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Synthesis and NMR study of a heptasaccharide, epitope of the stage-specific embryonic antigen-1 (SSEA-1)

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Abstract

This paper describes an efficient synthesis of the β-2-trimethylsilylethyl glycoside of lacto-*N*-fucoheptaose based on a highly stereo- and regioselective glycosylation between a Lewis^x trisaccharidic donor and a tetraol tetrasaccharidic acceptor. The title compound was characterized by high-resolution NMR spectroscopy.

Keywords: SSEA-1; Lewis^x; Glycosylation; Heptasaccharide; NMR spectroscopy; Conformation

1. Introduction

It has become evident in recent years that carbohydrates play an important role in intercellular recognition processes. Dramatic changes in specific carbohydrates have been observed at every step of ontogenic development, in oncogenic transformation, and their functional role in cell adhesion has been proposed [1,2]. Particular interest has focused on the family of oligosaccharides containing one or more non-reducing terminal Lewis^x group. In a mouse model of pre-implantation embryo, differential expression of 'stage-specific embryonic antigen 1' (SSEA-1) was observed at the 8–16 cell (morula) stage, which correlates approximately in time with the onset of compaction, and declined rapidly after compaction, being restricted to the inner cell mass of the blastocyst [3]. SSEA-1 was subsequently identified as Lewis^x [4,5]. The compaction process was inhibited and the once-compacted embryo 'decompacted' by Lewis^x, or more efficiently by LNFP III (Lewis^x pentasaccharide). Thus, compaction, the first cell-adhesion event of ontogenic development, is mediated in part by Lewis^x [6]. A glycolipid **1a**, one of the glycolipids that are reactive with anti-SSEA-1 antibody, has been isolated from human erythrocyte stroma [7] and chemically synthesized [8]. Because of the potential importance of the heptasaccharidic moiety of this glycolipid in glycobiology, we have undertaken a detailed study by NMR in order to explore its solution conformation. The β-2-trimethylsilylethyl glycoside was selected rather than a free sugar to simplify the NMR spectra; it can then be easily transformed into a glycosyl donor whenever the corresponding glycolipids are needed. We report herein an efficient total synthesis of this heptasaccharide **1b** based on our recent achievement for Lewis^x synthesis [9] and its NMR study.

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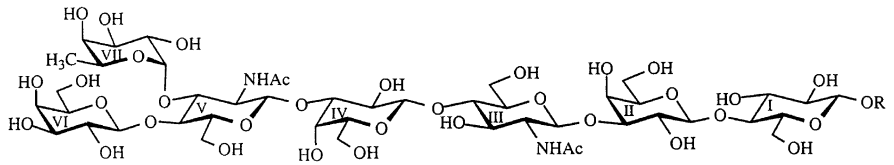


1a: R = Ceramide

1b: R = SE (2-trimethylsilylethyl)

2. Results and discussion

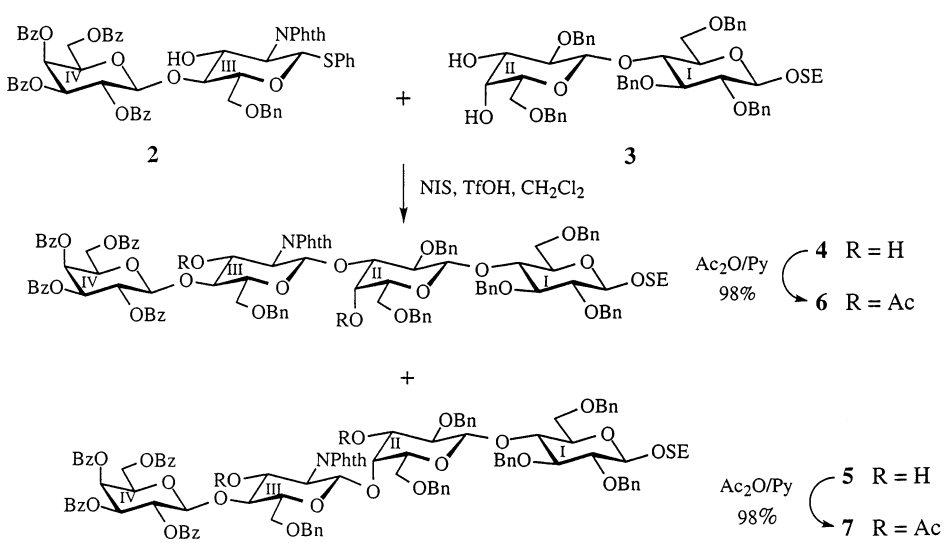
Chemical synthesis.—The selected synthetic strategy was a '2 + 2 + 3' block construction. Similar fragment condensations have been reported in the recent literature [10]. Thus, the coupling of the donor **2** [9] with the known lactoside **3** [11] was performed in dichloromethane, promoted by *N*-iodosuccinimide (NIS)-trifluoromethanesulfonic acid (TfOH) at −30 °C for 1 h, generating the desired tetrasaccharide **4** and its regioisomer **5** in 80 and 15% yield, respectively (Scheme 1).

The stereochemistry of the newly introduced linkage in tetrasaccharide **4** was determined to be β on the basis of the GlcN H-1, H-2 coupling constant ($J_{1^{III},2^{III}}$ 8.5 Hz). The regiochemistry of **4** was readily assigned from the ^{1}H NMR spectrum of **6**, obtained from **4** by acetylation, which revealed a deshielded signal for H-4 of the galactose unit (II ring) at 5.42 ppm (dd, $J_{4^{II},5^{II}} < 1$ Hz, $J_{3^{II},4^{II}}$ 3.5 Hz), indicating the position of the new glycosidic linkage in **4** to be at OH-3^{II} of the acceptor **3**.

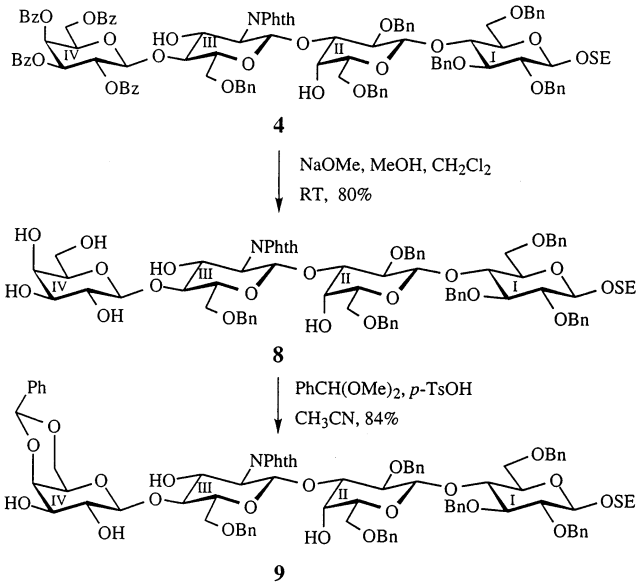
Similarly, the stereochemistry of the newly introduced linkage in tetrasaccharide **5** was determined to be β on the basis of the GlcN H-1, H-2 coupling constant ($J_{1^{III},2^{III}}$ 8.6 Hz). The regiochemistry of **5** was readily assigned from the ^{1}H NMR spectrum of **7**, obtained from **5** by acetylation, which revealed a deshielded signal for H-3 of the galactose unit (II ring) at 4.73 ppm (dd, $J_{3^{II},4^{II}}$ 2.5 Hz, $J_{2^{II},3^{II}}$ 10.3 Hz), indicating the position of new glycosidic linkage in **5** to be at OH-4^{II} of the acceptor **3**.

Treatment of **4** with sodium methoxide in methanol–dichloromethane gave **8** (80%) with the galactose moiety (IV ring) O-unprotected; regioselective 4^{IV},6^{IV}-*O*-benzylidenation by reaction with benzaldehyde dimethyl acetal in the presence of *p*-toluenesulfonic acid as catalyst furnished the tetrasaccharide **9** in 84% yield (Scheme 2).

When tetraol **9** was condensed with trisaccharide donor **10** [10] in the presence of NIS-TfOH, a sole regioisomer, the expected heptasaccharide **11** (glycosylation at 3^{IV} position), was formed in 87% yield (Scheme 3).



Scheme 1.

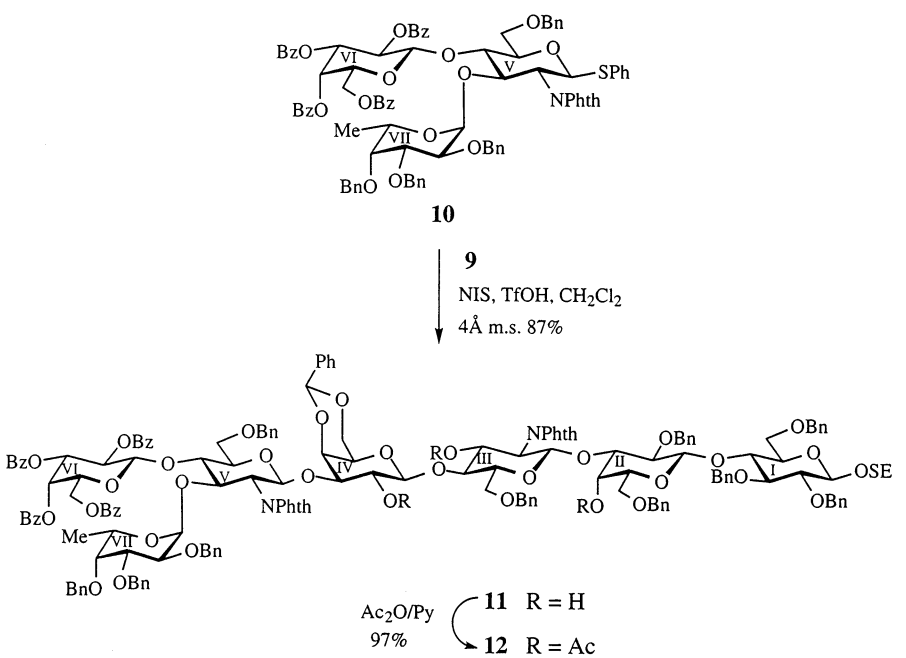


Scheme 2.

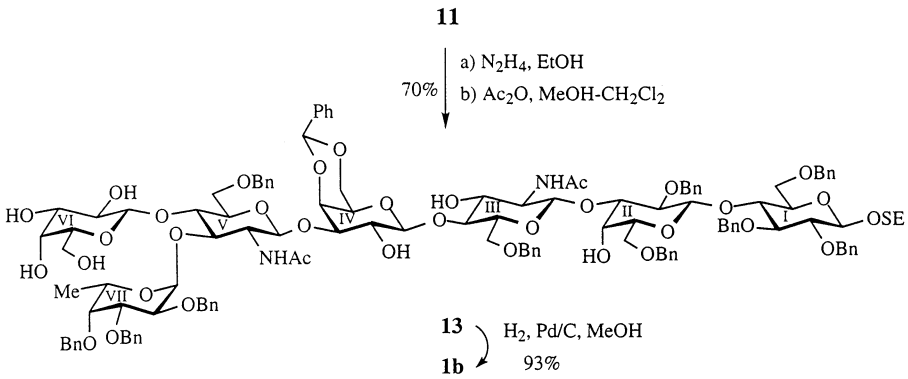
The stereochemistry of the newly introduced linkage in heptasaccharide **11** was determined to be β on the basis of the GlcN H-1, H-2 coupling constant ($J_{1^{III},2^{III}}$ and $J_{1^{V},2^{V}}$ 8.3 and 8.5 Hz). The regiochemistry of **11** was assigned from the ^{1}H NMR spectrum of **12**, obtained from **11** by acetylation, which revealed in $CDCl_3$ solvent a deshielded signal for H-3^{III} at 5.59 ppm (dd, $J_{3^{III},4^{III}}$ 9.0, $J_{2^{III},3^{III}}$ 10.7 Hz) and another deshielded signal for H-4^{II} at 5.45 ppm (dd, $J_{4^{II},5^{II}} < 1$, $J_{3^{II},4^{II}}$ 3.4 Hz); however the signal for H-2^{IV} was not clear due to overlapping of several signals. To clarify this, a ^{1}H NMR was taken in C_6D_6 and $CDCl_3$ solution. The spectrum then showed a well-resolved one-proton signal at 4.97 ppm for H-2^{IV} (dd, $J_{1^{IV},2^{IV}}$ 8.0 Hz, $J_{2^{IV},3^{IV}}$ 9.9 Hz). We therefore concluded that the position of the newly formed glycosidic linkage in **11** was OH-3^{IV} of the acceptor **9**, which confirmed the high stereo- and regioselectivity of this glycosylation.

Treatment of heptasaccharide **11** with hydrazine hydrate in refluxing ethanol, followed by N-acetylation using acetic anhydride in methanol and dichloromethane, led to the derivative **13** in 70% overall yield from **11**. Catalytic hydrogenolysis of **13** in methanol for 2 h and purification of the product on Sephadex G25-150 afforded, after freeze-drying, the desired heptasaccharide **1b** in 93% yield (Scheme 4).

NMR study.—NMR experiments have been performed in D_2O at 290 K and in $(CD_3)_2SO$ at 298 K. No peculiar conformational behavior of the heptasaccharide could be evidenced in these solvents, relative to the solution structure of the pentasaccharide LNFP III, which has recently been studied in detail by NMR spectroscopy [12]. For the heptasaccharide,



Scheme 3.



Scheme 4.

Table 1
600 MHz proton chemical shift assignment of **1b** in D_2O at 290 K

	Fuc^{VII}	Gal^{VI}	$GlcNAc^{V}$	Gal^{IV}	$GlcNAc^{III}$	Gal^{II}	$Glc^{I}OSE$
H-1	4.965	4.303	4.537	4.300	4.529	4.271	4.337
H-2	3.525	3.333	3.810	3.404	3.641	3.412	3.115
H-3	3.749	3.492	3.708	3.555	3.563	3.558	3.471
H-4	3.631	3.732	3.801	4.003	3.414	3.996	3.409?
H-5	4.689	3.435?					3.636?
H-6,H-6′	1.010						3.805?
Solvent	4.742						
Ac	1.860						
Ac	1.851						

Table 2
Proton chemical shifts of **1b** in $(CD_3)_2SO$ at 298 K

	Fuc^{VII}	Gal^{VI}	$GlcNAc^{V}$	Gal^{IV}	$GlcNAc^{III}$	Gal^{II}	$Glc^{I}OSE$
H-1	4.830	4.262	4.673	4.244	4.615	4.235	4.179
H-2	3.393	3.267	3.617	3.398	3.455	3.402	2.972
H-3	3.545	3.231	3.698	3.430	3.509	3.437	3.295
H-4	3.450	3.663	3.624	3.843	3.315	3.824	3.288
H-5	4.642	3.767 [a]	3.312		3.272		3.840 [a]

[a] From ROESY.

the presence of three galactose units and two glucosamine units rendered the proton assignment very difficult owing to severe spectral overlap, even at very high field. Moreover, the strong coupling conditions often encountered garbled some inter-proton distance constraints obtained from the NMR data. Nevertheless, to help overcome this first experimental difficulty, semi-soft experiments have been performed to allow both the intra- and inter-unit assignments. Selective excitation of the anomeric region by an 'excitation sculpting' sequence [13] was followed by the evolution delay. At the middle the composite pulse sequence 'hard 180° pulse–soft 180° pulse centered on the H-1' enabled the refocusing of the spin–spin coupling and then the recording of the proton-decoupled spectra in the dimension F1 [14], and therefore increased the apparent peak separation in this dimension.

Tables 1 and 2 give the ^1H chemical shifts of **1b** in D_2O and in $(CD_3)_2SO$, respectively. Addition of a few droplets of D_2O to the last solution allowed simplification of the spectrum by suppression of the OH signals and of the induced couplings. The H-6 signals could

not be unambiguously assigned, and are therefore not given. The safer assignment of the proton chemical shifts in $(CD_3)_2SO$ led us to use only this sample to analyze the conformational properties of the oligosaccharide. A previously described procedure using off-resonance ROESY experiments at different mixing times and spin-lock angles [15] has been shown to be very efficient in the case of oligosaccharides, avoiding the use of an internal distance reference and satisfyingly describing the local dynamics [16,17]. However, this approach seemed to us too time-consuming both for the acquisition and the processing in the view of the strong signal overlaps, which would prevent precise determination of the internal dynamics. We therefore decided to use the classical method (internal distance reference), using the off-resonance ROESY experiment instead of the ROESY method to avoid artifacts resulting from coherence transfer Hartmann–Hahn and offset effects. Analysis of ^{13}C relaxation would also have been helpful, but would have required higher oligosaccharide concentrations.

Off-resonance ROESY experiments were acquired at three mixing times (50, 100 and 200 ms) and at angle of 54.7° on a 18.8 Tesla spectrometer (protons resonating at 800 MHz). Fig. 1 displays the full contour plot of a semi-soft ROESY experiment.

Two other experiments (100 and 200 ms) were acquired with an angle of 45°. These last experiments are less sensitive to spin diffusion [18] and were consequently used to check the consistency of the data. The two-spin initial build-up rate approximation was used to derive dipolar cross-relaxation rates. The point for a vanishing mixing time was added and a second-order polynomial function was used to fit the data. The reference distance r_{ref} from which the other distances r_i were extracted via a simple proportionality rule between the cross-relaxation rates σ of the form $r_i = r_{ref}\ (\sigma_{ref}/\sigma_i)^{1/6}$ was Fuc-H-1/Fuc-H-2, taken to be equal to 2.6 Å. Forty H–H distance data obtained using this method were used as constraints in a simulated annealing procedure. Starting from a random non-aberrant structure, the following procedure was repeated 50 times. A 40 ps dynamics at 1000 K with a timestep of 4 fs was followed by slow cooling during 40 ps (same timestep). The force field had been parametrized for oligosaccharides and previously used for cyclodextrins or Lewis^x derivatives. In order to avoid the creation of virtual conformations during simulated annealing, dihedral angles defining the

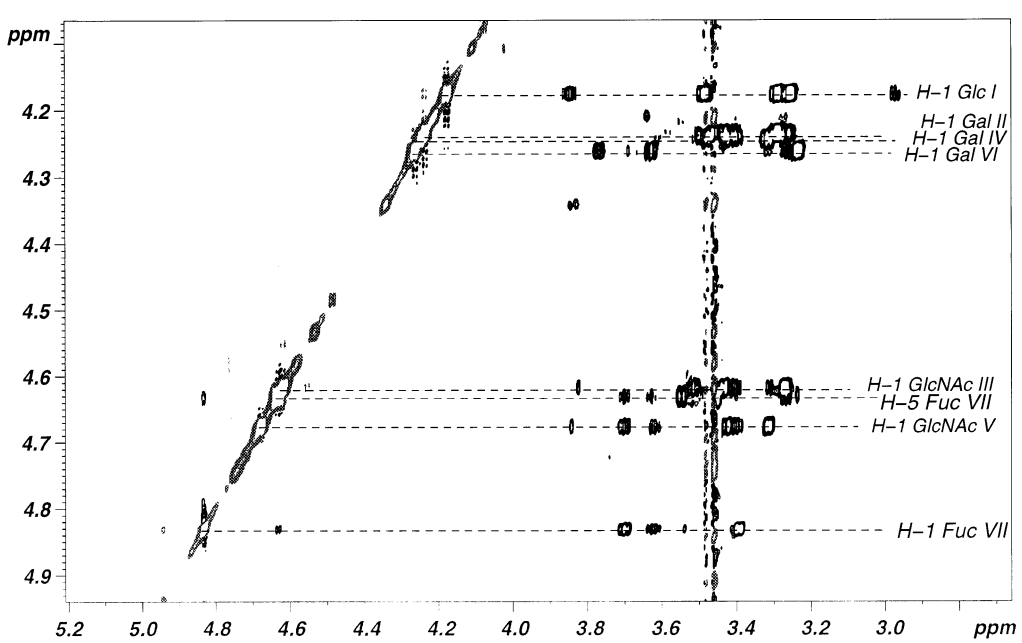


Fig. 1. Semi-soft ROESY map (mixing time, 100 ms; spin-lock angle, 54.7°). The complete contour plot is displayed and the H-1 rows (and Fuc H-5) are indicated. Complementary experiments with excitation of the non-anomeric protons have also been acquired.

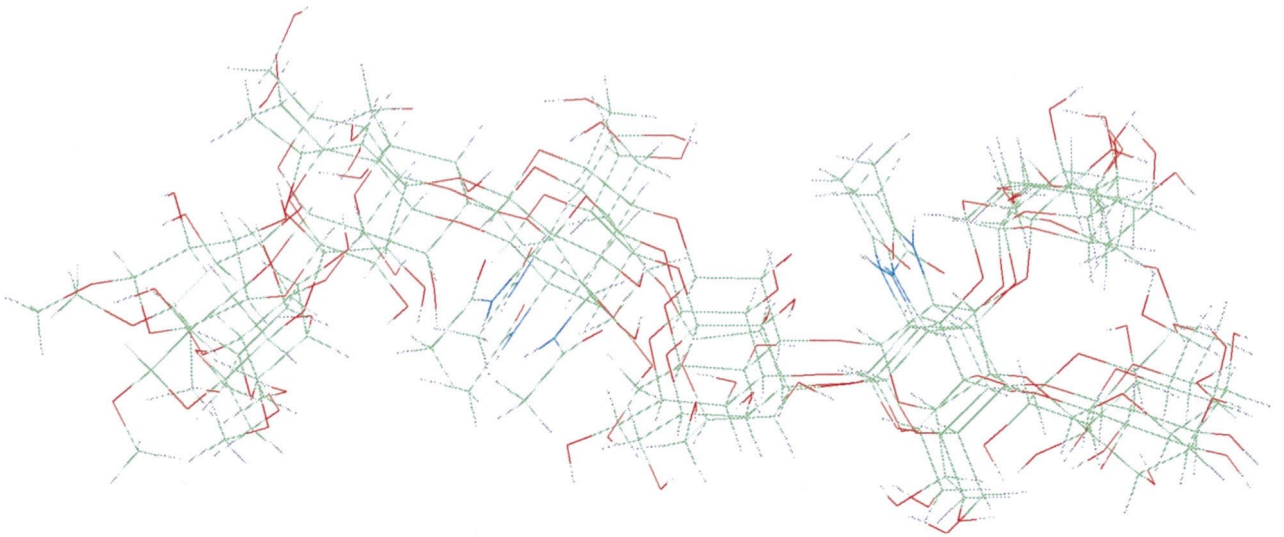


Fig. 2. Superposition of the four lowest-energy structures of the heptasaccharide **1b**.

pyranose units were replaced by impropers. During the high-temperature dynamics, progressive introduction of the distance constraints with low weight on the van der Waals terms ensured a large sampling of the conformational space. Among the 50 generated structures, only 18 were kept according to selection criteria based on the covalent energy and the number of nOe violations (the accepted structures had to exhibit less than three nOe violations). The reminiscent nOe violations invariably correspond to the inter-glycosidic linkage $GlcNAc^{V}$–Gal^{IV}. As in the case of the pentasaccharide LNFP III [12], this should arise from a fast exchange between two conformational families in solution. The rms deviation from the average of the coordinates of the heavy atoms was 1.5 Å, and 1.3 Å when the hydroxyl oxygens were removed from the calculation. The four lowest-energy structures are superimposed in Fig. 2.

The average interglycosidic angles for the $Lewis^{x}$ motif were measured at $\Phi 1 = -74°$, $\psi 1 = 132°$ for the Gal^{VI}–$GlcNAc^{V}$ bond and $\Phi 2 = -88°$, $\psi 2 = 151°$ for Fuc^{VII}–$GlcNAc^{V}$. A procedure has been applied to test the influence of the experimental constraints on the validity of the resulting structures. One of the structures has been energy-minimized after removal of these constraints. The force field used for this minimization was CFF91 [19], and 500 steps using the conjugate gradient algorithm have been performed. The rms deviation between the starting and the final structure was less than 0.5 Å, no interglycosidic dihedral angle deviates by more than 10°.

The overall solution structure of the heptasaccharide seems to adopt a helix shape, which has already been suggested for linear polysaccharides such as starch. A precise characterization of this helix is however prevented by the weakness of NMR spectroscopy in obtaining long-range constraints.

3. Experimental

General methods.—Melting points were determined with a Büchi model 510 m.p. apparatus and are uncorrected. Optical rotations were measured at 20 ± 2 °C with a Perkin–Elmer model 241 digital polarimeter, using a 10 cm, 1 mL cell. Chemical ionization mass spectra (CIMS ammonia) and fast atom bombardment mass spectra (FABMS) were obtained with a JMS-700 spectrometer. Elemental analyses were performed by Service de Microanalyse de l'Université Pierre et Marie Curie, 4 Place Jussieu, F-75005 Paris, France. ^{1}H NMR spectra were recorded with Bruker AC 250 and Bruker AM 400 spectrometers for solutions in $CDCl_3$ or D_2O at ambient temperature. Assignments were aided by COSY experiments. ^{13}C NMR spectra were recorded at 62.9 MHz with a Bruker AC 250 and at 100.6 MHz with a Bruker AM 400 for

solutions in $CDCl_3$ adopting 77.00 ppm for the central line of $CDCl_3$. Assignments were aided by the J-mod technique and proton–carbon correlation. Reactions were monitored by thin-layer chromatography (TLC) on a precoated plate of Silica Gel 60 F_{254} (layer thickness 0.2 mm, E. Merck, Darmstadt, Germany) and detection by charring with H_2SO_4. Flash column chromatography was performed on Silica Gel 60 (230–400 mesh, E. Merck). The characterization of the conformation of heptasaccharide **1b** was performed thanks to spectra acquired on Bruker DRX 500, DRX 600 and DRX 800 spectrometers. The molecular modelling was performed using X-PLOR software [20]. The energy minimization has been performed with the DISCOVER [21] program using the CFF91 force field (class II force field) with automatic assignment of the atom types and charges.

2-(Trimethylsilyl) ethyl 2,3,4,6-tetra-O-*benzoyl*-β-D-*galactopyranosyl*-(1→4)-*6*-O-*benzyl-2-deoxy-2-phthalimido*-β-D-*glucopyranosyl*-(1→3)-*2,6-di*-O-*benzyl*-β-D-*galactopyranosyl*(1→4)-*2,3,6-tri*-O-*benzyl*-β-D-*glucopyranoside* (**4**) *and 2-(trimethylsilyl) ethyl 2,3,4,6-tetra*-O-*benzoyl*-β-D-*galactopyranosyl*-(1→4)-*6*-O-*benzyl-2-deoxy-2-phthalimido*-β-D-*glucopyranosyl*-(1→4)-*2,6-di*-O-*benzyl*-β-D-*galactopyranosyl*-(1→4)-*2,3,6-tri*-O-*benzyl*-β-D-*glucopyranoside* (**5**).—A mixture of donor **2** (500 mg, 0.47 mmol), acceptor **3** (500 mg, 0.56 mmol), 4 Å powdered molecular sieves (2.5 g) and CH_2Cl_2 (30 mL) was stirred at room temperature (rt) for 30 min. NIS (225 mg, 1 mmol) was added at rt. The reaction mixture was cooled to −30 °C. Triflic acid (4.4 μL, 50 mmol) was added. The reaction mixture was stirred at −30 °C for 1 h, neutralized (Et_3N), filtered through Celite, washed with aq thiosulfate, water, brine, dried over $MgSO_4$ and concentrated. The residue obtained was submitted to flash chromatography (2:1 cyclohexane–EtOAc then 3:1 toluene–EtOAc) to afford **4** (697 mg, 80%) as an amorphous solid: $[\alpha]_D$ +62° (*c* 1, $CHCl_3$); TLC (3:2 cyclohexane–EtOAc): R_f 0.52; ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.15–6.85 (m, aromatic protons), 5.99 (dd, 1 H, $J_{4,5}<1$, $J_{3,4}$ 3.4 Hz, H-4^{IV}), 5.90 (dd, 1 H, $J_{1,2}$ 8.0, $J_{2,3}$ 10.5 Hz, H-2^{IV}), 5.63 (dd, 1 H, H-3^{IV}), 5.44 (d, 1 H, $J_{1,2}$ 8.5 Hz, H-1^{III}), 4.97, 4.71 (2 d, 2 H, *J* 11.9 Hz, CH_2Ph), 4.96 (d, 1 H, H-1^{IV}), 4.90, 4.72 (2 d, 2 H, *J* 11.1 Hz, CH_2Ph), 4.64 (ddd, 1 H, $J_{3,OH}$ 1.4, $J_{3,4}$ 8.3, $J_{2,3}$ 10.6 Hz, H-3^{III}), 4.47 (d, 1 H, OH-3^{III}), 4.44, 4.25 (2 d, 2 H, *J* 12.2 Hz, CH_2Ph), 4.38, 4.23 (2 d, 2 H, *J* 12.0 Hz, CH_2Ph), 4.13, 4.03 (2 d, 2 H, *J* 12.0 Hz, CH_2Ph), 4.02 (br, 1 H, H-4^{II}), 3.97 (m, 1 H, OCH_2CH$_2$Si), 3.83 (dd, 1 H, $J_{4,5}$ 9.4 Hz, H-4^{III}), 3.56 (m, 1 H, OCH_2CH$_2$Si), 2.78 (br, 1 H, OH-4^{II}); 1.02 (ddd, *J* 2.3, 6.7, 14.9 Hz, CH$_2$Si), 0.03 (s, 9 H, SiMe$_3$); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 168.27, 167.51 (2 C=O, Phth), 166.06, 165.37, 165.27, 164.90 (4 C=O, Bz), 139.06, 138.70, 138.42, 138.40, 138.23, 137.74, 131.19, 128.79, 128.53 (aromatic C), 133.74, 133.69, 133.38, 133.19, 129.93, 129.82, 129.77, 129.68, 128.64, 128.62, 128.38, 128.31, 128.29, 128.23, 128.16, 128.09, 127.92, 127.72, 127.66, 127.40, 127.38, 127.24, 127.07, 126.54, 126.25 (aromatic *C*H), 102.95, 101.99, 101.84, 98.62 (4 *C*H, C-1^I, C-1^{II}, C-1^{III}, C-1^{IV}), 83.93, 82.84, 82.83, 81.82, 77.96, 76.11, 74.63, 73.61, 72.57, 72.31, 71.28, 69.57, 69.44, 67.91, 67.23 (15 *C*H, ring C), 75.25, 74.82, 74.18, 73.19, 72.95, 72.87 (6 *C*H$_2$Ph), 68.54, 68.10, 67.82, 67.17 (4 *C*H$_2$, C-6^I, C-6^{II}, C-6^{III}, O*C*H$_2$-CH$_2$Si), 62.64 (C-6^{IV}), 55.69 (C-2^{III}), 18.36 (*C*H$_2$Si), −1.49 (SiMe$_3$); FABMS: *m*/*z* 1858 (63, $[M+Li]^+$). Anal. Calcd for $C_{107}H_{109}NO_{26}Si$: C, 69.35; H, 5.93; N, 0.76. Found: C, 69.30; H, 5.95; N, 0.68.

Second eluted was compound **5**, also as an amorphous solid: $[\alpha]_D$ +47° (*c* 1.6, $CHCl_3$); TLC (3:1 toluene–EtOAc): R_f 0.34; ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.11–6.90 (m, aromatic H), 5.98 (dd, 1 H, $J_{4,5}<1$, $J_{3,4}$ 3.5 Hz, H-4^{IV}), 5.89 (dd, 1 H, $J_{1,2}$ 8.0, $J_{2,3}$ 10.4 Hz, H-2^{IV}), 5.61 (dd, 1 H, H-3^{IV}), 5.39 (d, 1 H, $J_{1,2}$ 8.6 Hz, H-1^{III}), 5.09, 4.83 (2 d, 2 H, *J* 10.1 Hz, CH_2Ph), 5.00 (d, 1 H, H-1^{IV}), 4.95, 4.83 (2 d, 2 H, *J* 11.0 Hz, CH_2Ph), 4.87 (m, 1 H, H-3^{III}), 4.70 (dd, 1 H, H-6^{IV}), 4.45 (d, 1 H, $J_{3,OH}$ 1.1 Hz, OH-3^{III}), 4.06–3.97 (m, 3 H, OCH_2CH$_2$Si, H-4^{III}, H-4^{II}), 3.88 (dd, 1 H, $J_{3.4}=J_{4.5}$ 9.4 Hz, H-4^I), 3.78 (dd, 1 H, $J_{5,6}$ 6.3, $J_{6,6'}$ 10.1 Hz, H-6^{II}), 3.06 (dd, 1 H, $J_{1.2}$ 7.8, $J_{2.3}$ 9.6 Hz, H-2^{II}), 1.99 (d, 1 H, $J_{3,OH}$ 2.9 Hz, OH-3^{II}), 1.09–1.04 (m, 2 H, CH$_2$Si), 0.05 (s, 9 H, SiMe$_3$); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 168.49, 167.47 (2 C=O, Phth), 166.12, 165.40,

165.31, 165.04 (4 C=O, Bz), 138.89, 138.85, 138.78, 138.12, 137.83, 132.55, 131.88, 128.88, 128.81 (aromatic C), 133.71, 133.57, 133.52, 133.33, 133.17, 129.96, 129.76, 129.69, 129.06, 128.65, 128.51, 128.43, 128.27, 128.25, 128.18, 128.08, 127.99, 127.92, 127.66, 127.40, 127.34, 127.28, 127.25, 127.23, 127.19, 127.13 (aromatic *C*H), 103.07 (C-1^{I}), 102.10 (C-1^{II}), 102.00 (C-1^{IV}), 99.71 (C-1^{III}), 82.54, 82.17, 81.78, 80.88, 76.72, 76.30, 75.04, 73.83, 73.37, 73.13, 72.18, 71.40, 69.54, 69.04, 67.95 (15 *C*H, ring C), 75.84, 74.99, 74.91, 73.08, 73.04, 72.98 (6 CH_2Ph), 68.96 (C-6^{II}), 68.23 (C-6^{I}), 67.93 (C-6^{III}), 67.28 (OCH_2CH_2Si), 62.49 (C-6^{IV}), 56.29 (C-2^{III}), 18.40 (CH_2Si), −1.45 ($SiMe_3$); FABMS: *m*/*z* 1875 (100, $[M+Na]^+$). Anal. Calcd for $C_{107}H_{109}NO_{26}Si{\cdot}H_2O$: C, 68.68; H, 5.98; N, 0.75. Found: C, 68.65; H, 6.31; N, 0.79.

2-(Trimethylsilyl) ethyl 2,3,4,6-tetra-O-*benzoyl*-β-D-*galactopyranosyl*-(1→4)-*3*-O-*acetyl*-*6*-O-*benzyl*-*2*-*deoxy*-*2*-*phthalimido*-β-D-*glucopyranosyl*-(1→3)-*4*-O-*acetyl*-*2,6*-*di*-O-*benzyl*-β-D-*galactopyranosyl*-(1→4)-*2,3,6*-*tri*-O-*benzyl*-β-D-*glucopyranoside* (**6**).—Compound **4** (6 mg, 3.2 mmol) was acetylated with acetic anhydride (0.3 mL) in pyridine (0.6 mL) for 15 h at rt. After concentration, the residue was coevaporated with toluene to give **6** (6.2 mg) as a syrup: TLC (2:1 cyclohexane–EtOAc): R_f 0.33; ^{1}H NMR (400 MHz, $CDCl_3$): δ 5.94 (dd, 1 H, $J_{4,5}<1$, $J_{3,4}$ 3.4 Hz, H-4^{IV}), 5.83 (dd, 1 H, $J_{1,2}$ 8.0, $J_{2,3}$ 10.4 Hz, H-2^{IV}), 5.72 (dd, 1 H, $J_{3,4}$ 8.1, $J_{2,3}$ 10.2 Hz, H-3^{III}), 5.49 (d, 1 H, $J_{1,2}$ 8.5 Hz, H-1^{III}), 5.46 (dd, 1 H, H-3^{IV}), 5.42 (dd, 1 H, $J_{4,5}<1$, $J_{3,4}$ 3.5 Hz, H-4^{II}), 4.97 (d, 1 H, H-1^{IV}), 4.92, 4.68 (2 d, 2 H, *J* 10.7 Hz, CH_2Ph), 4.88, 4.71 (2 d, 2 H, *J* 11.1 Hz, CH_2Ph), 4.83, 4.48 (2 d, 2 H, *J* 12.1 Hz, CH_2Ph), 2.07 (s, 3 H, Ac), 1.92 (s, 3 H, Ac), 1.02 (m, 2 H, CH_2Si), 0.03 (s, 9 H, $SiMe_3$).

2-(Trimethylsilyl) ethyl 2,3,4,6-tetra-O-*benzoyl*-β-D-*galactopyranosyl*-(1→4)-*3*-O-*acetyl*-*6*-O-*benzyl*-*2*-*deoxy*-*2*-*phthalimido*-β-D-*glucopyranosyl*-(1→4)-*3*-O-*acetyl*-*2,6*-*di*-O-*benzyl*-β-D-*galactopyranosyl*-(1→4)-*2,3,6*-*tri*-O-*benzyl*-β-d-*glucopyranoside* (**7**).—Compound **5** (75 mg, 37.7 mmol) was acetylated with acetic anhydride (2.5 mL) in pyridine (5 mL) for 16 h at rt. After concentration, the residue was submitted to flash chromatography (2:1 cyclohexane–EtOAc) to afford **7** (74 mg, 94%) as an amorphous solid: $[\alpha]_D$ +45° (*c* 1.2, $CHCl_3$); TLC (1:1 cyclohexane–EtOAc): R_f 0.75; ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.68–6.65 (m, 4 H, aromatic H), 8.05–6.86 (m, 50 H, aromatic H), 6.11 (dd, 1 H, $J_{2,3}$ 9.0, $J_{3,4}$ 10.9 Hz, H-3^{III}), 5.93 (dd, 1 H, $J_{4,5}<1$, $J_{3,4}$ 3.4 Hz, H-4^{IV}), 5.72 (dd, 1 H, $J_{1,2}$ 8.0, $J_{2,3}$ 10.4 Hz, H-2^{IV}), 5.43 (dd, 1 H, H-3^{IV}), 5.27 (d, 1 H, $J_{1,2}$ 8.3 Hz, H-1^{III}), 5.11, 4.86 (2 d, 2 H, *J* 10.1 Hz, CH_2Ph), 4.96, 4.84 (2 d, 2 H, *J* 11.0 Hz, CH_2Ph), 4.92 (d, 1 H, H-1^{IV}), 4.73 (dd, 1 H, $J_{2,3}$ 10.3, $J_{3,4}$ 2.5 Hz, H-3^{II}), 3.86 (dd, 1 H, H-4^{I}), 3.03 (dd, 1 H, $J_{1,2}$ 7.5, $J_{2,3}$ 10.3 Hz, H-2^{II}), 2.12 (s, 3 H, Ac), 1.94 (s, 3 H, Ac), 1.06 (m, 2 H, CH_2Si), 0.05 (s, 9 H, $SiMe_3$); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 170.65, 169.99 (2 C=O, Ac), 167.74, 166.80 (2 C=O, Phth), 165.99, 165.47, 165.28, 164.59 (4 C=O, Bz), 139.06, 138.91, 138.66, 138.12, 137.90, 137.80, 131.91, 131.50, 129.13, 128.97, 128.86, 128.60 (aromatic C), 149.57, 136.14, 134.24, 134.07, 133.59, 133.38, 133.26, 129.89, 129.72, 129.63, 129.60, 129.10, 128.81, 128.65, 128.55, 128.47, 128.30, 128.26, 128.21, 128.12, 128.00, 127.93, 127.77, 127.48, 127.42, 127.39, 127.27, 127.18, 126.85, 123.80, 123.60, 123.28 (aromatic *C*H), 103.10 (C-1^{I}), 102.02 (C-1^{II}), 100.44 (C-1^{IV}), 98.28 (C-1^{III}), 82.63 (C-3^{I}), 81.75 (C-2^{I}), 78.60 (C-2^{II}), 77.04 (C-4^{I}), 75.35 (C-4^{III}), 74.95 (C-5^{I}), 74.74 (C-3^{II}), 73.89 (C-5^{III}), 73.84 (C-4^{II}), 72.94 (C-5^{II}), 71.79 (C-3^{IV}), 70.98 (C-5^{IV}), 69.99 (C-3^{III}), 69.81 (C-2^{IV}), 67.78 (C-4^{IV}), 75.86, 75.01, 74.71, 73.37, 72.99, 72.90 (6 CH_2Ph), 68.25 (C-6^{II}), 67.97 (C-6^{I}), 67.27 (C-6^{III}, OCH_2CH_2Si), 61.59 (C-6^{IV}), 20.74, 20.70 (2 C, Ac), 55.10 (C-2^{III}), 18.40 (CH_2Si), −1.44 ($SiMe_3$). Anal. Calcd for $C_{111}H_{113}NO_{28}Si$: C, 68.82; H, 5.88; N, 0.72. Found: C, 68.54; H, 6.22; N, 1.02.

2-(Trimethylsilyl) ethyl β-D-*galactopyranosyl*-(1→4)-*6*-O-*benzyl*-*2*-*deoxy*-*2*-*phthalimido*-β-D-*glucopyranosyl*-(1→3)-*2,6*-*di*-O-*benzyl*-β-D-*galactopyranosyl*-(1→4)-*2,3,6*-*tri*-O-*benzyl*-β-D-*glucopyranoside* (**8**).—To a solution of **4** (675 mg, 0.364 mmol) in 3:1 MeOH–CH_2Cl_2 (20 mL) was added 0.1 M NaOMe solution in MeOH (1.8 mL). After stirring for 2 h at rt, the mixture was neutralized by Amberlite resin (IR-120, H^+ form),

filtered and concentrated to dryness. The residue obtained was submitted to flash chromatography (10:1 CH_2Cl_2–MeOH) to afford **8** (418 mg, 80%) as an amorphous solid: TLC (7:1 CH_2Cl_2–MeOH): R_f 0.44; CIMS: m/z 1453 (35, $[M + NH_4]^+$). This compound was characterized as its 4,6-*O*-benzylidene derivative **9**.

*2-(Trimethylsilyl) ethyl 4,6-*O*-benzylidene-β-*D*-galactopyranosyl-(1→4)-6-O-benzyl-2-deoxy-2-phthalimido-β-*D*-glucopyranosyl-(1→3)-2,6-di-O-benzyl-β-*D*-galactopyranosyl-(1→4)-2,3,6-tri-O-benzyl-β-*D*-glucopyranoside* (**9**).—To a solution of **8** (780 mg, 0.543 mmol) in dry MeCN (20 mL) were added benzaldehyde dimethyl acetal (0.162 mL, 1.08 mmol) and *p*-toluenesulfonic acid (30 mg). The reaction mixture was stirred at rt for 2 h and solid potassium carbonate (200 mg) was added. The mixture was stirred for 0.5 h, filtered through Celite, and concentrated. The residue obtained was submitted to flash chromatography (1:4 cyclohexane–EtOAc) to afford **9** (696 mg, 84%) as an amorphous solid: $[\alpha]_D$ −18° (*c* 1, $CHCl_3$); TLC (1:4 cyclohexane–EtOAc): R_f 0.31; ^{1}H NMR (400 MHz, $CDCl_3$): δ 5.51 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1^{III}), 5.47 (s, 1 H, C*H*Ph), 5.00, 4.75 (2 d, 2 H, *J* 10.7 Hz, CH_2Ph), 4.93, 4.76 (2 d, 2 H, *J* 11.0 Hz, CH_2Ph), 4.64, 4.59 (2 d, 2 H, *J* 12.0 Hz, CH_2Ph), 1.06 (m, 2 H, CH_2Si), 0.06 (s, 9 H, $SiMe_3$); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 168.20, 167.90 (2 C=O, Phth), 139.03, 138.68, 138.38, 138.34, 138.25, 137.71, 137.25, 131.16 (aromatic C), 133.78, 129.14, 128.40, 128.21, 128.18, 128.14, 127.92, 127.91, 127.75, 127.71, 127.58, 127.45, 127.39, 127.35, 127.09, 126.55, 126.25, 126.23 (aromatic *C*H), 103.57, 102.93, 101.87, 101.10, 98.73 (5 *C*H, *C*HPh, C-1^{I}, C-1^{II}, C-1^{III}, C-1^{IV}), 83.79, 82.80, 82.25, 81.81, 77.98, 76.10, 74.75, 74.63, 73.55, 72.56, 72.48, 71.14, 69.44, 67.35, 66.69 (15 *C*H, ring C), 75.23, 74.80, 74.17, 73.35, 73.27, 72.84 (6 *C*H_2Ph), 68.94, 68.49, 68.49, 67.90, 67.16 (5 *C*H_2, C-6^{I}, C-6^{II}, C-6^{III}, C-6^{IV}, O*C*H_2CH_2Si), 55.89 (C-2^{III}), 18.34 (*C*H_2Si), −1.50 ($SiMe_3$); CIMS: m/z 1542 (100, $[M + NH_4]^+$). Anal. Calcd for $C_{86}H_{97}NO_{22}Si$: C, 67.74; H, 6.41; N, 0.92. Found: C, 67.62; H, 6.52; N, 0.93.

*2-(Trimethylsilyl) ethyl 2,3,4,6-tetra-*O*-benzoyl-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*-glucopyranosyl-(1→3)-4,6-O-benzylidene-β-*D*-galactopyranosyl-(1→4)-6-O-benzyl-2-deoxy-2-phthalimido-β-*D*-glucopyranosyl-(1→3)-2,6-di-O-benzyl-β-*D*-galactopyranosyl-(1→4)-2,3,6-tri-O-benzyl-β-*D*-glucopyranoside* (**11**).—A mixture of donor **10** (684 mg, 0.46 mmol), acceptor **9** (640 mg, 0.42 mmol), 4 Å powdered molecular sieves (3.5 g) and CH_2Cl_2 (35 mL) was stirred at rt for 30 min. NIS (225 mg, 1 mmol) was added at rt. The reaction mixture was cooled to −20 °C and triflic acid (4 μL, 46 μmol) was added. The reaction mixture was stirred at −20 °C for 1 h, then warmed to rt and stirred overnight. Triethylamine was added, the mixture was filtered through Celite, washed with aq sodium thiosulfate, water, brine, dried over $MgSO_4$ and concentrated. The residue obtained was submitted to flash chromatography (3:2 cyclohexane–EtOAc) to afford **11** (1.17 g, 87%) as an amorphous solid: $[\alpha]_D$ −5° (*c* 0.8, $CHCl_3$); TLC (3:2 cyclohexane–EtOAc): R_f 0.23; ^{1}H NMR (400 MHz, $CDCl_3$): δ 5.85 (d, 1 H, $J_{3,4}$ 3.6 Hz, H-4^{VI}), 5.77 (dd, 1 H, $J_{1,2}$ 8.2, $J_{2,3}$ 10.3 Hz, H-2^{VI}), 5.48, 5.43 (2 d, 2 H, $J_{1,2}$ 8.5 Hz, $J_{1,2}$ 8.3 Hz, H-1^{III}, H-1^{V}), 5.42 (dd, 1 H, H-3^{VI}), 5.35 (s, 1 H, C*H*Ph), 5.12 (d, 1 H, $J_{1,2}$ 3.5 Hz, H-1^{VII}), 5.03 (d, 1 H, H-1^{VI}), 4.95, 4.70 (2 d, 2 H, *J* 10.7 Hz, CH_2Ph), 4.89, 4.71 (2 d, 2 H, *J* 11.0 Hz, CH_2Ph), 1.44 (d, 3 H, $J_{5,6}$ 6.5 Hz, H-6^{VII}), 1.05 (m, 2 H, CH_2Si), 0.03 (s, 9 H, $SiMe_3$); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 167.93, 167.85 (C=O, Phth), 165.81, 165.67, 165.23, 164.64 (4 C=O, Bz), 139.02, 138.85, 138.73, 138.67, 138.40, 138.31, 138.22, 138.16, 137.57, 137.35, 131.62, 131.15, 129.35, 129.12, 128.92 (aromatic C), 133.89, 133.71, 133.50, 133.34, 133.28, 133.25, 129.79, 129.58, 128.70, 128.63, 128.37, 128.21, 128.17, 128.15, 128.13, 127.94, 127.89, 127.85, 127.83, 127.76, 127.73, 127.68, 127.58, 127.42, 127.37, 127.05, 126.57, 126.23, 125.87, 123.27 (aromatic *C*H), 103.74, 102.92, 101.84, 100.27, 99.88, 98.64, 98.63, 96.76 (8 *C*H, *C*HPh, C-1^{I}, C-1^{II}, C-1^{III}, C-1^{IV}, C-1^{V}, C-1^{VI}, C-1^{VII}), 83.75, 82.79, 82.63, 81.80, 79.13, 79.12, 78.40, 77.93, 76.09, 75.97, 75.34, 74.81, 74.75, 74.61, 73.41, 72.50, 72.49, 71.65,

71.34, 69.77, 69.60, 69.38, 68.17, 67.28, 66.74, 66.65 (26 *C*H, ring C), 75.21, 75.00, 74.78, 74.15, 73.69, 73.23, 73.14, 72.83, 72.59, 71.92 (10 *C*H_2Ph), 68.90, 68.43, 68.33, 67.99, 67.87, 67.12 (C-6^{I}, C-6^{II}, C-6^{III}, C-6^{IV}, C-6^{V}, O*C*H_2CH_2Si), 61.29 (C-6^{VI}), 56.06, 55.74 (C-2^{III}, C-2^{V}), 18.33 (*C*H_2Si), 16.80 (C-6^{VII}), −1.50 ($SiMe_3$); FABMS: *m*/*z* 2908 (100, $[M + Li]^+$). Anal. Calcd for $C_{168}H_{170}N_2O_{41}Si$: C, 69.55; H, 5.91; N, 0.97. Found: C, 69.42; H, 6.01; N, 1.05.

2-(Trimethylsilyl) ethyl 2,3,4,6-tetra-O-*benzoyl*-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-*glucopyranosyl*-(1→3)-*2*-O-*acetyl*-*4,6*-O-*benzylidene*-β-D-*galactopyranosyl*-(1→4)-*3*-O-*acetyl*-*6*-O-*benzyl*-*2*-*deoxy*-*2*-*phthalimido*-β-D-*glucopyranosyl*-(1→3)-*4*-O-*acetyl*-*2,6-di*-O-*benzyl*-β-D-*galactopyranosyl*-(1→4)-*2,3,6*-*tri*-O-*benzyl*-β-D-*glucopyranoside* (**12**).—Compound **11** (80 mg, 27 μmol) was acetylated with Ac_2O (2 mL) in pyridine (4 mL) for 3 days at rt. After concentration, the residue was submitted to flash chromatography (3:2 cyclohexane–EtOAc) to afford **12** (81 mg, 97%) as an amorphous solid: $[\alpha]_D$ −3° (*c* 0.8, $CHCl_3$); TLC (3:2 cyclohexane–EtOAc): R_f 0.5; ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.2–6.9 (m, aromatic H), 5.86 (dd, 1 H, $J_{4,5} < 1$, $J_{3,4}$ 3.6 Hz, H-4^{VI}), 5.78 (dd, 1 H, $J_{1,2}$ 8.2, $J_{2,3}$ 10.3 Hz, H-2^{VI}), 5.59 (dd, 1 H, $J_{3,4}$ 9.0, $J_{2,3}$ 10.7 Hz, H-3^{III}), 5.48, (d, 1 H, $J_{1,2}$ 8.0 Hz, H-1^{III}), 5.46 (dd, 1 H, H-3^{VI}), 5.45 (dd, 1 H, $J_{4,5} < 1$, $J_{3,4}$ 3.4 Hz, H-4^{II}), 5.40 (s, 1 H, C*H*Ph), 5.22 (d, 1 H, $J_{1,2}$ 8.3 Hz, H-1^{V}), 5.14 (d, 1 H, $J_{1,2}$ 3.2 Hz, H-1^{VII}), 5.09 (d, 1 H, H-1^{VI}), 2.07, 1.82, 1.60 (3 s, 9 H, 3 Ac), 1.43 (d, 3 H, $J_{5,6}$ 6.4 Hz, H-6^{VII}), 1.05 (m, 2 H, CH_2Si), 0.03 (s, 9 H, $SiMe_3$); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 170.28, 169.95, 168.17 (3 C=O, Ac), 167.45, 167.39 (C=O, Phth), 165.84, 165.66, 165.29, 164.68 (4 C=O, Bz), 139.01, 138.88, 138.76, 138.63, 138.35, 138.28, 138.10, 138.09, 138.04, 137.59, 131.80, 131.09, 129.31, 129.05, 129.03 (aromatic C), 133.85–133.25, 129.76, 129.62, 128.65, 128.24, 128.23, 128.15, 127.95, 127.93–127.71, 127.48, 127.37, 127.03, 126.88, 126.61, 126.36, 126.16 (aromatic *C*H), 102.96, 101.77, 100.57, 100.04, 99.75, 98.82, 98.40, 96.84 (*C*HPh, C-1^{I}, C-1^{II}, C-1^{III}, C-1^{IV}, C-1^{V}, C-1^{VI}, C-1^{VII}), 82.57, 81.66, 79.13, 79.05, 79.05, 78.57, 77.04, 76.40, 75.72, 75.56, 75.46, 74.76, 74.70, 74.52, 74.33, 72.81, 72.32, 71.71, 71.36, 70.26, 70.17, 69.95, 69.78, 68.14, 66.70, 66.28 (26 *C*H, ring C), 75.00, 74.99, 74.82, 74.24, 73.65, 73.36, 73.31, 72.92, 72.62, 71.88 (10 *C*H_2, *C*H_2Ph), 68.35, 68.31, 67.63, 67.22, 67.19, 67.18 (C-6^{I}, C-6^{II}, C-6^{III}, C-6^{IV}, C-6^{V}, O*C*H_2CH_2Si), 61.35 (C-6^{VI}), 55.75, 55.08 (C-2^{III}, C-2^{V}), 20.74, 20.32, 20.26 (3 Ac), 18.33 (*C*H_2Si), 16.84 (C-6^{VII}), -1.50 ($SiMe_3$); FABMS: *m*/*z* 3050 (100, $[M + Na]^+$). Anal. Calcd for $C_{174}H_{176}N_2O_{44}Si$: C, 69.03; H, 5.86; N, 0.92. Found: C, 68.89; H, 5.87; N, 0.85.

2-(Trimethylsilyl) ethyl β-D-*galactopyranosyl*-(1→4)-[*2,3,4*-*tri*-O-*benzyl*-α-L-*fucopyranosyl*-(1→3)]-*6*-O-*benzyl*-*2*-*acetamido*-*2*-*deoxy*-β-D-*glucopyranosyl*-(1→3)-*4,6*-O-*benzylidene*-β-D-*galactopyranosyl*-(1→4)-*6*-O-*benzyl*-*2*-*acetamido*-*2*-*deoxy*-β-D-*glucopyranosyl*-(1→3)-*2,6*-*di*-O-*benzyl*-β-D-*galactopyranosyl*-(1→4)-*2,3,6*-*tri*-O-*benzyl*-β-D-*glucopyranoside* (**13**).—Hydrazine hydrate (1 mL) was added to a stirred solution of compound **11** (240 mg, 82.7 μmol) in 95% aq EtOH (20 mL) and refluxed for 20 h. The solution was concentrated to dryness. The residue was then stirred with Ac_2O (1 mL) in 1:1 CH_2Cl_2–MeOH (10 mL) at rt for 20 h. The solution was concentrated and coevaporated with EtOH. The residue was submitted to flash chromatography on a column of silica gel (12:1 CH_2Cl_2–MeOH), followed by a column of Sephadex (LH20), using 1:1 CH_2Cl_2–MeOH as eluate to yield **13** (134 mg, 70%) as a white amorphous solid: $[\alpha]_D$ −38° (*c* 1, MeOH); TLC (10:1 CH_2Cl_2–MeOH): R_f 0.37; ^{1}H NMR (400 MHz, CD_3OD): δ 7.65–7.37 (m, aromatic H), 5.65 (s, 1 H, C*H*Ph), 5.52 (d, 1 H, $J_{1,2}$ 3.6 Hz, H-1^{VII}), 5.22, 4.81 (2 d, 2 H, *J* 10.6 Hz, C*H*$_2$Ph), 4.17 (dd, 1 H, $J_{2,3}$ 10.5 Hz, H-2^{VII}), 2.19 (s, 3 H, NHAc), 1.91 (s, 3 H, NHAc), 1.38 (d, 3 H, $J_{5,6}$ 6.4 Hz, H-6^{VII}), 1.16 (m, 2 H, CH_2Si), 0.20 (s, 9 H, $SiMe_3$); ^{13}C NMR (100.6 MHz, CD_3OD): δ 174.33, 174.21 (2 C=O, NHAc), 140.75, 140.68, 140.54, 140.35, 140.21, 139.97, 139.94, 139.82, 139.77, 139.69 (aromatic C), 130.02, 129.81, 129.74, 129.73, 129.63, 129.58, 129.55, 129.51, 129.48, 129.47, 129.41, 129.31, 129.24, 129.16, 129.01, 128.96, 128.87, 128.71, 127.66 (aromatic *C*H),

105.24, 104.58, 104.56, 104.42, 103.92, 103.90, 102.22, 98.14 (*C*HPh, C-1^{I}, C-1^{II}, C-1^{III}, C-1^{IV}, C-1^{V}, C-1^{VI}, C-1^{VII}), 84.18, 84.17, 83.33, 82.64, 81.38, 80.28, 80.26, 80.15, 77.85, 77.52, 77.37, 77.12, 76.83, 76.10, 75.92, 75.55, 75.19, 75.18, 75.15, 73.81, 73.08, 70.88, 70.62, 70.38, 68.37, 68.05 (26 *C*H, ring C), 76.71, 76.61, 76.18, 76.17, 74.86, 74.74, 74.61, 74.35, 73.95, 73.88 (10 CH_2Ph), 70.66, 70.25, 70.24, 69.35, 68.53 (C-6^{I}, C-6^{II}, C-6^{III}, C-6^{IV}, C-6^{V}, OCH_2CH_2Si), 63.71 (C-6^{VI}), 58.53, 57.16 (C-2^{III}, C-2^{V}), 24.00, 23.34 (2 CH_3, NHAc), 19.57 (CH_2Si), 17.25 (C-6^{VII}), −0.96 ($SiMe_3$); FABMS: *m*/*z* 2332 (100, $[M+Na]^+$). Anal. Calcd for $C_{128}H_{154}N_2O_{35}Si{\cdot}3H_2O$: C, 65.07; H, 6.82; N, 1.18. Found: C, 65.09; H, 6.64; N, 1.14.

*2-(Trimethylsilyl) ethyl β-*D*-galactopyranosyl-(1→4)-[α-*L*-fucopyranosyl-(1→3)]-2-acetamido-2-deoxy-β-*D*-glucopyranosyl-(1→3)-β-*D*-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-*D*-glucopyranosyl-(1→3)-β-*D*-galactopyranosyl-(1→4)-β-*D*-glucopyranoside* (**1b**). —A mixture of **13** (45 mg, 19.5 μmol), 10% Pd–C (20 mg) in MeOH (5 mL) was stirred for 2 h under hydrogen (150 kPa) and filtered through Celite. The Celite pad was rinsed with MeOH and water. The solution was concentrated and the residue was purified on a Sephadex column (G25-150), using water as eluate. After lyophilization, compound **1b** was obtained as a white amorphous solid (24 mg, 93%): $[\alpha]_D$ −27° (*c* 0.7, water); ^{1}H NMR: see Tables 1 and 2; ^{13}C NMR (100.6 MHz, D_2O): *δ* 175.26, 175.06 (2 C=O, NHAc), 103.28, 103.23, 103.11, 102.92, 102.12, 101.72, 98.97 (C-1^{I}, C-1^{II}, C-1^{III}, C-1^{IV}, C-1^{V}, C-1^{VI}, C-1^{VII}), 82.44, 82.39, 78.61, 78.46, 75.45, 75.26, 75.22, 75.21, 75.10, 75.09, 74.88, 74.87, 73.37, 73.16, 72.81, 72.51, 72.25, 71.39, 70.31, 70.31, 69.54, 68.69, 68.68, 68.64, 68.04, 67.05 (26 *C*H, ring C), 68.81 (OCH_2CH_2Si), 61.86, 61.31, 61.30, 60.37, 60.18, 59.96 (C-6^{I}, C-6^{II}, C-6^{III}, C-6^{IV}, C-6^{V}, C-6^{VI}), 56.30, 55.49 (C-2^{III}, C-2^{V}), 22.59, 22.51 (2 CH_3, NHAc), 17.91 (CH_2Si), 15.65 (C-6^{VII}), −2.20 ($SiMe_3$); FABMS (HR): *m*/*z* 1341.4956 (100, $[M+Na]^+$; calcd for $C_{51}H_{90}N_2O_{35}Si+Na$: 1341.4991).

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