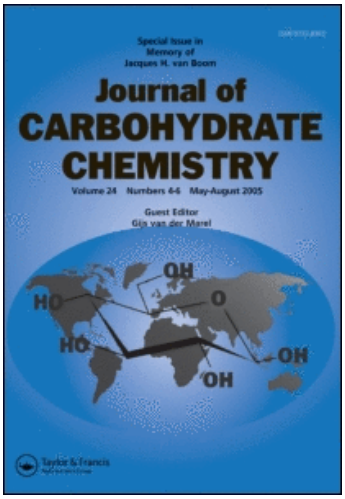

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Synthetic Studies on Sialoglycoconjugates 104: Synthesis of Kdn-Lewis X Ganglioside Analogs Containing Modified Reducing Terminal and L-Rhamnose in Place of L-Fucose

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SYNTHETIC STUDIES ON SIALOGLYCOCONJUGATES 104: SYNTHESIS OF KDN-LEWIS X GANGLIOSIDE ANALOGS CONTAINING MODIFIED REDUCING TERMINAL AND L-RHAMNOSE IN PLACE OF L-FUCOSE[1,2]

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ABSTRACT

KDN-Le^X ganglioside analogs (**10**, **13**, **16** and **19**) containing the modified reducing terminal and L-rhamnose in place of L-fucose have been synthesized. Glycosidation of methyl 2,3,4-tri-*O*-benzyl-1-thio-α-L-rhamnopyranoside (**1**) with 2-(trimethylsilyl)ethyl *O*-(2-acetamido-4,6-*O*-benzylidene-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (**2**), followed by reductive ring opening of the benzylidene acetal, gave 2-(trimethylsilyl)ethyl *O*-(2,3,4-tri-*O*-benzyl-α-L-rhamnopyranosyl)-(1→3)-*O*-(2-acetamido-6-*O*-benzyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (**4**). The tetrasaccharide **4** was coupled with methyl *O*-(methyl 4,5,7,8,9-penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-2,4,6-tri-*O*-benzoyl-1-thio-β-D-galactopyranoside (**5**), using dimethyl(methylthio)sulfonium triflate (DMTST), to give the hexasaccharide **6**, which was converted into compound **11** in the usual manner. Compounds **8** and **11** were transformed, *via* bromination of the reducing terminal, radical reduction, *O*-deacylation and saponification of the methyl ester, into the desired KDN-Le^X hexasaccharides (**10**, **13**). On the other hand, glycosylation of 2-

(tetradecyl)hexadecanol with α-trichloroacetimidates **14** and **17**, afforded the target ganglioside analogs **16** and **19**.

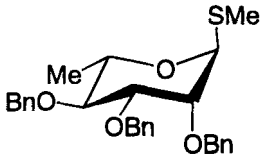
INTRODUCTION

3-Deoxy-D-*glycero*-D-*galacto*-2-nonulopyranosylonic acid (KDN), in which the acetamido group at C-5 of *N*-acetylneuraminic acid is replaced by a hydroxyl group, was first isolated[3] from rainbow trout eggs. Now a number of KDN-glycoconjugates have been reported[4] to occur in various living organisms ranging from bacterial to mammalian species. A unique feature of KDN-containing glycoconjugate is their complete resistance to the action of the known bacterial and viral exosialidases.[5]

We have systematically synthesized[6-9] various types of KDN-gangliosides and demonstrated[10,11] that the KDN-Lewis X ganglioside showed potent competitive inhibition activity against the selectin bindings with sialyl-Lewis X ganglioside. It has also been found that the deoxygenation[12] or 2-(tetradecyl)hexadecyl glycoside formation[13,14] at the reducing terminal is effective for increasing the affinity to selectins. In addition, the sialyl Le^x analogs containing the 2-epi- or 4-epi-L-fucose have been shown to be potent inhibitors[11] for selectin binding. As a part of our continuing efforts to elucidate the structure-function relationships of sialyl Le^x, we here describe the synthesis of novel KDN-Lewis X ganglioside analogs which contain 1,5-anhydro-D-glucitol and 2-(tetradecyl)hexadecyl β-D-glucopyranoside at the reducing terminal, and L-rhamnose (2,4-di-epi-L-fucose) in place of L-fucose.

RESULTS AND DISCUSSION

For the synthesis of the desired KDN-Lewis X ganglioside analogs which contain a modified reducing terminal, we prepared two kinds of key compounds; one is *O*-(methyl 4,5,7,8,9-penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2 → 3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl)-(1→ 3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glu-



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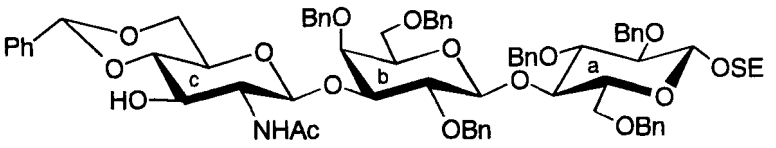

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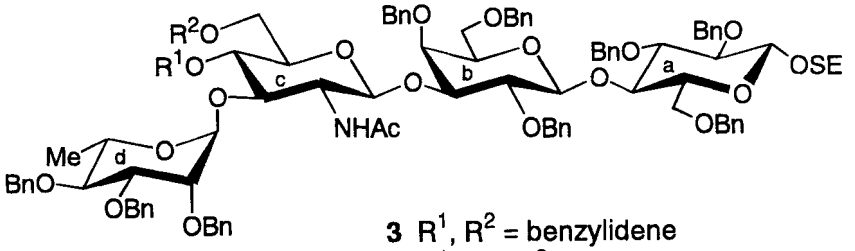
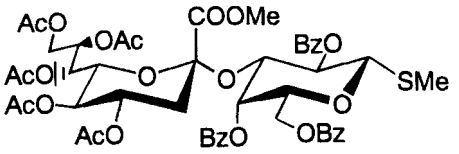


3 R^1, R^2 = benzylidene
4 R^1 = H, R^2 = Bn



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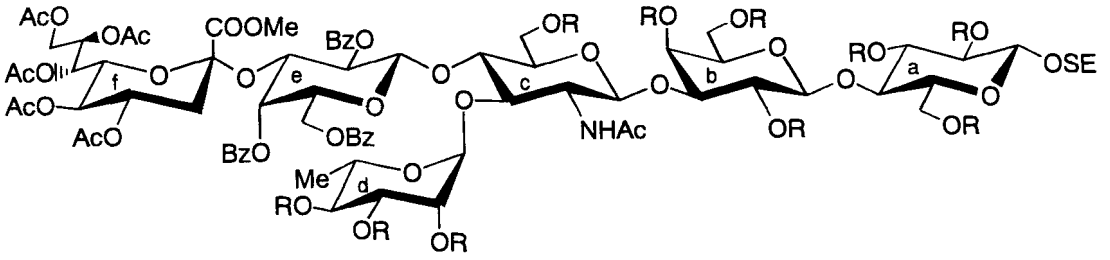


6 R = Bn
7 R = Ac

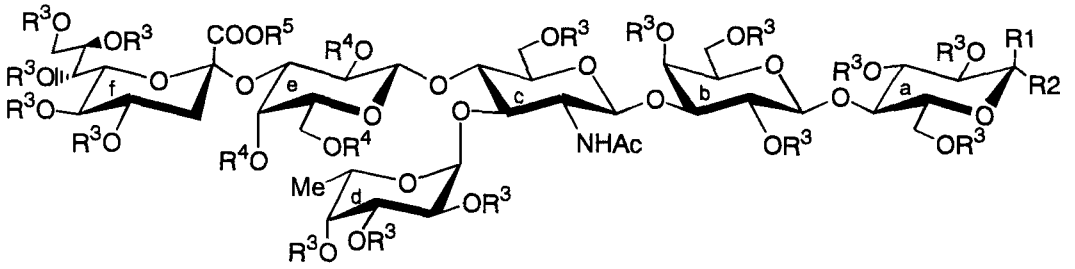
copyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-D-glucopyranose[7] (**8**), and the other *O*-(methyl 4,5,7,8,9-penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl)-(1→3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-D-glucopyranose (**11**).

To prepare compound **11** we employed the methylthio glycosides of rhamnose[15] (**1**) and KDN-α(2→3)-galactose[7] (**5**) as the glycosyl donors and the trisaccharide **2**[16] as a suitably protected glycosyl acceptor. Glycosylation[17] of **2** with **1**, in dry benzene in the presence of DMTST and molecular sieves 4Å (MS-4Å) for 5 h at 7 °C, gave exclusively the α-glycoside **3** in 85% yield; significant signals of the rhamnose residue in the ^{1}H NMR spectrum were a three-proton doublet at δ 1.33 ($J_{5,6}$ = 6.0 Hz, H-6) and one-proton doublet at δ 4.83 ($J_{1,2}$ = 1.2 Hz, H-1), indicating the structure assigned. Reductive ring-opening[18] of the benzylidene acetal in **3** with sodium cyanoborohydride-hydrogen chloride in dry ether afforded compound **4** as a syrup in 80% yield.

Glycosylation[7,17] of **4** with **5** in dichloromethane for 48 h at 7 °C in the presence of DMTST (4.0 equiv with respect to the glycosyl donor) and powdered MS-4Å gave the expected hexasaccharide derivative **6** in 62% yield; significant signals of KDN-Gal unit in the ^{1}H NMR spectrum of **6** [δ 4.88 (d, $J_{1,2}$ = 7.9 Hz, H-1e), 5.40 (broad d, $J_{3,4}$ = 2.8 Hz, H-4e), and 5.41 (dd, 1 H, $J_{2,3}$ = 8.3 Hz, H-2e)], supported the structure assigned.

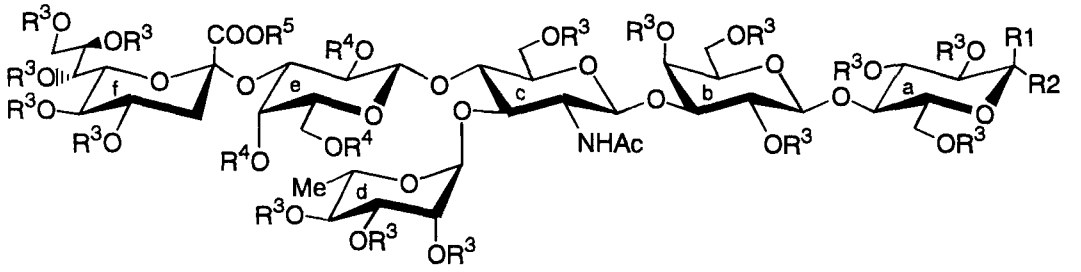
Catalytic hydrogenolysis (10% Pd-C) in ethanol-acetic acid of the benzyl groups in **6** and subsequent *O*-acetylation gave the per-*O*-acetyl derivative **7** (72%). Treatment of compound **7** with trifluoroacetic acid[19,20] in dichloromethane for 3 h at room temperature gave the corresponding 1-hydroxy compound **11** in quantitative yield.

Treatment[21] of **8** and **11** with carbon tetrabromide in dichloromethane in the presence of triphenylphosphine gave the 1-bromo derivatives, which were reduced[22] with tributyltin hydride in the presence of α,α′-azobis-isobutyronitrile (AIBN) in dry toluene to give the 1-deoxy derivatives **9** (72%) and **12** (70%). Significant signals in



8 R^1, R^2 = H, OH, R^3 = Ac, R^4 = Bz, R^5 = Me
9 $R^1 = R^2$ = H, R^3 = Ac, R^4 = Bz, R^5 = Me
10 $R^1 = R^2 = R^3 = R^4 = R^5$ = H

14 R^1 = $OC(=NH)CCl_3$, R^2 = H, R^3 = Ac, R^4 = Bz, R^5 = Me
15 R^1 = H, R^2 = B-30, OH, R^3 = Ac, R^4 = Bz, R^5 = Me
16 $R^1 = R^3 = R^4 = R^5$ = H, R^2 = B-30



11 R^1, R^2 = H, OH, R^3 = Ac, R^4 = Bz, R^5 = Me
12 $R^1 = R^2$ = H, R^3 = Ac, R^4 = Bz, R^5 = Me
13 $R^1 = R^2 = R^3 = R^4 = R^5$ = H

17 R^1 = $OC(=NH)CCl_3$, R^2 = H, R^3 = Ac, R^4 = Bz, R^5 = Me
18 R^1 = H, R^2 = B-30, OH, R^3 = Ac, R^4 = Bz, R^5 = Me
19 $R^1 = R^3 = R^4 = R^5$ = H, R^2 = B-30

B-30 = 2-(tetradecyl)hexadecyl

the ^{1}H NMR spectra were at δ 3.19 (t, $J_{gem} = J_{1ax,2}$ = 10.6 Hz, H-1ax) for **9** and δ 3.23 (t, $J_{gem} = J_{1ax,2}$ = 10.9 Hz, H-1ax) for **12**, which supported the structure assigned.

Treatment[23,24] of **11** with trichloroacetonitrile in dichloromethane in the presence of 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) for 2 h at 0 °C gave the α-trichloroacetimidate **17** (94%) (yield after column chromatography). Significant signals in the ^{1}H NMR spectrum were at δ 6.47 (d, $J_{1,2}$ = 3.7 Hz, H-1a) and 8.64 (C=NH) for **17**, which showed the imidate to be α. Glycosylation[23,25] of 2-(tetradecyl)hexadecanol with **14**[7] and **17** in dichloromethane in the presence of boron trifluoride etherate and MS-4Å (AW-300) for 5 h at room temperature gave the corresponding β-glycosides **15** (47%) and **18** (49%), respectively; the ^{1}H NMR data for the Glc unit [δ 4.50~4.60 (d, $J_{1,2}$ = 7.7~7.9 Hz, H-1a)] indicated the glycosidic linkages to be β.

Finally, *O*-deacylations of **9**, **12**, **15** and **18** with sodium methoxide in methanol, with subsequent saponification of the sialate methyl ester group, furnished the desired KDN-Lewis X ganglioside analogs **10**, **13**, **16** and **19** in high yields.

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 270 MHz with a Jeol JNM-GX 270 spectrometer. Preparative chromatography was performed on silica gel (Wako Chemical Co., 200 mesh) with the solvent systems specified. Concentrations were conducted *in vacuo*.

2-(Trimethylsilyl)ethyl *O*-(2,3,4-Tri-*O*-benzyl-α-L-rhamnopyranosyl)-(1→3)-*O*-(2-acetamido-4,6-*O*-benzylidene-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (3). To a solution of 2-(trimethylsilyl)ethyl *O*-(2,6-di-*O*-benzyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-

benzyl-β-D-glucopyranoside[16] (**2**, 1.0 g, 0.79 mmol) and methyl 2,3,4-tri-*O*-benzyl-1-thio-β-L-rhamnopyranoside[15] (**1**, 547 mg, 1.2 mmol) in benzene (20 mL) was added molecular sieves 4Å (MS-4Å, 4 g), and the mixture was stirred for 12 h at room temperature then cooled to 7 °C. To the cooled mixture were added, with stirring, dimethyl(methylthio)sulfonium triflate (DMTST; 914 mg, 3.54 mmol) and MS-4Å (365 mg), and the stirring was continued for 5 h at 7 °C. The precipitate was filtered off and washed thoroughly with CH_2Cl_2. The combined filtrate and washings were successively washed with M Na_2CO_3 and water, dried (Na_2SO_4) and concentrated. Column chromatography (1:3 EtOAc-hexane) of the residue on silica gel (300 g) gave **3** (1.13 g, 84.6%) as an amorphous mass; $[\alpha]_D$ -27.6° (*c* 0.6, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 1.00 (m, 2H, $Me_3SiCH_2CH_2$), 1.33 (d, 3H, $J_{5,6}$ = 6.1 Hz, H-6d), 1.40 (s, 3H, AcN), 4.83 (d, 1H, $J_{1,2}$ = 1.2 Hz, H-1d), 5.50 (s, H, C*H*Ph), and 7.09-7.25 (m, 50H, 10Ph).

Anal. Calcd for $C_{101}H_{115}NO_{20}Si$ (1691.1): C, 71.74; H, 6.85; N, 0.83. Found: C, 71.48; H, 6.62; N, 0.78.

2-(Trimethylsilyl)ethyl *O*-(2,3,4-Tri-*O*-benzyl-α-L-rhamnopyranosyl)-(1 → 3)-*O*-(2-acetamido-6-*O*-benzyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (4). To a solution of **3** (1.34 g, 0.79 mmol) in tetrahydrofuran (20 mL) was added MS-4Å (2 g), and the mixture was stirred for 5 h at room temperature. Sodium cyanoborohydride (2g, 31.8 mmol) was gradually added under a nitrogen atmosphere. After the reagent had dissolved, hydrogen chloride in ether was added at 0 °C until the evolution of gas ceased. TLC indicated that the reaction was complete after 5 min. The precipitate was filtered off and washed thoroughly with CH_2Cl_2. The combined filtrate and washings was successively washed with M Na_2CO_3 and water, dried (Na_2SO_4) and concentrated. Column chromatography (1:3 EtOAc-hexane) of the residue on silica gel (300 g) gave **4** (1.07 g, 80%) as an amorphous mass; $[\alpha]_D$ -5.4° (*c* 0.4, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 1.01 (m, 2H, $Me_3SiCH_2CH_2$), 1.32 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6d), 1.36 (s, 3H, AcN), 4.86 (d, 1H, $J_{1,2}$ = 1.1 Hz, H-1d), and 7.11-7.34 (m, 50H, 10Ph).

Anal. Calcd for $C_{101}H_{117}NO_{20}Si$ (1693.1): C, 71.65; H, 6.96; N, 0.83. Found: C, 71.62; H, 6.95; N, 0.61.

2-(Trimethylsilyl)ethyl *O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2 → 3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-benzyl-α-L-rhamnopyranosyl)-(1→3)]-*O*-(2-acetamido-6-*O*-benzyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)-(1 → 4)-2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (6). To a solution of methyl *O*-(methyl 4,5,7,8,9-penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-2,4,6-tri-*O*-benzoyl-1-thio-β-D-galactopyranoside[7] (**5**, 700 mg, 0.70 mmol) and **4** (793 mg, 0.47 mmol) in CH_2Cl_2 (6 mL) was added MS-4Å (2 g), and the mixture was stirred for 6 h at room temperature, then cooled to 0°C. A mixture of DMTST (725 mg, 0.28 mmol) and MS-4Å (295 mg) was added and the mixture was stirred for 48 h at 7 °C. The solids were collected and washed with CH_2Cl_2, and the combined filtrate was washed with M Na_2CO_3 and water, dried (Na_2SO_4), and concentrated. Column chromatography (2:3 EtOAc-hexane) of the residue on silica gel (200 g) gave **6** (767 mg, 62%) as an amorphous mass; $[\alpha]_D$ +4.8° (*c* 0.3, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 1.02 (m, 2H, $Me_3SiCH_2CH_2$), 1.17 (d, 3H, $J_{5,6}$ = 6.0 Hz, H-6d), 1.45-2.20 (6s, 18H, 5AcO, AcN), 2.51 (dd, 1H, J_{gem} = 12.5 Hz, $J_{3eq,4}$ = 4.6 Hz, H-3f*eq*), 3.87 (s, 3H, MeO), 4.88 (d, 1H, $J_{1,2}$ = 7.9 Hz, H-1e), 5.28 (dd, 1H, $J_{6,7}$ = 2.6 Hz, $J_{7,8}$ = 8.7 Hz, H-7f), 5.40 (br d, 1H, $J_{3,4}$ = 2.8 Hz, H-4e), 5.41 (dd, 1H, $J_{2,3}$ = 8.3 Hz, H-2e), 5.69 (m, 1H, H-8f), and 6.96-8.18 (m, 65H, 13Ph).

Anal. Calcd for $C_{148}H_{165}NO_{41}Si$ (2642.0): C, 67.28; H, 6.29; N, 0.53. Found: C, 67.05 H, 6.21; N, 0.40.

2-(Trimethylsilyl)ethyl *O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2 → 3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1 → 4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl)-(1 → 3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1 → 3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-β-D-glucopyranoside (7). A solution of **6** (596 mg,

0.23 mmol) in EtOH (60 mL) and acetic acid (10 mL) was hydrogenated in the presence of 10% Pd-C (1 g) for 4 days at 40 °C, then filtered and concentrated. The residue was acetylated with acetic anhydride (10 mL) and pyridine (20 mL) for 12 h at 40 °C. Column chromatography (50:1 CH_2Cl_2-MeOH) of the product on silica gel (50 g) gave **7** (351 mg, 72%) as an amorphous mass; $[\alpha]_D$ -1.1° (*c* 0.9, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.91 (m, 2H, $Me_3SiCH_2CH_2$), 1.15 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6d), 1.56-2.19 (16s, 48H, 15AcO, AcN), 2.40 (dd, 1H, J_{gem} = 12.4 Hz, $J_{3eq,4}$ = 4.4 Hz, H-3f*eq*), 3.77 (s, 3H, MeO), 5.25 (br d, 1H, $J_{3,4}$ = 3.3 Hz, H-4e), 5.49 (dd, 1H, $J_{1,2}$ = 8.2 Hz, $J_{2,3}$ = 9.1 Hz, H-2e), 5.70 (m, 1H, H-8f), 6.01 (d, 1H, $J_{2,NH}$ = 7.2 Hz, NH), and 7.44-8.17 (m, 15H, 3Ph).

Anal. Calcd for $C_{98}H_{125}NO_{51}Si$ (2161.1): C, 54.47; H, 5.83; N, 0.65. Found: C, 54.37; H, 5.77; N, 0.43.

***O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl)-(1→3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-1,5-anhydro-D-glucitol (9).** To a solution of *O*-(methyl 4,5,7,8,9-penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-fucopyransyl)-(1→3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-β-D-glucopyranose[7] (**8**, 100 mg, 48.5 μmol) in CH_2Cl_2, cooled to -15 °C, was added triphenylphosphine (31.8 mg, 0.12 mmol) and carbon tetrabromide (80 mg, 0.24 mmol). The reaction mixture was stirred for 12 h at room temperature then chromatographed directly on a column of silica gel (20 g) with 50 : 1 CH_2Cl_2-MeOH to give the 1-bromo compound. A solution of this compound in toluene (4 mL) was treated with tributyltin hydride (130 μL, 0.49 mmol) and AIBN (1.6 mg, 9.7 μmol) for 30 min under reflux and then concentrated. Column chromatography (50:1 CH_2Cl_2-MeOH) of the product on silica gel (50 g) gave **9** (351 mg, 72%) as an amorphous mass; $[\alpha]_D$ -14.0° (*c* 0.4, $CHCl_3$); ^1H NMR

($CDCl_3$) δ 1.20 (d, 3H, $J_{5,6}$ = 6.6 Hz, H-6d), 1.46-2.23 (16s, 48H, 5AcO, AcN), 2.45 (dd, 1H, J_{gem} = 11.8 Hz, $J_{3eq,4}$ = 4.8 Hz, H-3f*eq*), 3.19 (t, 1H, J_{gem} = $J_{1ax,2}$ = 10.6 Hz, H-1a*ax*), 3.87 (s, 3H, MeO), 5.33 (dd, 1H, $J_{1,2}$ = 8.0, $J_{2,3}$ = 9.9 Hz, H-2e), 5.37 (br d, 1H, $J_{3,4}$ = 3.0 Hz, H-4e), 5.70 (m, 1H, H-8f), 6.10 (d, 1H, $J_{2,NH}$ = 6.5 Hz, NH), and 7.38-8.18 (m, 15H, 3Ph).

Anal. Calcd for $C_{93}H_{113}NO_{50}$ (2044.9): C, 54.63; H, 5.57; N, 0.68. Found: C, 54.62; H, 5.37; N, 0.44.

***O*-(3-Deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic Acid)-(2→3)-*O*-(β-D-galactopyranosyl)-(1→4)-*O*-[α-L-fucopyranosyl-(1→3)]-*O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(β-D-galactopyraosyl)-(1→4)-1,5-anhydro-D-glucitol (10).** To a solution of **9** (50 mg, 24.5 μmol) in MeOH (5 mL) was added sodium methoxide (30 mg), and the mixture was stirred for 12 h at room temperature; the course of the reaction was monitored by TLC (4:2:1 butanol-ethanol-water). Sodium hydroxide (0.2 M, 5 mL) was added to the mixture, and this was stirred for 24 h at room temperature, neutralized with Dowex-50 (H^+) resin, and filtered. The resin was washed with methanol, and the combined filtrate and washings was concentrated to a syrup that was chromatographed on a column of Sephadex LH-20 (50 g) with MeOH to give **10** (24.2 mg, 91%) as an amorphous mass; $[\alpha]_D$ -4.8° (*c* 0.2, MeOH); ^1H NMR (DMSO-d_6) δ 0.95 (d, 3H, $J_{5,6}$ = 6.4 Hz, H-6d), 1.79 (s, 3H, AcN), 2.51 (dd, 1H, J_{gem} = 10.7 Hz, $J_{3eq,4}$ = 3.2 Hz, H-3f*eq*), 3.00 (t, 1H, J_{gem} = $J_{1ax,2}$ = 10.9 Hz, H-1a*ax*), and 5.08 (d, 1H, $J_{1,2}$ = 5.0 Hz, H-1d).

Anal. Calcd for $C_{41}H_{69}NO_{32}$ (1088.0): C, 45.26; H, 6.39; N, 1.29. Found: C, 44.98; H, 6.32; N, 1.20.

***O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl)-(1→3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-D-glucopyranose (11).** To a solution of **7** (300 mg, 0.14 mmol) in CH_2Cl_2 (4 mL)

cooled to 0 °C was added trifluoroacetic acid (6 mL), and the mixture was stirred for 3 h at room temperature. The reaction was monitored by TLC, and when it was complete ethyl acetate (10 mL) was added and the solution was concentrated. Column chromatography (20:1 CH_2Cl_2-MeOH) of the residue on silica gel (30 g) gave **11** (285 mg, quantitative) as an amorphous mass; ν 3400 (NH, OH), 1740 and 1230 (ester), 1680 and 1540 (amide), and 720 cm^{-1} (Ph).

Anal. Calcd for $C_{93}H_{113}NO_{51}$ (2060.9): C, 54.20; H, 5.53; N, 0.68. Found: C, 54.01; H, 5.32; N, 0.46.

***O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl)-(1→3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4, 6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)- 2,3,6-tri-*O*-acetyl-1,5-anhydro-D-glucitol (12).** Compound **11** (100 mg, 48.5 μmol) was deoxygenated as described for **9** to give **12** (69.5 mg, 70%) as an amorphous mass; $[\alpha]_D$ +12.2° (*c* 0.3, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 1.17 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6d), 1.57-2.20 (16s, 48H, 5AcO, AcN), 2.40 (dd, 1H, J_{gem} = 11.3 Hz, $J_{3eq,4}$ = 4.4 Hz, H-3f*eq*), 3.23 (t, 1H, J_{gem} = $J_{1ax,2}$ = 10.9 Hz, H-1a*ax*), 3.79 (s, 3H, MeO), 5.36 (dd, 1H, $J_{6,7}$ = 2.7, $J_{7,8}$ = 9.8 Hz, H-7f), 5.40 (br d, 1H, $J_{3,4}$ = 3.0 Hz, H-4e), 5.45 (dd, 1H, $J_{1,2}$ = 7.8 Hz, $J_{2,3}$ = 9.5 Hz, H-1e), 5.64 (m, 1H, H-8f), 6.01 (d, 1H, $J_{2,NH}$ = 6.3 Hz, NH), and 7.41-8.19 (m, 15H, 3Ph).

Anal. Calcd for $C_{93}H_{113}NO_{50}$ (2044.9): C, 54.63; H, 5.57; N, 0.68. Found: C, 54.33; H, 5.41; N, 0.44.

***O*-(3-Deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic Acid)-(2→3)-*O*-β-D-galactopyranosyl-(1→4)-*O*-[α-L-rhamnopyranosyl-(1→3)]-*O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(β-D-galactopyranosyl)-(1→4)-1,5-anhydro-D-glucitol (13).** Compound **12** (45 mg, 22.0 μmol) was treated with sodium methoxide and potassium hydroxide as described for **10** to give **13** (22.3 mg, 93%) as an amorphous mass; $[\alpha]_D$ -13.3° (*c* 0.2, MeOH); ^1H NMR (DMSO-d_6) δ 1.03 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6d), 1.79 (s, 3H, AcN), 2.55

(m, 1H, H-3f*eq*), 3.00 (t, 1H, $J_{gem} = J_{1ax,2} = 10.2$ Hz, H-1a*ax*), and 5.07 (d, 1H, $J_{1,2} = 5.0$ Hz, H-1d).

Anal. Calcd for $C_{41}H_{69}NO_{32}$ (1088.0): C, 45.26; H, 6.39; N, 1.29. Found: C, 45.05; H, 6.20; N, 1.10.

2-(Tetradecyl)hexadecyl *O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl)-(1→3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-β-D-glucopyranoside (15). To a solution of *O*-(methyl 4,5,7,8,9-penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl)-(1→3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-α-D-glucopyranosyl trichloroacetimidate[7] (**14**, 50.0 mg, 22.7 μmol) and 2-(tetradecyl)hexadecanol (20.0 mg, 45.6 μmol) in CH_2Cl_2 (1 mL) was added MS-4Å (500 mg), and the mixture was stirred for 6 h at room temperature then cooled to 0 °C. Boron trifluoride etherate (8.5 μL) was added to the mixture, and this was stirred for 8 h at room temperature. The precipitate was filtered off and washed with CH_2Cl_2. The filtrate and washings were combined, and the solution was successively washed with M Na_2CO_3 and water, dried (Na_2SO_4), and concentrated. Column chromatography (60:1 CH_2Cl_2-MeOH) of the residue on silica gel (20 g) gave **15** (26.4 mg, 47%) as an amorphous mass; $[\alpha]_D$ -1.1° (*c* 0.7, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.91 (t, 6H, 2C*H*$_3$CH$_2$), 1.15 (d, 3H, $J_{5,6} = 6.4$ Hz, H-6d), 1.25 (s, 52H, 26CH_2), 1.50-2.03 (16s, 48H, 15AcO, AcN), 2.44 (dd, 1H, $J_{gem} = 11.6$ Hz, $J_{3eq,4} = 4.8$ Hz, H-3f*eq*), 3.80 (s, 3H, MeO), 4.60 (d, 1H, $J_{1,2} = 7.7$ Hz, H-1a), 5.25 (br d, 1H, $J_{3,4} = 3.2$ Hz, H-4e), 5.68 (m, 1H, H-8f), 6.10 (d, 1H, $J_{2,NH} = 6.5$ Hz, NH), and 7.35-8.20 (m, 15H, 3Ph).

Anal. Calcd for $C_{123}H_{173}NO_{51}$ (2481.7): C, 59.53; H, 7.03; N, 0.56. Found: C, 59.44; H, 6.83; N, 0.56.

2-(Tetradecyl)hexadecyl *O*-(3-Deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic Acid)-(2→3)-*O*-(β-D-galactopyranosyl)-(1→4)-*O*-[α-L-fucopyranosyl-(1→3)]-*O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(β-D-galactopyranosyl)-(1→4)-β-D-glucopyranoside (16). Compound **15** (26.4 mg, 10.6 μmol) was treated with sodium methoxide and potassium hydroxide as described for **10** to give **16** (14.8 mg, 91%) after chromatography on a column of Sephadex LH-20 with (50 g) with 5:4:0.7 $CHCl_3$-MeOH-H_2O; amorphous mass; $[\alpha]_D$ -6.2° (*c* 0.3, 1:1 $CHCl_3$-MeOH) ^1H NMR (DMSO-*d*6): δ 0.84 (t, 6H, 2C*H*$_3$CH$_2$), 0.95 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6d), 1.22 (s, 52H, 26CH_2), 1.79 (s, 3H, AcN), 2.61 (m, 1H, H-3e) and 5.03 (d, 1H, $J_{1,2}$ = 5.0 Hz, H-1d).

Anal. Calcd for $C_{71}H_{129}NO_{33}$ (1524.8): C, 55.93; H, 8.53; N, 0.92. Found: C, 55.66; H, 8.37; N, 0.80.

***O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1→4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl)-(1→3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-α-D-glucopyranosyl Trichloroacetimidate (17).** To a stirred solution of **11** (100 mg, 48.5 μmol) in CH_2Cl_2 (2 mL) cooled to 0 °C were added trichloroacetonitrile (0.4 mL), DBU (8.1 mg), and Drierite (200 mg), then the mixture was stirred for 1 h at 0 °C and directly applied to a column of silica gel (20 g) eluted with 40:1 CH_2Cl_2-MeOH, to give **17** (101 mg, 94%) as an amorphous mass; $[\alpha]_D$ +22.3° (*c* 0.2, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 1.14 (d, 3H, $J_{5,6}$ = 6.1 Hz, H-6d), 1.82-2.20 (16s, 48H, 15AcO, AcN), 2.43 (dd, 1H, J_{gem} = 11.2 Hz, $J_{3eq,4}$ = 4.4 Hz, H-3f*eq*), 3.77 (s, 3H, MeO), 5.69 (m, 1H, H-8f), 6.04 (d, 1H, $J_{2,NH}$ = 7.2 Hz, NH), 6.47 (d, 1H, $J_{1,2}$ = 3.7 Hz, H-1a), 7.41-8.17 (m, 15H, 3Ph), and 8.64 (s, 1H, C=NH).

Anal. Calcd for $C_{95}H_{113}Cl_3N_2O_{51}$ (2205.3): C, 51.74; H, 5.16; N, 1.27. Found: C, 51.62; H, 4.94; N, 1.19.

2-(Tetradecyl)hexadecyl *O*-(Methyl 4,5,7,8,9-Penta-*O*-acetyl-3-deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2 →3)-*O*-(2,4,6-tri-*O*-benzoyl-β-D-galactopyranosyl)-(1 → 4)-*O*-[(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl)-(1 → 3)]-*O*-(2-acetamido-6-*O*-acetyl-2-deoxy-β-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-β-D-galactopyranosyl)-(1 →4)-2,3,6-tri-*O*-acetyl-β-D-glucopyranoside (18). Compound **17** (51.0 mg, 23.1 μmol) was condensed with 2-(tetradecyl)hexadecanol as described for **15** to give **18** (28.2 mg, 49%) as an amorphous mass; $[\alpha]_D$ -2.9° (*c* 0.6, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.91 (t, 6H, 2C*H*$_3$CH$_2$), 1.14 (d, 3H, $J_{5,6}$ = 6.0 Hz, H-6d), 1.26 (s, 52H, 26CH_2), 1.57-2.20 (16s, 48H, 15AcO, AcN), 2.46 (dd, 1H, J_{gem} = 11.5 Hz, $J_{3eq,4}$ = 4.7 Hz, H-3f*eq*), 3.78 (s, 3H, MeO), 4.54 (d, 1H, $J_{1,2}$ = 7.6 Hz, H-1a), 5.37 (br d, 1H, $J_{3,4}$ = 3.0 Hz, H-4e), 5.50 (dd, 1H, $J_{1,2}$ = 7.0 Hz, $J_{2,3}$ = 9.3 Hz, H-2e), 5.69 (m, 1H, H-8f), 5.98 (d, 1H, $J_{2,NH}$ = 6.8 Hz, NH), and 7.27-8.18 (m, 15H, 3Ph).

Anal. Calcd for $C_{123}H_{173}NO_{51}$ (2481.7): C, 59.53; H, 7.03; N, 0.56. Found: C, 59.28; H, 6.97; N, 0.40.

2-(Tetradecyl)hexadecyl *O*-(3-Deoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic Acid)-(2→3)-*O*-(β-D-galactopyranosyl)-(1 → 4)-*O*-[α-L-rhamnopyranosyl-(1→3)]-*O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1 → 3)-*O*-(β-D-galactopyranosyl)-(1 → 4)-β-D-glucopyranoside (19). Compound **18** (28.2 mg, 11.4 μmol) was treated with sodium methoxide and potassium hydroxide as described for **10** to give **19** (12.7 mg, 90%) after chromatography on a column of Sephadex LH-20 with 5:4:0.7 $CHCl_3$-MeOH-H_2O; amorphous mass; $[\alpha]_D$ -10.5° (*c* 0.1, 1:1 $CHCl_3$-MeOH); ^1H NMR (DMSO-d_6) δ 0.84 (t, 6H, 2C*H*$_3$CH$_2$), 1.03 (d, 3H, $J_{5,6}$ = 5.9 Hz, H-6d), 1.22 (s, 52H, 26CH_2), 1.79 (s, 3H, AcN), 2.61 (dd, 1H, J_{gem} = 10.4 Hz, $J_{3eq,4}$ = 3.9 Hz, H-3f*eq*) and 5.05 (br d, 1H, H-1d).

Anal. Calcd for $C_{71}H_{129}NO_{33}$ (1524.8): C, 55.93; H, 8.53; N, 0.92. Found: C, 55.88; H, 8.37; N, 0.66.

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