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Total synthesis of a modified ganglioside, de-*N*-acetyl GM_2

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Abstract

Regio- and stereo-selective glycosylation of a sialyl donor **6** that carries a *N*-phthaloyl protecting group at *C*-5 with a lactosyl acceptor **7** armed with a pivaloyl group at *O*-2a was performed to give the expected glycotrioside **5**. Subsequent glycosylation of **5** with 2-azido galactosyl donor **4** gave glycotetraosyl derivatives **18** and **19**. After conversion of **18** into imidates **25** and **26**, coupling with (2*S*,3*R*,4*E*)-3-*O*-benzoyl-2-*N*-tetracosanoylsphingenine (**2**) was executed to afford completely protected ganglioside analogues **27** and **28**. Selective cleavage of the methyl ester and *N*,*O*-deprotection gave the target de-*N*-acetyl GM_2 (**1**).

1. Introduction

Two recent reports have suggested the existence of small amounts of gangliosides with free amino groups on sialic acid in tumor cells [1,2]. This was confirmed by experiments using monoclonal antibody and radioisotopic tracer techniques. One of the biological functions of the de-*N*-acetyl GM_3 was demonstrated [1] to be its significant stimulatory effect on the epidermal growth factor (EGF) receptor of A-431 cells through tyrosine phosphorylation. We have previously reported the total synthesis of de-*N*-acetyl GM_3 and its analogues [3]. In this paper, we describe the total synthesis of de-*N*-acetyl GM_2 (**1**), the availability of which will provide an important tool for the investigation of the biological phenomena associated with it.

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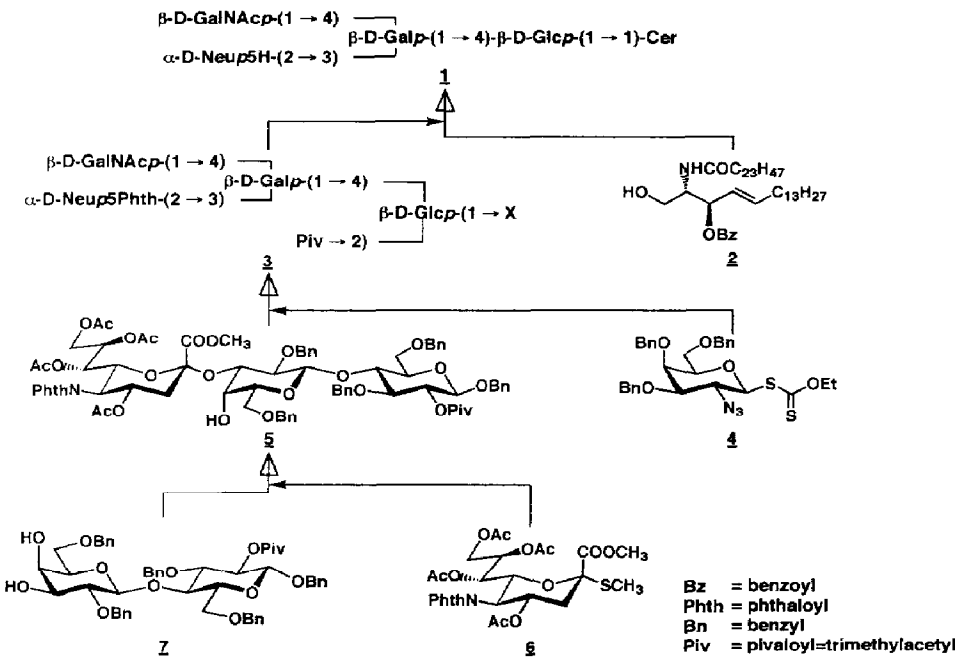


Scheme 1.

2. Results and discussion

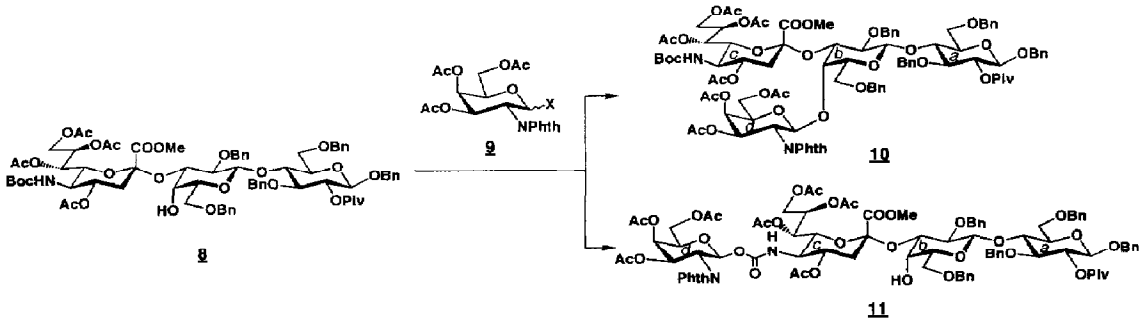
A retrosynthetic analysis of the target structure **1** suggested the construction from two parts, namely the benzoyl ceramide [4] **2** and glycotetraosyl donor **3**. This donor **3** was further divided into glycosyl donor [5] **4** and glycotrioside **5**. Finally, **5** was divided into the properly protected lactose derivative [6] **7** and neuraminyl donor **6** (Scheme 1).

In the preliminary experiments, glycosylation of **8**, which was previously reported in the synthesis of de-*N*-acetyl GM_3 [3], was carried out with *N*-phthaloyl galactosyl donors [7] **9** to give the glycotetraoside **10**, a precursor of **3**. But in those glycosylations, **11** was obtained as a major product. The *N-tert*-Butoxycarbonyl (Boc) group of **8** was attacked under the glycosylation conditions (Scheme 2 and Table 1).

Because of this result, we changed the amino protective group of neuraminic acid into the phthaloyl group. To differentiate the two amino protective groups in glycotetraoside **3**, compound **4** [5] with an azido group was chosen as the glycosyl donor.

Treatment of starting material **12** [3] with phthalic anhydride in methanol followed by acetylation with acetic anhydride in pyridine gave the carboxylic acid derivative **13** in two steps in 80% yield. Esterification with diazomethane in methanol afforded the appropriate glycosyl donor **6** in 91% yield.

Glycosylation of acceptor **7** [6] with donor **6** was performed in the presence of benzeneselenenyl triflate [8] as the promoter in propionitrile to give a 49% yield of a mixture of glycotriosides **5** and **14** (α:β 6:1) that was accompanied by 19% of byproduct **15**. This α selectivity was in accord with previous observations [9] performed with the Boc derivative of neuraminate [3].



Scheme 2.

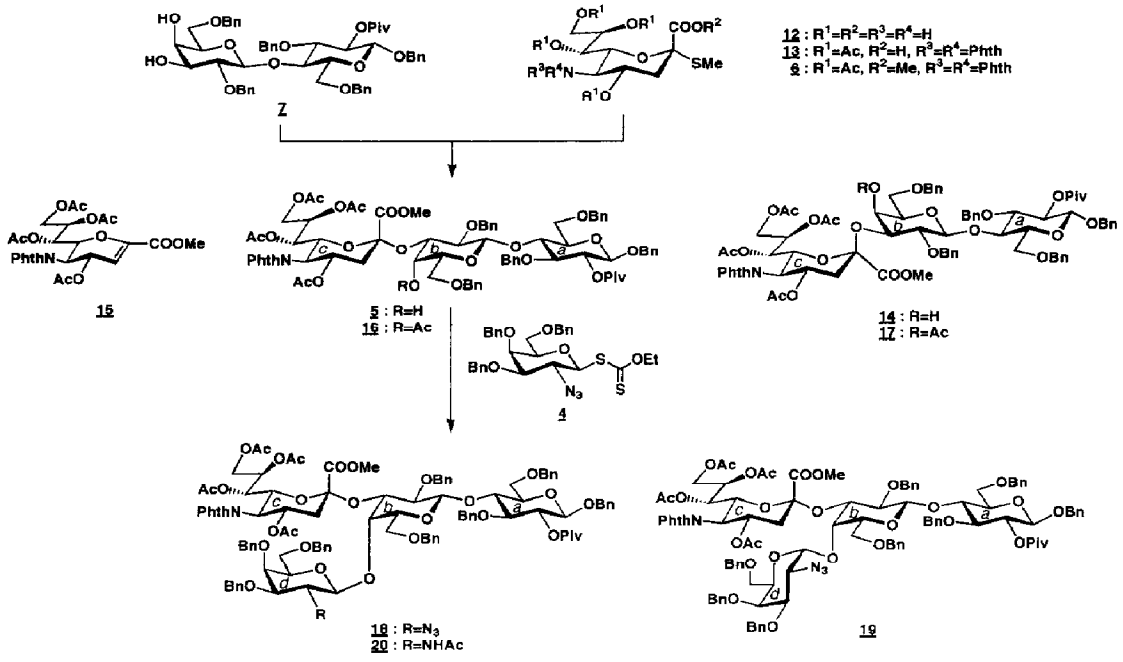
The regio- and stereo-chemistry of **5** and **14** were assigned as follows. Acetylation of **5** and **14** in acetic anhydride and pyridine afforded pentaacetates **16** and **17** in 76 and 94% yields, respectively. The ^{1} NMR data showed that H-4b in **16** and **17** was deshielded to δ 5.102 and 5.295, respectively, indicating that in **5** and **14** a new glycosidic linkage had been introduced at C-3b of the lactose residue. The configurations at C-2c in **5** and **14** were assigned as α and β, respectively, according to the ^{1}H NMR data. The signal for H-4c in **5** appeared at δ 5.495, while in **14** at δ 5.970, and the $J_{7c,8c}$ values for **5** and **14** were observed to be 8.8 and 2.6 Hz, respectively, in agreement with previous observations [10].

The crucial coupling of **5** and **4** [5] was performed in the presence of benzeneselenenyl triflate in propionitrile to give a 91% yield of a mixture of glycotetraosides **18** and **19** (β:α 5.5:1). The configurations of the newly formed glycosidic linkages in **18** and **19** were confirmed from the ^{1}H and ^{13}C NMR spectra. The signal for H-1d in **18** appeared at δ 4.474 with a coupling constant of 7.7 Hz, while that in **19** appeared at δ 4.926 with a coupling constant of 3.3 Hz, corresponding to β and α glycosidic linkages, respectively. The proton-coupled ^{13}C NMR spectra for C-1d of **18** and **19** show J_{CH} values of 162 and 175 Hz, confirming β and α configurations, respectively. Treatment of **18** with thioacetic acid [11] gave **20** in 84% yield. The ^{1}H NMR spectra of **20** are complex and show the presence of two rotamers in a ratio of ca. 2:1 ($CDCl_3$), as proven by recording spectra in CD_3OD (ratio of ca. 4:1) at an elevated temperature (40°C) (Scheme 3).

When **20** was subjected to hydrogenolysis with palladium hydroxide, followed by acetylation in acetic anhydride and pyridine, four products were obtained. The mixture could

Table 1
Glycosylation of galactosyl donors **9** with compound **8**

Entry	X	Promoter	Yield (%)	
			10	**11**
1	Br	AgOTf	13	45
2	Br	$HgBr_2$, $Hg(CN)_2$	20	55
3	Br	$Sn(OTf)_2$	5	39
4	SMe	PhSeOTf		15
5	SMe	MeOTf	5	49



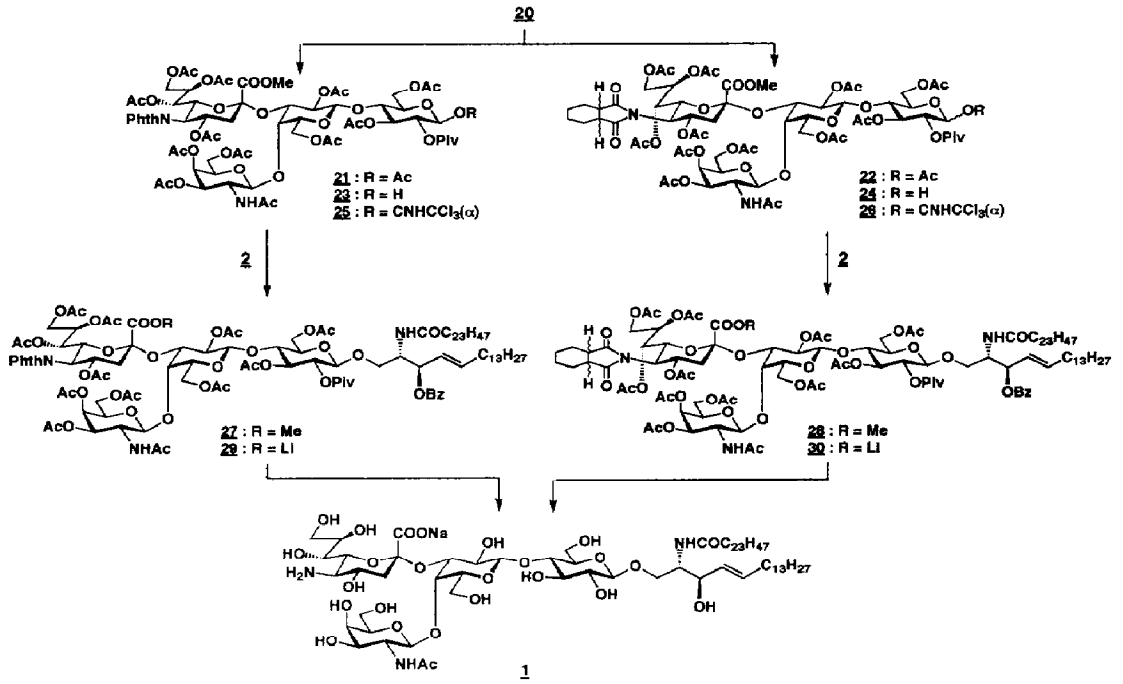
Scheme 3.

be separated by HPLC to give **21**α, **21**β, **22**α, and **22**β in 15, 13, 27, and 27% yields, respectively. The structures of these products were assigned on the basis of ^{1}H NMR spectroscopy. The minor compounds **21**α and **21**β were the desired products. On the other hand, the structure of major compounds **22**α and **22**β were assigned as cyclohexanedicarboxylimido derivatives because no signals were observed in the aromatic region. However, these cyclohexanedicarboxylimido group could be retained as the protective groups for the subsequent reaction sequences.

Regioselective deacetylation of the mixture of both pairs of **21**α, **21**β, and **22**α, **22**β with hydrazine acetate [12] in *N*,*N*-dimethylformamide afforded 81 and 70% yields of the hemiacetals **23** and **24**, respectively. These were then treated with trichloroacetonitrile [13] in the presence of 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) to give the α-imidates **25** and **26** in 99 and 94% yields, respectively. Boron trifluoride etherate-catalysed coupling of the ceramide derivative [4] **2** with the glycosyl donors **25** and **26** was performed in the presence of powdered 4A molecular sieves in chloroform to give the glycolipids **27** and **28** in 34 and 32% yields, respectively.

Selective cleavage of the methyl ester in **27** and **28** was carried out with lithium iodide in pyridine [14] to give **29** and **30** in 80 and 99% yields, respectively. Finally, deblocking of **29** and **30** with either methylamine (procedure A) or methyl hydrazine (procedure B) and subsequent purification by Sephadex LH-20 column chromatography gave the target compound **1** in 63 and 44% yields, respectively (Scheme 4).

The structure of compound **1** was confirmed by ^{1}H NMR spectra and mass spectrometry (Figs. 1 and 2). Three anomeric protons appeared at δ 4.844 (H-1d), 4.407 (H-1b), and 4.291 (H-1a) with $J_{H,H}$ values of 8.4, 8.1, and 7.7 Hz, respectively, indicating the β-



Scheme 4.

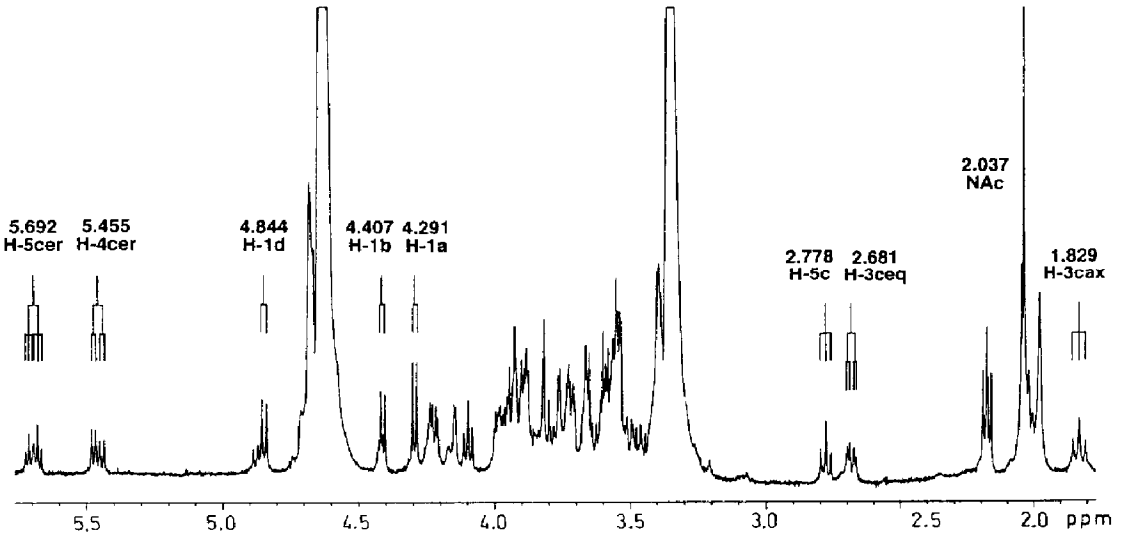


Fig. 1. 500-MHz ^{1}H NMR spectrum of synthetic de-*N*-acetyl GM_2 (**1**) in 1:1 $CDCl_3$-CD_3OD at 24°C.

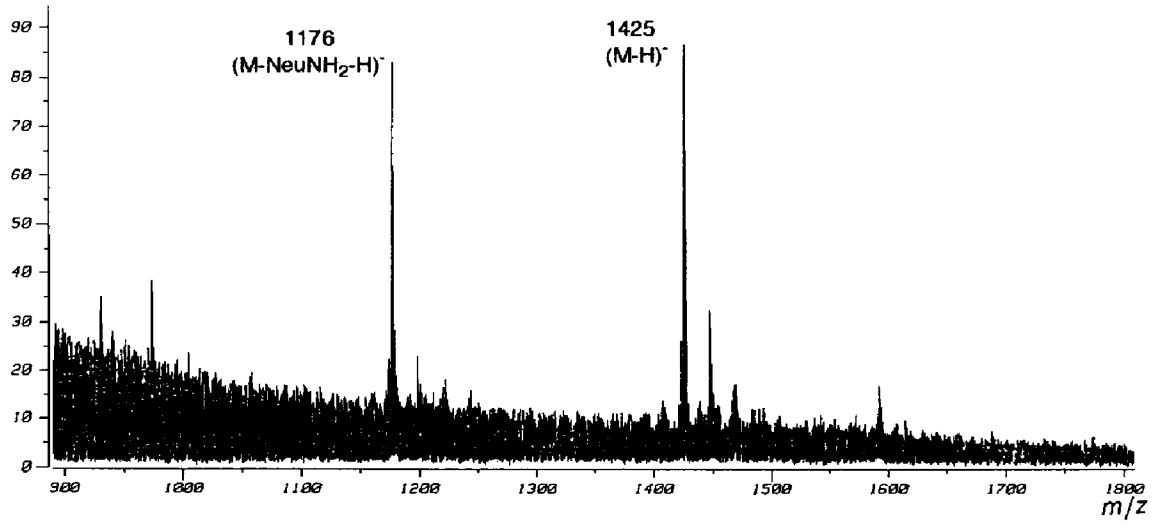


Fig. 2. Negative ion FAB-mass spectra of synthetic de-*N*-acetyl GM_2 (**1**) obtained by use of the triethanolamine matrix system.

glycosidic linkages. The signal at δ 2.778 was attributed to H-5c, supporting the absence of an acetyl group. In the negative ion FAB-mass spectrum of **1**, in a triethanolamine matrix, the pseudomolecular $(M-H)^-$ ion at m/z 1425 was observed abundantly.

In conclusion, de-*N*-acetyl GM_2 (**1**) was successfully sythesized using the glycosyl donor **6** via the key intermediate glycotrioside **5**.

3. Experimental

General.—Optical rotations were determined with a Jasco Model DIP-370 polarimeter for solutions in $CHCl_3$ at 25°C, unless noted otherwise. High-performance liquid chromatography (HPLC) was performed on a Inertsil PREP-SIL column (20.0×250 mm) with a flow rate of 15 mL/min using a GL-science pump (Model 572P). Flash chromatography was performed on columns of Wakogel C-300 (200–300 mesh). High-performance (HP) TLC was done on plates coated with Silica Gel 60 F_{254} (Merck). The 4A molecular sieves were purchased from Nakarai Chemicals. FAB-mass spectra were recorded with a Jeol HX-110 mass spectrometer. NMR spectra were recorded with a Jeol GX-500 (^1H at 500 MHz, ^{13}C at 125 MHz) spectrometer. Proton and ^{13}C chemical shifts are expressed in ppm downfield from internal Me_4Si for solutions in $CDCl_3$, unless noted otherwise.

Glycosylation of **8** *with* **9** *(X=Br) to give* **10** *and* **11** *(entry 2 in Table 1).*—To a mixture of **8** [3] (28 mg, 20 μmol), **9** [7] (21 mg, 41 μmol), and powdered 4A molecular sieves (0.50 g) in CH_2Cl_2 (0.70 mL) was added $HgBr_2$ (20 mg, 55 μmol) and $Hg(CN)_2$ (14 mg, 55 μmol) at 20°C under Ar. The mixture was stirred for 21 h, then diluted with $CHCl_3$ (20 mL) and filtered through Celite. The filtrate was washed with aq $NaHCO_3$ and aq NaCl, dried ($MgSO_4$), and evaporated in vacuo. The residue was chromatographed over Sephadex

LH-20 (90 mL) with MeOH and then chromatographed on silica gel (3.5 g) with 13:7 toluene–EtOAc to give **10** (7 mg, 20%) and **11** (19 mg, 55%).

Compound **10** had $[\alpha]_D$ −5.5° (*c* 0.53); R_f 0.45 in 1:1 toluene–EtOAc; NMR: δ_H 7.905 (d, 1 H, *J* 7.3 Hz, aromatic), 7.829 (d, 1 H, *J* 7.3 Hz, aromatic), 7.730 (t, 1 H, *J* 7.7 Hz, aromatic), 7.622 (t, 1 H, *J* 7.5 Hz, aromatic), 6.147 (dd, 1 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.5 Hz, H-3d), 5.536 (d, 1 H, H-4d), 5.394 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1d), 5.354 (dd, 1 H, $J_{7,6}$ 2.0, $J_{7,8}$ 9.3 Hz, H-7c), 5.052 (dd, 1 H, $J_{2,1}$ 8.4, $J_{2,3}$ 9.5 Hz, H-2a), 4.712 (ddd, 1 H, $J_{4,3eq}$ 4.5, $J_{4,5}$ 9.8, $J_{4,3ax}$ 12.4 Hz, H-4c), 4.605 (d, 1 H, H-2d), 4.449 (dd, 1 H, $J_{1,2}$ 7.7 Hz, H-1b), 4.401 (d, 1 H, H-1a), 4.065 (m, 2 H, H-9c, H-3b), 3.887 (s, 3 H, OMe), 3.878 (dd, 1 H, $J_{9',8}$ 4.8, $J_{9',9}$ 12.5 Hz, H-9′c), 3.854 (t, 1 H, $J_{4,3}=J_{4,5}=9.5$ Hz, H-4a), 3.601 (t, 1 H, H-3a), 2.822 (dd, 1 H, $J_{2,3}$ 9.7 Hz, H-2b), 2.778 (dd, 1 H, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq*), 2.233, 2.054, 2.045, 1.945, 1.929, 1.852, and 1.710 (7 s, 21 H, 7 OAc), 1.729 (t, 1 H, H-3c*ax*), 1.369 (s, 9 H, *t*-BuO), and 1.212 (s, 9 H, *t*-BuCO).

Compound **11** had $[\alpha]_D$ −3.9° (*c* 0.66); R_f 0.35 in 1:1 toluene–EtOAc; NMR: δ_H 7.840 (m, 2 H, aromatic), 7.745 (m, 2 H, aromatic), 6.215 (d, 1 H, $J_{1,2}$ 8.8 Hz, H-1d), 5.899 (dd, 1 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.5 Hz, H-3d), 5.501 (d, 1 H, H-4d), 5.382 (ddd, 1 H, $J_{8,9}$ 2.7, $J_{8,9'}$ 5.7, $J_{8,7}$ 8.4 Hz, H-8c), 5.326 (dd, 1 H, $J_{7,6}$ 2.2 Hz, H-7c), 5.103 (dd, 1 H, $J_{2,1}$ 7.7, $J_{2,3}$ 9.3 Hz, H-2a), 4.652 (d, 1 H, H-2d), 4.431 (d, 1 H, H-1a), 4.205 (dd, 1 H, $J_{9,9'}$ 12.5 Hz, H-9c), 4.053 (t, 1 H, $J_{4,3}=J_{4,5}=9.1$ Hz, H-4a), 3.984 (dd, 1 H, $J_{3,4}$ 3.3, $J_{3,2}$ 9.5 Hz, H-3b), 3.981 (dd, 1 H, H-9′c), 3.850 (dd, 1 H, $J_{6,5}$ 10.6 Hz, H-6c), 3.697 (s, 3 H, OMe), 3.603 (t, 1 H, H-3a), 3.590 (t, 1 H, $J_{5,4}=J_{5,6}=10.4$ Hz, H-5c), 3.491 (dd, 1 H, $J_{2,1}$ 7.7 Hz, H-2b), 2.372 (dd, 1 H, $J_{3eq,4}$ 4.8, $J_{3eq,3ax}$ 13.2 Hz, H-3c*eq*), 2.199, 2.060, 2.049, 1.973, 1.855, 1.842 and 1.483 (7 s, 21 H, 7 OAc), 1.119 (s, 9 H, *t*-Bu); δ_C 153.3 (*O*CONH), 102.4 (C-1b), 99.7 (C-1c), 98.1 (C-2c), and 91.4 (C-1d). Anal. Calcd for $C_{91}H_{104}N_2O_{34}$: C, 61.76; H, 5.92; N, 1.58. Found: C, 61.76; H, 5.95; N, 1.52.

Methyl (methyl 4,7,8,9-tetra-O-acetyl-5-phthalimido-3,5-dideoxy-2-thio-D-glycero-α-D-galacto-*2-nonulopyranosid)onate* (**6**).—To a solution of **12** [3] (297 mg, 1.0 mmol) in MeOH (10 ml) was added Et_3N (2.17 mL, 15.4 mmol) and phthalic anhydride (1.78 g, 12.0 mmol), and the mixture was stirred at room temperature for 5 days. After evaporation in vacuo, pyridine (10 mL), Ac_2O (10 mL), and 4-dimethylaminopyridine (DMAP) (122 mg, 1.0 mmol) were added to the residue, and stirred at room temperature for 20 h. The solvents were evaporated in vacuo and the residue was diluted with $CHCl_3$ and washed with 0.1 M HCl and aq NaCl, dried ($MgSO_4$), and evaporated in vacuo. The residue was chromatographed on silica gel (50 g) with 9:1 $CHCl_3$–MeOH to give **13** (474 mg, 79%); $[\alpha]_D$ +73.2° (*c* 1.0); R_f 0.40 in 4:1 $CHCl_3$–MeOH; NMR: δ_H 7.835 (m, 2 H, aromatic), 7.727 (m, 2 H, aromatic), 5.609 (dt, 1 H, $J_{4,3eq}$ 5.0, $J_{4,3ax}=J_{4,5}=10.9$ Hz, H-4), 5.478 (ddd, 1 H, $J_{8,9}$ 3.0, $J_{8,9'}$ 5.2, $J_{8,7}$ 8.5 Hz, H-8), 5.195 (dd, 1 H, $J_{7,6}$ 1.8 Hz, H-7), 4.983 (dd, 1 H, $J_{5,6}$ 10.6 Hz, H-6), 4.299 (dd, 1 H, $J_{9,9'}$ 12.5 Hz, H-9), 4.240 (t, 1 H, H-5), 4.084 (dd, 1 H, H-9′), 2.963 (dd, 1 H, $J_{3eq,3ax}$ 12.7 Hz, H-3*eq*), 2.238, 2.163, 2.146, 1.945, and 1.844 (5 s, 15 H, 4 OAc, SMe), and 2.003 (t, 1 H, H-3*ax*).

To a solution of **13** (42 mg, 70 μmol) in MeOH (3.0 mL) was added dropwise an ethereal solution of CH_2N_2 until TLC showed that the reaction was complete. After evaporation of the mixture in vacuo, the residue was chromatographed on silica gel (4.0 g) with 93:7 $CHCl_3$–THF to give **6** (39 mg, 91%); $[\alpha]_D$ +75.2° (*c* 1.0); R_f 0.49 in 9:1 $CHCl_3$–THF; NMR: δ_H 7.825 (m, 2 H, aromatic), 7.731 (m, 2 H, aromatic), 5.501 (dt, 1 H, $J_{4,3eq}$

5.1, $J_{4,3ax} = J_{4,5} = 10.6$ Hz, H-4), 5.454 (ddd, 1 H, $J_{8,9}$ 2.6, $J_{8,9'}$ 4.8, $J_{8,7}$ 9.1 Hz, H-8), 5.172 (dd, 1 H, $J_{7,6}$ 2.2 Hz, H-7), 4.873 (dd, 1 H, $J_{6,5}$ 10.6 Hz, H-6), 4.239 (dd, 1 H, $J_{9,9'}$ 12.5 Hz, H-9), 4.218 (t, 1 H, H-5), 3.899 (s, 3 H, OMe), 2.905 (dd, 1 H, $J_{3eq,3ax}$ 12.8 Hz, H-3*eq*), 2.195, 2.169, 2.151, 1.926, and 1.839 (5 s, 15 H, 4 OAc, SMe), and 1.988 (dd, 1 H, H-3*ax*). Anal. Calcd for $C_{27}H_{31}NO_{13}S$: C, 53.20; H, 5.13; N, 2.30. Found: C, 53.67; H, 5.16; N, 2.12.

Benzyl O-*(methyl 4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy-5-phthalimido*-D-glycero-α- (**5**) *and* -β-D-galacto-*2-nonulopyranosylonate)-(2→3)*-O-*(2,6-di*-O-*benzyl-β*-D-*galactopyranosyl)-(1→4)-3,6-di*-O-*benzyl-2*-O-*pivaloyl-β*-D-*glucopyranoside* (**14**).—A mixture of powdered 4A molecular sieves (0.90 g), **6** (275 mg, 0.45 mmol), and **7** [6] (791 mg, 0.90 mmol) in EtCN (10 mL) was stirred for 3 h at 20°C under Ar. After cooling to −50°C, a solution of AgOTf (234 mg, 0.90 mmol) in EtCN (2.0 mL), and PhSeCl (177 mg, 0.90 mmol) in EtCN (6.0 mL) were added. After stirring for 1 h at −50°C, the mixture was diluted with $CHCl_3$ (400 mL) and filtered through Celite. The filtrate was washed with aq $NaHCO_3$ and aq NaCl, dried ($MgSO_4$), and evaporated in vacuo. The residue was chromatographed over Sephadex LH-20 (130 mL) with MeOH and was further purified by HPLC with 93:7 $CHCl_3$–THF to give **5** (275 mg, 42%), **14** (44 mg, 7%), and **15** (49 mg, 19%).

Compound **5** had $[\alpha]_D$ +15.4° (*c* 1.0); R_F 0.23 in 19:1 $CHCl_3$–THF; NMR: δ_H 7.821 (m, 2 H, aromatic), 7.730 (m, 2 H, aromatic), 5.495 (m, 2 H, H-4c, H-8c), 5.161 (dd, 1 H, $J_{7,6}$ 2.6, $J_{7,8}$ 8.8 Hz, H-7c), 5.124 (dd, 1 H, $J_{2,1}$ 7.7, $J_{2,3}$ 9.2 Hz, H-2a), 5.026 (dd, 1 H, $J_{6,5}$ 10.6 Hz, H-6c), 4.575 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1b), 4.448 (d, 1 H, H-1a), 4.188 (dd, 1 H, $J_{9,8}$ 3.7, $J_{9,9'}$ 12.5 Hz, H-9c), 4.183 (t, 1 H, $J_{5,4}$ 10.6 Hz, H-5c), 4.143 (dd, 1 H, $J_{3,4}$ 3.3, $J_{3,2}$ 9.5 Hz, H-3b), 4.082 (t, 1 H, $J_{4,3} = J_{4,5} = 8.8$ Hz, H-4a), 3.937 (dd, 1 H, $J_{9',8}$ 5.5 Hz, H-9′c), 3.847 (s, 3 H, OMe), 3.829 (br s, 1 H, H-4b), 3.632 (t, 1 H, H-3a), 3.559 (dd, 1 H, H-2b), 2.721 (dd, 1 H, $J_{3eq,4}$ 5.1, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq*), 2.099, 1.929, 1.900, and 1.879 (4 s, 12 H, 4 OAc), 1.987 (t, 1 H, $J_{3ax,4}$ 12.5 Hz, H-3c*ax*), and 1.130 (s, 9 H, *t*-Bu). Anal. Calcd for $C_{78}H_{87}NO_{25}$: C, 65.13; H, 6.10; N, 0.97. Found: C, 65.00; H, 6.12; N, 0.91.

Compound **14** had $[\alpha]_D$ −3.7° (*c* 1.0); R_f 0.34 in 19:1 $CHCl_3$–THF; NMR: δ_H 7.827 (m, 2 H, aromatic), 7.727 (m, 2 H, aromatic), 5.970 (dt, 1 H, $J_{4,3eq}$ 5.1, $J_{4,3ax} = J_{4,5} = 11.4$ Hz, H-4c), 5.446 (dd, 1 H, $J_{6,7}$ 2.6, $J_{6,5}$ 10.6 Hz, H-6c), 5.280 (td, 1 H, $J_{8,7} = J_{8,9} = 2.6$, $J_{8,9'}$ 8.4 Hz, H-8c), 5.227 (t, 1 H, H-7c), 5.109 (dd, 1 H, $J_{2,1}$ 8.1, $J_{2,3}$ 9.2 Hz, H-2a), 4.888 (dd, 1 H, $J_{9,9'}$ 12.5 Hz, H-9c), 4.472 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1b), 4.448 (d, 1 H, H-1a), 4.233 (t, 1 H, H-5c), 4.050 (m, 2 H, H-4a, H-9′c), 3.618 (t, 1 H, $J_{3,4}$ 8.8 Hz, H-3a), 3.484 (s, 3 H, OMe), 2.794 (dd, 1 H, $J_{3eq,3ax}$ 13.2 Hz, H-3c*eq*), 2.093, 1.892, 1.866, and 1.836 (4 s, 12 H, 4 OAc), 1.899 (t, 1 H, H-3c*ax*), and 1.129 (s, 9 H, *t*-Bu). Anal. Calcd for $C_{78}H_{87}NO_{25}$: C, 65.13; H, 6.10; N, 0.97. Found: C, 65.00; H, 6.21; N, 0.96.

Compound **15** had $[\alpha]_D$ +80.4° (*c* 1.0); R_f 0.31 in 19:1 $CHCl_3$–THF; NMR: δ_H 7.855 (d, 1 H, *J* 5.5 Hz, aromatic), 7.845 (d, 1 H, *J* 5.5 Hz, aromatic), 7.751 (d, 1 H, *J* 5.5 Hz, aromatic), 7.745 (d, 1 H, *J* 5.5 Hz, aromatic), 6.139 (dd, 1 H, $J_{4,3}$ 2.6, $J_{4,5}$ 9.2 Hz, H-4), 6.003 (d, 1 H, H-3), 5.424 (dt, 1 H, $J_{8,9}$ 2.6, $J_{8,9'} = J_{8,7} = 6.2$ Hz, H-8), 5.252 (dd, 1 H, $J_{7,6}$ 2.0 Hz, H-7), 5.252 (dd, 1 H, $J_{6,5}$ 11.4 Hz, H-6), 4.541 (dd, 1 H, H-5), 4.500 (dd, 1 H, $J_{9,9'}$ 12.5 Hz, H-9), 4.116 (dd, 1 H, H-9′), 3.830 (s, 3 H, OMe), 2.096, 2.046, 1.964, and 1.930 (4 s, 12 H, 4 OAc). Anal. Calcd for $C_{26}H_{27}NO_{13}$: C, 55.62; H, 4.85; N, 2.49. Found: C, 56.04; H, 4.94; N, 2.55.

Benzyl O-*(methyl 4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy-5-phthalimido*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→)3*-O-*(4*-O-*acetyl-2,6-di*-O-*benzyl*-β-D-*galactopyranosyl)-(1→4)-3,6-di*-O-*benzyl-2*-O-*pivaloyl*-β-D-*glucopyranoside* (**16**).—A solution of **5** (15 mg, 10 μmol) and DMAP (3.0 mg, 24 μmol) in Ac_2O (50 μL) and pyridine (0.20 mL) was stirred for 17 h at 20°C. The mixture was evaporated in vacuo. The residue was chromatographed over Sephadex LH-20 (17 mL) with MeOH and then chromatographed on silica gel (1.8 g) with 24:1 $CHCl_3$–THF to give **16** (12 mg, 76%); $[\alpha]_D$ +17.0° (*c* 1.0); R_f 0.40 in 19:1 $CHCl_3$–THF; NMR: δ_H 7.811 (m, 2 H, aromatic), 7.722 (m, 2 H, aromatic), 5.666 (ddd, 1 H, $J_{8,9}$ 2.6, $J_{8,9'}$ 4.4, $J_{8,7}$ 9.2 Hz, H-8c), 5.576 (dt, 1 H, $J_{4,3eq}$ 5.1, $J_{4,3ax}=J_{4,5}=11.4$ Hz, H-4c), 5.177 (dd, 1 H, $J_{7,6}$ 2.6 Hz, H-7c), 5.116 (dd, 1 H, $J_{2,1}$ 8.4, $J_{2,3}$ 9.2 Hz, H-2a), 5.102 (br d, 1 H, $J_{4,3}$ 3.3 Hz, H-4b), 4.761 (dd, 1 H, $J_{6,5}$ 11.0 Hz, H-6c), 4.761 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1b), 4.618 (dd, 1 H, $J_{3,2}$ 9.9 Hz, H-3b), 4.425 (d, 1 H, H-1a), 4.182 (t, 1 H, H-5c), 4.165 (dd, 1 H, $J_{9,9'}$ 12.5 Hz, H-9c), 4.094 (t, 1 H, $J_{4,3}=J_{4,5}=9.2$ Hz, H-4a), 3.963 (dd, 1 H, H-9′c), 3.928 (s, 3 H, OMe), 3.628 (t, 1 H, H-3a), 3.465 (dd, 1 H, H-2b), 2.742 (dd, 1 H, $J_{3eq,3ax}$ 12.5 Hz, H-3c*eq*), 2.093, 2.002, 1.858, 1.828, and 1.815 (5 s, 15 H, 5 OAc), 1.790 (t, 1 H, H-3c*ax*), and 1.132 (s, 9 H, *t*-Bu). Anal. Calcd for $C_{80}H_{89}NO_{26}$: C, 64.90; H, 6.06; N, 0.95. Found: C, 64.40; H, 6.18; N, 0.99.

Benzyl O-*(methyl 4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy-5-phthalimido*-D-glycero-β-D-galacto-*2-nonulopyranosylonate)-(2→3)*-O-*(4*-O-*acetyl-2,6-di*-O-*benzyl*-β-D-*galactopyranosyl)-(1→4)-3,6-di*-O-*benzyl-2*-O-*pivaloyl*-β-D-*glucopyranoside* (**17**).—Compound **14** (6.9 mg) was converted into **17** (6.7 mg, 94%) as described for the synthesis of **16**. Compound **17** had $[\alpha]_D$ +6.1° (*c* 0.35); R_f 0.47 in 19:1 $CHCl_3$–THF; NMR: δ_H 7.805 (m, 2 H, aromatic), 7.703 (m, 2 H, aromatic), 5.835 (dt, 1 H, $J_{4,3eq}$ 4.8, $J_{4,3ax}=J_{4,5}=11.0$ Hz, H-4c), 5.585 (td, 1 H, $J_{8,9}=J_{8,7}=2.2$, $J_{8,9'}$ 9.5 Hz, H-8c), 5.300 (dd, 1 H, $J_{9,9'}$ 12.5 Hz, H-9c), 5.295 (d, 1 H, $J_{4,3}$ 3.7 Hz, H-4b), 5.268 (dd, 1 H, $J_{7,6}$ 2.2 Hz, H-7c), 5.122 (dd, 1 H, $J_{2,1}$ 8.1, $J_{2,3}$ 9.2 Hz, H-2a), 5.058 (dd, 1 H, $J_{6,5}$ 10.6 Hz, H-6c), 4.525 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1b), 4.432 (d, 1 H, H-1a), 4.271 (dd, 1 H, $J_{3,2}$ 9.9 Hz, H-3b), 4.155 (t, 1 H, H-5c), 3.878 (dd, 1 H, H-9′c), 3.653 (dd, 1 H, H-2b), 3.642 (t, 1 H, $J_{3,4}$ 9.0 Hz, H-3a), 3.355 (s, 3 H, OMe), 2.806 (dd, 1 H, $J_{3eq,3ax}$ 13.6 Hz, H-3c*eq*), 2.277, 2.105, 1.909, 1.850, and 1.800 (5 s, 15 H, 5 OAc), 1.930 (dd, 1 H, H-3c*ax*), and 1.126 (s, 9 H, *t*-Bu). Anal. Calcd for $C_{80}H_{89}NO_{26}$: C, 64.90; H, 6.06; N, 0.95. Found: C, 64.48; H, 6.21; N, 1.02.

Benzyl O-(*methyl* 4,7,8,9-*tetra*-O-*acetyl*-3,5-*dideoxy*-5-*phthalimido*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)*-O-*[2-azido-3,4,6-tri*-O-*benzyl-2-deoxy*-β- (**18**) *and* -α-D-*galactopyranosyl-(1→4)]*-O-*(2,6-di*-O-*benzyl*-β-D-*galactopyranosyl)-(1→4)-3,6-di*-O-*benzyl-2*-O-*pivaloyl*-β-D-*glucopyranoside* (**19**).—A mixture of 4A molecular sieves (1.04 g), compound **5** (133 mg, 0.092 mmol), and compound **4** [5] (268 mg, 0.463 mmol) in EtCN (2.6 mL) was stirred for 4 h at 20°C under Ar. After cooling to −78°, a solution of AgOTf (180 mg, 0.694 mmol) in EtCN (1.0 mL), and PhSeCl (136 mg, 0.696 mmol) in EtCN (1.0 mL) were added. After stirring for 6.5 h at −78°C, the mixture was diluted with $CHCl_3$ (100 mL) and filtered through Celite. The filtrate was washed with aq $NaHCO_3$ and aq NaCl, dried ($MgSO_4$), and evaporated in vacuo. The residue was chromatographed over Sephadex LH-20 (530 mL) with MeOH and then further purified on silica gel (17 g) with 4:1 toluene–EtOAc to give **18** (135 mg, 77%) and **19** (24 mg, 14%).

Compound **18** had $[\alpha]_D$ +3.5° (*c* 0.98); R_f 0.34 in 3:1 toluene–EtOAc; NMR: δ_H 7.830 (m, 2 H, aromatic), 7.731 (m, 2 H, aromatic), 5.430 (ddd, 1 H, $J_{4,3eq}$ 5.1, $J_{4,5}$ 10.6, $J_{4,3ax}$

11.7 Hz, H-4c), 5.344 (ddd, 1 H, $J_{8,9}$ 2.5, $J_{8,9'}$ 4.0, $J_{8,7}$ 9.5 Hz, H-8), 5.121 (dd, 1 H, $J_{7,6}$ 1.8 Hz, H-7c), 5.047 (dd, 1 H, $J_{2,1}$ 8.1, $J_{2,3}$ 9.5 Hz, H-2a), 4.656 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1b), 4.474 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1d), 4.429 (d, 1 H, H-1a), 4.174 (dd, 1 H, $J_{9,9'}$ 12.8 Hz, H-9c), 4.115 (t, 1 H, $J_{5,6}$ 10.3 Hz, H-5c), 3.955 (dd, 1 H, H-9′c), 3.894 (dd, 1 H, $J_{2,3}$ 10.6 Hz, H-2b), 3.840 (dd, 1 H, $J_{2,3}$ 9.5 Hz, H-2d), 3.768 (d, 1 H, $J_{4,3}$ 3.3 Hz, H-4b), 3.739 (s, 3 H, OMe), 3.577 (t, 1 H, $J_{3,4}$ 8.8 Hz, H-3a), 3.369 (dd, 1 H, H-3b), 2.580 (dd, 1 H, $J_{3eq,3ax}$ 13.6 Hz, H-3c*eq*), 2.086 (dd, 1 H, H-3c*ax*), 2.068, 1.912, 1.836, and 1.776 (4 s, 12 H, 4 OAc), 1.195 (s, 9 H, *t*-Bu); δ_C 103.0 ($J_{C,H}$ 164.4 Hz, C-1b), 102.3 ($J_{C,H}$ 161.9 Hz, C-1d), 99.9 ($J_{C,H}$ 161.9 Hz, C-1a), and 99.3 (C-2c). Anal. Calcd for $C_{105}H_{114}N_4O_{29}$: C, 66.51; H, 6.06; N, 2.95. Found: C, 65.89; H, 6.10; N, 2.95 (contamination with EtOAc).

Compound **19** had $[\alpha]_D$ +27.7° (*c* 0.91); R_f 0.40 in 3:1 toluene–EtOAc; NMR: δ_H 7.811 (m, 2 H, aromatic), 7.726 (m, 2 H, aromatic), 5.685 (ddd, 1 H, $J_{8,9}$ 2.6, $J_{8,9'}$ 4.4, $J_{8,7}$ 9.2 Hz, H-8), 5.506 (ddd, 1 H, $J_{4,3eq}$ 5.1, $J_{4,5}$ 10.6, $J_{4,3ax}$ 11.5 Hz, H-4c), 5.194 (dd, 1 H, $J_{7,6}$ 2.6 Hz, H-7c), 5.104 (dd, 1 H, $J_{2,1}$ 7.7, $J_{2,3}$ 9.5 Hz, H-2a), 4.926 (d, 1 H, $J_{1,2}$ 3.3 Hz, H-1d), 4.860 (dd, 1 H, $J_{6,5}$ 10.6 Hz, H-6c), 4.793 (d, 1 H, $J_{1,2}$ 7.3 Hz, H-1b), 4.412 (d, 1 H, H-1a), 4.224 (t, 1 H, H-5c), 4.125 (dd, 1 H, $J_{9,9'}$ 12.8 Hz, H-9c), 4.073 (br.s, 1 H, H-4d), 3.978 (t, 1 H, $J_{4,3}=J_{4,5}=9.9$ Hz, H-4a), 3.981 (dd, 1 H, H-9′c), 3.737 (s, 3 H, OMe), 3.681 (d, 1 H, $J_{4,3}$ 2.2 Hz, H-4b), 3.554 (dd, 1 H, $J_{2,3}$ 10.6 Hz, H-2b), 2.844 (dd, 1 H, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq*), 2.114, 1.875, 1.826, and 1.565 (4 s, 12 H, 4 OAc), 1.061 (s, 9 H, *t*-Bu); δ_C 103.2 ($J_{C,H}$ 162.7 Hz, C-1b), 99.3 ($J_{C,H}$ 161.0 Hz, C-1a), 98.2 ($J_{C,H}$ 174.5 Hz, C-1d), and 97.7 (C-2c). Anal. Calcd for $C_{105}H_{114}N_4O_{29}$: C, 66.51; H, 6.06; N, 2.95. Found: C, 66.72; H, 6.11; N, 3.04.

Benzyl O-*(methyl 4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy-5-phthalimido*-D-glycero-*α*-D-galacto-*2-nonulopyranosylonate)-(2→3)*-O-*[2-acetamido-3,4,6-tri*-O-*benzyl-2-deoxy-β*-D-*galactopyranosyl-(1→4)]*-O-*(2,6-di*-O-*benzyl-β*-D-*galactopyranosyl)-(1→4)-3,6-di*-O-*benzyl-2*-O-*pivaloyl-β*-D-*glucopyranoside* (**20**).—A solution of **18** (104 mg, 55 μmol) in thioacetic acid (1.0 mL) was stirred for 3 days at room temperature. The mixture was chromatographed over Sephadex LH-20 (90 mL) with MeOH, and then further purified on silica gel (12.0 g) with 19:1 $CHCl_3$–THF to give 20 (88 mg, 84%); $[\alpha]_D$ +16.2° (*c* 0.80); R_f 0.36 in 3:2 toluene–EtOAc; NMR ($CDCl_3$, 24°C): δ_H 7.836 (m, 2 H, aromatic), 7.741 (m, 2 H, aromatic), 6.124 (d, 0.65 H, $J_{NH,2}$ 9.5 Hz, NH), 6.018 (d, 0.65 H, $J_{NH,2}$ 10.3 Hz, NH), 5.664 (dt, 0.65 H, $J_{4,3eq}$ 5.5, $J_{4,3ax}=J_{4,5}=10.8$ Hz, H-4c), 5.647 (dt, 0.35 H, $J_{4,3eq}$ 5.5, $J_{4,3ax}=J_{4,5}=10.8$ Hz, H-4c), 5.228 (m, 1 H, H-8), 3.883 (s, 1.05 H, OMe), 3.763 (s, 1.95 H, OMe), 2.427 (dd, 0.65 H, $J_{3eq,3ax}$ 13.6 Hz, H-3c*eq*), 2.266 (dd, 0.35 H, $J_{3eq,3ax}$ 13.6 Hz, H-3c*eq*), 2.152 (dd, 0.65 H, H-3c*ax*), 2.136 (dd, 0.35 H, H-3c*ax*), 2.036 (1.05 H), 2.019 (1.95 H), 1.914 (1.05 H), 1.904 (1.95 H), 1.885 (1.05 H), 1.860 (1.95 H), 1.834 (1.05 H), 1.827 (1.95 H), 1.805 (1.95 H), and 1.786 (1.05 H) (10 s, 15 H, 4 OAc, NAc), 1.194 (s, 3.15 H, *t*-Bu), 1.187 (s, 5.85 H, *t*-Bu); (CD_3OD, 40°): δ_H 7.835 (m, 4 H, aromatic), 5.723 (dt, 0.2 H, $J_{4,3eq}$ 5.5, $J_{4,3ax}=J_{4,5}=10.8$ Hz, H-4c), 5.695 (dt, 0.8 H, $J_{4,3eq}$ 5.1, $J_{4,3ax}=J_{4,5}=10.8$ Hz, H-4c), 5.317 (td, 0.8 H, $J_{8,9}=J_{8,9'}=3.1$, $J_{8,7}$ 9.5 Hz, H-8c), 5.261 (td, 0.2 H, $J_{8,9}=J_{8,9'}$ 3.3, $J_{8,7}$ 9.5 Hz, H-8c), 5.082 (br d, 1 H, H-7c), 4.104 (br d, 1 H, $J_{9,9'}$ 13.4 Hz, H-9c), 3.887 (s, 0.6 H, OMe), 3.863 (s, 2.4 H, OMe), 2.520 (dd, 0.8 H, $J_{3eq,3ax}$ 13.9 Hz, H-3c*eq*), 2.367 (dd, 0.2 H, $J_{3eq,3ax}$ 14.0 Hz, H-3c*eq*), 2.095 (2.4 H), 2.082 (0.6 H), 1.920 (2.4 H), 1.883 (0.6 H), 1.869 (2.4 H), 1.825 (0.6 H), 1.805 (3 H), 1.801 (2.4 H), and 1.790 (0.6 H) (9 s, 15 H, 4 OAc, NAc), 1.188 (s, 7.2 H, *t*-Bu), and

1.175 (s, 1.8 H, *t*-Bu). Anal. Calcd for $C_{107}H_{118}N_2O_{30}$: C, 67.21; H, 6.22; N, 1.47. Found: C, 66.91; H, 6.23; N, 1.42.

Conversion of **20** *into the* **21α**, **21β**, **22α**, *and* **22β**.—A mixture of **20** (98 mg, 51 μmol) and 20% $Pd(OH)_2$–C (362 mg) in MeOH (10 mL) was stirred for 48 h at 20°C under H_2. After filtration, the solution was evaporated in vacuo to dryness. Without purification, a mixture of the residue, Ac_2O (1 mL), pyridine (1 mL), and DMAP (12 mg, 0.10 mmol) was stirred for 3 days at 20°C, and then evaporated in vacuo. The residual products were chromatographed over Sephadex LH-20 (130 mL) with MeOH and then further purified by HPLC with 7:3 toluene–THF and EtOAc to give **21α** (12 mg, 15%), **21β** (10 mg, 13%), **22α** (21 mg, 27%), and **22β** (21 mg, 27%).

Compound **21α** had $[\alpha]_D$ +40.9° (*c* 0.33); R_f 0.38 in 3:2 toluene–THF; R_f 0.38 in EtOAc; NMR: δ_H 7.819 (m, 2 H, aromatic), 7.730 (m, 2 H, aromatic), 6.305 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1a), 6.609 (d, 1 H, $J_{NH,2}$ 7.3 Hz, NH), 5.829 (dd, 1 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.0 Hz, H-3d), 5.575 (td, 1 H, $J_{8,9}=J_{8,9'}=2.9$, $J_{8,7}$ 9.9 Hz, H-8c), 5.498 (dt, 1 H, $J_{4,3eq}$ 5.6, $J_{4,3ax}=J_{4,5}=11.0$ Hz, H-4c), 5.480 (t, 1 H, $J_{3,2}=J_{3,4}=9.7$ Hz, H-3a), 5.367 (d, 1 H, H-4d), 5.224 (dd, 1 H, $J_{7,6}$ 2.2 Hz, H-7c), 5.123 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1d), 5.004 (dd, 1 H, $J_{2,1}$ 7.3, $J_{2,3}$ 10.6 Hz, H-2b), 4.987 (dd, 1 H, H-2a), 4.879 (dd, 1 H, $J_{6,5}$ 11.0 Hz, H-6c), 4.630 (d, 1 H, H-1b), 4.338 (br dd, 1 H, $J_{3,4}$ 2.4 Hz, H-3b), 4.284 (dd, 1 H, $J_{9,9'}$ 9.2 Hz, H-9c), 4.105 (t, 1 H, H-5c), 3.981 (dd, 1 H, H-9′c), 3.930 (s, 3 H, OMe), 3.596 (br.d, 1 H, H-4b), 3.432 (td, 1 H, H-2d), 2.955 (dd, 1 H, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq*), 2.258, 2.217, 2.146, 2.143, 2.109, 2.098, 2.091, 2.072, 2.030, 2.008, 1.994, 1.942, and 1.834 (13 s, 39 H, 12 OAc, NAc), 1.782 (t, 1 H, H-3c*ax*), and 1.135 (s, 9 H, *t*-Bu).

Compound **21β** had $[\alpha]_D$ +17.4° (*c* 0.56); R_f 0.42 in 3:2 toluene–THF; R_f 0.42 in EtOAc; NMR: δ_H 7.817 (m, 2 H, aromatic), 7.731 (m, 2 H, aromatic), 6.072 (d, 1 H, $J_{NH,2}$ 7.0 Hz, NH), 5.843 (dd, 1 H, $J_{3,4}$ 3.5, $J_{3,2}$ 11.2 Hz, H-3d), 5.712 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1a), 5.578 (ddd, 1 H, $J_{8,9}$ 2.9, $J_{8,9'}$ 4.0, $J_{8,7}$ 9.9 Hz, H-8c), 5.493 (dt, 1 H, $J_{4,3eq}$ 4.8, $J_{4,3ax}=J_{4,5}=11.7$ Hz, H-4c), 5.363 (d, 1 H, H-4d), 5.262 (t, 1 H, $J_{3,2}=J_{3,4}=9.5$ Hz, H-3a), 5.236 (dd, 1 H, $J_{7,6}$ 2.9 Hz, H-7c), 5.131 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1d), 5.069 (dd, 1 H, H-2a), 4.987 (dd, 1 H, $J_{2,1}$ 7.7, $J_{2,3}$ 9.9 Hz, H-2b), 4.876 (dd, 1 H, $J_{6,5}$ 11.0 Hz, H-6c), 4.618 (d, 1 H, H-1b), 4.329 (br dd, 1 H, $J_{3,4}$ 2.6 Hz, H-3b), 4.272 (dd, 1 H, $J_{9,9'}$ 12.8 Hz, H-9c), 4.103 (t, 1 H, H-5c), 3.973 (dd, 1 H, H-9′c), 3.916 (t, 1 H, $J_{4,5}$ 9.5 Hz, H-4a), 3.924 (s, 3 H, OMe), 3.755 (ddd, 1 H, $J_{5,6}$ 2.2, $J_{5,6'}$ 5.5 Hz, H-5a), 3.582 (br d, 1 H, H-4b), 3.399 (td, 1 H, H-2d), 2.955 (dd, 1 H, $J_{3eq,3ax}$ 13.0 Hz, H-3c*eq*), 2.246, 2.168, 2.137, 2.112, 2.099, 2.089, 2.071, 2.063, 2.027, 1.995, 1.988, 1.948, and 1.830 (13 s, 39 H, 12 OAc, NAc), 1.721 (t, 1 H, H-3c*ax*), and 1.144 (s, 9 H, *t*-Bu).

Compound **22α** had $[\alpha]_D$ +17.9° (*c* 0.66); R_f 0.37 in 3:2 toluene–THF; R_f 0.33 in EtOAc; mass spectrum (positive ion mode, 3-nitrobenzyl alcohol matrix): *m/z* 1533 $(M+H)^+$; NMR: δ_H 6.302 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1a), 6.025 (d, 0.4 H, $J_{NH,2}$ 7.3 Hz, NH*trans*), 5.990 (d, 0.6 H, $J_{NH,2}$ 7.3 Hz, NH*cis*), 5.840 (dd, 0.4 H, $J_{3,4}$ 3.7, $J_{3,2}$ 11.4 Hz, H-3d*trans*), 5.783 (dd, 0.6 H, $J_{3,4}$ 3.7, $J_{3,2}$ 11.4 Hz, H-3d*cis*), 5.537 (td, 0.6 H, $J_{8,9}=J_{8,9'}=3.3$, $J_{8,7}$ 9.5 Hz, H-8c*cis*), 5.359 (br s, 1 H, H-4d), 5.156 (dd, 0.6 H, $J_{7,6}$ 2.6 Hz, H-7c*cis*), 5.124 (dd, 0.4 H, $J_{7,6}$ 2.6, $J_{7,8}$ 9.9 Hz, H-7c*trans*), 5.091 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1d), 4.982 (dd, 1 H, $J_{2,3}$ 10.3 Hz, H-2a), 4.798 (dd, 0.4 H, $J_{6,5}$ 11.0 Hz, H-6c*trans*), 4.793 (dd, 0.6 H, $J_{6,5}$ 11.0 Hz, H-6c*cis*), 4.605 (d, 0.6 H, $J_{1,2}$ 8.1 Hz, H-1b*cis*), 4.594 (d, 0.4 H, $J_{1,2}$ 7.7 Hz, H-1b*trans*), 4.320 (br dd, 0.4 H, $J_{3,4}$ 2.4, $J_{3,2}$ 10.4 Hz, H-3b*trans*), 3.892

(s, 1.8 H, OMe*cis*), 3.881 (s, 1.2 H, OMe*trans*), 3.515 (br.d, 0.4 H, H-4b*trans*), 3.566 (br d, 0.6 H, H-4b*cis*), 3.454 (td, 0.6 H, H-2d*cis*), 3.401 (td, 0.6 H, H-2d*trans*), 2.930 (dd, 0.6 H, $J_{3eq,4}$ 4.8, $J_{3eq,3ax}$ 13.2 Hz, H-3c*eq-cis*), 2.892 (dd, 0.4 H, $J_{3eq,4}$ 5.1, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq-trans*), 2.818 (m, 1.2 H, cyclohexane ring H-1, 2*cis*), 2.711 (q, 0.4 H, J 7.0 Hz, cyclohexane ring H-1 or 2*trans*), 2.641 (q, 0.4 H, J 7.3 Hz, cyclohexane ring H-1 or 2*trans*), 2.228, 2.209, 2.162, 2.159, 2.148, 2.140, 2.097, 2.086, 2.060, 2.044, 2.030, 2.022, 2.015, 2.000, 1.990, 1.969, and 1.954 (17 s, 39 H, 12 OAc, NAc), and 1.131 (s, 9 H, *t*-Bu).

Compound **22β** had $[\alpha]_D$ −1.4° (*c* 1.23); R_f 0.42 in 3:2 toluene–THF; R_f 0.37 in EtOAc; mass spectrum (positive ion mode, 3-nitrobenzyl alcohol matrix): *m/z* 1533 $(M+H)^+$; NMR: δ_H 6.017 (d, 0.4 H, $J_{NH,2}$ 7.0 Hz, NH*trans*), 5.982 (d, 0.6 H, $J_{NH,2}$ 7.3 Hz, NH*cis*), 5.848 (dd, 0.4 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.7 Hz, H-3d*trans*), 5.794 (dd, 0.6 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.4 Hz, H-3d*cis*), 5.70 6 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1a), 5.538 (td, 0.6 H, $J_{8,9}=J_{8,9'}=2.6$, $J_{8,7}$ 10.3 Hz, H-8c*cis*), 5.477 (td, 0.4 H, $J_{8,9}=J_{8,9'}=2.6$, $J_{8,7}$ 9.6 Hz, H-8c*trans*), 5.425 (m, 1 H, H-4c), 5.355 (br s, 1 H, H-4d), 5.249 (br t, 1 H, $J_{3,2}=J_{3,4}=9.4$ Hz, H-3a), 5.149 (dd, 0.6 H, $J_{7,6}$ 2.6 Hz, H-7c*cis*), 5.116 (dd, 0.4 H, $J_{7,6}$ 2.2, H-7c*trans*), 5.094 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1d), 5.065 (t, 1 H, H-2a), 4.957 (t, 1 H, $J_{2,1}=J_{2,3}=8.1$ Hz, H-2b), 4.791 (m, 1 H, H-6c), 4.595 (d, 0.4 H, $J_{1,2}$ 7.3 Hz, H-1b*trans*), 4.584 (d, 0.6 H, $J_{1,2}$ 7.1 Hz, H-1b*cis*), 4.037 (dd, 0.6 H, $J_{9,9'}$ 12.8 Hz, H-9c*cis*), 3.886 (s, 1.8 H, OMe*cis*), 3.876 (s, 1.2 H, OMe*trans*), 3.739 (m, 1 H, H-5a), 3.554 (br s, 1 H, H-4b), 3.423 (td, 0.6 H, H-2d*cis*), 3.372 (td, 0.6 H, H-2d*trans*), 2.928 (dd, 0.6 H, $J_{3eq,4}$ 5.1, $J_{3eq,3ax}$ 12.5 Hz, H-3c*eq-cis*), 2.891 (dd, 0.4 H, $J_{3eq,4}$ 5.1, $J_{3eq,3ax}$ 13.2 Hz, H-3c*eq-trans*), 2.814 (m, 1.2 H, cyclohexane ring H-1, 2*cis*), 2.708 (q, 0.4 H, J 7.7 Hz, cyclohexane ring H-1 or 2*trans*), 2.637 (q, 0.4 H, J 6.8 Hz, cyclohexane ring H-1 or 2*trans*), 2.214, 2.197, 2.160, 2.145, 2.132, 2.101, 2.084, 2.069, 2.052, 2.044, 2.031, 2.019, 2.005, 1.984, 1.975, 1.966, and 1.952 (17 s, 39 H, 12 OAc, NAc), and 1.140 (s, 9 H, *t*-Bu).

Conversion of **21α** *and* **21β** *into the hemiacetal* **23**.—A mixture of **21α** and **21β** (combined weight 16 mg, 11 μmol) and $H_2NNH_2 \cdot AcOH$ (2.0 mg, 22 μmol) in Me_2NCHO (1.0 mL) was stirred for 30 min at 65°C, and then diluted with $CHCl_3$ (30 mL). The organic layer was washed with water. The aqueous layer was re-extracted with $CHCl_3$ and the combined organic layers washed with aq NaCl, dried ($MgSO_4$), and evaporated in vacuo. The residue was chromatographed on silica gel (1.8 g) with 97:3 $CHCl_3$–MeOH to give **23** (13 mg, 81%); $[\alpha]_D$ +32.8° (*c* 0.76); R_f 0.15 in 97:3 $CHCl_3$--MeOH; NMR: δ_H 7.819 (m, 2 H, aromatic), 7.735 (m, 2 H, aromatic), 6.303 (d, 0.7 H, $J_{NH,2}$ 7.0 Hz, NHα), 6.173 (d, 0.3 H, $J_{NH,2}$ 7.3 Hz, NHβ), 5.920 (dd, 0.7 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.0 Hz, H-3dα), 5.845 (dd, 0.7 H, $J_{3,4}$ 3.5, $J_{3,2}$ 11.0 Hz, H-3dβ), 5.577 (td, 1 H, $J_{8,9}=J_{8,9'}=3.7$, $J_{8,7}$ 9.5 Hz, H-8c), 5.538 (dd, 0.7 H, $J_{3,4}$ 9.2, $J_{3,2}$ 9.9 Hz, H-3aα), 5.491 (ddd, 1 H, $J_{4,3eq}$ 4.8, $J_{4,5}$ 10.3, $J_{4,3ax}$ 12.1 Hz, H-4c), 5.397 (br d, 0.7 H, $J_{1,2}$ 2.6 Hz, H-1aα), 5.370 (d, 0.7 H, H-4dα), 5.364 (d, 0.3 H, H-4dβ), 5.275 (t, 0.3 H, $J_{3,2}=J_{3,4}=9.5$ Hz, H-3aβ), 5.249 (dd, 0.7 H, $J_{7,6}$ 2.6 Hz, H-7cα), 5.241 (dd, 0.3 H, $J_{7,6}$ 2.9 Hz, H-7cβ), 5.159 (d, 0.7 H, $J_{1,2}$ 8.1 Hz, H-1da), 5.131 (d, 0.3 H, $J_{1,2}$ 8.4 Hz, H-1dβ), 5.007 (dd, 0.7 H, $J_{2,1}$ 8.1, $J_{2,3}$ 10.3 Hz, H-2bα), 4.987 (dd, 0.3 H, $J_{2,1}$ 8.1, $J_{2,3}$ 10.6 Hz, H-2bβ), 4.880 (dd, 1 H, $J_{6,5}$ 10.6 Hz, H-6c), 4.798 (dd, 0.7 H, H-2aα), 4.778 (dd, 0.3 H, $J_{2,1}$ 8.1 Hz, H-2aβ), 4.692 (br t, 0.3 H, $J_{1,2}=J_{1,OH}=7.7$ Hz, H-1aβ), 4.611 (d, 1 H, H-1b), 4.332 (br dd, 0.3 H, $J_{3,4}$ 2.8 Hz, H-3bβ), 4.329 (br dd, 0.7 H, $J_{3,4}$ 2.6 Hz, H-3bα), 4.100 (t, 1 H, H-5c), 3.926 (s, 0.9 H,

OMeβ), 3.923 (s, 2.1 H, OMeα), 3.584 (br d, 0.3 H, H-4bβ), 3.580 (br d, 0.7 H, H-4bα), 3.399 (td, 0.3 H, H-2dβ), 3.338 (td, 0.7 H, H-2dα), 2.975 (dd, 1 H, $J_{3eq,3ax}$ 12.6 Hz, H-3c*eq*), 2.246, 2.171, 2.138, 2.134, 2.109, 2.104, 2.101, 2.098, 2.071, 2.067, 2.028, 1.995, 1.991, 1.832, and 1.828 (15 s, 36 H, 11 OAc, NAc), 1.725 (t, 1 H, H-3c*ax*), and 1.190 (s, 9 H, *t*-Bu).

Conversion of **22α** *and* **22β** *into the hemiacetal* **24**.—A mixture of **22α** and **22β** (33 mg) was converted into **24** (22 mg, 70%) as described for the synthesis of **23**. Compound **24** had $[\alpha]_D$ +14.4° (*c* 0.88); R_f 0.39 and 0.36 in 19:1 $CHCl_3$–MeOH; NMR: δ_H 6.237 (d, 0.28 H, $J_{NH,2}$ 7.0 Hz, NHα*trans*), 6.209 (d, 0.42 H, $J_{NH,2}$ 7.0 Hz, NHα*cis*), 6.113 (d, 0.12 H, $J_{NH,2}$ 7.3 Hz, NHβ*trans*), 6.007 (d, 0.18 H, $J_{NH,2}$ 7.0 Hz, NHβ*cis*), 5.914 (dd, 0.28 H, $J_{3,4}$ 3.7, $J_{3,2}$ 11.4 Hz, H-3dα*trans*), 5.868 (dd, 0.42 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.0 Hz, H-3dα*cis*), 5.846 (dd, 0.12 H, $J_{3,4}$ 3.7, $J_{3,2}$ 11.0 Hz, H-3dβ*trans*), 5.792 (dd, 0.18 H, $J_{3,4}$ 3.7, $J_{3,2}$ 11.4 Hz, H-3dβ*cis*), 5.392 (d, 0.7 H, $J_{1,2}$ 3.7 Hz, H-1aα), 5.360 (br s, 1 H, H-4d), 5.155 (m, 1 H, H-7c), 4.975 (m, 1 H, H-2b), 4.795 (d, 0.7 H, $J_{2,3}$ 10.3 Hz, H-2aα), 4.585 (d, 0.4 H, $J_{1,2}$ 7.7 Hz, H-1b*trans*), 4.575 (d, 0.6 H, $J_{1,2}$ 7.7 Hz, H-1b*cis*), 3.886 and 3.876 (2s, 3 H, OMe), 3.554 (m, 1 H, H-4b), 3.375 (m, 1 H, H-2d), 2.931 (m, 1 H, H-3c*eq*), 2.816 (m, 1.2 H, cyclohexane ring H-1,2*cis*), 2.709 (q, 0.4 H, *J* 7.3 Hz, cyclohexane ring H-1 or 2*trans*), 2.638 (q, 0.4 H, *J* 6.6 Hz, cyclohexane ring H-1 or 2*trans*), 2.214, 2.196, 2.163, 2.148, 2.131, 2.096, 2.092, 2.083, 2.058, 2.055, 2.030, 2.020, 2.014, 1.999, 1.986, 1.971, 1.966, and 1.952 (18 s, 36 H, 11 OAc, NAc), 1.646 (m, 1 H, H-3c*ax*), and 1.187 (s, 9 H, *t*-Bu).

Conversion of **23** *into the trichloroacetimidate* **25**.—A solution of **23** (13 mg, 7.7 μmol), CCl_3CN (10 μL, 98 μmol), and DBU (1.8 μL, 12 μmol) in $(ClCH_2)_2$ (0.20 mL) was stirred for 3 h at 0°C. The mixture was directly chromatographed on silica gel (1.0 g) with 3:2 toluene–THF to give **25** (14 mg, 99%); $[\alpha]_D$ +40.2° (*c* 0.92); R_f 0.50 in 11:9 toluene–THF; NMR: δ_H 8.650(s, 1 H, C=NH), 7.818 (m, 2 H, aromatic), 7.735 (m, 2 H, aromatic), 6.522 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1a), 6.020 (d, 1 H, $J_{NH,2}$ 7.3 Hz, NH), 5.842 (dd, 1 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.0 Hz, H-3d), 5.573 (m, 2 H, H-8c, H-3a), 5.495 (ddd, 1 H, $J_{4,3eq}$ 4.8, $J_{4,5}$ 10.3, $J_{4,3ax}$ 12.1 Hz, H-4c), 5.366 (d, 1 H, H-4d), 5.224 (dd, 1 H, $J_{7,6}$ 2.9, $J_{7,8}$ 9.9 Hz, H-7c), 5.133 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1d), 5.076 (dd, 1 H, $J_{2,3}$ 10.3 Hz, H-2a), 5.016 (dd, 1 H, $J_{2,1}$ 7.7, $J_{2,3}$ 9.9 Hz, H-2b), 4.875 (dd, 1 H, $J_{6,5}$ 11.0 Hz, H-6c), 4.659 (d, 1 H, H-1b), 4.336 (br dd, 1 H, $J_{3,4}$ 2.6 Hz, H-3b), 4.267 (dd, 1 H, $J_{9,8}$ 5.5, $J_{9,9'}$ 11.4 Hz, H-9c), 4.102 (t, 1 H, H-5c), 3.929 (s, 3 H, OMe), 3.591 (br.d, 1 H, H-4b), 3.415 (td, 1 H, H-2d), 2.961 (dd, 1 H, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq*), 2.260, 2.173, 2.142, 2.110, 2.093, 2.073, 2.071, 2.029, 2.014, 1.992, 1.943, and 1.831 (12 s, 36 H, 11 OAc, NAc), 1.729 (t, 1 H, H-3c*ax*), and 1.141 (s, 9 H, *t*-Bu).

Conversion of **24** *into the trichloroacetimidate* **26**.—Compound **24** (22 mg) was converted into **26** (23 mg, 94%) as described for the synthesis of **25**. Compound **26** had $[\alpha]_D$ +26.3° (*c* 0.76); R_f 0.35 in 13:7 toluene–THF; NMR: δ_H 8.646 (s, 1 H, C=NH), 6.518 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1a), 5.981 (d, 0.4 H, $J_{NH,2}$ 7.3 Hz, NH*trans*), 5.948 (d, 0.6 H, $J_{NH,2}$ 7.3 Hz, NH*cis*), 5.856 (dd, 0.4 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.4 Hz, H-3d*trans*), 5.797 (dd, 0.6 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.0 Hz, H-3d*cis*), 5.568 (m, 1.6 H, H-3a, H-8c*cis*), 5.479 (td, 0.4 H, $J_{8,9}=J_{8,9'}=3.3$, $J_{8,7}$ 9.5 Hz, H-8c*trans*), 5.359 (br s, 1 H, H-4d), 5.153 (dd, 0.6 H, $J_{7,6}$ 2.9, $J_{7,8}$ 9.9 Hz, H-7c*cis*), 5.121 (dd, 0.4 H, $J_{7,6}$ 2.2 Hz, H-7c*trans*), 5.098 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1d), 5.072 (dd, 1 H, $J_{2,3}$ 10.3 Hz, H-2a), 4.989 (dd, 0.4 H, $J_{2,1}$ 7.3, $J_{2,3}$ 9.5 Hz, H-

2b*trans*), 4.984 (dd, 0.6 H, $J_{2,1}$ 7.7, $J_{2,3}$ 10.3 Hz, H-2b*cis*), 4.793 (dd, 0.4 H, $J_{6,5}$ 10.6 Hz, H-6c*trans*), 4.786 (dd, 0.6 H, $J_{6,5}$ 10.6 Hz, H-6c*cis*), 4.633 (d, 0.4 H, H-1b*trans*), 4.623 (d, 0.6 H, H-1b*cis*), 4.317 (br dd, 0.4 H, $J_{3,4}$ 2.6 Hz, H-3b*trans*), 4.275 (br.dd, 0.6 H, $J_{3,4}$ 2.6 Hz, H-3b*cis*), 3.890 (s, 1.8 H, OMe*cis*), 3.880 (s, 1.2 H, OMe*trans*), 3.563 (br s, 1 H, H-4b), 3.435 (td, 0.6 H, H-2d*cis*), 3.379 (td, 0.4 H, H-2d*trans*), 2.936 (dd, 0.6 H, $J_{3eq,4}$ 4.8, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq-cis*), 2.900 (dd, 0.4 H, $J_{3eq,4}$ 5.1, $J_{3eq,3ax}$ 13.2 Hz, H-3c*eq-trans*), 2.815 (m, 1.2 H, cyclohexane ring H-1,2*cis*), 2.709 (q, 0.4 H, *J* 7.7 Hz, cyclohexane ring H-1 or 2*trans*), 2.638 (q, 0.4 H, *J* 6.6 Hz, cyclohexane ring H-1 or 2*trans*), 2.356, 2.230, 2.211, 2.166, 2.151, 2.138, 2.099, 2.077, 2.066, 2.062, 2.022, 2.015, 2.000, 1.994, 1.989, 1.967, and 1.953 (17 s, 36 H, 11 OAc, NAc), 1.646 (m, 1 H, H-3c*ax*), and 1.139 (s, 9 H, *t*-Bu).

Glycosylation of **25** *with* **2** *to give* **27**.—To a mixture of **25** (14 mg, 8.5 μmol), **2** [4] (13 mg, 17.1 μmol), and powdered 4A molecular sieves (51 mg) in $CHCl_3$ (0.70 mL) was added $BF_3 \cdot OEt_2$ (2.1 μL, 17.1 μmol) at −18°C under Ar. The mixture was stirred for 2 h at 0°C, then diluted with $CHCl_3$ (40 mL) and filtered through Celite. The filtrate was washed with aq $NaHCO_3$ and aq NaCl, dried ($MgSO_4$), and evaporated in vacuo. The residue was chromatographed on silica gel (3.0 g) with 39:1 $CHCl_3$–MeOH to give **27** (6.4 mg, 34%); $[\alpha]_D$ +17.8° (*c* 0.39); R_f 0.34 in 13:7 toluene–THF; NMR: δ_H 7.993 (dd, 2 H, *J* 1.5, *J* 7.0 Hz, $C_6H_5C{=}O$), 7.819 (m, 2 H, aromatic), 7.735 (m, 2 H, aromatic), 7.541 (tt, 1 H, *J* 1.5, 7.3 Hz, $C_6H_5C{=}O$), 7.422 (t, 2 H, *J* 7.7 Hz, $C_6H_5C{=}O$), 6.236 (d, 1 H, $J_{NH,2}$ 7.3 Hz, NH-2d), 5.869 (td, 1 H, $J_{5,6} = J_{5,6'} = 7.0$, $J_{4,5}$ 15.4 Hz, H-5cer), 5.847 (dd, 1 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.4 Hz, H-3d), 5.756 (d, 1 H, $J_{NH,2}$ 9.5 Hz, NHcer), 5.569 (td, 1 H, $J_{8,9} = J_{8,9'} = 3.3$, $J_{8,7}$ 9.5 Hz, H-8c), 5.544 (t, 1 H, $J_{3,2} = J_{3,4} = 7.3$ Hz, H-3cer), 5.460 (dd, 1 H, H-4cer), 5.360 (d, 1 H, H-4d), 5.234 (dd, 1 H, $J_{7,6}$ 2.6 Hz, H-7c), 5.185 (t, 1 H, $J_{3,2} = J_{3,4} = 9.5$ Hz, H-3a), 5.125 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1d), 4.963 (dd, 1 H, $J_{2,1}$ 7.7, $J_{2,3}$ 10.3 Hz, H-2b), 4.926 (dd, 1 H, $J_{2,1}$ 7.7 Hz, H-2a), 4.878 (dd, 1 H, $J_{6,5}$ 10.6 Hz, H-6c), 4.574 (d, 1 H, H-1b), 4.456 (m, 1 H, H-2cer), 4.419 (d, 1 H, H-1a), 4.321 (br dd, 1 H, $J_{3,4}$ 2.6 Hz, H-3b), 4.095 (t, 1 H, $J_{5,4} = J_{5,6}$ 10.5 Hz, H-5c), 3.923 (s, 3 H, OMe), 3.829 (t, 1 H, $J_{4,3} = J_{4,5} = 10.5$ Hz, H-4a), 3.600 (dd, 1 H, $J_{1,2}$ 4.0, $J_{1,1'}$ 9.9 Hz, H-1cer), 3.577 (br d, 1 H, H-4b), 3.377 (td, 1 H, H-2d), 2.947 (dd, 1 H, $J_{3eq,4}$ 4.8, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq*), 2.210, 2.173, 2.132, 2.124, 2.091, 2.057, 2.027, 1.984, 1.976, 1.947, 1.893 and 1.828 (12 s, 36 H, 11 OAc, NAc), 1.714 (t, 1 H, H-3c*ax*), 1.151 (s, 9 H, *t*-Bu), 0.880 (t, 3 H, *J* 7.3 Hz, CH_2CH_3), and 0.876 (t, 3 H, *J* 7.0 Hz, CH_2CH_3).

Glycosylation of **26** *with* **2** *to give* **28**.—Compound **26** (22 mg) was coupled with **2** to give **28** (9.4 mg, 32%) as described for the synthesis of **27**. Compound **28** had $[\alpha]_D$ +3.9° (*c* 0.52); R_f 0.40 in 13:7 toluene–THF; NMR: δ_H 7.991 (d, 2 H, *J* 7.3 Hz, $C_6H_5C{=}O$), 7.542 (t, 1 H, *J* 7.7 Hz, $C_6H_5C{=}O$), 7.424 (t, 2 H, *J* 7.7 Hz, $C_6H_5C{=}O$), 6.183 (d, 0.4 H, $J_{NH,2}$ 7.7 Hz, NH-2d*trans*), 6.144 (d, 0.6 H, $J_{NH,2}$ 7.3 Hz, NH-2d*cis*), 5.867 (td, 1 H, $J_{5,6} = J_{5,6'} = 6.6$, $J_{4,5}$ 15.0 Hz, H-5cer), 5.797 (dd, 0.6 H, $J_{3,4}$ 3.3, $J_{3,2}$ 11.0 Hz, H-3d*cis*), 5.750 (d, 1 H, $J_{NH,2}$ 9.2 Hz, NHcer), 5.542 (t, 1 H, $J_{3,2} = J_{3,4} = 7.5$ Hz, H-3cer), 5.459 (dd, 1 H, H-4cer), 5.429 (m, 1 H, H-4c), 5.353 (br d, 1 H, $J_{4,3}$ 2.6 Hz, H-4d), 5.173 (t, 1 H, $J_{3,2} = J_{3,4} = 9.2$ Hz, H-3a), 5.089 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1d), 4.928 (m, 2 H, H-2a, H-2b), 4.799 (dd, 0.4 H, $J_{6,7}$ 2.6, $J_{6,5}$ 11.0 Hz, H-6c*trans*), 4.790 (dd, 0.6 H, $J_{6,7}$ 2.9, $J_{6,5}$ 10.6 Hz, H-6c*cis*), 4.554 (d, 0.4 H, $J_{1,2}$ 7.0 Hz, H-1b*trans*), 4.552 (d, 0.6 H, $J_{1,2}$ 8.1 Hz, H-1b*cis*), 4.454 (m, 1 H, H-2cer), 4.413 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1a), 4.301 (br d, 1 H, $J_{3,2}$ 10.3 Hz,

H-3b), 3.884 (s, 1.8 H, OMe*cis*), 3.875 (s, 1.2 H, OMe*trans*), 3.818 (t, 1 H, $J_{4,3}=J_{4,5}=9.7$ Hz, H-4a), 3.596 (dd, 1 H, $J_{1,2}$ 3.9, $J_{1,1'}$ 10.1 Hz, H-1cer), 3.551 (br s, 1 H, H-4b), 3.507 (m, 1 H, H-5a), 3.379 (m, 1 H, H-2d), 2.920 (dd, 0.6 H, $J_{3eq,4}$ 4.4, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq-cis*), 2.881 (dd, 0.4 H, $J_{3eq,4}$ 5.1, $J_{3eq,3ax}$ 12.8 Hz, H-3c*eq-trans*), 2.815 (m, 1.2 H, cyclohexane ring H-1, 2*cis*), 2.709 (q, 0.4 H, *J* 7.7 Hz, cyclohexane ring H-1 or 2*trans*), 2.637 (q, 0.4 H, *J* 6.6 Hz, cyclohexane ring H-1 or 2*trans*), 2.175, 2.164, 2.160, 2.156, 2.151, 2.128, 2.112, 2.074, 2.048, 2.046, 2.020, 2.006, 1.980, 1.962, 1.956, 1.954, 1.948, 1.889, and 1.885 (19 s, 36 H, 11 OAc, NAc), 1.147 (s, 9 H, *t*-Bu). 0.879 (t, 3 H, *J* 6.5 Hz, CH_2CH_3), and 0.878 (t, 3 H, *J* 7.0 Hz, CH_2CH_3).

Cleavage of methyl ester of **27** *to give* **29**.—A solution of **27** (4.0 mg, 1.8 μmol) in pyridine (0.20 mL) was added dropwise onto LiI (3.5 mg, 26 μmol, dried at 90°C for 21 h in vacuo), and the mixture was heated for 2 h at reflux under Ar. The crude mixture was chromatographed over Sephadex LH-20 (5.0 mL) with MeOH and then on silica gel (0.60 g) with 9:1 $CHCl_3$–MeOH to give **29** (3.2 mg, 80%); $[\alpha]_D$ +19.0° (*c* 0.21); R_f 0.53 in 17:3 $CHCl_3$–MeOH; NMR: δ_H (CD_3OD) 7.999 (d, 2 H, *J* 7.3 Hz, $C_6H_5C{=}O$), 7.862 (m, 2 H, aromatic), 7.806 (m, 2 H, aromatic), 7.591 (t, 1 H, *J* 7.3 Hz, $C_6H_5C{=}O$), 7.460 (t, 2 H, *J* 7.7 Hz, $C_6H_5C{=}O$), 5.872 (td, 1 H, $J_{5,6}=J_{5,6'}=7.3$, $J_{4,5}$ 15.0 Hz, H-5cer), 5.726 (dt, 1 H, $J_{4,3eq}$ 5.1, $J_{4,3ax}=J_{4,5}=11.0$ Hz, H-4c), 5.561 (t, 1 H, $J_{3,2}=J_{3,4}=6.8$ Hz, H-3cer), 5.507 (dd, 1 H, H-4cer), 5.311 (d, 1 H, $J_{4,3}$ 2.9 Hz, H-4d), 2.872 (dd, 1 H, $J_{3eq,3ax}$ 11.0 Hz, H-3c*eq*), 2.152, 2.146, 2.143, 2.110, 2.062, 2.038, 1.998, 1.987, 1.974, 1.912 (6 H), and 1.778 (11 s, 36 H, 11 OAc, NAc), 1.580 (t, 1 H, H-3c*ax*), 1.152 (s, 9 H, *t*-Bu), 0.896 (t, 3 H, *J* 7.3 Hz, CH_2CH_3), and 0.894 (t, 3 H, *J* 7.0 Hz, CH_2CH_3).

Cleavage of methyl ester of **28** *to give* **30**.—Compound **28** (5.9 mg) was converted into **30** (5.8 mg, 99%) as described for the synthesis of **29**. Compound **30** had $[\alpha]_D$ +12.9° (*c* 0.39); R_f 0.48 in 17:3 $CHCl_3$–MeOH; NMR: δ_H (CD_3OD) 7.997 (d, 2 H, *J* 8.0 Hz, $C_6H_5{=}O$), 7.592 (t, 1 H, *J* 7.3 Hz, $C_6H_5C{=}O$), 7.460 (t, 2 H, *J* 7.7 Hz, $C_6H_5C{=}O$), 5.880 (td, 1 H, $J_{5,6}=J_{5,6'}=6.2$, $J_{4,5}$ 15.4 Hz, H-5cer), 5.664 (m, 1 H, H-4c), 5.560 (t, 1 H, $J_{3,2}=J_{3,4}=7.7$ Hz, H-3cer), 5.497 (dd, 1 H, H-4cer), 5.434 (m, 0.6 H, H-8c*cis*), 5.348 (m, 0.4 H, H-8c*trans*), 5.305 (br d, 1 H, $J_{4,3}$ 2.6 Hz, H-4d), 4.635 (d, 2 H, $J_{1,2}$ 8.1 Hz, H-1a, H-1b), 2.166, 2.140, 2.129, 2.101, 2.073, 2.032, 2.029, 2.019, 2.001, 1.996, 1.965, 1.961, 1.934, and 1.914 (14 s, 36 H, 11 OAc, NAc), 1.149 (s, 9 H, *t*-Bu), 0.897 (t, 3 H, *J* 6.4 Hz, CH_2CH_3), and 0.895 (t, 3 H, *J* 6.4 Hz, CH_2CH_3).

O-(Sodium 5-amino-3,5-dideoxy-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2→3)*-O-*[2-acetamido-2-deoxy-β*-D-*galactopyranosyl-(1→4)]*-O-*β*-D-*galactopyranosyl-(1→4)*-O-*β*-D-*glucopyranosyl-(1→1)-(2S,3R,4E)-2*-N-*tetracosanoylsphingenine* (**1**).—*Procedure A*. A solution of **29** (1.7 mg, 0.8 μmol) in 40% $MeNH_2$–MeOH (0.40 mL) was stirred for 50 h at 20°C, and then evaporated in vacuo. After addition of N NaOH (16 μL), the crude mixture was purified by chromatography over Sephadex LH-20 (10 mL) with 5:4:1 $CHCl_3$–MeOH–H_2O to give **1** (0.7 mg, 63%).

Procedure B. A solution of **30** (2.8 mg, 1.3 μmol) in EtOH (0.20 mL) was added $MeNHNH_2$ (0.40 mL), and stirred for 48 h at 75°C, and then evaporated in vacuo. The residue was purified by HPLC with 5:4:1 $CHCl_3$–MeOH–H_2O. After addition of N NaOH (13 μL), the crude mixture was chromatographed over Sephadex LH-20 (10 mL) with 5:4:1 $CHCl_3$–MeOH–H_2O to give 1 (0.8 mg, 44%).

Compound **1** had $[\alpha]_D$ −8.8° (*c* 0.05, pyridine); R_f 0.29 in 5:4:1 $CHCl_3$–MeOH–H_2O; mass spectrum (negative ion mode, triethanolamine matrix): *m/z* 1176 $(M-NeuNH_2-H)^-$, 1425 $(M-H)^-$; NMR: δ_H (1:1 $CDCl_3$–CD_3OD) 5.692 (td, 1 H, $J_{5,6}=J_{5,6'}=6.6$, $J_{4,5}$ 15.4 Hz, H-5cer), 5.455 (dd, 1 H, $J_{4,3}$ 7.3 Hz, H-4cer), 4.844 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1d), 4.407 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1b), 4.291 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1a), 4.095 (t, 1 H, $J_{3,2}=J_{3,4}=7.7$ Hz, H-3cer), 2.778 (t, 1 H, $J_{5,4}=J_{5,6}=9.7$ Hz, H-5c), 2.681 (dd, 1 H, $J_{3eq,4}$ 4.8, $J_{3eq,3ax}$ 12.5 Hz, H-3c*eq*), 2.176 (t, 2 H, *J* 7.7 Hz, $NHCOCH_2$), 2.037 (s, 3 H, NAc), 1.829 (t, 1 H, $J_{3ax,4}=J_{3ax,3eq}=12.1$ Hz, H-3c*ax*), and 0.891 (t, 6 H, *J* 6.8 Hz, 2 CH_2CH_3).

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