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New β-cyclodextrin derivatives possessing biologically active saccharide antennae

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The synthesis of monosubstituted β-cyclodextrin derivatives having the monosaccharides, β-D-glucose, β-D-galactose, α-D-mannose, β-L and β-D-fucose linked to the macrocycle via a C9 spacer chain is described. The approach is based on the highly efficient coupling of the NCS sugar derivatives to monomethyl nonanedicarboxylate to generate a stable amido linkage. The NMR studies show the saccharide antennae to be oriented into the environment and not towards the CD cavity.

INTRODUCTION

The importance of oligosaccharide side chains in biological functions is well documented.[1] Oligosaccharides on cell surface glycoproteins[2] or lectins have been shown to be responsible for cell-cell recognition in biological processes and may be considered as ideal carried of biological information.[3a,3b]

The cyclodextrins (CDs) are a class of cyclic oligosaccharides well documented for their ability to complex and solubilise a wide range of organic molecules;[4] in view of this they have been widely applied in the bio-pharmaceutical field.[5] Whilst the transport of hydrophobic drugs can be performed using inclusion in β-cyclodextrin (β-CD), such action will be non-specific, due to an absence of effector groups on the CD motif capable of interacting with specific biological receptors.

We are interested in developing a chemical synthesis of artificial glycoconjugates in which the cyclodextrins play the role of molecular transport systems and saccharide antennae function as targeting devices, to be used in binding studies with lectins and ultimately in the 'intelligent' delivery *in vivo* of pharmacologically active molecules. Such systems must possess a cavity capable of solubilising and stabilising specified medications and a head group which is capable of interacting effectively with biological receptors; also, there must be no interactions between headgroup and the transport system which might adversely modify the inclusion properties of the CD cavity; finally, there must be no reduction of the transport cavity, in particular the solubility of the complex.

Based on the above, we have developed spacer arm-saccharide-CD glyconjugate systems. Previously used carbohydrate-protein conjugates may have several types of spacer arms, of which the C9 spacer chain has been frequently used;[6,7] in view of their efficiency we adopted a similar strategy. In a preceding paper we have reported the synthesis of a new glycoconjugate based on glucose (**A**), a monosaccharide which is widely available but inactive in biological recognition processes.[8] We here describe the synthesis of the new β-CD derivatives carrying bio-active saccharide antennae, β-galactose (**B**), α-mannose (**C**), β-D-fucose (**D**) and β-L-fucose (**E**) and containing the biologically inert C9 spacer, synthesised for examination of the effect of hydrophobicity of the spacer on the solubility of the novel glycoconjugate and also for the effects of differing saccharide stereochemistry on the geometry of the system. The obtained systems will be tested for their biological activity as part of a wider programme on cyclodextrin saccharide conjugates.[9]

The specific chemical synthesis involves the fixation of β-D-galactopyranoside, α-D-mannopyranoside, β-L-fucopyranoside and β-D-fucopyranoside antennae via amide bonds at one end of the spacer ligand and β-CD attached via an amide function at the other end. We consider that the relative stability of the amide bond will allow the transport and recognition functions to remain linked under normal extracellular conditions.

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RESULTS AND DISCUSSION

The N-glycosidic linkage is an extremely important glyco-protein linkage.[10,11] In the present work we are interested in the series of saccharides D-Glucose (**A**), D-Galactose (**B**), D-mannose (**C**), D-Fucose (**D**), and L-Fucose (**E**), the stereochemistries of which are shown below in Figure 1. Apart from glucose, all of the saccharides are important in the processes of biological recognition. The use of a wide variety of stereochemistries will allow the determination of the effect of the saccharide head group on the physical properties of the glycoconjugates and, in particular, verification of the hypothesis that the use of a spacer will prevent specific interactions with the cyclodextrin transport system.

The 6^A-deoxy-6^A-[(9-(β-D-galactopyranosylamino)-1,9-dioxononyl)amino]β-cyclodextrin **7B**, the 6^A-deoxy-6^A-[(9-(α-D-mannopyranosylamino)-1,9-dioxononyl)-amino]β-cyclodextrin **7C** and the 6^A-deoxy-6^A-[(9-(β-D or β-L-fucopyranosylamino)-1,9-dioxononyl)amino]-β-cyclodextrin **7D** or **7E** were synthesised from the corresponding O-acetyl glycosylisothiocyanate, **2B**, **2C**, **2D** or **2E**, in four steps (Figure 2 in which the Mannose stereochemistry is shown).

The synthetic route depends largely on the facile conversion of the saccharide isothiocyanate compounds into amides. The starting compounds are prepared in a phase transfer reaction of the corresponding halides **1** with KSCN and Bu_4NBr in acetonitrile in the presence of molecular sieves; after chromatography the compounds are obtained as crystalline solids from slow evaporation of a dichloromethane solution. Condensation of **2B**, **2C**,[12] **2D** or **2E**, with mono-

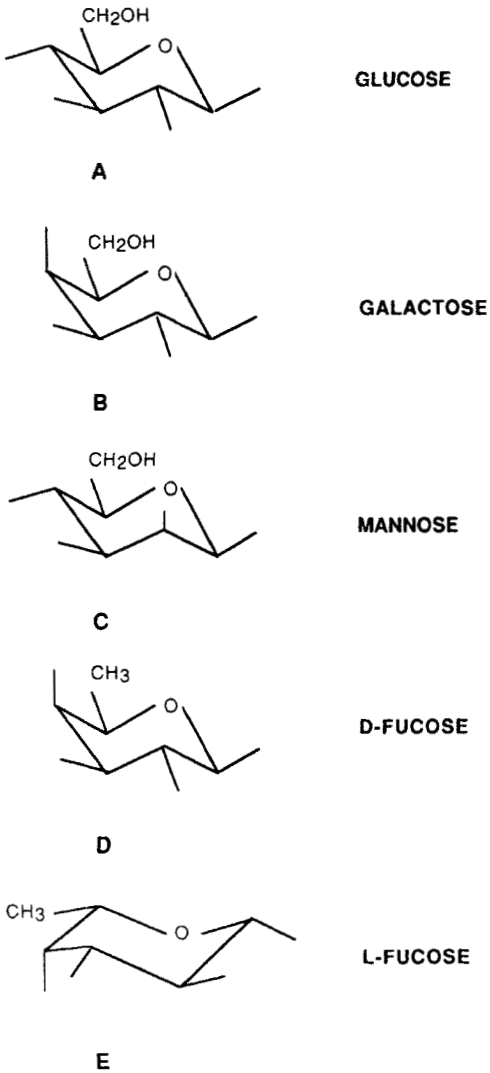

Figure 1 Schematic designs of the mono-saccharides that have been coupled to β-CD via the spacer arm.

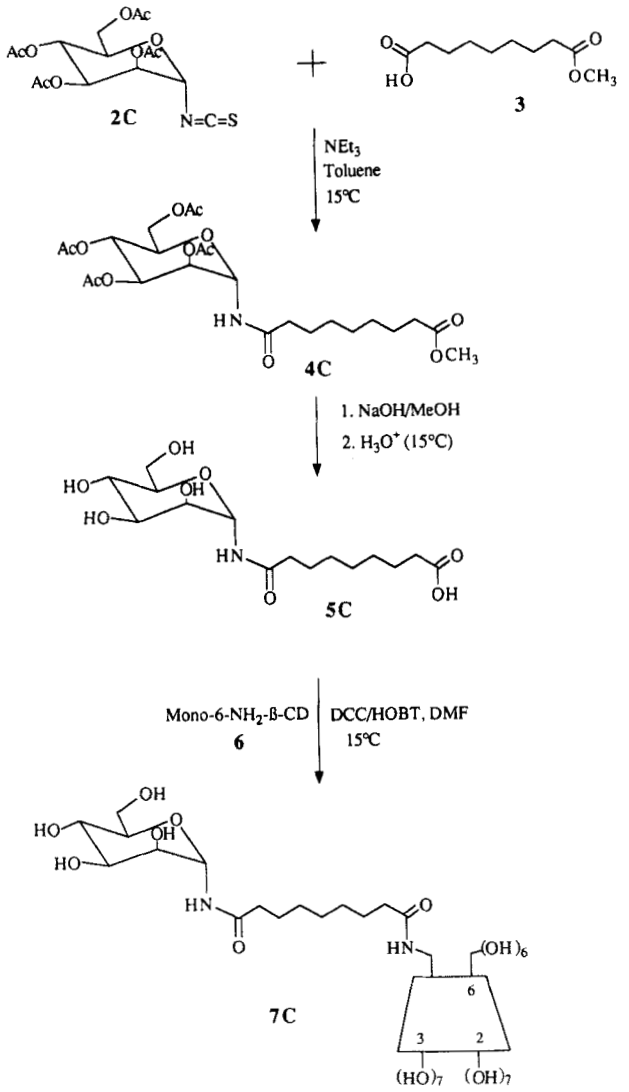

Figure 2 Synthesis of 6^A-deoxy-6^A-[(9-(α-D-manno(yranosylamino)-1,9-dioxononyl amino] β-cyclodextrin, (**7C**). This route is identical for **7B**, **7D** and **7E**.

methyl nonanedicarboxylate **3** in anhydrous toluene containing 0.1 molar equivalent of triethylamine, gave the protected glycosylamide **4B**, **4C**, **4D** or **4E** in 50% yield after chromatographic separation. The products are obtained as colourless oils which crystallise in standing at room temperature. **5B**, **5C**, **5D** and **5E** were prepared from the respective acetylated amido compounds by simultaneous deacetylation and de-esterification with methanolic sodium hydroxide (1 M). The extremely hygroscopic free acids, released by a simple neutralisation with 1 M hydrochloric acid, were immediately coupled to the previously reported 6^A-amino-6^A-deoxy-β-cyclodextrin[13] **6** using the standard peptide coupling system,[14] dicyclohexyl carbodiimide/hydroxybenzotriazole (DCC/HOBT) as the condensing agent. Extreme care must be used at this point to completely eliminate all traces of $NaCH_3CO_2$ formed during the saponification process. The final products are obtained in 40% to 45% yields after chromatography on silica gel (*n*-butanol: EtOH: H_2O). During certain early experiments, in which some $NaCH_3CO_2$ was present in the spacer, we observed preferential formation of mono-6-acetylamido-6-deoxy-β-CD **8**. The apparent higher reactivity of the acetate moiety leads to an enhancement in the percentage of **8** formed; removal of **8** is best achieved via reserved-phase HPLC.

The FAB MS of **7B**, **7C**, **7D** and **7E** confirm clean monosubstitution, with $[M+Na]^+$ peaks at 1487 for **7B**, **7C** and 1471 for **7D** and **7E**; in no case were peaks observed for products with higher degrees of substitution.

The structures of the compounds obtained were confirmed by ^1H and ^{13}C NMR; in particular the stereochemistries at the C1 carbon atoms of **4B**, **4C**, **4D** and **4E** were shown to be β and α,β,β respectively from the observed coupling constants (10 Hz, **4B**; 1.8 Hz, **4C**; 9.4 Hz, **4D**; 8.7 Hz, **4E**). NOE difference experiments show interactions between the Gal H-1 protons and the adjacent amide proton; in **7B** no NOE effects are seen between the Gal-amide proton and the β-cyclodextrin protons. The flexible spacer allows the hydrophilic saccharide antennae to be directed out into the aqueous environment. This is confirmed by the relatively small perturbation of the H-1 signals of the β-CD macrocycle; in cases in which there is strong interaction between the cavity and the pendant substitution the H-1 resonances are spread over up to 0.5 ppm.[13] The geometry of the conjugate then places the saccharide antenna at a long distance from the 'transporter' CD moiety, a stereochemistry which is favourable for antenna-lectin interactions.

All the β-CD derivatives, **7B**, **7C**, **7D** and **7E**, show enhanced aqueous solubilities with regard to the parent β-CD; thus the added hydrophilicity of the saccharide antenna is effectively more than balancing out the hydrophobicity of the C9 spacer.

Preliminary biological tests have shown that recognition of the Gal-spacer-CD system occurs.[15] We are currently synthesising molecules having other spacer systems and also with multiple substitutions to optimise the recognition process.

EXPERIMENTAL

General methods. Melting points were measured with a kofler apparatus and were not corrected. T,L,C, was performed on silica gel (F_{254}, MERCK) with detection by charring with H_2SO_4 (10%) and column chromatography was performed on silica gel 60. Optical rotations were measured in chloroform with a Perkin-Elmer Model 241 MC polarimeter. ^1H-NMR spectra were recorded with Bruker AC 200P and AM400 spectrometers. Fast-atom-bombardment mass spectra (FAB MS) were measured with a Kratos MS-80 mass spectrometer using the Magic Bullets technique.

-2,3,4,6-Tetra-O-acetyl-β-D-galactopyranosylisothyocyanate (2B)

Following the procedure of Camarasa *et al.*,[12] a mixture of potassium thiocyanate (4.89 g, 50.4 mmol), tetrabutylammonium bromide (7.85 g, 24.35 mmol) and molecular sieves (4 Å, 34 g) in anhydrous acetonitrile (1.120 mL) are stirred at room temperature for 2 h. 2,3,4,6-tetra-O-acetyl-α-D-galactopyranosyl bromide **1B** (10 g, 24.33 mmol) is added and the mixture is refluxed for 1 day. The mixture is filtered and the filtrate evaporated to dryness; column chromatography (toluene-acetone, 8:2) of the residue gave **2B**. R_f 0.39 (8.14 g, 86%); m.p. = 97–99 °C; $[\alpha]_D = +10°$ (c = 0.1, $CHCl_3$) (^1H-NMR ($CDCl_3$): δ_H 5.35 (dd, 1H, $J_{4,3}$ = 3.5 Hz, $J_{4,5}$ = 1.2 Hz, H-4), 5.28 (dd, 1H, $J_{2,3}$ = 10.3 Hz, $J_{2,1}$ = 8.6 Hz, H-2), 4.96 (dd, 1H, $J_{3,2}$ = 10.3 Hz, $J_{3,4}$ = 3.5 Hz, H-3), 4.93 (d, 1H, $J_{1,2}$ = 8.6 Hz, H-1), 4.08 (d, 2H, $J_{5,6a6b}$ = 6.2 Hz, H-6a and H-6b), 3.94 (m, 1H, $J_{5,6a6b}$ = 6.2 Hz, $J_{5,4}$ = 1.2 Hz, H-5), 2.12, 2.07, 2, 1.94 (s, 3H, CH_3-C=O).

-2,3,4-Tri-O-acetyl-β-D-fucopyranosylisothyocyanate (2D)

Using the procedure described for the preparation of **2B**, 2,3,4-tri-O-acetyl,6-deoxy-α-D-galactopyranosylchlorure (1 g, 3.24 mmol) and KSCN (630 mg, 6.48 mmol) and $(Bu)_4NBr$ (1.054 g, 3.24 mmol) and molecular sieves (4A, 3.35 g) in 120 mL acetonitrile gave **2D** after column chromatography (toluene-acetone, 8:2). R_f 0.59 (520 mg, 51%); mp = 94 °C; $[\alpha]_D = +20°$ (c = 0.1, $CHCl_3$). ^1H-NMR ($CDCl_3$): δ_H

5.34 – 5.22 (m, 2H, H-2, H-4), 5.03 (d, 1H, $J_{1,2}$ = 8.6 Hz, H-1); 4.93 (dd, 1H, $J_{3,2}$ = 8.9 Hz, $J_{3,4}$ = 3.3 Hz, H-3); 3.82 (q, 1H, $J_{H5,CH3}$ = 6.2 Hz, $J_{5,4}$ < 1 Hz, H-5); 2.18, 2.09, 1.97 (s, 3H, 3 CH_3-C=O); 1.2 (d, 3H, CH_3).

-2,3,4-Tri-O-acetyl-β-L-fucopyranosylisothyocyanate (2E)

Using the procedure for the preparation for **2B**, compound **2E** was prepared from the 2,3,4-tri-O-acetyl-6-deoxy α-L-galactopyranosylchlorure; purification by column chromatography (toluene-acetone, 8:2) gave 600 mg (56%) of **2E**. R_f 0.59; mp = 102–104 °C; ^{1}H-NMR ($CDCl_3$): δ_H 5.31 – 5.24 (m, 2H, H-2 and H-4), 5.04 – 4.94 (m, 2H, $J_{1,2}$ = 8.7 Hz, $J_{3,4}$ = 3.4 Hz, H-1 and H-3), 3.84 (q, 1H, $J_{H5,CH3}$ = 6.4 Hz, $J_{4,5}$ < 1 Hz, H-5), 2.20, 2.11, 1.99 (s, 3H, CH_3-C=O), 1.22 (d, 3H, $J_{CH3,H5}$ = 6.4 Hz, CH_3).

-1N-(8-Methoxycarbonyloctanoyl)-2,3,4,6-tetra-O-acetyl-β-D-galactopyranosylamine (4B)

To a solution of **2B** (16.2 g, 41.6 mmol) in dry toluene (200 mL) was added monomethyl nonanedicarboxylate **3** (12.12 g, 59.9 mmol) and 0.1 molar equivalent of triethylamine; the resultant solution was stirred for 5 days at room temperature. The organic phase was evaporated; flash-chromatography of the residue on a column of silica gel with 8:2 toluene-acetone gave **4B**. R_f 0.25 (12.6 g, 57%), m.p. = 89°, $[\alpha]_D$ = +25° (c = 0.48, $CHCl_3$). ^{1}H-NMR, COSY (acetone-d_6): δ_H 7.5 (d, 1H, $J_{NH,H1}$ = 10 Hz, NH), 5.32 (dd, 1H, $J_{4,3}$ = 3 Hz, $J_{4,5}$ = 1.2 Hz, H-4), 5.30 (dd, 1H, $J_{H1,NH}$ = 10 Hz, $J_{1,2}$ = 10 Hz, H-1), 5.16 (dd, 1H, $J_{3,2}$ = 10 Hz, $J_{3,4}$ = 3 Hz, H-3), 5.0 (dd, 1H, $J_{2,3}$ = 10 Hz, $J_{2,1}$ = 10 Hz, H-2), 4.2 (m, 1H, $J_{5,6a}$ = 6 Hz, $J_{4,5}$ = 1.2 Hz, H-5), 4.1 – 3.92 (m, 2H, $J_{6a,5}$ = $J_{6b,5}$ = 6 Hz, $J_{6a,6b}$ = 0 Hz, H-6a and H-6b), 3.5 (s, 3H, OCH_3), 2.25–2.05 (t, 4H, 2 CH_2 α to C=O), 2.02, 1.88, 1.80 (s, i 3/6/3, CH_3-C=O), 1.55 – 1.32 (m, 4H, 2 CH_2 β to C=O), 1.3 – 1.05 (m, 6H, 3 CH_2).

-1N-(8-Methoxycarbonyloctanoyl)-2,3,4,6-tetra-O-acetyl α-D-mannopyranosylamine (4C)

To a solution of 2,3,4,6-tetra-O-acetyl-α-D-mannopyranosylisothyocyanate **2C** (614 mg, 1.57 mmol) in dry toluene (7.5 mL) was added monomethyl nonanedicarboxylate **3** (447 mg, 2.2 mmol) and 0.1 molar equivalent of triethylamine the resultant solution was stirred for 3 days at room temperature. After removal of the solvent, the residue was chromatographed on silica gel with 8:2 toluene/acetone R_f 0.33 (335 mg, 40%) glass. ^{1}H-NMR (COSY, RCT) $CDCl_3$ δ_H 6.43 (d, 1H, $J_{NH,H1}$ = 9.5 Hz, NH), 5.51 (dd, 1H, $J_{H1,NH}$ = 9.5 Hz, $J_{1,2}$ = 1.8 Hz, H-1), 5.29 (d, 1H, $J_{2,1}$ = 1.8 Hz, $J_{2,3}$ = 0 Hz, H-2), 5.15 (m, 1H, H-4), 5.05 (m, 1H, $J_{3,4}$ = 12-5 Hz, H-3), 4.21/3.92 (m, 2H, $J_{6a,6b}$), 4.0 (d, 1H, $J_{3,4}$, = 12.5 Hz, H-3), 3.63 (m, 1H, H-5), 3.58 (s, 3H, OCH_3), 2.2 (t, 2H, CH_2 α to amide), 2.05 (t, 2H, CH_2 α to ester), 2.1, 2.0, 1.97, 1.89 (s, 3H, CH_3-C=O), 1.55 (m, 4H, 2 CH_2), 1.22 (m, 6H, 3 CH_2).

-1N-(8-Methoxycarbonyloctanyl)-2,3,4-tri-O-acetyl-β-D-fucopyranosylamine (4D)

Using the procedure for the preparation for **4B**, **4D** was prepared from **2D** (520 mg, 1.6 mmol), monomethyl nonanedicarboxylate **3** (577 mg, 2.2 mmol) and 0.1 molar equivalent of triethylamine. Purification by column chromatography (toluene-acetone, 8:2) gave **4D** (200 mg, 26%), mp = 92–96 °C, R_f = 0.35, $[\alpha]_D$ = +10° (c = 0.1, $CHCl_3$), ^{1}H-NMR (COSY, RCT) $CDCl_3$: δ_H 6.2 (d, 1H, $J_{NH,H1}$ = 8.7 Hz, NH), 5.28 (d, 1H, $J_{4,3}$ = 1.9 Hz, H-4), 5.17 (m, 1H, $J_{1,2}$ = 9.4 Hz, H-1), 5.17–5.02 (m, 2H, $J_{3,2}$ = 7.5 Hz, $J_{3,4}$ = 1.9 Hz, H-3 and H-2), 3.9 (q, 1H, $J_{H5,CH3}$ = 6.5 Hz, $J_{5,4}$ < 1 Hz, H-5), 3.65 (s, 3H, OCH_3), 2.3 (t, 2H, CH_2 α to ester), 2.2 (t, 2H, CH_2 α to amide), 2.16, 2.04, 1.99 (s, 3H, CH_3-C=O), 1.6 (m, 4H, 2CH_2), 1.29 (m, 6H, 3CH_2), 1.19 (d, 3H, CH_3).

-1N-(8-Methoxycarbonyloctanoyl)-2,3,4-tri-O-acetyl-β-L-fucopyranosylamine (4E)

Using the procedure for the preparation for **4B**, compound **4E** was prepared from **2E** (600 mg, 2.84 mmol), monomethyl nonanedicarboxylate **3** (820 mg, 3.98 mmol) and 0.1 molar equivalent of triethylamine. Purification by column chromatography (toluene-acetone, 9:1) gave **4E** (240 mg, 18%), R_f = 0.11. ^{1}H-NMR (COSY) $CDCl_3$: δ_H 6.23 (d, 1H, $J_{NH,H1}$ = 8.7 Hz, NH), 5.28 (d, 1H, $J_{4,3}$ = 1.8 Hz, $J_{4,5}$ < 1 Hz, H-4), 5.20 (t, 1H, $J_{1,2}$ = $J_{NH\text{-}H1}$ = 8.7 Hz, H-1), 5.11 – 5.07 (m, 2H, $J_{2,3}$ = 5.8 Hz, H-2 and H-3), 3.92 (q, 1H, $J_{H5,CH3}$ = 6.3 Hz, H-5), 3.66 (s, 3H, OCH_3), 2.29 (t, 4H, 2CH_2 α to C=O), 2.16, 2.04, 1.99 (s, 3H, CH_3-C=O), 1.6 (m, 4H, 2CH_2 β to CO), 1.3 (m, 6H, 3CH_2), 1.18 (d, 3H, $J_{CH3,H5}$ = 6.3 Hz, CH_3 fuc).

-1-N-(8-Carboxyoctanoyl)-β-D-galactopyranosylamine (5B)

Compound **4B** (3 g, 5.74 mmol) was O-deacetylated and de-esterified with methanolic sodium hydroxide (1 M) for 4 h at room temperature. The free acid **5B** (1.7 g, 85%) was released by a simple neutralisation with HCl (1 M) to yield the product after evaporation and treatment with methanol. mp = 128 °C. $[\alpha]_D$ = +15° (c = 0.2, MeOH). ^{1}H-NMR, COSY (DMSO-d_6): δ_H 8.5 (d, 1H, $J_{NH,1}$ = 10 Hz, NH), 4.6 (m, 1H, H-1), 3.65 (m, 1H, H-3), 3.60 – 3.35 (m, 4H glucos.), 3.30 (m,

1H, H-2), 2.05 (t, 4H, 2 CH_2 α to C=O), 1.45 (m, 4H, 2 CH_2 β to C=O), 1.3 (m, 6H, 3 CH_2).

-1-N-(8-Carboxyoctanoyl)-α-D-mannopyranosylamine (5C)

A mixture of **4C** (335 mg, 0.63 mmol) and a methanolic solution (0.5 M) in NaOH (6.4 mL) was stirred for 5 h at room temperature. After removal of the solvent, the residue was washed with hydrochloric acid (1 M) and dichloromethane. The aqueous phase was evaporated and the white product, **5C** washed with MeOH. (181 mg, 82%) glass. ^{1}H-NMR (DMSO-d_6): δ_H 8.0 (d, 1H, $J_{NH,H1}$ = 9.5 Hz, NH), 4.98 (dd, 1H, $J_{H1,NH}$ = 9.5 Hz, $J_{1,2}$ = 1.8 Hz, H-1), 3.55 (d, 1H, H-2), 3.5 – 3 (m, H man), 2.15 (t, 2H, CH_2 α to amide), 2.10 (t, 2H, CH_2 α to acide), 1.45 (m, 4H, 2 CH_2), 1.22 (m, 6H, 3 CH_2).

1-N-(8-Carboxyoctanoyl)-β-D-fucopyranosylamine (5D)

Using the procedure for the preparation for **5B**, compound **5D** was prepared from **4D** (80 mg, 0.169 mmol). The free acid was triturated with methanol (hygroscopic). ^{1}H-NMR COSY (D_2O): δ_H 8.7 (d, 1H, $J_{NH,H1}$ = 8.9 Hz), 4.9 (m, 1H, $J_{1,2}$ = 9.2 Hz, H-1), 3.9 – 3.5 (m, 4H, H-2, H-3, H-4, H-5), 2.4 – 2.3 (2t, 4H, 2CH_2), 1.6 (brd, 4H, 2CH_2), 1.3 (brd, 6H, 3CH_2), 1.2 (d, 3H, $J_{CH3,H5}$ = 6.4 Hz, CH_3 fuc).

1-N-(8-Carboxyoctanoyl)-β-L-fucopyranosylamine (5E)

Using the procedure for the preparation for **5B**, compound **5E** was prepared from **4E** (240 mg, 0.507 mmol). The free acid was treated with methanol (hygroscopic). ^{1}H-NMR COSY (DMSO-d_6): δ_H 8.3 (d, 1H, $J_{NH,H1}$ = 8.7 Hz, NH), 4.6 (t, 1H, H-1), 3.5 (q, 1H, $J_{CH3,H5}$ = 6.3 Hz, H-5), 3.5 – 3.3 (m, 3H, H-4 and H-3 and H-2), 2.25 (t, 2H, CH_2 α to C=O), 1.2 (m, 6H, 3CH_2), 1.02 (d, 3H, $J_{CH3,H5}$ = 6.3 Hz, CH_3 fuc).

-6^A-deoxy-6^A-[(9-(β-D-galactopyranosylamino)-1,9-dioxononyl)amino]-β-cyclodextrin (7B)

6^A-deoxy-6^A-amino β-cyclodextrin[13] **6** 2.84 g, 2.5 mmol was condensed with 1N-(8-carboxyoctanoyl)-β-D-galactopyranosylamine **5B** (876 mg, 2.50 mmol) in N,N-dimethylformamide ((DMF) 40 mL at 15 °C using dicyclohexylcarbodiimide/hydroxybenzotriazole (DCC/HOBT) as coupling reagents (619 mg, 3 mmol/405 mg, 3 mmol). After 4 days, evaporation of the solvent and chromatography of the product on a column with 5:4:3 *n*-butanol-EtOH-H_2O gave **7B** R_f 0.3 (1.4 g, 39%), mp dec. *180–182* °C, $[\alpha]_D$ = +200° (c = 0.1, MeOH). ^{1}H-NMR (COSY, HOHAHA 400 MHz) (DMSO-d_6): δ_H 8.26 (d, 1H, $J_{NH,1}$ = 10 Hz, NH gal), 7.55 (brd, 1H, NHβ-CD), 5.7 (brd, 16H, OH-2 and OH-3 β-CD and OH-2 and OH-3 gal), 4.85 (d, 7H, H-1, β-CD), 4.75 (d, 1H, H-1 gal), 4.45 (brd, 6H, OH-6 β-CD), 3.65 (m, 1H, H-3 gal), 3.6 (m, 7H, H-3, β-CD), 3.6 – 3.2 (brd, H gluc. β-CD and H-2 gal), 2.1 (t, 4H, 2 CH_2 α to C=O), 1.45 (m, 4H, 2 CH_2 β to C=O), 1.2 (m, 6H, 3 CH_2). FABMS: $(M + Na)^+$ = 1487.

-6^A-deoxy-6^A-[(9-(α-D-mannopyranosylamino)-1,9-dioxononyl)amino]β-cyclodextrin (7C)

Condensation of 1-N (8-carboxyoctanoyl)-α-D-mannopyranosylamine **5C** (171 mg, 0.49 mmol) with 6^A-deoxy-6^A-amino β-cyclodextrin[13] **6** (555 mg, 0.49 mmol) in anhydrous N,N-dimethylformamide (10 mL) in the presence of DCC (121 mg, 0.59 mmol/HOBT (79 mg, 0.59 mmol). After 3 days at room temperature, the insoluble material was removed by filtration and the filtrate was evaporated. Chromatography of the residue on a column of silica gel with 5:4:3 *n*-butanol-EtOH-H_2O gave **7C** (322 mg, 45%) mp = dec 250 °C. $[\alpha]_D$ = +114° (c = 0.88, MeOH). ^{1}H-NMR (COSY, RCT2) (DMSO-d_6): δ_H 7.90 (d, 1H, $J_{NH,H1}$ = 9.5 Hz, NH man), 7.65 (brd, 1H, NH-βCD), 5.7 (brd, OH-2 and OH-3 β-CD and man), 5.0 (d, 1H, H-1 man), 4.80 (d, 7H, H-1, β-CD), 4.5 (brd, 7H, OH-6βCD and H-4 man), 3.6 – 3.55 (brd, Hgluc. β-CD and man), 3.52 (m, 1H, H-2 man), 2.10 (m, 4H, 2CH_2 α to C=O), 1.45 (m, 4H, 2CH_2 β to C=O), 1.2 (m, 6H, 3CH_2). FABMS: $(M + Na)^+$ = 1487.

-6^A-deoxy-6^A-[(9-(β-D-fucopyranosylamino)-1,9-dioxononyl)amino]-β-cyclodextrin (7D)

6^A - deoxy - 6^A - amino β-cyclodextrin[13] **6** (264 mg, 0.23 mmol) was condensed with **5D** (77.5 mg, 0.23 mmol) using the process previously described. Purification by flash-chromatography (*n*-butanol-EtOH-H_2O, 5:4:3) gave **7D** (70 mg, 21%). mp = dec 190 °C. $[\alpha]_D$ = −437° (c = 0.63, H_2O). ^{1}H-NMR (COSY, RCT) (DMSO-d_6): δ_H 8.2 (d, 1H, $J_{NH,H1}$ = 8.9 Hz, NH fuc), 7.6 (brd, 1H, NHβ-CD), 5.8 – 5.6 (brd, 14H, OH-2 and OH-3β-CD), 4.8 (d, 7H, H-1, β-CD), 4.7 (m, 1H, OH-2 fuc), 4.6 (m, 1H, H-1 fuc), 4.4 (brd, 6H, OH-6 β-CD), 3.7 – 3.2 (m, Hgluc. β-CD and H-3, H-4 fuc), 3.5 (q, 1H, H-5 fuc), 3.3 (m, 1H, H-2 fuc), 3.25 (brd, 7H, H-2 β-CD), 2.05 (t, 2H, CH_2), 1.4 – 1.1 (brd, 12H, 6 CH_2), 1.05 (d, 3H, $J_{CH3,H5}$ = 6.4 Hz, CH3 fuc). FABMS: $(M + Na)^+$ = 1471.

-6^A-deoxy-6^A-[(9-(β-L-fucopyranosylamino)-1,9-dioxononyl)amino]-β-cyclodextrin (7E)

6^A - deoxy - 6^A - amino β-cyclodextrin[13] **6** (574 mg, 0.5 mmol) was condensed with **5E** (166.5 mg, 0.5 mmol) using the process previously described. Purification by flash-chromatography (*n*-butanol-EtOH-H_2O, 5:4:3) gave **7E** (294 mg, 40%). mp = dec 190 °C. $[\alpha]_D$ = +67° (c = 1.0, H_2O). ^{1}H-NMR (COSY, RCT) (DMSO-d_6):

δ_H 8.2 (d, 1H, $J_{NH,H1} = 8.2$ Hz, NH fuc), 7.55 (brd, 1H, NHβ-CD), 5.9 – 5.6 (brd, 16H, OH-2 and OH-3β-CD and OH-2 and OH-3 fuc), 4.8 (d, 7H, H-1, β-CD), 4.6 (m, 1H, H-1 fuc), 4.5 (m, 7H, H-4 β-CD), 3.7 (m, 7H, H-3 β-CD), 3.4 (q, 1H, H-5 fuc), 3.5 – 3.3 (brd, Hgluc β-CD and H-2 and H-3 fuc), 2.05 (t, 2H, CH2 α to C=O ester), 1.4 (m, 2H, CH_2 α to C=O amide), 1.3 (m, 10H, $5CH_2$), 1.10 (d, 3H, $J_{CH3,H5} = 6.5$ Hz, CH3 fuc) FABMS: $(M + Na)^+ = 1471$.

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