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A PRACTICAL SYNTHESIS OF 2-ACETAMIDO-2-DEOXY-3,4-DI-*O*-β-D-GALACTOPYRANOSYL-D-GALACTOPYRANOSE*

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ABSTRACT

Two different sugar derivatives having free hydroxyl groups have been employed for synthesis of the title trisaccharide. In one attempt, benzyl 2-acetamido-6-*O*-benzyl-2-deoxy-α-D-galactopyranoside (**8**) was treated with an excess of 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide (**11**), to give a mixture of products which, on fractionation, afforded benzyl 2-acetamido-6-*O*-benzyl-3,4-di-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-α-D-galactopyranoside (**15**) in 21% yield. However, in another, preferable approach, benzyl 2-acetamido-6-*O*-benzyl-2-deoxy-3-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-α-D-galactopyranoside (**13**) was treated with **11**, to produce **15** in 69% yield. Both **8** and **13** were conveniently prepared *via* reductive ring-opening of the respective 4,6-benzylidene acetals. *O*-Deacetylation of **15**, followed by hydrogenolysis, provided the title trisaccharide. The structure of the final product, and of various other intermediates, was established by ^{1}H- and ^{13}C-n.m.r. spectroscopy.

INTRODUCTION

The carbohydrate sequence β-Gal-(1→4)-GalNAc
3
↑
1
β-Gal

has been found to be a part of the carbohydrate moiety of the component of T-antigens[2]. The title trisaccharide is also reported to be a part of the core structure of the carbohydrate sequence of the blood-group, M-specific glycopentapeptide, which is further linked to neuraminic acid groups as follows[2].

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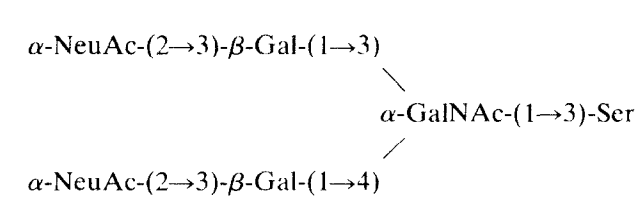


We had already accomplished the synthesis of 2-acetamido-2-deoxy-4-*O*-β-D-galactopyranosyl-D-galactose[3], and also reported that synthetic phenyl 2-acetamido-2-deoxy-3-*O*-β-D-galactopyranosyl-α-D-galactopyranoside acts as an acceptor for one of the sialyltransferases present in human serum[4]. The possibility of glycosyltransferases as tumor markers has been demonstrated by various investigators[5]. The title trisaccharide was needed in our laboratory for specificity testing of human sialyltransferases, and we now describe a practical and elegant synthesis of it.

RESULTS AND DISCUSSION

According to Bovin *et al.*[6], reaction of benzyl 2-acetamido-6-*O*-acetyl-2-deoxy-α-D-glucopyranoside with an equimolar proportion of 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide afforded the corresponding 3-*O*-substituted disaccharide derivative. In a recent review, Paulsen[7] mentioned that the title trisaccharide had been α-linked to $CH_2CH_2NHCO(CH_2)_4CO_2Me$ in his laboratory, and that the order in which the two D-galactosyl groups are attached to the GalNAc residue is important. The protected, disubstituted trisaccharide is obtained in high yield if the first galactosyl group is coupled at OH-3, and the second at OH-4.

Consequently, based on these two observations[6,7], we anticipated in our first approach that the reaction of an appropriately protected 2-acetamido-2-deoxy-D-galactose (having both OH-3 and -4 free) with an excess of compound **11** would afford the expected trisaccharide derivative. It is also apparent that the desired 6-*O*-substituted derivative, *e.g.*, benzyl 2-acetamido-6-*O*-benzyl(or 6-*O*-acetyl)-2-deoxy-α-D-galactopyranoside, can be obtained from benzyl 2-acetamido-2-deoxy-α-D-galactopyranoside in three steps, namely, acetonation to give the 3,4-*O*-isopropylidene derivative as the main product, protection of the primary hydroxyl group thereof by benzylation or acetylation, and *O*-deisopropylidenation.

Very recently, Garegg and Hultberg[8] reported a novel method of reductive ring-opening of carbohydrate benzylidene acetals with sodium cyanoborohydride in HCl–ether. Under these conditions, benzyl 2-acetamido-4,6-*O*-benzylidene-2-deoxy-α-D-galactopyranoside (**2**), preparable from **1**, gave **8** in 56% yield. It may also be pointed out that pure benzyl 2-acetamido-2-deoxy-α-D-galactopyranoside is obtained by removal of the 4,6-*O*-benzylidene group from **2**, as it has been reported[9] that, on treatment with benzyl alcohol containing a catalytic amount of dry HCl, commercially available 2-acetamido-2-deoxy-D-galactose gives a mixture of anomers that are conveniently separated by conversion into the 4,6-*O*-benzylidene

TABLE I

^{13}C-N M R [a] CHEMICAL SHIFTS[b]

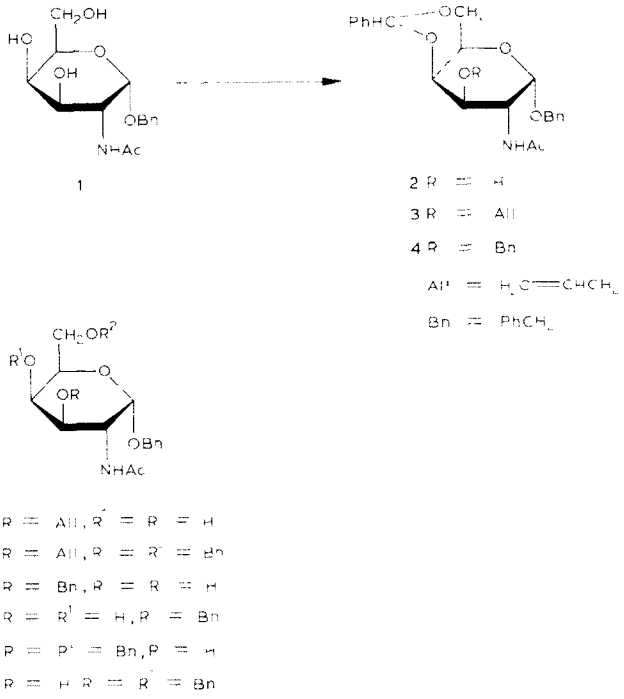
Atom	*Compound*				
	1	**7**	**8**	**9**	**10**
C-1	96.08	96.36	96.30	96.96	96.31
C-2	49.59	48.28	49.57	48.58	50.09
C-3	67 19	75.89	68.41	76.17	68.09
C-4	67.55	64.36	67.07	65 23	77.03
C-5	71.34	71 44	69.64	70.42	74.30
C-6	60.53	60.58	72.08	72.61	72.16
$COCH_3$	22.49	22.63	22.52	23.00	22 56
CH_2Ph	67.99	67.87	67.91	68 54	67.56
		69.88	69.55	70.03	68.91
					69.14
C=O	169.22	169.09	169.30	169.53	169 34

[a]At 25.2 MHz; solution in Me_2SO-d_6. [b]In p.p.m. downfield from Me_4Si (internal).

acetal followed by fractional recrystallization. In other words, the reductive-cleavage technique provides a rapid method for preparation of the diol **8**, required for further glycosylation. The structure of diol **8** was unambiguously supported by its ^{13}C-n.m.r. spectrum (see Table I). The pronounced, downfield shift of 11.55 p.p.m. exhibited by C-6 on benzylation, and the upfield shift (1.70 p.p.m.) of C-5, confirmed the position of substitution in **8**. In the present studies, we have also prepared benzyl 2-acetamido-3,6-di-*O*-benzyl-α-D-galactopyranoside[3] (**9**) by reductive cleavage of the benzylidene acetal of compound **4**, whereas, in a previous attempt[3], compound **9** was obtained by selective benzylation of benzyl 2-acetamido-3-*O*-benzyl-2-deoxy-α-D-galactopyranoside (**7**) by the phase-transfer-catalysis method.

The reaction of compound **2** with allyl bromide in *N*,*N*-dimethylformamide in the presence of barium oxide and barium hydroxide produced crystalline **3** which, on treatment with aqueous acetic acid at 100°, gave **5** in 81% yield. On benzylation, compound **5** afforded **6** which, on *O*-deallylation with potassium *tert*-butoxide in dimethyl sulfoxide[10], gave **10** in 76% yield. The structures of the benzylated derivatives **7**, **8**, **9**, and **10** were established by ^{13}C-n.m.r. spectroscopy (see Table I).

The coupling reaction of diol **8** with bromide **11** in dichloromethane was conducted in the presence of silver triflate and 1,1,3,3-tetramethylurea[11], the reaction being monitored by t.l.c. (4:1 chloroform–acetone) which, after 3 days, showed a major spot corresponding to benzyl 2-acetamido-6-*O*-benzyl-2-deoxy-3-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-α-D-galactopyranoside (**13**), trisaccharide derivative **15** as a minor product, and a significant amount of the starting material **8**. Additional amounts of the halide and catalyst were introduced, to give **15**; nevertheless, a considerable amount of starting material remained under these coupling conditions.



In another approach, we observed that the reductive cleavage[8] of benzyl 2-acetamido-4,6-*O*-benzylidene-2-deoxy-3-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-α-D-galactopyranoside (**12**) afforded **13**, having an *O*-benzyl group selectively at the primary position. *O*-Deacetylation of **13** provided benzyl 2-acetamido-6-*O*-benzyl-2-deoxy-3-*O*-β-D-galactopyranosyl-α-D-galactopyranoside (**14**) in 87% yield. The downfield shift of 1.58 p.p.m. exhibited by C-6 on benzylation (see Table II), and the upfield shift of C-5 (1.73 p.p.m.) confirmed the position of substitution in **14**. The complete absence of a C-6 signal in the region of 60–63 p.p.m. also confirmed that reductive cleavage of the benzylidene acetal **12** had given only the 6-*O*-benzyl derivative **13**. Treatment of alcohol **13** with bromide **11** in anhydrous dichloromethane in the presence of silver triflate and 1,1,3,3-tetramethylurea[11] afforded trisaccharide derivative **15** in 69% yield.

It is unclear why, on coupling with bromide **11**, diol **8** gives a low yield of expected compound **15**, although slow formation of disaccharide derivative **13** was observed. It is possible that, for glycosylation of diol **8**, changes in the reaction conditions, *e.g.*, in solvent and catalyst (probably a catalyst which is not hygroscopic) and use of a higher temperature, might provide compound **15** in appreciable yield. Nevertheless, based upon the present experimental observation, the use of "aglycon" hydroxide **13**, already having a 3-*O*-D-galactosyl unit, is strongly preferred for

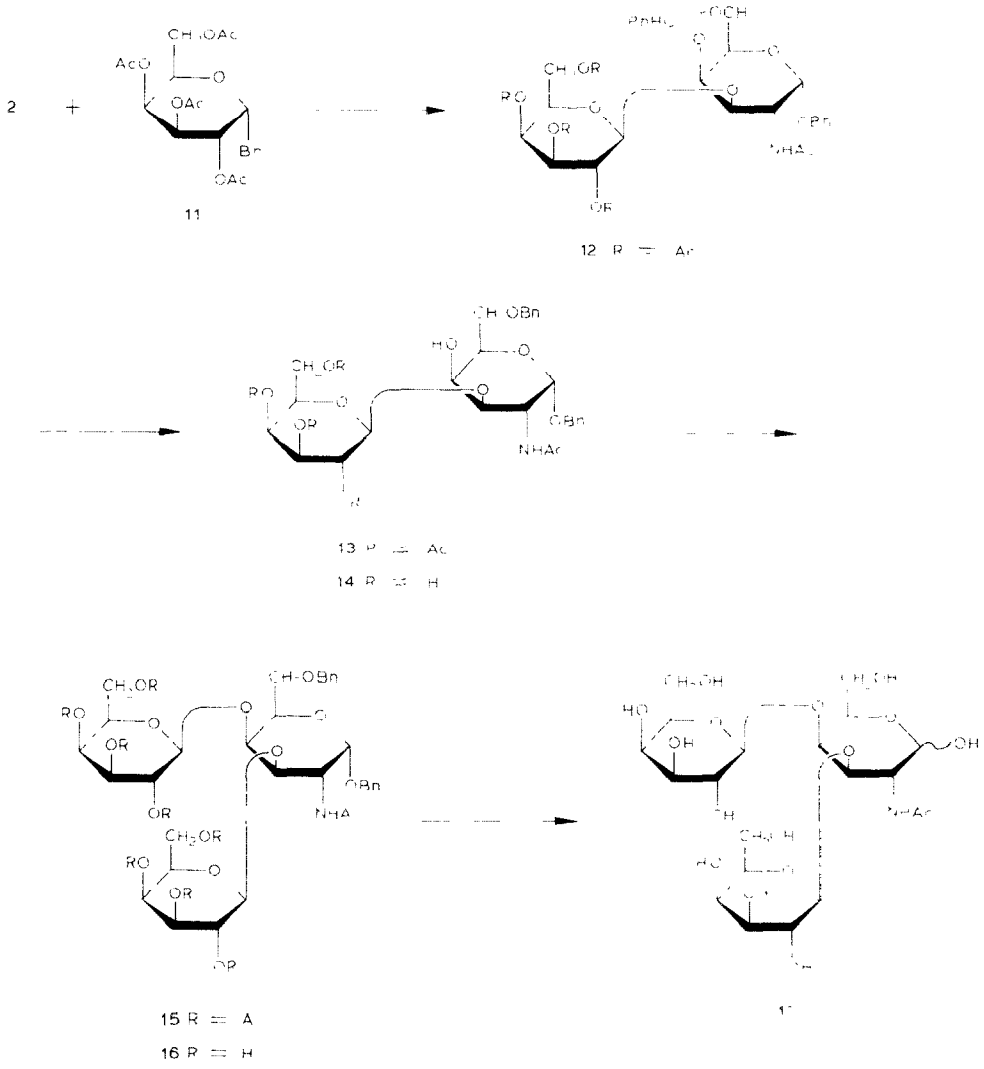
TABLE II

25.2-MHz, ^{13}C-N M R CHEMICAL SHIFTS[a]

Atom	*Compound*				
	14	**16**	**17α**		**17β**
C-1	96.46	97.93	92.37		96.20
C-2	48.34	50.65	50.57		53.99
C-3	75 53	77.86	77.62		80.64
C-4	67.58	77.32	76.95		76.05
C-5	69.61	71.73	70.89		75.25
C-6	72.11	72.33	61.76		61.53
C=O	169.48	173.91	175.62		175.94
CH_3	22.57	22.77	23.19		23.41
C-1′	103.55	104.78		104.24	
C-2′	70.64	73.13		71.72	
C-3′	73.24	74.51		73.59	
C-4′	68.05	70.22		69.74	
C-5′	75.21	76.23		75.86	
C-6′	60.42	62.40		62.13	
C-1″		106.35	105.92		106.22
C-2″		74.20		72.28	
C-3″		74.90		73.72	
C-4″		70.36		69.74	
C-5″		76.77		76.05	
C-6″		62.74		62.20	

[a]In p.p.m. downfield from Me_4Si. The solvent was Me_2SO-d_6 for **14**, CD_3OD for **16**, and D_2O for **17**. The reference (Me_4Si) was internal for solutions in Me_2SO-d_6 and CD_3OD, and external for that in D_2O.

preparation of compound **15**. An "aglycon" hydroxide, similar to **13** but having an ester as the protecting group on the primary hydroxyl group of the GalNAc residue, has been recommended for such glycosylation[12]; however, it is apparent that preparation of such a disaccharide "aglycon" hydroxide with a 6-ester group on the GalNAc unit is likely to be accomplished by selective acetylation of the corresponding 4,6-diol derivative. For example, should there be an interest in employing benzyl 2-acetamido-6-*O*-benzoyl-2-deoxy-3-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-α-D-galactopyranoside for further glycosylation at O-4 of GalNAc, this compound will have to be obtained by selectively benzoylating O-6 of the diol obtained by removal of the 4,6-*O*-benzylidene group of **12**. On the other hand, reductive cleavage[8] of **12** directly afforded, in one step, the desired, key intermediate having a 6-*O*-benzyl group on the GalNAc residue. The use of reductive cleavage of 4,6-*O*-benzylidene acetals, recently introduced into the field of carbohydrate chemistry, seems to be excellent for procurement of suitably protected sugars having free alcohol groups. To the best of our knowledge, the present investigation provides the first example of the technique of reductive ring-opening of a 4,6-benzylidene acetal in order to utilize the product successfully in the preparation of ap-



propriately protected disaccharides having a free alcohol group for further synthesis of higher saccharides.

O-Deacetylation[13] of **15** gave **16**, which, on catalytic hydrogenolysis in glacial acetic acid in the presence of 10% Pd–C, produced the title trisaccharide **17** as an amorphous material; its structure was confirmed by ^{13}C-n.m.r. spectroscopy.

EXPERIMENTAL

General methods. — Melting points were determined with a Fisher–Johns apparatus and are uncorrected. Optical rotations were measured with a Perkin–Elmer 241 polarimeter at room temperature. Ascending t.l.c. was conducted on

plates coated with a 0.25-mm layer of silica gel 60 PF-254 (E. Merck, Darmstadt, Germany); the components were located by exposure to u.v. light, or by spraying the plate with 5% sulfuric acid in ethanol, and heating. Elemental analyses were performed by Robertson Laboratory, Florham Park, New Jersey, U.S.A. N.m.r. spectra were recorded with Varian EM-390 and Varian XL-100 instruments, ^{1}H-n.m.r. (100 MHz) and ^{13}C-n.m.r. spectra (25.2 MHz) being obtained by use of the Fourier-transform (F.t.) mode, and the positions of the peaks expressed in δ from the signal for tetramethylsilane.

*Benzyl 2-acetamido-3-*O*-allyl-4,6-*O*-benzylidene-2-deoxy-α-*D*-galactopyranoside* (**3**). — A solution of compound **2** (3.0 g, 7.5 mmol) in *N*,*N*-dimethylformamide (50 mL) was stirred for 1 h at room temperature in the presence of barium oxide (5 g), barium hydroxide octahydrate (1.5 g), and allyl bromide (1.25 mL). The resulting, crystalline mass was then poured into cold, 20% acetic acid (100 mL) with stirring; stirring was continued for 15 min, and the white precipitate was filtered off, washed several times with cold water, and recrystallized from hot methanol, to give **3** (2.8 g) in 85% yield; m.p. 250–252°, $[\alpha]_D$ +176.1° (*c* 1.1, Me_2SO); t.l.c. (5:1 chloroform–acetone): R_F 0.8; ν_{max}^{KBr} 3300 (NH), 1650 (amide), and 700 cm^{-1} (aromatic).

Anal. Calc. for $C_{25}H_{29}NO_6$: C, 68.32; H, 6.65; N, 3.19. Found: C, 68.14; H, 6.80; N, 3.04.

*Benzyl 2-acetamido-3-*O*-allyl-2-deoxy-α-*D*-galactopyranoside* (**5**). — A suspension of **3** (2 g) in 60% acetic acid (100 mL) was stirred for 50 min at 100°. Evaporation, followed by several additions and evaporations of water, and then dry toluene, gave a solid mass which was recrystallized from methanol–ether, to give **5** in 81% yield (1.3 g); m.p. 180–181°, $[\alpha]_D$ +182.5° (*c* 0.6, Me_2SO); t.l.c. (9:1 chloroform–methanol): R_F 0.64; ν_{max}^{KBr} 3400 (OH), 3300 (NH), 1645 (amide), 730, and 695 cm^{-1} (aromatic); ^{1}H-n.m.r. data (Me_2SO-d_6): δ 1.84 (s, 3 H, NAc), 4.76 (d, 1 H, $J_{1,2}$ 4 Hz, H-1), 5.06–5.40 (m, 2 H, $=CH_2$), 5.77–6.15 (m, 1 H, –CH=), 7.40 (m, 5 H, aromatic), and 7.84 (d, 1 H, $J_{NH,2}$ 9 Hz, NH).

Anal. Calc. for $C_{18}H_{25}NO_6$: C, 61.52; H, 7.17; N, 3.99. Found: C, 61.50; H, 7.34; N, 3.86.

*Benzyl 2-acetamido-3-*O*-allyl-4,6-di-*O*-benzyl-2-deoxy-α-*D*-galactopyranoside* (**6**). — A solution of **5** (1.3 g, 3.7 mmol) in *N*,*N*-dimethylformamide (40 mL) was stirred for 2 days at room temperature in the presence of barium oxide (2.1 g), barium hydroxide octahydrate (0.75 g), and benzyl bromide (2.3 mL, 4 equiv.). After dilution with chloroform (150 mL), 50% acetic acid (50 mL) was added with stirring; stirring was continued for 15 min, and the chloroform layer was separated, washed successively with water, saturated aqueous sodium hydrogencarbonate, and water, dried (potassium carbonate), and evaporated to dryness. The residue crystallized from ethyl acetate–ether–hexane, to afford **6** in 76% yield (1.5 g); m.p. 155–156°, $[\alpha]_D$ +94.7° (*c* 1.3, chloroform); t.l.c. (9:1 chloroform–acetone): R_F 0.75; the i.r. spectrum showed the complete absence of hydroxyl group; ^{1}H-n.m.r. data (Me_2SO-d_6): δ 1.86 (s, 3 H, NAc), 4.78 (d, 1 H, $J_{1,2}$ 4 Hz,

H-1), 5.08–5.40 (m, 2 H, $=CH_2$), 5.80–6.16 (m, 1 H, –CH=), and 7.30–7.45 (m, 15 H, aromatic).

Anal. Calc. for $C_{32}H_{37}NO_6$: C, 72.29; H, 7.02; N, 2.64. Found: C, 72.23; H, 7.31; N, 2.61.

*Benzyl 2-acetamido-6-O-benzyl-2-deoxy-α-*D*-galactopyranoside* (**8**). — A mixture of **2** (1.6 g, 4 mmol) and sodium cyanoborohydride (2.262 g, 36 mmol) in dry oxolane (60 mL) containing 3A molecular sieves (8 g), was cooled to 0°. Hydrogen chloride in diethyl ether was added until the solution was acidic (pH paper, gas evolution). After 30 min at 0°, when t.l.c. (3:2 chloroform–acetone) indicated complete reaction, the mixture was poured into ice–water, and extracted with dichloromethane (5 × 40 mL). The combined extracts were successively washed with water, saturated aqueous sodium hydrogencarbonate, and water, dried (anhydrous magnesium sulfate), and evaporated. The solid mass was purified by chromatography on a column of silica gel, with elution with 3:2 chloroform–acetone, to give **8** in 56% yield (900 mg), amorphous; $[\alpha]_D$ +145.1° (*c* 1.2, Me_2SO); ^{1}H-n.m.r. data (Me_2SO-d_6): δ 1.93 (s, 3 H, NAc), 4.90 (d, 1 H, $J_{1,2}$ 4 Hz, H-1), and 7.33 (m, 10 H, aromatic).

Anal. Calc. for $C_{22}H_{27}NO_6$: C, 65.82; H, 6.78; N, 3.49. Found: C, 65.81; H, 6.86; N, 3.29.

*Benzyl 2-acetamido-3,6-di-O-benzyl-2-deoxy-α-*D*-galactopyranoside* (**9**). — Compound **4** (2.445 g, 5 mmol) was transformed into **9** as described for the preparation of **8**. The residue was purified by chromatography on a column of silica gel, eluting first with chloroform, then with 9:1 chloroform–acetone, and finally with 5:1 chloroform–acetone, to give **9** in 77% yield (1.9 g), amorphous; $[\alpha]_D$ +121.6° (*c* 1.4, chloroform), lit.[3] $[\alpha]_D$ +121.5° (*c* 1, chloroform); for ^{13}C-n.m.r. data, see Table I.

*Benzyl 2-acetamido-4,6-di-O-benzyl-2-deoxy-α-*D*-galactopyranoside* (**10**). — A solution of **6** (1.062 g, 2 mmol) and potassium *tert*-butoxide (1.48 g) in dimethyl sulfoxide (40 mL) was stirred for 3 h at 100° under a nitrogen atmosphere. After being cooled, the mixture was poured into ice–water (100 mL), extracted with chloroform (4 × 50 mL), and the extract washed with water, dried (anhydrous sodium sulfate), and evaporated to dryness. The colored, oily residue in 9:1 (v/v) acetone–water (40 mL) was stirred with yellow mercuric oxide (1 g), a solution of mercuric chloride (900 mg) in 9:1 (v/v) acetone–water (10 mL) was added dropwise, and the mixture was stirred for 3 h, the suspension filtered, and the filtrate evaporated to dryness. The residue was dissolved in chloroform (200 mL), and the solution was successively washed with 10% aqueous potassium iodide and water, dried (anhydrous sodium sulfate), and evaporated to dryness. The residue was purified by chromatography on a column of silica gel, with elution with 9:1 (v/v) chloroform–acetone, to afford crystalline **10** (750 mg, 76%); m.p. 176–177° (from acetone–ether–hexane), $[\alpha]_D$ +81.3° (*c* 0.4, Me_2SO); ^{1}H-n.m.r. data ($CDCl_3$): δ 1.98 (s, 3 H, NAc), 4.96 (d, 1 H, $J_{1,2}$ 4 Hz, H-1), 5.80 (d, 1 H, $J_{NH,2}$ 9 Hz, NH), and 7.30–7.50 (m, 15 H, aromatic).

Anal. Calc. for $C_{29}H_{33}NO_6$: C, 70.85; H, 6.77; N, 2.85. Found: C, 70.75; H, 6.81; N, 2.84.

Benzyl 2-acetamido-4,6-O-benzylidene-2-deoxy-3-O-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-α-D-galactopyranoside (**12**). — A solution of **2** (3.45 g, 8.65 mmol) in 1:1 (v/v) benzene–nitromethane (220 mL) was boiled until 60 mL of the solvent had been distilled. The temperature of the solution was adjusted to 40°, and mercuric cyanide (1.88 g) and a solution of bromide **11** (3.55 g, 8.65 mmol) in 1:1 (v/v) benzene–nitromethane (40 mL) were added. The mixture was stirred for 24 h at room temperature, mercuric cyanide (1.2 g) and bromide **11** (1.78 g, 4.33 mmol) were added, and the suspension was stirred for an additional 24 h. The solids were removed by filtration through a Celite pad, and washed with benzene (200 mL). The filtrate and washings were combined, successively washed twice with aqueous potassium iodide solution, saturated aqueous sodium hydrogencarbonate solution, and water, dried (anhydrous sodium sulfate), and evaporated. The residual syrup was purified by chromatography on a column of silica gel, with elution with 6:1 (v/v) chloroform–acetone, to give amorphous **12** in 81% yield (5.12 g); $[\alpha]_D$ +103.8° (*c* 1.6, Me_2SO); ν_{max}^{KBr} 3300 (NH), 1745 (OAc), 1655 (Amide I), 1525 (Amide II), 1220 (OAc), and 700 cm^{-1} (Ph); ^{1}H-n.m.r. data ($CDCl_3$): δ 1.90, 1.95, 2.0, 2.10 (s each, 15 H, 4 AcO + 1 NAc), 5.50 (s, 1 H, benzylic H), and 7.3–7.6 (m, 10 H, aromatic).

Anal. Calc. for $C_{36}H_{44}NO_{15}$: C, 59.17; H, 6.07; N, 1.92. Found: C, 59.17; H, 6.09; N, 1.91.

Benzyl 2-acetamido-6-O-benzyl-2-deoxy-3-O-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-α-D-galactopyranoside (**13**). — Compound **13** was prepared from **12** (1.46 g, 2 mmol) as described for **8**. The product was purified by chromatography on a column of silica gel, with elution with 6:1 (v/v) chloroform–acetone, to give **13** (920 mg, 63%); m.p. 150–152° (ethyl acetate–ether–hexane), $[\alpha]_D$ +66° (*c* 1.4, chloroform); t.l.c. in 3:1 chloroform–acetone: R_F 0.41; ^{1}H-n.m.r. data ($CDCl_3$): δ 1.90, 1.93, 2.0, 2.10 (4 s, 15 H, 4 AcO + NAc) and 7.20–7.40 (m, 10 H, aromatic); ^{13}C-n.m.r. data ($CDCl_3$): δ 20.47, 20.59 (OAc), 23.31 (NAc), 47.78 (C-2), 61.21 (C-6′), 78.02 (C-3), 97.18 (C-1), and 101.47 (C-1′).

Anal. Calc. for $C_{36}H_{45}NO_{15}$: C, 59.09; H, 6.20; N, 1.91. Found: C, 59.16; H, 5.93: N, 2.05.

Benzyl 2-acetamido-6-O-benzyl-2-deoxy-3-O-β-D-galactopyranosyl-α-D-galactopyranoside (**14**). — A solution of **13** (150 mg) in dry methanol (15 mL) was stirred overnight in the presence of a catalytic amount of the macroreticular[13] Amberlyst A-26 (OH^-). The resin was removed by filtration, and the filtrate was evaporated, to give a solid mass that was recrystallized from methanol–ether, to afford **14** in 87% yield (100 mg); m.p. 229–230°, $[\alpha]_D$ +102.5° (*c* 1.5, Me_2SO); the i.r. spectrum showed the complete absence of ester group.

Anal. Calc. for $C_{28}H_{37}NO_{11}$: C, 59.67; H, 6.62; N, 2.49. Found: C, 59.59; H, 6.36; N, 2.56.

Benzyl 2-acetamido-6-O-benzyl-2-deoxy-3,4-di-O-(2,3,4,6-tetra-O-acetyl-β-

D-*galactopyranoside* (**15**). — *Method a*. To a stirred solution of **13** (500 mg, 0.68 mmol) and bromide **11** (575 mg, 1.4 mmol) in dry dichloromethane (20 mL) was added 1,1,3,3-tetramethylurea (0.4 mL dissolved in 20 mL of dichloromethane). The flask was then wrapped in aluminum foil, silver triflate (0.36 g) was added, and stirring was continued for 3 days at room temperature. The suspension was filtered through a Celite pad, and the filtrate was successively washed with a saturated solution of sodium hydrogencarbonate and water, dried (anhydrous sodium sulfate), and evaporated. The syrupy product was purified by chromatography on a column of silica gel, with elution with 9:1 (v/v) chloroform–acetone, to give **15** (500 mg, 69%); m.p. 194–195° (chloroform–ether–hexane), $[\alpha]_D$ +33.6° (*c* 0.6, chloroform); t.l.c. (4:1 chloroform–acetone): R_F 0.36; ^{1}H-n.m.r. data ($CDCl_3$): δ 1.90–2.20 (cluster of singlets, 27 H, 8 AcO + NAc) and 7.2–7.3 (m, 10 H, aromatic). + NAc) and 7.2–7.3 (m, 10 H, aromatic).

Anal. Calc. for $C_{50}H_{63}NO_{24}$: C, 56.54; H, 5.98; N, 1.32. Found: C, 56.79; H, 5.73; N, 1.36.

Method b. In another experiment, diol **8** (401 mg, 1 mmol) was glycosylated exactly as described in (*a*), using the appropiate quantities of reagents, and the reaction was monitored by t.l.c. in 4:1 chloroform–acetone. After 3 days, t.l.c. showed a major spot corresponding to compound **13** and a very minor spot corresponding to compound **15**, but also a large amount of unchanged starting-material (**8**). More bromide **11** (300 mg) and silver triflate (150 mg) were added and stirring was continued for an additional 5 days. T.l.c. then showed that compound **13** was no longer present, but that large amounts of diol **8** still remained. After a total of 8 days, the mixture was processed, and the product purified as described in (*a*), to give, in 21% yield (220 mg), a pure compound that was identical to compound **15** on the basis of spectral data.

*Benzyl 2-acetamido-6-O-benzyl-2-deoxy-3,4-di-O-β-*D*-galactopyranosyl-α-*D*-galactopyranoside* (**16**). — *O*-Deacetylation of compound **15** (300 mg) as described for **14** gave amorphous **16** (180 mg, 88%); $[\alpha]_D$ +76.9° (*c* 1.1, methanol); the i.r. spectrum showed the complete absence of *O*-acetyl group; ^{1}H-n.m.r. data (MeOH-d_4): δ 1.92 (s, 3 H, NAc), 4.87 (d, 1 H, $J_{1,2}$ 4 Hz, H-1), and 7.27 (m, 10 H, aromatic).

Anal. Calc. for $C_{34}H_{47}NO_{16} \cdot H_2O$: C, 54.90; H, 6.64; N, 1.88. Found: C, 54.83; H, 6.71; N, 1.80.

*2-Acetamido-2-deoxy-3,4-di-O-β-*D*-galactopyranosyl-*D*-galactopyranose* (**17**). — A solution of **16** (150 mg) in glacial acetic acid (30 mL) was hydrogenolyzed in the presence of 10% Pd–C (150 mg) for 2 days, the suspension filtered, the filtrate evaporated, and the residue purified by chromatography on a column of silica gel, with elution with 11:9:2 (v/v/v) chloroform–methanol–water, to give amorphous **17** (95 mg, 84%); $[\alpha]_D$ +34.3° (*c* 1.2, water); t.l.c. in 11:9:2 chloroform–methanol–water: R_F 0.3. The purity of compound **17** was established by paper chromatography on Whatman No. 1 paper with 3.2:1 (v/v) butyl acetate–acetic acid–water, R_{Gal} 0.41 (silver nitrate reagent[14]); for ^{13}C-n.m.r. data, see Table II.

Anal. Calc. for $C_{20}H_{35}NO_{16} \cdot H_2O$: C, 42.63; H, 6.62; N, 2.49. Found: C, 42.39; H, 6.45; N, 2.39.

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