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Synthesis of novel trivalent amino acid glycoconjugates based on the cyclotriveratrylene ('CTV') scaffold

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The convenient synthesis of novel trivalent amino acid glycoconjugates based on cyclotriveratrylene ('CTV') is described. These constructs consist of the CTV scaffold, three oligoethylene glycol spacers of variable length connected to a glyco amino acid residue which can also be varied. The resulting library of trivalent glycoconjugates can be used for studying multivalent interactions.

Introduction

Binding of two molecules to each other, *e.g.* a substrate and an enzyme, constitutes the key event in many biochemical processes leading to the biological effect. In many cases each of the interacting partners has a single interaction site, and the resulting binding event is therefore called *monovalent*. Such an interaction can be remarkably effective: a striking example is the binding of biotin to streptavidin, which has a binding constant of approximately 2×10^{13} M^{-1}.[1] When two or more binding sites in one biomolecule bind to multiple receptor sites on another, the resulting interaction is called *multivalent*. Often, a multivalent interaction is orders of magnitude stronger than the corresponding monovalent system.[2] Multivalency plays a crucial role in many important biological interactions.[3] Outstanding examples in this respect are antibody–antigen interactions and cell–cell recognition processes.[4] In recent years, the affinity gain that can be achieved by transforming monovalent binding partners to multivalent systems has received considerable attention and has shown to be especially powerful for enhancing relatively weak ligand–receptor interactions.[5] A typical example was investigated in our group, where a dendrimer loaded with eight lactose moieties was found to bind the pentavalent cholera toxin 545 times as strong as monovalent lactose itself. This constitutes a 68-fold increase in affinity *per* lactose residue.[6a] A more recent example from our group describes rigidified multivalent lactose molecules and their interactions with mammalian galectins.[6b]

As part of our program aimed at designing molecules that can block the flow of nutrients through bacterial general porins, which may thus serve as a novel class of antibiotics, we decided to prepare trivalent "stopper" ("corks") constructs (Fig. 1) which could have a multivalent interaction with the trimeric general porins. As a trivalent scaffold, we chose the earlier introduced cyclotriveratrylene (CTV) scaffold,[7–9] which has a number of advantages: (i) it is readily accessible,[10] (ii) it can be easily derivatized, and (iii) its geometry is well-defined and assures that all three arms are oriented in parallel. As spacers connecting the scaffold and the interaction sites, oligoethylene glycol moieties were selected. These spacers have already proved their usefulness in a number of applications.[2d,11] They are water soluble, commercially available in a variety of lengths, and they can be easily derivatized. At the end of each spacer, a glyco amino acid was attached as a "bait". The carbohydrate moiety can be considered the natural "substrate" for general porins since these proteins allow the uptake of small peptides and carbohydrates. The amino acid residue was incorporated to possibly allow an extra ionic interaction with the charged amino acids which are abundant in the porin channel wall. As we have described before, such glyco amino acid residues can be conveniently prepared using different amino acids and carbohydrate moieties.[12]

It is shown here that all parts of the functional arms can be varied: (i) spacers of different lengths can be introduced (*e.g.* diethyleneglycol to tetraethyleneglycol), (ii) different functionalized amino acids can be used (*e.g.* alanine, glutamic acid, or lysine), and (iii) different carbohydrate residues can be attached (*e.g.* glucose, galactose, or the disaccharide lactose). This allows for the preparation of a combinatorial library of trivalent amino acid glycoconjugate constructs, which can be screened for optimal performance in the desired interaction.[13]

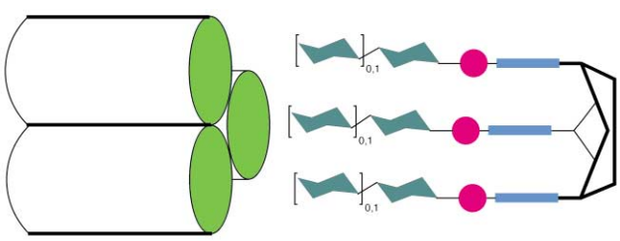

Fig. 1 General representation of trivalent glyco (green)–amino acid (pink) conjugates attached *via* a spacer (blue) to a suitable scaffold (black). Such a construct might fit into the trimer of a general porin (left).

Results and discussion

We thought that it was important to develop a synthesis which in principle could lead to a considerable diversity of the CTV molecular constructs. Therefore we decided to incorporate the possibility of varying the building blocks at all stages of the synthesis of the trivalent glycoconjugates. This might lead to libraries of compounds which is useful for an eventual biological evaluation.

First, the parallel synthesis of a small library of the novel trivalent glycoconjugates was realized by connection of oligoethyleneglycol spacers **1a**–**c** to the cyclotriveratrylene scaffold **3** (Scheme 1). In order to achieve this coupling *via* an ether linkage, the spacers had to be selectively furnished with one leaving group. Thus, a fourfold excess of oligoethyleneglycol moieties **1a**–**c** was treated with tosyl chloride. After reaction, the excess of spacer could be easily removed by an aqueous work-up. In all cases, the desired mono-tosylated products **2a**–**c** were formed exclusively. However, the reaction was never quantitative, and a small amount of tosyl chloride always remained which had to be removed by column chromatography. After

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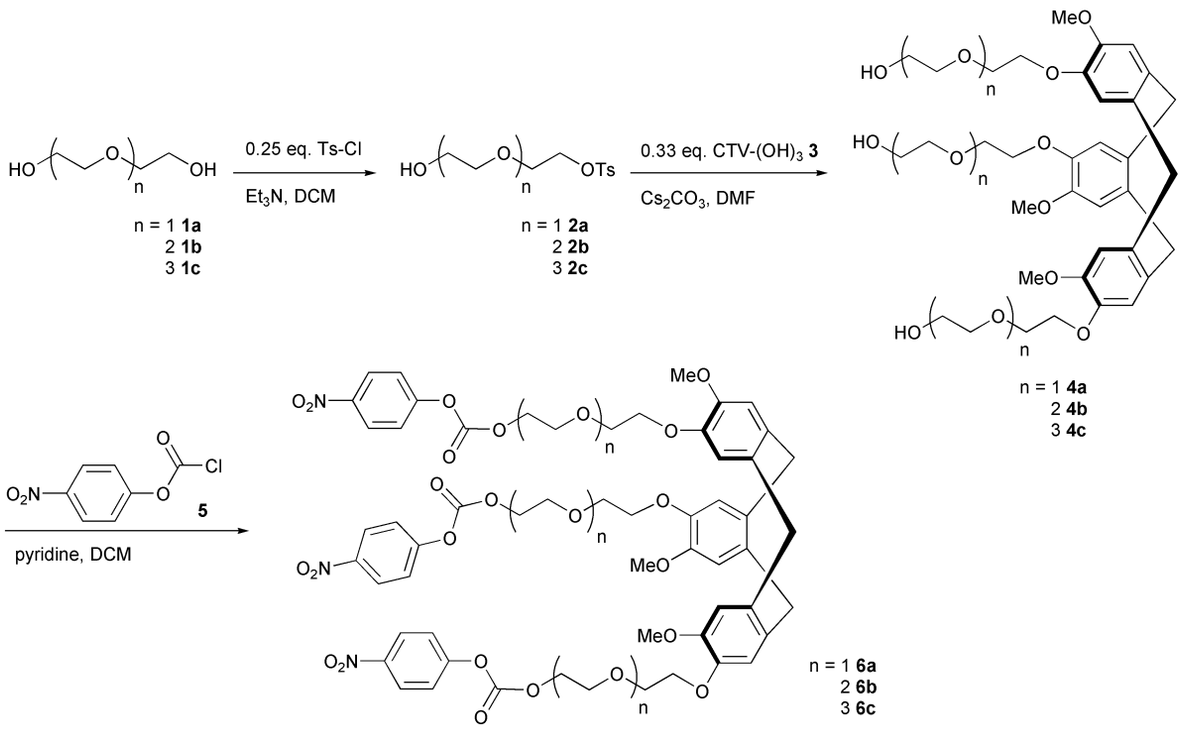


Scheme 1 Attachment of spacers to the CTV scaffold followed by conversion to the *para*-nitrophenylcarbonate.

purification, the mono-tosylated spacers were obtained in good yields (74–92%).

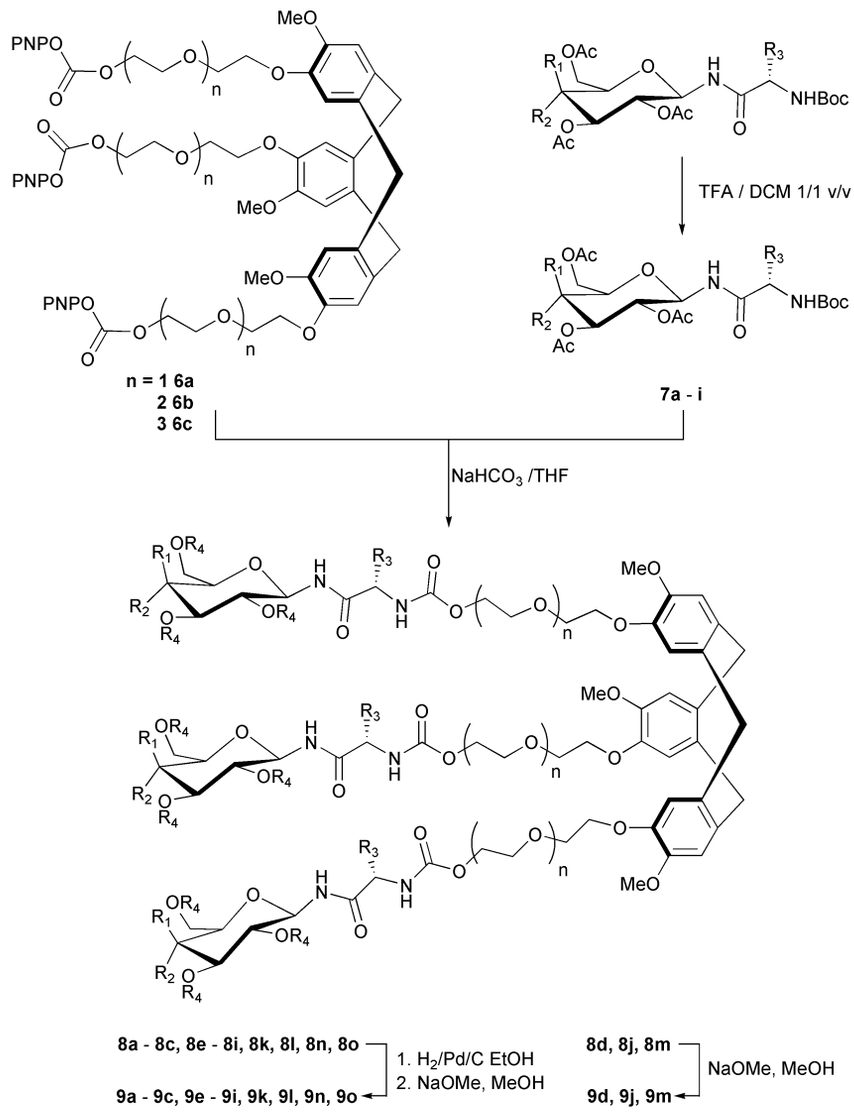
Then, three equivalents of tosylated spacers **2a**–**c** were added to a solution of CTV-$(OH)_3$ **3** in DMF. Using caesium carbonate as a base, the coupling reaction proceeded smoothly overnight. The resulting crude materials were subjected to aqueous work-up and column chromatography, after which the desired spacer-scaffold constructs **4a**–**c** were obtained in good yields (75–90%). In the next step, constructs **4a**–**c** were "activated" by transforming the three hydroxy groups into the corresponding *p*-nitrophenyl carbonate moieties. This was achieved by treatment of constructs **4a**–**c** with three equivalents of *p*-nitrophenyl chloroformate **5** using pyridine as a base. This reaction proceeded smoothly in one hour at room temperature, and after column chromatography the pure "activated" constructs **6a**–**c** were obtained in good yields (54–82%).

Subsequently, glyco amino acid monomers **7a**–**i**, which were prepared as described previously,[12] had to be coupled to spacer-scaffold constructs **6a**–**c** (Scheme 2). This reaction turned out to be problematic. The first step, *i.e.* removal of the Boc-group of monomers **7a**–**i** either with hydrochloric acid in diethyl ether or with TFA, posed no difficulties and gave quantitative yields of the desired deprotected glyco amino acids. However, coupling of the resulting salts to activated constructs **6a**–**c** was cumbersome and afforded the desired trivalent glycoconjugates **8a**–**o** in very poor yields. DiPEA, Et_3N, pyridine, and NMM were used as the base in this reaction, each giving similarly poor results even when large excesses of the nucleophile were used. In addition, the temperature was varied, by performing the coupling reaction in DCM or THF at reflux temperature. Still, only low yields of the products were obtained which had to be laboriously isolated from a complex mixture containing *inter alia* structures having only one or two complete arms. Next, it was decided to convert the salts of the glyco amino acids building blocks into free amines prior to coupling in order to enhance their nucleophilicity. This could be accomplished by dissolving the salt in DCM followed by washing with an aqueous sodium bicarbonate solution. The free amines coupled much more readily to activated constructs **6a**–**c**. Refluxing overnight in THF in the presence of nine equivalents of Et_3N led to the formation of appreciable amounts of glycoconjugates **8a**–**o**.

However, the high temperature required and the ensuing tedious purification by column chromatography were considered unsuitable for a combinatorial approach. Fortunately, we found that performing the coupling reaction in THF using sodium bicarbonate as the base led to smooth formation of trivalent glycoconjugates **8a**–**o**, even at room temperature. The reaction proceeded cleanly, and the products could be easily purified. In addition, these conditions also allowed the use of the TFA salts obtained from glyco amino acids **7a**–**i** after Boc deprotection, so that the extra step liberating the amine could be dispensed with.

Using the protocol described above, a small library of trivalent amino acid glycoconjugates could be prepared in a parallel fashion. The yields obtained were acceptable (Table 1) and could probably be improved by optimizing the column chromatography conditions for each compound. In view of the combinatorial nature of this synthesis, these adjustments were not carried out. Finally, trivalent glycoconjugates **8a**–**o** were deprotected in either one or two stages. In the compounds having a glutamic acid or lysine residue (**8a**–**c**,**e**–**i**,**k**–**l**,**n**–**o**), the benzyl ester or the Cbz group were first removed by catalytic hydrogenolysis in the presence of palladium on carbon. The acetyl protecting groups on the carbohydrate residues were removed by treatment of the conjugates with a solution of sodium methoxide in methanol. After neutralization of the reaction mixture with Dowex-H^+ ion exchange resin, the desired fully deprotected trivalent glycoconjugates ("sugar corks") **9a**–**o** were obtained. The yields in these deprotection steps were slightly disappointing (Table 1), particularly since product loss was not caused by the formation of side products as was judged by TLC. It was found that a significant loss was caused by binding of the products to the Dowex ion exchange resin. This was demonstrated in the case of the maltose derivatives **9m**–**o**, in which the resin was more thoroughly rinsed with water and the yields obtained were much improved. In order to demonstrate the quality of the final products, typical HPLC traces as obtained for **9m**–**o** are shown in Fig. 2.

Clearly, the HPLC traces of the sugar corks **9a**–**o** showed two peaks, which could be separated and both gave the same mass spectrum. This was caused by the chirality of



Scheme 2 Introduction of the *N*-linked glyco amino acids onto the activated CTV scaffold (R_1–R_4: Table 1).

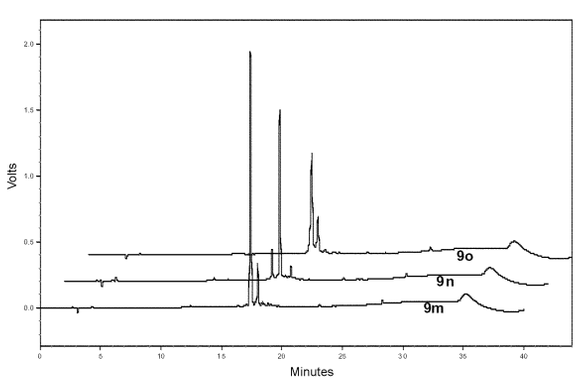


Fig. 2 HPLC traces of CTV amino acid glycoconjugates **9m**–**o**.

the cyclotriveratrylene molecule. Consequently, when enantiopure glyco amino acids were coupled to racemic scaffold, diastereomers were obtained. Typically diastereomeric ratios of 1 : 3 up to 1 : 7 were found, indicating that formation of one of both diastereomers was more favorable. It is known that homochiral CTV derivatives in solution racemize quickly when heated above room temperature. In order to determine the optical stability of the prepared CTV-derivative, a sample of **9m** in water was heated to 60 °C. However, even after 48 hours at this temperature, the diastereomeric ratio – as determined by HPLC – remained constant.

Conclusions

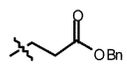
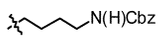
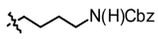
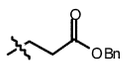
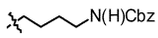
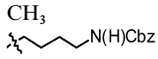
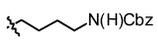
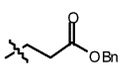
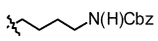
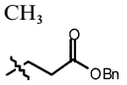
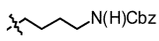
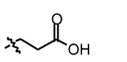
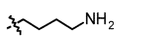
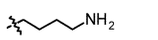
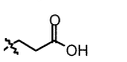
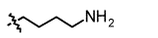
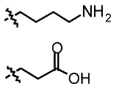
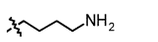
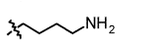
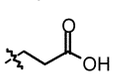
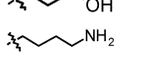
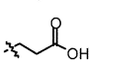
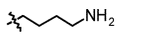
A convenient procedure for the preparation of novel cyclotriveratrylene based trivalent glycoconjugates was developed, demonstrating that CTV is a versatile scaffold for attachment of functional biomolecules. The key step in this synthesis was the coupling of an *N*-linked glyco amino acid building block to an alcohol "activated" as a *p*-nitrophenylcarbonate. It was found that this coupling could be effected using sodium bicarbonate as a base in THF. This synthesis was then used to prepare in parallel a small library of the trivalent constructs, in which the spacer length and the identity of the glyco amino acid were varied. This library can be used to select the optimal library member for a given multivalent interaction.

Experimental

General

Unless stated otherwise, chemicals were obtained from commercial sources and used without further purification. Solvents were purchased from Biosolve (Valkenswaard, The Netherlands) and were stored on molecular sieves (4 Å). DiPEA, Et_3N, and NMM were distilled from ninhydrin and KOH prior to use. Pyridine was distilled from KOH. Column chromatography was performed on ICN Silica 60 Å, 32–100 μm. TLC was performed on Merck precoated Silica 60 plates. Spots were visualized using UV-light, ninhydrin, and 10% sulfuric acid in methanol.

Table 1 Results of the CTV library synthesis

	n	R_1	R_2	R_3	R_4	Yield (%)
8a	3	OAc	H		Ac	34
8b	3	H	OAc		Ac	29
8c	3	OAc	H		Ac	47
8d	3	H	β-Gal(OAc)$_4$	CH_3	Ac	22
8e	3	H	β-Gal(OAc)$_4$		Ac	15
8f	3	H	β-Gal(OAc)$_4$		Ac	27
8g	2	OAc	H	CH_3	Ac	48
8h	2	H	OAc		Ac	24
8i	2	OAc	H		Ac	46
8j	2	H	β-Gal(OAc)$_4$	CH_3	Ac	39
8k	2	H	β-Gal(OAc)$_4$		Ac	43
8l	2	H	β-Gal(OAc)$_4$		Ac	48
8m	3	H	α-Glc(OAc)$_4$	CH_3	Ac	35
8n	3	H	α-Glc(OAc)$_4$		Ac	18
8o	3	H	α-Glc(OAc)$_4$		Ac	37
9a	3	OH	H		H	34
9b	3	H	OH		H	15
9c	3	OH	H		H	25
9d	3	H	β-Gal	CH_3	H	28
9e	3	H	β-Gal		H	38
9f	3	H	β-Gal		H	67
9g	2	OH	H		H	44
9h	2	H	OH		H	28
9i	2	OH	H		H	65
9j	2	H	β-Gal	CH_3	H	48
9k	2	H	β-Gal		H	65
9l	2	H	β-Gal		H	41
9m	3	H	α-Glc	CH_3	H	72
9n	3	H	α-Glc		H	77
9o	3	H	α-Glc		H	76

300 MHz NMR spectroscopy was performed on a Varian G-300 apparatus. ESI-MS experiments were performed on a Shimadzu LCMS QP8000 system. MALDI-TOF measurements were carried out on a Kratis Analytical Axima-CFR machine. Analytical HPLC was performed on a Shimadzu Class-VP automated HPLC using an analytical reversed-phase column (Alltech Adsorbosphere C8, 90 Å, 5 μm, 250 × 4.6 mm) and a UV detector operating at 220 nm and 254 nm. Elution was effected using an appropriate gradient from 0.1% TFA in water to 0.085% TFA in acetonitrile–water (1 : 1, v/v) using a flow rate of 1 ml min^{-1}.

Toluene-4-sulfonic acid 2-(2-hydroxyethoxy)ethyl ester 2a. Diethyleneglycol **1a** (4.24 g, 40 mmol) and triethylamine (2.79 mL, 20 mmol) were dissolved in DCM (50 mL). Then, tosyl chloride (1.91 g, 10 mmol) was added in one portion. The resulting mixture was stirred for one hour at room temperature. After washing with 1 M $KHSO_4$ and 5% $NaHCO_3$ and drying on Na_2SO_4, the crude product was obtained by evaporation and purification by column chromatography over silica (eluent: 5% MeOH–DCM). The pure mono-tosylated product was obtained as a clear, colorless oil (1.93 g, 74%). R_f (2% MeOH–DCM) = 0.19. δ (300 MHz; $CDCl_3$; Me_4Si) 2.38 (1H, br t, O*H*), 2.45 (3H, s, CH_3), 3.52 (2H, t, OCH_2), 3.67 (4H, m, 2 × OCH_2), 4.19 (2H, t, OCH_2), 7.35 (2H, d, Ar*H*, ^3J = 8.1 Hz), 7.80 (2H, d, Ar*H*, ^3J = 8.1 Hz); δ (75.5 MHz; $CDCl_3$; Me_4Si) 21.5 (*C*H$_3$), 61.4, 69.4, 69.1, 72.4 (O*C*H$_2$), 127.8, 129.8, 132.7, 144.9 (Ar*C*).

Toluene-4-sulfonic acid 2-[2-(2-hydroxyethoxy)ethoxy]ethyl ester 2b. This compound was prepared from triethyleneglycol **1b** (6.01 g, 10 mmol) following the procedure for **2a**. The mono-tosylated product was obtained as a clear, colorless oil (2.80 g,

92%). R_f (10% MeOH–DCM) = 0.56. δ (300 MHz; $CDCl_3$; Me_4Si) 2.45 (3H, s, CH_3), 2.54 (1H, br s, OH), 3.58 (6H, m, 3 × OCH_2), 3.70 (4H, m, 2 × OCH_2), 4.17 (2H, m, OCH_2), 7.35 (2H, d, ArH, ^3J = 8.2 Hz), 7.79 (2H, d, ArH, ^3J = 8.2 Hz); δ (75.5 MHz; $CDCl_3$; Me_4Si) 21.5 (CH_3), 61.5, 68.5, 69.1, 70.1, 70.6, 72.3 (OCH$_2$), 127.8, 129.7, 132.7, 144.8 (ArC).

Toluene-4-sulfonic acid 2-{2-[2-(2-hydroxyethoxy)ethoxy]-ethoxy}ethyl ester 2c. This compound was prepared from tetraethyleneglycol **1c** (7.77 g, 10 mmol) following the procedure for **2a**. The mono-tosylated product was obtained as a clear, colorless oil (1.87 g, 54%). R_f (10% MeOH–DCM) = 0.51. δ (300 MHz; $CDCl_3$; Me_4Si) 2.45 (3H, s, CH_3), 2.81 (1H, br s, OH), 3.64 (14H, 7 × OCH_2), 4.16 (2H, m, OCH_2), 7.35 (2H, d, ArH, ^3J = 8.2 Hz), 7.80 (2H, d, ArH, ^3J = 8.2 Hz); δ (75.5 MHz; $CDCl_3$; Me_4Si) 21.4 (CH_3), 61.4, 68.5, 69.1, 70.1, 70.2, 70.4, 70.5, 72.3 (OCH$_2$), 127.8, 129.7, 132.7, 144.7 (ArC).

CTV-*O*-tris(diethyleneglycol) 4a. Cyclotriveratrylene **3** (788 mg, 1.93 mmol) and toluene-4-sulfonic acid 2-(2-hydroxyethoxy)-ethyl ester **2a** (1.41 g, 6.38 mmol) were dissolved in DMF (5 mL). Caesium carbonate (2.20 g, 6.76 mmol) was added, and the resulting mixture was stirred overnight at room temperature. Subsequently, the solvent was evaporated, and the crude product was redissolved in DCM (10 mL) and 1 M $KHSO_4$ (10 mL) was added. The layers were separated, and the organic layer was dried on Na_2SO_4. Column chromatography over silica (eluent: 8% MeOH–DCM), gave the pure product as a clear, colorless oil (980 mg, 75%). R_f (10% MeOH–DCM) = 0.50. δ (300 MHz; $CDCl_3$; Me_4Si) 2.56 (3H, br s, OH), 3.52 (3H, d, Ar–CH', ^3J = 13.7 Hz), 3.67 (12H, m, OCH_2), 3.84 (15H, m, OCH_3, OCH_2), 4.13 (6H, m, OCH_2), 4.73 (3H, d, Ar–CH, ^3J = 13.7 Hz), 6.85 (6H, 2 × s, ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 36.4 (Ar–CH$_2$), 56.2 (OCH$_3$), 61.7, 69.0, 69.4, 72.5 (OCH$_2$), 113.7, 116.2, 131.8, 132.8, 146.8, 148.4 (ArC); *m*/*z* (ESI) 673.4 ([M + H]$^+$. $C_{36}H_{49}O_{12}$ requires 673.3).

CTV-*O*-tris(triethyleneglycol) 4b. This compound was prepared from cyclotriveratrylene **3** (674 mg, 1.65 mmol) and toluene-4-sulfonic acid 2-[2-(2-hydroxyethoxy)ethoxy]ethyl ester **2b** (1.66 g, 5.45 mmol) according to the procedure for **4a**. The product was obtained as a clear, colorless oil (1.14 g, 86%). R_f (10% MeOH–DCM) = 0.45. δ (300 MHz; $CDCl_3$; Me_4Si) 2.81 (3H, br s, OH), 3.59 (27H, m, Ar–CH', OCH_2), 3.83 (15H, m, OCH_3, OCH_2), 4.13 (6H, m, OCH_2), 4.73 (3H, d, ^3J = 13.8 Hz), 6.85 (6H, 2 × s, ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 36.4 (Ar–CH$_2$), 56.2 (OCH$_3$), 61.7, 68.8, 69.7, 70.2, 70.8, 72.5 (OCH$_2$), 113.8, 116.0, 131.9, 132.7, 146.8, 148.3 (ArC); *m*/*z* (ESI) 827.5 ([M + Na]$^+$. $C_{42}H_{60}O_{15}$Na requires 827.4).

CTV-*O*-tris(tetraethyleneglycol) 4c. This compound was prepared from cyclotriveratrylene **3** (633 mg, 1.55 mmol) and toluene-4-sulfonic acid 2-{2-[2-(2-hydroxyethoxy)ethoxy]-ethoxy}ethyl ester **2c** (1.78 g, 5.11 mmol) according to the procedure for **4a**. The product was obtained as a clear, colorless oil (1.30 g, 90%). R_f (10% MeOH–DCM) = 0.39. δ (300 MHz; $CDCl_3$; Me_4Si) 2.56 (3H, br s, OH), 3.52 (3H, d, Ar–CH', ^3J = 13.7 Hz), 3.67 (12H, m, OCH_2), 3.84 (15H, m, OCH_3, OCH_2), 4.13 (6H, m, OCH_2), 4.73 (3H, d, Ar–CH, ^3J = 13.7 Hz), 6.85 (6H, 2 × s, ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 36.4 (Ar–CH$_2$), 56.2 (OCH$_3$), 61.7, 69.0, 69.4, 72.5 (OCH$_2$), 113.7, 116.2, 131.8, 132.8, 146.8, 148.4 (ArC); *m*/*z* (ESI) 491.5 ([M + 2Na]$^{2+}$), 959.4 ([M + Na]$^+$. $C_{48}H_{72}O_{18}$Na requires 959.5).

CTV-*O*-tris(diethyleneglycol-*p*-nitrophenylcarbonate) 6a. CTV-*O*-tris(diethyleneglycol) **4a** (950 mg, 1.41 mmol) and *p*-nitrophenyl chloroformate **5** (853 mg, 4.23 mmol) were dissolved in DCM (5 mL). Then, pyridine (514 µL, 6.35 mmol) was added dropwise. After stirring at room temperature for 3 hours, DCM (5 mL) and water (10 mL) were added. The layers were separated, and the organic layer was washed with 1 M $KHSO_4$, and dried on Na_2SO_4. After evaporation to dryness, the crude product was purified by column chromatography over silica (eluent: 2% MeOH–DCM). This afforded the pure product as a light-yellow foam (1.35 g, 82%). δ (300 MHz; $CDCl_3$; Me_4Si) 3.53 (3H, d, Ar–CH', ^3J = 13.8 Hz), 3.81 (9H, s, OCH_3), 3.86 (12H, m, OCH_2), 4.16 (6H, m, OCH_2), 4.43 (6H, m, OCH_2), 4.73 (3H, d, Ar–CH, ^3J = 13.8 Hz), 6.90 (6H, 2 × s, CTV-ArH), 7.30 (6H, d, PNP-ArH, ^3J = 9.2 Hz), 8.19 (6H, d, PNP-ArH, ^3J = 9.2 Hz); δ (75.5 MHz; $CDCl_3$; Me_4Si) 36.3 (Ar–CH$_2$), 56.1 (OCH$_3$), 68.2, 68.7, 69.1, 69.7 (OCH$_2$), 113.8, 116.2, 121.7, 125.2, 131.8, 132.8, 145.2, 146.8, 148.3, 152.4 (ArC), 155.3 (C=O); *m*/*z* (ESI) 1168.4 ([M + H]$^+$. $C_{57}H_{58}N_3O_{24}$ requires 1168.3), 1190.4 ([M + Na]$^+$).

CTV-*O*-tris(triethyleneglycol-*p*-nitrophenylcarbonate) 6b. This compound was prepared from CTV-*O*-tris(triethyleneglycol) **4b** (487 mg, 0.53 mmol) and *p*-nitrophenyl chloroformate **5** (352 mg, 1.75 mmol) following the procedure for **6a**. This afforded the pure product as a light-yellow foam (535 mg, 78%). R_f (2% MeOH–DCM) = 0.09. δ (300 MHz; $CDCl_3$; Me_4Si) 3.51 (3H, d, Ar–CH', ^3J = 13.8 Hz), 3.79 (33H, m, OCH_2, OCH_3), 4.14 (6H, m, OCH_2), 4.37 (6H, m, OCH_2), 4.72 (3H, d, Ar–CH, ^3J = 13.8 Hz), 6.85 (6H, 2 × s, CTV-ArH), 7.34 (6H, d, PNP-ArH, ^3J = 9.0 Hz), 8.23 (6H, d, PNP-ArH, ^3J = 9.0 Hz); δ (75.5 MHz; $CDCl_3$; Me_4Si) 36.3 (Ar–CH$_2$), 56.1 (OCH$_3$), 68.2, 68.5, 68.7, 69.7, 70.5, 70.7 (OCH$_2$), 113.8, 116.0, 121.7, 125.1, 131.8, 132.6, 145.2, 146.8, 148.2, 152.4 (ArC), 155.4 (C=O); *m*/*z* (ESI) 1300.3 ([M + H]$^+$. $C_{63}H_{70}N_30_{27}$ requires 1300.4), 1322.8 ([M + Na]$^+$).

CTV-*O*-tris(tetraethyleneglycol-*p*-nitrophenylcarbonate) 6c. This compound was prepared from CTV-*O*-tris(tetraethyleneglycol) **4c** (930 g, 0.99 mmol) and *p*-nitrophenyl chloroformate **5** (659 mg, 3.27 mmol) following the procedure for **6a**. This afforded the pure product as a light-yellow foam (764 mg, 54%). R_f (10% MeOH–DCM) = 0.78. δ (300 MHz; $CDCl_3$; Me_4Si) 3.53 (3H, d, Ar–CH', ^3J = 13.8 Hz), 3.81 (9H, s, OCH_3), 3.86 (12H, m, OCH_2), 4.16 (6H, m, OCH_2), 4.43 (6H, m, OCH_2), 4.73 (3H, d, Ar–CH, ^3J = 13.8 Hz), 6.90 (6H, 2 × s, CTV-ArH), 7.30 (6H, d, PNP-ArH, ^3J = 9.2 Hz), 8.19 (6H, d, PNP-ArH, ^3J = 9.2 Hz); δ (75.5 MHz; $CDCl_3$; Me_4Si) 36.3 (Ar–CH$_2$), 56.1 (OCH$_3$), 68.2, 68.7, 69.1, 69.7 (OCH$_2$), 113.8, 116.2, 121.7, 125.2, 131.8, 132.8, 145.2, 146.8, 148.3, 152.4 (ArC), 155.3 (C=O); *m*/*z* (ESI) 1454.6 ([M + Na]$^+$, $C_{69}H_{81}N_3O_{30}$Na requires 1454.5).

8a. Boc–Glc(Bn)–Gal(OAc)$_4$ **7a** [12] (67 mg, 0.1 mmol) was dissolved in DCM (1 mL) and TFA (1 mL) was added. The resulting mixture was stirred at room temperature for one hour. After evaporation of the solvents, THF (2 mL), CTV-*O*-tris-(tetraethyleneglycol-*p*-nitrophenylcarbonate) **6c** (28.6 mg, 0.02 mmol), and $NaHCO_3$ (20 mg, 0.24 mmol) were added. This mixture was shaken overnight at room temperature. The solvents were evaporated and the crude product was redissolved in DCM (5 mL). After washing with 5% $NaHCO_3$ (3 ×) and 1 M $KHSO_4$, the organic layer was dried on Na_2SO_4. Column chromatography (eluent: 5% MeOH–DCM) afforded the product as a white foam (18.7 mg, 34%). R_f (300 MHz; $CDCl_3$; Me_4Si) = 0.17. δ (300 MHz, $CDCl_3$) 1.18–2.12 (42H, m, C$^{\beta}H_2$, C(O)CH_3), 2.41 (6H, m, C$^{\gamma}H_2$), 3.44 (3H, d, CTV-ArCHH$'$, ^3J = 13.3 Hz), 3.58 (30H, m, OCH_2), 3.73 (15H, m, OCH_2, OCH_3), 3.94–4.14 (27H, m, C^2H, C^5H, C$^6H_2$, C$^{\alpha}H$, OCH_2), 4.65 (3H, d, CTV-ArCHH', ^3J = 13.3 Hz), 5.04–5.15 (15H, m, C^1H, C^3H, NHCbz, Bn–ArCH_2), 5.35 (3H, d, C^4H, ^3J = 1.4 Hz), 5.49 (3H, br s, NHC$^{\alpha}$), 6.78 (6H, 2 × s, CTV-ArH), 6.99 (3H, br s, NHC^1), 7.27 (15H, m, Bn–ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.5, 20.6 (C(O)CH$_3$), 26.9 (C^{β}), 30.2 (C^{γ}), 36.4 (CTV-CH$_2$), 54.2 (C^{α}), 56.2 (OCH$_3$), 61.0, 64.5 (C^6, $C^{6'}$), 66.6, 68.8, 69.3,

69.6, 70.5, 70.9 (O*C*H$_2$), 67.0, 68.1, 70.7, 72.3 (*C*^{2}, *C*^{3}, *C*^{4}, *C*^{5}), 78.4 (*C*^{1}), 113.8, 116.0, 128.3, 128.3, 128.6, 131.8, 132.6, 135.6, 146.9, 148.3 (Ar*C*), 156.2, 169.8, 170.1, 170.4, 171.1, 171.7, 172.8 (*C*=O); *m*/*z* (MALDI) 2735 ([M + Na]$^+$. $C_{129}H_{168}N_6O_{57}$ requires 2736), 2752 ([M + K]$^+$).

8b. This compound was prepared from Boc–Lys(Cbz)–Glc(OAc)$_4$ **7b**[12] (71 mg, 0.1 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (16.3 mg, 29%). R_f (5% MeOH–DCM) = 0.25. δ (300 MHz; $CDCl_3$; Me_4Si) 1.31–1.84 (18H, m, C$^{\beta}H_2$, C$^{\gamma}H_2$, C$^{\delta}H_2$), 2.03 (36H, 4 × s, C(O)CH_3), 3.14 (6H, m, C$^{\varepsilon}H_2$), 3.45–3.68 (39H, C^5H, C$^{\alpha}H$, CTV-ArC*H*H′, OCH_2), 3.80 (15H, OCH_2, OCH_3), 4.04–4.30 (21H, C$^6H_2$, OCH_2), 4.73 (3H, d, CTV-ArCH*H*′, ^3J = 13.7 Hz), 4.96 (3H, t, C^4H, ^3J = 9.5 Hz), 5.02–5.13 (12H, m, C^2H, N*H*Cbz, Cbz–ArCH_2), 5.21–5.31 (6H, m, C^1H, C^3H), 5.54 (3H, br s, N*H*C$^{\alpha}$), 6.84 (6H, 2 × s, CTV-Ar*H*), 7.10 (3H, br s, N*H*C^1), 7.34 (15H, m, Cbz–Ar*H*); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.6, 20.7 (C(O)*C*H$_3$), 22.2 (*C*$^{\beta}$), 29.3 (*C*$^{\gamma}$), 30.3 (*C*$^{\delta}$), 36.4 (CTV-Ar*C*H$_2$), 40.3 (*C*$^{\varepsilon}$), 54.8 (*C*$^{\alpha}$), 56.2 (*O**C*H$_3$), 61.6, 64.5 (*C*^{6}, *C*$^{6'}$), 66.6, 68.8, 69.3, 69.6, 70.4, 70.5, 70.7 (O*C*H$_2$), 68.1, 72.8, 73.6 (*C*^{2}, *C*^{3}, *C*^{4}, *C*^{5}), 78.1 (*C*^{1}), 113.9, 116.0, 125.5, 128.1, 128.1, 128.5, 131.9, 132.7, 136.6, 146.9, 148.3 (Ar*C*), 156.6, 156.7, 169.5, 169.9, 170.6, 172.4 (*C*=O); *m*/*z* (MALDI) 2863 ([M + Na]$^+$. $C_{135}H_{183}N_9O_{57}Na$ requires 2865), 2880 ([M + K]$^+$).

8c. This compound was prepared from Boc–Lys(Cbz)–Gal(OAc)$_4$ **7c**[12] (71 mg, 0.1 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (26.8 mg, 47%). R_f (5% MeOH–DCM) = 0.20. δ (300 MHz; $CDCl_3$; Me_4Si) 1.30–1.85 (18H, m, C$^{\beta}H_2$, C$^{\gamma}H_2$, C$^{\delta}H_2$), 1.99–2.14 (36H, 4 × s, C(O)CH_3), 3.16 (6H, m, C$^{\varepsilon}H_2$), 3.52 (3H, d, CTV-ArC*H*H′, ^3J = 13.8 Hz), 3.61–3.76 (30H, m, OCH_2), 3.80 (15H, m, OCH_2, OCH_3), 4.01–4.21 (27H, m, C^2H, C^5H, C$^6H_2$, C$^{\alpha}H$, OCH_2), 4.73 (3H, d, CTV-ArCH*H*′, ^3J = 13.8 Hz), 5.00–5.23 (15H, m, C^1H, C^3H, N*H*Cbz, Cbz–ArCH_2), 5.42 (3H, s, C^4H), 5.49 (3H, br s, N*H*C$^{\alpha}$), 6.85 (6H, 2 × s, CTV-Ar*H*), 6.99 (3H, br s, N*H*C^1), 7.34 (15H, m, Cbz–Ar*H*); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.5, 20.6, 20.6 (C(O)*C*H$_3$), 22.3 (*C*$^{\beta}$), 29.3 (*C*$^{\gamma}$), 30.9 (*C*$^{\delta}$), 36.4 (CTV-*C*H$_2$), 40.3 (*C*$^{\varepsilon}$), 54.8 (*C*$^{\alpha}$), 56.2 (O*C*H$_3$), 61.0 (Cbz–*C*H$_2$), 64.5, 66.6 (*C*^{6}, *C*$^{6'}$), 67.0, 68.2, 70.9, 72.3 (*C*^{2}, *C*^{3}, *C*^{4}, *C*^{5}), 68.8, 69.3, 69.6, 70.4, 70.5, 70.7 (O*C*H$_2$), 78.4 (*C*^{1}), 113.8, 116.0, 128.1, 128.1, 128.5, 131.8, 132.6, 146.8, 148.3 (Ar*C*), 156.3, 156.6, 169.8, 170.1, 170.4, 171.1, 172.2 (*C*=O); *m*/*z* (MALDI) 2797 ([M − Ac + H]$^+$), 2863 ([M + Na]$^+$. $C_{135}H_{183}N_9O_{57}Na$ requires 2865), 2880 ([M + K]$^+$).

8d. This compound was prepared from Boc–Ala–Lac(OAc)$_7$ **7d**[12] (81 mg, 0.1 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (13.6 mg, 22%). R_f (5% MeOH–DCM) = 0.18. δ (300 MHz; $CDCl_3$; Me_4Si) 1.33 (9H, d, C$^{\beta}H_3$, ^3J = 6.9 Hz), 1.97–2.18 (63H, 7 × s, C(O)CH_3), 3.52 (3H, d, ArC*H*H′, ^3J = 13.7 Hz), 3.59–3.90 (54H, m, C$^{5'}H$, C$^{6'}H_2$, OCH_2, OCH_3), 4.04–4.23 (24H, C^5H, C$^6H_2$, C$^{5'}H$, C$^{6'}H_2$, C$^{\alpha}$H, OCH_2), 4.40–4.52 (6H, m, C^4H, C$^{1'}H$), 4.73 (3H, d, ArCH*H*′, ^3J = 13.7 Hz), 4.86 (3H, t, C^2H, ^3J =9.5 Hz), 4.94 (3H, dd, C$^{3'}H$, $^3J_{2',3'}$ = 10.3 Hz, $^3J_{3',4'}$ = 3.3 Hz), 5.08–5.31 (9H, C^1H, C^3H, C$^{2'}H$), 5.35 (6H, N*H*C^1, C$^{4'}H$), 6.85 (6H, 2 × s, CTV-Ar*H*), 6.98 (3H, br d, N*H*C^1); δ (75.5 MHz; $CDCl_3$; Me_4Si) 17.2 (*C*$^{\beta}$), 20.5, 20.6, 20.6, 20.7, 20.9 (C(O)*C*H$_3$), 36.4 (CTV-*C*H$_2$), 50.4 (*C*$^{\alpha}$), 56.2 (O*C*H$_3$), 60.8, 61.9 (*C*^{6}, *C*$^{6'}$), 64.5, 68.8, 69.3, 69.6, 70.5, 70.7, 70.9 (O*C*H$_2$), 66.6, 68.9, 71.0, 72.4, 74.5, 75.9 (*C*^{2}, *C*^{3}, *C*^{4}, *C*^{5}, *C*$^{2'}$, *C*$^{3'}$, *C*$^{4'}$, *C*$^{5'}$), 77.9 (*C*^{1}), 100.9 (*C*$^{1'}$), 113.9, 116.0, 131.8, 132.6, 146.9, 148.3 (Ar*C*), 156.1, 169.0, 169.4, 170.1, 170.2, 170.3, 170.9, 172.7 (*C*=O); *m*/*z* (MALDI) 3154 ([M + Na]$^+$. $C_{138}H_{192}N_6O_{75}Na$ requires 3156).

8e. This compound was prepared from Boc–Glu–Lac(OAc)$_7$ **7e**[12] (96 mg, 0.1 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (10.5 mg, 15%). R_f (10% MeOH–DCM) = 0.41. δ (300 MHz; $CDCl_3$; Me_4Si) 1.97–2.16 (69H, m, C$^{\beta}H_2$, C(O)CH_3), 2.44 (6H, m, C$^{\gamma}H_2$), 3.47 (3H, d, CTV-ArC*H*H′, ^3J = 13.7 Hz), 3.54–4.87 (54H, m, C$^{5'}H$, C$^{6'}H_2$, OCH_3, OCH_2), 4.04–4.17 (24H, m, C^5H, C$^6H_2$, C$^{5'}H$, C$^{6'}H_2$, C$^{\alpha}H$, OCH_2), 4.44 (6H, m, C^4H, C$^{1'}H$), 4.73 (3H, d, CTV-ArCH*H*′, ^3J = 13.7 Hz), 4.86 (3H, t, C^2H, ^3J = 9.6 Hz), 4.93 (3H, dd, C$^{3'}H$, $^3J_{3',2'}$ = 10.5 Hz, $^3J_{3',4'}$ = 2.7 Hz), 5.07–5.30 (18H, m, C^1H, C^3H, C$^{2'}H$, C$^{4'}H$, Bn–ArCH_2), 5.35 (3H, d, C$^{4'}H$, ^3J = 2.7 Hz), 5.57 (3H, br s, N*H*C$^{\alpha}$), 6.85 (6H, 2 × s, CTV-Ar*H*), 7.06 (3H, br s, N*H*C^1), 7.34 (15H, m, Bn–Ar*H*); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.5, 20.7, 20.8, 20.8 (C(O)*C*H$_3$), 27.0 (*C*$^{\beta}$), 30.2 (*C*$^{\gamma}$), 36.4 (CTV-Ar*C*H$_2$), 54.1 (*C*$^{\alpha}$), 56.2 (O*C*H$_3$), 60.8 (O*C*H$_2$), 61.8, 64.6 (*C*^{6}, *C*$^{6'}$), 66.6, 69.0, 70.7, 71.0, 72.5, 74.5, 75.9 (*C*^{2}, *C*^{3}, *C*^{4}, *C*^{5}, *C*$^{2'}$, *C*$^{3'}$, *C*$^{4'}$, *C*$^{5'}$), 66.6, 68.8, 69.1, 69.3, 70.5 (O*C*H$_2$), 77.9 (*C*^{1}), 100.9 (*C*$^{1'}$), 113.9, 115.9, 125.5, 128.3, 128.6, 131.8, 132.6, 133.4, 135.6, 146.9, 148.3 (Ar*C*), 158.0, 169.0, 170.1, 170.2, 170.4, 171.7, 172.9, 176.5 (*C*=O); *m*/*z* (MALDI) 3599 ([M + Na]$^+$. $C_{165}H_{216}N_6O_{81}Na$ requires 3600), 3617 ([M + K]$^+$).

8f. This compound was prepared from Boc–Lys(Cbz)-Lac(OAc)$_7$ **7f**[12] (102 mg, 0.1 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (20.1 mg, 27%). R_f (10% MeOH–DCM) = 0.45. δ (300 MHz; $CDCl_3$; Me_4Si) 1.32–1.85 (18H, m, C$^{\beta}H_2$, C$^{\gamma}H_2$, C$^{\delta}H_2$), 1.97–2.16 (63H, 7 × s, C(O)CH_3), 3.14 (6H, m, C$^{\varepsilon}H_2$), 3.49–3.89 (57H, C$^{5'}H$, C$^{6'}H_2$, OCH_3, CTV-ArC*H*H′, OCH_2), 4.04–4.18 (24H, m, C^5H, C$^6H_2$, C$^{5'}H$, C$^{6'}H_2$, C$^{\alpha}H$, OCH_2), 4.44 (6H, C^4H, C$^{1'}H$), 4.72 (3H, d, CTV-ArCH*H*′, ^3J = 13.7 Hz), 4.86 (3H, t, C^2H, ^3J = 9.5 Hz), 4.94 (3H, dd, C$^{3'}H$, $^3J_{2',3'}$ = 10.3 Hz, $^3J_{3',4'}$ = 3.4 Hz), 5.07–5.30 (18H, m, C^1H, C^3H, C$^{2'}H$, N*H*Cbz, Cbz–CH_2), 5.35 (3H, d, C$^{4'}H$, ^3J = 3.4 Hz), 5.48 (3H, br s, N*H*C$^{\alpha}$), 6.85 (6H, 2 × s, CTV-Ar*H*), 6.91 (3H, br s, N*H*C^1), 7.34 (15H, m, Cbz–Ar*H*); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.5, 20.6, 20.6, 20.7, 20.9 (C(O)*C*H$_3$), 22.1 (*C*$^{\beta}$), 29.3 (*C*$^{\gamma}$), 30.8 (*C*$^{\delta}$), 36.4 (CTV-Ar*C*H$_2$), 40.2 (*C*$^{\varepsilon}$), 54.7 (*C*$^{\alpha}$), 56.2 (O*C*H$_3$), 60.8, 61.7 (*C*^{6}, *C*$^{6'}$), 64.5 (Cbz–Ar*C*H$_2$), 68.8, 69.3, 69.6, 70.4, 70.7 (O*C*H$_2$), 69.0, 70.6, 70.8, 71.0, 72.5, 74.5, 75.9 (*C*^{2}, *C*^{3}, *C*^{4}, *C*^{5}, *C*$^{2'}$, *C*$^{3'}$, *C*$^{4'}$, *C*$^{5'}$), 77.9 (*C*^{1}), 100.9 (*C*$^{1'}$), 113.8, 116.0, 128.1, 128.5, 131.9, 132.7, 136.7, 146.8, 148.3 (Ar*C*), 156.3, 156.6, 168.9, 169.4, 170.1, 170.1, 170.3, 170.4, 171.0, 172.2 (*C*=O); *m*/*z* (MALDI) 3728 ([M + Na]$^+$. $C_{171}H_{231}N_9O_{81}Na$ requires 3729), 3744 ([M + K]$^+$).

8g. This compound was prepared from Boc–Glu(Bn)–Gal(OAc)$_4$ **7a**[12] (67 mg, 0.1 mmol) and CTV-*O*-tris(triethyleneglycol-*p*-nitrophenylcarbonate) **6b** (26.0 mg, 0.02 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (25 mg, 48%). R_f (10% MeOH–DCM) = 0.53. δ (300 MHz; $CDCl_3$; Me_4Si) 1.83–2.15 (42H, C$^{\beta}H_2$, C(O)CH_3), 2.41 (6H, m, C$^{\gamma}H_2$), 3.44 (3H, d, CTV-ArC*H*H′, ^3J = 13.6 Hz), 3.55–3.66 (24H, m, OCH_2), 3.72 (15H, m, OCH_2, OCH_3), 3.94–4.14 (21H, m, C^2H, C^5H, C$^6H_2$, C$^{\alpha}H$, OCH_2), 4.65 (3H, d, CTV-ArCH*H*′, ^3J = 13.6 Hz), 5.03–5.17 (12H, C^1H, C^3H, Bn–ArCH_2), 5.36 (3H, d, C^4H, ^3J = 1.4 Hz), 5.47 (3H, br d, N*H*C$^{\alpha}$), 6.78 (6H, 2 × s, CTV-Ar*H*), 6.99 (3H, br d, N*H*C^1), 7.27 (15H, m, Bn–Ar*H*); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.5, 20.6 (C(O)*C*H$_3$), 26.9 (*C*$^{\beta}$, 30.2 (*C*$^{\gamma}$), 36.4 (CTV-Ar*C*H$_2$), 54.1 (*C*$^{\alpha}$), 56.2 (O*C*H$_3$), 61.0 (*C*^{6}), 64.5, 66.6, 68.8, 69.3, 69.7, 70.5, 70.6 (O*C*H$_2$), 67.0, 68.1, 70.9, 72.3 (*C*^{2}, *C*^{3}, *C*^{4}, *C*^{5}), 78.4 (*C*^{1}), 113.9, 115.9, 128.3, 128.3, 128.6, 131.8, 132.6, 135.6, 146.8, 148.3 (Ar*C*), 156.2, 169.8, 170.1, 170.4, 171.1, 171.7, 172.8 (*C*=O); *m*/*z* (MALDI) 2603 ([M + Na]$^+$. $C_{123}H_{156}N_6O_{54}Na$ requires 2604), 2619 ([M + K]$^+$).

8h. This compound was prepared from Boc–Lys(Cbz)–Glc(OAc)$_4$ **7b**[12] (71 mg, 0.1 mmol) and CTV-*O*-tris(triethyleneglycol-*p*-nitrophenylcarbonate) **6b** (26.0 mg, 0.02 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (13 mg, 24%). R_f (10% MeOH–DCM) = 0.49. δ (300 MHz; $CDCl_3$; Me_4Si) 1.20–1.80 (18H, m, C$^{\beta}H_2$, C$^{\gamma}H_2$, C$^{\delta}H_2$), 2.03 (36H, 4 × s, C(O)CH_3), 3.12 (6H, m, C$^{\varepsilon}H_2$), 3.52 (3H, CTV-ArCHH′, ^3J = 13.8 Hz), 3.62–3.79 (39H, m, C^5H, C$^{\alpha}H$, OCH_3, OCH_2), 4.03–4.28 (18H, m, C$^6H_2$, OCH_2), 4.73 (3H, d, CTV-ArCHH′, ^3J = 13.8 Hz), 4.96 (3H, t, C^4H, ^3J = 9.5 Hz), 5.09 (9H, m, NHCbz, Cbz–ArCH_2), 5.26 (6H, m, C^1H, C^3H), 5.54 (3H, br s, NHC$^{\alpha}$), 6.85 (6H, 2 × s, CTV-ArH), 7.09 (3H, br s, NHC^1), 7.34 (15H, m, Cbz–ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.6, 20.7 (C(O)CH$_3$), 22.3 (C$^{\beta}$), 29.3 (C$^{\gamma}$), 30.3 (C$^{\delta}$), 36.4 (CTV-ArCH$_2$), 40.3 (C$^{\varepsilon}$), 54.8 (C$^{\alpha}$), 56.2 (OCH$_3$), 61.6 (C^6), 64.5, 66.6, 68.4, 68.8, 69.3, 70.6 (OCH$_2$), 68.1, 72.8, 73.6 (C^2, C^3, C^4, C^5), 78.1 (C^1), 113.9, 116.0, 128.1, 128.1, 128.5, 131.9, 132,7, 136.6, 146.9, 148.3 (ArC), 156.5, 156.6, 169.5, 169.9, 170.6, 172.5 (C=O); *m/z* (MALDI) 2732 ([M + Na]$^+$. $C_{129}H_{171}N_9O_{54}$Na requires 2733), 2748 ([M + K]$^+$).

8i. This compound was prepared from Boc–Lys(Cbz)–Gal(OAc)$_4$ **7c**[12] (71 mg, 0.1 mmol) and CTV-*O*-tris(triethyleneglycol-*p*-nitrophenylcarbonate) **6b** (26.0 mg, 0.02 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (25 mg, 46%). R_f (10% MeOH–DCM) = 0.48. δ (300 MHz; $CDCl_3$; Me_4Si) 1.22–1.81 (18H, m, C$^{\beta}H_2$, C$^{\gamma}H_2$, C$^{\delta}H_2$), 1.99–2.14 (36H, 4 × s, C(O)CH_3), 3.13 (6H, m, C$^{\varepsilon}H_2$), 3.52 (3H, d, CTV-ArCHH′, ^3J = 13.9 Hz), 3.63–3.76 (24H, m, OCH_2), 3.79 (15H, m, OCH_2, OCH_3), 4.01–4.22 (21H, C^2H, C^5H, C$^6H_2$, C$^{\alpha}H$, OCH_2), 4.73 (3H, d, CTV-ArCHH′, ^3J = 13.9 Hz), 5.01–5.22 (15H, m, C^1H, C^3H, NHCbz, Cbz–ArCH_2), 5.42 (3H, s, C^4H), 5.45 (3H, br s, NHC$^{\alpha}$), 6.85 (6H, 2 × s, CTV-ArH), 6.99 (3H, br s, NHC^1), 7.34 (15H, m, Bn–ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.6, 20.7 (C(O)CH_3), 22.3 (C$^{\beta}$), 25.6 (C$^{\gamma}$), 29.3 (C$^{\delta}$), 36.4 (CTV-ArCH$_2$), 44.1 (C$^{\varepsilon}$), 54.8 (C$^{\alpha}$), 56.2 (OCH$_3$), 61.0, 64.5 (C^6, C$^{6'}$), 66.6, 68.0, 68.8, 69.3, 69.7, 70.6 (OCH$_2$), 67.1, 68.2, 70.9, 72.4 (C^2, C^3, C^4, C^5), 78.4 (C^1), 113.9, 116.0, 128.1, 128.5, 131.9, 132.6, 136.6, 146.9, 148.3 (ArC), 156.6, 169.8, 170.1, 170.4, 171.2, 172.3 (C=O); *m/z* (MALDI) 2732 ([M + Na]$^+$. $C_{129}H_{171}N_9O_{54}$Na requires 2733), 2748 ([M + K]$^+$).

8j. This compound was prepared from Boc–Ala–Lac(OAc)$_7$ **7d**[12] (67 mg, 0.1 mmol) and CTV-*O*-tris(triethyleneglycol-*p*-nitrophenylcarbonate) **6b** (26.0 mg, 0.02 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (24 mg, 39%). R_f (10% MeOH–DCM) = 0.37. δ (300 MHz; $CDCl_3$; Me_4Si) 1.32 (9H, d, C$^{\beta}H_3$, ^3J = 6.9 Hz), 1.97–2.16 (63H, 7 × s, C(O)CH_3), 3.53 (3H, d, ArCHH′, ^3J = 13.7 Hz), 3.64–3.90 (42H, m, C$^{5'}H$, C$^{6'}H_2$, OCH_3, OCH_2), 4.04–4.31 (24H, m, C^5H, C$^6H_2$, C$^{5'}H$, C$^{6'}H_2$, C$^{\alpha}H$, OCH_2), 4.44 (6H, m, C^4H, C$^{1'}H$), 4.73 (3H, ArCHH′, ^3J = 13.5 Hz), 4.86 (3H, t, C^2H, ^3J = 9.5 Hz), 4.95 (3H, dd, C$^{3'}H$, $^3J_{2',3'}$ = 10.3 Hz, $^3J_{3',4'}$ = 3.4 Hz), 5.05–5.38 (15H, m, C^1H, C^3H, C$^{2'}H$, C$^{4'}H$, NHC$^{\alpha}$), 6.85 (6H, 2 × s, ArH), 6.99 (3H, d, NHC^1, ^3J = 9.0 Hz); δ (75.5 MHz; $CDCl_3$; Me_4Si) 17.1 (C$^{\beta}$), 20.4, 20.5, 20.6, 20.6, 20.9 (C(O)CH_3), 36.4 (CTV-CH$_2$), 50.4 (C$^{\alpha}$), 56.1 (OCH$_3$), 61.0, 61.8 (C^6, C$^{6'}$), 64.5, 68.3, 69.0, 69.6, 70.5, 70.7 (OCH$_2$), 67.6, 68.9, 69.5, 70.0, 71.0, 72.4, 74.0, 74.8 (C^2, C^3, C^4, C^5, C$^{2'}$, C$^{3'}$, C$^{4'}$, C$^{5'}$), 77.8 (C^1), 95.7 (C$^{1'}$), 113.9, 116.0, 131.8, 132.5, 146.9, 148.2 (ArC), 155.9, 169.3, 169.7, 170.1, 170.2, 170.4, 172.7 (C=O); *m/z* (ESI) 1526 ([M + 2H]$^{2+}$. $C_{132}H_{182}H_6O_{75}^{2+}$ requires 1526), 1536 ([M + H + Na]$^{2+}$), 1548 ([M + 2Na]$^{2+}$).

8k. This compound was prepared from Boc–Glu(Bn)–Lac(OAc)$_7$ **7e**[12] (67 mg, 0.1 mmol) and CTV-*O*-tris(triethyleneglycol-*p*-nitrophenylcarbonate) **6b** (26.0 mg, 0.02 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (30 mg, 43%). R_f (10% MeOH–DCM) = 0.42. δ (300 MHz; $CDCl_3$; Me_4Si) 1.97–2.16 (69H, m, C$^{\beta}H_2$, C(O)CH_3), 2.46 (6H, m, C$^{\gamma}H_2$), 3.51 (3H, d, CTV-ArCHH′, ^3J = 13.5 Hz), 3.63–3.90 (42H, m, C$^{5'}H$, C$^{6'}H_2$, OCH_3, OCH_2), 4.04–4.29 (24H, m, C^5H, C$^6H_2$, C$^{5'}H$, C$^{6'}H_2$, C$^{\alpha}H$, OCH_2), 4.39–4.51 (6H, m, C^4H, C$^{1'}H$), 4.73 (3H, d, CTV-ArCHH′, ^3J = 13.5 Hz), 4.84–4.98 (6H, m, C^2H, C$^{3'}H$), 5.04–5.30 (15H, m, C^1H, C^3H, C$^{2'}H$, Bn–ArCH_2), 5.35 (3H, s, C$^{4'}H$), 5.67 (3H, br s, NHC$^{\alpha}$), 6.85 (6H, 2 × s, CTV-ArH), 7.33 (18H, m, NHC^1, Bn–ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.5, 20.6, 20.6, 20.7, 20.8 (C(O)CH$_3$), 26.9 (C$^{\beta}$), 30.1 (C$^{\gamma}$), 36.4 (CTV-ArCH$_2$), 54.0 (C$^{\alpha}$), 56.2 (OCH$_3$), 60.8, 61.9 (C^6, C$^{6'}$), 64.5, 66.6, 68.7, 69.3, 69.6, 70.4 (OCH$_2$), 66.6, 68.9, 70.7, 71.0, 72.5, 74.4, 75.9 (C^2, C^3, C^4, C^5, C$^{2'}$, C$^{3'}$, C$^{4'}$, C$^{5'}$), 77.9 (C^1), 100.9 (C$^{1'}$), 113.9, 115.9, 128.2, 128.3, 128.5, 131.9, 132.6, 135.6, 146.8, 148.3 (ArC), 156.3, 169.0, 169.4, 170.1, 170.2, 170.4, 170.9, 171.7, 172.9 (C=O); *m/z* (MALDI) 3467 ([M + Na]$^+$. $C_{159}H_{204}N_6O_{78}$Na requires 3468), 3483 ([M + K]$^+$).

8l. This compound was prepared from Boc–Lys(Cbz)–Lac(OAc)$_7$ **7f**[12] (67 mg, 0.1 mmol) and CTV-*O*-tris(triethyleneglycol-*p*-nitrophenylcarbonate) **6b** (26.0 mg, 0.02 mmol) according to the procedure for **8a**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (35 mg, 48%). R_f (10% MeOH–DCM) = 0.49. δ (300 MHz; $CDCl_3$; Me_4Si) 1.25–1.82 (18H, m, C$^{\beta}H_2$, C$^{\gamma}H_2$, C$^{\delta}H_2$), 1.97–2.16 (63H, 7 × s, C(O)CH_3), 3.14 (6H, m, C$^{\varepsilon}H_2$), 3.53 (3H, d, CTV-ArCHH′, ^3J = 13.5 Hz), 3.61–3.96 (42H, C$^{5'}H$, C$^{6'}H_2$, OCH_3, OCH_2), 4.04–4.29 (24H, m, C^5H, C$^6H_2$, C$^{5'}H$, C$^{6'}H_2$, C$^{\alpha}H$, OCH_2), 4.46 (6H, m, C^4H, C$^{1'}H$), 4.73 (3H, CTV-ArCHH′, ^3J = 13.5 Hz), 4.81–4.99 (6H, m, C^2H, C$^{3'}H$), 5.04–5.30 (18H, m, C^1H, C^3H, C$^{2'}H$, NHCbz, Cbz–CH_2), 5.35 (3H, d, C$^{4'}H$, ^3J = 2.5 Hz), 5.50 (3H, br s, NHC$^{\alpha}$), 6.85 (6H, 2 × s, CTV-ArH), 6.98 (3H, br s, NHC^1), 7.34 (15H, m, Cbz–ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.5, 20.6, 20.7, 20.8 (C(O)CH$_3$), 22.1 (C$^{\beta}$), 29.2 (C$^{\gamma}$), 30.8 (C$^{\delta}$), 36.4 (CTV-ArCH$_2$), 40.2 (C$^{\varepsilon}$), 54.6 (C$^{\alpha}$), 56.2 (OCH$_3$), 60.8, 61.7 (C^6, C$^{6'}$), 64.4, 68.8, 69.3, 69.6, 70.4 (OCH$_2$), 66.6, 68.2, 68.9, 70.7, 70.9, 72.5, 74.5, 75.9 (C^2, C^3, C^4, C^5, C$^{2'}$, C$^{3'}$, C$^{4'}$, C$^{5'}$), 77.9 (C^1), 100.9 (C$^{1'}$), 113.9, 116.0, 125.5, 128.1, 128.5, 131.9, 132.7, 136.6, 146.8, 148.3 (ArC), 156.3, 156.6, 169.0, 169.4, 170.1, 170.2, 170.4, 171.0, 172.3 (C=O); *m/z* (MALDI) 3595 ([M + Na]$^+$. $C_{165}H_{219}N_9O_{78}$Na requires 3597).

8m. This compound was prepared from Boc–Ala–Malt-(OAc)$_7$ **7g**[12] (162 mg, 0.2 mmol) and CTV-*O*-tris(tetraethyleneglycol-*p*-nitrophenylcarbonate) **6c** (71 mg, 0.05 mmol) following the procedure for **8a**. Column chromatography (eluent: 5% MeOH–DCM) afforded the product as a white foam (55.3 mg, 35%). R_f (10% MeOH–DCM) = 0.55. δ (300 MHz; $CDCl_3$; Me_4Si) 1.33 (9H, d, C$^{\beta}H_3$, ^3J = 7.1 Hz), 1.99–2.14 (63H, 7 × s, C(O)CH_3), 3.53 (3H, d, ArCHH′, ^3J = 14.0 Hz), 3.58–3.69 (30H, m, OCH_2), 3.81 (15H, m, OCH_2, OCH_3), 3.91–4.27 (33H, OCH_2, C^4H, C^5H, C$^{5'}H$, C^6HH′, C$^{6'}H_2$, C$^{\alpha}H$), 3.42 (3H, d, C^{6}HH', ^3J = 10.4 Hz), 4.72–4.89 (9H, m, ArCHH′, C^2H, C$^{2'}H$), 5.06 (3H, t, C$^{4'}H$, ^3J = 9.8 Hz), 5.24–5.40 (15H, m, NHCO$_2$, C^1H, C$^{1'}H$, C^3H, C$^{3'}H$), 6.86 (6H, 2 × s, ArH), 6.97 (3H, br d, NHC^1); δ (75.5 MHz; $CDCl_3$; Me_4Si) 17.1 (C$^{\beta}$), 20.4, 20.5, 20.6, 20.8 (C(O)CH$_3$), 36.4 (CTV-ArCH$_2$), 50.4 (C$^{\alpha}$), 56.2 (OCH$_3$), 61.3, 62.6 (C^6, C$^{6'}$), 64.4, 68.7, 69.5, 70.4, 70.7 (OCH$_2$), 67.8, 68.4, 69.2, 69.9, 71.2, 72.5, 73.9, 75.1 (C^2, C^3, C^4, C^5, C$^{2'}$, C$^{3'}$, C$^{4'}$, C$^{5'}$), 77.6 (C^1), 95.5 (C$^{1'}$), 113.8, 116.0, 131.8, 132.6, 146.8, 148.2 (ArC), 156.1, 169.4, 169.7, 169.8, 170.4, 170.5, 170.6, 172.7 (C=O); *m/z* (MALDI) 3158 ([M + Na]$^+$. $C_{138}H_{192}N_6O_{75}$Na requires 3156), 1374 ([M + K]$^+$).

8n. This compound was prepared from Boc–Glu–Malt-(OAc)$_7$ **7h**[12] (192 mg, 0.2 mmol) and CTV-*O*-tris(tetra-

ethyleneglycol-*p*-nitrophenylcarbonate) **6c** (71 mg, 0.05 mmol) according to the procedure for **8m**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (32.5 mg, 18%). R_f (10% MeOH–DCM) = 0.51. δ (300 MHz; $CDCl_3$; Me_4Si) 1.79 (6H, m, $C^{\beta}H_2$), 1.95–2.13 (63H, 7 × s, C(O)CH_3), 2.43 (6H, m, $C^{\gamma}H_2$), 3.45–3.83 (60H, m, OCH_2, OCH_3, CTV-ArCHH′), 3.92–4.27 (27H, m, OCH_2, C^4H, C^5H, C^6HH′, $C^{5'}H$, $C^{6'}H_2$, $C^{\alpha}H$), 4.43 (3H, m, C^6HH'), 4.70–4.89 (9H, m, C^2H, $C^{2'}H$, CTV-ArCHH'), 5.07 (9H, m, Bn–ArCH_2, $C^{4'}H$), 5.20–5.41 (12H, m, C^1H, C^3H, $C^{1'}H$, $C^{3'}H$), 5.69 (1H, br s, NHCO_2), 6.82 (6H, 2 × s, CTV-ArH), 7.23 (1H, br s, NHC^1), 7.34 (15H, m, Bn–ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.6, 20.8 (C(O)CH$_3$), 26.7 (C^{β}), 30.3 (C^{γ}), 36.3 (CTV-ArCH$_2$), 54.2 (C^{α}), 56.2 (OCH$_3$), 61.3, 62.7 (C^6, $C^{6'}$), 66.5, 68.0, 68.1, 69.6, 71.2 (OCH$_2$), 67.9, 68.5, 69.2, 69.9, 70.5, 72.5, 73.9 (C^2, C^3, C^4, C^5, $C^{2'}$, $C^{3'}$, $C^{4'}$, $C^{5'}$), 75.1 (C^1), 95.5 ($C^{1'}$), 113.7, 116.0, 125.3, 128.3, 128.6, 131.9, 132.6, 136.5, 137.1, 148.4 (ArC), 157.0, 169.7, 169.9, 170.6, 170.7, 172.3 (C=O); *m*/*z* (MALDI) 3602 ([M + Na]$^+$. $C_{165}H_{216}N_6O_{81}Na$ requires 3600), 1619 ([M + K]$^+$).

8o. This compound was prepared from Boc–Lys(Cbz)–Malt(OAc)$_7$ **7i**[12] (200 mg, 0.2 mmol) and CTV-*O*-tris(tetraethyleneglycol-*p*-nitrophenylcarbonate) **6c** (71 mg, 0.05 mmol) according to the procedure for **8m**. After column chromatography (eluent: 5% MeOH–DCM), the product was obtained as a white foam (68.8 mg, 37%). R_f (10% MeOH–DCM) = 0.61. δ (300 MHz; $CDCl_3$; Me_4Si) 1.18–1.75 (18H, m, $C^{\beta}H_2$, $C^{\gamma}H_2$, $C^{\delta}H_2$), 1.92–2.03 (63H, 7 × s, C(O)CH_3), 3.08 (6H, m, $C^{\varepsilon}H_2$), 3.42–3.60 (33H, m, OCH_2, CTV-ArCHH′), 3.73 (15H, m, OCH_2, OCH_3), 3.84–4.20 (33H, m, OCH_2, C^4H, C^5H, C^6HH′, $C^{5'}H$, $C^{6'}H_2$, $C^{\alpha}H$), 4.38 (3H, d, C^6HH', ^3J = 11.0 Hz), 4.63–4.81 (9H, m, CTV-ArCHH', C^2H, $C^{2'}H$), 4.96–5.32 (18H, m, C^1H, $C^{1'}H$, C^3H, $C^{3'}H$, Cbz–ArCH_2), 5.51 (3H, br s, NHCO_2), 6.78 (6H, 2 × s, CTV-ArH), 6.94 (3H, br s, NHC^1), 7.27 (15H, m, Cbz–ArH); δ (75.5 MHz; $CDCl_3$; Me_4Si) 20.4, 20.5, 20.6, 20.7, 20.8 (C(O)CH$_3$), 22.0 (C^{β}), 29.1 (C^{γ}), 30.8 (C^{δ}), 36.3 (CTV-ArCH$_2$), 40.1 (C^{ε}), 54.6 (C^{α}), 56.2 (OCH$_3$), 61.3, 62.4 (C^6, $C^{6'}$), 64.4, 66.5, 68.7, 68.7, 69.5, 70.4, 70.4, 70.6 (OCH$_2$), 67.8, 68.4, 69.2, 69.9, 71.1, 72.3, 73.9, 75.1 (C^2, C^3, C^4, C^5, $C^{2'}$, $C^{3'}$, $C^{4'}$, $C^{5'}$), 77.5 (C^1), 95.4 ($C^{1'}$), 113.8, 115.9, 128.0, 128.1, 128.4, 131.8, 132.6, 136.6, 146.8, 146.8, 148.3 (ArC), 156.3, 156.6, 169.4, 169.7, 169.8, 170.4, 170.5, 170.6, 170.7, 172.2 (C=O); *m*/*z* (MALDI) 3730 ([M + Na]$^+$. $C_{171}N_{231}N_9O_{81}Na$ requires 3729), 3747 ([M + K]$^+$).

9a. Protected CTV-construct **8a** (9 mg, 3.3 μmol) was dissolved in ethanol, and 10% palladium on carbon (10 mg) was added. The resulting mixture was shaken during 72 hours at room temperature under a hydrogen atmosphere. The catalyst was removed by filtration, and the solvent was evaporated. Then, methanol (1 mL) and a 55% solution of sodium methoxide in methanol (10 μl) were added. After shaking this mixture overnight, the solution was neutralized with Dowex-H$^+$ ion exchange resin (25 mg). The resin was removed by filtration, and the volatiles were evaporated. After dissolving the product in water (5 mL) and lyophilization, the product was obtained as a white powder (2.2 mg, 34%). *m*/*z* (ESI) 971 ([M + 2H]$^{2+}$), 981 ([M + H + Na]$^{2+}$), 992 ([M + 2Na]$^{2+}$), 1940 ([M + H]$^+$. $C_{84}H_{127}O_6O_{45}$ requires 1939).

9b. This product was obtained from **8b** (8 mg, 2.8 μmol) according to the procedure for **9a**. This yielded the product as a white powder (0.8 mg, 15%). *m*/*z* (ESI) 969 ([M + 2H]$^{2+}$), 980 ([M + H+Na]$^{2+}$), 991 ([M + 2Na]$^{2+}$), 1937 ([M + H]$^+$. $C_{87}H_{142}N_9O_{39}$ requires 1937), 1959 ([M + Na]$^+$).

9c. This product was obtained from **8c** (13 mg, 4.6 μmol) according to the procedure for **9a**. This yielded the product as a white powder (2.2 mg, 25%). *m*/*z* (ESI) 969 ([M + 2H]$^{2+}$), 980 ([M + H+Na]$^{2+}$), 991 ([M + 2Na]$^{2+}$), 1937 ([M + H]$^+$. $C_{87}H_{142}N_9O_{39}$ requires 1937).

9d. Protected CTV-construct **8d** (7 mg, 2.2 μmol) was dissolved in methanol (1 mL) and a 55% solution of sodium methoxide in methanol (10 μl) was added. After shaking this mixture overnight, the solution was neutralized with Dowex-H$^+$ ion exchange resin (25 mg). The resin was removed by filtration, and the volatiles were evaporated. After dissolving the product in water (5 mL) and lyophilization, the product was obtained as a white powder (1.4 mg, 28%). *m*/*z* (ESI) 1126 ([M + 2H]$^{2+}$. $C_{96}H_{152}N_6O_{54}{}^{2+}$ requires 1126), 1138 ([M + H + Na]$^{2+}$), 1148 ([M + 2Na]$^{2+}$).

9e. This product was obtained from **8e** (5 mg, 1.4 μmol) according to the procedure for **9a**. This yielded the product as a white powder (1.3 mg, 38%). *m*/*z* (ESI) 1214 ([M + 2H]$^{2+}$. $C_{102}H_{158}N_6O_{60}{}^{2+}$ requires 1214), 1225 ([M + H + Na]$^{2+}$), 1236 ([M + 2Na]$^{2+}$).

9f. This product was obtained from **8f** (10 mg, 2.7 μmol) according to the procedure for **9a**. This yielded the product as a white powder (4.4 mg, 67%). *m*/*z* (ESI) 1212 ([M + 2H]$^{2+}$. $C_{105}H_{173}N_9O_{54}{}^{2+}$ requires 1212), 1224 ([M + H + Na]$^{2+}$), 1234 ([M + 2Na]$^{2+}$).

9g. This product was prepared from **8g** (12 mg, 4.6 μmol) according to the procedure for **9a**. This yielded the product as a white powder (3.7 mg, 44%). *m*/*z* (ESI) 904 ([M + 2H]$^{2+}$), 917 ([M + H+Na]$^{2+}$), 927 ([M + 2Na]$^{2+}$), 1808 ([M + H]$^+$, $C_{78}H_{115}N_6O_{42}$ requires 1808), 1830 ([M + Na]$^+$).

9h. This product was obtained from **8h** (6 mg, 2.2 μmol) according to the procedure for **9a**. This yielded the product as a white powder (1.1 mg, 28%). *m*/*z* (ESI) 903 ([M + 2H]$^{2+}$), 915 ([M + H+Na]$^{2+}$), 925 ([M + 2Na]$^{2+}$), 1805 ([M + H]$^+$. $C_{81}H_{130}N_9O_{36}$ requires 1805), 1827 ([M + Na]$^+$).

9i. This product was obtained from **8i** (12 mg, 4.4 μmol) according to the procedure for **9a**. This yielded the product as a white powder (5.2 mg, 65%). *m*/*z* (ESI) 903 ([M + 2H]$^{2+}$), 914 ([M + H+Na]$^{2+}$), 925 ([M + 2Na]$^{2+}$), 1804 ([M + H]$^+$. $C_{81}H_{130}N_9O_{36}$ requires 1805), 1827 ([M + Na]$^+$).

9j. This product was obtained from **8j** (12 mg, 3.9 μmol) according to the procedure for **9d**. This yielded the product as a white powder (4.1 mg, 48%). *m*/*z* (ESI) 1085 ([M + 2H]$^{2+}$. $C_{90}H_{140}N_6O_{54}{}^{2+}$ requires 1085), 1095 ([M + H + Na]$^{2+}$) 1107 ([M + 2Na]$^{2+}$).

9k. This product was obtained from **8k** (15 mg, 4.4 μmol) according to the procedure for **9a**. This yielded the product as a white powder (6.6 mg, 65%). *m*/*z* (ESI) 1171 ([M + 2H]$^{2+}$. $C_{96}H_{146}N_6O_{60}{}^{2+}$ requires 1172), 1183 ([M + H + Na]$^{2+}$), 1194 ([M + 2Na]$^{2+}$).

9l. This product was obtained from **8l** (17 mg, 4.7 μmol) according to the procedure for **9a**. This yielded the product as a white powder (4.5 mg, 41%). *m*/*z* (ESI) 1171 ([M + 2H]$^{2+}$. $C_{99}H_{161}N_9O_{54}{}^{2+}$ requires 1170), 1181 ([M + H + Na]$^{2+}$), 1192 ([M + 2Na]$^{2+}$).

9m. This product was obtained from **8m** (48.5 mg, 15 μmol) according to the procedure for **9d**. This yielded the product as a white powder (25.0 mg, 72%). *m*/*z* (ESI) 751 ([M + 3H]$^{3+}$), 1126 ([M + 2H]$^{2+}$. $C_{96}H_{152}N_6O_{54}{}^{2+}$ requires 1127), 1137 ([M + H + Na]$^{2+}$), 1149 ([M + 2Na]$^{2+}$).

9n. This product was obtained from **8n** (16.0 mg, 4.5 μmol) according to the procedure for **9a**. This yielded the product as a

white powder (8.3 mg, 77%). *m*/*z* (ESI) 809 ($[M + 3H]^{3+}$), 1214 ($[M + 2H]^{2+}$. $C_{102}H_{158}N_6O_{60}^{2+}$ requires 1214), 1224 ($[M + H + Na]^{2+}$), 1236 ($[M + 2Na]^{2+}$).

9o. This product was obtained from **8o** (58.9 mg, 16 μmol) according to the procedure for **9a**. This yielded the product as a white powder (29.2 mg, 76%). *m*/*z* (ESI) 808 ($[M + 3H]^{3+}$), 1212 ($[M + 2H]^{2+}$. $C_{105}H_{173}N_9O_{54}$ requires 1212), 1223 ($[M + H + Na]^{2+}$), 1234 ($[M + 2Na]^{2+}$).

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References

1 P. C. Weber, J. J. Wendoloski, M. W. Pantoliano and F. R. Salemme, *J. Am. Chem. Soc.*, 1992, **114**, 3197–3200.
2 For non-carbohydrate examples, see: (*a*) A. P. Tamiz, J. R. Zhang, M. Zhang, C. Z. Wang, K. M. Johnson and A. P. Kozikowski, *J. Am. Chem. Soc.*, 2000, **122**, 5393–5394; (*b*) A. A. Profit, T. R. Lee and D. S. Lawrence, *J. Am. Chem. Soc.*, 1999, **121**, 280–283; (*c*) N. Schaschke, G. Matschiner, F. Zettl, U. Marquardt, A. Bergner, W. Bode, C. P. Sommerhoff and L. Moroder, *Chem. Biol.*, 2001, **8**, 313–327; (*d*) G. Loidl, M. Groll, H.-J. Musiol, R. Huber and L. Moroder, *Proc. Natl. Acad. Sci. USA*, 1999, **96**, 5418–5422.
3 (*a*) M. Mammen, S.-K. Choi and G. M. Whitesides, *Angew. Chem., Int. Ed. Engl.*, 1998, **37**, 2754–2794; (*b*) L. L. Kiessling, L. E. Strong and J. E. Gestwicki, *Ann. Rep. Med. Chem.*, 2000, **35**, 321–330.
4 (*a*) Y. C. Lee and R. T. Lee, *Acc. Chem. Res.*, 1995, **28**, 321–327; (*b*) R. T. Lee and Y. C. Lee, *Glycoconjugate J.*, 2000, **17**, 543–551.
5 (*a*) R. Roy, *Carbohydr. Drug Res.*, 1997, 83–135; (*b*) H. C. Hansen, S. Haataja, J. Finne and G. Magnusson, *J. Am. Chem. Soc.*, 1997, **119**, 6974–6979; (*c*) P. I. Kitov, J. M. Sadowska, G. Mulvey, G. D. Armstrong, H. Ling, N. S. Pannu, R. J. Read and D. R. Bundle, *Nature*, 2000, **403**, 669–672; (*d*) E. K. Fan, Z. S. Zhang, W. E. Minke, Z. Hou, C. L. M. J. Verlinde and W. G. J. Hol, *J. Am. Chem. Soc.*, 2000, **122**, 2663–2664; (*e*) E. K. Fan, E. A. Merritt, C. L. M. J. Verlinde and W. G. J. Hol, *Curr. Opin. Struct. Biol.*, 2000, **10**, 680–686; (*f*) L. A. J. M. Sliedregt, P. C. N. Rensen, E. T. Rump, P. J. Van Santbrink, M. K. Bijsterbosch, A. R. P. M. Valentijn, G. A. Van der Marel, J. H. Van Boom, T. J. C. Van Berkel and E. A. L. Biessen, *J. Med. Chem.*, 1999, **42**, 609–618; (*g*) C. Grandjean, H. Gras-Masse and O. Melnyk, *Chem. Eur. J.*, 2001, **7**, 230–239.
6 (*a*) I. Vrasidas, N. J. De Mol, R. M. J. Liskamp and R. J. Pieters, *Eur. J. Org. Chem.*, 2001, 4685–4692; (*b*) I. Vrasidas, S. André, P. Valantini, C. Böck, M. Lensch, H. Kaltner, R. M. J. Liskamp, H-J. Gabius and R. J. Pieters, *Org. Biomol. Chem.*, 2003, **1**, 803–810.
7 For a review, see: A. Collet, *Tetrahedron*, 1987, **43**, 5725–5739.
8 A. M. A. Van Wageningen and R. M. J. Liskamp, *Tetrahedron Lett.*, 1999, **40**, 9347–9351.
9 E. T. Rump, D. T. S. Rijkers, H. W. Hilbers, P. G. De Groot and R. M. J. Liskamp, *Chem. Eur. J.*, 2002, **8**, 4613–4621.
10 J. Canceill, A. Collet and G. Gottarelli, *J. Am. Chem. Soc.*, 1984, **106**, 5997–6003.
11 (*a*) F. J. Dekker, N. J. De Mol, J. Van Ameijde, M. J. E. Fischer, R. Ruijtenbeek, F. A. M. Redegeld and R. M. J. Liskamp, *ChemBioChem.*, 2002, **3**, 238–242; (*b*) C. M. Dreef-Tromp, J. E. M. Basten, M. A. Broekhoven, T. G. Van Dinther, M. Petitou and C. A. A. Van Boeckel, *Bioorg. Med. Chem. Lett.*, 1998, **8**, 2081–2086; (*c*) S. Keil, C. Claus, W. Dippold and H. Kunz, *Angew. Chem., Int. Ed.*, 2001, **40**, 366–369; (*d*) J. Slama and R. R. Rando, *Carbohydr. Res.*, 1981, **88**, 213–221; (*e*) O. Seitz and H. Kunz, *Angew. Chem., Int. Ed. Engl.*, 1995, **34**, 803–805; (*f*) R. H. Kramer and J. W. Karpen, *Nature*, 1998, **395**, 710–713; (*g*) J. Mack, K. Falk, O. Rotzschke, T. Walk, J. L. Strominger and G. Jung, *J. Pept. Sci.*, 2001, **7**, 338–345.
12 J. Van Ameijde, H. B. A. Albada and R. M. J. Liskamp, *J. Chem. Soc., Perkin Trans. 1*, 2002, 1042–1049.
13 For a recent review on glycopeptides and glycoproteins see: B. G. Davis, *Chem. Rev.*, 2002, **102**, 579–602.