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THE SYNTHESIS OF 5-*O*-(2-ACETAMIDO-2-DEOXY-α-D-GLUCOPYRANOSYL)-β-D-GLUCOFURANOSE

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ABSTRACT

Condensation of dimeric 3,4,6-tri-*O*-acetyl-2-deoxy-2-nitroso-α-D-glucopyranosyl chloride (**1**) with 1,2-*O*-isopropylidene-α-D-glucofuranurono-6,3-lactone (**2**) gave 1,2-*O*-isopropylidene-5-*O*-(3,4,6-tri-*O*-acetyl-2-deoxy-2-hydroxyimino-α-D-*arabino*-hexopyranosyl)-α-D-glucofuranurono-6,3-lactone (**3**) Benzoylation of the hydroxyimino group with benzoyl cyanide in acetonitrile gave 1,2-*O*-isopropylidene-5-*O*-(3,4,6-tri-*O*-acetyl-2-benzoyloxyimino-2-deoxy-α-D-*arabino*-hexopyranosyl)-α-D-glucofuranurono-6,3-lactone (**4**) Compound **4** was reduced with borane in tetrahydrofuran, yielding 5-*O*-(2-amino-2-deoxy-α-D-glucopyranosyl)-1,2-*O*-isopropylidene-α-D-glucofuranose (**5**), which was isolated as the crystalline *N*-acetyl derivative (**6**) After removal of the isopropylidene acetal, the pure, crystalline title compound (**10**) was obtained

INTRODUCTION

Reducing disaccharides having a (1→5)-linkage and higher saccharides that contain such a structural element form a poorly studied group of substances The presence of (1→5)-linkages in carbohydrates may easily be overlooked, as methylation analysis and periodate oxidation do not discriminate between (1→4)- and (1→5)-linkages in linear or branched oligosaccharides Occasionally, the (1→4)-linkage is established without seriously considering the alternative On the other hand, conclusions favouring the (1→5)-linkage are based on qualitative results obtained from partial acid hydrolysis and "higher mobilities" on thin-layer chromatography (t l c) of species containing furanoid units[1], without support from instrumental techniques [n m r spectroscopy and mass spectrometry (m s)]

The few (1→5)-linked compounds described in the literature can be divided into three groups (*a*) leucrose (5-*O*-α-D-glucopyranosyl-D-fructopyranose)[2,3], consisting of two pyranoid rings, (*b*) disaccharides consisting of two furanoid rings, *e g* , 5-*O*-β-D-galactofuranosyl-D-galactofuranose[4], and (*c*) disaccharides consisting of one pyranoid and one furanoid ring at the non-reducing and reducing site, respectively.

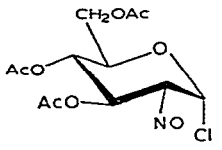
Disaccharides of type (*c*) are rare and are found in partial, acid hydrolysates of hetero-oligosaccharides as a result of the acid lability of furanosides[5] Watson[1] showed the presence of a 5-*O*-*β*-D-glucopyranosyl-D-galactofuranosyl unit in a hexasaccharide obtained from the type specific substance (S 33 B) from Pneumococcus type 33B 5-*O*-*β*-D-Glucopyranosyl-D-glucofuranose was found in hydrol, the residual mother liquor in the production of D-glucose by acid hydrolysis of corn starch[6]. Maghuin-Rogister and Jadot[7] reported the isolation of maniocose (5-*O*-*α*-D-glucopyranosyl-D-glucofuranose) from the roots of manioc The synthesis[8] of maniocose was also accomplished *via* condensation of 3,4,6-tri-*O*-acetyl-1,2-anhydro-*α*-D-glucose and 3,6-di-*O*-acetyl-1,2-*O*-isopropylidene-*α*-D-glucofuranose (**11**) However, in our laboratory, a sample of the isolated disaccharide proved to be mainly isomaltose Gas–liquid chromatography (g l c) showed that the retention times of per-*O*-Me_3Si-isomaltose and per-*O*-Me_3Si-"maniocose" and of their respective per-*O*-Me_3Si-alditols were identical Moreover, the per-*O*-Me_3Si-alditols of leucrose and maniocose were definitely different by g l c The ^{1}H-n m r spectra of isomaltose and "maniocose" in deuterium oxide and of their per-*O*-Me_3Si and per-*O*-Me derivatives in acetone-d_6 and acetonitrile-d_3 are indistinguishable In our hands, the synthesis according to Maghuin-Rogister[8] yielded isomaltose in substantial amounts because the conditions for the removal of the isopropylidene groups were such as to give an acid reversion mixture Apart from this acid reversion, evidence was obtained that the synthesis afforded a (1→3)-linked disaccharide up to the isopropylidene-protected stage as the result of a C-3→C-5 acetyl migration in **11** The C-3→C-5 acetyl migration in **11** was also encountered in methylation studies[9].

Previously[10], it was stated that differentiation between a (1→5)- and a (1→6)-linked per-*O*-Me_3Si-disaccharide by means of m s was impossible However, this conclusion was based on the use of a maniocose sample as the only (1→5)-linked compound Since maniocose is, in reality, isomaltose, it was considered necessary to synthesize a set of (1→5)-linked disaccharides, and to determine whether regularities in their mass spectra could be found to enable discrimination between the (1→5)- and (1→6)-linkages The most-promising approach to the synthesis of *α*-(1→5)-linked disaccharides seemed to be that of Lemieux *et al* [11 12]. These workers showed that condensation of dimeric 3,4,6-tri-*O*-acetyl-2-deoxy-2-nitroso-*α*-D-glucopyranosyl chloride (**1**) with suitably protected sugars yielded the corresponding 2-deoxy-2-hydroxyimino-*α*-D-*arabino*-hexopyranosides, which in turn give access to the *α*-D-linked amino-sugar disaccharides[13] as well as to the neutral *α*-D-linked disaccharides[14]. We now report the synthesis of a (1→5)-*α*-D-linked amino-sugar disaccharide

RESULTS AND DISCUSSION

As a suitably protected sugar aglycon having HO-5 free, the readily available 1,2-*O*-isopropylidene-*α*-D-glucofuranurono-6,3-lactone[15] (**2**) was selected This compound has the advantages that it does not possess protecting groups that are

likely to migrate (*cf.* **11**) and that borane reduction of **4** should lead to the formation of the anticipated monosaccharide moieties (2-amino-2-deoxy-D-glucose and D-glucose) in one step



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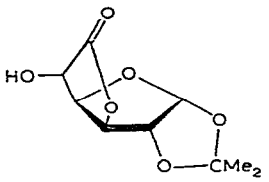

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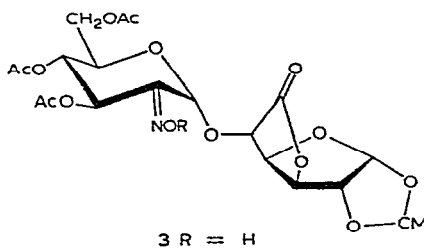


3 R = H
4 R = Bz

Condensation of **1** and **2** was carried out in *N*,*N*-dimethylformamide without an acid acceptor The presence of an acid acceptor is not recommended, as it is possible that α-hydroxyiminoglycoside formation may involve some anomerisation of initially formed β-hydroxyiminoglycoside[11] Indeed, Miyai and Jeanloz[16] synthesized mainly a β-D-linked disaccharide with *N*,*N*,2,6-tetramethylaniline as acid acceptor The condensation of **1** and **2** yielded a complex mixture (t l c) from which **3** was isolated by column chromatography at low temperature At room temperature, compound **3** was almost completely destroyed on silicic acid, and mainly **2** was eluted The coupling product **3** decomposed on prolonged standing in *N*,*N*-dimethylformamide The instability of **3** was more pronounced in both weak acid and alkaline media For this reason, the attempted conversion of **3** into the corresponding 3,4,6-tri-*O*-acetyl-α-D-*arabino*-hexopyranosid-2-ulose (a precursor for the synthesis of the neutral disaccharide[14]) with acetaldehyde in acetonitrile–hydrochloric acid or titanium(III) chloride failed Attempts to obtain the acetoxyimino derivative of **3**, which is a precursor for the synthesis of the amino-sugar disaccharide[13], by acetylation with acetic anhydride in pyridine also failed

Discrimination between α- or β-hydroxyiminoglycosides on the basis of the chemical shift of the H-1′ singlet is unreliable, because the chemical shifts of the anomeric hydrogen of the hydroxyiminoglycosyl unit varies over 0 5 p p m depending on the nature of the aglycon[12] A difference of only ~0 3 p p m is to be expected for H-1 of α- and β-analogues[11] Surprisingly, however, the electron paramagnetic resonance (e p r.) spectrum of the stable iminoxy radicals (>C=N–O) generated on oxidation of **3** is essentially the same as that of methyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-hydroxyimino-α-D-*arabino*-hexopyranoside, and is different from what has been interpreted to be that of the β isomer[17].

Acylation of the hydroxyimino group was accomplished with benzoyl cyanide in acetonitrile in the presence of a catalytic amount of triethylamine. This method was originally applied in the benzoylation of rather sensitive molecules in nucleoside and nucleotide chemistry[18] The structure of the stable benzoyloxyimino compound **4**

was proved by 220-MHz ^{1}H-n m r spectroscopy (Table I) The coupling constants and chemical shifts of H-3′ and H-4′ are of the same magnitude as those reported for 2-acetoxyimino-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-*arabino*-hexopyranosides[11]

TABLE I

^{1}H-N M R DATA OF **2** (100 MHz), **3** (60 MHz), AND **4** (220 MHz) IN CHLOROFORM-*d* WITH Me_4Si AS INTERNAL REFERENCE

Reducing unit

	H-1	*H-2*	*H-3*	*H-4*	*H-5*	$J_{1,2}$	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$
2	5 98	4 80	4 83	4 93	4 55	3 7	0 0	3 1	4 4
3	6 01	4 74	←4 86–5 05[a]→		4 64	3 7	—[a]	—[a]	4 4
4	6 03	4 80	4 84	5 07	4 61	3 7	0 0	3 1	4 4

Non-reducing unit

	H-1′	*H-3′*	*H-4′*	*H-5′*	*H-6a′*	*H-6b′*	$J_{3,4}$	$J_{4,5}$	$J_{5,6a}$	$J_{5,6b'}$	$J_{6a,6b}$
3	6 18	5 84	5 25	4 56[a]	←4 2–4 4[a]→		9 5	9 5	—[a]	—[a]	—[a]
4	6 26	5 99	5 39	4 49	4 31	4 16	10 0	10 0	3 7	2 5	−12 5

Other groups

	OH	CMe_2		*OAc*	*OBz*
2	3 4 (HO-5)	1 35	1 51	—	—
3	9 3 (oxime)	1 35	1 52	2 07 (max)	—
4	—	1 36	1 51	2 05 2 07 2 16	7 40–8 10

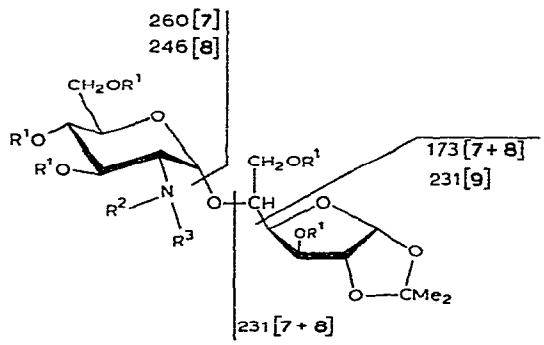
[a]Complex multiplet

The stability of **4** (in contrast to **3**) and the stable 3,4,6-tri-*O*-acetyl-2-deoxy-2-hydroxyimino-α-D-hexopyranosides described by Lemieux[12] imply that the driving force in the decomposition of **3** lies in the hydroxyimino group in combination with the present aglycon group.

Compound **4** was reduced with borane in tetrahydrofuran under the same conditions as for acetoxyiminoglycosides[13]. The ninhydrin-positive compound (**5**) thus obtained in solution was isolated as the crystalline *N*-acetyl derivative **6** [5-*O*-(2-acetamido-2-deoxy-α-D-glucopyranosyl)-1,2-*O*-isopropylidene-α-D-glucofuranose] The configuration of the inter-sugar glycosidic bond in **6** was unambiguously proved to be α by ^{1}H-n.m r spectroscopy

Methylation[26] of **6** resulted in a partially *N*-methylated mixture of the per-*O*-Me derivatives **7** and **8**. Although there is clear evidence from the fragment ions at *m/e* 173 and *m/e* 231 in the mass spectra of **7** and **8** for an exocyclic glycosidic

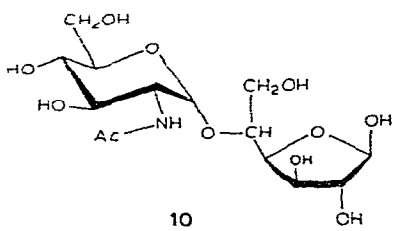
linkage, no discrimination was possible between the (1→5)- and (1→6)-linkages (Scheme 1). The same information with respect to the glycosidic linkage was given by the mass spectrum of the per-*O*-Me_3Si derivative (**9**), although the primary fragmentations were less pronounced than in the spectra of **7** and **8**. However, the relatively small peak at *m/e* 231 ($C_{10}H_{19}O_4Si$, calc. 231.1053, measured 231.1071) in the spectrum of **9** was due to to the 3-*O*-Me_3Si analogue of the 3-*O*-Me fragment (*m/e* 173) in the spectra of **7** and **8**, as shown by high-resolution mass spectrometry (Scheme 1)



5 $R^1 = R^2 = R^3 = H$
6 $R^2 = Ac, R^1 = R^3 = H$
7 $R^2 = Ac\ R^1 = R^3 = Me$
8 $R^2 = Ac, R^3 = H\ R^1 = Me$
9 $R^2 = Ac\ R^3 = H\ R^1 = Me_3Si$

Scheme 1

The permethylated mixture of **7** and **8** was further processed as described for alditol acetate analysis[20] using sodium borodeuteride in the reduction step The g l c–m s data for 1,2,4,5-tetra-*O*-acetyl-3,6-di-*O*-methyl-D-glucitol-d_1 (formula **12**) were the same as those earlier reported[21,22], except for the mass shift of one dalton in some fragment ions due to the introduction of the deuterium isotope. Apart from synthetic evidence, proof of the (1→5)-linkage in **6** is given by comparing the mass-spectral data of **7** and **8** and of the derived alditol acetate **12**



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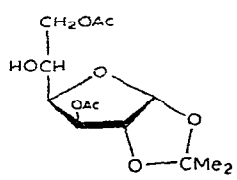

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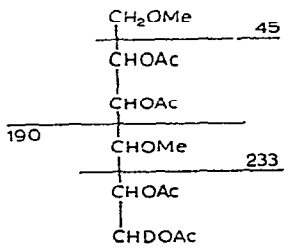

12

Because of the unknown acid-lability of the inter-sugar glycosidic linkage in **6**, 0 5M sulfuric acid was selected for the removal of the isopropylidene group under conditions that were easier to monitor than in trifluoroacetic acid–water[23] (9.1) The deprotected sugar (**10**) readily crystallized G l c analysis, after trimethylsilylation of the crystalline material, indicated that one anomer was present in the crystalline state The anomerisation of this furanose sugar is extremely rapid, the optical rotation ($\alpha_{t\infty}$) of **10** being constant directly after dissolution of crystalline **10** in water. However, the anomerisation of the crystalline disaccharide in pyridine-d_5 could be followed by ^{1}H-n m r spectroscopy. Thus, it was shown that the free sugar occurs as the β-D anomer in the crystalline state G l c analysis of trimethylsilylated, freeze-dried **10** showed two peaks in the ratio 1:1 for the two anomers (Table II) This finding was confirmed by the ^{1}H-n m r spectrum of **10** in deuterium oxide, which showed a 1·1 pair of doublets for H-1 (α, $J_{1,2}$ 3 7 Hz, β, $J_{1,2}$ 0 7 Hz) and also a 1 1 pair of doublets for H-1′ (both $J_{1',2'}$ 3 4 Hz, α) The small coupling constant of the β-anomeric hydrogen of the furanose moiety is in accordance with calculated and observed values of β-D-gluco- and β-D-galacto-furanosides[24]

Table II

G L C DATA[a] OF THE PER-*O*-TRIMETHYLSILYL DERIVATIVES OF **6**, **10**, AND THE ALDITOL OF **10**

	3% of OV-225 T_S	*3 8% of SE-30* T_S
Crystalline **6**	8 4 ±0 3	1 73 ±0 02
Mother liquor of **6**	8 4 ±0 3	1 73 ±0 02
	10 3 ±0 5[b]	1 08 ±0 01[b]
Crystalline **10**	4 4 ±0 2[c]	1 80 ±0 02[c]
Freeze-dried **10**	4 4 ±0 2[c]	1 80 ±0 02[c]
	5 0 ±0 2[d]	1 95 ±0 02[d]
Alditol[e] of **10**	4 5 ±0 2	2 57 ±0 02

[a]At 245°, the relative retention times (T_S) are given relative to the per-*O*-trimethylsilyl derivative of sucrose [b]Compound of unknown structure [c]Assigned as the β-anomeric form by time-dependent ^{1}H-n m r spectroscopy [d]α-Anomeric form [e]Prepared by sodium borodeuteride reduction of **10**

EXPERIMENTAL

Materials — Silylation Grade *N*,*N*-dimethylformamide was purchased from Pierce Chemical Company Nitrosyl chloride[12,16] (Matheson Gas Products) was distilled immediately before use A M borane solution in tetrahydrofuran was purchased from Aldrich Chemical Company Benzoyl cyanide of practical grade was obtained from Fluka A G. Indophenol Blue (Baker TLC reagent) was used as a reference material

General methods — Melting points are corrected Solutions were concentrated under diminished pressure (water aspirator) at 40° (bath). Optical rotations were

recorded at ambient temperature with a Perkin–Elmer 141 instrument. ^{1}H-N m r spectra (internal Me_4Si) were recorded with Varian A-60, HA-100, and HR-220 spectrometers Infrared spectra were recorded with a Beckman IR-8 instrument Trimethylsilylation of 1-mg samples of sugars was performed with 1,1,1,3,3,3-hexamethyldisilazane and chlorotrimethylsilane in pyridine[10]. Methanolysis of sugar samples followed by g l c analysis of the methyl per-*O*-Me_3Si-glycosides was performed by the procedure of Clamp[19] Directions for the generation of the iminoxy radicals (>C=N−O) from the parent oxime **3** and for the interpretation of the e p r spectra will be published elsewhere[17]

Chromatography — T l c was performed on silica gel (Schleicher & Schull TLC Ready Plastic Foil FR-1500) Mobilities are expressed as R_F, R_{GLC}, and $R_{INDOPHENOLBLUE}$ (R_I) values Detection was effected by spraying with 20% conc sulfuric acid in methanol and charring at 120° for 10 min The following solvents were used *A*, ethyl ether–light petroleum (b p 40–60°) (4 1), *B*, acetic acid–ethyl acetate–water–1-butanol (6 3 1 8), *C*, hexane–acetone (6 4), and *D*, 2-propanol–ethyl acetate–water (2 2 1)

G l c of per-*O*-Me and per-*O*-Me_3Si derivatives of sugars was carried out on a Pye 104 instrument equipped with flame-ionisation detector and glass columns (1.60 m × 4 mm) packed with 3 8% of SE-30 or 3% of OV-225 on Chromosorb W-AW DMCS (80–100 mesh) The gas flow rate for N_2 was 40 ml/min The retention times (T_S) are given relative to that of per-*O*-Me_3Si-sucrose

Mass spectrometry. — 70-eV Mass spectra were recorded on an AEI MS-902 mass spectrometer at an ion chamber temperature of 80–100° (trap current 500 μA, accelerating voltage 8 kV) High-resolution mass measurements were performed with a dynamic resolving power of 10,000 and scan speed of 16 sec per mass decade, with the spectrometer connected on-line with a Ferranti Argus 500 computer The exact masses were converted into element lists as described by Van't Klooster *et al*[25] 75-eV Mass spectra were recorded on a Jeol JGC-1100/JMS-07 combination (column material 3% of SE-30 on Chromosorb W-AW DMCS (80–100 mesh), oven temperature 158°, ion-source temperature 250°, accelerating voltage 3 kV, ionizing current 300 μA)

1,2-O-Isopropylidene-5-O-(3,4,6-tri-O-acetyl-2-deoxy-2-hydroxyimino-α-D-arabino-hexopyranosyl)-α-D-glucofuranurono-6,3-lactone (**3**) — Dimeric 3,4,6-tri-*O*-acetyl-2-deoxy-2-nitroso-α-D-glucopyranosyl chloride[12,16] (**1**, 8 00 g, 11 9 mmol) and 1,2-*O*-isopropylidene-α-D-glucofuranurono-6,3-lactone[15] (**2**; 8 00 g, 37 mmol) were dissolved in *N,N*-dimethylformamide (37 ml) The solution was flushed with dry nitrogen and kept, with exclusion of moisture, at 30° for 16 h in the dark The coloured reaction mixture was concentrated to 20 ml *in vacuo* using a vacuum capillary, diluted with chloroform (125 ml), washed with iced water (3 × 30 ml), dried (sodium sulfate), and concentrated The brown, syrupy residue was immediately applied to a column (1 25 m × 4 cm) of silica gel (Merck Kieselgel 60, 70–230 mesh) with a cooling jacket (4°) and eluted with solvent *A*. A mixture of products (8 40 g) having R_F values (t l.c, *A*) of 0.44, 0 39, 0.26 and 0.15, respectively, was eluted in three fractions

Preparative separation of these compounds failed because of their similar mobilities and tendency to decompose into a polymeric material and **2** The last fraction (3 70 g, 7.15 mmol, 30%) consisted mainly of **3** as judged from t l c (R_F 0 15, *A*) and spectroscopic analysis (^{1}H-n m r and e p r of the iminoxy radicals[17]) The material did not crystallize and was obtained as a brittle foam *in vacuo* Compound **3** could be stored at −20° over longer periods The ^{1}H-n m r spectrum (Table I) was consistent with the allocated structure, H-2, H-3, H-6a′, and H-6b′ gave unresolved resonances I r (film) data 3300 (OH, oxime), 1800 (C=O, lactone), and 1745 cm^{-1} (C=O, acetyl)

1,2-O-Isopropylidene-5-O-(3,4,6-tri-O-acetyl-2-benzoyloxyimino-2-deoxy-α-D-arabino-hexopyranosyl)-α-D-glucofuranurono-6,3-lactone (**4**) — A solution of **3** (850 mg, 1 64 mmol) and benzoyl cyanide (325 mg, 2 48 mmol) in dry acetonitrile (10 ml) was cooled to 0° and triethylamine (50 μl) was added The solution was allowed to reach room temperature during 10–15 min T.l c (*A*) then indicated the absence of the starting material Methanol (1 ml) was added and the solution was concentrated to a small volume The residue was twice recrystallized from ethanol–ether to yield 600 mg (0 97 mmol, 59%) of **4**, m p 159°, $[\alpha]_D^{20}$ +73 0° (*c* 2 7, chloroform) (Found C, 54 3, H, 5 15, N, 2.1, O, 38 2 $C_{28}H_{31}NO_{15}$ calc C, 54 1, H, 5 03, N, 2 25, O, 38 6%) The i r spectrum revealed the disappearence of the hydroxyl absorption of the oxime The structure of **4** was confirmed by 220-MHz ^{1}H-n m r spectroscopy (Table I)

5-O-(2-Acetamido-2-deoxy-α-D-glucopyranosyl)-1,2-O-isopropylidene-α-D-glucofuranose (**6**) — A solution of **4** (1 19 g, 1 92 mmol) in dry tetrahydrofuran (15 ml) was flushed with dry nitrogen and cooled to −60° A M solution of borane in tetrahydrofuran (18 ml) was slowly added with stirring and cooling below −40° The mixture was allowed to reach room temperature during 3 h Then methanol was added at −60° to destroy the excess of borane The resulting solution was concentrated, and methanol was distilled from the residue three times to remove boric acid The ninhydrin-positive material (**5**) was *N*-acetylated with acetic anhydride in 50% aqueous methanol[14] However, *O*-acetylation occurred to a small extent on concentration of the solution, as indicated by faster-moving bands on t l c (*B*) These bands disappeared on subsequent *O*-deacetylation with triethylamine in aqueous methanol[14] Crystallisation from methanol–ether gave **6** (214 mg, 26%), m p 215° (dec), $[\alpha]_D$ +95° (*c* 2 6, water), R_F 0 45, R_I 0 84, R_{GLC} 1 21 (t.l c , solvent *B*) G l c showed a purity of ≮97% (Table II) ^{1}H-N m r data (100 MHz, D_2O, external Me_4Si) δ 1 82, 1 98 (2 s, CMe_2), 2 30 (s, NAc), 5 14 (d, $J_{1,2}$ 3 7, $J_{2,3}$ ~0 Hz, H-2, partially masked by HOD signal), 5 52 (d, $J_{1',2}$ 3 4 Hz, H-1′), 6 42 (d, $J_{1,2}$ 3.7 Hz, H-1), the signals for H-1 and H-1′ were assigned by comparison with the signals for H-1 in methyl 2-acetamido-2-deoxy-α-D-glucopyranoside and 1,2-*O*-isopropylidene-α-D-glucofuranose (Found C, 48.55, H, 7 1, N, 3 22 $C_{17}H_{29}NO_{11}$ calc C, 48 22, H, 6 9, N, 3 31%)

Further structural evidence for **6** was obtained by methanolysis and g l c [19 24], 2-acetamido-2-deoxyglucose and glucose were found in a ratio of 1 1 1 0 No 2-acetamido-2-deoxymannose was present, and it was also absent from the mother

liquor of **6** Thus, the reduction of **4** is stereospecific Concentration of the mother liquor of **6** gave a syrup (250 mg) which consisted of **6** and an unknown compound in the ratio 7 4 (g l c , Table II)

Methylation and alditol-acetate analysis of **6**. — Kuhn methylation[26] of **6** (10 mg) gave a syrup which was purified by p l c on silica (solvent *C*, detection under u v. light after spraying with 1% of morin in methanol, followed by extraction with chloroform) The main components **7** and **8** (R_I 0 60 and 0.54) were isolated in one fraction, which on g l c (3% of OV-225, 225°) gave one peak with a shoulder on the negative slope ^{1}H-n m r data ($CDCl_3$) δ 1 34 and 1.48 (2 s, CMe_2), 2 10 (s, NAc), 2 14 (s, MeN*Ac*), 3 03 (s, *Me*NAc), 5 81 (d, $J_{1,2}$ 3 8 Hz, H-1), 4 54 (d, $J_{2,1}$ 3 8 Hz, H-2), 4 90 (d, $J_{1',2'}$ 4 0 Hz, H-1'), OMe resonances at 3.32, 3 41, 3.42, 3 50, and 3 54, other protons 2 8–4 2

The foregoing data and the m s data (formulae **7** and **8**) indicate partial *N*-methylation of **6** The mixture of **7** and **8** was hydrolyzed, reduced with sodium borodeuteride, and acetylated[21] to give 1,2,4,5-tetra-*O*-acetyl-3,6-di-*O*-methyl-glucitol-d_1 (**12**) with *T* 3 95 relative to that of 1,5-di-*O*-acetyl-2,3,4,6-tetra-*O*-methyl-glucitol on 3% of OV-225 at 158° (lit value[22] 3 73). The structure of **12** was confirmed by m s

*5-O-(2-Acetamido-2-deoxy-α-*D*-glucopyranosyl)-β-*D*-glucofuranose* (**10**) — A solution of **6** (120 mg, 0 28 mmol) in 0 5M sulfuric acid (6 ml) was kept for 16 h at 22° Most of the isopropylidene groups were then removed (t l c , solvent *B*, R_F 0 23, R_I 0 43, R_{GLC} 0 62), whereas only small amounts of glucose were detectable The solution was neutralized with Dowex 1 x2 (HCO_3^-) resin, filtered, and freeze-dried G l c of the crude product (100 mg) indicated a 7 1 mixture of **10** and **6**, corresponding to an 86% yield of **10** The mixture was eluted from a column (30 × 1 cm) of Silica H (Merck) with solvent *D* at ~0 5 ml/min to give **6** (11 mg, 9%) and **10** (65 mg, 61%) Compound **10** crystallized readily from ethanol–ether, consisted of only one anomer (g l c , Table II), and had m p 128°, $[\alpha]_D$ +101° (equil , *c* 1 8, water) (Found C, 40 37, H, 7 16, N, 3 20, O, 49 38 $C_{14}H_{25}NO_{11}$ $2H_2O$ calc C, 40 10, H, 6 97, N, 3 34, O, 49 59%) The ^{1}H-n m r spectrum of **10** is consistent with a 1 1 mixture of anomeric forms δ 5 92 ($J_{1,2}$ 3 7 Hz, H-1), 5 02 ($J_{1',2'}$ 3 4 Hz, αH-1'), 5 64 ($J_{1,2}$ 0 7 Hz, βH-1), 5 07 ($J_{1',2'}$ 3 4 Hz, αH-1') The β configuration of crystalline **10** was determined by time-dependent ^{1}H-n m r spectroscopy in pyridine-d_5 The signal of αH-1 (δ 6 20) reaches maximum intensity within 3 min after dissolution of crystalline **10**

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