Inhibitors of *endo*-α-mannosidase. Part III.[1] Congeners of 1-deoxy-3-*O*-(α-D-glucopyranosyl)-mannojirimycin modified in the glucose unit

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The syntheses of congeners of 1-deoxy-3-*O*-(α-D-glucopyranosyl)-mannojirimycin (**1**), a strong inhibitor of the glycoprotein-processing *endo*-mannosidase, are described. The chemical modifications of **1** involved all monodeoxygenations and mono-*O*-methylations of the glucose unit and the replacement of this unit by D-galactose, D-xylose, and 2-chloro-2-deoxy-D-glucose. As reported previously, none of the modifications of **1**, including deoxygenations and *O*- and *N*-methylations of the deoxymannojirimycin unit, improved the inhibitory properties, but demonstrated the high specificity in the recognition of **1** by the enzyme and allowed the assignment of intermolecular hydrogen bonds of the inhibitor · enzyme complex. Essential for complex formation were found NH-5, OH-2, OH-4, and OH-6 of the DMJ unit, as well as OH-3′, OH-4′, and CH_2-6′ of the glucose unit. The residual activities on deoxygenating the OH-2′ and OH-6′ groups of **1** suggest their involvement at the periphery of the binding site.

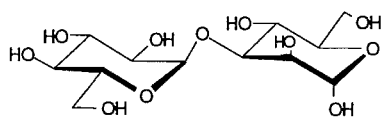
ULRIKE SPOHR et MIMI BACH. Can. J. Chem. **71**, 1943 (1993).

On décrit la synthèse de congénères de la 1-désoxy-3-*O*-(α-D-glucopyranosyl)mannojirimycine (**1**), un inhibiteur puissant de l'*endo*-mannosidase qui peut agir sur les glycoprotéines. Les modifications chimiques de **1** impliquent toutes des monodésoxygénations et des mono-*O*-méthylations de l'unité glucose et le remplacement de cette unité par du D-galactose, du D-xylose et du 2-chloro-2-désoxy-D-glucose. Comme il a été rapporté antérieurement, aucune des modifications du composé **1**, y compris les désoxygénations et les *O*- et *N*-méthylations de l'unité désoxymannojirimycine, n'améliore les propriétés inhibitrices; toutefois, ces modifications ont permis de démontrer la grande spécificité de la reconnaissance du composé **1** par l'enzyme et ont permis d'attribuer les liaisons hydrogènes intramoléculaires du complexe inhibiteur · enzyme. On a trouvé que les unités NH-5, OH-2, OH-4 et OH-6 de l'unité DMJ ainsi que les unités OH-3′, OH-4′ et CH_2-6′ de l'unité glucose sont essentielles à la formation de complexes. Les activités résiduelles observées lors de la désoxygénation des groupes OH-2′ et OH-6′ du composé **1** suggèrent qu'ils sont impliqués à la périphérie du site de liaison.

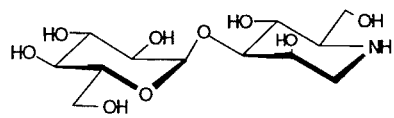
[Traduit par la rédaction]

Introduction

The glycoprotein-processing *endo*-α-D-mannosidase discovered by Spiro and Lubas (1, 2) releases the disaccharide αDGlc(1→3)DMan from immature *N*-linked glycoproteins containing the incompletely deglucosylated polymannose oligosaccharide $GlcMan_9GlcNAc_2$ with the formation of $Man_8GlcNAc_2$. The action of the enzyme was effectively inhibited by the disaccharide 1-deoxy-3-*O*-(α-D-glucopyranosyl)-mannojirimycin (**1**) (3).



αDGlc(1→3)DMan



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In the search for inhibitors with even greater potencies or better bioavailability all substituents of **1** were chemically modified, mainly by monodeoxygenation and mono-*O*-methylation and the effects of alteration on the inhibitory potencies observed, thus allowing for the assessment of intermolecular hydrogen bonds of the oligosaccharide · enzyme complex as demonstrated by Lemieux and coworkers (4, 5) for the complexation of oligosaccharides by lectins. The biological data of this investigation were published recently (3). While the chemical modifications of the deoxymannojirimycin unit of **1** are reported in the accompanying paper (6), the syntheses and properties of congeners of **1** modified in the glucose unit are described in this communication.

Discussion

Synthesis

The syntheses of congeners of αDGlc(1→3)-1-DMJ (**1**) (6) modified in the glucose unit are outlined in Schemes 1 and 2. The 1-deoxymannojirimycin derivative **2** (6, 7), which has a free hydroxyl group at the 3-position, was used as the glycosyl acceptor. The chloro sugars **3** (2-*O*-methyl), **5** (3-deoxy) (8), **6** (4-deoxy), **10** (4-*O*-methyl), **11** (galactosyl) (9), **12** (6-deoxy), **13** (xylosyl) (10), **50** (3-*O*-methyl), and **51** (6-*O*-acetyl) (11) were prepared from the corresponding hemiacetals by treatment with oxalyl chloride and *N*,*N*-dimethylformamide (12, 13). These precursors were de-

[1]For Parts I and II, see refs. 27 and 5.

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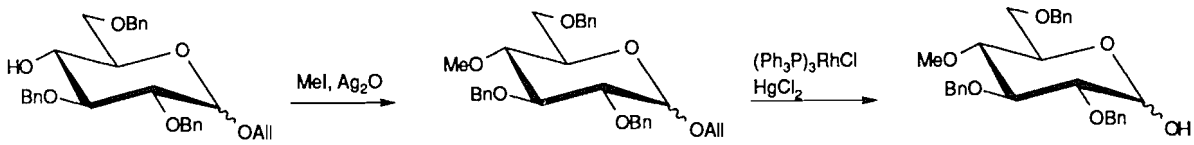


7 **8** **9**

scribed in the literature (14–19), except for 4-*O*-methyl **9**, which was synthesized from an anomeric mixture of **7** (20) by conventional methylation with methyl iodide and sodium hydride to give **8**, followed by deallylation (**9**).

The 2-chloro-2-deoxy and 2-deoxy chlorides **4** (21) and **49** (22) were prepared from 3,4,6-tri-*O*-benzyl-D-glucal as reported.

Since the glycosyl acceptor **2** was of low reactivity (6), all glycosidation reactions were promoted by silver trifluoromethanesulfonate in the presence of *sym*-collidine and 4 Å molecular sieves in dichloromethane at low temperature. Usually, an α:β mixture was produced with a ratio varying from 3:1 to 0.9:1 in a combined yield of 93–66%. Higher α:β ratios were observed for couplings employing galactosyl chloride **11**, 2-*O*-methyl, 3-*O*-methyl, and 4-*O*-methyl chlorides **3**, **50**, and **10** than for those employing 3-deoxy, 4-deoxy, 6-deoxy, and 2-chloro-2-deoxy halides **5, 6, 12,** and **4**. However, in the preparation of the 2′-deoxy disaccharide **52** (73% yield) (Scheme 2), no significant amount of the β-isomer was detected as this was likely to be instable under the reaction conditions. In most of the cases, the anomers could be separated by column chromatography, except for the 4′-*O*-methyl, 6′-deoxy, and xylosyl disaccharides **22, 26**, and **28**, which were isolated as anomeric mixtures, but could be separated readily after deacetylation.

The ^{1}H NMR spectra of the coupling products were broad and showed duplications of the acetyl signals due to the restricted rotation of the benzyloxycarbonyl group about the C—N amide bond (6). Signal duplication was also observed in the ^{13}C NMR spectra. For unambiguous identification, both α- and β-anomers were isolated and characterized. To ascertain the anomeric configurations, C—H coupled ^{13}C NMR spectra were recorded for all β-anomers and some α-anomers. The C-1′ coupling constants $J_{C1',H1'}$ for the β-anomers were found in the range 158.0–162.3 Hz and those for the α-anomers from 166.2 to 171.8 Hz, in agreement with earlier observations (23).

Deacetylations of α-anomers **14, 16, 18, 20, 24**, and the α,β-mixtures **22** + **23**, **26** + **27**, **28** + **29** (Scheme 1) were carried out with triethylamine in aqueous methanol for several days at room temperature to provide the alcohols **30–40** in overall yields of 75% as well as some unreacted starting acetate. A different approach was employed for the 2′-deoxy, 3′-*O*-methyl, and 6′-*O*-acetyl disaccharides **52, 53**, and **55** (Scheme 2). Treatment with methanolic sodium methoxide produced the oxazolidone derivatives **57, 58**, and **59** in high yields (6). 6′-OH disaccharide **59** was converted into the 6′-*O*-methyl derivative **60** (91% yield) by reaction with methyl iodide and silver oxide in toluene. The cyclic carbamates in **57, 58**, and **60** were readily saponified with alcoholic alkali (6) to afford the secondary amines **61, 62**, and **63** in an overall yield of 80% from the benzyloxycarbonyl precursors **52, 53**, and **55**. Catalytic hydrogenation over 5% palladium-on-carbon in the presence of 1 equivalent of hydrochloric acid provided the disaccharides **41–48** (Scheme 1) and **64–66** (Scheme 2) after gel filtration in high yields.

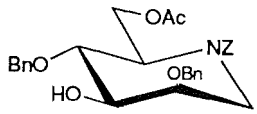
The ^{1}H and ^{13}C NMR data of all deblocked compounds are presented in Tables 1 and 2 and fully confirm the structural assignments. The ^{1}H NMR coupling constants are in agreement with the $^4C_1$ conformation of the pyranose rings. Comparison of the ^{1}H NMR chemical shifts of the DMJ unit of all congeners with those of the parent compound **1** shows that modification of the glucose unit has only minor effects on these shifts, indicating that the conformational preferences about the interglycosidic torsion angles are similar in all compounds. Conformational analysis based on HSEA calculations and nuclear Overhauser enhancements predicted a conformational preference of Φ/Ψ near −50°/−10° (6).

The recognition of αDGlc(1→3)-1-DMJ (1) by endo-*mannosidase*

While αDGlc(1→3)-1-DMJ (**1**) proved a strong inhibitor of the cleavage of $Glc Man_9 GlcNAc$ by *endo*-mannosidase with IC_{50} = 1.7 μmol, modifications of its substituents either led to essentially inactive or weaker compounds, as reported recently (3) and summarized in Fig. 1.

The data require that both OH-3′ and OH-4′ of the glucose unit are involved in strong polar interactions deep in the combining site, since deoxygenation at these positions to give **43** and **44** and *O*-methylation to give **65** and **45** abolished the activity. The polar interaction undergone by equatorial OH-4′ of **1** can be maintained by an axial OH-4′, since galactosyl congener **46** retained substantial activity (17%). That the CH_2-6′ grouping of the glucose unit is essential for providing the required complementarity for complex formation, is indicated by the inactivity displayed by the xylosyl derivative **48**, which has the CH_2OH-6′ grouping replaced by hydrogen. Although OH-6′ can be substituted for by hydrogen to give 6′-deoxy **47** while still retaining 17% activity, *O*-methylation led to inactive **66** for steric reasons. It is therefore expected that OH-6′ is hydrogen bonded to amino acids near the periphery of the binding site (4). Also, OH-2′ of this unit must be located at the periphery of the binding site and possibly interact with water, since deoxygenation (**64**), *O*-methylation (**41**), and chlorination (**42**) produced active, albeit weaker, inhibitors.

Equally intimate as the acceptance of the glucose unit of **1** is the binding of the DMJ unit. While *N*-methylation of **1** produced an inhibitor with a residual activity of around 10%, *N*-*n*-propylation led to an inactive compound, indicating a strong steric interference to binding. It has been observed that *N*-alkylation of 5-deoxy-5-imino sugar analogues may or may not increase the inhibitory properties (24–26). Deoxygenation or *O*-methylation at the 2-, 4-, and 6-positions of **1** virtually abolished the activity, although a marginal activity was detected for 4-deoxy and 4-*O*-methyl congeners. The strong



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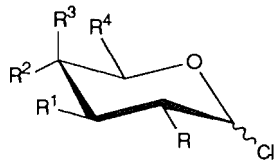
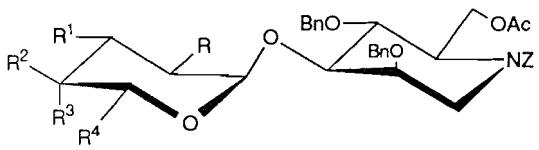


AgOTf →

	R	R^1	R^2	R^3	R^4
3	***OMe***	OBn	OBn	H	CH_2OBn
4	***Cl***	OBn	OBn	H	CH_2OBn
5	OBn	***H***	OBn	H	CH_2OBn
6	OBn	OBn	***H***	H	CH_2OBn
10	OBn	OBn	***OMe***	H	CH_2OBn
11	OBn	OBn	***H***	***OBn***	CH_2OBn
12	OBn	OBn	OBn	H	***CH_3***
13	OBn	OBn	OBn	H	***H***



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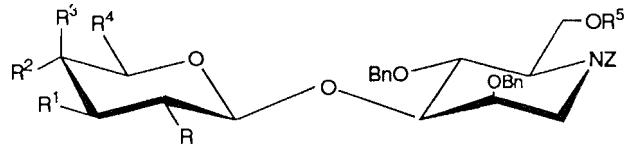


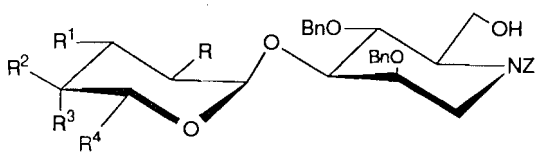
	R	R^1	R^2	R^3	R^4
14	***OMe***	OBn	OBn	H	CH_2OBn
16	***Cl***	OBn	OBn	H	CH_2OBn
18	OBn	***H***	OBn	H	CH_2OBn
20	OBn	OBn	***H***	H	CH_2OBn
(22	OBn	OBn	***OMe***	H	CH_2OBn)
24	OBn	OBn	***H***	***OBn***	CH_2OBn
(26	OBn	OBn	OBn	H	***CH_3***)
(28	OBn	OBn	OBn	H	***H***)

	R	R^1	R^2	R^3	R^4	R^5
15	***OMe***	OBn	OBn	H	CH_2OBn	Ac
17	***Cl***	OBn	OBn	H	CH_2OBn	Ac
19	OBn	***H***	OBn	H	CH_2OBn	Ac
21	OBn	OBn	***H***	H	CH_2OBn	Ac
(23	OBn	OBn	***OMe***	CH_2OBn	Ac)	
25	OBn	OBn	***H***	***OBn***	CH_2OBn	Ac
(27	OBn	OBn	OBn	H	***CH_3***	Ac)
(29	OBn	OBn	OBn	H	***H***	Ac)

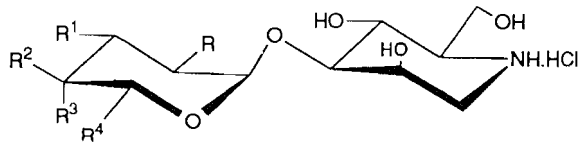
↓ NEt_3 (23, 27, 29)

35	OBn	OBn	***OMe***	H	CH_2OBn	H
38	OBn	OBn	OBn	H	***CH_3***	H
40	OBn	OBn	OBn	H	***H***	H

↓ NEt_3



H_2, Pd/C, HCl →

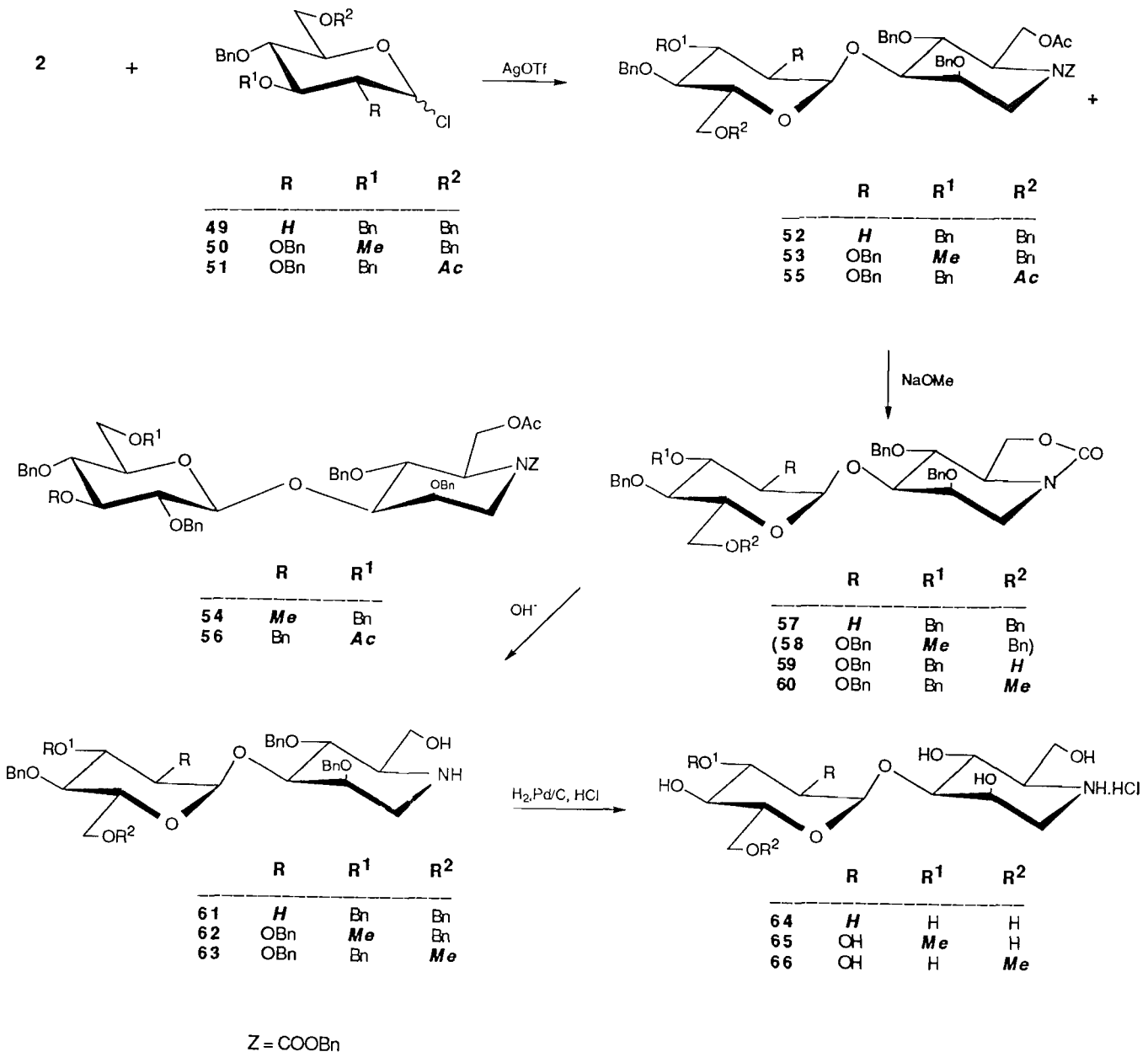


	R	R^1	R^2	R^3	R^4
30	***OMe***	OBn	OBn	H	CH_2OBn
31	***Cl***	OBn	OBn	H	CH_2OBn
32	OBn	***H***	OBn	H	CH_2OBn
33	OBn	OBn	***H***	H	CH_2OBn
34	OBn	OBn	***OMe***	H	CH_2OBn
36	OBn	OBn	***H***	***OBn***	CH_2OBn
37	OBn	OBn	OBn	H	***CH_3***
39	OBn	OBn	OBn	H	***H***

	R	R^1	R^2	R^3	R^4
41	***OMe***	OH	OH	H	CH_2OH
42	***Cl***	OH	OH	H	CH_2OH
43	OH	***H***	OH	H	CH_2OH
44	OH	OH	***H***	H	CH_2OH
45	OH	OH	***OMe***	H	CH_2OH
46	OH	OH	***H***	***OH***	CH_2OH
47	OH	OH	OH	H	***CH_3***
48	OH	OH	OH	H	***H***

Z = COOBn

SCHEME 1



	R	R^1	R^2
49	*H*	Bn	Bn
50	OBn	*Me*	Bn
51	OBn	Bn	*Ac*

	R	R^1	R^2
52	*H*	Bn	Bn
53	OBn	*Me*	Bn
55	OBn	Bn	*Ac*

	R	R^1
54	*Me*	Bn
56	Bn	*Ac*

	R	R^1	R^2
57	*H*	Bn	Bn
(58	OBn	*Me*	Bn)
59	OBn	Bn	*H*
60	OBn	Bn	*Me*

	R	R^1	R^2
61	*H*	Bn	Bn
62	OBn	*Me*	Bn
63	OBn	Bn	*Me*

	R	R^1	R^2
64	*H*	H	H
65	OH	*Me*	H
66	OH	H	*Me*

SCHEME 2

inhibitory properties towards glycosidases of glycosylamines, 5-amino-5-deoxyhexopyranoses, and 1,5-dideoxy-1,5-iminohexitols have been ascribed to the formation of an ion pair at the catalytic site of the protonated inhibitor and a negatively charged group of the protein likely involved in catalysis (27). In the case of the *endo*-mannosidase, the thus adjusted conformation of the catalytic site requires that all three hydroxyl groups of the DMJ unit of **1** are available for interaction with polar or charged groups of the protein. It is interesting that the active site adjusts differently in the complexation of the neutral O-5 analogue of **1**, namely, αDGlc(1→3)-1,5-anhydro-D-mannitol (3, 28), in which case the replacement of OH-2 by hydrogen increased the activity 40-fold.

Besides these polar interactions, it is to be expected that the complex is stabilized by hydrophobic interactions between nonpolar side chains of the protein and nonpolar patches on the sides of the pyranose rings, such as the α-side of the DMJ unit with CH-1, CH-2, CH-3, and CH-5.

Further synthetic and biological studies of the substrate specificity of *endo*-mannosidase are the subject of a different communication.[3]

Experimental

General methods

The ^{1}H NMR spectra were measured at 300 MHz and 360 MHz (Bruker AM 300 and WM 360) with tetramethylsilane as internal standard for $CDCl_3$ solutions. Reference standard for D_2O solutions was acetone (2.225 ppm). The ^{13}C NMR spectra were recorded at 75 MHz with dioxane (67.4 ppm) as reference for D_2O solutions and the $CDCl_3$ signal (77.0 ppm) as reference in $CDCl_3$

[3]U. Spohr, M. Bach, S.E.H. Moore, and R.G. Spiro. Manuscript in preparation.

TABLE 1. ^{1}H NMR chemical shifts (δ, ppm) and coupling constants (Hz) for αDGlc(1→3)-1-DMJ (**1**) and related structures[a]

	Derivatives of **1**											
	1	2′-Deoxy **64**	2′-OMe **41**	2′-Chloro-2′-deoxy **42**	3′-Deoxy **43**	3′-OMe **65**	4′-Deoxy **44**	4′-OMe **45**	4′-Epi **46**	6′-Deoxy **47**	6′-OMe **66**	5′-Dehydroxy-methylene **48**
1-DMJ unit												
H-1 ($J_{1,2}$)	3.42 (2.5)	3.38 (3.0)	3.40 (3.0)	3.38 (2.7)	3.45 (3.5)	3.44 (3.0)	3.44 (3.0)	3.42 (3.0)	3.44 (3.0)	3.42 (3.5)	3.41 (3.0)	3.42 (3.0)
H-1′ ($J_{1',1}$)	3.29 (13.5)	3.25 (13.5)	3.26 (13.5)	3.24 (13.5)	3.32 (13.5)	3.30 (13.5)	3.30 (13.5)	3.29 (13.5)	3.29 (13.5)	3.30 (13.5)	3.29 (13.5)	3.28 (13.5)
H-2 ($J_{1',2}$)	4.45 (<1)	4.38 (<1)	4.41 (<1)	4.41 (<1)	4.49 (<1)	4.45 (<1)	4.46 (1.0)	4.41 (<1)	4.48 (<1)	4.39 (<1)	4.41 (<1)	4.43 (<1)
H-3 ($J_{3,4}$)	3.81 (10.0)	3.75	3.80 (10.0)	3.81 (10.0)	3.85 (10.0)	3.81 (10.0)	3.80 (9.5)	3.79 (9.5)	3.82	3.78 (9.5)	3.77 (10.0)	3.78 (10.0)
H-4 ($J_{4,5}$)	4.08 (10.0)	3.97 (10.0)	4.05 (10.0)	4.05 (10.0)	4.09 (10.0)	4.09 (10.0)	4.08 (9.5)	4.06 (9.5)	4.06 (10.0)	4.07 (10.0)	4.07 (10.0)	4.08 (10.0)
H-5 ($J_{5,6}$)	3.21 (3.0)	3.17 (3.5)	3.18 (3.0)	3.15 (3.5)	3.25 (3.0)	3.24 (3.5)	3.24 (3.0)	3.25 (3.0)	3.23 (3.5)	3.21 (3.0)	3.22 (3.0)	3.21 (3.0)
H-6 ($J_{6,6'}$)	4.00 (12.5)	3.97	4.00 (12.5)	3.99 (12.5)	4.02 (12.5)	4.02 (13.0)	4.02 (12.5)	4.01 (12.0)	4.00	4.00 (12.5)	4.05 (13.0)	4.01 (13.5)
H-6′ ($J_{5,6'}$)	3.89 (6.7)	3.88 (6.5)	3.87 (6.5)	3.86 (6.5)	3.89 (6.5)	3.89 (6.2)	3.89 (6.5)	3.89 (6.0)	3.89 (6.5)	3.89 (6.5)	3.88 (6.0)	3.88 (6.0)
α**D**Glc unit												
H-1 ($J_{1,2}$)	5.25 (4.0)	5.27 (3.5)	5.46 (4.0)	5.34 (2.5)	5.15 (3.5)	5.23 (3.5)	5.28 (4.0)	5.23 (3.5)	5.27 (3.5)	5.18 (4.0)	5.23 (3.5)	5.22 (3.5)
H-2 ($J_{2,3}$)	3.58 (10.0)	1.72 (12.0)[b]	3.32 (9.5)	3.92	3.84 (11.5)	3.62 (9.5)	3.50 (10.0)	3.61 (9.5)	3.85 (10.0)	3.58 (10.0)	3.56 (10.0)	3.56 (9.5)
H-3 ($J_{3,4}$)	3.78 (9.5)	3.98 (9.0)	3.77 (9.5)	3.91 (9.5)	1.84 (11.5)[d]	3.54 (9.0)	4.01 (12.0)	3.87 (9.5)	3.94	3.72 (9.5)	3.79 (9.5)	3.71
H-4 ($J_{4,5}$)	3.43 (9.5)	3.38 (9.5)	3.43 (9.5)	3.49 (9.5)	3.67	3.49 (9.0)	1.46 (12.0)[e]	3.26 (9.5)	4.01	3.18 (9.5)	3.43 (9.5)	3.75–3.61[f]
H-5 ($J_{5,6}$)	3.81	3.76	3.78	3.91	3.75	3.83	4.10 (3.1)	3.81	4.10 (6.0)	3.89 (6.5)	3.90 (2.5)	
H-6 ($J_{6,6'}$)	3.86	*	3.84[c]	*	3.85	3.77[c]	3.68 (12.5)	3.81[c]	3.74 (0)	1.27	3.70 (11.0)	—
H-6′ ($J_{5,6'}$)	*	*	3.78[c]	3.77[c]	*	3.75	3.60 (6.5)	3.86[c]	3.74 (6.0)	—	3.68 (4.5)	—
CH_3O	—	—	3.50	—	—	3.62	—	3.59	—	—	3.39	—

[a]0.03 M solutions in D_2O with acetone as internal reference at 2.225 ppm were measured at 360 MHz and 295 K.
[b]H-2eq 2.29 ppm, $J_{2eq,2ax}$ 13.5 Hz, $J_{1,2eq}$ <1 Hz.
[c]Expected to be within ± 0.03 ppm.
[d]H-3eq 2.19 ppm, $J_{3eq,3ax}$ 11.5 Hz, $J_{3eq,4} \sim J_{2,3eq}$ 4.5 Hz.
[e]H-4eq 2.02 ppm, $J_{4eq,4ax}$ 12.7 Hz, $J_{3,4eq}$ 5.0 Hz, $J_{4eq,5}$ 2.0 Hz.
[f]H-5′ 3.75–3.61 ppm.
*Could not be assigned.

TABLE 2. ^{13}C NMR chemical shifts (δ, ppm) for αDGlc(1→3)-1-DMJ (**1**) and related structures[a]

		Derivatives of **1**									
	1	2′-Deoxy **64**	2′-OMe **41**	2′-Chloro-2′-deoxy **42**	3′-Deoxy **43**	3′-OMe **65**	4′-Deoxy **44**	4′-OMe **45**	4′-Epi **46**	6′-Deoxy **47**	6′-OMe **66**	5′-Dehydroxy-methylene **48**
1-DMJ unit												
C-1	48.44	48.37	48.34	48.28	48.31	48.30	48.26	48.32	48.35	48.31	48.28	48.35
C-2	66.37[b]	65.91[b]	66.06[b]	65.88[b]	65.95[b]	65.89[b]	65.91[b]	65.98[b]	66.21[b]	65.80[b]	66.64[b]	66.01[b]
C-3	81.67	80.53	81.22[c]	81.27	80.79	81.13	81.16	81.26	81.42	81.33	81.30	81.03
C-4	67.24[b]	66.40[b]	66.79[b]	66.29[b]	66.61[b]	66.55[b]	66.63[b]	66.70[b]	66.92[b]	66.67[b]	67.40[b]	66.73[b]
C-5	61.12	61.17	61.33	61.24	61.12	60.97	61.08	60.95	61.12	61.07	61.08	61.08
C-6	59.31	58.83	58.96	58.81	58.82	58.78	58.83	58.92	59.31	58.79	58.83	58.87
αDGlc unit												
C-1	101.50	100.92	98.61	101.06	100.30	101.55	102.27	101.35	101.71	101.49	101.53	101.60
C-2	72.49	37.59	80.87[c]	61.24	67.56	72.02	74.28	72.47[c]	69.43	72.72	72.39	72.48
C-3	73.57	68.82	72.57	73.56[c]	35.15	83.41	67.78	73.37[c]	70.07	73.32	73.44	73.74
C-4	70.35	71.81	70.40	71.12	65.03	69.78	34.92	80.10	70.07	75.75	70.39	70.11
C-5	73.27	73.80	73.08	73.47[c]	74.11	73.39	70.37	72.32[c]	72.39	69.30	72.01	62.63
C-6	61.34	61.51	61.35	61.26	61.42	61.23	64.44	61.08	62.05	17.35	71.79	—
CH_3O	—	—	58.36	—	—	61.08	—	60.95	—	—	59.49	—

[a]0.05 M solutions in D_2O with dioxane as internal reference at 67.4 ppm were measured at 75 MHz and 295 K. The assignments were made by inspection and are tentative.
[b,c] assignments are tentative and may be reversed.

solutions. Optical rotations were measured at room temperature (23 ± 1°C) in a 1-dm cell on a Perkin–Elmer 241 polarimeter. Thin-layer chromatography was performed on precoated plates of silica gel (60-F254, E. Merck, Darmstadt) and visualized by spraying with 5% sulfuric acid in ethanol followed by heating. For column chromatography silica gel 60 (230–400 mesh, E. Merck, Darmstadt) and distilled solvents were used. Solvents and reagents were purified and dried according to standard procedures. Melting points are uncorrected.

3,4,6-Tri-O-*benzyl-2*-O-*methyl-α,β-*D-*glucopyranosyl chloride (3)*

Oxalyl chloride (97 μL, 1.11 mmol) was added to a stirred solution of 3,4,6-tri-*O*-benzyl-2-*O*-methyl-D-glucopyranose (14) (385 mg, 0.83 mmol) and *N,N*-dimethylformamide (91 μL, 1.28 mmol) in dichloromethane (9 mL). The mixture was stirred for 20 min and then poured into ice-water. The organic solution was washed with ice-water, dried, and evaporated to provide chloride **3** (quant.) as a syrup. ^1H NMR ($CDCl_3$, α:β = 1:2.2) δ: 7.4–7.05 (m, 15H, 3Ph), 6.26 (d, $J_{1\alpha,2}$ 4.0 Hz, H-1α), 5.09 (d, $J_{1\beta,2}$ 8.5 Hz, H-1β), 4.08 (m, $J_{4,5}$ 10.0 Hz, H-5α), 3.96 (t, $J_{2,3} \sim J_{3,4}$ 9.5 Hz, H-3α), 3.67 (s, CH_3O-β), 3.54 (s, CH_3O-α).

2,4,6-Tri-O-*benzyl-3-deoxy-α-*D-ribo-*hexopyranosyl chloride (5) (8)*

2,4,6-Tri-*O*-benzyl-3-deoxy-D-*ribo*-hexopyranose (15) (251 mg, 0.58 mmol) was converted into chloride **5** just prior to use as reported for the preparation of **3**. ^1H NMR ($CDCl_3$, α:β = 3:1) δ: 7.45–7.20 (m, 15H, 3Ph), 6.19 (bd, 1H, $J_{1,2}$ 3.5 Hz, $J_{1,3eq} < 1$ Hz, H-1), 5.21 (d, $J_{1,2}$ 8.0 Hz, H-1β), 3.97 (ddd, $J_{4,5}$ 10.0 Hz, $J_{5,6}$ 3.5 Hz, $J_{5,6'}$ 2.5 Hz, H-5), 3.77 (dd, 1H, $J_{6,6'}$ 11.0 Hz, H-6), 3.67 (dt, overlapped, $J_{2,3eq} \sim 4.5$ Hz, H-2), 3.64 (H-6′), 2.35 (btd, 1H, $J_{3eq,3ax}$ 12.0 Hz, $J_{3eq,4} \sim 4.5$ Hz, H-3eq), 1.97 (q, 1H, $J_{3ax,4} \sim J_{2,3ax} \sim 11.5$ Hz, H-3ax).

2,3,6-Tri-O-*benzyl-4-deoxy-α-*D-xylo-*hexopyranosyl chloride (6)*

2,3,6-Tri-*O*-benzyl-4-deoxy-D-*xylo*-hexopyranose (16) (392.6 mg, 0.90 mmol) was converted into chloride **6** just prior to use as reported for the preparation of **3**. ^1H NMR ($CDCl_3$) δ: 7.40–7.20 (m, 15H, 3Ph), 6.09 (d, 1H, $J_{1,2}$ 3.5 Hz, H-1), 4.76 and 4.66 (ABq, 2H, $J_{A,B}$ 12.0 Hz, CH_2Ph), 4.74 and 4.70 (ABq, 2H, $J_{A,B}$ 12.0 Hz, CH_2Ph), 4.52 (s, 2H, CH_2Ph), 4.25 (m, 1H, H-5), 3.98 (ddd, 1H, $J_{2,3}$ 9.3 Hz, $J_{3,4eq}$ 5.0 Hz, $J_{3,4ax}$ 11.0 Hz, H-3), 3.61 (dd, 1H, H-2), 3.47 (d, 2H, $J_{6,5}$ 4.5 Hz, 2H-6), 2.10 (ddd, 1H, $J_{4eq,5}$ 2.0 Hz, $J_{4eq,4ax}$ 13.0 Hz, H-4eq), 1.61 (q, 1H, H-4ax).

Allyl 2,3,6-tri-O-*benzyl-4*-O-*methyl-α,β-*D-*glucopyranoside (8)*

Sodium hydride (528 mg, 22 mmol) was added to a stirred solution of allyl, 2,3,6-tri-*O*-benzyl-α,β-D-glucopyranoside (**7**, 5.51 g, 11.2 mmol) in *N,N*-dimethylformamide (70 mL) at ice-bath temperature. After 20 min, methyl iodide (1.37 mL, 22 mmol) was added and stirring continued for 4 h at room temperature. The excess of reagents was quenched by the addition of methanol (1 mL). The mixture was diluted with dichloromethane, poured into ice-water, and the aqueous layer was extracted with dichloromethane. The combined organic solutions were washed with water, dried, and evaporated to leave the title compound **8** (5.7 g, quant.) as an oil. The analytical sample was purified by column chromatography on silica gel (hexane–chloroform – ethyl acetate, 20:1:1). Evaporation of the first fraction provided the α-anomer, which was characterized; $[\alpha]_D$ +57 (*c* 1.0, chloroform). ^1H NMR ($CDCl_3$) δ: 7.4–7.2 (m, 15H, 3Ph), 5.93 (m, 1H, CH_2=C*H*), 5.31 and 5.20 (2m, 2H, CH_2=CH), 4.94 and 4.79 (ABq, 2H, $J_{A,B}$ 11.0 Hz, CH_2Ph), 4.81 (d, 1H, $J_{1,2}$ 3.5 Hz, H-1), 4.76 and 4.63 (ABq, 2H, $J_{A,B}$ 12.5 Hz, CH_2Ph), 4.63 and 4.50 (ABq, 2H, $J_{A,B}$ 12.5 Hz, CH_2Ph), 4.16 and 4.01 (2m, 2H, CH_2=CH-CH_2), 3.89 (t, 1H, $J_{2,3} \sim J_{3,4}$ 9.5 Hz, H-3), 3.72–3.61 (m, 3H, H-6, H-6′, H-5), 3.51 (dd, 1H, H-2), 3.46 (s, 3H, CH_3O), 3.34 (t, 1H, $J_{4,5}$ 9.0 Hz, H-4). Anal. calcd. for $C_{31}H_{36}O_6$: C 73.79, H 7.19; found: C 73.70, H 7.33.

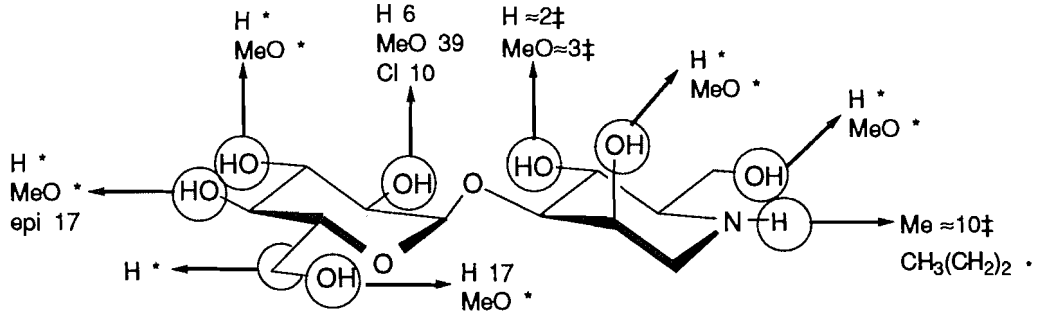


FIG. 1. The effect of chemical modification of **1** on the activity as inhibitor of *endo*-mannosidase in relative potencies (percentage of potency of **1**). ‡Approximate rel. potencies based on IC_{50} values as derived from inhibition data obtained for two inhibitor concentrations (3). *Denotes inactivity (rel. potency < 1).

2,3,6-Tri-O-benzyl-4-O-methyl-D-glucopyranose (9)

A mixture of crude **8** (5.7 g, 11.3 mmol), tris(triphenylphosphine)rhodium(I) chloride (805 mg, 0.87 mmol), 1,8-diazabicyclo[2.2.2]octane (342 mg, 3.05 mmol), and 95% ethanol–toluene–water (7:3:1, 77 mL) was boiled under reflux for 5 h. The solvent was evaporated and the remainder was taken up in acetone (70 mL) containing mercuric oxide (294 mg). A solution of mercuric chloride (14.4 g) in acetone–water (9:1, 50 mL) was added and the mixture stirred for 30 min. After solvent removal, the residue was taken up in dichloromethane, the solution washed with aqueous saturated potassium bromide and water, and the organic solution concentrated. Purification on a column of silica gel (hexane – ethyl acetate, 3:1) provided **9** (2.32 g, 44%) as a solid. The analytical sample was recrystallized from ether/hexane; mp 91–92°C; $[\alpha]_D$ +39 (*c* 0.3, chloroform). ^{1}H-nmr ($CDCl_3$, α:β = 1:0.6) δ: 7.42–7.24 (m, 15H, 3Ph), 5.20 (d, $J_{1\alpha,2}$ 3.5 Hz, H-1α), 5.03–3.97 (m, 3CH_2Ph, H-1β), 3.93 (ddd, $J_{4,5}$ 10.0 Hz, $J_{5,6}$ 2.5 Hz, $J_{5,6'}$ 3.5 Hz, H-5α), 3.85 (t, 1H, $J_{3,4}$ 9.5 Hz, H-3α), 3.72 (dd, $J_{6,6'}$ 11.0 Hz, $J_{6,5}$ 2.0 Hz, H-6β), 3.69 (dd, $J_{6,6'}$ 11.0 Hz, H-6α), 3.67 (H-6′β), 3.64 (dd, H-6′α), 3.54 (t, overlapped by H-2α, $J_{2,3} \sim J_{3,4}$ 9.0 Hz, H-3β), 3.53 (dd, H-2α), 3.47 (s, CH_3O-α), 3.465 (s, CH_3O-β), 3.43 (ddd, H-5β), 3.33 (dd, $J_{1\beta,2}$ 7.8 Hz, H-2β), 3.33 (t, H-4α), 3.27 (t, $J_{4,5}$ 9.5 Hz, H-4β). Anal. calcd. for $C_{28}H_{32}O_6$: C 72.39, H 6.94; found: C 71.78, H 6.92.

2,3,6-Tri-O-benzyl-4-O-methyl-α,β-D-glucopyranosyl chloride (10)

9 (550 mg, 1.18 mmol) was converted into **10** as described for the preparation of **3**. ^{1}H NMR ($CDCl_3$, α:β = 3:1) δ: 7.4–7.15 (m, 15H, 3Ph), 6.01 (d, 1H, $J_{1,2}$ 3.5 Hz, H-1), 3.95 (m, 1H, $J_{4,5}$ 10.0 Hz, H-5), 3.89 (t, $J_{2,3} \sim J_{3,4}$ 9.5 Hz, H-3), 3.46 (s, CH_3O), 3.41 (t, 1H, H-4).

2,3,4-Tri-O-benzyl-6-deoxy-α,β-D-glucopyranosyl chloride (12)

2,3,4-Tri-*O*-benzyl-6-deoxy-D-glucopyranose (17) (552.2 mg, 1.27 mmol) was converted into chloride **12** just prior to use as described for the preparation of **3**. ^{1}H NMR ($CDCl_3$, α:β = 2.2:1) δ: 7.45–7.2 (m, 15H, 3Ph), 5.98 (d, $J_{1\alpha,2}$ 4.0 Hz, H-1α), 5.19 (d, $J_{1\beta,2}$ 8.0 Hz, H-1β), 5.00–4.61 (m, 6H, 3CH_2Ph), 4.07 (m, H-5α), 4.00 (t, $J_{2,3} \sim J_{3,4}$ 9.0 Hz, H-3α), 3.69 (dd, H-2α), 3.60 (m, H-2β, H-3β), 3.49 (m, H-5β), 3.29 (t, $J_{4,5} \sim J_{3,4}$ 9.0 Hz, H-4β), 3.18 (t, $J_{4,5}$ 9.5 Hz, H-4α), 1.35 (d, $J_{5,6}$ 6.0 Hz, H_3-6β), 1.28 (d, $J_{5,6}$ 6.0 Hz, H_3-6α).

6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(3,4,6-tri-O-benzyl-2-O-methyl-α-D-glucopyranosyl)-D-mannitol (14) and -β-D-glucopyranosyl)-D-mannitol (15)

A solution of chloride **3** (0.82 mmol) in dichloromethane (5 mL) was added at −78°C to a stirred mixture of **2** (7) (210 mg, 0.40 mmol), silver trifluoromethanesulfonate (220 mg, 0.86 mmol), *sym*-collidine (43.9 μL, 0.33 mmol), and 4 Å molecular sieves (500 mg) in dichloromethane (1.5 mL). The temperature was gradually raised to 0°C and after 7 h the reaction was quenched by the addition of *sym*-collidine (75 μL, 0.57 mmol). The mixture was filtered through Celite, the organic solution washed with aqueous saturated sodium hydrogen carbonate and water, followed by evaporation. Column chromatography on silica gel (hexane – ethyl acetate, 5:1, 4:1) provided in the first main fraction the β-isomer **15** (90.1 mg, 23%); $[\alpha]_D$ −17.8 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.40–7.00 (m, 30H, 6Ph), 5.20–3.0 (complex m, 30H), 3.58 (s, 3H, CH_3O), 1.91 and 1.82 (2s, 3H, CH_3CO). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 170.62 (CH_3CO), 156.23 (OCO), 104.72 ($J_{C,H}$ 160.6 Hz, C-1′), 61.40 (61.21) (C-6), 60.59 (CH_3O), 52.97 (52.35) (C-5), (38.37) 37.83 (C-1), (20.74) 20.57 (CH_3CO). Evaporation of the second main fraction provided **14** (200 mg, 51%); $[\alpha]_D$ +34.1 (*c* 0.3, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.40–7.00 (m, 30H, 6Ph), 5.2–3.0 (complex m, 30H), 3.34 (s, 3H, CH_3O), 1.94 and 1.85 (2s, 3H, CH_3CO).

6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(3,4,6-tri-O-benzyl-2-chloro-2-deoxy-α-D-glucopyranosyl)-D-mannitol (16) and -β-D-glucopyranosyl)-D-mannitol (17)

A solution of chloride **4** (21) (0.60 mmol) in dichloromethane (2 mL) was added at −65°C to a stirred mixture of **2** (186 mg, 0.19 mmol), silver trifluoromethanesulfonate (165.9 mg, 0.65 mmol), *sym*-collidine (31 μL, 0.23 mmol), and 4 Å molecular sieves (800 mg) in dichloromethane (1.5 mL). The temperature was allowed to gradually rise to −20°C within 3 h. More *sym*-collidine (40 μL, 0.30 mmol) was added and, after 1 h at room temperature, the mixture was processed in the usual way. Column purification on silica gel (hexane – ethyl acetate, 5:1) provided, according to the ^{1}H NMR spectrum, an α:β mixture (231 mg, 67%) in the ratio 0.9:1. Separation of the anomers was achieved by repeated column chromatography on silica gel 60H (for thin-layer chromatography, E. Merck, Darmstadt) using hexane–chloroform – ethyl acetate, 14:14:1. Evaporation of the earlier fractions provided the α-anomer **16** (87.8 mg, 25%) as a syrup; $[\alpha]_D$ +45.6 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.0 (m, 30H, 6Ph), 5.3–2.95 (complex m, 27H), 1.93 and 1.86 (2s, 3H, CH_3CO). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 170.56 (CH_3CO), 156.09 (CO), 99.62 ($J_{C,H}$ 168.7 Hz, C-1′), 61.80 (61.66) (C-6), 60.21 (C-2), 52.51 (51.83) (C-5), (38.12), 37.49 (C-1), (20.74) 20.62 (CH_3CO). Anal. calcd. for $C_{57}H_{60}ClNO_{11}$: C 70.54, H 6.23, Cl 3.65, N 1.44; found: C 70.14, H 6.46, Cl 3.98, H 6.46. Continued development eluted the β-anomer **17** (75 mg, 22%) as a syrup; $[\alpha]_D$ −10.8 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.1 (m, 30H, 6Ph), 5.25–3.25 (complex m, 27H), 1.93 and 1.82 (2s, 3H, CH_3CO). ^{13}C nmr ($CDCl_3$, δ minor rotamer in parentheses) δ: 156.19 (CO), 101.88 (C-1′), 61.73 (C-2′), 61.50 (61.31) (C-6), 52.87 (52.16) (C-5), (38.08) 37.56 (C-1), (20.79) 20.61 (CH_3CO). Anal. calcd. for $C_{57}H_{60}ClNO_{11}$: C 70.54,

H 6.23, N 1.44; found: C 70.34, H 6.15, N 1.39. A second batch of α,β-mixture (42 mg) was separated by thin-layer chromatography (Whatman, silica gel 150A PLK5F, hexane–chloroform – ethyl acetate, 14:14:1, fourfold development). Elution of the higher zone provided the α-anomer **16** and elution of the lower zone the β-anomer **17** in slightly higher yields.

6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(2,4,6-tri-O-benzyl-3-deoxy-α-D-ribo-hexopyranosyl)-D-mannitol (18) and -β-D-ribo-hexopyranosyl)-D-mannitol (19)

A solution of chloride **5** (0.57 mmol) in dichloromethane (3 mL) was added at −78°C to a stirred mixture of **2** (166.8 mg, 0.32 mmol), silver trifluoromethanesulfonate (164 mg, 0.64 mmol), *sym*-collidine (30 μL, 0.23 mmol), and 4 Å molecular sieves (1 g) in dichloromethane (1.5 mL). Within 2 h the temperature was raised to −30°C and more *sym*-collidine (35 μL, 0.26 mmol) was added. After 1 h at room temperature, it was worked up in the usual way. Chromatographic separation on a column of silica gel (hexane – ethyl acetate, 5:1) provided in the earlier fractions the β-anomer **19** (121.2 mg, 40%) as a syrup; $[\alpha]_D$ +10 (*c* 0.6, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4 –7.05 (m, 30H, 6Ph), 5.24–3.15 (complex m, 26H), 2.46 (dt, 1H, $J_{3ax',3eq'}$ 13.0 Hz, $J_{2',3eq'} \sim J_{3eq',4'} \sim$ 4.5 Hz, H-3eq′), 1.92 and 1.82 (2s, 3H, CH_3CO), 1.51 (q, 1H, $J_{2',3ax'} \sim J_{3ax',4'} \sim$ 12 Hz, H-3ax′). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 170.8 (CH_3CO), 156.1 (CO), 106.8 ($J_{C,H}$ 161.7 Hz, C-1′), 61.44 (61.25) (C-6), 53.12 (52.48) (C-5), (38.50) 37.94 (C-1), 35.06 (C-3′), (20.81), 20.62 (CH_3CO). Anal. calcd. for $C_{57}H_{61}NO_{11}$: C 73.14, H 6.57, N 1.50; found: C 73.28, H 6.47, N 1.51. Continued elution provided the α-anomer **18** (140.0 mg, 47%) as a syrup; $[\alpha]_D$ +47 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.1 (m, 30H, 6Ph), 5.2–3.03 (complex m, 26H), 2.32 (m, 1H, $J_{3eq',3ax'}$ 13.0 Hz, H-3eq′), 1.85 and 1.74 (2s, 3H, CH_3CO), 1.75 (q, 1H, $J_{2',3ax'} \sim J_{4',3ax'} \sim$ 12 Hz, H-3ax′). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 170.5 (CH_3CO), 156.1 (CO), 96.48 ($J_{C,H}$ 166.2 Hz, C-1′), 61.96 (61.78) (C-6), 52.56 (51.94) (C-5), (38.10) 37.52 (C-1), 29.73 (C-3′), (20.68) 20.50 (CH_3CO). Anal. calcd. for $C_{57}H_{61}NO_{11}$: C 73.14, H 6.57, N 1.50; found: C 73.20, H 6.38, N 1.17.

6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(2,3,6-tri-O-benzyl-4-deoxy-α-D-xylo-hexopyranosyl)-D-mannitol (20) and -β-D-xylo-hexopyranosyl)-D-mannitol (21)

A solution of chloride **6** (0.90 mmol) in dichloromethane (4 mL) was added at −65°C to a stirred mixture of **2** (234.7 mg, 0.45 mmol), silver trifluoromethanesulfonate (256 mg, 1.0 mmol), *sym*-collidine (47.5 μL, 0.36 mmol), and 4 Å molecular sieves (1 g) in dichloromethane (2 mL). Within 2 h, the temperature was raised to −55°C. More *sym*-collidine (60 μL, 0.48 mmol) was added. After 1 h at room temperature, the mixture was worked up in the usual way, followed by column chromatography on silica gel (hexane – ethyl acetate, 5:1) to provide an anomeric mixture of **20** and **21**. A second chromatographic purification (carbon tetrachloride – ethyl acetate, 8:1) provided the α-anomer **20** (230.7 mg, 55%) as a syrup; $[\alpha]_D$ +54 (*c* 0.4, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.1 (m, 30H, 6Ph), 5.2–3.0 (complex m, 26H), 1.95 (unresolv. ddd, 1H, $J_{4eq',4ax'}$ 13.0 Hz, H-4eq′), 1.82 and 1.73 (2s, 3H, CH_3CO), 1.55 (q, 1H, H-4ax′). Anal. calcd. for $C_{57}H_{61}NO_{11}$: C 73.14, H 6.57, N 1.50; found: C 73.21, H 6.65, N 1.54. Continued development of the column eluted the β-anomer **21** (153.6 mg, 36%) as a syrup; $[\alpha]_D$ −19.8 (*c* 0.4, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.05 (m, 30H, 6Ph), 5.2–3.1 (complex m, 26H), 2.05 (unresolv. ddd, 1H $J_{4eq',4ax'}$ 12.5 Hz, H-4eq′), 1.89 and 1.80 (2s, 3H, CH_3CO), 1.38 (q, 1H, $J_{3',4ax'} \sim J_{4ax',5'} \sim$ 12 Hz, H-4ax′); ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 105.14 ($J_{C,H}$ 161.1 Hz, C-1′), 61.41 (61.22) (C-6), 53.04 (52.35) (C-5), (38.44) 37.89 (C-1), 33.76 (C-4′), 20.61 (CH_3CO). Anal. calcd. for $C_{57}H_{61}NO_{11}$: C 73.14, H 6.57, N 1.50; found: C 72.94, H 6.64, N 1.51.

6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(2,3,6-tri-O-benzyl-4-O-methyl-α-D-glucopyranosyl)-D-mannitol (22) and -β-D-glucopyranosyl)-D-mannitol (23)

A solution of chloride **10** (1.18 mmol) in dichloromethane (7 mL) was added to a stirred mixture of **2** (253 mg, 0.49 mmol), silver trifluoromethanesulfonate (380 mg, 1.48 mmol), *sym*-collidine (51 μL, 0.39 mmol), and 4 Å molecular sieves (500 mg) in dichloromethane (2.0 mL) at 0°C. Stirring was continued for 2.5 h. Then more *sym*-collidine (150 μL, 1.14 mmol) was added, followed by conventional work-up. Chromatographic isolation of the product on a column of silica gel (hexane – ethyl acetate 5:1, 4:1, 3:1) provided a 2.5:1 mixture of **22** and **23** (310 mg, 66%). This mixture was not further characterized, but was deacetylated.

6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(2,3,4,6-tetra-O-benzyl-α-D-galactopyranosyl)-D-mannitol (24) and -β-D-galactopyranosyl)-D-mannitol (25)

A solution of chloride **11** (9) (0.94 mmol) in dichloromethane (6.5 mL) was added to a stirred mixture of **2** (265 mg, 0.51 mmol), silver trifluoromethanesulfonate (310 mg, 1.20 mmol), *sym*-collidine (55 μL, 0.42 mmol), and 4 Å molecular sieves (500 mg) in dichloromethane (1.5 mL) at −78°C. The temperature was gradually raised to 0°C and, after 7 h, *sym*-collidine (180 μL, 1.36 mmol) and methanol (200 μL) were added. The mixture was processed in the usual way and the product mixture purified on a column of silica gel (hexane – ethyl acetate, 5:1). Evaporation of the first main fraction provided **24** (357.9 mg, 67%) as a tough syrup; $[\alpha]_D$ +38.5 (*c* 1.0, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.0 (m, 35H, 7Ph), 5.2–3.0 (complex m, 29H), 1.82 and 1.73 (2s, 3H, CH_3CO). Anal. calcd. for $C_{64}H_{67}NO_{12}$: C 73.76, H 6.48, N 1.34; found: C 73.60, H 6.49, N 1.41. Continued elution of the column furnished the β-anomer **25** (38.8 mg, 7.3%) as a tough syrup; $[\alpha]_D$ −18.9 (*c* 0.3, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.0 (m, 35H, 7Ph), 5.2–3.1 (complex m, 29H), 1.82 and 1.73 (2s, 3H, CH_3CO). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 170.4 (CH_3CO), 156.22 (CO), 104.83 ($J_{C,H}$ 160.7 Hz, C-1′), 61.22 (C-6), 52.85 (52.21) (C-5), (37.82) 37.35 (C-1), 20.62 (CH_3CO).

6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(2,3,4-tri-O-benzyl-6-deoxy-α-D-glucopyranosyl)-D-mannitol (26) and -β-D-glucopyranosyl)-D-mannitol (27)

A solution of chloride **12** (1.25 mmol) in dichloromethane (2 mL) was added at −40°C to a stirred mixture of alcohol **2** (287 mg, 0.55 mmol), silver trifluoromethanesulfonate (360 mg, 1.40 mmol), *sym*-collidine (58 μL, 0.44 mmol), and 4 Å molecular sieves (300 mg) in dichloromethane (1 mL). After 1 h at this temperature, the temperature was allowed to reach 0°C within 3 h. The reaction was quenched by the addition of *sym*-collidine (150 μL, 1.14 mmol) followed by usual work-up. Column chromatography on silica gel (hexane – ethyl acetate, 6:1) provided the anomeric mixture of **26** and **27** (464 mg, 90%), which was not further characterized, but deacetylated.

6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(2,3,4-tri-O-benzyl-α-D-xylopyranosyl)-D-mannitol (28) and -β-D-xylopyranosyl)-D-mannitol (29)

Chloride **13** (10) was prepared from 2,3,4-tri-*O*-benzyl-D-xylopyranose (18) as described for **3**. A solution of **13** (0.72 mmol) in dichloromethane (4.5 mL) was transferred into a stirred mixture of **2** (263 mg, 0.51 mmol), silver trifluoromethanesulfonate (334 mg, 1.3 mmol), *sym*-collidine (55 μL, 0.42 mmol), and 4 Å molecular sieves (0.5 g) in dichloromethane (1 mL) at −78°C. The mixture was allowed to reach 0°C within 4.5 h and more *sym*-col-

lidine (100 μL, 0.76 mmol) was added. It was processed as described for the preparation of **14**, followed by chromatography on a column of silica gel (hexane – ethyl acetate, 5:1, 4:1), which provided a 2:1 mixture of the anomers **28** and **29** (432.5 mg, 93%) according to the ^{1}H NMR spectrum. This mixture was not further characterized but deacetylated.

*2,4-Di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(3,4,6-tri-*O*-benzyl-2-*O*-methyl-α-D-glucopyranosyl)-D-mannitol (**30**)*

A solution of **14** (193 mg, 0.20 mmol) in methanol (5 mL), triethylamine (2 mL), and water (1 mL) was kept at room temperature for 4 days. It was evaporated to dryness and the resultant material was applied to a column of silica gel (hexane–dichloromethane – ethyl acetate, 5:1:1, 4:1:1) to provide the title compound **30** (144 mg, 78%) as a tough syrup; $[\alpha]_D$ +44.2 (*c* 0.2, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of rotamers) δ: 7.4–7.0 (m, 30H, 6Ph), 5.20–2.90 (complex m, 31H), 3.42 (s, 3H, CH_3O). Anal. calcd. for $C_{56}H_{61}NO_{11}$: C 72.79, H 6.65, N 1.52; found: C 73.05, H 6.68, N 1.45.

*2,4-Di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(3,4,6-tri-*O*-benzyl-2-chloro-2-deoxy-α-D-glucopyranosyl)-D-mannitol (**31**)*

A mixture of **16** (60 mg, 0.065 mmol), methanol (1.43 mL), triethylamine (572 μL), and water (286 μL) was kept at room temperature for 3 days. Evaporation, followed by column chromatography on silica gel (hexane – ethyl acetate, 2:1), provided syrupy **31** (54.6 mg, 95%); $[\alpha]_D$ +69 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of rotamers) δ: 7.4–7.0 (m, 30H, 6Ph), 5.15–2.9 (complex m, 28H). ^{13}C NMR ($CDCl_3$) δ: 99.25 (C-1′), 60.44 (C-6, C-2′), 55.59 (C-5), 38.0 (C-1). Anal. calcd. for $C_{55}H_{58}ClNO_{10}$: C 71.15, H 6.30, N 1.51; found: C 70.95, H 6.51, N 1.36.

*2,4-Di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(2,4,6-tri-*O*-benzyl-3-deoxy-α-D-*ribo*-hexopyranosyl)-D-mannitol (**32**)*

A mixture of **18** (121 mg, 0.13 mmol), methanol (3.2 mL), triethylamine (1.28 mL), and water (0.64 mL) was stirred at room temperature for 5 days. Evaporation and column chromatography on silica gel (hexane – ethyl acetate, 3:1) provided **32** (88.5 mg, 77%) as a tough syrup; $[\alpha]_D$ +36 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of rotamers) δ: 7.43–7.05 (m, 30H, 6Ph), 5.2–2.9 (complex m, 27H), 2.30 (m, 1H, H-3eq′), 1.71 (q, 1H, $J_{3ax',3eq'} \sim J_{2',3ax'} \sim J_{3ax',4'} \sim 12$ Hz, H-3ax′). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 156.10 (CO), 97.52 ($J_{C,H}$ 167.9 Hz, C-1′), 58.93 (C-6), 53.70 (53.26) (C-5), (39.19) 38.60 (C-1), 29.74 (C-3′). Anal. calcd. for $C_{55}H_{59}NO_{10}$: C 73.89, H 6.65, N 1.57; found: C 73.92, H 6.75, N 1.55.

*2,4-Di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(2,3,6-tri-*O*-benzyl-4-deoxy-α-D-*xylo*-hexopyranosyl)-D-mannitol (**33**)*

A solution of **20** (230.6 mg, 0.25 mmol) in methanol (6.1 mL), triethylamine (2.43 mL), and water (1.2 mL) was kept at room temperature for 6 days. The material resulting on evaporation was applied to a column of silica gel (hexane – ethyl acetate, 3:1) to provide syrupy **33** (172 mg, 78%); $[\alpha]_D$ +44 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of rotamers) δ: 7.4–7.05 (m, 30H, 6Ph), 5.2–2.9 (complex m, 27H), 2.01 (ddd, 1H, $J_{4ax',4eq'}$ 13.0 Hz, $J_{3',4eq'}$ 5.0 Hz, $J_{4eq',5'}$ 2.0 Hz, H-4eq′), 1.62 (q, 1H, H-4ax′). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 156.06 (CO), 99.71 ($J_{C,H}$ 166.9 Hz, C-1′), 58.06 (C-6), 53.90 (53.46) (C-5), (39.38) 38.75 (C-1), 33.09 (C-4′). Anal. calcd. for $C_{55}H_{59}NO_{10}$: C 73.89, H 6.65, N 1.57; found: C 73.70, H 6.65, N 1.62.

*2,4-Di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(2,3,6-tri-*O*-benzyl-4-*O*-methyl-α-D-glucopyranosyl)-D-mannitol (**34**) and -β-D-glucopyranosyl)-D-mannitol (**35**)*

An anomeric mixture of compounds **22** and **23** (295 mg, 0.31 mmol) in methanol (7.5 mL), triethylamine (3 mL), and water (1.5 mL) was kept for 7 days at room temperature. It was evaporated and the resultant material applied to a column of silica gel (hexane–dichloromethane – ethyl acetate, 4:1:1) to provide in the first fraction starting material **22** and **23** (58 mg, 20%). Evaporation of the second fraction gave α-isomer **34** (145.1 mg, 51%); $[\alpha]_D$ −3 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers) δ: 7.45–7.10 (m, 30H, 6Ph), 5.2–2.84 (complex m, 31H), 3.37 (s, 3H CH_3O). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 156.10 (CO), 99.28 ($J_{C,H}$ 168.5 Hz, C-1′), 60.42 (CH_3O), 59.12 (C-6), 53.87 (53.39) (C-5), (39.22) 38.65 (C-1). Anal. calcd. for $C_{56}H_{61}NO_{11}$: C 72.79, H 6.65, N 1.52; found: C 72.59, H 6.70, N 1.47. Continued elution of the column provided the β-isomer **35** (58.8 mg, 21%); $[\alpha]_D$ −6 (*c* 0.8, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers) δ: 7.4–7.0 (m, 30H, 6Ph), 5.2–3.0 (complex m, 31H), 3.40 (s, CH_3O). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 156.0 (CO), 105.61 ($J_{C,H}$ 158.0 Hz, C-1′), 60.64 (CH_3O), 59.71 (C-6), 54.85 (54.53) (C-5), (39.41) 38.98 (C-1).

*2,4-Di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-galactopyranosyl)-D-mannitol (**36**)*

A solution of acetate **24** (225 mg, 0.22 mmol) in methanol (5 mL), triethylamine (2 mL), and water (1 mL) was kept for 6 days at room temperature. It was evaporated and the resultant material applied to a column of silica gel (hexane–dichloromethane – ethyl acetate, 5:1:1). Evaporation of the first main fraction provided starting material **24** (51.8 mg, 23%). Continued elution gave **36** (138.4 mg, 64%) as a tough syrup; $[\alpha]_D$ +22.7 ^{1}H NMR ($CDCl_3$, broad, mixture of rotamers) δ: 7.4–7.0 (m, 35H, 7Ph), 5.15–2.90 (complex m, 29H). Anal. calcd. for $C_{62}H_{65}NO_{11}$: C 74.45, H 6.55, N 1.40; found: C 74.50, H 6.57, N 1.35.

*2,4-Di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(2,3,4-tri-*O*-benzyl-6-deoxy-α-D-glucopyranosyl)-D-mannitol (**37**) and -β-D-glucopyranosyl)-D-mannitol (**38**)*

An anomeric mixture of compounds **26** and **27** (450 mg, 0.48 mmol), triethylamine (4.7 mL), water (2.3 mL), and methanol (11.8 mL) was kept at room temperature for 5 days. It was evaporated and the resultant material applied to a column of silica gel (hexane – ethyl acetate, 3:1) to provide in the first main fraction α-anomer **37** (197 mg, 46%) as a syrup; $[\alpha]_D$ +33.4 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers) δ: 7.50–7.10 (m, 30H, 6Ph), 5.2–3.0 (complex m, 26H), 0.87 (d, 3H, $J_{5',6'}$ 6.2 Hz, H_3-6′). ^{13}C NMR (δ minor rotamer in parentheses) δ: 156.13 (CO), 98.95 ($J_{C,H}$ 171.8 Hz, C-1′), 59.25 (C-6), 53.94 (53.47) (C-5), (39.35) 38.76 (C-1), 17.45 (C-6′). Anal. calcd. for $C_{55}H_{59}NO_{10}$: C 73.89, H 6.65, N 1.57; found: C 73.88, H 6.39, N 1.57. Continued development eluted the syrupy β-isomer **38** (122.1 mg, 28%); $[\alpha]_D$ −16.5 (*c* 0.6, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers) δ: 7.45–7.05 (m, 30H, 6Ph), 5.2–2.8 (m, 26H), 1.13 (d, 3H, $J_{5',6'}$ 6.0 Hz, H_3-6′). ^{13}C NMR (δ minor rotamer in parentheses) δ: 156.28 (CO), 105.47 ($J_{C,H}$ 162.3 Hz, C-1′), (58.67) 58.44 (C-6), 54.51 (54.29) (C-5), 39.32 (38.84) (C-1), 17.72 (C-6′).

*2,4-Di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(2,3,4-tri-*O*-benzyl-α-D-xylopyranosyl)-D-mannitol (**39**) and -β-D-xylopyranosyl)-D-mannitol (**40**)*

A solution of the anomeric mixture of **28** and **29** (433 mg, 0.47 mmol) in methanol (10 mL), triethylamine (4 mL), and water (2 mL) was kept at room temperature for 7 days. It was evaporated and the product mixture separated on a column of silica gel (hexane–dichloromethane – ethyl acetate, 5:1:1 to 2:1:1) to provide in the first main fraction the α-anomer **39** (193 mg, 47%) as a tough syrup; $[\alpha]_D$ +22.6 (*c* 0.2, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers) δ: 7.40–7.10 (m, 30H, 6Ph), 5.20–3.0 (complex m, 27H). Anal. calcd. for $C_{54}H_{57}NO_{10}$: C 73.70, H 6.53, N 1.59; found: C 73.85, H 6.44, N 1.59. Continued development of the column eluted the β-anomer **40** (100.2 mg, 24%); $[\alpha]_D$ −22.1 (*c* 0.3, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture

of two rotamers) δ: 7.40–7.10 (m, 30H, 6Ph), 5.10–2.60 (complex m, 27H). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 156.25 (CO), 106.46 ($J_{C,H}$ 161.5 Hz, C-1′) (58.87) 58.55 (C-6), 54.69 (C-5), (39.19) 38.64 (C-1). Anal. calcd. for $C_{54}H_{57}NO_{10}$: C 73.70, H 6.53, N 1.59; found: C 73.49, H 6.50, N 1.54.

*1,5-Dideoxy-1,5-imino-3-*O*-(2-*O*-methyl-α-D-glucopyranosyl)-D-mannitol-hydrochloride (**41**)*

A mixture of **30** (131 mg, 0.14 mmol), 5% palladium-on-carbon (130 mg), and methanolic 0.12 N hydrochloric acid (1.3 mL, 0.156 mmol) in methanol (7 mL) was hydrogenated in a hydrogen stream for 2 h. Removal of the catalyst by filtration, evaporation, and gel filtration (Sephadex LH 20, ethanol–water, 1:1) provided **41** (50.9 mg, 96%) as a white solid after freeze-drying its aqueous solution; $[\alpha]_D$ +83.3 (*c* 0.2, water). The ^1H and ^{13}C NMR data are reported in Tables 1 and 2.

*3-*O*-(2-Chloro-2-deoxy-α-D-glucopyranosyl)-1,5-dideoxy-1,5-imino-D-mannitol-hydrochloride (**42**)*

A mixture of **31** (32 mg, 0.034 mmol), 5% palladium-on-carbon (30 mg), and methanolic 0.12 N hydrochloric acid (0.3 mL, 0.036 mmol) in methanol (4.5 mL) was hydrogenated in a hydrogen stream for 4 h. Removal of the catalyst by filtration, evaporation, and gel filtration (Sephadex LH 20, ethanol–water, 1:1) provided **42** (10.9 mg, 83%) as a white solid after lyophilization of an aqueous solution; $[\alpha]_D$ +48.5 (*c* 0.4, water). The ^1H and ^{13}C NMR data are reported in Tables 1 and 2.

*1,5-Dideoxy-3-*O*-(3-deoxy-α-D-*ribo*-hexopyranosyl)-1,5-imino-D-mannitol-hydrochloride (**43**)*

Compound **32** (93 mg, 0.104 mmol), 5% palladium-on-carbon (90 mg), and 0.12 N methanolic hydrochloric acid (1 mL, 0.12 mmol) in methanol (12 mL) were hydrogenated in the hydrogen stream for 1.5 h. Usual processing followed by gel filtration (Sephadex LH 20, ethanol–water, 1:1) provided **43** (32.4 mg, 90%) as a white solid; $[\alpha]_D$ +70.3 (*c* 0.3, water). The ^1H and ^{13}C NMR data are reported in Tables 1 and 2.

*1,5-Dideoxy-3-*O*-(4-deoxy-α-D-*xylo*-hexopyranosyl)-1,5-imino-D-mannitol-hydrochloride (**44**)*

A mixture of compound **33** (163.6 mg, 0.183 mmol), 5% palladium-on-carbon (150 mg), and methanolic 0.12 N hydrochloric acid (1.7 mL, 0.20 mmol) in methanol (18 mL) was hydrogenated in the hydrogen stream for 2.5 h. The catalyst was removed by filtration, followed by evaporation. Gel filtration of the resultant material on a column of Sephadex LH 20 (ethanol–water, 1:1) afforded the title compound **44** (45.6 mg, 72%) as a slightly yellowish powder; $[\alpha]_D$ +77.3 (*c* 0.3, water). The ^1H and ^{13}C NMR data are reported in Tables 1 and 2.

*1,5-Dideoxy-1,5-imino-3-*O*-(4-*O*-methyl-α-D-glucopyranosyl)-D-mannitol-hydrochloride (**45**)*

A mixture of **34** (114.3 mg, 0.12 mmol), 5% palladium-on-carbon (110 mg), and methanolic 0.12 N hydrochloric acid (1.13 mL, 0.14 mmol) in methanol (6 mL) was hydrogenated in the hydrogen stream for 2 h. The catalyst was removed by filtration and the solvent evaporated. The resultant material was passed through a column of Iatrobeads® (chloroform–methanol–water, 65:35:8) followed by gel filtration (Sephadex LH 20, ethanol–water, 1:1) to provide **45** (46.5 mg, 100%) as a solid; $[\alpha]_D$ +81 (*c* 0.4, water). The ^1H and ^{13}C NMR data are reported in Tables 1 and 2.

*1,5-Dideoxy-3-*O*-(α-D-galactopyranosyl)-1,5-imino-D-mannitol-hydrochloride (**46**)*

A mixture of **36** (128 mg, 0.128 mmol), 5% palladium-on-carbon (128 mg), and methanolic 0.12 N hydrochloric acid (1.1 mL, 0.13 mmol) in methanol (7 mL) was hydrogenated in a hydrogen stream for 3 h. The catalyst was removed by filtration, followed by evaporation. The resultant material was subjected to gel filtration (Sephadex LH 20, ethanol–water, 1:1) to provide, after freeze-drying an aqueous solution, title compound **46** (42.5 mg, 92%); $[\alpha]_D$ +91.6 (*c* 0.2, water). The ^1H and ^{13}C NMR data are reported in Tables 1 and 2.

*1,5-Dideoxy-3-*O*-(6-deoxy-α-D-glucopyranosyl)-1,5-imino-D-mannitol-hydrochloride (**47**)*

A mixture of **37** (183.1 mg, 0.21 mmol), 5% palladium-on-carbon (140 mg), and methanolic 0.12 N hydrochloric acid (1.87 mL, 0.224 mmol) in methanol (10 mL) was hydrogenated in the hydrogen stream for 2 h. The catalyst was removed by filtration and the solvent evaporated. The resultant material was passed through a column of Iatrobeads® (chloroform–methanol–water, 65:35:8) followed by gel filtration (Sephadex LH 20, ethanol–water, 1:1) to provide **47** (60.5 mg, 85%) as a white solid, $[\alpha]_D$ +64.4 (*c* 0.5, water). The ^1H and ^{13}C NMR data are reported in Tables 1 and 2.

*1,5-Dideoxy-1,5-imino-3-*O*-(α-D-xylopyranosyl)-D-mannitol-hydrochloride (**48**)*

A mixture of compound **39** (170 mg, 0.19 mmol), 5% palladium-on-carbon (170 mg), methanolic 0.12 N hydrochloric acid (1.6 mL, 0.19 mmol), and methanol (10 mL) was hydrogenated in a hydrogen stream for 3 h. The catalyst was removed by filtration, followed by evaporation and codistillation with water. The product was passed through a column of Sephadex LH 20 (ethanol–water, 1:1) to provide, after freeze-drying an aqueous solution, the title compound **48** (59.6 mg, 93%) as a white solid, $[\alpha]_D$ +63 (*c* 0.3, water). The ^1H and ^{13}C NMR data are reported in Tables 1 and 2.

*3,4,6-Tri-*O*-benzyl-2-deoxy-α-D-*arabino*-hexopyranosyl chloride (**49**) (22)*

49 was prepared just prior to use according to ref. 22 and characterized by ^1H NMR spectroscopy. ^1H NMR ($CDCl_3$) δ: 7.4–7.1 (m, 15H, 3Ph), 6.30 (d, 1H, $J_{1,2ax}$ 3.3 Hz, $J_{1,2eq}$ ~ 1 Hz, H-1), 4.16 (ddd, 1H, $J_{2ax,3}$ 11.0 Hz, $J_{2eq,3}$ 4.5 Hz, $J_{3,4}$ 9.5 Hz, H-3), 4.06 (ddd, 1H, $J_{4,5}$ 9.5 Hz, $J_{5,6}$ 3.7 Hz, $J_{5,6'}$ 1.8 Hz, H-5), 3.81 (dd, 1H, $J_{6,6'}$ 11.0 Hz, H-6), 3.71 (t, 1H, H-4), 3.65 (dd, 1H, H-6′), 2.51 (ddd, 1H, $J_{2eq,2ax}$ 13.5 Hz, H-2eq), 3.73 (ddd, 1H, H-2ax).

*2,4,6-Tri-*O*-benzyl-3-*O*-methyl-α,β-D-glucopyranosyl chloride (**50**)*

2,4,6-Tri-*O*-benzyl-3-*O*-methyl-D-glucopyranose (15) (550 mg, 1.18 mmol) was converted into chloride **50** as described for the preparation of **3**. ^1H NMR ($CDCl_3$, α:β = 2:1) δ: 7.4–7.1 (m, 15H, 3Ph), 5.99 (d, $J_{1\alpha,2}$ 3.5 Hz, H-1α), 5.13 (d, $J_{1\beta,2}$ 8.5 Hz, H-1β), 4.00 (m, $J_{4,5}$ 10.0 Hz, H-5α) 3.64 (s, CH_3O-α), 3.62 (s, CH_3O-β).

*6-*O*-Acetyl-2,3,4-tri-*O*-benzyl-α,β-D-glucopyranosyl chloride (**51**) (11)*

6-*O*-Acetyl-2,3,4-tri-*O*-benzyl-D-glucopyranose (19) (68 mg, 0.138 mmol) was converted into **51** as described for the preparation of chloride **3**. ^1H NMR ($CDCl_3$, α:β = 1.5:1) δ: 7.4–7.1 (m, 15H, 3Ph), 5.99 (d, $J_{1\alpha,2}$ 3.5 Hz, H-1α), 5.20 (d,$J_{1\beta,2}$ 8.0 Hz, H-1β), 4.33 (dd, $J_{6,6'}$ 12.0 Hz, $J_{5,6}$ ~ 1 Hz, H-6β), 4.25 (d, $J_{6,5}$ 3.2 Hz, 2H-6α), 4.18 (dd, $J_{5,6'}$ 4.5 Hz, H-6′β), 4.12 (dt, $J_{4,5}$ 10.0 Hz, H-5α), 4.04 (t, 1H, $J_{3,4}$ ~ $J_{2,3}$ 9.2 Hz, H-3α), 3.68 (dd, H-2α), 3.51 (dd, H-4α), 2.04, 2.00 (2s, CH_3CO-β, CH_3CO-α).

*6-*O*-Acetyl-2,4-di-*O*-benzyl-*N*-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-*O*-(3,4,6-tri-*O*-benzyl-2-deoxy-α-D-*arabino*-hexopyranosyl)-D-mannitol (**52**)*

A solution of chloride **49** (0.73 mmol) in dichloromethane (1 mL) was added at −65°C to a stirred mixture of **2** (145 mg, 0.28 mmol), silver trifluoromethanesulfonate (188.3 mg, 0.73 mmol), *sym*-collidine (34.9 μL, 0.26 mmol), and 4 Å molecular sieves (400 mg) in dichloromethane (2 mL). After 1.5 h at this temperature, triethylamine (80 μL, 0.61 mmol) was added and stirring continued for 30 min at room temperature. Usual work-up was followed by chromatography on a column of silica gel to afford **52** (189.3 mg, 73%) as a syrup; $[\alpha]_D$ +26.2 (*c* 0.5, chloroform). ^1H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.05 (m,

30H, 6Ph), 5.2–3.0 (complex m, 26H), 2.04 (ddd, 1H, $J_{2eq',2ax'}$ 13.5 Hz, $J_{2eq',3'}$ 4.5 Hz, H-2eq'), 1.94 and 1.85 (2s, 3H, CH_3CO), 1.67 (ddd, 1H, $J_{2ax',1'}$ 3.3 Hz, $J_{2ax',3'}$ 12.0 Hz, H-2ax'). Anal. calcd. for $C_{57}H_{61}NO_{11}$: C 73.14, H 6.57, N 1.50; found: C 73.32, H 7.01, N 1.59.

*6-O-Acetyl-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-3-O-(2,4,6-tri-O-benzyl-3-O-methyl-α-D-glucopyranosyl)-D-mannitol (**53**) and -β-D-glucopyranosyl)-D-mannitol (**54**)*

A solution of chloride **50** (1.18 mmol) in dichloromethane was added to a stirred mixture of **2** (276 mg, 0.53 mmol), silver trifluoromethanesulfonate (335 mg, 1.30 mmol), *sym*-collidine (56 μL, 0.42 mmol), and 4 Å molecular sieves (500 mg) in dichloromethane (2 mL) at −40°C. The mixture was gradually allowed to reach 0°C and, after 10 h, more *sym*-collidine (100 μL, 0.76 mmol) was added. Conventional work-up was followed by chromatography of the crude product on a column of silica gel (hexane–ethyl acetate, 5:1, 4:1, 3:1) to provide in the first main fraction the β-isomer **54** (111.1 mg, 22%); $[\alpha]_D$ −4.7 (*c* 0.7, chloroform). ^{1}H NMR (broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.0 (m, 30H, 6Ph), 5.2–3.1 (complex m, 30H), 3.58 (s, 3H, CH_3O), 1.88 and 1.79 (2s, 3H, CH_3CO). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 156.0 (CO), 104.86 ($J_{C,H}$ 160.9 Hz, C-1'), 61.35, 61.21 (CH_3O, C-6), 53.07 (52.44) (C-5), 38.40 (37.85) (C-1), 20.58 (CH_3CO). Continued elution gave **53** (277.4 mg, 54%); $[\alpha]_D$ +48.4 (*c* 0.7, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers in the ratio 3:2) δ: 7.4–7.0 (m, 30H, 6Ph), 5.2–3.0 (complex m, 30H), 3.65 (s, 3H, CH_3O), 1.82 and 1.74 (2s, 3H, CH_3CO). Anal. calcd. for $C_{58}H_{63}NO_{12}$: C 72.11, H 6.57, N 1.45; found: C 72.49, H 6.38, N 1.42.

*6-O-Acetyl-3-O-(6-O-acetyl-2,3,4-tri-O-benzyl-α- and -β-D-glucopyranosyl)-2,4-di-O-benzyl-N-benzyloxycarbonyl-1,5-dideoxy-1,5-imino-D-mannitol (**55** and **56**)*

A solution of chloride **51** (0.13 mmol) in dichloromethane (1.5 mL) was transferred into a stirred mixture of **2** (34 mg, 0.065 mmol), silver trifluoromethanesulfonate (43 mg, 0.17 mmol), *sym*-collidine (7.1 μL, 0.054 mmol), and 4 Å molecular sieves (100 mg) in dichloromethane (0.3 mL) at 0°C. After 1 h at this temperature, more *sym*-collidine (10 μL, 0.076 mmol) was added and the reaction mixture was processed as usual. The resultant material was applied to a column of silica gel (hexane – ethyl acetate, 5:1, 4:1) to provide in the first main fraction the β-anomer **56** (14.0 mg, 22%) as a tough syrup; $[\alpha]_D$ −18.9 (*c* 0.3, chloroform). ^{1}H NMR ($CDCl_3$, broad, mixture of two rotamers) δ: 7.4–7.0 (m, 30H, 6Ph). 5.2–3.1 (complex m, 27H), 1.98 (s, 3H, CH_3CO-6') 1.92 and 1.82 (2s, 3H, CH_3CO-6). ^{13}C NMR ($CDCl_3$, δ minor rotamer in parentheses) δ: 170.57 (CH_3CO), 156.40 (OCO), 104.91 ($J_{C,H}$ 161.5 Hz, C-1'), 62.51 (C-6'), 61.20 (61.04) (C-6), 52.90 (52.20) (C-5), 38.37 (37.84) (C-1), 20.77, 20.57 (CH_3CO). Continued elution of the column afforded **55** (40 mg, 62%) as a tough syrup; $[\alpha]_D$ +19.5 (*c* 0.2, chloroform). ^{1}H NMR (broad, mixture of two rotamers) δ: 7.4–7.1 (m, 30H, 6Ph), 5.2–3.0 (complex m, 27H), 1.91 (s, 3H, CH_3CO-6'), 1.83 and 1.75 (2s, 3H, CH_3CO-6). Anal. calcd. for $C_{59}H_{63}NO_{13}$: C 71.28, H 6.39, N 1.41; found: C 71.44, H 6.42, N 1.44.

*2,4-Di-O-benzyl-N,6-O-carbonyl-1,5-dideoxy-1,5-imino-3-O-(3,4,6-tri-O-benzyl-2-deoxy-α-D-*arabino*-hexopyranosyl)-D-mannitol (**57**)*

A solution of **52** (156.6 mg, 0.167 mmol) in methanolic 0.5 N sodium methoxide (1 mL) was kept at room temperature for 3.5 h. The remainder on evaporation was taken up in dichloromethane and the solution washed with water until neutral, followed by evaporation. Column chromatography provided the title compound **57** (108.4 mg, 83%) as a syrup; $[\alpha]_D$ +15 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$) δ: 7.4–7.1 (m, 25H, 5Ph), 5.22 (bd, 1H, $J_{1',2ax'}$ 3.0 Hz, $J_{1',2eq'} \sim 1$ Hz, H-1'), 4.97–4.37 (m, 10H, 5CH_2Ph), 4.28 (t, 1H, $J_{5,6A} \sim J_{6A,6B} \sim 8.5$ Hz, H-6A), 4.06 (dd, 1H, $J_{1A,1B}$ 14.0 Hz, $J_{1A,2}$ 2.0 Hz, H-1A), 4.01 (m, 1H, H-3'), 3.88 (overlapped, H-2), 2.71 (bd, 1H, H-1B), 2.25 (bdd, 1H, $J_{2eq',2ax'}$ 13.0 Hz, $J_{2eq',3'}$ 11.0 Hz, H-2eq') 1.72 (ddd, 1H, $J_{2ax',3'}$ 11.0 Hz, H-2ax'). ^{13}C NMR ($CDCl_3$) δ: 157.7 (CO), 100.03 ($J_{C,H}$ 169.4 Hz, C-1'), 65.58 (C-6), 57.53 (C-5), 41.12 (C-1), 35.77 (C-2'). Anal. calcd. for $C_{48}H_{51}NO_9$: C 73.35, H 6.54, N 1.78; found: C 73.46, H 6.88, N 1.64.

*2,4-Di-O-benzyl-N-6-O-carbonyl-1,5-dideoxy-1,5-imino-3-O-(2,3,4-tri-O-benzyl-α-D-glucopyranosyl)-D-mannitol (**59**)*

A solution of **55** (400 mg, 0.40 mmol) in methanolic 0.5 N sodium methoxide (4 mL) was kept at room temperature for 15 h. It was diluted with dichloromethane and the solution was washed with 1% hydrochloric acid and water. The product obtained on evaporation was purified on a column of silica gel (dichloromethane – ethyl acetate, 1:2) to provide **59** (294.9 mg, 91%) as a white foam; $[\alpha]_D$ +33.6 (*c* 0.3, chloroform). ^{1}H NMR ($CDCl_3$) δ: 7.4–7.0 (m, 25H, 5Ph), 4.99 (d, 1H, $J_{1',2'}$ 3.5 Hz, H-1'), 4.20 (dd, overlapped, 1H, $J_{1A,1B}$ 14.5 Hz, $J_{1A,2}$ 2.5 Hz, H-1A), 3.94 (bs, 1H, H-2), 3.46 (dd, 1H, $J_{2',3'}$ 10.0 Hz, H-2'), 2.82 (dd, 1H, $J_{1B,2} <$ 1 Hz, H-1B). ^{13}C NMR ($CDCl_3$) δ: 156.76 (CO), 100.21 (C-1'), 65.77 (C-6), 61.98 (C-6'), 57.47 (C-5), 40.93 (C-1). Anal. calcd. for $C_{48}H_{51}NO_{10}$: C 71.89, H 6.41, N 1.75; found: C 71.68, H 6.36, N 1.60.

*2,4-Di-O-benzyl-N-6-O-carbonyl-1,5-dideoxy-1,5-imino-3-O-(2,3,4-tri-O-benzyl-6-O-methyl-α-D-glucopyranosyl)-D-mannitol (**60**)*

A mixture of **59** (258 mg, 0.32 mmol), silver oxide (410 mg, 1.77 mmol), and methyl iodide (300 μL, 4.82 mmol) in toluene (6.3 mL) was stirred in the dark for 17 h. More methyl iodide (200 μL, 3.21 mmol) was added and stirring continued for another 4 h. Filtration and evaporation of the solvent left a crystalline residue, which was passed through a column of silica gel (dichloromethane – ethyl acetate, 1:1, 1:2) to give **60** (240.3 mg, 91%) as a white solid; $[\alpha]_D$ +22 (*c* 0.3, chloroform). ^{1}H NMR ($CDCl_3$) δ: 7.4–7.0 (m, 25H, 5Ph), 4.99 (d, 1H, $J_{1',2'}$ 3.5 Hz, H-1'), 4.21 (dd, 1H, $J_{1A,1B}$ 14.5 Hz, $J_{1A,2}$ 2.5 Hz, H-1A), 4.01 (m, 1H, H-2), 3.50 (overlapped, H-2'), 3.27 (s, 3H, CH_3O), 2.82 (dd, 1H, $J_{1B,2} <$ 1 Hz, H-1B). Anal. calcd. for $C_{49}H_{53}NO_{10}$: C 72.13, H 6.55, N 1.72; found: C 72.21, H 6.26, N 1.67.

*2,4-Di-O-benzyl-1,5-dideoxy-1,5-imino-3-O-(3,4,6-tri-O-benzyl-2-deoxy-α-D-*arabino*-hexopyranosyl)-D-mannitol (**61**)*

57 (85.5 mg, 0.109 mmol) in a 0.5 N potassium hydroxide solution in 90% ethanol (9.5 mL) was heated at 60°C for 3 h. It was neutralized with acetic acid, followed by solvent removal. The material was taken up in dichloromethane, and the solution was washed with a little water and evaporated. Chromatographic purification on a column of silica gel (dichloromethane – 3% methanol) afforded **61** (73.6 mg, 89%) as a syrup; $[\alpha]_D$ +20.6 (*c* 0.5, chloroform). ^{1}H NMR ($CDCl_3$) δ: 7.45–7.10 (m, 30H, 5Ph), 5.26 (unresolv. dd, 1H, $J_{1',2ax'}$ 3.0 Hz, $J_{1',2eq'} \sim 1$ Hz, H-1'), 4.02 (ddd, 1H, $J_{3',4'}$ 9.0 Hz, $J_{2ax',3'}$ 11.5 Hz, $J_{2eq',3'}$ 4.5 Hz, H-3'), 3.89 (dt, 1H, $J_{4',5'}$ 9.5 Hz, H-5'), 3.54 (overlapped, H-4'), 3.06 (dd, 1H, $J_{1A,2}$ 2.5 Hz, $J_{1A,1B}$ 14.2 Hz, H-1A), 2.55 (m, 1H, H-5), 2.47 (d, 1H, $J_{1B,2} <$ 1 Hz, H-1B), 2.25 (dd, 1H, $J_{2eq',2ax'}$ 13.0 Hz, H-2eq'), 1.71 (ddd, 1H, H-2ax'). ^{13}C NMR ($CDCl_3$) δ: 99.69 (C-1'), 62.57, 61.66 (C-5, C-6), 46.57 (C-1), 35.82 (C-2'). Anal. calcd. for $C_{47}H_{51}NO_8$: C 74.28, H 7.03, N 1.84; found: C 74.26, H 7.14, N 1.75.

*2,4-Di-O-benzyl-1,5-dideoxy-1,5-imino-3-O-(2,4,6-tri-O-benzyl-3-O-methyl-α-D-glucopyranosyl)-D-mannitol (**62**)*

A solution of **53** (230 mg, 0.24 mmol) in methanolic 0.5 N sodium methoxide (2 mL) was left at room temperature for 17 h. The solvent was evaporated and the remainder (**58**) was taken up in 0.5 N sodium hydroxide in 90% ethanol (4 mL) and heated at 70°C for 30 h. It was neutralized with acetic acid, followed by evaporation. The resultant material was taken up in dichloromethane and the solution washed with a little water, followed by drying and

evaporation of the organic solution. Chromatographic purification of the crude product on a column of silica gel (dichloromethane – 3% methanol) afforded **62** (163.5 mg, 87%) as a tough syrup; $[\alpha]_D$ +31.3 (*c* 0.2, chloroform). ^{1}H NMR ($CDCl_3$) δ: 7.4–7.1 (m, 25H, 5Ph), 4.02 (unres. ddd, 1H, $J_{4',5'}$ 10.0 Hz, H-5′), 3.82 (unres. m, 1H, H-2), 3.63 (s, 3H, CH_3O), 3.03 (dd, 1H, $J_{1A,1B}$ 14.5 Hz, $J_{1A,2}$ 4.5 Hz, H-1A), 2.64 (m, 1H, H-5), 2.52 (bd, 1H, H-1B). Anal. calcd. for $C_{48}H_{53}NO_9$: C 73.17, H 6.78, N 1.78; found: C 72.84, H 6.98, N 1.71.

*2,4-Di-*O*-benzyl-1,5-dideoxy-1,5-imino-3-*O*-(2,3,4-tri-*O*-benzyl-6-*O*-methyl-α-*D*-glucopyranosyl)-*D*-mannitol (**63**)*

Compound **60** (220 mg, 0.27 mmol) in 0.9 N potassium hydroxide solution in 90% ethanol (5 mL) was heated at 60°C for 7 h. The mixture was neutralized with glacial acetic acid and evaporated to dryness. The remainder was taken up in dichloromethane, followed by washing of the organic solution with a little water, drying, and evaporation. Column chromatography on silica gel (dichloromethane – 3–5% methanol) provided syrupy **63** (190 mg, 89%); $[\alpha]_D$ +44 (*c* 0.2, chloroform). ^{1}H NMR ($CDCl_3$) δ: 7.4–7.15 (m, 25H, 5Ph), 4.01 (m, 2H, H-3′, H-5′), 3.81 (m, 1H, H-2), 3.48 (dd, $J_{1',2'}$ 3.5 Hz, $J_{2',3'}$ 9.5 Hz, H-2′), 3.22 (s, 3H, CH_3O), 3.08 (dd, 1H, $J_{1A,1B}$ 14.5 Hz, $J_{1A,2}$ 5.0 Hz, H-1A), 2.69 (m, 1H, H-5), 2.58 (dd, 1H, $J_{1B,2}$ 1.8 Hz, H-1B).

*1,5-Dideoxy-3-*O*-(2-deoxy-α-*D*-*arabino*-hexopyranosyl)-1,5-imino-*D*-mannitol (**64**)*

Compound **61** (55.4 mg, 0.073 mmol), 5% palladium-on-carbon (50 mg), and methanolic 0.12 N hydrochloric acid (759 μL, 0.09 mmol) in methanol (6 mL) were hydrogenated in the hydrogen stream for 2 h. The excess of acid was neutralized by a minimal amount of triethylamine to avoid hydrolysis of the glycosidic linkage. Usual processing and gel filtration provided **64** (22.2 mg, 88%) as a solid after lyophilization of an aqueous solution; $[\alpha]_D$ +53 (*c* 0.3, water). The ^{1}H and ^{13}C NMR data are reported in Tables 1 and 2.

*1,5-Dideoxy-1,5-imino-3-*O*-(3-*O*-methyl-α-*D*-glucopyranosyl)-*D*-mannitol-hydrochloride (**65**)*

A mixture of **62** (140 mg, 0.18 mmol), 5% palladium-on-carbon (140 mg), and methanolic 0.12 N hydrochloric acid (1.9 mL, 0.23 mmol) in methanol (7 mL) was hydrogenated in a hydrogen stream for 2 h. Usual processing provided **65** (64.3 mg, 96%) as a white solid; $[\alpha]_D$ +72 (*c* 0.2, water). The ^{1}H and ^{13}C NMR data are reported in Tables 1 and 2.

*1,5-Dideoxy-1,5-imino-3-*O*-(6-*O*-methyl-α-*D*-glucopyranosyl)-*D*-mannitol-hydrochloride (**66**)*

Compound **63** (190 mg, 0.24 mmol), 5% palladium-on-carbon (190 mg), and 0.12 N methanolic hydrochloric acid (2.5 mL, 0.3 mmol) in methanol (8 mL) were hydrogenated in the hydrogen stream for 2 h. Usual processing followed by gel-filtration (Sephadex LH 20, ethanol–water, 1:1) provided **66** (82.9 mg, 92%) as a white solid; $[\alpha]_D$ +67.8 (*c* 0.2, water). The ^{1}H and ^{13}C NMR data are reported in Tables 1 and 2.

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