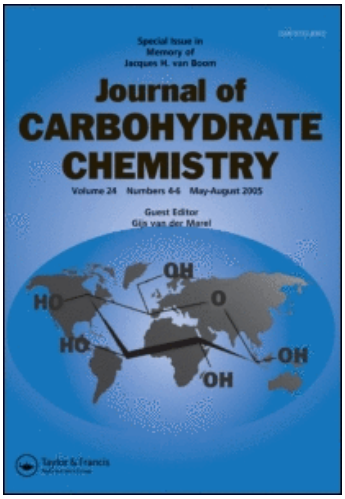

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SYNTHESIS OF THE β-METHYL GLYCOSIDE OF LACTO-*N*-FUCOPENTAOSE III[1]

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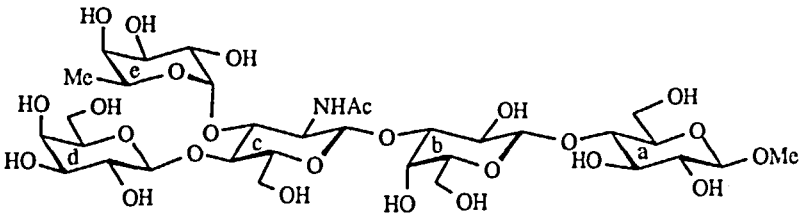
ABSTRACT

A total synthesis of the β-methyl glycoside of lacto-*N*-fucopentaose III (**1**) is described. Phenyl 2,3,4,6-tetra-*O*-benzoyl-β-D-galactopyranosyl-(1→4)-6-*O*-benzyl-2-deoxy-2-phthalimido-1-thio-β-D-glucopyranoside (**4**), a key intermediate prepared by condensation of 2,3,4,6-tetra-*O*-benzoyl-α-D-galactopyranosyl bromide (**2**) and phenyl 6-*O*-benzyl-2-deoxy-2-phthalimido-1-thio-β-D-glucopyranoside (**3**), was glycosylated with ethyl 2,3,4-tri-*O*-benzyl-1-thio-β-L-fucopyranoside (**5**) to give the trisaccharide donor **6**, which, on coupling with methyl 2,6-di-*O*-benzyl-β-D-galactopyranosyl-(1→4) 2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (**7**), afforded the pentasaccharide **8**. It was easily transformed into the target pentasaccharide **1** via hydrazinolysis, acetylation, *O*-deacetylation, and hydrogenolysis.

INTRODUCTION

A specific homotypic interaction between Lewis^x determinants involving either the trisaccharide Lewis^x *per se* or the Lewis^x containing pentasaccharide lacto-*N*-fucopentaose III (LNFP III) has been demonstrated by Hakomori *et al.*.[2] It is mediated by divalent metallic cations such as Ca^{2+}, Mg^{2+}, and Mn^{2+}. This seminal observation, largely based on the study of Lewis^x containing liposomes, may suggest the existence of a novel cell recognition system based on carbohydrate-carbohydrate interaction. Space-filling models and energy minimization calculations of Lewis^x associated with Ca^{2+} have been described,[3,4] but an NMR study[5] in aqueous solution of LNFP III failed to show any binding with Ca^{2+}. Because of the biological importance of this problem, we decided to

undertake the NMR study again and indeed have been able to demonstrate the binding of the synthetic β-methyl glycoside of lacto-*N*-fucopentaose III in methanol to divalent cations.[6] The β-methyl glycoside was selected rather than the free sugar[7] to simplify the NMR spectra. We describe herein a convenient total synthesis of the pentasaccharide **1** used in this NMR work.



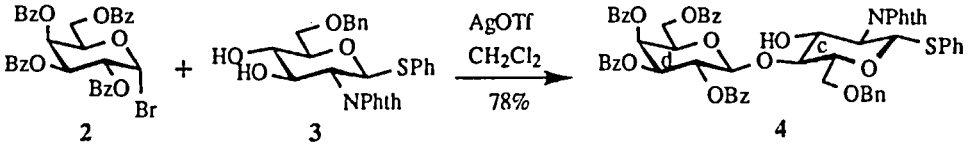
1

RESULTS AND DISCUSSION

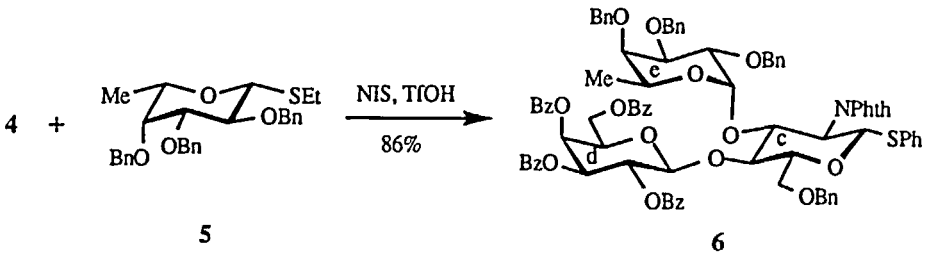
A key intermediate for the synthesis of the pentasaccharide **1** was phenyl 2,3,4,6-tetra-*O*-benzoyl-β-D-galactopyranosyl-(1→4)-6-*O*-benzyl-2-deoxy-2-phthalimido-1-thio-β-D-glucopyranoside (**4**). We found that condensation of 2,3,4,6-tetra-*O*-benzoyl-α-D-galactopyranosyl bromide (**2**)[8] with the diol derivative **3** of *N*-phthaloyl-D-glucosamine[9] in dichloromethane, in the presence of silver triflate at -10 °C for 1.5 h, regioselectively afforded the desired key disaccharide **4** in 78% yield (Scheme 1).

Such a selective behaviour of phenyl 6-*O*-benzyl-2-deoxy-2-phthalimido-1-thio-β-D-glucopyranoside (**3**) has previously been observed during its glycosylation either with 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl fluoride[9] or with 2,3,4-tri-*O*-benzoyl-6-*O*-benzyl-α-D-galactopyranosyl bromide.[10] A very recent example for this type of regioselective glycosylation was reported using phenyl 2,3,4-tri-*O*-acetyl-6-*O*-benzyl-1-thio-β-D-galactopyranoside.[11] The ^{1}H NMR spectrum of **4** showed the presence of H-3 of the glucosamine unit at δ 4.64 (ddd, $J_{3c,OH}$ =1.2 Hz, $J_{3c,4c}$ = 8.6 Hz, $J_{2c,3c}$ = 10.4 Hz), indicating the position of the newly introduced glycosidic linkage in **4** to be at OH-4 of the acceptor **3**. Its stereochemistry was determined to be β on the basis of the Gal H-1, H-2 coupling constant ($J_{1d,2d}$ = 8.1 Hz).

The fucosylation of **4** with ethyl 2,3,4-tri-*O*-benzyl-1-thio-β-L-fucopyranoside **5**[12] in the presence of *N*-iodosuccinimide (NIS) - trifluoromethanesulfonic acid (TfOH)[13] in toluene for 10 min at room temperature provided the expected trisaccharide **6** in 86% yield (Scheme 2). The stereochemistry of the newly introduced glycosidic linkage was determined to be α on the basis of the low value of the Fuc H-1, H-2 coupling constant ($J_{1e,2e}$ = 3.5 Hz).



Scheme 1



Scheme 2

The glycosylation of the diol 7[14] with the donor **6** was achieved under the conditions described above, affording the pentasaccharide **8** in 86% yield (Scheme 3).

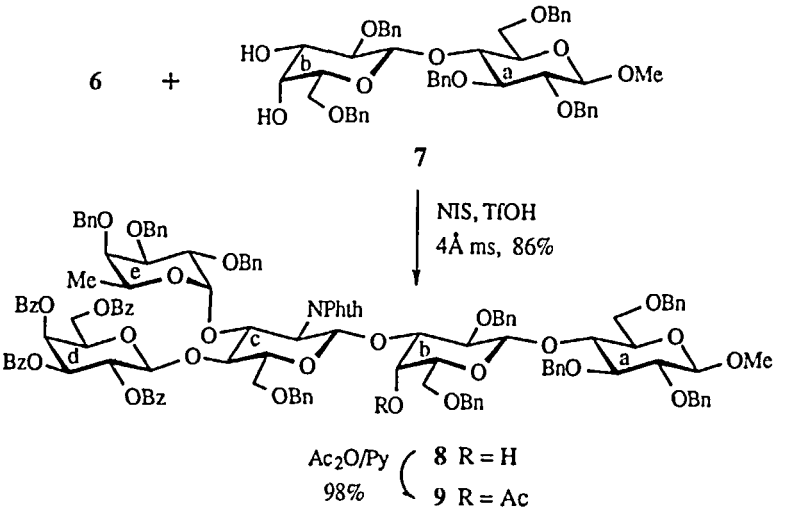
The stereochemistry of the newly introduced linkage was determined to be β on the basis of the GlcN H-1, H-2 coupling constant ($J_{1c,2c}$ = 8.5 Hz). The regiochemistry of **8** was readily assigned from the ^{1}H NMR spectrum of pentasaccharide **9**, obtained from **8** by acetylation, which revealed a deshielded signal for H-4 of the galactose unit (**b** ring) at 5.42 ppm (dd, $J_{4b,5b}$ < 1 Hz, $J_{3b,4b}$ = 3.5 Hz), indicating the position of a new glycosidic linkage in **8** to be at OH-3b of the acceptor **7**.

Treatment of pentasaccharide **8** with hydrazine in boiling ethanol, followed by acetylation, then by *O*-deacetylation, led to the derivative **10** in 81% overall yield from **8**. Catalytic hydrogenolysis of **8** and purification of the product on Sephadex G25-150 afforded the desired pentasaccharide **1** in almost quantitative yield (Scheme 4).

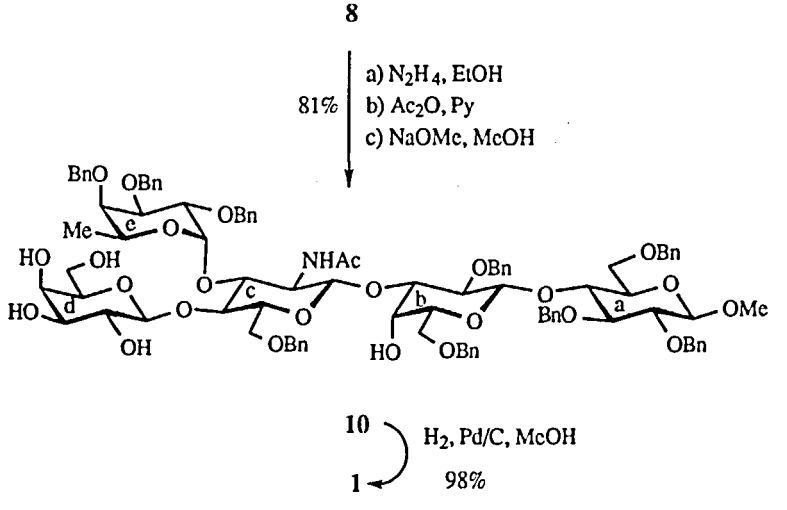
A ^{1}H NMR study of **1** has been published elsewhere.[6]

EXPERIMENTAL

General methods. Melting points were determined with a Büchi model 510 mp 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 Ionisation (CI,



Scheme 3



Scheme 4

ammonia) and FAB mass spectra were obtained with a Nermag R10-10 and a JMS-700 spectrometer, respectively. Elemental analyses were performed by Service Central d'Analyse du C.N.R.S., BP 22, 69390 Vernaison, France, or by Service de Microanalyse de l'Université Pierre et Marie Curie, 4, Place Jussieu, 75005 Paris, France. ^{1}H NMR spectra were recorded with a Bruker AC 250 or a Bruker AM 400 spectrometer at ambient

temperature. Assignments were aided by COSY experiments. ^{13}C NMR spectra were recorded at 62.9 MHz with a Bruker AC 250 or 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 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 sulfuric acid. Flash column chromatography was performed on silica gel 60 (230-400 mesh, E. Merck).

Phenyl 6-*O*-benzyl-2-deoxy-2-phthalimido-4-*O*-(2,3,4,6-tetra-*O*-benzoyl-β-D-galactopyranosyl)-1-thio-β-D-glucopyranoside (4). A mixture of **2** (3.68 g, 5.58 mmol), **3** (2.74 g, 5.58 mmol), 4Å powdered molecular sieves (6.4 g) and dry CH_2Cl_2 (64 mL) was stirred at room temperature for 30 min, then cooled to -10 °C under argon. Silver triflate (1.73 g, 6.73 mmol, 1.2 equiv) was added. Stirring was continued at -10 °C for 1.5 h. The mixture was neutralized with triethylamine, filtered through celite (eluting with CH_2Cl_2), washed with water, aqueous sodium thiosulfate, water, brine, dried over $MgSO_4$ and concentrated. The residue was flash chromatographed (toluene/AcOEt 10/1) to give **4** (4.67 g, 78%): mp 180-181 °C (from ether); $[\alpha]_D$ +99° (*c* 1.0, chloroform); ^1H NMR (400 MHz, $CDCl_3$): δ 8.08-7.19 (m, 34H, arom.), 5.96 (dd, 1H, $J_{4,5} < 1$ Hz, $J_{3,4} = 3.4$ Hz, H-4d), 5.87 (dd, 1H, $J_{1,2} = 8.1$ Hz, $J_{2,3} = 10.5$ Hz, H-2d), 5.60 (d, 1H, $J_{1,2} = 10.4$ Hz, H-1c), 5.59 (dd, 1H, H-3d), 4.94 (d, 1H, H-1d), 4.69 (m, 1H, H-6d), 4.64 (ddd, 1H, $J_{3,OH} = 1.4$ Hz, $J_{3,4} = 8.6$ Hz, $J_{2,3} = 10.4$ Hz, H-3c), 4.47 (d, 1H, OH-3c), 4.37 (dd, 1H, H-2c), 4.36 (m, 2H, H-5d, H-6'd), 4.26, 4.17 (2d, 2H, J = 12.0 Hz, PhC$\underline{H}_2$), 3.87 (dd, $J_{4,5} = 9.7$ Hz, H-4c), 3.72 (ddd, $J_{5,6} = 2.5$ Hz, $J_{5,6'} = 3.2$ Hz, H-5c), 3.53 (m, 2H, H-6c, H-6'c); ^{13}C NMR (62.9 MHz, $CDCl_3$): δ 168.20, 167.52 (2 C=O, Phth), 167.52, 166.16, 165.50, 165.06 (4 C=O, Bz), 138.28, 134.08, 133.80, 133.69, 133.45, 133.30, 132.41, 132.27, 131.78, 131.62, 130.27, 130.00, 129.90, 129.73, 129.07, 128.92, 128.86, 128.72, 128.66, 128.53, 128.48, 128.37, 128.26, 127.83, 127.63, 127.46, 125.34, 123.64, 123.30 (aromatic C, aromatic $\underline{C}$H), 102.02 (C-1d), 83.47 (C-1c), 82.69 (C-4c), 77.88 (C-5c), 72.95 (Ph$\underline{C}H_2$), 72.39 (C-5d), 71.51 (C-3d), 70.98 (C-3c), 69.57 (C-2d), 66.33 (C-6c), 66.14 (C-4d), 62.82 (C-6d), 55.10 (C-2c). MS (CI, NH_3): *m/z* 1087 $(M+NH_4)^+$.

Anal. Calcd for $C_{61}H_{51}NO_{15}S$ (1070.143): C, 68.46; H, 4.80; N, 1.31. Found C, 68.48; H, 4.73; N, 1.32.

Phenyl 6-*O*-benzyl-2-deoxy-2-phthalimido-4-*O*-(2,3,4,6-tetra-*O*-benzoyl-β-D-galactopyranosyl)-1-thio-3-*O*-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyl)-β-D-glucopyranoside (6). A mixture of **4** (2.49 g, 2.33 mmol), **5** (2.56 g, 5.35 mmol, 2.3 equiv), 4Å powdered molecular sieves (5.1 g) and dry toluene (125 mL) was stirred at room temperature for 30 min. NIS (1.21 g, 5.37 mmol) then TfOH (48 μL, 0.54 mmol) were added at room temperature (**4** is not soluble in toluene at lower temperature). The reaction

mixture was neutralized (Et_3N) after 10 min at room temperature, diluted with dichloromethane, filtered through celite, washed with aqueous thiosulfate, water, brine, dried over $MgSO_4$ and concentrated. The residue was flash chromatographed (toluene/AcOEt 10/1 then cyclohexane/AcOEt 3/1) to give **6** (2.99 g, 86%) as an amorphous powder: $[\alpha]_D$ +17° (*c* 1.0, chloroform); ^1H NMR (400 MHz, $CDCl_3$): δ 8.16-7.18 (m, 49H, arom.), 5.86 (dd, 1H, $J_{4,5}$ < 1 Hz, $J_{3,4}$ = 3.6 Hz, H-4d), 5.77 (dd, 1H, $J_{1,2}$ = 8.3 Hz, $J_{2,3}$ = 10.3 Hz, H-2d), 5.50 (d, 1H, $J_{1,2}$ = 10.6 Hz, H-1c), 5.44 (dd, 1H, H-3d), 5.12 (d, 1H, $J_{1,2}$ = 3.5 Hz, H-1e), 5.10 (d, 1H, H-1d), 4.88 (d, 1H, J = 12.0 Hz, PhC$\underline{H}_2$), 4.87 (dd, 1H, $J_{3,4}$ = 8.7 Hz, $J_{2,3}$ = 10.1 Hz, H-3c), 4.75 (dq, 1H, $J_{4,5}$ < 1 Hz, $J_{5,6}$ = 6.5 Hz, H-5e), 4.72 (d, 1H, J = 11.3 Hz, PhC$\underline{H}_2$), 4.55-4.47 (m, 4H, H-2c, H-6d, PhC$\underline{H}_2$), 4.42-4.37 (m, 3H, H-6'd, PhC$\underline{H}_2$), 4.35 (d, 1H, J = 12.3 Hz, PhC$\underline{H}_2$), 4.31 (dd, 1H, $J_{4,5}$ = 10.1 Hz, H-4c), 4.26 (d, 1H, J = 11.1 Hz, PhC$\underline{H}_2$), 3.97-3.92 (m, 3H, H-3e, H-5d, H-6c), 3.85 (dd, 1H, $J_{2,3}$ = 10.2 Hz, H-2e), 3.74 (m, 1H, H-6'c), 3.62 (dd, 1H, $J_{4,5}$ < 1 Hz, $J_{3,4}$ = 2.5 Hz, H-4e), 3.49 (m, 1H, H-5c), 1.45 (d, 3H, H-6e); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 165.79, 165.71, 165.27, 164.67 (4 C=O, Bz), 138.88, 138.73, 138.15, 137.78, 131.62, 129.39, 128.90 (aromatic C), 134.09, 133.48, 133.45, 133.30, 133.24, 132.27, 132.23, 129.81, 129.61, 129.58, 128.71, 128.67, 128.63, 128.59, 128.48, 128.22, 128.17, 128.00, 127.96, 127.95, 127.87, 127.76, 127.67, 127.17, 127.08, 126.90, 126.62, 123.56 (aromatic $\underline{C}$H), 99.81 (C-1d), 96.87 (C-1e), 84.02 (C-1c), 79.15 (C-3e), 79.13 (C-4e), 78.93 (C-5c), 75.40 (C-4c), 75.29 (C-2e), 73.51 (C-3c), 71.71 (C-3d), 71.30 (C-5d), 69.74 (C-2d), 68.20 (C-4d), 66.69 (C-5e), 75.04, 73.61, 72.75, 71.89 (4 Ph$\underline{C}H_2$), 67.67 (C-6c), 61.23 (C-6d), 55.34 (C-2c), 16.80 (C-6e). MS (CI, NH_3): *m/z* 1504 $(M+NH_4)^+$.

Anal. Calcd for $C_{88}H_{79}NO_{19}S$ (1486.661): C, 71.10; H, 5.36; N, 0.94. Found C, 71.12; H, 5.29; N, 0.96.

Methyl (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)-(2,6-di-*O*-benzyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (8). A mixture of **6** (2.85 g, 1.92 mmol), **7** (2.32 g, 2.87 mmol, 1.5 equiv), 4Å powdered molecular sieves (5.2 g) and CH_2Cl_2 (52 mL) was stirred at room temperature for 30 min. NIS (1.3 g, 5.77 mmol) was added at room temperature. The reaction mixture was cooled at -60 °C. Triflic acid (77 μL, 0.86 mmol) was added. The reaction mixture was stirred at -60 °C for 1 h, neutralized (Et_3N), filtered through celite, washed with aqueous thiosulfate, water, brine, dried ($MgSO_4$) and concentrated. The residue was flash chromatographed (toluene/AcOEt 6.5/1) to give **8** (3.62 g, 86%) as an amorphous powder: $[\alpha]_D$ +10° (*c* 1.0, chloroform); ^1H NMR (400 MHz, $CDCl_3$): δ 8.20-6.80 (m, 69H, arom.), 5.87 (dd, 1H, $J_{4,5}$ < 1 Hz, $J_{3,4}$ = 3.6 Hz, H-4d), 5.77 (dd, 1H, $J_{1,2}$ = 8.2 Hz, $J_{2,3}$ = 10.3 Hz, H-2d), 5.45 (dd, 1H, H-3d), 5.32 (d, 1H, $J_{1,2}$ = 8.5 Hz, H-1c), 5.11

(d, 1H, $J_{1,2}$ = 3.4 Hz, H-1e), 5.07 (d, 1H, H-1d), 4.94 (d, 1H, J = 10.5 Hz, PhC$\underline{H}_2$), 4.85 (d, 1H, J = 10.7 Hz, PhC$\underline{H}_2$), 4.82 (d, 1H, J = 11.4 Hz, PhC$\underline{H}_2$), 4.73-4.67 (m, 4H, H-5e, PhC$\underline{H}_2$), 4.50-4.40 (m, 7H, H-2c, H-6d, H-6'd, PhC$\underline{H}_2$), 4.25 (d, 1H, $J_{1,2}$ = 7.5 Hz, H-1a), 4.17 (d,1H, $J_{1,2}$ = 7.6 Hz, H-1b), 4.04-4.00 (m, 2H, H-4b, H-5d), 3.81 (dd, 1H, $J_{2,3}$ = 10.1 Hz, H-2e), 3.51 (s, 3H, OMe), 2.72 (br, 1H, OH-4b), 1.43 (d, 3H, $J_{5,6}$ = 6.5 Hz, H-6e); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 165.88, 165.76, 165.34, 164.73 (4 C=O, Bz), 138.45, 133.35, 129.66, 128.81, 128.36, 128.22, 128.20, 128.01, 127.98, 127.92, 127.86, 127.85, 127.75, 127.70, 127.46, 127.44, 126.67, 123.24 (aromatic C), 104.48 (C-1b), 101.90 (C-1a), 99.85 (C-1d), 98.82 (C-1c), 96.80 (C-1e), 83.27, 82.68, 81.70, 79.14, 79.11, 77.97, 75.75, 75.63, 75.58, 74.59, 74.53, 72.60, 72.50, 71.75, 71.42, 69.87, 68.22, 67.48, 66.77 (19 ring $\underline{C}$H), 75.36, 75.09, 74.81, 74.07, 73.86, 73.28, 72.92, 72.75, 71.92 (9 Ph$\underline{C}H_2$), 68.38, 67.80, 67.68 (C-6a, C-6b, C-6c), 61.39 (C-6d), 56.91 (OMe), 56.17 (C-2c), 16.86 (C-6e). MS (CI, NH_3): *m/z* 2201 $(M+NH_4)^+$.

Anal. Calcd for $C_{130}H_{127}NO_{30}$ (2183.444): C, 71.51; H, 5.86; N, 0.64. Found: C, 71.56; H, 5.95; N, 0.59.

Methyl (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-*O*-acetyl-2,6-di-*O*-benzyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (9). Compound **8** (38 mg, 17.4 μmol) was acetylated with acetic anhydride (1 mL) in pyridine (2 mL) for 18 h at room temperature. After concentration, the residue was flash chromatographed (cyclohexane/AcOEt 2.5/1) to give **9** (38 mg, 98%) as an amorphous powder: $[\alpha]_D$ +2.5° (*c* 1.1, chloroform); ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.14-6.89 (m, 69H, arom.), 5.91 (dd, 1H, $J_{4,5}$ < 1 Hz, $J_{3,4}$ = 3.6 Hz, H-4d), 5.78 (dd, 1H, $J_{1,2}$ = 8.3 Hz, $J_{2,3}$ = 10.3 Hz, H-2d), 5.48 (dd, 1H, H-3d), 5.42 (dd, 1H, $J_{4,5}$ < 1 Hz, $J_{3,4}$ = 3.6 Hz, H-4b), 5.25 (d, 1H, $J_{1,2}$ = 8.2 Hz, H-1c), 5.20 (d, 1H, H-1d), 5.08 (d, 1H, $J_{1,2}$ = 3.5 Hz, H-1e), 5.01, 4.58 (2d, 2H, J = 12.0 Hz, PhC$\underline{H}_2$), 4.94, 4.69 (2d, 2H, J = 10.5 Hz, PhC$\underline{H}_2$), 4.72 (m, 1H, H-5e), 4.49, 4.23 (2d, 2H, J = 12.2 Hz, PhC$\underline{H}_2$), 4.43 (dd, 1H, $J_{2,3}$ = 10.8 Hz, H-2c), 4.30-4.25 (m, 5H, H-1a, PhC$\underline{H}_2$), 4.18 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1b), 3.90 (dd, 1H, $J_{3,4}$ = 2.8 Hz, $J_{2,3}$ = 10.2 Hz, H-3e), 3.81 (dd, 1H, H-2e), 3.52 (s, 3H, OMe), 3.47 (dd, 1H, $J_{3,4}$ = 3.6 Hz, $J_{2,3}$ = 10.1 Hz, H-3b), 2.11 (s, 3H, OAc), 1.45 (d, 3H, $J_{5,6}$ = 6.5 Hz, H-6e); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 169.90 (C=O, Ac), 165.75, 165.74, 165.33, 164.76 (4 C=O, Bz), 138.97, 138.94, 138.82, 138.60, 138.27, 138.21, 138.17, 138.09 (aromatic C), 133.70, 133.43, 133.36, 133.21 (aromatic $\underline{C}$H), 131.26 (aromatic C), 129.87, 129.68, 129.60 (aromatic $\underline{C}$H), 129.46, 129.26, 129.02, 128.69 (aromatic C), 128.62, 128.57, 128.52, 128.22, 128.20, 128.08, 128.02, 127.98, 127.95, 127.92, 127.86, 127.81, 127.80, 127.74, 127.72, 127.48, 127.41, 127.05, 126.69, 126.23, 123.18 (aromatic $\underline{C}$H), 104.50 (C-1b), 101.85 (C-1a), 99.77 (C-1d), 98.85 (C-1c),

96.53 (C-1e), 82.46, 81.56, 79.19, 79.16, 78.96, 78.73, 75.50, 75.35, 75.28, 74.85, 74.52, 72.54, 72.15, 71.82, 71.27, 69.80, 69.79, 68.19, 66.63 (19 ring-CH), 75.10, 75.07, 74.82, 74.12, 73.60, 73.43, 72.96, 72.57, 71.98 (9 Ph CH_2), 68.26, 67.53, 67.40 (C-6a, C-6b, C-6c), 61.20 (C-6d), 56.92 (OMe), 56.48 (C-2c), 20.76 (Ac), 16.84 (C-6e). MS (CI, NH_3): *m/z* 2243 $(M+NH_4)^+$.

Anal. Calcd for $C_{132}H_{129}NO_{31}$ (2225.472): C, 71.24; H, 5.84; N, 0.63. Found: C, 71.04; H, 5.89; N, 0.57.

Methyl (β-D-galactopyranosyl)-(1→4)-[(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyl)-(1→3)] - (2-acetamido - 6 -*O*-benzyl-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 (10). A solution of **8** (2.3 g, 1.05 mmol) water (4 mL), hydrazine (4 mL) and ethanol (90 mL) was refluxed for 16 h. TLC showed a complete disappearance of **8**, the reaction mixture was concentrated. A solution of the residue in CH_2Cl_2/MeOH (1/1) was filtered and concentrated. To the residue, acetic anhydride (10 mL) and pyridine (20 mL) was added and the solution was stirred overnight at room temperature, then concentrated. The residue was then stirred for 24 h in 0.1 M NaOMe in methanol, neutralized (IR-120, H^+ form), filtered, concentrated. The residue was flash chromatographed (toluene/acetone 1/1.2) to give **10** (1.44 g, 81%) as an amorphous powder: $[\alpha]_D$ -19° (*c* 1.0, chloroform); ^1H NMR (400 MHz, $CDCl_3$): δ 7.5-7.2 (m, 45H, arom.), 5.78 (d, 1H, $J_{2c,NH}$ = 7.1 Hz, NH), 5.18 (d, 1H, $J_{1,2}$ = 7.4 Hz, H-1), 5.08 (d, 1H, $J_{1,2}$ = 3.5 Hz, H-1e), 5.02, 4.77 (2d, 2H, J = 10.7 Hz, PhCH_2), 4.95, 4.59 (2d, 2H, J = 11.5 Hz, PhCH_2), 4.91, 4.68 (2d, 2H, J = 11.8 Hz, PhCH_2), 4.89, 4.74 (2d, 2H, J = 10.9 Hz, PhCH_2), 4.52-4.41 (m, 5H, 2xH-1, PhCH_2), 4.29 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1), 4.15 (dq, 1H, $J_{4,5}$ < 1 Hz, $J_{5,6}$ = 6.4 Hz, H-5e), 4.08 (dd, 1H, $J_{2,3}$ = 10.2 Hz, H-2e), 3.92 (dd, 1H, $J_{3,4}$ = 2.6 Hz, H-3e), 3.70 (dd, 1H, $J_{4,5}$ < 1 Hz, H-4e), 3.58 (s, 3H, OMe), 3.47 (dd, 1H, $J_{3,4}$ = 3.6 Hz, $J_{2,3}$ = 10.1 Hz, H-3b), 1.40 (s, 3H, NAc), 1.14 (d, 3H, H-6e); ^{13}C NMR (100.6 MHz, $CDCl_3$): δ 170.91 (C=O, NAc), 138.95, 138.75, 138.59, 138.39, 138.35, 138.31, 138.18, 138.07, 137.45 (aromatic C), 128.49, 128.41, 128.35, 128.30, 128.23, 128.19, 128.16, 128.14, 128.06, 127.95, 127.91, 127.72, 127.62, 127.57, 127.44, 127.35, 127.33, 127.05 (aromatic CH), 104.49, 102.06, 100.68, 99.83 (4C-1), 97.81 (C-1e), 82.67, 81.69, 81.68, 79.24, 79.03, 77.25, 76.25, 76.17, 75.82, 75.17, 74.92, 74.91, 74.48, 73.86, 72.89, 71.51, 69.13, 67.81, 67.21 (19 ring-CH), 75.29, 74.85, 74.79, 74.57, 73.86, 73.29, 73.28, 73.06, 72.24 (9 Ph CH_2), 69.48, 68.70, 68.05 (C-6a, C-6b, C-6c), 62.56 (C-6d), 56.94 (OMe), 56.93 (C-2c), 22.84 (NAc), 16.60 (C-6e). MS (CI, NH_3): *m/z* 1697 $(M+NH_4)^+$.

Anal. Calcd for $C_{96}H_{111}NO_{25}\cdot 1.5H_2O$ (1705.960): C, 67.59; H, 6.74; N, 0.82. Found: C, 67.60; H, 6.78; N, 0.69.

Methyl (β-D-galactopyranosyl) - (1→4) - [(α-L-fucopyranosyl) - (1→3)] - (2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1→3) - (β-D-galactopyranosyl)-(1→4)-β-D-glucopyranoside (1). A mixture of **10** (1.08 g), 10% Pd/C (0.2 g) and methanol (45 mL) was stirred for 2 h under hydrogen (150 kPa), then filtered through celite. The celite pad was rinsed with methanol and water. The solution was concentrated, and the residue was purified on a Sephadex column (G25-150), using water as eluant. After lyophilization, compound **1** was obtained as a white amorphous solid (0.55 g, 98%): $[\alpha]_D$ -49° (*c* 0.83, methanol); By use of a combination of homonuclear and heteronuclear 2D NMR techniques (COSY, RELAYs, TOCSY, HMQC, HMBC), the ^1H and ^{13}C spectra of compound **1** were completely assigned.[6] HRMS (FAB) Calcd for $C_{33}H_{57}NO_{25}Na$ $(M+Na)^+$: 890.3117. Found: 890.3123.

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