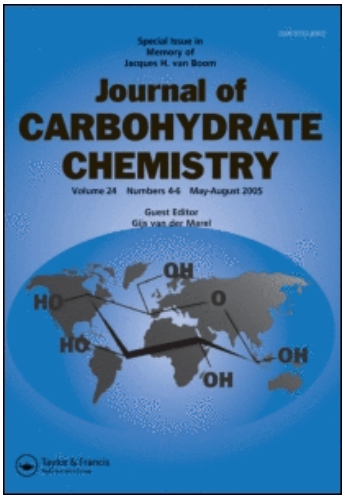

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Synthetic Studies on Sialoglycoconjugates 89: Synthesis of Ganglioside GM_3 and KDN-GM_3 Containing Different Carbon-Chain Length Fatty Acyl Groups at the Ceramide Residue

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SYNTHETIC STUDIES ON SIALOGLYCOCONJUGATES 89: SYNTHESIS OF GANGLIOSIDE GM3 AND KDN-GM3 CONTAINING DIFFERENT CARBON-CHAIN LENGTH FATTY ACYL GROUPS AT THE CERAMIDE RESIDUE

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ABSTRACT

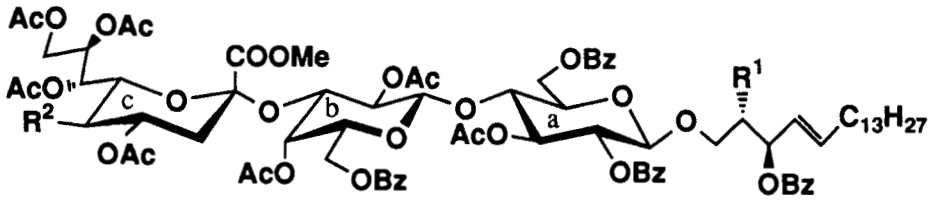
Ganglioside GM3 and KDN-ganglioside GM3, containing hexanoyl, decanoyl, and hexadecanoyl groups at the ceramide moiety have been synthesized. Selective reduction of the azido group in *O*-(methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-(3-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol (**1**) and *O*-(methyl 4,5,7,8,9-penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-(3-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol (**2**), coupling with hexanoic, decanoic, and hexadecanoic acids, *O*-deacylation, and de-esterification gave the title gangliosides GM3 (**11~13**) and KDN-GM3 (**14~16**) in good yields. On the other hand, *O*-deacylation of **1** and subsequent de-esterification gave 2-azido-sphingosine containing-GM3 analogue **17**, which was converted into lyso-GM3, in which no fatty acyl group was substituted at the sphingosine residue, by selective reduction of the azido group.

INTRODUCTION

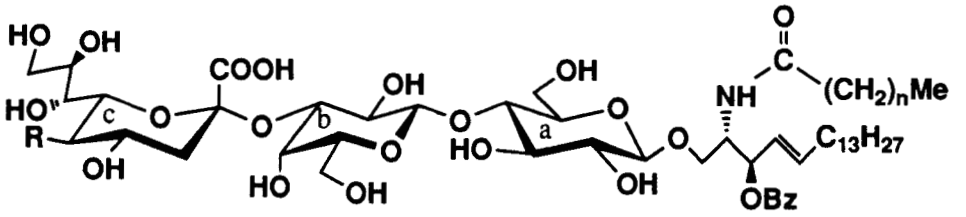
Ganglioside GM3 has various types of important biological functions,[1] and those activities are strictly related to the structure of sialic acid, oligosaccharide chain and fatty-acyl residue at the ceramide moiety. Gangliosides are potent inhibitors of cellular immune responses, and the structures of gangliosides greatly influence their activity.[2,3] Recent studies using chemically synthesized gangliosides GM3 and GM4 and their analogues have shown[4,5] the influence of certain structural details of their immunosuppressive activity as follows: 1) fatty acyl chain length at the ceramide moiety is an important factor for the activity 2) hydroxylation of the fatty acyl group decreases immunosuppressive activity 3) substitution of an *S*-glycosidic linkage for an *O*-glycosidic linkage in the sialic acid linkage does not alter its activity and 4) modification of the sialic acid structure variably influence the activity, since KDN (3-deoxy-D-*glycero*-D-*galacto*-2-nonulopyranosonic acid)-GM3 and -GM4 analogues retain potent activity, while other modifications such as 8-epi-GM3 and 9-deoxy-GM3 reduce immunosuppressive activity. In view of these facts, we describe here the synthesis of GM3 and KDN-GM3 each containing three different fatty-acyl groups hexanoyl, decanoyl, or hexadecanoyl at the ceramide moiety in order to clarify the relationship between fatty acyl chain length and immunosuppressive activity.

RESULTS AND DISCUSSION

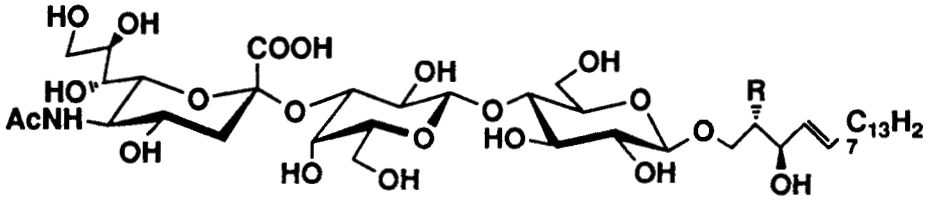
For the synthesis of the desired ganglioside GM3 and KDN-GM3, we employed the intermediates (**1** and **2**) developed for the synthesis of GM3[6] and KDN-GM3.[7] Selective reduction[6,8] of the azido group in compounds **1** and **2** with H_2S in 5:1 pyridine-water for 48 h at 0 °C gave the corresponding amines **3** and **4**, which on condensation with hexanoic, decanoic , and hexadecanoic acids, respectively, using 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (WSC) in CH_2Cl_2, gave the corresponding GM3 and KDN-GM3 derivatives **5~10** in high yields. Finally, *O*-deacylation of **5~10** with sodium methoxide in methanol, and subsequent saponification of the methyl ester group, yielded almost quantitatively the



1	$R^1 = N_3$, R^2 = NHAc
2	$R^1 = N_3$, R^2 = OAc
3	$R^1 = NH_2$, R^2 = NHAc
4	$R^1 = NH_2$, R^2 = OAc
5, 6, 7	$R^1 = NHCO(CH_2)_nMe$, R^2 = NHAc, n = 4, 8, 14
8, 9, 10	$R^1 = NHCO(CH_2)_nMe$, R^2 = OAc, n = 4, 8, 14



11, 12, 13	R = NHAc, n = 4, 8, 14
14, 15, 16	R = OH, n = 4, 8, 14



17	R = N_3
18	R = NH_2

corresponding ganglioside GM3 (**11~13**) and KDN-GM3 (**14~16**) containing different fatty acyl groups at the ceramide moiety.

On the other hand, selective reduction of the azido group with H_2S in **17** prepared from **1** by *O*-deacylation and saponification of the methyl ester group, gave lyso-GM3 (**18**) in 80% yield, in which no acyl group was substituted at the sphingosine moiety.

EXPERIMENTAL

General Procedures. Specific rotations were determined with a Union PM-201 polarimeter at 25 °C. ^1H NMR spectra were recorded with a JEOL JNM-GX 270 spectrometer. Preparative chromatography was performed on silica gel (Wako Chemical Co., 200 mesh) with the solvent systems specified. Concentrations were conducted *in vacuo*.

***O*-(Methyl 5-Acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-(3-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-3-*O*-benzoyl-2-hexanamido-4-octadecene-1,3-diol (5).** Hydrogen sulfide was bubbled through a stirred solution of **16** (100 mg, 0.06 mmol) in aq 83% pyridine (6 mL) for 48 h at 0 °C. The reaction was monitored by TLC. After completion of the reaction, the mixture was concentrated, and the residue was stirred with hexanoic acid (21 mg, 0.18 mmol) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (WSC) (57 mg, 0.3 mmol) in dry CH_2Cl_2 (5 mL) overnight at room temperature. Dichloromethane (30 mL) was added, and the mixture was washed with water, dried (Na_2SO_4) and concentrated. Column chromatography (60:1 CH_2Cl_2-MeOH) of the residue on silica gel (30 g) gave **5** (85 mg, 82%) as an amorphous mass: $[\alpha]_D$ +7.1° (*c* 0.7, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.85 (t, 6H, 2*Me*CH_2), 1.24 (s, 32H, 16CH_2), 1.84~2.18 (8s, 24H, 7AcO, AcN), 2.57 (dd, 1H, J_{gem} = 12.6 Hz, $J_{3eq,4}$ = 4.4 Hz, H-3b*eq*), 3.70 (s, 3H, MeO), 4.58 (dd, 1H, $J_{2,3}$ = 10.3 Hz, $J_{3,4}$ = 3.3 Hz, H-3b), 4.61 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1a), 4.84 (d, 1H, $J_{1,2}$ = 8.1 Hz, H-1b), 5.02 (d, 1H, H-4b), 5.15

(dd, 1H, H-2b), 5.19 (dd, 1H, $J_{2,3}$ = 8.0 Hz, H-2a), 5.38 (dd, 1H, $J_{6,7}$ = 2.9 Hz, $J_{7,8}$ = 8.8 Hz, H-7c), 5.47 (br t, 1H, $J_{3,4}$ = 9.7 Hz, H-3a), 5.65 (d, 1H, $J_{5,NH}$ = 9.0 Hz, NH), 5.76 (m, 1H, H-5 of sphingosine), and 7.25-8.06 (m, 20H, 4Ph).

Anal. Calcd for $C_{90}H_{116}N_2O_{32}$ (1737.0): C, 62.21; H, 6.73; N, 1.61. Found: C, 62.33; H, 6.81; N, 1.56.

***O*-(Methyl 5-Acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-(3-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-3-*O*-benzoyl-2-decanamido-4-octadecene-1,3-diol (6).** The azido group in **1** (100 mg, 0.06 mmol) was converted into amine as described for **5**, which was then condensed with decanoic acid (31 mg, 0.18 mmol) in the presence of WSC (57 mg, 0.3 mmol), to give **6** (85 mg, 79%) as an amorphous mass: $[\alpha]_D$ +6.5° (*c* 0.8, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.89 (t, 6H, 2*Me*CH_2), 1.28 (s, 40H, 20CH_2), 1.75 (t, 1H, J_{gem} = $J_{3ax,4}$ = 12.5 Hz, H-3c*ax*), 1.85~2.27 (8s, 24H, 7AcO, AcN), 2.67 (dd, 1H, $J_{3eq,4}$ = 4.9 Hz, H-3c*eq*), 3.67 (dd, 1H, $J_{5,6}$ = 10.4 Hz, $J_{6,7}$ = 3.6 Hz, H-6c), 3.78 (s, 3H, MeO), 4.67 (dd, 1H, $J_{2,3}$ = 9.7 Hz, $J_{3,4}$ = 3.1 Hz, H-3b), 4.70 (d, 1H, $J_{1,2}$ = 7.5 Hz, H-1a), 4.93 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1b), 5.02 (d, 1H, H-4b), 5.24 (dd, 1H, H-2b), 5.33 (t, 1H, $J_{2,3}$ = 7.5 Hz, H-2a), 5.45 (dd, 1H, $J_{7,8}$ = 6.8 Hz, H-7c), 5.58 (br t, 1H, $J_{3,4}$ = 7.0 Hz, H-3a), 5.76 (d, 1H, $J_{5,NH}$ = 9.8 Hz, NH), 5.84 (m, 1H, H-5 of sphingosine), and 7.34-8.14 (m, 20H, 4Ph).

Anal. Calcd for $C_{94}H_{124}N_2O_{32}$ (1794.0): C, 62.93; H, 6.97; N, 1.56. Found: C, 62.77; H, 6.95; N, 1.49.

***O*-(Methyl 5-Acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-(3-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-3-*O*-benzoyl-2-hexadecanamido-4-octadecene-1,3-diol (7).** Reduction of the azido group in **1** (100 mg, 0.06 mmol) and subsequent coupling with hexadecanoic acid (46 mg, 0.18 mmol) using WSC (57 mg, 0.3 mmol), according to the procedure described for **5**, gave compound **7** (95.5 mg, 85%) as an amorphous mass: $[\alpha]_D$ +6.2° (*c* 1.9, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.96 (t, 6H, 2*Me*CH_2), 1.30 (s, 52H, 26CH_2), 1.91-2.26 (8s,

24H, 7AcO, AcN), 1.78 (t, 1H, $J_{gem} = J_{3ax,4}$ = 12.5 Hz, H-3c*ax*), 2.64 (dd, 1H, $J_{3eq,4}$ = 4.4 Hz, H-3c*eq*), 3.67 (dd, 1H, $J_{5,6}$ = 10.8 Hz, $J_{6,7}$ = 2.7 Hz, H-6c), 3.77 (s, 3H, MeO), 4.68 (dd, 1H, $J_{2,3}$ = 9.9 Hz, $J_{3,4}$ = 3.5 Hz, H-3b), 4.70 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1a), 4.93 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1b), 5.07 (d, 1H, H-4b), 5.24 (dd, 1H, H-2b), 5.33 (dd, 1H, $J_{2,3}$ = 8.1 Hz, H-2a), 5.45 (dd, 1H, $J_{7,8}$ = 7.1 Hz, H-7), 5.57 (dd, 1H, $J_{3,4}$ = 7.0 Hz, H-3a), 5.85 (m, 1H, H-5 of sphingosine), and 7.36-8.14 (m, 20H, 4Ph).

Anal. Calcd for $C_{100}H_{136}N_2O_{32}$ (1878.2): C, 63.95; H, 7.30; N, 1.49. Found: C, 63.72; H, 7.45; N, 1.50.

***O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-(3-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-3-*O*-benzoyl-2-hexanamido-4-octadecene-1,3-diol (8).** Reduction of the azido group in 2^7 (111.6 mg, 0.07 mmol) and subsequent coupling with hexanoic acid (16.5 mg, 0.14 mmol) using WSC (27 mg), according to the procedure described for **5**, gave compound **8** (86 mg, 70%) as an amorphous mass: $[\alpha]_D$ +11.5° (*c* 0.4, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.86 (t, 6H, 2*Me*CH_2), 1.25 (s, 32H, 16CH_2), 1.96-2.15 (8s, 24H, 8AcO), 2.63 (dd, 1H, J_{gem} = 12.8 Hz, $J_{3eq,4}$ = 4.3 Hz, H-3c*eq*), 3.74 (s, 3H, MeO), 4.60 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1a), 5.19 (dd, 1H, $J_{2,3}$ = 9.8 Hz, H-2a), 5.57 (m, 1H, H-8c), 5.78 (dt, 1H, $J_{4,5}$ = 14.8 Hz, $J_{5,6} = J_{5,6'}$ = 6.6 Hz, H-5 of sphingosine), and 7.25-8.06 (m, 20H, 4Ph).

Anal. Calcd for $C_{90}H_{115}NO_{33}$ (1738.9): C, 62.17; H, 6.67; N, 0.81. Found: C, 62.03; H, 6.85; N, 0.97.

***O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-(3-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-3-*O*-benzoyl-2-decanamido-4-octadecene-1,3-diol (9).** Reduction of the azido group in **2** (102 mg, 0.061 mmol) and subsequent coupling with decanoic acid (21 mg, 0.12 mmol) using WSC (23.5 mg), according to the procedure described for **5**, gave compound **9** (76 mg, 69%) as an amorphous mass: $[\alpha]_D$ +9.2° (*c* 0.3, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.89 (t, 6H, 2*Me*CH_2), 1.26 (s, 40H, 20CH_2), 1.87-2.21 (8s, 24H, 8AcO), 2.65 (dd, 1H,

J_{gem} = 12.5 Hz, $J_{3eq,4}$ = 4.5 Hz, H-3c*eq*), 3.78 (s, 3H, MeO), 4.63 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1a), 4.91 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1b), 5.22 (dd, 1H, $J_{2,3}$ = 9.1 Hz, H-2a), 5.45 (dd, 1H, $J_{6,7}$ = 2.0 Hz, $J_{7,8}$ = 7.9 Hz, H-7c), 5.57 (m, 1H, H-8c), 5.80 (m, 1H, H-5 of sphingosine), and 7.31~8.09 (m, 20H, 4Ph).

Anal. Calcd for $C_{94}H_{123}NO_{33}$ (1795.0): C, 62.90; H, 6.91; N, 0.78. Found: C, 62.86; H, 7.15; N, 0.83.

***O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-(3-*O*-acetyl-2,6-di-*O*-benzoyl-β-D-glucopyranosyl)-(1→1)-(2*S*,3*R*,4*E*)-3-*O*-benzoyl-2-hexadecanamido-4-octadecene-1,3-diol (10).** Reduction of the azido group in **2** (182.5 mg, 0.11 mmol) and subsequent coupling with hexadecanoic acid (56 mg, 0.22 mmol) using WSC (42 mg, 0.22 mmol), according to the procedure for **5**, gave compound **10** (162 mg, 79%) as an amorphous mass: $[\alpha]_D$ +10.1° (*c* 0.5, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.88 (t, 6H, 2*Me*CH_2), 1.26 (s, 52H, 26CH_2), 1.97~2.17 (8s, 24H, 8AcO), 2.64 (dd, 1H, J_{gem} = 12.7 Hz, H-3c*eq*), 3.74 (s, 3H, MeO), 4.63 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1a), 4.90 (d, 1H, $J_{1,2}$ = 8.2 Hz, H-1b), 5.22 (dd, 1H, $J_{2,3}$ = 9.3 Hz, H-2b), 5.57 (m, 1H, H-8c), 5.78 (dt, 1H, $J_{4,5}$ = 15.2 Hz, $J_{5,6}$ = $J_{5,6'}$ = 6.6 Hz, H-5 of sphingosine), and 7.25-8.07 (m, 20H, 4Ph).

Anal. Calcd for $C_{100}H_{135}NO_{33}$ (1879.2): C, 63.92; H, 7.24; N, 0.75. Found: C, 64.11; H, 7.20; N, 0.81.

***O*-(5-Acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-*O*-β-D-galactopyranosyl-(1→4)-*O*-β-D-glucopyranosyl-(1→1)-(2*S*,3*R*,4*E*)-2-hexanamido-4-octadecene-1,3-diol (11).** To a solution of **5** (85.1 mg, 0.05 mmol) in MeOH (5 mL) was added NaOMe (20 mg) and the mixture was stirred overnight at 40 °C; the course of the reaction was monitored by TLC. Water (0.2 mL) was added to the mixture, and this was stirred overnight at room temperature and then treated with Amberlite IR-120 (H^+) resin and filtered. The resin was washed with 1:1 $CHCl_3$-MeOH, and the combined filtrate and washings was concentrated. Column chromatography (1:1 $CHCl_3$-MeOH) of the residue on Sephadex LH-20 (20 g) gave **11** (48.6 mg, 98%) as an amorphous mass: $[\alpha]_D$ +6.5°

(*c* 0.5, 1:1 $CHCl_3$-MeOH); ^1H NMR (1:2 $CDCl_3$-CD_3OD) δ 0.99 (t, 6H, 2*Me*CH_2), 1.38 (s, 32H, 16CH_2), 2.10 (s, 3H, AcN), 2.27 (br t, 1H, H-3c*eq*), 4.15 (d, 1H, $J_{1,2}$ = 7.3 Hz, H-1a), 4.40 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1b), and 5.74 (m, 1H, H-5 of sphingosine).

Anal. Calcd for $C_{47}H_{84}N_2O_{21}$ (1013.2): C, 55.72; H, 8.36; N, 2.77. Found: C, 55.77; H, 8.49; N, 2.70.

***O*-(5-Acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-*O*-β-D-galactopyranosyl-(1→4)-*O*-β-D-glucopyranosyl-(1→1)-(2*S*,3*R*,4*E*)-2-decanamido-4-octadecene-1,3-diol (12).** The *O*-acyl and methyl ester groups in **6** (40 mg, 0.02 mmol) were removed, as described for **11**, to give compound **12** (22.4 mg, 93%) as an amorphous mass: $[\alpha]_D$ +4.5° (*c* 0.5, 1:1 $CHCl_3$-MeOH); ^1H NMR (1:2 $CDCl_3$-CD_3OD) δ 0.99 (t, 6H, 2*Me*CH_2), 1.39 (s, 40H, 20CH_2), 1.68 (br t, 1H, H-3c*ax*), 2.11 (s, 3H, AcN), 2.27 (br dd, 1H, H-3c*eq*), 3.99 (d, 1H, $J_{1,2}$ = 7.3 Hz, H-1a), 4.40 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1b), and 5.66 (m, 1H, H-5 of sphingosine).

Anal. Calcd for $C_{51}H_{92}N_2O_{21}$ (1069.3): C, 57.29; H, 8.67; N, 2.62. Found: C, 57.13; H, 8.90; N, 2.62.

***O*-(5-Acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-*O*-β-D-galactopyranosyl-(1→4)-*O*-β-D-glucopyranosyl-(1→1)-(2*S*,3*R*,4*E*)-2-hexadecanamido-4-octadecene-1,3-diol (13).** The *O*-acyl and methyl ester groups in **7** (95.4 mg, 0.05 mmol) were removed, as described for **11**, to give compound **13** (59 mg, quantitative) as an amorphous mass: $[\alpha]_D$ +1.5° (*c* 1.2, 1:1 $CHCl_3$-MeOH); ^1H NMR (1:2 $CDCl_3$-CD_3OD) δ 0.89 (t, 6H, 2*Me*CH_2), 1.24 (s, 52H, 26CH_2), 1.67 (br t, 1H, H-3c*ax*), 1.91 (s, 3H, AcN), 2.07 (br dd, 1H, H-3c*eq*), 3.95 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1a), 4.20 (d, 1H, $J_{1,2}$ = 7.7 Hz, H-1b), and 5.30~5.61 (m, 2H, H-4,5 of sphingosine).

Anal. Calcd for $C_{57}H_{104}N_2O_{21}$ (1153.5): C, 59.35; H, 9.09; N, 2.43. Found: C, 59.20; H, 9.18; N, 2.55.

***O*-(3-Deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-*O*-β-D-galactopyranosyl-(1→4)-*O*-β-D-glucopyranosyl-(1→1)-(2*S*,3*R*,4*E*)-2-hexanamido-4-octadecene-1,3-diol (14).** The *O*-acyl and

methyl ester groups in **8** (60.5 mg, 0.035 mmol) were removed, as described for **11**, to give compound **14** (34.3 mg, quantitative) as an amorphous mass: $[\alpha]_D$ -7.1° (*c* 0.8, 1:1 $CHCl_3$-MeOH); ^1H NMR [100:1 $(CD_3)_2SO$-H_2O] δ 0.88 (t, 6H, 2*Me*CH_2), 1.24 (s, 32H, 16CH_2), 2.06 (br t, 1H, H-3c*ax*), 2.43 (br dd, 1H, H-3c*eq*), 4.16 (d, 1H, $J_{1,2}$ = 7.5 Hz, H-1a), 4.24 (d, 1H, $J_{1,2}$ = 7.6 Hz, H-1b), 5.36 (dd, 1H, $J_{3,4}$ = 6.8 Hz, $J_{4,5}$ = 14.8 Hz, H-4 of sphingosine), and 5.57 (dt, 1H, $J_{5,6}$ = $J_{5,6'}$ = 6.1 Hz, H-5 of sphingosine).

Anal. Calcd for $C_{45}H_{81}NO_{21}$ (972.2): C, 55.60; H, 8.40; N, 1.44. Found: C, 55.43; H, 8.59; N, 1.41.

***O*-(3-Deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-*O*-β-D-galactopyranosyl-(1→4)-*O*-β-D-glucopyranosyl-(1→1)-(2*S*,3*R*,4*E*)-2-decanamido-4-octadecene-1,3-diol (15).** *O*-Deacylation and saponification of **9** (58 mg, 0.032 mmol), as described for **11**, yielded amorphous **15** (37.5 mg, quantitative): $[\alpha]_D$ -7.9° (*c* 0.8, 1:1 $CHCl_3$-MeOH); ^1H NMR [100:1 $(CD_3)_2SO$-H_2O] δ 0.87 (t, 6H, 2*Me*CH_2), 1.24 (s, 40H, 20CH_2), 2.05 (br t, 1H, H-3c*ax*), 2.63 (br dd, 1H, H-3c*eq*), 4.16 (d, 1H, $J_{1,2}$ = 7.6 Hz, H-1a), 4.20 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1b), 5.35 (dd, 1H, $J_{3,4}$ = 6.0 Hz, $J_{4,5}$ = 15.4 Hz, H-4 of sphingosine), and 5.56 (dt, 1H, $J_{5,6}$ = $J_{5,6'}$ = 6.1 Hz, H-5 of sphingosine).

Anal. Calcd for $C_{49}H_{89}NO_{21}$ (1028.3): C, 57.23; H, 8.72; N, 1.36. Found: C, 57.05; H, 8.93; N, 1.35.

***O*-(3-Deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-*O*-β-D-galactopyranosyl-(1→4)-*O*-β-D-glucopyranosyl-(1→1)-(2*S*,3*R*,4*E*)-2-hexadecanamido-4-octadecene-1,3-diol (16).** *O*-Deacylation and saponification of **10** (70.6 mg, 0.038 mmol), as described for **11**, yielded amorphous **16** (39.8 mg, 94%): $[\alpha]_D$ -7.3° (*c* 0.8, 1:1 $CHCl_3$-MeOH); ^1H NMR [100:1 $(CD_3)_2SO$-H_2O] δ 0.86 (t, 6H, 2*Me*CH_2), 1.23 (s, 52H, 26CH_2), 2.03 (br t, 1H, H-3c*ax*), 2.53 (br dd, 1H, H-3c*eq*), 4.16 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1a), 4.23 (d, 1H, $J_{1,2}$ = 7.5 Hz, H-2b), 5.35 (dd, 1H, $J_{3,4}$ = 6.1 Hz, $J_{4,5}$ = 15.2 Hz, H-4 of sphingosine), and 5.54 (dt, 1H, $J_{5,6}$ = $J_{5,6'}$ = 6.4 Hz, H-5 of sphingosine).

Anal. Calcd for $C_{55}H_{101}NO_{21}$ (1112.4): C, 59.39; H, 9.15; N, 1.26. Found: C, 59.42; H, 9.33; N, 1.24.

***O*-(5-Acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-*O*-β-D-galactopyranosyl-(1→4)-*O*-β-D-glucopyranosyl-(1→1)-(2*S*,3*R*,4*E*)-2-amino-4-octadecene-1,3-diol (lyso-GM3) (18).** Hydrogen sulfide was bubbled through a stirred solution of **17**, prepared from **1** by *O*-deacylation and subsequent de-esterification (49.5 mg, 0.06 mmol) in aq 83% pyridine (12 mL) for 3 days at room temperature. The reaction was monitored by TLC. After completion of the reaction, the mixture was concentrated to a syrup which was chromatographed on a column of Sephadex LH-20 (20 g) with 1:1 $CHCl_3$-MeOH to give **18** (39 g, 80%) as an amorphous mass: $[\alpha]_D$ -4.5° (*c* 0.5, 1:1 $CHCl_3$-MeOH); ^1H NMR (1:1 $CDCl_3$-CD_3OD) δ 0.86 (t, 3H, 2*Me*CH_2), 1.25 (s, 22H, 11CH_2), 1.91 (s, 3H, AcN), 2.78 (br dd, 1H, H-3c*eq*), 4.20 (d, 1H, $J_{1,2}$ = 7.5 Hz, H-1a), 4.25 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1b), 5.44 (dd, 1H, $J_{3,4}$ = 6.0 Hz, $J_{4,5}$ = 15.2 Hz, H-4 of sphingosine), and 5.73 (dt, 1H, $J_{5,6}$ = $J_{5,6'}$ = 6.6 Hz, H-5 of sphingosine).

Anal. Calcd for $C_{41}H_{74}N_2O_{20}$ (915.0): C, 53.82; H, 8.15; N, 2.62. Found: C, 53.61; H, 8.20; N, 2.56.

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