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A simple procedure for the synthesis of the methyl and benzyl glycosides of Neu5Ac and 4-*epi*-Neu5Ac. Conversion of the benzyl and methyl glycosides of Neu5Ac into *N*-trifluoroacetylneuraminic acid benzyl glycosides *

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ABSTRACT

From the reaction products of the Kuhn–Bashang synthesis of Neu5Ac, 5-acetamido-3,5-dideoxy-α- and -β-D-*glycero*-D-*talo*-2-nonulopyranosonic acid (4-*epi*-Neu5Ac) were isolated as the acetylated methyl esters (**1** and **2**). Treatment of methyl (5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-D-*galacto*-2-nonulopyranosyl bromide)onate (**5**) with an excess of methanol gave a high yield of a 9:1 α,β-mixture of the methyl glycosides (**13** and **14**). Likewise, with benzyl alcohol, **5** gave a 63:32 α,β-mixture of the benzyl glycosides (**17** and **18**). Treatment of methyl (5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-β-D-*glycero*-D-*talo*-2-nonulopyranosyl bromide)onate (**7**) with an excess of benzyl alcohol gave a 3:1 α,β-mixture of the benzyl glycosides (**21** and **22**) together with methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-2,6-anhydro-3,5-dideoxy-D-*glycero*-D-*talo*-non-2-enonate (**9**). Condensation of bromide **7** in chloroform with benzyl alcohol in the presence of silver carbonate afforded a 7:1 α,β-mixture of benzyl glycosides together with **9**. The benzyl glycosides **17** and **18** were converted into their respective *N*-trifluoroacetyl derivatives **27** and **28** by saponification and then *N*-trifluoroacetylation. Methanolysis of Neu5Ac followed by *N*-trifluoroacetylation and *O*-acetylation afforded methyl (methyl 4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-5-trifluoroacetamido-β-D-*glycero*-D-*galacto*-2-nonulopyranosid)onate (**30**), which was converted into the benzyl glycosides (**32** and **33**) via the 2-bromide (**31**). A simplified preparation of the protected 2-halogeno derivatives of Neu5Ac and 4-*epi*-Neu5Ac is described. The conversion of neuraminic acid methyl glycoside into the corresponding 2-bromide derivative by the action of hydrogen bromide is demonstrated.

INTRODUCTION

X-ray analysis of the complex of the hemagglutinin (HA) of the influenza virus with sialyl-lactose revealed[2] three substantial binding sites of 5-acetamido-3,5-di-

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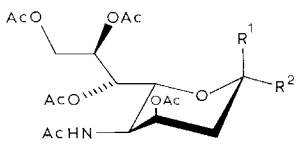
deoxy-D-*glycero*-D-*galacto*-2-nonulopyranosonic acid (Neu5Ac), namely, the trihydroxypropyl fragment C-7,8,9, NAc-5, and the hydroxycarbonyl function C-1,2. According to these data, HO-4 and the aglycon moiety project into the water phase and do not participate in binding with HA. The three-dimensional structure of the complex has been resolved only for two viral strains, both of the H3N2 subtype. However, other types, subtypes, and strains of influenza virus may not necessarily have the same subsite specificity and it is of interest to investigate and compare the fine specificity of the various viral HA. In this context, we now report the synthesis of neuraminic acid derivatives modified at C-2, C-4, and C-5.

Benzyl[3] and substituted benzyl[4] glycosides of α-Neu5Ac are considerably more active inhibitors in binding with HA than methyl glycosides. Therefore, benzyl glycosides modified at C-5 and C-4, namely, amino, *N*-trifluoroacetyl, and 4-*epi*-derivatives, have been synthesised. Considering the known[5] resistance of 4-*O*-acetylated neuraminic acid glycosides towards neuraminidases and the non-participation of HO-4 with HA, it was anticipated that α-glycosides of 5-acetamido-3,5-dideoxy-D-*glycero*-D-*talo*-2-nonulopyranosonic acid (4-*epi*-Neu5Ac) would also be resistant to neuraminidase whilst still binding to HA. For the purposes of comparison, β-glycosides and glycals of Neu5Ac and 4-*epi*-Neu5Ac were also prepared as compounds with an altered position of the carboxylic group which plays a substantial role[2] in binding with HA of influenza virus.

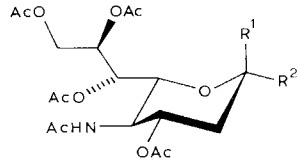
The results on the adhesion of the compounds described to the influenza virus have been reported[6].

RESULTS AND DISCUSSION

After crystallisation of Neu5Ac from the products of the Kuhn–Bashang synthesis[7] (i.e., condensation of 2-acetamido-2-deoxy-D-mannose with the potassium salt of the di-*tert*-butyl ester of oxalacetic acid in methanol followed by decarboxylation of the resulting lactones), the residual mixture of isomeric nonulosonic acids was methanolysed and then treated with acetic anhydride in pyridine. Chromatography of the products on silica gel gave the pure α and β derivatives of 4-*epi*-Neu5Ac (**1** and **2**) and of Neu5Ac (**3** and **4**).



1 and 2



3 and 4

1 and 3 R^1 = COOMe, R^2 = OAc

2 and 4 R^1 = OAc, R^2 = COOMe

TABLE I

^{1}H NMR data [a] for methyl esters of acetylated Neu5Ac and 4-*epi*-Neu5Ac

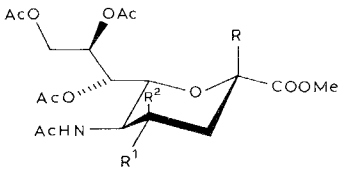
Compound	H-3*ax* (dd)	H-3*eq* (dd)	H-4 (ddd)	H-5 (ddd)	H-6 (dd)	H-7 (dd)	H-8 (ddd)	H-9a (dd)	H-9b (dd)
1	2.16	2.65	5.12	4.36	5.06	5.42	5.28	4.09	4.44
2	2.23	2.68	5.09	4.45	4.39	5.44	5.09	4.15	4.57
3 [b]	2.08	2.57	5.02	4.16	4.70	5.58	5.20	4.36	4.06
4 [b]	2.10	2.55	5.00	4.17	4.08	5.38	5.08	4.50	4.12

	$J_{3eq,3ax}$	$J_{3eq,4}$	$J_{3ax,4}$	$J_{4,5}$	$J_{5,6}$	$J_{6,7}$	$J_{7,8}$	$J_{8,9a}$	$J_{8,9b}$	$J_{9a,9b}$
1	15	3	3	3	11	2.2	6	2.2	6.5	12.7
2	15.8	3	3.5	3	10.5	2.3	4.3	2.2	7.3	12.5
3 [b]	13.5	4.7	12.0	10.3	n.d. [c]	2.5	7.0	2.5	5.7	12.5
4 [b]	13.2	5.0	11.5	n.d.	n.d.	1.5	5.3	2.6	6.6	12.5

[a] See Experimental for data on NH, COOMe, and Ac. [b] Data from ref. 8. [c] Not determined.

The structures of **1** and **2** were confirmed by comparison of their ^{1}H NMR data, particularly the $J_{3ax,4}$, $J_{3eq,4}$, and $J_{5,6}$ values, with those[8] of **3** and **4** (see Table I), which also indicated a $^2C_5$ conformation of the pyranose ring. It was not possible to ascertain anomeric configurations on the basis of the ^{1}H NMR data, and the anomer with the higher mobility in TLC was assumed to be α, as for the corresponding Neu5Ac derivatives **3** and **4**. Two different strategies for the synthesis of 4-*epi*-Neu5Ac have been reported[9]. The 2-bromo and 2-chloro derivatives of Neu5Ac (**5** and **6**) and 4-*epi*-Neu5Ac (**7** and **8**) were obtained by a simplified mild procedure[10] for the preparation of unstable acetohalogenoses, which involved the reaction of **1**–**4** severally with hydrogen chloride (or bromide) generated from acetyl chloride (or bromide) and methanol in chloroform or dichloromethane. As also found by Paulsen and Tietz[11], who used $TiCl_4$ or $TiBr_4$ to obtain **5** and **6**, there is no need to employ drastic conditions such as acetyl chloride saturated with hydrogen chloride. The prerequisite for the reaction to proceed under mild conditions is for the starting acetate to be the fully acetylated methyl ester of Neu5Ac (**3** or **4**). Marra and Sinaÿ[8] confirmed the earlier finding[12] that the product obtained by the action of acetic anhydride on Neu5Ac methyl ester in the presence of perchloric acid differed from that obtained in the presence of pyridine in having HO-2 unsubstituted.

The ^{1}H NMR data for **5**–**8** in the protonated and unprotonated forms are given in Table II. The bromide **5** and the *epi*-chloride **8** are stable in both forms, whereas the *epi*-bromide **7** is unstable and yields the glycal **9** and the products of its reaction with hydrogen bromide (^{1}H NMR data). The ^{1}H NMR spectra of protonated **5** and **7**, obtained after concentration of the reaction mixture, and those of the samples obtained after treatment with anhydrous sodium acetate before concentration, showed that NAc-5 in the former compound was protonated. Thus, the signal (d) for NH at 5–6 ppm was absent, that (ddd, $J_{5,NH}$ 10 Hz) for H-5 was shifted upfield by 0.07–0.16 ppm, and those of H-4 and H-6 were shifted



	R	R^1	R^2
5	Br	OAc	H
6	Cl	OAc	H
7	Br	H	OAc
8	Cl	H	OAc

downfield by 0.09–0.2 and 0.32–0.36 ppm. After deprotonation, the NH doublet returns to its normal position (5–6 ppm) and the NAc signal is shifted upfield from 2.00 to 1.93–1.96 ppm. The analogous relationship was observed for the protonated and unprotonated forms of the *epi*-glycal **9** (see Table II).

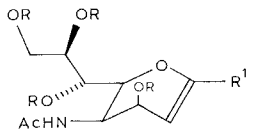
TABLE II

^{1}H NMR data for the protonated and unprotonated forms of 2-halogeno derivatives and glycals of Neu5Ac and 4-*epi*-Neu5Ac

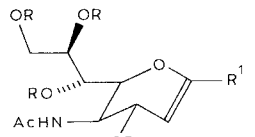
Compound	H-3*ax* (dd)	H-3*eq* (dd)	H-4 (ddd)	H-5 (ddd)	H-6 (dd)	H-7 (dd)	H-8 (ddd)	H-9a (dd)	H-9b (dd)	NH (d)
5	2.25	2.92	5.44	4.24	4.29	5.49	5.17	4.43	4.07	5.53
5 [a]	n.d.	2.99	5.54	4.17	4.65	5.50	5.23	4.41	4.10	n.d. [f]
6 [b]	2.28	2.80	5.40	4.20	4.30	5.47	5.18	4.40	4.08	5.60
7	2.44	3.13	5.08	4.49	4.50	5.54	5.21	4.50	4.11	5.64
7 [a]	n.d.	3.08	5.22	4.36	4.86	5.56	5.27	4.49	4.12	n.d.
8	2.41	2.97	5.07	4.46	4.65	5.54	5.23	4.51	4.11	5.59
9 [a]	6.17 [c]		5.36 [d]	4.44	4.61	5.55	5.43	4.72	4.20	n.o.
9	6.22 [c]		5.16 [d]	4.60	4.29	5.51	5.33	4.79	4.20	5.54
11	6.01 [c]		5.51 [d]	4.40	4.40	5.51	5.37	4.60	4.20	5.53

	$J_{3ax,3eq}$	$J_{3ax,4}$	$J_{3eq,4}$	$J_{4,5}$	$J_{5,6}$	$J_{6,7}$	$J_{7,8}$	$J_{8,9a}$	$J_{8,9b}$	$J_{9a,9b}$	$J_{NH,5}$
5	14	11	5	10	11	2	7	3	6	12.5	10
5 [a]	14	n.d.	4.5	10	11	2	8	2.5	6	12.5	10
6 [b]	13.8	10.6	4.9	10.7	10.4	2.1	6.8	2.5	5.9	12.5	–
7	16.5	3.5	2.5	3	11	2	6.5	n.d.	6	12.5	10
7 [a]	16	n.d.	2	2.5	11	1.5	6	2	6	12.5	10
8	16	3.5	2.5	3	11	2	6.5	2.5	6	12.5	10
9 [a]	6 [e]			4	11	2	n.d.	2.5	7.5	12.5	10
9	6 [e]			4	11	2	4.5	2.5	7.5	12.5	10.5
11	6 [e]			9	n.d.	2.5	5	3	7	12.5	10

[a] Protonated forms. [b] Data from ref. 26. [c] H-3 (d). [d] H-4 (dd). [e] Data for $J_{3,4}$. Additional signals: 3.81–3.90 (COOMe), 1.93–1.96 (NAc of the unprotonated forms), 2.05–2.17 (OAc and NAc of the protonated forms). [f] Not determined.



9 and 10



11 and 12

9 and 11 R = Ac, R^1 = COOMe

10 and 12 R = H, R^1 = COOH

Treatment of the bromide **5** with methanol containing *sym*-collidinium salt gave 89% of a mixture of the methyl glycosides **13** and **14** with an α,β-ratio of 9:1 (^{1}H NMR data). Zemplén *O*-deacetylation of the mixture followed by saponification and chromatography gave the methyl α- (**15**, 75%) and β-glycoside (**16**, 7%), which were characterised by ^{1}H NMR spectroscopy (see Experimental). Replacement of methanol by benzyl alcohol in the above reaction gave the benzyl α- (**17**, 63%) and β-glycoside (**18**, 32%), which, together with the respective deprotected derivatives **19** and **20**, were characterised by their ^{1}H NMR spectra (see Tables III and IV). The chloride **6** did not react with benzyl alcohol under these conditions.

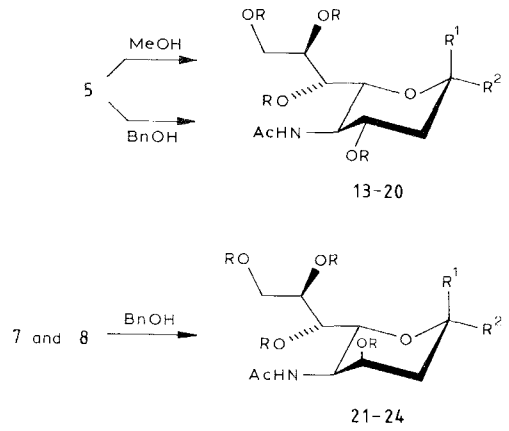
These glycosylations with simple alcohols are adaptations of the methyl 1,2-*trans*-glycosylation procedure[13], except that *sym*-collidinium salts are present, which may affect the α-selectivity. In a series of experiments (details not given), **5**, **7**, and **8** were reacted variously with benzyl alcohol in dichloromethane in the presence (*A*) and absence (*B*) of collidinium salts and in the presence of *sym*-collidine (*C*) and silver carbonate (*D*, Koenigs–Knorr) and the results are shown in Table IV. The results demonstrated a great tendency for the *epi*-bromide **7** to form

TABLE III

^{1}H NMR data [a] for the protected benzyl glycosides of Neu5Ac and its analogues

Compound	H-3*ax* (dd)	H-3*eq* (dd)	H-4 (ddd)	H-5 (ddd)	H-6 (dd)	H-7 (dd)	H-8 (ddd)	H-9a (dd)	H-9b (dd)	CHa (d)	CHb (d)
17	n.d. [b]	2.67	4.89	4.10	4.15	5.36	5.47	4.34	4.12	4.82	4.44
18	1.93	2.56	5.30	4.15	4.00	5.41	5.28	4.85	4.13	4.56	4.49
21	n.d.	2.84	5.11	4.36	4.49	5.41	5.53	4.43	4.15	4.79	4.36
22	2.02	2.72	4.97	4.38	4.38	5.46	5.29	4.89	4.16	4.60	4.41
32	n.d.	2.73	5.03	4.00	4.32	5.32	5.48	4.32	4.15	4.83	4.45
33	1.95	2.62	5.45	4.08	4.17	5.36	5.33	4.77	4.14	4.57	4.52
	$J_{3ax,3eq}$	$J_{3ax,4}$	$J_{3eq,4}$	$J_{4,5}$	$J_{5,6}$	$J_{6,7}$	$J_{7,8}$	$J_{8,9a}$	$J_{8,9b}$	$J_{9a,9b}$	$J_{CHa,Hb}$
17	12.5	12	4.5	10	10.5	2	8	3	6	12.5	12
18	13	11.5	5	10	11	2	4	3	8	12.5	12
21	15	3	3.5	3	11	2	8	2.5	5.5	12.5	12
22	15.5	3.5	3	n.d.	n.d.	2	4	2	8	12.5	11.5
32	13	n.d.	5	10	10	2	9	2	5	12.5	11.5
33	13	13	5	10	10	2	4.5	2	6.5	12.5	12

[a] See Experimental for data on NH, Ph, COOMe, and Ac. [b] Not determined.



	R	R^1	R^2
13	Ac	COOMe	OMe
14	Ac	OMe	COOMe
15	H	COOH	OMe
16	H	OMe	COOH
17	Ac	COOMe	OBn
18	Ac	OBn	COOMe
19	H	COOH	OBn
20	H	OBn	COOH
21	Ac	COOMe	OBn
22	Ac	OBn	COOMe
23	H	COOH	OBn
24	H	OBn	COOH

the glycal **9**, which is produced even on treatment of the *epi*-acetates (**1** and **2**) with hydrogen bromide (see above). The Koenigs–Knorr condensation gave the highest α,β-ratio for the *epi*-chloro derivative (entry 5). The most convenient procedure to achieve the highest α,β-ratio from the *epi*-bromide **7** was *B* (compare entries 3 and 4). Column chromatography of the products for entry 6 gave 88% of an inseparable 52:36 mixture of the α anomer **21** and the glycal **9**, and 7% of the β-glycoside **22**, identified by their ^{1}H NMR spectra (see Tables II and III). Deprotection of the mixture of **21** and **9** and column chromatography gave the α-glycoside **23** and the glycal **10**. Deprotection of **22** gave the β-glycoside **24**. The benzyl β-glycoside (**20**) of Neu5Ac, obtained by Faillard et al.[14] by the reaction of Neu5Ac penta-acetate with benzyl alcohol in the presence of $ZnCl_2$ at 130–150°C and deprotection of the product, was characterised as neuraminidase-resistant. The ^{1}H NMR data of the protected (**17**, **18**, **21**, and **22**) and unprotected (**19**, **20**, **23**, and **24**) benzyl glycosides of Neu5Ac and 4-*epi*-Neu5Ac are listed in Tables III and V. For the purposes of comparison, the protected glycal **11** and its unprotected congener **12** were prepared. The ^{1}H NMR data for **11** and **12** and their 4-*epi*-analogues (**9** and **10**) are given in Tables II and VI. The main differences in the spectra of **9** and **11** are the position of the H-4 signal (dd) at 5.16 and 5.506 ppm respectively. The $J_{4,5}$ values (4 and 9 Hz, respectively) demonstrate the H-4*eq*,5*ax* and H-4*ax*,5*ax* relationships and confirm the configuration of the 4-*epi*-glycal (**9**).

The assignments of the anomeric configurations of the benzyl glycosides of *epi*-Neu5Ac in the $^2C_5$ conformation were made on the assumption that the relationship between the signals for H-3*eq* and H-3*ax*, which are indicative[15,16] of the configuration of the glycosidic linkage of Neu5Ac, is identical with that of 4-*epi*-Neu5Ac.

The *N*-trifluoroacetyl benzyl glycosides **27** (77%) and **28** (80%) were obtained from the corresponding *N*-acetyl derivatives (**17** and **18**) by hydrolysis under basic

TABLE IV

Glycosidation of 2-halogeno derivatives of Neu5Ac and 4-*epi*-Neu5Ac with benzyl alcohol–dichloromethane

Entry	Halide	Procedure [a]	α,β-ratio	Yield (%) [b]		
				α-Anomer	β-Anomer	Glycal
1	**5**	*A*	2:1	63	32	–
2	**5**	*C*	1.3:1	40	30	–
3	**7**	*A*	1:1	38	39	23
4	**7**	*B*	3:1	56	18	26
5	**8**	*D*	14:1	82	6	12
6	**7**	*D*	7:1	52	7	36

[a] See Results and Discussion. [b] Isolated yield for entries 1, 2, and 6, and based on the ^{1}H NMR spectra of the mixture after deprotection for entries 3–5.

TABLE V

^{1}H NMR data for the unprotected benzyl glycosides of Neu5Ac and its analogues (Na^+ salts)

Compound	H-3*ax* (dd)	H-3*eq* (dd)	H-4 (ddd)	H-5 (dd)	H-6 (dd)	H-7 (dd)	H-8 (ddd)	H-9a (dd)	H-9b (dd)	CHa (d)	CHb (d)	NAc (s)	Ph (m)
19	1.70	2.80	3.71	3.84	3.75	3.61	3.82	3.87	3.65	4.77	4.54	2.05	7.44
20	1.69	2.41	4.06	3.92	3.99	3.58	3.96	3.86	3.69	4.62	4.30	2.05	7.46
23	1.93	2.67	4.19	4.07	4.41	3.61	3.86	3.88	3.66	4.74	4.47	2.04	7.42
24	1.98	2.31	4.07	4.13	4.24	3.58	3.98	3.85	3.68	4.67	4.32	2.02	7.44
27 [a]	1.77	2.79	3.82	4.02	4.00	3.57	3.84	3.86	3.63	4.79	4.57	–	7.43
28 [a]	1.71	2.43	4.17	4.05	4.17	3.54	3.96	3.87	3.67	4.62	4.30	–	7.47
25	1.72	2.83	3.78	3.20	4.02	3.80	3.87	3.89	3.72	4.76	4.55	–	7.44

	$J_{3ax,3eq}$	$J_{3ax,4}$	$J_{3eq,4}$	$J_{4,5}$	$J_{5,6}$	$J_{6,7}$	$J_{7,8}$	$J_{8,9a}$	$J_{8,9b}$	$J_{9a,9b}$	$J_{Ha,Hb}$
19	12.5	12.5	4.5	10	10	1.5	9	2.5	6	12	11
20	13	12	5	10	10.5	< 2	9	2.5	5.5	12	10
23	14	3	3.5	3	11	< 2	9	2	6	12	11
24	15	3.5	3	3	11	< 2	9	3	6	12	11
27 [a]	12	12	5	10	10.5	2	9	3	6	12	11
28 [a]	12.5	12.5	5	10	10.5	< 2	10	2.5	6	12	10
25	12.5	12.5	5	10	10	2	9	2.5	5.5	12	11

[a] Free acids.

TABLE VI

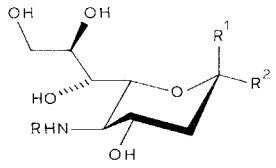
^{1}H NMR data for deprotected glycals (**10** and **12**) of 4-*epi*-Neu5Ac and Neu5Ac (Na^+ salts)

Compound	H-3 (d)	H-4 (dd)	H-5 (dd)	H-6 (dd)	H-7 (dd)	H-8 (ddd)	H-9a (dd)	H-9b (dd)	NAc (s)
10	5.88	4.26	4.22	4.22	3.64	4.00	3.91	3.67	2.08
12	5.67	4.45	4.02	4.19	3.59	3.92	3.86	3.63	2.05

	$J_{3,4}$	$J_{4,5}$	$J_{5,6}$	$J_{6,7}$	$J_{7,8}$	$J_{8,9a}$	$J_{8,9b}$	$J_{9a,9b}$
10	6	3	n.d. [a]	< 1	9	2.5	6.5	12
12	2.5	8.5	11	< 2	9	3	6	12

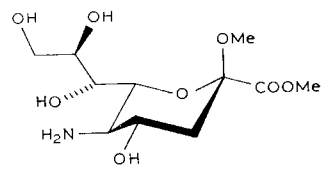
[a] Not determined.

conditions (to **25** and **26**) followed by treatment with methyl trifluoroacetate and methanol in the presence of triethylamine.

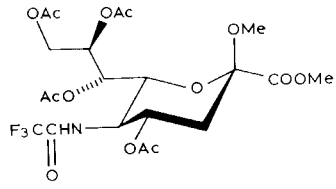


	R	R^1	R^2
25	H	COOH	OBn
26	H	OBn	COOH
27	$COCF_3$	COOH	OBn
28	$COCF_3$	OBn	COOH

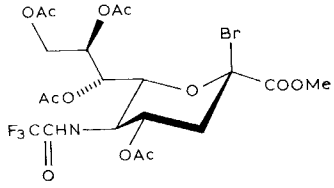
The protected derivatives **32** and **33** were prepared by another route. The known[17] amine **29**, obtained by methanolysis of Neu5Ac, was *N*-trifluoroacetylated, as described above, then *O*-acetylated to give the methyl glycoside **30**. Treatment of **30** with an excess of hydrogen bromide in chloroform gave the 2-bromo derivative **31**, which, without isolation, was reacted with an excess of benzyl alcohol in the presence of *sym*-collidine to give a 1:2 mixture (32% from **30**, not optimised) of the α- (**32**) and β-glycoside (**33**) from which the components were isolated by column chromatography. The ^1H NMR data of the protected and unprotected *N*-trifluoroacetyl derivatives are listed in Tables III and V, respectively. Comparison of the pairs of protected anomeric benzyl glycosides **17–18**, **21–22**, and **32–33** reveals $J_{7,8}$ values of 8–9 Hz for the α anomers and 4–4.5 Hz for the β anomers, indicative of different conformations of the C-7,8,9 moieties. The analogous observation was made for the neuraminyl oligosaccharides[16] and the anomeric *p*-substituted benzyl glycosides[1] of Neu5Ac. For the pairs of unprotected derivatives, the $J_{7,8}$ value is 9–10 Hz for each anomer. For the protected anomers, the chemical shifts of the resonances of the benzyl CH_2 group differed by ~ 0.4 ppm for the α anomers and 0.05–0.2 ppm for the β anomers.



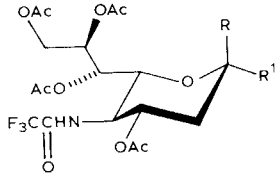
29



30



31



32 R = COOMe, R^1 = OBn

33 R = OBn, R^1 = COOMe

EXPERIMENTAL

General methods.—The ^{1}H NMR spectra (δ in ppm, relative to Me_4Si) were recorded with a WM-500 Bruker spectrometer for solutions in $CDCl_3$ (protected derivatives) and D_2O (unprotected derivatives), and assignments were confirmed by spin decoupling techniques. HPLC was performed on a column of Partisil 10 ODS-3 (Whatman) with a water–acetonitrile gradient and UV detection (200–254 nm). Optical rotations were measured with a DIP-360 (JASCO) polarimeter. Column chromatography was performed on Silica Gel L 40–100 μm (Czechoslovakia) and TLC on Kieselgel 60 (Merck, 5553), using *A*, 1-propanol–acetone–water (4:3:2); *B*, ethyl acetate; 1-propanol–ethyl acetate–water, 2:3:1 (*C*) and 4:3:2 (*D*); *E*, toluene–ethyl acetate, 1:2; hexane–chloroform–2-propanol, 4:2:1 (*F*) and 8:4:1 (*G*); *H*, chloroform–2-propanol, 25:1; *I*, toluene–ethyl acetate, 3:1; and detection by charring with H_3PO_4. Organic solutions were dried by filtration through a cotton plug. Satisfactory elemental analyses were not obtained for amorphous compounds, but their purity was proved by TLC and ^{1}H NMR data.

*Methyl 5-acetamido-2,4,7,8,9-penta-*O*-acetyl-3,5-dideoxy-α- and -β-*D-glycero-D-talo-*2-nonulopyranosonate* (**1** *and* **2**).—From the mixture (16 g) of nonulosonic acids obtained as described[7], Neu5Ac (12 g) was isolated by crystallisation from acetic acid. The mother liquors were concentrated and the residue, which contained ~30% of Neu5Ac and ~20% of 4-*epi*-Neu5Ac [TLC, R_F 0.47 and 0.60 (solvent *A*)] was treated with methanolic HCl and the product was acetylated as described[1]. Column chromatography (toluene with 20 → 70% of EtOAc) gave the mixtures **1** + **3** and **2** + **4** [R_F 0.44 and 0.35, respectively (solvent *B*)]. Each pair was subjected to repeated column chromatography [hexane–$CHCl_3$ (2:1) with 2 → 10% of 2-propanol] to give pure **3**, **1**, **4**, and **2** [R_F 0.41, 0.31, 0.36, and 0.23, respectively (solvent *F*)].

Compound **1** was amorphous and had $[\alpha]_D^{20}$ −5.2° (*c* 1.5, $CHCl_3$). ^{1}H NMR data: Table I and δ 1.92, 2.05, 2.06, 2.08, 2.13, 2.14 (6 s, each 3 H, 6 Ac), 5.43 (d, 1 H, $J_{NH,5}$ 9.5 Hz, NH), 3.74 (s, 3 H, COOMe).

Compound **2** was amorphous and had $[\alpha]_D^{20}$ −51° (*c* 1.0, $CHCl_3$). ^{1}H NMR data: Table I and δ 1.92, 2.05, 2.09, 2.11, 2.13, 2.15 (6 s, each 3 H, 6 Ac), 5.42 (d, 1 H, $J_{NH,5}$ 10 Hz, NH), 3.79 (s, 3 H, COOMe).

*Methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-2,6-anhydro-3,5-dideoxy-*D-glycero-D-talo-*non-2-enonate* (**9**).—To a solution of **1** (116 mg, 0.2 mmol) in dry $CHCl_3$ (4 mL) containing acetyl bromide (600 μL, 8.1 mmol) at 0°C was added MeOH (160 μL, 4 mmol), and the mixture was stored for 2.5 h at 10°C, then concentrated to dryness in vacuo. A solution of the residue in dry CH_2Cl_2 (4 mL) was treated with *sym*-collidine (0.9 mL) at room temperature for 15 h, diluted with $CHCl_3$, washed with 2 M HCl, H_2O, satd aq $NaHCO_3$, and H_2O, filtered, dried, and concentrated. Column chromatography ($CHCl_3$, with 0.5 → 2% of 2-propanol) of the residue gave amorphous **9** (77 mg, 81%), R_F 0.38 (solvent *H*), $[\alpha]_D^{20}$ −120° (*c* 1.5, $CHCl_3$): lit.[18] mp 95–97°C, $[\alpha]_D^{30}$ −121°. ^{1}H NMR data: Table II and δ 1.96, 2.07–2.12 (5 Ac), 3.81 (s, 3 H, COOMe).

*Methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-2,6-anhydro-3,5-dideoxy-*D-glycero-D-galacto-*non-2-enonate* (**11**).—To a solution of the mixture (100 mg, 0.187 mmol) of **3** and **4** in dry $CHCl_3$ (2 mL) containing acetyl bromide (420 μL, 5.7 mmol) at −10°C was added MeOH (76 μL, 1.9 mmol). The mixture was stored at 10°C for 2.5 h, then concentrated. The residue was treated with $CHCl_3$ (2 mL) and 1,8-diazabicyclo[5,4,0]undec-7-ene (DBU, 56 μL) for 2 h at room temperature. The mixture was worked-up as described above. Column chromatography (toluene–EtOAc) of the product gave amorphous **11** (70 mg, 79%), R_F 0.36 (solvent *B*), $[\alpha]_D^{20}$ +68° (*c* 0.5, $CHCl_3$); lit. [19] mp 126–127°C, $[\alpha]_D$ +79.9° ($CHCl_3$). ^{1}H NMR data: Table II and δ 1.95, 2.07, 2.08, 2.10, 2.14 (5 s, each 3 H, 5 Ac), 3.82 (s, 3 H, COOMe).

*Sodium 5-acetamido-2,6-anhydro-3,5-dideoxy-*D-glycero-D-galacto-*non-2-enonate* (**12**).—A solution of **11** (50 mg, 0.106 mmol) in methanolic 0.2 M NaOMe (6 mL) was stored at room temperature for 1 h. Water (2 mL) was added, and, after 2 h, the solvents were evaporated. A solution of the residue in water (2 mL) was treated with KU-2 (H^+) resin (1.5 mL), then concentrated. A solution of the residue in water was neutralised with 0.2 M NaOH and concentrated. Elution of the residue from a column of Silica Gel 60 (6 g, 63–100 mesh, Merck) with 2-propanol–EtOAc–H_2O (2:2:1 → 4:3:2) gave amorphous **12** (28 mg, 85%), R_F 0.56 (solvent *D*), $[\alpha]_D^{20}$ +31° (*c* 0.5, H_2O); lit.[20] for the free acid, mp 137–140°C, $[\alpha]_D$ +41.6° (H_2O).

*Methyl (methyl 5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-α,β-*D-glycero-D-galacto-*2-nonulopyranosid)onate* (**13** *and* **14**).—To a solution of **4** (213 mg, 0.4 mmol) in CH_2Cl_2 (4 mL) at 0°C was added acetyl bromide (185 μL, 2.5 mmol) followed by a solution of MeOH (100 μL, 2.5 mmol) in CH_2Cl_2 (1 mL). The mixture was stored at 0°C for 2 h, a solution of *sym*-collidine (278 μL, 2.1 mmol) in MeOH (5 mL) was added, and the mixture was stored at 0°C for 2 h. Pyridine (100 μL) was added, and the mixture was diluted with $CHCl_3$ (30 mL), washed with aq 1% HCl, H_2O, aq $NaHCO_3$, and H_2O, dried, and concentrated in vacuo to afford a crude mixture (180 mg, 89%) of **13** and **14** in the ratio 9:1 (^{1}H NMR data), R_F 0.36 (solvent *B*).

*Sodium (methyl 5-acetamido-3,5-dideoxy-α- and -β-*D-glycero-D-galacto-*2-nonulopyranosid)onate* (**15** and **16**).—To a solution of the above mixture (100 mg) of **13** and **14** in MeOH (4.5 mL) was added methanolic 2 M NaOMe (0.5 mL), and the mixture was stored for 1 h at room temperature, then concentrated. The residue was treated with 2 M NaOH (5 mL) for 16 h at room temperature, and the cooled solution was neutralised with KU-2 (H^+) resin, filtered, and concentrated. Column chromatography with 2-propanol–EtOAc–H_2O (2:2:1 → 4:3:2) of the residue on silica gel (6 g) gave, first, amorphous **15** (51 mg, 75%), R_F 0.49 (solvent *D*), $[\alpha]_D^{20}$ +3.6° (*c* 1.0, H_2O); lit.[21] for the free acid, $[\alpha]_D^{25}$ −13° (H_2O), and lit.[22] $[\alpha]_D$ −9.5° (H_2O). ^{1}H NMR data: δ 1.64 (dd, 1 H, $J_{3ax,4}$ 12 Hz, H-3*ax*), 2.04 (s, 3 H, NAc), 2.73 (dd, 1 H, $J_{3eq,3ax}$ 12.5, $J_{3eq,4}$ 5 Hz, H-3*eq*), 3.35 (s, 3 H, OMe), 3.60 (dd, 1 H, $J_{7,8}$ 9 Hz, H-7), 3.66 (dd, 1 H, $J_{9b,8}$ 6.5 Hz, H-9b), 3.69 (ddd, 1 H, H-4), 3.72,

(dd, 1 H, $J_{6,5}$ 10, $J_{6,7}$ 1.5 Hz, H-6), 3.82 (dd, 1 H, H-5), 3.88 (dd, 1 H, $J_{9a,9b}$ 12, $J_{9a,8}$ 2 Hz, H-9a), 3.90 (ddd, 1 H, H-8).

Eluted second was amorphous **16** (5 mg, 7%), R_F 0.33 (solvent *D*), $[\alpha]_D^{20}$ −44° (*c* 1, H_2O); lit.[22] for the free acid, $[\alpha]_D$ −46° (H_2O). ^1H NMR data: 1.68 (dd, 1 H, $J_{3ax,4}$ 12 Hz, H-3*ax*), 2.06 (s, 3 H, NAc), 2.35 (dd, 1 H, $J_{3eq,3ax}$ 13, $J_{3eq,4}$ 5 Hz, H-3*eq*), 3.23 (s, 3 H, Me), 3.58 (dd, 1 H, $J_{7,8}$ 9 Hz, H-7), 3.67 (dd, $J_{9b,8}$ 6 Hz, H-9b), 3.81 (dd, 1 H, $J_{6,5}$ 10, $J_{6,7}$ 2 Hz, H-6), 3.84 (dd, 1 H, $J_{9a,9b}$ 12, $J_{9a,8}$ 2.5 Hz, H-9a), 3.90 (dd, 1 H, $J_{5,4} = J_{5,6} = 10$ Hz, H-5), 3.91 (ddd, 1 H, H-8), 4.02 (ddd, 1H, H-4).

Methyl (benzyl 5-acetamido-4,7,8,9-tetra-O*-acetyl-3,5-dideoxy-α- and -β-*D-glycero-D-galacto-*2-nonulopyranosid)onate* (**17** and **18**).—To a solution of **4** (534 mg, 1 mmol) and acetyl bromide (370 μL, 5 mmol) in CH_2Cl_2 (8 mL) at 0°C was added benzyl alcohol (415 μL, 4 mmol), and the mixture was stored at 0°C for 2 h to give bromide **5** [R_F 0.47 (solvent *B*); R_F 0.33 for **4**]. A solution of *sym*-collidine (530 μL, 4 mmol) in benzyl alcohol (10 mL) was added, the mixture was stored for 16 h at room temperature, then diluted with $CHCl_3$, washed with H_2O, dried, and concentrated in vacuo. Column chromatography (2:1 hexane–$CHCl_3$ with 0.5 → 3% of 2-propanol) of the residue on silica gel (50 g) gave, first, amorphous **18** (185 mg, 32%), R_F 0.34 (solvent *G*), $[\alpha]_D^{20}$ −19° (*c* 1.5, $CHCl_3$); lit.[23] mp 135.5–138.5°C, $[\alpha]_D^{20}$ −13° (MeOH). ^1H NMR data: Table III and δ 1.87, 1.96, 2.01, 2.04, 2.17 (5 s, each 3 H, 5 Ac), 3.72 (s, 3 H, COOMe), 5.40 (d, 1 H, $J_{NH,5}$ 10 Hz, NH), 7.36 (m, 5 H, Ph).

Eluted second was amorphous **17** (364 mg, 63%), R_F 0.27 (solvent *G*), $[\alpha]_D^{20}$ −0.53° (*c* 1.5, $CHCl_3$); lit.[24] mp 85–89°C, $[\alpha]_D^{25}$ −3.5° (MeOH). ^1H NMR data: Table III and δ 1.91, 2.04, 2.06, 2.16, 2.18 (5 s, each 3 H, 5 Ac), 3.68 (s, 3 H, COOMe), 5.17 (d, 1 H, $J_{NH,5}$ 10 Hz, NH), 7.35 (m, 5 H, Ph).

*Sodium (benzyl 5-acetamido-3,5-dideoxy-α-*D-glycero-D-galacto-*2-nonulopyranosid)onate* (**19**).—To a solution of **17** (58 mg, 0.1 mmol) in dry MeOH (2 mL) was added methanolic 2 M NaOMe (200 μL), and the mixture was stored for 1 h at room temperature, then concentrated. A solution of the residue in 0.2 M NaOH (2 mL) was stored overnight at room temperature, then neutralised with KU-2 (H^+) resin, filtered, and concentrated in vacuo. HPLC of the residue gave amorphous **19** (39 mg, 93%), R_F 0.40 (solvent *C*), $[\alpha]_D^{20}$ −23° (*c* 1.5, H_2O); lit.[25] for the free acid, mp 162–164°C, $[\alpha]_D^{21}$ −16° (H_2O).

*Sodium (benzyl 5-acetamido-3,5-dideoxy-β-*D-glycero-D-galacto-*2-nonulopyranosid)onate* (**20**).—Treatment of **18**, as described for **19**, gave amorphous **20** (95%), R_F 0.3 (solvent *C*), $[\alpha]_D^{20}$ +8.3° (*c* 1.5, H_2O).

Methyl (benzyl 5-acetamido-4,7,8,9-tetra-O*-acetyl-3,5-dideoxy-α- and -β-*D-glycero-D-talo-*2-nonulopyranosid)onate* (**21** and **22**).—(*a*) *From the bromide* **7**. To a solution of **1** (133 mg, 0.25 mmol) in dry $CHCl_3$ (5 mL) containing acetyl bromide (740 μL, 10 mmol) at 0°C was added MeOH (200 μL, 5 mmol), and the mixture was stored for 2.5 h at 10°C, then concentrated to dryness. A solution of the residue in $CHCl_3$ (5 mL) was added to a stirred mixture of benzyl alcohol (520 μL, 5 mmol), silver carbonate (414 mg, 1.5 mmol), powdered molecular sieves 4A (1 g),

and CH_2Cl_2 (15 mL). The mixture was stirred for 19 h at room temperature, filtered, washed with M sodium thiosulphate and water, dried, and concentrated to dryness in vacuo. Column chromatography (2:1 hexane–$CHCl_3$ with $1 \rightarrow 4\%$ of 2-propanol) of the residue gave, first, amorphous **22** (10 mg, 7%), R_F 0.43 (solvent *F*), $[\alpha]_D^{20}$ −39° (*c* 0.5, $CHCl_3$). ^{1}H NMR data: Table III and δ 1.91, 1.92, 2.04, 2.15 (×2) (5 Ac), 3.79 (s, 3 H, COOMe), 5.52 (d, 1 H, $J_{NH,5}$ 10 Hz, NH), 7.38 (m, 5 H, Ph).

Eluted second was a 52:36 mixture (118 mg) of **21** and the glycal **9** (^{1}H NMR data), R_F 0.36 (solvent *F*). Based on the $[\alpha]_D^{20}$ of **9**, the $[\alpha]_D^{20}$ value ($CHCl_3$) of **21** was calculated to be −27°. ^{1}H NMR data of **21**: Table III and δ 1.94, 2.04–2.23 (15 H, 5 Ac), 3.60 (s, 3 H, COOMe), 5.30 (d, 1 H, $J_{NH,5}$ 10 Hz, NH), 7.34 (m, 5 H, Ph).

(*b*) *From the chloride* **8**. To a solution of **2** (10.7 mg, 0.02 mmol) in $CHCl_3$ (1 mL) containing MeOH (40 μL, 1 mmol) at 0°C was added acetyl chloride (142 μL, 2 mmol), and the mixture was stored at 10°C for 72 h, then concentrated in vacuo. A solution of the residue in $CHCl_3$ (1 mL) was added to a stirred mixture of benzyl alcohol (104 μL, 1 mmol), silver carbonate (28 mg, 0.1 mmol), molecular sieves 4A (100 mg), and $CHCl_3$ (1 mL). The mixture was stirred at room temperature for 48 h, filtered, washed with M sodium thiosulphate and water, dried, and concentrated in vacuo to give a crude 82:6:12 mixture of **21**, **22**, and **9**.

*Sodium (benzyl 5-acetamido-3,5-dideoxy-α-*D*-glycero-*D*-talo-2-nonulopyranosid)onate* (**23**) *and sodium 5-acetamido-2,6-anhydro-3,5-dideoxy-*D*-glycero-*D*-talo-non-2-enonate* (**10**).—A solution of the above mixture (60 mg) of **21** and **9** in methanolic 0.2 M NaOMe (5 mL) was stored at room temperature for 3 h. Water (1.5 mL) was added, the mixture was stored overnight, then concentrated, and a solution of the residue in water (2 mL) was applied to a column of KU-2 (H^+) resin (1.5 mL) and then concentrated. A solution of the residue in water was neutralised with 0.2 M NaOH and concentrated. Column chromatography (2-propanol–EtOAc–H_2O, $2:5:1 \rightarrow 2:3:1$) of the residue gave amorphous **23** (24 mg), R_F 0.40 (solvent *C*), $[\alpha]_D^{20}$ −60° (*c* 0.5, H_2O), and amorphous **10** (13 mg), R_F 0.22, $[\alpha]_D^{20}$ −133° (*c* 0.5, H_2O).

*Sodium (benzyl 5-acetamido-3,5-dideoxy-β-*D*-glycero-*D*-talo-2-nonulopyranosid)onate* (**24**).—Compound **22** (10 mg) was deprotected, as described in the foregoing experiment, to give amorphous **24** (6.5 mg, 90%), R_F 0.33 (solvent *C*), $[\alpha]_D^{20}$ −29° (*c* 0.35, H_2O).

*Benzyl 5-amino-3,5-dideoxy-α-*D*-glycero-*D*-galacto-2-nonulopyranosidonic acid* (**25**).—Compound **17** (58 mg, 0.1 mmol) was treated with methanolic 0.2 M NaOMe (3 mL) for 3 h. The solution was concentrated, and a solution of the residue in 2 M NaOH (3 mL) was heated in a sealed tube for 10 h at 110°C, the pH of the cooled solution was adjusted to 7 with 2 M HCl, and the solution was concentrated. A solution of the residue in water was passed through a column (2.5 × 40 cm) of TSK-Gel Toyopearl HW-40F, then subjected to column chro-

matography (2-propanol–EtOAc–H_2O, 2:5:1 → 2:3:1) to give amorphous **25** (31 mg, 87%), ninhydrin positive, R_F 0.58 (solvent *D*), $[\alpha]_D^{20}$ −41° (*c* 0.5, H_2O).

*Sodium (benzyl 3,5-dideoxy-5-trifluoroacetamido-α-*D*-glycero-*D*-galacto-2-nonulopyranosid)onate* (**27**).—A solution of **17** (87 mg, 0.15 mmol) in methanolic 0.2 M NaOMe (4 mL) was kept for 3 h at room temperature, then concentrated. A solution of the residue in 2 M NaOH (4 mL) was heated in a sealed tube at 110°C for 10 h, then cooled, the pH was adjusted to 7 with 2 M HCl, the solution was concentrated, and the residue was dried in vacuo. Dry MeOH (3 mL), triethylamine) (1 mL), and methyl trifluoroacetate (3 mL) were added to the residue, and the mixture was kept overnight at room temperature. The solution was evaporated to dryness and the residue was extracted with EtOH. The extract was filtered and concentrated, and a solution of the residue in water was applied to column (2 × 25 cm) of Sephadex A-25-DEAE equilibrated with 2 mM pyridinium acetate. The column was eluted with pyridinium acetate (linear gradient, 0.002 → 0.2 M), the eluate was concentrated, and the residue was dried in vacuo to afford the crude acid (58 mg, 85%), R_F 0.60 (solvent *C*), which was converted into the sodium salt and subjected to HPLC to give amorphous **27** (55 mg, 77%), $[\alpha]_D^{20}$ −25° (*c* 0.7, H_2O).

*Sodium (benzyl 3,5-dideoxy-5-trifluoroacetamido-β-*D*-glycero-*D*-galacto-2-nonulopyranosid)onate* (**28**).—Compound **18** was treated as described above for **17**, to give amorphous **28** (80%), R_F 0.53 (solvent *C*), $[\alpha]_D^{20}$ +5.4° (*c* 0.7, H_2O).

*Methyl (methyl 4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-5-trifluoroacetamido-β-*D*-glycero-*D*-galacto-2-nonulopyranosid)onate* (**30**).—Neu5Ac (77 mg, 0.25 mmol) was dissolved in a solution prepared by the addition of acetyl chloride (0.4 mL) to MeOH (5 mL) at 0°C, and then heated in a sealed tube at 100°C for 3 h as described[17]. The solution was then concentrated, MeOH was evaporated repeatedly from the residue to afford the crude amine **29**, to which were added methyl trifluoroacetate (3 mL), MeOH (3 mL), and triethylamine (0.4 mL). The mixture was stored for 3 h at room temperature, then concentrated, and pyridine (5 mL) was evaporated from the residue, which was then treated with acetic anhydride (3 mL) and pyridine (3 mL) at room temperature overnight. Methanol (2 mL) was added to the cooled solution, which, after 10 min, was concentrated, and a solution of the residue in $CHCl_3$ was washed with M HCl, H_2O, aq $NaHCO_3$, and H_2O, dried, and concentrated. Column chromatography (toluene with increasing proportions of EtOAc) of the residue on silica gel (25 g) gave amorphous **30** (50 mg, 36% based on Neu5Ac), R_F 0.3 (solvent *I*), $[\alpha]_D^{20}$ −8.6° (*c* 1, $CHCl_3$). ^1H NMR data: δ 1.90 (dd, 1 H, $J_{3ax,3eq} = J_{3ax,4} = 13$ Hz, H-3*ax*), 2.02, 2.04, 2.09, 2.16 (4 s, each 3 H, 4 Ac), 2.49 (dd, 1 H, $J_{3eq,4}$ 5 Hz, H-3*eq*), 3.28 (s, 3 H, OMe), 3.83 (s, 3 H, COOMe), 4.05–4.12 (m, 2 H, H-5,6), 4.15 (dd, 1 H, $J_{9b,9a}$ 12, $J_{9b,8}$ 7.5 Hz, H-9b). 4.79 (dd, 1 H, $J_{9a,8}$ 2.5 Hz, H-9a), 5.25 (m, 1 H, H-8), 5.39 (dd, 1 H, $J_{7,8}$ 5 Hz, H-7), 5.40 (m, 1 H, H-4), 6.79 (d, 1 H, $J_{NH,5}$ 9.5 Hz, NH).

*Methyl (benzyl 4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-5-trifluoroacetamido-α-* (**32**) *and -β-*D*-glycero-*D*-galacto-2-nonulopyranosid)onate* (**33**).—To a solution of **30** (17 mg,

0.03 mmol) and acetyl bromide (155 μL, 2.1 mmol) in $CHCl_3$ (0.3 mL) at 0°C was added MeOH (24 μL, 0.6 mmol). The mixture was stored at room temperature for 48 h during which time **30** reacted [TLC, R_F 0.3 (solvent *1*)] to give the bromide **31**, R_F 0.44. The mixture was cooled to 0°C and treated with a solution of *sym*-collidine (318 μL, 2.4 mmol) in benzyl alcohol (0.5 mL) for 30 min at room temperature. Pyridine (150 μL) was added, and the mixture was diluted with $CHCl_3$ (10 mL), washed with 2 M HCl, H_2O, aq $NaHCO_3$, and H_2O, dried, and concentrated in vacuo. Column chromatography (toluene with increasing proportions of EtOAc) of the residue on silica gel (3 g) gave, first, **33** (4 mg), R_F 0.43 (solvent *1*), then a ~4:1 mixture (2 mg) of **32** and **33**, R_F 0.38. The overall yield of **32** and **33** was 32%. ^1H NMR data: Table III and for **32**, δ 2.03, 2.05, 2.16, 2.19 (4 s, each 3 H, 4 Ac), 3.71 (s, 3 H, COOMe), 6.29 (d, 1 H, $J_{NH,5}$ 10 Hz, NH), 7.34 (m, 5 H, Ph); for **33**, δ 1.95, 2.02, 2.05, 2.17 (4 s, each 3 H, 4 Ac), 3.75 (s, 3 H, COOMe), 6.49 (d, 1 H, $J_{NH,5}$ 10 Hz, NH), 7.37 (m, 5 H, Ph).

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