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# SYNTHESIS OF GLYCOSIDES OF α-D-MANNOPYRANOSE 6-(α-D-GLUCOPYRANOSYL PHOSPHATE). THE PUTATIVE LIGATIN RECEPTOR

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## ABSTRACT

*p*-Nitrophenyl α-D-mannopyranoside 6-(α-D-glucopyranosyl phosphate) (**7**) and 6-(α-D-galactopyranosyl phosphate) (**8**) were synthesized by condensation of the appropriate peracetylated glycosyl phosphate with *p*-nitrophenyl 2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside followed by alkaline deprotection. $^1$H-, $^{31}$P-, and $^{13}$C-n.m.r. spectroscopy were used to establish the structures of **7** and **8**, and to examine the conformational preferences about the phosphoric diester linkage.

## INTRODUCTION

The complex carbohydrate components of the glycoconjugates are known to mediate a wide variety of biological recognition phenomena. These ubiquitous structures represent many of the human blood-group antigenic determinants[1] and are known to function as receptors for both toxins[2] and bacteria[3]. The clearance of serum asialoglycoproteins by the hepatic D-galactose–2-acetamido-2-deoxy-D-galactose receptor[4], and the transport and uptake of lysozomal enzymes mediated by the D-mannose 6-phosphate marker[5] are but two striking and well-established examples of vital processes directed by "carbohydrate messages". In addition, the changes in cell-surface carbohydrates observed in both normal and abnormal cellular development[6] suggest, for these structures, important roles in intercellular recognition[7].

Recently, Koro and Marchase[8,9] have suggested that oligosaccharide determinants may also be involved in the localization of cell-surface glycoproteins. Evidence was obtained that retinal ligatin, a plasma membrane protein, functions as a baseplate for the attachment of peripheral glycoproteins to the cell surface by binding with high-mannose type oligosaccharide chains terminating in phosphoric diester-linked D-glucose. We report herein the synthesis of a glycoside of α-D-mannopyranose 6-(α-D-glucopyranosyl phosphate), the putative recognition marker. This phosphoric diester has also been found to be present as an important immunochemical determinant of the yeast *Hanselua polymorpha*[10]. We also report the synthesis of the 4′-epimer (where the D-glucosyl residue has been replaced by

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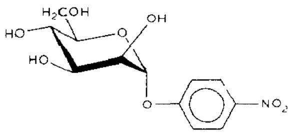
galactose) which will serve in characterizing the structural requirements for ligatin-binding. These phosphoric diesters were synthesized as the *p*-nitrophenyl glycosides **7** and **8** to allow for the preparation of affinity chromatography matrices for use in the isolation and study of ligatin.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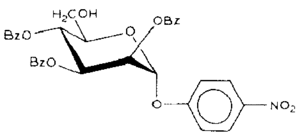
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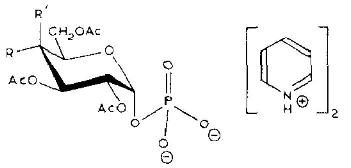
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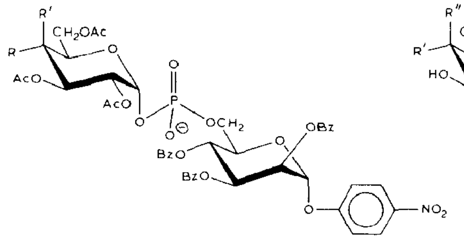
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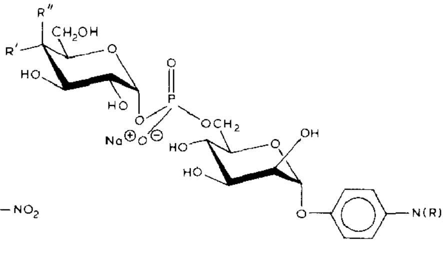
7 R = O, R' = OH, R'' = H
8 R = O, R' = H, R'' = OH
9 R = H, R' = OH, R'' = H

## RESULTS AND DISCUSSION

Our synthesis of the phosphoric diesters **7** and **8** is based on that of Cawley and Letters[11] who have reported the preparation of methyl α-D-mannopyranoside 6-(α-D-mannopyranosyl phosphate). This route takes advantage of the ready access to the anomerically pure peracetylated glycosyl phosphates provided by the MacDonald procedure[12]. A similar method has been used by Warren *et al.*[13] in the preparation of other carbohydrate-derived phosphoric diesters.

Successive treatment of *p*-nitrophenyl α-D-mannopyranoside with chloro-4-methoxyphenyldiphenylmethane and benzoyl chloride, followed by hydrolysis of the 4-methoxytrityl group in aqueous acetic acid at room temperature proved to be a convenient route to a mannose derivative **2** (57%) having OH-6 free for phosphoric diester formation. The primary position of the free hydroxyl group in **2** could be readily established from its $^1$H-n.m.r. spectrum for a solution in ($^2$H)chloroform where the signal for the OH proton ($D_2O$-exchangeable) appeared as doublet of doublets due to coupling with the two H-6 protons. Condensation[11] of **2** with the pyridinium salt of 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl phosphate[13] (**3**) in the presence of dicyclohexylcarbodiimide (DCC) provided only a low yield ($\sim$30%) of the protected phosphoric diester **5** which proved difficult to purify by chromatography. Under the same conditions, reaction of **2** with the peracetylated D-galactosyl phosphate[14] **4** furnished the diester **6** in a corresponding low yield.

A substantial improvement could be achieved by application of the more powerful condensing agent 3-nitro-1-(2,4,6-triisopropylbenzenesulfonyl)-1,2,4-triazole[15], which increased the yields of **5** and **6** to 68 and 71%, respectively. Also, it considerably simplified the chromatographic purification which was complicated, in the case of the DCC-promoted reactions, owing to the large quantity of dicyclohexylurea produced.

Deacylation of **5** and **6** was effected by triethylamine in aqueous methanol for 16 h at 4°. The $^1$H-n.m.r. spectra of the crude products from these reactions showed no evidence for either phosphate migration or hydrolysis despite the basic conditions used. The *p*-nitrophenyl glycosides **7** and **8** were obtained after desalting on Bio-Gel P-2 and conversion to the sodium salts by passage through Dowex 50 ($Na^+$) resin.

To provide the free amine **9**, the nitro group of **7** was reduced by catalytic hydrogenation in the presence of Adams' catalyst. The immobilization of **9** and the use of the resulting affinity support in the isolation and characterization of ligatin are reported elsewhere[16,17].

The position of the phosphate group in **7** and **8** could be conclusively established by the observation of the expected $^{31}$P–$^1$H and $^{31}$P–$^{13}$C coupling in the n.m.r. spectra. In the $^1$H-n.m.r. spectrum of **7**, $J_{5,6a}$ and $J_{5,6b}$ were 2.2 and 4.5 Hz, respectively, similar to the coupling constants observed[18] in the spectrum of methyl α-D-mannopyranoside and suggesting[19] important contributions of both the so called gg (H-5 antiperiplanar to O-6) and gt (C-4 antiperiplanar to O-6) hydroxymethyl group rotamers to the conformational equilibrium of the D-mannose residue. $J_{P,H\text{-}6a}$ and $J_{P,H\text{-}6b}$ were 5.2 and 6.2 Hz, respectively, suggesting very similar dihedral angles between the phosphorus atom and both the hydroxymethyl group protons, which requires the phosphorus atom to be nearly antiperiplanar to C-5. Indeed, application of the expression proposed by Lee and Sarma[20] for the dependence of $^3J_{P,O,CH}$ on the dihedral P–H angle $\theta$ ($J = 18.1 \cos^2\theta - 4.5 \cos\theta$) leads to the prediction that the conformational equilibrium about the C-6–O-6 bond involves the conformer with C-5 and P antiperiplanar to the extent of 80–85%. A coupling of 7.0 Hz was observed between the phosphorus atom and H-1′ of the D-glucosyl residue of **7** which would indicate[20] a dihedral angle of near 40° according to the expression just mentioned. The observation of a large long-range coupling between H-2′ and P ($^4J_{P,H\text{-}2'}$ 2.8 Hz) is strongly suggestive[21–24] of the near co-planarity (W-arrangement)[25] of the four bonds joining these atoms and requires the phosphorus atom to be nearly antiperiplanar to C-2′. Taken together, these data suggest that the phosphoric diester **7** is "stretched out" in aqueous solution, rather than having the D-glucosyl residue folded back towards the α-D-mannosyl unit which, in the natural-glycoprotein structures bound by ligatin, is at the end of a high-mannose oligosaccharide chain[8,9]. In this conformation, the phosphoric diester terminal group would be fully exposed to the solvent, making it an ideal candidate for a recognition marker.

## EXPERIMENTAL

*General methods.* — Melting points are uncorrected. Optical rotations were measured with a Perkin–Elmer 241 polarimeter at ambient temperatures (22 ±2°). Thin-layer chromatography was performed on precoated plates of silica gel (60-$F_{254}$, E. Merck, Darmstadt) with detection by quenching of fluorescence, or by charring, or both, after spraying with 5% $H_2SO_4$ in ethanol. Column chromatography was performed on Silica gel Merck 60 (40–63 $\mu$m) and Bio-Gel P-2 (200–400 mesh, Bio-Rad). $^1$H-N.m.r. spectra were recorded at 360 MHz (Bruker WM-360) with either tetramethylsilane ($Me_4Si$, $\delta$ 0 in $CDCl_3$) or acetone ($\delta$ 2.225 in $D_2O$) as internal standards at ambient temperature. $^{13}$C-N.m.r. spectra were recorded at 100 MHz (Bruker WH-400) with either internal $Me_4Si$ ($\delta$ 0 in $CDCl_3$) or external 1,4-dioxane ($\delta$ 67.4 in $D_2O$) as reference standards. $^{31}$P-N.m.r. spectra were recorded at 145 MHz (Bruker WM-360) with external 85% $H_3PO_4$ in $D_2O$ as reference ($\delta$ 0). $^{31}$P-decoupled $^1$H-n.m.r. spectra were recorded with a Bruker WH-400 equipped with a B-SV3-BX heteronuclear decoupler. The microanalyses were carried out by the Analytical Services Laboratory of this department.

p-*Nitrophenyl 2,3,4-tri-O-benzoyl-α-*D*-mannopyranoside* (**2**). — *p*-Nitrophenyl α-D-mannopyranoside[26] (**1**; 5.0 g, 16.6 mmol) was dissolved in anhydrous pyridine (50 mL) and added to chloro-4-methoxyphenyldiphenylmethane (*p*-anisylchlorodiphenylmethane; 7.7 g, 24.9 mmol). After 24 h, benzoyl chloride (7.0 mL, 60.3 mmol) was added and stirring was continued for an additional 5 h at room temperature. Methanol (20 mL) was then added to destroy the excess of reagents and, after 0.5 h, the solution was taken to dryness. The residual syrup was dissolved in dichloromethane (200 mL) and washed sequentially with water (200 mL), 5% HCl (200 mL), water (200 mL), saturated $NaHCO_3$ (200 mL), and finally twice with water (2 × 200 mL) before concentration to a syrup. This was dissolved in dichloromethane (25 mL) and 80% aqueous acetic acid (750 mL) was added. After 20 h at room temperature, the solvents were evaporated and the residue was purified by chromatography on silica gel in 1:3 ethyl acetate–hexane as eluant. Pure **2** (5.8 g, 57%) crystallized from ethyl acetate–hexane, m.p. 150–152°, $[\alpha]_D^{22}$ +5.7° (*c* 0.9, chloroform); t.l.c. $R_F$ 0.40 in 1:1 ethyl acetate–hexane; $^1$H-n.m.r. ($CDCl_3$): $\delta$ 8.3–7.2 (m, 19 H, arom.), 6.200 (dd, 1 H, $J_{2,3}$ 3.2, $J_{3,4}$ 10.0 Hz, H-3), 5.986 (dd, 1 H, $J_{4,5}$ 10.0 Hz, H-4), 5.970 (d, 1 H, $J_{1,2}$ 1.8 Hz, H-1), 5.903 (dd, 1 H, H-2), 4.089 (m, 1 H, H-5), 3.86–3.71 (m, 2 H, H-6a,6b), and 2.733 (dd, 1 H, $J_{OH,6a}$ 6.0, $J_{OH,6b}$ 8.0 Hz, $D_2O$ exchangeable, OH).

*Anal.* Calc. for $C_{33}H_{27}NO_{11}$: C, 64.60; H, 4.44; N, 2.28. Found: C, 64.45; H, 4.42; N, 2.34.

p-*Nitrophenyl 2,3,4-tri-O-benzoyl-α-*D*-mannopyranoside 6-(triethylammonium 2,3,4,6-tetra-O-acetyl-α-*D*-glucopyranosyl phosphate)* (**5**). — (*a*) *Using dicyclohexylcarbodiimide (DCC).* The pyridinium salt of 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl phosphate[13] (**3**, 530 mg, 0.90 mmol) was dried by repeated addition and evaporation of anhydrous pyridine and dissolved in the same solvent (5 mL).

To this was added the alcohol **2** (500 mg, 0.82 mmol), followed by DCC (1.35 g, 6.54 mmol), and the mixture was stirred for 18 h at 37°. After addition of 1:1 pyridine–water (10 mL) to eliminate the excess of DCC, the precipitated 1,3-dicyclohexylurea was removed by filtration and washed with pyridine. The filtrate was evaporated to dryness and the solid residue extracted with dichloromethane (20 × 5 mL). The combined extracts were evaporated and the residue was chromatographed on silica gel in 1:9 methanol–dichloromethane as eluent. The fractions containing the phosphoric diester **5** ($R_F$ 0.50 in 1:5 methanol–dichloromethane) were pooled and evaporated to provide a product (350 mg) whose $^1$H-n.m.r. spectrum showed 10–20% by weight of contaminating 1,3-dicyclohexylurea. Further purification of this material was not attempted.

(*b*) *Using 3-nitro-1-(2,4,6-triisopropylbenzenesulfonyl)-1,2,4-triazole (TPSNT)*. An anhydrous pyridine solution of **3** (420 mg, 0.72 mmol) was prepared as described under (*a*). To this solution was added the alcohol **2** (400 mg, 0.65 mmol), followed by the TPSNT reagent (620 mg, 1.63 mmol), and the reaction was kept for 7 h at 37°. After addition of 1:1 pyridine–water (10 mL), the solvent was evaporated and the residue chromatographed directly on silica gel in 1:9 methanol–dichloromethane as eluent. The fractions containing **5** were pooled and evaporated. The residue, dissolved in dichloromethane (10 mL), was passed through a column of Dowex 50-X8 ($H^+$) cation-exchange resin and the eluate immediately made neutral with triethylamine and evaporated to provide **5** (500 mg, 68%), $[\alpha]_D^{22}$ +51.3° (*c* 1.0, chloroform); $^1$H-n.m.r. ($CDCl_3$): $\delta$ 8.3–7.1 (m, 19 H, arom.), 6.039 (dd, 1 H, $J_{2,3}$ 3.5, $J_{3,4}$ 10.0 Hz, H-3), 5.928 (dd, 1 H, $J_{4,5}$ 10.0 Hz, H-4), 5.897 (d, $J_{1,2}$ 2.0 Hz, H-1), 5.872 (dd, 1 H , H-2), 5.676 (dd, 1 H, $J_{1',2'}$ 3.5, $J_{1',P}$ 7.5 Hz, H-1′), 5.502 (dd, 1 H, $J_{2',3'}$ 10.0, $J_{3',4'}$ 10.0 Hz, H-3′), 5.086 (dd, 1 H, $J_{4',5'}$ 10.0 Hz, H-4′), 4.882 (ddd, 1 H, $J_{2',P}$ 2.2 Hz, H-2′), 4.145 (m, 1 H, H-5′), 4.29–4.00 (5 H, H-5,6a,6b,6′a,6′b), 2.950 (q, 6 H, 3 $NCH_2CH_3$), 2.013, 1.982, 1.961, 1.909 (all s, 4 $COCH_3$), and 1.211 (t, 9 H, 3 $NCH_2CH_3$); $^{13}$C-n.m.r. ($CDCl_3$): $\delta$ 96.25 (C-1) and 91.91 ($J_{C,P}$ 5.1 Hz, C-1′); $^{31}$P-n.m.r. ($CDCl_3$): $\delta$ −4.6.

*Anal*. Calc. for $C_{53}H_{61}N_2O_{23}P$: C, 56.57; H, 5.47; N, 2.49. Found: C, 56.28; H, 5.30; N, 2.44.

p-*Nitrophenyl 2,3,4-tri-*O*-benzoyl-α-*D*-mannopyranoside 6-(triethylammonium 2,3,4,6-tetra-*O*-acetyl-α-*D*-galactopyranosyl phosphate)* (**6**). — (*a*) *Using DCC*. Reaction of the pyridinium salt of 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl phosphate[14] (**4**) with the alcohol **2**, as described for the corresponding preparation of **5**, afforded, after chromatography, the 1,3-dicyclohexylurea-contaminated phosphoric diester in only 31% yield.

(*b*) *Using TPSNT*. Condensation of **2** (300 mg, 0.49 mmol) and **4** (315 mg, 0.54 mmol) in the presence of TPSNT (465 mg, 1.22 mmol), as described for the preparation of **5**, provided, after conversion to its triethylammonium salt, the phosphoric diester **6** ($R_F$ 0.40 in 1:5 methanol–dichloromethane) as a hygroscopic, white solid (400 mg, 71%), $[\alpha]_D^{22}$ +40.4° (*c* 1.3, chloroform); $^1$H-n.m.r. ($CDCl_3$): $\delta$ 8.4–7.2 (m, 19 H, arom.), 6.020 (dd, 1 H, $J_{2,3}$ 3.2, $J_{3,4}$ 10.0 Hz, H-3), 5.908 (dd, 1 H, $J_{4,5}$

10.0 Hz, H-4), 5.907 (d, 1 H, $J_{1,2}$ 2 Hz, H-1), 5.857 (dd, 1 H, H-2), 5.713 (dd, 1 H, $J_{1',2'}$ 3.5, $J_{1',P}$ 7.5 Hz, H-1'), 5.384 (m, 2 H, H-3',4'), 5.126 (m, 1 H, H-2'), 4.429 (m, 2 H, H-5,5'), 4.153 (m, 2 H, H-6a,6b), 4.011 (dd, 1 H, $J_{5',6'a}$ 8.0, $J_{6'a,6'b}$ 11.0 Hz, H-6'a), 3.942 (dd, 1 H, $J_{5',6'b}$ 6.0 Hz, H-6'b), 2.900 (q, 6 H, 3 $\mathrm{NC}H_2\mathrm{CH_3}$), 2.130, 1.965, 1.948, 1.943 (all s, 12 H, 4 $\mathrm{OCOC}H_3$), and 1.189 (t, 9 H, 3 $\mathrm{NCH_2C}H_3$); $^{13}$C-n.m.r. ($CDCl_3$): $\delta$ 96.17 (C-1), 92.39 ($J_{C,P}$ 4.8 Hz, C-1'); $^{31}$P-n.m.r. ($CDCl_3$): $\delta$ −4.30.

*Anal.* Calc. for $C_{53}H_{61}N_2O_{23} \cdot H_2O$: C, 55.69; H, 5.55; N, 2.45. Found: C, 55.74; H, 5.37; N, 2.50.

p-*Nitrophenyl α*-D-*mannopyranoside 6-(sodium α*-D-*glucopyranosyl phosphate)* (**7**). — The protected phosphoric diester **5** (300 mg, 0.27 mmol) was kept in 1:2:1 triethylamine–methanol–water for 16 h at 4°, then the solvent was evaporated. The residue was freed from triethylamine by addition and evaporation of ethanol (3 × 10 mL) and was desalted by passage through a column (75 × 2 cm i.d.) of Bio-Gel P-2 in 10% aqueous ethanol as eluent. After evaporation of the solvent of the carbohydrate-containing fractions, the residue was converted into the sodium salt by passage through Dowex 50-X8 ($Na^+$) cation-exchange resin (10 g) and the aqueous solution lyophilized to provide **7** ($R_F$ 0.47 in 5:1 2-propanol–water) as a white powder (125 mg, 83%), $[\alpha]_D^{22}$ +123.1° (*c* 0.9, water); $^1$H-n.m.r. ($D_2O$): $\delta$ 8.267 (m, 2 H, $^3J \approx 9.5$ Hz, arom.), 7.286 (m, 2 H, $^3J \approx 9.5$ Hz, arom.), 5.761 (d, 1 H, $J_{1,2}$ 1.9 Hz, H-1), 5.440 (dd, 1 H, $J_{1',2'}$ 3.5, $J_{1',P}$ 7.0 Hz, H-1'), 4.202 (dd, 1 H, $J_{2,3}$ 3.5 Hz, H-2), 4.154–4.018 [3 H, including H-6a (4.122, ddd, $J_{5,6a}$ 4.5, $J_{6a,6b}$ 11.5, $J_{6a,P}$ 6.2 Hz), H-3 (4.079, dd, $J_{3,4}$ 9.7 Hz), and H-6b (4.042, ddd, $J_{5,6b}$ 2.2, $J_{6b,P}$ 5.2 Hz)], 3.888 (dd, 1 H, $J_{4,5}$ 9.8 Hz, H-4), 3.817–3.700 [5 H, including H-3' (3.726, dd, $J_{2',3'}$ 9.8, $J_{3',4'}$ 9.4 Hz) and H-5,5',6'a,6'b], 3.534 (ddd, 1 H, $J_{2',P}$ 2.8 Hz, H-2'), and 3.460 (dd, 1 H, H-4'); $^{13}$C-n.m.r. ($D_2O$): $\delta$ 161.79 ($CNO_2$), 142.30 (phenolic *C*O), 126.91 and 117.76 (arom.), 98.89 (C-1), 96.11 ($J_{C,P}$ 6 Hz, C-1'), 73.62, 73.45 (2 × C), 72.25 ($J_{C,P}$ 8 Hz, C-5), 71.03, 70.49, 70.06, 66.86, 65.12 ($J_{C,P}$ < 3 Hz, C-6), and 61.23; $^{31}$P-n.m.r. ($D_2O$): $\delta$ 0.05.

*Anal.* Calc. for $C_{18}H_{25}NNaO_{16}P \cdot H_2O$: C, 37.06; H, 4.66; N, 2.40. Found: C, 37.31; H, 4.57; N, 2.70.

p-*Nitrophenyl α*-D-*mannopyranoside 6-(sodium α*-D-*galactopyranosyl phosphate)* (**8**). — The phosphoric diester **6** (250 mg, 0.22 mmol) was deprotected and purified as described for the preparation of **7**, yielding **8** ($R_F$ 0.42 in 5:1 2-propanol–water) as a white lyophilized powder (105 mg, 84%), $[\alpha]_D^{22}$ +87.9° (*c* 0.9, water); $^1$H-n.m.r. ($D_2O$): $\delta$ 8.269 (m, 2 H, $^3J \approx 9$ Hz, arom.), 7.293 (m, 2 H, $^3J \approx 9$ Hz, arom.), 5.756 (d, 1 H, $J_{1,2}$ 1.9 Hz, H-1), 5.473 (dd, 1 H, $J_{1',2'}$ 3.0, $J_{1',P}$ 6.9 Hz, H-1'), 4.191 (dd, 1 H, $J_{2,3}$ 3.5 Hz, H-2), 4.12–3.99 [4 H, including H-6a,6b, H-3 (4.084, dd, $J_{3,4}$ 9.0 Hz) and H-5' (4.006, ddd, $J_{4',5'}$ 1, $J_{5',6'a}$ 6.0 $J_{5',6'b}$ 6.0 Hz)], 3.975 (dd, 1 H, $J_{3',4'}$ 3.0 Hz, H-4'), 3.869 (dd, 1 H, $J_{4,5}$ 9.5 Hz, H-4), 3.82–3.74 (3 H, H-2',3',5), and 3.670 (d, 2 H, $J_{5',6'a} = J_{5',6'b} = 6.0$ Hz, H-6'a,6'b); $^{13}$C-n.m.r. ($D_2O$): $\delta$ 161.32 ($CNO_2$), 142.70 (phenolic *C*O), 126.40 and 117.23 (arom.), 98.42 (C-1), 95.81 ($J_{C,P}$ 7 Hz, C-1'), 73.00 ($J_{C,P}$ 7 Hz, C-5), 72.06, 70.53, 69.99, 69.47, 69.42, 68.57 ($J_{C,P}$ 8

Hz, C-2'), 66.41, 64.69 ($J_{C,P}$ 4 Hz, C-6), and 61.25; $^{31}$P-n.m.r. ($D_2O$): $\delta$ 0.18.

*Anal.* Calc. for $C_{18}H_{25}NNaO_{16}P \cdot H_2O$: C, 37.06; H, 4.66; N, 2.40. Found: C, 36.70; H, 4.47; N, 2.49.

p-*Aminophenyl α-*D*-mannopyranoside 6-(sodium α-*D*-glucopyranosyl phosphate* (**9**). — The nitrophenyl glycoside **7** (50 mg, sodium salt) was dissolved in 20% ethanol (2 mL) and Adams' catalyst ($PtO_2$, 30 mg) was added. After being stirred under hydrogen at 0.1 MPa for 40 min, the catalyst was removed by filtration on paper and the solvent was evaporated. The product was passed through a column of Bio-Gel P-2 (in 10% ethanol as eluent). Fractions containing **9** ($R_F$ 0.24 in 5:1 2-propanol–water) were pooled, evaporated, and converted into the sodium salt by passage through Dowex 50-X8 ($Na^+$) cation-exchange resin to provide a clear solution which darkened somewhat on evaporation. The residue was obtained as a pale brown solid (41 mg) after lyophilization; $^1$H-n.m.r. ($D_2O$): $\delta$ *(inter alia)* 7.15–6.85 (4 H, arom.), 5.505 (dd, 1 H, $J_{1',2'}$ 3.5, $J_{1',P}$ 6.7 Hz, H-1'), 5.465 (d 1 H, $J_{1,2}$ 1.8 Hz, H-1), 4.118 (ddd, 1 H, $J_{5,6a}$ 2.4, $J_{6a,6b}$ 11.5, $J_{6a,P}$ 5.0 Hz, H-6a), and 3.571 (ddd, 1 H, $J_{2',3'}$ 9.5, $J_{2',P}$ 3.0 Hz, H-2'); the $^1$H-n.m.r. spectrum indicated a level of purity exceeding 95% (in carbohydrate).

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