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# Synthesis of the putative minimal FGF binding motif heparan sulfate trisaccharides by an orthogonal protecting group strategy☆,☆☆

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ABSTRACT

The synthesis of two trisaccharides, the putative minimal heparan sulfate sequences responsible for binding to acidic and basic fibroblast growth factors, respectively, is described from a common protected intermediate using an orthogonal protecting group strategy.



## 1. Introduction

Heparin (H) and heparan sulfate (HS) are sulfated glycosaminoglycans (GAGs) built up of linear chains of alternating D-glucosamine and hexuronic acid units.[1–4] The carbohydrate backbone, consisting of →4)-α-L-Ido*p*A-(1→4)-α-D-Glc*p*N-(1→ and →4)-β-D-Glc*p*A-(1→4)-α-D-Glc*p*N-(1→ disaccharide units, is substituted at certain positions. The variations of the hexuronic acid units (L-iduronic or D-glucuronic acid), the substitutions of the amino groups (with acetyl or sulfate groups), and the sulfations of hydroxyl groups (O-3 and O-6 of the D-glucosamine, O-2 of the uronic acids) result in an enormous structural diversity. Heparin and heparan sulfate interact with a large number of proteins including enzymes, protease inhibitors, growth factors, chemokines, and several others.[5–8] These glycosaminoglycan/protein interactions regulate a series of important biological processes. Though the binding of heparin and heparan sulfate to some proteins is nonspecific, simply ionic by nature, many of the biologically relevant interactions of H/HS show a great deal of specificity. It is commonly assumed, though not rigorously proven, that specific oligosaccharide sequences with a unique sulfation pattern are required to mediate the activities of individual heparin-binding proteins.[5,9]

Fibroblast growth factors (FGFs) are a family of more than 20 structurally related signaling polypeptides involved in processes such as cell proliferation, differentiation, and migration.[10–12] The biological activities of FGFs are tightly regulated by heparin and heparan sulfate.[13–16] Heparan sulfate interacts both with FGFs and their cell surface receptors (FGFRs); FGF signaling requires the formation of ternary complex between FGF, FGFR, and HS.[17] The structural requirements of heparin and heparan sulfate to modulate FGF signaling are intensely studied, and a great deal of information in relation to the best studied acidic (FGF-1) and basic fibroblast growth factors (FGF-2) has emerged.[13,14] It was determined that FGF-1 and FGF-2 have distinct O-sulfation requirements for activation:[18] both 2-O-sulfation [Ido*p*A(2-$OSO_3$)] and 6-O-sulfation [Glc*p*N(6-$OSO_3$)] are needed for binding to FGF-1,[19] whereas FGF-2 requires 2-O-sulfation [Ido*p*A(2-$OSO_3$)] only, 6-O-sulfation is not essential for binding.[20] In addition to the gross structural features of H/HS required for FGF-1 and FGF-2 binding, a number of different oligosaccharide structures having high affinity for these FGFs have been proposed.[20–30] Recently, the heparan sulfate sequence **1** has been identified as the minimal binding motif for FGF-1, and sequence **2** as a high affinity binding epitope for FGF-2 (Fig. 1).[31]

These structures were deduced from the study of a heparan sulfate octasaccharide library, the actual trisaccharides were not tested as they were not available. The trisaccharides **1** and **2**, with

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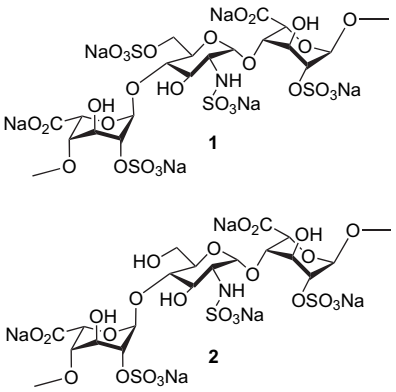



**Figure 1.** Minimal heparan sulfate binding motifs of FGF-1 and FGF-2.

L-iduronic acid at the reducing end, are not obtainable by chemical or enzymatic degradation of heparin, as the current methods afford only oligosaccharides with the amino sugar (or its degradation product) at the reducing end. The chemical synthesis of the suggested trisaccharide ligands has not been accomplished yet.

The chemical synthesis of H/HS oligosaccharides is intensely studied[32–36] and a number of oligosaccharides have been synthesized in relation to FGFs by different groups.[37–42] Syntheses of heparin oligosaccharides are elaborate, multi-step processes, and a common feature of the previous syntheses is that they afford a single heparin oligosaccharide after a long synthetic sequence.

In a program on the synthesis of glycosaminoglycan oligosaccharides, we have previously developed[43,44] a synthesis strategy based on orthogonal protection, which allows the synthesis of multiple sulfated oligosaccharides from a single protected precursor. We now report on the synthesis of the methyl α-glycosides (**3** and **4**) of the putative FGF binding trisaccharides (**1** and **2**) using this orthogonal protection-based methodology.

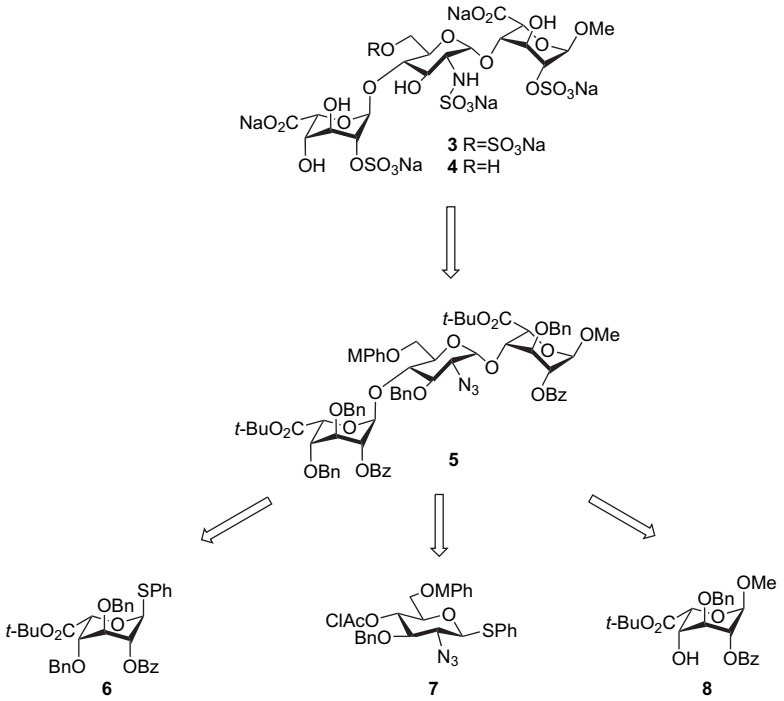
## 2. Results and discussion

### 2.1. Retrosynthesis of the trisaccharides

The target trisaccharides (**3** and **4**) share the same carbohydrate backbone and differ only in their sulfation pattern. Retrosynthesis of **3** and **4** (Scheme 1) revealed that both compounds could be accessed from a single common intermediate, if the O-2 positions of the L-iduronic acid units and O-6 of the D-glucosamine residue are protected by orthogonal protecting groups. The benzoyl group was selected for the O-2 protection of the L-iduronic acids, due to its advantageous properties in glycosylation reactions[45] and the (4-methoxy)phenyl (MPh) group[46] for the protection of O-6 of the D-glucosamine residue. The latter group, which can be removed with ceric ammonium nitrate (CAN) similarly to the commonly used 4-methoxybenzyl residue, was preferred due to its more robust, acid-resistant character. We have shown previously[43,44] that these groups are not only orthogonal, they are also stable under O-sulfations and can be selectively removed without affecting the sensitive *O*-sulfate groups. The orthogonally protected trisaccharide **5** should be available from the monosaccharide building blocks **6**, **7**, and **8**.

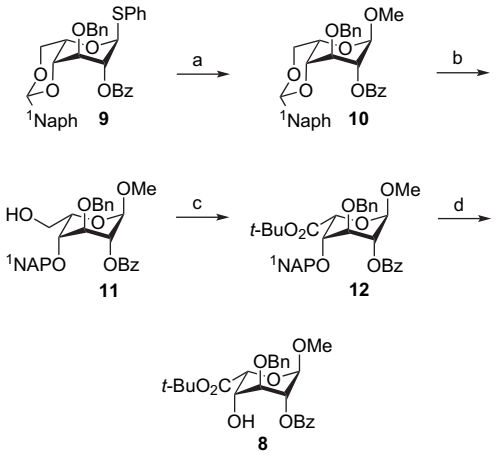
### 2.2. Synthesis of the L-iduronic acid building blocks

We have recently described[47] an efficient synthesis for the non-reducing end L-iduronic acid thioglycoside **6**. For the synthesis of **8** (Scheme 2), the thioglycoside **9**[47] was chosen as starting material. Glycosylation of **9** with methanol in dichloromethane in the presence of dimethyl(methylthio)sulfonium triflate[45] (DMTST) afforded an anomeric mixture of the methyl glycosides, from which the α-glycoside **10** was obtained in 49% yield. A significant amount (35%) of the β-glycoside was also isolated. Although various L-iduronic acid donors generally give high α stereoselectivity in reactions with complex glycosyl acceptors in oligosaccharide syntheses, formation of anomeric mixtures with a high proportion of the β-glycoside are not unprecedented in glycosylations of simple alcohols[48–50] such as




**Scheme 1.** Retrosynthesis of trisaccharides **3** and **4**.

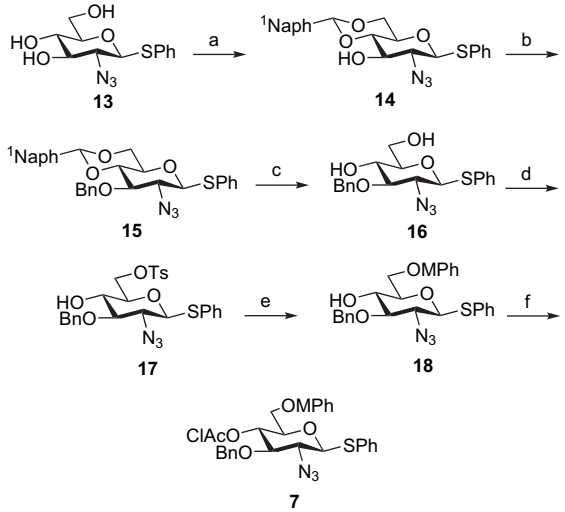
methanol or isopropyl alcohol. Regioselective reductive opening of the (1-naphthyl)methylene acetal **10** with $BH_3 \cdot THF/TMSOTf$[51] gave the corresponding 4-*O*-(1-naphthyl)methyl derivative **11**. Oxidation of **11** using pyridinium dichromate and acetic anhydride in the presence of *tert*-butanol[52] afforded the *tert*-butyl uronate **12**. The L-iduronic acid glycosyl acceptor **8** was obtained by oxidative removal of the (1-naphthyl)methyl group with CAN to give **8** in 76% yield.




**Scheme 2.** Synthesis of L-iduronic acid glycosyl acceptor **8**; (a) DMTST, MeOH, $CH_2Cl_2$, 49%; (b) $BH_3 \cdot THF$, TMSOTf, $CH_2Cl_2$, 66%; (c) PDC, $Ac_2O$, *t*-BuOH, $CH_2Cl_2$, 67%; (d) CAN, MeCN, $H_2O$, 76%.

## 2.3. Synthesis of the D-glucosamine building block

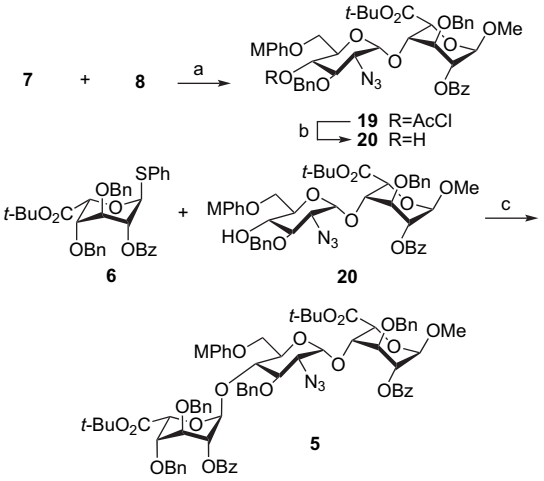
The known triol **13**[53] was reacted with 1-naphthaldehyde dimethyl acetal in DMF solution in the presence of camphorsulfonic acid to afford **14** (Scheme 3). After benzylation of the free hydroxyl group, acid hydrolysis of the (1-naphthyl)methylene acetal of **15** led to **16**. Regioselective tosylation of the primary hydroxyl group of **16** and nucleophilic replacement of the resulting tosylate **17** with sodium (4-methoxy)phenoxide gave the 6-*O*-(4-methoxy)phenyl derivative **18** in 83% yield. Chloroacetylation of **18** then afforded the D-glucosamine building block **7**.




**Scheme 3.** Synthesis of the D-glucosamine building block **7**; (a) $^1NaphCH(OMe)_2$, CSA, DMF, 75%; (b) BnBr, NaH, DMF, 86%; (c) 60% aq AcOH, 90%; (d) TsCl, DMAP, $Et_3N$, $CH_2Cl_2$; (e) 4-methoxyphenol, NaH, DMF, 83% for two steps; (f) $(ClCH_2CO)_2O$, pyridine, $CH_2Cl_2$, 81%.

## 2.4. Synthesis of the orthogonally protected trisaccharide

DMTST-promoted glycosylation[45] of the L-iduronic acid acceptor **8** with the D-glucosamine thioglycoside **7** resulted stereoselectively in the α-linked disaccharide **19** (Scheme 4). Removal of the temporary chloroacetyl protecting group by hydrazinedithiocarbonate[54] afforded the alcohol **20**. This disaccharide acceptor (**20**) was coupled with the L-iduronic acid thioglycoside **6** in the presence of DMTST to give the orthogonally protected trisaccharide **5** in 61% yield.




**Scheme 4.** Synthesis of the orthogonally protected trisaccharide **5**; (a) DMTST, $Et_2O$, $CH_2Cl_2$, 47%; (b) HDTC, DMF, 80%; (c) DMTST, $CH_2Cl_2$, 61%.

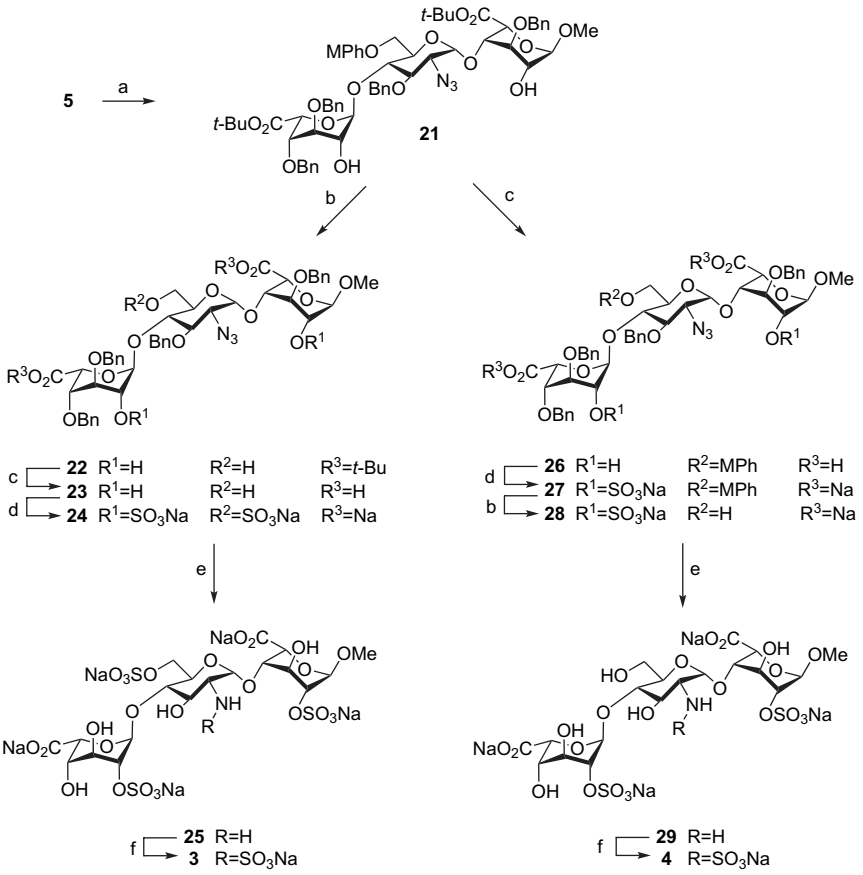
## 2.5. Preparation of heparan sulfate trisaccharides

Debenzoylation of compound **5** (Scheme 5) using mild Zemplén deacylation prevented possible β-elimination of the base-sensitive L-iduronic residues. Though the reaction was sluggish due to the isolated nature of the benzoyl groups[55] it provided the diol **21** in high yield. Cleavage of the (4-methoxy)phenyl group by treatment with CAN afforded triol **22**. After ester hydrolysis, the free acid **23** was sulfated with sulfur trioxide pyridine complex in DMF and the product was converted into pentasodium salt (**24**) by Dowex 50 ($Na^+$ form) resin. Catalytic hydrogenation with palladium on carbon removed the benzyl groups and reduced the azido group. Chemoselective N-sulfation of **25** using sulfur trioxide trimethylamine complex in water at pH 9.8 afforded the putative FGF-1 binding trisaccharide (**3**).

A different sequence of deprotection and sulfation steps starting from the same orthogonally protected trisaccharide (**5**) also afforded the putative FGF-2 binding trisaccharide (**4**). First the *tert*-butyl esters were cleaved from the debenzoylated trisaccharide **21** to give **26**. The dihydroxy uronic acid, still having the (4-methoxy)phenyl group, was first sulfated, and the (4-methoxy)phenyl group was removed from **27** by treatment with CAN to give **28** having the sulfate esters intact (**28**). Hydrogenolysis followed by N-sulfation of the unprotected trisaccharide **29** resulted in the putative FGF-2 binding trisaccharide (**4**).

## 3. Conclusion

In summary, we have synthesized the heparan sulfate trisaccharides **3** and **4**, the putative minimal binding epitopes of FGF-1 and FGF-2, respectively. Both target compounds were readily prepared from the same orthogonally protected trisaccharide **5**. The synthesis relies on a new pair of orthogonal protecting groups [benzoyl and (4-methoxy)phenyl], which, in addition to their




**Scheme 5.** Synthesis of the putative FGF binding trisaccharides **3** and **4**; (a) NaOMe, MeOH 93%; (b) CAN, MeCN, $H_2O$, 84% for **22**, 83% for **28**; (c) $CF_3CO_2H$, $CH_2Cl_2$, 98% for **23**, 92% for **26**; (d) Py·$SO_3$, DMF, 68% for **24**, 99% for **27**; (e) Pd/C, THF, $H_2O$; (f) $NMe_3$·$SO_3$, NaOH, $H_2O$, 45% for two steps for **3**, 68% for two steps for **4**.

orthogonality, can also be removed selectively in the presence of *O*-sulfate groups.

The synthesis also demonstrates that the preparation of multiple heparin/heparan sulfate oligosaccharides is more efficient and less time consuming by our orthogonal protection strategy, than synthesizing these oligosaccharides individually.

## 4. Experimental

### 4.1. General methods

Organic solutions were dried over $MgSO_4$ and concentrated under reduced pressure at 40 °C. Thin-layer chromatography (TLC) was performed on Silica gel 60 $F_{254}$ plates (E. Merck, Darmstadt), the compounds were detected under UV light and by spraying the plates with a 0.02 M solution of resorcinol in 20% methanolic $H_2SO_4$ solution followed by heating. For column chromatography, silica gel 60 (0.040–0.063 mm) (E. Merck) was employed. Melting points were determined in capillary tubes on a Griffin melting point apparatus and are uncorrected. Optical rotations were measured at 23 °C with an Optical Activity AA-10R polarimeter. The NMR spectra were recorded on a Varian Gemini 2000 ($^1$H: 200 MHz; $^{13}$C: 50 MHz) and Varian Unity-Inova ($^1$H: 400 MHz; $^{13}$C: 100 MHz) spectrometers at ambient temperature. The chemical shifts were referenced to TMS (0.00 ppm for $^1$H) and to the central line of $CDCl_3$ (77.16 ppm for $^{13}$C) for solutions in $CDCl_3$, to the central line of the solvent (3.31 ppm for $^1$H, 49.00 ppm for $^{13}$C) for solutions in $CD_3OD$, and to the methyl signal of acetone (2.22 ppm for $^1$H, 30.89 ppm for $^{13}$C) for solutions in $D_2O$, as internal standards. Elemental analyses were performed with an Elementar Vario EL III instrument at the Analytical Department of the Chemical Research Center, Hungarian Academy of Sciences.

### 4.2. Methyl 2-*O*-benzoyl-3-*O*-benzyl-4,6-*O*-(1-naphthyl)-methylidene-α-L-idopyranoside (10)

A mixture of **9**[47] (4.0 g, 6.6 mmol), MeOH (2.6 mL, 66.0 mmol), and 4 Å molecular sieves (3 g) was stirred in dry $CH_2Cl_2$ (30 mL) under argon for 30 min, then DMTST (5.1 g, 19.8 mmol) was added. After 1 day, the reaction was quenched with triethylamine (5 mL). The mixture was filtered through a pad of Celite, the filtrate was diluted with $CH_2Cl_2$ (500 mL) and washed with 2 M aq HCl (100 mL), saturated aq $NaHCO_3$ (100 mL), and water (100 mL), dried, and concentrated. The residue was purified by column chromatography (95:5 toluene/EtOAc) to give first **10** (1.7 g, 49%); mp 140–141 °C (from EtOAc/hexanes); $[\alpha]_D$ −102 (*c* 0.4, $CHCl_3$); $^1$H NMR (400 MHz, $CDCl_3$): $\delta$ 8.42 (m, 1H, aromatic), 7.87–7.80 (m, 4H, aromatic), 7.71 (dd, 1H, aromatic), 7.45–7.24 (m, 9H, aromatic), 7.11–7.06 (m, 2H, aromatic), 6.06 (s, 1H, $^1$NaphCH), 5.29 (ddd, 1H, $J_{1,2}$ 1.7 Hz, $J_{2,3}$ 2.9 Hz, $J_{2,4}$ 0.8 Hz, H-2), 5.06 (d, 1H, $J_{1,2}$ 1.7 Hz, H-1), 4.88 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.70 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.43 (dd, 1H, $J_{5,6a}$ 1.6 Hz, $J_{6a,6b}$ 12.5 Hz, H-6a), 4.25 (dd, 1H, $J_{5,6b}$ 1.8 Hz, $J_{6a,6b}$ 12.5 Hz, H-6b), 4.16 (ddd, 1H, $J_{2,4}$ 0.8 Hz, $J_{3,4}$ 2.5 Hz, $J_{4,5}$ 1.5 Hz, H-4), 4.09 (ddd, 1H, $J_{4,5}$ 1.5 Hz, $J_{5,6a}$ 1.6 Hz, $J_{5,6b}$ 1.8 Hz, H-5), 3.81 (dd, 1H, $J_{2,3}$ 2.9 Hz, $J_{3,4}$ 2.5 Hz, H-3), 3.52 (s, 3H, $OCH_3$); $^{13}$C NMR (100 MHz, $CDCl_3$): $\delta$ 165.8 (*O*C(O)Ph), 137.7, 133.9, 133.4, 133.0, 130.6, 130.1, 129.7, 129.5, 128.5, 128.3, 128.1, 127.92, 127.88, 126.3, 125.6, 125.0, 124.93, 124.86 (aromatic), 101.2 ($^1$NaphCH), 100.0 (C-1, $J_{C\text{-}1,H\text{-}1}$ 168.4 Hz), 74.6 (C-3), 74.1 (C-4), 72.1 ($PhCH_2$), 70.0 (C-6), 66.8 (C-2), 59.8 (C-5), 55.8 ($OCH_3$). Anal. Calcd for $C_{32}H_{30}O_7$: C, 72.99; H, 5.74. Found: C, 72.76; H, 5.79.

Eluted second was the β-anomer, methyl 2-*O*-benzoyl-3-*O*-benzyl-4,6-*O*-(1-naphthyl)methylidene-β-L-idopyranoside (1.2 g, 35%) as a syrup; $[\alpha]_D$ +40 (*c* 0.4, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$): δ 8.20 (m, 1H, aromatic), 8.01–7.95 (m, 2H, aromatic), 7.85–7.70 (m, 3H, aromatic), 7.52–7.34 (m, 8H, aromatic), 7.25–7.09 (m, 3H, aromatic), 6.09 (s, 1H, $^1$NaphCH), 5.35 (ddd, 1H, $J_{1,2}$ 1.6 Hz, $J_{2,3}$ 2.8 Hz, $J_{2,4}$ 0.6 Hz, H-2), 4.92 (d, 1H, $J_{1,2}$ 1.6 Hz, H-1), 4.86 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.73 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.52 (dd, 1H, $J_{5,6a}$ 2.0 Hz, $J_{6a,6b}$ 12.4 Hz, H-6a), 4.26 (dd, 1H, $J_{5,6b}$ 1.5 Hz, $J_{6a,6b}$ 12.4 Hz, H-6b), 4.09 (ddd, 1H, $J_{2,4}$ 0.6 Hz, $J_{3,4}$ 2.4 Hz, $J_{4,5}$ 1.5 Hz, H-4), 4.01 (dd, 1H, $J_{2,3}$ 2.8 Hz, $J_{3,4}$ 2.4 Hz, H-3), 3.87 (ddd, 1H, $J_{4,5}$ 1.5 Hz, $J_{5,6a}$ 2.0 Hz, $J_{5,6b}$ 1.5 Hz, H-5), 3.60 (s, 3H, $OCH_3$); $^{13}C$ NMR (100 MHz, $CDCl_3$): δ 166.5 (O*C*(O)Ph), 137.4, 133.7, 133.1, 132.8, 130.5, 130.3, 129.9, 129.5, 128.6, 128.4, 128.2, 128.0, 127.8, 126.3, 125.5, 124.9, 124.7, 124.2 (aromatic), 100.0 ($^1$NaphCH), 99.2 (C-1, $J_{C\text{-}1,H\text{-}1}$ 156.8 Hz), 75.9 (C-3), 73.3 (C-4), 72.8 ($PhCH_2$), 69.9 (C-6), 67.0 (C-5), 66.7 (C-2), 57.1 ($OCH_3$). Anal. Calcd for $C_{32}H_{30}O_7$: C, 72.99; H, 5.74. Found: C, 72.75; H, 5.72.

### 4.3. Methyl 2-*O*-benzoyl-3-*O*-benzyl-4-*O*-(1-naphthyl)-methyl-α-L-idopyranoside (11)

To a solution of **10** (2.6 g, 4.9 mmol) in dry $CH_2Cl_2$ (50 mL), 1 M $BH_3$·THF in THF (9.8 mL, 9.8 mmol) and TMSOTf (0.09 mL, 0.49 mmol) were added. The mixture was stirred for 2 h at room temperature, cooled to 0 °C, and quenched with triethylamine (5 mL) and MeOH (25 mL). The solution was concentrated in vacuo and co-evaporated three times with MeOH (25 mL). The residue was purified by column chromatography (98:2 → 9:1 toluene/acetone) to give **11** (1.7 g, 66%) as a syrup; $[\alpha]_D$ −23 (*c* 0.4, $CHCl_3$); $^1H$ NMR (200 MHz, $CDCl_3$): δ 8.04–7.72 (m, 5H, aromatic), 7.57–7.08 (m, 12H, aromatic), 5.22 (ddd, 1H, $J_{1,2}$ 1.6 Hz, $J_{2,3}$ 2.9 Hz, $J_{2,4}$ 0.7 Hz, H-2), 4.94 (d, 1H, *J* 11.7 Hz, $ArCH_2$), 4.88 (d, 1H, $J_{1,2}$ 1.6 Hz, H-1), 4.83 (d, 1H, *J* 11.7 Hz, $ArCH_2$), 4.68 (d, 1H, *J* 12.1 Hz, $ArCH_2$), 4.62 (d, 1H, *J* 12.1 Hz, $ArCH_2$), 4.23 (ddd, 1H, $J_{4,5}$ 2.8 Hz, $J_{5,6a}$ 4.4 Hz, $J_{5,6b}$ 6.6 Hz, H-5), 3.96 (dd, 1H, $J_{2,3}$ 2.9 Hz, $J_{3,4}$ 3.0 Hz, H-3), 3.88 (dd, 1H, $J_{5,6a}$ 4.4 Hz, $J_{6a,6b}$ 8.8 Hz, H-6a), 3.61 (dd, $J_{5,6b}$ 6.6 Hz, $J_{6a,6b}$ 8.8 Hz, 1H, H-6b), 3.56 (ddd, 1H, $J_{2,4}$ 0.7 Hz, $J_{3,4}$ 3.0 Hz, $J_{4,5}$ 2.8 Hz, H-4), 3.44 (s, 3H, $OCH_3$), 1.96 (m, 1H, OH); $^{13}C$ NMR (50 MHz, $CDCl_3$): δ 165.7 (O*C*(O)Ph), 137.8, 133.8, 133.3, 132.8, 131.6, 130.1, 129.7, 129.1, 128.7, 128.6, 128.5, 128.2, 128.1, 127.2, 126.5, 126.0, 125.2, 123.8 (aromatic), 99.7 (C-1), 73.8 (C-4), 72.4, 71.9 ($2ArCH_2$), 70.5 (C-3), 68.7, 67.7 (C-2, C-5), 62.7 (C-6), 55.7 ($OCH_3$). Anal. Calcd for $C_{32}H_{32}O_7$: C, 72.71; H, 6.10. Found: C, 72.53; H, 6.07.

### 4.4. *tert*-Butyl [methyl 2-*O*-benzoyl-3-*O*-benzyl-4-*O*-(1-naphthyl)methyl-α-L-idopyranoside]uronate (12)

To a solution of **11** (1.7 g, 3.3 mmol) in $CH_2Cl_2$ (40 mL) pyridinium dichromate (2.5 g, 6.6 mmol), $Ac_2O$ (3.1 mL, 33.0 mmol), and *tert*-butyl alcohol (6.2 mL, 66.0 mmol) were added. The mixture was stirred for 6 h at room temperature and it was then applied on the top of a silica gel column in EtOAc, with a 10 cm layer of EtOAc on top of the gel. The chromium compounds were allowed to precipitate in the presence of EtOAc and after 30 min the product was eluted with EtOAc. After evaporating the solvent, the residue was purified by column chromatography (98:2 toluene/acetone) to give **12** as a syrup (1.3 g, 67%); $[\alpha]_D$ −42 (*c* 0.3, $CHCl_3$); $^1H$ NMR (200 MHz, $CDCl_3$): δ 7.91–7.72 (m, 5H, aromatic), 7.47–7.14 (m, 12H, aromatic), 5.14 (ddd, 1H, $J_{1,2}$ 2.8 Hz, $J_{2,3}$ 3.3 Hz, $J_{2,4}$ 0.8 Hz, H-2), 5.06 (dd, 1H, $J_{1,2}$ 2.8 Hz, $J_{1,3}$ 0.7 Hz, H-1), 4.96 (d, 1H, *J* 12.0 Hz, $ArCH_2$), 4.89 (d, 1H, *J* 12.0 Hz, $ArCH_2$), 4.78 (d, 1H, *J* 11.9 Hz $ArCH_2$), 4.71 (d, 1H, $J_{4,5}$ 3.1 Hz, H-5), 4.57 (d, 1H, *J* 11.9 Hz $ArCH_2$), 4.02 (ddd, 1H, $J_{2,4}$ 0.8 Hz, $J_{3,4}$ 3.9 Hz, $J_{4,5}$ 3.1 Hz, H-4), 3.90 (ddd, 1H, $J_{1,3}$ 0.7 Hz, $J_{2,3}$ 3.3 Hz, $J_{3,4}$ 3.9 Hz, H-3), 3.49 (s, 3H, $OCH_3$), 1.41 (s, 9H, $C(CH_3)_3$); $^{13}C$ NMR (50 MHz, $CDCl_3$): δ 168.5 (C-6), 165.6 (O*C*(O)Ph), 137.8, 133.5, 133.2, 133.0, 131.3, 129.9, 129.6, 128.47, 128.45, 128.41, 128.2, 128.1, 127.9, 126.2, 126.1, 125.7, 125.1, 123.7 (aromatic), 99.8 (C-1), 82.0 ($C(CH_3)_3$), 75.2 (C-4), 73.5 (C-3), 72.6, 70.9 ($2ArCH_2$), 68.9 (2C, C-2,5), 56.2 ($OCH_3$), 28.1 ($C(CH_3)_3$). Anal. Calcd for $C_{36}H_{38}O_8$: C, 72.22; H, 6.40. Found: C, 72.03; H, 6.41.

### 4.5. *tert*-Butyl (methyl 2-*O*-benzoyl-3-*O*-benzyl-α-L-idopyranoside)uronate (8)

To a solution of **12** (1.5 g, 2.4 mmol) in MeCN (20 mL) and water (5 mL), CAN (4.0 g, 7.2 mmol) was added. The mixture was stirred at room temperature for 3 h, neutralized with saturated aq $NaHCO_3$ (10 mL), diluted with $CH_2Cl_2$ (500 mL), washed with saturated aq $NaHCO_3$ (150 mL) and water (150 mL), dried, and concentrated. The residue was purified by column chromatography (98:2 toluene/EtOAc) to give **8** as a syrup (0.85 g, 76%); $[\alpha]_D$ −17.1 (*c* 0.3, $CHCl_3$); $^1H$ NMR (200 MHz, $CDCl_3$): δ 8.02–7.97 (m, 2H, aromatic), 7.63–7.27 (m, 8H, aromatic), 5.20 (ddd, 1H, $J_{1,2}$ 2.9 Hz, $J_{2,3}$ 3.2 Hz, $J_{2,4}$ 0.8 Hz, H-2), 4.99 (dd, 1H, $J_{1,2}$ 2.9 Hz, $J_{1,3}$ 1.0 Hz, H-1), 4.86 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.74 (d, 1H, $J_{4,5}$ 1.5 Hz, H-5), 4.66 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.02 (dddd, 1H, $J_{2,4}$ 0.8 Hz, $J_{3,4}$ 3.9 Hz, $J_{4,5}$ 1.5 Hz, $J_{4,OH}$ 11.4 Hz, H-4), 3.84 (ddd, 1H, $J_{1,3}$ 1.0 Hz, $J_{2,3}$ 3.2 Hz, $J_{3,4}$ 3.9 Hz, H-3), 3.51 (s, 3H, $OCH_3$), 2.75 (d, 1H, $J_{4,OH}$ 11.4 Hz, OH), 1.51 (s, 9H, $C(CH_3)_3$); $^{13}C$ NMR (50 MHz, $CDCl_3$): δ 168.6 (C-6), 165.1 (O*C*(O)Ph), 137.7, 133.7, 129.9, 129.2, 128.7, 128.6, 128.1, 128.0 (aromatic), 100.0 (C-1), 82.5 ($C(CH_3)_3$), 75.0 (C-3), 72.3 ($PhCH_2$), 68.3, 67.7, 67.6 (C-2,4,5), 56.4 ($OCH_3$), 28.2 ($C(CH_3)_3$). Anal. Calcd for $C_{25}H_{30}O_8$: C, 65.49; H, 6.60. Found: C, 65.33; H, 6.57.

### 4.6. Phenyl 2-azido-2-deoxy-4,6-*O*-(1-naphthyl)methylidene-1-thio-β-D-glucopyranoside (14)

To a solution of **13**[53] (3.7 g, 12.5 mmol) in dry DMF (30 mL), 1-naphthaldehyde dimethyl acetal (4.0 mL, 18.8 mmol), and (±)-camphor-10-sulfonic acid (0.3 g, 1.3 mmol) were added. The mixture was stirred at 50 °C under reduced pressure (40 kPa) for 5 h, neutralized with saturated aq $NaHCO_3$ (10 mL), and concentrated. To the residue, hexanes (100 mL) and water (100 mL) were added, and the mixture was stirred overnight. The precipitated crystalline material was filtered off and crystallized from EtOH to afford **14** as a white solid (4.1 g, 75%); mp 135–136 °C; $[\alpha]_D$ −42 (*c* 0.5, $CHCl_3$); $^1H$ NMR (200 MHz, $CDCl_3$): δ 8.22–7.32 (m, 12H, aromatic), 5.99 (s, 1H, $^1$NaphCH), 4.40 (dd, 1H, $J_{5,6a}$ 4.8 Hz, $J_{6a,6b}$ 10.6 Hz, H-6a), 4.34 (d, 1H, $J_{1,2}$ 9.9 Hz, H-1), 3.81 (dd, 1H, $J_{5,6b}$ 10.3 Hz, $J_{6a,6b}$ 10.6 Hz, H-6b), 3.65–3.30 (m, 4H, H-3, H-4, H-5, OH), 3.24 (dd, 1H, $J_{1,2}$ 9.9 Hz, $J_{2,3}$ 8.8 Hz, H-2); $^{13}C$ NMR (50 MHz, $CDCl_3$): δ 134.0, 133.8, 132.1, 131.1, 130.4, 130.3, 129.2, 128.9, 128.8, 126.5, 125.9, 125.1, 125.0, 124.2 (aromatic), 101.6 ($^1$NaphCH), 86.9 (C-1), 80.8 (C-4), 74.1 (C-3), 70.4 (C-5), 68.8 (C-6), 65.2 (C-2). Anal. Calcd for $C_{23}H_{21}N_3O_4S$: C, 63.43; H, 4.86; N, 9.65; S, 7.36. Found: C, 63.16; H, 4.81; N, 9.62; S, 7.40.

### 4.7. Phenyl 2-azido-3-*O*-benzyl-2-deoxy-4,6-*O*-(1-naphthyl)methylidene-1-thio-β-D-glucopyranoside (15)

To a stirred solution of **14** (4.1 g, 9.2 mmol) in dry DMF (40 mL), 60% NaH suspension (0.75 g, 18.4 mmol) was added at 0 °C. After 30 min, benzyl bromide (1.6 mL, 13.8 mmol) was added dropwise. Stirring was continued for 2 h, then MeOH (2 mL) was added. After 30 min, the mixture was diluted with $CH_2Cl_2$ (300 mL), washed with water (100 mL), dried, and concentrated. The residue was crystallized from EtOAc/hexanes to afford **15** as a white solid (4.2 g, 86%); mp 159–160 °C; $[\alpha]_D$ −79 (*c* 0.3, $CHCl_3$); $^1H$ NMR (200 MHz, $CDCl_3$): δ 8.12–7.23 (m, 17H, aromatic), 6.10 (s, 1H, $^1$NaphCH), 4.78 (d, 1H, *J* 11.0 Hz, $PhCH_2$), 4.66 (d, 1H, *J* 11.0 Hz, $PhCH_2$), 4.52 (d, 1H, $J_{1,2}$ 10.3 Hz, H-1), 4.50 (dd, 1H, $J_{5,6a}$ 4.8 Hz, $J_{6a,6b}$ 10.6 Hz, H-6a), 3.90, 3.78, 3.66 (3H, 3 t, H-3,4,6b), 3.55 (dd, 1H, $J_{4,5}$ 9.5 Hz, $J_{5,6a}$ 4.8 Hz, $J_{5,6b}$ 9.5 Hz, H-5), 3.39 (dd, 1H, $J_{1,2}$ 10.3 Hz, $J_{2,3}$ 8.8 Hz, H-2); $^{13}C$ NMR

(50 MHz, $CDCl_3$): δ 137.5, 134.2, 133.9, 132.3, 130.7, 130.6, 130.0, 129.3, 128.9, 128.8, 128.6, 128.5, 128.1, 126.5, 125.9, 125.2, 124.1, 124.0 (aromatic), 100.4 ($^1$NaphCH), 86.8 (C-1), 81.9, 81.1 (C-3,4), 75.4 ($PhCH_2$), 70.7 (C-5), 68.9 (C-6), 64.9 (C-2). Anal. Calcd for $C_{30}H_{27}N_3O_4S$: C, 68.55; H, 5.18; N, 7.99; S, 6.10. Found: C, 68.09; H, 5.15; N, 8.04; S, 6.05.

### 4.8. Phenyl 2-azido-3-*O*-benzyl-2-deoxy-1-thio-β-D-glucopyranoside (16)

A solution of **15** (2.3 g, 4.4 mmol) in 60% aq AcOH (40 mL) was heated under reflux for 6 h. The mixture was cooled to room temperature and concentrated. The residue was dissolved in dry MeOH (50 mL) and a catalytic amount of NaOMe was added. The mixture was stirred overnight at room temperature, neutralized with Amberlite IR120 ($H^+$) resin, filtered, and concentrated. The residue was purified by column chromatography (9:1→4:1 toluene/EtOAc) to give **16** as a syrup (1.5 g, 90%); $[\alpha]_D$ −72 (*c* 0.3, $CHCl_3$); $^1$H NMR (200 MHz, $CDCl_3$): δ 7.56–7.15 (m, 10H, aromatic), 4.92 (d, 1H, *J* 11.4 Hz, $PhCH_2$), 4.75 (d, 1H, *J* 11.4 Hz, $PhCH_2$), 4.46 (m, 1H, H-1), 3.85 (dd, 1H, $J_{5,6a}$ 3.3 Hz, $J_{6a,6b}$ 12.1 Hz, H-6a), 3.74 (dd, 1H, $J_{5,6b}$ 4.4 Hz, $J_{6a,6b}$ 12.1 Hz, H-6b), 3.54 (m, 1H, H-5), 3.38–3.24 (m, 3H, H-2,3,4), 2.90 (s, 1H, OH), 2.44 (s, 1H, OH); $^{13}$C NMR (50 MHz, $CDCl_3$): δ 137.9, 133.4, 131.4, 129.3, 129.2, 128.8, 128.6, 128.4, 128.3, 125.4 (aromatic), 86.6 (C-1), 84.8, 79.5, 75.6, 70.3 (C-3,4,5, $PhCH_2$), 65.1 (C-2), 62.5 (C-6). Anal. Calcd for $C_{19}H_{21}N_3O_4S$: C, 58.90; H, 5.46; N, 10.85; S, 8.28. Found: C, 58.48; H, 5.45; N, 10.83; S, 8.27.

### 4.9. Phenyl 2-azido-3-*O*-benzyl-2-deoxy-6-*O*-(4-methoxy)phenyl-1-thio-β-D-glucopyranoside (18)

To a solution of **16** (1.5 g, 3.8 mmol) in dry $CH_2Cl_2$ (40 mL), triethylamine (1.1 mL, 7.6 mmol), 4-dimethylaminopyridine (0.02 g, 0.2 mmol), and tosyl chloride (0.9 g, 4.6 mmol) were added. The mixture was stirred overnight at room temperature. Ice was added, after 1 h the mixture was diluted with $CH_2Cl_2$ (300 mL), washed with ice-cold 2 M aq HCl (100 mL), saturated aq $NaHCO_3$ (100 mL), and water (100 mL), dried, and concentrated to give crude **17**. The solution of the residue in dry DMF (20 mL) was added to a mixture of 4-methoxyphenol (1.2 g, 9.4 mmol) and 60% NaH suspension (0.38 g, 9.4 mmol) in dry DMF (20 mL). The mixture was stirred at 50 °C for 3 h, cooled to room temperature, diluted with $CHCl_3$ (500 mL), then was washed with water (150 mL), saturated aq $NaHCO_3$ (150 mL), and water (150 mL), dried, and concentrated. The residue was purified by column chromatography (98:2 toluene/acetone) to give **18** (1.6 g, 83%); mp 103–104 °C (from EtOAc/hexanes); $[\alpha]_D$ −40 (*c* 0.5, $CHCl_3$); $^1$H NMR (200 MHz, $CDCl_3$): δ 7.62–6.76 (m, 14H, aromatic), 4.95 (d, 1H, *J* 11 Hz, $PhCH_2$), 4.77 (d, 1H, *J* 11 Hz, $PhCH_2$), 4.47 (m, 1H, H-1), 4.23 (dd, 1H, $J_{5,6a}$ 2.9 Hz, $J_{6a,6b}$ 10.3 Hz, H-6a), 4.14 (dd, 1H, $J_{5,6b}$ 4.8 Hz, $J_{6a,6b}$ 10.3 Hz, H-6b), 3.76 (s, 3H, $OCH_3$), 3.70–3.53 (m, 2H, H-4,5), 3.42–3.29 (m, 2H, H-2,3), 2.50 (d, 1H, $J_{2,OH}$ 2.9 Hz, OH); $^{13}$C NMR (50 MHz, $CDCl_3$): δ 154.3, 152.8, 137.9, 133.6, 131.4, 129.1, 128.9, 128.5, 128.4, 128.3, 115.9, 114.8 (aromatic), 86.5 (C-1), 84.9, 78.0, 75.6, 70.7 (C-3,4,5, $PhCH_2$), 68.4 (C-6), 64.8 (C-2), 55.9 ($OCH_3$). Anal. Calcd for $C_{26}H_{27}N_3O_5S$: C, 63.27; H, 5.51; N, 8.51; S, 6.50. Found: C, 63.03; H, 5.48; N, 8.54; S, 6.53.

### 4.10. Phenyl 2-azido-3-*O*-benzyl-4-*O*-chloroacetyl-2-deoxy-6-*O*-(4-methoxy)phenyl-1-thio-β-D-glucopyranoside (7)

A solution of 90% chloroacetic anhydride (0.90 g, 0.75 mmol) in $CH_2Cl_2$ (12 mL) was added dropwise to a stirred solution of **18** (1.6 g, 3.2 mmol) in dry $CH_2Cl_2$ (46 mL) and dry pyridine (12 mL) at −20 °C. After 20 min, the reaction was quenched with water (10 mL) and stirred for 20 min. The mixture was diluted with $CH_2Cl_2$ (200 mL), washed with 2 M aq HCl (75 mL), saturated aq $NaHCO_3$ (75 mL), and water (75 mL), dried, and concentrated. The residue was purified by column chromatography (95:5 toluene/acetone) to give **7** (1.5 g, 81%); mp 75–76 °C (from EtOAc/hexanes); $[\alpha]_D$ −35 (*c* 0.3, $CHCl_3$); $^1$H NMR (200 MHz, $CDCl_3$): δ 7.60–6.80 (m, 14H, aromatic), 5.09 (dd, 1H, $J_{3,4}$ 9.1 Hz, $J_{4,5}$ 9.5 Hz, H-4), 4.87 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.64 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.49 (d, 1H, $J_{1,2}$ 9.9 Hz, H-1), 4.07–3.93 (m, 2H, H-6a, H-6b), 3.76 (m, 1H, H-5), 3.75 (s, 1H, $OCH_3$), 3.72 (d, 1H, *J* 7.3 Hz, $C(O)CH_2Cl$), 3.57 (dd, 1H, $J_{2,3}$ 9.9 Hz, $J_{3,4}$ 9.1 Hz, H-3), 3.43 (t, 1H, $J_{1,2}$ 9.9 Hz, $J_{2,3}$ 9.9 Hz, H-2); $^{13}$C NMR (50 MHz, $CDCl_3$): δ 166.2 ($C(O)CH_2Cl$), 154.5, 152.6, 137.6, 133.9, 130.8, 129.2, 128.8, 128.7, 128.2, 115.9, 114.9 (aromatic), 86.4 (C-1), 82.5, 76.5, 75.6, 72.3 (C-3,4,5, $PhCH_2$), 68.2 (C-6), 65.1 (C-2), 55.8 ($OCH_3$), 40.5 ($C(O)CH_2Cl$). Anal. Calcd for $C_{28}H_{28}ClN_3O_6S$: C, 58.99; H, 4.95; N, 7.37; S, 5.62. Found: C, 58.64; H, 4.92; N, 7.35; S, 5.65.

### 4.11. Methyl [2-azido-3-*O*-benzyl-4-*O*-chloroacetyl-2-deoxy-6-*O*-(4-methoxy)phenyl-α-D-glucopyranosyl]-(1→4)-(*tert*-butyl 2-*O*-benzoyl-3-*O*-benzyl-α-L-idopyranosideuronate) (19)

A mixture of **8** (0.72 g, 1.56 mmol), **7** (1.07 g, 1.87 mmol), and 4 Å molecular sieves (3 g) was stirred in dry $CH_2Cl_2$ (5 mL) and dry $Et_2O$ (15 mL) at 0 °C under argon for 30 min, then DMTST (2.00 g, 7.80 mmol) was added. After 1 day, the reaction was quenched with triethylamine (3 mL). The mixture was filtered through a pad of Celite, the filtrate was diluted with $CH_2Cl_2$ (500 mL), and washed with 2 M aq HCl (150 mL), saturated aq $NaHCO_3$ (150 mL), and water (150 mL), dried, and concentrated. The residue was purified by column chromatography (98:2→9:1 toluene/EtOAc) to give **19** as a syrup (0.67 g, 47%); $[\alpha]_D$ +7 (*c* 0.3, $CHCl_3$); $^1$H NMR (400 MHz, $CDCl_3$): δ 8.16–8.09 (m, 2H, aromatic), 7.60–7.12 (m, 13H, aromatic), 6.82–6.78 (m, 4H, aromatic), 5.24 (dd, 1H, $J_{3',4'}$ 9.2 Hz, $J_{4',5'}$ 10.2 Hz, H-4′), 5.15 (dd, 1H, $J_{1,2}$ 3.0 Hz, $J_{2,3}$ 3.2 Hz, H-2), 5.14 (dd, 1H, $J_{1,2}$ 3.0 Hz, $J_{1,3}$ 1.7 Hz, H-1), 4.98 (d, 1H, $J_{1',2'}$ 3.6 Hz, H-1′), 4.87 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.80 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.68 (d, 1H, $J_{4,5}$ 3.3 Hz, H-5), 4.45 (d, 1H, *J* 11.3 Hz, $PhCH_2$), 4.29 (ddd, 1H, $J_{4',5'}$ 10.2 Hz, $J_{5',6a'}$ 4.2 Hz, $J_{5',6b'}$ 3.4 Hz, H-5′), 4.24 (ddd, 1H, $J_{1,3}$ 1.7 Hz, $J_{2,3}$ 3.2 Hz, $J_{3,4}$ 4.8 Hz, H-3), 4.23 (d, 1H, *J* 11.3 Hz, $PhCH_2$), 4.12 (dd, 1H, $J_{3,4}$ 4.8 Hz, $J_{4,5}$ 3.3 Hz, H-4), 4.04 (dd, 1H, $J_{5',6a'}$ 4.2 Hz, $J_{6a',6b'}$ 10.5 Hz, H-6a′), 3.90 (dd, 1H, $J_{5',6b'}$ 3.4 Hz, $J_{6a',6b'}$ 10.5 Hz, H-6b′), 3.77 (dd, 1H, $J_{2',3'}$ 10.0 Hz, $J_{3',4'}$ 9.2 Hz, H-3′), 3.77 (d, 1H, *J* 14.3 Hz, $C(O)CH_2Cl$), 3.75 (s, 3H, Ar–$OCH_3$), 3.72 (d, 1H, *J* 14.3 Hz, $C(O)CH_2Cl$), 3.50 (s, 3H, $OCH_3$), 3.35 (dd, 1H, $J_{1',2'}$ 3.6 Hz, $J_{2',3'}$ 10.0 Hz, H-2′), 1.52 (s, 9H, $C(CH_3)_3$); $^{13}$C NMR (100 MHz, $CDCl_3$): δ 168.2 (C-6), 165.7 ($OC(O)CH_2Cl$), 165.5 ($OC(O)Ph$), 154.3, 152.6, 137.6, 137.5, 133.4, 130.0, 129.8, 129.7, 128.6, 128.5, 128.4, 128.1, 127.9, 127.7, 115.9, 114.7 (aromatic), 100.3 (C-1), 98.3 (C-1′), 82.8 ($C(CH_3)_3$), 77.9 (C-3′), 74.7 ($PhCH_2$), 74.4 (C-4), 74.1 (C-3), 73.3 ($PhCH_2$), 72.5 (C-4′), 70.1 (C-2), 69.1 (C-5), 68.8 (C-5′), 67.4 (C-6′), 63.3 (C-2′), 56.2 ($OCH_3$), 55.7 ($ArOCH_3$), 40.4 ($C(O)CH_2Cl$), 28.3 ($C(CH_3)_3$). Anal. Calcd for $C_{47}H_{52}ClN_3O_{14}$: C, 61.47; H, 5.71; N, 4.58. Found: C, 61.03; H, 5.66; N, 4.49.

### 4.12. Methyl [2-azido-3-*O*-benzyl-2-deoxy-6-*O*-(4-methoxy)-phenyl-α-D-glucopyranosyl]-(1→4)-(*tert*-butyl 2-*O*-benzoyl-3-*O*-benzyl-α-L-idopyranosideuronate) (20)

To a solution of **19** (0.67 g, 0.74 mmol) in dry DMF (7 mL), a solution of hydrazinedithiocarbonate[54] (2.21 mmol) in 5.3 mL of ethanol/water/dioxane (4:2:1) was added. The mixture was stirred at room temperature for 1 h, diluted with $CH_2Cl_2$ (250 mL), washed with 2 M aq HCl (50 mL), saturated aq $NaHCO_3$ (50 mL), and water (50 mL), dried, and concentrated. The residue was purified by column chromatography (9:1 toluene/EtOAc) to give **20** as a syrup (0.49 g, 80%); $[\alpha]_D$ +2 (*c* 0.9, $CHCl_3$); $^1$H NMR (200 MHz, $CDCl_3$): δ 8.15–8.10 (m, 2H, aromatic), 7.58–7.10 (m, 13H, aromatic), 6.89–6.77 (m, 4H, aromatic), 5.15 (dd, 1H, $J_{1,2}$ 2.7 Hz, $J_{2,3}$ 3.2 Hz, H-2), 5.14 (d, 1H, $J_{1,2}$ 2.7 Hz, H-1), 4.96 (d, 1H, $J_{1',2'}$ 3.3 Hz, H-1′), 4.89 (d, 1H, *J*

11.7 Hz, $PhCH_2$), 4.78 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.65 (d, 1H, $J_{4,5}$ 3.3 Hz, H-5), 4.46 (d, 1H, *J* 11.0 Hz, $PhCH_2$), 4.34 (d, 1H, *J* 11.0 Hz, $PhCH_2$), 4.25–4.08 (m, 5H, H-3,4,3′,5′,6a′), 3.83 (dd, 1H, $J_{5',6b'}$ 3.3 Hz, $J_{6a',6b'}$ 8.8 Hz, H-6b′), 3.75 (s, 3H, $ArOCH_3$), 3.62 (ddd, 1H, $J_{3',4'}$ 9.9 Hz, $J_{4',5'}$ 9.9 Hz, $J_{4',OH}$ 3.3 Hz, H-4′), 3.49 (s, 3H, $OCH_3$), 3.20 (dd, 1H, $J_{1',2'}$ 3.3 Hz, $J_{2',3'}$ 9.9 Hz, H-2′), 2.41 (d, 1H, $J_{4',OH}$ 3.3 Hz, OH), 1.49 (s, 9H, $C(CH_3)_3$); $^{13}C$ NMR (50 MHz, $CDCl_3$): δ 168.2 (C-6), 165.6 (O*C*(O)Ph), 154.2, 152.9, 138.1, 137.7, 133.4, 130.1, 129.8, 129.1, 128.7, 128.6, 128.5, 128.3, 128.1, 128.0, 127.9, 125.4, 115.8, 114.7 (aromatic), 100.3 (C-1), 98.9 (C-1′), 82.8 (*C*$(CH_3)_3$), 80.0 (C-3′), 74.8, 74.52, 74.47 (C-3,4, $PhCH_2$), 73.3 ($PhCH_2$), 71.3, 70.6, 70.1, 69.2 (C-2,5,4′,5′), 67.8 (C-6′), 63.2 (C-2′), 56.3 ($OCH_3$), 55.8 ($ArOCH_3$), 28.3 (C(*C*$H_3)_3$). Anal. Calcd for $C_{45}H_{51}N_3O_{13}$: C, 64.20; H, 6.11; N, 4.99. Found: C, 63.81; H, 6.03; N, 4.86.

### 4.13. Methyl (*tert*-butyl 2-*O*-benzoyl-3,4-di-*O*-benzyl-α-L-idopyranosyluronate)-(1→4)-[2-azido-3-*O*-benzyl-2-deoxy-6-*O*-(4-methoxy)phenyl-α-D-glucopyranosyl]-(1→4)-(*tert*-butyl 2-*O*-benzoyl-3-*O*-benzyl-α-L-idopyranosideuronate) (5)

A mixture of **6** (0.45 g, 0.72 mmol), **20** (0.40 g, 0.48 mmol), and 4 Å molecular sieves (1.0 g) was stirred in dry $CH_2Cl_2$ (10 mL) at 0 °C under argon for 30 min, then DMTST (0.62 g, 2.40 mmol) was added. After 3 days, the reaction was quenched with triethylamine (1 mL). The mixture was filtered through a pad of Celite, the filtrate was diluted with $CH_2Cl_2$ (250 mL) and washed with 2 M aq HCl (50 mL), saturated aq $NaHCO_3$ (50 mL), and water (50 mL), dried, and concentrated. The residue was purified by column chromatography (95:5→9:1 toluene/EtOAc) to give **5** as a syrup (0.40 g, 61%); $[\alpha]_D$ +3 (*c* 0.3, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$): δ 8.14–8.05 (m, 2H, aromatic), 7.86–7.78 (m, 2H, aromatic), 7.59–7.42 (m, 4H, aromatic), 7.37–7.12 (m, 22H, aromatic), 6.91–6.82 (m, 2H, aromatic), 6.77–6.68 (m, 2H, aromatic), 5.55 (d, 1H, $J_{1'',2''}$ 5.6 Hz, H-1″), 5.14 (ddd, 1H, $J_{1'',2''}$ 5.6 Hz, $J_{2'',3''}$ 4.4 Hz, $J_{2'',4''}$ 0.7 Hz, H-2″), 5.12 (ddd, 1H, $J_{1,2}$ 3.3 Hz, $J_{2,3}$ 4.2 Hz, $J_{2,4}$ 0.5 Hz, H-2), 5.09 (d, 1H, $J_{1,2}$ 3.3 Hz, H-1), 4.90 (d, 1H, $J_{1',2'}$ 3.7 Hz, H-1′), 4.84 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.77 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.74 (d, 1H, *J* 11.1 Hz, $PhCH_2$), 4.73 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.62 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.57 (d, 1H, $J_{4,5}$ 3.6 Hz, H-5), 4.50 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.48 (d, 1H, $J_{4'',5''}$ 5.1 Hz, H-5″), 4.42 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.22 (dd, 1H, $J_{3',4'}$ 9.0 Hz, $J_{4',5'}$ 9.8 Hz H-4′), 4.18 (dd, 1H, $J_{2,3}$ 4.2 Hz, $J_{3,4}$ 4.8 Hz, H-3), 4.12 (d, 1H, *J* 11.1 Hz, $PhCH_2$), 4.1–4.05 (m, 3H, H-5′,6a′,6b′), 4.03 (ddd, 1H, $J_{2,4}$ 0.5 Hz, $J_{3,4}$ 4.8 Hz, $J_{4,5}$ 3.6 Hz, H-4), 3.88 (ddd, 1H, $J_{2'',4''}$ 0.7 Hz, $J_{3'',4''}$ 5.6 Hz, $J_{4'',5''}$ 5.1 Hz, H-4″), 3.82 (dd, 1H, $J_{2'',3''}$ 4.4 Hz, $J_{3'',4''}$ 5.6 Hz, H-3″), 3.73 (s, 3H, Ar–$OCH_3$), 3.64 (dd, 1H, $J_{2',3'}$ 10.2 Hz, $J_{3',4'}$ 9.0 Hz, H-3′), 3.45 (s, 3H, $OCH_3$), 3.30 (dd, 1H, $J_{1',2'}$ 3.7 Hz, $J_{2',3'}$ 10.2 Hz, H-2′), 1.41 (s, 9H, $C(CH_3)_3$), 1.39 (s, 9H, $C(CH_3)_3$); $^{13}C$ NMR (100 MHz, $CDCl_3$): δ 168.6 (C-6″), 168.2 (C-6), 165.5 (O*C*(O)Ph), 165.3 (O*C*(O)Ph), 154.0, 152.7, 138.4, 137.7, 137.4, 133.4, 133.2, 129.99, 129.96, 129.6, 129.5, 128.7, 128.4, 128.3, 128.1, 128.04, 128.00, 127.9, 127.82, 127.77, 127.75, 127.7, 127.3, 116.3, 114.4 (aromatic), 100.1 (C-1), 98.8 (C-1′), 98.0 (C-1″), 82.7 (*C*$(CH_3)_3$), 81.8 (*C*$(CH_3)_3$), 78.4 (C-3′), 76.1 (C-4″), 76.0 (C-4′), 75.8 (C-3″), 74.8 (C-4), 74.7 ($PhCH_2$), 74.5 (C-3), 73.3 ($PhCH_2$), 73.2 ($PhCH_2$), 73.1 ($PhCH_2$), 72.3 (C-2″), 72.0 (C-5″), 70.8 (C-5′), 69.9 (C-2), 69.2 (C-5), 66.4 (C-6′), 63.4 (C-2′), 56.2 ($OCH_3$), 55.7 ($ArOCH_3$), 28.2 (C(*C*$H_3)_3$), 28.1 (C(*C*$H_3)_3$). Anal. Calcd for $C_{76}H_{83}N_3O_{20}$: C, 67.19; H, 6.16; N, 3.09. Found: C, 66.98; H, 6.19; N 3.02.

### 4.14. Methyl (*tert*-butyl 3,4-di-*O*-benzyl-α-L-idopyranosyluronate)-(1→4)-[2-azido-3-*O*-benzyl-2-deoxy-6-*O*-(4-methoxy)phenyl-α-D-glucopyranosyl]-(1→4)-(*tert*-butyl 3-*O*-benzyl-α-L-idopyranosideuronate) (21)

To a solution of **5** (0.39 g, 0.29 mmol) in dry MeOH (5 mL), a catalytic amount of NaOMe was added. The mixture was stirred for one week at room temperature, neutralized with Amberlite IR120 ($H^+$) resin, filtered, and concentrated to give **21** as a syrup (0.31 g, 93%); $[\alpha]_D$ −32 (*c* 0.4, $CHCl_3$); $^1H$ NMR (200 MHz, $CDCl_3$): δ 7.40–7.14 (m, 22H, aromatic), 6.89–6.75 (m, 2H, aromatic), 5.26 (d, 1H, $J_{1'',2''}$ 2.6 Hz, H-1″), 5.06 (d, 1H, $J_{1',2'}$ 3.7 Hz, H-1′), 5.00 (d, 1H, *J* 10.3 Hz, $PhCH_2$), 4.94 (s, 1H, H-1), 4.74 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.73 (d, 1H, *J* 12.0 Hz, $PhCH_2$), 4.62 (d, 1H, $J_{4,5}$ 2.9 Hz, H-5), 4.60 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.59 (d, 1H, *J* 10.6 Hz, $PhCH_2$), 4.57 (d, 1H, *J* 12.0 Hz, $PhCH_2$), 4.57 (d, 1H, $J_{4'',5''}$ 1.8 Hz, H-5″), 4.53 (d, 1H, *J* 12.4 Hz, $PhCH_2$), 4.47 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.25–4.16 (m, 2H, H-3,4′), 4.15–4.06 (m, 3H, H-5′,6a′,6b′), 4.01 (t, 1H, $J_{3'',4''}$ 1.8 Hz, $J_{4'',5''}$ 1.8 Hz, H-4″), 3.91 (t, 1H, $J_{3,4}$ 2.9 Hz, $J_{4,5}$ 2.9 Hz, H-4), 3.80 (m, 1H, H-3″), 3.75 (s, 3H, $ArOCH_3$), 3.73–3.68 (m, 2H, H-2,3′), 3.61 (dd, 1H, $J_{1',2'}$ 3.7 Hz, $J_{2',3'}$ 10.3 Hz, H-2′), 3.51 (m, 1H, H-2″), 3.48 (s, 3H, $OCH_3$), 1.53 (s, 9H, $C(CH_3)_3$), 1.32 (s, 9H, $C(CH_3)_3$); $^{13}C$ NMR (50 MHz, $CDCl_3$): δ 169.0, 168.6 (C-6, C-6″), 154.2, 152.7, 138.0, 137.70, 137.66, 137.0, 129.1, 128.7, 128.62, 128.57, 128.3, 128.22, 128.18, 128.14, 128.09, 128.0, 127.6, 125.4, 116.1, 114.7 (aromatic), 102.9, 100.9, 95.3 (C-1,1′,1″), 82.6 (*C*$(CH_3)_3$), 82.0 (*C*$(CH_3)_3$), 80.0 (C-3′), 75.8, 75.5, 74.71, 74.70, 73.4, 73.0, 72.8, 72.5, 71.4, 70.8, 69.3, 68.6, 68.0, 67.7, 66.6 (C-6′), 64.0 (C-2′), 56.2, 55.8 ($OCH_3$, $ArOCH_3$), 28.3 (C(*C*$H_3)_3$), 28.1 (C(*C*$H_3)_3$). Anal. Calcd for $C_{62}H_{75}N_3O_{18}$: C, 64.74; H, 6.57; N, 3.65. Found: C, 64.38; H, 6.52; N, 3.59.

### 4.15. Methyl (*tert*-butyl 3,4-di-*O*-benzyl-α-L-idopyranosyluronate)-(1→4)-(2-azido-3-*O*-benzyl-2-deoxy-α-D-glucopyranosyl)-(1→4)-(*tert*-butyl 3-*O*-benzyl-α-L-idopyranoside-uronate) (22)

To a solution of **21** (0.27 g, 0.24 mmol) in MeCN (4.5 mL) and water (0.5 mL), ceric ammonium nitrate (0.39 g, 0.72 mmol) was added. The mixture was stirred at room temperature for 1 h, neutralized with saturated aq $NaHCO_3$ (2 mL), diluted with $CHCl_3$ (300 mL), washed with saturated aq $NaHCO_3$ (50 mL) and water (50 mL), dried, and concentrated. The residue was purified by column chromatography (9:1→4:1 toluene/acetone) to give **22** (0.21 g, 84%) as a syrup; $[\alpha]_D$ −40 (*c* 0.3, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$): δ 7.41–7.15 (m, 20H, aromatic), 5.30 (d, 1H, $J_{1'',2''}$ 3.5 Hz, H-1″), 5.00 (d, 1H, $J_{1',2'}$ 3.7 Hz, H-1′), 4.94 (d, 1H, *J* 11.6 Hz, $PhCH_2$), 4.92 (d, 1H, $J_{1,2}$ 1.8 Hz, H-1), 4.75 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.67 (d, 1H, $J_{4,5}$ 2.4 Hz, H-5), 4.65 (s, 1H, $PhCH_2$), 4.63 (s, 1H, $PhCH_2$), 4.62 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.60 (d, 1H, $J_{4'',5''}$ 2.5 Hz, H-5″), 4.58 (d, 1H, *J* 11.6 Hz, $PhCH_2$), 4.57 (d, 1H, *J* 11.6 Hz, $PhCH_2$), 4.54 (d, 1H, *J* 11.6 Hz, $PhCH_2$), 4.14 (dd, 1H, $J_{3,4}$ 3.8 Hz, $J_{4,5}$ 2.4 Hz, H-4), 3.90 (dd, 1H, $J_{2,3}$ 3.2 Hz, $J_{3,4}$ 3.8 Hz, H-3), 3.89 (dd, 1H, $J_{3',4'}$ 8.8 Hz, $J_{4',5'}$ 9.8 Hz, H-4′), 3.84 (m, 2H, H-6a′,6b′), 3.81 (dd, 1H, $J_{3'',4''}$ 5.0 Hz, $J_{4'',5''}$ 2.5 Hz, H-4″), 3.80 (dd, 1H, $J_{2'',3''}$ 5.4 Hz, $J_{3'',4''}$ 5.0 Hz, H-3″), 3.78 (m, 1H, H-5′), 3.72 (dd, 1H, $J_{2',3'}$ 10.0 Hz, $J_{3',4'}$ 8.8 Hz, H-3′), 3.71 (ddd, 1H, $J_{1,2}$ 1.8 Hz, $J_{2,3}$ 3.2 Hz, $J_{2,OH}$ 10.8 Hz, H-2), 3.58 (ddd, 1H, $J_{1'',2''}$ 3.5 Hz, $J_{2'',3''}$ 5.4 Hz, $J_{2'',OH}$ 7.0 Hz, H-2″), 3.51 (dd, 1H, $J_{1',2'}$ 3.7 Hz, $J_{2',3'}$ 10.0 Hz, H-2′), 3.47 (s, 3H, $OCH_3$), 3.42 (d, 1H, $J_{2,OH}$ 10.8 Hz, C-2-OH), 3.08 (d, 1H, $J_{2'',OH}$ 7.0 Hz, C-2″-OH), 2.20 (br s, 1H, C-6′-OH), 1.52 (s, 9H, $C(CH_3)_3$), 1.34 (s, 9H, $C(CH_3)_3$); $^{13}C$ NMR (100 MHz, $CDCl_3$): δ 169.1 (C-6), 168.8 (C-6″), 138.2, 137.81, 137.77, 137.2, 128.83, 128.76, 128.7, 128.44, 128.40, 128.37, 128.33, 128.25, 128.2, 128.1, 127.7 (aromatic), 103.1 (C-1), 100.8 (C-1″), 95.5 (C-1′), 83.0 (*C*$(CH_3)_3$), 82.4 (*C*$(CH_3)_3$), 79.8 (C-3′), 76.3 (C-4″), 75.6 ($PhCH_2$), 75.3 (C-3″), 75.0 (C-4′), 73.5 ($PhCH_2$), 73.4 ($PhCH_2$), 73.1 (C-3), 72.7 ($PhCH_2$), 72.4 (C-5′), 71.7 (C-4), 69.9 (C-5″), 69.4 (C-2″), 68.0 (C-5), 67.8 (C-2), 64.2 (C-2′), 61.6 (C-6′), 56.3 ($OCH_3$), 28.4 (C(*C*$H_3)_3$), 28.2 (C(*C*$H_3)_3$). Anal. Calcd for $C_{55}H_{69}N_3O_{17}$: C, 63.27; H, 6.66; N, 4.02. Found: C, 62.91; H, 6.62; N, 3.96.

### 4.16. Methyl (3,4-di-*O*-benzyl-α-L-idopyranosyluronic acid)-(1→4)-(2-azido-3-*O*-benzyl-2-deoxy-α-D-glucopyranosyl)-(1→4)-(3-*O*-benzyl-α-L-idopyranosideuronic acid) (23)

A solution of compound **22** (0.19 g, 0.19 mmol) in a 20% solution of $CF_3CO_2H$ in $CH_2Cl_2$ (6 mL) was stirred for 1 h, the mixture was concentrated, and the residue was co-evaporated with toluene

(50 mL) to give **23** (0.17 g, 98%) as a syrup; $[\alpha]_D$ −30 (*c* 0.3, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$): 7.40–7.10 (m, 20H, aromatic), 5.22 (d, 1H, $J_{1'',2''}$ 2.0 Hz, H-1″), 4.94 (s, 1H, H-1), 4.83 (br s, 1H, H-5), 4.77 (d, 1H, $J_{1',2'}$ 4.1 Hz, H-1′), 4.71 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.70 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.65 (br s, 1H, H-5″), 4.62 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.56 (d, 1H, *J* 11.6 Hz, $PhCH_2$), 4.55 (d, 1H, *J* 11.6 Hz, $PhCH_2$), 4.53 (d, 1H, *J* 11.5 Hz, $PhCH_2$), 4.51 (s, 2H, $PhCH_2$), 4.14 (s, 1H, H-4), 3.89–3.67 (m, 10H), 3.49 (dd, 1H, $J_{1',2'}$ 4.1 Hz, $J_{2',3'}$ 10.1 Hz, H-2′); $^{13}C$ NMR (100 MHz, $CDCl_3$): δ 171.4, 171.1 (C-6,6″), 137.44, 137.37, 137.2, 136.5, 128.6, 128.41, 128.37, 128.3, 128.19, 128.15, 127.9, 127.67, 127.65 (aromatic), 103.2 (C-1), 99.9 (C-1″), 95.7 (C-1′), 78.9, 75.41, 75.40 ($PhCH_2$), 75.2, 74.6, 73.9 ($PhCH_2$), 73.0 ($PhCH_2$), 72.9, 72.2, 71.8, 71.9 ($PhCH_2$), 68.9, 68.8, 66.2, 66.0, 64.0, 61.4 (C-6′), 56.3 ($OCH_3$).

### 4.17. Methyl (2-*O*-sulfonato-α-L-idopyranosyluronic acid)-(1→4)-(2-deoxy-2-sulfonatamido-6-*O*-sulfonato-α-D-glucopyranosyl)-(1→4)-(2-*O*-sulfonato-α-L-idopyranosideuronic acid) hexasodium salt (3)

To a solution of **23** (0.15 g, 0.16 mmol) in dry DMF (3.0 mL), sulfur trioxide pyridine complex (0.15 g, 0.96 mmol) was added. The mixture was stirred at room temperature for 3 h, neutralized with saturated aq $NaHCO_3$ (1 mL), and concentrated. The residue was purified by column chromatography (4:1→2:1 $CH_2Cl_2$/MeOH), and the product was eluted from a column of Dovex-50W X8 resin using MeOH to give **24** (0.14 g, 68%) as a foam; $^1H$ NMR (200 MHz, $CD_3OD$): δ 7.41–7.08 (m, 20H, aromatic), 5.84, 5.33 (2s, 2H, H-1,1″), 5.10–3.70 (m, 23H), 3.53 (dd, 1H, $J_{1',2'}$ 3.7 Hz, $J_{2',3'}$ 10.1 Hz, H-2′), 3.39 (s, 3H, $OCH_3$); $^{13}C$ NMR (50 MHz, $CD_3OD$): δ 177.0, 176.9 (C-6,6″), 140.0, 139.0, 138.9, 138.5, 130.1, 129.4, 129.20, 129.16, 129.0, 128.9, 128.8, 128.6, 128.4 (aromatic), 101.6, 96.0, 94.2 (C-1,1′,1″), 77.0, 75.9, 75.0, 73.8, 72.8, 72.6, 71.8, 70.97, 70.95, 70.9, 70.2, 70.1, 69.3, 68.3, 66.1, 56.4 ($OCH_3$); ESIMS (−): *m*/*z* 1235.6 $[M-2Na+H]^-$.

A solution of **24** (0.14 g, 0.11 mmol) in THF (2.5 mL) and water (2.5 mL) was hydrogenated with 10% Pd/C (0.05 g) under atmospheric pressure at room temperature. After 2 days, the mixture was filtered through a pad of Celite and concentrated to give **25**. No aromatic signal was visible in the NMR spectra.

The pH of a solution of **25** in water (3 mL) was adjusted to 9.8 with 1 M aq NaOH, then sulfur trioxide trimethylamine complex (0.092 g, 0.66 mmol) was added in six portions hourly, the pH being maintained at 9.8 by the addition of 0.5 M aq NaOH. After 6 h, the mixture was neutralized with 1 M aq HCl and concentrated. The residue was purified by column chromatography (2:2:1 EtOAc/MeOH/$H_2O$) to give trisaccharide fractions, which were eluted from a column of Dovex-50W X8 resin using $H_2O$, and desalted on a column of Sephadex G-25 using water as eluent to afford **3** (0.049 g, 45%) as a foam; $^1H$ NMR (400 MHz, $D_2O$): δ 5.23 (d, 1H, $J_{1',2'}$ 3.3 Hz, H-1′), 5.19 (s, 1H, H-1″), 5.00 (s, 1H, H-1), 4.88 (s, 1H, H-5″), 4.50 (d, 1H, $J_{4,5}$ 2.5 Hz, H-5), 4.25–4.14 (m, 5H, H-2,3,6a′,6b′,2″), 4.04 (t, 1H, $J_{2'',3''}$ 3.2 Hz, $J_{3'',4''}$ 3.2 Hz, H-3″), 3.98 (t, 1H, $J_{3,4}$ 2.5 Hz, $J_{4,5}$ 2.5 Hz, H-4), 3.96–3.89 (m, 2H, H-5′,4″), 3.74–3.64 (m, 2H, H-3′,4′), 3.34 (s, 3H, $OCH_3$), 3.19 (dd, 1H, $J_{1',2'}$ 3.3 Hz, $J_{2',3'}$ 9.8 Hz, H-2′); $^{13}C$ NMR (100 MHz, $D_2O$): δ 175.8 (C-6″), 174.4 (C-6), 99.9 (C-1), 99.4 (C-1″), 97.9 (C-1′), 76.8 (C-4′), 76.3 (C-4), 74.8 (C-2), 74.3 (C-2″), 69.8 (C-3′), 69.4 (C-5′), 69.2 (C-4″), 69.2 (C-5″), 68.9 (C-3″), 67.7 (C-3), 67.5 (C-5), 66.5 (C-6′), 58.2 (C-2′), 55.8 ($OCH_3$); HRMS (ESI) calcd for $C_{19}H_{25}NO_{29}S_4Na_6$ $[M-6Na+4H]^{2-}$ 431.4860. Found: 431.4871.

### 4.18. Methyl (3,4-di-*O*-benzyl-α-L-idopyranosyluronic acid)-(1→4)-[2-azido-3-*O*-benzyl-2-deoxy-6-*O*-(4-methoxy)-phenyl-α-D-glucopyranosyl]-(1→4)-(3-*O*-benzyl-α-L-idopyranosideuronic acid) (26)

Compound **21** (0.22 g, 0.19 mmol) was hydrolyzed as described for compound **23** to give **26** (0.18 g, 92%) as a syrup; $[\alpha]_D$ −29 (*c* 0.2, $CHCl_3$); $^1H$ NMR (200 MHz, $CDCl_3$): δ 7.39–7.11 (m, 20H, aromatic), 6.89–6.72 (m, 4H, aromatic), 5.16, 4.96 (2s, 2H, H-1,1″), 4.91 (d, 1H, $J_{1',2'}$ 3.7 Hz, H-1′), 4.87 (d, 1H, $J_{4,5}$ 1.1 Hz, H-5), 4.71 (d, 2H, *J* 11.7 Hz, $PhCH_2$), 4.71 (d, 1H, $J_{4'',5''}$ 2.6 Hz, H-5″), 4.65 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.59 (d, 1H, *J* 11.0 Hz, $PhCH_2$), 4.55 (d, 1H, *J* 12.1 Hz, $PhCH_2$), 4.54 (d, 1H, *J* 11.4 Hz, $PhCH_2$), 4.45 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.44 (d, 1H, *J* 11.4 Hz, $PhCH_2$), 4.23–4.03 (m, 4H), 3.96–3.71 (m, 5H), 3.68 (s, 3H, $ArOCH_3$), 3.68–3.63 (m, 2H), 3.59 (dd, 1H, $J_{1',2'}$ 3.7 Hz, $J_{2',3'}$ 9.9 Hz, H-2′), 3.43 (s, 3H, $OCH_3$); $^{13}C$ NMR ($CDCl_3$): δ 171.7, 170.9 (C-6,6″), 154.4, 152.5, 138.0, 137.5, 137.4, 137.1, 136.4, 129.1, 128.7, 128.5, 128.4, 128.3, 128.2, 127.9, 127.8, 127.5, 125.4, 116.1, 114.8 (aromatic), 103.4 (C-1), 100.6 (C-1″), 94.9 (C-1), 79.1 (C-3′), 75.3, 75.2, 74.9, 73.9, 73.7, 72.6, 72.1, 71.3, 71.1 (2C), 68.5, 67.8, 66.7, 66.5, 66.0 (C-6′), 64.1 (C-2′), 56.5, 55.7 ($ArOCH_3$, $OCH_3$).

### 4.19. Methyl (2-*O*-sulfonato-α-L-idopyranosyluronic acid)-(1→4)-(2-deoxy-2-sulfonatamido-α-D-glucopyranosyl)-(1→4)-(2-*O*-sulfonato-α-L-idopyranosideuronic acid) pentasodium salt (4)

Compound **26** (0.18 g, 0.17 mmol) was *O*-sulfated as described for compound **24**. The product was purified by column chromatography (12:2:1 EtOAc/MeOH/$H_2O$), then it was eluted from a column of Dovex-50W X8 resin using MeOH to give **27** (0.22 g, 99%) as a foam; $^1H$ NMR (200 MHz, $CD_3OD$): δ 7.42–7.07 (m, 20H, aromatic), 6.91–6.75 (m, 4H, aromatic), 5.90, 5.28, 5.07 (3s, 3H, H-1,1′,1″), 5.10 (d, 1H, *J* 11.0 Hz, $PhCH_2$), 5.00 (d, 1H, *J* 11.0 Hz, $PhCH_2$), 4.80 (s, 1H), 4.79 (d, 1H, *J* 11.7 Hz, $PhCH_2$), 4.77 (s, 1H), 4.70–4.18 (m, 11H), 4.11 (s, 1H), 4.03 (s, 2H), 3.88 (s, 2H), 3.70 (s, 3H, $ArOCH_3$), 3.64 (dd, 1H, $J_{1',2'}$ 2.9 Hz, $J_{2',3'}$ 9.2 Hz, H-2′), 3.39 (s, 3H, $OCH_3$); $^{13}C$ NMR (50 MHz, $CD_3OD$): δ 175.9, 175.2 (C-6,6″), 153.8, 153.0, 138.1, 137.03, 136.96, 136.1, 128.5, 128.1, 128.0, 127.9, 127.6, 127.4, 127.3, 127.1, 115.6, 114.5 (aromatic), 99.8, 94.0, 93.0 (C-1,1′,1″), 77.2, 75.8, 75.5, 73.8, 72.4, 71.7 (2C), 71.0 (2C), 70.3, 69.8, 69.4, 69.4, 68.9, 67.7, 66.9, 64.2, 55.2 (2C, $ArOCH_3$, $OCH_3$); ESIMS (−): *m*/*z* 1253.9 $[M-3Na+2NH_4]^-$.

To a solution of **27** (0.22 g, 0.17 mmol) in MeCN (4.5 mL) and water (0.5 mL), ceric ammonium nitrate (0.27 g, 0.50 mmol) was added. The mixture was stirred at room temperature for 3 h, neutralized with saturated aq $NaHCO_3$ (2 mL), and concentrated. The residue was purified by column chromatography (12:2:1 EtOAc/MeOH/$H_2O$), and the product was eluted from a column of Dovex-50W X8 resin using MeOH to give **28** (0.16 g, 83%) as a foam; $^1H$ NMR (200 MHz, $CD_3OD$): δ 7.40–7.11 (m, 20H, aromatic), 5.82, 5.19 (2s, 2H, H-1,1″), 5.08 (d, 1H, *J* 11.0 Hz, $PhCH_2$), 5.01–4.86 (m, 2H), 4.82–3.90 (m, 16H), 3.78–3.51 (m, 3H), 3.46 (dd, 1H, $J_{1',2'}$ 3.3 Hz, $J_{2',3'}$ 9.9 Hz, H-2′), 3.38 (s, 3H, $OCH_3$); $^{13}C$ NMR (50 MHz, $CD_3OD$): δ 177.6, 176.9 (C-6,6″), 140.0, 139.1, 138.8, 138.3, 130.0, 129.5, 129.4, 129.3, 129.2, 128.92, 128.87, 128.7, 128.4 (aromatic), 101.7, 95.9, 94.7 (C-1,1′,1″), 78.2, 77.6, 75.70, 75.69, 73.61, 73.60, 72.69 (2C), 72.68 (2C), 71.89, 71.87, 70.75, 70.73, 69.1, 65.9, 63.1, 56.3 ($OCH_3$); ESIMS (−): *m*/*z* 1112.7 $[M-3Na+2H]^-$.

Compound **28** (0.15 g, 0.13 mmol) was hydrogenated, and *N*-sulfated as described for compound **3**, to give **4** (0.084 g, 68%) as a foam; $[\alpha]_D$ +3 (*c* 0.3, $H_2O$); $^1H$ NMR (400 MHz, $D_2O$): δ 5.23 (d, 1H, $J_{1',2'}$ 3.5 Hz, H-1′), 5.07 (dd, 1H, $J_{1'',2''}$ 1.4 Hz, $J_{1'',3''}$ 1.0 Hz, H-1″), 4.96 (dd, 1H, $J_{1,2}$ 2.0 Hz, $J_{1,3}$ 3.0 Hz, H-1), 4.76 (d, 1H, $J_{4'',5''}$ 2.2 Hz, H-5″), 4.36 (d, 1H, $J_{4,5}$ 2.5 Hz, H-5), 4.21 (ddd, 1H, $J_{1'',2''}$ 1.4 Hz, $J_{2'',3''}$ 3.0 Hz, $J_{2'',4''}$ 1.0 Hz, H-2″), 4.15 (ddd, 1H, $J_{1,3}$ 3.0 Hz, $J_{2,3}$ 3.0 Hz, $J_{3,4}$ 2.5 Hz, H-3), 4.14 (ddd, 1H, $J_{1,2}$ 2.0 Hz, $J_{2,3}$ 3.0 Hz, $J_{2,4}$ 1.5 Hz, H-2), 3.99 (ddd, 1H, $J_{1'',3''}$ 1.0 Hz, $J_{2'',3''}$ 3.0 Hz, $J_{3'',4''}$ 3.4 Hz, H-3″), 3.94 (ddd, 1H, $J_{2,4}$ 1.5 Hz, $J_{3,4}$ 2.5 Hz, $J_{4,5}$ 2.5 Hz, H-4), 3.90 (ddd, 1H, $J_{2'',4''}$ 1.0 Hz, $J_{3'',4''}$ 3.4 Hz, $J_{4'',5''}$ 2.2 Hz, H-4″), 3.79 (m, 1H, H-5′), 3.78 (dd, 1H, $J_{5',6a'}$ 2.2 Hz, $J_{6a',6b'}$ 12.4 Hz, H-6a′), 3.73 (dd, 1H, $J_{5',6b'}$ 3.6 Hz, $J_{6a',6b'}$ 12.4 Hz, H-6b′), 3.63 (dd, 1H, $J_{2',3'}$ 10.0 Hz, $J_{3',4'}$ 8.8 Hz, H-3′), 3.60 (dd, 1H, $J_{3',4'}$ 8.8 Hz, $J_{4',5'}$ 9.7 Hz, H-4′), 3.33 (s, 3H, $OCH_3$), 3.15 (dd, 1H, $J_{1',2'}$ 3.5 Hz, $J_{2',3'}$ 10.0 Hz, H-2′); $^{13}C$ NMR (100 MHz, $D_2O$): δ 176.7 (C-6″), 175.2 (C-6),

99.9 (C-1), 99.5 (C-1″), 97.4 (C-1′), 78.0 (C-4′), 76.1 (C-4), 75.3 (C-2), 73.8 (C-2″), 71.3 (C-5′), 69.7 (C-3′), 69.0 (2C, C-3″,4″), 68.6 (C-5″), 68.2 (C-3), 68.1 (C-5), 60.0 (C-6′), 58.6 (C-2′), 55.6 ($OCH_3$); HRMS (ESI) calcd for $C_{19}H_{26}NO_{26}S_3Na_5$ $[M-5Na+3H]^{2-}$ 391.5075. Found: 391.5090.

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