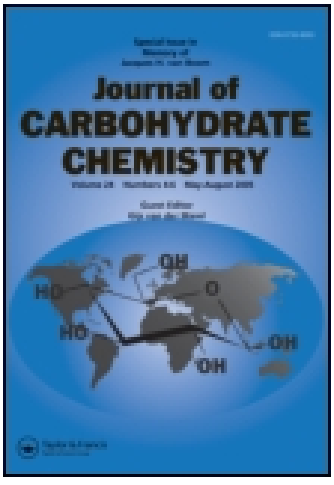


Journal of Carbohydrate Chemistry
Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/lcar20

Synthesis of C-Glycoconjugates from Readily Available Unprotected C-Allyl Glycosides by Chemoselective Ligation

Francisco Cardona [a] & Barbara La Ferla [a]
[a] Department of Biotechnology and Bioscience , University of Milano Bicocca , Milano, Italy
Published online: 27 Jun 2008.

To cite this article: Francisco Cardona & Barbara La Ferla (2008) Synthesis of C-Glycoconjugates from Readily Available Unprotected C-Allyl Glycosides by Chemoselective Ligation, Journal of Carbohydrate Chemistry, 27:4, 203-213

To link to this article: http://dx.doi.org/10.1080/07328300802082457

Journal of Carbohydrate Chemistry, 27:203–213, 2008

ISSN: 0732-8303 print 1532-2327 online
DOI: 10.1080/07328300802082457

Synthesis of C-Glycoconjugates from Readily Available Unprotected C-Allyl Glycosides by Chemoselective Ligation

Francisco Cardona and Barbara La Ferla

Department of Biotechnology and Bioscience, University of Milano Bicocca, Milano, Italy

Keywords C-glycosides, Neoglycoconjugates, Chemoselective ligation

INTRODUCTION

Glycoconjugates such as glycoproteins, glycolipids, and proteoglycans are key components of the living system and are involved in various recognition processes such as cell adhesion, migration, and signaling,[1] and in many pathological events such as autoimmune disease,[2] bacterial and viral invasion of the host,[3] and tumor-cell motility and progression.[4] Understanding how and why these highly specific interactions come about is a hard task due to the difficulties associated with the "microheterogenity" of natural glycoconjugates. Thus, there is an increasing demand for synthetic glycoconjugates for a better comprehension of these biological processes and to expedite the development of therapeutic agents.

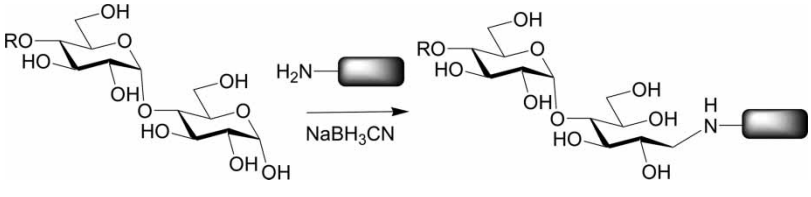
Received March 20, 2008; accepted March 26, 2008.
Address correspondence to Barbara La Ferla, Department of Biotechnology and Bioscience, University of Milano Bicocca, Piazza della Scienza 2, I-20126 Milano, Italy. E-mail: barbara.laferla@unimib.it

The synthesis of glycoconjugates presents two main problems: the complexity of the glycidic entity, which requires stereochemical and regiochemical control, and the glycosylation of a polyfunctionalized aglycon, such as a peptide or a protein, which requires chemoselectivity. This last problem has been faced in different ways, among which is the reductive amination that allows one to link the anomeric center of an aldose to primary amines such as the residues of lysine (Sch. 1). Since no aldehydo or keto groups are present in natural amino acids, this process is chemoselective, and the conjugation is metabolically and chemically stable. The procedure, however, generates glycoconjugate mimics (neoglycoconjugates) where the sugar directly linked to the amino group loses its natural cyclic structure.[5]

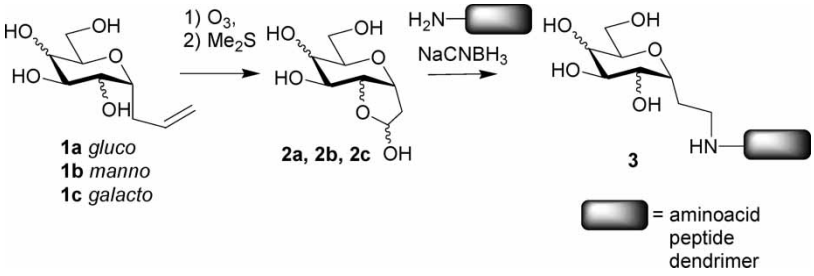
This problem can be avoided through a laborious manipulation of the carbohydrate moiety to introduce a linker at the reducing end bearing a terminal aldehyde group.[6]

RESULTS AND DISCUSSION

In order to generate neoglycoconjugates maintaining the cyclic structure of the sugar, by exploiting the chemoselective reductive amination approach, we speculated that instead of using the anomeric aldehydes, the sugar should contain an anomeric appendage with the aldehyde. Furthermore, if the aldehyde generates a hemiacetal with the hydroxyl group at C-2, the structures would be more stable. Following this idea we synthesized compounds **2** (Sch. 2) bearing a masked aldehyde linked to the anomeric center through a carbon chain (a C-glycoside). Such hemiacetals have already been synthesized from C-glycosides in order to generate amphiphilic liposaccharides[7] and their reactivity investigated.[8] The reaction of compounds **2** with an amino acid, a peptide, or a dendrimer containing a primary amino group, under reductive amination conditions, afforded neoglycoconjugates **3**. It is interesting to note that the glycoconjugation process can be performed on fully deprotected substrates in media that are compatible with biomaterials, and the synthesis of the glycidic structures **2** does not require protection-deprotection steps.



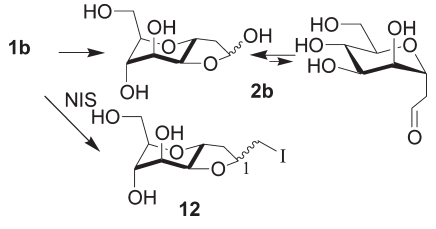
Scheme 1: Glycoconjugation by reductive amination.



Scheme 2: Synthesis of bicyclic hemiacetals **2** and their conjugation.

Deprotected C-allyl α-D-glucoside (**1a**), α-D-mannoside (**1b**), and α-D-galactoside (**1c**) were stereoselectively synthesized according to a procedure described by Gray and coworkers.[9]

The hemiacetals gluco, manno, and galacto derivatives **2** were readily obtained from the corresponding C-allyl glycosides **1**, a solution of which (7.3 mmol) in $MeOH/H_2O$ (100 mL) was cooled to −78°C and saturated with ozone. After 75 min the pale blue solution was purged with oxygen and then with nitrogen until colorless. Then Me_2S (10 equiv.) was added and the reaction mixture was warmed to rt and left stirring overnight. The solvent was then removed under reduced pressure affording compounds **2** (78–81% yield) as a diastereoisomeric mixture of the bicyclic hemiacetalic structure, along with compounds **2** ^{1}H NMR, which showed the formation of the methyl acetal derivatives as minor products (22–19%). Surprisingly, despite the unfavorable orientation of the C-2 OH, also the C-allyl α-D-mannoside afforded the hemiacetals as major product, probably with a distorted conformation (Sch. 3) in equilibrium with the monocyclic aldehyde, as evidenced by the ^{1}H NMR aldehyde signal at 9.4 ppm. To better understand this behavior, we reacted compound **2b** with *N*-iodosuccinimide in MeOH and the reaction afforded the product of the iodocyclization **12** (Sch. 3) as evidenced by the chemical shift at 4.1 ppm of H1 confirmed through COSY experiments.



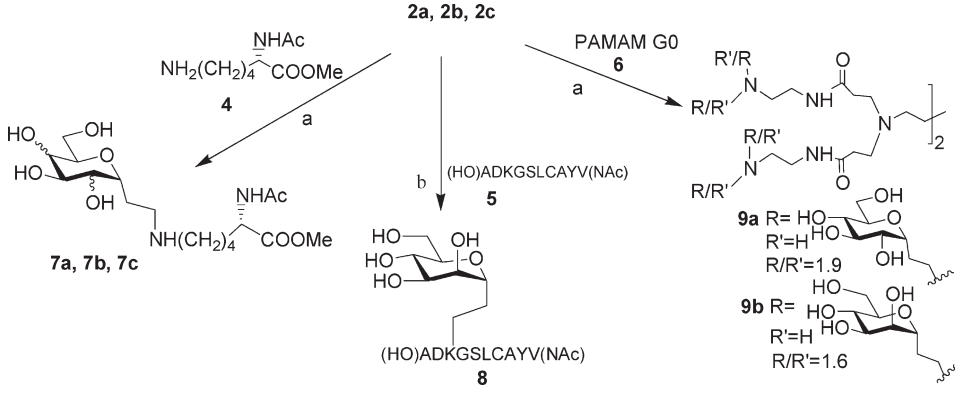
Scheme 3: Conformational change for mannose derivatives.

Reductive amination of compounds **2** (MeOH or MeOH/H_2O, $NaBH_3CN$ and AcOH) with the amino group of protected lysine **4**, with peptide **5**, and with PAMAM G0 dendrimer **6** generated the corresponding neoglycosyl aminoacids **7a**, **7b**, and **7c**, neoglycosyl peptide **8**, and neoglycodendrimers **9a** and **9b** (Sch. 4).

The neoglycosyl aminoacids **7** were obtained heating crude **2** (0.48 mmol) at 60°C with *N*-acetyl L-lysine methyl ester **4** (0.24 mmol) in MeOH (2 mL) AcOH (28 μL, 2 equiv.) and then adding $NaCNBH_3$ (0.48 mmol). After 3 h, usual workup and chromatography (AcOEt/MeOH/H_2O/AcOH v/v 6/2/1/1) afforded compounds **7** (**7a** 54%, **7b** 70%, and **7c** 55% yield). The neoglycosyl peptide **8** was generated by adding **2b** (0.019 mmol) dissolved in MeOH (100 μL) to a solution of peptide **5** (0.0018 mmol) in degassed MeOH (100 μL), and then by adding a solution of $NaCNBH_3$ (0.048 mmol) in MeOH (50 μL) and heating at 50°C for 1 h. The reaction was monitored by HPLC and the product characterized by mass spectrometry (Fig. 1).

The neoglycodendrimers **9a** and **9b** were obtained by reaction of PAMAM (G0) dendrimer **6** (0.04 mmol) and crude **2a** (0.24 mmol) in acetate buffer pH 4 (2 mL) with $NaCNBH_3$ (0.26 mmol) for 1 h. After workup, the recovered solid was subjected to two more reaction cycles. After the third cycle the solid was purified by dialysis against water for 48 h and then lyophilized. Glucodendrimer **9a** (98% yield) presented an average of 7.7 sugar moieties around 1.9 for each terminal amino group, as determined by ^1H NMR signal integration (Fig. 2), while mannodendrimer **9b** (76% yield) carried an average of 6.4 sugar residues.

An alternative chemoselective approach for the synthesis of neoglycoconjugates relies on the radical reaction between the thiol group and a terminal olefinic double bond. This reaction has already been used for the generation of glycodendrimers,[10] using thioglycosides and dendrimer cores bearing



Scheme 4: Synthesis of derivatives **7**, **8**, and **9** by reductive amination. Reagents and conditions: a) MeOH, $NaCNBH_3$, AcOH; b) MeOH/H_2O, $NaCNBH_3$, AcOH.

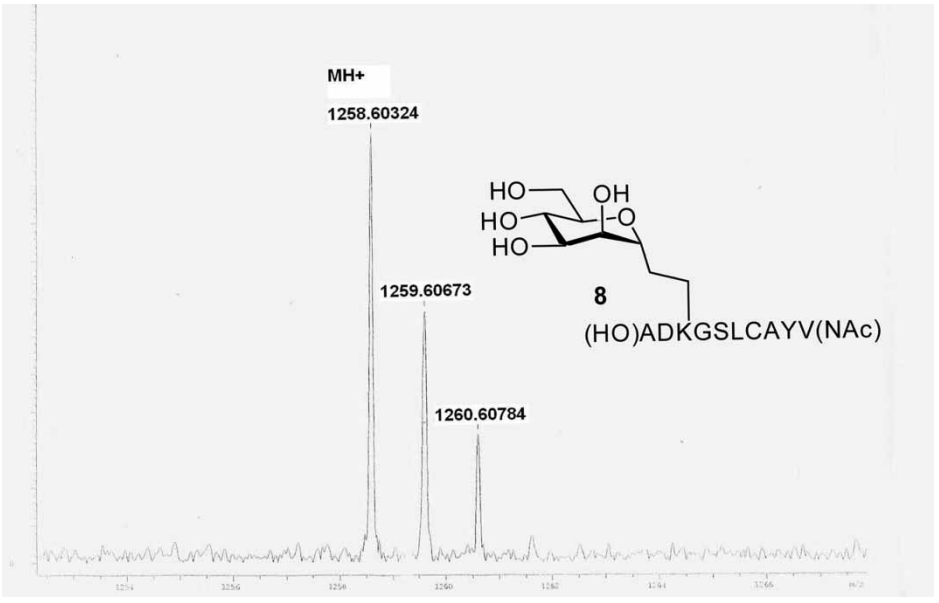


Figure 1: Mass spectra of glycopeptide **8**. Calculated mass = 1258.57512 Da, observed mass = 1258.60324 Da.

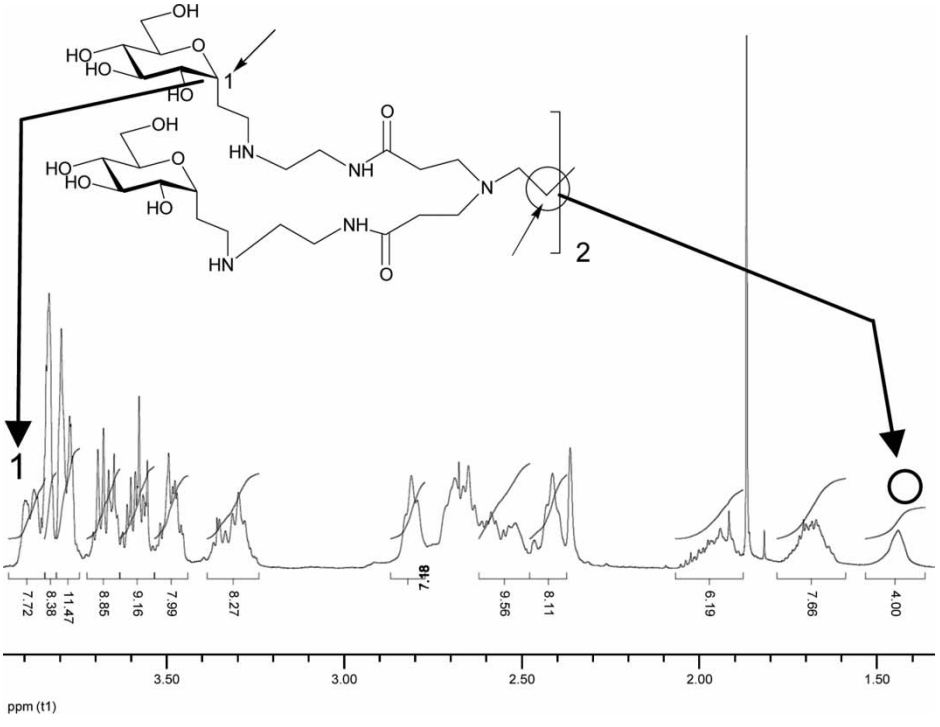


Figure 2: ^{1}H NMR of signal integration of glucodendrimer **9a**.

olefinic terminal groups, or in the synthesis of carbohydrate microarray.[11] The availability of allyl *C*-glycosides **1** suggested to us that we perform the reaction with aglycons containing a thiol group, such as a cysteine residue (Sch. 5).

This reaction was carried out by irradiation ($h\nu = 254$ nm) of compounds **1** (0.49 mmol) and *N*-acetyl L-cysteine methyl ester **10** (0.98 mmol), in degassed MeOH/H_2O (3 mL). After 1 h the solvent was removed under reduced pressure and the product purified by flash chromatography (AcOEt/MeOH v/v 9/1) affording **11** (**11a** 86%, **11b** 87%, and **11c** 84% yield) as amorphous white solids.

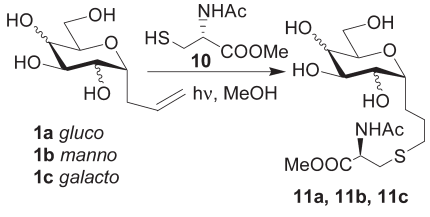
CONCLUSION

In summary, we have presented an efficient and direct method for the generation of neoglycoconjugates without the use of protecting groups, and exploited two chemoselective approaches, the reductive amination and photochemical addition of a thiol to a double bond. Both methods take advantage of the easy and stereoselective preparation of deprotected *C*-allyl glycosides **1**.

EXPERIMENTAL SECTION

General Remarks

All solvents were dried with molecular sieves, for at least 24 h prior to use. Thin layer chromatography (TLC) was performed on silica gel 60 F254 plates (Merck) with detection using UV light when possible, or by charring with a solution of concd. H_2SO_4/EtOH/H_2O (5:45:45) or a solution of $(NH_4)_6Mo_7O_{24}$ (21 g), $Ce(SO_4)_2$ (1 g), and concd. H_2SO_4 (31 mL) in water (500 mL). Flash column chromatography was performed on silica gel 230–400 mesh (Merck). 1H, 13C NMR spectra were recorded at 25°C with a Varian Mercury 400 MHz instrument using $CDCl_3$ as the solvent unless otherwise stated. Chemical shift assignments, reported in ppm, are referenced to the corresponding solvent peaks. Peptide coupling was performed on reversed phase analytical HPLC on Waters HPLC system equipped with a Merck LiChrospher 100



Scheme 5: Chemoselective synthesis of cystein conjugates **11**.

RP18 column (250 × 4 mm; 5 μm), eluting at a flow rate 1 mL/min. Detection was at 214 and 280 nm with an absorbance detector Waters 2487 dual λ. Mass spectra were recorded with an ESI positive Fourier Transform Ion Ciclotron Resonance Mass Spectrometer APEX II (Bruker Daltonics) — 4.7T magnet (Magnex). Optical rotations were measured at rt using a Krüss P3002 electronic polarimeter and are reported in units of 10^{-1} deg/cm^2/g^{-1}. Peptide **5** was prepared with SPPS approach using Fmoc protected amino acids and 2-chlorotrityl chloride resin as solid support. HBTU and DIPEA were used as coupling agents, a solution of piperidine 20% in DMF was used for Fmoc deprotection, and the terminal amino group was acetylated prior to deprotection and cleavage from the resin. PAMAM Dendrimer (G0) 1,4-diaminobutane core was purchased from Sigma-Aldrich.

3,7-Anhydro-2-deoxy-α,β-D-glycero-D-ido-octofuranose (1,4) (2a)

A solution of allyl-C-glucoside **1a** (1.5 g, 7.3 mmol) in MeOH/H_2O (100 mL) was cooled to −78°C and saturated with ozone. After 75 min the pale blue solution was purged with oxygen and then with nitrogen until colorless. Then Me_2S (73 mmol, 10 equiv.) was added and the reaction mixture was warmed to rt and left stirring overnight. The solvent was then removed under reduced pressure affording crude compound **2a** as a colorless oil. Crude compound **2a**, a mixture of hemiacetalic diastereoisomers, was used without purification.

3,7-Anhydro-2-deoxy-α,β-D-glycero-D-talo-octofuranose (1,4) (2b)

The same procedure was used as for the synthesis of **2a**. Starting material allyl-C-mannoside **1b** (1.8 g, 8.8 mmol) afforded crude compound **2b** as a colorless oil. Crude compound **2b**, a mixture of hemiacetalic diastereoisomer containing also the aldehyde form, was used without purification.

3,7-Anhydro-2-deoxy-α,β-D-glycero-L-gluco-octofuranose (1,4) (2c)

The same procedure was used as for the synthesis of **2a**. Starting material allyl-C-galactoside **1c** (2.0 g, 9.8 mmol) afforded crude compound **2c** as a colorless oil. The crude **2c**, a mixture of hemiacetalic diastereoisomer containing also the two methyl acetal derivatives, was used without purification.

*N*2-Acetyl-*N*5-((2-glucopyranosyl)ethyl)-L-lysine Methyl Ester (7a)

To a solution of crude **2a** (100 mg, 0.48 mmol) and Ac-Lys-OMe **4** (57 mg, 0.24 mmol) in MeOH (2 mL), AcOH (28 μL, 2 equiv.) was added and the reaction was heated to 60°C. Then $NaCNBH_3$ (30 mg, 0.48 mmol) was added and the mixture was stirred at 60°C for 3 h. The solvent was then removed under reduced pressure and the product purified by flash chromatography (AcOEt/MeOH/H_2O/AcOH v/v 6/2/1/1) yielding **7a** (50 mg, 54% yield) as amorphous white solid. $[\alpha]_d^{25}$ +13.6 (c = 0.25, CH_3OH); ^{1}H NMR (400 MHz, D_2O) δ: 4.33 (dd, 1H, $J = 9.0$, $J = 5.1$ Hz), 4.12–4.04 (m, 1H), 3.80 (dd, 1H, $J = 12.0$, $J = 2.1$ Hz), 3.71–3.68 (m, 4H), 3.63 (dd, 1H, $J = 12.3$, $J = 6.3$ Hz), 3.55 (t, 1H, $J = 9.0$ Hz), 3.48–3.43 (m, 1H), 3.29 (t, 1H, $J = 9.0$ Hz), 3.13–3.08 (m, 2H), 3.02 (bt, 2H), 2.12–2.00 (m, 1H), 2.00–1.78 (m, 5H), 1.77–1.60 (m, 3H), 1.48–1.35 (m, 2H); ^{13}C NMR (100 MHz) δ: 174.4 (C), 174.3 (C), 73.64 (CH), 73.03 (CH), 73.03 (CH), 70.51 (CH), 69.99 (CH), 60.97 (CH_2), 52.90, 52.54 (CH, CH_3), 47.44 (CH_2), 44.92 (CH_2), 29.86 (CH_2), 24.95 (CH_2), 22.07 (CH_2), 21.53 (CH_3), 21.22 (CH_2); HRMS calcd for $C_{17}H_{32}N_2O_8$ (MH+) 393.22314, found 393.22246; calcd (MNa+) 415.20509, found 415.20514.

*N*2-Acetyl-*N*5-((2-mannopyranosyl)ethyl)-L-lysine Methyl Ester (7b)

The same procedure was used as for the synthesis of **7a**. Starting material **2b** (100 mg, 0.48 mmol). Product obtained after purification by flash chromatography (AcOEt/MeOH/H_2O/AcOH v/v 6/2/1/1) **7b** (65 mg, 70% yield) as amorphous white solid. $[\alpha]_d^{25}$ +6.5 (c = 0.4, H_2O); ^{1}H NMR (400 MHz, D_2O) δ: 4.33 (dd, 1H, $J = 9.1$, $J = 5.0$ Hz), 4.00–3.96 (m, 1H); 3.86–3.68 (m, 7H), 3.60 (t, 1H, $J = 8.7$ Hz), 3.53–3.46 (m, 1H), 3.12, (bt, 2H), 3.01 (bt, 2H), 2.24–2.10 (m, 1H), 2.00 (s, 3H), 1.90–1.80 (m, 2H), 1.78–1.60 (m, 3H), 1.45, 1.35 (m, 2H); ^{13}C NMR (100 MHz) δ: 176.9 (C), 176.8 (C), 77.88 (CH), 77.14 (CH), 73.45 (CH), 73.21 (CH), 70.12 (CH), 63.62 (CH_2), 55.33, 55.69 (CH, CH_3), 50.28 (CH_2), 47.79 (CH_2), 32.70 (CH_2), 27.85 (CH_2), 27.24 (CH_2), 24.96 (CH_2), 24.38 (CH_3); HRMS calcd for $C_{17}H_{32}N_2O_8$ (MH+) 393.22314, found 393.22251; calcd (MNa+) 415.20509, found 415.20512.

*N*2-Acetyl-*N*5-((2-galactopyranosyl)ethyl)-L-lysine Methyl Ester (7c)

The same procedure was used as for the synthesis of **7a**. Starting material **2c** (50 mg, 0.24 mmol). Product obtained after purification by flash chromatography (AcOEt/MeOH/H_2O/AcOH v/v 6/2/1/1) **7c** (30 mg, 55% yield) as

amorphous white solid. $[\alpha]_d^{25}$ +26.7 (c = 0.15, CH_3OH); ^1H NMR (400 MHz, D_2O) δ: 4.37 (dd, 1H, $J = 9.1$, $J = 5.2$ Hz), 4.18–4.12 (m, 1H), 4.00–3.95 (m, 2H), 3.79–3.69 (m, 7H), 3.16, (bt, 1H), 3.04 (bt, 1H), 2.14–2.05 (m, 1H), 2.02 (s, 3H), 2.02–1.94 (m, 1H), 1.93–1.84 (m, 1H), 1.79–1.65 (m, 3H), 1.44–1.38 (m, 2H); ^{13}C NMR (100 MHz) δ: 174.4 (C), 174.3 (C), 73.61 (CH), 72.51 (CH), 69.75 (CH), 68.93 (CH), 67.93 (CH), 61.28 (CH_2), 53.15, 52.83 (CH, CH_3), 48.91 (CH_2), 47.73 (CH_2), 30.15 (CH_2), 26.49 (CH_2), 25.29 (CH_2), 22.35 (CH_2), 21.86 (CH_3); HRMS calcd for $C_{17}H_{32}N_2O_8$ (MNa+) 415.20509, found 415.20515.

C-Glycopeptide (8)

Peptide **5** (2 mg, 0.0018 mmol) was dissolved in degassed MeOH (100 μL) and **2b** (4 mg, 0.019 mmol) dissolved in MeOH (100 μL) was added. Then a solution of $NaCNBH_3$ (3 mg, 0.048 mmol) in MeOH (50 μL) was added and the reaction mixture was heated to 50°C for 1 h. HPLC profile using gradient 10% A, 90% B to 50% A, 50% B in 30 min (A = MeCN/H_2O/TFA 90/10/0.01 and B = H_2O/TFA 0.1%) showed complete conversion of peptide **5** to glycopeptide **8** (retention time 17, 35 min) and small amount of a second peak not identified. **8** HRMS calcd for $C_{53}H_{86}N_{12}O_{21}S$ (M+) 1258.57512, found 1258.60324.

Glucodendrimer (9a)

PAMAM Dendrimer (G0) 1,4-diaminobutane core (22 mg, 0.04 mmol) was dissolved in acetate buffer pH 4 (2 mL) and crude **2a** (50 mg, 0.24 mmol, 6 equiv.) was added. Then $NaCNBH_3$ (16 mg, 0.26 mmol) was added and the mixture was stirred for 1 h. The solvent was removed under reduced pressure and the solid dissolved in MeOH, precipitated with cold Et_2O, and then filtered. The solid recovered was subjected to two more equivalent reaction cycles. After the third cycle the solid was purified by dialysis against water for 48 h. The final product was lyophilized affording 79 mg of glucodendrimer with 7.7 sugar moieties (98% yield). ^1H NMR (400 MHz, D_2O) δ: 3.95–3.86 (m, 7.7H), 3.85–3.82 (m, 8.4H), 3.82–3.76 (m, 11.5H), 3.72–3.64 (m, 8.8H), 3.63–3.55 (m, 9.2H), 3.53–3.46 (m, 8H), 3.48–3.26 (m, 8.3H), 2.85–2.76 (m, 7.2H), 2.73–2.64 (m, 13.3H), 2.63–2.49 (m, 9.5H), 2.48–2.33 (m, 11.3H), 2.07–1.87 (m, 6.2H), 1.77–1.61 (m, 7.7H), 1.52–1.40 (bs, 4H).

Mannodendrimer (9b)

The same procedure was used as for the synthesis of **9a**. Final lyophilization afforded 50 mg of mannodendrimer with an average of 6.4 sugar moieties (76% yield). ^1H NMR (400 MHz, D_2O) δ: 4.04–3.96 (m, 6.4H),

3.87–3.78 (m, 7.0H), 3.72–3.57 (m, 23.7H), 3.55–3.47 (m, 5.7H), 3.39–3.29 (m, 12.6H), 2.86–2.78 (m, 7.8H), 2.76–2.55 (m, 17.3H), 2.54–2.49 (m, 4.6H), 2.48–2.38 (m, 8.1H), 2.38–2.32 (m, 4.4H), 1.93–1.80 (m, 12.2H), 1.51–1.42 (bs, 4H).

N-Acetyl-S-((3-glucopyranosyl)propyl)-L-cysteine Methyl Ester (11a)

Compound **1a** (100 mg, 0.49 mmol) and cysteine L-methyl ester (174 mg, 0.98 mmol) was dissolved in degassed MeOH/H_2O (3 mL) and irradiated with UV lamp hν = 254 nm. After 1 h the solvent was removed under reduced pressure and the product purified by flash chromatography (AcOEt/MeOH v/v 9/1) affording **11a** (160 mg, 86% yield) as amorphous white solid. $[\alpha]_d^{25}$ +24.4 (c = 0.5, CH_3OH); ^{1}H NMR (400 MHz, D_2O) δ: 4.63 (dd, 1H, J = 8.1, J = 5.0 Hz), 4.01–3.96 (m, 1H); 3.82 (dd, 1H, J = 12.2, J = 2.2 Hz), 3.77 (s, 3H), 3.71–3.63 (m, 2H), 3.61 (t, 1H, J = 8.1 Hz), 3.52–3.46 (m, 1H), 3.32, (bt, 1H), 3.07 (dd, 1H, J = 14.7, J = 5.0 Hz), 3.92 (dd, 1H, J = 14.7, J = 8.1 Hz), 2.70–2.60 (m, 2H), 2.05 (s, 3H), 1.72–1.53 (m, 4H); ^{13}C NMR (100 MHz) δ: 176.7 (C), 175.3 (C), 78.20 (CH), 76.07 (CH), 75.25 (CH), 74.00 (CH), 73.10 (CH), 63.90 (CH_2), 55.96, 55.47 (CH, CH_3), 35.17 (CH_2), 34.23 (CH_2), 27.60 (CH_2), 25.60 (CH_2), 24.57 (CH_3); HRMS calcd for $C_{15}H_{27}NO_8S$ (MNa+) 404.13496, found 404.13425.

N-Acetyl-S-((3-mannopyranosyl)propyl)-L-cysteine Methyl Ester (11b)

The same procedure was used as for the synthesis of **11a** starting from **1b**. Purification by flash chromatography (AcOEt/MeOH v/v 9/1) afforded **11b** (162 mg, 87% yield) as amorphous white solid. $[\alpha]_d^{25}$ +24.4 (c = 0.25, CH_3OH); ^{1}H NMR (400 MHz, D_2O) δ: 4.61 (dd, 1H, J = 8.2, J = 5.0 Hz), 3.95–3.89 (m, 1H), 3.88–3.77 (m, 6H), 3.71 (dd, 1H, J = 12.1, J = 6.1 Hz), 3.62 (t, 1H, J = 9.3 Hz), 3.53–3.48 (m, 1H), 3.07 (dd, 1H, J = 14.0, J = 5.0 Hz), 2.92 (dd, 1H, J = 14.0, J = 8.2 Hz), 2.63 (bt, 2H), 1.90–1.80 (m, 1H), 1.80–1.53-1.53 (m, 3H); ^{13}C NMR (100 MHz) δ: 176.7 (C), 175.3 (C), 80.70 (CH), 76.34 (CH), 74.30 (CH), 73.62 (CH), 70.13 (CH), 64.10 (CH_2), 55.97, 55.47 (CH, CH_3), 35.19 (CH_2), 34.16 (CH_2), 29.29 (CH_2), 27.97 (CH_2), 24.59 (CH_3); HRMS calcd for $C_{15}H_{27}NO_8S$ (MNa+) 404.13496, found 404.13440.

N-Acetyl-S-((3-glucopyranosyl)propyl)-L-cysteine Methyl Ester (11c)

The same procedure was used as for the synthesis of **11a** starting from **1c**. Purification by flash chromatography (AcOEt/MeOH v/v 8/2) afforded

11c (156 mg, 84% yield) as amorphous white solid. $[\alpha]_d^{25}$ +65.3 (c = 0.15, CH_3OH); ^1H NMR (400 MHz, D_2O) δ: 4.61 (dd, 1H, $J = 8.2$, $J = 5.0$ Hz), 4.06–3.99 (m, 1H); 3.98–3.90 (m, 2H), 3.79–3.67 (m, 7H), 3.06 (dd, 1H, $J = 14.0$, $J = 5.0$ Hz), 2.90 (dd, 1H, $J = 14.0$, $J = 8.2$ Hz), 2.68–2.55 (m, 2H), 2.06 (s, 3H), 1.80–1.50 (m, 4H); ^{13}C NMR (100 MHz) δ: 176.7 (C), 175.3 (C), 77.86 (CH), 74.43 (CH), 72.46 (CH), 71.90 (CH), 71.05 (CH), 63.99 (CH_2), 55.93, 55.44 (CH, CH_3), 35.10 (CH_2), 34.24 (CH_2), 27.73 (CH_2), 25.46 (CH_2), 24.52 (CH_3); HRMS calcd for $C_{15}H_{27}NO_8S$ (MNa+) 404.13496, found 404.13483.

3,7-Anhydro-2-deoxy-C-iodomethyl-α,β-D-glycero-D-talo-octofuranoside (1,4) (12) Major Isomer

^1H NMR (400 MHz, CD_3OD) δ: 4.18–4.00 (m, 4H), 3.94–3.86 (m, 2H,), 3.76 (d, 1H, $J = 3.0$ Hz), 3.58 (dd, 1H, $J = 12.0$, $J = 4.6$ Hz), 3.41–3.37 (m, 2H), 2.46–2.36 (m, 1H), 1.80–1.61 (m, 1H); ^{13}C NMR (100 MHz) δ: 86.12 (CH), 81.67 (CH), 80.89 (CH), 74.13 (CH), 73.00 (CH), 70.97 (CH), 63.65 (CH_2), 40.45 (CH_2), 13.95 (CH_2); MS calcd for $C_9H_{16}IO_5$ (MH+) 331.0, found 331.3.

REFERENCES

[1] (a) Dwek, R.A. Chem. Rev. **1996**, *96*, 683–720; (b) Fukuda, M. Bioorg. Med. Chem. **1995**, *3*, 207–215; (c) Varki, A. Glycobiology **1993**, *3*, 97–130; (d) Feizi, T. Glycoconj. J. **2000**, *17*, 553–565; (e) Bertozzi, C.; Kiessling, L. Science **2001**, *291*, 2357–2364; (f) Varki, A.; Cummings, R.; Esko, J.; Freeze, H.; Hart, G.; Marth, J. *Essentials of Glycobiology*; Cold Spring Harbor Laboratory Press: Cold Spring Harbor, NY, 1999.

[2] Lowe, J.B. Cell **2001**, *104*, 809–812.

[3] Karlsson, K.A. Curr. Opin. Struct. Biol. **1995**, *5*, 622–635.

[4] Hakomori, S.; Zhang, Y. Chem. Biol. **1997**, *4*, 97–104.

[5] (a) Wang, P.; Hill, T.G.; Wartchow, A.; Huston, M.E.; Oehler, L.M.; Bradley Smith, M.; Bednarski, M.D.; Callstrom, M.R. J. Am. Chem. Soc. **1992**, *114*, 378–380; (b) Wartchow, C.A.; Wang, P.; Bednarski, M.D.; Callstrom, M.R. J. Org. Chem. **1995**, *60*, 2216–2226.

[6] (a) Wong, C.-H.; Moris-Varas, F.; Hung, S.-C.; Marron, T.G.; Lin, C.C.; Gong, K.W.; Weitz-Schmidt, G. J. Am. Chem. Soc. **1997**, *119*, 8152–8158; (b) Arya, P.; Kutterer, K.M.K.; Barkley, A. J. Comb. Chem. **2000**, *2*, 120–126; (c) Arya, P.; Barkley, A.; Randell, K.D. J. Comb. Chem. **2002**, *4*, 193–198; (d) Clausen, M.H.; Madsen, R. Carbohydr. Res. **2004**, *339*, 2159–2169.

[7] Cabaret, D.; Wakselman, M. Carbohydr. Res. **1989**, *189*, 341–348.

[8] Cabaret, D.; Wakselman, M. Can. J. Chem. **1990**, *68*, 2253–2257.

[9] Bennek, J.A.; Gray, G.R. J. Org. Chem. **1987**, *52*, 892–897.

[10] Lubineau, A.; Malleron, A.; Le Narvor, C. Tetrahedron Lett. **2000**, *41*, 8887–8891.

[11] Qian, X.; Metallo, S.J.; Choi, I.S.; Wu, H.; Liang, M.N.; Whitesides, G.M. Anal. Chem. **2002**, *74*, 1805–1810.