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Note

Syntheses of derivatives of *p*-aminophenyl 3-*O*-(3,6-dideoxy-α-D-*arabino*- and α-D-*ribo*-hexopyranosyl)-α-D-mannopyranosides by use of hydroxyglycal intermediates

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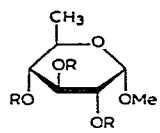
Disaccharides consisting of 3,6-dideoxyhexosyl units, with the D-*arabino*, D-*ribo*, or D-*xylo* configuration, α-(1→3)-linked to *p*-aminophenyl α-D-mannopyranoside are of interest in the diagnosis of *Salmonella* infections[1–3]. The *p*-aminophenyl unit can be joined, *via* a thiourea linkage, to an immunogenic protein carrier containing free amino groups, thereby producing an artificial antigen. In the syntheses so far described, we have condensed suitably protected 3,6-dideoxyhexosyl bromides with the mannoside **4**, which has HO-3 unsubstituted[4–6]. We now report an efficient synthesis of the disaccharide **9** (containing 3,6-dideoxy-D-*ribo*-hexose) by the hydroxyglycal procedure. This type of glycosidation, which was developed by Ferrier and co-workers[7], has previously been used[8] in the synthesis of an analogue of **9** containing methyl mannoside instead of the *p*-trifluoroacetamidophenyl mannoside unit in **9**.

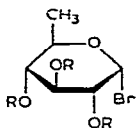
Methyl 2,3,4-tri-*O*-benzoyl-6-deoxy-α-D-glucopyranoside[9] (**1**) was converted into the bromo sugar **2** (99%) which, on elimination of hydrogen bromide, gave **3** (76%). Reaction of **3** with the mannoside **4** gave the disaccharide derivative **5** (80%). Conversion of the nitro group in **5** into an amino group by catalytic hydrogenation over Adams' catalyst was followed by *N*-trifluoroacetylation to give **6** (80%). Hydrogenation of **6** over palladium-on-carbon removed benzyl and benzylidene groups and hydrogenated the 2-enopyranose double-bond to give a mixture of **7** (60%) and **8** (18%). Debenzoylations of **7** and **8** gave the desired compounds **9** (86%) and **10** (82%), from which the amino group is easily liberated by treatment with methanolic ammonia[4–6]. The overall yield (23% for **1**→**9**) is substantially higher than that obtained by an alternative route[6].

In our initial attempts at synthesizing **9**, a similar sequence was carried out starting from methyl 2,3,4-tri-*O*-acetyl-6-deoxy-α-D-glucopyranoside[10] (**1a**). A yield of **6a** similar to that for **6** was obtained. However, catalytic hydrogenation of **6a** afforded a mixture of **7a** and **8a** in which the ratio of paratose (in **7a**) to tyvelose (in **8a**) was 0.7. This route is therefore inferior to that described above for obtaining **9**.

However, since chromatographic separation of **9** and **10** is possible, the sequence is useful for obtaining both disaccharides **9** and **10**.



1 R = Bz
1a R = Ac



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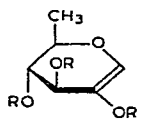
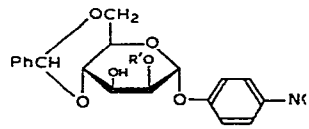
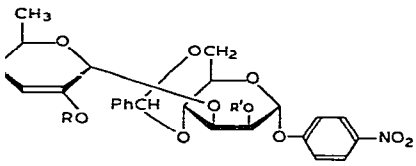
2a R = Ac



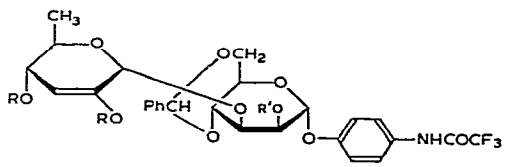
3 R = Bz
3a R = Ac



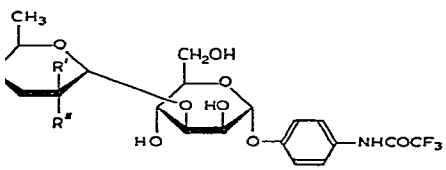
4 R' = Bzl



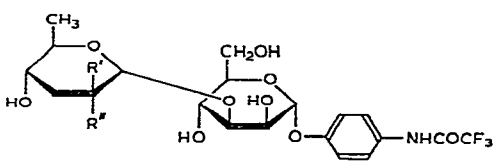
5 R = Bz, R' = Bzl
5a R = Ac, R' = Bzl



6 R = Bz, R' = Bzl
6a R = Ac, R' = Bzl



7 R = Bz, R' = H, R'' = OBz
7a R = Ac, R' = H, R'' = OAc
8 R = Bz, R' = OBz, R'' = H
8a R = Ac, R' = OAc, R'' = H



9 R' = H, R'' = OH
10 R' = OH, R'' = H

EXPERIMENTAL

General. — General methods were the same as those reported in a recent paper[6], except that ^{1}H-n.m.r. spectra were recorded with a Jeol JNM FX 100 instrument operating at 99.55 MHz in the Fourier-transform mode. ^{1}H-N.m.r. spectra were recorded for all new compounds and were in agreement with postulated structures. Only especially significant n.m.r. data are presented. The purity of syrupy new compounds, for which elemental analyses were not performed, was carefully ascertained by chromatography in solvent systems that gave R_F values of ~0.5, and the substances were rechromatographed until they were pure. Assignments of anomeric configurations are based upon optical rotations and n.m.r. data.

2,3,4-Tri-O-benzoyl-6-deoxy-α-D-glucopyranosyl bromide (**2**). — A saturated solution of hydrogen bromide in glacial acetic acid (250 ml) was added to a solution of methyl 2,3,4-tri-*O*-benzoyl-6-deoxy-α-D-glucopyranoside[9] (**1**, 9.8 g) in dichloromethane (30 ml). After the mixture had been kept at room temperature overnight,

t.l.c. (toluene–ethyl acetate, 8:1) showed the presence of one compound only, R_F 0.66. The solution was diluted with dichloromethane, washed with ice–water, cold, saturated, aqueous sodium hydrogen carbonate, and ice–water, dried ($MgSO_4$), filtered, and concentrated to yield **2** (10.7 g). When crystallized from diethyl ether, **2** had m.p. 155–161°, $[\alpha]_{578}$ +118° (*c* 1.02, chloroform). N.m.r. data ($CDCl_3$): δ 1.39 (d, 3 H, $J_{5,6}$ 6.3 Hz, H-6), 4.40–4.57 (m, 1 H, H-5), 5.29 (dd, 1 H, $J_{1,2}$ 3.9, $J_{2,3}$ 9.8 Hz, H-2), 5.46 (dd, 1 H, $J_{3,4} = J_{4,5} = 9.8$ Hz, H-4), 6.20 (t, 1 H, $J_{3,4} = J_{2,3} =$ 9.8 Hz, H-3), 6.84 (d, 1 H, $J_{1,2}$ 3.9 Hz, H-1), and 7.18–8.03 (15 H, aromatic H).

Anal. Calc. for $C_{27}H_{23}BrO_7$: C, 60.1; H, 4.3; Br, 14.8. Found: C, 60.2; H, 4.4; Br, 15.0.

*1,5-Anhydro-2,3,4-tri-*O*-benzoyl-6-deoxy-*D-arabino*-hex-1-enitol* (**3**)[17]. — A solution of **2** (10 g) and sodium iodide (5 g) in acetone (55 ml) was stirred at room temperature for 15 min, diethylamine (9 ml) was then added, and the solution was stirred at room temperature for 2 h. The solution was diluted with chloroform, washed with ice-cold solutions of 2% aqueous hydrochloric acid, saturated, aqueous sodium hydrogen carbonate, and water, dried ($MgSO_4$), filtered, concentrated, decolourized on a short column of silica gel (toluene–ethyl acetate, 8:1), and concentrated to yield **3** (6.43 g), R_F 0.63 (t.l.c., same solvent); when crystallized from ethanol, **3** had m.p. 93–95°, $[\alpha]_{578}$ −182° (*c* 1.0, chloroform). N.m.r. data ($CDCl_3$): δ 1.58 (d, 3 H, $J_{5,6}$ 6.0 Hz, H-6), 4.54 (quin, 1 H, $J_{4,5} = J_{5,6} = 6.0$ Hz, H-5), 5.54 (dd, 1 H, $J_{3,4}$ 4.4, $J_{4,5}$ 6.0 Hz, H-4), 6.08 (d, 1 H, $J_{3,4}$ 4.4 Hz, H-3), and 6.86 (s, 1 H, H-1).

Anal. Calc. for $C_{27}H_{22}O_7$: C, 70.7; H, 4.8. Found: C, 70.9, H, 4.9.

*p-Nitrophenyl 2-*O*-benzyl-4,6-*O*-benzylidene-3-*O*-(2,4-di-*O*-benzoyl-3,6-dideoxy-*α*-*D*-erythro-hex-2-enopyranosyl)-*α*-*D*-mannopyranoside* (**5**). — Boron trifluoride etherate (freshly distilled, 0.5 ml) was added to a solution of **3** (4.6 g) and *p*-nitrophenyl 2-*O*-benzyl-4,6-*O*-benzylidene-*α*-D-mannopyranoside (**4**, 4.8 g) in benzene (100 ml) with stirring at room temperature. After 15 min, excess of sodium carbonate was added and the mixture was poured into ice–water. The benzene layer was washed with saturated, aqueous sodium hydrogen carbonate and water, dried ($MgSO_4$), filtered, and concentrated to a syrup that contained **5**. The syrup was purified on a prepacked column of silica gel (toluene–ethyl acetate, 8:1) to yield **5** (6.5 g), R_F 0.60 (t.l.c., same solvent), $[\alpha]_{578}$ +119° (*c* 1.1, chloroform). N.m.r. data: δ 1.35 (d, 3 H, $J_{5,6}$ 6.3 Hz, H-6, hexenopyranose residue), 3.74–4.33 (m, 6 H; H-4, H-5, and H-6, mannose residue; H-5, hexenopyranose residue), 4.57 (dd, 1 H, $J_{2,3}$ 2.9 Hz, $J_{3,4}$ 10.3 Hz, H-3, mannose residue), 4.84 and 4.95 (AB spectrum, 2 H, $J_{H,H}$ 12.2 Hz, PhCH_2), 5.33 (s, 1 H, PhC*H*), 5.53–5.66 (m, 3 H; H-1, mannose residue; H-3, H-4, hexenopyranose residue), 6.01 (d, 1 H, $J_{1,3}$ 2.0 Hz, H-1, hexenopyranose residue), and 6.95–8.3 (24 H, aromatic H).

*p-Trifluoroacetamidophenyl 2-*O*-benzyl-4,6-*O*-benzylidene-3-*O*-(2,4-di-*O*-benzoyl-3,6-dideoxy-α-D-*erythro*-hex-2-enopyranosyl)-α-D-mannopyranoside* (**6**). — A solution of **5** (6.0 g) in ethyl acetate (200 ml) was hydrogenated over Adams' catalyst (400 mg) at room temperature and atmospheric pressure. When sufficient hydrogen ($NO_2 \rightarrow$

NH_2) had been consumed, trifluoroacetic anhydride (13.5 ml) and pyridine (32 ml) were added, and the mixture was kept at 60° for 30 min, and then filtered and concentrated. A solution of the residue in toluene was washed with water, dried (Na_2SO_4), filtered, and concentrated to a syrup that was purified on a prepacked column of silica gel (toluene–ethyl acetate, 8:1) to yield **6** (5.2 g). R_F 0·55 (t.l.c., same solvent), $[\alpha]_{578}$ +91° (*c* 1.0, chloroform). N.m.r. data ($CDCl_3$): δ 1.32 (d, 3 H, $J_{5,6}$ 6.5 Hz, H-6, hexenopyranose residue), 3.65–4.33 (m, 6 H; H-4, H-5, H-6, mannose residue; H-5, hexenopyranose residue), 4.55 (dd, 1 H, $J_{2,3}$ 2.9, $J_{3,4}$ 10.0 Hz, H-3, mannose residue), 4.84 and 4.92 (AB spectrum, 2 H, $J_{H,H}$ 12.2 Hz, PhCH_2), 5.30 (s, 1 H, PhC*H*), 5.41–5.65 (m, 3 H; H-1, mannose residue; H-3, H-4, hexenopyranose residue), 5.97 (d, 1 H, $J_{1,3}$ 2.0 Hz, H-1, hexenopyranose residue), and 6.84–8.09 (24 H, aromatic H).

p-*Trifluoroacetamidophenyl 3*-O-(*2,4-di*-O-*benzoyl-3,6-dideoxy*-α-D-ribo-*hexopyranosyl*)-α-D-*mannopyranoside* (**7**) *and* p-*trifluoroacetamidophenyl 3*-O-(*2,4-di*-O-*benzoyl-3,6-dideoxy*-α-D-arabino-*hexopyranosyl*)-α-D-*mannopyranoside* (**8**). — A solution of **6** (2.2 g) in ethanol (100 ml) and tetrahydrofuran (10 ml) was hydrogenated over 10% palladium-on-charcoal (1.5 g). When no more hydrogen was consumed (36 h), the mixture was filtered and concentrated to a syrup that gave two spots in t.l.c. (chloroform–methanol, 9:1). Fractionation on a column of silica gel gave **7** (1.05 g) and **8** (0.33 g). Disaccharide **7** had R_F 0.43 (t.l.c., same solvent), $[\alpha]_{578}$ +118° (*c* 1.1, chloroform). N.m.r. data (CD_3OD): δ 1.26 (d, 3 H, $J_{5,6}$ 6.0 Hz, H-6, paratose residue), 2.15–2.61 (m, 2 H, H-3, paratose residue), and 7.07–8.15 (14 H, aromatic H). Disaccharide **8** had R_F 0.35 (same solvent), $[\alpha]_{578}$ +86° (*c* 1.1, chloroform). N.m.r. data (CD_3OD): δ 1.31 (d, 3 H, $J_{5,6}$ 5.9 Hz, H-6, tyvelose residue), 2.20–2.42 (m, 2 H, H-3, tyvelose residue), 7.10–8.17 (14 H, aromatic H).

Anal. Calc. for $C_{34}H_{34}F_3NO_{12}$: C, 57.9; H, 4.9; F, 8.1; N, 2.0. Found for **7**: C, 57.8; H, 4.8; F, 7.9; N, 2.0. Found for **8**: C, 57.7; H, 4.8; F, 8.2; N, 2.1.

p-*Trifluoroacetamidophenyl 3*-O-(*3,6-dideoxy*-α-D-ribo-*hexopyranosyl*)-α-D-*mannopyranoside* (**9**). — A solution of **7** (100 mg) in methanol (25 ml) containing a catalytic amount of sodium was kept at room temperature for 12 h, neutralized with Dowex 50 (H^+) resin, filtered, and concentrated. The residue was purified by preparative t.l.c. (ethyl acetate–methanol–water, 85:10:5) to yield **9** (60 mg), R_F 0.50 (t.l.c., same solvent), $[\alpha]_{578}$ +150° (*c* 1.0, water). N.m.r. data (CD_3OD): δ 1.22 (d, 3 H, $J_{5,6}$ 6.3 Hz, H-6, paratose residue), 5.04 (d, 1 H, $J_{1,2}$ 3.4 Hz, H-1, paratose residue), 5.47 (d, 1 H, $J_{1,2}$ 1.5 Hz, H-1, mannose residue), 7.13 and 7.53 (2 d, each 2 H, $J_{H,H}$ 8.8 Hz, *p*-$CF_3CONHC_6H_4O$).

p-*Trifluoroacetamidophenyl 3*-O-(*3,6-dideoxy*-α-D-arabino-*hexopyranosyl*)-α-D-*mannopyranoside* (**10**). — Compound **8** (95 mg) in methanol was debenzoylated, worked-up, and purified, as described for **7**, to give **10** (54 mg), R_F 0.41 (t.l.c.; ethyl acetate–methanol–water, 85:10:5), $[\alpha]_{578}$ +145° (*c* 1.1, water). N.m.r. data (CD_3OD): δ 1.26 (d, 3 H, $J_{5,6}$ 5.9 Hz, H-6, tyvelose residue), 4.92 (d, 1 H, $J_{1,2}$ 1.5 Hz, tyvelose residue), 5.47 (d, 1 H, $J_{1,2}$ 2.0 Hz, H-1, mannose residue), 7.13 and 7.53 (2 d, each 2 H, $J_{H,H}$ 9.3 Hz, *p*-$CF_3CONHC_6H_4O$).

Hydrolysis of **9** and **10** with 0.25M sulfuric acid for 12 h at 100°, followed by reduction with sodium borohydride and treatment with acetic anhydride in pyridine, gave paratitol tetra-acetate and mannitol hexa-acetate from **9**, and tyvelitol tetra-acetate and mannitol hexa-acetate from **10**, indistinguishable from authentic samples in g.l.c.–m.s.[11,12]. Methylation[13] of **9** and **10**, followed by hydrolysis, reduction, and acetylation as described above, gave *O*-methylalditol acetates that were indistinguishable by g.l.c.–m.s.[14] from authentic 1,5-di-*O*-acetyl-3,6-dideoxy-2,4-di-*O*-methyl-D-*ribo*-hexitol and 1,3,5-tri-*O*-acetyl-2,4,6-tri-*O*-methyl-D-mannitol from **9**, and 1,5-di-*O*-acetyl-3,6-dideoxy-2,4-di-*O*-methyl-D-*arabino*-hexitol and 1,3,5-tri-*O*-acetyl-2,4,6-tri-*O*-methyl-D-mannitol from **10**.

p-*Nitrophenyl 2*-O-*benzyl-4,6*-O-*benzylidene-3*-O-(*2,4-di*-O-*acetyl-3,6-dideoxy*-α-D-erythro-*hex-2-enopyranosyl*)-α-D-*mannopyranoside* (**5a**). — A solution of **3a**[11] (2.5 g, prepared[12] from **2a**) and **4** (4.5 g) in benzene (90 ml) was treated as described for **3** and **4**. After work-up and purification on silica gel (toluene–ethyl acetate, 8:1), **5a** (3.7 g) was obtained; R_F 0.56 (t.l.c., same solvent), $[\alpha]_{578}$ +103° (*c* 1.1, chloroform). N.m.r. data ($CDCl_3$): δ 1.22 (d, 3 H, $J_{5,6}$ 6.0 Hz, H-6, hexenopyranose residue), 1.63 (s, 3 H, OAc), 2.10 (s, 3 H, OAc), 4.77 and 4.89 (AB spectrum, 2 H, $J_{H,H}$ 11.5 Hz, PhCH_2), 5.19 (m, 1 H, H-4, hexenopyranose residue), 5.36 (d, 1 H, $J_{1,3}$ 0.5 Hz, H-1, hexenopyranose residue), 5.57 (d+s, 2 H, H-1, mannose residue and PhC*H*), 5.64 (d, 1 H, $J_{3,4}$ 2.0 Hz, H-3, hexenopyranose residue), 7.10 and 8.18 (2 d, each 2 H, $J_{H,H}$ 9.2 Hz, *p*-$NO_2C_6H_4O$), and 7.24–7.56 (m, 10 H, aromatic H).

p-*Trifluoroacetamido 2*-O-*benzyl-4,6*-O-*benzylidene-3*-O-(*2,4-di*-O-*acetyl-3,6-dideoxy*-α-D-erythro-*hex-2-enopyranosyl*)-α-D-*mannopyranoside* (**6a**). — Compound **5a** (3.0 g) was hydrogenated over Adams' catalyst. *N*-Trifluoroacetylation of the product and work-up as described for **5**, followed by purification on silica gel (toluene–ethyl acetate, 4:1), yielded **6a** (2.6 g), R_F 0.52 (t.l.c. same solvent), $[\alpha]_{578}$ +103° (*c* 1.0, chloroform). N.m.r. data ($CDCl_3$): δ 1.21 (d, 3 H, $J_{5,6}$ 6.3 Hz, H-6, hexenopyranose residue), 1.64 (s, 3 H, OAc), 2.10 (s, 3 H, OAc), 4.78 and 4.88 (AB spectrum, 2 H, $J_{H,H}$ 12.0 Hz, PhCH_2). 5.21 (m, 1 H, H-4, hexenopyranose residue), 5.37 (s, 1 H, H-1, hexenopyranose residue), 5.51 (d, 1 H, $J_{1,2}$ 1.5 Hz, H-1, mannose residue), 5.58 (s, 1 H, PhC*H*), 5.66 (d, 1 H, $J_{3,4}$ 2.0 Hz, H-3, hexenopyranose residue), 7.05 and 7.49 (2 d, each 2 H, $J_{H,H}$ 9.0 Hz, *p*-$CF_3CONHC_6H_4O$), 7.12–7.54 (m, 10 H, aromatic H).

Anal. Calc. for $C_{38}H_{38}F_3NO_{12}$: C, 60.2; H, 5.1; F, 7.5; N, 1.9. Found: C, 60.3; H, 5.1; F, 7.4; N, 1.8.

p-*Trifluoroacetamidophenyl 3*-O-(*2,4-di*-O-*acetyl-3,6-dideoxy*-α-D-ribo-*hexopyranosyl*)-α-D-*mannopyranoside* (**7a**) *and* p-*trifluoroacetamidophenyl 3*-O-(*2,4-di*-O-*acetyl-3,6-dideoxy*-α-D-arabino-*hexopyranosyl*)-α-D-*mannopyranoside* (**8a**). — A solution of **6a** (2.0 g) in ethanol (100 ml) was hydrogenated over 10% palladium-on-charcoal (2.0 g) and worked-up as described for **6**. Purification on silica gel (chloroform–methanol, 7.5:1) gave a mixture of **7a** and **8a** (0.8 g, not separated by t.l.c.), R_F 0.20 (t.l.c., same solvent), $[\alpha]_{578}$ +148° (*c* 0.5, chloroform). N.m.r. data (CD_3OD):

δ 1.16 and 1.19 (two overlapping d, $J_{5,6}$ 5.9 and 6.3 Hz, H-6 from dideoxy sugars), 7.16 and 7.56 (2 d, each 2 H, $J_{H,H}$ 9.0 Hz, *p*-$CF_3CONHC_6H_4O$).

p-*Trifluoroacetamidophenyl 3*-O-(*3,6-dideoxy*-α-D-ribo-*hexopyranosyl*)-α-D-*mannopyranoside* (**9**) *and* p-*trifluoroacetamidophenyl 3*-O-(*3,6-dideoxy*-α-D-arabino-*hexopyranosyl*)-α-D-*mannopyranoside* (**10**). — A solution of **7a** and **8a** (680 mg) in methanol (40 ml) containing a catalytic amount of sodium was kept at room temperature for 12 h. Work-up as described for **9** gave a mixture of **9** and **10**. Separation on a column of silica gel (ethyl acetate–methanol–water, 85:10:5) gave pure **9** (150 mg) and pure **10** (240 mg).

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