Synthesis of Blood-group Substances. Part 11.[1] Synthesis of the Trisaccharide *O*-α-D-Galactopyranosyl-(1 → 3)-*O*-β-D-galactopyranosyl-(1 → 4)-2-acetamido-2-deoxy-D-glucopyranose

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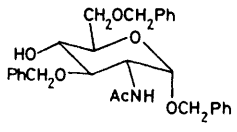
Reaction of 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide with benzyl 2-acetamido-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside in 1,2-dichloroethane in the presence of mercuric bromide and molecular sieves (4 Å) provided after *O*-deacetylation crystalline benzyl 2-acetamido-3,6-di-*O*-benzyl-2-deoxy-4-*O*-(β-D-galactopyranosyl)-α-D-glucopyranoside. Acetonation followed by benzylation and mild acid hydrolysis provided crystalline benzyl 2-acetamido-4-*O*-(2,6-di-*O*-benzyl-β-D-galactopyranosyl)-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside. Reaction of this diol with 1-*O*-(*N*-methyl)acetimidyl-2,3,4,6-tetra-*O*-benzyl-β-D-galactopyranose in nitromethane in the presence of toluene-*p*-sulphonic acid regiospecifically gave the derivative (15). The title trisaccharide was obtained after catalytic hydrogenolysis. The alcohol benzyl 2-acetamido-4-*O*-(2,3,6-tri-*O*-benzyl-β-D-galactopyranosyl)-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside was found to be unreactive under the same conditions.

EXAMINATION of the serologically active fragments isolated from human blood-group active glycoproteins reveals that there are two types of carbohydrate chain endings: type 1 chain, β-Gal-(1→3)-GNAc; and type 2 chain, β-Gal-(1→4)-GNAc; these form the basis of the blood-group active structures.[2] The A, B, and H determinants can be based on either the type 1 or type 2 chains. In contrast, all variants of glycosphingolipid antigens found in human erythrocytes contain only type 2 chain.[3] With a convenient synthesis of *N*-acetyllactosamine in hand[4] (particularly suitable for the synthesis of selectively protected derivatives) we started a few years ago a programme aimed at the chemical synthesis of the type 2 blood-group antigenic determinants. *O*-α-L-Fucopyranosyl-(1→2)-*O*-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-D-glucopyranose is the H-type 2 antigenic determinant and was synthesized by us in 1976.[5] The corresponding H-type 1 trisaccharide, in which *N*-acetyl-D-glucosamine is substituted by D-galactose at position 3, was prepared by Kochetkov *et al.*[6] More recently, the 'imidate procedure'[7] enabled a synthesis of the human type 2 tetrasaccharide which is the blood-group B antigenic determinant.[8] A synthesis of the homologous type 1 tetrasaccharide has also been reported by Paulsen *et al.*[9]

Painter *et al* in 1963 isolated a type 2 active trisaccharide from the products of partial acid hydrolysis of a human blood-group B specific glycoprotein.[10] This trisaccharide was believed to the *O*-α-D-galactopyranosyl-(1→3)-*O*-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-D-glucopyranose. It is also the terminal block of α-galactosyl-paragloboside.[11] This paper describes a synthetic approach to this trisaccharide. The protected *N*-acetyl-lactosamine derivative (2) has been previously prepared[12] in 75% yield when benzyl 2-acetamido-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside[12] (1) was condensed with 3,4,6-tri-*O*-acetyl-α-D-galactopyranose-1,2-(t-butyl orthoacetate). This synthesis has now been simplified in our laboratory. It was found that a satisfactory reaction was achieved when the alcohol (1) was condensed with the easily available 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide in 1,2-dichloroethane in the presence of mercuric bromide and molecular sieves (4 Å).[4] For purification purposes



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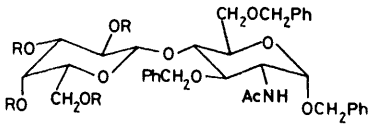
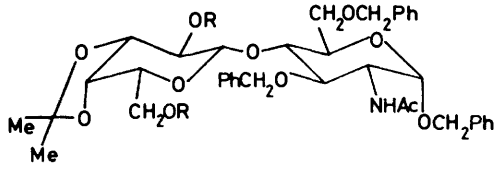


(2) R = Ac
(3) R = H

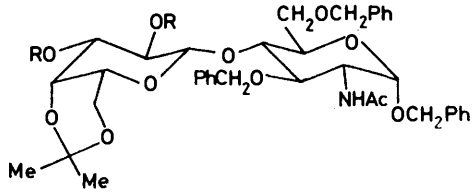
the reaction mixture was *O*-deacetylated and chromatographed on silica gel to obtain the amorphous β-linked protected disaccharide (3) (87%) which could not be crystallised easily at this step. Acetylation of this compound gave nicely crystalline benzyl 2-acetamido-3,6-di-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-2-deoxy-α-D-glucopyranoside (2). *O*-Deacetylation of (2) provided crystalline benzyl 2-acetamido-3,6-di-*O*-benzyl-4-*O*-(β-D-galactopyranosyl)-2-deoxy-α-D-glucopyranoside (3). Furthermore, the alcohol (1) is now prepared by a new and very high yielding sequence.[13] A combination of these improvements, together with the catalytic hydrogenolysis of (3) previously described,[12] results in a fairly efficient route to this major disaccharide, which compares well with other syntheses.[14]

Acetonation of the disaccharide (3) was achieved in excellent yield with acetone in the presence of toluene-*p*-sulphonic acid. The mixture of acetals (4) and (6), which is inseparable on silica gel t.l.c. at this stage, was benzylated with benzyl bromide in *NN*-dimethylformamide in the presence of barium oxide and barium hydroxide octahydrate to give two derivatives (5) and (7), easily separable on silica gel. The major and faster migrating product was the wanted benzyl 2-acetamido-4-

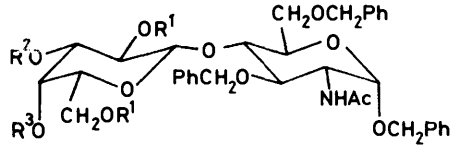
O-(2,6-di-*O*-benzyl-3,4-*O*-isopropylidene-β-D-galactopyranosyl)-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside (5) and the by-product was benzyl 2-acetamido-4-*O*-(2,3-di-*O*-benzyl-4,6-*O*-isopropylidene-β-D-galactopyranosyl)-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside (7). Under such conditions, the thermodynamic product (5) is to be expected.[15] Indeed, when acetonation of (3) was performed in *NN*-dimethylformamide with 2,2-dimethoxypropane containing a trace of toluene-*p*-sulphonic acid,[16] the kinetically controlled product (7) was isolated in good yield after the aforementioned benzylation. Mild acidic treatment of (5) converted it



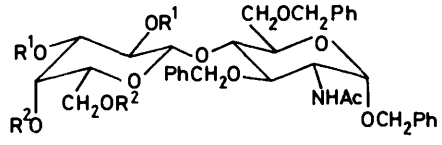
(4) R = H
(5) R = CH_2Ph



(6) R = H
(7) R = CH_2Ph



(8) $R^1 = CH_2Ph$, $R^2 = R^3 = H$
(9) $R^1 = CH_2Ph$, $R^2 = R^3 = Ac$
(10) $R^1 = R^2 = CH_2Ph$, $R^3 = H$
(11) $R^1 = R^2 = CH_2Ph$, $R^3 = Ac$



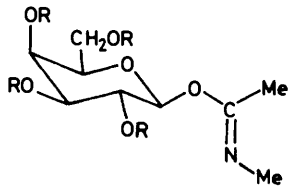
(12) $R^1 = CH_2Ph$, $R^2 = H$
(13) $R^1 = CH_2Ph$, $R^2 = Ac$

into crystalline benzyl 2-acetamido-4-*O*-(2,6-di-*O*-benzyl-β-D-galactopyranosyl)-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside (8). Acetylation of (8) provided crystalline acetate (9). Similar reactions were performed on acetal (7) to give the crystalline alcohol (12) and the amorphous acetate (13), respectively. Examination of the ^1H n.m.r. spectra of compounds (9) and (13) unambiguously indicates the proposed structures. In acetate (9), both protons H-3′ and H-4′ are deshielded as expected [H-3′, δ 4.85; and H-4′, δ 5.38; $J_{3'4'}$ 4 and $J_{4'5'}$ 1 Hz], whereas in acetate (13), only H-4′ is deshielded (δ 5.39, $J_{3'4'}$ 3 and $J_{4'5'}$ 1 Hz), H-3′ being shielded (δ 3.40).

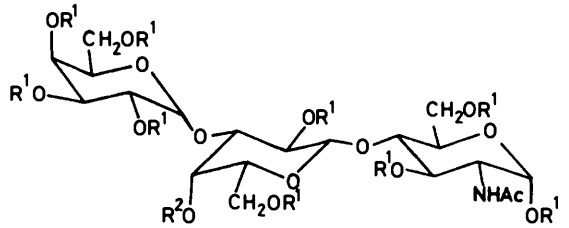
Treatment of the diol (8) in anhydrous nitromethane at room temperature for 24 h with an excess of 1-*O*-(*N*-methyl)acetimidyl-2,3,4,6-tetra-*O*-benzyl-β-D-galactopyranose[17] (14) in the presence of toluene-*p*-sulphonic acid gave, after chromatography, 74% of the crystalline trisaccharide (15). No other isomer was isolated from the reaction mixture. Acetylation of (15) provided the amorphous monoacetate (16). Examination of the ^1H n.m.r. spectrum of (16) demonstrates the proposed structure unambiguously: the proton H-4′ appears as a deshielded doublet of doublets (δ 5.54, $J_{3'4'}$ 3 and $J_{4'5'}$ 1 Hz), H-3′ being shielded as expected for a glycosylated position (δ 3.70). On the other hand, H-1″ appears as a low-field doublet with a small coupling constant (δ 5.32, $J_{1''2''}$ 2.5 Hz).

The explanation of the regiospecificity of the galactosidation reaction is in our opinion not straightforward, because it has been shown [18,19] that the axial orientation of the 4-hydroxy-group of galactopyranosides is no hindrance to α-disaccharide synthesis. In order to shed some light on this matter, we decided to prepare the disaccharide (10), where only the 4-hydroxy-group is available for galactosidation. It is now well established [20,21] that treatment of stannylene derivatives of axial-equatorial, vicinal diols in the galactopyranose series gives almost exclusively monosubstitution products, with very high regioselectivity for reaction at the equatorial oxygen. Thus, reaction of the 3,4-*O*-dibutylstannylene derivative of (8) with benzyl bromide in *NN*-dimethylformamide gave 75% of the crystalline benzyl 2-acetamido-4-*O*-(2,3,6-tri-*O*-benzyl-β-D-galactopyranosyl)-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside (10). The structure of this compound was ascertained after *O*-acetylation, whereupon H-4′ becomes typically deshielded (δ 5.55), H-3′ appearing at a much higher field (δ 3.47). No trace of condensation product could be isolated when the imidate (14) was partnered with the alcohol (10), in conditions where this imidate smoothly reacts with benzyl 2,3,6-tri-*O*-benzyl-β-D-galactopyranoside or methyl 2,3,6-tri-*O*-benzyl-α-D-galactopyranoside to give an α-linked disaccharide.[18] It thus appears that the axial hydroxy-group of *N*-acetyl-lactosamine is much

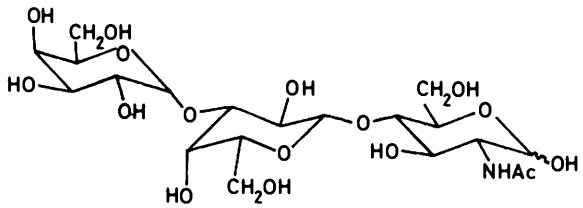
less reactive than the axial hydroxy-group of the isolated galactopyranose moiety. It must be emphasized in this respect that we have to date been able to glycosylate every equatorial hydroxy-position of suitably protected *N*-acetyl-lactosamine derivatives.[5,22]



(14) R = CH_2Ph



(15) R^1 = CH_2Ph, R^2 = H
(16) R^1 = CH_2Ph, R^2 = Ac



(17)

Product (15) was debenzylated to the title trisaccharide by hydrogenolysis in acetic acid in the presence of palladium–carbon. M.p. and optical rotation are identical with those values reported by Painter *et al.*[10]

EXPERIMENTAL

Melting points were determined on a Büchi apparatus. Specific optical rotations were measured at 22—24 °C with a Perkin-Elmer model 141 polarimeter. I.r. spectra were recorded with a Jouan-Jasco IRA-1 spectrometer. ^1H N.m.r. spectra were obtained for solutions in deuteriochloroform (tetramethylsilane as internal standard) unless otherwise stated; indices refer respectively to: primary, glucosamine; secondary, galactose; tertiary, external galactose in trisaccharide derivatives. Gas–liquid chromatography (g.l.c.) of the per-*O*-(trimethylsilyl) derivatives was performed with a Girdel 3000 apparatus provided with a flame-ionization detector and a 3.40-m Pyrex column (4% OV 17 on Gas-Chrom Q, 80—100 mesh), programmed for a rise of 10 °C min^{-1} from 150 to 300 °C; R_t is given relative to that of per-*O*-(trimethylsilyl)myoinositol. Purity of products was determined by thin layer chromatography {t.l.c. on silica gel 60 F 254 (E. Merck)}. Components were located by spraying with sulphuric acid in ethanol (50% solution) and charring. Colum chromatography was performed on silica gel Merck 60 (powder, 0.063—0.200 mm) which was used without pre-treatment. Elemental analyses were obtained from the Service Central de Microanalyse du Centre National de la Recherche Scientifique.

Benzyl 2-Acetamido-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (2).—A solution of benzyl 2-acetamido-3,6-di-*O*-benzyl-2-deoxy-α-D-glucopyranoside [13] (1) (5.05 g) in 1,2-dichloroethane (150 ml) containing anhydrous powdered mercuric bromide (1.8 g) and activated powdered molecular sieves (4 Å) (10 g) was heated under a dry atmosphere of nitrogen. After solvent (30 ml) had been distilled off a freshly prepared solution of 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide (9.6 g) in 1,2-dichloroethane (50 ml) was added dropwise and more solvent (50 ml) was distilled off. The mixture was stirred at 80 °C during 36 h. After cooling, the reaction mixture was filtered, washed with 10% aqueous potassium iodide and water, and then dried ($CaCl_2$) and evaporated. The residue was *O*-deacetylated (sodium methoxide in methanol) and chromatographed on silica gel (200 g); elution with chloroform–methanol (9 : 1) gave the amorphous disaccharide (3) (5.22 g, 77.8%). Acetylation (acetic anhydride–pyridine) gave the disaccharide (2) (5.22 g, 95%), m.p. 109—110.5 °C, $[\alpha]_D$ +66.5° (*c* 1 in $CHCl_3$) {lit.,[12] m.p. 110—111 °C, $[\alpha]_D$ +66° (*c* 1 in $CHCl_3$)}.

Benzyl 2-Acetamido-4-O-(β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (3).—The disaccharide (2) (5.28 g) was *O*-deacetylated (sodium methoxide in methanol) to give *compound* (3) [4.7 g, 70% from (1)], m.p. 154—155 °C (from methanol–water), $[\alpha]_D$ +86° (*c* 1 in MeOH) (Found: C, 63.3; H, 6.6; N, 2.0. $C_{35}H_{43}NO_{11}\cdot H_2O$ requires C, 63.1; H, 6.7; N, 2.0%).

Benzyl 2-Acetamido-4-O-(2,6-di-O-benzyl-3,4-O-isopropylidene-β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-D-galactopyranoside (5).—A suspension of disaccharide (3) (500 mg) was stirred for 3 h at room temperature in dry acetone (25 ml) in the presence of toluene-*p*-sulphonic acid (50 mg). The mixture was then stirred during 15 min with an aqueous saturated sodium hydrogencarbonate solution and evaporated. The solid residue was extracted with chloroform (100 ml). The chloroform extract was washed with aqueous saturated sodium hydrogencarbonate and water, dried (Na_2SO_4), and evaporated. A portion (400 mg) of the crude residue of (4) and (6) (506 mg, 95%) was dissolved in *NN*-dimethylformamide (20 ml) and benzylated with benzyl bromide (0.56 ml) at room temperature during 36 h in the presence of barium oxide (1.36 g) and barium hydroxide octahydrate (0.35 g). After destruction of the excess of benzyl bromide with methanol (5 ml), the reaction mixture was diluted with chloroform (100 ml), washed with ice-cold 50% aqueous acetic acid, water, aqueous saturated sodium hydrogencarbonate and water, dried (Na_2SO_4), and evaporated. The residue was chromatographed on silica gel (40 g); elution with ethyl acetate–hexane (2 : 1) gave two compounds. The first compound eluted was the *acetal* (5) (381 mg, 76%), m.p. 151 °C (from ethyl acetate–hexane), $[\alpha]_D$ +78° (*c* 1 in $CHCl_3$); δ 1.32 and 1.40 (6 H, 2s, Me), 1.79

(3 H, s, Ac), 4.97 (1 H, d, J 4 Hz, H-1), 5.32 (1 H, d, J 8.5 Hz, NH), and 7.31 (25 H, s, Ph) (Found: C, 71.9; H, 6.7; N, 1.5. $C_{52}H_{59}NO_{11}$ requires C, 71.5; H, 6.8; N, 1.6%).

Benzyl 2-Acetamido-4-O-(2,3-di-O-benzyl-4,6-O-isopropylidene-β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (7).—The second compound eluted during the aforementioned chromatography was the *acetal* (7) (87 mg, 17%), $[\alpha]_D$ +74.5° (*c* 1 in $CHCl_3$); δ 1.40 (6 H, s, Me), 1.78 (3 H, s, Ac), 2.91 (1 H, br s, H-5'), 3.32 (1 H, dd, $J_{2'3'}$ 10, $J_{3'4'}$ 4 Hz, H-3'), 4.98 (1 H, d, J_{12} 4 Hz, H-1), 5.30 (1 H, d, J 9.5 Hz, NH), and 7.23—7.35 (25 H, 3s, Ph) (Found: C, 71.1; H, 6.7; N, 1.4. $C_{52}H_{59}NO_{11}$ requires C, 71.5; H, 6.8; N, 1.6%).

This acetal was selectively formed under kinetic conditions: a solution of the disaccharide (3) (1.291 g) in *NN*-dimethylformamide (20 ml) containing 2,2-dimethoxypropane (0.367 ml) and toluene-*p*-sulphonic acid (10 mg) was left at room temperature during 1.5 h. Addition of triethylamine (1 ml) was followed by evaporation. The residue was taken up in chloroform (100 ml), washed with aqueous saturated sodium hydrogencarbonate and water, dried (Na_2SO_4), and evaporated. A solution of the dried residue (1.32 g, 96%) in *NN*-dimethylformamide (20 ml) was stirred at room temperature during 4 days in the presence of benzyl bromide (0.9 ml), barium oxide (2.72 g), and barium hydroxide octahydrate (0.7 g). The work-up described for the synthesis of (5) gave a residue which was chromatographed on silica gel (50 g); elution with ethyl acetate–hexane (3 : 1) gave the acetal (7) (1.33 g, 89%), identical to the one previously prepared.

Benzyl 2-Acetamido-4-O-(2,6-di-O-benzyl-β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (8).—A solution of acetal (5) (350 mg) in 80% aqueous acetic acid (15 ml) was heated for 30 min at 95 °C. After cooling, the mixture was evaporated to give the *disaccharide* (8) (301 mg, 90%), m.p. 164—165 °C (from ethyl acetate–hexane), $[\alpha]_D$ +80° (*c* 1 in $CHCl_3$); δ 1.78 (3 H, s, Ac), 4.96 (1 H, d, J_{12} 4 Hz, H-1), 5.33 (1 H, d, J 8.5 Hz, NH), and 7.30 (25 H, s, Ph) (Found: C, 70.3; H, 6.7; N, 1.7. $C_{49}H_{55}NO_{11}$ requires C, 70.6; H, 6.7; N, 1.7%).

Benzyl 2-Acetamido-4-O-(3,4-di-O-acetyl-2,6-di-O-benzyl-β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (9).—The disaccharide (8) (90 mg) was acetylated (acetic anhydride–pyridine) to give *compound* (9) (88 mg, 89%), m.p. 133—134 °C (from ether-hexane), $[\alpha]_D$ +53° (*c* 1 in $CHCl_3$); δ 1.82 (3 H, s, NAc), 1.92 and 1.95 (6 H, 2 s, OAc), 4.85 (1 H, dd, H-3'), 5.0 (1 H, d, J_{12} 4 Hz, H-1), 5.38 (1 H, dd, $J_{3'4'}$ 4, $J_{4'5'}$ 1 Hz, H-4'), 5.41 (1 H, d, J 9 Hz, NH), and 7.30 (25 H, s, Ph) (Found: C, 69.6; H, 6.4; N, 1.5. $C_{53}H_{59}NO_{13}$ requires C, 69.3; H, 6.5; N, 1.5%).

Benzyl 2-Acetamido-4-O-(2,3-di-O-benzyl-β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-glucopyranoside (12).—A solution of acetal (7) (180 mg) in 80% aqueous acetic acid (10 ml) was heated for 20 min at 95 °C. After cooling, the mixture was evaporated to give the *disaccharide* (12) (147 mg, 86%), m.p. 182—183 °C (methanol–hexane), $[\alpha]_D$ 75.5° (*c* 1 in $CHCl_3$); δ 1.81 (3 H, s, Ac), 2.95 (2 H, s, OH), 3.32 (1 H, dd, $J_{2'3'}$ 10, $J_{3'4'}$ 4 Hz, H-3'), 3.96 (1 H, dd, $J_{4'5'}$ 1 Hz, H-4'), 4.96 (1 H, d, J_{12} 4 Hz, H-1), 5.36 (1 H, d, J 9 Hz, NH), and 7.28—7.40 (25 H, m, Ph) (Found: C, 70.6; H, 6.7; N, 1.7. $C_{49}H_{55}NO_{11}$ requires C, 70.6; H, 6.7; N, 1.7%).

Benzyl 2-Acetamido-4-O-(4,6-di-O-acetyl-2,3-di-O-benzyl-β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (13).—The disaccharide (12) (100 mg) was acetylated (acetic anhydride–pyridine) to give *compound* (13) (100 mg, 91%), $[\alpha]_D$ +78° (*c* 1 in $CHCl_3$); δ 1.78 (3 H, s, NAc), 1.96 and 2.03 (6 H, 2 s, OAc), 3.40 (2 H, m, H-3' and H-5'), 4.96 (1 H, d, J_{12} 4 Hz, H-1), 5.32 (1 H, d, J 9 Hz, NH), 5.39 (1 H, dd, $J_{3'4'}$ 3 $J_{4'5'}$ 1 Hz, H-4'), and 7.25—7.35 (25 H, m, Ph) (Found: C, 68.3; H, 6.7; N, 1.7. $C_{53}H_{59}NO_{13}\cdot H_2O$ requires C, 67.9; H, 6.5; N, 1.5%).

Benzyl 2-Acetamido-4-O-{2,6-di-O-benzyl-3-O-(2,3,4,6-tetra-O-benzyl-α-D-galactopyranosyl)-β-D-galactopyranosyl}-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (15).—A solution of disaccharide (8) (278 mg) and 1-*O*-(*N*-methyl)-acetimidyl-2,3,4,6-tetra-*O*-benzyl-β-D-galactopyranose (14) (400 mg) in anhydrous nitromethane (10 ml) was stirred at room temperature under nitrogen for 3 h with activated powdered molecular sieves (4 Å) (500 mg). A solution of anhydrous toluene-*p*-sulphonic acid (86 mg) in anhydrous nitromethane (2 ml) was then added and the stirring was resumed for 21 h. The mixture was diluted with chloroform (10 ml), filtered, washed with aqueous saturated sodium hydrogencarbonate and water, dried (Na_2SO_4), and evaporated. The residue was chromatographed on silica gel (50 g); elution with ethyl acetate–hexane (3 : 2) gave first the *trisaccharide* (15) (335 mg, 71%), m.p. 122—123 °C (from ether–hexane), $[\alpha]_D$ +63° (*c* 1 in $CHCl_3$); δ 1.79 (3 H, Ac), 4.96 (1 H, d, J_{12} 4 Hz, H-1), 5.27 (1 H, d, J 9 Hz, NH), and 3.20—3.40 (45 H, m, Ph) (Found: C, 73.5; H, 6.6; N, 1.0. $C_{83}H_{89}NO_{16}$ requires C, 73.5; H, 6.6; N, 1.0%). Further elution gave a small amount of starting material (8) (14 mg, 5%).

Benzyl 2-Acetamido-4-O-{4-O-acetyl-2,6-di-O-benzyl-3-O-(2,3,4,6-tetra-O-benzyl-α-D-galactopyranosyl)-β-D-galactopyranosyl}-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (16).—The trisaccharide (15) (50 mg) was acetylated (acetic anhydride–pyridine) to give *compound* (16) (45 mg, 89%), $[\alpha]_D$ +57° (*c* 1 in $CHCl_3$); δ 1.80 and 1.82 (6 H, 2 s, Ac), 3.70 (H-3' with other signals), 4.96 (1 H, d, J_{12} 4 Hz, H-1), 5.32 (1 H, d, $J_{1'2'}$ 2.5 Hz, H-1''), 5.50 (1 H, d, J 9 Hz, NH), 5.54 (1 H, dd, $J_{3'4'}$ 3, $J_{4'5'}$ 1 Hz, H-4'), and 3.30 (45 H, s, Ph) (Found: C, 72.5; H, 6.5. $C_{85}H_{91}NO_{17}$ requires C, 72.9; H, 6.6%).

O-α-D-Galactopyranosyl-(1→3)-O-β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-D-glucopyranose (17).—Compound (15) (130 mg) in acetic acid (8 ml) was hydrogenolysed with Pd–C (10% 100 mg) for 24 h. The mixture was filtered and evaporated to give the *trisaccharide* (17) (47 mg, 96%), m.p. 236—239 °C (decomp.) (from methanol–acetone–water), $[\alpha]_D$ +104°→+100° [24 h; *c* 1 water–methanol (19 : 1)] (Found: C, 42.9; H, 6.6; N, 2.9. $C_{20}H_{35}NO_{16}\cdot H_2O$ requires 42.6; H, 6.6; N, 2.5%) {lit.,[10] m.p. 235—240 °C (decomp.), $[\alpha]_D$ +99° (eq., *c* 1.0 in water)}.

After reduction with sodium borohydride followed by per-*O*-trimethylsilylation, this compound was homogeneous on g.l.c. (R_t 3.43).

Benzyl 2-Acetamido-4-O-(2,3,6-tri-O-benzyl-β-D-galactopyranosyl)-3,6-di-O-benzyl-2-deoxy-α-D-glucopyranoside (10).—A suspension of the diol (8) (229 mg) and dibutyltin oxide (75 mg) in methanol (18 ml) was heated for 1.5 h under reflux, then the solvent was slowly distilled off after addition of benzene (10 ml). The resulting dibutylstannylene derivative was dried under vacuum, taken up in *NN*-dimethylformamide (10 ml), and treated with benzyl bromide (0.11 ml). The mixture was heated for 1.5 h at 100 °C. Following evaporation of the solvent, the residue was chromatographed on silica gel (10 g); elution with ethyl acetate–hexane (5 : 2) gave the *alcohol* (10) (198 mg, 75%), m.p. 141.5—142.5 °C (from ethyl acetate–hexane), $[\alpha]_D$ +72°

(*c* 1 in $CHCl_3$); δ 1.78 (3 H, s, Ac), 2.52 (1 H, d, OH), 3.34 (1 H, dd, $J_{2'3'}$ 10, $J_{3'4'}$ 4 Hz, H-3'), 5.00 (1 H, d, J_{12} 4 Hz, H-1), 5.33 (1 H, d, *J* 9 Hz, NH), and 7.23—7.40 (30 H, m, Ph) (Found: C, 72.8; H, 6.7; N, 1.5. $C_{56}H_{61}NO_{11}$ requires C, 72.8; H, 6.7; N, 1.5%).

In order to record a ^1H n.m.r. spectrum, the disaccharide (10) (100 mg) was acetylated (acetic anhydride–pyridine) to give *compound* (11) (75 mg, 72%); δ 1.82 (3 H, s, NAc), 2.01 (3 H, s, OAc), 3.47 (H-3'), 5.00 (1 H, d, J_{12} 4 Hz, H-1), 5.42 (1 H, d, *J* 9 Hz, NH), 5.55 (1 H, dd, $J_{3'4'}$ 3 Hz, $J_{4'5'}$ 1 Hz, H-4'), and 7.20—7.40 (30 H, m, Ph).

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