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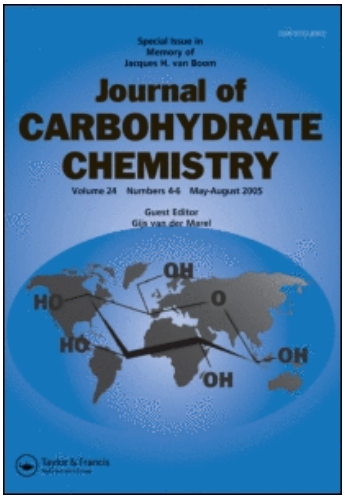

Journal of Carbohydrate Chemistry
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713617200

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Makoto Kiso[a]; Akerai Nakamura[a]; Akira Hasegawa[a]
[a] Department of Agricultural Chemistry, Gifu University, Gifu, Japan

To cite this Article Kiso, Makoto , Nakamura, Akerai and Hasegawa, Akira(1987) 'Synthesis of 1-O-(N-Acetyl-α- and -β-D-Neuraminyl)-Ceramides', Journal of Carbohydrate Chemistry, 6: 3, 411 — 422
To link to this Article: DOI: 10.1080/07328308708057929
URL: http://dx.doi.org/10.1080/07328308708057929

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J. CARBOHYDRATE CHEMISTRY, 6(3), 411-422 (1987)

SYNTHESIS OF 1-O-(N-ACETYL-α- AND -β-D-NEURAMINYL)-CERAMIDES

Makoto Kiso, Akemi Nakamura and Akira Hasegawa

Department of Agricultural Chemistry
Gifu University, Gifu 501-11, Japan

Received February 5, 1987 - Final Form April 27, 1987

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ABSTRACT

3-O-Protected D-erythro-ceramides, [2(S),3(R),4E]-3-O-acetyl-2-octadecanamido-4-octadecene-1,3-diol (8), [2(S),3(R),4E]-3-O-benzoyl-2-octadecanamido- and -tetracosanamido-4-octadecene-1,3-diol (9 and 10) were each coupled with methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-D-glycero-β-D-galacto-2-nonulopyranosylchlorid)-onate (11), and the resulting glycosides were deprotected in a stepwise manner to give the desired 1-O-(N-acetyl-α- and -β-D-neuraminyl)-ceramides (17α,β and 18α,β), respectively.

INTRODUCTION

Sialic acid[1] and ceramide[2] are the essential constituents of gangliosides[3,4] which participate in the important biological functions on cell surfaces serving as antigens, receptors of viruses, toxins and hormones, and as mediators of cell growth control. Some gangliosides have also been established as tumor-specific markers.

In a synthetic approach to investigate the relationship between the molecular structure and the function (biological activity) of gangliosides and related glycolipids, we have recently developed new procedures for the synthesis of D-erythro-sphingosine and -ceramide[5], and a variety of thioglycosides of N-acetylneuraminic

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acid.[6] The present paper describes a synthesis of 1-O-sialyl ceramide derivatives, the minimal structural units of gangliosides. These compounds may be useful for the modification of cell surfaces.

RESULTS AND DISCUSSION

The D-erythro-ceramides,[5] [2(S),3(R),4E]-2-octadecanamido-4-octadecene-1,3-diol (1), and [2(S),3(R),4E]-2-tetracosanamido-4-octadecene-1,3-diol (2) were tritylated in the usual way, to give 3[7] and 4[8], which were then acylated with acetic anhydride or benzoyl chloride in pyridine. The resulting 3-O-acetyl-1-O-trityl derivatives (5-7) were treated with aqueous acetic acid at 45-50 °C to afford the corresponding 3-O-protected ceramides 8-10, respectively. When the detritylation from 5 was conducted at a higher temperature, acetyl migration occurred to give the 1-O-acetyl derivative as a major by-product.

Silver triflate promoted glycosylations of 8-10 with methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-2-chloro-2,3,5-trideoxy-D-glycero-β-D-galacto-2-nonulopyranosonate (11)[9] were performed by the procedure reported by van der Vleugel et al.[10], to give the corresponding 12α,β, 13α,β and 14α,β as approximately 1:1 mixtures of the α- and β-glycosides in 46-57% yield. In the ^{1}H NMR spectra of these latter compounds, the resonances characteristic of both the sialic acid (Neu5Ac) and ceramide moieties were clearly observed, and H-3e of the Neu5Ac moiety appeared as a doublet of doublets ($J_{3a,3e}$ = 13, $J_{3e,4}$ = 4-5 Hz) at δ 2.61 (for 12α), 2.51 (for 12β), 2.59 (for 13α and 14α) and 2.46 ppm (for 13β and 14β), respectively. Since the synthetic objective was to obtain both the α- and β-glycosides of ceramides simultaneously, other glycosylation methods were not examined.

Treatment of 12α,β, 13α,β and 14α,β with methanolic sodium methoxide gave 15α,β and 16α,β quantitatively. The isomeric forms of 15 and 16 were then obtained by preparative thin layer chromatography (TLC). The desired final products (17α,β and 18α,β) were obtained by saponification of the methyl ester from 15α,β and 16α,β, respectively.

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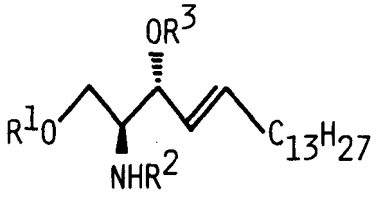
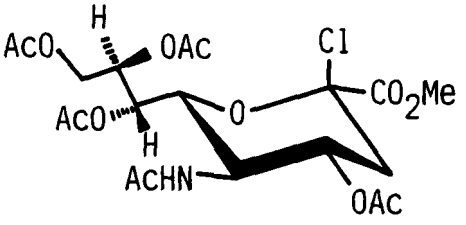


	R^1	R^2	R^3
1	H	Ste	H
2	H	Lig	H
3	Tr	Ste	H
4	Tr	Lig	H
5	Tr	Ste	Ac
6	Tr	Ste	Bz
7	Tr	Lig	Bz
8	H	Ste	Ac
9	H	Ste	Bz
10	H	Lig	Bz



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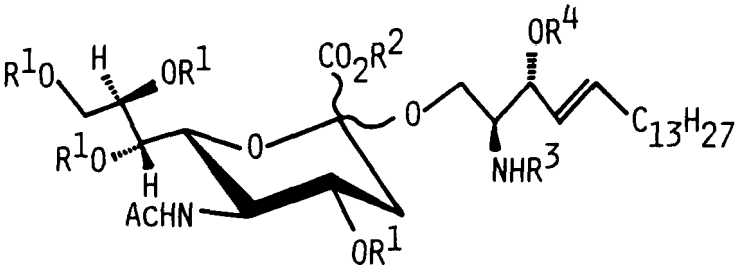

	R^1	R^2	R^3	R^4
12α, 12β	Ac	Me	Ste	Ac
13α, 13β	Ac	Me	Ste	Bz
14α, 14β	Ac	Me	Lig	Bz
15α, 15β	H	Me	Ste	H
16α, 16β	H	Me	Lig	H
17α, 17β	H	H	Ste	H
18α, 18β	H	H	Lig	H

Ste = $CH_3(CH_2)_{16}CO$, Lig = $CH_3(CH_2)_{22}CO$

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Recently a variety of new biological activities of sphingosine[11] and glycosphingolipids[12,13] have been found. Therefore it is of interest to determine what biological activities are expressed by artificial glycolipids such as 17α,β and 18α,β.

EXPERIMENTAL

General Procedures. Melting points were determined with a Yanagimoto micro melting-point apparatus and are uncorrected. Specific rotations were determined with a Union PM-201 polarimeter, and IR spectra were recorded with a Jasco A-100 spectrophotometer. ^{1}H NMR spectra were recorded at 270 MHz with a JEOL JNM-GX270 spectrometer. Preparative TLC was performed on silica gel 60 (Merck Co.), and column chromatography on silica gel (Wako Co.; 200 mesh) was accomplished with the solvent systems (v/v) specified. Concentrations and evaporations were conducted in vacuo.

[2(S),3(R),4E]-2-Octadecanamido-1-O-trityl-4-octadecene-1,3-diol (3). A solution of 1[5] (0.5 g) in dry pyridine (7 mL) was stirred at 90 °C with trityl chloride (0.32 g). After completion of the reaction (TLC, 40:1 dichloromethane-methanol), the mixture was cooled, and methanol was added in order to decompose excess reagent. Solvents were evaporated, and a solution of the residual syrup in dichloromethane was successively washed with ice-cold M hydrochloric acid, water, 10% sodium carbonate, and water, and then dried over anhydrous sodium sulfate, and concentrated. The residue was chromatographed on a column of silica gel with 500:1 dichloromethane-methanol, to give 3 (0.64 g, 93%); $[\alpha]_D$ -1° (c 0.87, chloroform); ^{1}H NMR ($CDCl_3$): δ 3.30, 3.39 (2dd, 2H, J_{gem} = 10, $J_{1,2}$ = 4 and 3.7 Hz, H-1,1'), 4.05 (m, 1H, H-2), 4.18 (broad m, 1H, H-3), and 7.1-7.45 (m, 15H, Ph).

Anal. Calcd for $C_{55}H_{85}NO_3$ (808.24): C, 81.73; H, 10.60; N, 1.73. Found: C, 81.52; H, 10.49; N, 1.71.

[2(S),3(R),4E]-3-O-Acetyl-2-octadecanamido-1-O-trityl-4-octadecene-1,3-diol (5) and [2(S),3(R),4E]-3-O-Benzoyl-2-

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octadecanamido-1-O-trityl-4-octadecene-1,3-diol (6). 3-O-Acetylation and benzoylation of 3 were performed in the usual manner with acetic anhydride or benzoyl chloride in pyridine, respectively, to give 5 and 6 in near quantitative yield.

Compound 5 had $[\alpha]_D$ -13° (c 1, chloroform): IR (film) 1740 (ester), and 1650 and 1540 cm^{-1} (amide); ^1H NMR ($CDCl_3$) δ 3.13, 3.30 (2dd, 2H, H-1,1'), 4.33 (m, 1H, H-2), 5.33 (dd, 1H, $J_{4,5}$ = 15, $J_{3,4}$ = 7 Hz, H-4), 5.44 (t, 1H, H-3), 5.63 (d, 1H, J = 9.5 Hz, NH), 5.76 (m, 1H, H-5), and 7.1-7.5 (m, 15H, Ph).

Anal. Calcd for $C_{57}H_{87}NO_4$ (850.28): C, 80.51; H, 10.31; N, 1.65. Found: C, 80.24; H, 10.25; N, 1.63.

Compound 6 had $[\alpha]_D$ +7° (c 0.65, chloroform): IR (film) 1740 (ester), and 1650 and 1560 cm^{-1} (amide); ^1H NMR ($CDCl_3$) δ 3.20, 3.44 (2dd, 2H, H-1,1'), 4.49 (m, 1H, H-2), 5.45 (dd, 1H, $J_{4,5}$ = 15.4, $J_{3,4}$ = 7.7 Hz, H-4), 5.65 (d, 1H, NH), 5.70 (t, 1H, H-3), 5.87 (m, 1H, H-5), and 7.1-8.0 (m, 20H, Ph).

Anal. Calcd for $C_{62}H_{89}NO_4$ (912.35): C, 81.62; H, 9.83; N, 1.54. Found: C, 81.36; H, 10.01; N, 1.40.

[2(S),3(R),4E]-3-O-Benzoyl-2-tetracosanamido-1-O-trityl-4-octadecene-1,3-diol (7). Benzoylation of 4 was performed as described for 6 to give 7: $[\alpha]_D$ +10° (c 0.5, chloroform); IR and ^1H NMR spectra were similar to those of 6.

Anal. Calcd for $C_{68}H_{101}NO_4$ (996.50): C, 84.00; H, 7.98; N, 1.44. Found: C, 84.32; H, 7.84; N, 1.57.

[2(S),3(R),4E]-3-O-Acetyl-2-octadecanamido-4-octadecene-1,3-diol (8) and [2(S),3(R),4E]-3-O-Benzoyl-2-octadecanamido-4-octadecene-1,3-diol (9). To a solution of 5 or 6 (0.6 g) in acetic acid (30 mL) was added water (2 mL), the mixture was stirred for 3 h at 45 °C and then concentrated to dryness. The residue was crystallized from ethanol to give 8 or 9, respectively.

Compound 8 had mp 87-88 °C: $[\alpha]_D$ -21° (c 1.2, 50:1 chloroform-methanol); IR (Nujol) 1740 (ester), and 1640 and 1540 cm^{-1} (amide); ^1H NMR ($CDCl_3$) δ 0.88 (t, 6H, Me), 1.0-1.4 and 1.5-1.7 (m, 50H + 2H, $-CH_2-$), 2.03 (near q, 2H, $-CH=CH-CH_2-$), 2.10 (s, 3H, AcO), 2.17 (m, 2H, $-COCH_2-$), 2.81 (broad s, 1H, OH), 3.55-3.75 (m, 2H, H-1,1'), 4.12 (m, 1H, H-2), 5.29 (t, 1H, H-3), 5.46 (near dd, 1H, $J_{4,5}$ = 15,

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$J_{3,4}$ = 7.7, $J_{4,6(6')}$ = 1.5 Hz, H-4), 5.77 (m, 1H, $J_{5,6(6')}$ = 7 Hz, H-5), and 5.95 (d, 1H, J = 8.4 Hz, NH).

Anal. Calcd for $C_{38}H_{73}NO_4$ (608.00): C, 75.07; H, 12.10; N, 2.30. Found: C, 75.37; H, 11.95; N, 2.30.

Compound 9 had mp 86.5-87.5 °C: $[\alpha]_D$ +16° (c 0.5, chloroform); IR (Nujol): 1730 (ester), and 1640 and 1540 cm^{-1} (amide); ^1H NMR ($CDCl_3$) 2.05 (near q, 2H, -CH=CH-CH_2-), 2.19 (m, 2H, -$COCH_2$-), 3.6-3.8 (m, 2H, H-1,1'), 4.27 (m, 1H, H-2), 5.53 (t, 1H, $J_{2,3}$ = $J_{3,4}$ 7.3 Hz, H-3), 5.60 (near dd, 1H, H-4; partly overlapping with H-3), 5.86 (m, 1H, $J_{4,5}$ = 15, $J_{5,6(6')}$ = 7 Hz, H-5), 6.06 (near d, 1H, NH), and 7.4-8.1 (m, 5H, Ph).

Anal. Calcd for $C_{43}H_{75}NO_4$ (670.07): C, 77.08; H, 11.28; N, 2.09. Found: C, 76.86; H, 11.09; N, 2.03.

[2(S),3(R),4E]-3-O-Benzoyl-2-tetracosanamido-4-octadecene-1,3-diol (10). Treatment of 7 (0.75 g) with aqueous acetic acid at 50 °C as described for 9 gave 10[8,14] (quantitative) which crystallized from ethanol; mp 88.5-89.5 °C (lit.[14] 86-88 °C); $[\alpha]_D$ +20° (c 0.5, 50:1 chloroform-methanol) (lit.[8] +16.5°, lit.[14] +17.7°); ^1H NMR similar to that of 9 except for the number of methylene protons.

[2(S),3(R),4E]-3-O-Acetyl-1-O-(methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-D-glycero-α- and -β-D-galacto-2-nonulopyranosylonate)-2-octadecanamido-4-octadecene-1,3-diol (12α and 12β). To a stirred solution of 8 (0.2 g, 0.33 mmol) in dichloromethane (5 mL) were successively added 4 A molecular sieves (0.2 g), chloride 11 (0.502 g, 1 mmol), 2,4,6-trimethylpyridine (0.16 mL) in dry 1:1 ether-nitromethane (1 mL), and silver triflate (0.385 mg). Stirring was continued overnight in the dark at room temperature. The suspension was filtered and washed with chloroform. The filtrate and washings were combined, successively washed with ice-cold M hydrochloric acid, 5% sodium thiosulphate, 10% sodium carbonate, and water, and then dried, and concentrated. The products were isolated by preparative TLC (Kieselgel 60 F_{254} PTLC, 30:1 ether-methanol) to give 12α (0.097 g, 27% based on 8) and 12β (0.106 g, 30%).

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Compound 12α (smaller Rf) had $[\alpha]_D$ -8° (c 0.65, chloroform): ^1H NMR ($CDCl_3$); Neu5Ac unit δ 1.88 (s, 3H, AcN), 2.61 (dd, 1H, $J_{3a,3e}$ = 13, $J_{3e,4}$ = 4.6 Hz, H-3e), and 3.80 (s, 3H, MeO); ceramide unit δ 0.88 (t, 6H, Me), 1.0-1.4 (m, 50H, $-CH_2-$), and 5.29 (t, 1H, H-3); other protons: δ 2.03, 2.04(6H), 2.12, 2.14 (4s, 15H, AcO).

Anal. Calcd for $C_{58}H_{100}N_2O_{16}$ (1081.43): C, 64.42; H, 9.32; N, 2.59. Found: C, 64.23; H, 9.49; N, 2.60.

Compound 12β (larger Rf) had $[\alpha]_D$ -11° (c 0.53, chloroform): ^1H NMR ($CDCl_3$); Neu5Ac unit δ 1.91 (s, 3H, AcN), 2.51 (dd, 1H, $J_{3a,3e}$ = 13, $J_{3e,4}$ = 5 Hz, H-3e), 3.71 (dd, 1H, $J_{5,6}$ = 10.6, $J_{6,7}$ = 2.6 Hz, H-6), 3.78 (s, 3H, MeO), 4.01, 4.82 (2dd, 2H, J_{gem} = 12.5, $J_{8,9}$ = 8.4 and 2.2 Hz, H-9,9'), 4.19 (q, 1H, $J_{4,5} = J_{5,6} = J_{5,NH}$ 10 Hz, H-5), 5.09 (m, 1H, $J_{7,8}$ = 2.6 Hz, H-8), and 5.34 (t, 1H, H-7); ceramide unit δ 0.88 (t, 6H, Me), 1.0-1.4 (m, 50H, $-CH_2-$), 3.33, 3.55 (2dd, 2H, J_{gem} = 10, $J_{1,2}$ = 2.7 and 2.5 Hz, H-1,1'), 4.33 (m, 1H, H-2), 5.45 (near q, 1H, $J_{4,5}$ = 15, $J_{3,4}$ = 8.4 Hz, H-4), 5.56 (t, 1H, J = 8.6 Hz, H-3), and 5.80 (m, 1H, $J_{5,6(6')}$ = 7 Hz, H-5); other protons δ 2.02, 2.03, 2.06, 2.16, 2.18 (5s, 15H, AcO), and 5.95-6.05 (2d, 2H, NH).

Anal. Found: C, 64.62; H, 9.46; N, 2.63.

[2(S),3(R),4E]-3-O-Benzoyl-1-O-(methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-D-glycero-α- and -β-D-galacto-2-nonulopyranosylonate)-2-octadecanamido-4-octadecene-1,3-diol (13α and 13β). Condensation of 9 (0.3 g, 0.45 mmol) and 11 (0.455 g, 0.9 mmol) was performed as described for 12α and 12β, to give 13α (0.133 g, 26% based on 9) and 13β (0.159 g, 31%).

Compound 13α had $[\alpha]_D$ +0.4° (c 0.9, chloroform): ^1H NMR ($CDCl_3$); Neu5Ac unit δ 1.87 (s, 3H, AcN), 2.59 (dd, 1H, $J_{3a,3e}$ = 13, $J_{3e,4}$ = 4.6 Hz, H-3e), 3.57 (s, 3H, MeO), 4.30 (dd, 1H, J_{gem} = 12.7 Hz, H-9), and 4.86 (m, 1H, H-4); ceramide unit δ 0.88 (t, 6H, Me), 1.0-1.7 (m, 52H, $-CH_2-$), 3.52, 3.87 (2dd, 2H, J_{gem} = 9.8, $J_{1,2}$ = 4.9 and 3.9 Hz, H-1,1'), 4.51 (m, 1H, H-2), and 7.4-8.1 (m, 5H, Ph); other protons δ 2.03(6H), 2.07, 2.12 (3s, 12H, AcO).

Anal. Calcd for $C_{63}H_{102}N_2O_{16}$ (1143.51): C, 66.17; H, 8.99; N, 2.45. Found: C, 66.39; H, 8.84; N, 2.33.

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Compound 13β had $[\alpha]_D$ +1° (c 0.6, chloroform): ^{1}H NMR ($CDCl_3$); Neu5Ac unit δ 2.46 (dd, 1H, $J_{3a,3e}$ = 13, $J_{3e,4}$ = 5.4 Hz, H-3e), 3.61 (dd, 1 H, $J_{5,6}$ = 10.7, $J_{6,7}$ = 2.4 Hz, H-6), 3.77 (s, 3H, MeO), 3.90, 4.64 (2dd, 2H, J_{gem} = 12.7, $J_{8,9}$ = 7.3 and 2.2 Hz, H-9,9'), 4.11 (q, 1H, $J_{4,5} = J_{5,6} = J_{5,NH}$ = 10.4 Hz, H-5), 5.22 (m, 1H, $J_{3a,4}$ = 11 Hz, H-4); ceramide unit δ 0.88 (t, 6H, Me), 1.1-1.7 (m, 52H, $-CH_2-$), 3.41, 3.72 (2dd, 2H, J_{gem} = 10, $J_{1,2}$ = 2.4 Hz, H-1,1'), 4.52 (m, 1H, H-2), 5.56 (dd, 1H, $J_{4,5}$ = 15, $J_{3,4}$ = 8.3 Hz, H-4), 5.72 (t, 1H, J = 8.3 Hz, H-3), 5.91 (m, 1H, $J_{5,6(6')}$ = 7 Hz, H-5), and 7.45-8.15 (m, 5H, Ph); other protons δ 1.85, 1.88, 1.96, 2.02, 2.11 (5s, 15H, AcN and 4AcO), and 5.31 and 6.17 (2d, 2H, NH).

Anal. Found: C, 65.98; H, 9.06; N, 2.41.

[2(S),3(R),4E]-3-O-Benzoyl-1-O-(methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-D-glycero-α- and -β-D-galacto-2-nonulopyranosylonate)-2-tetracosanamido-4-octadecene-1,3-diol (14α and 14β). Condensation of 10 (0.39 g, 0.52 mmol) with 11 (0.526 g, 1 mmol) was carried out as described for 12α,β and 13α,β, to give 14α (0.134 g, 21% based on 10) and 14β (0.158 g, 25%).

Compound 14α had $[\alpha]_D$ -2° (c 0.5, chloroform): ^{1}H NMR spectrum was quite similar to that of 13α except for the number of methylene protons in the ceramide unit.

Anal. Calcd for $C_{69}H_{114}N_2O_{16}$ (1227.62): C, 67.51; H, 9.36; N, 2.28. Found: C, 67.70; H, 9.27; N, 2.34.

Compound 14β had $[\alpha]_D$ +2.4° (c 1.4, chloroform): ^{1}H NMR spectrum was almost the same as that of 13β, except for the number of methylene protons in the ceramide unit and the chemical shift of one NH proton (δ 5.25).

Anal. Found: C, 67.73; H, 9.48; N, 2.26.

[2(S),3(R),4E]-1-O-(Methyl 5-acetamido-3,5-dideoxy-D-glycero-α- and -β-D-galacto-2-nonulopyranosylonate)-2-octadecanamido-4-octadecene-1,3-diol (15α and 15β). O-Deacylations of 12α and 13α, or 12β and 13β were performed with a catalytic amount of sodium methoxide in methanol solution. The resulting 15α and 15β were each

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purified by chromatography on a column of silica gel with 30:1 dichloromethane-methanol.

Compound 15α had mp 95-96 °C; $[\alpha]_D$ +3° (c 0.5, methanol): IR (KBr) 3600-3100 (OH), 1720 (ester), and 1650 and 1550 cm^{-1} (amide); ^1H NMR (CD_3OD); Neu5Ac unit δ 1.79 (near t, 1H, $J_{3a,3e}$ = 13, $J_{3a,4}$ = 12 Hz, H-3a), 2.0 (s, 3H, AcN), 2.69 (dd, 1H, $J_{3e,4}$ = 4.4 Hz, H-3e), and 3.83 (s, 3H, MeO); ceramide unit δ 0.90 (near t, 6H, Me), 1.0-1.7 (m, 52H, $-CH_2-$), 1.9-2.1 (m, 2H, $-CH=CH-CH_2-$), 2.18 (near t, 2H, $-COCH_2-$), 5.44 (dd, 1H, $J_{4,5}$ = 15.4, $J_{3,4}$ = 7.3 Hz, H-4), and 5.67 (m, 1H, $J_{5,6(6')}$ = 6-7 Hz, H-5).

Anal. Calcd for $C_{48}H_{90}N_2O_{11}$ (871.25): C, 66.17; H, 10.41; N, 3.22. Found: C, 65.98; H, 10.32. N, 3.16.

Compound 15β had mp 159-160 °C; $[\alpha]_D$ -4° (c 0.5, methanol): IR (KBr) 3600-3100 (OH), 1730 (ester), and 1650 and 1550 cm^{-1} (amide); ^1H NMR (CD_3OD); Neu5Ac unit δ 1.65 (near t, 1H, $J_{3a,3e}$ = 12.8, $J_{3a,4}$ = 11.4 Hz, H-3a), 2.01 (s, 3H, AcN), 2.42 (dd, 1H, $J_{3e,4}$ = 4.8 Hz, H-3e), and 3.77 (s, 3H, MeO); ceramide unit δ 0.90 (near t, 6H, Me), 1.0-1.7 (m, 52H, $-CH_2-$), 2.20 (near t, 2H, $-COCH_2-$), 5.46 (dd, 1H, $J_{4,5}$ = 15.4, $J_{3,4}$ = 7 Hz, H-4), and 5.71 (m, 1H, $J_{5,6(6')}$ = 6-7 Hz, H-5).

Anal. Found: C, 66.03; H, 10.22; N, 3.34.

[2(S),3(R),4E]-1-O-(Methyl 5-acetamido-3,5-dideoxy-D-glycero-α- and -β-D-galacto-2-nonulopyranosylonate)-2-tetracosanamido-4-octadecene-1,3-diol (16α and 16β). O-Deacetylations of 14α and 14β were carried out as described for the preparation of 15α and 15β to afford 16α and 16β, respectively.

Compound 16α had mp 76-77.5 °C; $[\alpha]_D$ -2° (c 0.6, methanol): ^1H NMR (CD_3OD); Neu5Ac unit δ 1.79 (near t, $J_{3a,3e}$ = 13, $J_{3a,4}$ = 12 Hz, H-3a), 2.0 (s, 3H, AcN), 2.68 (dd, 1H, $J_{3e,4}$ = 4.4 Hz, H-3e), and 3.83 (s, 3H, MeO); ceramide unit δ 0.90 (near t, 6H, Me), and (m, 64H, $-CH_2-$); other protons similar to those of 15α.

Anal. Calcd for $C_{54}H_{102}N_2O_{11}$ (955.41): C, 67.89; H, 10.76; N, 2.93. Found: C, 68.17; H, 10.55; N, 3.04.

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Compound 16β had mp 134-135.5 °C; $[\alpha]_D$ -4° (c 0.5, 3:1 methanol-chloroform): ^{1}H NMR (CD_3OD + $CDCl_3$); Neu5Ac unit δ 1.70 (near t, 1H, $J_{3a,3e}$ = 13, $J_{3a,4}$ = 11-12 Hz, H-3a), 2.04 (s, 3H, AcN), 2.44 (dd, 1H, $J_{3e,4}$ = 4.8 Hz, H-3e), and 3.80 (s, 3H, MeO); ceramide unit δ 0.89 (near t, 6H, Me), 1.0-1.7 (m, 64H, $-CH_2-$), 5.46 (dd, 1H, H-4), and 5.72 (m, 1H, H-5).

Anal. Found: C, 68.10; H, 10.68; N, 2.91.

[2(S),3(R),4E]-1-O-(5-Acetamido-3,5-dideoxy-D-glycero-α- and -β-D-galacto-2-nonulopyranosylonic acid)-2-octadecanamido-4-octadecene-1,3-diol (17α and 17β). To a solution of 15α or 15β (0.03 g) in methanol (3 mL) was added 0.1 M potasium hydroxide (0.43 mL). The mixture was stirred for 3 h at 0 °C, 3 h at room temperature, and then treated with Amberlite IR-120 (H^+) resin to remove the base. The suspension was filtered, and the resin was washed with methanol. The filtrate and washings were combined, and concentrated to a residue, which crystallized from 1,4-dioxane.

Compound 17α had mp 120-121.5 °C; $[\alpha]_D$ +5° (c 0.4, 3:1 methanol-chloroform): ^{1}H NMR (CD_3OD + $CDCl_3$); Neu5Ac unit δ 2.05 (s, 3H, AcN), 2.75 (dd, 1H, $J_{3a,3e}$ = 13, $J_{3e,4}$ = 4.4 Hz, H-3e), and complete disappearance of CO_2CH_3; ceramide unit δ 0.89 (near t, 6H, Me), 1.0-1.7 (m, 52H, $-CH_2-$), 5.43 (m, 1H, $J_{4,5}$ = 15.4, $J_{3,4}$ = 7-8 Hz, H-4), and 5.68 (m, 1H, $J_{5,6(6')}$ = 6-7 Hz, H-5).

Anal. Calcd for $C_{47}H_{88}N_2O_{11}$ (857.22): C, 65.85; H, 10.35; N, 3.27. Found: C, 66.15; H, 10.21; N, 3.18.

Compound 17β had mp 163-164.5 °C; $[\alpha]_D$ -3° (c 0.5, 3:1 methanol-chloroform): ^{1}H NMR (CD_3OD + $CDCl_3$); Neu5Ac unit δ 2.05 (s, 3H, AcN), 2.43 (dd, 1H, $J_{3a,3e}$ = 13, $J_{3e,4}$ = 4.4 Hz, H-3e), and complete disappearance of CO_2CH_3; ceramide unit δ 0.89 (near t, 6H, Me), 1.0-1.7 (m, 52H, $-CH_2-$), 5.48 (dd, 1H, $J_{4,5}$ = 15.4, $J_{3,4}$ = 7.3 Hz, H-4), and 5.72 (m, 1H, $J_{5,6(6')}$ = 6.6 Hz, H-5).

Anal. Found: C, 66.17; H, 10.13; N, 3.20.

[2(S),3(R),4E]-1-O-(5-Acetamido-3,5-dideoxy-D-glycero-α- and -β-D-galacto-2-nonulopyranosylonic acid)-2-tetracosanamido-4-octadecene-1,3-diol (18α and 18β). Saponification of the methyl

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ester in 16α and 16β was performed as described for the preparation of 17α and 17β, to give 18α and 18β, respectively, in 95-98% yield.

Compound 18α had mp 98-99.5 °C; $[\alpha]_D$ -4° (c 0.4, 3:1 methanol-chloroform): ^{1}H NMR (CD_3OD + $CDCl_3$) Neu5Ac unit δ 2.04 (s, 3H, AcN), 2.73 (dd, 1H, H-3e), and complete disappearance of CO_2CH_3; ceramide unit δ 0.89 (near t, 6H, Me), 1.0-1.7 (m, 64H, $-CH_2-$), 5.46 (dd, 1H, H-4), and 5.69 (m, 1H, H-5).

Anal. Calcd for $C_{53}H_{100}N_2O_{11}$ (941.39): C, 67.62; H, 10.71; N, 2.98. Found: C, 67.34; H, 10.51; N, 3.08.

Compound 18β had mp 164-165.5 °C; $[\alpha]_{DD}$ -13° (c 0.4, 3:1 methanol-chloroform): ^{1}H NMR (CD_3OD + $CDCl_3$); Neu5Ac unit δ 2.05 (s, 3H, AcN), 2.43 (dd, 1H, H-3e), and complete loss of CO_2CH_3; ceramide unit δ 0.89 (near t, 6H, Me), 1.0-1.7 (m, 64H, $-CH_2-$), 5.48 (dd, 1H, $J_{4,5}$ = 15.4, $J_{3,4}$ = 7.3 Hz, H-4), and 5.72 (m, 1H, H-5).

Anal. Found: C, 67.32; H, 10.96; N, 2.82.

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