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A first total synthesis of a novel sulfated ganglioside, 3′-*O*-sulfo-GM1b☆

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In honor of Professor Guido Tettamanti

Abstract

A first total synthesis of a novel sulfated ganglioside, 3′-*O*-sulfo-GM1b, is described. The suitably protected gangliotriose (GgOSe3) derivative, 2-(trimethylsilyl)ethyl (2-acetamido-4,6-*O*-benzylidene-2-deoxy-β-D-galactopyranosyl)-(1→4)-(2,6-di-*O*-benzyl-3-*O*-*p*-methoxybenzyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-D-glucopyranoside was glycosylated with the α-NeuAc-(2→3)-galactose donor to give the protected GM1b oligosaccharide (95%). After proper manipulation of the protecting groups, the oligosaccharide was converted into the target ganglioside by the successive introduction of the ceramide and sulfo groups, followed by complete deprotection. © 2002 Elsevier Science Ltd. All rights reserved.

Keywords: Sulfated ganglioside; 3′-*O*-sulfo-GM1b; Chemical synthesis

1. Introduction

Gangliosides, sialic acid-containing glycosphingolipids, are a class of structurally diverse molecules commonly present in cell-surface membranes and are particularly rich in tissues of central nervous system. It has been widely recognized that gangliosides are involved in many biological processes such as cell growth, cell differentiation, cell adhesion, immune response, oncogenesis, and many other receptor-mediated reactions.[2–5] We have succeeded in the total syntheses of a variety of gangliosides including their analogs and derivatives, and have contributed to the elucidation of their biological functions at the molecular level.[6,7] Recently, a novel series of gangliosides that contain both sialic acid and sulfate were isolated from mammals. One is sulfated GM1a [HSO_3-3Galβ1-3GalNAcβ1-4(NeuGcα2-3)Galβ1-4Glcβ1-1Cer] from rat kidney,[8] and another is 3′-*O*-sulfo-GM1b (**1**) from bovine cauda equina.[9] However, the biological functions of these molecules have not been elucidated because of insufficient availability of materials. In this paper, as a part of our study to contribute to glycobiology by a chemical approach, a first total synthesis of the 3′-*O*-sulfo-GM1b ganglioside is described.

2. Results and discussion

Fig. 1 illustrates the synthetic plan (retrosynthesis) for the target compound. Lactose acceptor **6** and galactosamine donor **4** were obtained in high yield from the known compound **5**[10] and **2**,[10] respectively. Sialylgalactose donor **9**[11] has already been successfully applied to the syntheses of a series of gangliosides.[6,7]

Coupling of **6** with **4** was carried out in the presence of dimethyl(methylthio)sulfonium triflate (DMTST)[12] in CH_2Cl_2 at room temperature to give **7** in 65% yield (Scheme 1). The *N*-phthaloyl and *O*-acetyl group in **7** were simultaneously cleaved by treatment with hydrazine monohydrate in ethanol, and the resulting free amino group was selectively acetylated to afford the trisaccharide acceptor **8** in high yield. Stereoselective glycosylation at O-3 of GalNAc residue in **8** was car-

☆ Synthetic studies on sialoglycoconjugates, Part 127. For Part 126, see Ref. 1.

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ried out by employing **9** as the glycosyl donor and DMTST as the glycosyl promotor in dichloromethane to yield the desired pentasaccharide **10** in 95% yield. After hydrolytic removal of the benzylidene group (**11**) and the successive O-acetylation (**12**), the *p*-methoxybenzyl group was cleaved by treatment with ceric ammonium nitrate (CAN) in acetonitrile–water.[13,14] The resulting **13** was protected with the levulinyl group[15] with levulynic anhydride and 4-dimethylaminopyridine (DMAP) in pyridine, which can be chemoselectively removed at the final stage of the synthetic strategy to undergo sulfation.

Hydrogenolytic removal of the benzyl group in **14** over $Pd(OH)_2$ in ethanol, followed by complete acetylation of the resulting free hydroxyls with acetic anhydride, DMAP and pyridine, afforded the fully acylated oligosaccharide **16** (Scheme 2). Selective removal of the 2-(trimethylsilyl)ethyl (SE) group[16] was achieved by treatment of **16** with trifluoroacetic acid in dichloromethane to give the 1-hydroxy compound **17** (100%), which upon further treatment[17] with trichloroacetonitrile in the presence of 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) in dichloromethane, gave the trichloroacetimidate **18** in a quantitative yield.

Coupling of **18** with (2*S*,3*R*,4*E*)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol (**19**)[18,19] was carried out in the presence of borontrifluoride etherate and 4 Å molecular sieves (AW-300) in dichloromethane to give **20** in 31% yield. Selective reduction[20] of the azido function in **20** with hydrogen sulfide in 83% aqueous pyridine for 48 h at 0 °C, and subsequent condensation with octadecanoic acid using 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (a water-soluble carbodiimide) in dichloromethane, furnished the corresponding ganglioside derivative **21** in 68% yield. Selective removal[21] of the levulinyl group from **21** was carried out in ethanol with hydrazine monoacetate at room temperature to give **22** in 80% yield. Treatment[22] of **22** with the sulfur trioxide·pyridine complex in DMF for 24 h at room temperature afforded the corresponding sulfate **23** in 80% yield as their pyridinium salt. O-Deacylation of **23** with sodium methoxide in methanol, with subsequent saponification of the sialic acid methyl ester group, yielded the desired 3′-*O*-sulfo-GM1b ganglioside in 89% yield as their sodium salts. The NMR data were consistent with that reported[8] for the natural product.

In conclusion, an efficient total synthesis of 3′-*O*-sulfo-GM1b ganglioside was achieved based on the rationally designed, synthetic strategy.

3. Experimental

General methods.—Specific rotations were determined with a Horiba SEPA-300 high sensitive polarimeter at 25 °C, and ^{1}H NMR spectra were recorded on Varian Unity Inova (400 and 500 MHz) spectrometers

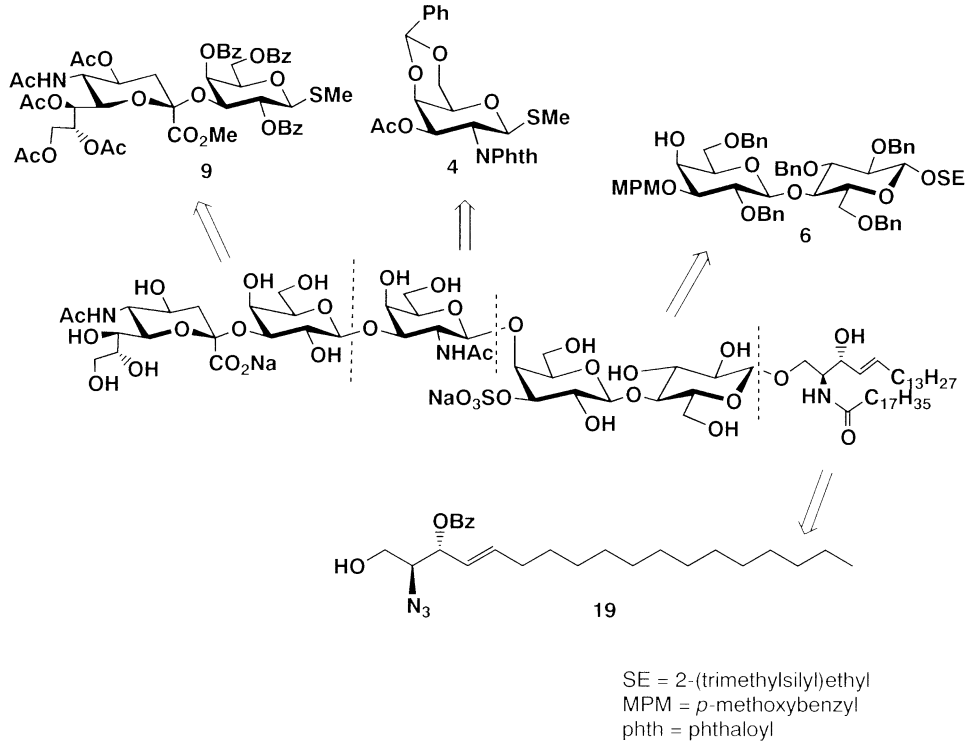
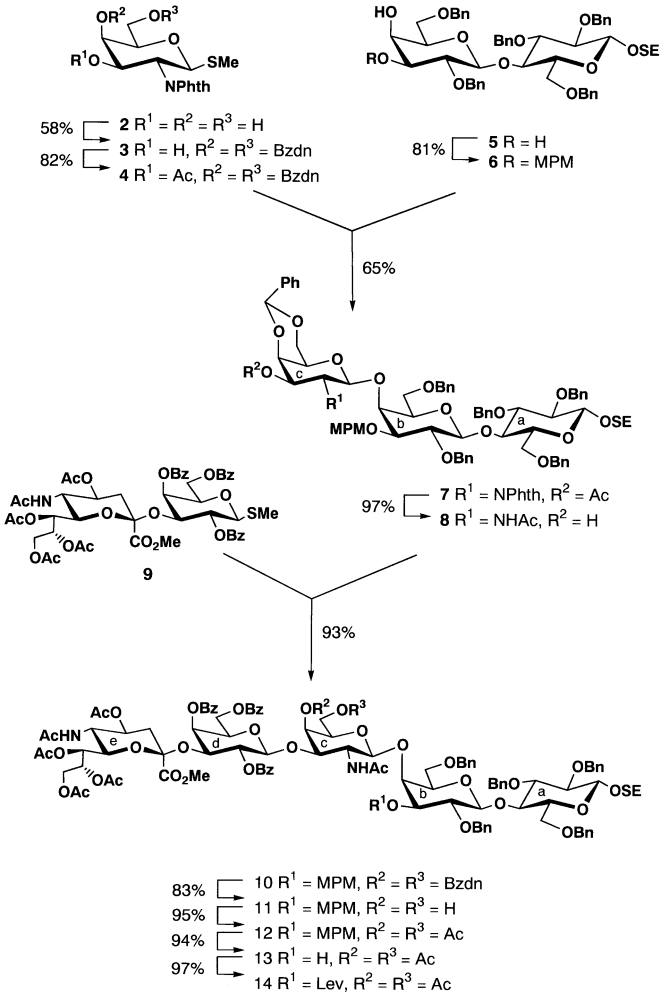


Figure 1. Retrosynthetic analysis for the synthesis of 3′-*O*-sulfo-GM1b (**1**).

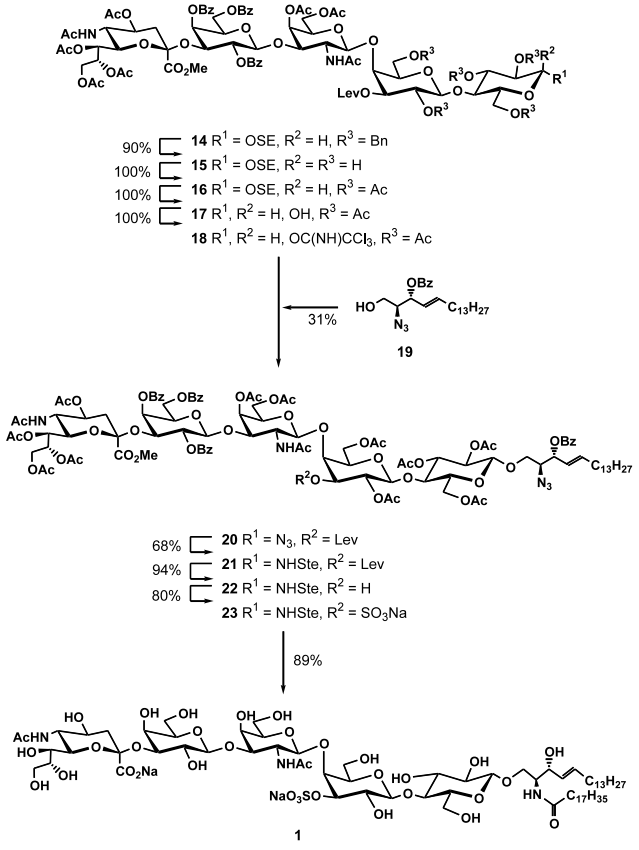


Scheme 1.

with TMS as the internal standard. FABMSs were recorded on a JEOL JMS-SX 120A mass spectrometer/JMA-DA 7000 data system. Preparative thin-layer chromatography (TLC) was performed on Silica Gel 60 (E. Merck), and column chromatography on Silica Gel (Fuji Silysia Co., 300 mesh) was performed with the specified solvent systems (v/v). Concentrations and evaporations were conducted in vacuo.

*2-(Trimethylsilyl)ethyl (3-*O*-acetyl-4,6-*O*-benzylidene-2-deoxy-2-phthalimido-β-*D*-galactopyranosyl)-(1→4)-(2,6-di-*O*-benzyl-3-*O*-p-methoxybenzyl-β-*D*-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-*D*-glucopyranoside* (**7**).—To a solution of **5** (100 mg, 98.6 μmol) and **4** (116 mg, 246 μmol), in dry CH_2Cl_2 (10 mL) was added 4 Å molecular sieves (216 mg), and the mixture was stirred for 1 h at room temperature, then cooled to 0 °C. Dimethyl(methylthio)sulfonium triflate (DMTST; 53.8% (w/w), 355 mg, 1.37 mmol) was added to the mixture, and the resultant mixture was stirred for 2 h at room temperature. After dilution with $CHCl_3$, the precipitate was filtered off and washed with $CHCl_3$. The filtrate and washings were combined and successively washed with M Na_2CO_3 and water, dried (Na_2SO_4) and concentrated. Column chromatography (1:3 EtOAc–hexane) of the residue on silica gel afforded **7** (91.6 mg, 65%) as an amorphous mass: $[\alpha]_D$ +24.3° (*c* 1.4, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 0.12 (s, 9 H, Me_3Si), 1.01–1.06 (m, 2 H, $SiCH_2CH_2O$-), 1.98 (s, 3 H, OAc), 3.78 (d, 1 H, $J_{3,4}$ 7.0 Hz H-4b), 3.81 (s, 3 H, $PhOCH_3$), 4.85 (t, 1 H, $J_{1,2} = J_{2,3}$ 8.4 Hz, H-2c), 5.37 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1c), 5.60 (s, 1 H, benzylidene), 6.07 (m, 1 H, H-3c), 7.15–7.89 (m, 38 H, 8 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz: δ 170.6 (C=O), 186.5 (C=O), 167.3 (C=O), 139.0, 138.9, 137.9, 133.9, 133.7, 132.8, 131.6, 130.3, 130.1, 129.2, 128.8, 128.3, 128.2, 128.1, 128.0, 127.5, 127.4, 127.3, 127.2, 126.2, 123.4, 123.1, 113.7, 103.2 (O-C-O), 102.3 (O-C-O), 100.1 (O-C-O), 99.4 (O-C-O), 82.9, 81.9, 80.3, 80.1, 76.7, 75.2, 75.1, 74.5, 74.3, 73.1, 73.1, 72.9, 72.8, 72.3, 68.7, 68.3, 67.2,66.4, 55.3, 51.3, 20.9, 18.5. Anal. Calcd for $C_{18}H_{91}NO_{19}Si$ (1434.72): C, 69.49; H, 6.39; N, 0.98. Found: C, 69.48; H, 6.30; N, 0.85.

*2-(Trimethylsilyl)ethyl (2-acetamido-4,6-*O*-benzylidene-2-deoxy-β-*D*-galactopyranosyl)-(1→4)-(2,6-di-*O*-benzyl-3-*O*-p-methoxybenzyl-β-*D*-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-*D*-glucopyranoside* (**8**).—A solution of **7** (70 mg, 49.1 μmol) in EtOH (7 mL) and hydrazine monohydrate (47.6 μL, 951 μmol) was heated



Scheme 2.

for 30 h under reflux. After cooling, insoluble materials were filtered off and washed with EtOH. The filtrate and washings were combined and concentrated to dryness. After acetylation with acetic anhydride and pyridine in methanol, column chromatography (3:2 EtOAc–hexane) of the residue on silica gel gave **8** (59.4 mg, 93%) as an amorphous mass: $[\alpha]_D$ +67.6° (*c* 0.6, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 0.02 (s, 9 H, Me_3Si), 1.01–1.06 (m, 2 H, $SiCH_2CH_2O$-), 1.65 (s, 3 H, NHAc), 3.56 (m, 1 H, H-2c), 3.81 (s, 3 H, $PhOCH_3$), 4.13 (m, 1 H, H-3c), 4.50 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1c), 5.60 (s, 1 H, benzylidene), 5.82 (d, 1 H, $J_{2,NH}$ 2.9 Hz, NH), 7.17–7.58 (m, 34 H, 7 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 173.8 (C=O), 160.1, 138.9, 138.6, 138.4, 138.2, 137.9, 130.4, 128.9, 128.7, 128.5, 128.4, 128.3, 128.2, 127.9, 127.7, 127.7, 127.6, 127.5, 127.4, 127.4, 126.6, 114.2, 103.2 (O-C-O), 103.2 (O-C-O), 102.7 (O-C-O), 101.4 (O-C-O), 82.8, 82.2, 82.0, 77.0, 76.1, 75.6, 75.4, 75.2, 75.1, 74.8, 74.7, 73.2, 73.1, 72.8, 69.0, 68.3, 68.0, 67.4, 67.2, 55.4, 55.3, 22.5, 18.5. Anal. Calcd for $C_{75}H_{89}NO_{17}Si$ (1304.61): C, 69.05; H, 6.88; N, 1.07. Found: C, 68.86; H, 6.58; N, 1.05.

2-(Trimethylsilyl)ethyl (methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6*-O-*benzylidene-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*benzyl-3*-O-p-*methoxybenzyl-β*-D-*galactopyranosyl)-(1→4)-2,3,6-tri*-O-*benzyl-β*-D-*glucopyranoside* (**10**).—To a solution of **8** (450 mg, 344 μmol) and **9** (515 mg, 517 μmol) in dry CH_2Cl_2 (20 mL) was added 4 Å molecular sieves (965 mg), and the mixture was stirred for 1 h at room temperature, then cooled to 0 °C. DMTST (53.8% (w/w), 745 mg, 5.35 mmol) was added to the mixture, and the resultant mixture was stirred for 1 h at 0 °C. After dilution with $CHCl_3$, the precipitate was filtered off and washed with $CHCl_3$. The filtrate and washings were combined and successively washed with 1 M Na_2CO_3 and water, dried (Na_2SO_4) and concentrated. Column chromatography (toluene) of the residue on Bio-Beads SX-II afforded **10** (736 mg, 95%) as an amorphous mass: $[\alpha]_D$ +39.6° (*c* 0.6, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.02 (s, 9 H, Me_3Si), 0.99–1.03 (m, 2 H, $SiCH_2CH_2O$-), 1.19–2.18 (6 s, 18 H, 4 OAc, 2 NHAc), 1.59 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.4 Hz, H-3eax), 2.42 (dd, 1 H, $J_{3eq,4}$ 4.4 Hz, H-3eeq), 3.65 (m, 1 H, H-6e), 3.78 (s, 3 H, $PhOCH_3$), 3.78 (m, 1 H, H-5e), 3.82 (s, 3 H, COOMe), 4.37 (d, 1 H, $J_{3,4}$ 3.4 Hz, H-4c), 5.10 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1d), 5.22 (m, 1 H, H-9e), 5.42 (s, 1 H, benzylidene), 5.49 (dd, 1 H, $J_{1,2}$ 7.8 Hz, H-2d), 7.06–8.11 (m, 49 H, 10 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 172.2 (C=O), 171.6 (C=O), 169.6 (C=O), 167.3 (C=O), 167.2 (C=O), 139.8, 134.7, 131.9, 131.6, 131.4, 131.1, 131.0, 130.7,130.0, 129.9, 129.8, 129.7, 129.7, 129.6, 129.6, 129.6, 129.4, 129.4, 129.0, 129.0, 128.9, 128.7, 128.6, 127.5 115.0, 104.6 (O-C-O), 104.5 (O-C-O), 102.3 (O-C-O), 101.2 (O-C-O), 101.7 (O-C-O), 98.2, 83.3, 78.7, 78.4, 78.2, 78.1, 76.6, 76.4, 74.4, 73.3, 72.0, 69.7, 68.7, 56.6, 54.6, 31.1, 24.6, 24.4, 22.9, 22.3, 21.9, 19.9, 1.40. Anal. Calcd for $C_{122}H_{138}N_2O_{37}Si$ (2252.51): C, 65.05; H, 6.18; N, 1.24. Found: C 64.93; H, 6.09; N, 1.19.

2-(Trimethylsilyl)ethyl (methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-D-*galactopyranosyl)-(1→3)-(2-acetamido-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*benzyl-3*-O-p-*methoxybenzyl-β*-D-*galactopyranosyl)-(1→4)-2,3,6-tri*-O-*benzyl-β*-D-*glucopyranoside* (**11**).—A solution of **10** (127 mg, 56.5 μmol) in 80% aq HOAc (5 mL) was heated, with stirring, for 24 h at 50 °C, then concentrated. Column chromatography (120:1 $CHCl_3$–MeOH) of the residue on silica gel gave **11** (101 mg, 83%) as an amorphous mass: $[\alpha]_D$ +39.6° (*c* 0.6, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.01 (s, 9 H, Me_3Si), 1.01–1.04 (m, 2 H, $SiCH_2CH_2O$-), 0.89–2.21 (6 s, 18 H, 4 OAc, 2 NHAc), 1.57 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.6 Hz, H-3eax), 2.42 (dd, 1 H, $J_{3eq,4}$ 4.6 Hz, H-3eeq), 3.40 (m, 1 H, H-2c), 3.61 (m, 1 H, H-6e), 3.73 (m, 1 H, *J* 10.5 Hz, H-5e), 3.80 (s, 3 H, -COOMe), 3.85 (s, $PhOCH_3$), 5.17 (m, 1 H, H-7e), 5.25 (m, 1 H, H-9e), 5.32 (d, 1 H, $J_{3,4}$ 3.2 Hz, H-4d), 5.61 (m, 1 H, H-8e), 7.15–8.13 (m, 44 H, 9 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): 172.1 (C=O), 169.6 (C=O), 168.7 (C=O), 167.3 (C=O), 166.3 (C=O), 161.7 (C=O), 160.6 (C=O), 146.8, 143.6, 140.0, 135.2, 134.8, 132.5, 131.5, 131.1, 130.4, 130.0, 129.6, 128.9, 115.5, 104.6 (C-O-C), 102.7 (C-O-C), 101.4 (C-O-C), 100.5 (C-O-C), 98.2 (C-O-C), 84.3, 83.3, 81.2, 74.4, 73.3, 72.8, 71.9, 70.7, 69.7, 69.1, 68.7, 67.7, 63.7, 56.7, 54.7, 50.2, 31.1, 24.6, 23.7, 23.0, 22.3, 21.9, 19.9, 11.7, 5.42. Anal. Calcd for $C_{115}H_{134}N_2O_{37}Si$ (2164.40): C, 63.82; H, 6.24; N, 1.29. Found: C, 63.73; H, 6.20; N, 1.07.

2-(Trimethylsilyl)ethyl (methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-*acetyl-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*benzyl-3*-O-p-*methoxybenzyl-β*-D-*galactopyranosyl)-(1→4)-2,3,6-tri*-O-*benzyl-β*-D-*glucopyranoside* (**12**).—Compound **11** (564 mg, 261 μmol) was acetylated with Ac_2O (3 mL) and pyridine (8 mL) in the presence of 4-dimethylaminopyridine (DMAP; 1.7 mg, 13.9 μmol) for 20 h at 40 °C. The mixture was concentrated, and a solution of the residue in EtOAc was successively washed with 2 M HCl and M Na_2CO_3, dried (Na_2SO_4) and concentrated. Column chromatography (120:1 $CHCl_3$–MeOH) of the residue on silica gel gave **12** (556 mg, 95%) as an amorphous mass: $[\alpha]_D$ +23.5° (*c* 0.40, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.01 (s, 9 H, Me_3Si), 1.01–1.05 (m, 2 H, $SiCH_2CH_2O$-), 1.29–2.15 (8 s, 24 H, 6 OAc, 2 NHAc), 1.63 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.3 Hz, H-3eax), 2.45 (dd, 1 H, $J_{3eq,4}$ 4.3 Hz, H-3eeq), 3.30

(m, 1 H, H-2c), 3.59 (dd, 1 H, *J* 5.13 Hz, H-6e), 3.80 (s, 3 H, -COOMe), 3.82 (s, 3 H, $PhOCH_3$), 5.60 (m, 1 H, H-8e), 7.18–8.12 (m, 44 H, 9 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 172.2 (C=O), 171.8 (C=O), 171.6 (C=O), 171.1 (C=O), 170.9 (C=O), 170.0 (C=O), 167.1 (C=O), 166.1 (C=O), 162.0 (C=O), 160.7 (C=O), 140.2, 139.8, 134.4, 131.4, 131.2, 130.9, 130.5, 130.2, 129.8, 129.6, 129.4, 128.9, 128.7, 115.1, 104.5 (O-C-O), 102.8 (O-C-O), 100.9 (O-C-O), 98.2 (O-C-O), 95.8 (O-C-O), 84.3, 83.3, 82.5, 78.7, 78.4, 78.1, 77.0, 76.6, 75.1, 74.8, 74.6, 73.2, 72.7, 72.3, 71.8, 70.8, 70.5, 69.2, 68.8, 67.9, 64.1, 63.2, 56.6, 56.3, 54.6, 50.2, 38.8, 31.1, 24.4, 22.8, 22.3, 22.1, 21.8, 19.9, 3.70, 2.46, 1.40. Anal. Calcd for $C_{119}H_{138}N_{39}Si$ (2248.47): C, 63.57; H, 6.19; N, 1.25. Found: C, 63.48; H, 6.15; N, 1.10.

2-(Trimethylsilyl)ethyl (methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-*α*-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6-di*-O-*acetyl-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*benzyl-β*-D-*galactopyranosyl)-(1→4)-2,3,6-tri*-O-*benzyl-β*-D-*glucopyranoside* (**13**).—To a solution of **12** (414 mg, 184 μmol) in CH_3CN (10 mL) and water (1 mL) was added ceric ammonium nitrate (CAN; 404 mg, 737 μmol), and the mixture was stirred for 3 h at room temperature and extracted with $CHCl_3$. The extract was successively washed with cold 1 M Na_2CO_3 and water, dried (Na_2SO_4) and concentrated. Column chromatography (35:1 toluene–MeOH) of the residue on silica gel gave **13** (368 mg, 94%) as an amorphous mass: $[\alpha]_D$ + 34.3° (*c* 1.4, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 0.03 (s, 9 H, Me_3Si), 1.01–1.03 (m, 2 H, $SiCH_2CH_2O$-), 1.33–2.15 (8 s, 24 H, 6 OAc, 2 NHAc), 1.57 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.5 Hz, H-3eax), 2.43 (dd, 1 H, $J_{3eq,4}$ 4.3 Hz, H-3eeq), 3.01 (d, 1 H, $J_{3,OH}$ 7.3 Hz, -OH), 3.20 (m, 1 H, H-2b), 3.31 (m, 1 H, H-2c), 3.45 (m, 1 H, H-3b), 3.58 (m, H-6e), 3.76 (m, 1 H, H-5e), 3.83 (s, 3 H, -COOMe), 5.52 (d, 1 H, $J_{3,4}$ 2.9 Hz H-4d), 5.60 (m, 1 H, H-8e), 7.18–8.14 (m, 40 H, 8 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 172.6 (C=O), 172.2 (C=O), 172.0 (C=O), 171.9 (C=O), 171.6 (C=O), 171.5 (C=O), 170.9 (C=O), 169.5 (C=O), 167.1 (C=O), 177.0 (C=O), 166.2 (C=O), 140.4, 140.2, 140.0, 139.8, 139.6, 134.7, 134.5, 131.6, 131.5, 131.1, 130.0, 129.7, 129.7, 129.4, 129.0, 128.9, 104.5 (O-C-O), 103.7 (O-C-O), 102.3 (O-C-O), 101.2 (O-C-O), 101.2 (O-C-O), 98.2 (O-C-O), 84.2, 83.3, 82.2, 78.7, 78.6, 78.4, 78.1, 77.7, 77.3, 76.8, 76.5, 76.4, 75.6, 75.3, 74.6, 73.2, 72.8, 72.7, 72.0, 70.7, 70.6, 69.8, 69.2, 68.8, 68.5, 68.0, 64.0, 63.2, 56.5, 54.6, 50.1, 38.7, 31.1, 24.5, 24.4, 22.8, 22.1, 21.8, 19.8. Anal. Calcd for $C_{111}H_{130}N_2O_{38}Si$ (2128.32): C, 62.64; H, 6.16; N, 1.32. Found: C, 62.48; H, 6.10; N, 1.30.

2-(Trimethylsilyl)ethyl (methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-*α*-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6-di*-O-*acetyl-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*benzyl-3*-O-*levulinyl-β*-D-*galactopyranosyl)-(1→4)-2,3,6-tri*-O-*benzyl-β*-D-*glucopyranoside* (**14**).—To a solution of **13** (368 mg, 173 μmol) and DMAP (1.1 mg, 9.0 μmol) in pyridine (10 mL), cooled to 0 °C, was added levulinic anhydride (111 mg, 518 μmol). The mixture was stirred for 3 h at 50 °C, then MeOH (2 mL) was added. The solution was stirred for 30 min, concentrated and then extracted with EtOAc. The extract was successively washed with 2 M HCl, M Na_2CO_3 and water, dried (Na_2SO_4) and concentrated. Column chromatography (40:1 toluene–MeOH) of the residue on silica gel gave **14** (330 mg, 81%) as an amorphous mass: $[\alpha]_D$ + 34.1° (*c* 1.3, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 0.03 (s, 9 H, Me_3Si), 1.01–1.06 (m, 2 H, $SiCH_2CH_2O$-), 1.25–2.22 (9 s, 27 H, 7 $COCH_3$, 2 NHAc), 1.60 (t, 1 H, $J_{gem} = J_{3ax,4}$ 9.52 Hz, H-3eax), 2.41 (m, 2 H, -$COCH_2CH_2$-), 2.42 (dd, 1 H, $J_{3eq,4}$ 4.39 Hz, H-3eeq), 2.62 (m, 2 H, -$COCH_2CH_2$), 3.56 (m, 1 H, H-6e), 3.75 (m, 1 H, H-5e), 4.41 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1c), 4.78 (m, 1 H, H-4e), 4.92 (d, 1 H, NH), 5.19 (m, 1 H, H-7e), 5.53 (m, 1 H, H-8e), 6.16 (d, 1 H, NH), 7.07–8.06 (m, 40 H, 8 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 176.6 (C=O), 174.2 (C=O), 172.2 (C=O), 171.9 (C=O), 171.4 (C=O), 169.5 (C=O), 168.3 (C=O), 167.1 (C=O), 148.0, 140.2, 139.8, 139.6, 136.2, 134.8, 134.4, 131.4, 131.1, 130.7, 130.5, 129.8, 129.2, 129.0, 128.8, 128.5, 104.5 (O-C-O), 103.9 (O-C-O), 102.8 (O-C-O), 101.4 (O-C-O), 98.2 (O-C-O), 84.0, 83.1, 79.2, 78.7, 78.4, 78.1, 77.1, 76.4, 75.5, 74.5, 73.1, 72.7, 72.3, 71.7, 70.8, 69.2, 68.7, 68.5, 67.7, 64.1, 63.6, 63.1, 54.5, 52.3, 50.2, 39.3, 38.8, 31.3, 29.0, 24.5, 24.3, 22.7, 22.1, 21.8, 19.9, 1.39. Anal. Calcd for $C_{116}H_{136}N_2O_{40}Si$ (2226.42): C, 62.58; H, 6.16; N, 1.26. Found: C, 62.54; H, 6.14; N, 1.23.

2-(Trimethylsilyl)ethyl (methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-*α*-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6-di*-O-*acetyl-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(3*-O-*levulinyl-β*-D-*galactopyranosyl)-(1→4)-β*-D-*glucopyranoside* (**15**).—A solution of **14** (310 mg, 139 μmol) in EtOH (12 mL) was vigorously stirred with $Pd(OH)_2$ (310 mg) for 48 h at room temperature under H_2. The catalyst was collected and washed with EtOH. The combined filtrate and washings were concentrated. Column chromatography (25:1 $CHCl_3$–MeOH) of the residue on silica gel gave **15** (223 mg, 90%) as an amorphous mass: $[\alpha]_D$ + 27.1° (*c* 0.77, MeOH); ^1H NMR (CD_3OD, 400 MHz): δ 0.03 (s, 9 H, Me_3Si), 0.94–1.10 (m, 2 H, $SiCH_2CH_2O$-), 1.42–2.29 (9 s, 27 H, 7 $COCH_3$, 2 NHAc), 1.48 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.8 Hz, H-3eax), 2.45 (dd, 1 H, $J_{3eq,4}$ 4.0 Hz, H-3eeq), 2.73 (m, 2 H, -$COCH_2CH_2$), 3.03 (m, 2 H, -$COCH_2CH_2$), 3.65 (m, 1 H, H-5e), 5.07 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1d), 5.35 (dd, $J_{1,2}$ 7.7, $J_{2,3}$ 5.4 Hz, H-2d), 5.54 (d, 1 H, $J_{3,4}$ 2.9

Hz, H-4d), 5.59 (m, 1 H, H-8), 7.45–8.15 (m, 15 H, 3 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 173.3 (C=O), 172.2 (C=O), 172.0 (C=O), 171.2 (C=O), 171.0 (C=O), 170.4 (C=O), 170.0 (C=O), 168.2 (C=O), 166.9 (C=O), 165.8 (C=O), 165.1 (C=O), 136.2, 133.3, 130.1, 129.8, 129.5, 129.3, 128.3, 110.8, 103.5 (O-C-O), 102.3 (O-C-O), 101.6 (O-C-O), 100.2 (O-C-O), 96.8 (O-C-O), 86.0, 79.7, 79.0, 77.9, 77.6, 77.3, 75.8, 74.9, 74.4, 73.2, 71.3, 70.4, 69.8, 69.1, 68.0, 67.2, 66.5, 63.0, 62.0, 60.7, 52.7, 48.4, 48.2, 47.8, 47.6, 37.8, 37.2, 29.4, 27.5, 22.0, 20.9, 20.0, 18.0, 7.93. Anal. Calcd for $C_{81}H_{106}N_2O_{40}Si$ (1775.80): C, 54.79; H, 6.02; N, 1.58. Found: C, 54.60; H, 5.89; N, 1.34.

2-(Trimethylsilyl)ethyl (methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6-di*-O-*acetyl-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*acetyl-3*-O-*levulinyl-β*-D-*galactopyranosyl)-(1→4)-2,3,6-tri*-O-*acetyl-β*-D-*glucopyranoside* (**16**).—A solution of **15** (223 mg, 125 μmol) in Ac_2O (5 mL) and pyridine (10 mL) was heated for 24 h at 40 °C, then cooled to 0 °C. MeOH (5 mL) was added, the mixture was concentrated, and the residue was extracted with EtOAc and successively washed with cold 2 M HCl, M Na_2CO_3 and water, dried (Na_2SO_4) and concentrated. Column chromatography (24:1 toluene–MeOH) of the residue on silica gel gave **16** (249 mg, quant.) as an amorphous mass: $[\alpha]_D$ +17.1° (*c* 0.3, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 0.01 (s, 9 H, Me_3Si), 0.84–0.97 (m, 2 H, $SiCH_2CH_2O$-), 1.54–2.09 (14 s, 42 H, 12 $COCH_3$, 2 NHAc), 1.58 (nt, 1 H, $J_{gem} = J_{3ax,4}$ 12.1 Hz, H-3eax), 2.37 (m, 2 H, -$COCH_2CH_2$), 2.42 (dd, 1 H, $J_{3eq,4}$ 4.0 Hz, H-3eeq), 2.65 (m, 2 H, -$COCH_2CH_2$), 3.00 (t, 1 H, H-2c), 3.57 (dd, 1 H, H-6c), 3.77 (q, 1 H, H-5e), 3.78 (d, 1 H, $J_{3,4}$ 4.4 Hz, H-4b), 3.83 (s, 3 H, -COOMe), 4.24 (dd, 1 H, H-9e), 4.36 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1b), 4.41 (d,1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.88 (t, 1 H, $J_{1,2}$ 7.7, $J_{2,3}$ 8.1 Hz, H-2), 4.95 (dd, 1 H, H-3c), 4.96 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1d), 5.03 (dd, 1 H, $J_{1,2}$ 7.7, $J_{2,3}$ 8.1 Hz, H-2b), 5.09 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1c), 5.12 (t, 1 H, $J_{2,3}$ 9.5 Hz, H-3), 5.21 (m, 1 H, H-7e), 5.34 (d, 1 H, $J_{3,4}$ 3.3 Hz, H-4d), 5.36 (t, 1 H, $J_{1,2} = J_{2,3}$ 7.7 Hz, H-2d), 5.50 (m, 1 H, H-8e), 5.84 (d, 1 H, *J* 6.9 Hz, NH), 7.40–8.17 (m, 15 H, 3 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 172.9 (C=O), 172.0 (C=O), 170.8 (C=O), 170.6 (C=O), 170.4 (C=O), 170.3 (C=O), 170.1 (C=O), 169.7 (C=O), 169.6 (C=O), 169.5 (C=O), 168.2 (C=O), 165.7 (C=O), 164.9 (C=O), 133.3, 133.0, 130.2, 130.1, 129.8, 129.5, 128.7, 128.5, 128.3, 101.4 (O-C-O), 100.6 (O-C-O), 100.0 (O-C-O), 97.9 (O-C-O), 96.9 (O-C-O), 77.4, 77.1, 76.7, 76.1, 73.6, 73.0, 72.9, 72.7, 72.2, 71.8, 71.7, 71.4, 71.2, 71.0, 70.4, 69.6, 69.4, 69.3, 67.9, 67.5, 67.2, 66.5, 63.0, 62.7, 61.9, 55.5, 53.2, 48.8, 37.7, 29.7, 27.4, 23.2, 23.0, 21.4, 20.9, 20.9, 20.8, 20.7, 20.5, 17.9, 1.38. Anal. Calcd for $C_{91}H_{116}N_2O_{45}Si$ (1985.98): C, 55.04; H, 5.89; N, 1.41. Found: C, 54.60; H, 5.64; N, 1.16.

(Methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6-di*-O-*acetyl-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*acetyl-3*-O-*levulinyl-β*-D-*galactopyranosyl)-(1→4)-2,3,6-tri*-O-*acetyl*-D-*glucopyranosyl trichloroacetimidate* (**18**).—To a solution of **16** (210 mg, 118 μmol) in CH_2Cl_2 (10 mL) was added CF_3CO_2H (5 mL), and the mixture was stirred for 2 h at 0 °C, then for 4 h at room temperature, and concentrated. Column chromatography (60:1 $CHCl_3$–MeOH) of the residue on silica gel gave the free 1-OH derivative, which was treated with CCl_3CN (353 μL, 2.45 mmol) and DBU (19.5 μL, 128 μmol) in dry CH_2Cl_2 (10 mL) for 2 h at room temperature. After concentration, the residue was chromatographed on a column of silica gel (20:1 $CHCl_3$–MeOH) to afford amorphous **18** (232 mg, quant.); ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.84–0.97 (m, 2 H, $SiCH_2CH_2O$-), 1.54–2.09 (14 s, 42 H, 12 $COCH_3$, 2 NHAc), 1.59 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.6 Hz, H-3eax), 2.40 (m, 2 H, -$COCH_2CH_2$), 2.67 (dd, 1 H, $J_{3eq,4}$ 4.0 Hz, H-3eeq), 2.68 (m, 2 H, -$COCH_2CH_2$), 3.01 (dt, 1 H, H-2c), 3.57 (m, 1 H, H-6e), 3.77 (m, 1 H, H-5e), 3.83 (s, 3 H, -COOMe), 4.24 (m, 1 H, H-9e), 4.88 (t, 1 H, $J_{1,2}$ 7.7, $J_{2,3}$ 8.1 Hz, H-2b), 4.96 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1d), 5.21 (m, 1 H, H-7e), 5.36 (t, 1 H, $J_{1,2}$ 7.6, $J_{2,3}$ 7.7 Hz, H-2d), 5.51 (d, 1 H, $J_{3,4}$ 3.3 Hz, H-4d), 5.60 (m, 1 H, H-8e), 5.84 (d, 1 H, *J* 6.9 Hz, -$NHCOCl_3$), 7.39–8.19 (m, 15 H, 3 Ph).

(Methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl-β*-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6-di*-O-*acetyl-2-deoxy-β*-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*acetyl-3*-O-*levulinyl-β*-D-*galactopyranosyl)-(1→4)-(2,3,6-tri*-O-*acetyl-β*-D-*glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-2-azido-3*-O-*benzoyl-4-octadecene-1,3-diol* (**20**).—To a solution of **18** (209 mg, 114 μmol) and **19** (97.8 mg, 228 μmol) in dry CH_2Cl_2 (3 mL) were added 4 Å molecular sieves (type AW 300; 300 mg), and the mixture was stirred for 1 h at 0 °C. $BF_3{\cdot}OEt_2$ (28.9 μL, 204 μmol) was added to the mixture, and this was stirred for 12 h at 0 °C and filtered. The residue was washed with $CHCl_3$, and the combined filtrate washings were concentrated. Column chromatography (30:1 toluene–MeOH) of the residue on silica gel afforded **20** (73.4 mg, 31%) as an amorphous mass: $[\alpha]_D$ +7.2° (*c* 1.2, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.87 (t, 3 H, -CH_2CH_3 of sphingoshine), 1.24 (m, 52 H, 12 CH_2), 1.55–2.16 (14 s, 42 H, 12 $COCH_3$, 2 NHAc), 1.56 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.5 Hz, H-3eax), 2.45 (dd, 1 H, $J_{3eq,4}$ 4.0 Hz, H-3eeq), 2.39 (m, 2 H, -$COCH_2CH_2$), 2.65 (m, 2 H, -$COCH_2CH_2$), 3.01 (dd, 1 H, $J_{1,2}$ 8.1 Hz, H-2c), 3.57 (m, 1 H, H-6e), 3.76

(dt, 1 H, H-5e), 4.36 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1b), 4.49 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1a), 4.97 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1d), 5.09 (d, 1 H, $J_{1,2}$ 8.2 Hz, H-1c), 5.21 (dd, 1 H, J 2.5, J 7.1 Hz, H-7e), 5.50 (d, 1 H, $J_{3,4}$ 3.4 Hz, H-4d), 5.58 (m, 1 H, H-8e), 5.95 (d, 1H, NH), 7.41–8.18 (m, 15 H, 3 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 172.8 (C=O), 172.0 (C=O), 170.8 (C=O), 170.6 (C=O), 170.2 (C=O), 170.0 (C=O), 169.7 (C=O), 169.5 (C=O), 168.1 (C=O), 165.6 (C=O), 165.1 (C=O), 164.9 (C=O), 139.0, 133.2, 133.0, 129.7, 129.5, 129.0, 128.6, 128.5, 128.2, 122.6, 101.4 (O-C-O), 100.7 (O-C-O), 100.3 (O-C-O), 98.0 (O-C-O), 96.8 (O-C-O), 77.3, 77.0, 76.7, 75.9, 74.7, 73.6, 72.9, 72.6, 72.2, 71.8, 71.3, 71.0, 70.4, 69.6, 69.4, 69.2, 68.3, 67.8, 67.1, 66.4, 63.5, 63.0, 62.7, 62.3, 61.8, 55.4, 53.1, 48.8, 37.7, 37.5, 32.4, 31.9, 29.7, 29.4, 29.2, 28.7, 27.4, 23.2, 22.9, 22.7, 21.4, 20.7, 20.5, 14.1. Anal. Calcd for $C_{111}H_{141}N_5O_{47}$ (2297.34): C, 58.03; H, 6.19; N, 3.05. Found: C, 57.94; H, 5.99; N, 2.91.

(Methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-di-deoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl*-β-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6-di*-O-*acetyl-2-deoxy*-β-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*acetyl-3*-O-*levul-inyl*-β-D-*galactopyranosyl)-(1→4)-2,3,6-tri*-O-*acetyl*-β-D-*glucopyranosyl-(1→1)-(2*S,*3*R,*4*E)-*3*-O-*benzoyl-2-octadecanamido-4-octadecene-1,3-diol* (**21**).—H_2S was bubbled through a stirred solution of **20** (70 mg, 33 μmol) in pyridine (4.2 mL) and water (0.85 mL) for 48 h at 0 °C. The mixture was concentrated, and the residual syrup was treated with octadecanoic acid (28.4 mg, 99.8 μmol) and 3-(3-dimethylaminopropyl)-1-ethylcarbodiimide hydrochloride (19.2 mg, 100 μmol) in CH_2Cl_2 (3 mL) for 2 h at room temperature. The mixture was extracted with $CHCl_3$, and the extract was successively washed with water, dried (Na_2SO_4) and concentrated. Column chromatography (90:1 $CHCl_3$–MeOH) of the residue on silica gel gave **21** (53 mg, 68%) as an amorphous mass: $[\alpha]_D$ +7.2° (*c* 1.2, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz): 0.86 (t, 6 H, 2 CH_2CH_3), 1.23 (s, 52 H, 26 CH_2), 1.55–2.16 (14 s, 42 H, 12 $COCH_3$, 2 NHAc), 1.58 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.2 Hz, H-3eax), 2.39 (m, 2 H, -$COCH_2CH_2$), 2.43 (dd, 1 H, $J_{3eq,4}$ 3.7 Hz, H-3eeq), 2.67 (m, 2 H, -$COCH_2CH_2$), 3.02 (dd, 1 H, $J_{1,2}$ 8.1 Hz, H-2c), 3.57 (m, 1 H, H-6e), 3.82 (s, 3 H, -COOMe), 4.47 (d, 1 H, H-1a), 4.80 (m, 1 H, H-4e), 4.81 (d, 1 H, H-1b), 4.82 (m, 1 H, H-3), 4.92 (d, 1 H, NH), 5.00 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1d), 5.09 (d, 1 H, $J_{1,2}$ 8.5 Hz, H-1c), 5.21 (dd, 1 H, H-7e), 5.36 (dd, 1 H, H-2d), 5.50 (d, 1 H, $J_{3,4}$ 3.4 Hz, H-4d), 5.58 (m, 1 H, H-8e), 6.00 (d, 1 H, NH), 7.39–8.17 (m, 15 H, 3 Ph). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 172.7 (C=O), 172.0 (C=O), 170.8 (C=O), 170.6 (C=O), 170.2 (C=O), 169.9 (C=O), 169.4 (C=O), 168.4 (C=O), 165.7 (C=O), 165.2 (C=O), 164.9 (C=O), 137.6, 133.2, 131.0, 130.2, 130.1, 129.8, 128.6, 128.2, 124.6, 101.4 (O-C-O), 100.6 (O-C-O), 100.3 (O-C-O), 97.9 (O-C-O), 96.8 (O-C-O), 77.4, 77.0, 76.7, 75.8, 74.1, 73.7, 72.8, 72.4, 72.2, 71.8, 71.2, 71.0, 70.4, 69.6, 69.4, 69.2, 67.8, 67.3, 67.2, 66.5, 63.0, 62.7, 62.4, 61.9, 55.4, 53.2, 50.6, 48.8, 37.7, 36.9, 32.2, 31.9, 29.7, 29.4, 28.9, 27.4, 25.7, 23.2, 22.9, 22.7, 21.4, 20.8, 20.6, 20.5, 14.1. Anal. Calcd for $C_{129}H_{177}N_3O_{48}$ (2537.81): C, 61.05; H, 7.03; N, 1.66. Found: C, 60.80; H, 6.82; N, 1.41.

(Methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-di-deoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)-(2,4,6-tri*-O-*benzoyl*-β-D-*galactopyranosyl)-(1→3)-(2-acetamido-4,6-di*-O-*acetyl-2-deoxy*-β-D-*galactopyranosyl)-(1→4)-(2,6-di*-O-*acetyl*-β-D-*galactopyranosyl)-(1→4)-(2,3,6-tri*-O-*acetyl*-β-D-*glucopyranosyl)-(1→1)-(2*S,*3*R,*4*E)-*3*-O-*benzoyl-2-octadecanamido-4-octadecene-1,3-diol* (**22**).—To a solution of **21** (43 mg, 18.2 μmol) in EtOH (3 mL) was added hydrazine monoacetate (1.85 mg, 20 μmol), and the mixture was stirred for 2 h at 0 °C and then concentrated. Column chromatography (20:1 $CHCl_3$–MeOH) of the residue on silica gel afforded **22** (38.4 mg, 94%) as an amorphous mass: $[\alpha]_D$ +30.1° (*c* 0.76, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz): δ 0.87 (t, 6 H, 2 CH_2CH_3), 1.23 (s, 52 H, 26 CH_2), 1.56–2.17 (13 s, 39 H, 11 OAc, 2 NHAc), 1.58 (t, 1 H, $J_{gem} = J_{3ax,4}$ 12.2 Hz, H-3eax), 2.43 (dd, 1 H, $J_{3eq,4}$ 3.7 Hz, H-3eeq), 3.11 (m, 1 H, H-2c), 3.58 (m, 1 H, H-6e), 3.83 (s, 3 H, COOMe), 4.00 (m, 1 H, H-5e), 4.80 (m, 1 H, H-4e), 4.94 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1d), 5.20 (dd, 1 H, H-7e), 5.34 (d, 1 H, $J_{3,4}$ 3.2 Hz, H-4d), 5.61 (m, 1 H, H-8e), 7.40–8.18 (m, 15 H, 3 Ph). ^{13}C NMR ($CDCl_3$, 126 MHz): δ 174.0 (C=O), 173.4 (C=O), 171.5 (C=O), 171.0 (C=O), 170.4 (C=O), 168.8 (C=O), 166.4 (C=O), 165.8 (C=O), 138.3, 133.7, 130.4, 129.0, 125.3, 102.2 (O-C-O), 101.7 (O-C-O), 101.1 (O-C-O), 99.7 (O-C-O), 97.6 (O-C-O), 77.7, 76.7, 76.0, 75.3, 74.7, 73.6, 73.0, 71.9, 71.4, 73.6, 73.0, 71.9, 71.4, 70.1, 68.6, 67.9, 67.3, 64.3, 63.3, 62.6, 56.6, 53.9, 51.3, 49.5, 38.1, 37.6, 32.6, 30.4, 29.7, 26.5, 23.4, 22.1, 21.6, 14.8. Anal. Calcd for $C_{124}H_{171}N_3O_{46}$ (2439.71): C, 61.05; H, 7.07; N, 1.72. Found: C, 60.98; H, 6.89; N, 1.50.

(5-Acetamido-3,5-dideoxy-D-galacto-*2-nonulopyran-osylonate)-(2→3)-(*β-D-*galactopyranosylonic acid)-(1→3)-(2-acetamido-2-deoxy*-β-D-*galactopyranosyl)-(1→4)-(3*-O-*sulfo*-β-D-*galactopyranosyl)-(1→4)-(*β-D-*glu-copyranosyl)-(1→1)-(2*S,*3*R,*4*E)-*2*-octadecanamido-4-octadecene-1,3-diol* (**1**).—To a solution of **22** (30 mg, 12 μmol) in DMF (1 mL) was added sulfur trioxide·pyridine complex (10.6 mg, 67 μmol), and the mixture was stirred for 2 h at room temperature. The mixture was concentrated. Column chromatography (MeOH) of the residue on Sephadex LH-20 gave the sulfated product **23** (25.8 mg, 80%). To a solution of **23** (18 mg, 7.4 μmol) in MeOH–dioxane (1 mL) was added, catalytic amount of 28% NaOMe in MeOH, and the mixture was stirred for 72 h at room temperature. Water (1 mL) was added, and the mixture was stirred for 48 h at room temperature, and then concentrated. Column chromatography (MeOH) of the residue on

Sephadex LH-20 gave **17** (10.9 mg, 89%) as an amorphous mass. $[\alpha]_D$ +3.4° (*c* 0.6, 5:4:0.7 $CHCl_3$–CH_3OH, H_2O); ^1H NMR (1:1 $CDCl_3$–CD_3OD, 500 MHz): δ 0.85 (t, 6 H, 2 CH_2CH_3), 1.24 (s, 52 H, 26 CH_2), 1.46 (br m, 2 H, $COCH_2CH_2$), 2.45 (dd, 1 H, $J_{3eq,4}$ 4.8, J_{gem} 12 Hz), 4.22 (d, 1 H, $J_{1,2}$ 7.9 Hz, H-1a), 4.26 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1b), 4.28 (d, 1 H, $J_{1,2}$ 7.3 Hz, H-1d), 4.49 (d, 1 H, $J_{1,2}$ 8.0 Hz, H-1c), 5.37 (dd, 1 H, $J_{3,4}$ 7.0, $J_{4,5}$ 15.4 Hz, H-4 of sphingosine) and 5.55 (m, 1 H, H-5 of sphingosine), FABMS (negative-ion mode, triethanolamine matrix), *m*/*z* 1646 $(M - Na)^-$ and 1545 $(M - SO_3Na + H)^-$.

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