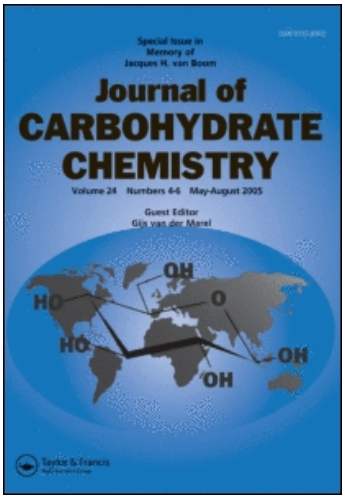

Journal of Carbohydrate Chemistry

Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713617200

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To cite this Article Norberg, Thomas , Walding, Marianne and Westman, Erik(1988) 'Synthesis of the Methyl, 1-Octyl, and D-Trifluoro-Acetamidophentlethyl α-Glycosides of 3,6-Di-O-(α-D-Galactopyranosyl)-D-Glucopyranose and an Acyclic Analogue Thereof', Journal of Carbohydrate Chemistry, 7: 2, 283 — 292
To link to this Article: DOI: 10.1080/07328308808058925
URL: http://dx.doi.org/10.1080/07328308808058925

J. CARBOHYDRATE CHEMISTRY, 7(2), 283-292 (1988)

SYNTHESIS OF THE METHYL, 1-OCTYL, AND p-TRIFLUORO-ACETAMIDOPHENYLETHYL α-GLYCOSIDES OF 3,6-DI-O-(α-D-GALACTOPYRANOSYL)-D-GLUCOPYRANOSE AND AN ACYCLIC ANALOGUE THEREOF

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Received June 30, 1987 - Final Form October 22, 1987

ABSTRACT

The title trisaccharides were synthesized from a common trisaccharide thioglycoside derivative, which was, in turn, prepared from monosaccharide thioglycoside precursors. An acyclic analogue, methyl 3-O-(α-D-galactopyranosyl)-6-O-[(2'-hydroxyethyl)oxymethyl]-α-D-glucopyranoside, which carries a 2'-hydroxyethyloxymethyl group in place of the 6-O-galactosyl residue, was also synthesized.

INTRODUCTION

The core region of the lipopolysaccharide from Salmonella bacteria has the structure[1] shown in FIG. 1. This oligosaccharide is recognized as a binding site by bacteriophages at the initial stage of infection. Thus, mutant Salmonella bacteria lacking certain structural elements in the core region are resistant to infection by phages. To study in more detail the specific binding between bacteriophages and the core oligosaccharide, synthetic oligosaccharides were needed. We have synthesized the methyl trisaccharide 10 and its acyclic analogue 14 to be used in equilibrium dialysis binding experiments for comparing the binding of the rigid 10, its more flexible partial structure 14, and the entire core oligosaccharide. In connection with this work, the octyl (11) and

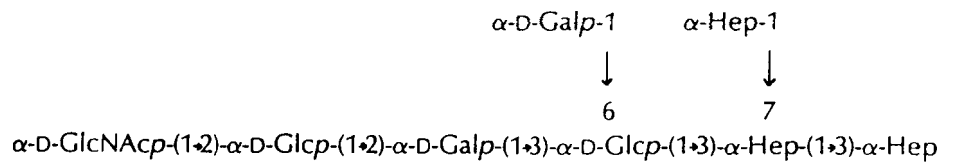


Figure 1. The structure of the Salmonella core oligosaccharide. Hep = L-*glycero*-D-mannoheptopyranoside.

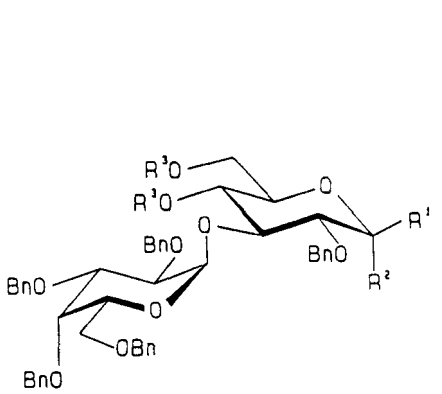
p-trifluoroacetamidophenylethyl (12) glycosides were also synthesized for studies requiring attachment of the synthetic oligosaccharide to liposomes or proteins. The results of the binding studies will be published elsewhere. The 8-methoxycarbonyloctyl α-glycoside of 3,6-di-O-(α-D-galactopyranosyl)-D-glucopyranose has been synthesized before,[2] by another route than ours.

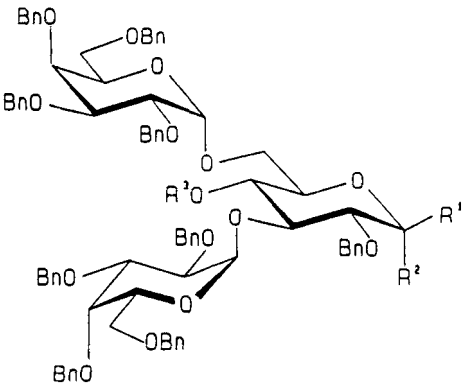
RESULTS AND DISCUSSION

The synthetic strategy adopted was based on methyl 1-thioglycoside intermediates.[3] Thus, the methyl, 1-octyl, and p-trifluoroacetamidophenylethyl glycosides 10, 11, and 12 were synthesized from the protected trisaccharide 1-thioglycosides 5 or 6. The methyl disaccharide 4, used in the preparation of the acyclic analog 14, was prepared from the corresponding disaccharide 1-thioglycoside 1. The oligosaccharide 1-thioglycosides were in turn prepared from known monosaccharide 1-thioglycoside derivatives. The following synthetic steps were carried out:

2,3,4,6-Tetra-O-benzyl-α-D-galactopyranosyl bromide, prepared from the corresponding 1-thioglycoside,[4] was used in a halide-ion promoted glycosidation[5] of methyl 4,6-O-benzylidene-2-O-benzyl-1-thio-β-D-glucopyranoside.[6] This gave the disaccharide derivative 1 (87% yield), which, when treated with aqueous acetic acid, gave the disaccharide diol 2 in 84% yield. A second halide-ion promoted glycosidation, using the same glycosyl donor and the diol 2 as an acceptor gave the trisaccharide 5 in 89% yield. Acetylation of 5 with acetic anhydride-pyridine gave 6 (98% yield). The NMR signal for H-4 (5.11 ppm) in 6 was shifted downfield compared to the corresponding signal in 5 (3.58 ppm). This shift in the NMR signal indicated the position of acetylation in 6, and thus indirectly that glycosidation of

5 had occured in the 6-position. Treatment of 6, first with bromine, and then with tetraethylammonium bromide and methanol gave the trisaccharide glycoside 7 in 63% yield. Similar treatment of 5, first with bromine and then with 1-octanol or p-trifluoroacetamidophenylethanol,[7] gave 8 (79% yield) and 9 (82% yield), respectively. Deprotection of 7 (treatment with methanolic sodium methoxide followed by catalytic hydrogenolysis with palladium on charcoal) as well as of 8 and 9 (catalytic hydrogenolysis) gave the target trisaccharide glycosides 10, 11, and 12 in 98, 98, and 92% yields, respectively. The ^{13}C NMR signals for C-3 and C-6 in these derivatives were shifted significantly downfield as compared to the corresponding signals in methyl α-D-glucopyranoside.[8] This is consistent with the 3,6-substitution pattern.

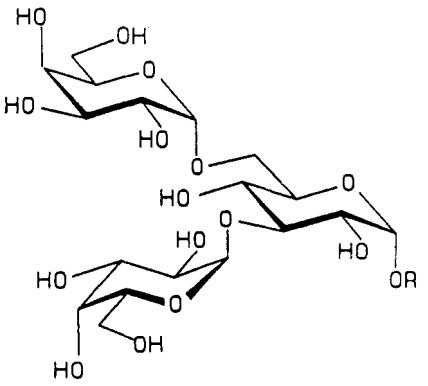




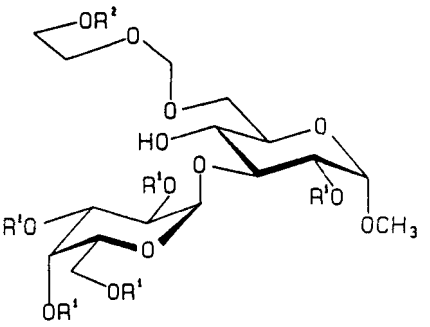
1) $R^1 = SCH_3$ $R^2 = H$ $R^3 = PhCH<$
2) $R^1 = SCH_3$ $R^2 = H$ $R^3 = H$
3) $R^1 = H$ $R^2 = OCH_3$ $R^3 = PhCH<$
4) $R^1 = H$ $R^2 = OCH_3$ $R^3 = H$

5) $R^1 = SCH_3$ $R^2 = H$ $R^3 = H$
6) $R^1 = SCH_3$ $R^2 = H$ $R^3 = Ac$
7) $R^1 = H$ $R^2 = OCH_3$ $R^3 = Ac$
8) $R^1 = H$ $R^2 = O(CH_2)_7CH_3$ $R^3 = H$
9) $R^1 = H$ $R^2 = OCH_2CH_2PhNHCOCF_3$ $R^3 = H$

For preparation of the acyclic analog 14, the disaccharide 1 was treated first with bromine, then with tetraethylammonium bromide and methanol to give 3 in 62% yield. This derivative was then treated with aqueous acetic acid to give the diol 4 (96% yield), which was reacted with 2'-(benzoyloxyethyl)-oxymethyl chloride[9] and molecular sieves to give the monosubstitution product 13 in 67% yield. Deprotection (catalytic hydrogenolysis with palladium on charcoal followed by treatment with methanolic sodium methoxide) gave the target compound 14 (95% yield). The 3,6 substitution pattern was established by the ^{13}C NMR spectrum of 14 in a manner similar to that used for 10.



10) R = CH_3
11) R = $(CH_2)_7CH_3$
12) R = $CH_2CH_2PhNHCOCF_3$



13) R' = Bn R² = Bz
14) R' = R² = H

EXPERIMENTAL

General Procedures. Melting points are corrected. Concentrations were performed at 1-2 kPa at <40°C (bath). Optical rotations were recorded for 0.5-1.0 % solutions in chloroform, unless otherwise stated, using a Perkin-Elmer 241 polarimeter. NMR spectra were recorded at 25 °C for solutions in $CDCl_3$ unless otherwise stated using JEOL JNM FX-100 or Bruker AM 500 instruments. The following reference signals were used: TMS δ 0.00 (^{13}C and ^1H in $CDCl_3$), Me_2CO δ 2.225 (^1H in D_2O), dioxane δ 67.4 (^{13}C in D_2O). Only selected NMR data are reported. All ^1H assignments were corroborated by 2-D COSY or decoupling experiments. In the NMR assignments below, atoms of glucose carry no superscript, while atoms of 3-linked galactose and 6-linked galactose carry the ' and " superscripts, respectively. The FAB-MS spectra were recorded with a VG ZAB-SE mass spectrometer. The primary beam consisted of xenon atoms with a maximum energy of 8 keV. The samples were dissolved in thioglycerol and the positive ions were extracted and accelerated over a potential of 10 kV. TLC was performed on silica gel F_{254} (Merck) with detection by UV light when applicable or by spraying with 5% sulfuric acid and then heating. Column chromatography was performed on silica gel 60 (0.04-0.063 mm, Merck) with loadings in the range 1/25-1/100 and elution with toluene-ethyl acetate mixtures unless otherwise stated. Organic solutions were dried over $MgSO_4$. Molecular sieves (4Å, Union Carbide) were desiccated in a vacuum at 300°C and ground immediately before use.

Dowex-50 (H^+) ion exchange resin was washed extensively with methanol before use.

Methyl 2-O-Benzyl-3-O-(2,3,4,6-tetra-O-benzyl-α-D-galactopyranosyl)--4,6-O-benzylidene-1-thio-β-D-glucopyranoside (1). Bromine (0.30 mL, 5.84 mmol) in dichloromethane (2 mL) was added to a stirred and cooled (ice) mixture of methyl 2,3,4,6-tetra-O-benzyl-1-thio-β-D-galactopyranoside[4] (3.0 g, 5.26 mmol), dichloromethane (26 mL) and molecular sieves (3.0 g). After 45 min, the mixture was filtered, concentrated, and co-concentrated with toluene. The residue was mixed with a solution of methyl 2-O-benzyl--4,6-O-benzylidene-1-thio-β-D-glucopyranoside[7] (0.68 g, 1.79 mmol) and tetraethylammonium bromide (1.25 g, 5.95 mmol) in dichloromethane (14 mL). Molecular sieves (3.0 g) were added, and the mixture was stirred overnight at room temperature. Pyridine (1.0 mL) was added, and the mixture was filtered. The filtrate was washed with aqueous 1M sulfuric acid, aqueous sodium hydrogen carbonate, and water, dried, and concentrated. The residue was purified by column chromatography to give syrupy 1 (1.39 g, 87%), $[\alpha]_D$ +22°. ^{13}C NMR data: δ 12.9 (SCH_3), 68.6, 68.8, 69.8, 71.9, 72.7, 73.0, 74.8, 75.3, 75.4, 75.5, 75.7, 76.0, 78.0, 79.6, 82.4 (C-2,3, 4,5,6, C-2',3',4',5',6', $PhCH_2$), 86.1 (C-1), 96.4 (PhCH), 101.6 (C-1'). ^1H NMR data: δ 2.24 (s, SCH_3), 4.41 (d, $J_{1,2}$ 9.7 Hz, H-1), 5.45 (s, PhCH), 5.70 (d, $J_{1',2'}$ 3.4 Hz, H-1').

Methyl 2-O-Benzyl-3-O-(2,3,4,6-tetra-O-benzyl-α-D-galactopyranosyl)--1-thio-β-D-glucopyranoside (2). A solution of 1 (0.59 g) in 80 % aqueous acetic acid (20 mL) was stirred at 60 °C for 2 h. Then water was added and the mixture was extracted twice with dichloromethane. The organic layer was washed with water, aqueous sodium hydrogen carbonate, and water, dried, and concentrated. The residue was crystallized from methanol-water to give 2 (0.44 g, 84%), mp 102-103 °C, $[\alpha]_D$ +28°. ^{13}C NMR data: δ 13.2 (SCH_3), 88.4 (C-1), 101.4 (C-1').
Anal. Calcd for $C_{48}H_{54}O_{10}S$: C, 70.1; H, 6.6; S, 3.9. Found: C, 69.8; H, 6.6, S, 3.9.

Methyl 2-O-Benzyl-3-O-(2,3,4,6-tetra-O-benzyl-α-D-galactopyranosyl)--4,6-O-benzylidene-α-D-glucopyranoside (3). Bromine (0.044 mL, 0.86 mmol) was added to a stirred and cooled (ice) mixture of 1 (530 mg, 0.59 mmol), dichloromethane (10 mL), and molecular sieves (1.5 g). After 1 h, 1-hexene (0.05 mL) was added, then tetraethylammonium bromide (980 mg, 4.67 mmol). Stirring was continued at room temperature for 1 h, then methanol (0.1 mL, 2.47 mmol) was added, and stirring was continued overnight. Pyridine (1.0 mL) was added, and the mixture was processed as

described for the preparation of 1. After column chromatography, pure 3 (324 mg, 62%) was obtained. Crystallization from toluene-hexane gave material with mp 116 °C, $[\alpha]_D$ +53°. ^{13}C NMR data: δ 55.2 (OCH_3), 61.8, 68.7, 72.4, 75.2, 75.7, 78.2, 78.3, 83.0 (C-2,3,4,5, C-2', 3', 4', 5'), 68.4, 69.2, 71.6, 72.9, 73.0, 73.5, 74.9 (C-6, C-6', $PhCH_2$) 96.7 (PhCH), 98.8 (C-1), 101.8 (C-1'). ^1H NMR data: δ 3.33 (s, OCH_3), 4.55 (d, J 3.6 Hz, H-1), 5.42 (s, PhCH), 5.61 (d, J 3.7 Hz, H-1').
Anal. Calcd for $C_{55}H_{58}O_{11}$: C, 73.8; H, 6.5. Found: C, 73.4; H, 6.5.

Methyl 2-O-Benzyl-3-O-(2,3,4,6-tetra-O-benzyl-α-D-galactopyranosyl)--α-D-glucopyranoside (4). Compound 3 (849 mg) was treated essentially as described for preparation of 2 to give, after column chromatography, syrupy 4 (739 mg, 96%), $[\alpha]_D$ +38°. ^{13}C NMR data: δ 55.0 (OCH_3), 98.4 (C-1), 101.1 C-1').

Methyl 2-O-Benzyl-3,6-di-O-(2,3,4,6-tetra-O-benzyl-α-D-galacto-pyranosyl)-1-thio-β-D-glucopyranoside (5). Methyl 2,3,4,6-tetra-O-benzyl-1-thio-β-D-galactopyranoside[4] (1.04 g, 1.82 mmol) was treated with bromine (0.13 mL, 2.53 mmol) in dichloromethane (15 mL) essentially as described for the preparation of 1 and the resulting crude bromide was mixed with dichloromethane (15 mL), 2 (0.675 g, 0.82 mmol), tetraethylammonium bromide (1.38 g, 6.57 mmol) and molecular sieves (3.0 g). After stirring overnight, pyridine (1.0 mL) was added and the mixture was processed as described for the preparation of 1. After column chromatography, pure 5 (983 mg, 89%) was obtained. After crystallization from toluene-hexane, the material had mp 98-99 °C, $[\alpha]_D$ +41°. ^{13}C NMR data: δ 12.9 (SCH_3), 68.8, 70.2, 70.5, 74.6, 75.1, 76.4, 76.7, 78.5, 78.6, 79.1, 79.7, 85.2, 87.7 (C-1,2,3,4,5, C-2', 3', 4', 5', C-2", 3", 4", 5"), 66.6, 68.4, 68.7, 72.5, 72.6, 72.9, 73.0, 73.1, 73.9, 74.5, 74.7 (C-6, C-6', C-6", $PhCH_2$), 97.4 (C-1"), 100.7 (C-1'). ^1H NMR data: δ 2.10 (s, SCH_3), 3.10 (dd, $J_{1,2}$ 9.7, $J_{2,3}$ 8.8 Hz, H-2), 3.51 (dd, $J_{2,3}$ 8.8, $J_{3,4}$ 8.8 Hz, H-3), 3.58 (m, H-4), 4.27 (d, $J_{1,2}$ 9.7 Hz, H-1), 4.80 (d, 4-OH), 5.01, 5.11 (doublets, J 3.7 and 3.4 Hz, H-1' and H-1'').
Anal. Calcd for $C_{82}H_{86}O_{15}S$: C, 73.2; H, 6.6, S, 2.4. Found: C, 73.4; H, 6.6; S, 2.2.

Methyl 4-O-Acetyl-2-O-benzyl-3,6-di-O-(2,3,4,6-tetra-O-benzyl-α-D--galactopyranosyl)-1-thio-β-D-glucopyranoside (6). A solution of 5 (286 mg) in pyridine (3 mL) and acetic anhydride (1.5 mL) was stirred at room temperature overnight. Water (1 mL) was added, and stirring was continued for 1h. The mixture was then diluted with dichloromethane and washed with 1 M sulfuric acid, aqueous sodium hydrogen carbonate, and water, dried,

and concentrated. The residue was purified by column chromatography to give syrupy 6 (288 mg, 98%), $[\alpha]_D$ +43°. ^{1}H NMR data: δ 2.06 (s, CH_3COO), 3.39 (dd, $J_{1,2}$ 9.5, $J_{2,3}$ 8.7 Hz, H-2), 3.59 (m, H-5), 3.85 (dd, $J_{3,4}$ 8.8 Hz, H-3), 4.26 (d, H-1), 5.11 (dd, $J_{4,5}$ 8.8 Hz, H-4).

Methyl 4-O-Acetyl-2-O-benzyl-3,6-di-O-(2,3,4,6-tetra-O-benzyl-α-D--galactopyranosyl)-α-D-glucopyranoside (7). Bromine (0.008 mL, 0.16 mmol) was added to a stirred and cooled (ice) mixture of 6 (135 mg, 0.097 mmol), dichloromethane (3.0 mL), and molecular sieves (1.5 g). After 1 h, 1-hexene (0.05 mL) was added, then tetraethylammonium bromide (167 mg, 0.80 mmol). Stirring was continued at room temperature for 3 h, then methanol (0.1 mL, 2.47 mmol) was added, and stirring was continued overnight. Pyridine (1.0 mL) was added, and the mixture was processed as described for the preparation of 1. After column chromatography, pure 7 (84 mg, 63%) was obtained, $[\alpha]_D$ +49°. ^{13}C NMR data: δ 21.0 (CH_3COO), 55.0 (OCH_3), 97.4, 97.6, 98.1 (C-1, 1', 1"). ^{1}H NMR data: δ 1.71 (s, CH_3COO), 3.16 (s, CH_3), 3.46 (dd, $J_{1,2}$ 3.6, $J_{2,3}$ 9.1 Hz, H-2), 4.08 (dd, $J_{3,4}$ 9.0 Hz, H-3), 4.31 (d, H-1), 4.76 (d, $J_{1,2}$ 3.6 Hz, H-1"), 5.01 (dd, $J_{4,5}$ 10.2 Hz, H-4), 5.30 (d, $J_{1,2}$ 3.3 Hz, H-1').

1-Octyl 2-O-Benzyl-3,6-di-O-(2,3,4,6-tetra-O-benzyl-α-D-galacto-pyranosyl)-α-D-glucopyranoside (8). Bromine (0.009 mL, 0.175 mmol) was added to a stirred and cooled (ice) mixture of 5 (200 mg, 0.149 mmol), dichloromethane (3.3 mL), and molecular sieves (1.5 g). After 45 min, 1-hexene (0.05 mL) was added, then tetraethylammonium bromide (66 mg, 0.31 mmol), 1-octanol (0.5 mL, 3.80 mmol) and dichloromethane (2.3 mL). Stirring was continued at room temperature for 20 h, then pyridine (1.0 mL) was added, and the mixture was processed as described for the preparation of 1. After column chromatography, pure 8 (167 mg, 79%) was obtained, $[\alpha]_D$ +53°. ^{13}C NMR data: δ 14.2, 22.7, 26.3, 29.3, 29.4, 29.5, 31.9 (octyl C-2,3,4,5,6,7,8), 96.7, 97.8, 100.6 (C-1,1',1"). ^{1}H NMR data: δ 4.48 (d, $J_{1,2}$ 3.7 Hz, H-1), 4.84 (d, $J_{1,2}$ 3.3 Hz, H-1"), 5.06 (d, $J_{1,2}$ 3.2 Hz, H-1').

2-(p-Trifluoroacetamidophenyl)ethyl 2-O-Benzyl-3,6-di-O-(2,3,4,6--tetra-O-benzyl-α-D-galactopyranosyl)-α-D-glucopyranoside (9). Bromine (0.004 mL, 0.078 mmol) was added to a stirred and cooled (ice) mixture of 5 (84 mg, 0.063 mmol), dichloromethane (1.4 mL), and molecular sieves (0.3 g). After 1 h, 1-hexene (0.05 mL) was added, then tetraethylammonium bromide (28 mg, 0.13 mmol), 2-(p-trifluoroacetamidophenyl)ethanol[7] (50 mg, 0.21 mmol), N,N-dimethylformamide (0.14 mL), and dichloromethane (1.0

mL). Stirring was continued at room temperature for 48 h, then pyridine (0.5 mL) was added, and the mixture was processed as described for the preparation of 1. After column chromatography, pure 9 (78 mg, 82%) was obtained, $[\alpha]_D$ +50°. ^{13}C NMR data: δ 35.2 ($PhCH_2CH_2$), 96.8, 97.5, 100.6 (C-1, 1', 1"). ^1H NMR data: δ 2.82 (t, J 7.3 Hz, $PhCH_2CH_2$), 4.55 (d, $J_{1,2}$ 3.7 Hz, H-1), 4.86 (d, $J_{1,2}$ 3.6 Hz, H-1"), 5.14 (d, $J_{1,2}$ 3.6 Hz, H-1').

Methyl 3,6-Di-O-(α-D-galactopyranosyl)-α-D-glucopyranoside (10). A solution of 7 (210 mg) in dichloromethane-methanol (3:2, 20 mL) was treated with methanolic sodium methoxide (0.1 M, 3 mL). After 2h, the mixture was neutralized with Dowex-50, filtered, and concentrated. A solution of the residue in ethyl acetate-ethanol-water (12:3:2, 30 mL) was hydrogenated over Pd/C (100 mg) at 400 kPa for 16 h, then filtered and concentrated. The residue was purified by gel filtration on a column of BioGel P2. Elution with water gave pure, amorphous 10 (78 mg, 98%), $[\alpha]_D$ +214° (H_2O). ^{13}C NMR data (D_2O): δ 55.9 (OCH_3), 61.6, 61.9 (C-6', 6"), 66.1 (C-6), 81.1 (C-3), 98.8 (C-1"), 100.0 (C-1'), 100.1 (C-1). ^1H NMR data (D_2O): δ 3.43 (s, OCH_3), 3.69 (dd, $J_{1,2}$ 3.9, $J_{2,3}$ 9.2 Hz, H-2), 4.23 (m, H-5'), 4.84 (d, $J_{1,2}$ 3.9 Hz, H-1), 4.98 (d, $J_{1,2}$ 3.7 Hz, H-1"), 5.36 (d, $J_{1,2}$ 3.9 Hz, H-1'). The FAB-MS of 10 showed an $(M+H)^+$ ion at m/z = 519.

1-Octyl 3,6-Di-O-(α-D-galactopyranosyl)-α-D-glucopyranoside (11). A solution of 8 (144 mg) in ethyl acetate-ethanol-water (12:3:2, 30 mL) was hydrogenated over Pd/C (70 mg) at 400 kPa for 16 h, then filtered and concentrated. The residue was purified by gel filtration on a column of BioGel P2. Elution with water gave pure, amorphous 11 (61 mg, 98%), $[\alpha]_D$ +184° (H_2O). ^{13}C NMR data (D_2O): δ 14.4, 23.1, 26.4, 29.4, 29.5, 29.6, 32.1 (octyl C-2,3,4,5,6,7,8), 62.0, 62.1 (C-6', 6"), 66.3 (C-6), 69.5 (octyl C-1), 81.5 (C-3), 99.0 (C-1"), 99.2 (C-1), 100.2 (C-1'). ^1H NMR data (D_2O): δ 3.67 ($J_{1,2}$ 3.9, $J_{2,3}$ 9.4 Hz, H-2), 4.24 (m, H-5'), 4.93 (d, $J_{1,2}$ 3.9 Hz, H-1), 4.97 (d, $J_{1,2}$ 3.7 Hz, H-1"), 5.36 (d, $J_{1,2}$ 3.9 Hz, H-1'). The FAB-MS of 11 showed an $(M+H)^+$ ion at m/z = 617.

2-(p-Trifluoroacetamidophenyl)ethyl 3,6-di-O-(α-D-galactopyranosyl)-α-D-glucopyranoside (12). A solution of 9 (75 mg) in ethyl acetate-ethanol-water (12:3:2, 15 mL) was hydrogenated over Pd/C (30 mg) at 400 kPa for 16 h, then filtered and concentrated. The residue was purified by gel filtration on a column of BioGel P2. Elution with water gave pure, amorphous 12 (33 mg, 92%), $[\alpha]_D$ +183° (H_2O). ^{13}C NMR data (D_2O): δ 35.6 ($PhCH_2CH_2$), 61.7, 61.8 (C-6', 6"), 65.7 (C-6),

81.2 (C-3), 98.6, 98.7, 100.0 (C-1, 1', 1"), 123.2, 130.7, 133.8, 139.3 (aromatic C), 157.7 (q, $COCF_3$). ^1H NMR data (D_2O): δ 2.96 (t, J 6.1 Hz, $PhCH_2CH_2$), 3.01 (dq, $J_{4,5}$ 9.8, $J_{5,6a}$ 3.6, $J_{5,6b}$ 2.1 Hz, H-5), 3.36 (dd, $J_{6a,6b}$ 11.5 Hz, H-6b), 3.61 (dd, $J_{1,2}$ 3.8, $J_{2,3}$ 9.2 Hz, H-2), 3.65 (dd, $J_{3,4}$ 9.8 Hz, H-4), 3.70 (dd, H-6a), 3.71 (dd, H-3), 4.22 (m, H-5'), 4.82 (d, $J_{1,2}$ 3.3 Hz, H-1"), 4.89 (d, H-1), 5.32 (dd, $J_{1,2}$ 3.8 Hz, H-1'). The FAB-MS of 12 showed an $(M+H)^+$ ion at m/z = 720.

Methyl 2-O-Benzyl-3-O-(2,3,4,6-tetra-O-benzyl-α-D-galactopyranosyl-6-O-[(2'-benzoyloxyethyl)oxymethyl]-α-D-glucopyranoside (13). A solution of 4 (283 mg), 2'-(benzoyloxyethyl)-oxymethyl chloride[9] (3 mL), and molecular sieves (0.5 g) in dichloromethane (3.0 mL) was stirred at room temperature for 16 h. Pyridine (1 mL) was added while stirring and cooling in an ice bath. After 30 min, the mixture was diluted with dichloromethane, washed with water, dried, and concentrated. The residue was purified by column chromatography to give 13 (232 mg, 67%), $[\alpha]_D$ +33°. ^{13}C NMR data: δ 55.0 (OCH_3), 64.1, 65.4 (CH_2CH_2), 66.3 (C-6'), 68.2 (C-6), 84.1 (C-3), 95.5 (OCH_2O), 98.4 (C-1), 101.1 (C-1'), 166.4 (C=O).

Methyl 3-O-(α-D-Galactopyranosyl)-6-O-[(2'-hydroxyethyl)oxymethyl]--α-D-glucopyranoside (14). A solution of 13 (220 mg) in ethyl acetate-ethanol-water (12:3:2, 20 mL) was hydrogenated over Pd/C (100 mg) at 400 kPa for 16 h, then filtered and concentrated. The residue was dissolved in methanol (10 mL) and treated with methanolic sodium methoxide (0.1 M, 2.0 mL). After 1h, the mixture was neutralized with Dowex-50 and concentrated. The residue was purified by gel filtration on a column of BioGel P2. Elution with water gave pure, amorphous 14 (92 mg, 95%), $[\alpha]_D$ +162° (H_2O). ^{13}C NMR data (D_2O): δ 56.0 (OCH_3), 61.3, 61.7 (C-6', OCH_2CH_2OH), 67.0, 69.9 (C-6, OCH_2CH_2OH), 69.4, 69.9, 70.1, 70.6, 70.7, 70.9, 71.6 (C-2,4,5, C-2',3', 4', 5'), 80.5 (C-3), 96.0 (OCH_2O), 99.9, 100.2 (C-1, C-1'). ^1H NMR data (D_2O): δ 3.43 (s, OCH_3), 3.67 (dd, $J_{1,2}$ 3.9, $J_{2,3}$ 9.4 Hz, H-2), 3.69 (dd, $J_{3,4}$ 9.0, $J_{4,5}$ 9.8 Hz, H-4), 3.82 (dd, $J_{2,3}$ 9.4, $J_{3,4}$ 9.0 Hz, H-3), 3.84 (dd, $J_{1',2'}$ 4.0, $J_{2',3'}$ 10.3 Hz, H-2'), 3.84 (dd, $J_{5,6a}$ 4.5, $J_{6a,6b}$ 11.2 Hz, H-6a), 3.89 (dd, $J_{5,6b}$ 2.3, $J_{6a,6b}$ 11.2 Hz, H-6b), 3.90 (dd, $J_{2',3'}$ 10.3, $J_{3',4'}$ 3.2 Hz, H-3'), 4.00 (dd, $J_{3',4'}$ 3.2, $J_{4',5'}$ 1.0 Hz, H-4'), 4.23 (m, H-5'), 4.80, 4.81 (two doublets, J 6.8 Hz, OCH_2O), 4.81 (d, $J_{1,2}$ 3.9 Hz, H-1), 5.38 (d, $J_{1',2'}$ 4.0 Hz, H-1'). The FAB-MS of 14 showed an $(M+H)^+$ ion at m/z = 431.

ACKNOWLEDGEMENTS

We thank Mr. Gunnar Grönberg (Analytical Department, BioCarb AB) for recording and assigning of NMR spectra and Mr. Stefan Strömberg (Analytical Department, BioCarb AB) for recording the mass spectra.

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