Tetrahedron Vol. 43, No. 24, pp. 5919 to 5928, 1987
Printed in Great Britain.

0040-4020/87 $3.00+.00

AN EFFECTIVE SYNTHESIS OF α-GLYCOSIDES OF N-ACETYLNEURAMINIC ACID DERIVATIVES BY USE OF 2-DEOXY-2β-HALO-3β-HYDROXY-4,7,8,9-TETRA-O-ACETYL-N-ACETYLNEURAMINIC ACID METHYL ESTER[1]

KAORU OKAMOTO,[†] TADAO KONDO,[††*] and TOSHIO GOTO[*]

Laboratory of Organic Chemistry, Faculty of Agriculture; and [††]Chemical Instrument Center; Nagoya University, Chikusa, Nagoya 464, Japan

(*Received in UK 7 September* 1987)

Abstract - Condensation of the acetyl protected 2-deoxy-2β-halo-3β-hydroxy-N-acetylneuraminic acid methyl ester with various acceptors such as properly protected glucose, galactose, and lactose derivatives in the presence of silver triflate gave the NeuAc α-glycosides in preference to the corresponding β-glycosides. By this method NeuAc(α2-6)Glc, NeuAc-(α2-3)Gal, and NeuAc(α2-3)Gal(β1-4)Glc derivatives were obtained in moderate yields.

In the course of glycosylation study of N-acetylneuraminic acid (NeuAc), we have already reported that the acetyl protected 2-deoxy-2β,3α-dibromo-NeuAc[2,3] and 2-deoxy-2β,3β-epoxy-NeuAc[4] methyl esters, which were prepared[5] by functionalization of the 2-deoxy-2,3-dehydro-NeuAc, gave stereospecifically NeuAc β-glycosides. We now report here the glycosylation of the acetyl protected 2-deoxy-2β-halo(Cl, Br)-3β-hydroxy-NeuAc methyl esters with various acceptors such as 6-unprotected glucose, 3-unprotected galactose, and 3'-unprotected lactose derivatives to give 2α-glycosides of NeuAc in preference to the corresponding β-glycosides.[6,7]

It was reported that the 5-acetamido-4,7,8,9-tetra-O-acetyl-2,5-dideoxy-2-fluoro-β-D-glycero-D-galacto-2-nonulopyranosonic acid allyl ester was effective for glycosylation of 1,2;3,4-di-O-isopropylidene-α-D-galactopyranose in the presence of boron trifluoride-ether complex to form the corresponding disaccharides (α:β=1:5).[8] When the above glycosylation condition was applied to the condensation of methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-2,5-dideoxy-2-fluoro-β-D-erythro-L-gluco-2-nonulopyranosonate (1) with methyl 2,3,4-tri-O-benzyl-α-D-glucopyranoside (4)[9], the desired glycosides were not obtained but the starting materials were recovered unchanged (Scheme 1, Table 1, Entry 1). The other catalysts such as silver trifluoromethanesulfonate (AgOTf) and trimethylsilyl trifluoromethanesulfonate (TMSOTf) gave similar results (Entry 2). These results suggested that the C-F bond was too strong for the glycosylation.

Glycosylation of 4 with 2β-chloro-3β-hydroxy-NeuAc derivative 2 in the presence of AgOTf (1.0 equiv) in benzene at room temperature gave a mixture of α- and β-glycosides, which could be separated by silica gel column chromatography to afford the α-glycoside 5 (21% yield) and the β-glycoside 8[4] (18% yield) (Entry 3). When two equivalents of the acceptor 4 were used in this condensation, improvement in the yield of the glycosides was observed (Entry 4). Determination of anomeric configuration of the α-glycoside 5 was made by analysis of its ^{1}H-NMR spectrum.[4,10] Thus, the coupling constant $J_{7,8}$ (8.9 Hz) and $\Delta\delta|$H-9'-H-9$|$ value (0.27 ppm) of the NeuAc unit clearly indicated that the anomeric configuration is α. The corresponding values of the β-anomer 8 were 1.5 Hz and

[†]Present address: Institute of Bio-Active Science (IBAS), Nippon Zoki Pharmaceutical Co., Ltd. (Kinashi, Yashiro-cho, Kato-gun, Hyogo 673-14, Japan).

Scheme 1

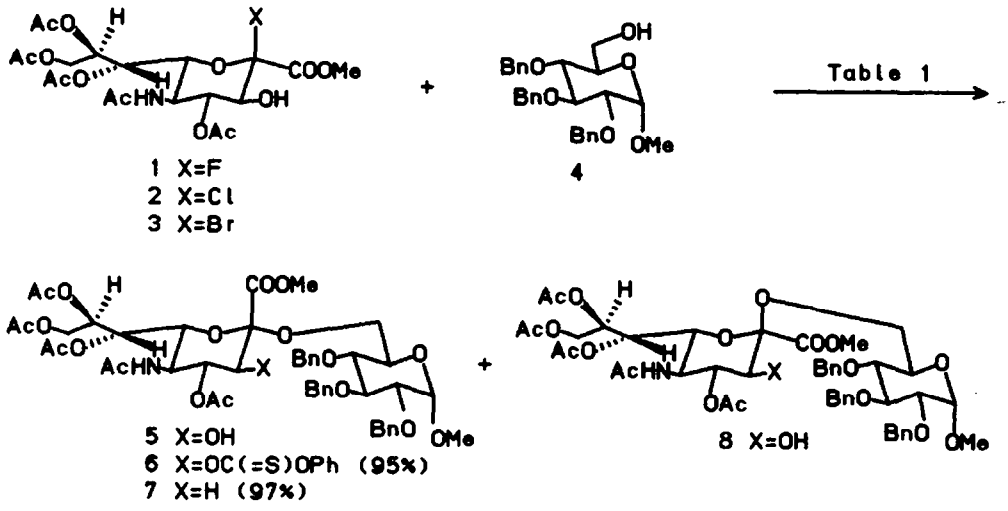


Table 1. Glycosylation of 4 with 1, 2, or 3

Entry	Donor	Acceptor 4 (mol. equiv)	Catalyst (mol. equiv)	Solvent	Reaction Temp.	Reaction Time	%Yield[a] of Glycosides 5(α form)	8(β form)
1	1	2.0	$BF_3 \cdot OEt_2$ (10.0), M.S.4A[b]	$ClCH_2CH_2Cl$	rt	2.0 h	c	
2	1	2.0	AgOTf (1.0), Na_2HPO_4	benzene	rt	1.0 h	c	
3	2	1.0	AgOTf (1.0), Na_2HPO_4	benzene	rt	0.5 h	21	18
4	2	2.0	AgOTf (1.0), Na_2HPO_4	benzene	rt	0.5 h	33	25
5	3	2.0	AgOTf (1.0), Na_2HPO_4	benzene	rt	10 min	38	50
6	3	2.0	$Hg(CN)_2$ (1.3), $HgBr_2$ (1.1)	$ClCH_2CH_2Cl$	rt	2.5 days	6	32
7	3	2.0	Ag_2CO_3 (5.0), Drierite	$ClCH_2CH_2Cl$	rt	4.0 days	d	
8	3	1.0	AgOTf (1.0), Na_2HPO_4	benzene	rt	10 min	28	53
9	3	1.0	AgOTf (1.0), Na_2HPO_4	toluene	-10°C	25 min	64	15

[a]Isolated yield. [b]Molecular sieves 4A. [c]Starting materials were recovered. [d]2,3-Epoxy-NeuAc derivative[5] was isolated in 67% yield.

0.94 ppm, respectively.[4] Moreover, by phenoxythiocarbonylation followed by reduction of the hydroxyl group at 3 position, 5 afforded 7, which was identical with the authentic α-glycoside.[2] Although the chloro derivative 2 was more reactive than the fluoride 1, glycosylation of the secondary alcohols such as methyl 2,4,6-tri-O-benzyl-β-D-galactopyranoside (9)[11] or methyl 2,6-di-O-benzyl-β-D-galactopyranoside (10)[11] with 2 did not proceed.

Glycosylation of 4 with the methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-2-bromo-2,5-dideoxy-β-D-*erythro*-L-*gluco*-2-nonulopyranosonate (3) was carried out in benzene, 1,2-dichloroethane, or toluene (Table 1). In the case of condensation using two equivalents of the acceptor 4 in the presence of AgOTf at room temperature, α- and β-glycosides were obtained in 38% and 50% yield, respectively, after chromatographic separation (Entry 5). This result indicated that the bromo derivative 3 was more reactive than the chloro derivative 2. The use of mercury(II) cyanide - mercury(II) bromide catalyst system, instead of AgOTf, resulted in lowering the yield (38%) and the preference of the β-glycoside (Entry 6). On the other hand, in the presence of silver carbonate, no glycoside was obtained but 2,3-epoxy-NeuAc compound[4], the precursor of 3, was produced (Entry 7). These results indicated that the best catalyst for this condensation was AgOTf. The use of only one equimolar

Scheme 2

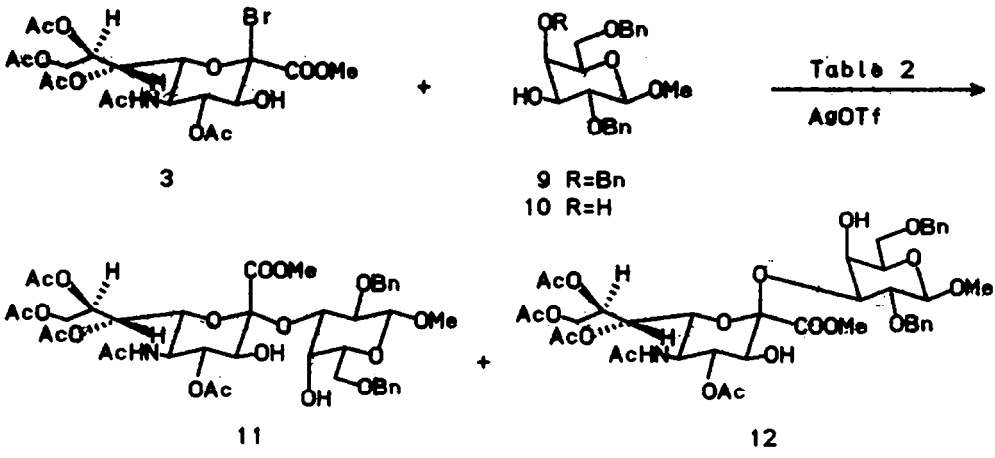


Table 2. Glycosylation of **10** with **3**[a]

Entry	Additive (equiv)	Solvent	Reaction Temp.	Time	%Yield[b] of Glycosides 11(α form)	12(β form)
1	Na_2HPO_4	benzene	rt	10 min	23	48
2	Na_2HPO_4	toluene	-15°C-rt	30 min	37	15
3	Na_2HPO_4	toluene-$ClCH_2CH_2Cl$ (1:1)	0 °C	10 min	33	17
4	Na_2HPO_4	toluene-$ClCH_2CH_2Cl$ (1:6)	0 °C	15 min	13	44
5	Ag_2CO_3 (5.0)	toluene	0 °C	20 min	27	27
6	Ag_2CO_3 (2.6)	toluene-CH_3NO_2 (1:7)	0 °C	30 min	c	
7	2,4,6-collidine	toluene	0 °C	30 min	c	

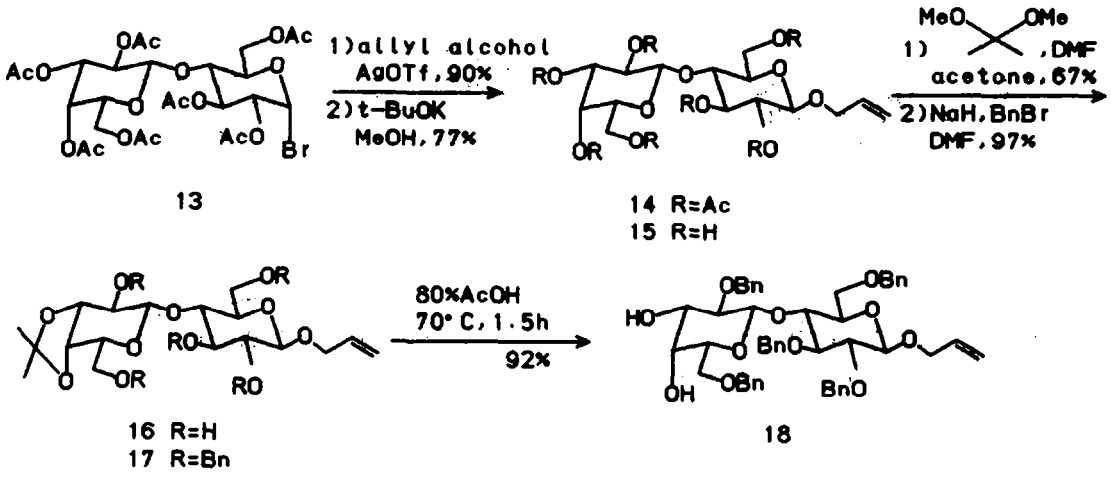
[a]Glycosylation was carried out using 1.0 equivalent of the acceptor **10** under argon atmosphere. [b]Determined by ^{1}H-NMR. [c]2,3-Epoxy-NeuAc derivative[5] was obtained in 70% yield (Entry 6) or 32% yield (Entry 7).

amount of **4** caused the yield to slightly decrease as expected (Entry 8).

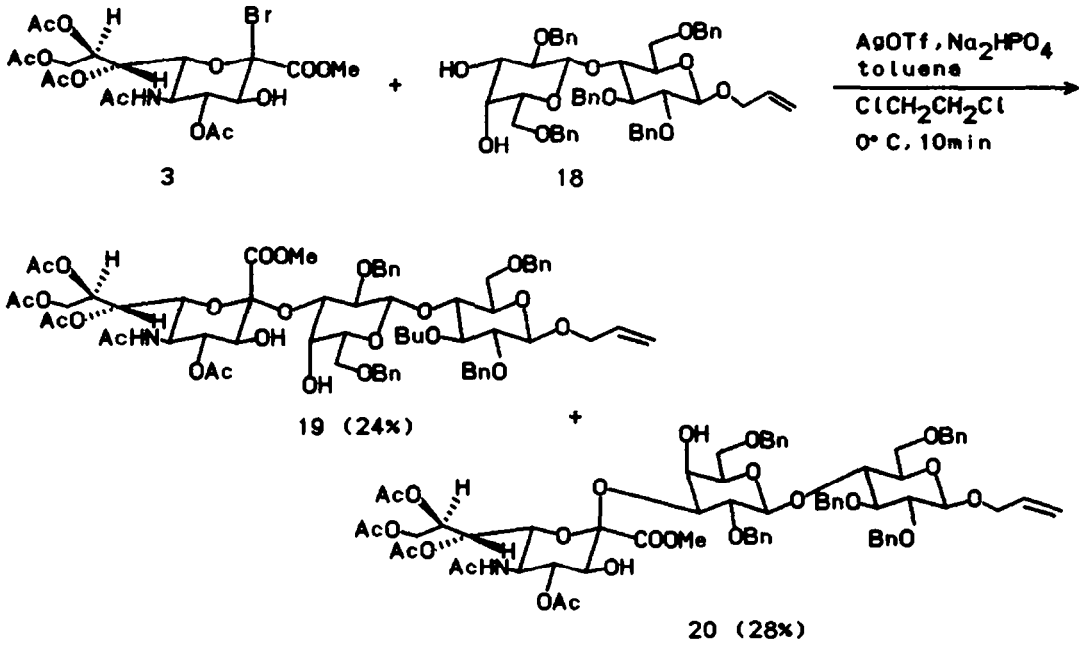
If this glycosylation can be performed under kinetically controlled condition, preferential formation of the more desired α-glycoside is expected. Indeed, as shown in Entry 9, the α-glycoside **5** was obtained in 64% yield in preference to the β-glycoside **8** (15% yield) when the condensation was run at -10 °C in toluene. Successful application of this method to the synthesis of a NeuAc(α2-3)-Gal derivative, which is related in natural gangliosides, is shown as follows.

Glycosylation of the galactose derivative **9** with the bromohydrin **3** in the presence of AgOTf gave no glycosides because the 3-hydroxyl group in **9** was sterically hindered by the 4-O-benzyl group (Scheme 2). By use of the 3,4-unprotected galactopyranoside **10** (1.0 equiv), the desired glycoside was obtained in 71% yield as a mixture of α- and β-anomers, which could not be separated by the usual silica gel column chromatography but was easily separated by ODS HPLC (methanol:water=70:30) giving the α-glycoside **11** (23% yield) and the β-glycoside **12** (48% yield) (Table 2, Entry 1). The anomeric configuration was deduced from the ^{1}H-NMR data: The coupling consatant $J_{7,8}$ values of **11** and **12** were 8.1 and 2.3 Hz, respectively, and $\Delta\delta|H\text{-}9'\text{-}H\text{-}9|$ values of them were 0.32 and 0.97 ppm, respectively. These values agreed with the empirical rule for determination of the anomeric position.[4] In this case, the β-glycoside **12** was in preference to the α-glycoside **11** but the total

Scheme 3



Scheme 4



yield of the both glycosides (71% yield) was superior than that (15% yield) reported by Ogawa et al.[12] As mentioned above, the yield of the α-anomer **11** was much improved (α:β=37:15) when the condensation was carried out in toluene at a lower temperature (-15 °C) (Entry 2). The same result was obtained when a mixture of toluene and 1,2-dichloroethane in a ratio of 1:1 was used as a solvent but the formation of the β-anomer **12** increased in proportion to the addition of 1,2-dichloroethane (Entries 3, 4).

Throughout the above glycosylation, disodium hydrogenphosphate (Na_2HPO_4) was used to neutralize trifluoromethanesulfonic acid that was formed by condensation. Instead of Na_2HPO_4, the use of silver carbonate in toluene resulted in the formation of a mixture of α- and β-isomers in a ratio of 1:1 (Entry 5), whereas the only 2,3-epoxy-NeuAc derivative[5] was yielded in nitromethane (a small amount of toluene was used to dissolve AgOTf) (Entry 6). Interestingly, the use of 2,4,6-collidine also gave the epoxy compound[5] (Entry 7). In conclusion, the α-glycoside **11** could be obtained in

preference to the β-glycoside **12** by the use of Na_2HPO_4 as a buffer and toluene or toluene-1,2-dichloroethane (1:1) as a solvent at a lower temperature in the presence of AgOTf.

Next, glycosylation of the 3',4'-unprotected lactose derivative **18** with the bromohydrin **3** was carried out to form the NeuAc(α2-3)Gal(β1-4)Glc linkage which was a part of ganglioside sugar chain. The acceptor **18** was prepared from the peracetyl α-lactosyl bromide **13**[13] by the following procedure (Scheme 3). Treatment of **13** with allyl alcohol in benzene in the presence of AgOTf afforded in 90% yield the allyl glycoside **14**, which was deacetylated with potassium t-butoxide in methanol to give allyl β-lactoside (**15**). The 3',4'-O-isopropylidene compound **16** was obtained in 2,2-dimethoxypropane-acetone-DMF/H_2SO_4 system as a main product. In this reaction **16** was scarcely formed without acetone as the solvent but the 4',6'-O-isopropylidene isomer was a main product. Benzylation of **16** with sodium hydride and benzyl bromide afforded **17**, which was treated with 80% acetic acid at 70 °C for 1.5 h to give the 3',4'-unprotected lactose derivative **18** as white crystals.

Glycosylation of **18** with the bromohydrin **3** in toluene-1,2-dichloroethane (1:1) in the presence of AgOTf at 15 °C gave a mixture of the 3'-O-(3β-hydroxy-2-neuraminyl)lactosides which was separated by ODS HPLC (methanol-water, 88:12) to give the α-anomer (24% yield) and the corresponding β-anomer (28% yield) (Scheme 4). The anomeric configuration could be also determined by ^{1}H-NMR spectra. The fact that the $J_{7,8}$ and $\Delta\delta|$H-9'-H-9$|$ values of the α-glycoside **19** were 7.9 Hz and 0.31 ppm, respectively, and those of the β-glycoside **20** were 1.8 Hz and 0.86 ppm, respectively, confirmed the anomeric configuration of the glycosides. Since the proton at 4 position of the galactose unit in **19** and **20** was coupled with a hydroxyl proton, 3 position of the galactose unit was glycosylated. In this condensation, treatment at a lower temperature (-15 °C) resulted in a lower yield (23%).

Using the bromohydrin **3** as a donor, various glycosides were obtained in good yields. However, when the hydroxyl group at 3 position of **3** was acetylated, glycosylation did not proceed at all. Therefore, the 3β-hydroxyl group may assist the catalyst in coordination and also prevent dehydrobromination reaction.

As reported earlier,[4] the Robins' method[14] has applied for removal of the 3β-hydroxyl group in the glycosides obtained above. This method could be used for the α-glycoside **5** to transform into **7**. The 3'-O-(3β-hydroxy-2-neuraminyl)lactose derivatives **19** and **20**, however, have two hydroxyl groups in each molecule, and it is required that the 3β-hydroxyl group of the NeuAc unit must be reduced selectively for ganglioside synthesis. When the α-glycoside **19** was treated with phenyl chlorocarbonothioate and 4-dimethylaminopyridine (DMAP) in N,N-dimethylformamide (DMF), a single product was obtained in 85% yield. In ^{1}H-NMR spectrum, the axial proton at 3 position of the NeuAc unit was coupled with a hydroxyl proton, suggesting the presence of 3-OH in NeuAc unit unchanged. And the fact that the methyl protons of ester group disappeared and the equatorial proton at 4 position of the galactose unit shifted to a lower field (δ 4.80) confirmed the structure to be the lactone **21** illustrated in Fig. 1. Similarly, the β-glycoside **20** was converted in 83% yield into the lactone **22** whose proton at 4 position of the galactose unit appeared in δ 4.96. The lactones **21** and **22** did not change by retreatment with phenyl chlorocarbonothioate in dimethylsulfoxide. From these results, reduction of 3β-hydroxyl group of **19** and **20** must be carried out stepwise.[15]

In conclusion, we found that the 2β-bromo-3β-hydroxy-NeuAc **3** can be used as a prominent glycosyl donor to obtain the 2α-glycosides of neuraminic acid such as NeuAc(α2-3)Gal and NeuAc(α2-3')Lac in preference to the corresponding 2β-glycosides in moderate yields.

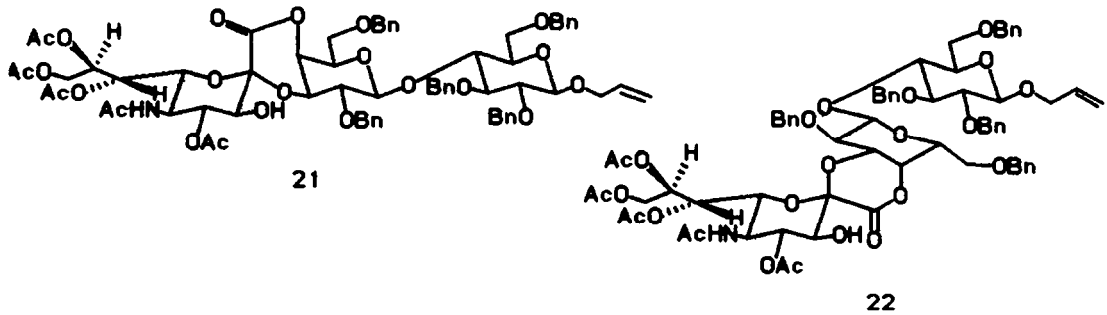


Figure 1

EXPERIMENTAL

General. Melting points were taken on a Mitamura Riken flat-bulb thermometer with a heating metal block and uncorrected. Elemental analyses were done on a Perkin-Elmer 240C elemental analyzer. Nuclear magnetic resonance spectra (NMR) were obtained with a JEOL GX-500 instrument in the FT mode. Chemical shifts (δ) were expressed in parts per milion from internal tetramethylsilane unless otherwise noted. Coupling constants are in hertz (Hz) and splitting pattern abbreviations are: s, singlet; d, doublet; dd, doublet of doublets; ddd, doublet of double doublets; m, mutiplet; br, broad. Mass spectra (MS) were obtained on a JEOL DX-300 spectrometer. Infrared spectra (IR) were recorded on a JASCO A-3 spectrophotometer. Optical rotations $[\alpha]_D$ were recorded on a JASCO DIP-181 digital polarimeter.

Preparative high performance liquid chromatography (HPLC) was carried out on a JASCO Trirotor III, IV, or BIP liquid chromatography system and UVIDEC-III, IV, or V as a UV (254 nm) detector by use of a reversed-phase silica-gel (ODS, 10-20 μ Develosil, NOMURA Chemical Co. Ltd.) in a stainless column (10 φ X 250 mm).

Analytical thin layer chromatography (TLC) was conducted on precoated TLC glass sheets (silica gel 60F-254, layer thickness 0.25 mm) manufactured by E. Merck. Detection was effected by dipping into 2% concentrated sulfuric acid ethanol solution followed by heating on a hot plate (ca 120 °C).

Column chromatography was performed with Merck silica gel 60 (70-230 mesh).

^{1}H-NMR data were summarized in Table 3 - 8 and MS, elemental analyses, Mp, Rf, $[\alpha]_D$, and IR data were in Table 9.

Condensation of 2 with the glucose derivative 4. To a mixture of 2 (50 mg, 0.095 mmol), 4 (45 mg, 0.097 mmol), and anhydrous Na_2HPO_4 (50 mg) in benzene (1.0 ml) was added a solution of AgOTf (25 mg, 0.097 mmol) in benzene (1.0 ml) at room temp under argon. The mixture was stirred for 30 min, diluted with ethyl acetate, filtered through a glass filter, and the solid was washed well with ethyl acetate. The combined filtrates and washings were condensed to a residue, which was dissolved in ethyl acetate, washed with 5% $Na_2S_2O_3$, 5% $NaHCO_3$, and brine, dried over anhydrous Na_2SO_4, and evaporated *in vacuo* to give a syrup, which was chromatographed on a silica gel column (benzene-acetone, 3:2). The fast migrating zone was methyl 2,3,4-tri-O-benzyl-6-O-(methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-5-deoxy-β-D-*erythro*-L-*gluco*-2-nonulopyranosylonate)-α-D-glucopyranoside (**8**) (16 mg, 18%), and the slow one was the α-isomer **5** (19 mg, 21%), both of which were obtained as a syrup.

Condensation of 3 with the glucose derivative 4.

A (AgOTf catalyst: Typical procedure for Table 1, Run 5, 8, 9). To a stirred mixture of **3** (70 mg, 0.12 mmol), **4** (57 mg, 0.12 mmol), and anhydrous Na_2HPO_4 (80 mg) in dry toluene (1.5 ml) was added a solution of AgOTf (32 mg, 0.12 mmol) in toluene (1.0 ml) at -10 °C under argon. The mixture was stirred for 10 min at -10 °C and for additional 15 min at room temp and worked up in the same manner as described above to give **5** (75 mg, 64%) and **8** (18 mg, 15%).

B (Hg^{++} catalyst: Table 1, Run 6). A mixture of **3** (70 mg, 0.12 mmol), **4** (114 mg, 0.24 mmol), mercury(II) cyanide (40 mg, 0.16 mmol), mercury(II) bromide (50 mg, 0.14 mmol), and powdered molecular sieves 4A (100 mg) in 1,2-dichloroethane (3 ml) was stirred for 2.5 days at room temp under argon in the dark. The reaction mixture was worked up in the same manner as described above and the glycosides **5** (7 mg, 6%) and **8** (37 mg, 32%) were obtained.

C (Ag_2CO_3 catalyst: Table 1, Run 7). A mixture of **3** (70 mg, 0.24 mmol), **4** (114 mg, 0.24 mmol), Ag_2CO_3 (170 mg, 0.62 mmol), and Drierite (200 mg) in 1,2-dichloroethane (3 ml) was stirred for 4.0 days at room temp under argon. The mixture was filtered and washed with chloroform. The chloroform solution was evaporated *in vacuo* to give a syrup, which was chromatographed on a silica gel column (benzene-acetone, 3:2) to give methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-2,3-anhydro-5-deoxy-β-D-*erythro*-L-*gluco*-2-nonulopyranosonate[5] (41 mg, 67%) as white crystals.

Condensation of 3 with the galactose derivative 10.

A (Typical procedure for Table 2, Run 1-4). To a stirred mixture of **3** (130 mg, 0.23 mmol), **10** (90 mg, 0.24 mmol), and anhydrous Na_2HPO_4 (120 mg) in toluene (4 ml) was added a solution of AgOTf (60 mg, 0.23 mmol) in toluene (2 ml) at -15 °C under argon. The mixture was stirred for 15 min at the same temp and allowed to warm to room temp. After stirring for further 15 min, the mixture was filtered and washed with ethyl acetate. The combined filtrates and washings were washed with 5% $Na_2S_2O_3$, 5% $NaHCO_3$, and brine, dried over anhydrous Na_2SO_4, and evaporated *in vacuo* to give a crude material, which was chromatographed on a silica gel column (benzene-acetone, 3:2) to give methyl 2,6-di-O-benzyl-3-O-(methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-5-deoxy-α and β-D-*erythro*-L-*gluco*-2-nonulopyranosylonate)-β-D-galactopyranoside (**11**) and (**12**) (102 mg, 52%) as an inseparable syrup in a ratio of **11**:**12**=37:15. This syrup was separated by preparative ODS HPLC (methanol-water, 70:30 at 40 °C). The fast eluted compound was α-isomer **11** and the slow one β-isomer **12**.

B (Ag_2CO_3 as a buffer in toluene: Table 2, Run 5). A solution of **3** (80 mg, 0.14 mmol) and **10** (55 mg, 0.15 mmol) in toluene (3 ml) were added to a mixture of Ag_2CO_3 (195 mg, 0.71 mmol) and Drierite (250 mg) in dry toluene (3 ml). To this was added a solution of AgOTf (36 mg, 0.14 mmol) in toluene (1 ml) at 0 °C under argon. After stirring for 20 min, the mixture was worked up in the same manner as **A** to give a mixture of **11** and **12** (65 mg, 54%) in a ratio of **11**:**12**=27:27.

C (Ag_2CO_3 as a buffer in toluene and nitromethane: Table 2, Run 6). To a stirred mixture of **3** (80 mg, 0.14 mmol), **10** (55 mg, 0.15 mmol), Ag_2CO_3 (100 mg, 0.36 mmol), and powdered molecular sieves 4A (200 mg) in nitromethane (3.5 ml) was added a solution of AgOTf (36 mg, 0.14 mmol) in toluene (0.5 ml) at 0 °C under argon. The mixture was stirred for 30 min and worked up in the same manner as **A** to give the 2,3-epoxy-NeuAc derivative[5] (48 mg, 70%).

Table 3. ^{1}H-NMR Data for Non-reducing (NeuAc) Unit in Chloroform-d

Compound	Chemical shifts, δ (multiplicities)												
	H-3ax (dd)	H-4 (dd)	H-5 (ddd)	H-6 (dd)	H-7 (dd)	H-8 (ddd)	H-9 (dd)	H-9' (dd)	Me ester (s)	NH (d)	OH-3 (d)	O-Ac, N-Ac (s)	Phenyl (m)
5	3.82	5.12	4.15	4.56	5.21	5.33	3.85	4.12	3.73	5.38	2.97	1.86,2.01,2.04,2.05,2.17	
6	5.88[a]	5.43	4.41	4.64	5.28	5.37	3.98	4.22	3.65	5.40		1.95,1.96,2.02,2.03,2.04	6.90-7.50
11	3.92	5.28	4.15	4.37	5.19	5.18	3.89	4.21	3.79	5.50	2.51	1.87,1.99,1.99,2.00,2.03	
12	4.02	5.36	4.19	4.35	5.37	5.15	3.95	4.92	3.48	6.07	3.87	1.77,2.00,2.02,2.06,2.11	
19	3.90	5.30	4.19	4.38	5.21	5.18	3.88	4.19	3.78	5.54	2.38	1.87,1.91,1.98,2.03,2.04	
20	3.97	5.30	4.17	4.52	5.30	5.22	3.96	4.82	3.67	5.20	3.27	1.79,1.91,2.03,2.06,2.10	
21	3.65	5.71	4.32	3.87	5.27	5.10	3.93	4.38		5.92	2.74	1.86,1.94,2.00,2.12,2.15	
22	3.85	5.12	4.53	4.03	5.32	5.55	4.13	4.57		5.78	3.76	1.81,1.86,1.91,1.91,1.91	

Compound	First-order coupling constants, Hz									
	$J_{3ax,4}$	$J_{4,5}$	$J_{5,6}$	$J_{5,NH}$	$J_{6,7}$	$J_{7,8}$	$J_{8,9}$	$J_{8,9'}$	$J_{9,9'}$	$J_{OH,3ax}$
5	10.1	10.1	11.0	10.4	2.1	8.9	5.8	2.5	-12.5	4.9
6	9.2	10.4	11.0	10.1	2.1	8.9	5.8	2.7	-12.5	
11	10.1	10.1	10.3	10.1	1.5	8.1	3.0	2.1	-12.5	4.5
12	9.8	9.8	10.4	9.5	2.1	2.3	8.9	2.1	-12.5	7.3
19	9.8	9.4	11.7	10.1	1.8	7.9	6.1	2.4	-12.2	5.2
20	9.8	10.0	11.4	10.5	2.7	1.8	8.7	2.1	-12.2	6.0
21	10.1	10.1	10.7	10.2	2.1	7.3	7.0	3.0	-11.0	8.9
22	9.5	10.1	10.5	9.0	3.1	3.8	1.8	1.5	-12.0	9.2

[a]Multiplicity: d.

Table 4. ^{1}H-NMR Data for Reducing (Glc, Gal) Unit in Chloroform-d

Compound	Chemical shifts, δ (multiplicities)								First-order coupling constants, Hz						
	H-1 (d)	H-2 (dd)	H-3 (dd)	H-4 (dd)	H-5 (ddd)	H-6 (dd)	H-6' (dd)	OH-4 (br.s)	$J_{1,2}$	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$	$J_{5,6}$	$J_{5,6'}$	$J_{6,6'}$
5	4.61	3.51	3.97	3.54	3.82	3.77	4.14		3.4	9.8	9.5	9.2	2.3	3.8	-10.5
6	4.61	3.48	3.99	3.44	3.83	3.85	4.04		3.7	9.9	9.5	9.2	1.2	6.9	-10.4
11	4.35	3.71	3.88	4.00	3.63	3.77	3.87	2.95	7.6	9.8	2.9	0.5	5.8	5.8	-10.1
12	4.32	3.85	4.42	4.13	3.69	3.74	3.78	4.15	7.6	9.8	3.1	0	5.8	5.2	-10.1
19	4.43	3.45	3.58	4.09	3.41	3.80	3.83		7.7	8.9	9.0	9.5	1.7	4.0	-11.0
20	4.48	3.43	3.56	4.02	3.30	3.73	3.78		7.9	9.2	9.5	9.5	1.5	4.1	-11.1
21	4.40	3.37	3.58	3.91	3.23	3.52	3.66		7.7	9.2	10.1	10.1	1.0	4.5	-9.8
22	4.41	3.43	3.55	4.04	3.24	3.63	3.68		7.9	9.2	8.9	9.8	1.5	3.6	-9.8

Table 5. ^{1}H-NMR Data for Central (Gal) Unit in Chloroform-d

Compound	Chemical shifts, δ (multiplicities)								First-order coupling constants, Hz							
	H-1 (d)	H-2 (dd)	H-3 (dd)	H-4 (dd)	H-5 (ddd)	H-6 (dd)	H-6' (dd)	OH-4 (d)	$J_{1,2}$	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$	$J_{5,6}$	$J_{5,6'}$	$J_{6,6'}$	$J_{OH,4}$
19	4.52	3.69	3.74	4.04	3.47	3.56	3.78	2.94	7.3	9.3	3.1	0	6.0	5.5	-9.8	2.4
20	4.52	3.72	4.17	4.08	3.35	3.53	3.63	3.62	7.9	9.3	2.4	0	5.2	5.6	-10.1	2.1
21	4.42	3.37	4.09	4.80	3.47	3.20	3.62		7.9	9.2	3.7	0	6.7	5.4	-10.7	
22	4.48	3.77	4.18	4.96	3.56	3.46	3.58		7.6	9.8	3.0	0	4.9	7.9	-8.5	

D (2,4,6-Collidine as a buffer in toluene: Table 2, Run 7). To a mixture of **3** (80 mg, 0.14 mmol), **10** (55 mg, 0.15 mmol), 2,4,6-collidine (21 μl, 0.16 mmol), and powdered molecular sieves 4A (100 mg) in toluene (2.5 ml) was added a solution of AgOTf (36 mg, 0.14 mmol) in toluene (0.5 ml) at 0 °C under argon. The resulting mixture was treated in the same manner as **C** to give the 2,3-epoxy-NeuAc derivative[5] (22 mg, 32%).

Allyl 2,3,6-tri-O-acetyl-6-O-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-β-D-glucopyranoside (14). To a mixture of the peracetyl α-lactosyl bromide 13[13] (70 g, 100 mmol) and anhydrous Na_2HPO_4 (50 g) in allyl alcohol (130 ml) and benzene (200 ml) was added dropwise a solution of AgOTf (26 g, 101 mmol) in benzene (100 ml) at room temp under argon. After stirring for 30 min, the reaction mixture was filtered and the solid was washed well with ethyl acetate. The combined filtrates and washings were evaporated *in vacuo* to give a residue, which was dissolved in ethyl acetate, washed with 5% $Na_2S_2O_3$, 5% $NaHCO_3$, and brine, dried over anhydrous Na_2SO_4, and evaporated *in vacuo* to give a syrup. This was chromatographed on a silica gel column (benzene-ethyl acetate, 2:1) to give **14** (61 g, 90%) as a syrup.

Table 6. ^{1}H-NMR Data for Other Groups in Chloroform-d

Compound	Chemical shifts, δ (multiplicities)				Coupling constants, Hz
	PhCH$_2$ (AB quartet)	Ph (m)	OMe (s)	O-allyl (m)	J_{gem}
5	4.66 & 4.80, 4.77 & 4.81, 4.78 & 4.94	7.25-7.45	3.38		-12.2, -10.7, -11.0
6	4.65 & 4.77, 4.71 & 4.87, 4.81 & 4.96	6.90-7.50	3.36		-12.2, -10.7, -11.0
11	4.58 & 4.61, 4.61 & 5.00	7.25-7.40	3.60		-11.9, -11.3
12	4.60, 4.63 & 4.89	7.25-7.40	3.47		-10.8
19	4.38 & 4.48, 4.54 & 4.72, 4.62 & 4.88 4.73 & 4.92, 4.75 & 5.01	7.20-7.48		4.13, 4.41, 5.20 5.33, 5.96	-11.9, -12.5, -11.6 -11.0, - 9.4
20	4.39 & 4.44, 4.47 & 4.58, 4.70 & 4.89 4.73 & 4.84, 4.76 & 4.99	7.20-7.42		4.11, 4.38, 5.18 5.32, 5.95	-12.5, -12.2, -10.7 -11.3, -11.0
21	4.13 & 4.35, 4.49 & 4.66, 4.37 & 4.52 4.73 & 4.89, 4.83 & 4.94	7.10-7.45		3.99, 4.28, 5.18 5.32, 5.93	-11.9, -11.6, -12.2 -10.7, -10.7
22	4.32 & 4.41, 4.46 & 4.54, 4.67 & 4.79 4.75 & 4.93, 4.78 & 4.95	7.10-7.45		4.12, 4.40, 5.20 5.35, 5.96	-12.2, -11.9, -11.9 -10.7, -10.4

Table 7. ^{1}H-NMR Data for Lactose Derivatives **14** - **18** in Chloroform-d

(Glucose Unit)

Compound	Chemical shifts, δ (multiplicities)							First-order coupling constants, Hz						
	H-1 (d)	H-2 (dd)	H-3 (dd)	H-4 (dd)	H-5 (ddd)	H-6 (dd)	H-6' (dd)	$J_{1,2}$	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$	$J_{5,6}$	$J_{5,6'}$	$J_{6,6'}$
14	4.52	4.92	5.20	3.81	3.60	4.10	4.49	7.9	9.5	9.5	9.5	5.0	2.2	-11.3
15[a]	4.52	3.32	3.63	3.62	3.58	3.79	3.96	7.9	9.2	b	b	5.2	2.1	-12.2
16[a]	4.52	3.32	3.62	3.63	3.57	3.77	3.94	7.9	9.2	b	b	5.3	2.1	-12.5
17	4.43	3.41	3.57	3.96	3.40	3.73	3.80	7.9	9.2	9.2	9.5	4.3	1.5	-11.0
18	4.44	3.46	3.59	4.00	3.38	3.75	3.81	7.9	9.2	9.2	9.5	1.5	4.0	-11.0

(Galactose Unit)

Compound	Chemical shifts, δ (multiplicities)									First-order coupling constants, Hz								
	H-1 (d)	H-2 (dd)	H-3 (dd)	H-4 (dd)	H-5 (ddd)	H-6 (dd)	H-6' (dd)	OH-3 (d)	OH-4 (d)	$J_{1,2}$	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$	$J_{5,6}$	$J_{5,6'}$	$J_{6,6'}$	$J_{3,OH}$	$J_{4,OH}$
14	4.49	5.10	4.95	5.34	3.88	4.08	4.13			7.9	10.4	3.7	1.2	7.5	6.4	-11.0		
15[a]	4.43	3.52	3.65	3.91	3.71	b	b			7.9	9.1	b	0	b	b	b		
16[a]	4.48	3.49	4.20	4.35	4.08	3.80	3.84			8.7	7.8	5.2	1.9	8.2	4.0	-12.2		
17	4.40	3.34	4.02	4.10	3.67	3.53	3.68			8.2	7.7	5.5	1.0	9.5	b	-13.1		
18	4.43	3.42	3.43	3.93	3.34	3.49	3.61	2.44	2.52	7.9	b	3.3	0	5.2	6.4	-10.0	4.2	3.4

[a]Measured in D_2O (t-BuOH=1.23 ppm). [b]Not assigned owing to the complexity of the spectrum.

Table 8. ^{1}H-NMR Data for Other Groups of Lactose Derivatives **14** - **18** in Chloroform-d

Compound	Chemical shifts, δ (multiplicities)				Coupling constants, Hz
	PhCH$_2$ (AB quartet)	Ph (m)	O-allyl (m)	acetonide	J_{gem} of PhCH$_2$
14	1.96, 2.04, 2.05, 2.05 2.06, 2.13, 2.15[a]		4.08,4.31,5.19 5.26,5.85		
15[b]			4.22,4.38,5.28 5.38,5.98		
16[b]			4.22,4.48,5.27 5.37,5.97	1.37,1.53	
17	4.31 & 4.50, 4.42 & 4.58, 4.67 & 4.79 4.71 & 4.90, 4.74 & 4.93	7.25-7.45	4.13,4.41 5.19 5.33,5.96	1.35,1.40	-13.2,-12.2,-11.6 -10.7,-10.4
18	4.38 & 4.45, 4.43 & 4.60, 4.67 & 4.81 4.72 & 4.91, 4.78 & 4.98	7.25-7.45	4.13,4.42 5.95 5.33,5.95		-11.9,-12.2,-11.6 -10.7,-11.0

[a]Acetyl groups. [b]Measured in D_2O (t-BuOH=1.23 ppm).

Allyl 6-O-(β-D-galactopyranosyl)-β-D-glucopyranoside (15). To a solution of **14** (60 g, 89 mmol) in absolute methanol (300 ml) was added potassium t-butoxide (1.0 g) at room temp under argon. The mixture was stirred for 1 h and acidified with Dowex 50W-X8 [H^+]. The resin was removed by filtration and washed well with water. After removal of the solvent, the residue was crystallized from water-methanol-ether to give **15** (26 g, 77%) as white crystals.

Allyl 6-O-(3,4-O-isopropylidene-β-D-galactopyranosyl)-β-D-glucopyranoside (16). To a mixture of the finely powdered **15** (10 g, 26 mmol) in 2,2-dimethoxypropane (20 ml), acetone (200 ml), and N,N-dimethylformamide (DMF) (50 ml) was added conc. sulfuric acid (cat. amount). The mixture was refluxed for 15 h, neutralized with anhydrous sodium carbonate, and filtered through a Celite 545 bed. The filtrate was evaporated in vacuo to give a crude material, which was chromatographed on a silica gel column (chloroform-methanol, 5:1) and recrystallized from acetone-methanol to give **16** (7.4 g, 67%) as white crystals.

Allyl 2,3,6-tri-O-benzyl-6-O-(2,6-di-O-benzyl-3,4-O-isopropylidene-β-D-galactopyranosyl)-β-D-glucopyranoside (17). To a suspension of sodium hydride (50% in oil, 4.8g, 100 mmol) in dry DMF (80 ml) was added a solution of **16** (5.0 g, 12 mmol) in DMF (20 ml) at 0 °C under argon. The mixture was stirred for 0.5 h at room temp and cooled again to 0 °C. To this was added dropwise benzyl bromide (12 ml, 101 mmol) with stirring. The resulting mixture was stirred for additional 6 h at room temp and methanol (10 ml) was added carefully to decompose the remaining sodium hydride and benzyl bromide. After addition of ethyl acetate (200 ml), the mixture was worked up as usual to give a crude oil, which was chromatographed on a silica gel column (hexane-ethyl aceate, 3:1) to give **17** (10.0 g, 97%) as an oil.

Allyl 2,3,6-tri-O-benzyl-6-O-(2,6-di-O-benzyl-β-D-galactopyranosyl)-β-D-glucopyranoside (18). A solution of **17** (2.4 g, 2.8 mmol) in 80% aqueous acetic acid (25 ml) was heated at 70 °C for 1.5 h and condensed to a syrup, which was crystallized from hexane-ethyl acetate to give **18** (2.1 g, 92%) as white crystals.

Condensation of 3 with the lactose derivative 18. To a stirred mixture of **3** (200 mg, 0.35 mmol), **18** (320 mg, 0.38 mmol), and anhydrous Na_2HPO_4 (200 mg) in toluene (10 ml) was added a solution of AgOTf (100 mg, 0.39 mmol) in toluene (2 ml) at 15 °C under argon. After stirring for 15 min, the mixture was worked up as described before to give a syrup, which was chromatographed on a silica gel column (benzene-acetone, 2:1) to give allyl 2,3,6-tri-O-benzyl-4-O-[2,6-di-O-benzyl-3-O-(methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-5-deoxy-α and β-D-erythro-L-gluco-2-nonulopyranosylonate)-β-D-galactopyranosyl]-β-D-glucopyranoside (**19**) and (**20**) (259 mg, 52%) as an inseparable mixture in a ratio of **19**:**20**=24:28. The mixture was separated by preparative ODS HPLC (methanol-water, 44:6 at 37 °C). The fast eluted compound was α-isomer **19** and the slow one β-isomer **20**.

Methyl 2,3,4-tri-O-benzyl-6-O-(methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-5-deoxy-3-[phenoxy(thiocarbonyl)]-α-D-erythro-L-gluco-2-nonulopyranosylonate)-α-D-glucopyranoside (6). To a solution of **5** (55 mg, 0.058 mmol) and DMAP (30 mg, 0.25 mmol) in dry acetonitrile (1.5 ml) was added phenyl chlorocarbonothioate (16 µl, 0.12 mmol) under argon. The mixture was stirred for 24 h at room temp and condensed to a residue, which was partitioned between ethyl acetate and water. The ethyl acetate layer was worked up as usual to give a crude material, which was chromatographed on a silica gel column (benzene-acetone, 3:1) to give **6** (60 mg, 95%) as a syrup.

Reaction of 19 with phenyl chlorocarbonothioate in the presence of DMAP. To a solution of **19** (40 mg, 0.030 mmol) and DMAP (15 mg, 0.12 mmol) in dry DMF (0.5 ml) was added phenyl chlorocarbonothioate (6 µl, 0.043 mmol) under argon. The mixture was stirred at 80 °C for 4 h and condensed to a residue. A solution of the residue in ethyl acetate was worked up as usual to give a syrup, which was chromatographed on a silica gel column (benzene-acetone, 2:1) to give lactone **21** (33 mg, 85%) as a syrup.

Reaction of 20 with phenyl chlorocarbonothioate in the presence of DMAP. A mixture of **20** (105 mg, 0.079 mmol), DMAP (40 mg, 0.33mmol), and phenyl chlorocarbonothioate (22 µl, 0.16 mmol) in dry DMF (1 ml) was stirred at 80 °C for 12 h under argon and worked up in the same manner as described above to give a solid, which was recrystallized from hexane-ethyl acetate to give lactone **22** (85 mg, 83%) as white crystals.

Methyl 2,3,4-tri-O-benzyl-6-O-(methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-α-D-glycero-D-galacto-2-nonulopyranosylonate)-α-D-glucopyranoside (7). To a solution of **6** (110 mg, 0.10 mmol) in toluene (1.5 ml) were added tributylstannane (54 µl, 0.20 mmol) and AIBN (cat. amount) under argon. The mixture was heated at 110 °C for 10 min and condensed to a residue, which was chromatographed on a silica-gel column (benzene-acetone, 2:1) to give a syrup. The syrup was triturated with hexane-ethyl acetate and washed with hexane to give **7** (92 mg, 97%) as a white powder.

Table 9. MS, Elemental Analyses, Mp, Rf, $[\alpha]_D$, and IR Data

Compound	Formula	MS [a] (M+H)	Anal.	% C	% H	% N	Mp (°C)	Rf	$[\alpha]_D$(c)[b] (Temp)	ν_{max}^{KBr} NH,OH	ester	amide I	II
5	$C_{48}H_{59}NO_{19}$	955	Calcd.	60.43	6.23	1.47	---[c]	0.34[d]	- 8.9°(1.0) (21°C)	3400	1743	1660	1540
			Found	60.57	6.55	1.23							
6	$C_{55}H_{63}NO_{20}S$		Calcd.	60.60	5.82	1.29	---[c]	0.57[e]	- 4.9°(2.1) (20°C)	3400	1745	1663	1538
			Found	60.38	5.69	1.29							
11	$C_{41}H_{53}NO_{19}$	865	Calcd.	57.00	6.18	1.62	---[c]	0.35[d]	-19.7°(1.0) (21°C)	3430	1740	1660	1540
			Found	57.36	6.01	1.87							
12	$C_{41}H_{53}NO_{19}$	865	Calcd.	57.00	6.18	1.62	---[c]	0.35[d]	-23.3°(1.2) (21°C)	3430	1740	1660	1540
			Found	56.79	5.99	1.43							
14	$C_{29}H_{40}O_{18}$		Calcd.	51.48	5.96		---[c]	0.33[f]	-11.7°(1.8) (24°C)		1750		
			Found	51.45	6.12								
15	$C_{15}H_{26}O_{11}$	383	Calcd.	47.12	6.85		168-170[g]	0.29[h]	+ 2.1°(2.4)[i] (24°C)	3450			
			Found	47.33	6.71								
16	$C_{18}H_{30}O_{11}$	423	Calcd.	51.18	7.16		185-186[j]	0.35[k]	+11.8°(1.5)[i] (24°C)	3450			
			Found	50.97	7.25								
17	$C_{53}H_{60}O_{11}$		Calcd.	72.91	6.93		---[c]	0.60[l]	+18.5°(1.6) (24°C)				
			Found	73.11	6.84								
18	$C_{50}H_{56}O_{11}$		Calcd.	72.09	6.78		110-111[m]	0.53[f]	+18.0°(1.2) (24°C)	3455			
			Found	72.21	6.86								
19	$C_{70}H_{83}NO_{24}$	1323	Calcd.	63.58	6.33	1.06	---[c]	0.68[n]	- 4.4°(0.9) (13°C)	3425	1747	1660	1540
			Found	63.45	6.38	1.13							
20	$C_{70}H_{83}NO_{24}$	1323	Calcd.	63.58	6.33	1.06	---[c]	0.70[n]	- 5.8°(1.1) (13°C)	3440	1745	1665	1540
			Found	63.35	6.59	1.02							
21	$C_{69}H_{79}NO_{23}$		Calcd.	64.22	6.17	1.09	---[c]	0.46[e]	- 8.5°(1.5) (12°C)	3420	1750	1660	1540
			Found	64.31	6.35	1.24							
22	$C_{69}H_{79}NO_{23}$		Calcd.	64.22	6.17	1.09	150-152[l]	0.36[e]	-37.8°(1.0) (12°C)	3400	1745	1660	1550
			Found	64.56	6.19	1.33							

[a]Fast atom bomberdment method. [b]Measured in chloroform. [c]Viscous syrup. [d]Solvent system is benzene-acetone (3:2). [e]Solvent system is benzene-acetone (2:1). [f]Solvent system is benzene-ethyl acetate (2:1). [g]Recrystallized from water-methanol-ether. [h]Solvent system is butanol-acetic acid-water (3:1:1). [i]Measured in water. [j]Recrystallized from acetone-methanol. [k]Solvent system is chloroform-methanol (5:1). [l]Solvent system is hexane-ethyl acetate (2:1). [m]Recrystallized from hexane-ethyl acetate. [n]Solvent system is benzene-acetone (1:1).

REFEREMCES AND NOTES

1. Synthetic Studies on Gangliosides 7.
2. K. Okamoto, T. Kondo, and T. Goto, Chemistry Lett., **1986**, 1449.
3. K. Okamoto, T. Kondo, and T. Goto, Tetrahedron, in contribution.
4. K. Okamoto, T. Kondo, and T. Goto, Bull. Chem. Soc. Jpn., **60**, 637 (1987).
5. K. Okamoto, T. Kondo, and T. Goto, Bull. Chem. Soc. Jpn., **60**, 631 (1987).
6. K. Okamoto, T. Kondo, and T. Goto, Tetrahedron Lett., **27**, 5229 (1986).
7. K. Okamoto, T. Kondo, and T. Goto, Tetrahedron Lett., **27**, 5233 (1986).
8. H. Kunz and H. Waldmann, J. Chem. Soc., Chem. Commun., **1985**, 638.
9. P. Kovac, J. Alfödi, and B. Kosik, Chem. Zveski, **28**, 820 (1974).
10. H. Paulsen and H. Tietz, Carbohydr. Res., **125**, 47 (1984).
11. H. M. Flowrers, Carbohydr. Res., **39**, 245 (1975).
12. T. Ogawa and M. Sugimoto, Carbohydr. Res., **135**, C5-C9 (1985).
13. C. S. Hudson and A. Kunz, J. Am. Chem. Soc., **47**, 2052 (1925).
14. M. J. Robins and J. S. Wilson, J. Am. Chem. Soc., **103**, 932 (1981).
15. Reduction of 3β-hydroxyl group will be reported elsewhere.