Organic & Biomolecular Chemistry

PAPER

View Article Online
View Journal | View Issue

Cite this: *Org. Biomol. Chem.*, 2014, **12**, 6250

6-Azido hyacinthacine A_2 gives a straightforward access to the first multivalent pyrrolizidine architectures†

Giampiero D'Adamio,[a,b] Camilla Parmeggiani,[a,b,c] Andrea Goti,*[a] Antonio J. Moreno-Vargas,[d] Elena Moreno-Clavijo,[d] Inmaculada Robina[d] and Francesca Cardona*[a]

Received 30th May 2014,
Accepted 18th June 2014

DOI: 10.1039/c4ob01117a

www.rsc.org/obc

The synthesis of the first multivalent pyrrolizidine iminosugars is reported. The key azido intermediates **4** and **31** were prepared after suitable synthetic elaboration of the cycloadduct obtained from 1,3-dipolar cycloaddition of D-arabinose derived nitrone to dimethylacrylamide. The key step of the strategy was the stereoselective installation of an azido moiety at C-6 of the pyrrolizidine skeleton. The click reaction with different monovalent and dendrimeric alkyne scaffolds allowed the preparation of a library of new mono- and multivalent pyrrolizidine compounds that were preliminarily assayed as glycosidase inhibitors towards a panel of commercially available glycosyl hydrolases.

Introduction

The multivalent display of carbohydrate ligands on a scaffold is currently an area of great interest, since carbohydrates bind only weakly to their complementary proteins, while modulation and enhancement of the biological response can be achieved by means of multiple interactions established by multivalent carbohydrates. The multivalent or cluster glycoside effect is defined as the affinity enhancement obtained with multivalent ligands compared to their monovalent counterparts.[1] This concept, widely investigated for studying carbohydrate–lectin interactions,[2] has remained essentially unexplored concerning specific glycosidase inhibition using iminosugars[3] as glycomimetics up to 2010.

Some examples of relatively low valent (2–4 iminosugar units) monocyclic iminosugars were already reported in the literature, but early glycosidase inhibition tests were not encouraging.[4] The first promising example of a multivalent effect on a glycosidase was recently reported for a trivalent 1-nojirimycin derivative that showed a 6-fold affinity enhancement towards jack bean α-mannosidase.[5] Much greater effects were demonstrated for fullerene decorated with up to 12 deoxynojirimycin iminosugars[6] and for cyclodextrin-based scaffolds decorated with 7 and 14 deoxynojirimycin units.[7] In both cases, the highest inhibition enhancements were found with jack bean α-mannosidase. Remarkably high multivalent effects were determined recently with relatively low valent (4 epitopes) porphyrin-based 1-deoxynojirimycin (1-DNJ) inhibitors, showing the importance of the spatial presentation of the DNJ epitopes in modulating the activity (that is, the compounds with highest valencies are not necessarily the best inhibitors).[8] The multivalent approach towards α-L-fucosidase inhibition was recently explored by some of us with trivalent pyrrolidine derivatives.[9,10] Two mechanisms have been proposed for the rationalization of the multivalent effect:[11] the "statistical rebinding" or "proximity effect"[12] and the standard "chelation effect". The former appears more appropriate for glycosidases since these types of enzymes have usually a single binding site and therefore an increased propensity for ligand rebinding may occur when additional ligands are in close proximity. However, very recent studies highlighted the existence of interactions of the multivalent inhibitors with non-glycone sites,[13] and the formation of aggregates of receptors with different sizes and shapes.[8]

A strong multivalent effect was also observed with deoxynojirimycin-decorated cyclodextrins towards a β-glucosidase of therapeutic interest, namely human GCase, thus showing the applicability of the multivalence concept in the development

[a]*Dipartimento di Chimica "Ugo Schiff", Università di Firenze, Via della Lastruccia 3-13, 50019 Sesto Fiorentino (FI), Italy. E-mail: francesca.cardona@unifi.it, andrea.goti@unifi.it*
[b]*European Laboratory for Non Linear Spectroscopy (LENS), via Nello Carrara 1, 50019 Sesto Fiorentino (FI), Italy*
[c]*CNR-INO, U.O.S. Sesto Fiorentino, via Nello Carrara 1, 50019 Sesto Fiorentino (FI), Italy*
[d]*Departamento de Química Orgánica, Facultad de Química, Universidad de Sevilla, c/Prof. García González 1, E-41012 Sevilla, Spain*
†Electronic supplementary information (ESI) available: ^{1}H NMR and ^{13}C NMR spectra of all new compounds, IC_{50} and Lineweaver–Burk plots of active compounds towards amyloglucosidase. See DOI: 10.1039/c4ob01117a

of chaperones for the treatment of Gaucher's disease.[14] Conversely, in this regard it was recently shown that the compounds displaying the best affinity for GCase are not necessarily the best chaperones, the best results in increasing the enzyme activity being observed with relatively low valent (tri- and tetravalent) deoxynojirimycin analogues. This could be reasonably explained by a lower cellular permeability of cyclodextrin-based compounds with respect to smaller dendrimeric analogues.[15] Relatively small trivalent and tetravalent *N*-butyl deoxynojirimycin analogues were also recently found to act as more potent CFTR (cystic fibrosis transmembrane conductance regulator) correctors than the corresponding monovalent iminosugars, thus representing promising new candidates for the treatment of cystic fibrosis (CF).[16] Finally, the porphyrin-based tetravalent 1-DNJ inhibitor recently described was found to selectively inhibit Golgi α-mannosidase ManIIb (GM) over lysosomal α-mannosidase LManII (LM), thus representing a promising compound for cancer chemotherapy.[8]

All these results illustrate the relatively newly born interest of the scientific community in this field[17] and the importance of building new and diversified multivalent iminosugars in the search for more selective glycosidase inhibitors and/or more active molecular chaperones.

Due to our experience in the synthesis of pyrrolizidine alkaloids and related structural analogues,[18] and based on our recent findings that hyacinthacine and casuarine derivatives (Fig. 1) are tight inhibitors of glycosidases of therapeutic interest (*e.g.* human maltase glucoamylase)[19] or with agrochemical potential application (*e.g.*, trehalases),[20] we report in this work the synthesis of the per-*O*-benzyl protected and unprotected 6-azido-6-*epi*-6,7-dideoxycasuarine key intermediates **4** and **31**, exploiting the cyclic polyhydroxylated nitrone cycloaddition chemistry of nitrone **1**.[21] These compounds, prepared by stereoselective installation of an azido moiety at C-6, allowed a straightforward and versatile functionalization of the pyrrolizidine skeleton by the Cu^{I}-catalyzed azide–alkyne cycloaddition (CuAAC)[22,23] and the preparation of the first examples of multivalent (3 to 9 pyrrolizidine units) 6-*epi*-6,7-dideoxycasuarine-like pyrrolizidine iminosugars. Preliminary biological evaluation towards a panel of commercial glycosidases is also reported herein.

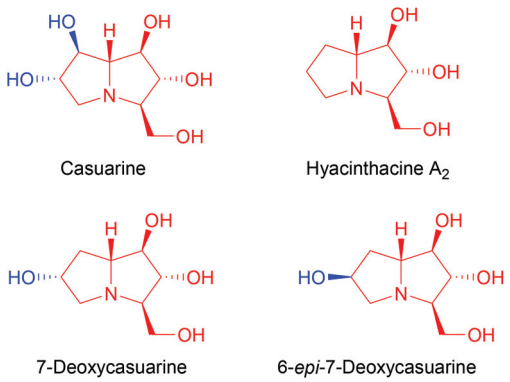
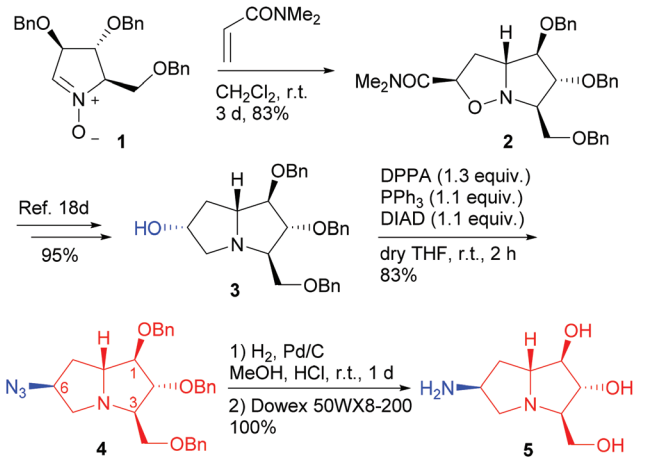


Fig. 1 Iminosugar pyrrolizidine glycosidase inhibitors.

Results and discussion

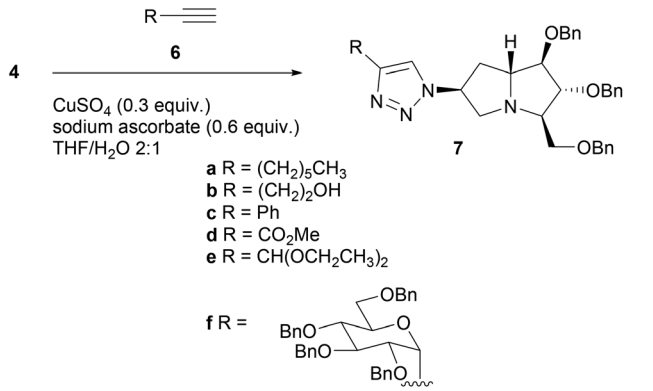
Cycloaddition of D-arabinose derived nitrone **1**[24] to *N*,*N*-dimethylacrylamide afforded isoxazolidine **2**[24a,18d] with improved 83% yield. This compound, after N–O bond cleavage with Zn in acetic acid and reduction of the C=O bond with $LiAlH_4$ in THF afforded compound **3**[18d] with an optimized 95% overall yield. Straightforward introduction of the azido group was achieved by the direct Mitsunobu reaction of **3** with 1.3 equivalents of diphenylphosphoryl azide (DPPA) in the presence of 1.1 equiv. of PPh_3 and diisopropyl azodicarboxylate (DIAD) at room temperature in dry THF,[25] which furnished the 6-azido intermediate **4** in 83% yield (Scheme 1). Confirmation of the occurred inversion of configuration at C-6 was given by 1D NOESY correlations between H-6 and H-1 and between H-6 and H-3 observed for compound **4**. Catalytic hydrogenation in acidic MeOH followed by basic treatment furnished (6*S*)-6-aminohyacinthacine A_2 (**5**) in quantitative yield (Scheme 1).

With the "6-*epi*-6,7-dideoxycasuarine like" azide **4** in hand, we tested the CuAAC cycloaddition in the presence of alkynes **6** bearing different substituents (Table 1). The reaction was performed in a 2 : 1 THF–H_2O mixture with 0.3 equivalents of $CuSO_4$ and 0.6 equivalents of sodium ascorbate,[26] and readily afforded after 2–3 hours at room temperature the 1,4-disubstituted triazole adducts **7a–e** in excellent yields after purification over silica gel.[27] Only in the case of glucosyl alkyne **6f**, synthesized in three steps from commercially available tetrabenzylated glucose,[28] no conversion of the starting material was observed at room temperature, and the reaction was successfully performed in a MW reactor at 80 °C for 45 minutes. Under these conditions, the triazole glucosyl adduct **7f** was obtained in 86% yield after isolation by flash column chromatography (FCC) (Table 1).



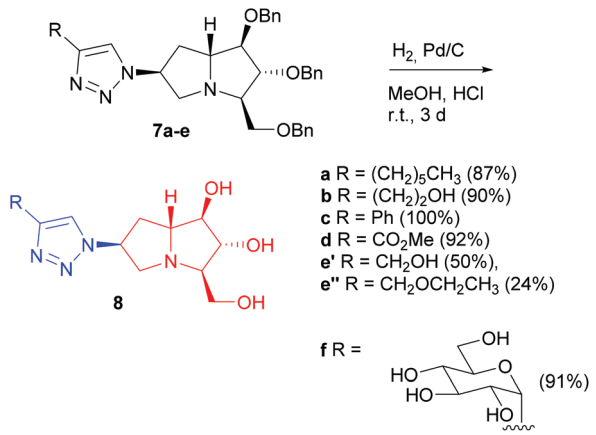
Scheme 1 Synthesis of azido intermediate **4** and of (6*S*)-6-amino-hyacinthacine A_2 (**5**).

Table 1 The CuAAC cycloaddition of **4** with alkynes **6**



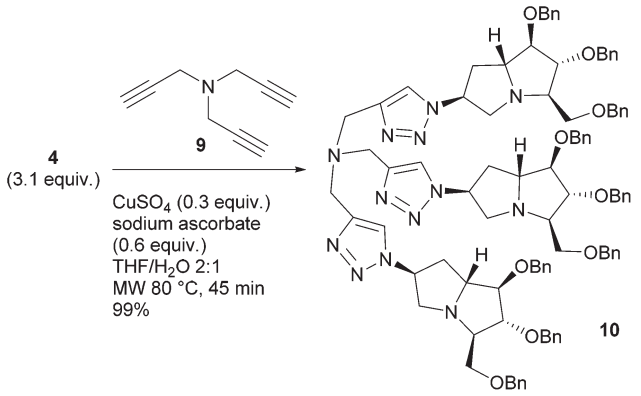
Entry	Alkyne	Temp. (°C)	Time	Product	Yield[a] (%)
1	**6a**	25	2 h	**7a**	83
2	**6b**	25	2 h	**7b**	81
3	**6c**	25	2 h	**7c**	80
4	**6d**	25	3 h	**7d**	81
5	**6e**	25	3 h	**7e**	80
6	**6f**	80[b]	45 min	**7f**	86

[a] Isolated yield after FCC. [b] Reaction performed in a MW reactor.



Scheme 2 Deprotection of the adducts. Synthesis of pyrrolizidine triazoles **8**.

Catalytic hydrogenation with Pd/C in acidic MeOH afforded pyrrolizidine derivatives **8a–d** and **8f** in excellent yields (Scheme 2). Unfortunately, the acetal protecting group for the aldehyde moiety in **7e** did not tolerate these acidic conditions, and catalytic hydrogenation afforded compounds **8e′** and **8e″** in 50% and 24% yields, respectively (Scheme 2). Compounds **8**, obtained as hydrochloride salts after catalytic hydrogenation, were obtained as pure free amines either by treatment with ion exchange resin Dowex 50WX8-200 or by FCC (see the Experimental section). Compound **8f** represents an interesting new example of an aza-*C*-disaccharide mimic[29] in which the iminosugar is linked to a common sugar by a non-hydrolyzable bio-compatible triazole linkage.[30]



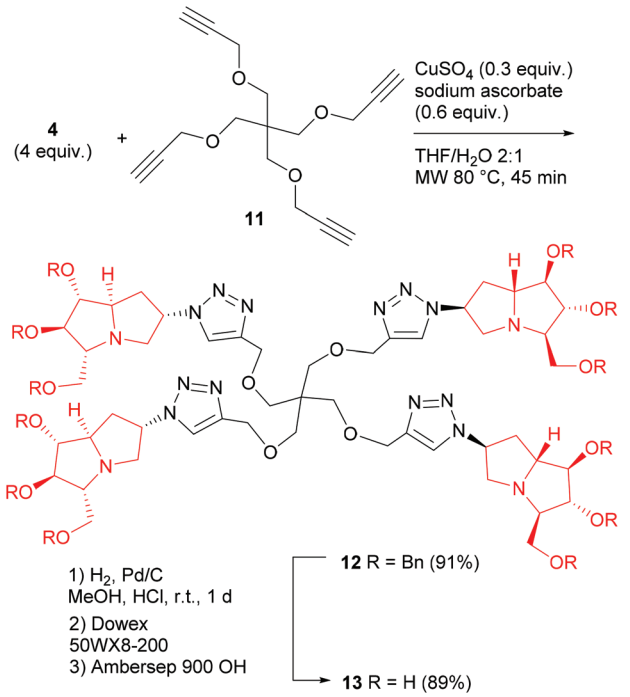
Scheme 3 Synthesis of the trivalent pyrrolizidine **10**.

Successively, the synthesis of multivalent pyrrolizidine structures was targeted by employing different scaffolds bearing terminal alkyne moieties. The CuAAC reaction of compound **4** (3.1 equivalents) with commercially available tripropargyl amine (**9**) performed with $CuSO_4$ (0.3 equiv.)/sodium ascorbate (0.6 equiv.) in THF–H_2O 2 : 1 in a MW reactor at 80 °C for 45 minutes[31] gave a very clean crude mixture, from which the trivalent pyrrolizidine compound **10** was isolated in 99% yield after flash column chromatography (Scheme 3). However, catalytic hydrogenation of **10** in acidic MeOH was troublesome, and all attempts to isolate the pure debenzylated compound failed.[32] Despite the advantage of commercial availability of this scaffold, the problems in the deprotection step prompted us to turn our attention to other polyfunctionalized alkynes on which our azide **4** is ligated.

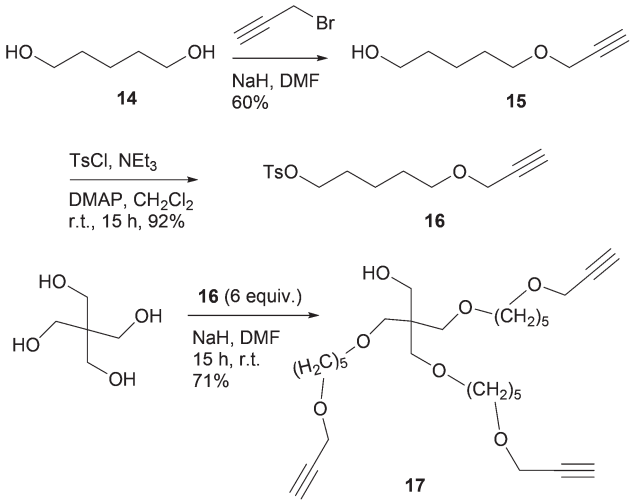
The reaction of 4 equivalents of **4** with tetravalent scaffold **11**, synthesized by propargylation of pentaerythritol with propargyl bromide and NaH as previously reported,[33] afforded the expected tetravalent iminosugar derivative **12** in 91% yield (Scheme 4). Deprotection of the perbenzylated adduct **12** was not a trivial task, due to the high basicity and hydrophilicity of the deprotected compound. Catalytic hydrogenation of **12** in acidic MeOH with Pd/C gave a mixture of hydrochloride salts that were passed onto an ion exchange resin Dowex 50WX8-200 eluting successively with MeOH, H_2O and 6% aqueous ammonia. However, part of the compound was eluted in the first fraction with MeOH as hydrochloride salt, and part with ammonia in the final fraction as free amine. After treatment of the methanolic fraction with strongly basic resin Ambersep 900 OH, pure **13** was obtained in 89% overall yield (Scheme 4).

With the aim of exploring the role of the tether in imposing a different spatial arrangement of the multivalent iminosugar, we synthesized a trivalent ligand with a longer alkyl chain. Pentane-1,5-diol (**14**) was therefore propargylated with propargyl bromide and NaH, affording alcohol **15** in 60% yield (Scheme 5).[34]

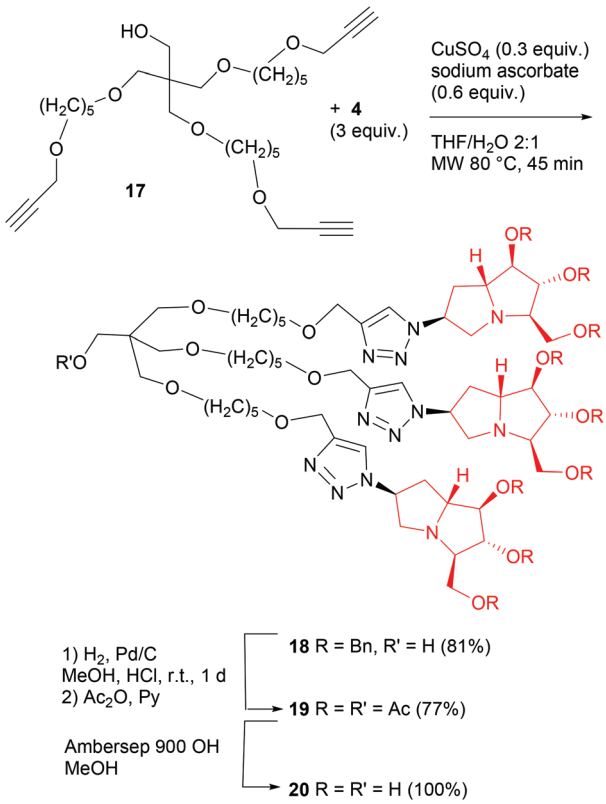
Tosylation of the primary alcohol gave tosylate **16** in 92% yield, and final treatment with pentaerythritol (6 equivalents of **16**) and NaH gave the trivalent scaffold **17** in 71% yield instead of the expected tetravalent compound. With this novel



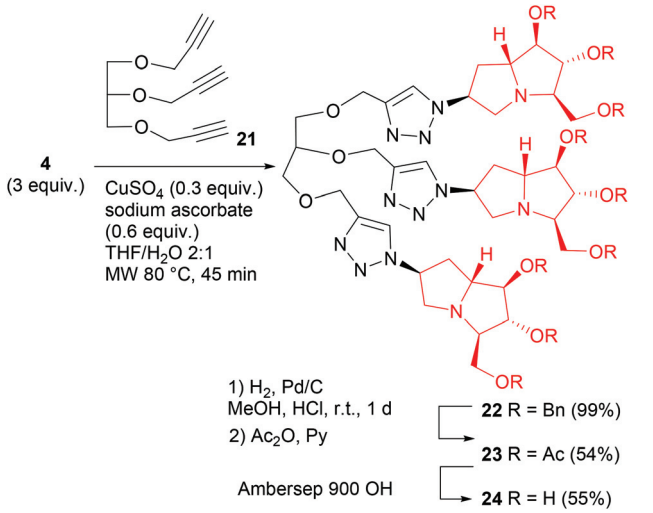
Scheme 4 Synthesis of the tetravalent pyrrolizidine **12**–**13**.



Scheme 5 Synthesis of the trivalent scaffold **17**.



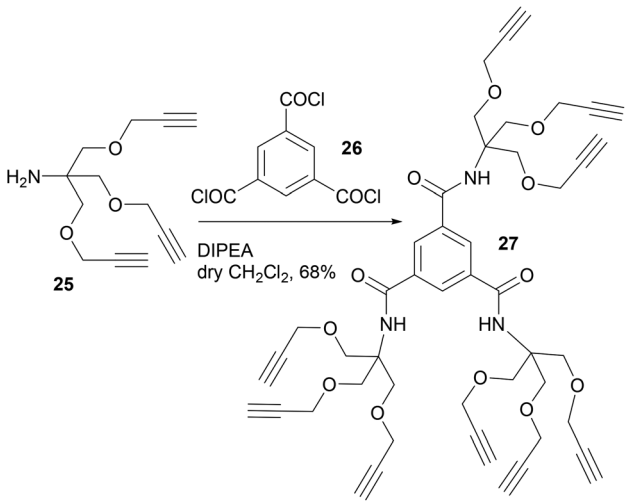
Scheme 6 Synthesis of the trivalent pyrrolizidines **19** and **20**.



Scheme 7 Synthesis of the trivalent pyrrolizidines **23** and **24**.

compound in hand, the CuAAC performed as usual and readily afforded trivalent pyrrolizidine **18** in 81% yield (Scheme 6). After catalytic hydrogenation in acidic MeOH, the crude mixture was directly reacted with excess of acetic anhydride in pyridine affording peracetylated compound **19** in 77% over two steps. Final treatment with strongly basic resin Ambersep 900 OH in MeOH overnight gave pure **20** in quantitative yield (Scheme 6).

Propargylation of glycerol with NaH and propargyl bromide gave **21**, another good and inexpensive trivalent scaffold[35] to be used in the CuAAC reaction. The reaction of **4** (3.3 equivalents) with **21** performed as described above afforded the trivalent pyrrolizidine **22** in 99% yield (Scheme 7). Catalytic hydrogenation with Pd/C in acidic MeOH followed by treatment with excess of acetic anhydride and pyridine at room temperature yielded the peracetylated compound **23** in 54% yield over two steps. Reaction of **23** with strongly basic Ambersep 900 OH in MeOH overnight gave compound **24** that needed further purification by size exclusion chromatography to yield pure **24** in 55% overall yield (Scheme 7). It should be



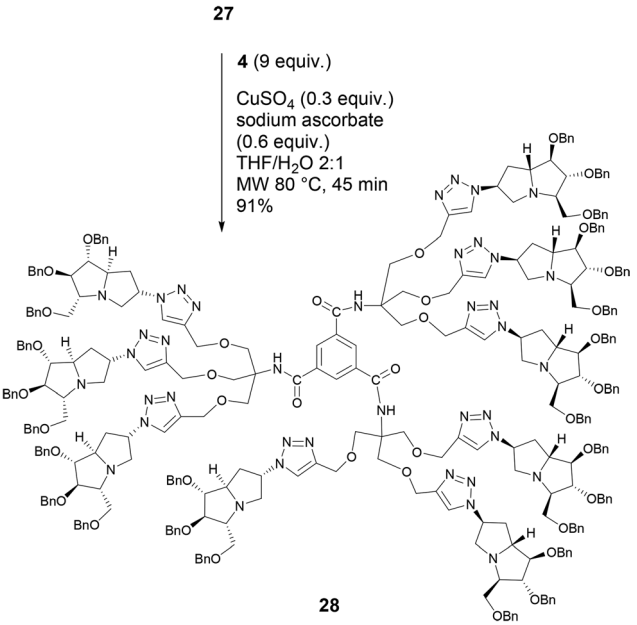
Scheme 8 Synthesis of the dendrimeric nonavalent scaffold **27**.

noted that peracetylated compounds **19** and **23** are also interesting for biological applications, in particular for use as pharmacological chaperones, since they could act as pro-drugs *in vivo* by facilitating cellular uptake, as recently reported.[15]

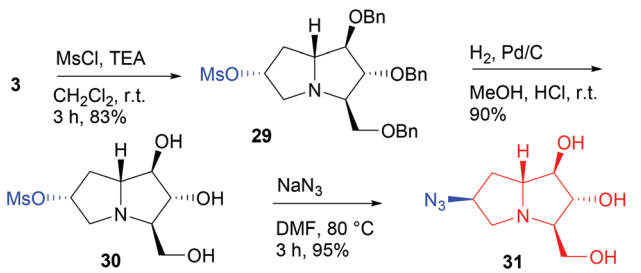
The possibility of generating higher valent pyrrolizidine iminosugars by employing a dendrimeric scaffold was finally investigated. Tris[(propargyloxy)methyl]aminomethane (**25**), which was prepared in a three step sequence from tris (hydroxymethyl)aminomethane as previously reported,[36] was reacted with trimesoyl chloride (**26**), in turn synthesized from trimesic acid,[37] in the presence of DIPEA in CH_2Cl_2 at room temperature, affording dendrimeric nonavalent scaffold **27** in 68% yield (Scheme 8).[38]

CuAAC reaction of **27** with 9 equivalents of **4** gave nonavalent pyrrolizidine **28** in 91% yield (Scheme 9). However, attempted deprotection of the benzyl groups of **28** failed. Indeed, catalytic hydrogenation in acidic MeOH afforded a mixture of salts that were treated with excess Ac_2O and Py affording the peracetylated derivative, which decomposed either upon treatment with the strongly basic resin Ambersep 900 OH or with the less basic Amberlyst A21, due to the labile amide bond.[39]

Taking into account the relatively low stability of the dendrimeric nonavalent pyrrolizidine **28**, performing the click reaction as the last step was envisioned as a better strategy. To this aim, pyrrolizidine azide with free hydroxy groups was needed as the key precursor. This turned out to be quite troublesome, since catalytic hydrogenation of **4** or treatment with BCl_3 not only removed the benzyl groups but also reduced the azide moiety to the corresponding amine, while an oxidative cleavage with $Na_2S_2O_4$ and $NaBrO_3$ [40] aimed to preserve the azido group resulted in decomposition of the compound. Therefore, a different strategy was considered, with introduction of the azido moiety after deprotection of the benzyl groups *via* a mesylate precursor. Alcohol **3** was then reacted with MsCl in CH_2Cl_2 in the presence of NEt_3 to afford mesylate **29** in 83% yield (Scheme 10).



Scheme 9 Synthesis of the nonavalent pyrrolizidine derivative **28**.

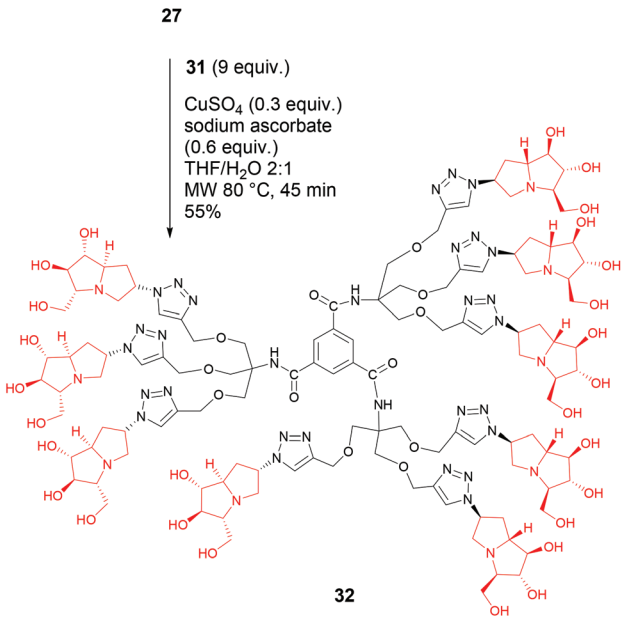


Scheme 10 Synthesis of the intermediate azide **31**.

Catalytic hydrogenation of **29** performed as usual cleanly gave compound **30** in 90% yield. Subsequent reaction with NaN_3 in DMF at 80 °C afforded azide **31** in 95% yield. Finally, the reaction of dendrimeric nonavalent scaffold **27** with 9 equivalents of azide **31** yielded pure nonavalent pyrrolizidine **32** in 55% yield after purification by FCC (Scheme 11).

Preliminary biological evaluation of these new pyrrolizidine derivatives and multivalent pyrrolizidine iminosugars was carried out by measuring their enzyme inhibitory activity. Compounds **5**, **8a–f**, **13**, **20**, **24**, **31** and **32** as well as 6-*epi*-7-deoxycasuarine were evaluated towards a panel of eleven commercially available glycosidases.[41] The preparation and biological evaluation as an enzyme inhibitor of 6-*epi*-7-deoxycasuarine has been reported by Behr.[42] Nevertheless, for comparative purpose, we have evaluated it under the same conditions as the other compounds.

The assessed compounds did not show inhibition towards β-galactosidase from *Escherichia coli* or from *Aspergillus oryzae* at 1 mM concentration and under optimal pH. Table 2 shows the inhibition results towards α-L-fucosidase from bovine kidney, α-galactosidase from coffee beans, α-glucosidase from



Scheme 11 Synthesis of the nonavalent pyrrolizidine **32**.

rice and yeast (*Saccharomyces cerevisiae*), amyloglucosidase from *Aspergillus niger*, β-glucosidase from almonds, α-mannosidase from jack beans, β-mannosidase from snail and β-*N*-acetylglucosaminidase from jack beans. It is worth mentioning that most of the new compounds showed good, competitive (see ESI† for Lineweaver–Burk plots) and specific inhibitory activity towards amyloglucosidase in the low μM range. These results are in agreement with those given by other stereochemically related pyrrolizidine alkaloids. Nevertheless some interesting structure–activity relationships can be derived from their enzyme inhibitory data. The comparison of the inhibitory properties towards amyloglucosidases of 6-*epi*-7-deoxy-casuarine (IC_{50} = 2.5 μM, K_i = 2.8 μM) and compound **5** (IC_{50} = 15.3 μM, K_i 18.2 = μM) indicates that the replacement of a hydroxy with an amino group at position 6 of the pyrrolizidine framework is detrimental, while this molecule gains moderate inhibition towards β-*N*-acetylglucosaminidase (Table 2, entries 1 and 2). However, the presence of an azido group at position 6 of the bicyclic iminosugar, as in **31**, improves the inhibition towards amyloglucosidases [(IC_{50} = 3.4 μM, K_i = 3.4 μM)] and becomes similar to 6-*epi*-7-deoxycasuarine (Table 2, entries 1 and 3). When a triazole linker is attached to position 6, a slight improvement in the inhibitory activity towards amyloglucosidases is detected (entries 4–10). The substitution at the triazole linker has little effect on the inhibitory properties against the same enzyme, given that compounds **8a–f** (IC_{50} = 1.7–2.6 μM) present similar values and behaviour. Only the hexyl derivative **8a** shows additionally a weak-to-moderate inhibition towards β-*N*-acetylglucosaminidase.

Concerning the multivalent structures, the trimeric structure **24** showed similar inhibitory potency towards amyloglucosidase (IC_{50} = 1.6 μM, K_i = 2.4 μM) compared to the most similar monovalent triazole **8e″** (Table 2, entries 9 and 13). The trimeric tethered compound **20** presented even worse inhibitory properties, thus indicating that the length of the spacer has a detrimental effect on the inhibitory values; the arms in the conjugate should move away the pyrolizidine moieties from the active site. The tetrameric structure **13** presented only a three-to-fourfold increase on the inhibitory properties towards amyloglucosidase with reference to the monovalent triazole **8e″** (Table 2, entries 9 and 11). Thus, an

Table 2 Inhibitory activities of 6-*epi*-7-deoxycasuarine and compounds **5**, **8(a–f)**, **13**, **20**, **24**, **31** and **32** towards glycosidases. Percentage of inhibition at 1 mM, IC_{50} (in parenthesis, μM) and K_i (bold, μM) if measured. Optimal pH, 37 °C[a,b,c]

		Enzymes							
Entry	Compounds	α-L-Fuc-ase	α-Gal-ase	α-Glc-ase	Amylogluc-ase	β-Glc-ase	α-Man-ase	β-Man-ase	β-*N*-Acetylglucosaminidase
1	6-*epi*-7-Deoxycasuarine	—	—	44[d]	98 (2.5) **2.8**	—	—	—	—
2	**5**	—	—	—	98 (15.3) **18.2**	24	—	—	44
3	**31**	—	—	—	98 (3.4) **3.4**	39	39	20	17
4	**8a**	—	—	—	98 (1.7) **2.2**	—	—	—	51
5	**8b**	23	—	—	98 (2.3) **3.7**	—	—	—	19
6	**8c**	—	—	—	99 (2.0)	—	—	—	—
7	**8d**	—	—	—	98 (1.9) **4.5**	—	—	—	—
8	**8e′**	—	—	—	97 (2.1) **1.9**	—	—	—	—
9	**8e″**	—	—	—	96 (2.3) **2.5**	—	—	—	—
10	**8f**	20	—	—	99 (2.6)	—	—	17	21
11	**13**	—	—	—	94 (1.5) **0.7**	—	—	—	—
12	**20**	—	29	—	83	—	16	—	—
13	**24**	—	—	—	97 (1.6) **2.4**[e]	—	26	—	—
14	**32**	21	49	51[f]	97 (0.7) **0.7**	—	—	—	—

[a] For conditions of measurements see ref. 41 and the Experimental section. [b] — No inhibition was detected at 1 mM concentration of the corresponding compound. [c] α-L-Fucosidase from bovine kidney, α-galactosidase from coffee beans, α-glucosidase from yeast (*Saccharomyces cerevisiae*), and rice, Amyloglucosidase from *Aspergillus niger*, β-glucosidase from almonds, α-mannosidase from jack beans, β-mannosidase from snail and β-*N*-acetylglucosaminidase from jack beans. [d] α-Glucosidase from yeast (*Saccharomyces cerevisiae*). [e] Mixed inhibition: the K_i value corresponding to the competitive term is given, and the non-competitive inhibition has an apparent inhibition constant K_i' = 7.6 μM. [f] α-Glucosidase from rice.

inhibitory multivalent effect is not observed, because an increase in the inhibitory potency is not over the statistical value when compared with the related monovalent compound. The selectivity observed in monovalent triazole **8e″** is maintained on the tetramer **13**. Nonaderivative **32** displayed the same inhibitory value against amyloglucosidase than **13**, although lower selectivity is observed since new weak-to-moderate inhibition is found against α-galactosidase (49%) and α-glucosidase (51%), which was not present in **13**, or in any of the monovalent compounds.

Conclusions

In conclusion, the preparation of two new azido intermediates **4** and **31**, in turn synthesized by 1,3-dipolar cycloaddition of a D-arabinose derived nitrone followed by N–O ring cleavage and stereoselective installation of an azido moiety at C-6, allowed the preparation of the first examples of multivalent pyrrolizidine iminosugars with up to 9 bioactive molecules on the same scaffold. The exploitation of the CuAAC cycloaddition on compound **4** also enabled the preparation of a small library of functionalized pyrrolizidines possessing the triazole ring at C-6. The data from the biological evaluation of these compounds revealed interesting results regarding inhibition towards amyloglucosidase.

First of all, it is worth mentioning that activity towards this enzyme is not lost despite the complexity and variety of the new molecules tested. In the monovalent pyrrolizidines studied, the presence of an azido group or a substituted triazole linker determines the same effect of the inhibitory activity towards amyloglucosidase than a hydroxyl group, while an amino group is clearly detrimental. For the multivalent structures, no multivalent effect is observed towards amyloglucosidase, the enzyme most influenced by this set of compounds. The inhibitory properties of the tetramer **13** concur approximately with the statistical effect of binding four pyrrolizidine moieties. The same value is obtained for nonavalent compound **32**. It is worth mentioning the role of the spacer in the inhibitory potency: a longer linker, although detrimental in the corresponding conjugate (**20** *vs.* **24**), provides structural information about the binding of the pyrrolizidine moieties to the enzyme active site. It is important to stress that, although from the results presented here the existence of a multivalent effect on amyloglucosidase inhibition cannot be deduced, the structure–activity relationships obtained for the monovalent and multivalent structures provide the basis for further studies in the rising field of multivalency and the action of glycosidases. Work is underway to evaluate the inhibition of these compounds towards human glycosidases and their properties as pharmacological chaperones towards lysosomal glycosidases.

Experimental section

Commercial reagents were used as received. All reactions were carried out under magnetic stirring and monitored by TLC on 0.25 mm silica gel plates (Merck F_{254}). Column chromatographies were carried out on Silica Gel 60 (32–63 μm) or on silica gel (230–400 mesh, Merck). Yields refer to spectroscopically and analytically pure compounds unless otherwise stated. ^{1}H NMR spectra were recorded on a Varian Mercury-400 or on a Varian INOVA 400 instrument at 25 °C. ^{13}C NMR spectra were recorded on a Varian Gemini-200 or on a Varian Gemini-300. Chemical shifts are reported relative to TMS (^{1}H: δ = 0.00 ppm) and $CDCl_3$ (^{13}C: δ = 77.0 ppm). Integrals are in accordance with assignments, coupling constants are given in Hz. For detailed peak assignments 2D spectra were measured (COSY, HSQC, NOESY, and NOE as necessary). Small scale microwave-assisted syntheses were carried out in a CEM Discover microwave apparatus for synthesis with an open reaction vessel and an external surface sensor. IR spectra were recorded with a BX FT-IR Perkin-Elmer System spectrophotometer. ESI-MS spectra were recorded with a Thermo Scientific™ LCQ Fleet Ion Trap Mass Spectrometer. Elemental analyses were performed with a Perkin-Elmer 2400 analyzer. Optical rotation measurements were performed on a JASCO DIP-370 polarimeter.

Cycloaddition of nitrone 1 to *N*,*N*-dimethylformamide[18d]

A solution of nitrone **1** (600 mg, 1.44 mmol) and *N*,*N*-dimethylformamide (223 μL, 2.16 mmol) in 4 mL of CH_2Cl_2 was stirred at room temp. for 3 d, until TLC analysis (AcOEt–PE 3 : 1) showed the disappearance of the starting material (R_f = 0.39) and the appearance of a new product (R_f = 0.30). At completion of the reaction, the solvent was removed under reduced pressure and the crude mixture was purified by FCC (AcOEt–PE 3 : 1) affording pure **2** (R_f = 0.30, 620 mg, 1.2 mmol, 83% yield) as a white solid.

N–O ring cleavage of cycloadduct 2 and reduction of lactam to alcohol 3 [18d]

To a solution of **2** (190 mg, 0.37 mmol) in 3 mL of a 9 : 1 CH_3COOH–H_2O mixture, Zn dust (97 mg, 1.47 mmol) was added. The suspension was stirred at 65 °C for 3 h, then TLC analysis (AcOEt–PE, 3 : 1) showed the disappearance of the starting material (R_f = 0.30) and formation of a new product (R_f = 0.44). After filtration through cotton the mixture was concentrated at reduced pressure and saturated aqueous solution of $NaHCO_3$ was added at 0 °C until basic pH was attained. After extraction with AcOEt (3 × 25 mL), the organic layers were dried over Na_2SO_4 and concentrated at reduced pressure affording pure lactam as a white solid that was dissolved in 10 mL of dry THF and, under a nitrogen atmosphere at 0 °C, $LiAlH_4$ (1 m solution in THF, 1.22 mL, 1.22 mmol) was added dropwise. The mixture was raised to room temp. and heated at reflux temperature for 2 h, until TLC analysis (AcOEt–PE, 3 : 1) showed the disappearance of the starting material (R_f = 0.37) and the formation of a new product (R_f = 0.00). The reaction was then quenched at 0 °C with a saturated aqueous solution of Na_2SO_4. After extraction with AcOEt (3 × 20 mL), the organic layers were dried on Na_2SO_4 and concentrated at reduced pressure affording pure **3** (161 mg, 0.35 mmol, 95% yield over two steps) as a white solid.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-1,2-Bis(benzyloxy)-3-[(benzyloxy)methyl]-6-azido-hexahydro-1*H*-pyrrolizidine (4)

To a solution of **3** (666 mg, 1.45 mmol) in dry THF (6 mL) under a nitrogen atmosphere triphenylphosphine (422 mg, 1.60 mmol) was added. The reaction mixture was cooled to 0 °C and DIAD (315 μL, 1.60 mmol) was added dropwise, forming a yellow precipitate. After addition of diphenylphosphorilazide (520 mg, 1.89 mmol), the suspension was raised to room temp. and stirred under a nitrogen atmosphere for 2 h, until TLC analysis (CH_2Cl_2–MeOH 10 : 1) showed the disappearance of the starting material (R_f = 0.22) and formation of a new product (R_f = 0.95). The solvent was removed under reduced pressure and the crude was purified by FCC (PE–AcOEt 4 : 1) affording pure **4** (R_f = 0.21, 582 mg, 1.20 mmol, 83% yield) as a yellow oil. $[\alpha]_D^{27}$ = +3.96 (*c* = 1.06 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.30–7.18 (m, 15H, H-Ar), 4.60–4.48 (AB system, *J* = 11.7 Hz, 2H, H-Bn), 4.50–4.41 (AB system, *J* = 11.7 Hz, 2H, H-Bn), 4.48–4.42 (AB system, *J* = 12.2 Hz, 2H, H-Bn), 4.10–4.06 (m, 1H, H-6), 3.96 (dd, *J* = 6.3, 5.4 Hz, 1H, H-2), 3.76 (t, *J* = 4.9 Hz, 1H, H-1), 3.59–3.54 (m, 1H, H-7a), 3.50 (dd, *J* = 9.2, 5.3 Hz, 1H, Ha-8), 3.43 (dd, *J* = 9.2, 6.3 Hz, 1H, Hb-8), 3.07 (dd, *J* = 11.7, 3.4 Hz, 1H, Ha-5), 2.98 (dd, *J* = 11.7, 5.4 Hz, 1H, Hb-5), 2.93 (q, *J* = 6.3 Hz, 1H, H-3), 1.96 (ddd, *J* = 13.2, 6.8, 3.4 Hz, 1H, Ha-7), 1.81 (ddd, *J* = 13.6, 8.3, 6.3 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 138.4, 138.2, 137.9 (s, C-Ar), 128.4–127.5 (d, 15C, C-Ar), 88.3 (d, C-1), 85.9 (d, C-2), 73.3 (t, C-Bn), 72.4 (t, 2C, C-Bn, C-8), 71.9 (t, C-Bn), 69.2 (d, C-3), 66.7 (d, C-7a), 62.1 (d, C-6), 59.9 (t, C-5), 37.0 (t, C-7); IR ($CDCl_3$): ν = 3081, 3064, 3032, 2926, 2864, 2102, 1496, 1453, 1365, 1264, 1099 cm^{-1}; MS (ESI): *m*/*z* calcd for $C_{29}H_{32}N_4O_3$ + H^+ 485.25 $[M + H]^+$; found: 485.33; elemental analysis calcd (%) for $C_{29}H_{32}N_4O_3$ (484.59): C 71.88, H 6.66, N 11.56; found: C 71.45, H 6.54, N 11.68.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-Amino-3-hydroxymethylhexahydro-1*H*-pyrrolizidine-1,2-diol (5)

To a solution of **4** (112 mg, 0.23 mmol) in 10 mL of methanol 56 mg of 10% Pd/C and two drops of 37% HCl were added under a nitrogen atmosphere, and then the mixture was stirred under a hydrogen atmosphere at room temp. for 1 day. TLC analysis (PE–AcOEt 3 : 1) showed the disappearance of the starting material (R_f = 0.35) and formation of a new product (R_f = 0.00). The mixture was filtered through Celite® and the solvent was removed under reduced pressure affording a crude yellow oil (117 mg). Free amine was obtained by passing the hydrochloride through a Dowex 50WX8 ion-exchange resin. Elution with 6% NH_4OH afforded the free base **5** (43 mg, 0.23 mmol, 100% yield over two steps) as a waxy brown solid. $[\alpha]_D^{23}$ = +16.7 (*c* = 1.01 in MeOH); ^{1}H-NMR (400 MHz, D_2O): δ = 3.66–3.59 (m, 3H, H-1, H-2, Ha-8), 3.49 (dd, *J* = 11.7, 6.3 Hz, 1H, Hb-8), 3.46–3.40 (m, 1H, H-6), 3.15 (td, *J* = 7.8, 3.9 Hz, 1H, H-7a), 2.85 (dd, *J* = 11.7, 6.3 Hz, 1H, Ha-5), 2.59 (ddd, *J* = 9.8, 6.3, 3.9 Hz, 1H, H-3), 2.47 (dd, *J* = 11.7, 7.8 Hz, 1H, Hb-5), 1.96 (ddd, *J* = 12.7, 6.4, 3.9 Hz, 1H, Ha-7), 1.62 (dt, *J* = 12.7, 8.3 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, D_2O): δ = 80.3 (d, C-1) 76.6 (d, C-2), 69.4 (d, C-3), 65.1 (d, C-7a), 62.9 (t, C-8), 60.9 (t, C-5), 49.4 (d, C-6), 37.7 (t, C-7); MS (ESI): *m*/*z* calcd for $C_8H_{16}N_2O_3$ + H^+ 189.12 $[M + H]^+$; found: 189.09; elemental analysis calcd (%) for $C_8H_{16}N_2O_3$ (188.22): C 51.05, H 8.57, N 14.88; found: C 51.66, H 8.98, N 15.08.

General procedure for the synthesis of 1,4-disubstituted triazole adducts 7a–f

To a solution of **4** (1 equiv.) in a 2 : 1 THF–H_2O mixture were added $CuSO_4$ (30 mol%), sodium ascorbate (60 mol%) and alkyne **6a–f** (1 equiv.). The reaction mixture was stirred at room temp[43] until TLC analysis (EP–AcOEt 3 : 1) showed the disappearance of the starting material (R_f = 0.49) and formation of the desired product. After filtration through Celite®, the solvent was removed under reduced pressure and the crude was purified by FCC to obtain the 1,4-disubstituted triazole adducts **7a–f**.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-1,2-Bis(benzyloxy)-3-[(benzyloxy)methyl]-6-[(4-hexyl)-1*H*-1,2,3-triazol-1-yl]-hexahydro-1*H*-pyrrolizidine (7a). Obtained as a colorless oil in 83% yield on 0.12 mmol of alkyne **6a** after 2 h. R_f = 0.14 (PE–AcOEt 3 : 1); $[\alpha]_D^{26}$ = +8.99 (*c* = 1.48 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.41 (s, 1H, H-triazole), 7.36–7.25 (m, 15H, H-Ar), 5.29–5.23 (m, 1H, H-6), 4.70–4.59 (AB system, *J* = 11.7 Hz, 2H, H-Bn), 4.54–4.48 (AB system, *J* = 11.7 Hz, 2H, H-Bn), 4.53 (s, 2H, H-Bn), 4.08 (dd, *J* = 5.9, 5.0 Hz, 1H, H-2), 3.90 (t, *J* = 5.0 Hz, 1H, H-1), 3.66 (td, *J* = 7.7, 5.0 Hz, 1H, H-7a), 3.60 (dd, *J* = 9.4, 5.3 Hz, 1H, Ha-8), 3.52 (dd, *J* = 9.4, 6.4 Hz, 1H, Hb-8), 3.38 (d, *J* = 6.0 Hz, 2H, H-5), 3.13 (q, *J* = 6.0 Hz, 1H, H-3), 2.68 (t, *J* = 7.8 Hz, 2H, *CH*$_2$CH=CH), 2.35 (ddd, *J* = 13.4, 7.3, 3.5 Hz, 1H, Ha-7), 2.25–2.18 (m, 1H, Hb-7), 1.65 (pquint, *J* = 7.6 Hz, 2H, *CH*$_2$CH$_2$CH=CH), 1.40–1.25 (m, 6H, CH_2), 0.88 (t, *J* = 6.9 Hz, 3H, CH_3); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 148.2 (s, C-triazole), 138.0, 137.7, 137.4 (s, C-Ar), 128.1–127.2 (d, 15C, C-Ar), 118.9 (d, C-triazole), 88.1 (d, C-1), 85.5 (d, C-2), 73.0 (t, C-Bn), 72.1 (t, 2C, C-Bn, C-8), 71.7 (t, C-Bn), 68.8 (d, C-3), 66.5 (d, C-7a), 61.1 (d, C-6), 60.1 (t, C-5), 37.9 (t, C-7), 31.3 (t, CH_2), 29.2 (t, *CH*$_2$CH$_2$CH=CH), 28.7 (t, CH_2), 25.5 (t, *CH*$_2$CH=CH), 22.3 (t, CH_2), 13.8 (q, CH_3); IR ($CDCl_3$): ν = 3088, 3065, 3031, 2928, 2859, 1708, 1454, 1361, 1223, 1110, 1094 cm^{-1}; MS (ESI): *m*/*z* calcd for $C_{37}H_{46}N_4O_3$ + H^+ 595.36 $[M + H^+]$; found: 595.43; elemental analysis calcd (%) for $C_{37}H_{46}N_4O_3$ (594.79): C 74.72, H 7.80, N 9.42; found: C 74.40, H 8.03, N 8.95.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-1,2-Bis(benzyloxy)-3-[(benzyloxy)methyl]-6-[4-(2-hydroxyethyl)-1*H*-1,2,3-triazol-1-yl]-hexahydro-1*H*-pyrrolizidine (7b). Obtained as a colorless oil in 81% yield on 0.09 mmol of alkyne **6b** after 2 h. R_f = 0.36 (CH_2Cl_2–MeOH 12 : 1); $[\alpha]_D^{25}$ = +14.0 (*c* = 0.92 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.53 (s, 1H, H-triazole), 7.35–7.26 (m, 15H, H-Ar), 5.30–5.23 (m, 1H, H-6), 4.71–4.56 (AB system, *J* = 11.7 Hz, 2H, H-Bn), 4.54–4.47 (AB system, *J* = 11.7 Hz, 2H, H-Bn), 4.52 (s, 2H, H-Bn), 4.08 (dd, *J* = 5.9, 5.0 Hz, 1H, H-2), 3.96–3.92 (m, 2H, *CH*$_2$OH), 3.90 (t, *J* = 5.0 Hz, 1H, H-1), 3.65 (td, *J* = 7.9, 5.3 Hz, 1H, H-7a), 3.59 (dd, *J* = 9.4, 5.3 Hz, 1H, Ha-8), 3.51 (dd, *J* = 9.4, 6.4 Hz, 1H, Hb-8), 3.40–3.38 (m, 2H, H-5), 3.14 (q, *J* = 6.2 Hz, 1H, H-3), 2.92 (t, *J* = 5.9 Hz, 2H, *CH*$_2$CH$_2$OH), 2.68 (bs, 1H,

OH), 2.36 (ddd, J = 13.4, 7.3, 3.5 Hz, 1H, Ha-7), 2.22 (ddd, J = 13.5, 7.9, 6.8 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 145.6 (s, C-triazole), 138.3, 138.0, 137.7 (s, C-Ar), 128.5–127.6 (d, 15C, C-Ar), 120.3 (d, C-triazole), 88.4 (d, C-1), 85.9 (d, C-2), 73.3 (t, C-Bn), 72.4 (t, C-Bn), 72.3 (t, C-Bn), 72.0 (t, C-8), 69.1 (d, C-3), 66.8 (d, C-7a), 61.6 (d, C-6), 61.5 (t, CH_2OH), 60.4 (t, C-5), 38.1 (t, CH_2CH_2OH), 28.7 (t, C-7); IR ($CDCl_3$): ν = 3622, 3469, 3088, 3066, 3032, 2865, 2247, 1496, 1453, 1365, 1112, 1062 cm^{-1}; MS (ESI): m/z calcd for $C_{33}H_{38}N_4O_4$ + H^+ 555.29 [M + H^+]; found: 555.40; elemental analysis calcd (%) for $C_{33}H_{38}N_4O_4$ (554.68): C 71.46, H 6.91, N 10.10; found: C 71.06, H 6.98, N 10.19.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-1,2-Bis(benzyloxy)-3-[(benzyloxy)methyl]-6-[(4-phenyl)-1*H*-1,2,3-triazol-1-yl]-hexahydro-1*H*-pyrrolizidine (7c). Obtained as colorless oil in 80% yield on 0.11 mmol of alkyne **6c** after 2 h. R_f = 0.28 (PE–AcOEt 2 : 1); $[\alpha]_D^{25}$ = +5.33 (c = 1.05 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.92 (s, 1H, H-triazole), 7.83–7.80 (m, 2H, H-Ar), 7.43–7.39 (m, 2H, H-Ar), 7.36–7.26 (m, 15H, H-Ar), 7.25 (s, 1H, H-Ar), 5.35 (dq, J = 6.4, 3.5 Hz, 1H, H-6), 4.71–4.60 (AB system, J = 11.7 Hz, 2H, H-Bn), 4.58–4.52 (AB system, J = 11.7 Hz, 2H, H-Bn), 4.54–4.48 (AB system, J = 11.7 Hz, 2H, H-Bn), 4.11 (dd, J = 5.9, 4.9 Hz, 1H, H-2), 3.93 (t, J = 4.9 Hz, 1H, H-1), 3.70 (td, J = 7.8, 4.9 Hz, 1H, H-7a), 3.62 (dd, J = 9.3, 5.4 Hz, 1H, Ha-8), 3.54 (dd, J = 9.3, 6.4 Hz, 1H, Hb-8), 3.48 (dd, J = 11.7, 3.4 Hz, 1H, Ha-5), 3.42 (dd, J = 11.7, 5.4 Hz, 1H, Hb-5), 3.17 (q, J = 5.9 Hz, 1H, H-3), 2.40 (ddd, J = 13.6, 7.3, 3.4 Hz, 1H, Ha-7), 2.26 (ddd, J = 13.7, 8.3, 6.8 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 147.5 (s, C-triazole), 138.0, 137.7, 137.4 (s, C-Ar), 130.4 (s, C-Ar), 128.4–127.2 (d, 19C, C-Ar), 125.4 (d, C-Ar), 118.1 (d, C-triazole), 88.1 (d, C-1), 85.7 (d, C-2), 73.0 (t, C-Bn), 72.2 (t, C-Bn), 72.0 (t, C-8), 71.8 (t, C-Bn), 68.9 (d, C-3), 66.5 (d, C-7a), 61.6 (d, C-6), 60.1 (t, C-5), 38.0 (t, C-7); IR ($CDCl_3$): ν = 3087, 3066, 3031, 2928, 2865, 1496, 1454, 1264, 1206, 1097, 1075, 1027 cm^{-1}. MS (ESI): m/z calcd for $C_{37}H_{38}N_4O_3$ + H^+ 587.29 [M + H^+]; found: 587.42; elemental analysis calcd (%) for $C_{37}H_{38}N_4O_3$ (586.72): C 75.74, H 6.53, N 9.55; found: C 75.33, H 6.42, N 9.10.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-1,2-Bis(benzyloxy)-3-[(benzyloxy)methyl]-6-[(4-methylcarboxylate)-1*H*-1,2,3-triazol-1-yl]-hexahydro-1*H*-pyrrolizidine (7d). Obtained as a colorless oil in 81% yield on 0.11 mmol of alkyne **6d** after 3 h. R_f = 0.17 (PE–AcOEt 2 : 1); $[\alpha]_D^{24}$ = +17.1 (c = 1.18 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 8.27 (s, 1H, H-triazole), 7.34–7.24 (m, 15H, H-Ar), 5.35–5.31 (m, 1H, H-6), 4.67–4.57 (AB system, J = 11.7 Hz, 2H, H-Bn), 4.51 (s, 2H, H-Bn), 4.48 (s, 2H, H-Bn), 4.08 (dd, J = 5.5, 5.0 Hz, 1H, H-2), 3.94 (s, 3H, OMe), 3.89 (t, J = 5.0 Hz, 1H, H-1), 3.59 (td, J = 8.2, 5.1 Hz, 1H, H-7a), 3.58 (dd, J = 9.4, 5.5 Hz, 1H, Ha-8), 3.50 (dd, J = 9.3, 6.6 Hz, 1H, Hb-8), 3.40–3.39 (m, 2H, H-5), 3.16 (q, J = 6.1 Hz, 1H, H-3), 2.30 (ddd, J = 13.7, 7.2, 2.9 Hz, 1H, Ha-7), 2.22 (ddd, J = 13.7, 8.4, 6.6 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 161.2 (s, C═O), 140.0 (s, C-triazole), 138.3, 138.0, 137.7 (s, C-Ar), 128.5–127.6 (d, 15C, C-Ar), 126.5 (d, C-triazole), 88.4 (d, C-1), 86.1 (d, C-2), 73.3 (t, C-Bn), 72.4 (t, C-Bn), 72.3 (t, C-8), 72.1 (t, C-Bn), 69.1 (d, C-3), 66.6 (d, C-7a), 62.4 (d, C-6), 60.3 (t, C-5), 52.1 (q, OMe), 38.1 (t, C-7); IR ($CDCl_3$): ν = 3088, 3066, 3031, 2951, 2864, 1730, 1496, 1454, 1326, 1262, 1240, 1208, 1100 cm^{-1}; MS (ESI): m/z calcd for $C_{33}H_{36}N_4O_5$ + H^+ 569.27 [M + H^+]; found: 569.38; elemental analysis calcd (%) for $C_{33}H_{36}N_4O_5$ (568.66): C 69.70, H 6.38, N 9.85; found: C 69.21, H 5.98, N 10.17.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-1,2-Bis(benzyloxy)-3-[(benzyloxy)methyl]-6-[(4-diethoxymethyl)-1*H*-1,2,3-triazol-1-yl]-hexahydro-1*H*-pyrrolizidine (7e). Obtained as a colorless oil in 80% yield on 0.11 mmol of alkyne **6e** after 3 h. R_f = 0.28 (PE–AcOEt 1 : 1); $[\alpha]_D^{23}$ = +9.2 (c = 0.9 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.65 (s, 1H, H-triazole), 7.28–7.18 (m, 15H, H-Ar), 5.63 (s, 1H, $CH(OCH_2CH_3)_2$), 5.22–5.17 (m, 1H, H-6), 4.54–4.51 (AB system, J = 11.7 Hz, 2H, H-Bn), 4.47–4.41 (m, 4H, H-Bn), 4.00 (dd, J = 5.8, 4.9 Hz, 1H, H-2), 3.83 (t, J = 4.9 Hz, 1H, H-1), 3.67–3.50 (m, 6H, Ha-8, H-7a, $CH(OCH_2CH_3)_2$), 3.44 (dd, J = 9.3, 6.3 Hz, 1H, Hb-8), 3.33 (d, J = 5.4 Hz, 2H, H-5), 3.06 (q, J = 5.8 Hz, 1H, H-3), 2.29 (ddd, J = 13.6, 7.3, 3.9 Hz, 1H, Ha-7), 2.20–2.13 (m, 1H, Hb-7), 1.17 (t, J = 7.1 Hz, 3H, CH_3), 1.16 (t, J = 7.1 Hz, 3H, CH_3); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 147.2 (s, C-triazole), 138.3, 138.1, 137.7 (s, C-Ar), 128.5–127.6 (d, 15C, C-Ar), 120.6 (d, C-triazole), 97.0 (d, $CH(OCH_2CH_3)_2$), 88.4 (d, C-1), 85.9 (d, C-2), 73.3 (t, C-Bn), 72.4 (t, 2C, C-Bn, C-8), 72.0 (t, C-Bn), 69.1 (d, C-3), 66.8 (d, C-7a), 61.8 (t, 2C, $CH(OCH_2CH_3)_2$), 61.5 (d, C-6), 60.4 (t, C-5), 38.1 (t, C-7), 15.2 (q, 2C, CH_3); IR ($CDCl_3$): ν = 3031, 2978, 1496, 1454, 1364, 1230, 1206, 1027, 1001 cm^{-1}; MS (ESI): m/z calcd for $C_{36}H_{44}N_4O_5$ + H^+ 613.33 [M + H^+]; found: 613.45; elemental analysis calcd (%) for $C_{36}H_{44}N_4O_5$ (612.76): C 70.56, H 7.24, N 9.14; found: C 70.37, H 7.18, N 9.05.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-1,2-Bis(benzyloxy)-3-[(benzyloxy)methyl]-6-[(4-tetrabenzylglucosyl)-1*H*-1,2,3-triazol-1-yl]-hexahydro-1*H*-pyrrolizidine (7f). Obtained as a colorless oil in 86% yield on 0.06 mol of alkyne **6f** after heating to 80 °C in a MW reactor for 45 minutes. R_f = 0.21 (PE–AcOEt 3 : 2); $[\alpha]_D^{22}$ = +26.3 (c = 1.28 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.73 (s, 1H, H-triazole), 7.34–7.18 (m, 33H, H-Ar), 7.14–7.12 (m, 2H, H-Ar), 5.26 (d, J = 5.9 Hz. 1H, H-1′), 5.24–5.19 (m, 1H, H-6), 4.92 (d, J = 10.7 Hz. 1H, H-Bn), 4.83–4.78 (AB system, J = 10.7 Hz, 2H, H-Bn), 4.70–4.44 (m, 11H, H-Bn), 4.20–4.15 (m, 1H, H-3′), 4.04 (dd, J = 5.9, 4.9 Hz, 1H, H-2), 3.98 (dd, J = 9.2, 5.8 Hz, 1H, H-2′), 3.89 (t, J = 4.9 Hz, 1H, H-1), 3.77 (d, J = 5.3 Hz, 2H, H-4′, H-5′), 3.72–3.63 (m, 3H, H-7a, H-6′), 3.56 (dd, J = 9.3, 5.4 Hz, 1H, Ha-8), 3.45 (dd, J = 9.3, 6.8 Hz, 1H, Hb-8), 3.43 (dd, J = 11.7, 5.9 Hz, 1H, Ha-5), 3.37 (dd, J = 11.7, 4.4 Hz, 1H, Hb-5), 3.12 (q, J = 5.8 Hz, 1H, H-3), 2.39 (ddd, J = 13.2, 7.3, 3.9 Hz, 1H, Ha-7), 2.25 (dt, J = 13.7, 7.3 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 143.8 (s, C-triazole), 138.8–137.8 (s, 7C, C-Ar), 128.5–127.6 (d, 35C, C-Ar), 122.9 (d, C-triazole), 88.5 (d, C-1), 85.9 (d, C-2), 82.4 (d, C-3′), 79.8 (d, C-2′), 78.0 (d, C-5′), 75.4–72.0 (d or t, 9C, 7 C-Bn, C-4′, C-8), 69.2 (d, C-1′), 69.1 (d, C-3), 68.7 (t, C-6′), 66.9 (d, C-7a), 61.1 (d, C-6), 60.4 (t, C-5), 38.0 (t, C-7); IR ($CDCl_3$): ν = 3088, 3065, 3031, 2920, 2867, 1495, 1454, 1361, 1207, 1087, 1074, 1027 cm^{-1}; MS (ESI): m/z calcd for $C_{65}H_{68}N_4O_8$ + H^+ 1033.50 [M + H^+]; found: 1033.57; elemental analysis calcd (%) for $C_{65}H_{68}N_4O_8$ (1033.26): C 75.56, H 6.63, N 5.42; found: C 75.71, H 6.99, N 5.26.

General procedure for the synthesis of pyrrolizidine derivatives 8a–d and 8f

To a solution of compounds **7a–d** and **7f** (1 equiv.) in methanol, 10% Pd/C (50% weight) and two drops of 37% HCl were added under a nitrogen atmosphere, then the mixture was stirred under a hydrogen atmosphere at room temp. At completion of the reaction, the mixture was filtered through Celite® and the solvent was removed under reduced pressure affording a crude oil. Free amines were obtained by purification either with the ion exchange resin Dowex 50WX8-200 eluting successively with MeOH, H_2O and 6% NH_4OH (**8a**,**8b**,**8f**) or by FCC using mixtures of CH_2Cl_2–MeOH as eluents with addition (1% v/v) of 6% NH_4OH (**8c**,**8d**,**8e′**,**8e″**).

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-[(4-Hexyl)-1*H*-1,2,3-triazol-1-yl]-3-(hydroxymethyl)-hexahydro-1*H*-pyrrolizidine-1,2-diol (8a). Obtained as a pale yellow solid in 87% yield on 0.05 mmol of adduct **7a** after 3 days. M.p. 113–115 °C; $[\alpha]_D^{24}$ = +15.8 (*c* = 0.55 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 7.85 (s, 1H, H-triazole), 5.30 (quint, *J* = 5.6 Hz, 1H, H-6), 3.88 (t, *J* = 7.3 Hz, 1H, H-2), 3.82 (t, *J* = 6.8 Hz, 1H, H-1), 3.75 (dd, *J* = 11.2, 3.4 Hz, 1H, Ha-8), 3.61 (dd, *J* = 11.2, 5.8 Hz, 1H, Hb-8), 3.56 (q, *J* = 7.1 Hz, 1H, H-7a), 3.47 (dd, *J* = 11.7, 4.4 Hz, 1H, Ha-5), 3.41 (dd, *J* = 11.7, 5.9 Hz, 1H, Hb-5), 2.86 (ddd, *J* = 9.3, 6.4, 3.5 Hz, 1H, H-3), 2.67 (t, *J* = 7.8 Hz, 2H, CH_2CH=CH), 2.51 (ddd, *J* = 13.2, 7.8, 5.1 Hz, 1H, Ha-7), 2.41 (dt, *J* = 13.2, 6.8 Hz, 1H, Hb-7), 1.65 (pquint, *J* = 7.4 Hz, 2H, CH_2CH_2CH=CH), 1.40–1.28 (m, 6H, CH_2), 0.89 (t, *J* = 6.8 Hz, 3H, CH_3); ^{13}C-NMR (50 MHz, CD_3OD): δ = 148.0 (s, C-triazole), 120.8 (d, C-triazole), 81.2 (d, C-1), 77.7 (d, C-2), 71.8 (d, C-3), 67.0 (d, C-7a), 62.2 (t, C-8), 60.7 (d, C-6), 59.6 (t, C-5), 36.5 (t, C-7), 31.3 (t, CH_2), 29.1 (t, CH_2CH_2CH=CH), 28.5 (t, CH_2), 24.9 (t, CH_2CH=CH), 22.2 (t, CH_2), 12.9 (q, CH_3); MS (ESI): *m*/*z* calcd for $C_{16}H_{28}N_4O_3$ + Na^+ 347.21 $[M + Na]^+$; found: 347.28; elemental analysis calcd (%) for $C_{16}H_{28}N_4O_3$ (324.42): C 59.24, H 8.70, N 17.27; found: C 59.18, H 8.48, N 17.09.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-[4-(2-Hydroxyethyl)-1*H*-1,2,3-triazol-1-yl]-3-(hydroxymethyl)-hexahydro-1*H*-pyrrolizidine-1,2-diol (8b). Obtained as a colorless oil in 90% yield on 0.05 mmol of adduct **7b** after 3 days. $[\alpha]_D^{23}$ = +11.9 (*c* = 0.74 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 7.90 (s, 1H, H-triazole), 5.29 (quint, *J* = 5.4 Hz, 1H, H-6), 3.86 (t, *J* = 7.3 Hz, 1H, H-2), 3.79 (t, *J* = 7.3 Hz, 1H, H-1), 3.78 (t, *J* = 6.6 Hz, 2H, CH_2OH), 3.73 (dd, *J* = 11.2, 3.2 Hz, 1H, Ha-8), 3.58 (dd, *J* = 11.2, 5.9 Hz, 1H, Hb-8), 3.50 (q, *J* = 7.3 Hz, 1H, H-7a), 3.41 (dd, *J* = 11.7, 4.8 Hz, 1H, Ha-5), 3.36 (dd, *J* = 11.7, 5.9 Hz, 1H, Hb-5), 2.89 (t, *J* = 6.6 Hz, 2H, CH_2CH_2OH), 2.79 (ddd, *J* = 8.8, 5.8, 3.4 Hz, 1H, H-3), 2.49 (ddd, *J* = 13.2, 7.8, 5.4 Hz, 1H, Ha-7), 2.38 (dt, *J* = 13.2, 6.8 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, CD_3OD): δ = 143.9 (s, C-triazole), 120.7 (d, C-triazole), 80.3 (d, C-1), 76.8 (d, C-2), 70.8 (d, C-3), 66.0 (d, C-7a), 61.3 (t, C-8), 59.9 (d, C-6), 59.8 (t, CH_2OH), 58.7 (t, C-5), 35.6 (t, C-7), 27.7 (t, CH_2CH_2OH); MS (ESI): *m*/*z* calcd for $C_{12}H_{20}N_4O_4$ + Na^+ 307.14 $[M + Na]^+$; found: 307.25; elemental analysis calcd (%) for $C_{12}H_{20}N_4O_4$ (284.31): C 50.69, H 7.09, N 19.71; found: C 50.23, H 6.97, N 19.27.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-[4-(Phenyl)-1*H*-1,2,3-triazol-1-yl]-3-(hydroxymethyl)-hexahydro-1*H*-pyrrolizidine-1,2-diol (8c). Obtained as a white solid in 100% yield on 0.15 mmol of adduct **7c** after 6 h. R_f = 0.29 (CH_2Cl_2–MeOH 4 : 1 + 1% v/v of 6% NH_4OH); M.p. 136–137 °C; $[\alpha]_D^{22}$ = +14.8 (*c* = 0.27 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 8.52–8.50 (m, 1H, H-triazole), 7.83 (d, *J* = 7.8 Hz, 2H, H-Ar), 7.43 (t, *J* = 7.3 Hz, 2H, H-Ar), 7.34 (t, *J* = 7.3 Hz, 1H, H-Ar), 5.57–5.56 (m, 1H, H-6), 4.07–3.98 (m, 3H, H-1, H-2, H-7a), 3.94 (dd, *J* = 12.7, 3.9 Hz, 1H, Ha-5), 3.90–3.76 (m, 3H, Hb-5, H-8), 3.34–3.30 (m, 1H, H-3), 2.82–2.76 (m, 1H, Ha-7), 2.74–2.67 (m, 1H, Hb-7); ^{13}C-NMR (50 MHz, CD_3OD): δ = 147.8 (s, C-triazole), 130.1 (s, C-Ar), 128.6 (d, 2C, C-Ar), 128.1 (d, C-Ar), 125.4 (d, 2C, C-Ar), 120.8 (d, C-triazole), 79.3, 77.2 (d, C-1, C-2), 74.2 (d, C-3), 70.5 (d, C-7a), 60.3 (d, C-6), 59.8, 59.4 (t, C-5, C-8), 35.3 (t, C-7); MS (ESI): *m*/*z* calcd for $C_{16}H_{20}N_4O_3$ + Na^+ 339.14 $[M + Na]^+$; found: 339.30; elemental analysis calcd (%) for $C_{16}H_{20}N_4O_3$ (316.35): C 60.75, H 6.37, N 17.71; found: C 60.37, H 6.21, N 17.58.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-[4-(Methylcarboxylate)-1*H*-1,2,3-triazol-1-yl]-3-(hydroxymethyl)-hexahydro-1*H*-pyrrolizidine-1,2-diol (8d). Obtained as a waxy yellow solid in 92% yield on 0.09 mmol of adduct **7d** after 16 h. R_f = 0.36 (CH_2Cl_2–MeOH 4 : 1 + 1% v/v of 6% NH_4OH); $[\alpha]_D^{24}$ = +15.4 (*c* = 0.59 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 8.65 (s, 1H, H-triazole), 5.47–5.42 (pquint., *J* = 4.9 Hz, 1H, H-6), 3.92–3.85 (m, 2H, H-1, H-2), 3.91 (s, 3H, OMe), 3.77 (dd, *J* = 11.2, 3.4 Hz, 1H, Ha-8), 3.68–3.57 (m, 3H, Hb-8, Ha-5, H-7a), 3.52–3.48 (m, 1H, Hb-5), 2.98–2.90 (m, 1H, H-3), 2.62–2.56 (m, 1H, Ha-7), 2.49 (dt, *J* = 13.6, 6.8 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, CD_3OD): δ = 160.0 (s, C=O), 138.2, (s, C-triazole), 126.6 (d, C-triazole), 79.9 (d, C-1), 76.8 (d, C-2), 71.4 (d, C-3), 66.6 (d, C-7a), 60.9 (t, C-8), 60.6 (d, C-6), 58.8 (t, C-5), 50.2 (q, OMe), 35.3 (t, C-7); MS (ESI): *m*/*z* calcd for $C_{12}H_{18}N_4O_5$ + Na^+ 321.12 $[M + Na]^+$; found: 321.16; elemental analysis calcd (%) for $C_{12}H_{18}N_4O_5$ (298.30): C 48.32, H 6.08, N 18.78; found: C 48.46, H 6.32, N 18.92.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-[4-(Glucosyl)-1*H*-1,2,3-triazol-1-yl]-3-(hydroxymethyl) hexahydro-1*H*-pyrrolizidine-1,2-diol (8f). Obtained as a waxy white solid in 91% yield on 0.04 mmol of adduct **7f** after 1 day. $[\alpha]_D^{22}$ = +62.3 (*c* = 0.44 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 8.12 (s, 1H, H-triazole), 5.32 (quint, *J* = 5.6 Hz, 1H, H-6), 5.18 (d, *J* = 5.4 Hz. 1H, H-1′) 3.87–3.83 (m, 3H, H-2, H-2′, H-3′), 3.80 (t, *J* = 7.1 Hz, 1H, H-1), 3.76–3.72 (m, 2H, Ha-8, Ha-6′), 3.66 (dd, *J* = 12.2, 4.9 Hz, 1H, Hb-6′), 3.58 (dd, *J* = 11.7, 6.3 Hz, 1H, Hb-8), 3.52 (q, *J* = 6.8 Hz, 1H, H-7a), 3.45–3.36 (m, 4H, H-5, H-4′ and H-5′), 2.80 (ddd, *J* = 8.8, 6.4, 3.4 Hz, 1H, H-3), 2.52 (ddd, *J* = 13.2, 7.8, 5.4 Hz, 1H, Ha-7), 2.40 (dt, *J* = 13.2, 6.6 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, CD_3OD): δ = 142.8 (s, C-triazole), 122.9 (d, C-triazole), 80.4 (d, C-1), 76.8 (d, C-2), 74.1 (d, C-5′), 73.2 (d, C-2′), 70.6 (d, C-3), 70.5 (d, C-3′), 69.9 (d, C-1′), 69.7 (d, C-4′), 65.7 (d, C-7a), 61.7 (t, C-8), 60.6 (d. C-6), 59.8 (t, C-6′), 58.7 (t, C-5), 35.7 (t, C-7); MS (ESI): *m*/*z* calcd for $C_{16}H_{26}N_4O_8$ + Na^+ 425.16 $[M + Na]^+$; found: 425.25; elemental analysis calcd (%) for $C_{16}H_{26}N_4O_8$ (402.40): C 47.76, H 6.51, N 13.92; found: C 47.89, H 6.60, N 13.48.

Catalytic hydrogenation of 1,4-disubstituted triazole adduct 7e. To a solution of **7f** (59 mg, 0.096 mmol) in 10 mL of ethanol, 10% Pd/C (50% weight) and two drops of 37% HCl were added under a nitrogen atmosphere, and then the

mixture was stirred under a hydrogen atmosphere at room temp for 5 days. TLC analysis (EP–AcOEt 1 : 1) showed the disappearance of the starting material (R_f = 0.36) and formation of a new product (R_f = 0.00). The mixture was filtered through Celite® and the solvent was removed under reduced pressure. Then, the crude was purified by FCC (CH_2Cl_2–MeOH 4 : 1 + 1% v/v of 6% NH_4OH) affording pure **8e′** (R_f = 0.17, 13 mg, 0.048 mmol, 50% yield) and **8e″** (R_f = 0.23, 7 mg, 0.024 mmol, 24% yield) as a colorless oils.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-[4-(Hydroxymethyl)-1*H*-1,2,3-triazol-1-yl]-3-(hydroxymethyl)-hexahydro-1*H*-pyrrolizidine-1,2-diol (8e′). $[\alpha]_D^{23}$ = +61.2 (*c* = 0.26 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 8.03 (s, 1H, H-triazole), 5.33 (quint, *J* = 5.4 Hz, 1H, H-6), 4.67 (s, 2H, O*CH*$_2$triazole), 3.87 (t, *J* = 7.8 Hz, 1H, H-2), 3.80 (t, *J* = 7.1 Hz, 1H, H-1), 3.74 (dd, *J* = 11.2, 3.4 Hz, 1H, Ha-8), 3.59 (dd, *J* = 11.7, 5.9 Hz, 1H, Hb-8), 3.50 (q, *J* = 6.8 Hz, 1H, H-7a), 3.41 (dd, *J* = 11.7, 4.9 Hz, 1H, Ha-5), 3.41–3.36 (m, 1H, Hb-5), 2.79 (ddd, *J* = 8.8, 5.9, 3.5 Hz, 1H, H-3), 2.49 (ddd, *J* = 12.9, 7.3, 4.9 Hz, 1H, Ha-7), 2.39 (dt, *J* = 13.4, 6.7 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, CD_3OD): δ = 147.6 (s, C-triazole), 121.8 (d, C-triazole), 81.3 (d, C-1), 77.8 (d, C-2), 71.4 (d, C-3), 66.5 (d, C-7a), 62.5 (t, C-8), 60.9 (d, C-6), 59.6 (t, C-5), 55.1 (t, O*CH*$_2$triazole), 36.6 (t, C-7); MS (ESI): *m*/*z* calcd for $C_{11}H_{18}N_4O_4$ + Na^+ 293.12 $[M + Na]^+$; found: 293.18; elemental analysis calcd (%) for $C_{11}H_{18}N_4O_4$ (270.29): C 48.88, H 6.71, N 20.73; found: C 48.65, H 6.24, N 20.34.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-[4-(Ethoxymethyl)-1*H*-1,2,3-triazol-1-yl]-3-(hydroxymethyl)-hexahydro-1*H*-pyrrolizidine-1,2-diol (8e″). $[\alpha]_D^{23}$ = +27.5 (*c* = 0.12 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 8.08 (s, 1H, H-triazole), 5.39–5.34 (m, 1H, H-6), 4.57 (s, 2H, O*CH*$_2$triazole), 3.89 (t, *J* = 7.3 Hz, 1H, H-2), 3.84 (t, *J* = 6.9 Hz, 1H, H-1), 3.76 (dd, *J* = 11.2, 3.4 Hz, 1H, Ha-8), 3.64–3.54 (m, 4H, Hb-8, H-7a, CH_3CH_2O), 3.53–3.42 (m, 2H, H-5), 2.89–2.86 (m, 1H, H-3), 2.53 (ddd, *J* = 12.9, 7.8, 5.1 Hz, 1H, Ha-7), 2.47–2.41 (m, 1H, Hb-7), 1.19 (t, *J* = 7.1 Hz, 3H, CH_3); ^{13}C-NMR (50 MHz, CD_3OD): δ = 144.7 (s, C-triazole), 122.7 (d, C-triazole), 81.3 (d, C-1), 77.8 (d, C-2), 71.6 (d, C-3), 66.7 (d, C-7a), 66.5 (t, CH_3CH_2O), 63.0 (t, O*CH*$_2$triazole), 62.3 (t, C-8), 61.0 (d, C-6), 59.6 (t, C-5), 36.6 (t, C-7), 13.9 (q, CH_3); MS (ESI): *m*/*z* calcd for $C_{13}H_{22}N_4O_4$ + Na^+ 321.15 $[M + Na]^+$; found: 321.22; elemental analysis calcd (%) for $C_{13}H_{22}N_4O_4$ (298.34): C 52.34, H 7.43, N 18.78; found: C 52.25, H 7.38, N 18.72.

Benzylated trivalent iminosugar 10 from tripropargyl amine (9)

To a solution of **4** (150 mg, 0.31 mmol) in 1.5 mL of a 2 : 1 THF–H_2O mixture, $CuSO_4$ (30 mol%, 5 mg, 0.03 mmol), sodium ascorbate (60 mol%, 12 mg, 0.06 mmol) and tripropargyl amine (**9**) (14 μL, 0.1 mmol) were added dropwise. The reaction mixture was heated in a MW reactor at 80 °C for 45 min, until TLC analysis (PE–AcOEt 3 : 1) showed the disappearance of the starting material (R_f = 0.35) and formation of a new product (R_f = 0.00). After filtration through Celite®, the solvent was removed under reduced pressure and the crude was purified by FCC (CH_2Cl_2–MeOH 10 : 1 + 1% v/v of 6% NH_4OH) affording pure **10** (R_f = 0.47, 157 mg, 0.1 mmol, 99%) as a yellow oil. $[\alpha]_D^{22}$ = +6.15 (*c* = 0.26, $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.83 (s, H-triazole), 7.70 (s, H-triazole), 7.67 (s, H-triazole), 7.32–7.24 (m, 45H, H-Ar), 5.28–5.17 (m, 3H, H-6), 4.69–4.47 (m, 18H, H-Bn), 4.09–4.04 (m, 3H, H-2), 3.90–3.88 (m, 3H, H-1), 3.86, 3.84, 3.76 (s, 6H, N*CH*$_2$), 3.73–3.63 (m, 3H, H-7a), 3.60–3.51 (m, 6H, H-8), 3.45–3.37 (m, 6H, H-5), 3.16–3.09 (m, 3H, H-3), 2.45–3.32 (m, 3H, Ha-7), 2.30–2.19 (m, 3H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 144.2, 144.0, 143.5 (s, C-triazole), 138.0–137.4 (s, 9C, C-Ar), 128.1–127.2 (d, 45C, C-Ar), 122.3, 121.6, 121.3 (d, C-triazole), 88.0 (d, 3C, C-1), 85.4 (d, 3C, C-2), 73.5–71.7 (t, 9C, C-Bn), 71.6 (t, 3C, C-8), 68.8 (d, 3C, C-3), 66.5 (d, 3C, C-7a), 61.2, 61.0, 60.6 (d, C-6), 47.9, 47.7, 47.0 (t, N*CH*$_2$), 41.7 (t, 3C, C-5), 37.9, 37.7, 37.6 (t, C-7); MS (ESI): *m*/*z* calcd for $C_{96}H_{105}N_{13}O_9$ + Na^+ 1606.81 $[M + Na]^+$; found: 1606.83; elemental analysis calcd (%) for $C_{96}H_{105}N_{13}O_9$ (1584.94): C 72.75, H 6.68, N 11.49; found: C 72.81, H 6.18, N 11.24.

Synthesis of tetravalent scaffold 11

A solution of pentaerythritol (200 mg, 1.47 mmol) in 15 mL of dry DMF was cooled to 0 °C and NaH (60% w/w in mineral oil, 1.53 g, 38.2 mmol) was added. After stirring for 30 min. at 0 °C, propargyl bromide (80% w/w in toluene, 1.31 g, 8.82 mmol) was slowly added dropwise over 20 minutes. The colour of the solution changed to brown and formation of a precipitate was observed. Then, the reaction mixture was stirred under a nitrogen atmosphere at room temp. for 16 h, until TLC analysis (PE–AcOEt 3 : 1) showed the disappearance of the starting material (R_f = 0.00) and formation of a new product (R_f = 0.59). The reaction was quenched with methanol and the solvent was removed under reduced pressure. After extraction with AcOEt (3 × 20 mL), the combined organic layers were dried on Na_2SO_4, concentrated at reduced pressure and the crude mixture was purified by FCC (hexane–AcOEt 5 : 1) affording pure **11** (R_f = 0.19, 254 mg, 0.88 mmol, 60% yield) as a white solid.

Benzylated tetravalent iminosugar 12

To a solution of **4** (58 mg, 0.12 mmol) in 1.5 mL of a 2 : 1 THF–H_2O mixture, $CuSO_4$ (30 mol%, 2 mg, 0.009 mmol), sodium ascorbate (60 mol%, 4 mg, 0.018 mmol) and **11** (10 mg, 0.03 mmol) were added. The reaction mixture was heated in a MW reactor at 80 °C for 45 min, until TLC analysis (PE–AcOEt 3 : 1) showed the disappearance of the starting material (R_f = 0.59) and formation of a new product (R_f = 0.00). After filtration through Celite®, the solvent was removed under reduced pressure and the crude was purified by FCC (CH_2Cl_2–MeOH 15 : 1) affording pure **12** (R_f = 0.32, 61 mg, 0.023 mmol, 91%) as a yellow oil. $[\alpha]_D^{22}$ = +3.43 (*c* = 0.35 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.71 (s, 4H, H-triazole), 7.35–7.22 (m, 60H, H-Ar), 5.24–5.18 (m, 4H, H-6), 4.68–4.45 (m, 24H, H-Bn), 4.50 (s, 8H, O*CH*$_2$triazole), 4.10–4.03 (m, 4H, H-2), 3.89 (t, *J* = 4.9 Hz, 4H, H-1), 3.69 (td, *J* = 7.4, 4.8 Hz, 4H, H-7a), 3.58 (dd, *J* = 9.3, 5.4 Hz, 4H, Ha-8), 3.54–3.50 (m, 4H, Hb-8), 3.50 (s, 8H, C*CH*$_2$O), 3.36 (d, *J* = 5.3 Hz, 8H, H-5), 3.14–3.09 (m, 4H, H-3),

2.42–2.35 (m, 4H, Ha-7), 2.21 (td, J = 13.6, 7.3 Hz, 4H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 145.2 (s, 4C, C-triazole), 138.4, 138.2, 137.9 (s, 12C, C-Ar), 128.4–127.6 (d, 60C, C-Ar), 121.6 (d, 4C, C-triazole), 88.4 (d, 4C, C-1), 85.8 (d, 4C, C-2), 73.2 (t, 4C, C-Bn), 72.4 (t, 4C, C-8), 72.3 (t, 4C, C-Bn), 71.9 (t, 4C, C-Bn), 69.3, 69.0 (t, 4C, CCH_2O), 68.9 (d, 4C, C-3), 66.7 (d, 4C, C-7a), 65.1 (t, 4C, OCH_2triazole), 60.9 (d, 4C, C-6), 60.2 (t, 4C, C-5), 45.7 (s, CCH$_2$O), 37.9 (t, 4C, C-7); IR ($CDCl_3$): ν = 3088, 3066, 3032, 2909, 2866, 1496, 1453, 1308, 1255, 1206, 1098 cm^{-1}; MS (ESI): m/z calcd for $[(C_{133}H_{148}N_{16}O_{16} + 2Na)/2]^{2+}$ 1135.57 $[(M + 2Na)/2]^+$; found: 1136.10; elemental analysis calcd (%) for $C_{133}H_{148}N_{16}O_{16}$ (2226.70): C 71.74, H 6.70, N 10.06; found: C 71.26, H 6.27, N 9.85.

Polyhydroxylated tetravalent iminosugar 13

To a solution of **12** (96 mg, 0.043 mmol) in 10 mL of methanol, 48 mg of 10% Pd/C and two drops of 37% HCl were added under a nitrogen atmosphere, and then the mixture was stirred under a hydrogen atmosphere at room temp. for 4 days. TLC analysis (CH_2Cl_2–MeOH 10 : 1) showed the disappearance of the starting material (R_f = 0.57) and formation of a new product (R_f = 0.00). The mixture was filtered through Celite® and the solvent was removed under reduced pressure affording a crude yellow oil. Then, the hydrochloride form was dissolved in MeOH and passed onto an ion exchange resin Dowex 50WX8-200 eluting successively with MeOH, H_2O and 6% NH_4OH. Part of the compound was eluted in the first methanolic fraction as hydrochloride salt, and a small part in the final elution with ammonia as pure free amine **13** (6 mg, 0.005 mmol, 12% yield). After treatment of the methanolic fraction with strongly basic resin Ambersep 900 OH pure **13** (38 mg, 0.033 mmol, 77% yield) was recovered as a waxy white solid (89% overall yield). $[\alpha]_D^{21}$ = +13.8 (c = 0.32 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 8.05 (s, 4H, H-triazole), 5.33 (pquint, J = 5.7 Hz. 4H, H-6), 4.49 (s, 8H, OCH_2triazole), 3.87 (t, J = 7.3 Hz, 4H, H-2), 3.82 (t, J = 7.3 Hz, 4H, H-1), 3.74 (dd, J = 11.2, 2.9 Hz, 4H, Ha-8), 3.59 (dd, J = 11.2, 5.3 Hz, 4H, Hb-8), 3.49 (q, J = 6.8 Hz, 4H, H-7a), 3.43 (dd, J = 6.4, 2.0 Hz, 4H, Ha-5), 3.42 (s, 8H, CCH_2O), 3.38 (dd, J = 5.9, 2.0 Hz, 4H, Hb-5), 2.80 (ddd, J = 7.8, 5.8, 3.4 Hz, 4H, H-3), 2.49 (ddd, J = 13.2, 7.8, 5.6 Hz, 4H, Ha-7), 2.39 (dt, J = 13.2, 6.6 Hz, 4H, Hb-7); ^{13}C-NMR (50 MHz, CD_3OD): δ = 143.8 (s, 4C, C-triazole), 121.8 (d, 4C, C-triazole), 80.4 (d, 4C, C-1), 76.7 (d, 4C, C-2), 70.5 (d, 4C, C-3), 67.6 (t, 4C, CCH_2O), 65.8 (s, CCH$_2$O), 65.7 (d, 4C, C-7a), 63.0 (t, 4C, OCH_2triazole), 61.6 (t, 4C, C-8), 59.8 (d, 4C, C-6), 58.7 (t, 4C, C-5), 35.7 (t, 4C, C-7). MS (ESI): m/z calcd for $C_{49}H_{76}N_{16}O_{16}$ + Na$^+$ 1167.55 $[M + Na]^+$; found: 1167.78; elemental analysis calcd (%) for $C_{49}H_{76}N_{16}O_{16}$ (1145.23): C 51.39, H 6.69, N 19.57; found: C 51.81, H 6.65, N 19.52.

5-(Prop-2-in-1-yloxy)pentyl 4-methylbenzenesulfonate (16)

To a solution of **15** (320 mg, 2.25 mmol) in 30 mL of dry CH_2Cl_2 were added dry NEt_3 (1.57 mL, 11.25 mmol), DMAP (6.5 μL, 54 μmol) and tosyl chloride (1.29 g, 6.75 mmol). The reaction mixture was stirred under a nitrogen atmosphere for 15 h. Then, TLC analysis (AcOEt–PE 3 : 1) showed the disappearance of the starting material (R_f = 0.26) and formation of a new product (R_f = 0.92). After washing with a saturated aqueous solution of $NaHCO_3$ (3 × 25 mL) and H_2O (25 mL), the combined organic layers were dried on Na_2SO_4, concentrated at reduced pressure and the crude mixture was purified by FCC (PE–AcOEt 5 : 1) affording pure **16** (R_f = 0.25, 614 mg, 2.07 mmol, 92% yield) as a colorless oil. ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.78 (d, J = 8.3 Hz, 2H, H-Ar), 7.34 (d, J = 7.8 Hz, 2H, H-Ar), 4.10 (d, J = 2.4 Hz, 2H, OCH_2C≡CH), 4.02 (t, J = 6.3 Hz, 2H, H-1), 3.46 (t, J = 6.3 Hz, 2H, H-5), 2.45 (s, 3H, CH_3), 2.41 (t, J = 2.4 Hz, 1H, OC≡CH), 1.71–1.64 (m, 2H, H-2), 1.59–1.51 (m, 2H, H-4), 1.43–1.35 (m, 2H, H-3); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 144.7 (s, C-S), 133.1 (s, C-Ar), 129.8 (d, 2C, C-Ar), 127.8 (d, 2C, C-Ar), 79.9 (s, OC≡CH), 74.2 (d, OCH≡CH), 70.4 (t, C-1), 69.6 (t, C-5), 58.0 (t, OCH_2C≡CH), 28.7 (t, C-4), 28.5 (t, C-2), 22.0 (t, C-3), 21.6 (q, CH_3); IR ($CHCl_3$): ν = 3306, 3031, 3019, 3010, 2957, 2867, 1598, 1495, 1455, 1358, 1188, 1175, 1097 cm^{-1}; MS (ESI): m/z calcd for $C_{15}H_{20}O_4S$ + Na$^+$ 319.10 $[M + Na]^+$; found: 319.05; elemental analysis calcd (%) for $C_{15}H_{20}O_4S$ (296.38): C 60.79, H 6.80, S 10.82; found: C 60.67, H 6.72, S 10.69.

Synthesis of trivalent scaffold 17 from pentaerythritol

A solution of pentaerythritol (8 mg, 0.059 mmol) in 3 mL of dry DMF was cooled to 0 °C and a solution of NaH (60% w/w in mineral oil, 60 mg, 1.44 mmol) and **16** (107 mg, 0.36 mmol) in 3 mL of dry DMF was added dropwise. The reaction mixture was stirred under a nitrogen atmosphere at room temp. for 20 h, until TLC analysis (PE–AcOEt 1 : 1) showed the disappearance of the starting material (R_f = 0.00) and formation of a new product (R_f = 0.73). The reaction was quenched with methanol and the solvent was removed under reduced pressure. After addition of AcOEt (15 mL), the combined organic layers were washed with H_2O (2 × 15 mL), dried on Na_2SO_4 and concentrated under reduced pressure. The crude mixture was purified by FCC (PE–AcOEt 3 : 1) affording pure **17** (R_f = 0.18, 21 mg, 0.042 mmol, 71% yield) as a colorless oil. ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 4.12 (d, J = 2.4 Hz, 6H, OCH_2C≡CH), 3.68 (s, 2H, CH_2OH), 3.50 (t, J = 6.6 Hz, 6H, CH_2), 3.42 (s, 6H, CCH_2O), 3.38 (t, J = 6.6 Hz, 6H, CH_2), 2.42–2.40 (m, 3H, OC≡CH), 1.64–1.53 (m, 12H, CH_2), 1.43–1.35 (m, 2H, CH_2); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 80.0 (s, 3C, OC≡CH), 74.1 (d, 3C, OCH≡CH), 71.5 (t, 3C, CCH_2O), 71.5, 70.0 (t, 6C, CH_2), 66.4 (t, CH_2OH), 58.0 (t, 3C, OCH_2C≡CH), 44.7 (s, CCH$_2$O), 29.3, 29.2 (t, 6C, CH_2), 22.7 (t, 3C, CH_2); IR ($CHCl_3$): ν = 3488, 3306, 3009, 2942, 2865, 1459, 1358, 1094 cm^{-1}; MS (ESI): m/z calcd for $C_{29}H_{48}O_7$ + H + Na$^+$ 532.33 $[M + H + Na]^+$; found: 532.58. Anal. Calcd for $C_{29}H_{48}O_7$ (508.69): C, 68.47; H, 9.51. Found C, 68.39; H, 9.41.

Benzylated trivalent iminosugar 18

To a solution of **4** (78 mg, 0.16 mmol) in 3 mL of a 2 : 1 THF–H_2O mixture, $CuSO_4$ (30 mol%, 2 mg, 0.015 mmol), sodium ascorbate (60 mol%, 6 mg, 0.03 mmol) and **17** (27 mg, 0.05 mmol) were added. The reaction mixture was heated in a MW reactor at 80 °C for 45 min, until TLC analysis (PE–AcOEt 3 : 1) showed the disappearance of the starting material (R_f =

0.18) and formation of a new product (R_f = 0.00). After filtration through Celite®, the solvent was removed under reduced pressure and the crude was purified by FCC (CH_2Cl_2–MeOH 10 : 1) affording pure **18** (R_f = 0.26, 78 mg, 0.041 mmol, 81%) as a waxy white solid. $[\alpha]_D^{21}$ = +8.0 (c = 0.3 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.63 (s, 3H, H-triazole), 7.28–7.17 (m, 45H, H-Ar), 5.22–5.18 (m, 3H, H-6), 4.61–4.50 (AB system, J = 11.7 Hz, 6H, H-Bn), 4.51 (s, 6H, OCH_2triazole), 4.47–4.40 (AB system, J = 11.7 Hz, 6H, H-Bn), 4.46 (s, 6H, H-Bn), 4.00 (t, J = 5.4 Hz, 3H, H-2), 3.83 (t, J = 4.6 Hz, 3H, H-1), 3.61 (s, 2H, CH_2OH), 3.55–3.43 (m, 6H, H-8), 3.46–3.43 (m, 3H, H-7a), 3.45 (t, J = 6.6 Hz, 6H, CH_2), 3.34–3.28 (m, 6H, H-5), 3.34 (s, 6H, CCH_2O), 3.30 (t, J = 6.6 Hz, 6H, CH_2), 3.10–3.06 (m, 3H, H-3), 2.32–2.28 (m, 3H, Ha-7), 2.19–2.15 (m, 3H, Hb-7), 1.58–1.44 (m, 12H, CH_2), 1.35–1.26 (m, 6H, CH_2); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 145.3 (s, 3C, C-triazole), 138.3, 138.0, 137.7 (s, 9C, C-Ar), 128.5–127.6 (d, 45C, C-Ar), 121.3 (d, 3C, C-triazole), 88.3 (d, 3C, C-1), 85.8 (d, 3C, C-2), 73.3, 72.4 (t, 6C, C-Bn), 72.3 (t, 3C, C-8), 72.0 (t, 3C, C-Bn), 71.6 (t, 6C, C-5, CH_2), 70.8 (d or t, 6C, C-7a, CH_2), 69.1 (d, 3C, C-3), 66.8 (t, CH_2OH), 64.3 (t, 3C, OCH_2triazole), 61.4 (d, 3C, C-6), 60.2 (t, 3C, CCH_2O), 44.7 (s, CCH$_2$O), 38.0 (t, 3C, C-7), 29.4 (t, 6C, CH_2), 22.7 (t, 3C, CH_2); IR ($CHCl_3$): ν = 3644, 3005, 2934, 2864, 1453, 1214, 1097 cm^{-1}; MS (ESI): m/z calcd for $C_{116}H_{144}N_{12}O_{16}$ + Na^+ 1984.07 $[M + Na]^+$; found: 1984.88; elemental analysis calcd (%) for $C_{116}H_{144}N_{12}O_{16}$ (1962.46): C 70.99, H 7.40, N 8.56; found: C 71.0, H 7.68, N 8.45.

Peracetylated trivalent iminosugar 19

To a solution of **18** (67 mg, 0.035 mmol) in 15 mL of methanol, 34 mg of 10% Pd/C and four drops of 37% HCl were added under a nitrogen atmosphere, then the mixture was stirred under a hydrogen atmosphere at room temp. for 5 days. After that TLC analysis (CH_2Cl_2–MeOH 10 : 1) showed the disappearance of the starting material (R_f = 0.26) and formation of a new product (R_f = 0.00), the mixture was filtered through Celite® and the solvent was removed under reduced pressure. The crude yellow oil was dissolved in pyridine (3 mL) and acetic anhydride (2 mL) was added. The solution was stirred at room temp. for two days. After concentration under reduced pressure, the crude mixture was purified by FCC (CH_2Cl_2–MeOH 30 : 1) affording pure **19** (R_f = 0.31, 40 mg, 0.027 mmol, 77%) as a pale yellow oil. $[\alpha]_D^{24}$ = +6.9 (c = 0.35 in $CHCl_3$); ^{1}H-NMR (400 MHz, CD_3OD): δ = 7.99 (s, 3H, H-triazole), 5.41–5.35 (m, 3H, H-6), 5.34–5.31 (m, 3H, H-2), 5.10 (t, J = 5.4 Hz, 3H, H-1), 4.55 (s, 6H, OCH_2triazole), 4.21 (dd, J = 12.0, 5.7 Hz, 3H, Ha-8), 4.08 (dd, J = 11.8, 5.9 Hz, 3H, Hb-8), 4.04 (d, J = 4.9 Hz, 2H, CH_2OAc), 3.70 (td, J = 7.8, 5.4 Hz, 3H, H-7a), 3.52–3.35 (m, 12H, H-5, CCH_2O), 3.50 (t, J = 6.6 Hz, 6H, CH_2), 3.38–3.34 (m, 6H, CH_2), 3.29–3.26 (m, 3H, H-3), 2.55–2.51 (m, 6H, H-7), 2.08–2.00 (m, 30H, OAc), 1.62–1.49 (m, 12H, CH_2), 1.43–1.37 (m, 6H, CH_2); ^{13}C-NMR (50 MHz, CD_3OD): δ = 170.2 (s, C═O), 170.1 (s, 3C, C═O), 169.7 (s, 3C, C═O), 169.2 (s, 3C, C═O), 143.9 (s, 3C, C-triazole), 121.5 (d, 3C, C-triazole), 79.8 (d, 3C, C-1), 73.4 (d, 3C, C-2), 70.0, 69.3, 68.1 (t, 9C, CH_2, CCH_2O), 66.0 (d, 3C, C-3), 65.7 (d, 3C, C-7a), 63.1 (t, 3C, C-8), 62.6 (t, CH_2OAc), 62.4 (t, 3C, OCH_2triazole), 60.4 (d, 3C, C-6), 58.6 (t, 3C, C-5), 45.4 (s, CCH$_2$O), 35.8 (t, 3C, C-7), 28.2 (t, 6C, CH_2), 21.7 (t, 3C, CH_2), 18.5 (q, 10C, OAc); IR ($CHCl_3$): ν = 3033, 3002, 2929, 2859, 1370, 1236, 1212 cm^{-1}; MS (ESI): m/z calcd for $C_{73}H_{110}N_{12}O_{26}$ + Na^+ 1593.75 $[M + Na]^+$; found: 1593.72; elemental analysis calcd (%) for $C_{73}H_{110}N_{12}O_{26}$ (1571.72): C 55.78, H 7.05, N 10.69; found: C 55.66, H 6.67, N 10.35.

Polyhydroxylated trivalent iminosugar 20

A suspension of **19** (36 mg, 0.024 mmol) and ion exchange resin Ambersep 900 OH (600 mg) in 10 mL of methanol was slowly stirred at room temp. for 16 h. After filtration of the resin on Celite®, the solvent was removed under reduced pressure affording pure **20** (28 mg, 0.024 mmol, 100% yield) as a yellow oil. $[\alpha]_D^{22}$ = +10.0 (c = 0.3 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 7.99 (s, 3H, H-triazole), 5.32–5.29 (m, 3H, H-6), 4.47 (s, 6H, OCH_2triazole), 3.84–3.76 (m, 6H, H-1, H-2), 3.69 (dd, J = 11.7, 3.4 Hz, 3H, Ha-8), 3.61–3.48 (m, 9H, Hb-8, H-7a, Ha-5), 3.46 (s, 2H, CH_2OH), 3.46–3.38 (m, 3H, Hb-5), 3.42 (t, J = 6.6 Hz, 6H, CH_2), 3.29–3.26 (m, 12H, CH_2, CCH_2O), 2.91–2.88 (m, 3H, H-3), 2.48 (ddd, J = 12.7, 7.6, 5.1 Hz, 3H, Ha-7), 2.40 (dt, J = 12.7, 6.8 Hz, 3H, Hb-7), 1.53–1.48 (m, 12H, CH_2), 1.34–1.28 (m, 6H, CH_2); ^{13}C-NMR (50 MHz, CD_3OD): δ = 144.9 (s, 3C, C-triazole), 123.0 (d, 3C, C-triazole), 80.5 (d, 3C, C-1), 77.5 (d, 3C, C-2), 72.5 (d, 3C, C-3), 71.0, 70.2 (t, 6C, CH_2), 69.5 (t, 3C, CCH_2O), 68.0 (d, 3C, C-7a), 63.2 (t, 3C, OCH_2triazole), 62.0 (t, CH_2OH), 61.3 (t, 3C, C-8), 60.7 (d, 3C, C-6), 59.8 (t, 3C, C-5), 45.2 (s, CCH$_2$O), 36.2 (t, 3C, C-7), 29.0 (t, 6C, CH_2), 22.6 (t, 3C, CH_2); MS (ESI): m/z calcd for $C_{53}H_{90}N_{12}O_{16}$ + Na^+ 1173.65 $[M + Na]^+$; found: 1173.59; elemental analysis calcd (%) for $C_{53}H_{90}N_{12}O_{16}$ (1151.35): C 55.29, H 7.88, N 14.60; found: C 55.11, H 7.65, N 14.32.

Benzylated trivalent iminosugar 22

To a solution of **4** (141 mg, 0.291 mmol) in 3 mL of a 2 : 1 THF–H_2O mixture, $CuSO_4$ (30 mol%, 4 mg, 0.026 mmol), sodium ascorbate (60 mol%, 10 mg, 0.052 mmol) and **21** (18 mg, 0.087 mmol) were added. The reaction mixture was heated in a MW reactor at 80 °C for 45 min, until TLC analysis (PE–AcOEt 5 : 1) showed the disappearance of the starting material (R_f = 0.55) and formation of a new product (R_f = 0.00). After filtration on Celite®, the solvent was removed under reduced pressure and the crude was purified by FCC (CH_2Cl_2–MeOH 12 : 1) affording pure **22** (R_f = 0.37, 143 mg, 0.086 mmol, 99%) as a yellow oil. $[\alpha]_D^{24}$ = +6.0 (c = 0.3 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.76 (s, 3H, H-triazole), 7.34–7.23 (m, 45H, H-Ar), 5.30–5.20 (m, 3H, H-6), 4.76 (s, 2H, OCH_2triazole), 4.67–4.45 (m, 22H, H-Bn, OCH_2triazole), 4.08–4.05 (m, 3H, H-2), 3.92–3.88 (m, 3H, H-1), 3.82 (pquint, J = 5.0 Hz, 1H, CH_2CHCH_2), 3.76–3.50 (m, 13H, H-7a, H-8, OCH_2CHCH_2O), 3.44–3.36 (m, 6H, H-5), 3.20–3.10 (m, 3H, H-3), 2.44–2.34 (m, 3H, Ha-7), 2.26–2.16 (m, 3H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 144.9 (s, 3C, C-triazole), 138.4, 138.1, 137.8 (s, 9C, C-Ar), 128.5–127.6 (d, 45C, C-Ar), 121.7 (d, 3C, C-triazole), 88.4 (d, 3C, C-1), 85.9 (d, 3C, C-2), 77.4 (d,

CH_2CHCH_2), 73.3, 72.4, 72.0 (t, 11C, C-Bn, OCH_2CHCH_2O), 70.4 (t, 3C, C-8), 69.1 (d, 3C, C-3), 66.8 (d, 3C, C-7a), 64.9, 64.8, 63.9 (t, OCH_2triazole), 61.3 (d, 3C, C-6), 60.3 (t, 3C, C-5), 38.0 (t, 3C, C-7); IR ($CHCl_3$): ν = 3088, 3066, 3032, 2866, 1496, 1453, 1365, 1249, 1207, 1111 cm^{-1}. Elemental analysis calcd (%) for $C_{99}H_{110}N_{12}O_{12}$ (1660.01): C 71.63, H 6.68, N 10.13; found: C 71.51, H 6.60, N 10.08.

Peracetylated trivalent iminosugar 23

To a solution of **22** (109 mg, 0.066 mmol) in 30 mL of methanol, 60 mg of 10% Pd/C and four drops of 37% HCl were added under a nitrogen atmosphere, then the mixture was stirred under a hydrogen atmosphere at room temp. for 5 days. TLC analysis (CH_2Cl_2–MeOH 10 : 1) showed the disappearance of the starting material (R_f = 0.67) and formation of a new product (R_f = 0.00) The mixture was filtered through Celite® and the solvent was removed under reduced pressure. The crude yellow oil was dissolved in pyridine (3 mL), acetic anhydride (2 mL) was added and the reaction mixture was stirred at room temp. for two days. Then, after concentration under reduced pressure, the crude mixture was purified by FCC (CH_2Cl_2–MeOH 10 : 1) affording pure **23** (R_f = 0.43, 44 mg, 0.036 mmol, 54%) as a pale yellow oil. $[\alpha]_D^{22}$ = −2.3 (c = 0.94 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.79 (s, H-triazole), 7.73 (s, H-triazole), 7.72 (s, H-triazole), 5.32 (t, J = 5.3 Hz, 6H, H-6, H-2), 5.03 (t, J = 5.2 Hz, 3H, H-1), 4.74 (s, 2H, OCH_2triazole), 4.60 (s, 4H, OCH_2triazole), 4.17–4.06 (m, 6H, H-8), 3.81 (pquint, J = 5.1 Hz, 1H, CH_2CHCH_2), 3.69–3.48 (m, 7H, H-7a, OCH_2CHCH_2O), 3.45–3.33 (m, 6H, H-5), 3.19 (pq, J = 5.9 Hz, 3H, H-3), 2.54–2.36 (m, 6H, H-7), 2.06–2.04 (s, 27H, OAc); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 170.7, 170.4, 169.7 (s, 9C, C═O), 145.4, 145.0, 144.9 (s, C-triazole), 121.8, 121.6, 121.5 (d, C-triazole), 80.8 (d, 3C, C-1), 78.6 (d, 3C, C-2), 77.3 (d, CH_2CHCH_2), 70.2 (t, 2C, OCH_2CHCH_2O), 67.3 (d, 3C, C-3), 66.5 (d, 3C, C-7a), 64.8 (t, 4C, C-8 and OCH_2triazole), 64.6, 63.8 (t, OCH_2triazole), 61.1 (d, 3C, C-6), 60.0 (t, 3C, C-5), 37.4 (t, 3C, C-7), 20.9 (q, 9C, CH_3); IR ($CHCl_3$): ν = 2942, 2867, 1740, 1626, 1453, 1373, 1233, 1108, 1046 cm^{-1}; MS (ESI): m/z calcd for $C_{54}H_{74}N_{12}O_{21}$ + Na^+ 1249.50 $[M + Na]^+$; found: 1249.50; elemental analysis calcd (%) for $C_{54}H_{74}N_{12}O_{21}$ (1660.01): C 52.85, H 6.08, N 13.70; found: C 52.87, H 5.94, N 13.72.

Polyhydroxylated trivalent iminosugar 24

A suspension of **23** (25 mg, 0.02 mmol) and ion exchange resin Ambersep 900 OH (1000 mg) in 10 mL of methanol was slowly stirred at room temp. for 16 h. After filtration through Celite®, the solvent was removed under reduced pressure and the crude was purified through size exclusion chromatography by Sephadex G-10 (eluting with H_2O) affording pure **24** (9 mg, 0.011 mmol, 55% yield) as a yellow oil. $[\alpha]_D^{25}$ = +20.8 (c = 0.48 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 8.10–8.09 (m, 3H, H-triazole), 5.33 (quint, J = 5.7 Hz, 3H, H-6), 4.73–4.71 (m, 2H, OCH_2triazole), 4.60–4.58 (m, 4H, OCH_2triazole), 3.87 (t, J = 7.4 Hz, 3H, H-2), 3.80 (t, J = 7.2 Hz, 3H, H-1), 3.78–3.75 (m, 1H, OCH_2CHCH_2O), 3.73 (dd, J = 11.3, 3.1 Hz, 3H, Ha-8), 3.64–3.54 (m, 4H, OCH_2CHCH_2O), 3.58 (dd, J = 11.3, 5.8 Hz, 3H, Hb-8), 3.50 (pq, J = 6.8 Hz, 3H, H-7a), 3.42–3.36 (m, 6H, H-5), 2.80 (ddd, J = 8.4, 5.5, 3.1 Hz, 3H, H-3), 2.52–2.44 (m, 3H, Ha-7), 2.44–2.34 (m, 3H, Hb-7); ^{13}C-NMR (50 MHz, CD_3OD): δ = 145.9 (s, 3C, C-triazole), 124.4 (d, 3C, C-triazole), 82.6 (d, 3C, C-1), 79.0 (d, 3C, C-2), 78.9 (d, OCH_2CHCH_2O), 72.9 (d, 3C, C-3), 71.2 (t, 2C, OCH_2CHCH_2O), 68.1 (d, 3C, C-7a), 65.2, 65.1, 64.2 (t, OCH_2triazole), 63.8 (t, 3C, C-8), 62.3 (d, 3C, C-6), 61.0 (t, 3C, C-5), 38.0 (t, 3C, C-7); MS (ESI): m/z calcd for $C_{36}H_{56}N_{12}O_{12}$ + Na^+ 871.40 $[M + Na]^+$; found: 871.50; elemental analysis calcd (%) for $C_{36}H_{56}N_{12}O_{12}$ (848.90): C 50.93, H 6.65, N 19.80; found: C 50.53, H 6.45, N 19.65.

Synthesis of dendrimeric nonavalent scaffold 27

To a solution of **25** (75 mg, 0.32 mmol) and DIPEA (139 μL, 0.81 mmol) in dry CH_2Cl_2 (1 mL), cooled to 0 °C and under a nitrogen atmosphere, a solution of 26 (24 mg, 0.09 mmol) in dry CH_2Cl_2 (1 mL) was added. The reaction mixture was stirred at room temp. for 15 h, until TLC analysis (CH_2Cl_2–MeOH 10 : 1) showed the disappearance of the starting material (R_f = 0.96) and formation of a new product (R_f = 0.84). After washing with HCl 0.5 M (10 mL) and H_2O (2 × 10 mL), the combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The crude mixture was purified by FCC (hexane–AcOEt 2 : 1) affording pure **27** (R_f = 0.17, 53 mg, 0.060 mmol, 68% yield) as a white solid.

Benzylated nonavalent iminosugar 28

To a solution of **4** (218 mg, 0.45 mmol) in 2.4 mL of a 2 : 1 THF–H_2O mixture. $CuSO_4$ (30 mol%, 2 mg, 0.014 mmol), sodium ascorbate (60 mol%, 6 mg, 0.029 mmol) and **27** (41 mg, 0.048 mmol) were added. The reaction mixture was heated in a MW reactor at 80 °C for 45 min, until TLC analysis (PE–AcOEt 3 : 1) showed the disappearance of the starting material (R_f = 0.35) and formation of a new product (R_f = 0.00). After a filtration through Celite®, the solvent was removed under reduced pressure and the crude was purified by FCC (CH_2Cl_2–MeOH 20 : 1) affording pure **28** (R_f = 0.31, 214 mg, 0.041 mmol, 91%) as a yellow oil. $[\alpha]_D^{22}$ = +3.21 (c = 0.53 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 8.34 (s, 9H, H-triazole), 7.69 (s, 3H, H-Ar), 7.32–7.19 (m, 135H, H-Ar), 5.19 (m, 9H, H-6), 4.66–4.55 (AB system, J = 12.2 Hz, 18H, Bn), 4.54–4.49 (m, 36H, H-Bn, OCH_2triazole), 4.53–4.42 (AB system, J = 12.2 Hz, 18H, Bn), 4.05 (t, J = 5.4 Hz, 9H, H-2), 3.94 (s, 18H, CCH_2O), 3.89–3.86 (m, 9H, H-1), 3.69–3.66 (m, 9H, H-7a), 3.56 (dd, J = 9.3, 5.4 Hz, 9H, Ha-8), 3.50 (dd, J = 9.2, 6.4 Hz, 9H, Hb-8), 3.34–3.32 (m, 18H, H-5), 3.11–3.08 (m, 9H, H-3), 2.39–2.34 (m, 9H, Ha-7), 2.23–2.15 (m, 9H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 166.4 (s, 3C, C═O), 144.7 (s, 9C, C-triazole), 138.4, 138.2, 137.9 (s, 30C, C-Ar), 135.9 (s, 3C, Ar), 128.4–127.6 (d, 138C, C-Ar), 121.9 (d, 9C, C-triazole), 88.4 (d, 9C, C-1), 85.8 (d, 9C, C-2), 73.2 (t, 9C, C-Bn), 72.4 (t, 9C, C-Bn), 72.2 (t, 9C, C-Bn), 71.9 (t, 9C, C-8), 68.9 (t or d, 18C, CCH_2O, C-3), 66.8 (d, 9C, C-7a), 64.8 (t, 9C, OCH_2triazole), 61.0 (d, 9C, C-6), 60.8, (s, 3C, HNC-), 60.2 (t, 9C, C-5), 37.8 (t, 9C, C-7); IR ($CHCl_3$): ν = 3088, 3065, 3031, 2923, 2866, 1667, 1497, 1454, 1346, 1111, 1095 cm^{-1}. Elemental analysis calcd (%) for $C_{309}H_{339}N_{39}O_{39}$

(5223.24): C 71.05, H 6.54, N 10.46; found: C 70.54, H 6.15, N 10.13.

(1*R*,2*R*,3*R*,6*R*,7a*R*)-1,2-Bis(benzyloxy)-3-[(benzyloxy)methyl]-6-(methylsulfonyloxy)-hexahydro-1*H*-pyrrolizine (29)

To a stirred solution of **3** (326 mg, 0.71 mmol) in dry CH_2Cl_2 (12 mL), NEt_3 (298 μL, 2.13 mmol) was added under a nitrogen atmosphere, and, at 0 °C, MsCl (71 μL, 0.92 mmol) was added dropwise. The solution was left stirring at room temp. for 2 h. TLC analysis (AcOEt–PE 3 : 1) showed the disappearance of the starting material (R_f = 0.16) and formation of a new product (R_f = 0.43). Then 20 mL of H_2O was added, the mixture was extracted with CH_2Cl_2 and the organic layers were dried on Na_2SO_4. Filtration through cotton and evaporation under reduced pressure afforded crude **29** that was purified by FCC (AcOEt–PE 1 : 1) affording pure **29** (R_f = 0.23, 317 mg, 0.59 mmol, 83% yield) as a colorless oil. $[\alpha]_D^{25}$ = −11.5 (c = 0.2 in $CHCl_3$); ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.37–7.25 (m, 15H, H-Ar), 5.20 (ddd, J = 8.3, 5.9, 2.9 Hz, 1H, H-6), 4.71–4.56 (AB system, J = 11.7 Hz, 2H, H-Bn), 4.59 (s, 2H, H-Bn), 4.57–4.50 (AB system, J = 11.7 Hz, 2H, Bn), 4.05–3.99 (m, 2H, H-1 e H-2), 3.59 (dd, J = 9.8, 3.9 Hz, 1H, Ha-8), 3.52 (dt, J = 8.3, 5.4 Hz, 1H, H-7a), 3.56 (dd, J = 9.8, 6.3 Hz, 1H, Hb-8), 3.33 (dt, J = 13.7, 2.0 Hz, 1H, Ha-5), 3.29–3.25 (m, 1H, H-3), 3.21 (dd, J = 13.7, 4.9 Hz, 1H, Hb-5), 2.81 (s, 3H, Ms), 2.43 (ddd, J = 13.7, 7.8, 6.8 Hz, 1H, Ha-7), 1.80 (dt, J = 13.7, 6.3 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, $CDCl_3$): δ = 138.1, 138.1, 138.0 (s, C-Ar), 128.4–127.8 (d, 15C, C-Ar), 88.0 (d, C-1), 85.0 (d, C-2), 82.8 (d, C-6), 73.4 (t, C-Bn), 72.8 (t, C-Bn), 72.3 (t, C-Bn), 72.3 (t, C-8), 67.9 (d, C-3), 66.5 (d, C-7a), 59.5 (t, C-5), 38.7 (q, OMs), 37.8 (t, C-7); IR ($CHCl_3$): ν = 3088, 3053, 2986, 2831, 1951, 1808, 1606, 1422, 1268, 1153; MS (ESI): m/z calcd for $C_{30}H_{35}NO_6S$ + H^+ 538.22 $[M + H]^+$; found: 538.42; elemental analysis calcd (%) for $C_{30}H_{35}NO_6S$ (537.67): C 67.02, H 6.56, N 2.61; found: C 67.47, H 6.87, N 2.31.

(1*R*,2*R*,3*R*,6*R*,7a*R*)-6-Methansulfonyloxy-3-hydroxymethylhexahydro-1*H*-pyrrolizidine-1,2-diol (30)

To a solution of **29** (264 mg, 0.49 mmol) in 10 mL of methanol, 132 mg of 10% Pd/C and two drops of 37% HCl were added under a nitrogen atmosphere, then the mixture was stirred under a hydrogen atmosphere at room temp. for 16 h. ^{1}H NMR control showed the disappearance of the starting material, then the mixture was filtered through Celite® and the solvent was removed under reduced pressure affording a crude yellow oil (168 mg). Free amine was obtained by passing the hydrochloride salt through a Dowex 50WX8 ion-exchange resin. Elution with 6% NH_4OH afforded the free base **30** (118 mg, 0.44 mmol, 90% yield) as a colourless oil. $[\alpha]_D^{29}$ = +19.0 (c = 0.625 in MeOH); ^{1}H-NMR (400 MHz, CD_3OD): δ = 5.26–5.23 (m, 1H, H-6), 3.95 (t, J = 8.0 Hz, 1H, H-1), 3.75 (dd, J = 11.2, 3.1 Hz, 1H, Ha-8), 3.71 (dd, J = 9.0, 8.3 Hz, 1H, H-2), 3.55 (dd, J = 11.2, 5.9 Hz, 1H, Hb-8), 3.36 (dt, J = 13.9, 1.6 Hz, 1H, Ha-5), 3.34 (dd, J = 8.0, 3.7 Hz, 1H, H-7a), 3.16 (dd, J = 13.9, 3.7 Hz, 1H, Hb-5), 3.12 (s, 3H, OMs), 3.02 (ddd, J = 9.1, 6.0, 3.1 Hz, 1H, H-3), 2.35–2.23 (m, 2H, H-7); ^{13}C-NMR (50 MHz, CD_3OD): δ = 83.7 (d, C-6), 80.8 (d, C-1), 76.8 (d, C-2), 70.2 (d, C-3), 66.5 (d, C-7a), 62.8 (t, C-8), 59.4 (t, C-5), 37.0 (q, OMs), 36.2 (t, C-7); MS (ESI): m/z calcd for $C_9H_{17}NO_6S$ + Na^+ 290.07 $[M + Na]^+$; found: 290.00; elemental analysis calcd (%) for $C_9H_{17}NO_6S$ (267.30): C 40.44, H 6.41, N 5.24; found: C 40.31, H 6.87, N 4.75.

(1*R*,2*R*,3*R*,6*S*,7a*R*)-6-Azido-3-hydroxymethylhexahydro-1*H*-pyrrolizidine-1,2-diol (31)

To a solution of **30** (108 mg, 0.40 mmol) in 4 mL of dry DMF sodium azide (52 mg, 0.80 mmol) was added. The reaction mixture was stirred at 80 °C for 3 h, until TLC analysis (CH_2Cl_2–MeOH 3 : 1) showed the disappearance of the starting material (R_f = 0.58) and formation of a new product (R_f = 0.50). Then, the solvent was removed under reduced pressure and the crude was purified by FCC (CH_2Cl_2–MeOH 4 : 1) affording pure **31** (R_f = 0.38, 82 mg, 0.38 mmol, 95%) as a yellow oil. $[\alpha]_D^{24}$ = +17.5 (c = 0.53 in MeOH); ^{1}H-NMR (400 MHz, D_2O): δ = 4.31–4.27 (m, 1H, H-6), 3.73–3.66 (m, 2H, H-1, H-2), 3.63 (dd, J = 11.7, 3.4 Hz, 1H, Ha-8), 3.46 (dd, J = 11.7, 6.9 Hz, 1H, Hb-8), 3.17 (pq, J = 7.1 Hz, 1H, H-7a), 2.95 (dd, J = 11.7, 2.9 Hz, 1H, Ha-5), 2.80 (dd, J = 11.7, 4.9 Hz, 1H, Hb-5), 2.63 (ddd, J = 9.2, 7.1, 3.5 Hz, 1H, H-3), 2.02 (ddd, J = 13.1, 6.8, 3.4 Hz, 1H, Ha-7), 1.88 (ddd, J = 13.1, 7.8, 5.3 Hz, 1H, Hb-7); ^{13}C-NMR (50 MHz, D_2O) δ = 80.3 (d, C-1), 77.9 (d, C-2), 70.0 (d, C-3), 65.2 (d, C-7a), 62.8 (d, C-6), 62.4 (t, C-8), 59.1 (t, C-5), 35.3 (t, C-7); MS (ESI): m/z calcd for $C_8H_{14}N_4O_3$ + Na^+ 237.10 $[M + Na]^+$; found: 237.08; elemental analysis calcd (%) for $C_8H_{14}N_4O_3$ (214.22): C 44.85, H 6.59, N 26.15; found: C 44.74, H 6.45, N 26.09.

Polyhydroxylated nonavalent iminosugar 32

To a solution of **31** (82 mg, 0.38 mmol) in 3 mL of a 2 : 1 THF–H_2O mixture, $CuSO_4$ (30 mol%, 2 mg, 0.012 mmol), sodium ascorbate (60 mol%, 5 mg, 0.024 mmol) and **27** (34 mg, 0.04 mmol) were added. The reaction mixture was heated in a MW reactor at 100 °C for 2 h, until TLC analysis (EP–AcOEt 2 : 1) showed the disappearance of the starting material (R_f = 0.26) and formation of a new product (R_f = 0.00). After a filtration through Celite®, the solvent was removed under reduced pressure and the crude was purified by FCC (MeOH–CH_2Cl_2–NH_4OH 2 : 1 : 1) affording pure **32** (R_f = 0.26, 62 mg, 0.022 mmol, 55%) as a waxy white solid. $[\alpha]_D^{30}$ = +3.0 (c = 0.5 in H_2O); ^{1}H-NMR (400 MHz, D_2O): δ = 7.87 (s, 9H, H-triazole), 7.79 (s, 3H, H-Ar), 5.08–5.05 (m, 9H, H-6), 4.45 (s, 18H, OCH_2-triazole), 3.76 (t, J = 7.4 Hz, 9H, H-1), 3.72–3.69 (m, 9H, H-2), 3.70 (s, 18H, CCH_2O), 3.62 (dd, J = 11.8, 3.4 Hz, 9H, Ha-8), 3.47 (dd, J = 11.8, 6.4 Hz, 9H, Hb-8), 3.33 (q, J = 6.8 Hz, 9H, H-7a), 3.22 (dd, J = 12.2, 6.1 Hz, 9H, Ha-5), 3.13 (dd, J = 12.0, 5.8 Hz, 9H, Hb-5), 2.75–2.71 (m, 9H, H-3), 2.34–2.22 (m, 18H, H-7); ^{13}C-NMR (50 MHz, D_2O): δ = 168.2 (s, 3C, C═O), 144.0 (s, 9C, C-triazole), 135.3 (s, 3C, Ar), 129.1 (d, 3C, Ar), 124.0 (d, 9C, C-triazole), 80.1 (d, 9C, C-1), 76.9 (d, 9C, C-2), 70.1 (d, 9C, C-3), 67.2 (t, 9C, CCH_2O), 66.0 (d, 9C, C-7a), 63.5 (t, 9C, OCH_2-triazole), 62.0 (t, 9C, C-8), 60.7 (s, 3C, CCH$_2$O), 60.1 (d, 9C, C-6), 59.0 (t, 9C, C-5), 35.6 (t, 9C, C-7); MS (ESI): m/z calcd for $[(C_{120}H_{177}N_{39}O_{39} + 3Na)/3]^{3+}$ 952.42 $[(M + 3Na)/3]^+$; found: 953.17; elemental analysis calcd (%) for $C_{120}H_{177}N_{39}O_{39}$

(2789.93): C 51.66, H 6.39, N 19.59; found: C 51.59, H 6.33, N 19.42.

Glycosidase inhibition assays

The experiments were performed essentially as previously described.[41] The percentage of inhibition towards the corresponding glycosidase was determined in the presence of 1 mM of the inhibitor on the well. Each enzymatic assay (final volume 0.12 mL) contained 0.01 to 0.5 units mL^{-1} of the enzyme and 10 mM aqueous solution of the appropriate *p*-nitrophenyl glycoside substrate buffered to the optimal pH of the enzyme. The enzyme and the inhibitor were preincubated for 5 min at rt, and the reaction started by the addition of the substrate. After 20 min of incubation at 37 °C, the reaction was stopped by the addition of 0.1 mL of sodium borate buffer (pH 9.8). The *p*-nitrophenolate formed was measured by visible absorption spectroscopy at 405 nm. Under these conditions, the *p*-nitrophenolate released led to optical densities linear with both reaction time and concentration of the enzyme. For the best inhibitors (% inhibition ≥ 80), the IC_{50} value (concentration of inhibitor required for 50% inhibition of enzyme activity) and K_i towards the corresponding glycosidase were calculated. IC_{50} values were calculated from plots of percentage of inhibition *versus* inhibitor concentration. The K_i values were determined from the Lineweaver–Burk plots (see ESI†). Each experiment (%, IC_{50} and K_i) was performed in duplicate and the average values were given.

Acknowledgements

We thank MIUR-Italy (PRIN 2008, 200824M2HX, and PRIN 2010-2011, 2010L9SH3 K 006) and Ente Cassa di Risparmio di Firenze (CRF, 2009/3141) for financial support. The research leading to these results has received funding from the European Research Council under the European Union's Seventh Framework Programme (FP7/2007-2013)/ERC grant agreement no. [291349] on photonic micro robotics. We thank Ministerio de Economia y Competitividad (CTQ2012-31247) of Spain, Junta de Andalucía (FQM 345) and European Community FP7 program (HEALTH-F2-2011-256986-project acronym PANACREAS) for financial support. We also thank Mr Simone Pisano for preliminary experiments.

Notes and references

1 (*a*) Y. C. Lee and T. R. Lee, *Acc. Chem. Res.*, 1995, **28**, 321–327; (*b*) M. Mammen, S.-K. Choi and G. M. Whitesides, *Angew. Chem., Int. Ed.*, 1998, **37**, 2754–2794; (*c*) J. J. Lundquist and E. J. Toone, *Chem. Rev.*, 2002, **102**, 555–578; (*d*) C. Fasting, C. A. Schalley, M. Weber, O. Seitz, S. Hecht, B. Koksch, J. Dernedde, C. Graf, E.-W. Knapp and R. Haag, *Angew. Chem., Int. Ed.*, 2012, **51**, 10472–10498.

2 Selected reviews: (*a*) S.-K. Choi, in *Synthetic Multivalent molecules: concepts and biomedical applications*, John Wiley & Sons, Inc., Hoboken, New Jersey, 2004; (*b*) L. L. Kiessling, J. E. Gestwicki and L. E. Strong, *Angew. Chem., Int. Ed.*, 2006, **45**, 2348–2368; (*c*) A. Imberty, Y. M. Chabre and R. Roy, *Chem. – Eur. J.*, 2008, **14**, 7490–7499; (*d*) D. Deniaud, K. Julienne and S. G. Gouin, *Org. Biomol. Chem.*, 2011, **9**, 966–979.

3 P. Compain and O. R. Martin, in *Iminosugars: from Synthesis to Therapeutic Applications*, Wiley VCH, New York, 2007.

4 (*a*) B. A. Johns and C. R. Johnson, *Tetrahedron Lett.*, 1998, **39**, 749–752; (*b*) A. Lohse, K. B. Jensen, K. Lundgren and M. Bols, *Bioorg. Med. Chem.*, 1999, **7**, 1965–1971; (*c*) T. Wennekes, R. J. B. H. N. Van der Berg, K. M. Bonger, W. E. Donker-Koopman, A. Ghisaidoobe, G. A. Van der Marel, A. Strijland, J. M. F. G. Aerts and H. S. Overkleeft, *Tetrahedron: Asymmetry*, 2009, **20**, 836–846.

5 J. Diot, M. I. García-Moreno, S. G. Gouin, C. Ortiz Mellet, K. Haupt and J. Kovensky, *Org. Biomol. Chem.*, 2009, **7**, 357–363.

6 P. Compain, C. Decroocq, J. Iehl, M. Holler, D. Hazelard, T. Mena Barragán, C. Ortiz Mellet and J.-F. Nierengarten, *Angew. Chem., Int. Ed.*, 2010, **49**, 5753–5756.

7 (*a*) C. Decroocq, D. Rodríguez-Lucena, V. Russo, T. Mena Barragán, C. Ortiz Mellet and P. Compain, *Chem. – Eur. J.*, 2011, **17**, 13825–13831; (*b*) C. Decroocq, A. Joosten, R. Sergent, T. Mena Barragán, C. Ortiz Mellet and P. Compain, *ChemBioChem*, 2013, **14**, 2038–2049.

8 Y. Brissonet, C. Ortiz Mellet, S. Morandat, M. I. Garcia Moreno, D. Deniaud, S. E. Matthews, S. Vidal, S. Šesták, K. El Kirat and S. G. Gouin, *J. Am. Chem. Soc.*, 2013, **135**, 18427–18435.

9 E. Moreno-Clavijo, A. T. Carmona, A. J. Moreno-Vargas, L. Molina, D. W. Wright, G. J. Davies and I. Robina, *Eur. J. Org. Chem.*, 2013, 7328–7336.

10 For our preliminary efforts aimed to synthesizing multivalent pyrrolidine-based ligands on calixarene cores, see: (*a*) M. Marradi, S. Cicchi, F. Sansone, A. Casnati and A. Goti, *Beilstein J. Org. Chem.*, 2012, **8**, 951–957; (*b*) F. Cardona, G. Isoldi, F. Sansone, A. Casnati and A. Goti, *J. Org. Chem.*, 2012, **77**, 6980–6988.

11 R. J. Pieters, *Org. Biomol. Chem.*, 2009, **7**, 2013–2025.

12 (*a*) J. E. Gestwicki, C. W. Cairo, L. E. Strong, K. A. Oetjen and L. L. Kiessling, *J. Am. Chem. Soc.*, 2002, **124**, 14922–14933; (*b*) M. L. Wolfenden and M. J. Cloninger, *Bioconjugate Chem.*, 2006, **17**, 958–966; (*c*) T. K. Dam and C. F. Brewer, *Biochemistry*, 2008, **47**, 8470–8476.

13 R. Rísquez-Cuadro, J. M. García Fernández, J.-F. Nierengarte and C. Ortiz Mellet, *Chem. – Eur. J.*, 2013, **19**, 16791–16803.

14 C. Decroocq, D. Rodríguez-Lucena, K. Ikeda, N. Asano and P. Compain, *ChemBioChem*, 2012, **13**, 661–664.

15 A. Joosten, C. Decroocq, J. de Sousa, J. Schneider, E. Etamé, A. Bodlenner, T. J. Butters and P. Compain, *ChemBioChem*, 2014, **15**, 309–319.

16 P. Compain, C. Decroocq, A. Joosten, J. de Sousa, D. Rodríguez-Lucena, T. D. Butters, J. Bertrand,

R. Clément, C. Boinot, F. Becq and C. Norez, *ChemBioChem*, 2013, **14**, 2050–2058.

17 For a recent review see: P. Compain and A. Bodlenner, *ChemBioChem*, 2014, **15**, 1239–1251.

18 (*a*) C. Parmeggiani, F. Cardona, L. Giusti, H.-U. Reissig and A. Goti, *Chem. – Eur. J.*, 2013, **19**, 10595–10604; (*b*) D. Martella, F. Cardona, C. Parmeggiani, F. Franco, J. A. Tamayo, I. Robina, E. Moreno-Clavijo, A. J. Moreno-Vargas and A. Goti, *Eur. J. Org. Chem.*, 2013, 4047–4056; (*c*) G. D'Adamio, A. Goti, C. Parmeggiani, E. Moreno-Clavijo, I. Robina and F. Cardona, *Eur. J. Org. Chem.*, 2011, 7155–7162; (*d*) C. Bonaccini, M. Chiocchioli, C. Parmeggiani, F. Cardona, D. Lo Re, G. Soldaini, P. Vogel, C. Bello, A. Goti and P. Gratteri, *Eur. J. Org. Chem.*, 2010, 5574–5585; (*e*) C. Parmeggiani, D. Martella, F. Cardona and A. Goti, *J. Nat. Prod.*, 2009, **72**, 2058–2060.

19 F. Cardona, C. Parmeggiani, E. Faggi, C. Bonaccini, P. Gratteri, L. Sim, T. M. Gloster, S. Roberts, G. J. Davies, D. R. Rose and A. Goti, *Chem. – Eur. J.*, 2009, **15**, 1627–1636.

20 (*a*) F. Cardona, A. Goti, C. Parmeggiani, P. Parenti, M. Forcella, P. Fusi, L. Cipolla, S. M. Roberts, G. J. Davies and T. M. Gloster, *Chem. Commun.*, 2010, **46**, 2629–2631; (*b*) M. Forcella, F. Cardona, A. Goti, C. Parmeggiani, L. Cipolla, M. Gregori, R. Schirone, P. Fusi and P. Parenti, *Glycobiology*, 2010, **20**, 1186–1195.

21 A. Brandi, F. Cardona, S. Cicchi, F. M. Cordero and A. Goti, *Chem. – Eur. J.*, 2009, **15**, 7808–7821.

22 (*a*) H. C. Kolb, M. G. Finn and K. B. Sharpless, *Angew. Chem., Int. Ed.*, 2001, **40**, 2004–2021; (*b*) V. C. Rostovtsev, L. G. Green, V. V. Fokin and K. B. Sharpless, *Angew. Chem., Int. Ed.*, 2002, **41**, 2596–2599; (*c*) C. W. Tornøe, C. Christensen and M. Meldal, *J. Org. Chem.*, 2002, **67**, 3057–3064.

23 For reviews, see: (*a*) V. D. Bock, H. Hiemstra and J. H. van Maarseveen, *Eur. J. Org. Chem.*, 2006, 51–68; (*b*) J. F. Lutz, *Angew. Chem., Int. Ed.*, 2007, **46**, 1018–1025; (*c*) M. V. Gil, M. J. Arevalo and O. Lopez, *Synthesis*, 2007, 1589–1620; (*d*) J. E. Moses and A. D. Moorhouse, *Chem. Soc. Rev.*, 2007, **36**, 1249–1262; (*e*) P. Wu and V. V. Fokin, *Aldrichimica Acta*, 2007, **40**, 7–17; (*f*) M. Meldal and C. W. Tornøe, *Chem. Rev.*, 2008, **108**, 2952–3015.

24 (*a*) F. Cardona, E. Faggi, F. Liguori, M. Cacciarini and A. Goti, *Tetrahedron Lett.*, 2003, **44**, 2315–2318; (*b*) A. T. Carmona, R. H. Wightman, I. Robina and P. Vogel, *Helv. Chim. Acta*, 2003, **86**, 3066–3073; (*c*) S. Desvergnes, S. Py and Y. Vallée, *J. Org. Chem.*, 2005, **70**, 1459–1462; (*d*) J. Revuelta, S. Cicchi, A. Goti and A. Brandi, *Synthesis*, 2007, 485–504; (*e*) E.-L. Tsou, Y.-T. Yeh, P.-H. Liang and W.-C. Cheng, *Tetrahedron*, 2009, **65**, 93–100; (*f*) X.-G. Hu, Y.-M. Jia, J. Xiang and C.-Y. Yu, *Synlett*, 2010, 982–986.

25 K. Kamikawa, T. Watanabe, A. Daimon and M. Uemura, *Tetrahedron*, 2000, **56**, 2325–2337.

26 (*a*) Y. Xia, Z. Fan, J. Yao, Q. Liao, W. Li, F. Qu and L. Peng, *Bioorg. Med. Chem. Lett.*, 2006, **16**, 2693–2698; (*b*) Y. Xia, W. Li, F. Qu, Z. Fan, X. Liu, C. Berro, E. Rauzy and L. Peng, *Org. Biomol. Chem.*, 2007, **5**, 1695–1701.

27 For a recent related example of CuAAC cycloaddition with an azidoindolizidine derivative, see: F. M. Cordero, P. Bonanno, S. Neudeck, C. Vurchio and A. Brandi, *Adv. Synth. Catal.*, 2009, **351**, 1155–1161.

28 T. Toba, K. Murata, T. Yamamura, S. Miyake and H. Annoura, *Tetrahedron Lett.*, 2005, **46**, 5043–5047.

29 For a review on the synthesis of aza-*C*-disaccharides, see: I. Robina and P. Vogel, *Synthesis*, 2005, 675–702.

30 For a review on the bioactivity and biocompatibility of the triazole linker, see: H. C. Kolb and K. B. Sharpless, *Drug Discovery Today*, 2003, **8**, 1128–1137.

31 Only traces of the desired adduct were detected in the crude mixture after 2 hours when the reaction was performed at room temperature.

32 Treatment of the crude mixture after hydrogenation with ion exchange resin DOWEX 50WX8 gave impure deprotected trivalent compound, while peracetylation with Ac_2O and Py afforded the acetylated derivative in low yield. Subsequent treatment with strongly basic resin Ambersep 900 OH again afforded impure deprotected final compound.

33 I. Papp, J. Dernedde, S. Enders and R. Haag, *Chem. Commun.*, 2008, 5851–5853.

34 (*a*) Y. Yamamoto, K. Kurihara, A. Yamada, M. Takahashi, Y. Takahashi and N. Miyaura, *Tetrahedron*, 2003, **59**, 537–542; (*b*) I. Kadota, M. Kawada, V. Gevorgyan and Y. Yamamoto, *J. Org. Chem.*, 1997, **62**, 7439–7446.

35 M. Mourer, F. Hapiot, S. Tilloy, E. Monflier and S. Menuel, *Eur. J. Org. Chem.*, 2008, 5723–5730.

36 Y. M. Chabre, C. Contino-Pépin, V. Placide, T. C. Shiao and R. Roy, *J. Org. Chem.*, 2008, **73**, 5602–5605.

37 M. C. Davis, *Synth. Commun.*, 2007, **37**, 1457–1462.

38 (*a*) Y. M. Chabre, D. Giguère, B. Blanchard, J. Rodrigue, S. Rocheleau, M. Neault, S. Rauthu, A. Papadopoulos, A. A. Arnold, A. Imberty and R. Roy, *Chem. – Eur. J.*, 2011, **17**, 6545–6562; (*b*) For a recent example of dendrimeric piperidine iminosugars see: A. Joosten, J. P. Schneider, M. L. Lepage, C. Tarnus, A. Bodlenner and P. Compain, *Eur. J. Org. Chem.*, 2014, 1866–1872.

39 Attempted purification of the crude mixture of hydrochloride salts after hydrogenation by size exclusion chromatography failed.

40 M. Niemietz, L. Perkams, J. Hoffman, S. Eller and C. Unverzagt, *Chem. Commun.*, 2011, **47**, 10485–10487.

41 (*a*) R. Saul, J. P. Chambers, R. J. Molyneux and A. D. Elbein, *Arch. Biochem. Biophys.*, 1983, **221**, 593–597; (*b*) A. Brandi, S. Cicchi, F. M. Cordero, R. Frignoli, A. Goti, S. Picasso and P. Vogel, *J. Org. Chem.*, 1995, **60**, 6806–6812.

42 (*a*) J.-B. Behr, A. Erard and G. Guillerm, *Eur. J. Org. Chem.*, 2002, 1256–1262; (*b*) J.-B. Behr, A. Gainvors-Claisse and A. Belarbi, *Nat. Prod. Res.*, 2007, **21**, 76–82.

43 The reaction to yield **7f** was performed in a MW reactor at 80 °C for 45 min.

