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Note

Synthesis of trisaccharide glycosides related to nigeran

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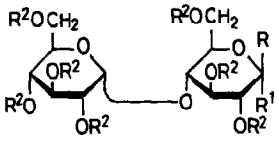
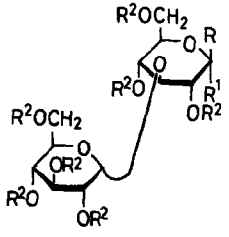
Nigeran, a polysaccharide produced intracellularly by *Aspergillus niger*, is an unbranched α-D-glucan containing mainly alternating (1→3) and (1→4) linkages[1,2]. We now report the synthesis of the trisaccharide glycosides methyl *O*-α-D-glucopyranosyl-(1→4)-*O*-α-D-glucopyranosyl-(1→3)-α-D-glucopyranoside (**17**) and methyl *O*-α-D-glucopyranosyl-(1→3)-*O*-α-D-glucopyranosyl-(1→4)-α-D-glucopyranoside (**20**), which are structurally related to nigeran. The reducing trisaccharides corresponding to **17** and **20** have been isolated after partial acid hydrolysis of nigeran[2].

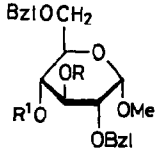
Condensation of hepta-*O*-acetyl-α-maltosyl bromide (**1**) with allyl alcohol in the presence of mercuric cyanide gave the allyl glycoside **2** (76%), *O*-deacetylation of which afforded allyl β-maltoside (**3**). Treatment of **3** with benzyl bromide and sodium hydride in *N*,*N*-dimethylformamide[3] gave the hepta-*O*-benzyl derivative **4**, which was *O*-deallylated with palladium chloride–sodium acetate[4] in aqueous



1 R = H, R^1 = Br, R^2 = Ac
2 R = OAll, R^1 = H, R^2 = Ac
3 R = OAll, R^1 = R^2 = H
4 R = OAll, R^1 = H, R^2 = Bzl
5 R, R^1 = H, OH; R^2 = Bzl
6 R = H, R^1 = Cl, R^2 = Bzl



7 R = H, R^1 = Br, R^2 = Ac
8 R = OAll, R^1 = H, R^2 = Ac
9 R = OAll, R^1 = R^2 = H
10 R = OAll, R^1 = H, R^2 = Bzl
11 R, R^1 = H, OH; R^2 = Bzl
12 R = H, R^1 = Cl, R^2 = Bzl



13 R = H, R^1 = Bzl
14 R = Bzl, R^1 = H

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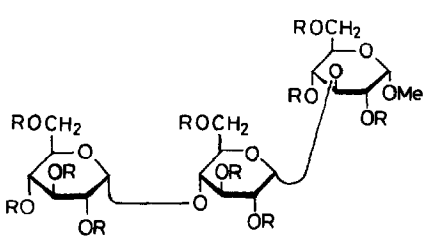
acetic acid. The resulting product (**5**) was converted into the corresponding α-glycosyl chloride **6** with oxalyl chloride in dichloromethane in the presence of a catalytic amount of *N,N*-dimethylformamide[5].

Glycosylation of methyl 2,4,6-tri-*O*-benzyl-α-D-glucopyranoside[6] (**13**) with **6** in ether in the presence of silver perchlorate[7–9] and a molecular sieve[8,9] afforded 64% of the trisaccharide derivative **15** after column chromatography. The ^{13}C-n.m.r. spectrum of **15** contained signals for anomeric carbons at 97.7, 96.9, and 96.6 p.p.m., indicating[10] the configuration at each anomeric carbon atom to be α. Catalytic hydrogenolysis (Pd/C) of **15**, followed by acetylation, gave the deca-acetate **16**, which was *O*-deacetylated to furnish **17**.

The reaction of hepta-*O*-acetyl-α-nigerosyl bromide[11] (**7**) with allyl alcohol, as for **1**, gave the allyl glycoside **8** (78%), which was transformed into the α-glycosyl chloride **12** by a reaction sequence (→**9**→**10**→**11**→**12**) similar to that described above. Condensation of methyl 2,3,6-tri-*O*-benzyl-α-D-glucopyranoside[12] (**14**) with **12**, as for the preparation of **15**, gave the trisaccharide derivative **18** (62% after column chromatography), the ^{13}C-n.m.r. spectrum of which contained signals for anomeric carbons at 97.6, 97.1, and 95.5 p.p.m., in accord[10] with the α configuration at each anomeric center. Hydrogenolysis of **18**, followed by acetylation (→**19**), and then *O*-deacetylation furnished **20**.



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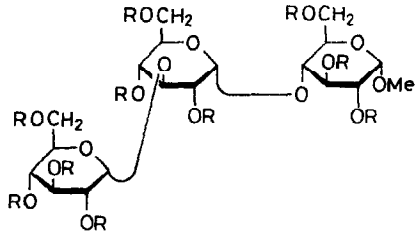
16 R = Ac
17 R = H



18 R = Bzl
19 R = Ac
20 R = H

EXPERIMENTAL

General. — These were the same as those described previously[4]. The solvent systems hexane–ethyl acetate (*A*, 4:1; *B*, 2:1; and *C*, 2:3) were used for chromatography.

Allyl 2,3,6-tri-O-acetyl-4-O-(2,3,4,6-tetra-O-acetyl-α-D-glucopyranosyl)-β-D-glucopyranoside (**2**). — A mixture of **1** (10.5 g, 15 mmol) and mercuric cyanide (3.79 g, 15 mmol) in dry allyl alcohol (50 mL) was stirred at 50° for 1 h, and then concentrated to a syrup which was extracted with dichloromethane. The extract was washed successively with water, aqueous potassium bromide, and water, dried,

and concentrated. The residue was recrystallised twice from ethanol to give **2** (7.75 g, 76%), m.p. 109–110°, $[\alpha]_D^{26}$ +47° (*c* 1.5, chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 170.2–169.1 (C=O), 133.2 and 117.4 ($CH=CH_2$), 99.0 (C-1), 95.5 (C-1′), and 20.85–20.5 ($COCH_3$).

Anal. Calc. for $C_{29}H_{40}O_{18}$: C, 51.48; H, 5.96. Found: C, 51.42; H, 5.90.

Allyl 4-O-α-D-glucopyranosyl-β-D-glucopyranoside (**3**). — A solution of **2** (6.85 g) in methanol (70 mL) was treated with M sodium methoxide (1 mL). The solution was kept for 2 h at room temperature, neutralised with Amberlite IR-120 (H^+) resin, filtered, and concentrated, to give amorphous **3** (3.36 g, 94%), $[\alpha]_D^{26}$ +63° (*c* 3.3, water). ^{13}C-N.m.r. data (D_2O): δ 135.8 and 121.1 ($CH=CH_2$), 103.5 (C-1), and 102.1 (C-1′).

Anal. Calc. for $C_{15}H_{26}O_{11}$: C, 47.12; H, 6.85. Found: C, 47.05; H, 6.96.

2,3,6-Tri-O-benzyl-4-O-(2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-D-glucopyranose (**5**). — A solution of **3** (2.21 g) in *N,N*-dimethylformamide (50 mL) was stirred with sodium hydride (4.86 g; 50% mineral oil) for 1 h at room temperature and then cooled to 0°. Benzyl bromide (9.5 mL) was added dropwise and the mixture was stirred overnight at room temperature. Methanol was then added to decompose the excess of hydride, most of the solvent was evaporated, and a solution of the residue in dichloromethane was washed with water, dried, and concentrated. The residue was subjected to column chromatography (solvent *A*), to give allyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-β-D-glucopyranoside (**4**), isolated as a syrup (5.38 g, 92%), $[\alpha]_D^{26}$ +28° (*c* 3, chloroform).

A mixture of **4** (3.84 g), palladium chloride (0.74 g), and sodium acetate (1.72 g) in acetic acid–water (20:1, 30 mL) was stirred overnight at room temperature. Insoluble material was collected on a Celite pad and washed with methanol, and the combined filtrate and washings were concentrated. A solution of the residue in dichloromethane was washed successively with water, aqueous sodium hydrogencarbonate, and water, dried, and concentrated. Column chromatography (solvent *B*) of the residue gave amorphous **5** (2.88 g, 78%), $[\alpha]_D^{26}$ +39.5° (*c* 1.5, chloroform).

Anal. Calc. for $C_{61}H_{64}O_{11}$: C, 75.29; H, 6.63. Found: C, 75.37; H, 6.72.

Methyl O-α-D-glucopyranosyl-(1→4)-O-α-D-glucopyranosyl-(1→3)-α-D-glucopyranoside (**17**). — A solution of oxalyl chloride (0.65 mL) in dichloromethane (5 mL) was added dropwise at 0° to a solution of **5** (2.45 g) in dichloromethane (20 mL) containing *N,N*-dimethylformamide (0.06 mL). The mixture was kept for 2 h at room temperature and then concentrated. A solution of the residue in hexane–ethyl acetate (1:1, 30 mL) was filtered through a layer of silica gel (8 g) which was washed with hexane–ethyl acetate (1:1, 10 mL). The combined filtrate and washings were concentrated to give 2,3,6-tri-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-α-D-glucopyranosyl chloride (**6**) as an amorphous powder (2.37 g, 95%), $[\alpha]_D^{26}$ +89.5° (*c* 1.6, chloroform), which was used in the glycosylation step without purification. ^{1}H-N.m.r. data ($CDCl_3$): δ 6.07 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1) and 5.63 (d, 1 H, $J_{1',2'}$ 3.5 Hz, H-1′).

A solution of **6** (2.20 g, 2.2 mmol) in ether (20 mL) was added dropwise during 30 min at −10° to a stirred mixture of **13** (0.68 g, 1.5 mmol), silver perchlorate (0.51 g, 2.5 mmol), and powdered molecular sieve Type 4A (5 g) in ether (30 mL). After being stirred at 0° for 1 h, the mixture was filtered through a Celite pad, which was then washed with ether. The combined filtrate and washings were washed successively with water, aqueous sodium hydrogencarbonate, and water, dried, and concentrated. Column chromatography (solvent *B*) of the residue gave methyl *O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-α-D-glucopyranosyl)-(1→3)-2,4,6-tri-*O*-benzyl-α-D-glucopyranoside (**15**), isolated as a syrup (1.33 g, 64%), $[\alpha]_D^{26}$ +72° (*c* 1.3, chloroform).

A solution of **15** (1.16 g) in acetic acid (20 mL) was hydrogenated in the presence of 10% Pd/C (2 g) at normal pressure for 3 days at room temperature, and then filtered through a Celite pad which was washed with methanol. The combined filtrate and washings were concentrated and the residue was treated with acetic anhydride–pyridine (1:1, 10 mL) overnight at room temperature. Column chromatography (solvent *C*) of the product gave methyl *O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-acetyl-α-D-glucopyranosyl)-(1→3)-2,4,6-tri-*O*-acetyl-α-D-glucopyranoside (**16**; 0.70 g, 91%), $[\alpha]_D^{26}$ +150° (*c* 1.7, chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 96.8, 95.1, and 94.9 (C-1,1′,1″).

O-Deacetylation of **16** (0.65 g), as described for **2**, afforded **17** as an amorphous powder (0.34 g, 94%), $[\alpha]_D^{26}$ +200.5° (*c* 1.9, water). ^{13}C-N.m.r. data (D_2O): δ 102.3, 102.0, and 101.4 (C-1,1′,1″), 82.5 (C-3), 79.6 (C-4′), 75.9, 75.5, 75.2, 74.4, 74.1, 74.0, 73.0, 72.6, 72.5, 72.0, 63.2 and 63.05 (2 C) (C-6,6′,6″), and 57.7 (OMe).

Anal. Calc. for $C_{19}H_{34}O_{16}$: C, 44.02; H, 6.61. Found: C, 43.80; H, 6.79.

*Allyl 2,4,6-tri-*O*-acetyl-3-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)-β-D-glucopyranoside* (**8**). — A mixture of **7** (4.5 g, 6.4 mmol) and mercuric cyanide (1.63 g, 6.5 mmol) in allyl alcohol (20 mL) and dry benzene (20 mL) was stirred for 1 h at 60°. The mixture was processed as described for the preparation of **2**, and the product was recrystallised twice from ethanol to give **8** (3.39 g, 78%), m.p. 106–107°, $[\alpha]_D^{26}$ +57° (*c* 1.4, chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 170.3–168.7 (C=O), 133.2 and 117.2 ($CH{=}CH_2$), 99.5 (C-1), 96.0 (C-1′), and 20.7–20.5 ($COCH_3$).

Anal. Calc. for $C_{29}H_{40}O_{18}$: C, 51.48; H, 5.96. Found: C, 51.57; H, 6.04.

*Allyl 3-*O*-α-D-glucopyranosyl-β-D-glucopyranoside* (**9**). — *O*-Deacetylation of **8** (3.11 g), as described for **2**, afforded **9** (1.57 g, 89%), m.p. 185–186° (from ethanol), $[\alpha]_D^{26}$ +83° (*c* 2.1, water). ^{13}C-N.m.r. data (D_2O): δ 135.7 and 120.55 ($CH{=}CH_2$), and 102.0 and 101.9 (C-1,1′).

Anal. Calc. for $C_{15}H_{26}O_{11}$: C, 47.12; H, 6.85. Found: C, 47.21; H, 6.90.

*2,4,6-Tri-*O*-benzyl-3-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-D-glucopyranose* (**11**). — Compound **9** was treated in *N*,*N*-dimethylformamide (30 mL) with sodium hydride (3.19 g; 50% mineral oil) and benzyl bromide (6.3 mL), and processed as described for the preparation of **4**. Column chromatography (solvent *A*) of the product gave allyl 2,4,6-tri-*O*-benzyl-3-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-β-D-glucopyranoside (**10**), isolated as a syrup (3.50 g, 91%), $[\alpha]_D^{26}$ +33° (*c* 2.2, chloroform).

Compound **10** (3.39 g) was treated in acetic acid–water (20:1, 25 mL) with palladium chloride (0.65 g) and sodium acetate (1.5 g), and processed as described for the preparation of **5**. Column chromatography (solvent *B*) of the product afforded amorphous **11** (2.51 g, 77%), $[\alpha]_D^{26}$ +30.5° (*c* 1.4, chloroform).

Anal. Calc. for $C_{61}H_{64}O_{11}$: C, 75.29; H, 6.63. Found: C, 75.40; H, 6.70.

Methyl O-α-D-*glucopyranosyl-(1→3)*-O-α-D-*glucopyranosyl-(1→4)*-α-D-*glucopyranoside* (**20**). — A solution of **11** (2.35 g) in dichloromethane (20 mL) and *N,N*-dimethylformamide (0.06 mL) was treated with a solution of oxalyl chloride (0.62 mL) in dichloromethane (5 mL), and processed as described for the preparation of **6**, to give 2,4,6-tri-*O*-benzyl-3-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-α-D-glucopyranosyl chloride (**12**) as an amorphous powder (2.25 g, 94%), $[\alpha]_D^{26}$ +107° (*c* 1, chloroform), which was used in the coupling reaction without purification. ^{1}H-N.m.r. data ($CDCl_3$): δ 6.16 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1) and 5.54 (d, 1 H, $J_{1',2'}$ 3.5 Hz, H-1').

A solution of **12** (2.15 g, 2.2 mmol) in ether (20 mL) was added at −10° to a stirred mixture of **14** (0.67 g, 1.4 mmol), silver perchlorate (0.49 g, 2.4 mmol), and powdered molecular sieve Type 4A (5 g) in ether (30 mL), as described for the preparation of **15**. Column chromatography (solvent *B*) of the product gave methyl *O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-benzyl-α-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (**18**), isolated as a syrup (1.27 g, 62%), $[\alpha]_D^{26}$ +57° (*c* 1.2, chloroform).

Hydrogenolysis of **18** (1.12 g), as described for **15**, followed by acetylation of the product and column chromatography (solvent *C*), as described for the preparation of **16**, gave methyl *O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)-(1→3)-*O*-(2,4,6-tri-*O*-acetyl-α-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-acetyl-α-D-glucopyranoside (**19**) as an amorphous powder (0.66 g, 89%), $[\alpha]_D^{26}$ +144° (*c* 2.2, chloroform). ^{13}C-N.m.r. data ($CDCl_3$): δ 96.6, 95.6, and 95.4 (C-1,1',1'').

O-Deacetylation of **19** (0.61 g), as described for **2**, gave amorphous **20** (0.31 g, 91%), $[\alpha]_D^{26}$ +203.5° (*c* 0.9, water). ^{13}C-N.m.r. data (D_2O): δ 102.6 and 101.6 (2 C) (C-1,1',1''), 82.35 (C-3'), 79.9 (C-4), 76.0, 75.5, 75.0, 74.3, 74.25, 73.6, 73.0, 72.7, 72.3, 72.0, 63.1 (3 C, C-6,6',6''), and 57.7 (OMe).

Anal. Calc. for $C_{19}H_{34}O_{16}$: C, 44.02; H, 6.61. Found: C, 43.91; H, 6.74.

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