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Note

Syntheses of *p*-trifluoroacetamidophenyl 4-*O*-α-D-glucopyranosyl-α-D-galactopyranoside and *p*-trifluoroacetamidophenyl 6-*O*-α-D-glucopyranosyl-α-D-galactopyranoside

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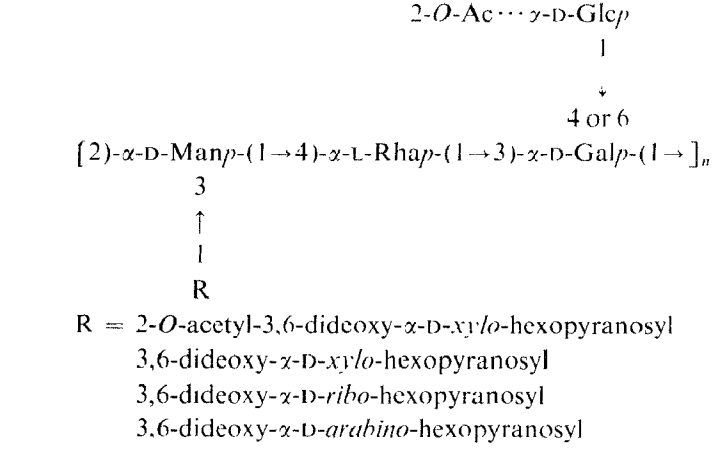
A generalised structure for the immunogenic polysaccharide part of *Salmonella* cell-wall lipopolysaccharides belonging to serogroups A, B, and D_1 is as follows[1].



R = 2-*O*-acetyl-3,6-dideoxy-α-D-*xylo*-hexopyranosyl
3,6-dideoxy-α-D-*xylo*-hexopyranosyl
3,6-dideoxy-α-D-*ribo*-hexopyranosyl
3,6-dideoxy-α-D-*arabino*-hexopyranosyl

The title compounds **9** and **12**, representing the glucosylgalactose branching part(s) of this structure, were required for immunological studies, and their synthesis is now reported.

p-Nitrophenyl 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranoside[2] (**1**) was converted *via* deacetylation[2,3], benzylidenation[4], and acetylation into **4**. Hydrogenation of **4** followed by *N*-trifluoroacetylation yielded the key compound *p*-trifluoroacetamidophenyl 2,3-di-*O*-acetyl-4,6-*O*-benzylidene-α-D-galactopyranoside (**5**). Reduction of **5** with sodium cyanoborohydride–hydrogen chloride[5] in tetrahydrofuran yielded **6** having HO-4 free. Partial, acidic hydrolysis of **5**, on the other hand, yielded **7** having both HO-4 and HO-6 free. The diol **7** was glucosylated with 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl bromide, using halide-ion assistance[6], to yield, after deacetyla-

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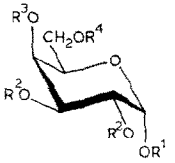
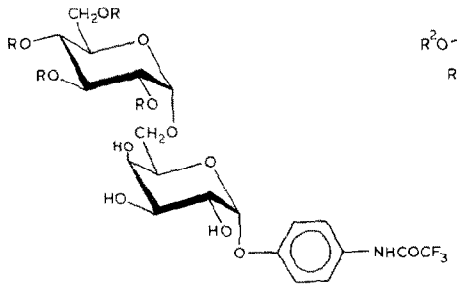
tion, 54% of the (1→6)-α-linked disaccharide derivative **8**. Catalytic hydrogenation of **8** then afforded the target compound **9** (20% from **2**).

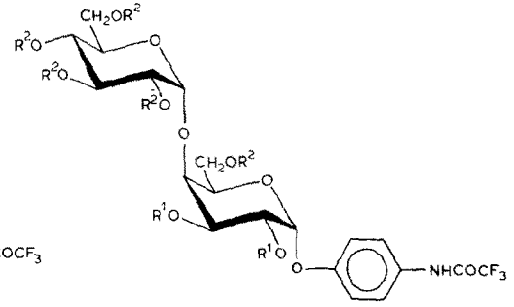
Glucosylation of **6** with 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl chloride, using silver triflate as promoter[7], yielded 51% of the (1→4)-α-linked disaccharide derivative **10**. Deprotection of **10** *via* **11** then afforded the target compound **12** (22% from **2**).



1 R^1 = C_6H_4-*p*-NO_2, R^2 = R^3 = R^4 = Ac
2 R^1 = C_6H_4-*p*-NO_2, R^2 = R^3 = R^4 = H
3 R^1 = C_6H_4-*p*-NO_2, R^2 = H, R^3 and R^4 = CHPh
4 R^1 = C_6H_4-*p*-NO_2, R^2 = Ac, R^3 and R^4 = CHPh
5 R^1 = C_6H_4-*p*-$NHCOCF_3$, R^2 = Ac, R^3 and R^4 = CHPh
6 R^1 = C_6H_4-*p*-$NHCOCF_3$, R^2 = Ac, R^3 = H, R^4 = Bzl
7 R^1 = C_6H_4-*p*-$NHCOCF_3$, R^2 = Ac, R^3 = R^4 = H



8 R = Bzl
9 R = H



10 R^1 = Ac, R^2 = Bzl
11 R^1 = H, R^2 = Bzl
12 R^1 = R^2 = H

EXPERIMENTAL

General methods. — These were the same as those previously reported[8]. Column chromatography on silica gel was performed in the flash mode[9]. Sugar[10,11] and methylation[12] analyses were in agreement with the postulated structures. ^{13}C-N.m.r. shifts are given in p.p.m. downfield from that of internal Me_4Si for solutions in $CDCl_3$, and otherwise from external Me_4Si.

p-*Nitrophenyl 4,6-O-benzylidene-α-D-galactopyranoside* (**3**). — A mixture[4] of

benzaldehyde (20 mL), formic acid (20 mL), and *p*-nitrophenyl α-D-galactopyranoside[2,3] (**2**, 5.0 g) was stirred at room temperature for 15 min, and then poured into a stirred mixture of water (75 mL) and light petroleum (75 mL). The resulting crystals were collected, and washed with saturated, aqueous sodium hydrogencarbonate and light petroleum. Recrystallisation from toluene yielded **3** (5.8 g, 90%), m.p. 213°, $[\alpha]_D$ +126° (*c* 1, *N*,*N*-dimethylformamide), which contained toluene (0.25 mol/mol, ^{1}H-n.m.r. data). Recrystallisation from ethanol–water gave material with m.p. 213°, $[\alpha]_D$ +129° (*c* 1.9, *N*,*N*-dimethylformamide). ^{13}C-N.m.r. data (25 MHz, pyridine-d_6): δ 65.3 (C-6), 69.3, 69.5, 69.7, 77.8, (C-2,3,4,5), 99.9 (C-1), 101.5 (PhCH), 117.0, 126.1, 127.1, 128.5, 129.2, 139.5, 142.6, and 163.0 (aromatic C).

Anal. Calc. for $C_{19}H_{19}NO_8$: C, 58.6; H, 4.92; N, 3.60. Found: C, 58.6; H, 5.02; N, 3.50.

p-*Nitrophenyl 2,3-di-O-acetyl-4,6-O-benzylidene-α-D-galactopyranoside* (**4**). — Treatment of **3** (3.4 g) with pyridine (5 mL) and acetic anhydride (5 mL) at room temperature for 2 h, followed by concentration, several co-concentrations with toluene, and crystallisation of the residue from ethanol, yielded **4** (3.9 g, 95%), m.p. 167–169°, $[\alpha]_D$ +197° (*c* 1.2, chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 20.7, 20.8 (2 CH_3CO), 64.2 (C-6), 68.1, 68.8, 69.2, 74.1 (C-2,3,4,5), 96.0 (C-1), 101.4 (PhCH), 117.0, 126.2, 126.7, 128.6, 129.5, 138.0, 143.6, 161.8 (aromatic C), 170.2, and 170.6 (2 CH_3CO).

Anal. Calc. for $C_{23}H_{23}NO_{10}$: C, 58.4; H, 4.90; N, 2.96. Found: C, 58.2; H, 4.95; N, 2.87.

p-*Trifluoroacetamidophenyl 2,3-di-O-acetyl-4,6-O-benzylidene-α-D-galactopyranoside* (**5**). — A solution of **4** (3.4 g) in ethyl acetate (100 mL) was hydrogenated over 5% Pd/C at atmospheric pressure, filtered, and concentrated, to yield a syrup that was treated with trifluoroacetic anhydride (1.5 mL) in ice-cold pyridine (30 mL). After 30 min at room temperature, the mixture was diluted with toluene–ether (10:1), washed with water and saturated aqueous sodium hydrogencarbonate, dried (Na_2SO_4), filtered, and concentrated. Chromatography of the syrupy residue on a column of silica gel (toluene–ethyl acetate, 3:1) yielded **5** (3.6 g, 93%), $[\alpha]_D$ +146° (*c* 0.9, chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 20.7, 20.8 (2 CH_3CO), 63.3 (C-6), 67.9, 68.6, 69.0, 73.8 (C-2,3,4,5), 95.5 (C-1), 101.0 (PhCH), 117.1, 122.6, 126.3, 128.3, 129.2, 130.3, 137.6, 154.6 (aromatic C), 170.3, and 170.8 (2 CH_3CO); the $COCF_3$ signals were not recorded.

p-*Trifluoroacetamidophenyl 2,3-di-O-acetyl-6-O-benzyl-α-D-galactopyranoside* (**6**). — Sodium cyanoborohydride (1.5 g) was added at room temperature to a stirred solution of **5** (1.5 g) in tetrahydrofuran[5]. A saturated solution of hydrogen chloride in ether was then added until gas evolution ceased. After an additional 10 min, Dowex 50 (H^+) resin was added, and the mixture was filtered through Celite, diluted with dichloromethane, washed with saturated aqueous sodium hydrogencarbonate and water, dried (Na_2SO_4), filtered, and concentrated. Chromatography of the residue on a column of silica gel (toluene–ethyl acetate, 2:1) yielded **6** (1.15 g, 76%) which, after crystallisation from ethyl acetate–hexane, had m.p. 90–95°, $[\alpha]_D$ +144° (*c* 1,

chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 20.7, 20.9 (2 CH_3CO), 68.1 (C-6), 68.9, 69.0, 69.2, 70.3, 73.7 (C-2,3,4,5, PhCH_2), 95.4 (C-1), 117.5, 122.5, 127.7, 128.0, 128.5, 130.4, 137.3, 154.4 (aromatic C), 170.6, and 170.7 (2 CH_3CO); CF_3 signals were recorded at 110.2 and 121.7. A satisfactory elemental analysis was not obtained for this compound, but its purity was established by t.l.c. (toluene–ethyl acetate, 2:1) and by ^{1}H- and ^{13}C-n.m.r. spectroscopy.

p-*Trifluoroacetamidophenyl 2,3-di*-O-*acetyl*-α-D-*galactopyranoside* (**7**). — A mixture of **5** (0.90 g) and 90% trifluoroacetic acid (10 mL) was stirred at room temperature for 5 min. Toluene (50 mL) was then added and the mixture was concentrated. Chromatography of the residue on a column of silica gel (toluene–ethyl acetate, 1:3) yielded **7** (0.58 g, 77%), $[\alpha]_D$ +174° (*c* 1.2, ethanol). ^{13}C-N.m.r. data (CD_3OD): δ 20.6, 20.8 (2 CH_3CO), 62.0 (C-6), 68.3, 69.4, 71.9, 72.8 (C-2,3,4,5), 97.0 (C-1), 118.5, 123.8, 132.4, 155.9 (aromatic C), and 172.1 (2 CH_3CO); CF_3 signals were recorded at 111.7 and 124.0.

p-*Trifluoroacetamidophenyl* 6-O-(*2,3,4,6-tetra*-O-*benzyl*-α-D-*glucopyranosyl*)-α-D-*galactopyranoside* (**8**). — A solution of 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl bromide[6,13] (1.2 g) in dichloromethane (3 mL) was added, with stirring, at room temperature to a solution of **7** (0.45 g) in *N,N*-dimethylformamide (2 mL) containing tetraethylammonium bromide (0.35 g) and crushed molecular sieves (4 Å). After being stirred overnight, the mixture was diluted with dichloromethane, filtered, and concentrated. Chromatography of the residue on a column of silica gel (toluene–ethyl acetate, 4:1) yielded a crude product that was deacetylated with a catalytic amount of sodium methoxide in methanol. When the reaction was complete (t.l.c.; toluene–ethyl acetate, 1:2), the mixture was neutralised with Dowex 50 (H^+) resin, filtered, and concentrated. Crystallisation of the residue from ethanol–water yielded **8** (0.48 g, 54%), m.p. 158–161°, $[\alpha]_D$ +85° (*c* 0.9, chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 67.3, 68.4, 69.1, 69.8, 69.9, 70.5, 73.4, 75.1, 75.5, 77.6, 79.6, 81.9 (C-2,3,4,5,6, C-2′,3′,4′,5′,6′, 4 PhCH_2, some signal overlap), 97.3, 98.4 (C-1,1′), 117.6, 122.8, 127.7, 128.0, 128.4, 129.8, 137.8, 137.9, 138.3, 138.7, and 155.0 (aromatic C); $COCF_3$ signals were not recorded.

Anal. Calc. for $C_{48}H_{48}F_3NO_{12}$: C, 64.9, H, 5.45; N, 1.58. Found: C, 64.7; H, 5.65; N, 1.46.

p-*Trifluoroacetamidophenyl* 6-O-α-D-*glucopyranosyl*-α-D-*galactopyranoside* (**9**). — Catalytic hydrogenation of a solution of **8** (0.20 g) in acetic acid (15 mL) over 5% Pd/C and chromatography of the product on a column of silica gel (ethyl acetate–methanol–water, 85:10:5) and then on a column of Biogel P2 yielded **9** (0.085 g, 71%), $[\alpha]_D$ +130° (*c* 0.8, water). ^{13}C-N.m.r. data (CD_3OD): δ 62.5 (C-6′), 67.8 (C-6), 69.9, 70.8, 71.2, 71.3, 71.6, 73.4, 73.6, 74.9 (C-2,3,4,5 and C-2′,3′,4′,5′), 99.9 (C-1,1′), 118.8, 124.1, 131.9, and 156.6 (aromatic C); CF_3 signals were observed at 118.8 and 123.4.

p-*Trifluoroacetamidophenyl 2,3-di*-O-*acetyl*-6-O-*benzyl*-4-O-(*2,3,4,6-tetra*-O-*benzyl*-α-D-*glucopyranosyl*)-α-D-*galactopyranoside* (**10**). — A solution of 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl chloride[14] (1.2 g) in toluene (5 mL) was added with

stirring at room temperature to a solution of **6** (0.60 g) in toluene (5 mL) and dichloromethane (3 mL) containing silver triflate (0.60 g) and 2,4,6-trimethylpyridine (0.16 g). After 15 min, the mixture was diluted with toluene, filtered through Celite, and concentrated. Chromatography of the residue on a column of silica gel (toluene–ethyl acetate, 15:1) yielded **10** (0.60 g, 51%), which, after crystallisation from ethanol, had m.p. 162°, $[\alpha]_D$ +123° (*c* 1.5, chloroform); and the corresponding β-linked disaccharide derivative (0.30 g, 25%) containing traces of the α anomer, $[\alpha]_D$ +88° (*c* 0.7, chloroform). ^{13}C-N.m.r. data for **10** ($CDCl_3$): δ 20.7, 21.2, (2 CH_3CO), 68.4, 69.6, 70.4, 71.3, 72.9, 73.6, 74.0, 75.2, 75.6, 76.7, 77.9, 80.4, 81.9 (C-2,3,4,5,6, C-2′,3′,4′,5′,6′, 5 $PhCH_2$, some signal overlap), 95.4 (C-1), 100.1 (C-1′), 117.8, 122.2, 127.4, 127.6, 127.8, 128.0, 128.4, 130.2, 137.9, 138.0, 138.2, 138.3, 138.8, 154.6 (aromatic C), 170.2, and 170.7 (2 CH_3CO); $COCF_3$ signals were not recorded.

Anal. Calc. for $C_{59}H_{60}F_3NO_{14}$: C, 66.6; H, 5.68; N, 1.32. Found: C, 66.5; H, 5.71; N, 1.27.

The C-1′ signal for the (1→4)-β-linked isomer had δ 104.1.

p-*Trifluoroacetamidophenyl* 6-O-*benzyl*-4-O-(2,3,4,6-*tetra*-O-*benzyl*-α-D-*glucopyranosyl*)-α-D-*galactopyranoside* (**11**). — Compound **10** (0.40 g) was deacetylated, as described above in the preparation of **8**, to yield **11** (0.34 g, 93%) which, after crystallisation from toluene–light petroleum, had m.p. 148–151°, $[\alpha]_D$ +83° (*c* 1.3, chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 69.0, 69.3, 69.8, 70.5, 71.0, 71.6, 72.9, 73.7, 75.2, 75.7, 78.0, 79.9, 81.7 (C-2,3,4,5,6, C-2′,3′,4′,5′,6′, 5 $PhCH_2$, one signal overlap), 97.8 (C-1), 99.9 (C-1′), 117.8, 122.2, 127.3, 127.5, 127.7, 127.9, 128.1, 128.2, 128.5, 129.9, 137.5, 137.8, 138.0, 138.2, 138.6, and 154.7 (aromatic C); $COCF_3$ signals were not recorded.

Anal. Calc. for $C_{55}H_{54}F_3NO_{12}$: C, 67.5; H, 5.57; N, 1.43. Found: C, 67.4; H, 5.69; N, 1.43.

p-*Trifluoroacetamidophenyl* 4-O-α-D-*glucopyranosyl*-α-D-*galactopyranoside* (**12**). — Compound **11** (0.20 g) was hydrogenated and the product was isolated, as described above for the preparation of **9**, to yield **12** (85 mg, 78%), $[\alpha]_D$ +170° (*c* 1, water). ^{13}C-N.m.r. data (D_2O): δ 61.4 (C-6,6′), 69.4, 70.0, 70.2, 70.6, 73.0, 73.2, 73.9, 79.8 (C-2,3,4,5, C-2′,3′,4′,5′), 98.8, 101.1 (C-1,1′), 119.1, 124.9, 130.9, and 155.8 (aromatic C); $COCF_3$ signals were not recorded.

Methylation analysis[12]. — Methylation analysis of **9** and **12** afforded 1,5,6-tri-*O*-acetyl-2,3,4-tri-*O*-methylgalactitol and 1,5-di-*O*-acetyl-2,3,4,6-tetra-*O*-methylglucitol from **9**, and 1,4,5-tri-*O*-acetyl-2,3,6-tri-*O*-methylgalactitol and 1,5-di-*O*-acetyl-2,3,4,6-tetra-*O*-methylglucitol from **12**.

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