DOI: 10.1002/ejoc.201201412

Efficient Synthesis of Thiolactoside Glycoclusters by Ruthenium-Catalyzed Cycloaddition Reaction of Disubstituted Alkynes on Carbohydrate Scaffolds

Alejandro J. Cagnoni,[a,b] Oscar Varela,[a] María Laura Uhrig,*[a] and José Kovensky*[b]

Keywords: Carbohydrates / Glycoconjugates / Glycoclusters / Click reaction / Ruthenium / Calorimetry

Lectin-carbohydrate interactions are responsible for several cellular processes involved in the immune system and the development of certain types of cancer. To further understanding of the cellular responses triggered by these interactions, complex carbohydrates are designed and prepared. Here, we describe the synthesis of a family of multivalent glycoclusters based on carbohydrate cores bearing thiolactosides or thiogalactosides as recognition elements with structural valencies ranging from 2 to 8. The synthetic strategy involves a key ruthenium-catalyzed cycloaddition reaction between symmetric disubstituted alkynes bearing two thiosugar units and azide-containing carbohydrate scaffolds. This methodology afforded high-valency glycoconjugates in good to excellent yields. Binding affinities of the synthetic β-thiolactosides for peanut lectin were measured by isothermal titration calorimetry. These titrations revealed micromolar affinities as well as a multivalent effect. A tetravalent glycoconjugate based on a trehalose scaffold displayed the highest binding affinity.

Introduction

The synthesis of multivalent ligands over the last years has been addressed in many ways in the search for improved methodologies to obtain high-valency glycoclusters through high-yielding optimized processes.[1] Indeed, glycoclusters that interfere with carbohydrate-protein recognition processes are seen as promising chemotherapeutics owing to the relevance of these interactions in triggering many cellular recognition processes such as viral and bacterial infections, inflammation and tumor metastasis.[2] The multivalent display of sugar residues, leading to the so-called "cluster effect", is used by natural systems to overcome the usually weak binding affinities displayed in these processes.[3]

The study of protein-carbohydrate interactions and the development of new chemotherapeutic agents have prompted the synthesis of a variety of multivalent glycoconjugates such as glycoclusters,[1a] glycodendrimers[1a] and glycopolymers.[4] Thus, owing to the availability of these synthetic multivalent ligands, the "cluster effect" has been studied in detail for several lectin systems over recent years.[5]

Scaffolds of diverse properties, structure, flexibility, and valency have been employed for the preparation of multivalent ligands useful for biomedical applications.[6] Small C_4-symmetric calix[4]arenes[7] and porphyrin[8] cores were used as scaffolds of multivalent ligands of pharmaceutical interest. In addition, large-sized dendrimers,[9] nanoparticles[10] and highly-flexible supramolecular platforms like rotaxanes[11] and polymers[12] have also been employed for the synthesis of promising anti-adhesive agents for toxins and other relevant lectins. Carbohydrates, including monosaccharides, disaccharides, oligosaccharides and cyclodextrins, have been used as scaffolds in the synthesis of multivalent ligands.[13] These materials may be more appropriate for in vivo applications than aromatic or polymeric scaffolds, owing to their biocompatibility.

In order to connect the different sugar epitopes or recognition elements to the scaffold, the copper-catalyzed alkyne–azide cycloaddition has become a powerful tool in bioorganic chemistry, because this reaction leads exclusively to 4-substituted 1,2,3-triazoles in high yields.[14] Using this reaction, we have successfully coupled a variety of carbohydrate epitopes armed with alkynyl linkers to azide-functionalized sugar scaffolds affording multivalent glycoclusters: multimannosides,[13c] multilactosides,[15] multimeric heptylmannosides,[16] and multi thiogalactosides.[17]

Furthermore, it has been reported that ruthenium-catalyzed alkyne-azide cycloaddition (RuAAC) reactions readily engage internal alkynes under catalysis with Cp*RuCl-$(PPh_3)_2$ or Cp*RuCl(COD) providing access to fully-substi-

[a] CIHIDECAR-CONICET, Departamento de Química Orgánica, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Ciudad Universitaria, Pab II, 3er piso, 1428 Ciudad de Buenos Aires, Argentina
Fax: +54-1145763352
E-mail: mluhrig@qo.fcen.uba.ar
Homepage: http://www.qo.fcen.uba.ar
[b] Laboratoire des Glucides, CNRS FRE 3517, Université de Picardie Jules Verne
33 Rue Saint Leu, 80039 Amiens, France
Fax: +33-3-22827568
E-mail: jose.kovensky@u-picardie.fr
Homepage: http://www.u-picardie.fr/labo/LG/
Supporting information for this article is available on the WWW under http://dx.doi.org/10.1002/ejoc.201201412.

tuted 1,2,3-triazoles.[18] The RuAAC reaction could be useful for the synthesis of high-valency glycoclusters from scaffolds having a limited number of azide groups.

We report here the cycloaddition reaction of internal (disubstituted) alkynes with azides by using Ru^{II} catalysis as a key step in the synthesis of high-valency glycoclusters. This methodology overcomes the limitation imposed by the number of azido groups on the oligosaccharide scaffold as "clickable" epitopes. Consequently, the number of recognition elements coupled to the scaffold can be doubled in one reaction step. The usefulness of this new approach is demonstrated here by the synthesis of multi-thiogalactosides and multi-thiolactosides. So far, the RuAAC reaction has been primarily used for simple azides and terminal alkynes. To the best of our knowledge, there are only two examples in which RuAAC has been applied to connect sugar azides to terminal alkynes, but not disubstituted ones.[19] In this work we use RuAAC as a key step for the preparation of multivalent glycoclusters.

Results and Discussion

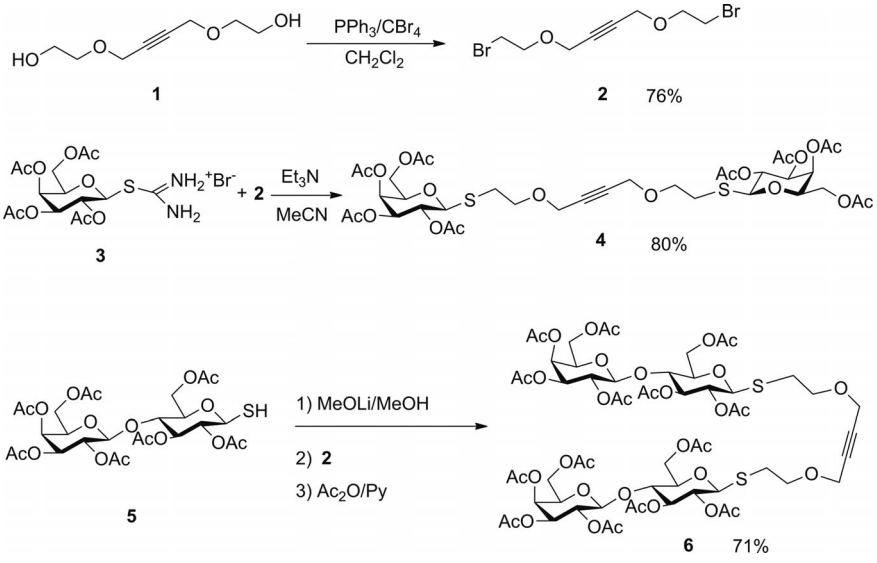
The strategy involves the synthesis of a symmetric alkyne linker functionalized with two sugar residues (Scheme 1). Starting diol **1** was converted into dibromide **2** by reaction with PPh_3–CBr_4 (76% yield).[20] The disubstituted alkyne carrying two thiogalactose units was prepared by reaction of **2** with isothiouronium salt **3** to give **4** (80% yield).[17] On the other hand, the preparation of **6** from thiogalactoside **5** and dibromide **2** was achieved in 71% yield under stronger basic conditions, such as treatment with MeOLi-MeOH, followed by acetylation.[21]

The simplicity of the NMR spectra of compounds **4** and **6** confirmed the advantages of symmetric precursors in terms of characterization. For example, the NMR spectra of **4** and **6** showed a single set of carbohydrate signals corresponding to the symmetric galactose and lactose epitopes. For these compounds and their derivatives, the signal of the anomeric carbon linked to the sulfur (ca. 85 ppm in the ^{13}C NMR spectrum) was considered diagnostic, confirming the presence of such residues in the final products.

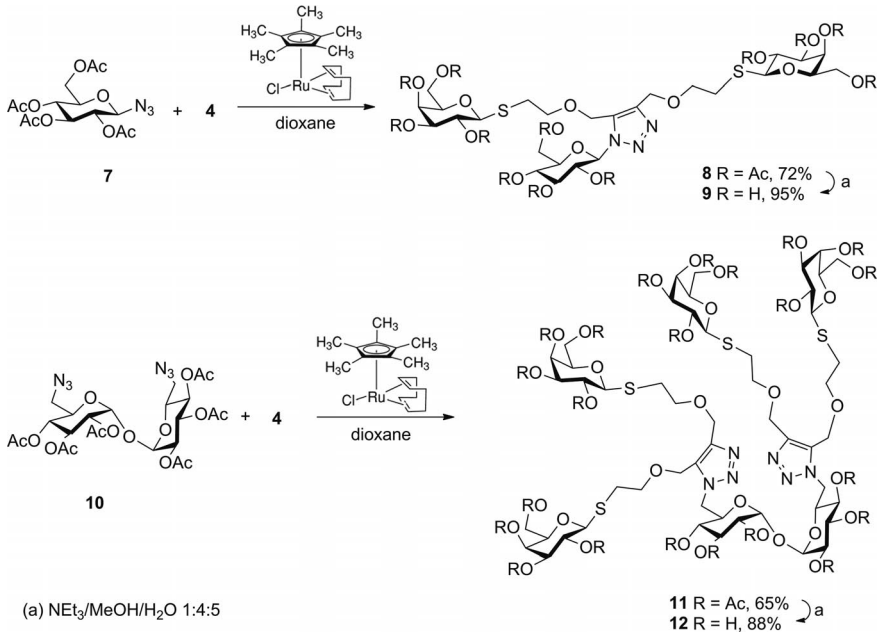
These dimers were coupled to two different azide-containing scaffolds previously prepared: azidoglucose **7** and diazidotrehalose **10**.[17] Dithiogalactoside **4** was used in the first instance for optimization of the ruthenium-catalyzed cycloaddition. In this reaction, it was shown that the alkyne should be added first, followed by the addition of the azide, because the latter may react with the ruthenium catalyst forming inactive [Cp*RuCl] tetraazadiene complexes.[18c] Alternatively, a solution of both the azide and the alkyne can be added to the catalyst dissolved in the chosen solvent. Thus, a dioxane solution of **4** and **7** was added to the Cp*RuCl(COD) catalyst dissolved in dioxane under an argon atmosphere. The reaction mixture was stirred at room temperature until TLC indicated complete consumption of the starting material. In this way, dithiogalactoside **8** was obtained in 72% yield after work-up and purification by column chromatography. The reaction between **4** and **10** carried out under the same conditions afforded tetrathiogalactoside **11** in 65% yield. Deacetylation of **8** and **11** led to free glycoclusters **9** and **12**, respectively, in excellent yields (Scheme 2).

Reaction of **7** and **10** with dithiolactoside **6** afforded di- and tetra-thiolactosides **14** and **16**, respectively (Scheme 3).

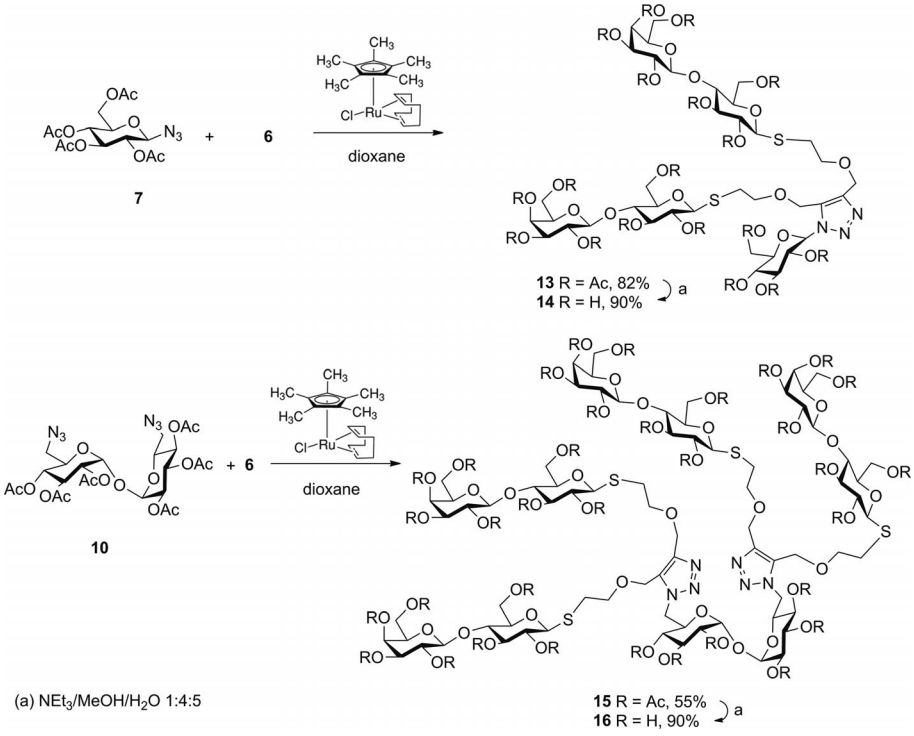
These results confirmed that the proposed strategy had high potential for the preparation of the target glycoclusters. Higher-valency derivatives could be easily accessed by simple modification of the scaffolds by using the same strategy to double the number of clickable positions. Thus, mono-azide scaffold **7** was converted into diazide **19**, by a RuAAC reaction, using diol **17** followed by functional group manipulation (Scheme 4). The same sequence applied to 6,6′-diazidotrehalose (**10**) afforded tetraazido derivative



Scheme 1. Synthesis of symmetric alkyne linkers **4** and **6**.



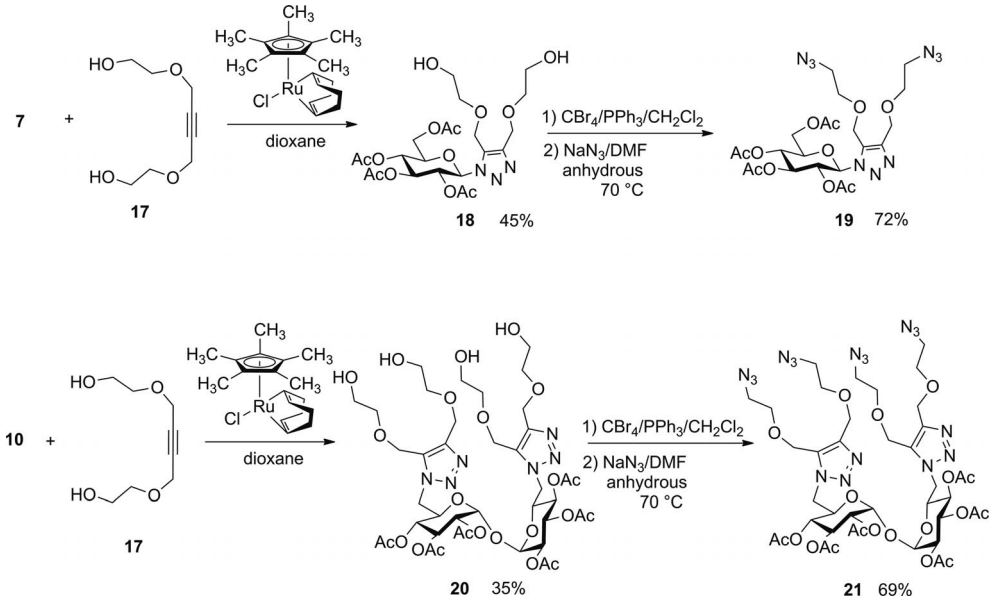
Scheme 2. RuAAC of symmetric dithiogalactoside **4** to azide-containing sugar scaffolds **7** and **10**.



Scheme 3. RuAAC of symmetric dithiolactoside **6** to azide-containing sugar scaffolds **7** and **10**.

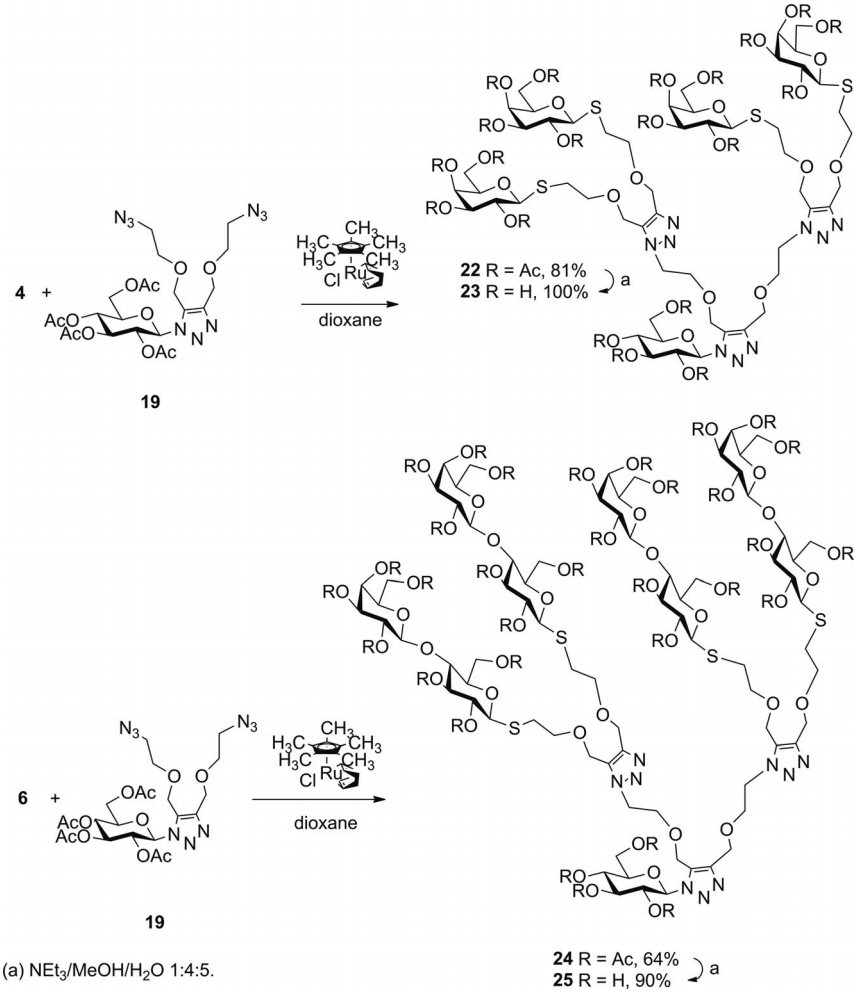
21. It should be mentioned that the first attempts to perform the dipolar cycloaddition reaction directly between **7** and dibromide **2** were unsuccessful. Probably dibromide **2** reacts with the ruthenium catalyst rendering it inactive for the azide–alkyne cycloaddition.

Reaction of scaffold **19** with dithiogalactoside derivative **4**, in the presence of ruthenium catalyst, led to tetravalent compound **22** in 81% yield. Although dithiolactoside **6** showed a lower reactivity than **4**, tetralactoside **24** was obtained in 64% yield (Scheme 5).

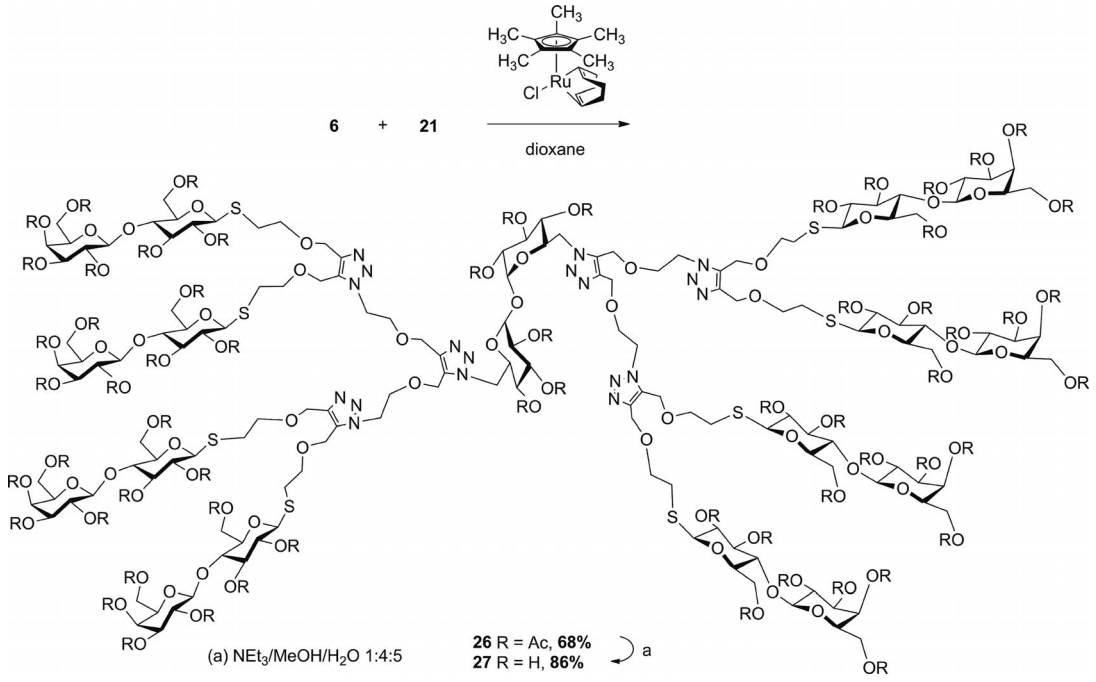


Scheme 4. Strategy used to double the number of clickable positions: synthesis of **19** and **21**.

The NMR spectra of **22** showed that the four β-thiogalactose residues are not exactly equivalent, and the signals for the anomeric carbon atoms in the ^{13}C NMR spectrum appeared as two close peaks. This fact could be ascribed to some conformational restriction or a different chiral environment leading to differentiation of the four resi-



Scheme 5. Synthesis of tetravalent glycoclusters **23** and **25**.



Scheme 6. Synthesis of octavalent glycocluster **27**.

dues in two pairs. The same pattern was observed in the ^{13}C NMR spectra (anomeric region) of tetrathiolactose glycocluster **24**.

The most relevant result was the synthesis of octavalent thiolactoside **26**, which was obtained by reaction of **21** and **6** in a 1:4 ratio. After 6 h of stirring at room temperature, the cycloaddition reaction occurred on the four azide groups attached to the trehalose scaffold, leading to **26** in 68% yield, in a single step (Scheme 6).

Remarkably, the NMR spectra of octavalent glycoclusters **26** and **27** were quite simple, owing to the symmetry of the molecules, showing that the lactose residues are almost equivalent. Nevertheless, the molecular weight, indicating the presence of 18 monosaccharides and 6 triazole rings, was confirmed by mass spectrometry.

Isothermal Titration Calorimetry

Peanut (*Arachis Hypogaea*) agglutinin (PNA) is a homotetrameric legume lectin, specific to galactose at the monosaccharide level, although it displays a higher affinity towards lactose derivatives.[22] It has previously been used as a model lectin to study the cluster effect in multivalent ligands.[22c] In particular, isothermal titration calorimetry (ITC) analysis is a useful tool for the determination of the thermodynamic parameters of binding (K, ΔG, ΔH, ΔS and n). Glycoclusters **14**, **16**, **25** and **27**, bearing thiolactose residues as recognition elements, were evaluated as ligands for PNA by using lactose as reference compound (Table 1).

Divalent derivative **14** showed a multivalent effect and its binding affinity was 18.8-fold higher than lactose (9-fold higher on a lactose molar basis).

The values obtained for tetravalent derivatives **16** and **25** showed the influence of the scaffold, which determines the ligand presentation, on the affinity for PNA. Tetrathiolactose **25** binds to PNA with a relative potency of 96.3 relative to lactose, which means a 24-fold enhancement on a lactose molar basis.

On the other hand, tetravalent compound **16** (Figure 1), based on the trehalose scaffold, showed a 137.8-fold higher binding affinity (34-fold on a lactose basis). This scaffold may provide a different arrangement of thiolactosides fac-

Table 1. Termodynamic binding parameters of the synthetic glycoclusters related to lactose that were used as reference. Val refers to the structural valency of the ligand; n is the stoichiometry of the binding; Pot rel is the relative potency of the ligand referred to the lactose reference; Pot rel/lac is the corrected potency on a lactose molar basis.

Compound	val	n	K_a $\times 10^{-3}$ M^{-1}	ΔH [kcal mol^{-1}]	$T\Delta S$ [kcal mol^{-1}]	ΔG [kcal mol^{-1}]	Pot rel	Pot rel/lac
Lactose	1	0.98	2.46	–10.9	–6.3	–4.6	1	1
14	2	0.52	46.2	–18.8	–12.5	–7.3	18.8	9
16	4	0.26	339	–49.3	–38.7	–10.6	137.8	34
25	4	0.43	237	–34.7	–23.8	–10.9	96.3	24
27	8	0.18	296	–79.5	–68.5	–11.0	120.3	15

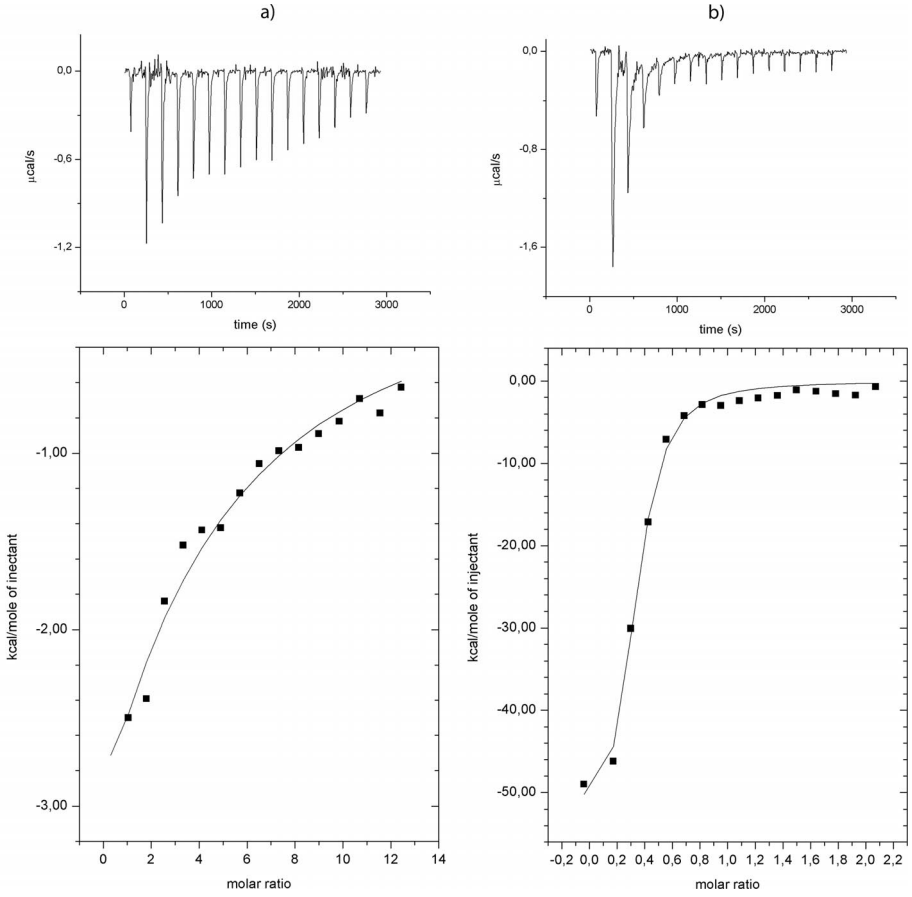


Figure 1. Experimental calorimetric data associated with the isothermal titration at 298 K of PNA (50 μM) with (a) lactose (3 mM) and (b) compound **16** (1 mM).

ing different directions (two of them opposite to the other two), which could explain the differences observed. The *n* values suggest that four lectin molecules would bind to compound **16** (trehalose scaffold, $n = 0.26$), whereas compound **25** (glucose scaffold, $n = 0.43$) would only accommodate two or three lectin molecules. These facts suggest that some of the sugar epitopes in **25** do not participate in the binding process. The affinity constants values observed $K_a = 339$ and 237 mM for **16** and **25**, respectively, are consistent with this assumption.

The binding enthalpies of multivalent glycoclusters to the lectin increase almost linearly with the number of thiolactose residues. As shown in Table 1, the entropic terms increase concomitantly. The relatively narrow range of binding free-energies provides binding curves consistent with a single site model, indicating a compensation of enthalpy and entropy factors for these glycoclusters.

Octathiolactoside **27** showed, however, a lower binding affinity than tetravalent glycocluster **16**, with a 96.3-fold increase relative to lactose (15-fold on a lactose basis). The optimal valency was four thiolactoside epitopes for the trehalose scaffold.

Conclusions

In conclusion, we have developed an efficient and atom-economic strategy for the synthesis of multivalent thiogalactosides and thiolactosides with valencies that range from 2 to 8. This methodology overcomes the limitations regarding the number of recognition elements imposed by the number of clickable groups on the scaffolds. A renewed synthetic potential for the oligosaccharide scaffolds can be envisaged, because the number of sugar epitopes connected in a single cycloaddition step is now doubled. Moreover, these high-valency glycoclusters, based on oligosaccharide scaffolds carrying free hydroxy groups, are expected to display high solubility and compatibility in biological media.

The parameters associated to the interaction between the synthetic ligands and the peanut lectin, determined by ITC measurements, provided insights into the binding mode. All glycoclusters exhibited higher binding affinities than lactose used as reference. Moreover, a significant binding improvement was observed in the case of tetravalent glycoclusters relative to divalent ones (24- or 34-fold on a lactose basis). The differences observed between two tetravalent glycoclusters **16** and **25** may be because of the spatial arrangement of the sugar epitopes, suggesting the effect of subtle structural differences of the scaffold on the presentation of the sugar epitopes to the lectin. The binding affinity shown by octavalent compound **27** was higher than that of tetravalent glycocluster **25**, but lower than that of **16**.

Although PNA is a tetrameric lectin, a chelate effect does not seem possible because, from our preliminary calcula-

tions, the distance between the sugar epitopes is approximately 20 Å which is much less than the shortest separation of the lectin binding sites (70 Å).[22] Thus, the increment in binding affinity observed may be attributed either to a multivalent effect of internal diffusion (sliding mechanism), in which the proximity of the recognition elements allow their subsequent binding and recapture by a unique protein; or to an aggregative process, in which the distance between the ligands is long enough to allow different lectins to bind simultaneously to the same glycocluster without steric constraints.[23,24]

Experimental Section

General Procedures: Analytical thin layer chromatography (TLC) was performed on Silica Gel 60 F254 aluminum supported plates (layer thickness 0.2 mm) with the solvent systems given in the text. Visualization of the spots was achieved by exposure to UV light or by charring with a solution of 5% (v/v) sulfuric acid in EtOH, containing 0.5% *p*-anisaldehyde. Column chromatography was carried out with Silica Gel 60 (230–400 mesh). Optical rotations were measured using a 343 Perkin–Elmer instrument at 20 °C in a 10 cm cell in the stated solvent. $[\alpha]_D$ values are given in 10^{-1} deg cm^{-1} g^{-1} (concentration *c* given as g/100 mL). High-resolution mass spectra (HRMS) were obtained by electrospray ionization (ESI) using a Micromass-Waters Q-TOF Ultima Global instrument. ^{1}H and ^{13}C nuclear magnetic resonance (NMR) spectra were recorded at 25 °C at 500 and 125 MHz, respectively, using a Bruker AC-500 or at 600 and 150 MHz, respectively, using a Bruker AC-600 spectrometer. Chemical shifts are reported relative to tetramethylsilane or a residual solvent peak ($CHCl_3$: ^{1}H: δ = 7.26 ppm; ^{13}C: δ = 77.2 ppm). Assignments of ^{1}H and ^{13}C were assisted by 2D ^{1}H-COSY and 2D ^{1}H-^{13}C CORR experiments. Peak multiplicity is reported as: singlet (s), doublet (d), triplet (t), quartet (q), multiplet (m), and broad (br.). The capital letters "G" and "L" refer to galactose and lactose signals, respectively. Microwave irradiation was performed with a CEM Discover apparatus (300 W).

Compound 2: Tetrabromomethane (420 mg, 1.265 mmol) was added to a solution of 1,4-bis(2-hydroxyethoxy)-2-butyne (100 mg, 0.574 mmol) in dichloromethane (1 mL) under an argon atmosphere. The reaction mixture was cooled to 0 °C, and a solution of triphenylphosphane (330 mg, 1.265 mmol) in dichloromethane (2 mL) was added dropwise. After stirring for 3 h, the solvent was evaporated and the residue was purified by flash chromatography (hexane/EtOAc, 9:1) to obtain pure **2** (131 mg, 76%). R_f = 0.56 (hexane/EtOAc, 3:1). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 4.26 (s, 4 H, CH_2–C≡C), 3.84 (t, *J* = 6.2 Hz, 4 H, CH_2O), 3.48 (t, *J* = 6.2 Hz, 4 H, CH_2Br) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 82.4 (*C*≡C), 69.7 (*C*H$_2$–C≡C), 58.5 (*C*H$_2$O), 30.0 (*C*H$_2$Br) ppm. $C_8H_{12}Br_2O_2$ (299.99): calcd. C 32.03, H 4.03, Br 53.27; found C 32.28, H 4.31. HRMS (ESI): calcd. for $C_8H_{12}Br_2O_2$ [M + Na]$^+$ 320.9096; found 320.9109.

Compound 4: Triethylamine (189 μL, 1.35 mmol) was added dropwise to a solution of 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosylisothiouronium bromide (292 mg, 0.6 mmol) and dibromide **2** (90 mg, 0.3 mmol) in CH_3CN (3 mL) at room temperature under an argon atmosphere. After 8 h, the solvent was evaporated and the residue was purified by column chromatography (hexane/EtOAc, 2:3) to afford **4** (208 mg, 80%). $[\alpha]_D^{20}$ = –20.6 (*c* = 0.3 in $CHCl_3$). R_f = 0.30 (hexane/EtOAc, 1:1.5). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.44 (dd, $J_{4,5}$ = 1.0, $J_{3,4}$ = 3.4 Hz, 1 H, 4-H), 5.22 (t, $J_{1,2}$ = $J_{2,3}$ = 10.0 Hz, 1 H, 2-H), 5.07 (dd, $J_{3,4}$ = 3.4, $J_{2,3}$ = 10.0 Hz, 1 H, 3-H), 4.57 (d, $J_{1,2}$ = 10.0 Hz, 1 H, 1-H), 4.23 (s, 2 H, CH_2–C≡C), 4.16 (dd, $J_{5,6a}$ = 6.7, $J_{6a,6b}$ = 11.3 Hz, 1 H, 6-Ha), 4.13 (dd, $J_{5,6b}$ = 7.6, $J_{6a,6b}$ = 11.3 Hz, 1 H, 6-Hb), 3.95 (ddd, $J_{4,5}$ = 1.0, $J_{5,6a}$ = 6.7, $J_{5,6b}$ = 7.6 Hz, 1 H, 5-H), 3.78–3.65 (m, 2 H, CH_2O), 2.98, 2.83 (2m, 2 H, CH_2S), 2.16, 2.08, 2.05, 1.99 (4s, 12 H, 4 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.3, 170.1, 170.0, 169.5 (–*C*OCH$_3$), 84.0 (C-1), 82.2 (–*C*≡C–), 74.4 (C-5), 71.7 (C-3), 69.5 (*C*H$_2$O), 67.2 (×2) (C-2,C-4), 61.3 (C-6), 58.3 (*C*H$_2$–C≡C), 29.31 (*C*H$_2$S), 20.8 (×2), 20.6 (×2) (*C*H$_3$CO) ppm. $C_{36}H_{50}O_{20}S_2$ (866.90): calcd. C 49.88, H 5.81, S 7.40; found C 49.67, H 5.97, S 7.61. HRMS (ESI): calcd. for $C_{36}H_{50}O_{20}S_2$ [M + Na]$^+$ 889.2229; found 889.2208.

Compound 6: A 1 M LiOMe/MeOH solution (0.5 mL), cooled to 0 °C, was added to 4-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-1-thio-2,3,6-triacetate β-D-glucopyranose (80 mg, 0.123 mmol). After 30 min, dibromide **2** (18.5 mg, 0.0613 mmol) was added and the reaction mixture was stirred for 2 h at room temperature. The mixture was neutralized with acetic acid, concentrated, and the residue was treated overnight with acetic anhydride (1 mL) in pyridine (1 mL) at room temperature. Upon concentration and purification by column chromatography (hexane/EtOAc, 3:7) compound **6** was obtained (63 mg, 71%). $[\alpha]_D^{20}$ = –37.6 (*c* = 0.5, $CHCl_3$). R_f = 0.30 (hexane/EtOAc, 1:2). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.36 (dd, $J_{4',5'}$ = 0.9, $J_{3',4'}$ = 3.4 Hz, 1 H, 4′-H), 5.22 (t, $J_{2,3}$ = $J_{3,4}$ = 9.2 Hz, 1 H, 3-H), 5.12 (dd, $J_{1',2'}$ = 7.9, $J_{2',3'}$ = 10.4 Hz, 1 H, 2′-H), 4.98 (dd, $J_{3',4'}$ = 3.5, $J_{2',3'}$ = 10.4 Hz, 1 H, 3′-H), 4.93 (t, $J_{1,2}$ = $J_{2,3}$ = 9.7 Hz, 1 H, 2-H), 4.58 (d, $J_{1,2}$ = 10.0 Hz, 1 H, 1-H), 4.53 (dd, $J_{5,6a}$ = 2.0, $J_{6a,6b}$ = 12.0 Hz, 1 H, 6-Ha), 4.50 (d, $J_{1',2'}$ = 7.9 Hz, 1 H, 1′-H), 4.22 (s, 2 H, CH_2–C≡C–), 4.16 (dd, $J_{5',6'a}$ = 6.3, $J_{6'a,6'b}$ = 11.2 Hz, 1 H, 6′-Ha), 4.13 (dd, $J_{5',6'b}$ = 7.4, $J_{6'a,6'b}$ = 11.1 Hz, 1 H, 6′-Hb), 4.11 (dd, $J_{5,6b}$ = 5.1, $J_{6a,6b}$ = 12.0 Hz, 1 H, 6-Hb), 3.90 (ddd, $J_{4',5'}$ = 0.9, $J_{5',6'a}$ = 6.3, $J_{5',6'b}$ = 7.4 Hz, 1 H, 5′-H), 3.80 (t, $J_{3,4}$ = $J_{4,5}$ = 9.5 Hz, 1 H, 4-H), 3.72–3.57 (m, 3 H, 5-H, CH_2O), 2.92, 2.80 (2m, 2 H, CH_2S), 2.17, 2.14, 2.08, 2.07, 2.06 (× 2), 1.98 (7s, 21 H; 7 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.3, 170.2, 170.1, 170.0, 169.7, 169.6, 169.0 (–*C*OCH$_3$), 101.1 (C-1′), 83.4 (C-1), 82.3 (–C≡C–), 76.7 (C-5), 76.2 (C-4), 73.7 (C-3), 71.0 (C-3′), 70.7 (C-5′), 70.4 (C-2), 69.7 (*C*H$_2$O), 69.1 (C-2′), 66.6 (C-4′), 62.2 (C-6), 60.8 (C-6′), 58.4 (CH_2–C≡C–), 29.5 (*C*H$_2$S), 20.9 (× 2), 20.7, 20.6, 20.5, 20.4, 20.3 (*C*H$_3$CO) ppm. $C_{60}H_{82}O_{36}S_2$ (1443.41): calcd. C 49.93, H 5.73, S 4.44; found C 49.70, H 5.44, S 4.69. HRMS (ESI): calcd. for $C_{60}H_{82}O_{36}S_2$ [M + Na]$^+$ 1465.3919; found 1465.3975.

General Procedure for Ru^{II}-Catalyzed Cycloaddition Reaction: Exemplified for the synthesis of compound **8**. Cp*RuCl(COD) (4.0 mg, 0.010 mmol) was added to a tube with a septa cap. The tube was sealed, then evacuated, and filled with argon. This procedure was repeated three times. Dioxane (5 mL) was added followed by a dioxane solution of compound **4** (433 mg, 0.50 mmol) and azide **7** (187 mg, 0.50 mmol). The reaction was stirred at room temperature until TLC analysis indicated complete consumption of the starting materials (2–4 h). The solvent was evaporated and the mixture was adsorbed onto silica and purified by flash chromatography (hexane/EtOAc) to afford the pure product. (Note: in all reactions azide and alkyne were dissolved in the reaction solvent and added to the solution of the catalyst; azide should not be added first.)

Compound 8: Yield 72%. $[\alpha]_D^{20}$ = –12.2 (*c* = 0.2 in $CHCl_3$). R_f = 0.25 (hexane/EtOAc, 1:1). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.91 (d, $J_{1,2}$ = 9.4 Hz, 1 H, 1-H), 5.82 (t, $J_{1,2}$ = $J_{2,3}$ = 9.4 Hz, 1 H, 2-H), 5.37 (m, 3 H, 3-H, 4′-H, 4′′-H), 5.22 (t, $J_{3,4}$ = $J_{4,5}$ = 9.8 Hz, 1

H, 4-H), 5.15, 5.13 (2t, $J_{1',2'} = J_{2',3'} = J_{2'',3''} = 10.0$ Hz, 2 H, 2′-H, 2′′-H), 5.01, 4.99 (2dd, $J_{3',4'} = J_{3'',4''} = 3.4$, $J_{2',3'} = J_{2'',3''} = 9.9$ Hz, 2 H, 3′-H, 3′′-H), 4.70, 4.60 (2s, 4 H, CH_2-triazole), 4.49 (d, $J_{1',2'} = J_{1'',2''} = 9.9$ Hz, 2 H, 1′-H, 1′′-H), 4.24 (dd, $J_{5,6a} = 4.7$, $J_{6a,6b} = 12.6$ Hz, 1 H, 6-Ha), 4.13 (dd, $J_{5,6b} = 2.0$, $J_{6a,6b} = 12.6$ Hz, 1 H, 6-Hb), 4.10–3.96 (m, 5 H, 5-H, 6′-Ha, 6′-Hb, 6′′-Ha, 6′′-Hb), 3.90 (m, 2 H, 5′-H, 5′′-H), 3.65 (m, 4 H, 2× CH_2O), 2.94, 2.80 (2m, 4 H, CH_2S), 2.09 (× 2), 2.02, 2.01, 2.00, 1.99, 1.98, 1.97, 1.96, 1.92, 1.91, 1.81 (12s, 36 H; 12 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.3, 170.2, 170.0, 169.9, 169.5, 169.2, 168.6 (–*C*OCH$_3$), 143.7, 132.2 (*C*-triazole), 85.2 (C-1), 83.9, 83.7 (C-1′, C-1′′), 74.7 (C-5), 74.4, 74.3 (C-5′, C-5′′), 73.1 (C-3), 71.7, 71.6 (C-3′, C-3′′), 70.1, 70.0, 69.8 (C-2, 2× *C*H$_2$O), 67.5 (C-4), 67.2 (× 2), 67.1, 67.0 (C-2′, C-2′′, C-4′, C-4′′), 63.5, 60.0 (2× *C*H$_2$-triazole), 61.5, 61.2, 61.0 (C-6, C-6′, C-6′′), 29.4, 29.1 (2× *C*H$_2$S), 20.8 (× 2), 20.7, 20.6, 20.5, 20.4, 20.3, 20.2, 20.1 (*C*H$_3$CO) ppm. $C_{50}H_{69}N_3O_{29}S_2$ (1240.22): calcd. C 48.42, H 5.61, N 3.39, S 5.17; found C 48.17, H 5.44, N 3.28, S 4.94. HRMS (ESI): calcd. for $C_{50}H_{69}N_3O_{29}S_2$ [M + H]$^+$ 1240.3532; found 1240.3531.

Compound 11: Yield 65%. $[\alpha]_D^{20}$ = +8.8 (c = 0.1 in $CHCl_3$). R_f = 0.33 (hexane/EtOAc, 1:5). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.43 (t, $J_{2,3} = J_{3,4} = 9.5$ Hz, 1 H, 3-H), 5.37 (m, 2 H, 2× 4-HG), 5.13 (2t, $J_{1G,2G} = J_{2G,3G} = 10.0$ Hz, 2 H, 2× 2-HG), 5.00 (m, 3 H, 2-H, 2× 3-HG), 4.92 (t, $J_{3,4} = J_{4,5} = 9.5$ Hz, 1 H, 4-H), 4.67 (d, $J_{1,2} = 3.7$ Hz, 1 H, 1-H), 4.57–4.47 (m, 6 H, 5-H, 2× 1-HG, 2× CH_2-triazole), 4.45 (d, $J_{1G,2G} = 10.0$ Hz, 1 H, 1-HG), 4.40 (dd, $J_{5,6a} = 8.9$, $J_{6a,6b} = 14.4$ Hz, 1 H, 6-Ha), 4.33 (dd, $J_{5,6b} = 3.0$, $J_{6a,6b} = 14.4$ Hz, 1 H, 6-Hb), 4.10–4.00 (m, 4 H, 2× 6-HaG, 2× 6-HbG), 3.90 (ddd, $J_{4G,5G} = 1.0$, $J_{5G,6aG} = 6.7$, $J_{5G,6bG} = 7.8$ Hz, 2 H, 2× 5-HG), 3.65, 3.55 (2m, 4 H, 2× CH_2O), 2.87, 2.74 (2m, 4 H, 2× CH_2S), 2.12, 2.11, 2.10, 2.00, 1.99, 1.98, 1.97, 1.96, 1.92, 1.91, 1.90 (11s, 33 H, 11 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.0, 169.9, 169.5, 169.2, 168.6 (–*C*OCH$_3$), 142.8, 132.5 (*C*-triazole), 91.2 (C-1), 83.9, 83.6 (2× C-1G), 74.3, 74.2 (2× C-5G), 71.7 (2× C-3G), 70.4 (C-4), 69.9 (C-3), 69.9, 69.8 (2× *C*H$_2$O), 68.6 (C-5), 68.5 (C-2), 67.2 (× 2), 67.1, 67.0 (2× C-2G, 2× C-4G), 63.5, 59.8 (2× *C*H$_2$-triazole), 61.2, 61.1 (2× C-6G), 49.1 (C-6), 29.4, 29.0 (2× *C*H$_2$S), 21.0, 20.8, 20.7, 20.6, 20.5, 20.4, 20.3, 20.2, 20.1 (*C*H$_3$CO) ppm. $C_{96}H_{132}N_3O_{55}S_4$ (2336.33): calcd. C 48.48, H 5.59, N 3.53, S 5.39; found C 48.33, H 5.41, N 3.31, S 5.61. HRMS (ESI): calcd. for $C_{96}H_{132}N_3O_{55}S_4$ [M + H]$^+$ 2377.6672; found 2377.6683.

Compound 13: Yield 82%. $[\alpha]_D^{20}$ = +12.2 (c = 0.3 in $CHCl_3$). R_f = 0.25 (hexane/EtOAc, 1:4). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.96 (d, $J_{1,2} = 9.4$ Hz, 1 H, 1-H), 5.85 (t, $J_{1,2} = J_{2,3} = 9.4$ Hz, 1 H, 2-H), 5.41 (t, $J_{2,3} = J_{3,4} = 9.4$ Hz, 1 H, 3-H), 5.33 (m, 2 H, 2× 4′-HL), 5.26 (t, $J_{3,4} = J_{4,5} = 9.8$ Hz, 1 H, 4-H), 5.19, 5.17 (2t, $J_{2L,3L} = J_{3L,4L} = 9.2$ Hz, 2 H, 2× H-3L), 5.08, 5.06 (2dd, $J_{1'L,2'L} = 8.0$, $J_{2'L,3'L} = 10.4$ Hz, 2 H, H-2′L), 4.98, 4.97 (2dd, $J_{3'L,4'L} = 3.4$, $J_{2'L,3'L} = 10.4$ Hz, 2 H, 2× H-3′L), 4.92, 4.89 (2t, $J_{2L,3L} = 9.2$, $J_{1L,2L} = 9.5$ Hz, 2 H, 2× H-2L), 4.73, 4.64 (2s, 4 H, CH_2-triazole), 4.53, 4.51, 4.47 (m, 6 H, 2× H-1L, 2× H-1′L, 2× H-6aL), 4.28 (dd, $J_{5,6a} = 4.7$, $J_{6a,6b} = 12.6$ Hz, 1 H, 6-Ha), 4.17 (dd, $J_{5,6b} = 2.1$, $J_{6a,6b} = 12.6$ Hz, 1 H, 6-Hb), 4.12–3.99 (m, 7 H, 5-H, 2× H-6bL, 2× H-6a′L, 2× H-6b′L), 3.89 (m, 2 H, 2× H-5′L), 3.81, 3.79 (2t, $J_{3L,4L} = J_{4L,5L} = 9.5$ Hz, 2 H, 2× H-4L), 3.72–3.57 (m, 6 H, 2× H-5L, 2× CH_2O), 2.89, 2.76 (2m, 4 H, 2× CH_2S), 2.13, 2.09 (× 2), 2.06, 2.05, 2.04, 2.03, 2.02, 2.01, 2.00, 1.99, 1.98, 1.97, 1.96, 1.92, 1.91, 1.85, 1.81 (18s, 54 H, 18 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 171.1, 170.5, 170.4, 170.3, 170.2, 170.1, 170.0, 169.9, 169.6, 169.5, 169.3, 169.2, 168.6 (–*C*OCH$_3$), 143.8, 132.6 (*C*-triazole), 101.1 (2× C-1′L), 85.1 (C-1), 83.9, 83.7 (2× C-1L), 76.7 (2× C-5L), 76.1, 76.0 (2× C-4L), 74.7 (C-5), 73.7, 73.6 (2× C-3L), 73.2 (C-3), 71.0, 70.9 (2× C-3′L), 70.6 (2× C-5′L), 70.3, 70.2, 70.1, 70.0, 69.9 (C-2, 2× C-2L, 2× *C*H$_2$O), 69.1 (C-2′L), 67.6 (C-4), 66.6 (2× C-4′L), 62.1, 61.9 (2× C-6L), 61.6 (C-6), 63.4, 60.0 (2× *C*H$_2$-triazole), 60.7, 60.6 (2× C-6′L), 29.5, 29.3 (2× *C*H$_2$S), 20.8 (× 2), 20.7, 20.6, 20.5, 20.4, 20.3, 20.2, 20.1, 20.0 (*C*H$_3$CO) ppm. $C_{74}H_{101}N_3O_{45}S_2$ (1816.73): calcd. C 48.92, H 5.60, N 2.31, S 3.53; found C 48.70, H 5.44, N 2.40, S 3.69. HRMS (ESI): calcd. for $C_{74}H_{101}N_3O_{45}S_2$ [M + Na]$^+$ 1838.5041; found 1238.5012.

Compound 15: Yield 55%. $[\alpha]_D^{20}$ = +8.8 (c = 0.4 in $CHCl_3$). R_f = 0.19 (hexane/EtOAc, 1:5). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.47 (t, $J_{2,3} = J_{3,4} = 9.5$ Hz, 1 H, 3-H), 5.34 (m, 2 H, 2× H-4′L), 5.17 (t, $J_{2L,3L} = J_{3L,4L} = 9.2$ Hz, 2 H, 2× H-3L), 5.12–5.04 (m, 3 H, 2-H, 2× H-2′L), 4.98 (m, 3 H, 4-H, 2× H-3′L), 4.89 (t, $J_{1L,2L} = J_{2L,3L} = 9.7$ Hz, 2 H, H-2L), 4.69 (d, $J_{1,2} = 3.7$ Hz, 1 H, 1-H), 4.62–4.35 (m, 13 H, 5-H, 6-Ha, 6-Hb, 2× H-1L, 2× H-6aL, 2× H-1′L, 2× CH_2-triazole), 4.15–4.02 (m, 6 H, 2× H-6bL, 2× H-6a′L, 2× H-6b′L), 3.89 (ddd, $J_{4'L,5'L} = 1.0$, $J_{5'L,6a'L} = 6.7$, $J_{5'L,6b'L} = 7.8$ Hz, 2 H, 2× H-5′L), 3.79 (m, 2 H, 2× H-4L), 3.70–3.54 (m, 6 H, 2× H-5L, 2× CH_2O), 2.87, 2.74 (2m, 4 H, 2× CH_2S), 2.16, 2.14 (2×), 2.10, 2.08, 2.05 (2×), 2.04 (2×), 2.03, 2.02, 2.01, 2.00, 1.99, 1.96, 1.95, 1.94 (17s, 51 H, 17 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.3, 170.2, 170.1, 170.0, 169.9, 169.5, 169.2, 168.6 (–*C*OCH$_3$), 142.8, 132.7 (*C*-triazole), 101.1, 101.0 (2× C-1′L), 91.3 (C-1), 83.3, 83.1 (2× C-1L), 76.7 (2× C-5L), 76.2, 76.0 (2× C-4L), 73.7 (2× C-3L), 70.9, 70.6, 70.5, 70.4, 70.1, 70.0, 69.7, 69.1, 68.5 (C-2, C-3, C-4, C-2L, C-2′L, C-3′L, C-5′L, 2× *C*H$_2$O), 66.6 (2× C-4′L), 63.6, 62.1, 61.9, 60.7, 60.6, 60.3, 59.8 (C-5, 2× C-6L, 2× C-6′L, 2× *C*H$_2$-triazole), 49.0 (C-6), 29.6, 29.5 (2× *C*H$_2$S), 21.0, 20.9, 20.8, 20.7, 20.6, 20.5, 20.4, 20.3, 20.2, 20.1 (*C*H$_3$CO) ppm. $C_{144}H_{196}N_6O_{87}S_4$ (3531.36): calcd. C 48.98, H 5.59, N 2.38, S 3.63; found C 48.69, H 5.84, N 2.41, S 3.69. HRMS (ESI): calcd. for $C_{144}H_{196}N_6O_{87}S_4$ [M + 2Na]$^{2+}$ 1787.4882; found 1788.4860.

Compound 18: Yield 45%. $[\alpha]_D^{20}$ = –16.1 (c = 0.2 in $CHCl_3$). R_f = 0.29 (EtOAc/MeOH, 9:1). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 6.01 (d, $J_{1,2} = 9.3$ Hz, 1 H, 1-H), 5.84 (t, $J_{1,2} = J_{2,3} = 9.4$ Hz, 1 H, 2-H), 5.42 (t, $J_{2,3} = J_{3,4} = 9.5$ Hz, 1 H, 3-H), 5.27 (t, $J_{3,4} = J_{4,5} = 9.8$ Hz, 1 H, 4-H), 4.82, 4.72 (2s, 4 H, 2× CH_2-triazole), 4.32 (dd, $J_{5,6a} = 4.9$, $J_{6a,6b} = 12.6$ Hz, 1 H, 6-Ha), 4.17 (dd, $J_{5,6b} = 2.2$, $J_{6a,6b} = 12.6$ Hz, 1 H, 6-Hb), 4.02 (ddd, $J_{5,6a} = 2.2$, $J_{5,6b} = 4.9$, $J_{4,5} = 10.1$ Hz, 1 H, 5-H), 3.80–3.62 (m, 8 H, 2× CH_2O–, 2× CH_2OH), 3.21, 2.88 (2 br. s, 2 H, 2× O*H*), 2.09, 2.07, 2.03, 1.88 (4s, 12 H, 4 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.4, 170.1, 169.3, 169.1 (–*C*OCH$_3$), 144.5, 132.8 (*C*-triazole), 85.9 (C-1), 75.0 (C-5), 72.9 (C-3), 72.9, 72.6 (2× *C*H$_2$O–), 70.4 (C-2), 67.6 (C-4), 63.8, 60.4 (2× *C*H$_2$-triazole), 61.6, 61.6, 61.5 (C-6, 2× *C*H$_2$OH), 20.7, 20.5, 20.4, 20.3 (*C*H$_3$CO) ppm. $C_{22}H_{33}N_3O_{13}$ (547.52): calcd. C 48.26, H 6.08, N 7.67; found C 48.17, H 5.94, N 7.48. HRMS (ESI): calcd. for $C_{22}H_{33}N_3O_{13}$ [M + H]$^+$ 548.2086; found 548.2086.

Compound 18b: Tetrabromomethane (120 mg, 0.361 mmol) was added to a solution of **18** (50 mg, 0.091 mmol) in dichloromethane (1 mL) under an argon atmosphere. The reaction mixture was cooled to 0 °C and a solution of triphenylphosphane (95 mg, 0.362 mmol) in dichloromethane (1 mL) was added dropwise. After stirring for 3 h, the solvent was evaporated and the residue was purified by flash chromatography (hexane/EtOAc, 1:1.5) to obtain pure **18b** (50 mg, 81%). $[\alpha]_D^{20}$ = +8.0 (c = 0.5 in $CHCl_3$). R_f = 0.60 (hexane/EtOAc, 1:2). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 6.05 (d, $J_{1,2} = 9.3$ Hz, 1 H, 1-H), 5.94 (t, $J_{1,2} = J_{2,3} = 9.4$ Hz, 1 H, 2-H), 5.44 (t, $J_{2,3} = J_{3,4} = 9.5$ Hz, 1 H, 3-H), 5.31 (t, $J_{3,4} = J_{4,5} = 9.8$ Hz, 1 H, 4-H), 4.86, 4.75 (2s, 4 H, 2× CH_2-triazole), 4.30 (dd, $J_{5,6a}$ =

4.9, $J_{6a,6b}$ = 12.6 Hz, 1 H, 6-Ha), 4.22 (dd, $J_{5,6b}$ = 2.2, $J_{6a,6b}$ = 12.6 Hz, 1 H, 6-Hb), 4.05 (ddd, $J_{5,6a}$ = 2.2, $J_{5,6b}$ = 4.8, $J_{4,5}$ = 10.1 Hz, 1 H, 5-H), 3.92–3.82 (m, 4 H, 2× CH_2O–), 3.55, 3.51 (2t, $J_{CH2Br,CH2O}$ = 5.8 Hz, 4 H, 2× CH_2Br), 2.11, 2.09, 2.05, 1.90 (4s, 12 H, 4 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.5, 170.2, 169.3, 168.6 (–COCH$_3$), 143.8, 132.6 (*C*-triazole), 85.3 (C-1), 74.9 (C-5), 73.2 (C-3), 70.4, 70.2 (2× CH$_2$O–), 70.0 (C-2), 67.6 (C-4), 63.9, 60.2 (2× CH$_2$-triazole), 61.6 (C-6), 30.7, 30.3 (2× CH$_2$Br), 20.8, 20.6, 20.5, 20.4 (CH$_3$CO) ppm. $C_{22}H_{31}Br_2N_3O_{11}$ (673.31): calcd. C 39.24, H 4.64, N 6.24; found C 39.17, H 6.44, N 6.28. HRMS (ESI): calcd. for $C_{22}H_{31}Br_2N_3O_{11}$ [M + Na]$^+$ 694.0218; found 694.0211.

Compound 19: Compound **18b** (130 mg, 0.193 mmol) and sodium azide (50 mg, 0.769 mmol) were dissolved in DMF (4 mL), and the mixture was stirred at 70 °C for 2 h. The solvent was evaporated and the residue was purified by column chromatography (hexane/EtOAc, 1.5:1) to give **19** (103 mg, 89%). $[\alpha]_D^{20}$ = +5.6 (*c* = 0.6 in $CHCl_3$). R_f = 0.50 (hexane/EtOAc, 1:2). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 6.00 (d, $J_{1,2}$ = 9.3 Hz, 1 H, 1-H), 5.93 (t, $J_{1,2}$ = $J_{2,3}$ = 9.3 Hz, 1 H, 2-H), 5.42 (t, $J_{2,3}$ = $J_{3,4}$ = 9.4 Hz, 1 H, 3-H), 5.33 (t, $J_{3,4}$ = $J_{4,5}$ = 9.8 Hz, 1 H, 4-H), 4.83, 4.71 (2s, 4 H, 2× CH_2-triazole), 4.32 (dd, $J_{5,6a}$ = 4.9, $J_{6a,6b}$ = 12.6 Hz, 1 H, 6-Ha), 4.19 (dd, $J_{5,6b}$ = 2.2, $J_{6a,6b}$ = 12.6 Hz, 1 H, 6-Hb), 4.01 (ddd, $J_{5,6a}$ = 2.3, $J_{5,6b}$ = 4.9, $J_{4,5}$ = 10.0 Hz, 1 H, 5-H), 3.75–3.62 (m, 4 H, 2× CH_2O–), 3.50, 3.40 (2m, 4 H, 2× CH_2N$_3$), 2.09, 2.07, 2.04, 1.88 (4s, 12 H, 4 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.2, 170.1, 169.9, 169.8 (–COCH$_3$), 143.9, 132.4 (*C*-triazole), 85.7 (C-1), 75.0 (C-5), 73.1 (C-3), 70.1 (C-2), 69.3, 69.0 (2× CH$_2$O–), 67.6 (C-4), 63.8, 60.2 (2× CH$_2$-triazole), 61.6 (C-6), 50.8, 50.7 (2× CH$_2$N$_3$), 20.8, 20.7, 20.6, 20.3 (CH$_3$CO) ppm. $C_{22}H_{31}N_9O_{11}$ (597.54): calcd. C 44.22, H 5.23, N 21.10; found C 44.17, H 5.44, N 21.28. HRMS (ESI): calcd. for $C_{22}H_{31}N_9O_{11}$ [M + Na]$^+$ 620.2035; found 620.2017.

Compound 20: The RuAAC reaction applied to **10** and **17** afforded **20**. Yield 35%. $[\alpha]_D^{20}$ = –30.2 (*c* = 0.6 in $CHCl_3$). R_f = 0.15 (EtOAc/MeOH, 4:1). ^{1}H NMR (500 MHz, $CDCl_3$ + [D$_6$]DMSO): δ = 5.05 (t, $J_{2,3}$ = $J_{3,4}$ = 9.5 Hz, 1 H, 3-H), 4.70 (dd, $J_{1,2}$ = 3.7, $J_{2,3}$ = 9.2 Hz, 1 H, 2-H), 4.65 (t, $J_{3,4}$ = $J_{4,5}$ = 9.3 Hz, 1 H, 4-H), 4.30 (m, 5 H, 1-H, 2× CH_2-triazole), 4.16–4.03 (m, 3 H, 6-Ha, 6-Hb, 5-H), 3.29 (m, 4 H, 2× CH_2OH), 3.16 (m, 4 H, 2× CH_2O–), 2.89–2.63 (br. s, 2 H, OH + [D$_6$]DMSO), 1.80, 1.72, 1.64 (3s, 9 H, 3 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$ + [D$_6$]DMSO): δ = 174.5, 174.4, 174.2 (–COCH$_3$), 147.8, 136.2 (*C*-triazole), 95.9 (C-1), 76.8, 76.5 (2× CH$_2$O–), 74.9, 74.8 (C-3, C-4), 73.9 (C-5), 73.1 (C-2), 68.4, 64.8 (2× CH$_2$-triazole), 65.8, 65.7 (2× CH$_2$OH), 53.5 (C-6), 25.8, 25.5, 25.4 (CH$_3$CO) ppm. $C_{40}H_{60}N_6O_{23}$ (992.94): calcd. C 48.38, H 6.09, N 8.46; found C 48.33, H 5.91, N 8.31. HRMS (ESI): calcd. for $C_{40}H_{60}N_6O_{23}$ [M + Na]$^+$ 1015.3602; found 1015.3588.

Compound 21: Tetrabromomethane (254 mg, 0.766 mmol) was added to a solution of **20** (95 mg, 0.096 mmol) in dichloromethane (2 mL) under an argon atmosphere. The reaction mixture was cooled to 0 °C and a solution of triphenylphosphane (200 mg, 0.766 mmol) in dichloromethane (1 mL) was added dropwise. After stirring for 4 h the solvent was evaporated and the residue was dissolved DMF (3 mL). Sodium azide (50 mg, 0.766 mmol) was added to the solution and the reaction mixture was stirred at 70 °C for 5 h. The mixture was concentrated and the residue was purified by flash chromatography (hexane/EtOAc, 1.5:8.5) to obtain pure **21** (72 mg, 69%). $[\alpha]_D^{20}$ = –11.4 (*c* = 0.8 in $CHCl_3$). R_f = 0.32 (hexane/EtOAc, 1:4). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.42 (t, $J_{2,3}$ = $J_{3,4}$ = 9.7 Hz, 1 H, 3-H), 5.01 (dd, $J_{1,2}$ = 2.6, $J_{2,3}$ = 10.1 Hz, 1 H, 2-H), 4.90 (t, $J_{3,4}$ = $J_{4,5}$ = 9.7 Hz, 1 H, 4-H), 4.60 (m, 5 H, 1-H, 2× CH_2-triazole), 4.42 (m, 1 H, 5-H), 4.36 (m, 2 H, 6-Ha, 6-Hb), 3.75–3.62 (m, 4 H, 2× CH_2O–), 3.32 (m, 4 H, 2× CH_2N$_3$), 2.14, 2.03, 1.95 (3s, 9 H, 3 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 169.9, 169.8, 169.6 (–COCH$_3$), 143.9, 132.4 (*C*-triazole), 91.4 (C-1), 70.4 (C-4), 69.9 (C-3), 69.4, 68.9 (2× CH$_2$O), 69.1 (C-5), 68.5 (C-2), 63.8, 60.2 (2× CH$_2$-triazole), 50.7, 50.6 (2× CH$_2$N$_3$), 49.2 (C-6), 20.8, 20.7, 20.6 (CH$_3$CO) ppm. $C_{40}H_{56}N_{18}O_{19}$ (1092.99): calcd. C 43.96, H 5.16, N 23.07; found C 43.87, H 5.23, N 22.88. HRMS (ESI): calcd. for $C_{40}H_{56}N_{18}O_{19}$ [M + Na]$^+$ 1115.3867; found 1115.3870.

Compound 22: Yield 81%. $[\alpha]_D^{20}$ = –22.2 (*c* = 0.4 in $CHCl_3$). R_f = 0.29 (EtOAc). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.80 (d, $J_{1,2}$ = 9.6 Hz, 1 H, 1-H), 5.42 (m, 4 H, 4× 4-HG), 5.37 (t, $J_{1,2}$ = $J_{2,3}$ = 9.4 Hz, 1 H, 2-H), 5.24 (t, $J_{2,3}$ = $J_{3,4}$ = 9.9 Hz, 1 H, 3-H), 5.17 (4t, $J_{1G,2G}$ = $J_{2G,3G}$ = 10.0 Hz, 4 H, 4× H-2G), 5.06 (m, 4 H, 4× H-3G), 4.66 (m, 12 H, 6× CH_2-triazole), 4.55 (m, 8 H, 4× H-1G, 2× CH_2-N), 4.28 (dd, $J_{5,6a}$ = 4.4, $J_{6a,6b}$ = 12.7 Hz, 1 H, 6-Ha), 4.16–4.04 (m, 10 H, 5-H, 6-Hb, 4× H-6aG, 4× H-6bG), 4.00–3.90 (m, 9 H, 4-H, 4× H-5G, 2× CH_2O), 3.77–3.62 (m, 8 H, 4× CH_2O–), 2.95, 2.83 (2m, 8 H, 4× CH_2S), 2.15 (2×), 2.14 (2×), 2.07, 2.05 (2×), 2.03 (4×), 2.02 (4×), 1.97 (4×), 1.84 (8s, 60 H, 20 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 170.4, 170.3 (3×), 170.2 (2×), 170.1 (2×), 170.0 (2×), 169.9, 169.6, 169.5, 169.2, 168.6 (–COCH$_3$), 143.4, 143.2, 142.9 (3× C-4 triazole), 132.3, 132.2, 132.1 (3× C-5 triazole), 84.7 (C-1), 84.0 (2×), 83.8, 83.7 (4× C-1G), 74.5 (C-5), 74.4 (4× C-5G), 73.1 (C-2), 71.8, 71.7 (2×), 71.6 (C-3, 4× C-3G), 69.9 (2×), 69.8, 69.7 (2×), 68.6, 68.5 (C-4, 6× CH$_2$O–), 67.5, 67.3 (2×), 67.2 (3×), 67.1 (4× C-2G, 4× C-4G), 63.8, 63.7, 63.5, 60.0, 59.9, 59.8 (6× CH$_2$-triazole), 61.4, 61.3 (2×), 61.2, 61.1 (C-6, 4× C-6G), 48.4, 48.1 (2× CH$_2$N), 29.6 (2×), 29.5, 29.4 (4× CH$_2$S), 20.8 (3×), 20.7 (4×), 20.6 (3×), 20.4, 20.3 (CH$_3$CO) ppm. $C_{94}H_{131}N_9O_{51}S_4$ (2331.34): calcd. C 48.43, H 5.66, N 5.41, S 5.50; found C 48.17, H 5.44, N 5.28, S 5.54. HRMS (ESI): calcd. for $C_{94}H_{131}N_9O_{51}S_4$ [M + H]$^+$ 2330.6890; found 2330.6887.

Compound 24: Yield 64%. $[\alpha]_D^{20}$ = –68.9 (*c* = 0.3 in $CHCl_3$). R_f = 0.47 (EtOAc). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 5.80 (d, $J_{1,2}$ = 9.6 Hz, 1 H, 1-H), 5.37 (t, $J_{1,2}$ = $J_{2,3}$ = 9.4 Hz, 1 H, 2-H), 5.33 (m, 4 H, 4× H-4′L), 5.24 (t, $J_{2,3}$ = $J_{3,4}$ = 9.9 Hz, 1 H, 3-H), 5.19 (m, 4 H, 4× H-3L), 5.09 (m, 4 H, 4× H-2′L), 4.95 (m, 4 H, 4× H-3′L), 4.87 (m, 4 H, 4× H-2L), 4.70–4.45 (m, 24 H, 4× H-1L, 4× H-1′L, 2× CH$_2$N, 6× CH$_2$-triazole), 4.28 (dd, $J_{5,6a}$ = 4.4, $J_{6a,6b}$ = 12.7 Hz, 1 H, 6-Ha), 4.17–4.03 (m, 18 H, 5-H, 6-Hb, 4× H-6aL, 4× H-6bL, 4× H-6a′L, 4× H-6b′L), 4.00–3.90 (m, 8 H, 4-H, 4× H-5′L, 2× CH_2O), 3.80 (m, 4 H, 4× H-4L), 3.77–3.62 (m, 5 H, 4-H, 2× CH_2O), 3.60 (m, 8 H, 4× H-5L, 4× CH_2O), 2.95–2.73 (m, 8 H, 4× CH_2S), 2.15 (4×), 2.14 (2×), 2.09, 2.08, 2.07, 2.06, 2.05 (2×), 2.03 (4×), 2.02 (4×), 1.97 (4×), 1.94, 1.84, 1.82 (13s, 96 H; 32 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 171.1, 170.5, 170.4, 170.3 (3×), 170.2 (2×), 170.1 (2×), 170.0 (2×), 169.9, 169.8, 169.7, 169.6, 169.5, 169.3, 169.2, 169.1, 168.6 (–COCH$_3$), 143.4, 143.1, 142.8 (3× C-4 triazole), 132.4, 132.3, 132.2 (3× C-5 triazole), 101.0 (4× C-1′L), 84.6 (C-1), 83.4 (2×), 83.2, 83.1 (4× C-1L), 76.8, 76.7 (4× C-5L), 76.1, 76.0 (4× C-4L), 74.5 (C-5), 73.7 (4× C-3L), 73.1 (C-2), 70.9, 70.8, 70.7, 70.6, 70.3, 70.2, 69.9, 69.8, 69.7, 60.6, 69.5 (C-3, 4× C-2L, 4× C-3′L, 4× C-5′L, 6× CH$_2$O–), 69.1 (4× C-2′L), 68.6 (C-4), 66.6 (4× C-4′L), 63.7, 63.6, 63.5, 62.1, 62.0, 61.4, 60.7, 60.6, 60.3, 59.9, 59.8 (C-6, 4× C-6L, 4× C-6′L, 6× CH$_2$-triazole), 48.4, 48.1 (2× CH$_2$N), 29.7 (2×), 29.5, 29.4 (4× CH$_2$S), 20.9, 20.8 (3×), 20.7 (4×), 20.6 (3×), 20.5, 20.4, 20.3 (CH$_3$CO) ppm. $C_{142}H_{195}N_9O_{83}S_4$ (3484.35): calcd. C 48.95, H 5.64, N 3.62, S 3.68; found C 49.27, H 5.88, N

3.58, S 3.55. HRMS (ESI): calcd. for $C_{142}H_{195}N_9O_{83}S_4$ $[M + Na]^+$ 3505.0090; found 3505.0100.

Compound 26: Yield 68%. $[\alpha]_D^{20}$ = –55.7 (*c* = 0.4 in $CHCl_3$). R_f = 0.46 (EtOAc/MeOH, 98:2). ^1H NMR (500 MHz, $CDCl_3$): δ = 5.40 (t, $J_{2,3} = J_{3,4}$ = 9.6 Hz, 1 H, 3-H), 5.31 (m, 4 H, 4× H-4′L), 5.16 (m, 4 H, 4× H-3L), 5.04 (m, 4 H, 4× H-2′L), 4.95 (m, 5 H, 2-H, 4× H-3′L), 4.84 (m, 5 H, 4-H, 4× H-2L), 4.64 (d, $J_{1,2}$ = 3.1 Hz, 1 H, 1-H), 4.63–4.33 (m, 25 H, 5-H, 4× H-1L, 4× H-1′L, 6× CH_2-triazole, 2× CH_2-N), 4.23 (m, 2 H, 6-Ha, 6-Hb), 4.12–4.00 (m, 16 H, 4× H-6aL, 4× H-6bL, 4× H-6′aL, 4× H-6′bL), 3.87 (m, 4 H, 4× H-5′L), 3.76 (m, 4 H, 4× H-4L), 3.60 (m, 16 H, 4× H-5L, 6× CH_2O), 2.83, 2.73 (2m, 8 H, 4× CH_2S), 2.12, 2.11, 2.07, 2.06, 2.05, 2.04, 2.03, 2.02, 2.01, 2.00, 1.99, 1.98, 1.97, 1.91 (14s, 93 H; 31 CH_3CO) ppm. ^{13}C NMR (125 MHz, $CDCl_3$): δ = 171.1, 170.2, 170.1, 170.0, 169.9, 169.8, 169.7, 169.6, 169.5, 169.4, 169.0, 168.9 (–*C*OCH$_3$), 143.0, 142.8, 142.4 (3× C-4 triazole), 132.5, 132.2, 132.1 (3× C-5 triazole), 101.0 (4× C-1′L), 91.3 (C-1), 83.4 (2×), 83.2, 83.1 (4× C-1L), 76.7 (4× C-5L), 76.1 (4× C-4L), 73.7, 73.6 (4× C-3L), 70.9, 70.6, 70.3, 70.2, 70.0, 69.7, 69.6, 69.1, 68.2 (C-2 to C-5, 4× C-2L, 4× C-2′L, 4× C-3′L, 4× C-5′L, 6× *C*H$_2$O), 66.6 (4× C-4′L), 63.6, 63.5, 62.1, 62.0, 60.7, 60.6, 60.3, 59.9, 59.8, 59.7 (4× C-6L, 4× C-6′L, 6× *C*H$_2$-triazole), 48.8 (C-6), 48.3, 48.0 (2× *C*H$_2$N), 29.7, 29.5 (4× *C*H$_2$S), 20.9, 20.8, 20.7, 20.6, 20.5, 20.4, 20.3 (*C*H$_3$CO) ppm. $C_{280}H_{384}N_{18}O_{163}S_8$ (6866.62): calcd. C 48.98, H 5.64, N 3.67, S 3.74; found C 49.17, H 5.34, N 3.31, S 3.44. HRMS (ESI): calcd. for $C_{280}H_{384}N_{18}O_{163}S_8$ $[M + 2Na]^{2+}$ 3453.9931; found 3453.9995.

General Procedure for *O*-Deacetylation: Exemplified for the synthesis of compound **9**. Compound **8** (0.10 mmol) was suspended in a mixture of MeOH/Et_3N/H_2O (4:1:5; 10 mL) and stirred at room temperature. The solid was progressively dissolved and after 4–6 h TLC (EtOAc or EtOAc/MeOH, 9:1) showed complete consumption of the starting material. The solution was concentrated and the residue was dissolved in water (1 mL) and then passed through a column filled with Dowex M$R\bar{3}$C mixed bed ion-exchange resin. The eluant was concentrated and purified by filtration through an octadecyl C18 minicolumn. Evaporation of the solvent afforded the free product, which showed a single spot by TLC (*n*-BuOH/EtOH/H_2O, 2.5:1:1).

Compound 9: Yield 95%. $[\alpha]_D^{20}$ = –50.6 (*c* = 0.5 in H_2O). R_f = 0.31 (*n*BuOH/EtOH/H_2O, 2.5:1:1). ^1H NMR (500 MHz, D_2O): δ = 5.63 (d, $J_{1,2}$ = 9.2 Hz, 1 H, 1-H), 4.70, 4.60 (2s, 4 H, CH_2-triazole), 4.34 (d, $J_{1',2'} = J_{1'',2''}$ = 9.7 Hz, 2 H, 1′-H, 1′′-H), 4.13 (t, $J_{1,2} = J_{2,3}$ = 9.3 Hz, 1 H, 2-H), 3.81 (m, 2 H, 4′-H, 4′′-H), 3.77 (dd, $J_{5,6a}$ = 1.0, $J_{6a,6b}$ = 11.7 Hz, 1 H, 6-Ha), 3.69–3.46 (m, 16 H, 3-H, 4-H, 5-H, 6-Hb, 3′-H, 3′′-H, 5′-H, 5′′-H, 6′-Ha, 6′-Hb, 6′′-Ha, 6′′-Hb, 2× CH_2O), 3.40 (2t, $J_{1',2'} = J_{2',3'} = J_{2'',3''}$ = 9.6 Hz, 2 H, 2′-H, 2′′-H), 2.91–2.75 (m, 4 H, 2× CH_2S) ppm. ^{13}C NMR (125 MHz, D_2O): δ = 143.0, 134.1 (*C*-triazole), 85.7 (2×) (C-1′, C-1′′), 85.6 (C-1), 79.0 (2×) (C-5′, C-5′′), 78.9 (C-5), 76.0 (C-3), 73.9 (2×) (C-3′, C-3′′), 71.6 (C-2), 69.6 (2×) (C-2′, C-2′′), 69.8, 69.5, 69.0 (C-4, 2× *C*H$_2$O), 67.2 (× 2) (C-4′, C-4′′), 62.0, 58.8 (2× *C*H$_2$–C≡C), 61.0 (2×) (C-6′, C-6′′), 60.5 (C-6), 29.4, 29.3 (2× *C*H$_2$S) ppm. $C_{26}H_{45}N_3O_{17}S_2$ (735.77): calcd. C 42.44, H 6.16, N 5.71, S 8.72; found C 42.66, H 6.24, N 5.61, S 8.44. HRMS (ESI): calcd. for $C_{26}H_{45}N_3O_{17}S_2$ $[M + H]^+$ 736.2263; found 736.2266.

Compound 12: Yield 88%. $[\alpha]_D^{20}$ = +33.6 (*c* = 0.5 in H_2O). R_f = 0.10 (*n*BuOH/EtOH/H_2O, 2.5:1:1, after two successive developments). ^1H NMR (500 MHz, D_2O): δ = 4.73 (dd, $J_{5,6a}$ = 2.1, $J_{6a,6b}$ = 14.8 Hz, 1 H, 6-Ha), 4.63, 4.57 (2s, 4 H, 2× CH_2-triazole), 4.47 (dd, $J_{5,6b}$ = 8.5, $J_{6a,6b}$ = 14.8 Hz, 1 H, 6-Hb), 4.34, 4.32 (2d, $J_{1G,2G}$ = 9.8 Hz, 2 H, 2× H-1G), 4.27 (d, $J_{1,2}$ = 3.7 Hz, 1 H, 1-H), 3.88 (ddd, $J_{5,6a}$ = 2.0, $J_{5,6b}$ = 8.5, $J_{4,5}$ = 10.0 Hz, 1 H, 5-H), 3.82 (m, 2 H, 2× H-4G), 3.65–3.45 (m, 13 H, 3-H, 2× H-3G, 2× H-5G, 2× H-6aG, 2× H-6bG, 2× CH_2O), 3.41 (2t, $J_{1G,2G} = J_{2G,3G}$ = 9.6 Hz, 2 H, 2× H-2G), 3.32 (dd, $J_{1,2}$ = 3.8, $J_{2,3}$ = 9.9 Hz, 1 H, 2-H), 3.19 (t, $J_{3,4} = J_{4,5}$ = 9.5 Hz, 1 H, 4-H), 2.85, 2.76 (2m, 4 H, 2× CH_2S) ppm. ^{13}C NMR (125 MHz, D_2O): δ = 143.7, 132.2 (*C*-triazole), 92.8 (C-1), 85.9, 85.8 (2× C-1G), 78.9 (2×) (2× C-5G), 73.9 (2×) (2× C-3G), 72.6 (C-3), 71.0 (C-4), 70.7 (C-5), 70.7 (C-2), 69.7, 69.4 (2× *C*H$_2$O), 69.6 (2×) (2× C-2G), 68.8, 68.7 (2× C-4G), 62.2, 59.4 (2× *C*H$_2$-triazole), 61.0 (2×) (2× C-6G), 49.5 (C-6), 29.3 (2×) (2× *C*H$_2$S) ppm. $C_{52}H_{88}N_6O_{33}S_4 \cdot 0.5H_2O$: calcd. C 42.70, H 6.13, N 5.75, S 8.77; found C 42.66, H 6.18, N 5.69, S 8.76. HRMS (ESI): calcd. for $C_{52}H_{88}N_6O_{33}S_4$ $[M + 2H]^{2+}$ 727.2210; found 727.2235.

Compound 14: Yield 90%. $[\alpha]_D^{20}$ = +10.6 (*c* = 0.5 in H_2O). R_f = 0.23 (*n*BuOH/EtOH/H_2O, 2.5:1:1, after two successive developments). ^1H NMR (500 MHz, D_2O): δ = 5.70 (d, $J_{1,2}$ = 9.2 Hz, 1 H, 1-H), 4.79, 4.68 (2s, 4 H, CH_2-triazole), 4.50 (d, $J_{1L,2L}$ = 9.8 Hz, 2 H, 2× H-1L), 4.37 (d, $J_{1'L,2'L}$ = 9.8 Hz, 2 H, 2× H-1′L), 4.20 (t, $J_{1,2} = J_{2,3}$ = 9.2 Hz, 1 H, 2-H), 3.90–3.45 (m, 31 H, 3-H to 5-H, 6-Ha, 6-Hb, 2× H-3L to H-5L, 2× H-6aL, 2× H-6bL, 2× H-2′L to H-5′L, 2× H-6′aL, 2× H-6′bL, 2× CH_2O), 3.30 (t, $J_{1L,2L} = J_{2L,3L}$ = 9.8 Hz, 2 H, 2× H-2L), 2.99–2.83 (m, 4 H, 2× CH_2S) ppm. ^{13}C NMR (125 MHz, D_2O): δ = 143.1, 134.1 (*C*-triazole), 102.9 (2× C-1′L), 85.8 (C-1), 85.3, 85.2 (2× C-1L), 79.0, 78.6 (2×), 78.1 (2×), 76.0, 75.8, 75.7, 75.3 (2×), 72.5 (2×), 72.1, 72.0, 71.7, 71.0 (2×), 69.7, 69.6, 69.1, 68.6 (2×) (C-2 to C-5, 2× C-2L to C-5L, 2× C-2′L to C-5′L, 2× *C*H$_2$O), 62.1, 58.8 (2× *C*H$_2$-triazole), 61.0 (2×), 60.6, 60.2 (2×) (C-6, 2× C-6L, 2× C-6′L), 29.3 (2×) (2× *C*H$_2$S) ppm. $C_{38}H_{65}N_3O_{27}S_2$ (1060.06): calcd. C 43.05, H 6.18, N 3.96, S 6.05; found C 42.96, H 6.24, N 3.61, S 6.24. HRMS (ESI): calcd. for $C_{38}H_{65}N_3O_{27}S_2$ $[M + Na]^+$ 1082.3139; found 1082.3158.

Compound 16: Yield 90%. $[\alpha]_D^{20}$ = +19.0 (*c* = 0.2 in H_2O). R_f = 0.12 (*n*BuOH/EtOH/H_2O, 2.5:1:1, after two successive developments). ^1H NMR (500 MHz, D_2O): δ = 4.79 (dd, $J_{5,6a}$ = 2.1, $J_{6a,6b}$ = 14.5 Hz, 1 H, 6-Ha), 4.74, 4.66 (2s, 4 H, 2× CH_2-triazole), 4.56 (dd, $J_{5,6b}$ = 8.3, $J_{6a,6b}$ = 14.5 Hz, 1 H, 6-Hb), 4.50, 4.48 (2d, $J_{1L,2L}$ = 9.9 Hz, 2 H, 2× H-1L), 4.38 (2d, $J_{1'L,2'L}$ = 9.9 Hz, 2 H, 2× H-1′L), 4.34 (d, $J_{1,2}$ = 3.7 Hz, 1 H, 1-H), 3.95 (ddd, $J_{5,6a}$ = 2.0, $J_{5,6b}$ = 8.5, $J_{4,5}$ = 10.0 Hz, 1 H, 5-H), 3.90–3.45 (m, 27 H, 3-H, 2× H-3L, 2× H-4L, 2× H-5L, 2× H-6aL, 2× H-6bL, 2× H-2′L, 2× H-3′L, 2× H-4′L, 2× H-5′L, 2× H-6′aL, 2× H-6′bL, 2× CH_2O), 3.39 (dd, $J_{1,2}$ = 3.7, $J_{2,3}$ = 9.9 Hz, 1 H, 2-H), 3.30 (2d, $J_{1L,2L} = J_{2L,3L}$ = 9.6 Hz, 2 H, 2× H-2L), 3.25 (t, $J_{3,4} = J_{4,5}$ = 9.5 Hz, 1 H, 4-H), 2.92, 2.85 (2m, 4 H, 2× CH_2S) ppm. ^{13}C NMR (125 MHz, D_2O): δ = 142.8, 133.8 (*C*-triazole), 102.9 (2× C-1′L), 92.7 (C-1), 85.3, 85.2 (2× C-1L), 78.7, 78.2, 78.1, 75.7, 75.4, 72.5, 72.0, 71.9, 71.1, 71.0, 70.7, 70.6, 69.7, 69.4, 68.6 (C-2 to C-5, 2× C-2L to C-5L, 2× C-2′L to C-5′L, 2× *C*H$_2$O), 62.2, 59.4 (2× *C*H$_2$-triazole), 61.0, 60.2 (2× C-6L, 2× C-6′L), 49.5 (C-6), 29.3 (2× *C*H$_2$S) ppm. $C_{76}H_{128}N_6O_{53}S_4$ (2102.10): calcd. C 43.42, H 6.14, N 4.00, S 6.10; found C 43.66, H 6.18, N 3.79, S 5.96. HRMS (ESI): calcd. for $C_{76}H_{128}N_6O_{53}S_4$ $[M + 2H]^{2+}$ 1051.3267; found 1051.3238.

Compound 23: Yield 100%. $[\alpha]_D^{20}$ = –56.5 (*c* = 0.5 in H_2O). R_f = 0.73 (*n*BuOH/EtOH/H_2O, 2.5:1:1). ^1H NMR (500 MHz, D_2O): δ = 5.43 (d, $J_{1,2}$ = 9.1 Hz, 1 H, 1-H), 4.65–4.49 (m, 16 H, 2× CH_2N, 6× CH_2-triazole), 4.55 (4d, $J_{1G,2G}$ = 9.7 Hz, 4 H, 4× H-1G), 4.08 (t, $J_{1,2} = J_{2,3}$ = 9.2 Hz, 1 H, 2-H), 3.88 (m, 4 H, 4× H-4G), 3.69 (dd, $J_{5,6a}$ = 1.1, $J_{6a,6b}$ = 11.7 Hz, 1 H, 6^a-H-6), 3.75–3.50 (m, 32 H, 3-H, 4-H, 5-H, 6-Hb, 4× H-3G, 4× H-5G, 4× H-6aG, 4× H-6bG, 6× CH_2O), 3.47 (m, 4 H, 4× H-2G), 2.90 (m, 8 H, 4×

CH_2S) ppm. ^{13}C NMR (125 MHz, D_2O): δ = 142.9, 142.8, 142.7 (3× C-4 triazole), 133.9, 133.4, 133.3 (3× C-5 triazole), 85.9 (4× C-1G), 85.8 (C-1), 78.9 (C-5, 4× C-5G), 76.0 (C-3), 73.9 (4× C-3G), 71.6 (C-2), 69.7, 69.6, 69.5, 69.0, 68.9, 68.8, 68.7, 68.4, 68.0 (C-4, 6× CH_2O–, 4× C-2G, 4× C-4G), 62.3, 62.2, 62.1, 59.2, 59.1, 59.0 (6× CH_2-triazole), 61.0 (4× C-6G), 60.5 (C-6), 48.9, 48.6 (2× CH_2N), 29.3, 29.2 (4× CH_2S) ppm. $C_{54}H_{91}N_9O_{31}S_4$ (1490.59): calcd. C 43.51, H 6.15, N 8.46, S 8.60; found C 43.37, H 6.44, N 8.28, S 8.94. HRMS (ESI): calcd. for $C_{54}H_{91}N_9O_{31}S_4$ $[M + Na]^+$ 1512.4602; found 1512.4535.

Compound 25: Yield 90%. $[\alpha]_D^{20}$ = –45.6 (c = 0.5 in H_2O). R_f = 0.10 (nBuOH/EtOH/H_2O, 2.5:1:1, after three successive developments). ^1H NMR (500 MHz, D_2O): δ = 5.39 (d, $J_{1,2}$ = 9.2 Hz, 1 H, 1-H), 4.66, 4.45 (2m, 12 H, 6× CH_2-triazole), 4.52 (m, 4 H, 2× CH_2N), 4.40 (m, 4 H, 4× H-1L), 4.32 (m, 4 H, 4× H-1′L), 4.04 (t, $J_{1,2}$ = $J_{2,3}$ = 9.2 Hz, 1 H, 2-H), 3.85–3.40 (m, 61 H, 3-H to 5-H, 6-Ha, 6-Hb, 4× H-3L to H-5L, 4× H-6aL, 4× H-6bL, 4× H-2′L to H-5′L, 4× H-6′aL, 4× H-6′bL, 6× CH_2O), 3.24 (m, 4 H, 4× H-2L), 2.99–2.83 (m, 8 H, 4× CH_2S) ppm. ^{13}C NMR (125 MHz, D_2O): δ = 143.1, 142.9 (2×), 134.1, 133.9, 133.4 (*C*-triazole), 102.9 (4× C-1′L), 85.8 (C-1), 85.3 (4× C-1L), 79.0, 78.7, 78.6 (2×), 78.2, 78.1 (2×), 76.0, 75.8, 75.4, 72.6 (2×), 72.1, 71.7, 71.0 (2×), 69.5, 69.0, 68.6 (2×) (C-2 to C-5, 4× C-2L to C-5L, 4× C-2′L to C-5′L, 6× CH_2O), 62.4, 61.1, 60.6, 60.3, 59.2 (6× CH_2-triazole, C-6, 4× C-6L, 4× C-6′L), 29.4 (2×) (4× CH_2S) ppm. $C_{78}H_{131}N_9O_{51}S_4$ (2139.16): calcd. C 43.79, H 6.17, N 5.89, S 6.00; found C 44.06, H 6.44, N 6.21, S 5.90. HRMS (ESI): calcd. for $C_{78}H_{131}N_9O_{51}S_4$ $[M + Na]^+$ 2160.6709; found 2160.6680.

Compound 27: Yield 86%. $[\alpha]_D^{20}$ = –78.6 (c = 0.5 in H_2O). R_f = 0.10 (nBuOH/EtOH/H_2O, 2.5:1:1, after four successive developments). ^1H NMR (500 MHz, D_2O): δ = 4.65–4.60 (m, 18 H, 6× CH_2-triazole, 2× CH_2N, 6-Ha, 6-Hb), 4.50 (m, 5 H, 1-H, 4× H-1L), 4.40 (m, 4 H, 4× H-1′L), 3.95–3.45 (m, 31 H, 2-H, 3-H, 5-H, 4× H-3L to H-5L, 4× H-6aL, 4× H-6bL, 4× H-2′L to H-5′L, 4× H-6′aL, 4× H-6′bL, 6× CH_2O), 3.30 (t, 5 H, 4-H, 4× H-2L), 2.96–2.70 (m, 8 H, 4× CH_2S) ppm. ^{13}C NMR (125 MHz, D_2O): δ = 142.9, 142.8, 142.5, 133.5, 133.3, 133.3 (*C*-triazole), 102.9 (4× C-1′L), 93.1 (C-1), 85.3 (4× C-1L), 79.3, 78.7, 78.2, 76.0, 75.8, 75.4, 72.6, 72.1, 71.7, 71.6, 71.0, 70.6, 69.7, 69.6, 69.5, 69.3, 68.8, 68.7, 68.6, 67.9 (C-2 to C-5, 4× C-2L to C-5L, 4× C-2′L to C-5′L, 6× CH_2O), 62.5, 62.3 (2×), 59.5, 59.2, 59.1 (6× CH_2-triazole), 61.1, 60.5, 60.4, 60.3 (4× C-6L, 4× C-6′L), 49.2, 48.9, 48.6 (2× CH_2N, C-6), 29.4, 29.3, 29.2 (4× CH_2S) ppm. $C_{156}H_{260}N_{18}O_{101}S_8$ (4260.31): calcd. C 43.98, H 6.15, N 5.92, S 6.02; found C 44.36, H 5.90, N 6.21, S 6.34. HRMS (ESI): calcd. for $C_{156}H_{260}N_{18}O_{101}S_8$ $[M + 2Na]^{2+}$ 2151.6656; found 2151.6612.

Isothermal Titration Calorimetry: Peanut agglutinin from *Arachis hypogaea* was purchased from Sigma (lyophilized powder, affinity-purified, agglutination activity < 0.1 μg mL^{-1}). A VP-ITC (Microcal) instrument was used for the titrations at 298 K. Respective concentrations and molar ratios in needle and cell, injection volumes, and time intervals between injections were varied to obtain (1) inflection and saturation about halfway through the experiment, (2) sufficient heat production per injection to allow good peak integration, and (3) sufficient time between the injections to allow a return to equilibrium. A typical titration involved 16 injections at 3-min intervals of 2.5 μL aliquots of ligand solution into the sample cell (200 μL) containing PNA (50 μM). The solutions were prepared by dissolving the ligand in 20 mM phosphate buffer, pH 7.4, and 150 mM NaCl at 298 K. The concentrations used in each ITC experiment are given in Figures S1 and S2. The titration cell was continuously stirred at 400 rev/min. The heats of dilution of the ligands in the buffer were subtracted from the titration data. Fitting was performed using the Origin software to determine the binding stoichiometry (n), association constant, and the enthalpy change (ΔH).

Supporting Information (see footnote on the first page of this article): NMR spectra of the products and intermediates and ITC data.

Acknowledgments

Support for this work by the National Agency for Promotion of Science and Technology (ANPCyT) (project PICT Bicentenario 2010-1080), the National Research Council of Argentina (CONICET) (PIP 0064), the Centre National de la Recherche Scientifique (CNRS), the Ministère Délégué à l'Enseignement Supérieur et à la Recherche and the Conseil Régional de Picardie (France) is gratefully acknowledged. M. L. U. and O. V. are research members of CONICET.

[1] For reviews, see: a) Y. M. Chabre, R. Roy, *Adv. Carbohydr. Chem. Biochem.* **2010**, *63*, 165–393; b) N. Jayaraman, *Chem. Soc. Rev.* **2009**, *38*, 3463–3483.
[2] a) H. Lis, N. Sharon, *Chem. Rev.* **1998**, *98*, 637–674; b) H.-J. Gabius, S. André, H. Kaltner, H.-C. Siebert, *Biochim. Biophys. Acta Gen. Subj.* **2002**, *1572*, 165–177; c) H.-J. Gabius, *Adv. Drug Delivery Rev.* **2004**, *56*, 421–424; d) M. Ambrosi, N. R. Cameron, B. G. Davis, *Org. Biomol. Chem.* **2005**, *3*, 1593–1608.
[3] G. A. Rabinovich, M. A. Toscano, S. S. Jackson, G. R. Vasta, *Curr. Opin. Struct. Biol.* **2007**, *17*, 513–520.
[4] S. L. Flitsch, *Curr. Opin. Chem. Biol.* **2000**, *4*, 619–625.
[5] a) Y. C. Lee, R. T. Lee, *Acc. Chem. Res.* **1995**, *28*, 321–327; b) M. Mammen, S.-K. Choi, G. M. Whitesides, *Angew. Chem.* **1998**, *110*, 2908–2953; *Angew. Chem. Int. Ed.* **1998**, *37*, 2754–2794; c) R. T. Lee, Y. C. Lee, *Glycoconjugate J.* **2000**, *17*, 543–551; d) J. Lundquist, E. J. Toone, *Chem. Rev.* **2002**, *102*, 555–578; e) I. Vrasidas, S. André, P. Valentini, C. Böck, M. Lensch, H. Kaltner, R. M. J. Liskamp, H.-J. Gabius, R. Pieters, *Org. Biomol. Chem.* **2003**, *1*, 803–810.
[6] V. Martos, P. Castreño, J. Valero, J. de Mendoza, *Curr. Opin. Chem. Biol.* **2008**, *12*, 698–706.
[7] a) S. Fletcher, A. D. Hamilton, *J. R. Soc. Interface* **2006**, *3*, 215–233; b) H. Zhou, D. Wang, L. Baldini, E. Ennis, R. Jain, A. Carie, S. M. Sebti, A. D. Hamilton, *Org. Biomol. Chem.* **2006**, *4*, 2376–2386.
[8] a) S. Hershberger, S.-G. Lee, J. Chmielewski, *Curr. Top. Med. Chem.* **2007**, *7*, 928–942; b) D. M. Tagore, K. I. Sprinz, S. Fletcher, J. Jayawickramarajah, A. D. Hamilton, *Angew. Chem.* **2007**, *119*, 227–229; *Angew. Chem. Int. Ed.* **2007**, *46*, 223–225.
[9] a) D. Specker, V. Wittman, *Top. Curr. Chem.* **2007**, *267*, 65–107; b) N. Röckendorf, T. K. Lindhorst, *Top. Curr. Chem.* **2001**, *217*, 201–238.
[10] M.-C. Bowman, T. E. Ballard, C. J. Ackerson, D. L. Feldheim, D. M. Margolis, C. Melander, *J. Am. Chem. Soc.* **2008**, *130*, 6896–6897.
[11] N. Yui, T. Ooya, *Chem. Eur. J.* **2006**, *12*, 6730–6737.
[12] a) S. R. S. Ting, G. Chen, M. H. Stenzel, *Polym. Chem.* **2010**, *1*, 1392–1412; b) V. Ladmiral, G. Mantovani, G. J. Clarkson, S. Cauet, J. L. Irwin, D. M. Haddleton, *J. Am. Chem. Soc.* **2006**, *128*, 4823–4830.
[13] a) C. Ortiz Mellet, J. Defaye, J. M. García Fernández, *Chem. Eur. J.* **2002**, *8*, 1982–1990; b) O. Sperling, A. Fuchs, T. K. Lindhorst, *Org. Biomol. Chem.* **2006**, *4*, 3913–3922; c) S. G. Gouin, E. Vanquelef, J.-M. García Fernández, C. Ortiz Mellet, F.-Y. Dupradeau, J. Kovensky, *J. Org. Chem.* **2007**, *72*, 9032–9045; d) M. Ortega-Muñoz, F. Perez-Balderas, J. Morales-Sanfrutos, F. Hernández-Mateo, J. Isac-García, F. Santoyo-Gonzalez, *Eur. J. Org. Chem.* **2009**, 2454–2473.

[14] a) V. V. Rostovtsev, L. G. Green, V. V. Fokin, K. B. Sharpless, *Angew. Chem.* **2002**, *114*, 2708–2711; *Angew. Chem. Int. Ed.* **2002**, *41*, 2596–2599; b) C. W. Tornoe, C. Christensen, M. Meldal, *J. Org. Chem.* **2002**, *67*, 3057–3064.
[15] S. G. Gouin, J.-M. García Fernández, E. Vanquelef, F.-Y. Dupradeau, E. Salomonsson, H. Leffler, M. Ortega-Muñoz, U. J. Nilsson, J. Kovensky, *ChemBioChem* **2010**, *11*, 1430–1442.
[16] a) S. G. Gouin, A. Wellens, J. Bouckaert, J. Kovensky, *ChemMedChem* **2009**, *5*, 749–755; b) M. Almant, V. Moreau, J. Kovensky, J. Bouckaert, S. G. Gouin, *Chem. Eur. J.* **2011**, *17*, 10029–10038.
[17] A. J. Cagnoni, O. Varela, S. G. Gouin, J. Kovensky, M. L. Uhrig, *J. Org. Chem.* **2011**, *76*, 3064–3077.
[18] a) L. Zhang, X. Chen, P. Xue, H. H. Y. Sun, I. D. Williams, K. B. Sharpless, V. V. Fokin, G. Jia, *J. Am. Chem. Soc.* **2005**, *127*, 15998–15999; b) M. M. Majireck, S. M. Weinreb, *J. Org. Chem.* **2006**, *71*, 8680–8683; c) B. C. Boren, S. Narayan, L. K. Rasmussen, L. Zhang, H. Zhao, Z. Lin, G. Jia, V. V. Fokin, *J. Am. Chem. Soc.* **2008**, *130*, 8923–8930.
[19] a) D. Crich, F. Yang, *Angew. Chem.* **2009**, *121*, 9058–9061; *Angew. Chem. Int. Ed.* **2009**, *48*, 8896–8899; b) A. J. Salmon, M. L. Williams, A. Maresca, C. T. Supuran, S.-A. Poulsen, *Bioorg. Med. Chem. Lett.* **2011**, *21*, 6058–6061.
[20] T. W. Baughman, J. C. Sworen, K. B. Wagener, *Tetrahedron* **2004**, *60*, 10943–10948.
[21] V. E. Manzano, M. L. Uhrig, O. Varela, *J. Org. Chem.* **2008**, *73*, 7224–7235.
[22] a) R. Barenjee, S. C. Mande, V. Ganesh, K. Das, V. Dhanaraj, S. K. Mahanta, K. Suguna, A. Surolia, M. Vijayan, *Proc. Natl. Acad. Sci. USA* **1994**, *91*, 227–231; b) R. Ravishankar, C. J. Thomas, K. Suguna, A. Surolia, M. Vijayan, *Proteins* **2001**, *43*, 260–270; c) M. Ambrosi, N. R. Cameron, B. G. Davis, S. Stolnik, *Org. Biomol. Chem.* **2005**, *3*, 1476–1480.
[23] S.-K. Choi, *Synthetic multivalent molecules. Concepts and Biomedical Aplications*, Wiley-Interscience, New Jersey, **2004**, pp. 1–22.
[24] a) T. K. Dam, C. F. Brewer, *Biochemistry* **2008**, *47*, 8470–8476; b) E. Fan, Z. Zhang, W. E. Minke, Z. Hou, C. L. M. J. Verlinde, W. G. J. Hol, *J. Am. Chem. Soc.* **2000**, *122*, 2663–2664; c) S. M. Dimick, S. C. Powell, S. A. McMahon, D. N. Moothoo, J. H. Naismith, E. J. Toone, *J. Am. Chem. Soc.* **1999**, *121*, 10286–10296.

Received: October 23, 2012
Published Online: December 13, 2012

