ELSEVIER

Carbohydrate Research 272 (1995) 159–178

CARBOHYDRATE RESEARCH

Synthetic and structural studies of α-sialyl-(2 → 6) and α-sialyl-(2 → 3) 1-deoxynojirimycin derivatives potentially useful for biomedical applications ☆

Makoto Kiso *, Keiko Ando, Haruko Inagaki, Hideharu Ishida, Akira Hasegawa *

Department of Applied Bioorganic Chemistry, Gifu University, Gifu 501-11, Japan

Received 28 March 1994; accepted in revised form 22 December 1994

Abstract

Suitably protected derivatives of 1-deoxynojirimycin (1,5-dideoxy-1,5-imino-D-glucitol, DNJ) and its D-*galacto* analog were coupled with 2-thioglycosides of *N*-acetylneuraminic acid. The resulting disaccharides were converted into a variety of α-sialyl-(2 → 6)-and α-sialyl-(2 → 3)-DNJ derivatives, including the cyclic lactams 6-*O*-(5-acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylono-1′,5-lactam)-1,5-dideoxy-1,5-imino-D-glucitol and -D-galactitol. The structural features of the synthetic compounds were investigated by ion-spray mass and ^{1}H NMR spectrometry. The $^1C_4$ conformation of *N-tert*-butoxycarbonyl-DNJ, a synthetic intermediate having the *gluco* configuration, was confirmed by X-ray crystallography.

Keywords: Synthetic and structural studies; α-Sialyl-(2 → 6); α-Sialyl-(2 → 3); 1-Deoxynojirimycin derivatives; Biomedical applications

1. Introduction

Cell-surface sialoglycoconjugates participate in a variety of biological functions. In infection by influenza virus, for example, the recognition of sialic acid by hemagglutinin

☆ Synthetic Studies on Sialoglycoconjugates. Part 64. For Part 63, see ref. [1].

* Corresponding authors.

SSDI 0008-6215(95)00068-2

is an essential process [2,3], after which the sialic acid residue is cleaved by viral sialidase [4,5]. Similarly, *Trypanosoma cruzi* trypomastigotes, that cause Chagas' disease in humans, acquire sialic acid from host sialoglycoconjugates by means of a plasma membrane-associated trans-sialidase [6,7]. Our interest in the biology of sialic acid has led us to synthesize a series of gangliosides (sialoglycolipids) [8], and also to explore the preparation of conjugates of sialic acid with other biologically active molecules. In this connection our attention has been focussed on the potent glucosidase inhibitor deoxynojirimycin (DNJ) [9] and its analogs, some of which have been evaluated as potential antidiabetic, antitumor, and anti-HIV agents. As a continuation of our work on DNJ-containing sialo-oligosaccharides [10], we describe here synthetic and structural studies on novel α-sialyl-(2 → 6)- and α-sialyl-(2 → 3)-linked 1-deoxynojirimycin derivatives.

2. Results and discussion

Synthesis and conformation of the DNJ derivatives.—*N-tert*-Butoxycarbonyl (*N*-Boc, **1**) and *N*-benzyloxycarbonyl (*N*-Z, **2**) 1-deoxynojirimycin derivatives have served as convenient intermediates for the synthesis of various DNJ analogs as well as DNJ-containing oligosaccharides. As described in a previous paper [11], it was found that a series of *N*-Boc-and *N*-Z-DNJ derivatives have the $^1C_4$ conformation in solution, based on ^{1}H NMR analysis. To confirm this finding, we undertook the X-ray crystallographic analysis of the parent compound **1**, which deposited as well-formed plates from methanol–water solution. As shown in the resulting two molecular structures I and II in the asymmetric unit (Fig. 1), the substituents at C-2–C-5 of the piperidine ring are all axially disposed indicating that **1** has the $^1C_4$ conformation also in the crystal. The coplanar arrangement of C-1, C-5, N-5, C-7, O-7 and O-8 is a significant structural feature, which may be attributed to some double bond character [12] between the ring nitrogen (N-5) and the carbonyl carbon (C-7) in the carbamate structure. In fact, the

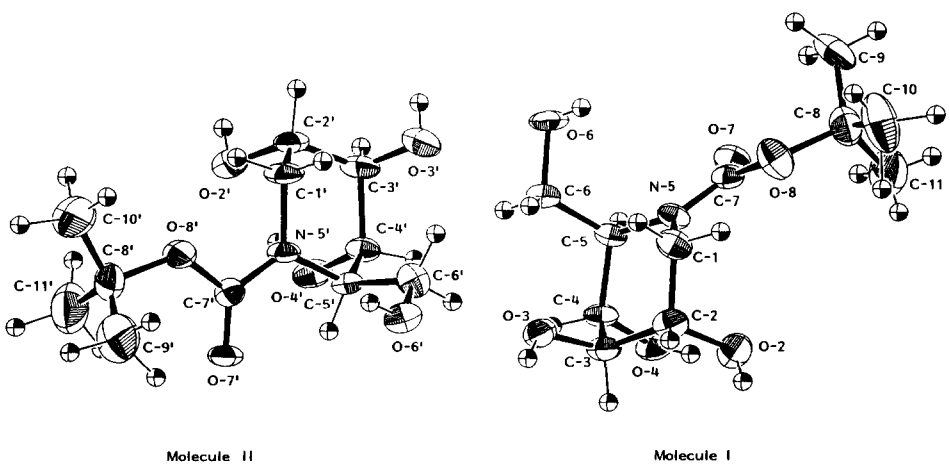
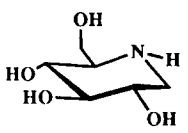
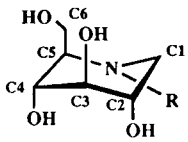


Fig. 1. Molecular structures I and II

C-7–N-5 bond length is 1.349 Å for molecule I and 1.331 Å for molecule II, much closer to the normal C = C double bond length of 1.33 Å than to the normal C–N single bond length of 1.47 Å. This molecular structure is basically the same as the minimum-energy conformation we calculated for **1** and **2** (see Experimental), suggesting considerable steric hindrance between the *N*-Boc or *N*-Z group and the vicinal substituent. The hydrogen bond between O*H*-4 and O-2 may be another factor in stabilizing the $^1C_4$ conformation. 4,6-*O*-Benzylidenation of **2**, however, caused a dramatic conformational change, from $^1C_4$ to $^4C_1$, to give **3** [11].

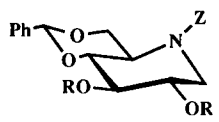


1-deoxynojirimycin
(DNJ)

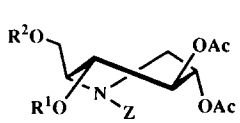


	R
1	Boc
2	Z

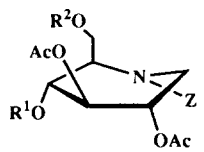
Boc = *tert*-butoxycarbonyl
Z = benzyloxycarbonyl



	R
3	H
4	Ac

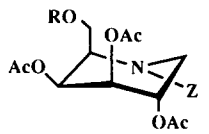


or



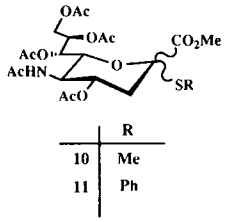
	R^1	R^2
5	H	H
6	H	TBDPS
7	Tf	TBDPS

TBDPS = *tert*-butyldiphenylsilyl
Tf = trifluoromethylsulfonyl

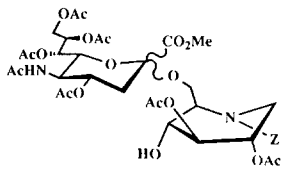


	R
8	TBDPS
9	H

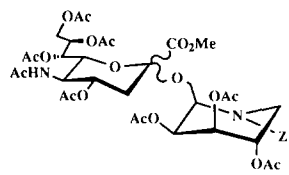
Acetylation of **3** and the following debenzylidenation gave **5**, which was selectively protected by the *tert*-butyldiphenylsilyl (TBDPS) group to afford **6** in high yield. The ^{1}H NMR data of **5** ($J_{1ax,2} = J_{1eq,2} = 3$–4, $J_{2,3}$ 5.7, and $J_{3,4}$ 7.3 Hz), and **6** ($J_{1ax,2}$ 3, $J_{1eq,2}$ 2, $J_{2,3}$ 4, $J_{3,4}$ 7, and $J_{4,5}$ 5.5 Hz) strongly suggest a significant conformational change involving flexible skew–boat type conformations. 4-*O*-Trifluoromethylsulfonylation of **6**, and treatment of the resulting **7** with cesium acetate in acetonitrile in the presence of 1,4,7,10,13,16-hexaoxacyclooctadecane (18-crown-6) at room temperature gave a 67% yield (two steps) of the *galacto* isomer **8**. The TBDPS group in **8** was then removed by treatment with BF_3 etherate in dichloromethane to give **9**. The preferred conformation of **8** and **9** seems to be near $^1C_4$ based on the ^{1}H NMR data ($J_{1ax,2} = J_{1eq,2} = 2$–4, $J_{2,3} = J_{3,4} = 3$–4, and $J_{4,5}$ 6 Hz).



	R
10	Me
11	Ph



12 α, β



13 α, β

Synthesis and structural analysis of the α-sialyl-(2 → 6)-DNJ derivatives.—The glycosylation of **5** with **10** [13] was performed with dimethyl(methylthio)sulfonium triflate (DMTST) [14] as a promoter in acetonitrile, to give **12**α,β in an 85% yield ($\alpha:\beta = 3:1$). The *galacto*-type DNJ derivative **9** was coupled with **10** in the presence of *N*-iodosuccinimide (NIS) and trifluoromethanesulfonic acid (TfOH) [15] in acetonitrile to afford an 80% yield of **13**α,β ($\alpha:\beta = 7:3$). When this coupling was carried out by using the phenyl 2-thioglycoside **11** [16] as the glycosyl donor, **13**α and **13**β were obtained in 53% and 22% yields, respectively (details not presented). The desired disaccharides **12**α and **13**α were each subjected to hydrogenolysis for 30 min over 10% palladium-on-carbon in methanol, followed by *O*-deacetylation and saponification of the methyl ester group, to give novel lactams **14** and **16** as the major products, respectively, together with the α-sialyl-(2 → 6)-DNJ derivatives **15**, **17**, and **18** (Scheme 1).

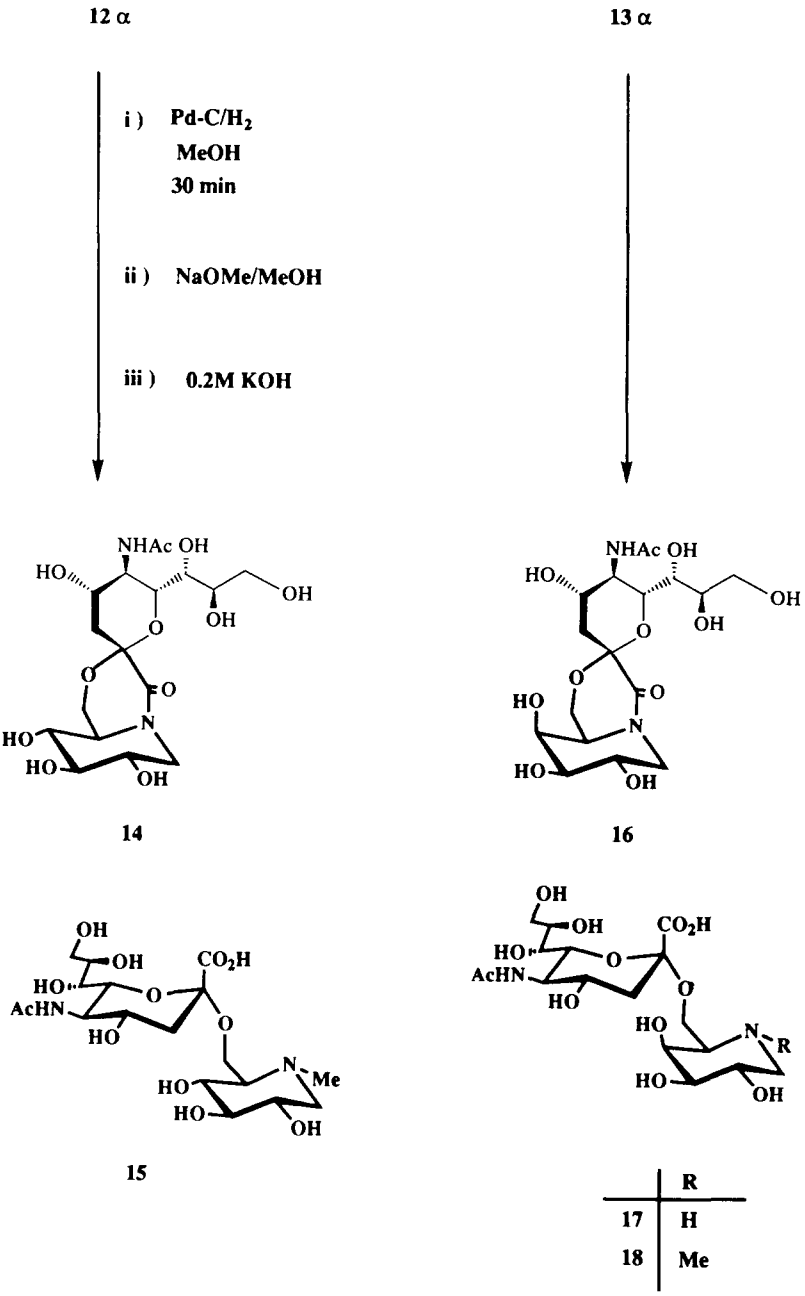
The structures of compounds **14–18** were investigated by mass and ^{1}H NMR spectroscopy. Ion-spray and tandem (MS/MS) mass spectrometry were particularly effective [17]. In the mass spectra of **14** and **16**, the characteristic molecular ions $[M + H]^+$, $[M + Na]^+$, and $[M + NH_4]^+$ were clearly detected at *m/z* 437, 459, and 454, respectively, indicating an average mass of 436.42 ($C_{17}H_{28}N_2O_{11}$). The MS/MS spectra were somewhat complex, but characteristic daughter ions were observed at *m/z* 419 $[M - H_2O + H]^+$, 257 (Neu5Ac fragment $-H_2O$), 239 (Neu5Ac fragment -2 H_2O), and 125 (DNJ fragment -2 H_2O). As described in a previous communication [10c], the lactam ring structure was indicated by the *gauche–gauche* conformation of the C-6 protons of the DNJ moiety, as shown by 2D NMR.

We have found that *N*-benzyloxycarbonyl-DNJ derivatives are readily hydrogenolyzed over palladium catalysts in methanol to give the corresponding *N*-methyl-DNJ derivatives [10b]. The mechanism is believed [18] to involve the dehydrogenation of methanol by palladium to form formaldehyde, which then reacts with the free amine. The resulting iminium ion is finally reduced to yield the *N*-methyl derivative. In the present experiment, however, the major part of the free amines formed in the initial stage seem to be efficiently trapped as lactam derivatives (**14** or **16**).

The mixture of **17** and **18** gave the respective molecular ion pairs at *m/z* 455.5 $[M + H]^+$ and 476.9 $[M + Na]^+$ for **17**, and *m/z* 469.0 $[M + H]^+$ and 490.9 $[M + Na]^+$ for **18**. The MS/MS spectra of P [†] = 455.5 and P = 469.0 showed characteristic daughter ions at *m/z* 163.6 (protonated DNJ fragment) and *m/z* 177.6 (protonated *N*-methyl-DNJ fragment), respectively, together with common fragment ions at *m/z*

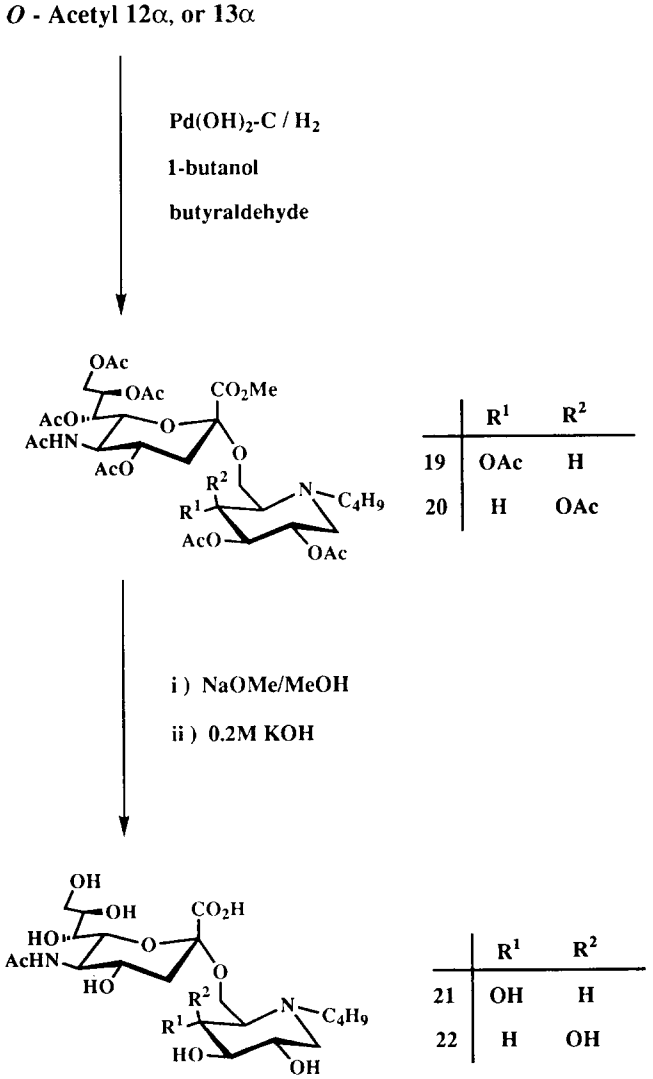
[†] Parent peak subjected to fragmentation.

292 (Neu5Ac moiety) and 274 (Neu5Ac moiety $-H_2O$). These spectral features are compatible with those reported for **15** [17].



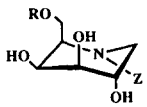
Scheme 1.

Based on these findings just described, the 4-*O*-acetyl derivative of **12**α and the corresponding **13**α were each hydrogenolyzed over palladium hydroxide-on-carbon in 1-butanol in the presence of butyraldehyde, to give **19** and **20** almost quantitatively, which were then converted, by *O*-deacetylation and saponification of the methyl ester group, to the corresponding α-sialyl-(2 → 6)-*N*-butyl-DNJ derivatives **21** and **22** (Scheme 2). These structures were elucidated from ion-spray MS and MS/MS spectra. Characteristic molecular and daughter ions were clearly observed at m/z 511 $[M + H]^+$, 493 $[M - H_2O + H]^+$, 292 (Neu5Ac fragment), 274 (Neu5Ac fragment $-H_2O$), 220 (protonated *N*-butyl-DNJ fragment), and 202 ($220 - H_2O$), providing the unambiguous evidence for the *N*-butylated structures assigned.

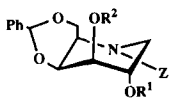


Scheme 2.

Synthesis of the α-sialyl-(2 → 3)-DNJ derivatives.—*O*-Deacetylation of **8** and removal of the TBDPS group gave **24**, which was then benzylidenated. The resulting **25** was treated with 1.2 equiv of chloroacetyl chloride at −20°C in a mixture of dichloromethane and 2,6-lutidine to give the 3-*O*-chloroacetyl derivative (**26**) in 67% yield. After 2-*O*-acetylation, the chloroacetyl group was selectively removed by treatment with pyridine–water to afford **28**. Glycosylation of **28** with **11** was performed in the presence of NIS and TfOH in acetonitrile to provide **29** in about 50% yield based on **28**.

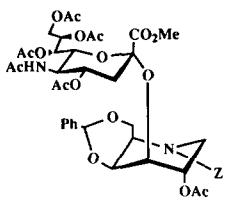


	R
23	TBDPS
24	H

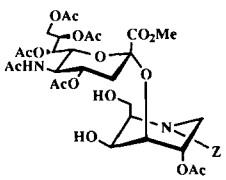


	R^1	R^2
25	H	H
26	H	CAc
27	Ac	CAc
28	Ac	H

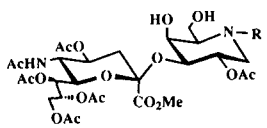
CAc = chloroacetyl



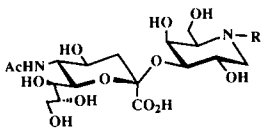
29



30



	R
31	Me
32	C_4H_9



	R
33	H
34	Me
35	C_4H_9

When **29** was directly hydrogenolyzed in the presence of palladium black catalyst in acetic acid, followed by *O*-deacetylation and saponification of the methyl ester group, the corresponding α-sialyl-(2 → 3)-1,5-dideoxy-1,5-imino-D-galactitol (**33**) was formed as a single product. This experiment showed that the presence of methanol in the hydrogenolysis reaction was critical for the *N*-methylation. For making the *N*-alkylated derivatives of **33**, the benzylidene group of **29** was first cleaved by 80% acetic acid to give **30**, which was then hydrogenolyzed over palladium hydroxide-on-carbon in the presence of formaldehyde or butyraldehyde and methanol or 1-butanol, respectively. The expected *N*-alkyl derivatives **31** and **32** were formed in very good yields, and converted to α-sialyl-(2 → 3)-1,5-alkylimino-1,5-dideoxy-D-galactitols (**34** and **35**).

3. Experimental

General methods.—Optical rotations were determined with a Union PM-201 Polarimeter at 25°C and ^{1}H NMR spectra were recorded at 270 or 400 MHz using JEOL

JNM-GX 270 and JNM-GX 400 spectrometers [internal standards: Me_4Si in $CDCl_3$ and CD_3OD, acetone (2.225 ppm) in D_2O]. Preparative chromatography was performed on silica gel (Wako Chemical Co., 200 mesh) with the solvent systems specified. Concentrations were conducted in vacuo.

Mass spectrometry.—Electrospray mass spectra were recorded on an API-III triple-quadrupole mass spectrometer (Perkin–Elmer Sciex Instruments, Thornhill, Canada) fitted with an atmospheric pressure ionization source. The mass spectrometer was operated in the positive mode; the ion-spray voltage was set to 4500 V or 4600 V, and the interface plate voltage was 650 V. The orifice voltage was 60–100 V. The pressure of the nebulizing gas was 30 psi and the flow rate was 0.8 L/min. The collisionally activated dissociated (CAD) spectrum was measured with argon as the collision gas, and the collision gas pressure was set at 300×10^{12} atoms/cm^3. The collision energy was 120 eV.

X-ray crystallography.—Crystal data for **1** are as follows: $C_{11}H_{21}O_6N$, $M = 263.29$, triclinic, space group $P1$, $a = 18.414$ (12), $b = 6.418$ (6), $c = 6.387$ (5) Å, $\alpha = 112.94$ (8)°, $\beta = 91.49$ (9)°, $\gamma = 78.22$ (6)°, $U = 679$ (1) Å^3, $Z = 2$, $D_c = 1.288$ g cm^{-3}, $\mu = 1.95$ cm^{-1}. Reflections with $2\theta \leq 50.0°$, 2678 in all, were recorded on a Rigaku AFC-6A four-circle diffractometer using graphite-monochromated Mo$K\alpha$ radiation. Of these, 1909 with $F > 3\sigma(F)$ were judged as observed. The structure was solved using SHELX-86 [19]. Full-matrix least-squares refinement with anisotropic temperature factors for nonhydrogen atoms and isotropic hydrogens converged to $R = 0.062$ and $R_w = 0.086$ [20], where R and R_w are $\Sigma| \, |F_0| - |F_c| \, |/\Sigma \, |F_0|$ and $[\Sigma w(|F_0| - |F_c|)^2/\Sigma w(F_0)^2]^{1/2}$, respectively. The function minimized during least-squares refinement was $\Sigma w(|F_0| - |F_0|)^2$ with $w = [\sigma^2(F_0) + 0.01(F_0)^2]^{-1}$. Final atomic parameters for the non-hydrogen atoms are listed in Table 1. The bond lengths, angles, and torsion angles are shown in Table 2. ‡

Molecular modeling.—The molecular modeling was performed using a Nemesis molecular modeling system with the COSMIC force field of Oxford Molecular Ltd.

2,3-Di-O-acetyl-4,6-O-benzylidene-1,5-benzyloxycarbonylimino-1,5-dideoxy-D-glucitol (**4**).—Acetylation of 4,6-*O*-benzylidene-1,5-benzyloxycarbonylimino-1,5-dideoxy-D-glucitol (**3**) [11] with Ac_2O and pyridine gave the title compound **4** (quant.); $[\alpha]_D$ −18.7° (*c* 1, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 1.98, 2.07 (2 s, 6 H, AcO), 3.32 (dd, 1 H, J_{gem} 14, $J_{1ax,2}$ 8 Hz, H-1*ax*), 3.47 (ddd, 1 H, $J_{4,5} = J_{5,6ax} = 10$, $J_{5,6eq}$ 4.6 Hz, H-5), 3.88 (t, 1 H, $J_{3,4} = J_{4,5} = 10$ Hz, H-4), 4.07 (dd, 1 H, $J_{1eq,2}$ 4 Hz, H-1*eq*), 4.22 (~ t, 1 H, J_{gem} 11.4, $J_{5,6ax}$ 10 Hz, H-6*ax*), 4.83 (dd, 1 H, J_{gem} 11.4, $J_{5,6eq}$ 4.6 Hz, H-6*eq*), 4.93 (m, $J_{1ax,2}$ 8, $J_{1eq,2}$ 4, $J_{2,3}$ 6.6 Hz, H-2), 5.15 (dd, 1 H, $J_{2,3}$ 6.6, $J_{3,4}$ 10 Hz, H-3), 5.10, 5.16 (2 d, 2 H, J_{gem} 12.3 Hz, OCH_2Ph), 5.53 (s, 1 H, PhC*H*), and 7.3–7.5 (m, 5 H, Ph-H). Anal. Calcd for $C_{25}H_{27}NO_8$ (469.47): C, 63.95; H, 5.80; N, 2.98. Found: C, 63.70; H, 5.76; N, 3.27.

‡ Tables 1 and 2 and a table of observed and calculated structure factors have been deposited with the Cambridge Crystallographic Data Centre. They may be obtained, on request, from the Director, Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge, CB2 1EZ, UK.

Table 1
Fractional coordinates and mean square displacement parameters (in Å^2) for N-Boc-1-deoxynojirimycin (**1**) [a,b]

Atom	x	y	z	$U11$ or U	$U22$	$U33$	$U12$	$U13$	$U23$
Molecule I									
C-1	8241 (3) $\times 10^{-4}$	12722 (9) $\times 10^{-4}$	4230 (9) $\times 10^{-4}$	460 (3) $\times 10^{-4}$	330 (3) $\times 10^{-4}$	350 (3) $\times 10^{-4}$	−70 (2) $\times 10^{-4}$	50 (2) $\times 10^{-4}$	150 (2) $\times 10^{-4}$
C-2	8165 (3)	13664 (9)	6811 (9)	460 (3)	210 (3)	330 (3)	30 (2)	−40 (2)	60 (2)
C-3	7469 (3)	13226 (9)	7648 (8)	440 (3)	330 (2)	220 (2)	−20 (2)	−30 (2)	90 (2)
C-4	7452 (3)	10690 (9)	6560 (8)	450 (3)	410 (3)	200 (2)	−100 (2)	−10 (2)	160 (2)
C-5	7591 (3)	9595 (8)	3949 (8)	380 (3)	260 (2)	200 (2)	−60 (2)	40 (2)	90 (2)
C-6	6924 (3)	10224 (9)	2694 (8)	440 (3)	330 (2)	200 (2)	−70 (2)	10 (2)	80 (2)
C-7	8821 (3)	8579 (8)	2034 (8)	400 (3)	360 (3)	220 (2)	−70 (2)	−40 (2)	150 (2)
C-8	10049 (4)	8210 (1)	390 (1)	370 (3)	580 (4)	580 (3)	−20 (3)	40 (3)	260 (3)
C-9	9974 (5)	6480 (2)	−1970 (1)	600 (5)	1040 (6)	470 (4)	−100 (4)	260 (3)	50 (4)
C-10	10411 (5)	10080 (2)	280 (2)	490 (5)	1000 (7)	1330 (9)	−190 (4)	230 (5)	570 (6)
C-11	10458 (5)	7030 (2)	1870 (2)	540 (5)	1210 (8)	850 (6)	10 (5)	−70 (4)	580 (6)
N-5	8250 (2)	10243 (7)	3314 (7)	380 (2)	370 (2)	260 (2)	−90 (2)	60 (2)	140 (2)
O-2	8800 (2)	12593 (7)	7662 (7)	440 (2)	350 (2)	470 (2)	−40 (2)	−90 (2)	90 (2)
O-3	6822 (2)	14545 (6)	7151 (6)	400 (2)	400 (2)	320 (2)	50 (2)	−50 (2)	70 (2)
O-4	7977 (2)	9444 (7)	7568 (6)	490 (2)	440 (2)	280 (2)	−60 (2)	−70 (2)	230 (2)
O-6	7017 (2)	8787 (7)	313 (6)	580 (2)	490 (2)	150 (1)	−160 (2)	−20 (1)	50 (1)
O-7	8873 (2)	6522 (6)	1511 (6)	490 (2)	310 (2)	350 (2)	−40 (2)	50 (2)	90 (2)
O-8	9317 (3)	9521 (7)	1443 (8)	470 (3)	460 (2)	610 (3)	−70 (2)	130 (2)	220 (2)
Molecule II									
C-1′	4273 (3) $\times 10^{-4}$	7559 (9) $\times 10^{-4}$	2044 (8) $\times 10^{-4}$	580 (4) $\times 10^{-4}$	430 (3) $\times 10^{-4}$	160 (2) $\times 10^{-4}$	20 (3) $\times 10^{-4}$	−20 (2) $\times 10^{-4}$	150 (2) $\times 10^{-4}$
C-2′	4360 (3)	4950 (1)	1058 (8)	460 (3)	530 (3)	160 (2)	−20 (3)	−30 (2)	110 (2)
C-3′	5052 (3)	3773 (9)	1846 (8)	590 (4)	270 (2)	190 (2)	−20 (2)	30 (2)	20 (2)
C-4′	5040 (3)	4769 (8)	4488 (8)	440 (3)	260 (2)	230 (2)	−40 (2)	10 (2)	110 (2)
C-5′	4866 (3)	7421 (7)	5592 (7)	370 (3)	230 (2)	180 (2)	−50 (2)	−40 (2)	70 (2)
C-6′	5544 (3)	8387 (9)	5470 (1)	400 (3)	370 (3)	470 (3)	−80 (2)	30 (2)	200 (2)
C-7′	3661 (3)	9961 (8)	5913 (8)	330 (3)	280 (2)	320 (2)	−40 (2)	−40 (2)	130 (2)
C-8′	2443 (3)	12250 (1)	5620 (1)	330 (3)	640 (4)	540 (3)	10 (3)	−40 (3)	270 (3)
C-9′	2505 (5)	14570 (1)	7460 (2)	660 (5)	600 (5)	930 (6)	270 (4)	−50 (5)	150 (4)
C-10′	2109 (5)	12570 (2)	3540 (2)	700 (6)	1130 (7)	650 (5)	270 (5)	−100 (4)	490 (5)
C-11′	2021 (5)	10950 (2)	6490 (2)	510 (5)	1180 (7)	990 (7)	−220 (5)	−70 (4)	580 (6)
N-5′	4231 (3)	8413 (7)	4581 (6)	410 (2)	320 (2)	160 (2)	30 (2)	−10 (2)	100 (2)
O-2′	3723 (2)	4359 (8)	1763 (7)	480 (3)	640 (3)	310 (2)	−190 (2)	−90 (2)	110 (2)
O-3′	5694 (3)	4166 (7)	1040 (7)	550 (3)	470 (2)	310 (2)	−30 (2)	100 (2)	90 (2)
O-4′	4505 (2)	3867 (6)	5278 (6)	580 (2)	390 (2)	300 (2)	−150 (2)	−80 (2)	220 (2)
O-6′	5423 (2)	10810 (6)	6689 (6)	530 (2)	280 (2)	360	−150 (2)	−80 (2)	130 (1)
O-7′	3588 (2)	10497 (7)	7974 (6)	450 (2)	480 (2)	160 (1)	−10 (2)	0 (1)	90 (1)
O-8′	3174 (2)	10848 (8)	4695 (6)	410 (2)	670 (3)	300 (2)	130 (2)	−20 (2)	250 (2)

[a] Standard deviations of the least significant figures are given in parentheses. [b] The U_{ij} coefficients are given by the expression $\exp[-2\pi^2(U_{11}h^2a^{*2} + U_{22}k^2b^{*2} + U_{33}l^2c^{*2} + 2U_{12}hka^*b^* + 2U_{13}hla^*c^* + 2U_{23}klb^*c^*)]$. U values are in the form $\exp[-2\pi^2U(2\sin\theta/\lambda)^2]$.

*2,3-Di-*O*-acetyl-1,5-benzyloxycarbonylimino-1,5-dideoxy-*D*-glucitol* (**5**).—Compound **4** (0.72 g) was treated with 80% aq AcOH (20 mL) for 6 h at 45°C. The mixture was concentrated to a syrup, which was chromatographed on a column of silica gel with 3:1

Table 2
Molecular geometry of **1**

Bond lengths (Å)			
Molecule I		Molecule II	
C-1–C-2	1.518 (8)	C-1′–C-2′	1.518 (8)
C-1–N-5	1.462 (7)	C-1′–N-5′	1.493 (6)
C-2–C-3	1.519 (9)	C-2′–C-3′	1.521 (8)
C-2–O-2	1.436 (7)	C-2′–O-2′	1.436 (9)
C-3–C-4	1.507 (7)	C-3′–C-4′	1.553 (7)
C-3–O-3	1.419 (7)	C-3′–O-3′	1.408 (8)
C-4–C-5	1.541 (6)	C-4′–C-5′	1.533 (6)
C-4–O-4	1.429 (7)	C-4′–O-4′	1.430 (8)
C-5–C-6	1.521 (7)	C-5′–C-6′	1.520 (9)
C-5–N-5	1.473 (8)	C-5′–N-5′	1.472 (7)
C-6–O-6	1.431 (5)	C-6′–O-6′	1.413 (6)
C-7–N-5	1.349 (6)	C-7′–N-5′	1.331 (6)
C-7–O-7	1.212 (7)	C-7′–O-7′	1.228 (6)
C-7–O-8	1.328 (8)	C-7′–O-8′	1.354 (7)
C-8–C-9	1.508 (10)	C-8′–C-9′	1.516 (10)
C-8–C-10	1.512 (15)	C-8′–C-10′	1.518 (13)
C-8–C-11	1.522 (15)	C-8′–C-11′	1.509 (16)
C-8–O-8	1.455 (7)	C-8′–O-8′	1.451 (7)
Bond angles (°)			
Molecule I		Molecule II	
C-2–C-1–N-5	109.1 (5)	C-2′–C-1′–N-5′	108.6 (5)
C-1–C-2–C-3	111.7 (5)	C-1′–C-2′–C-3′	111.7 (5)
C-1–C-2–O-2	110.0 (4)	C-1′–C-2′–O-2′	109.6 (4)
C-3–C-2–O-2	108.6 (5)	C-3′–C-2′–O-2′	108.5 (6)
C-2–C-3–C-4	110.3 (4)	C-2′–C-3′–C-4′	110.4 (4)
C-2–C-3–O-3	110.7 (5)	C-2′–C-3′–O-3′	110.3 (5)
C-4–C-3–O-3	109.8 (4)	C-4′–C-3′–O-3′	107.9 (5)
C-3–C-4–C-5	114.6 (5)	C-3′–C-4′–C-5′	113.6 (5)
C-3–C-4–O-4	109.5 (4)	C-3′–C-4′–O-4′	107.7 (4)
C-5–C-4–O-4	109.9 (4)	C-5′–C-4′–O-4′	110.6 (4)
C-4–C-5–C-6	113.4 (4)	C-4′–C-5′–C-6′	111.5 (4)
C-4–C-5–N-5	109.5 (4)	C-4′–C-5′–N-5′	111.4 (4)
C-6–C-5–N-5	110.8 (5)	C-6′–C-5′–N-5′	111.2 (5)
C-5–C-6–O-6	111.3 (4)	C-5′–C-6′–O-6′	112.6 (4)
N-5–C-7–O-7	124.7 (6)	N-5′–C-7′–O-7′	124.2 (5)
N-5–C-7–O-8	110.2 (5)	N-5′–C-7′–O-8′	111.2 (4)
O-7–C-7–O-8	125.0 (5)	O-7′–C-7′–O-8′	124.6 (4)
C-9–C-8–C-10	110.5 (8)	C-9′–C-8′–C-10′	110.6 (8)
C-9–C-8–C-11	111.4 (7)	C-9′–C-8′–C-11′	111.9 (7)
C-9–C-8–O-8	109.9 (6)	C-9′–C-8′–O-8′	110.7 (6)
C-10–C-8–C-11	112.2 (8)	C-10′–C-8′–C-11′	112.0 (7)
C-10–C-8–O-8	102.0 (6)	C-10′–C-8′–O-8′	101.6 (6)
C-11–C-8–O-8	110.4 (7)	C-11′–C-8′–O-8′	109.6 (6)
C-1–N-5–C-5	115.7 (4)	C-1′–N-5′–C-5′	116.0 (4)
C-1–N-5–C-7	124.4 (5)	C-1′–N-5′–C-7′	123.9 (5)
C-5–N-5–C-7	119.8 (4)	C-5′–N-5′–C-7′	120.1 (4)
C-7–O-8–C-8	122.8 (5)	C-7′–O-8′–C-8′	123.0 (5)

Table 2 (continued)

Torsion angles (°)			
Molecule I		Molecule II	
C-1–C-2–C-3–C-4	54.2 (6)	C-1′–C-2′–C-3′–C-4′	55.6 (6)
C-2–C-3–C-4–C-5	−49.8 (6)	C-2′–C-3′–C-4′–C-5′	−48.9 (7)
C-3–C-4–C-5–N-5	48.1 (6)	C-3′–C-4′–C-5′–N-5′	46.0 (6)
C-4–C-5–N-5–C-1	−53.1 (6)	C-4′–C-5′–N-5′–C-1′	−51.4 (6)
C-5–N-5–C-1–C-2	58.6 (6)	C-5′–N-5′–C-1′–C-2′	57.6 (6)
N-5–C-1–C-2–C-3	−57.6 (6)	N-5′–C-1′–C-2′–C-3′	−58.6 (6)
N-5–C-1–C-2–O-2	63.1 (6)	C-4′–C-3′–C-2′–O-2′	−65.3 (6)
C-4–C-3–C-2–O-2	−67.4 (5)	N-5′–C-1′–C-2′–O-2′	61.6 (6)
C-1–C-2–C-3–O-3	−67.5 (6)	C-1′–C-2′–C-3′–O-3′	−63.5 (5)
C-5–C-4–C-3–O-3	72.4 (6)	C-5′–C-4′–C-3′–O-3′	71.6 (6)
C-3–C-4–C-5–C-6	−76.2 (6)	C-2′–C-3′–C-4′–O-4′	74.0 (6)
C-1–N-5–C-5–C-6	72.7 (5)	N-5′–C-5′–C-4′–O-4′	−75.3 (5)
C-4–C-5–C-6–O-6	−167.1 (5)	C-3′–C-4′–C-5′–C-6′	−78.9 (6)
N-5–C-5–C-6–O-6	69.3 (6)	C-1′–N-5′–C-5′–C-6′	73.6 (6)
C-2–C-1–N-5–C-7	−118.9 (5)	C-2′–C-1′–N-5′–C-7′	−121.9 (6)
C-4–C-5–N-5–C-7	124.6 (5)	C-4′–C-5′–N-5′–C-7′	128.0 (5)

EtOAc–hexane to give **5** (quant.); $[\alpha]_D$ −1.7° (*c* 1, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 1.95, 2.10 (2 s, 6 H, AcO), 4.86 (~ q, 1 H, $J_{1ax,2} = J_{1eq,2} = 4$, $J_{2,3}$ 5.7 Hz, H-2), 5.03 (dd, 1 H, $J_{2,3}$ 5.7, $J_{3,4}$ 7.3 Hz, H-3), 5.10, 5.21 (2 d, 2 H, J_{gem} 12.3 Hz, OCH_2Ph), and complete disappearance of PhC*H*. Anal. Calcd for $C_{18}H_{23}NO_8$ (381.37): C, 56.68; H, 6.08; N, 3.67. Found: C, 56.49; H, 5.80; N, 3.69.

*2,3-Di-*O*-acetyl-1,5-benzyloxycarbonylimino-6-*O-tert*-butyldiphenylsilyl-1,5-dideoxy-*D*-glucitol* (**6**).—To a solution of **5** (1.31 g) in pyridine (25 mL), cooled to 0°C, was added *tert*-butyldiphenylsilyl chloride (2.7 mL, 3 mol equiv) and the mixture was stirred overnight at 20°C. Dichloromethane was added, and the solution was washed with ice-cold 2 M HCl and water, dried (Na_2SO_4), and concentrated to a syrup, which was chromatographed on a column of silica gel with 1:2 EtOAc–hexane to give **6** (93%); $[\alpha]_D$ +2.6° (*c* 1, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 1.04 (s, 9 H, *t*-Bu), 1.94, 1.98 (2 s, 6 H, 2 AcO), 2.54 (d, 1 H, $J_{4,OH}$ 7.9 Hz, 4-O*H*), 3.25 (dd, 1 H, J_{gem} 15.6, $J_{1ax,2}$ 3 Hz, H-1*ax*), 3.84 (m, 1 H, H-5), 3.90 (dd, 1 H, J_{gem} 11, $J_{5,6}$ 5.3 Hz, H-6), 4.04 (~ q, 1 H, $J_{3,4}$ 7, $J_{4,5}$ 5.5, $J_{4,OH}$ 7.9 Hz, H-4), 4.18 (~ d, 1 H, J_{gem} 15.6 Hz, H-1*eq*), 4.25 (dd, 1 H, J_{gem} 11, $J_{5,6'}$ 6 Hz, H-6′), 4.89 (br s, 1 H, H-2), 4.98 (dd, 1 H, $J_{2,3}$ 4, $J_{3,4}$ 7 Hz, H-3), 5.02–5.17 (OCH_2Ph), and 7.2–7.7 (m, 15 H, Ph-H). Anal. Calcd for $C_{34}H_{41}NO_8Si$ (619.74): C, 65.89; H, 6.67; N, 2.26. Found: C, 65.98; H, 6.54; N, 2.25.

*2,3,4-Tri-*O*-acetyl-1,5-benzyloxycarbonylimino-6-*O-tert*-butyldiphenylsilyl-1,5-dideoxy-*D*-galactitol* (**8**).—To a solution of **6** (1.05 g) in 2:1 CH_2Cl_2–pyridine (45 mL), cooled to −15°C, was added dropwise a solution of trifluoromethanesulfonic anhydride (0.58 mL, 2 mol equiv) in CH_2Cl_2 (10 mL). The mixture was stirred for 3 h at −15°C and neutralized with Et_3N. Dichloromethane was added, and the solution was washed with ice-cold water, 2 M HCl, and water, dried (Na_2SO_4), and concentrated to a syrup of **7**, which was then dissolved in dry MeCN (40 mL) and stirred with molecular sieves

3 Å (120 mg) for 1 h. Cesium acetate (1.63 g, 5 mol equiv) and 1,4,7,10,13,16-hexaoxacyclooctadecane (0.67 g, 1.5 mol equiv) were added, and the mixture was stirred overnight at room temperature. The solids were filtered off and washed with CH_2Cl_2. The filtrate and washings were combined and concentrated to a syrup, which was chromatographed on a column of silica gel with 5:1 hexane–EtOAc to give **8** (0.75 g, 67%); $[\alpha]_D$ +0.1° (*c* 0.9, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 0.99 (s, 9 H, *t*-Bu), 1.83 (s, 3 H, AcO), 1.91 (s, 6 H, 2 AcO), 2.96 (dd, 1 H, J_{gem} 15.6, $J_{1ax,2}$ 2.4 Hz, H-1*ax*), 3.71 (m, 1 H, H-5), 3.95 (~ t, 1 H, J_{gem} 12, $J_{5,6}$ 9.2 Hz, H-6), 4.08 (br d, 1 H, H-1*eq*), 4.65-4.8 (m, 2 H, H-2, 6′), 5.04 (t, 1 H, $J_{2,3} = J_{3,4}$ = 3–4 Hz, H-3), 5.10 (br s, 2 H, OCH_2Ph), 5.34 (dd, 1 H, $J_{3,4}$ 3.1, $J_{4,5}$ 6.2 Hz, H-4), and 7.29–7.61 (m, 15 H, Ph-H). Anal. Calcd for $C_{36}H_{43}NO_9Si$ (661.77): C, 65.33; H, 6.55; N, 2.12. Found: C, 65.34; H, 6.66; N, 2.01.

*2,3,4-Tri-*O*-acetyl-1,5-benzyloxycarbonylimino-1,5-dideoxy-*D*-galactitol* (**9**).—To a solution of **8** (0.63 mg) in CH_2Cl_2 (40 mL), cooled to 0°C, was added BF_3 etherate (7 mL), and the mixture was stirred for 10 h at room temperature. Dichloromethane was added, and the solution was washed with ice-cold M Na_2CO_3 and water, dried (Na_2SO_4), and concentrated to a syrup which was chromatographed on a column of silica gel with 200:1 CH_2Cl_2–MeOH to give **9** (0.324 g, 81%); $[\alpha]_D$ +20° (*c* 1.6, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 1.91, 2.05, 2.09 (3 s, 9 H, 3 AcO), 3.51 (br d, 1 H, H-1*ax*), 3.78 (dd, 1 H, J_{gem} 12, $J_{5,6}$ 4.2 Hz, H-6), 4.02 (dd, 1 H, J_{gem} 12, $J_{5,6'}$ 9.4 Hz, H-6′), 4.15 (br d, 1 H, H-1*eq*), 4.56 (m, 1 H, H-5), 4.89 (m, 1 H, $J_{1ax,2} = J_{1eq,2}$ = 2–3, $J_{2,3}$ 4.5 Hz, H-2), 5.15 (~ t, 1 H, $J_{2,3}$ 4.5, $J_{3,4}$ 3.3 Hz, H-3), 5.09, 5.23 (2 d, 2 H, J_{gem} 12.1 Hz, OCH_2Ph), 5.39 (dd, 1 H, $J_{3,4}$ 3.3, $J_{4,5}$ 5.8 Hz, H-4), and 7.35 (s, 5 H, Ph-H). Anal. Calcd for $C_{20}H_{25}NO_9$ (423.41): C, 56.73; H, 5.95; N, 3.31. Found: C, 56.63; H, 6.11; N, 3.32.

O-*(Methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-*D*-*glycero*-α-*D*-galacto-2-nonulopyranosylonate)-(2 → 6)-2,3-di-*O*-acetyl-1,5-benzyloxycarbonylimino-1,5-dideoxy-*D*-glucitol* (**12α**) *and its β-linked anomer*.—To a solution of **5** (0.12 g) and methyl (methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-2-thio-*D*-*glycero*-*D*-*galacto*-2-nonulopyranosid)onate [13] (**10**, 0.328 g, 2 mol equiv) in dry MeCN (10 mL) was added molecular sieves 3 Å (0.5 g), and the mixture was stirred overnight at room temperature, then cooled to −15°C. Dimethyl(methylthio)sulfonium triflate (0.435 g) was added, and the mixture was stirred overnight at 0°C. The solids were filtered off and washed with CH_2Cl_2. The combined filtrate and washings was washed with ice-cold M Na_2CO_3 and water, dried (Na_2SO_4), and concentrated. Column chromatography (70:1 CH_2Cl_2–MeOH) of the residue on silica gel gave **12**α (64% based on acceptor) and the corresponding β-glycoside **12**β (21%). Compound **12**α had $[\alpha]_D$ −16° (*c* 0.8, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 1.70, 1.90, 2.02, 2.04, 2.12, 2.13, 2.16 (7 s, 21 H, AcN and 6 AcO), 2.56 (dd, 1 H, J_{gem} 13, $J_{3eq,4}$ 5 Hz, H-3*eq* of Neu5Ac), 3.14 (d, 1 H, $J_{4,OH}$ 4.9 Hz, 4-O*H* of DNJ), 3.42 (dd, 1 H, J_{gem} 15.6, $J_{1ax,2}$ 3.5 Hz, H-1*ax* of DNJ), 3.73 (s, 3 H, CO_2CH_3), 4.18 (~ d, 1 H, J_{gem} 15.6 Hz, H-1*eq* of DNJ), 4.31 (dd, 1 H, *J* 12.3, 2.2 Hz), 4.90 (m, 1 H, H-4 of Neu5Ac), 4.92 (narrow m, 1 H, H-2 of DNJ), 5.03 (narrow m, 1 H, H-3 of DNJ), 5.09, 5.23 (2 d, 2 H, OCH_2Ph), 5.26, 5.40 (2 m, 2 H, H-7 and H-8 of Neu5Ac), and 7.35 (~ s, 5 H, Ph-H). Anal. Calcd for $C_{38}H_{50}N_2O_{20}$ (854.80): C, 53.39; H, 5.90; N, 3.28. Found: C, 53.52; H, 5.85; N, 3.18.

^{1}H NMR ($CDCl_3$) data for compound **12**β : δ 2.35 (dd, 1 H, J_{gem} 13, $J_{3eq,4}$ 5 Hz, H-3*eq* of Neu5Ac), 3.13 (d, 1 H, $J_{4,OH}$ 6.6 Hz, 4-O*H* of DNJ), 3.40 (dd, 1 H, J_{gem} 15.6, $J_{1ax,2}$ 2.5 Hz, H-1*ax* of DNJ), 4.87, 5.01 (2 narrow m, 2 H, $J_{2,3} = J_{3,4} = 4.5$ Hz, H-2 and H-3 of DNJ), and 7.37 (~ s, 5 H, Ph-H).

O-*(Methyl 5-acetamido-4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2 → 6)-2,3,4-tri*-O-*acetyl-1,5-benzyloxycarbonylimino-1,5-dideoxy*-D-*galactitol* (**13**α) *and its* β*-linked anomer*.—To a solution of **9** (0.147 g) and **10** (0.308 g, 1.7 mol equiv) in dry MeCN (15 mL) was added molecular sieves 3 Å (0.5 g), and the mixture was stirred overnight at room temperature, then cooled to −40°C. Powdered *N*-iodosuccinimide (0.266 g, 3.4 mol equiv) and trifluoromethanesulfonic acid (10 mL, 0.34 mol equiv) were added, and the mixture was stirred overnight at −40°C. The solids were filtered off and washed with CH_2Cl_2. The combined filtrate and washings was washed with ice-cold M Na_2CO_3, $Na_2S_2O_3$, and water, dried (Na_2SO_4), and concentrated. Column chromatography (50:1 toluene–MeOH) of the residue on silica gel gave **13**α (56% based on acceptor) and the corresponding β-glycoside **13**β (24%). Compound **13**α had $[\alpha]_D$ +24° (*c* 0.5, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 1.8–2.2 (25 H, AcN, 7 AcO and H-3*ax* of Neu5Ac), 2.47 (br dd, 1 H, H-3*eq* of Neu5Ac), 3.48 (~ d, J_{gem} 15.6 Hz, 1 H, H-1*ax* of DNJ), 3.74 (br s, 3 H, CO_2CH_3), 4.68 (m, 1 H, H-5 of DNJ), 4.82–4.95 (m, 2 H, H-4 of Neu5Ac and H-2 of DNJ), 5.16 (~ t, 1 H, $J_{2,3} = J_{3,4} = 3$–4 Hz, H-3 of DNJ), 5.09, 5.27 (2 d, 2 H, OCH_2Ph), 5.26, 5.39 (2 m, 2 H, H-7 and H-8 of Neu5Ac), and 7.3–7.4 (m, 5 H, Ph-H). Anal. Calcd for $C_{40}H_{52}N_2O_{21}$ (896.83): C, 53.57; H, 5.84; N, 3.12. Found: C, 53.76; H, 5.68; N, 2.98.

^{1}H NMR ($CDCl_3$) data for compound **13**β : δ 2.36 (dd, 1 H, J_{gem} 13, $J_{3eq,4}$ 5 Hz, H-3*eq* of Neu5Ac), 3.38 (~ d, 1 H, J_{gem} 15.4 Hz, H-1*ax* of DNJ), 3.79 (s, 3 H, CO_2CH_3), 4.27 (~ d, 1 H, J_{gem} 15.4 Hz, H-1*eq* of DNJ), 4.86 (narrow m, 1 H, H-2 of DNJ), 5.01 (~ s, 2 H, OCH_2Ph), 5.14 (m, 1 H, H-4 of Neu5Ac), and 5.24 (t, 1 H, *J* 3.2 Hz, H-3 of DNJ).

6-O-*(5-Acetamido-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylono-1',5-lactam)-1,5-dideoxy-1,5-imino*-D-*glucitol* (**14**) *and* O-*(5-acetamido-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonic acid)-(2 → 6)-1,5-dideoxy-1,5-methylimino*-D-*glucitol* (**15**).—Compound **12**α (0.39 g) in MeOH (15 mL) was hydrogenolyzed over 10% Pd–C (0.4 g) for 30 min at room temperature. The solids were filtered off and washed with MeOH. The combined filtrate and washings was concentrated, and the residue was treated with NaOMe in methanol overnight at room temperature. Potassium hydroxide (0.2 M, 4 mL) was added and the mixture was again stirred overnight at room temperature, then neutralized with Amberlite IR-120 (H^+) resin. The resin was filtered off and washed with MeOH–water. The combined filtrate and washings was concentrated, and the residue was chromatographed on a column of Sephadex LH-20 to give novel lactam **14** (81%) and the *N*-methylated product **15** (19%), respectively. Compound **14** had $[\alpha]_D$ +38° (*c* 0.8, MeOH); ^{1}H NMR (CD_3OD): δ (for the DNJ moiety) 2.48 (dd, 1 H, J_{gem} 13, $J_{1ax,2}$ 10 Hz, H-1*ax*), 3.04 (dd, 1 H, $J_{4,5}$ 10, $J_{5,6}$ 0, $J_{5,6'}$ 4.4 Hz, H-5), 3.29 (t, 1 H, $J_{2,3} = J_{3,4} = 8.8$ Hz, H-3), 3.33 (m, 1 H, $J_{1ax,2}$ 10, $J_{1eq,2}$ 5 Hz, H-2), 3.41 (dd, 1 H, $J_{3,4}$ 8.8, $J_{4,5}$ 10 Hz, H-4), 4.05 (d, 1 H, J_{gem} 12, $J_{5,6}$ 0 Hz, H-6), 4.48 (dd, 1 H, J_{gem} 12, $J_{5,6'}$ 4.4 Hz, H-6'), and 4.49 (dd, 1 H, J_{gem} 13, $J_{1eq,2}$ 5 Hz,

H-1*eq*); δ (for the Neu5Ac moiety) 1.56 (dd, 1 H, J_{gem} 13.2, $J_{3ax,4}$ 11 Hz, H-3*ax*), 2.01 (s, 3 H, AcN), 2.27 (dd, 1 H, J_{gem} 13.2, $J_{3eq,4}$ 5.9 Hz, H-3*eq*), 3.45 (dd, 1 H, $J_{6,7}$ 1.5, $J_{7,8}$ 9.5 Hz, H-7), 3.61 (dd, 1 H, J_{gem} 11.4, $J_{8,9}$ 5.5 Hz, H-9), 3.72 (m, 1 H, H-8), 3.77 (dd, 1 H, $J_{8,9'}$ 2.9 Hz, H-9′), 3.79 (t, 1 H, $J_{4,5} = J_{5,6}$ = 10–11 Hz, H-5), 3.91 (dd, 1 H, $J_{5,6}$ 11, $J_{6,7}$ 1.5 Hz, H-6), and 4.47–4.54 (m, 1 H, H-4); see also the previous communication [10c]; all data were confirmed by 2D NMR. Ion-spray MS (positive-ion mode): *m/z* 459.0 $[M + Na]^+$, 437.2 $[M + H]^+$ (base peak), and 419.2 $[M - H_2O + H]^+$; MS/MS (P = 437): *m/z* 418.6 $[M - H_2O + H]^+$, 256.9 ($C_{11}H_{15}NO_6^+$, Neu5Ac fragment $-H_2O$), 239.0 ($C_{11}H_{13}NO_5^+$, Neu5Ac fragment $-2\ H_2O$), and 125.1 (DNJ fragment $-H_2O$). Anal. Calcd for $C_{17}H_{28}N_2O_{11}$ (436.41): C, 46.79; H, 6.47; N, 6.42. Found: C, 46.65; H, 6.64; N, 6.24.

Compound **15** had $[\alpha]_D$ +10° (*c* 0.5, 2:3 H_2O–EtOH); ^1H NMR (CD_3OD): δ 1.68 (~ t, 1 H, $J_{gem} = J_{3ax,4}$ = 11.6 Hz, H-3*ax* of Neu5Ac), 2.02 (s, 3 H, AcN), 2.75–2.90 (m, 2 H, H-3*eq* of Neu5Ac and H-1*ax* of DNJ, overlapping with *N*-CH_3), and 2.85 (s, 3 H, *N*-CH_3); ion-spray MS (positive-ion mode): *m/z* 469.2 $[M + H]^+$ (base peak) and 491.2 $[M + Na]^+$; (negative-ion mode): 466.8 $[M - H]^-$ (base peak); average molecular weight from the results, 468.46 ($C_{18}H_{32}N_2O_{12}$); MS/MS (P = 469.2): *m/z* 292.1 ($C_{11}H_{18}NO_8^+$, Neu5Ac fragment), 273.9 [292 $-H_2O$], and 177.0 ($C_7H_{15}NO_4^+$, protonated *N*-methyl DNJ fragment); for further details, see ref. [17].

6-O-(5-Acetamido-3,5-dideoxy-D-glycero-α-D-galacto-*2-nonulopyranosylono-1′,5-lactam)-1,5-dideoxy-1,5-imino*-D-*galactitol* (**16**), *the free acid* O-*(5-acetamido-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonic acid)-(2 → 6)-1,5-dideoxy-1,5-imino*-D-*galactitol* (**17**), *and its* N-*methyl derivative* (**18**).—Compound **12**α (174 mg) was hydrogenolyzed over 10% Pd–C (0.2 g) in MeOH for 30 min. The mixture was processed as just described for **12**α, to give **16** (87%) as a major product; $[\alpha]_D$ +36° (*c* 1.4, MeOH); ^1H NMR (CD_3OD): δ 1.52 (~ t, 1 H, J_{gem} 13, $J_{3ax,4}$ 11 Hz, H-3*ax* of Neu5Ac), 1.99 (s, 3 H, AcN), and 2.33–2.45 (m, 2 H, H-3*eq* of Neu5Ac and H-1*ax* of DNJ); ion-spray MS (positive-ion mode): *m/z* 459.0 $[M + Na]^+$, 437.2 $[M + H]^+$ (base peak), and 419.2 $[M - H_2O + H]^+$; MS/MS (P = 437): *m/z* 418.8 $[M - H_2O + H]^+$, 256.9 ($C_{11}H_{15}NO_6^+$, Neu5Ac fragment $-H_2O$), and 125.1 (DNJ fragment $-H_2O$). Anal. Calcd for $C_{17}H_{28}N_2O_{11}$ (436.41): C, 46.79; H, 6.47; N, 6.42. Found: C, 46.76; H, 6.27; N, 6.34.

The minor product (13%) was a mixture of **17** and **18**; ion-spray MS (positive-ion mode): *m/z* 455.5 $[M + H]^+$ of **17**, 469.0 $[M + H]^+$ of **18**; MS/MS (P = 455): *m/z* 291.7 ($C_{11}H_{18}NO_8^+$, Neu5Ac fragment), 273.8 (291.7 $-H_2O$), and 163.6 (protonated DNJ fragment); MS/MS (P = 469): *m/z* 291.9 (Neu5Ac fragment), 273.7 [291.9 $-H_2O$], and 177.6 (protonated *N*-methyl-DNJ fragment).

O-(Methyl 5-acetamido-4,7,8,9-tetra-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate)-(2 → 6)-2,3,4-tri*-O-*acetyl-1,5-butylimino-1,5-dideoxy*-D-*glucitol* (**19**).—The 4-*O*-acetyl derivative of **12**α (116 mg), which was readily obtained by treatment of **12a** with Ac_2O–pyridine, was dissolved in 1-butanol (10 mL). Acetic acid (10 μL), water (1.5 mL), butyraldehyde (0.23 mL, about 20 mol equiv), and then palladium hydroxide-on-carbon (120 mg) were added. The mixture was stirred overnight in a hydrogen atmosphere. The catalyst was removed by filtration and washed with MeOH. The combined filtrate and washings was concentrated. Column chromatography

of the residue on silica gel with 250:4 CH_2Cl_2–MeOH gave **19** almost quantitatively; $[\alpha]_D$ +13° (*c* 2, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ (for the DNJ moiety) 0.92 (t, 3 H, *J* 7 Hz, CH_3 of *N*-butyl), 1.2–1.5 [m, 4 H, (CH_2)$_2$CH$_3$], 2.31 (~ t, 1 H, J_{gem} 12, $J_{1ax,2}$ 11 Hz, H-1*ax*), 2.5–2.75 (m, 2 H, N-CH_2), 3.20 (dd, 1 H, J_{gem} 12, $J_{1eq,2}$ 4.6 Hz, H-1*eq*), 4.98 (m, 1 H, H-2), 5.01, and 5.12 (2 t, 2 H, $J_{2,3} = J_{3,4} = J_{4,5}$ = 9–10 Hz, H-3, 4); δ (for the Neu5Ac moiety) 1.95 (t, 1 H, *J*12.6 Hz, H-3*ax*), 2.60 (dd, 1 H, J_{gem} 13, $J_{3eq,4}$ 4.5 Hz, H-3*eq*), 3.78 (s, 3 H, CO_2CH_3), and 4.85 (m, 1 H, H-4). The acetyl protons (8 s, 24 H, AcN and AcO) were clearly observed at δ 1.87, 2.01, 2.012, 2.02, 2.04, 2.07, 2.13, and 2.14. Anal. Calcd for $C_{36}H_{54}N_2O_{19}$ (818.81): C, 52.80; H, 6.65; N, 3.42. Found: C, 52.76; H, 6.62; N, 3.24.

O-(*Methyl 5-acetamido-4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate*)-(*2* → *6*)-*2,3,4-tri*-O-*acetyl-1,5-butylimino-1,5-dideoxy*-D-*galactitol* (**20**).—A mixture of **13**α (59 mg), 1-butanol (5 mL), AcOH (6.3 μL), water (0.11 mL), butyraldehyde (0.12 mL), and palladium hydroxide-on-carbon (60 mg) was stirred overnight in a hydrogen atmosphere as just described for **19**. Workup and column chromatography on silica gel with 30:1 CH_2Cl_2–MeOH gave **20** (90%), $[\alpha]_D$ −3° (*c* 1, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ (for the DNJ moiety) 0.93 (t, 3 H, *J* 7 Hz, CH_3 of *N*-butyl), 1.2–1.5 [m, 4 H, (CH_2)$_2$CH$_3$], 2.35 (t, 1 H, *J* 10–11 Hz, H-1*ax*), 2.5–2.75 (m, 2 H, N-CH_2), 2.87 (narrow t, 1 H, H-5), 3.18 (dd, 1 H, J_{gem} 11.4, $J_{1eq,2}$ 4.6 Hz, H-1*eq*), 4.87 (dd, 1 H, $J_{2,3}$ 10, $J_{3,4}$ 3.5 Hz, H-3), 5.16 (m, 1 H, $J_{1ax,2} = J_{2,3}$ = 10, $J_{1eq,2}$ 4.6 Hz, H-2), and 5.48 (~ s, 1 H, H-4); δ (for the Neu5Ac moiety) 2.53 (dd, 1 H, J_{gem}13, $J_{3eq,4}$ 4.3 Hz, H-3*eq*), 3.78 (s, 3 H, CO_2CH_3), and 4.84 (m, 1 H, H-4). The acetyl protons (8 s, 24 H, AcN and AcO), appeared at δ 1.88, 1.98, 2.02, 2.03, 2.034, 2.11, 2.12, and 2.15. Anal. Calcd for $C_{36}H_{54}N_2O_{19}$ (818.81): C, 52.80; H, 6.65; N, 3.42. Found: C, 52.73; H, 6.56; N, 3.41.

O-(*5-Acetamido-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonic acid*)-(*2* → *6*)-*1,5-butylimino-1,5-dideoxy*-D-*glucitol* (**21**).—Compound **19** (105 mg) was successively treated with NaOMe in MeOH and 0.2 M KOH as described for **14** and **15**, followed by neutralization and purification on a Sephadex LH-20 column. The resulting title compound **21** (almost quantitative) had $[\alpha]_D$ −6° (*c* 0.4, MeOH); ^{1}H NMR (D_2O): δ (for the DNJ moiety) 0.93 (t, 3 H, *J* 7.4 Hz, CH_3 of *N*-butyl), 1.38, 1.68 [2 m, 4 H, (CH_2)$_2$CH$_3$], 3.07 (t, 1 H, *J* 12 Hz, H-1*ax*), 3.15–3.35 (m, 2 H, N-CH_2); δ (for the Neu5Ac moiety) 1.74 (t, 1 H, *J* 12 Hz, H-3*ax*), 2.02 (s, 3 H, AcN), and 2.70 (dd, 1 H, J_{gem} 12, $J_{3eq,4}$ 4.5 Hz, H-3*eq*); ion-spray MS (positive-ion mode) (0.5% HCOOH −50% CH_3CN): *m/z* 511.3 $[M + H]^+$; (1 mM NH_4OAc −50% CH_3CN): *m/z* 511.2 $[M + H]^+$ and 533.2 $[M + Na]^+$; MS/MS (P = 511): *m/z* 492.7 $[M - H_2O + H]^+$, 291.9 (Neu5Ac fragment), 274.0 (Neu5Ac fragment $-H_2O$), 219.6 (protonated *N*-butyl-DNJ fragment), and 201.6 [219.6 $-H_2O$]. Anal. Calcd for $C_{21}H_{38}N_2O_{12}$ (510.53): C, 49.40; H, 7.50; N, 5.49. Found: C, 49.28; H, 7.29; N, 5.20.

O-(*5-Acetamido-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonic acid*)-(*2* → *6*)-*1,5-butylimino-1,5-dideoxy*-D-*galactitol* (**22**).—Compound **20** (46 mg) was deprotected as described for **21**, to give **22** in almost quantitative yield, $[\alpha]_D$ −8° (*c* 0.65, MeOH); ^{1}H NMR (CD_3OD): δ 0.97 (t, 3 H, CH_3 of *N*-*n*-butyl), 1.25–1.7 [m, 5 H, (CH_2)$_2$CH$_3$ of *N*-*n*-butyl and H-3*ax* of Neu5Ac], 1.92 (s, 3 H, AcN), 2.44 (t, 1 H, *J* 11–12 Hz, H-1*ax* of DNJ), 2.7–2.9 (m, 3 H, N-CH_2 of *N*-*n*-butyl and H-3*eq* of

Neu5Ac), and 3.09 (dd, 1 H, J_{gem} 11.6, $J_{1eq,2}$ 4.3 Hz, H-1*eq* of DNJ). Anal. Calcd for $C_{21}H_{38}N_2O_{12}$ (510.53): C, 49.40; H, 7.50; N, 5.49. Found: C, 49.14; H, 7.46; N, 5.27.

*1,5-Benzyloxycarbonylimino-6-*O*-tert-butyldiphenylsilyl-1,5-dideoxy-*D*-galactitol* (**23**).—A mixture of **8** (150 mg) and a catalytic amount of sodium methoxide in MeOH was stirred for 2.5 h at 0°C, then neutralized with Amberlite IR-120 (H^+) resin. The resin was filtered off and washed with MeOH. The combined filtrate and washings was concentrated to a syrup, which was chromatographed on a column of silica gel with 100:1 CH_2Cl_2–MeOH to give **23** (quant.), $[\alpha]_D$ −26° (*c* 0.5, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 1.03 (s, 9 H, *t*-butyl), 3.34 (dd, 1 H, J_{gem} 15, $J_{1ax,2}$ 2.5 Hz, H-1*ax*), 3.81, 3.89 (2 narrow m, H-2, 3), 3.93 (~ d, 1 H, J_{gem} 15 Hz, H-1*eq*), 3.98, 4.03 (2 dd, J_{gem} 11, $J_{5,6} = J_{5,6'} = 5$ Hz, H-6, 6′), 4.22 (dd, 1 H, $J_{3,4}$ 3, $J_{4,5}$ 6 Hz, H-4), 4.46 (~ q, 1 H, H-5), 5.03, 5.08 (2 d, 2 H, J_{gem} 12.3 Hz, OCH_2Ph), and 7.2–7.7 (m, 15 H, Ph-H). Anal. Calcd for $C_{30}H_{37}NO_6Si$ (535.66): C, 67.26; H, 6.96; N, 2.62. Found: C, 67.41; H, 7.07; N, 2.61.

*4,6-*O*-Benzylidene-1,5-benzyloxycarbonylimino-1,5-dideoxy-*D*-galactitol* (**25**).—BF_3 etherate (1 mL) was added, at 0°C, to a solution of **23** (389 mg) in CH_2Cl_2 (30 mL). The mixture was stirred for 2 h at room temperature, and washed successively with ice-cold M Na_2CO_3 and water, dried, and concentrated. The residue was chromatographed on a column of silica gel with 15:1 CH_2Cl_2–MeOH to give **24** (215 mg). This was dissolved in dry acetonitrile (30 mL), then Drierite (250 mg), benzaldehyde dimethyl acetal (0.22 mL, about 2 mol equiv) and a catalytic amount of *p*-toluenesulfonic acid were added, and the mixture was stirred overnight at room temperature. After neutralization with triethylamine the mixture was filtered and the solids were washed with MeOH. The combined filtrate and washings was concentrated, and the residue was chromatographed on a column of silica gel with 100:1 CH_2Cl_2–MeOH to afford **25** in 80% yield, $[\alpha]_D$ −69° (*c* 0.3, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 3.97 (dd, 1 H, J_{gem} 14, $J_{1ax,2}$ 2–3 Hz, H-1*ax*), 4.11 (~ d, 1 H, J_{gem} 14 Hz, H-1*eq*), 5.1, 5.15 (2 d, 2 H, OCH_2Ph), 5.44 (s, 1 H, C*H*Ph), and 7.25–7.45 (m, 10 H, Ph-H). Three narrow signals were clearly observed at δ 3.58 (dd, 1 H, *J* 4 and 2 Hz), 3.66 (~ s, 1 H, *J* 2–4 Hz), and 4.33 (~ t, 1 H, *J* 2–3 Hz), respectively, showing the $^1C_4$ conformation. Anal. Calcd for $C_{21}H_{23}NO_6$ (385.42): C, 65.44; H, 6.02; N, 3.63. Found: C, 65.19; H, 5.98; N, 3.82.

*4,6-*O*-Benzylidene-1,5-benzyloxycarbonylimino-3-*O*-chloroacetyl-1,5-dideoxy-*D*-galactitol* (**26**).—A mixture of **25** (149 mg) and 2,6-lutidine (0.27 mL) in CH_2Cl_2 (40 mL) was cooled to −20°C and chloroacetyl chloride (37 μL, 1.2 mol equiv) in CH_2Cl_2 (5 mL) was added. The mixture was stirred for 1 h at −20°C, washed successively with ice-cold M HCl and water, dried, and concentrated. The residue was chromatographed on a column of silica gel with 250:1 CH_2Cl_2–MeOH to give **26** (67%), $[\alpha]_D$ −28° (*c* 1, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 3.09 (br s, 1 H, OH-2), 4.14 (s, 2 H, COCH_2Cl), 4.27 (d, 1 H, J_{gem} 14.6 Hz, H-1*eq*), 4.54 (dd, 1 H, $J_{3,4}$ 2.4, $J_{4,5}$ 3.7 Hz, H-4), 4.76 (dd, 1 H, $J_{2,3}$ 5, $J_{3,4}$ 2.4 Hz, H-3), 5.14, 5.20 (2 d, 2 H, OCH_2Ph), 5.48 (s, 1 H, C*H*Ph), and 7.3–7.5 (m, 10 H, Ph-H). Anal. Calcd for $C_{23}H_{24}NO_7Cl$ (461.89): C, 59.80; H, 5.24; N, 3.03. Found: C, 59.67; H, 5.41; N, 2.85.

*2-*O*-Acetyl-4,6-*O*-benzylidene-1,5-benzyloxycarbonylimino-1,5-dideoxy-*D*-galactitol* (**28**).—Acetyl chloride (75 μL, 2 mol equiv) was added to a mixture of **26** (242 mg)

and pyridine (0.5 mL) in CH_2Cl_2 (20 mL) at 0°C. The mixture was stirred for 45 min at 0°C, diluted with CH_2Cl_2, successively washed with ice-cold M HCl and water, dried, and concentrated. The residue was chromatographed on a column of silica gel with 500:1 CH_2Cl_2–MeOH to afford **27** (264 mg), ^{1}H NMR ($CDCl_3$): δ 1.96 (s, 3 H, AcO), 4.12 (s, 2 H, COCH_2Cl), 4.60 (narrow m, 1 H, H-4), 5.03 (dd, 1 H, $J_{2,3}$ 6, $J_{3,4}$ 2.3 Hz, H-3), 5.08, 5.27 (2 d, 2 H, J_{gem} 12.3 Hz, OCH_2Ph), 5.19 (m, 1 H, H-2), 5.50 (s, 1 H, C*H*Ph), and 7.3-7.5 (m, 10 H, Ph-H).

A mixture of **27** (30 mg), pyridine (15 mL), and water (2 mL) was stirred overnight at room temperature, and then extracted with CH_2Cl_2. The extract was washed with ice-cold M HCl and water, dried, and concentrated. The residue was chromatographed on a column of silica gel with 100:1 CH_2Cl_2–MeOH to give the title compound **28** (24 mg), $[\alpha]_D$ −53° (*c* 0.5, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 2.02 (s, 3 H, AcO), 3.75 (dd, 1 H, $J_{2,3}$ 5, $J_{3,4}$ 2.4 Hz, H-3), 4.11 (dd, 1 H, J_{gem} 15, $J_{1ax,2}$ 4.4 Hz, H-1*ax*), 4.28 (d, 1 H, J_{gem} 15 Hz, H-1*eq*), 4.88 (t, 1 H, $J_{1ax,2} = J_{2,3}$ 4-5 Hz, H-2), 5.07, 5.26 (2 d, 2 H, J_{gem} 12.5 Hz, OCH_2Ph), 5.52 (s, 1 H, C*H*Ph), and 7.3–7.5 (m, 10 H, Ph-H). Anal. Calcd for $C_{23}H_{25}NO_7$ (427.44): C, 64.62; H, 5.90; N, 3.28. Found: C, 64.38; H, 5.86; N, 3.37.

O-(*Methyl 5-acetamido-4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate*)-(2 → 3)-2-O-*acetyl-4,6*-O-*benzylidene-1,5-benzyloxycarbonylimino-1,5-dideoxy*-D-*galactitol* (**29**).—A mixture of **28** (196 mg), **11** (535 mg, 1.7 mol equiv), and powdered molecular sieves 3 Å (750 mg) in CH_3CN was stirred for 6 h at room temperature, and then cooled to −10°C. *N*-Iodosuccinimide (351 mg, 3.4 mol equiv) and trifluoromethanesulfonic acid (14 μL, 0.34 mol equiv) were added, and the mixture was stirred overnight at −10°C. The solids were filtered off and washed with CH_2Cl_2. The combined filtrate and washings was successively washed with ice-cold M Na_2CO_3, $Na_2S_2O_3$, and water, dried, and concentrated. The residue was chromatographed on a column of silica gel with 3:2 AcOEt–hexane to give **29** (195 mg, 47% based on **28**), $[\alpha]_D$ −12° (*c* 1.5, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ 2.60 (dd, 1 H, J_{gem} 13, $J_{3eq,4}$ 4.6 Hz, H-3*eq* of Neu5Ac), 3.70 (s, 3 H, CO_2CH_3), 4.88 (m, 1 H, $J_{3ax,4}$ 12, $J_{3eq,4}$ 4.6, $J_{4,5}$ 10 Hz, H-4 of Neu5Ac), 5.08, 5.26 (2 d, 2 H, OCH_2Ph), 5.45 (s, 1 H, C*H*Ph), and 7.3–7.5 (m, 10 H, Ph-H). Anal. Calcd for $C_{43}H_{52}N_2O_{19}$ (900.86): C, 57.33; H, 5.82; N, 3.11. Found: C, 57.25; H, 5.79; N, 3.01.

O-(*Methyl 5-acetamido-4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate*)-(2 → 3)-2-O-*acetyl-1,5-dideoxy-1,5-methylimino*-D-*galactitol* (**31**).—Compound **29** (196 mg) was treated with 80% acetic acid (10 mL) overnight at 45°C. The mixture was concentrated and the residue was chromatographed on a column of silica gel with 3:1 AcOEt–hexane to give the debenzylidenated product, *O*-(methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2 → 3)-2-*O*-acetyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-D-galactitol (**30**) in almost quantitative yield, $[\alpha]_D$ +8° (*c* 2.5, CH_2Cl_2). To a solution of **30** (61 mg) in MeOH (5 mL) was added formaldehyde (0.11 mL of 37 wt% solution in water) and palladium hydroxide-on-carbon (60 mg). The mixture was stirred overnight in a hydrogen atmosphere. The catalyst was filtered off and washed with MeOH. The combined filtrate and washings was concentrated to a syrup, which was chromatographed on a column of silica gel with 15:1 CH_2Cl_2–MeOH to afford **31** (93%), $[\alpha]_D$ −4.5° (*c* 1, CH_2Cl_2); ^{1}H NMR ($CDCl_3$): δ (for the *N*-methyl-DNJ moiety) 2.23

(t, 1 H, $J_{gem} = J_{1ax,2} = 11$ Hz, H-1*ax*), 2.36 (s, 3 H, N-CH_3), 3.07 (dd, 1 H, J_{gem} 11, $J_{1eq,2}$ 5 Hz, H-1*eq*), and 5.24 (m, 1 H, $J_{1ax,2} = J_{2,3} = 10–11$ Hz, $J_{1eq,2}$ 5 Hz, H-2); δ (for the Neu5Ac moiety) 1.95 (t, 1 H, J 12–13 Hz, H-3*ax*), 2.64 (dd, 1 H, J_{gem} 12.8, $J_{3eq,4}$ 4.4 Hz, H-3*eq*), 3.82 (s, 3 H, CO_2CH_3), 4.37 (dd, 1 H, J_{gem} 12.3, $J_{8,9}$ 2.4 Hz, H-9), 4.86 (m, 1 H, $J_{3ax,4}$ 12, $J_{3eq,4}$ 4.4, $J_{4,5}$ 10 Hz, H-4), 5.34 (dd, 1 H, $J_{7,8}$ 8.2, $J_{6,7}$ 2.4 Hz, H-7), and 5.47 (m, 1 H, $J_{7,8}$ 8.2, $J_{8,9}$ 2.4, $J_{8,9'}$ 5.9 Hz, H-8). Six singlets for acetyl protons appeared at δ 1.89, 2.03, 2.05, 2.10, 2.14, and 2.15, respectively. Anal. Calcd for $C_{29}H_{44}N_2O_{17}$ (692.66): C, 50.28; H, 6.40; N, 4.04. Found: C, 50.07; H, 6.16; N, 3.98.

O-(*Methyl 5-acetamido-4,7,8,9-tetra*-O-*acetyl-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonate*)-(*2 → 3*)-*2*-O-*acetyl-1,5-butylimino-1,5-dideoxy*-D-*galactitol* (**32**). —A mixture of **30** (40 mg), 1-butanol (5 mL), butyraldehyde (87 μL), acetic acid (4.7 μL), water (80 μL), and palladium hydroxide-on-carbon (40 mg) was stirred overnight in a hydrogen atmosphere, and worked up as just described for **31**. The product was purified by column chromatography on silica gel with 10:1 CH_2Cl_2–MeOH to give **32** (90%), $[\alpha]_D$ −4.7° (*c* 0.3, CH_2Cl_2) ; ^{1}H NMR ($CDCl_3$): δ (for the *N*-*n*-butyl-DNJ moiety) 0.91 (t, 3 H, CH_3 of *N*-butyl), 1.2-1.55 [m, 4 H, $(CH_2)_2CH_3$], 2.35 (t, 1 H, $J_{gem} = J_{1ax,2} = 11$ Hz, H-1*ax*), 2.45–2.83 (m, 2 H, N-CH_2), 3.13 (dd, 1 H, J_{gem} 11, $J_{1eq,2}$ 5 Hz, H-1*eq*), and 5.19 (m, 1 H, $J_{1ax,2} = J_{2,3} = 10–11$ Hz, $J_{1eq,2}$ 5 Hz, H-2); δ (for the Neu5Ac moiety) 1.96 (t, 1 H, J 12–13 Hz, H-3*ax*), 2.64 (dd, 1 H, J_{gem} 13, $J_{3eq,4}$ 4.8 Hz, H-3*eq*), 3.81 (s, 3 H, CO_2CH_3), 4.37 (dd, 1 H, J_{gem} 12.3, $J_{8,9}$ 2.4 Hz, H-9), 4.86 (m, 1 H, $J_{3ax,4}$ 12, $J_{3eq,4}$ 4.8, $J_{4,5}$ 10 Hz, H-4), 5.34 (dd, 1 H, $J_{7,8}$ 8.6, $J_{6,7}$ 2.6 Hz, H-7), and 5.47 (m, 1 H, $J_{7,8}$ 8.6, $J_{8,9}$ 2.4, $J_{8,9'}$ 6 Hz, H-8). Six singlets for acetyl protons appeared at δ 1.88, 2.03, 2.04, 2.11, 2.136, and 2.143, respectively. Anal. Calcd for $C_{32}H_{50}N_2O_{17}$ (734.74): C, 52.31; H, 6.86; N, 3.81. Found: C, 52.19; H, 6.65; N, 3.58.

O-(*5-Acetamido-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonic acid*)-(*2 → 3*)-*1,5-dideoxy-1,5-imino*-D-*galactitol* (**33**).—Compound **29** (44 mg) in acetic acid (10 mL) was hydrogenolyzed in the presence of palladium black catalyst prepared from 80 mg of palladium chloride. The catalyst was filtered off and the filtrate was concentrated. *O*-Deacetylation and saponification of the methyl ester group, followed by chromatography on a column of Sephadex LH-20, gave **33** in quantitative yield, $[\alpha]_D$ +23° (*c* 0.5, 2:3 H_2O–EtOH); ^{1}H NMR (D_2O): δ 1.80 (t, 1 H, J 12 Hz, H-3*ax* of Neu5Ac), 2.03 (s, 3 H, AcN), and 2.75 (dd, 1 H, J_{gem} 12, $J_{3eq,4}$ 4.4 Hz, H-3*eq* of Neu5Ac); ion-spray MS (positive-ion mode): *m/z* 455.2 $[M + H]^+$ (base peak), and 477.1 $[M + Na]^+$; MS/MS (P = 455.2): *m/z* 291.2 (Neu5Ac fragment), 273 (Neu5Ac fragment $-H_2O$), and 163.5 (protonated DNJ fragment); for further details see ref. [17]. Anal. Calcd for $C_{17}H_{30}N_2O_{12}$ (454.43): C, 44.93; H, 6.65; N, 6.17. Found: C, 44.75; H, 6.63; N, 6.20.

O-(*5-Acetamido-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonic acid*)-(*2 → 3*)-*1,5-dideoxy-1,5-methylimino*-D-*galactitol* (**34**).—*O*-Deacetylation of **31** (48 mg) and saponification of the methyl ester group were performed as for the preparation of **21** and **22**, and the resulting **34** (quant.) was purified by column chromatography on Sephadex LH-20, $[\alpha]_D$ +22° (*c* 0.6, 2:1 MeOH–H_2O); ^{1}H NMR (D_2O): δ (for the *N*-methyl-DNJ moiety) 3.09 (s, 3 H, N-CH_3), 3.18 (t, 1 H, $J_{gem} = J_{1ax,2} = 11$-12 Hz,

H-1*ax*), 3.45 (narrow t, 1 H, H-5), 3.71 (dd, 1 H, J_{gem} 12, $J_{1eq,2}$ 5 Hz, H-1*eq*), 4.25 (dd, 1 H, $J_{2,3}$ 10, $J_{3,4}$ 2.6 Hz, H-3), 4.32 (m, 1 H, $J_{1ax,2}$ 11, $J_{2,3}$ 10, $J_{1eq,2}$ 5 Hz, H-2), and 4.46 (~ s, 1 H, H-4); δ (for the Neu5Ac moiety) 1.99 (t, 1 H, $J_{gem} = J_{3ax,4} = 12$ Hz, H-3*ax*), 2.11 (s, 3 H, AcN), and 2.95 (dd, 1 H, J_{gem} 12.4, $J_{3eq,4}$ 4.5 Hz, H-3*eq*). Anal. Calcd for $C_{18}H_{32}N_2O_{12}$ (468.45): C, 46.15; H, 6.89; N, 5.98. Found: C, 45.97; H, 6.81; N, 5.97.

O-(*5-Acetamido-3,5-dideoxy*-D-glycero-α-D-galacto-*2-nonulopyranosylonic acid*)-(2 → *3*)-*1,5-butylimino-1,5-dideoxy*-D-*galactitol* (**35**).—Compound **32** (40 mg) was deprotected and purified as just described for **34**, to give **35** in a quantitative yield, $[\alpha]_D$ +16° (*c* 0.6, MeOH); ^{1}H NMR (CD_3OD): δ 1.00 (t, 3 H, *J* 7 Hz, CH_3 of *n*-butyl), 1.41 (m, 2 H, CH_2CH$_3$), 1.65–1.85 (m, 3 H, CH_2CH$_2$CH$_3$ and H-3*ax* of Neu5Ac), 2.02 (s, 3 H, AcN), 2.82 (dd, 1 H, J_{gem} 13, $J_{3eq,4}$ 4.3 Hz, H-3*eq* of Neu5Ac), and 2.97 (br t, 1 H, *J* 11 H, H-1*ax* of DNJ). Anal. Calcd for $C_{21}H_{38}N_2O_{12}$ (510.53): C, 49.40; H, 7.50; N, 5.49. Found: C, 49.38; H, 7.33; N, 5.43.

Acknowledgements

This work was supported in part by grants-in-aid for Scientific Research (C) and for Developmental Scientific Research from the Japanese Ministry of Education, Science and Culture (Nos. 05660386 and 05556019) and by the Naito Foundation. The authors thank Professor K. Hirotsu, Faculty of Science, Osaka City University for the X-ray crystallographic analysis.

References

[1] H.-K. Ishida, H. Ishida, M. Kiso, and A. Hasegawa, *Carbohydr. Res.*, 260 (1994) C1–C6.

[2] N.K. Sauter, J.E. Hanson, G.D. Glick, J.H. Brown, R.L. Crowther, S.-J. Park, J.J. Skehel, and D.C. Wiley, *Biochemistry*, 31 (1992) 9609–9621, and references therein.

[3] Y. Suzuki, T. Nakao, T. Ito, N. Watanabe, Y. Toda, X. Guiyun, T. Suzuki, T. Kobayashi, Y. Kimura, A. Yamada, K. Sugawara, H. Nishimura, F. Kitame, K. Nakamura, E. Deya, M. Kiso, and A. Hasegawa, *Virology*, 189 (1992) 121–131.

[4] M. von Itzstein, W.-Y. Wu, G.B. Kok, M.S. Pegg, J.C. Dyason, B. Jin, T.V. Phan, M.L. Smythe, H.F. White, S.W. Oliver, P.M. Colman, J.N. Varghese, D.M. Ryan, J.M. Woods, R.C. Bethell, V.J. Hotham, J.M. Cameron, and C.R. Penn, *Nature*, 363 (1993) 418–423, and references therein.

[5] (a) G. Xu, T. Suzuki, G. Hanagata, E. Deya, M. Kiso, A. Hasegawa, and Y. Suzuki, *J. Biochem. (Tokyo)*, 113 (1993) 304–307; (b) Y. Suzuki, K. Sato, M. Kiso, and A. Hasegawa, *Glycoconjugate J.*, 7 (1990) 349–356.

[6] S. Schenkman, M.-S. Jiang, G.W. Hart, and V. Nussenzweig, *Cell*, 65 (1991) 1117–1125.

[7] F. Vandekerckhove, S. Schenkman, L. Pontes de Carvalho, S. Tomlinson, M. Kiso, M. Yoshida, A. Hasegawa, and V. Nussenzweig, *Glycobiology*, 2 (1992) 541–548.

[8] (a) A. Hasegawa and M. Kiso, in H. Ogura, A. Hasegawa, and T. Suami (Eds.), *Carbohydrates -Synthetic Methods and Applications in Medicinal Chemistry*, Kodansha/VCH, Tokyo/Weinheim, 1992, pp 243–266; (b) B.K. Brandley, M. Kiso, S. Abbas, P. Nikrad, O. Srivastava, C. Foxall, Y. Oda, and A. Hasegawa, *Glycobiology*, 3 (1993) 633–639.

[9] (a) Y. Yoshikuni, *Trends Glycosci. Glycotechnol. (TIGG)*, 3 (1991) 184–192; (b) Y. Nishimura, in Atta-ur-Rahmann (Ed.), *Studies in Natural Products Chemistry*, Vol. 10, Elsevier, Amsterdam, 1992, pp

495–583; (c) G.B. Karlsson, T.D. Butters, R.A. Dwek, and F.M. Platt, *J. Biol. Chem.*, 268 (1993) 570–576.

[10] (a) H. Furui, M. Kiso, and A. Hasegawa, *Carbohydr. Res.*, 229 (1992) C1–C4; (b) M. Kiso, H. Furui, K. Ando, and A. Hasegawa, *J. Carbohydr. Chem.*, 12 (1993) 673–677; (c) M. Kiso, K. Ando, H. Furui, H. Ishida, and A. Hasegawa, *ibid.*, 12 (1993) 985–990; (d) M. Kiso, H. Furui, K. Ando, H. Ishida, and A. Hasegawa, *Bioorg. Med. Chem.*, 2 (1994) 1295–1308.

[11] M. Kiso, H. Katagiri, H. Furui, and A. Hasegawa, *J. Carbohydr. Chem.*, 11 (1992) 627–644.

[12] (a) H. Paulsen and K. Todt, *Adv. Carbohydr. Chem.*, 23 (1968) 115–232; (b) H. Paulsen, M. Matzke, B. Orthen, R. Nuck, and W. Reutter, *Liebigs Ann. Chem.*, (1990) 953–963.

[13] A. Hasegawa, H. Ohki, T. Nagahama, H. Ishida, and M. Kiso, *Carbohydr. Res.*, 212 (1991) 277–281.

[14] (a) P. Fügedi and P.J. Garegg, *Carbohydr. Res.*, 149 (1986) C9–C12; (b) O. Kanie, M. Kiso, and A. Hasegawa, *J. Carbohydr. Chem.*, 7 (1988) 501–506; (c) T. Murase, H. Ishida, M. Kiso, and A. Hasegawa, *Carbohydr. Res.* 184 (1988) c1–c4.

[15] (a) G.H. Veeneman, S.H. van Leeuwen, and J.H. van Boom, *Tetrahedron Lett.*, 31 (1990) 1331–1334; (b) P. Konradsson, D.R. Mootoo, R.E. McDevitt, and B. Fraser-Reid, *J. Chem. Soc., Chem. Commun.*, (1990) 270–272.

[16] A. Marra and P. Sinaÿ, *Carbohydr. Res.*, 187 (1989) 35–42.

[17] A. Kondo, K. Ando, H. Ishida, I. Kato, A. Hasegawa, and M. Kiso, *J. Carbohydr. Chem.*, 13 (1994) 545–554.

[18] (a) R.G. Rice and E.J. Kohn, *J. Am. Chem. Soc.*, 77 (1955) 4052–4054; (b) C. Ainsworth, *J. Am. Chem. Soc.*, 78 (1956) 1635–1636.

[19] G.M. Sheldrick, SHELX86: Programs for the Automatic Solution of Crystal Structures, University of Göttingen, Germany, 1986.

[20] W.R. Busing, K.O. Martin, and H.S. Levy, ORFLS, Oak Ridge National Laboratory Report, ORNL-TM-305, Oak Ridge, TN, USA, 1965.