ELSEVIER

www.elsevier.nl/locate/carres

Carbohydrate Research 329 (2000) 561–577

CARBOHYDRATE
RESEARCH

Chemical synthesis of sulfated oligosaccharides with a β-D-Gal-(1 → 3)-[β-D-Gal-(1 → 4)-(α-L-Fuc-(1 → 3)-β-D-GlcNAc-(1 → 6)]-α-D-GalNAc sequence

Jie Xia, T. Srikrishnan, James L. Alderfer, R.K. Jain, Conrad F. Piskorz, Khushi L. Matta *

Molecular and Cellular Biophysics, Roswell Park Cancer Institute, Elm and Carlton Streets, Buffalo, NY 14263, USA

Received 22 May 2000; accepted 7 July 2000

Abstract

The syntheses of two sulfated pentasaccharides: β-D-Gal6SO_3Na-(1 → 3)-[β-D-Gal-(1 → 4)-α-L-Fuc-(1 → 3)-β-D-GlcNAc-(1 → 6)]-α-D-GalNAc → OMe (**1**) and β-D-Gal6SO_3Na-(1 → 3)-[β-D-Gal-(1 → 4)-α-L-Fuc-(1 → 3)-β-D-GlcNAc6SO_3Na-(1 → 6)]-α-D-GalNAc → OMe (**2**) by using Lewis^X trisaccharides **12** and **16** as glycosyl donors are described. Sulfated oligosaccharides **1**–**2** and intermediate compounds are fully characterized by 2D ^{1}H–^{1}H DQF-COSY and 2D ROESY experiments. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Chemical synthesis; 2D DQF-COSY; 2D ROESY; Sulfated oligosaccharides; Crystallography

1. Introduction

Mammalian lectins mediate the early steps of recruitment of leukocytes from the bloodstream in a variety of normal and pathological situations [1–4]. In the last decade there has been a tremendous interest in the synthesis of high affinity molecules that target the selectin family of cell-surface proteins (L, P and E) because these molecules can become future drugs for treatment of various diseases such as chronic and acute inflammation. Since the natural selectin ligands (GlyCAM-1 and PSGL-1) carry sialyl Le^X or NeuAcα-(2 → 3)-Galβ-(1 → 3)-GalNAc oligosaccharide unit as part of core 2 O-linked structures, much effort has been made for the synthesis of sialylated and sulfated Lewis^X, and mimics of SLe^X [5,6]. Recently, Kiessling and co-workers [7] have examined the inhibition of L-selectin with various disulfated Le^X derivatives. We became interested in the synthesis of sialyl and sulfated core 2 analogs for the binding studies of these selectins because the role of core 2 analogs as inhibitors for these selectins has become evident [8]. Our interest in core 2 branched structures which contain sulfates has also stemmed from our study of sulfotransferase using the core 2 branched structures as acceptors [9]. We hereby report the synthesis of branched structures **1** and **2**.

---

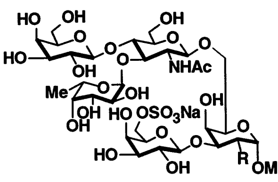
* Corresponding author. Tel.: + 1-716-8452300; fax: + 1-716-8453458.

E-mail address: klmatta@yahoo.com (K.L. Matta).

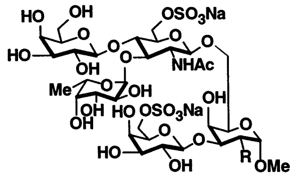
PII: S0008-6215(00)00211-1

2. Results and discussion

Synthesis of trisaccharide building blocks **12** *and* **16**.—In order to synthesize two sulfated oligosaccharides **1** and **2**,

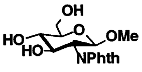


1: R = NHAc

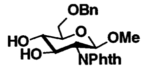


2: R = NHAc

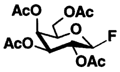
key building blocks **12**, **16**, **21** and **27** were prepared starting from monosaccharide building blocks



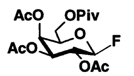
4



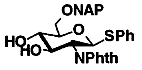
5



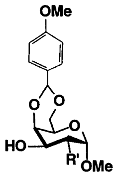
6



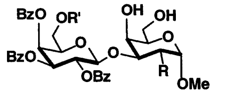
13



14



20: R′ = NHAc

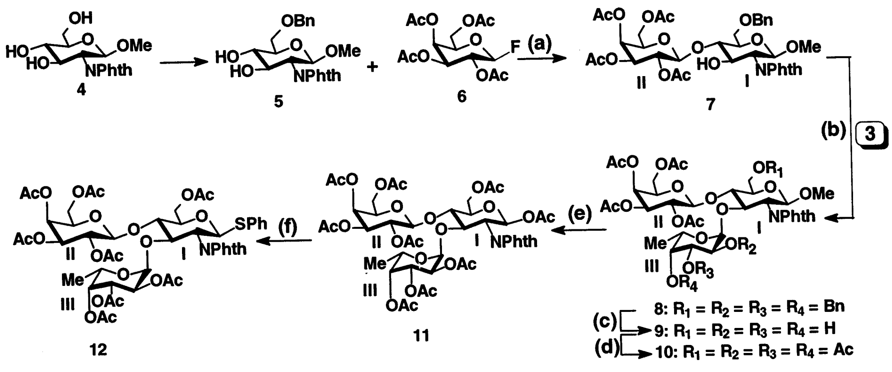


27: R = NHAc, R′ = BrCHCO

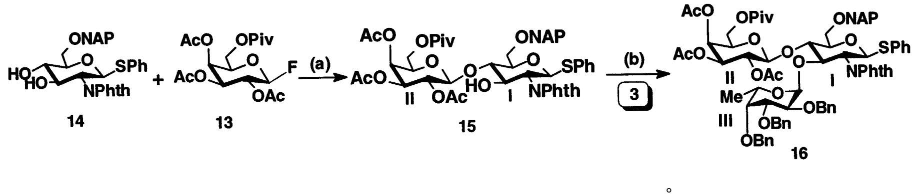
according to Schemes 1–3.

The HO-4 of diol **5** was regioselectively glycosylated with donor **6** in the presence of silver triflate–tin chloride as catalyst [10] to give disaccharide **7** in good yield (73%). The stereochemical outcome of both the glycosylic and position connectivity of disaccharide **7** was well established through homonuclear correlation experiments (2D $^1H-^1H$ DQF-COSY: Fig. 1 and 2D ROESY: Fig. 2). The (1 → 4)-linkage of disaccharide **7** was indicated by the observation of a strong NOE cross peak between H-1^{II} and H-4^{I} in the 2D ROESY spectrum. A 1,2-*trans* glycoside of disaccharide **7** was confirmed by a larger coupling constant of $^3J_{1,2}$ (J 8.0 Hz). The structure of disaccharide **7** was further characterized by X-ray diffraction analysis (Fig. 3).

Disaccharide **7** was fucosylated with methyl 2,3,4-tri-*O*-benzyl-1-thio-β-L-fucoside (**3**) in the presence of tetrabutylammonium bromide–copper bromide as catalyst [11], affording trisaccharide **8** in good yield (76%). The removal of benzyl groups from compound **8** was achieved in the presence of 10% palladium-on-carbon catalyst under a hydrogen atmosphere. Compound **9** was then treated with 1:1 acetic anhydride–pyridine in the presence



Scheme 1. Reagents and conditions. (a) 5:1 AgOTf–$SnCl_2/CH_2Cl_2$–toluene, 4 Å 10% MS, −15 to 0 °C, 12 h, 73%; (b) **3**, $CuBr_2$–n-Bu_4NBr, 5:1 $ClCH_2CH_2Cl$–DMF, 4 Å MS, rt, 16–24 h, 91%; (c) 10% H_2/Pd–C, 1:1 CH_2Cl_2–CH_3OH, rt, 12 h, 96%; (d) 1:1 Ac_2O–pyridine, DMAP, rt, 12 h, 96%; (e) H_2SO_4–Ac_2O, rt, 12 h, 76%; (f) PhSH/CH_2Cl_2, $BF_3{\cdot}Et_2O$, rt, N_2, 16 h, 67%.



Scheme 2. Reagents and conditions: (a) AgOTf–$SnCl_2$/5:1 CH_2Cl_2–toluene, 4 Å MS, −15 to 0 °C, 12 h, 75%; (b) **3**, $CuBr_2$–n-Bu_4NBr, 5:1 $ClCH_2CH_2Cl$–DMF, 4 Å MS, rt, 16–24 h, 76%.

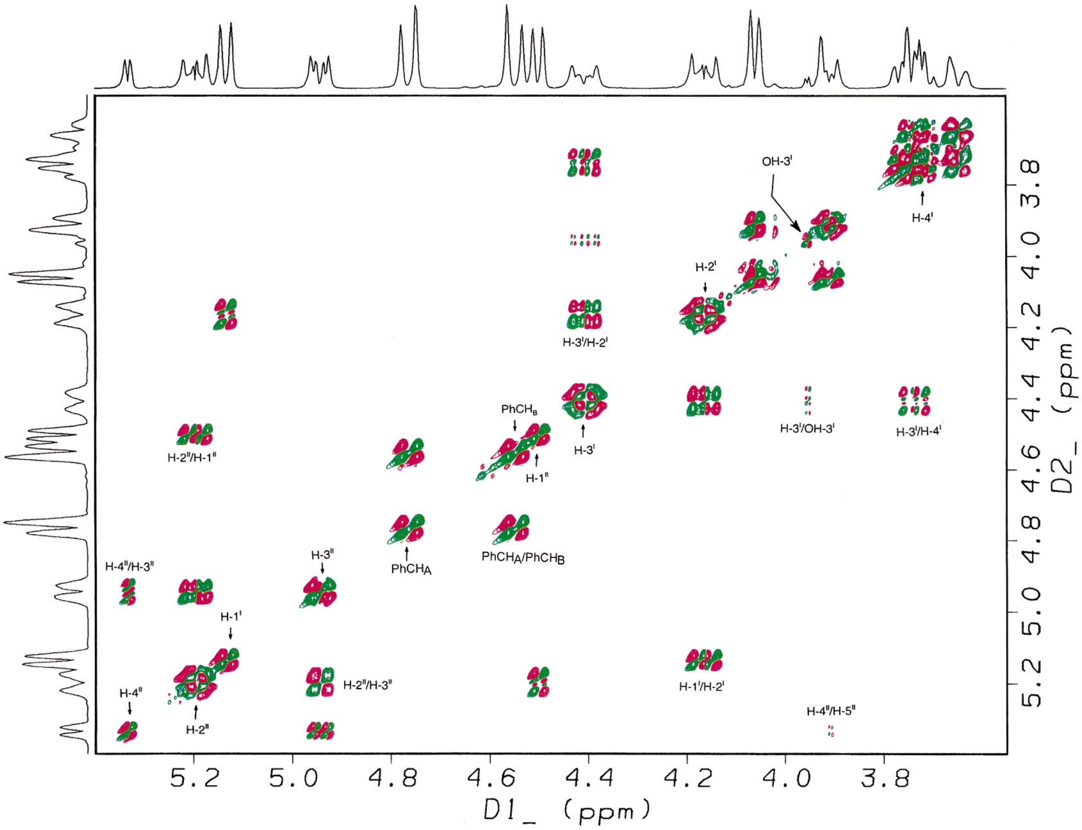


Fig. 1. 400 MHz 2D ^1H–^1H DQF-COSY spectrum of disaccharide **7** recorded in $CDCl_3$ at 303.0 K.

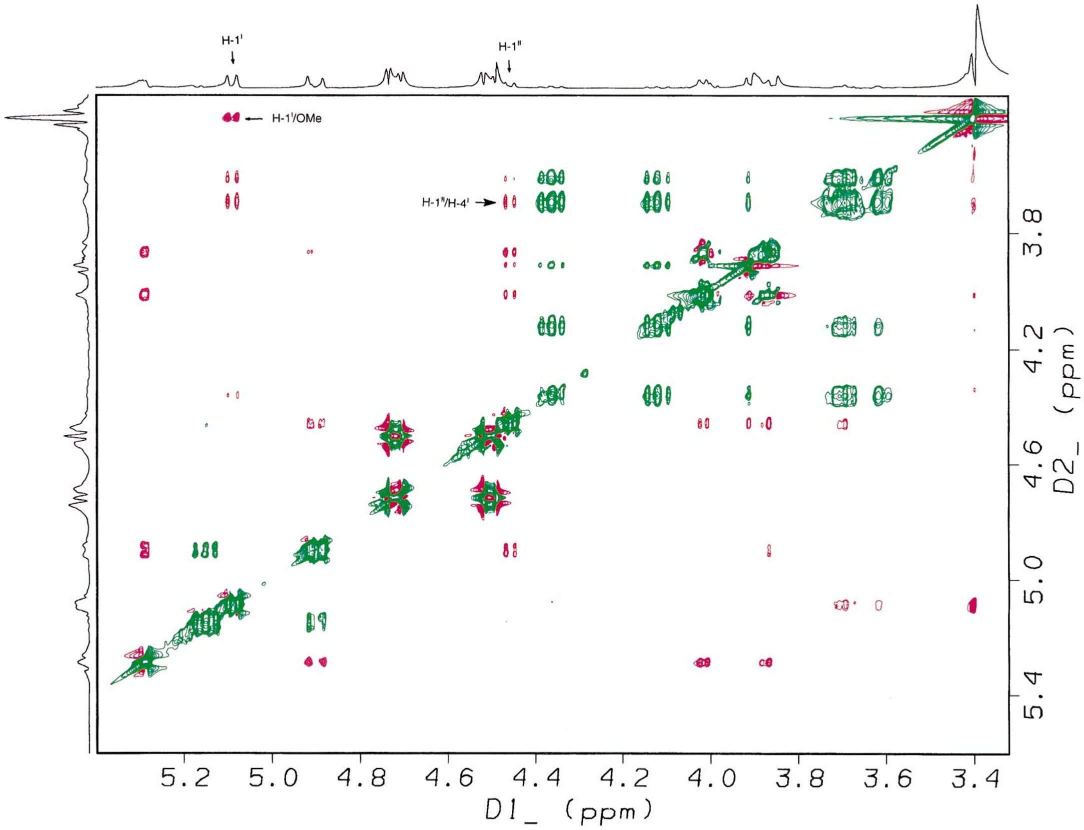
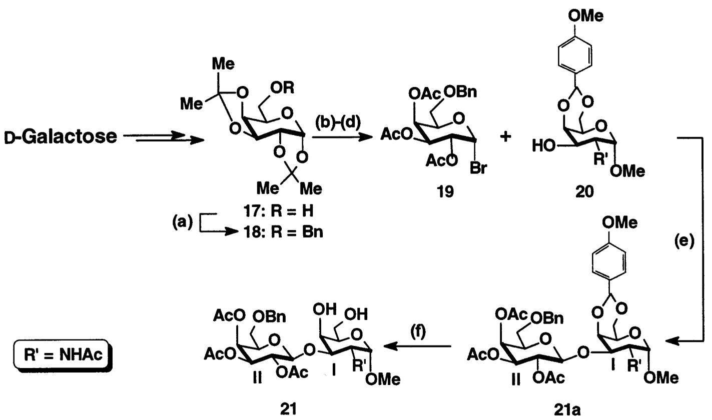


Fig. 2. 400 MHz 2D ROESY spectrum of disaccharide **7** recorded in $CDCl_3$ at 303.0 K.



Scheme 3. Reagents and conditions: (a) BnBr/KOH, 18-crown-6, THF, rt, 12 h, 86%; (b) 1 N HCl–1,4-dioxane, 4 h, 75%; (c) 1:1 pyridine–Ac_2O, 65–70 °C, 3 h, 76%; (d) $Me_3SiBr/BiBr_3$, CH_2Cl_2, rt, 12 h, 59%; (e) $Hg(CN)_2$–$HgBr_2/ClCH_2CH_2Cl$, 4 Å MS, rt, 12 h, 66%; (f) 60% HOAc, 60–65 °C, 1.5 h, 76%.

of a catalytic amount of DMAP, providing compound **10**. The conversion of compound **10** into compound **11** was successfully performed by treatment of **10** with acetic anhydride [12] and a catalytic amount of concentrated sulfuric acid. Trisaccharide donor **12** was obtained by treatment of compound **11** with thiophenol in the presence of BF_3Et_2O [13] under a nitrogen atmosphere (Scheme 1). The structure of trisaccharide donor **12** was fully confirmed by a combination of 2D DQF-COSY and 2D ROESY experiments. For preparation of trisaccharide donor **16**, a strategy of sequential glycosylation is employed. Use of three different leaving groups (fluorine, SMe and SPh) which exhibit different reactivities in different coupling reactions allows us to carry out the direct glycosylation of monosaccharide acceptor **13**. The construction of trisaccharide donor **16** was outlined in Scheme 2. Regioselective glycosylation of diol **13** with donor **14** was performed in the presence of silver triflate–tin chloride as catalyst to afford disac-

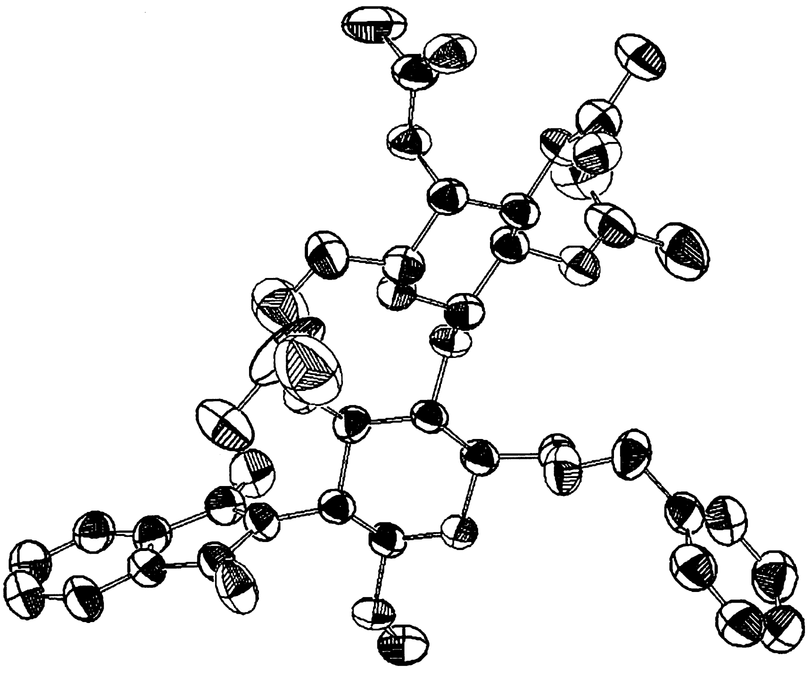
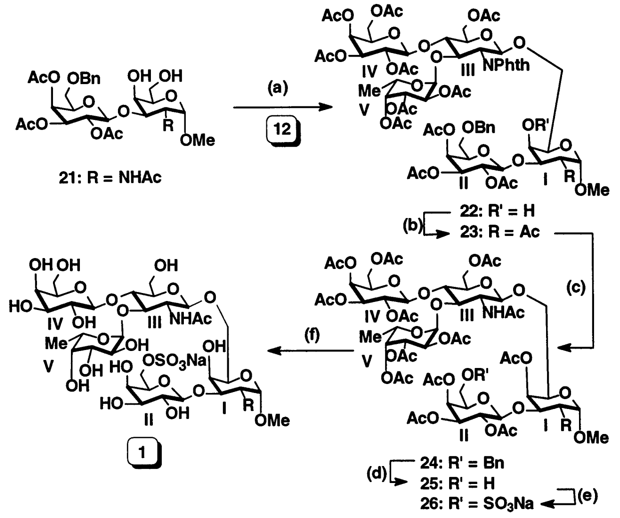
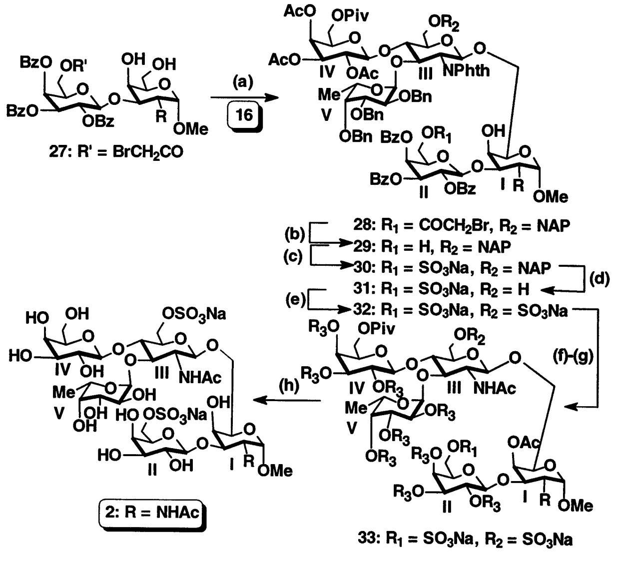


Fig. 3. An ORTEP [22] diagram showing the molecular structure of disaccharide **7**.



Scheme 4. Reagents and conditions: (a) NIS–TfOH (cat)/CH_2Cl_2, 4 Å MS, −65 to −60 °C, 3–4 h, 44%; (b) 1:1 Ac_2O–pyridine, DMAP, rt, 12 h, 86%; (c) 1:5 $NH_2NH_2 \cdot xH_2O$–CH_3OH, 85–90 °C, 4 h, then, 1:1 Ac_2O–pyridine, DMAP, rt, 12 h, 76%; (d) H_2, 10% Pd–C, CH_2Cl_2–CH_3OH, rt, 6–8 h, quantitative; (e) $SO_3 \cdot$pyridine–pyridine, 0–25 °C, 6 h, 79%; (f) 1 M, 1:1 CH_3ONa–CH_3OH (cat)/CH_3OH–water, rt, 24 h, 36%.



Scheme 5. Reagents and conditions: (a) NIS–TfOH (cat)/CH_2Cl_2, 4 Å MS, −75 to −70 °C, 1.5 h, 46%; (b)HN_2CSNH_2/2,6-lutidine, 60–65 °C, 18 h, 87%; (c) $SO_3 \cdot$pyridine–pyridine, 0–5 °C, 12 h, 79%; (d) DDQ/4:1 CH_2Cl_2–CH_3OH, rt, 20 h, 79%; (e) $SO_3 \cdot$pyridine–pyridine, 0–5 °C, 12 h, 78%; (f) H_2/10% Pd–C, 4:1 CH_2Cl_2–MeOH, rt, 10 h, 87%; (g) 1:5 $NH_2NH_2 \cdot xH_2O$–CH_3OH, 85–90 °C, 4–5 h, then, 1:1 Ac_2O–pyridine, DMAP, rt, 12 h, 76%; (h) 1 M, CH_3ONa–CH_3OH (cat)/1:1 CH_3OH–water (1:1), rt, 12 h, 26%.

charide **15** in good yield (75%). Disaccharide **15** was further fucosylated with donor **3** to give trisaccharide **16** in excellent yield (76%). The structure of trisaccharide **16** was established by 2D NMR experiments (2D DQF-COSY and 2D ROESY). The α-L-fucopyranoside of trisaccharide **16** was indicated by a small coupling constant of $^3J_{1,2}$ (J 3.3 Hz), which is a characteristic feature of a 1,2-*cis*-α-L-fucopyranoside.

Synthesis of disaccharide building block **21**.—Disaccharide building block **27** was prepared according to documented protocol [14]. The construction of disaccharide acceptor **21** is illustrated in Scheme 3. Compound **17** was prepared according to literature procedures [15]. Monosaccharide **17** was treated with potassium hydroxide in the presence of a phase-transfer catalyst [16] (18-crown-6) in dry THF, followed by benzyl bromide, to afford compound **18** in excellent yield (86%). Compound **18** was treated with 1 N HCl in 1,4-dioxane at reflux, resulting in removal of the *O*-isopropylidene group from **18**. The resulting 6-*O*-benzyl-D-galactose was further acetylated with 1:1 acetic anhydride–pyridine at 65–70 °C to give 6-*O*-benzyl-1,2,3,4-tetra-*O*-acetyl-D-galactopyranose, which was then treated with bromotrimethylsilane in the presence of bismuth(III) bromide [17] to give donor **19**. Disaccharide **21a** was obtained by treatment of compound **20** with glycosyl donor **19** in the presence of mercury cyanide and mercury bromide as catalysts [18]. Compound **21a** was treated with 60% acetic acid at 60–65 °C to give the desired disaccharide acceptor **21** in good yield (76%).

The construction of target sulfated oligosaccharides **1** *and* **2**.—The target sulfated oligosaccharides **1** and **2** were constructed as shown in Schemes 4 and 5. On account of the much higher reactivity of the primary hydroxyl group in acceptors **21** and **27**, totally regioselective glycosylation of the primary HO-6 of disaccharide acceptor **21** with trisaccharide donor **12** was achieved under controlled glycosylation conditions to give **22** as the only glycosylation product. The β-(1 → 6)-linkage of **22** was indicated by observation of strong NOE cross peaks between H-1^{III} of sugar residue **III** and H-$6a^{I}$, H-$6b^{I}$ of sugar residue **I** by 2D ROESY spectroscopy. Compound **22** was acetylated with 1:1 acetic anhydride–pyridine in the presence of a catalytic amount of DMAP to give compound **23**. Compound **23** was then treated with 1:5 $NH_2NH_2 \cdot H_2O$–methanol at 85–90 °C, which resulted in the removal of the phthalimido group, followed with 1:1 acetic anhydride–pyridine in the presence of a catalytic amount of DMAP at room temperature to afford compound **24**. Compound **24** was treated with 10% palladium-on-carbon in a hydrogen atmosphere to give compound **25**. Compound **26** was obtained by treatment of compound **25** with sulfur trioxide–pyridine complex in dry pyridine. O-Deacetylation of **26** was performed by treatment with 1 M sodium methoxide to give compound **1**. The structure of compound **1** was well characterized by 2D ^1H–^1H DQF-COSY, 2D ROESY (Fig. 4), and ^{13}C NMR spectroscopy and by FABMS. Similarly, the synthesis of target oligosaccharide **2** was carried out as outlined in Scheme 5. Regioselective glycosylation of HO-6 of diol **27** was achieved by treatment of diol **27** with trisaccharide donor **16** under controlled glycosylation conditions.

Compound **28** was obtained in reasonable yield (46%). The β-(1→6)-linkage of compound **28** was indicated by observation of strong NOE cross peaks between H-1^{III} of sugar residue **III** and H-$6a^{I}$, H-$6b^{I}$ of sugar residue **I** by 2D ROESY spectroscopy. The α-bromoacetyl group was removed by treatment of compound **28** with thiourea and 2,6-lutidine at reflux in dichloromethane to give compound **29**. Compound **29** was then treated with the sulfur trioxide–pyridine complex in dry pyridine to give compound **30**. The next reaction was the selective removal of the 2-naphthymethyl (NAP) protecting group from compound **30**. The NAP protecting group was recently introduced as a hydroxyl protecting group [19] and can be removed by DDQ in the presence of benzyl groups [20]. Thus, compound **30** was carefully treated with DDQ, resulting in the removal of NAP protecting group. Compound **31** was then treated with the sulfur trioxide–pyridine complex in dry pyridine to give **32**. Compound **32** was deprotected by the following procedure to give target oligosaccharide **2**: (a) removal of benzyl

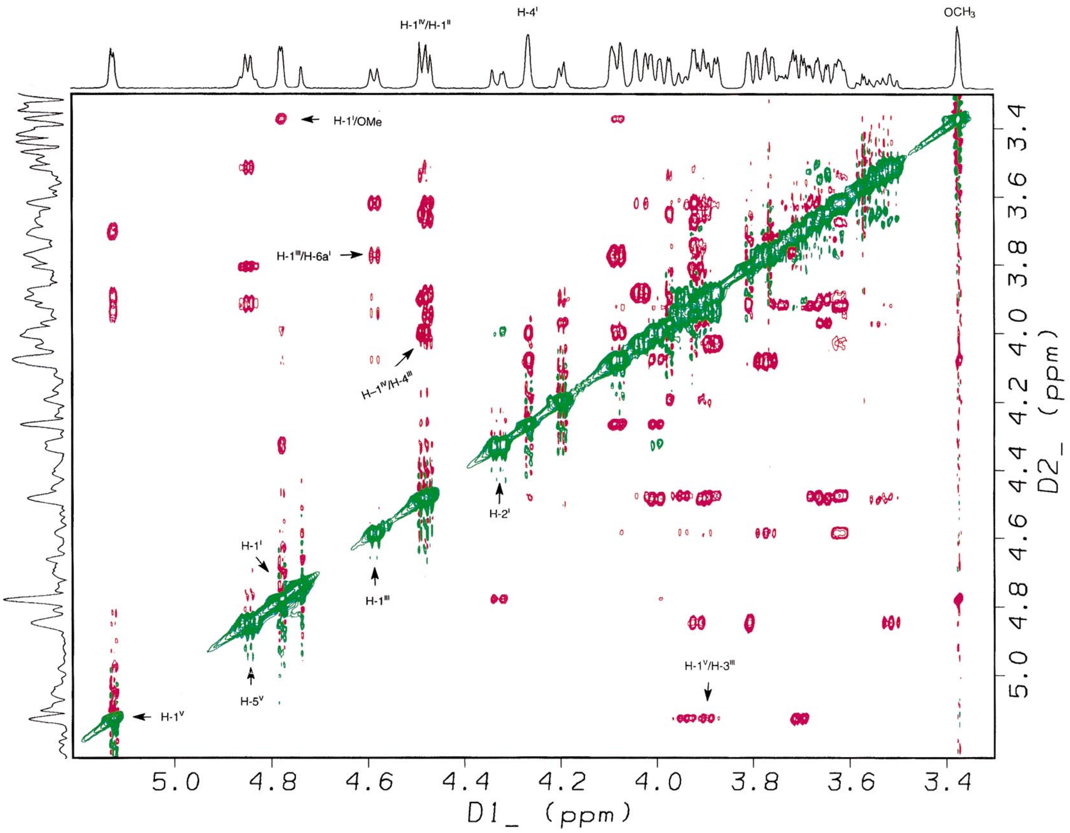


Fig. 4. 600 MHz 2D ROESY spectrum of pentasaccharide **1** recorded in D_2O at 303.0 K.

protecting groups; (b) removal of the phthalimido group, followed with acetic anhydride–pyridine and DMAP; (c) removal of acetyl and trimethylacetyl groups, which is shown in Scheme 5. Target oligosaccharide **2** was well characterized with 2D ^{1}H–^{1}H DQF-COSY, 2D ROESY, and ^{13}C NMR spectroscopy and by FABMS.

In summary, we describe the convergent syntheses of two sulfated pentasaccharides **1** and **2** through a potentially general strategy of regio- and stereoselective glycosylation by the use of unprotected or partially protected acceptors.

3. Experimental

General procedures.—TLC was conducted on glass plates precoated with a 0.25-mm layer of Silica Gel 60 F-254 (Analtech, GHLF uniplates). The components were located either by exposure to UV light or by spraying with 10% H_2SO_4, and 0.2% *p*-anisaldehyde in EtOH soln. Solutions were concd under reduced pressure. The silica gel used for column chromatography was Baker Analyzed (60–200 mesh ASTM). Optical rotations were measured at 25 °C with a Perkin–Elmer 241 polarimeter. ^{1}H NMR spectra were recorded at 303.0 K with either a Bruker AM 400 (400 MHz) or AMX 600 (600 MHz) spectrometer. The values of δ (ppm) are given relative to the signal (δ 0) for internal Me_4Si for solns in $CDCl_3$, CD_2Cl_2, or CD_3OD. ^{13}C NMR spectra were recorded at 303.0 K with a Bruker AM 400 (100.6 MHz) spectrometer using $CDCl_3$ (77.0 ppm), CD_2Cl_2 (54.15 ppm), CD_3OD (49.15 ppm), or acetone-d_6 (206.0 or 29.8 ppm) as reference. First-order chemical shifts and coupling constants (J/Hz) were obtained from one-dimensional spectra, and assignments of protons were based on 2D DQF ^{1}H–^{1}H COSY and 2D ROESY experiments. Two-dimensional double-quantum filtered phase sensitive ^{1}H–^{1}H correlated spectra (DQF ^{1}H–^{1}H COSY), rotating-frame nuclear Overhauser enhancement spectroscopy (ROESY) were recorded at 303.0 K using a Bruker AM 400 (400 MHz) spectrometer and a Bruker AMX 600 (600 MHz) spectrometer. For ROESY experiments, the mixing time was set at 400 ms. All samples submitted for elemental analyses were dried under vacuum over P_2O_5 at rt. Elemental analyses were carried out by Robertson Laboratory, Madison, NJ. *p*-Toluenesulfonic acid monohydrate (*p*-TsOH·H_2O) was dried by co-evaporating with dry MeCN three times at 80 °C before use. Dichloromethane, MeCN, MeOH, benzene, and DMF were kept dry over 4 Å MS, pyridine was redistilled over KOH, and nitromethane was freshly distilled over P_2O_5.

*Methyl (2,3,4,6-tetra-*O*-acetyl-β-*D*-galactopyranosyl)-(1→4)-6-*O*-benzyl-2-deoxy-2-phthalimido-β-*D*-glucopyranoside* (**7**).—A soln of compound **5** (1.18 g, 3.38 mmol), compound **6** (2.37 g, 5.75 mmol), and $SnCl_2$ (1.13 g, 4.39 mmol) in dry 5:1 CH_2Cl_2–toluene (43 mL) containing 4 Å MS (8 g) was stirred at − 15 °C for 2 h under a N_2 atmosphere. Silver triflate (832 mg, 4.39 mmol) was added, and the mixture was stirred overnight at − 15 to 0 °C under a N_2 atmosphere, then neutralized with Et_3N. The solids were filtered off, and the organic layer was washed with satd $NaHCO_3$ soln, water, and dried (Na_2SO_4). The organic solvent was evaporated under reduced pressure to give a crude mixture, which was then applied to a column of silica gel and eluted with 1:1 hexane–EtOAc to give pure compound **7** (1.84 g, 73%) as a white solid. R_f 0.34 (1:1 hexane–EtOAc); $[\alpha]_D$ + 8.8° (c 0.5, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 7.84–7.82 (m, 2 H, ArH), 7.72–7.70 (m, 2 H, ArH), 7.41–7.32 (m, 5 H, ArH), 5.33–5.32 (d, 1 H, J 3.2 Hz, H-4^{II}), 5.21–5.16 (dd, 1 H, J 8.0, 10.4 Hz, H-2^{II}), 5.13–5.11 (d, 1 H, $J_{1,2}$ 8.8 Hz, H-1^{I}), 4.95–4.92 (dd, 1 H, J 3.6, 10.2 Hz, H-3^{II}), 4.77–4.74 (d, 1 H, J_{gem} 12.4 Hz, OCH_APh, ABq), 4.55–5.52 (d, 1 H, J_{gem} 12.0 Hz, OCH_BPh, ABq), 4.50–4.48 (d, 1 H, $J_{1,2}$ 8.0 Hz, H-1^{II}), 4.42–4.37 (t, 1 H, H-3^{I}), 4.18–4.13 (dd, 1 H, J 8.8 Hz, H-2^{I}), 4.06–4.04 (m, 2 H, H-6b^{II}, H-6a^{II}), 3.95 (d, 1 H, OH-3^{I}), 3.91–3.88 (t, 1 H, J 7.2, 6.4 Hz, H-5^{II}), 3.77–3.69 (m, 3 H, H-5^{I}, H-4^{I}, H-6b^{I}), 3.65–3.62 (m, 1 H, H-6a^{I}), 3.44 (s, 3 H, OCH_3), 2.11 (s, 3 H, Ac), 2.00 (s, 3 H, Ac), 1.95, (s, 3 H, Ac), 1.91 (s, 3 H, Ac); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 170.46 (C=O), 170.06 (C=O), 169.92 (C=O), 169.15 (C=O), 138.18, 133.99,

Table 1
X-ray crystal data and structure refinement for **7**

Compound	Methyl (2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-(1 → 4)-6-*O*-benzyl-2-deoxy-2-phthalimido-β-D-glucopyranoside (**7**)
Color/shape	white/rectangular needle
Empirical formula	$C_{36}H_{41}NO_{16}$
Formula weight	743.7
Temperature (°C)	22 ± 3
Crystal system	monoclinic
Space group	$P2_1$
Unit cell dimensions	
a (Å)	12.529 (3)
b (Å)	14.765 (5)
c (Å)	10.728 (3)
α (°)	90.0
β (°)	113.98 (2)
γ (°)	90.0
Volume (Å^3)	1813.3(9)
Z	2 (two molecules per unit cell)
D_{calcd} (g cm^{-3})	1.362
Absorption coefficient (mm^{-1})	0.915
Difftractometer	CAD-4
Radiation ($\lambda = 1.5418$ Å)	Cu K$_\alpha$
F(000)	784
Crystal size (mm)	$0.15 \times 0.45 \times 0.25$
2θ Range for data collection (°)	0–150
Index ranges (°)	$-10 \leq h \leq 10$, $0 \leq k \leq 17$, $-18 \leq l \leq 18$
Reflections collected	4099
Independent/observed reflections	3892 (3820 with $I \leq 3\sigma$)
Absorption correction	semiempirical from psi scans
Range of relative transmission factors	0.99 and 0.90
Refinement method	full-matrix least-squares on F^2
Computing	SHELXS-96
Data/restraints/parameters	3820/0/643
Goodness-of-fit on F^2	1.120
Function minimized	$\Sigma[\lvert F_o^2\rvert - (1/k)\lvert F_c^2\rvert]$
Final R indices	0.0879 for all 3892 reflections 0.064 for 3186 [$I \geq 4\sigma(I)$]
Final extinction coefficient	5.59×10^{-7}
Large difference peak and hole (e Å^{-3})	0.15 and −0.15

131.92, 128.55, 127.91, 127.86, 123.34, 101.54, 99.26, 82.06, 74.27, 73.74, 71.22, 70.81, 69.69, 68.80, 67.97, 66.90, 61.47, 56.75, 55.94, 20.72 (Ac), 20.56 (Ac), 20.50 (Ac), 20.33 (Ac); Anal Calcd for $C_{36}H_{41}NO_{16}$: C, 58.12; H, 5.56; N, 1.88. Found: C, 58.00; H, 5.59; N, 1.81.

X-ray crystallography.—Crystals of **7** were obtained by a slow evaporation of the compound dissolved in hexanes–EtOAc. The crystals were rectangular needles. A crystal of dimensions $0.15 \times 0.45 \times 0.25$ mm was used for the X-ray diffraction. The crystals belonged to a monoclinic system, space group $P2_1$. Table 1 gives the crystal data and structure refinement parameters.

Complete three-dimensional data were collected on an Enraf–Nonius CAD-4 computer-controlled diffractometer. A total of 4099 reflections ($2\theta_{\max} = 150°$) were collected by the $\omega/2\theta$ method, out of which 3820 were considered significant ($I \geq 3\sigma$). The crystal structure was solved by a routine and straightforward application of the SHELX-96 program on a Silicon Graphics R10,000 computer. The structure was refined using the SHELX-96 package [21] of programs. All hydrogen atoms were located on a difference Fourier map. Refinements were carried out with anisotropic thermal parameters for the nonhydrogen atoms and isotropic thermal parameters for the hydrogen atoms. The final reliability index (R-factor) was 0.0879 for all the observed 3892 reflections and 0.064 for 3186 reflections ($I \geq 4\sigma$) with a goodness of fit parameter $S =$ 1.120. Table 2 gives the final positional parameters for the nonhydrogen atoms in the structure. The crystal structural investigation confirms the structure of compound **7**. The disaccharide has the sugar residues, the galactopyranosyl and the glucopyranoside in the $^4C_1$ chair conformation. The ring-to-ring conformation can be described as $(\phi, \psi) = (159.7, -126.3)$. The conformation across the C-5–C-6 bond is trans–gauche for the galactopyranosyl ring and gauche–gauche for the glucopyranosyl ring. The crystal structure is stabilized by a network of O–H···O hydrogen bonds.

*Methyl (2,3,4,6-tetra-*O*-acetyl-β-*D*-galactopyranosyl)-(1 → 4)-[(2,3,4-tri-*O*-benzyl-α-*L*-fucopyranosyl)-(1 → 3)]-6-*O*-benzyl-2-deoxy-2-phthalimido-β-*D*-glucopyranoside* (**8**).—A soln of compound **7** (320 mg, 0.36 mmol), methyl 2,3,4-tri-*O*-benzyl-1-thio-β-L-fucopyranoside

Table 2
Final fractional positional parameters for nonhydrogen atoms with estimated standard deviations given in parentheses for **7**

Atom	x	y	z	U_{eq} [a]
O-1	0.0784(3)	0.5082(3)	0.7758(4)	0.0480(8)
O-2	0.1769(3)	0.3972(2)	0.9223(4)	0.0487(9)
O-3	−0.0120(3)	0.7273(3)	0.8543(4)	0.0508(9)
O-4	0.0887(4)	0.8505(3)	0.9695(4)	0.0523(9)
O-5	−0.0927(4)	0.3807(3)	0.6207(4)	0.0587(10)
O-6	−0.0191(4)	0.2016(3)	0.6622(4)	0.0553(10)
O-7	0.2190(4)	0.2272(3)	0.8293(5)	0.0595(11)
O-8	0.3041(5)	0.7413(5)	1.2883(5)	0.0765(15)
O-9	0.2848(4)	0.5702(3)	1.0130(5)	0.0598(10)
O-10	−0.1547(4)	0.5666(4)	0.8231(5)	0.0711(13)
O-11	−0.0860(5)	0.1083(3)	0.7786(7)	0.0686(13)
O-12	0.1886(5)	0.0781(3)	0.8058(7)	0.0840(20)
O-13	0.3417(4)	0.8014(4)	0.8940(5)	0.0714(13)
O-14	−0.0515(7)	0.3525(7)	0.4401(6)	0.1130(20)
O-15	0.3475(8)	0.3787(6)	1.1611(9)	0.1220(30)
O-16	0.2713(11)	0.3584(11)	1.3027(11)	0.1760(50)
N-1	0.2987(4)	0.7553(3)	1.0723(5)	0.0466(10)
C-1	0.0170(5)	0.2641(4)	0.7725(6)	0.0499(12)
C-2	0.0793(5)	0.5834(3)	0.8617(6)	0.0441(11)
C-3	0.1733(6)	0.3142(4)	0.9879(6)	0.0520(13)
C-4	0.1874(5)	0.7107(4)	1.0002(6)	0.0442(11)
C-5	0.0977(5)	0.7733(4)	0.9023(6)	0.0458(11)
C-6	0.0652(5)	0.4242(4)	0.8260(6)	0.0475(12)
C-7	0.1981(5)	0.6268(4)	0.9215(6)	0.0486(12)
C-8	0.1329(5)	0.2379(4)	0.8859(6)	0.0507(12)
C-9	−0.0132(5)	0.6506(4)	0.7745(6)	0.0503(12)
C-10	0.0228(5)	0.3563(4)	0.7127(6)	0.0491(12)
C-11	0.3474(6)	0.7676(4)	1.2134(6)	0.0547(14)
C-12	−0.0712(8)	0.1259(6)	0.6711(11)	0.0640(20)
C-13	0.0335(8)	0.9240(4)	0.8793(8)	0.0620(20)
C-14	0.5617(6)	0.8844(5)	1.1259(9)	0.0680(20)
C-15	−0.3636(6)	0.5634(4)	0.7549(8)	0.0610(20)
C-16	0.3661(5)	0.7984(4)	1.0152(6)	0.0516(13)
C-17	0.2945(7)	0.3002(5)	1.0978(9)	0.0720(20)
C-18	0.4672(5)	0.8372(4)	1.1276(7)	0.0553(14)
C-19	0.2375(6)	0.1436(5)	0.7931(8)	0.0670(20)
C-20	0.4576(5)	0.8172(4)	1.2462(7)	0.0561(14)
C-21	0.6384(7)	0.8890(7)	1.3696(10)	0.0830(20)
C-22	−0.2430(14)	0.3994(12)	0.4055(12)	0.1130(50)
C-23	−0.1078(10)	0.0681(7)	0.5539(10)	0.0870(30)
C-24	−0.1212(8)	0.3753(6)	0.4867(7)	0.0770(20)
C-25	−0.5262(8)	0.6569(7)	0.6200(10)	0.0850(20)
C-26	0.3151(11)	0.3873(11)	1.2405(14)	0.1330(60)
C-27	−0.5626(8)	0.6626(7)	0.7244(11)	0.0880(20)
C-28	−0.4034(8)	0.5695(6)	0.8603(9)	0.0760(20)
C-29	−0.4275(7)	0.6083(6)	0.6365(8)	0.0740(20)
C-30	−0.2567(6)	0.5123(6)	0.7767(10)	0.0750(20)
C-31	0.6463(7)	0.9103(6)	1.2502(10)	0.0760(20)
C-32	0.5421(6)	0.8417(6)	1.3724(8)	0.0740(20)
C-33	0.3241(14)	0.1464(10)	0.7335(20)	0.1250(70)
C-34	0.3829(11)	0.4979(7)	1.3227(10)	0.0890(20)
C-35	−0.5004(9)	0.6193(7)	0.8430(11)	0.0890(20)
C-36	−0.1354(6)	0.6116(5)	0.7199(7)	0.0604(15)

[a] U_{eq} is defined as the trace of the orthogonalized U_{ij} tensor.

(**3**, 338 mg, 0.72 mmol), and n-Bu$_4$NBr (232 mg, 0.72 mmol) in dry 5:1 1,2-dichloroethane–DMF (15 mL) containing 4 Å MS (2 g) was stirred for 2 h at rt under a N$_2$ atmosphere. Copper bromide (161 mg, 0.72 mmol) was added, and the mixture was stirred overnight at rt. An additional portion of compound **3** (790 mg), CuBr$_2$ (101 mg) and n-Bu$_4$NBr (145 mg) were added, and the stirring was continued for a total of 24 h. The solids were filtered off, the organic layer was washed with satd NaHCO$_3$ soln, and water, and then it was dried (Na$_2$SO$_4$) and concd. The crude product was applied to a short column of silica gel and eluted with 2:1 hexane–EtOAc to give compound **8** (380 mg, 0.32 mmol, 91%) as a white solid. R_f 0.17 (2:1 hexane–EtOAc); $[\alpha]_D$ + 5.4° (c 0.7, CHCl$_3$); ^{1}H NMR (CDCl$_3$, 600 MHz): δ 7.67–7.66 (m, 2 H, ArH), 7.41–7.39 (m, 2 H, ArH), 7.26–7.01 (m, 20 H, ArH), 5.23–5.22 (d, $J_{1,2}$ 3.2 Hz, H-4$^{\mathrm{II}}$), 5.01–4.99 (m, 2 H, H-2$^{\mathrm{II}}$, H-1$^{\mathrm{I}}$, $J_{1,2}$ 8.4 Hz), 4.85–4.73 (m, 4 H, OCHPh, H-1$^{\mathrm{III}}$, OCHPh, H-3$^{\mathrm{II}}$), 4.71–4.62 (m, 2 H, H-1$^{\mathrm{II}}$, H-3$^{\mathrm{I}}$), 4.61–4.59 (dd, 2 H, OCH$_2$Ph), 4.58–4.55 (d, 1 H, J_{gem} 12.0 Hz, OCHPh, ABq), 4.51–4.48 (d, 1 H, J_{gem} 12.0 Hz, OCHPh, ABq), 4.42–4.33 (m, 2 H, OCHPh, H-2$^{\mathrm{I}}$), 4.26–4.23 (d, 1 H, J_{gem} 12.0 Hz, ABq), 4.15–4.09 (m, 2 H, H-4$^{\mathrm{I}}$, H-6b$^{\mathrm{II}}$), 4.00–3.94 (dd, 1 H, H-6a$^{\mathrm{II}}$), 3.92–3.80 (m, 4 H, H-6a$^{\mathrm{I}}$, H-3$^{\mathrm{III}}$, H-6b$^{\mathrm{I}}$, H-2$^{\mathrm{III}}$), 3.61 (d, 1 H, J 3.2 Hz, H-4$^{\mathrm{III}}$), 3.55–3.45 (m, 2 H, H-5$^{\mathrm{I}}$, H-5$^{\mathrm{II}}$), 3.28 (s, 3 H, OCH$_3$), 2.01 (s, 3 H, Ac), 2.00 (s, 3 H, Ac), 1.94 (s, 3 H, Ac), 1.84 (s, 3 H, Ac), 1.21–1.19 (d, 3 H, J 6.4 Hz, CH$_3^{\mathrm{III}}$); ^{13}C NMR (CDCl$_3$, 100.6 MHz): δ 170.56 (C=O), 170.36 (C=O), 170.09 (C=O), 169.84 (C=O), 168.79 (C=O), 139.93, 139.85, 138.89, 138.62, 134.93, 134.82, 129.28, 128.78, 128.66, 128.59, 128.51, 128.07, 127.83, 127.79, 124.19, 100.47, 99.95, 98.32, 80.42, 78.90, 75.97, 75.81, 75.34, 75.26, 74.18, 73.56, 73.16, 72.94, 71.69, 71.30, 69.58, 68.53, 67.63, 67.18, 61.21, 57.25, 56.97, 21.90 (Ac), 21.87 (Ac), 21.67 (Ac), 20.83 (Ac), 16.89 (CH$_3$); Anal Calcd for C$_{63}$H$_{68}$NO$_{20}$: C, 65.27; H, 5.91; N, 1.21. Found: C, 64.57; H, 5.94; N, 1.11.

*Phenyl (2,3,4,6-tetra-*O*-acetyl-β-*D*-galactopyranosyl)-(1→4)-[(2,3,4-tri-*O*-acetyl-α-*L*-*

*fucopyranosyl)-(1 → 3)]-6-*O*-acetyl-2-deoxy-2-phthalimido-1-thio-β-*D*-glucopyranoside* (**12**). —A soln of compound **8** (310 mg, 0.32 mmol), and 10% Pd–C (320 mg) in a mixture of MeOH (5 mL) and CH_2Cl_2 (5 mL) was stirred overnight at rt under a H_2 atmosphere. The catalyst was filtered off, and the filtrate was concd under reduced pressure to give crude **9**, which was then treated with 1:1 Ac_2O–pyridine (6 mL) in the presence of a catalytic amount of DMAP (3 mg) at rt overnight. The reaction mixture was then concd. The crude product was applied to a column of silica gel and eluted with 1:2 hexane–EtOAc to give pure compound **10** (250 mg, 0.21 mmol, 96%) in two steps. To a cold (0 °C) soln of compound **10** (320 mg, 0.33 mmol) in Ac_2O (3.6 mL), was added Ac_2O (3.6 mL) and concd H_2SO_4 (80 μL). The reaction mixture was stirred for 16–20 h at the same temperature, then neutralized with satd $NaHCO_3$ soln and concd to give a crude residue, which was extracted with CH_2Cl_2 (30 mL). The organic extract was washed with satd NaCl soln (3 × 100 mL), dried over anhyd Na_2SO_4 and concd to give a crude product. BF_3Et_2O (120 μL) was added to a soln of the crude product and thiophenol (0.3 mL) in dry CH_2Cl_2 (6 mL), and the mixture was stirred for 16 h at rt under a N_2 atmosphere, then neutralized with Et_3N and concd to a mixture, which was applied to a column of silica gel and eluted with 1:1 hexane–EtOAc to give pure compound **12** (260 mg, 67%) as an amorphous solid. $[\alpha]_D$ $-38.8°$ (*c* 0.5, $CHCl_3$); ^1H NMR ($CDCl_3$, 600 MHz): δ 7.75–7.00 (m 9 H, ArH), 5.55–5.40 (m, 2 H, H-1$^{\mathrm{I}}$, $J_{1,2}$ 7.8 Hz, H-4$^{\mathrm{II}}$, *J* 2.8 Hz), 5.40–5.30 (d, 1 H, *J* 3.2 Hz, H-4$^{\mathrm{III}}$), 5.29–5.20 (m, 2 H, H-3$^{\mathrm{III}}$, H-2$^{\mathrm{II}}$), 5.19–5.15 (dd, 1 H, H-3$^{\mathrm{II}}$), 5.15–5.10 (dd, 1 H, H-5$^{\mathrm{III}}$), 4.95–4.90 (d, 1 H, *J* 3.1 Hz, H-1$^{\mathrm{III}}$), 4.85–4.80 (dd, 1 H, H-2$^{\mathrm{III}}$), 4.75–4.65 (m, 2 H, H-3$^{\mathrm{I}}$, H-6b$^{\mathrm{I}}$), 4.60–4.50 (m, 2 H, H-1$^{\mathrm{II}}$, H-6b$^{\mathrm{II}}$), 4.40–4.30 (m, 2 H, H-2$^{\mathrm{I}}$, H-6b$^{\mathrm{I}}$), 4.20–4.19 (dd, 1 H, H-6a$^{\mathrm{I}}$), 4.00–3.90 (m, 2 H, H-4$^{\mathrm{I}}$, H-5$^{\mathrm{II}}$), 3.80–3.70 (dd, 1 H, H-5$^{\mathrm{I}}$), 2.30 (s, 3 H, Ac), 2.29 (s, 3 H, Ac), 2.27 (s, 3 H, Ac), 1.99 (s, 3 H, Ac), 1.20 (d, 3 H, *J* 6.4 Hz, CH_3^{III}); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 170.90 (C=O), 170.86 (C=O), 170.80 (C=O), 170.69 (C=O), 170.35 (C=O), 170.20 (C=O), 169.90 (C=O), 169.15 (C=O), 134.75, 133.0, 132, 129.00, 128.25, 124.00, 101.00, 95.50, 84.30, 77.30, 75.50, 72.55, 71.89, 71.45, 71.40, 69.00, 68.30, 67.88, 66.89, 64.50, 62.20, 61.00, 56.50, 21.60 (Ac), 21.20 (Ac), 20.98 (Ac), 20.88 (Ac), 20.84 (Ac), 20.81(Ac), 20.70 (2 Ac), 16.45 (CH_3); Anal Calcd for $C_{48}H_{55}NO_{23}S$: C, 55.10; H, 5.30; N, 1.34, S, 3.07. Found: C, 55.38; H, 5.29; N, 1.26, S, 2.94.

*Phenyl (2,3,4-tri-*O*-acetyl-6-*O*-trimethylacetyl-β-*D*-galactopyranosyl)-(1 → 4)-2-deoxy-6-*O*-naphthymethyl-2-phthalimido-1-thio-β-*D*-glucopyranoside* (**15**).—A soln of compound **13** (4.74 g, 8.73 mmol), compound **14** (6.83 g, 17.5 mmol), and $SnCl_2$ (4.97 g, 26.2 mmol) in dry 5:1 CH_2Cl_2–toluene (100 mL) containing 4 Å MS (25 g) was stirred at -15 °C for 1.5–2 h under a N_2 atmosphere. Silver triflate (6.73 g, 26.2 mmol) was added, and the reaction mixture was stirred overnight at -15 to 0 °C under a N_2 atmosphere, then neutralized with Et_3N. The solids were filtered off, the organic layer was washed with satd $NaHCO_3$ soln, water, and then dried (Na_2SO_4). The solvent was evaporated under reduced pressure to give a crude mixture that was applied to a column of silica gel and eluted with 1:1 hexane–EtOAc to give pure **15** (5.97 g, 75%) as an amorphous solid. R_f 0.54 (1:1 hexane–EtOAc); $[\alpha]_D$ $+35.0°$ (*c* 0.5, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 7.86–7.80 (m, 6 H, ArH), 7.68–7.64 (m, 2 H, ArH), 7.52–7.40 (m, 5 H, ArH), 7.28–7.16 (m, 3 H, ArH), 5.65–5.63 (d, 1 H, $J_{1,2}$ 10.4 Hz, H-1$^{\mathrm{I}}$), 5.32–5.31 (d, 1 H, *J* 2.0 Hz, H-4$^{\mathrm{II}}$), 5.20–5.16 (t, 1 H, *J* 10.0, 8.8 Hz, H-2$^{\mathrm{II}}$), 4.94–4.90 (dd, 1 H, H-3$^{\mathrm{II}}$), 4.89–4.86 (d, 1 H, *J* 12.0 Hz, $OCH_AC_{10}H_7$, ABq), 4.72–4.69 (d, 1 H, *J* 12.0 Hz, $OCH_BC_{10}H_7$, ABq), 4.53–4.51 (d, 1 H, *J* 8.0 Hz), 4.47–4.42 (t, 1 H), 4.05–4.03 (m, 4 H), 3.87–3.71 (m, 4 H), 2.14–2.10 (s, 3 H, Ac), 1.96 (s, 3 H, Ac), 1.94 (s, 3 H, 2 Ac), 1.12 (s, 9 H, *t*-Bu); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 177.90 (C=O), 169.93 (C=O), 169.83 (C=O), 169.20 (C=O), 168.30 (C=O), 16.84 (C=O), 135.63, 134.14, 133.09, 132.64, 128.91, 128.39, 127.94, 127.81, 126.71, 126.34, 126.09, 125.95, 123.60, 123.28, 100.42, 83.57, 81.51, 78.30, 73.82, 71.43, 70.79, 70.78, 68.80, 67.99, 66.69, 61.82, 55.17, 38.67, 27.09 (3 CH_3), 20.63 (Ac), 20.52 (Ac), 20.33 (Ac); Anal Calcd for $C_{48}H_{51}NO_{15}S$: C, 63.08; H, 5.62; N, 1.53. Found: C, 62.90; H, 6.06; N, 1.98.

Phenyl (2,3,4-tri-O-acetyl-6-O-trimethylacetyl-β-D-galactopyranosyl)-(1 → 4)-[(2,3,4-tri-O-benzyl-α-L-fucopyranosyl)-(1 → 3)]-2-deoxy-6-O-naphthymethyl-2-phthalimido-1-thio-β-D-glucopyranoside (**16**).—A soln of compound **15** (5.2 g, 5.7 mmol), methyl 2,3,4-tri-*O*-benzyl-β-L-fucopyranoside (**3**, 10.63 g, 22.8 mmol), and *n*-Bu_4NBr (7.36 g, 22.8 mmol) in dry 5:1 1,2-dichloroethane–DMF (30 mL) containing 4 Å MS (25 g) was stirred for 2 h at rt under a N_2 atmosphere. Copper bromide (1.25 g, 5.6 mmol) was then added, and the mixture was stirred for 16–24 h at the same temperature. The solids were filtered off, the organic layer washed with satd $NaHCO_3$ soln, and water, dried (Na_2SO_4) and concd. The crude product was applied to a column of silica gel and eluted with 2:1 hexane–EtOAc to give compound **16** (87%) as a white solid. R_f 0.17 (2:1 hexane–EtOAc); $[\alpha]_D$ + 10.2° (*c* 0.54, $CHCl_3$); ^1H NMR ($CDCl_3$, 600 MHz): δ 7.93–7.87 (m, 4 H, ArH), 7.70–7.60 (m, 3 H, ArH), 7.56–7.44 (m, 4 H, ArH), 7.40–7.36 (m, 3 H, ArH), 7.28–7.16 (m, 14 H, ArH), 6.98–6.92 (m, 3 H, ArH), 5.50–5.45 (d, 1 H, *J* 7.8 Hz, H-1$^{\mathrm{I}}$), 5.24–5.18 (d, 1 H, *J* 2.8 Hz, H-4$^{\mathrm{III}}$), 5.10–5.00 (dd, 1 H, H-2$^{\mathrm{II}}$), 4.97–4.92 (d, 1 H, J_{gem} 11.6 Hz, OCHAr, ABq), 4.88–4.86 (dd, 1 H, *J* 3.6, 10.4 Hz, H-3$^{\mathrm{II}}$), 4.82–4.80 (m, 2 H, OCHAr, H-1$^{\mathrm{III}}$, $J_{1,2}$ 3.3 Hz), 4.80–4.78 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1$^{\mathrm{II}}$), 4.74–4.70 (m, 2 H, H-3$^{\mathrm{I}}$, OCHAr, ABq), 4.68–4.60 (m, 2 H, H-5$^{\mathrm{III}}$, J_{gem} 11.8 Hz, OCHAr, ABq), 4.40–4.32 (t, 1 H, *J* 10.4 Hz, H-2$^{\mathrm{I}}$), 4.32–4.20 (dd, 4 H, 2 OCH_2Ar), 4.19–4.16 (t, 1 H, *J* 9.2 Hz, H-4$^{\mathrm{I}}$), 4.14–4.00 (dd, 1 H, H-6b$^{\mathrm{II}}$), 4.00–3.84 (m, 4 H, H-6a$^{\mathrm{II}}$, H-6b$^{\mathrm{I}}$, H-6a$^{\mathrm{I}}$, H-3$^{\mathrm{III}}$), 3.80 (dd, 1 H, H-2$^{\mathrm{III}}$), 3.76–3.60 (m, 3 H, H-4$^{\mathrm{III}}$, H-5$^{\mathrm{I}}$, H-5$^{\mathrm{II}}$), 1.94 (s, 3 H, Ac), 1.92 (s, 6 H, 2 Ac), 1.20 (d, 3 H, *J* 6.4 Hz, CH_3^{III}), 1.16 (s, 9 H, *t*-Bu); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 170.09 (C=O), 169.94 (C=O), 168.89 (C=O), 138.84, 138.35, 135.49, 134.42, 133.43, 133.25, 132.65, 131.89, 129.04, 128.70, 128.38, 128.34, 128.27, 128.19, 128.15, 128.09, 128.03, 127.99, 127.61, 127.42, 127.34, 127.17, 126.80, 126.49, 126.21, 126.01, 123.84, 99.77, 97.85, 84.53, 79.95, 79.77, 75.29, 74.75, 74.40, 73.88, 73.83, 73.19, 72.52, 71.20, 70.60, 69.20, 68.05, 66.90, 66.74, 60.30, 55.76, 20.80 (3 Ac), 20.67 (Ac), 16.89 (CH_3); Anal Calcd for $C_{75}H_{79}NO_{19}S \cdot H_2O$: C, 66.80; H, 5.90; N, 1.04, S, 2.38. Found: C, 66.33; H, 6.02; N, 0.70, S, 2.24.

2,3,4-Tri-O-acetyl-6-O-benzyl-α-D-galactopyranosyl bromide (**19**).—Compound **18** was obtained in quantitative yield by treatment of compound **17** with BnBr in the presence of powdered KOH and 18-crown-6 at rt for 4–5 h in dry THF. The removal of the *O*-isopropylidene group was achieved by treatment of compound **18** with 1 N HCl in refluxing 1,4-dioxane, and the product was acetylated with 1:1 pyridine–Ac_2O and followed by treatment of the acetylated compound with $BiBr_3$ as described in the literature [17]. After purification by silica gel chromatography (2:1 hexane–EtOAc), compound **19** was obtained in 59% yield. R_f 0.45 (2:1 hexane–EtOAc); ^1H NMR ($CDCl_3$, 400 MHz): δ 7.42–7.26 (m, 5 H, ArH), 6.76–6.72 (d, 1 H, H-1), 5.58–5.52 (d, 1 H, *J* 2.8 Hz, H-4), 5.44–5.36 (dd, 1 H, H-2), 5.10–5.00 (dd, 1 H, H-3), 4.56–4.50 (d, 1 H, *J* 12.6 Hz, OCHPh, ABq), 4.50–4.40 (d, 1 H, OCHPh), 3.50–3.40 (m, 3 H, H-6a, H-6b), 2.05 (s, 3 H, Ac), 2.03 (s, 3 H, Ac), 1.99 (s, 3 H, Ac); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 170.30 (C=O), 170.10 (C=O), 169.59 (C=O), 137.44, 128.73, 128.21, 108.51, 73.86, 72.65, 70.46, 69.31, 67.29, 67.05, 20.90 (Ac), 20.81 (Ac), 20.77 (Ac).

Methyl 2-acetamido-2-deoxy-4,6-O-(p-methoxybenzylidene)-α-D-galactopyranoside (**20**).—*p*-Toluenesulfonic acid monohydrate (769 mg) was added to a soln of methyl 2-acetamido-2-deoxy-α-D-galactopyranoside (3.35 g, 14.3 mol) and α,α-dimethoxy(*p*-methyloxy)toluene (3.90 g, 21.39 mmol) in dry MeCN (105 mL). The mixture was stirred overnight at rt and quenched with Et_3N, and concd under reduced pressure to give a crude product, which was then applied to a column of silica gel and eluted with 30:1 CH_2Cl_2–MeOH to give pure compound **20** (4.40 g, 87%) as a white solid. R_f 0.39 (1:1 hexane–EtOAc); ^1H NMR ($CDCl_3$, 400 MHz): δ 7.46–7.43 (d, 2 H, *J* 8.4 Hz, ArH), 6.91–6.89 (d, 2 H, *J* 8.0 Hz, ArH), 5.74–5.72 (d, 1 H, *J* 8.0 Hz, NHAc), 5.53 (s, 1 H, PhCHO_2), 4.84–

4.83 (d, 1 H, *J* 2.8 Hz, H-1), 4.48–4.43 (m, 1 H), 4.28–4.19 (m, 3 H), 4.07–4.04 (d, 1 H), 3.81 (s, 3 H, OCH_3), 3.64 (s, 1 H), 3.40 (s, 3 H, OCH_3), 1.98 (s, 3 H, Ac);); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 160.91 (C=O), 130.34, 127.93, 113.87, 101.49, 99.67, 76.92, 69.54, 69.35, 63.05, 55.69, 55.56, 50.66, 23.67 (Ac).

*Methyl (2,3,4-tri-*O*-acetyl-6-*O*-benzyl-β-D-galactopyranosyl)-(1→3)-2-acetamido-2-deoxy-α-D-galactopyranoside* (**21**).—To a mixture of compound **20** (2.20 g, 6.23 mmol), powered $Hg(CN)_2$ (7.6 g) and $HgBr_2$ (760 mg) in dry CH_2Cl_2 (60 mL) containing 4 Å MS (20 g) was added 2,3,4-tri-*O*-acetyl-6-*O*-benzyl-α-D-galactopyranosyl bromide **19** (4.27 g, 9.30 mmol), and the mixture was stirred overnight at rt. The reaction mixture was diluted with benzene and washed with satd aq $NaHCO_3$, 10% KI soln, and water. The organic layer was dried with Na_2SO_4 and concd. The crude residue was applied to a column of silica gel and eluted with 20:1 CH_2Cl_2–MeOH to give compound **21a** (2.3 g, 51%) as an amorphous solid. ^{1}H NMR ($CDCl_3$, 400 MHz): δ 7.48–7.42 (d, 2 H, *J* 8.7 Hz, ArH), 7.40–7.28 (d, 5 H, ArH), 6.96–6.88 (d, 2 H, *J* 8.6 Hz, ArH), 5.70–5.60 (d, 1 H, *J* 7.8 Hz, NHAc), 5.47 (s, 1 H, PhC*H*O_2), 5.42–5.40 (d, 1 H, *J* 4.4 Hz, H-4$^{\mathrm{II}}$), 5.20–5.10 (dd, 1 H, H-2$^{\mathrm{II}}$), 5.05–4.95 (dd, 1 H, H-3$^{\mathrm{II}}$), 4.80–4.70 (d, 1 H, *J* 4.0 Hz, H-1$^{\mathrm{I}}$), 4.70–4.60 (d, 1 H, $J_{1,2}$ 8.0 Hz, H-1$^{\mathrm{II}}$), 4.56–4.44 (dd, 2 H, OCH_2Ph, AB), 4.30–4.20 (d, 1 H), 4.20–4.10 (dd, 1 H), 4.00–3.85 (m, 3 H), 3.85–3.75 (m, 4 H, OCH_3), 3.65–3.55 (dd, 1 H), 3.55–3.50 (m, 2 H), 3.42 (s, 3 H, OCH_3), 2.08 (s, 3 H, Ac), 2.02 (s, 3 H, Ac), 1.95 (s, 3 H, Ac), 1.94 (s, 3 H, Ac); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 171.12 (C=O), 171.10 (C=O), 170.40 (C=O), 161.50 (C=O), 131.80, 129.50, 128.83, 128.80, 128.40, 114.50, 103.18, 101.72, 101.51, 76.71, 76.46, 74.44, 73.27, 72.22, 70.06, 69.82, 69.02, 68.60, 63.85, 56.22, 48.99, 24.17 (NAc), 21.39 (Ac), 21.33 (Ac), 21.13 (Ac); Anal Calcd for $C_{36}H_{45}NO_{15}$: C, 59.07; H, 6.20; N, 1.92. Found: C, 58.97; H, 6.19; N, 1.61.

Compound **21a** was treated with 60% HOAc at 60–65 °C for 1.5 h and purified by silica gel column to give pure compound **21** (76%) as an amorphous solid. R_f 0.47 (20:1 CH_2Cl_2–MeOH); $[\alpha]_D$ + 5.7° (*c*, 0.1, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 7.60–7.20 (m, 5 H, ArH), 5.60–5.52 (m, 1 H, *J* 8.6 Hz, NHAc), 5.44–5.40 (d, 1 H, *J* 3.0 Hz), 5.20–5.12 (dd, 1 H), 5.04–4.96 (dd, 1 H), 4.80–4.72 (d, 1 H, *J* 3.2 Hz, H-1$^{\mathrm{I}}$), 4.60–4.56 (d, 1 H, *J* 7.8 Hz, H-1$^{\mathrm{II}}$), 4.54–4.48 (d, 1 H, J_{gem} 12.6 Hz, OCHPh, ABq), 4.48–4.40 (d, 1 H, J_{gem} 12.4 Hz, OCHPh, ABq), 4.10 (s, 1 H), 4.00–3.80 (m, 3 H), 3.80–3.70 (m, 4 H), 3.60–3.45 (m, 3 H), 3.44 (s, 3 H, OCH_3), 2.08 (s, 3 H, Ac), 2.06 (s, 3 H, Ac), 1.99 (s, 3 H, Ac), 1.98 (s, 3 H, Ac); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 170.50 (C=O), 170.30 (C=O), 169.70 (C=O), 128.75, 128.25, 128.00, 101.75, 99.30, 74.10, 72.85, 71.00, 69.83 (2 C), 69.21, 68.20, 67.85, 63.00, 55.21, 48.00, 23.60 (NAc), 20.90 (Ac), 20.83 (Ac), 20.75 (Ac); Anal Calcd for $C_{28}H_{39}NO_{14}$: C, 54.81; H, 6.41; N, 2.28. Found: C, 54.28; H, 6.19; N, 1.98.

*Methyl [(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-[(2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl)-(1→3)]-6-*O*-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl)-(1→6)-[(2,3,4-tri-*O*-acetyl-6-*O*-benzyl-β-D-galactopyranosyl)]-(1→3)-2-acetamido-4-*O*-acetyl-2-deoxy-α-D-galactopyranoside* (**23**).—A soln of trisaccharide donor **12** (264 mg, 0.21 mmol), disaccharide acceptor **21** (233 mg, 0.19 mmol), and *N*-iodosuccinimide (NIS, 126 mg, 0.62 mmol) in dry CH_2Cl_2 (12 mL) containing 4 Å MS (8 g) was stirred for 2 h at − 65 to − 60 °C under a N_2 atmosphere. Trifluoromethanesulfonic acid (TfOH) (20 μL) in dry CH_2Cl_2 (2 mL) was then added dropwise, and the mixture was stirred for 2 h at the same temperature. The mixture was then neutralized with satd aq $NaHCO_3$ soln. The solids were filtered off, the organic layer was washed with satd $NaHCO_3$ soln, 10% $Na_2S_2O_3$, and water, and then it was dried (Na_2SO_4) and concd. The crude mixture was applied to a column of silica gel and eluted with 30:1 CH_2Cl_2–MeOH to give pure compound **22** (200 mg, 41%) as a glassy white solid. Compound **22** was further acetylated with 1:1 pyridine–Ac_2O (10 mL) in the presence of a catalytic amount of DMAP (5 mg). The soln was concd and the crude product was purified by silica gel column chromatography (30:1 CH_2Cl_2–MeOH) to give compound **23** in quantitative yield as an amorphous solid. R_f 0.46 (30:1 CH_2Cl_2–MeOH); ^{1}H NMR

(CDCl$_3$, 600 MHz): δ 7.88–7.68 (m, 4 H, ArH), 7.40–7.24 (m, 5 H, ArH), 5.53–5.50 (d, 1 H, *J* 9.2 Hz, NHAc), 5.47–5.46 (d, 1 H, *J* 3.2 Hz, H-4^{II}), 5.38–5.37 (d, 1 H, *J* 2.8 Hz, H-4^{IV}), 5.34–5.33 (d, 1 H, *J* 2.0 Hz, H-4^{V}), 5.17–5.08 (m, 3 H, H-4^{I}, H-3^{V}, H-2^{II}), 5.05–4.88 (m, 6 H, H-2^{IV}, H-1^{III}, H-3^{II}, H-5^{V}, H-1^{V}, H-3^{IV}), 4.81–4.75 (dd, 1 H, *J* 4.0, 11.0 Hz, H-2^{V}), 4.72–4.60 (m, 2 H, H-3^{III}, H-6b^{III}), 4.57–4.40 (m, 5 H, OCHPh, ABq, H-1^{II}, H-1^{IV}, H-6b^{II}, OCHPh, ABq), 4.32–4.16 (m, 5 H, H-1^{I}, H-6a^{II}, H-2^{I}, H-2^{III}, H-6a^{III}), 3.97–3.86 (m, 2 H, H-4^{III}, H-5^{II}), 3.84–4.71 (m, 4 H, H-6b^{I}, H-3^{I}, H-5^{I}, H-5^{IV}), 3.63–3.60 (dd, 1 H, H-5^{III}), 3.56–3.48 (dd, 1 H, H-6b^{IV}), 3.46–3.40 (dd, 1 H, H-6a^{IV}), 3.28–3.18 (dd, 1 H, H-6a^{I}), 2.89 (s, 3 H, OCH$_3$), 2.18 (s, 3 H, Ac), 2.17 (s, 9 H, 3 Ac), 2.10 (s, 3 H, Ac), 2.09 (s, 3 H, Ac), 2.05 (s, 6 H, 2 Ac), 2.03 (s, 3 H, Ac), 2.02 (s, 3 H, Ac), 1.97 (s, 3 H, Ac), 1.90 (s, 3 H, Ac), 1.89 (s, 3 H, Ac), 1.21 (d, 3 H, *J* 6.4 Hz, CH$_3^{V}$); ^{13}C NMR (CDCl$_3$, 100.6 MHz): δ 170.82 (C=O), 170.73 (C=O), 170.68 (C=O), 170.59 (C=O), 170.37 (C=O), 170.33 (C=O), 170.30 (C=O), 170.23 (C=O), 170.11 (C=O), 169.84 (C=O), 169.68 (C=O), 169.64 (C=O), 169.07 (C=O), 134.47, 128.66, 128.09, 127.99, 100.84, 100.80, 98.77, 98.29, 95.28, 75.36, 73.81, 73.76, 73.36, 72.85, 72.52, 71.54, 71.24, 71.06, 70.17, 69.92, 69.09, 68.51, 68.23, 67.99, 67.75, 67.50, 66.81, 64.41, 61.72, 60.87, 56.33, 54.70, 48.96, 31.02 (Ac), 23.46 (Ac), 20.96 (Ac), 20.92 (Ac), 20.87 (Ac), 20.80 (Ac), 20.76 (Ac), 20.64 (Ac), 15.99 (CH$_3$); Anal Calcd for $C_{72}H_{90}N_2O_{38}$: C, 54.34; H, 5.70; N, 1.76. Found: C, 54.49; H, 5.81; N, 1.64.

Methyl (2,3,4,6-tetra-O-*acetyl-β*-D-*galactopyranosyl)-(1→4)-[(2,3,4-tri*-O-*acetyl-α*-L-*fucopyranosyl)-(1→3)]-2-acetamido-6*-O-*acetyl-2-deoxy-β*-D-*glucopyranosyl)-(1→6)-[(2,3,4-tri*-O-*acetyl-6*-O-*benzyl-β*-D-*galactopyranosyl)-(1→3)]-2-acetamido-4*-O-*acetyl-2-deoxy-α*-D-*galactopyranoside* (**24**).—Compound **23** (80 mg, 0.05 mmol) in a mixture of MeOH (5 mL) and NH$_2$NH$_2$·H$_2$O (1 mL) was stirred for 4–5 h at 80–85 °C. The reaction mixture was concd under reduced pressure to give a crude mixture, which was co-evaporated with toluene and acetylated with 1:1 Ac$_2$O–pyridine (2 mL) with a catalytic amount of DMAP at rt overnight. The acetylated mixture was concd and passed through a short column of silica gel and eluted with 30:1 CH$_2$Cl$_2$–MeOH to give compound **24** (65 mg, 86%) as an amorphous solid. $[\alpha]_D$ −16.6° (*c* 0.2, CHCl$_3$); ^{1}H NMR (CDCl$_3$, 600 MHz): δ 7.40–7.25 (m, 5 H, ArH), 6.00–5.90 (d, 1 H, *J* 8.3 Hz, NHAc), 5.80–5.78 (d, 1 H, *J* 8.4 Hz, NHAc), 5.43–5.37 (m, 4 H, H-1^{III}, H-1^{V}, H-4^{II}, H-4^{IV}), 5.26–5.24 (d, 1 H, *J* 2.8 Hz, H-4^{I}), 5.22–5.18 (dd, 1 H, *J* 3.2, 11.2 Hz), 5.12–4.95 (m, 5 H, H-2^{IV}, H-2^{II}, H-2^{V}, H-3^{IV}, H-3^{II}), 4.95–4.88 (dd, 1 H, H-5^{V}), 4.74–4.73 (d, 1 H, $J_{1,2}$ 3.6 Hz, H-1^{I}), 4.59–4.42 (m, 7 H, H-1^{II}, H-6b^{IV}, OCH$_A$Ph, H-6b^{III}, H-1^{IV}, H-2^{I}, OCH$_B$Ph), 4.32–4.29 (dd, 1 H, H-6a^{III}), 4.14–4.10 (dd, 1 H, H-6a^{I}), 4.00–3.99 (dd, 1 H, H-3^{I}), 3.97–3.93 (m, 1 H, H-5^{I}), 3.87 (t, 1 H, H-5^{III}), 3.82–3.79 (m, 4 H, H-5^{II}, H-6b^{I}), 3.60–3.50 (m, 2 H, H-6b^{II}, H-5^{IV}), 3.50–3.40 (m, 2 H, H-6a^{II}, H-6a^{I}), 3.39 (s, 3 H, OCH$_3$), 2.19 (s, 3 H, Ac), 2.18 (s, 3 H, Ac), 2.15 (s, 3 H, Ac), 2.09 (s, 3 H, Ac), 2.08 (s, 3 H, Ac), 2.07 (s, 3 H, Ac), 2.06 (s, 3 H, Ac), 1.98 (s, 3 H, Ac), 1.97 (s, 3 H, Ac), 1.95 (s, 3 H, Ac), 1.94 (s, 3 H, Ac), 1.69 (s, 6 H, 2 Ac), 1.24–1.20 (d, 3 H, *J* 6.4 Hz, CH$_3^{V}$); ^{13}C NMR (CDCl$_3$, 100.6 MHz): δ 171.85 (C=O), 171.20 (C=O), 171.15 (C=O), 170.90 (C=O), 170.80 (C=O), 170.60 (C=O), 170.49 (C=O), 170.43 (C=O), 170.40 (C=O), 170.35 (C=O), 170.20 (C=O), 170.15 (C=O), 170.10 (C=O), 169.40 (C=O), 137.52, 128.83, 128.33, 128.25, 100.75 (2 C), 100.50, 98.90, 95.50, 74.85, 73.89, 73.25, 72.60, 72.20, 71.60, 71.40, 71.30, 71.20, 69.90, 69.25, 69.00, 68.60, 68.45, 68.25, 67.90, 67.60, 66.98, 64.50, 62.25, 61.00, 55.80, 49.85, 23.57 (NAc), 23.40 (NAc), 21.16 (Ac), 21.06 (Ac), 21.02 (2 Ac), 20.98 (Ac), 20.94 (Ac), 20.86 (2 Ac), 20.82 (2 Ac), 20.76 (Ac), 20.72 (Ac), 16.20 (CH$_3$); Anal Calcd for $C_{66}H_{90}N_2O_{37}$: C, 52.73; H, 6.03; N, 1.86. Found: C, 53.16; H, 5.30; N, 1.56.

Methyl (2,3,4,6-tetra-O-*acetyl-β*-D-*galactopyranosyl)-(1→4)-[(2,3,4-tri*-O-*acetyl-α*-L-*fucopyranosyl)-(1→3)]-2-acetamido-6*-O-*acetyl-2-deoxy-β*-D-*glucopyranosyl)-(1→6)-[(2,3,4-tri*-O-*acetyl-6*-O-*sulfo-β*-D-*galactopyranosyl)-(1→3)]-2-acetamido-4*-O-*acetyl-2-deoxy-α*-D-*galactopyranoside sodium salt* (**26**).—A soln of compound **24** (25 mg, 0.017 mmol) and 10% Pd–C (25 mg) in 1:4 MeOH–

CH_2Cl_2 (3 mL) was stirred for 2.5 h at rt under a H_2 atmosphere. The solids were filtered off, and the organic layer was concd. The crude residue was treated for 6–9 h at 0–25 °C with the SO_3·pyridine complex (10 mg) in pyridine (2 mL), then quenched with MeOH and treated with Amberlite IR 120 (Na^+) cation-exchange resin in MeOH at rt for 1.5 h. The filtrate was concd and purified by a short column of silica gel eluted with 10:1 CH_2Cl_2–MeOH to give pure compound **26** (25 mg, 97% in two steps) as an amorphous solid. ^1H NMR (CD_3OD, 600 MHz): δ 5.48–5.40 (m, 3 H), 5.40–5.30 (m, 2 H), 5.28–5.20 (dd, 1 H), 5.16–4.4.92 (m, 5 H), 4.75–4.70 (d, 1 H), 4.70–4.60 (m, 3 H), 4.48–4.32 (m, 2 H), 4.32–4.28 (m, 2 H), 4.24–4.04 (m, 4 H), 4.04–3.92 (m, 3 H), 3.92–3.60 (m, 6 H), 3.52–3.44 (m, 1 H), 3.38 (s, 3 H, OCH_3), 2.17 (s, 3 H, Ac), 2.16 (s, 3 H, Ac), 2.15 (s, 3 H, Ac), 2.14 (s, 3 H, Ac), 2.09 (s, 6 H, 2 Ac), 2.08 (2s, 6 H, 2 Ac), 2.04 (s, 3 H, Ac), 2.00 (s, 3 H, Ac), 1.96 (2s, 6 H, 2 Ac), 1.93 (s, 3 H, Ac), 1.90 (s, 3 H, Ac), 1.20 (d, 3 H, J 6.4 Hz, CH_3); ^{13}C NMR (CD_3OD, 100.6 MHz): δ 173.38 (C=O), 173.21 (C=O), 172.57 (C=O), 172.55 (C=O), 172.33 (C=O), 172.23 (C=O), 172.11 (C=O), 171.60 (C=O), 171.16 (C=O), 103.03, 102.36, 101.79, 100.14, 96.99, 76.66, 75.48, 74.43, 74.15, 73.01, 72.60, 72.42, 72.32, 72.26, 72.02, 70.60, 70.52, 70.34, 69.82, 69.63, 69.38, 68.81, 68.70, 66.07, 65.45, 63.56, 62.52, 56.87, 56.14, 55.69, 23.65 (NAc), 23.22 (NAc), 21.47 (Ac), 21.35 (Ac), 21.30 (Ac), 21.21 (Ac), 21.17 (Ac), 21.07 (Ac), 20.99 (Ac), 20.96 (Ac), 20.93 (Ac), 16.70 (CH_3).

Methyl (β-D-galactopyranosyl)-(1 → 4)-[(α-L-fucopyranosyl)-(1 → 3)]-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1 → 6)-[(6-O-sulfo-β-D-galactopyranosyl)-(1 → 3)]-2-acetamido-2-deoxy-α-D-galactopyranoside sodium salt (**1**). —Sodium methoxide (80 μL of a 1 M soln) was added to a soln of compound **26** (25 mg, 0.017 mmol) in 1:1 MeOH–water (3 mL). The reaction mixture was stirred for 24 h at rt and concd. The crude product was applied to a short column of silica gel and eluted with 3:1:1 n-C_3H_7OH–HOAc–water to give pure compound **1** (8 mg, 48%) as an amorphous solid. R_f 0.26 (3:1:1 n-C_3H_7OH–HOAc–water); ^1H NMR (D_2O, 600 MHz): δ 5.16–5.08 (d, 1 H, $J_{1,2}$ 3.0 Hz, H-1^{V}), 4.88–4.78 (dd, 1 H, H-5^{V}), 4.78–4.60 (d, 1 H, $J_{1,2}$ 3.0 Hz, H-1^{I}), 4.60–4.52 (d, 1 H, $J_{1,2}$ 8.8 Hz, H-1^{II}), 4.52–4.46 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1^{IV}), 4.46–4.44 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1^{II}), 4.36–4.28 (dd, 1 H, H-2^{I}), 4.28–4.24 (d, J 2.8 Hz, H-4^{I}), 4.22–4.18 (d, 2 H), 4.14–3.84 (m, 11 H, H-5^{I}, H-4^{III}, H-4^{II}, H-2^{III}, H-4^{IV}, H-3^{III}), 3.84–3.46 (m, 12 H, H-4^{V}, H-6a^{I}, H-3^{IV}, H-3^{II}, H-5^{III}, H-2^{IV}, H-2^{II}), 3.36 (s, 3 H, OCH_3), 2.13 (s, 3 H, Ac), 2.02 (s, 3 H, Ac), 1.20 (d, 3 H, J 6.4 Hz, CH_3^{V}); ^{13}C NMR (D_2O, 100.6 MHz): δ 173.50 (C=O), 173.20 (C=O), 103.40, 100.45, 100.22, 97.45, 97.20, 76.48, 74.45, 73.90, 73.83, 72.80, 71.55, 71.53, 71.50, 70.98, 70.00, 69.53 (2 C), 68.54, 68.52, 67.90, 67.40, 67.30, 66.75, 66.25, 65.63, 60.50, 58.90, 54.98, 54.00, 47.50, 21.25 (Ac), 21.00 (Ac), 14.25 (CH_3^{V}); FABMS (m/z) (positive ion mode): Calcd for $C_{35}H_{59}N_2NaO_{28}S$: 1012; found: 1034.0 [M^+ + Na], 1012 [M^+].

Methyl (2,3,4-tri-O-acetyl-6-O-trimethylacetyl-β-D-galactopyranosyl)-(1 → 4)-[(2,3,4-tri-O-benzyl-α-L-fucopyranosyl)-(1 → 3)]-2-deoxy-6-O-naphthylmethyl-2-phthalimido-β-D-glucopyranosyl)-(1 → 6)-[(2,3,4-tri-O-benzoyl-β-D-galactopyranosyl)-(1 → 3)]-2-acetamido-2-deoxy-α-D-galactopyranoside (**29**).—A soln of trisaccharide donor **16** (722 mg, 0.54 mmol), disaccharide acceptor **27** (460 mg, 0.50 mmol), and N-iodosuccinimide (NIS, 365 mg, 0.62 mmol) in dry CH_2Cl_2 (12 mL) containing 4 Å MS (10 g) was stirred for 2 h at − 65 to − 70 °C under a N_2 atmosphere. Trifluoromethanesulfonic acid (TfOH) (60 μL) in dry CH_2Cl_2 (2 mL) was then added dropwise, and the mixture was stirred for 2 h at the same temperature. The mixture was neutralized with satd aq $NaHCO_3$. The solids were filtered off, and the organic layer was washed with satd $NaHCO_3$ soln, 10% $Na_2S_2O_3$, and water. The organic extract was dried (Na_2SO_4) and concd under reduced pressure. The crude mixture was applied to a column of silica gel and eluted with 1:1 hexane–EtOAc to give pure compound **28** (490 mg, 46%) as an amorphous solid. Compound **28** (490 mg, 0.23 mmol), 2,6-lutidine (10 μL) and thiourea (52 mg) in 1:1 EtOH–CH_2Cl_2 (30 mL) were stirred overnight at 60–65 °C. The reaction mixture was then concd to give a crude product, which was applied to a column of silica gel and eluted with 1:3 hexane–EtOAc to give pure compound **29** (390 mg, 84%) as an amorphous solid. R_f 0.47 (1:3 hexane — EtOAc); $[\alpha]_D$ +70.8° (*c* 0.6, CHCl$_3$); ^{1}H NMR (CDCl$_3$, 600 MHz): δ 8.10–7.82 (m, 10 H, ArH), 7.80–7.77 (m, 3 H, ArH), 7.60–7.50 (m, 2 H, ArH), 7.50–7.30 (m, 10 H, ArH), 7.30–7.10 (m, 15 H, ArH), 7.00–6.90 (m, 1 H, ArH), 5.75–5.15 (m, 2 H, H-4^{II}, H-2^{II}), 5.60–5.50 (dd, 1 H), 5.30–5.20 (d, 1 H), 5.15–5.05 (m, 2 H, H-4^{V}, H-1^{III}, $J_{1,2}$ 8.6 Hz), 5.01–4.98 (dd, 1 H, H-2^{IV}), 4.96–4.92 (d, 2 H, J_{gem} 12.6 Hz, 2 OCHAr), 4.90–4.84 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1^{II}), 4.84–4.75 (m, 3 H, OCHAr, H-1^{I}, H-3^{IV}), 4.75–4.64 (m, 3 H, H-1^{IV}, H-3^{III}, OCHAr), 4.64–4.60 (m, 2 H, H-5^{V}), 4.60–4.52 (d, 2 H, J_{gem} 12.4 Hz, 2 OCHPh), 4.40–4.32 (m, 2 H, H-2^{III}), 4.32–4.24 (dd, 2 H, OCH$_2$Ph), 4.20–4.12 (t, 1 H, H-4^{III}), 4.12–4.04 (m, 2 H), 4.04–3.96 (t, 1 H, H-5^{II}), 3.92–3.84 (m, 2 H, H-3^{I}), 3.84–3.76 (m, 1 H), 3.76–3.69 (dd, 1 H, H-2^{I}), 3.69–3.48 (m, 7 H, H-5^{III}), 3.12–3.04 (t, 1 H), 2.90–2.84 (t, 1 H), 2.84 (s, 3 H, OCH$_3$), 1.92 (s, 3 H, Ac), 1.84 (s, 3 H, Ac), 1.80 (s, 3 H, Ac), 1.79 (s, 3 H, Ac), 1.20 (d, 3 H, J 6.4 Hz, CH$_3^{V}$), 1.12 (s, 9 H, *t*-Bu); ^{13}C NMR (CDCl$_3$, 100.6 MHz): δ 177.85 (C=O), 170.20 (C=O), 170.00 (C=O), 169.90 (C=O), 168.90 (C=O), 166.85 (C=O), 165.78 (C=O), 165.00 (C=O), 138.85, 138.45, 135.10, 134.23, 134.00, 133.60, 133.48, 133.41, 133.25, 130.27, 130.00, 129.86, 128.89, 128.76, 128.47, 128.40, 128.30, 128.24, 128.18, 128.16, 120.05, 127.82, 127.60, 127.36, 127.25, 126.60, 126.30, 126.04, 123.60, 102.30, 99.86, 99.00, 98.50, 97.80, 80.00, 79.86, 75.66, 75.45, 75.20, 75.00, 74.50, 74.20, 73.21, 72.55, 71.89, 71.40, 70.50, 70.34, 69.30, 69.20, 68.80, 68.50, 68.00, 67.78, 67.00, 66.70, 61.00, 60.30, 56.50, 54.80, 48.00, 38.85, 27.50, 22.50 (Ac), 20.58 (3 Ac), 16.85 (CH$_3$); Anal Calcd for C$_{105}$H$_{110}$N$_2$O$_{33}$: C, 65.60; H, 5.65; N, 1.43. Found: C, 65.35; H, 5.81; N, 1.64.

*Methyl (2,3,4-tri-*O*-acetyl-6-*O*-trimethylacetyl-β-*D*-galactopyranosyl)-(1→4)-[(2,3,4-tri-*O*-benzyl-α-*L*-fucopyranosyl)-(1→3)]-2-deoxy-6-*O*-naphthylmethyl-2-phthalimido-β-*D*-glucopyranosyl)-(1→6)-[(2,3,4-tri-*O*-benzoyl-6-*O*-sulfo-β-*D*-galactopyranosyl)-(1→3)]-2-acetamido-2-deoxy-α-*D*-galactopyranoside sodium salt* (**30**).—Sulfur trioxide–pyridine (35 mg, 0.23 mmol) was added to a cold (ice bath) soln of compound **29** (300 mg, 0.15 mmol) in dry pyridine (2 mL). The mixture was stirred for 18–24 h at the same temperature then quenched with MeOH and treated in MeOH with Amberlite IR 120 (Na$^+$) cation-exchange resin. The filtrate was concd to give a crude residue, which was applied to a short column of silica gel and eluted with 30:1 CH$_2$Cl$_2$–MeOH to give pure compound **30** (260 mg, 82%) as an amorphous solid. $[\alpha]_D$ +38° (*c* 0.2, CHCl$_3$); ^{1}H NMR (CD$_3$OD, 600 MHz): δ 8.15–7.86 (m, 10 H, ArH), 7.90–7.78 (m, 2 H, ArH), 7.60–7.50 (m, 3 H, ArH), 7.50–7.40 (m, 10 H, ArH), 7.30–7.15 (m, 14 H, ArH), 7.00–6.90 (m, 2 H, ArH), 5.33–5.26 (m, 2 H, H-4^{II}, H-2^{II}), 5.70–5.55 (dd, 1 H), 5.25–5.20 (d, 1 H), 5.15–5.08 (m, 2 H, H-4^{IV}, H-1^{III}, $J_{1,2}$ 8.6 Hz), 5.05–4.98 (dd, 1 H, H-2^{IV}), 4.96–4.94 (d, 1 H, J_{gem} 12.6 Hz, OCHAr), 4.90–4.84 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1^{II}), 4.84–4.77 (m, 4 H, 2 OCHAr, H-1^{I}, H-3^{IV}), 4.76–4.68 (m, 3 H, H-1^{IV}, H-3^{III}, OCHAr), 4.64–4.60 (m, 2 H, H-5^{V}), 4.60–4.52 (d, 2 H, J_{gem} 12.4 Hz, 2 OCHPh), 4.48–4.32 (m, 2 H, H-2^{III}), 4.32–4.24 (dd, 2 H, OCH$_2$Ph), 4.20–4.12 (t, 1 H, H-4^{V}), 4.12–4.04 (m, 2 H), 4.04–3.96 (t, 1 H), 3.92–3.84 (m, 2 H, H-3^{I}), 3.84–3.76 (m, 1 H), 3.76–3.69 (dd, 1 H, H-2^{I}), 3.69–3.48 (m, 7 H, H-5^{III}), 3.12–3.04 (m, 1 H), 2.90–2.84 (m, 1 H), 2.84 (s, 3 H, OCH$_3$), 1.92 (s, 3 H, Ac), 1.88 (s, 3 H, Ac), 1.87 (s, 3 H, Ac), 1.80 (s, 3 H, Ac), 1.21 (d, 3 H, J 6.4 Hz, CH$_3^{V}$), 1.12 (s, 9 H, *t*-Bu); ^{13}C NMR (CDCl$_3$, 100.6 MHz): δ 177.86 (C=O), 170.26 (C=O), 170.05 (C=O), 169.98 (C=O), 168.93 (C=O), 166.88 (C=O), 165.79 (C=O), 165.00 (C=O), 138.95, 138.45, 135.10, 134.23, 134.08, 133.60, 133.48, 133.41, 133.26, 130.27, 130.05, 129.85, 128.86, 128.76, 128.37, 128.30, 128.28, 128.24, 128.15, 128.11, 120.05, 127.88, 127.65, 127.36, 127.35, 126.80, 126.37, 126.24, 123.60, 102.30, 99.86, 99.00, 98.50, 97.80, 80.00, 79.86, 75.66, 75.45, 75.20, 75.08, 74.50, 74.20, 73.21, 72.55, 71.89, 71.46, 70.55, 70.34, 69.34, 69.20, 68.85, 68.50, 68.00, 67.86, 67.80, 66.70, 61.06, 60.30, 56.50, 54.20, 48.25, 38.85, 27.50, 22.60 (Ac), 20.59 (3 Ac), 16.45 (CH$_3$).

*Methyl (2,3,4-tri-*O*-acetyl-6-*O*-trimethylacetyl-β-*D*-galactopyranosyl)-(1→4)-[(2,3,4-tri-*O*-benzyl-α-*L*-fucopyranosyl)-(1→3)]-2-*

deoxy-2-phthalimido-6-O-sulfo-β-D-glucopyranosyl)-(1 → 6)-[(2,3,4-tri-O-benzoyl-6-O-sulfo-β-D-galactopyranosyl)-(1 → 3)]-2-acetamido-2-deoxy-α-D-galactopyranoside disodium salt (**32**).—To a soln of compound **30** (235 mg, 0.12 mmol) in 4:1:trace CH_2Cl_2–MeOH–water (10 mL) was added DDQ (40 mg). After stirring for 3.5 h at rt, an additional portion of DDQ (20 mg) was added, and the mixture was stirred at the same temperature for a total of 16 h. The mixture was concd under reduced pressure to give a crude residue, which was dissolved in CH_2Cl_2 (100 mL) and washed with satd $NaHCO_3$ soln (3 × 100 mL). The organic extract was dried over Na_2SO_4 and concd. The crude product was applied to a column of silica gel and eluted with 20:1 CH_2Cl_2–MeOH to give pure compound **31** (120 mg, yield: 51%) as an amorphous solid. Compound **31** was treated with SO_3·pyridine for 12 h in dry pyridine at 0–5 °C, then purified by a short column of silica gel that was eluted with 15:1 CH_2Cl_2–MeOH to give pure compound **32** (79%) as an amorphous solid. $[\alpha]_D$ + 59° (*c* 0.5, $CHCl_3$); ^{1}H NMR (D_2O, 600 MHz): δ 8.10–7.82 (m, 10 H, ArH), 7.80–7.77 (m, 3 H, ArH), 7.70–7.59 (m, 2 H, ArH), 7.56–7.30 (m, 6 H, ArH), 7.30–7.15 (m, 12 H, ArH), 7.00–6.90 (m, 1 H, ArH), 5.70–5.65 (m, 2 H, H-4^{II}, H-2^{II}), 5.60–5.50 (dd, 1 H, H-3^{II}), 5.30–5.20 (d, 1 H), 5.15–5.05 (m, 2 H, H-4^{IV}, H-1^{III}, $J_{1,2}$ 8.6 Hz), 5.01–4.98 (dd, 1 H, H-2^{IV}), 4.96–4.92 (d, 1 H, J_{gem} 12.6 Hz, OCHAr), 4.90–4.84 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1^{II}), 4.84–4.75 (m, 3 H, OCHAr, H-1^{I}, H-3^{IV}), 4.75–4.64 (m, 3 H, H-1^{IV}, H-3^{III}, OCHAr), 4.64–4.60 (m, 2 H, H-5^{V}), 4.60–4.52 (d, 1 H, J_{gem} 12.4 Hz, OCHPh), 4.04–4.32 (m, 2 H, H-2^{III}), 4.32–4.24 (dd, 2 H, OCH_2Ph), 4.20–4.12 (t, 1 H, H-4^{III}), 4.12–4.04 (m, 2 H), 4.04–3.96 (t, H-5^{II}), 3.92–3.84 (m, 2 H, H-3^{I}), 3.84–3.76 (m, 1 H), 3.76–3.69 (dd, 1 H, H-2^{I}), 3.69–3.48 (m, 7 H, H-5^{III}), 3.12–3.04 (t, 1 H), 2.90–2.84 (t, 1 H), 2.84 (s, 3 H, OCH_3), 1.92 (s, 3 H, Ac), 1.84 (s, 3 H, Ac), 1.80 (s, 3 H, Ac), 1.79 (s, 3 H, Ac), 1.20 (d, 3 H, *J* 6.4 Hz, CH$_3^{V}$), 1.12 (s, 9 H, *t*-Bu); ^{13}C NMR ($CDCl_3$, 100.6 MHz): δ 176.85 (C=O), 170.25 (C=O), 170.00 (C=O), 169.93 (C=O), 168.91 (C=O), 166.85 (C=O), 165.78 (C=O), 165.01 (C=O), 135.13, 134.23, 134.00, 133.61, 133.48, 133.41, 133.25, 130.27, 130.01, 129.86, 128.89, 128.76, 128.47, 128.40, 128.32, 128.24, 128.18, 128.16, 128.05, 127.82, 127.62, 127.36, 127.25, 126.63, 126.31, 126.04, 123.61, 102.35, 99.86, 99.08, 98.52, 97.81, 80.08, 79.86, 75.66, 75.45, 75.21, 75.05, 74.52, 74.23, 73.21, 72.55, 71.89, 71.40, 70.50, 70.34, 69.35, 69.20, 68.83, 68.57, 68.08, 67.86, 67.81, 66.75, 61.08, 60.31, 56.56, 54.87, 48.50, 38.85, 27.51, 22.53 (Ac), 20.58 (3 Ac), 16.35 (CH_3).

Methyl (β-D-galactopyranosyl)-(1 → 4)-[(α-L-fucopyranosyl)-(1 → 3)]-2-acetamido-2-deoxy-6-O-sulfo-β-D-glucopyranosyl)-(1 → 6)-[(6-O-sulfo-β-D-galactopyranosyl)-(1 → 3)]-2-acetamido-2-deoxy-α-D-galactopyranoside disodium salt (**2**).—A soln of compound **32** (90 mg) and 10% Pd–C (200 mg) in 3:1 CH_2Cl_2–MeOH (6 mL) was stirred overnight at rt under a H_2 atmosphere. The filtrate was concd under reduced pressure to give a crude residue that was treated for 4–6 h in 1:5 NH_2NH_2·H_2O–MeOH (12 mL) at 80–85 °C. The reaction mixture was then concd and treated overnight with 1:1 Ac_2O–pyridine (6 mL) in the presence of a catalytic amount of DMAP at rt. The mixture was concd and purified by a short column of silica gel and eluted with 6:1 CH_2Cl_2–MeOH to give pure compound **33**. Compound **33** was dissolved in 1:1 MeOH–water (3 mL), treated for 24 h with 1 M CH_3ONa (80 μL) at rt, and then concd. The crude residue was applied to a short column of silica gel and eluted with 3:1:1 *n*-C_3H_7OH–HOAc–water to give compound **2** (6 mg, 26%) as an amorphous solid. R_f 0.16 (3:1:1 *n*-C_3H_7OH–HOAc–water); ^{1}H NMR (D_2O, 600 MHz): δ 5.16–5.10 (d, 1 H, $J_{1,2}$ 3.5 Hz, H-1^{V}), 4.90–4.79 (dd, 1 H, H-5^{V}), 4.78–4.60 (d, 1 H, $J_{1,2}$ 3.0 Hz, H-1^{I}), 4.64–4.60 (d, 1 H, $J_{1,2}$ 8.8 Hz, H-1^{III}), 4.60–4.58 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1^{IV}), 4.46–4.44 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1^{II}), 4.44–4.40 (d 1 H, *J* 2.8 Hz, H-4^{IV}), 4.40–4.30 (dd, 1 H, H-2^{I}), 4.30–4.28 (dd, 1 H, *J* 2.8 Hz, H-4^{I}), 4.22–4.18 (m, 2 H, H-6b^{II}, H-5^{I}), 4.14–3.80 (m, 11 H, H-3^{I}, H-6b^{I}, H-4^{II}, H-2^{III}, H-3^{V}, H-6a^{I}, H-5^{IV}, H-4^{V}), 3.80–3.46 (m, 12 H, H-5^{III}, H-2^{V}, H-5^{II}, H-3^{IV}, H-3^{II}, H-4^{III}, H-2^{IV}, H-2^{II}), 3.36 (s, 3 H, OCH_3), 2.13 (s, 3 H, Ac), 2.02 (s, 3 H, Ac), 1.20 (d, 3 H, *J* 6.4 Hz, CH$_3^{V}$); ^{13}C NMR (D_2O, 100.6 MHz): δ 173.50 (C=O), 173.20 (C=O), 104.50, 102.30, 101.60, 98.60, 98.40, 77.52, 75.20, 75.00, 73.30, 72.90, 72.75, 72.65 (2 C), 72.25, 71.50, 71.25, 70.60, 69.50 (2 C), 69.25, 68.60, 68.50, 68.00, 67.50, 67.00, 66.30, 62.65, 56.00, 55.28, 48.90, 23.00 (Ac), 22.50 (Ac), 15.50 (CH_3^V); FABMS (*m*/*z*) (positive ion mode): Calcd for $C_{35}H_{58}O_{31}N_2Na_2S_2$: 1112.3; found: 1113.9 [M^+ + 1].

Acknowledgements

The authors thank Dr S. Dutta for mass spectrometry analyses. This work was supported by Grant No. CA 35329 and in part by Grant No. P30CA16056, both awarded by National Cancer Institute.

References

[1] S.D. Rosen, C.R. Bertozzi, *Curr. Biol.*, 6 (1996) 261–264.

[2] A. Lasky, *Annu. Rev. Biochem.*, 64 (1995) 113–139.

[3] A. Varki, *J. Clin. Invest.*, 99 (1997) 158–162.

[4] J.B. Lowe, P.A. Ward, *J. Clin. Invest.*, 99 (1997) 822–826.

[5] T. Feizi, C. Galustian, *Trends Biochem. Sci.*, 24 (1999) 124–128.

[6] C. Galustian, A.M. Lawson, S. Komba, H. Ishida, M. Kiso, T. Feizi, *Biochem. Biophys. Res. Commun.*, 240 (1997) 748–751.

[7] W.J. Sander, E.J. Gordon, O. Dwir, P.J. Beck, R. Alon, L.L. Kiessling, *J. Biol. Chem.*, 274 (1999) 5271–5278.

[8] (a) E.E. Shimanel, G.J. McGarvey, A.J. Jablonowski, C.-H. Wong, *Chem. Rev.*, 98 (1998) 833–862; (b) C.-H. Wong, *Acc. Chem. Res.*, 32 (1999) 376–385.

[9] R.K. Jain, C.F. Piskorz, B.-G. Huang, R.D. Locke, H.-L. Han, A. Koening, A. Varki, K.L. Matta, *Glycobiology*, 8 (1998) 707–717.

[10] T. Mukaiyama, Y. Murai, S. Shaoda, *Chem. Lett.*, (1981) 431–432.

[11] S. Sato, M. Mori, Y. Ito, T. Ogawa, *Carbohydr. Res.*, 155 (1986) C6–C10.

[12] T. Ogawa, K. Sasajima, *Tetrahedron*, 37 (1981) 2787–2792.

[13] N. Khiar, M. Martin-Lomas, *J. Org. Chem.*, 60 (1995) 7017–7021.

[14] A. Koening, R.K. Jain, R. Vig, K.E. Norgard-Sumnicht, K.L. Matta, A. Varki, *Glycobiology*, 7 (1997) 79–93.

[15] S, Ogawa, K. Hirai, T. Odagiri, N. Matsunaga, T. Yamazaki, A. Nakajima, *Eur. J. Org. Chem.*, (1998) 1099–1152.

[16] M. Bessodes, J. Shamsazar, K. Antonakis, *Synthesis*, (1988) 560–562.

[17] J.-L. Montero, J.-Y. Winum, A. Leydet, M. Kamal, A.A Pavia, J.-P. Roque, *Carbohydr. Res.*, 297 (1997) 175–180.

[18] R.K. Jain, C.F. Piskorz, E.V. Chandrasekaran, K.L. Matta, *Carbohydr. Res.*, 271 (1995) 247–251.

[19] (a) A.K. Sarkar, K.S. Rostand, R.K. Jain, K.L. Matta, J.D. Esko, *J. Biol. Chem.*, 272 (1997) 25608–25616. (b) M.J. Gaunt, J. Yu, J.B. Spencer, *J. Org. Chem.*, 63 (1998) 4172–4173.

[20] (a) J. Xia, S.A. Abbas, R.D. Locke, C.F. Piskorz, J.L. Alderfer, K.L. Matta, *Tetrahedron Lett.*, 41 (2000) 169–173. (b) J. Xia, C.F. Piskorz, J.L. Alderfer, R.D. Locke, K.L. Matta, *Tetrahedron Lett.*, 41 (2000) 2773–2776. (c) J. Xia, J.L. Alderfer, C.F. Piskorz, K.L. Matta, *Chem. Eur. J.*, (in press).

[21] G.M. Sheldrick, SHELXL-96: Program for the Solution of Crystal Structures, University of Göttingen, Germany, 1996.

[22] C.K. Johnson, ORTEP II Report ORNL-5138, 1976, Oak Ridge National Laboratory, Oak Ridge, TN, USA.