187–207.

[2] a) A. O. Tzianabos, R. W. Finberg, Y. Wang, M. Chan, A. B. Onderdonk, H. J. Jennings, D. L. Kaper, *J. Biol. Chem.* **2000**, *275*, 6733–6740; b) J. O. Brubaker, Q. Li, A. O. Tzianabos, D. L. Kasper, R. W. Finberg, *J. Immunol.* **1999**, *162*, 2235–2242; c) P. J. Baker, *J. Infect.*

Dis. **1992**, *165*, S44–S48; d) N. M. Zirk, S. F. Hashmi, H. K. Ziegler, *Infect. Immun.* **1999**, *67*, 319–326.
[3] J. Duan, F. Y. Avci, D. L. Kasper, *Proc. Natl. Acad. Sci. USA* **2008**, *105*, 5183–5188.
[4] a) A. O. Tzianabos, A. B. Onderdonk, D. F. Zaleznik, R. S. Smith, D. L. Kasper, *Infect. Immun.* **1994**, *62*, 4881–4886; b) S. Gallorini, F. Berti, P. Parente, R. Baronio, S. Aprea, U. D′Oro, M. Pizza, J. L. Telford, A. Wack, *J. Immunol.* **2007**, *179*, 8208–8215.
[5] W. M. Kalka-Moll, A. O. Tzianabos, Y. Wang, V. J. Carey, R. W. Finberg, A. B. Onderdonk, D. L. Kasper, *J. Immunol.* **2000**, *164*, 719–724.
[6] Y.-H. Choi, M. H. Roehrl, D. L. Kasper, J. Y. Wang, *Biochemistry* **2002**, *41*, 15144–15151.
[7] A. O. Tzianabos, J. Y. Wang, J. C. Lee, *Proc. Natl. Acad. Sci. USA* **2001**, *98*, 9365–9370.
[8] L. J. van den Bos, T. J. Boltje, T. Provoost, J. Mazurek, H. S. Overkleeft, G. A. van der Marel, *Tetrahedron Lett.* **2007**, *48*, 2697–2700.
[9] L. J. van den Bos, Ph.D. Thesis, Leiden University (The Netherlands), **2007**.
[10] a) J.-H. Kim, H. Yang, G.-J. Boons, *Angew. Chem.* **2005**, *117*, 969–971; *Angew. Chem. Int. Ed.* **2005**, *44*, 947–949; b) J.-H. Kim, H. Yang, J. Park, G.-J. Boons, *J. Am. Chem. Soc.* **2005**, *127*, 12090–12097.
[11] a) Y. Nakahara, T. Ogawa, *Tetrahedron Lett.* **1987**, *28*, 2731–2734; b) L. J. Van Den Bos, J. D. C. Codee, R. E. J. N. Litjens, J. Dinkelaar, H. S. Overkleeft, G. A. Van der Marcel, *Eur. J. Org. Chem.* **2007**, 3963–3976; c) M. Haller, G.-J. Boons, *J. Chem. Soc. Perkin Trans. 1* **2001**, 814–822.
[12] a) H. Lönn, J. Lonngren, *J. Carbohydr. Chem.* **1984**, *132*, 39–44; b) N. J. Davis, S. L. Flitsch, *Tetrahedron Lett.* **1993**, *34*, 1181–1184.
[13] J. Madaj, M. Jankowska, A. Wiśniewski, *Carbohydr. Res.* **2004**, *339*, 1293–1300.
[14] K.-H. Jung, M. Müller, R. R. Schmidt, *Chem. Rev.* **2000**, *100*, 4423–4442.
[15] J.-H. Kim, H. Yang, J. Park, G.-J. Boons, *J. Am. Chem. Soc.* **2005**, *127*, 12090–12097.
[16] J.-H. Kim, H. Yang, G.-J. Boons, *Angew. Chem.* **2005**, *117*, 969–971; *Angew. Chem. Int. Ed.* **2005**, *44*, 947–949.
[17] a) H. Paulsen, C. Kolar, W. Stenzel, *Chem. Ber.* **1978**, *111*, 2358–2369; b) R. U. Lemieux, R. M. Lemieux, *Can. J. Chem.* **1979**, *57*, 1244–1251; c) For review of synthesis of oligosaccharide of 2-amino-2-deoxy sugars: J. Banoub, P. Boullanger, D. Lafont, *Chem. Rev.* **1992**, *92*, 1167–1195.
[18] S. Manabe, K. Ishii, Y. Ito, *J. Am. Chem. Soc.* **2006**, *128*, 10666–10667.
[19] a) D. Crich, T. Hu, F. Cai, *J. Org. Chem.* **2008**, *73*, 8942–8953; b) Y.-P. Cheng, H.-T. Chen, C.-C. Lin, *Tetrahedron Lett.* **2002**, *43*, 7721–7723; c) A. V. Demchenko, E. Rousson, G.-J. Boons, *Tetrahedron Lett.* **1999**, *40*, 6523–6526.
[20] a) A. Stadelmaier, Ph.D. Thesis, Konstanz University (Germany), **2003**; b) G. Grundler, R. R. Schmidt, *Liebigs Ann. Chem.* **1984**, 1826–1847.
[21] L. Jiang, T.-H. Chan, *Tetrahedron Lett.* **1998**, *39*, 355–358.
[22] R. V. Stick, D. Tilbrook, G. Mattew, *Aust. J. Chem.* **1990**, *43*, 1643–1655.
[23] P. B. Alper, S.-C. Hung, C.-H. Wong, *Tetrahedron Lett.* **1996**, *37*, 6029–6032.
[24] M. Grathwohl, R. R. Schmidt, *Synthesis* **2001**, 2263–2272.
[25] a) M. V. Chiesa, R. R. Schmidt, *Eur. J. Org. Chem.* **2000**, 3541–3554; b) K. C. Nicolaou, C. A. Veale, C.-K. Hwang, J. Hutchinson, C. V. C. Prasad, W. W. Ogilvie, *Angew. Chem.* **1991**, *103*, 304–308; *Angew. Chem. Int. Ed. Engl.* **1991**, *30*, 299–303.
[26] A. B. Smith III, G. R. Ott, *J. Am. Chem. Soc.* **1996**, *118*, 13095–13096.
[27] a) R. R. Schmidt, *Angew. Chem.* **1986**, *98*, 213–236; *Angew. Chem. Int. Ed. Engl.* **1986**, *25*, 212–235; b) R. R. Schmidt, W. Kinzy, *Adv. Carbohydr. Chem. Biochem.* **1994**, *50*, 21–123.
[28] E. Petrakova, U. Spohr, R. U. Lemieux, *Can. J. Chem.* **1992**, *70*, 233–240.
[29] B. Liessem, A. Giannis, K. Sandhoff, M. Nieger, *Carbohydr. Res.* **1993**, *250*, 19–30.
[30] J. Xue, Z. Guo, *J. Am. Chem. Soc.* **2003**, *125*, 16334–16339.
[31] P. J. Garegg, *Adv. Carbohydr. Chem. Biochem.* **1997**, *52*, 179–205.
[32] K. Bock, C. J. Pedersen, *J. Chem. Soc. Perkin Trans. 1* **1974**, *2*, 293–297.
[33] T. Sato, J. Otera, H. Nozaki, *J. Org. Chem.* **1992**, *57*, 2166–2169.
[34] L. Huang, N. Teumelsan, X. Huang, *Chem. Eur. J.* **2006**, *12*, 5246–5252.
[35] a) P. L. Barili, G. Berti, D. Bertozzi, G. Catelani, F. Colonna, T. Corsetti, F. D′Andrea, *Tetrahedron* **1990**, *46*, 5365–5376; b) H. M. Branderhorst, R. J. Liskamp, R. J. Pieters, *Tetrahedron* **2007**, *63*, 4290–4296.
[36] P. J. Garegg, *Pure. Appl. Chem.* **1984**, *56*, 845–858.
[37] T. A. Boebel, D. Y. Gin, *Angew. Chem.* **2003**, *115*, 6054–6057; *Angew. Chem. Int. Ed.* **2003**, *42*, 5874–5877.

Received: September 6, 2009
Published online: February 9, 2010

