Syntheses of the 2′-Acetate and Some Deoxy Analogues of p-Trifluoroacetamidophenyl 3-O-(3,6-Dideoxy-α-d-xylo-hexopyranosyl)-α-d-mannopyranoside

BJÖRN CLASSON, PER J. GAREGG and THOMAS NORBERG

Department of Organic Chemistry, Arrhenius Laboratory, University of Stockholm, S-106 91 Stockholm, Sweden

Syntheses are described of derivatives of p-trifluoroacetamidophenyl 3-O-(3,6-dideoxy-α-d-xylo-hexopyranosyl)-α-d-mannopyranoside carrying an acetyl group in the 2-position of the abequosyl group, corresponding to the Salmonella O-antigen 5 and also of various deoxy derivatives of the parent compound.

In our continuing programme aimed at obtaining improved diagnostic methods for diseases caused by infections with Salmonella bacteria, several di- and oligosaccharide derivatives have been synthesized and evaluated in immunological systems. ¹ In the present paper we present a facile synthesis of the title O-acetylated disaccharide fragment corresponding to the Salmonella O-antigen 5, linked to a moiety suitable for attachment to a protein. ²

Hydrophobic interactions have been proposed to provide the main driving force for the binding between oligosaccharides and proteins. ³,⁴ One way of examining this hypothesis is to remove hydroxyl groups from various positions of an oligosaccharide hapten and thus obtain a series of deoxy derivatives. The binding properties of the compounds obtained are tested and compared to those of the parent compound. This approach has yielded important information of the nature on the interactions of blood group antigens with antibodies and lectins. ⁴,⁵ In the present work we describe syntheses of deoxy derivatives of p-

* 3,6-Dideoxy-α-xylo-hexose is henceforth termed abequose.

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protecting groups, in 27% yield from methyl \( \alpha \)-abequoside. Compound 2 was converted into the glycosyl bromide 3 which was used immediately in a halide-assisted \(^6\) glycosylation with \( p \)-nitrophenyl \( 2-O \)-benzyl-4,6-\( O \)-benzylidene-\( \alpha \)-\( D \)-mannopyranoside \(^9\) to give the disaccharide derivative 4 in 71% yield. Isomerization and removal \(^7\)\(^,\)\(^11\) of the allyl group of 4 to give 5 was followed by acetylation to give 6 in 68% yield from 4. The nitro group of 6 was reduced to an amino group and the benzyl and benzylidene protecting groups were removed from 6 by catalytic hydrogenation to give 7 which was converted into the more stable derivative 8 for storage.

3,4-Di-\( O \)-acetyl-1,2,6-trideoxy-\( \alpha \)-lyxo-hex-1-enopyranose \(^1\) was treated with iodine, mercury-(II) chloride and silver imidazolate \(^13\) and then reacted with \( p \)-trifluoroacetamidophenyl \( 2-O \)-benzyl-4,6-\( O \)-benzylidene-\( \alpha \)-\( D \)-mannopyranoside \(^14\) to yield the disaccharide derivative 9 in 49% yield. Reductive elimination from 9 by treatment with zinc dust in ethanol containing acetic acid \(^15\) yielded 10. Catalytic hydrogenation of 10 gave upon catalytic hydrogenation, gave 15 in a total yield of 67%. Treatment of 15 with bromo-trimethylsilane afforded the glycosyl bromide 16 which was used directly in a halide-assisted glycosylation \(^8\) with \( p \)-trifluoroacetamidophenyl \( 2-O \)-benzyl-4,6-\( O \)-benzylidene-\( \alpha \)-\( D \)-mannopyranoside \(^14\) to yield the disaccharide derivative 17 in 50% yield. Deprotection of 17 by means of catalytic hydrogenation afforded the target compound 18.

Finally, a deoxy function was introduced at C-6 of the parent molecule \( p \)-trifluoroacetamidophenyl 3-\( O \)-abequosyl-\( \alpha \)-\( D \)-mannopyranoside. Thus the benzylidene group was removed by careful partial hydrolysis of \( p \)-trifluoroacetamidophenyl \( 2-O \)-benzyl-3-\( O \)-(2,4-di-\( O \)-benzyl)-3,6-
dideoxy-α-D-xylo-hexopyranosyl)-4,6-O-benzylidene-α-D-mannopyranoside. The product was then treated with triphenylphosphine, iodine and imidazole to give the 6-deoxy-6-iodo derivative. This upon hydrogenation yielded the target compound in 68% yield from the disaccharide starting material.

**EXPERIMENTAL**

**General methods.** Melting points are corrected. Concentrations were performed at reduced pressure (1–2 kPa) at <40 °C (bath). Optical rotations were recorded for solutions in chloroform unless otherwise stated at 22–24 °C using a Perkin-Elmer 241 polarimeter. NMR spectra (100 or 400 MHz for 1H and 25 MHz for 13C) were recorded at 25 °C unless otherwise stated, using a JEOL JNM FX-100 or a JEOL GX-400 instrument. Chemical shifts are given in ppm downfield from those of internal (CDCl3 or CD3OD solutions) or external (D2O solutions) Me4Si. NMR spectra for all new compounds were in accordance with the postulated structures and only selected data are reported. TLC was performed on silica gel F254 (Merck) with detection by UV light and/or charring with sulfuric acid. Column chromatography was performed on silica gel 60 (0.04–0.063 mm, Merck) in the flash mode using, unless otherwise stated, a suitable toluene-ethyl acetate mixture for elution. The loading was in the range 1/25–1/100. Organic solutions were dried over anhydrous magnesium sulfate. Molecular sieves (4Å, Union Carbide) were desiccated in vacuo at 300 °C overnight and ground immediately before use.

**Methyl 2-O-allyl-3,6-dideoxy-α-D-xylo-hexopyranoside (1).** Sodium hydride (630 mg, 26.3 mmol) was added to a stirred and cooled (ice bath) solution of methyl 3,6-dideoxy-α-D-xylo-hexopyranoside (4.0 g, 24.7 mmol) in N,N-dimethylformamide (80 ml). After 30 min, allyl bromide (3.0 g, 24.7 mmol) was added. After 6 h at 0 °C, ethanol (2 ml) was added and the mixture was filtered and concentrated (100 Pa). TLC of the residual syrup showed three minor spots tentatively assigned to starting material, the 2,4-dialyl and the 4-allyl derivative and a main spot corresponding to compound 1. Purification by column chromatography, using 1:4 ethanol-hexane for elution gave pure, syrupy I (1.75 g, 35%), [α]D +109° (c 1.0). 13C NMR (CDCl3): δ 16.3 (C-6), 32.2 (C-3), 55.2 (OCH3), 65.7, 69.3, 70.1, 70.6 (C-2,4,5 and allyl CH2—CH=CH2), 98.0 (C-1), 117.3, 134.9 (allyl olefinic C). Acetylation of I with acetic anhydride in pyridine produced a single compound, whose 1H NMR spectrum showed, *inter alia*, a multiplet, not present in the spectrum of I at δ 4.98, assigned by spin decoupling to H-4. The position of acetylation and allylation is therefore proved.

**Methyl 2-O-allyl-4-O-benzyl-3,6-dideoxy-α-D-xylo-hexopyranoside (2).** Compound 1 (1.48 g) was treated with benzy1 bromide (1.17 ml) and sodium hydride (180 mg) in N,N-dimethyl formamide (30 ml) essentially as described above for the analogous allylation. Pure, syrupy 2 (1.61 g, 75%) was obtained after column chromatography, [α]D +35° (c 1.0). 13C NMR (CDCl3): δ 16.4 (C-6), 27.7 (C-3), 54.9 (OCH3), 97.8 (C-1), 117.3, 134.9 (allyl olefinic C).

**2-O-Allyl-4-O-benzyl-3,6-dideoxy-α-D-xylo-hexopyranosyl bromide (3).** Compound 2 (560 mg) was dissolved in bromotrimethylsilane (5.0 ml) at room temperature. After 3 h, the solution was concentrated and subsequently co-concentrated twice with dry dichloromethane. The unstable syrup obtained (650 mg) was used directly in the
next step. 1H NMR (CDCl₃): δ 6.69 (d, J₁,₂ 2Hz, H-1).  

p-Nitrophenyl 2-O-benzyl-4,6-O-benzylidene-3-O-(2-O-allyl-4-O-benzyl-3,6-dideoxy-α-d-xylono-hexopyranosyl)-α-d-mannopyranoside (4). A solution of freshly prepared 3 (650 mg) in 9:1 dichloromethane-N,N-dimethyleformamide (4 ml) was added to a stirred and cooled (−25 °C) solution of p-nitrophenyl 2-O-benzyl-4,6-O-benzylidene-α-d-mannopyranoside ⁹,1⁰ (984 mg) and tetraethylammonium bromide (420 mg) in the same solvent (5 ml) containing molecular sieves. The mixture was allowed to attain room temperature and was then stirred overnight, filtered, diluted with dichloromethane, washed with aqueous sodium hydrogencarbonate and concentrated. Purification by column chromatography gave pure, syrupy 4 (1.0 g, 71 %). 1H NMR (CDCl₃): δ [a]D +112° (c 0.7), 13C NMR (CDCl₃): δ 16.8 (C-6'), 28.1 (C-3'), 97.2, 97.5 (C-1, 1'), 102.3 (benzylidene acetal C).  

p-Nitrophenyl 2-O-benzyl-4,6-O-benzylidene-3-O-((4-O-benzyl-3,6-dideoxy-α-d-xylono-hexopyranosyl)-α-d-mannopyranoside (5). A solution of 4 (1.66 g) in 8:3:1 ethanol–benzene–water (83 ml) containing tris(triphenylphosphine)-rhodium(I) chloride (325 mg) was boiled under reflux for 24 h. The concentrated mixture was dissolved in 10:1 acetone–water (60 ml). Mercury(II) oxide (832 mg) and mercury(II) chloride were added with stirring at room temperature. After 16 h, the mixture was concentrated. The residue was dissolved in chloroform, washed with 5 % aqueous EDTA solution and water, concentrated, and purified by column chromatography. Pure 5 (1.23 g, 78 %) was obtained. Crystallized from chloroform–diethyl ether–hexane it had m.p. 121 °C, [a]D +114° (c 1.0). 13C NMR (CDCl₃): δ 16.8 (C-6'), 30.8 (C-3'), 96.9 (C-1), 101.1 (C-1'), 101.9 (benzylidene acetal C). Anal: C₉₀H₇₀NO₁₁·H₂O.  

p-Nitrophenyl 3-O-(2-O-acetyl-4-O-benzyl-3,6-dideoxy-α-d-xylono-hexopyranosyl)-2-O-benzyl-4,6-O-benzylidene-α-d-mannopyranoside (6). Compound 5 (1.10 g) was dissolved in 2:1 pyridine–acetic anhydride. After 16 h at room temperature, the mixture was poured into ice-water and the crude product was filtered off. Purification by column chromatography gave pure, syrupy 6 (1.02 g, 87 %), [a]D +102° (c 1.0). 13C NMR (CDCl₃): δ 16.7 (C-6'), 20.7 (acetyl Me), 26.8 (C-3'), 96.7 (C-1), 97.7 (C-1'), 102.0 (benzylidene acetal C).  

p-Aminophenyl 3-O-(2-O-acetyl-3,6-dideoxy-α-d-xylono-hexopyranosyl)-α-d-mannopyranoside (7). A solution of 6 (250 mg) in absolute ethanol (5 ml) was added to a suspension of 10 % Pd/C (150 mg) in absolute ethanol (10 ml) containing concentrated hydrochloric acid (0.05 ml). The mixture was hydrogenated at 400 kPa for 16 h. Chromatographic purification (solvent system: ethyl acetate–methanol–water, 85:10:5) of the filtered and concentrated material gave fractions containing 7. These were pooled and concentrated to about 5 ml then diluted with 10 ml of water and lyophilized to give pure 7 (130 mg, 87 %), [a]D +150° (c 0.5, water). NMR (D₂O, 50 °C): 1H NMR (CDCl₃, δ 16.8 (C-6'), 21.9 (acetyl Me), 31.6 (C-3'), 99.4, 100.8 (C-1, C-1'), 119.4, 120.3, 142.1, 150.6 (p-aminophenyl C), 174.8 (acetyl C=O). 1H NMR (CDCl₃): δ 1.03 (d, J₆',₇ 6.9 Hz, H-6'), 1.96 (s, acetyl Me), 4.88 (m, H-2'), 5.09 (d, J₆',₇ 3.4 Hz, H-1'), 5.24 (d, J₆',₇ 1.4 Hz, H-1).  

p-Trifluorooacetamidophenyl 3-O-(2-O-acetyl-3,6-dideoxy-α-d-xylono-hexopyranosyl)-α-d-mannopyranoside (8). Trifluoroacetic anhydride (0.07 ml) was added in portions to a solution of 7 (44 mg) in pyridine (1 ml) while stirring and cooling in ice. After 1 h, water (0.1 ml) was added and the mixture was stirred at room temperature for 16 h. The residue after concentration was subjected to column chromatography (solvent system: ethyl acetate–methanol, 95:5, yielding pure 8 (30 mg, 56 %), [a]D +128° (c 0.5, water). 13C NMR (D₂O): δ 16.6 (C-6'), 21.6 (acetyl Me), 31.3 (C-3'), 61.7 (C-6), 66.8, 68.1, 68.3, 69.3, 71.2, 75.2, 80.0 (C-2,3,4,5, C-2', 4', 5'), 99.3, 99.4 (C-1, 1'), 118.7, 125.1, 130.7, 154.9 (p-trifluorooacetamidophenyl aromatic C). 174.5 (acetyl C=O). 1H NMR (D₂O, 85 °C): δ 1.15 (d, J₆',₇ 6.8 Hz, H-6'), 1.87–2.34 (m, H-3'ax, H-3'ex), 2.09 (s, acetyl Me), 4.96–5.17 (m, H-2'), 5.26 (d, J₆',₇ 3.9 Hz, H-1'), 5.56 (d, J₆',₇ 1.9 Hz H-1'), 7.14, 7.23, 7.43, 7.52 (aromatic H).  

p-Trifluorooacetamidophenyl 3-O-(3,4-di-O-acetyl-2-deoxy-2-ido-α-p-talo-hexopyranosyl)-2-O-benzyl-4,6-O-benzylidene-α-d-mannopyranoside (9). A mixture of silver imidazolate ¹³ (1.09 g), mercury(II) chloride (2.54 g) and molecular sieves was stirred in acetonitrile (38 ml) for 1 h, then 3,4-di-O-acetyl-1,2,6-trideoxy-d-lxyo-hex-1-enopyranose ¹⁴ (1.00 g) was added, followed by iodine (1.59 g). After 1 h, p-trifluorooacetamidophenyl 2-O-benzyl-4,6-O-benzylidene-α-d-mannopyranoside ¹⁴ (2.55 g) was added and stirring was continued at room temperature for 4 h, then more iodine (0.40 g) and silver imidazolate (0.27 g) were added. After another 4 h, triethylamine (9 ml) was added and the mixture was diluted with toluene–ethyl acetate (1:1) and filtered. The filtrate was washed with 5 % aqueous EDTA solution, aqueous sodium hydrogencarbonate and brine. The residue after drying and concentrating was purified by column chromatography to give pure, syrupy 9 (2.02 g,  

methane (50 ml) was stirred and cooled in an ice bath while triphosphene (1.18 ml, 15.4 mmol) was added dropwise. After 30 min, 15 g of silica gel was added and the mixture was concentrated. The residue was slurried in 7:3 toluene—ethyl acetate and applied to a column of silica gel. Elution with the same solvent mixture gave pure 16 (3.8 g, 95%). Crystallized from diethyl ether—hexane it had m.p. 91–92 °C, [α]_D + 48° (c 0.6). Anal. C_{15}H_{18}O_{5}S: C, H, S.

Methyl 2-O-benzyl-3,4,6-trIDEOxy-β-D-erythro-
hex-3-enopyranoside (14). A solution of 13 (3.6 g) in trimethylphosphate (10 ml) was kept at 110 °C for 40 h, and then concentrated. The residue was purified by column chromatography to give an oil (3.8 g) which, according to TLC, still contained non-carbohydrate impurities. It was used directly in the next step. 13C NMR (CDCl₃): δ 21.0 (C-6), 75.0 (OCH₃), 71.7, 72.6, 76.5 (C-2,5 and benzyl CH₂), 106.5 (C-1); 1H, δ 1.16 (d, J=6.5 Hz, H-6), 3.44 (s, OCH₃), 4.12 (d, J=1.2 Hz, H-1), 4.61 (AB q, benzyl CH₂).

2-O-Benzyl-3,4,6-trIDEOxy-α-D-erythro-hexo-
pyranosyl bromide (16). Compound 15 (200 mg) was dissolved in bromotrimethylsilane (1.5 ml) at room temperature. After 10 min, 20 ml of dry toluene was added and the solution was concentrated. After two more concentrations with toluene, the unstable oil obtained (210 mg) was taken up in dichloromethane and used directly in the next step. NMR (CDCl₃): 13C, δ 20.5 (C-6), 25.1, 31.1 (C-3,4), 69.5, 73.0, 74.7 (C-2,5 and benzyl CH₂), 95.8 (C-1); 1H, δ 6.65 (d, J=3.4 Hz, H-1).

p-Trifluoroacacetidamophenyl 2-O-benzyl-4,6-O-
benzylidene-3-O-(2-O-benzyl-3,4,6-trIDEOxy-
hexopyranosyl)-α-D-mannopyranoside (17). A solution of 16 (freshly prepared from 830 mg, 3.5 mmol of 15) in dichloromethane (2 ml) was added, at room temperature, to a stirred solution of p-trifluoroacacetidamophenyl 2-O-
benzyl-4,6-O-benzylidene-α-D-mannopyranoside (700 mg, 1.3 mmol) and tetraethylammonium bromide (500 mg) in 9:1 dichloromethane-N,N-dimethylformamide (5.5 ml) containing molecular sieves. After 6 h, triethylamine (0.5 ml) was added
added, and the mixture was diluted with dichloromethane and filtered. After concentration of the filtrate to a fourth of the original volume, diethyl ether was added and the solution was washed with water, 2M sulfuric acid, and aqueous sodium hydroxide carbonato. The residue after concentration was purified twice by column chromatography to give pure, syrupy 17 (480 mg, 50 %). [α]D +129° (c 0.4). NMR data (CDCl₃): ¹³C, δ 212.2 (C-6'), 24.6, 32.6 (C-3', 4'), 64.9, 65.1, 68.7, 69.4, 72.2, 73.9, 74.1, 77.8, 79.5, (C-2, 3, 4, 5, 6, C-2', 5'), and benzyl CH₂). 97.2, 97.7 (C-1-C-1'), 102.5 (benzylidene O-C=O); ¹H, δ 1.14 (d, J₅₆, 6.5 Hz, H-6'), 5.42 (d, J₁₂, 3.2 Hz, H-1'), 5.48 (s, benzylidene acetal H), 5.59 (d, J₁₂, 1.4 Hz, H-1').

p-Trifluoroacetamidophenyl 3-O-(3,4,6-trideoxy-α-D-erythro-hexopyranosyl)-a-D-mannopyranoside (18). A solution of 17 (240 mg) in 5:1 ethanol-ethyl acetate (24 ml) was hydrogenated over Pd/C (10 %, 150 mg) at 400 kPa for 20 h. The residue after filtration and concentration waspartitioned between diethyl ether and water. The aqueous phase was extracted twice with diethyl ether, and the combined etheral extracts were concentrated. The residue was partitioned between hexane and water. The aqueous phase was lyophilized to give pure 18 (120 mg, 78 %) as an amorphous powderr, [α]D +145° (c 0.3, water). ¹³C NMR (CD₂OD): δ 21.3 (C-6'), 27.8 (C-4'), 33.4 (C-3'), 62.5, 66.1, 67.3, 70.0, 71.8, 75.4, 80.7 (C-2, 3, 4, 5, 6, and C-2', 5'), 100.3 (C-1'), 102.5 (C-1'), 118.2, 123.7, 132.0, 155.5 (p-trifluoroacetamidophenyl aromatic C). ¹H NMR (CD₂OD, 85 °C): δ 1.11 (d, J₅₆, 6.4 Hz, H-6'), 5.10 (d, J₁₂, 3.4 Hz, H-1'), 5.57 (d, J₁₂, 2.0 Hz, H-1').

p-Trifluoroacetamidophenyl 2-O-benzyl-3-O-(2,4-di-O-benzyl-3,6-dideoxy-α-D-xylopyranosyl)-6-deoxy-6-ido-a-D-mannopyranoside (19). A solution of p-trifluoroacetamidophenyl 2-O-benzyl-3-O-(2,4-di-O-benzyl-3,6-dideoxy-α-D-xylopyranosyl)-4,6-O-benzylidene-a-D-mannopyranoside ¹⁴ (260 mg) in 80 % aqueous acetic acid (2 ml) was kept at 80 °C for 2 h and was then evaporated. The residue was taken up in 2.1 toluene-acetonitrile (3 ml) and added, at 70 °C, to a mixture of triphenylphosphine (120 mg), iodine (120 mg) and imidazole (60 mg) in 2.1 toluene-acetonitrile (7 ml). After 1 h at 70 °C, the mixture was washed with water, dried and evaporated. The residue was purified by column chromatography to give compound 19 (250 mg, 92 %) as a syrup, which, crystallized from dichloromethane-hexane, had m.p. 170-173 °C, [α]D +60° (c 0.5). Anal: C₄₁H₂₄F₆I₅N₇O₉: C, H, N. NMR (CDCl₃): ¹³C, δ 6.36 (C-6), 16.7 (C-6'), 27.0 (C-3'), 67.3, 70.9, 71.2, 71.5, 71.9, 72.9, 73.4, 75.6, 77.9, 81.9 (C-2, 3, 4, 5, C-2', 3', 5', and benzyl CH₂), 96.9, 101.1 (C-1, C-1').

p-Trifluoroacetamidophenyl 3-O-(3,6-dideoxy-α-D-xylopyranosyl)-6-deoxy-a-D-mannopyranoside (20). A solution of 19 (134 mg) in 3:1 ethanol-ethyl acetate (12 ml) was hydrogenated at 400 kPa for 20 h in the presence of 10 % Pd/C (200 mg) and powdered 4A molecular sieves (750 mg). The residue after filtration and evaporation was purified by column chromatography. The main band eluted was, according to ¹³C NMR data, the tribenzyl ether of 20. Hydrogenation as above over fresh Pd/C (100 mg) but without molecular sieves gave, after purification by gel filtration on a column of Bio-gel P-2, compound 20 (53 mg, 74 %). Crystallized from water it had m.p. 228-231 °C, [α]D +137° (c 0.2, water). Anal: C₃₉H₂₅F₄I₇O₇·H₂O: C, H, N, I. NMR (CD₂OD, 85 °C): δ 1.15 (d, J₅₆, 6.6 Hz, H-6'), 1.24 (d, J₅₆, 6.1 Hz, H-6), 1.98 (m, J₃₅₆, 2.04), 2.04 (m, J₃₅₆, 3.70 (t, J₀₃, 9.8 Hz, H-4'), 3.83 (q, J₅₆, 6.1 Hz, H-5), 3.85 (m, H-4'), 4.02 (m, H-2'), 4.04 (dd, J₅₆, 9.8 Hz, H-3), 4.11 (dd, J₅₆, 6.6, J₅₆, 1.5 Hz, H-5'), 4.24 (dd, J₁₂, 2.0, J₅₆, 3.4 Hz, H-2'), 5.15 (d, J₁₂, 3.9 Hz, H-1'), 5.53 (d, J₁₂, 2.0 Hz, H-1), 7.18, 7.20, 7.49, 7.51 (aromatic H).

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REFERENCES


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