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Carbohydrate Research 300 (1997) 207–217

CARBOHYDRATE RESEARCH

A synthetic approach to the c-series gangliosides containing sialyl-α(2 → 8)sialyl-α(2 → 8)sialic acid: Synthesis of ganglioside GT4, α(2 → 6)GT4 and GT3 [1]

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Received 8 October 1996; accepted 27 January 1997

Abstract

Trimeric sialic acid [Neu5Acα(2 → 8)Neu5Acα(2 → 8)Neu5Ac, **1**] residue-containing gangliosides, GT4, α(2 → 6)GT4 and GT3, have been synthesized for the first time. Methyl [phenyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-2-thio-D-*glycero*-D-*galacto*-2-nonulopyranosid]onate (**3**) was prepared from **1**, via lactonization, methyl esterification of the carboxyl group at the reducting end, *O*-acetylation and conversion of the anomeric acetoxy group into a phenylthio group. Iodonium-promoted glycosylation of **3** with 2-(trimethylsilyl)ethyl 2,6-di-*O*-benzyl-β-D-galactopyranoside (**5**), 2-(trimethylsilyl)ethyl 3-*O*-benzyl-β-D-galactopyranoside (**6**), 2-(trimethylsilyl)ethyl 2-*O*-benzoyl-3-*O*-benzyl-β-D-galactopyranoside (**9**), and 2-(trimethylsilyl)ethyl 2,3-di-*O*-benzyl-β-D-galactopyranoside (**11**) gave the corresponding tetrasaccharides (**13–15**, **17**) having the $(Neu5Ac)_3$-Gal structure. The peracylated oligosaccharides **18** and **24** derived from **13** and **17**, and the previously reported lactose derivative **29** were converted into the α-trichloroacetimidates **20**, **26** and **31**, and coupled with (2*S*,3*R*,4*E*)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol (**21**) to afford the corresponding β-glycosides **22**, **27** and **32**. These protected azidosphingosine derivatives were each transformed into the target gangliosides GT4, α(2 → 6)GT4 and GT3 via selective reduction of the azido group, subsequent coupling with octadecanoic acid, *O*-deacylation and saponification of the methyl ester and lactone groups. © 1997 Elsevier Science Ltd.

Keywords: C-series gangliosides, synthesis; Trimeric sialic acid; Polysialo gangliosides

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[1] Synthetic Studies on Sialoglycoconjugates, Part 92. For Part 91, see ref. [1].

[2] Deceased 10 October 1996.

PII S0008-6215(97)00051-7

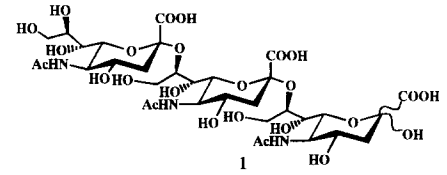
1. Introduction

c-Series gangliosides, which are distinguished from other gangliosides in that they contain a sialyl-$\alpha(2 \rightarrow 8)$sialyl-$\alpha(2 \rightarrow 8)$sialic acid residue as a constituent, were first isolated from fish brain by Ishizuka et al. [2]. This type of ganglioside is also expressed in the mammalian neuronal system [3–5], and recently, their expression in the brain of a patient suffering from Alzheimer's disease has been reported [6]. In view of these facts, it is speculated that c-series gangliosides play important roles in the neuronal functions. Therefore, the pure ganglioside as a single molecule has been required for the clarification of their multifunctionality in the complicated nuronal system at the molecular level. The chemical synthesis of gangliosides would fulfill this requirement because of the structural diversity among constituents such as sialic acid residues and ceramide parts in gangliosides in nature.

Recently, we have reported the synthesis of various polysialo-gangliosides, such as GQ1b [7], GQ1bα [8] and others [9,10], which were accomplished by regio- and stereoselective condensation of the phenylthioglycoside of the sialyl-$\alpha(2 \rightarrow 8)$sialic acid derivative carrying a 1′,9-lactone group with oligosaccharide derivatives at appropriate stages. And also, we have examined the coupling of the phenylthioglycoside of the 1″,9′:1′,9-lactonated trimeric sialic acid derivative with the suitably protected galactose and lactose derivatives in order to establish the systematic synthesis of c-series gangliosides [11]. We describe herein the α-glycosylation of the suitably protected galactose derivatives with the trimeric sialic acid donor and the synthesis of ganglioside GT4, $\alpha(2 \rightarrow 6)$GT4 and GT3.

2. Results and discussion

For the synthesis of ganglioside GT4 and $\alpha(2 \rightarrow 6)$GT4, having a galactose residue at the reducing end, the effectiveness of the following galactose derivatives as the glycosyl acceptor, 2-(trimethylsilyl)ethyl 2,6-di-*O*-benzyl-, 3-*O*-benzyl-, 2-*O*-benzoyl-3-*O*-benzyl-, and 2,3-di-*O*-benzyl-β-D-galactopyranoside (**5** [12], **6** [13], **9** and **11**), were examined. Methyl {phenyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-2-thio-D-*glycero*-D-*galacto*-2-nonulopyranosid}onate (**3** [11]) was selected as the glycosyl donor.



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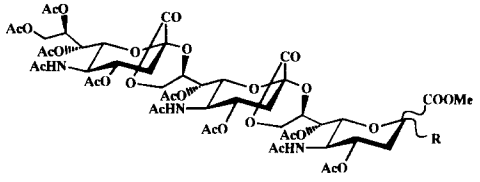
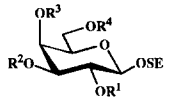


2 R = OAc
3 R = SPh



	R^1	R^2	R^3	R^4
4	H	H	H	Bz
5	Bn	H	H	Bn
6	H	Bn	H	H
7	H	Bn	isopropylidene	
8	Bz	Bn	isopropylidene	
9	Bz	Bn	H	H
10	Bn	Bn	isopropylidene	
11	Bn	Bn	H	H

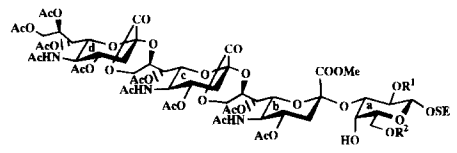
Bn = benzyl
Bz = benzoyl
SE = 2-(trimethylsilyl)ethyl

4,6-*O*-Isopropylidenation of 2-(trimethylsilyl)ethyl 3-*O*-benzyl-β-D-galactopyranoside (**6**) with 2,2-dimethoxypropane quantitatively gave the galactose derivative **7**, which was transformed into **8** by benzoylation with benzoic anhydride in pyridine, and into the 2,3-di-*O*-benzyl derivative **10** by benzylation. Treatment of compounds **8** and **10** with 80% AcOH afforded the diol galactose derivatives **9** and **11**, respectively, which were used as the glycosyl acceptors for the glycosylation to C-6 hydroxyl groups.

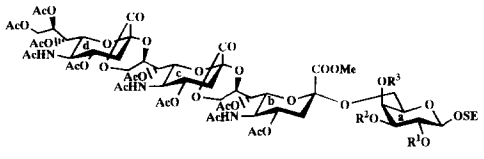
In our previous report [11], methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1′,9-lactone]-2,4,7-tri-*O*-acetyl-3,5-dideoxy-D-

glycero-D-*galacto*-2-nonulopyranosonate (**2**), a precursor of trimeric sialic acid donor **3**, was prepared from 5-acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid-(2 → 8)-5-acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid-(2 → 8)-5-acetamido-3,5-dideoxy-D-*glycero*-D-*galacto*-2-nonulopyranosonic acid (**1**), which was readily obtained by a limited acidic hydrolysis of colominic acid, via methyl esterification and 1′,9-lactonation in MeOH in the presence of Amberlite IR-120 (H^+) and subsequent *O*-acetylation in 50% overall yield. In the first treatment of **1** in MeOH under acidic conditions, methyl esterification and lactonization occurred simultaneously, and so, di- or tri-methyl-esterified derivatives of trimeric sialic acid were eventually formed as byproducts in almost equal amounts relative to the target compound **2**. In order to circumvent this result, we first prepared 1″,9′:1′,9-lactonated derivative of trimeric sialic acid in the presence of Drierite under acidic conditions by heating at 40 °C in *N*,*N*-dimethylformamide, and then, methyl esterification of the product with methyl *p*-toluenesulfonate and triethylamine, and subsequent *O*-acetylation gave **2** in 70% overall yield. Compound **2** was converted into the corresponding phenylthioglycoside derivative **3** according to the established procedure.

Glycosylation [9,14] of **5** with **3**, employing *N*-iodosuccinimide (NIS)–trifluoromethanesulfonic acid (TfOH) as a glycosyl promoter in CH_3CN at −30 °C in the presence of 3 Å molecular sieves gave the α-trisialylated oligosaccharide **13** in 47% yield (Table 1, entry 2). The ^1H NMR spectrum of the compound showed significant signals at δ 2.41 (dd, 1 H, J_{gem} 12.8 Hz, $J_{3eq,4}$ 5.5 Hz, H-3beq), and 4.69 (m, 1 H, H-4b), indicating the newly formed linkage to be α [11]. In essentially the same way, glycosylation of **6**, **9** and **11** with **3** gave the corresponding α-glycosides bearing trimeric sialic acid at the C-6 hydroxyl group **14**, **15** and **17** (23–46%) together with the β-glycoside, respectively (Table 1, entries 3–5). The stereochemistry of the newly formed glycosidic linkage was established by ^1H NMR spectroscopy. The spectra showed significant signals at δ 4.72–4.88 (m, 1 H, H-4b), 5.31–5.38 (m, H-4d), and 5.46–5.55 (m, H-4c), indicating an α-glycoside [11]. As a result, the dibenzylated galactose acceptors **5** and **11** were found to give good yields of the desired products. Therefore, we adopted compound **5** as the building block for ganglioside GT4 synthesis and **11** for ganglioside α(2 → 6)GT4 synthesis, respectively.



12 R^1= H, R^2= Bz
13 R^1=R^2= Bn



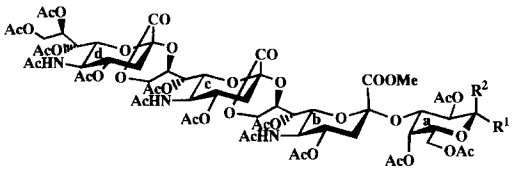
14 R^1=R^3= H, R^2= Bn
15 R^1= Bz, R^2= Bn, R^3= H
16 R^1=R^3= Bz R^2= Bn
17 R^1=R^2= Bn R^3= H

Hydrogenolytic removal of the benzyl groups in **13**, **17** with 10% Pd–C in AcOH and EtOH at 45 °C and subsequent *O*-acetylation gave the peracylated oligosaccharides **18** (92%) and **24** (94%), respectively.

Table 1
Regioselective glycosylation of the galactose acceptors with the trimeric sialic acid donor in CH_3CN by use of NIS–TfOH

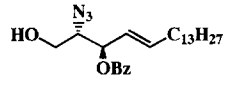
Entry	Acceptor	Temperature	Product (α)	Yield (α/β) (%)
1 [a]	**4**	−30 °C	**12**	30/0
2	**5**	−30 °C	**13**	47/0
3	**6**	−25 °C	**14**	23/2
4	**9**	−35 °C	**15**	30/19[b]
5	**11**	−35 °C	**17**	46/15

[a] Ref. [11].
[b] Calculated from α/β ratio of **16**.



18 R^1 = OSE, R^2 = H
19 R^1 = R^2 = H, OH
20 R^1 = H, R^2 = OC(=NH)CCl_3

Treatment [15] of compounds **18**, **24** or **29** [11] with CF_3CO_2H in CH_2Cl_2 at room temperature afforded the 1-hydroxy compounds **19**, **25** and **30** in good yields, which were treated with trichloroacetonitrile in the presence of 1,8-diazabicyclo[5.4.0]undec-7-ene [16,17] to give the corresponding α-trichloroacetimidates **20**, **26** and **31**, exclusively. Significant signals in ^{1}H NMR spectra of **20**, **26** and **31** were a one-proton doublet at δ 6.47–6.48 ($J_{1,2}$ 3.5 Hz, H-1a) and a one-proton singlet at δ 8.67–8.68 (C=NH), which indicated the imidates to be α.



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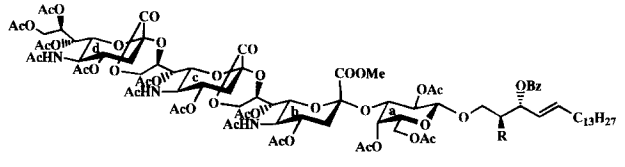
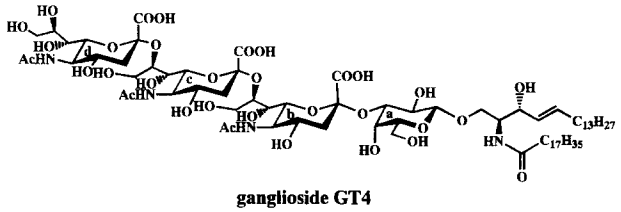
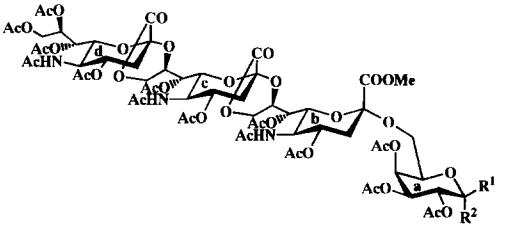


22 R = N_3
23 R = $NHCOC_{17}H_{35}$

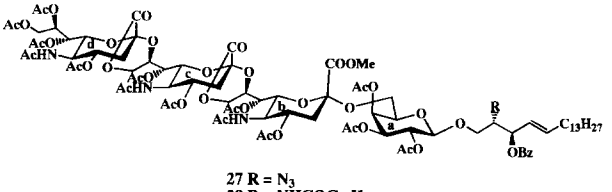
Glycosylation of (2*S*,3*R*,4*E*)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol [18] (**21**) with **20**, **26** and **31** was performed at 0 °C in the presence of trimethylsilyl trifluoromethanesulfonate (TMSOTf) and 4 Å molecular sieves (AW-300) to give the desired β-glycoside **22** (72%), **27** (56%) and **32** (66%), respectively. Selective reduction of the azido group in **22**, **27** and **32** with hydrogen sulfide in 83% aqueous pyridine at 0 °C gave the amines, which were successively applied to the coupling with octadecanoic acid in the presence of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (WSC) to afford the protected ganglioside derivatives **23** (73%), **28** (77%) and **33** (77%), respectively. Finally, *O*-deacylation of the ganglioside derivatives thus obtained with sodium methoxide in methanol and subsequent saponification of methyl esters and lactones yielded gangliosides GT4, α(2 → 6)GT4 and GT3 in almost quantitative yields.



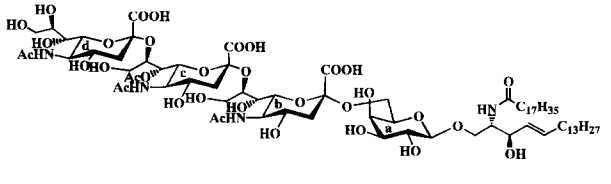
ganglioside GT4



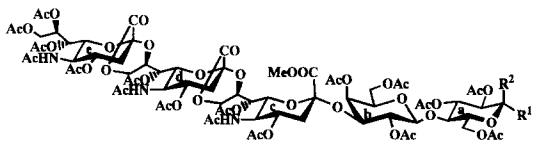
24 R^1 = OSE, R^2 = H
25 R^1 = R^2 = H, OH
26 R^1 = H, R^2 = OC(=NH)CCl_3



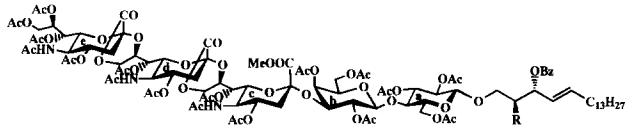
27 R = N_3
28 R = $NHCOC_{17}H_{35}$



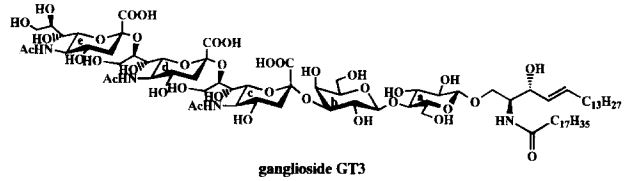
ganglioside α(2→6)GT4



29 R^1 = OSE, R^2 = H
30 R^1 = R^2 = H, OH
31 R^1 = H, R^2 = OC(=NH)CCl_3



32 R = N_3
33 R = $NHCOC_{17}H_{35}$



ganglioside GT3

In conclusion, trimeric sialic acid containing gangliosides were efficiently synthesized by regio- and stereoselective glycosylation of the suitably protected galactose and lactose derivatives with the phenyl 2-thioglycosides of 1″,9′:1′,9-lactonated trimeric sialic acid derivative as glycosyl donor in CH_3CN in the presence of NIS–TfOH, indicating a possibility for the systematic synthesis of c-series gangliosides.

3. Experimental

General methods.—Optical rotations were determined with a Union PM-201 Polarimeter at 25 °C and IR spectra were recorded with a Jasco IRA-100 spectrophotometer. ^{1}H NMR spectra were recorded at 200, 270 and 500 MHz with JEOL JNM-GX, Varian GEMINI 2000 and UNITY INOVA 500 spectrometer, respectively. Preparative chromatography was performed on silica gel (Fuji Silysia Co., 300) or Sephadex (Pharmacia, LH-20) with the solvent system specified. Concentrations were conducted in vacuo.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-2,4,7-tri-*O*-acetyl-3,5-dideoxy-*D*-glycero-*D*-galacto-2-nonulopyranosonate* (**2**).—To a suspension of 5-acetamido-3,5-dideoxy-*D*-*glycero*-α-*D*-*galacto*-2-nonulopyranosylonic acid-(2 → 8)-5-acetamido-3,5-dideoxy-*D*-*glycero*-α-*D*-*galacto*-2-nonulopyranosylonic acid-(2 → 8)-5-acetamido-3,5-dideoxy-*D*-*glycero*-*D*-*galacto*-2-nonulopyranosonic acid (**1**; 1.0 g, 1.0 mmol) in *N,N*-dimethylformamide (DMF) (10 mL) were added Amberlite IR-120 (H^+) resin (3 g) and Drierite (1 g), and the mixture was stirred for 2 days at 40 °C. The resin was filtered off and washed with MeOH. The filtrate and washings were combined and concentrated. To a solution of the residue in DMF were added methyl *p*-toluenesulfonate (650 mg, 3.5 mmol) and triethylamine (0.6 mL, 4.4 mmol), and the mixture was stirred for 38 h at room temperature and concentrated. To a solution of the residue in Ac_2O (3.0 mL) was added dropwise pyridine (3.0 mL) at 0 °C, and the mixture was stirred for 24 h at 40 °C. Methanol (3.0 mL) was added to the cooled mixture that was then concentrated. A solution of the residue in CH_2Cl_2 (50 mL) was washed with 2 M HCl and M Na_2CO_3, dried (Na_2SO_4) and concentrated. Column chromatography (30:1 CH_2Cl_2–MeOH) of the residue on silica gel gave **2** (940 mg, 70%) as an amorphous mass. Signals in ^{1}H NMR spectrum at 270 MHz ($CDCl_3$) and IR spectrum were identical with those of an authentic sample **2**.

*2-(Trimethylsilyl)ethyl 3-*O*-benzyl-4,6-*O*-isopropylidene-β-*D*-galactopyranoside* (**7**).—To a solution of **6** (2.0 g, 5.4 mmol) in CH_3CN (10 mL) were added 2,2-dimethoxypropane (1.3 mL, 10.8 mmol) and *p*-toluenesulfonic acid monohydrate (200 mg), and the mixture was stirred for 4 h at room temperature. The mixture was neutralized with triethylamine and concentrated. Column chromatography (50:1 CH_2Cl_2–MeOH) of the residue on silica gel gave **7** (2.2 g, 99%) as an amorphous mass: $[\alpha]_D$ −1.7° (*c* 0.8, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 200 MHz: δ 0.95 (m, 2 H, $Me_3SiCH_2CH_2$), 1.53 and 1.64 (2 s, 6 H, Me_2C), 3.60 (m, 2 H, $Me_3SiCH_2CH_2$), 3.75 (dd, 1 H, $J_{2,3}$ 10.1, $J_{3,4}$ 3.7 Hz, H-3), 4.12 (m, 2 H, H-6 and H-6′), 4.32 (dd, 1 H, $J_{4,5}$ 0.7 Hz, H-4), 4.62 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.78 and 4.86 (2 d, 2 H, $PhCH_2$), 5.66 (dd, 1 H, H-2), and 7.52–8.25 (m, 5 H, Ph). Anal. Calcd for $C_{21}H_{34}O_6Si$ (410.6): C, 61.43; H, 8.35. Found: C, 61.27; H, 8.17.

*2-(Trimethylsilyl)ethyl 2-*O*-benzoyl-3-*O*-benzyl-4,6-*O*-isopropylidene-β-*D*-galactopyranoside* (**8**).—To a solution of **7** (1.1 g, 2.6 mmol) in pyridine (5 mL)

was added benzoic anhydride (1.2 g, 5.2 mmol), and the mixture was stirred for 2 h at room temperature. Usual workup gave a crude product. Purification of the product by silica gel column chromatography (100:1 CH_2Cl_2–MeOH) gave **8** (1.3 g, quantitative) as an amorphous mass: $[\alpha]_D$ +0.22° (*c* 3.7, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 200 MHz: δ 0.95 (m, 2 H, $Me_3SiCH_2CH_2$), 1.53 and 1.64 (2 s, 6 H, Me_2C), 3.60 (m, 2 H, $Me_3SiCH_2CH_2$), 3.75 (dd, 1 H, $J_{2,3}$ 10.1, $J_{3,4}$ 3.7 Hz, H-3), 4.12 (m, 2 H, H-6 and H-6′), 4.32 (dd, 1 H, $J_{4,5}$ 0.7 Hz, H-4), 4.62 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.78 and 4.86 (2 d, 2 H, $PhCH_2$), 5.66 (dd, 1 H, H-2), and 7.52–8.25 (m, 10 H, Ph). Anal. Calcd for $C_{28}H_{38}O_7Si$ (514.7): C, 65.34; H, 7.44. Found: C, 65.31; H, 7.38.

2-(Trimethylsilyl)ethyl 2-O-benzoyl-3-O-benzyl-β-D-galactopyranoside (**9**).—A solution of **8** (1.1 g, 2.6 mmol) in aq 80% AcOH (5 mL) was stirred for 18 h at 40 °C and concentrated. Column chromatography (2:1 AcOEt–hexane) of the residue on silica gel gave **9** (1.2 g, quantitative) as an amorphous mass: $[\alpha]_D$ +16.6° (*c* 1.1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 200 MHz: δ 0.95 (m, 2 H, $Me_3SiCH_2CH_2$), 3.76 (dd, 1 H, $J_{2,3}$ 10.1, $J_{3,4}$ 3.7 Hz, H-3), 4.12 (m, 2 H, H-6 and H-6′), 4.29 (dd, 1 H, $J_{4,5}$ 0.7 Hz, H-4), 4.63 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.70 and 4.86 (2 d, 2 H, $PhCH_2$), 5.57 (dd, 1 H, H-2), and 7.52–8.25 (m, 10 H, 2 Ph). Anal. Calcd for $C_{25}H_{34}O_7Si$ (474.6): C, 63.27; H, 7.22. Found: C, 63.13; H, 6.97.

2-(Trimethylsilyl)ethyl 2,3-di-O-benzyl-4,6-O-isopropylidene-β-D-galactopyranoside (**10**).—To a solution of **7** (1.3 g, 2.6 mmol) in DMF (5 mL) was added sodium hydride (1.2 g, 5.2 mmol), and the mixture was stirred for 30 min at 0 °C. To the stirring mixture was added benzyl bromide (0.46 mL, 3.9 mmol), and the mixture was continuously stirred for 2 h at room temperature. Usual workup gave a crude product. Purification of the product by silica gel column chromatography (100:1 CH_2Cl_2–MeOH) gave **10** (1.3 g, quantitative) as an amorphous mass: $[\alpha]_D$ +4.2° (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 200 MHz: δ 0.95 (m, 2 H, $Me_3SiCH_2CH_2$), 1.53 and 1.64 (2 s, 6 H, Me_2C), 3.60 (m, 2 H, $Me_3SiCH_2CH_2$), 3.75 (dd, 1 H, $J_{2,3}$ 10.1, $J_{3,4}$ 3.7 Hz, H-3), 4.12 (m, 2 H, H-6 and H-6′), 4.32 (dd, 1 H, $J_{4,5}$ 0.7 Hz, H-4), 4.54 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), and 7.08–7.18 (m, 10 H, 2 Ph). Anal. Calcd for $C_{28}H_{46}O_6Si$ (474.6): C, 67.17; H, 8.05. Found: C, 66.91; H, 7.80.

2-(Trimethylsilyl)ethyl 2,3-di-O-benzyl-β-D-galactopyranoside (**11**).—A solution of **10** (1.3 g, 2.6 mmol) in aq 80% AcOH (5 mL) was stirred for 16 h at 40 °C and concentrated. Column chromatography (2:1 AcOEt–hexane) of the residue on silica gel gave **11** (1.2 g, quantitative) as an amorphous mass: $[\alpha]_D$ −0.8° (*c* 1.1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 200 MHz: δ 0.95 (m, 2 H, $Me_3SiCH_2CH_2$), 3.76 (dd, 1 H, $J_{2,3}$ 10.1, $J_{3,4}$ 3.7 Hz, H-3), 4.12 (m, 2 H, H-6 and H-6′), 4.29 (dd, 1 H, $J_{4,5}$ 0.7 Hz, H-4), 4.63 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.70 and 4.86 (2 d, 2 H, $PhCH_2$), 5.57 (dd, 1 H, H-2), and 7.09–7.18 (m, 10 H, 2 Ph). Anal. Calcd for $C_{25}H_{36}O_6Si$ (460.6): C, 63.27; H, 7.22. Found: C, 63.13; H, 6.97.

2-(Trimethylsilyl)ethyl {methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosyl-ono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate}-(2 → 3)-2,6-di-O-benzyl-β-D-galactopyranoside (**13**).—To a solution of **3** (200 mg, 0.15 mmol) and 2-(trimethylsilyl)ethyl 2,6-di-*O*-benzyl-β-D-galactopyranoside (**5**; 177 mg, 0.39 mmol) in CH_3CN (5 mL) were added 3 Å molecular sieves (1 g), and the suspension was stirred for 12 h at room temperature. To the mixture were then added, with stirring, *N*-iodosuccinimide (NIS; 139 mg, 0.63 mmol) and trifluoromethanesulfonic acid (TfOH; 5.4 μL, 0.063 mmol), and the stirring was continued for 6 h at −30 °C. The solids were filtered off and washed with CH_2Cl_2. The combined filtrate and washings was washed with M Na_2CO_3 and M $Na_2S_2O_3$, dried (Na_2SO_4) and concentrated. Column chromatography (30:1 CH_2Cl_2–MeOH) of the residue on silica gel gave **13** (120 mg, 47%) as an amorphous mass: $[\alpha]_D$ −15° (*c* 0.9, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 500 MHz: δ 0.95 (m, 2 H, $Me_3SiCH_2CH_2$), 1.88–2.18 (11 s, 42 H, 3 AcN and 8 AcO), 2.30 (dd, 1 H, J_{gem} 12.9, $J_{3eq,4}$ 5.5 Hz, H-3deq), 2.41 (dd, 1 H, J_{gem} 12.8, $J_{3eq,4}$ 5.5 Hz, H-3beq), 2.57 (dd, 1 H, J_{gem} 12.8, $J_{3eq,4}$ 5.5 Hz, H-3ceq), 3.49 (s, 3 H, MeO), 4.69 (m, 1 H, H-4b), 5.13 (m, 1 H, H-8d), 5.40 (m, 1 H, H-4d), 5.58 (m, 1 H, H-4c), and 7.04–7.18 (m, 10 H, 2 Ph). Anal. Calcd for $C_{75}H_{101}N_3O_{36}Si$ (1648.7): C, 54.64; H, 6.18; N, 2.55. Found: C, 54.59; H, 5.98; N, 2.25.

2-(Trimethylsilyl)ethyl {methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-D-glyc-ero-α-D-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate}-(2 → 6)-3-O-benzyl-β-D-galactopyranoside (**14**) *and corresponding β*

isomer.—Condensation of **3** (200 mg, 0.15 mmol) with **6** (143 mg, 0.39 mmol) in the presence of NIS (104 mg, 0.46 mmol) and TfOH (4.1 μL, 46 μmol) in CH_3CN (5 mL) at −35 °C, as described for **13**, gave **14** (57 mg, 23%) and some of the β isomer (5 mg, 2%), followed by chromatography of the amorphous residue with 15:1 toluene–MeOH. Compound **14** had $[\alpha]_D$ −29.8° (*c* 0.8, $CHCl_3$); ^1H NMR ($CDCl_3$) at 500 MHz: δ 0.86 (m, 2 H, $Me_3SiCH_2CH_2$), 1.82–2.15 (11 s, 33 H, 3 AcN and 8 AcO), 2.38 (m, 3 H, H-3beq, H-3ceq and H-3deq), 3.41 (m, 1 H, $Me_3SiCH_2CH_2$), 3.81 (s, 3 H, MeO), 4.05 (dd, 1 H, $J_{8,9}$ 5.3, J_{gem} 12.6 Hz, H-9d), 4.37 (dd, 1 H, $J_{6,7}$ 2.4, $J_{7,8}$ 9.7 Hz, H-7b), 4.72 (m, 1 H, H-4b), 4.73–4.81 (2 d, 2 H, $PhCH_2$), 5.03 (dd, 1 H, $J_{6,7}$ 2.4, $J_{7,8}$ 9.5 Hz, H-7c), 5.13 (m, 1 H, H-8d), 5.03 (dd, 1 H, $J_{6,7}$ 1.8, $J_{7,8}$ 7.8 Hz, H-7d), 5.38 (m, 1 H, H-4d), 5.46 (m, 1 H, H-4c), and 7.28–7.47 (m, 5 H, Ph). Anal. Calcd for $C_{68}H_{95}N_3O_{36}Si$ (1558.6): C, 52.40; H, 6.14; N, 2.70. Found: C, 52.22; H, 6.06; N, 2.64.

*2-(Trimethylsilyl)ethyl {methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosyl-ono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylonate}-(2 → 6)-2,4-di-O-benzoyl-3-O-benzyl-β-*D*-galactopyranoside* (**16**) *and corresponding β isomer*.—To a solution of **3** (200 mg, 0.15 mmol) and **9** (182 mg, 0.39 mmol) in CH_3CN (5 mL) was added 3 Å molecular sieves (1 g), and the suspension was stirred for 12 h at room temperature. To the mixture were then added, with stirring, NIS (104 mg, 0.63 mmol) and TfOH (4.1 μL, 0.063 mmol), and the stirring was continued for 2 h at −20 °C. Similar workup, as described for **13**, gave an anomeric mixture of product **15**. Then, to a solution of the product in pyridine (2 mL) were added benzoic anhydride (176 mg, 0.78 mmol) and catalytic amount of 4,4-dimethylaminopyridine, and the mixture was stirred for 3 h at room temperature. Usual workup gave a crude product. Purification of the product by silica gel column chromatography (30:1 CH_2Cl_2–MeOH) gave **16** (75 mg, 30%) and β isomer (17 mg, 19%) as an amorphous mass: Compound **16** had $[\alpha]_D$ −23.7° (*c* 0.9, $CHCl_3$); ^1H NMR ($CDCl_3$) at 500 MHz: δ 0.96 (m, 2 H, $Me_3SiCH_2CH_2$), 1.72–2.28 (11s, 33 H, 3 AcN and 8 AcO), 2.59 (dd, 1 H, J_{gem} 12.8, $J_{3eq,4}$ 5.13 Hz, H-3beq), 2.71 (dd, 1 H, J_{gem} 12.8, $J_{3eq,4}$ 5.13 Hz, H-3ceq), 3.49 (s, 3 H, MeO), 3.59 (m, 1 H, $Me_3SiCH_2CH_2$), 3.74 (dd, 1 H, $J_{5,6}$ 10.1, $J_{6,7}$ 2.9 Hz, H-6b), 4.69–4.75 (2d, 2 H, J_{gem} 11.7 Hz, $PhCH_2$), 4.88 (m, 1 H, H-4b), 5.13 (m, 1 H, H-8d), 5.31 (m, 1 H, H-4d), 5.55 (m, 1 H, H-4c), and 7.04–8.13 (m, 15 H, 3 Ph). Anal. Calcd for $C_{82}H_{103}N_3O_{38}Si$ (1766.8): C, 55.75; H, 5.88; N, 2.38. Found: C, 55.61; H, 5.79; N, 2.37.

*2-(Trimethylsilyl)ethyl {methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosyl-ono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylonate}-(2 → 6)-2,3-di-O-benzyl-β-*D*-galactopyranoside* (**17**) *and corresponding β isomer*.—Condensation of **3** (200 mg, 0.15 mmol) with **11** (182 mg, 0.39 mmol) in the presence of NIS (104 mg, 0.46 mmol) and TfOH (4.1 μL, 46 μmol) in CH_3CN (5 mL) at −20 °C, as described for **13**, gave **17** (115 mg, 46%), and β isomer (51 mg, 20%) followed by chromatography of the amorphous residue with 15:1 toluene–MeOH. Compound **17** had $[\alpha]_D$ −23.7° (*c* 0.9, $CHCl_3$); ^1H NMR ($CDCl_3$) at 270 MHz: δ 1.00 (m, 2 H, $Me_3SiCH_2CH_2$), 1.88–2.18 (11 s, 33 H, 3 AcN and 8 AcO), 2.33–2.50 (m, 3 H, H-3beq, H-3ceq and H-3deq), 3.86 (s, 3 H, MeO), 4.76–4.89 (2 d, 2 H, $PhCH_2$), 5.18 (m, 1 H, H-8d), and 7.23–7.33(m, 10 H, 2 Ph). Anal. Calcd for $C_{75}H_{101}N_3O_{36}Si$ (1648.7): C, 54.64; H, 6.18; N, 2.55. Found: C, 54.50; H, 5.90; N, 2.28.

*2-(Trimethylsilyl)ethyl {methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosyl-ono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylonate}-(2 → 3)-2,4,6-tri-O-acetyl-β-*D*-galactopyranoside* (**18**).—A solution of **13** (430 mg, 0.26 mmol) in EtOH (20 mL) and AcOH (20 mL) was hydrogenated in the presence of 10% Pd–C (430 mg) for 24 h at 45 °C. The catalyst was filtered off and washed with EtOH. The combined filtrate and washings was concentrated. The residue was acetylated with Ac_2O (4 mL) and pyridine (4 mL) for 24 h at 40 °C. Usual workup gave a crude product. Column chromatography (20:1 CH_2Cl_2–MeOH) of the product on silica gel gave **18** (367 mg, 92%) as an amorphous mass: $[\alpha]_D$ +19.4° (*c* 0.7, $CHCl_3$); ^1H NMR ($CDCl_3$) at 500 MHz: δ 1.00 (m, 2 H, $Me_3SiCH_2CH_2$), 1.86–2.20 (14 s, 42 H, 3 AcN and 11 AcO), 3.85 (s, 3 H, MeO), 5.15 (d, 1 H, $J_{3,4}$ 3.5 Hz, H-4a), 5.57 (m, 1 H, H-4d), 5.72 (m, 1 H, H-4c), and 6.11 (d, 1 H, $J_{5,NH}$ 10.3 Hz, NH). Anal. Calcd

for $C_{67}H_{95}N_3O_{39}Si$ (1594.6): C, 50.47; H, 6.01; N, 2.64. Found: C, 50.25; H, 5.90; N, 2.60.

*Methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylonate-(2 → 3)-2,4,6-tri-O-acetyl-*D*-galactopyranose* (**19**).—To a solution of **18** (254 mg, 0.16 mmol) in CH_2Cl_2 (1.8 mL) was added CF_3CO_2H (3.5 mL) at 0 °C, and the solution was stirred for 2 h at room temperature. Ethyl acetate was added to the mixture, and it was concentrated to a syrup that was chromatographed on silica gel with 30:1 CH_2Cl_2–MeOH to give **19** (226 mg, 95%) as an amorphous mass: ν 3600–3100 (NH, OH), 1730 and 1220 (ester), 1650 and 1540 cm^{-1} (amide). Anal. Calcd for $C_{62}H_{83}N_3O_{39}$ (1494.3): C, 49.83; H, 5.60; N, 2.81. Found: C, 49.63; H, 5.53; N, 2.62.

*Methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylonate-(2 → 3)-2,4,6-tri-O-acetyl-α-*D*-galactopyranosyl trichloroacetimidate* (**20**).—To a solution of **19** (226 mg, 0.15 mmol) in CH_2Cl_2 (2.0 mL) and trichloroacetonitrile (0.63 mL) was added 1,8-diazabicyclo[5.4.0]undec-7-ene (27 μL, 0.18 mmol), and the mixture was stirred for 1.5 h at 0 °C. Column chromatography (30:1 CH_2Cl_2–MeOH) of the mixture on silica gel gave **20** (160 mg, 65%) as an amorphous mass: ^{1}H NMR ($CDCl_3$) at 270 MHz: δ 1.83–2.19 (14 s, 42 H, 3 AcN and 11 AcO), 3.89 (s, 3 H, MeO), 6.20 (d, 1 H, $J_{5,NH}$ 9.4 Hz, NH), 6.20 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1a), and 8.68 (s, 1 H, C=NH). Anal. Calcd for $C_{64}H_{83}Cl_3N_4O_{39}$ (1638.7): C, 46.91; H, 5.11; N, 3.42. Found: C, 46.76; H, 5.09; N, 3.22.

*Methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylonate-(2 → 3)-2,4,6-tri-O-acetyl-β-*D*-galactopyranosyl-(1 → 1)-(2S,3R,4E)-2-azido-3-O-benzoyl-4-octadecene-1,3-diol* (**22**).—To a solution of **20** (160 mg, 98 μmol) and (2*S*,3*R*,4*E*)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol (**21**; 84 mg, 195 mmol) in CH_2Cl_2 (0.5 mL) were added 4 Å molecular sieves (AW-300; 750 mg), and the mixture was stirred for 3 h at room temperature, then cooled to 0 °C. To the stirring mixture was added TMSOTf (38 μL, 195 μmol), and the stirring was continued for 16 h at 0 °C. The solids were filtered off and washed with CH_2Cl_2. The combined filtrate and washings was washed with M Na_2CO_3 and H_2O, dried (Na_2SO_4) and concentrated. Column chromatography (30:1 CH_2Cl_2–MeOH) of the residue on silica gel gave **22** (134 mg, 72%) as an amorphous mass: $[\alpha]_D$ −26.5° (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 270 MHz: δ 0.87 (t, 3 H, $J_{Me,CH2}$ 7.0, *Me*CH_2), 1.24 (s, 22 H, 11 CH_2), 1.85–2.19 (14 s, 42 H, 3 AcN and 11 AcO), 3.86 (s, 3 H, MeO), 5.93 (m, 1 H, H-5 of sphingosine), and 7.43–8.09 (m, 5 H, Ph). Anal. Calcd for $C_{87}H_{121}N_6O_{41}$ (1905.9): C, 54.80; H, 6.40; N, 4.41. Found: C, 54.48; H, 6.29; N, 4.41.

*Methyl 5-acetamido-8-O-[5-acetamido-8-O-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-O-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylonate-(2 → 3)-2,4,6-tri-O-acetyl-β-*D*-galactopyranosyl-(1 → 1)-(2S,3R,4E)-3-O-benzoyl-2-otcadecanamido-4-octadecene-1,3-diol* (**23**).—Hydrogen sulfide was bubbled through a solution of **22** (134 mg, 70 μmol) in aq 83% pyridine (12 mL) for 3 days while the solution was stirred at 0 °C. The mixture was concentrated to a syrup, which was used for next reaction without further purification. A solution of the residue in CH_2Cl_2 (2 mL) was treated with octadecanoic acid (60 mg, 210 μmol) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (WSC; 40 mg, 210 μmol), and the mixture was stirred for 18 h at room temperature. The mixture was washed with water, dried (Na_2SO_4) and concentrated. Column chromatography (30:1 CH_2Cl_2–MeOH) of the residue on silica gel gave **23** (110 mg, 73%) as an amorphous mass: $[\alpha]_D$ −11.6° (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 270 MHz: δ 0.88 (t, 6 H, $J_{Me,CH2}$ 7.0 Hz, 2 *Me*CH_2), 1.26 (s, 52 H, 26 CH_2), 1.84–2.19 (14 s, 42 H, 3 AcN and 11 AcO), 3.84 (s, 3 H, MeO), 5.85 (m, 1 H, H-5 of ceramide), 5.99 (d, 1 H, $J_{5,NH}$ 10.1 Hz, NH), and 7.43–8.09 (m, 5 H, Ph). Anal. Calcd for $C_{105}H_{156}N_4O_{42}$ (2146.4): C, 58.76; H, 7.33; N, 2.61. Found: C, 58.56; H, 7.13; N, 2.57.

Ganglioside GT4.—To a solution of **23** (110 mg, 0.068 mmol) in MeOH (2 mL) was added NaOMe, and the mixture was stirred for 12 h at room temperature. Potassium hydroxide (0.2 M) was added, and

the mixture was stirred for an additional 12 h, then neutralized with Amberlite IR-120 (H^+) resin. The resin was filtered off and 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 gave ganglioside GT4 (60 mg, 98%) as an amorphous mass: $[\alpha]_D$ −11.6° (*c* 1.0, 1:1 $CHCl_3$–MeOH); ^1H NMR ($CDCl_3$–CD_3OD) at 500 MHz: δ 0.88 (t, 6 H, $J_{Me,CH2}$ 7.0 Hz, 2 *Me*CH_2), 1.19 (s, 52 H, 26 CH_2), 1.84–1.94 (3 s, 9 H, 3 AcN), 5.78 (m, 1 H, H-5 of ceramide). Anal. Calcd for $C_{75}H_{132}N_4O_{32}$ (1601.88): C, 56.24; H, 8.31; N, 3.50. Found: C, 56.23; H, 8.22; N, 3.21.

*2-(Trimethylsilyl)ethyl {methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylonate}-(2 → 6)-2,3,4-tri-*O*-acetyl-β-*D*-galactopyranoside* (**24**).—Reductive removal of the benzyl groups in the presence of 10% Pd–C (60 mg) in AcOH and EtOH at 45 °C for 16 h, subsequent *O*-acetylation of **17** (115 mg, 70 μmol), and usual workup gave **24** (104 mg, 94%) as an amorphous mass: $[\alpha]_D$ −26.6° (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$) at 270 MHz: δ 0.89 (m, 2 H, $Me_3SiCH_2CH_2$), 1.86–2.18 (14 s, 42 H, 3 AcN and 11 AcO), 2.34–2.60 (m, 3 H, H-3beq, H-3ceq and H-3deq), 3.85 (s, 3 H, MeO), and 6.01 (d, 1 H, $J_{5,NH}$ 10.3 Hz, NH). Anal. Calcd for $C_{67}H_{95}N_3O_{39}Si$ (1594.6): C, 50.47; H, 6.01; N, 2.64. Found: C, 50.19; H, 5.72; N, 2.41.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate-(2 → 6)-2,3,4-tri-*O*-acetyl-D-galactopyranose* (**25**).—Removal of the 2-(trimethylsilyl)ethyl group on **24** (220 mg, 0.14 mmol) in CH_2Cl_2 by use of CF_3CO_2H (3.0 mL) at 0 °C for 2 h and similar workup, as described for **19**, gave **25** (194 mg, 94%) as an amorphous mass: ν 3600–3100 (OH, NH), 1730 and 1220 (ester), and 1650 and 1540 (amide). Anal. Calcd for $C_{62}H_{83}N_3O_{39}$ (1494.3): C, 49.83; H, 5.60; N, 2.81. Found: C, 49.77; H, 5.60; N, 2.81.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1′,9-lactone-(2 → 6)-2,3,4-tri-*O*-acetyl-α-D-galactopyranosyl trichloroacetimidate* (**26**).—Treatment of **25** (194 mg, 0.13 mmol) with trichloroacetonitrile (0.53 mL) and DBU (24 μL, 0.16 mmol) at 0 °C for 2 h, as described for **20**, gave **26** (177 mg, 83%) as an amorphous mass: $[\alpha]_D$ +3.6° (*c* 1.1, $CHCl_3$); ^1H NMR ($CDCl_3$) at 270 MHz: δ 1.83–2.18 (14 s, 42 H, 3 AcN and 11 AcO), 3.89 (s, 3 H, MeO), 5.18 (m, 1 H, H-8d), 6.58 (d, 1 H, $J_{1,2}$ 2.9 Hz, H-1a), and 8.67 (s, 1 H, C=NH). Anal. Calcd for $C_{64}H_{83}Cl_3N_4O_{39}$ (1638.7): C, 46.91; H, 5.11; N, 3.42. Found: C, 46.76; H, 5.00; N, 3.26.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate-(2 → 6)-2,3,4-tri-*O*-acetyl-β-D-galactopyranosyl-(1 → 1)-(2S,3R,4E)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol* (**27**).—Condensation of **26** (177 mg, 0.11 mmol) with **21** (93 mg, 0.22 mmol) by use of TMSOTf (41 μL, 0.22 mmol) in CH_2Cl_2 (1.0 mL) in the presence of 4 Å molecular sieves (AW-300; 500 mg) at 0 °C for 16 h and similar workup, as described for **22**, gave **27** (115 mg, 56%): $[\alpha]_D$ −23.8° (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$) at 270 MHz: δ 0.88 (t, 3 H, $J_{Me,CH2}$ 7.0 Hz, *Me*CH_2), 1.24 (s, 22 H, 11 CH_2), 1.86–2.18 (14 s, 42 H, 3 AcN and 11 AcO), 3.79 (s, 3 H, MeO), 5.93 (m, 1 H, H-5 of sphingosine), and 7.43–8.09 (m, 5 H, Ph). Anal. Calcd for $C_{87}H_{121}N_6O_{41}$ (1905.9): C, 54.80; H, 6.40; N, 4.41. Found: C, 54.56; H, 6.29; N, 4.16.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1′,9-lactone-(2 → 6)-2,3,4-tri-*O*-acetyl-β-D-galactopyranosyl-(1 → 1)-(2S,3R,4E)-3-O-benzoyl-2-octadecanamido-4-octadecene-1,3-diol* (**28**).—Selective reduction of the azido group in **27** (115 mg, 0.06 mmol) and subsequent coupling with octadecanoic acid (52 mg, 0.18 mmol), as described for **23**, gave **28** (101 mg, 77%) as an amorphous mass: $[\alpha]_D$ −11.6° (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$) at 270 MHz: δ 0.88 (t, 6 H, $J_{Me,CH2}$ 7.0 Hz, 2 *Me*CH_2), 1.26 (s, 52 H, 26 CH_2), 1.84–2.19 (14 s, 42 H, 3

AcN and 11 AcO), 3.84 (s, 3 H, MeO), 5.85 (m, 1 H, H-5 of ceramide), 5.99 (d, 1 H, $J_{5,NH}$ 10.1 Hz, NH), and 7.43–8.09 (m, 5 H, Ph). Anal. Calcd for $C_{105}H_{156}N_4O_{42}$ (2146.4): C, 58.76; H, 7.33; N, 2.61. Found: C, 58.56; H, 7.13; N, 2.57.

Ganglioside α(2 → 6)GT4.—*O*-Deacylation and saponification of methyl ester and lactone groups in **28** (101 mg, 0.047 mmol), as described for ganglioside GT4, gave ganglioside α(2 → 6)GT4 (52 mg, 95%) as an amorphous mass: $[\alpha]_D$ −11.6° (*c* 1.0, 1:1 $CHCl_3$–MeOH); ^{1}H NMR ($CDCl_3$–CD_3OD) at 500 MHz: δ 0.88 (t, 6 H, $J_{Me,CH2}$ 7.0 Hz, 2 *Me*CH_2), 1.20 (s, 52 H, 26 CH_2), 1.84–1.96 (3 s, 9 H, 3 AcN), 5.81 (m, 1 H, H-5 of ceramide). Anal. Calcd for $C_{75}H_{132}N_4O_{32}$ (1601.88): C, 56.24; H, 8.31; N, 3.50. Found: C, 55.95; H, 8.02; N, 3.25.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-*D*-glycero-α-*D*-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate-(2 → 3)-2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl-(1 → 4)-2,3,6-tri-*O*-acetyl-D-glucopyranose* (**30**).—Removal of 2-(trimethylsilyl)ethyl group of 2-(trimethylsilyl)ethyl {methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosonate}-(2 → 3)-2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl-(1 → 4)-2,3,6-tri-*O*-acetyl-β-D-glucopyranoside (**29**; 350 mg, 0.19 mmol) in CH_2Cl_2 with CF_3CO_2H (4.0 mL) at 0 °C for 1.5 h and similar workup, as described for **19**, gave **30** (320 mg, 97%) as an amorphous mass: ν 3600–3100 (OH, NH), 1730 and 1220 (ester), and 1650 and 1540 (amide). Anal. Calcd for $C_{62}H_{83}N_3O_{39}$ (1782.6): C, 49.80; H, 5.60; N, 2.36. Found: C, 49.71; H, 5.34; N, 2.26.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate-(2 → 3)-2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl-(1 → 4)-2,3,6-tri-*O*-acetyl-α-D-glucopyranosyl trichloroacetimidate* (**31**).—Imidate formation of **30** (330 mg, 0.19 mmol) by the treatment with trichloroacetonitrile (0.74 mL) and DBU (33 μL, 0.22 mmol) in CH_2Cl_2 (3 mL) at 0 °C for 1.5 h, as described for **20**, gave **31** (300 mg, 84%) as an amorphous mass: $[\alpha]_D$ +10.0° (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 270 MHz: δ 1.82–2.19 (17 s, 51 H, 3 AcN and 14 AcO), 3.86 (s, 3 H, MeO), 5.16 (m, 1 H, H-8e), 5.42 and 6.00 (2 d, 2 H, 2 NH), 6.49 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1a), and 8.66 (s, 1 H, C=NH). Anal. Calcd for $C_{76}H_{99}Cl_3N_4O_{47}$ (1927.0): C, 47.37; H, 5.18; N, 2.91. Found: C, 47.30; H, 4.94; N, 2.91.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate-(2 → 3)-2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl-(1 → 4)-2,3,6-tri-*O*-acetyl-β-D-glucopyranosyl-(1 → 1)-(2S,3R,4E)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol* (**32**).—Condensation of **31** (258 mg, 0.13 mmol) with **21** (115 mg, 0.27 mmol) by use of TMSOTf (51 μL, 0.26 mmol) in CH_2Cl_2 (1.0 mL) in presence of 4 Å molecular sieves (AW-300; 1.0 g) at 0 °C for 1 day and similar workup, as described for **22**, gave **32** (193 mg, 66%): $[\alpha]_D$ −12.2° (*c* 0.9, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 270 MHz: δ 0.87 (t, 3 H, $J_{Me,CH2}$ 7.0 Hz, *Me*CH_2), 1.24 (s, 22 H, 11 CH_2), 1.83–2.19 (17 s, 51 H, 3 AcN and 14 AcO), 3.86 (s, 3 H, MeO), 5.93 (m, 1 H, H-5 of sphingosine), and 7.42–8.06 (m, 5 H, Ph). Anal. Calcd for $C_{99}H_{136}N_6O_{49}$ (2194.2): C, 54.19; H, 6.25; N, 3.83. Found: C, 54.13; H, 6.17; N, 3.74.

*Methyl 5-acetamido-8-*O*-[5-acetamido-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1″,9′-lactone)-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylono-1′,9-lactone]-4,7-di-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate-(2 → 3)-2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl-(1 → 4)-2,3,4-tri-*O*-acetyl-β-D-glucopyranosyl-(1 → 1)-(2S,3R,4E)-2-azido-3-*O*-benzoyl-4-octadecanamido-1,3-diol* (**33**).—Selective reduction of the azido group in **32** (95 mg, 0.043 mmol) and subsequent coupling with octadecanoic acid (37 mg, 0.13 mmol), as described for **23**, gave **33** (81 mg, 77%) as an amorphous mass: $[\alpha]_D$ −3.3° (*c* 0.9, $CHCl_3$); ^{1}H NMR ($CDCl_3$) at 270 MHz: δ 0.88 (t, 6 H, $J_{Me,CH2}$ 7.0 Hz, 2 *Me*CH_2), 1.25 (s, 52 H, 26 CH_2), 1.83–2.19 (17 s, 51 H, 3 AcN and 14 AcO), 3.85 (s, 3 H, MeO), 5.85 (m, 1 H, H-5 of ceramide), and 7.40–8.01 (m, 5 H, Ph). Anal. Calcd for $C_{117}H_{172}N_4O_{49}$ (2418.6): C, 58.10; H, 7.17; N, 2.32. Found: C, 57.87; H, 7.01; N, 2.25.

Ganglioside GT3.—*O*-Deacylation and saponification of methyl ester and lactone group in **33** (101 mg, 0.041 mmol), as described for ganglioside GT4, gave ganglioside GT3 (45 mg, 98%) as an amorphous mass: $[\alpha]_D$ −11.6° (*c* 1.0, 1:1 $CHCl_3$–MeOH); ^1H NMR ($CDCl_3$–CD_3OD) at 400 MHz: δ 0.89 (t, 6 H, $J_{Me,CH2}$ 7.0 Hz, 2 $MeCH_2$), 1.23 (s, 52 H, 26 CH_2), 1.84–1.96 (3 s, 9 H, 3 AcN), 5.81 (m, 1 H, H-5 of ceramide). Anal. Calcd for $C_{81}H_{142}N_4O_{36}$ (1746.0): C, 55.72; H, 8.08; N, 3.21. Found: C, 55.67; H, 8.05; N, 3.01.

Acknowledgements

This work was supported in part by Grants-in-Aid (no. 05274102) for the Scientific Research on Priority Areas and Scientific Research (B) (no.07456162) from the Ministry of Education, Science and Culture of Japan. The authors thank Dr. Mitsuo Kawase of NGK Insulators, Ltd. for his kind supply of trimeric sialic acid.

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