Synthesis and Structural Analysis of Cyclic Oligomers Consisting of Furanoid and Pyranoid ε-Sugar Amino Acids

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Cyclic oligomers composed of amide-linked furanoid (i.e., **1**, **3**) and pyranoid (i.e., **2**, **4**) ε-sugar amino acids (SAAs) were prepared by a cyclization/cleavage approach with use of the oxime resin. These cyclic homooligomers were constructed by use of the known *N*-Boc protected furanoid ε-SAA **11** and the novel pyranoid hydroxymethylene homologue **22**. Conformational analysis of cyclic trimer **1** by an unrestrained simulated annealing technique showed that the furanoid rings of the residues **I** flip between a twist (north, $P = 0°$) and an envelope (south, $P = 167°$) conformation. Furthermore, the side chains connecting the carbonyl functionality (i.e. C2) proved to be rigid, while the other side chains (C7) are conformationally flexible. Similar conformational behaviour is observed for the side chains of the pyranoid ε-SAA **II** residues in trimer **2**, but the pyranoid ring chair conformation remains stable during the calculation. These conformational details may have important implications in the future design of SAA-based artificial receptors or peptidomimetics.

Introduction

Naturally occurring cyclic structures provide an impetus for the design and synthesis of molecular receptors.[1] One type of cyclic oligomers found in nature are the cyclodextrins. These cyclic oligomers, composed of carbohydrate units, have been used in the areas of drug delivery, asymmetric synthesis, and chromatography, and also as enzyme mimetics.[2] Novel host molecules with differently shaped internal cavities have been obtained by replacement of the natural glycosidic bonds connecting the individual carbohydrate derivatives by other types of linkages. Vasella and co-workers reported the development of cyclic oligomers composed of acetylene-linked carbohydrate residues, which were shown to accommodate a nucleoside.[3] In addition, a first example of oligomers containing amide bonds instead of glycosidic linkages was disclosed recently.[4,5] These latter compounds, containing pyranoid δ-sugar amino acids (SAAs),[6] were prepared by the assembly of the linear sequences on a tritylchloropolystyrene resin by a Fmoc-based peptide synthesis technique, followed by cleavage from the resin and cyclization under high-dilution conditions.

As part of a program directed towards the synthesis and conformational analysis of SAA-containing oligomers, we present here the utility of a cyclization/cleavage strategy for the solid-phase synthesis of the novel cyclic SAA oligomers **1**−**4** (Figure 1). The cyclic trimers (**1**, **2**) and tetramers (**3**, **4**) contain the known 3,6-anhydro-amino-2,7-dideoxy-D-*allo*-

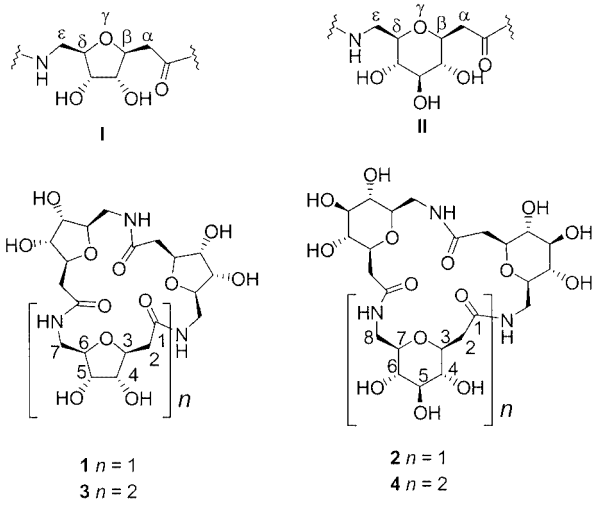

Figure 1. Cyclic homooligomers composed of furanoid ε-SAAs or pyranoid ε-SAAs

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heptulosonic acid (furanoid ε-SAA **I**)[7,8] residue and the new 3,7-anhydro-8-amino-2,8-dideoxy-D-*gulo*-D-*glycero*-octulonic acid (pyranoid ε-SAA **II**) residue. As furanoid and pyranoid carbohydrates[9] are known to adopt different conformations, we investigated the conformational behaviour of the trimers **1** and **2** by unrestrained simulated annealing calculations[10] in conjunction with NMR analysis.

Results

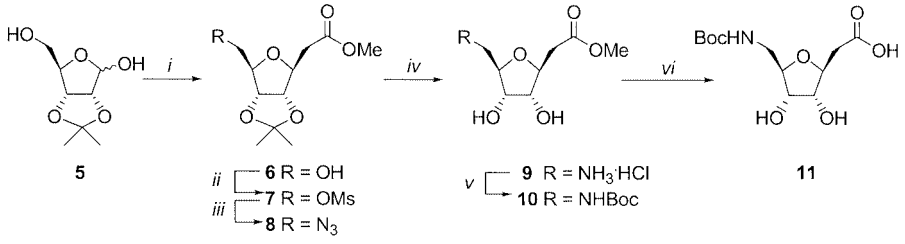
Synthesis of SAA Building Blocks 11 and 22

The synthesis of the Boc-protected furanoid SAA **11**[7] started with the Wittig olefination of 2,3-*O*-isopropylidene-protected ribose **5** with methyl (triphenylphosphoranylidene)acetate to afford the methyl ester **6** in 81% yield (Scheme 1).[11] Treatment of **6** with methanesulfonyl chloride in pyridine, followed by substitution of the intermediate mesylate with sodium azide, furnished the fully protected SAA **8** in a yield of 87%. Palladium-catalysed hydrogenation of compound **8** in the presence of hydrochloric acid resulted in the reduction of the azide functionality with concomitant cleavage of the isopropylidene protective group to give the amine **9** as its HCl salt. Finally, treatment of **9** with Boc_2O under Schotten−Baumann conditions and subsequent saponification of the methyl ester yielded the desired, suitably protected furanoid ε-SAA **11** in 73% yield over the last three steps.

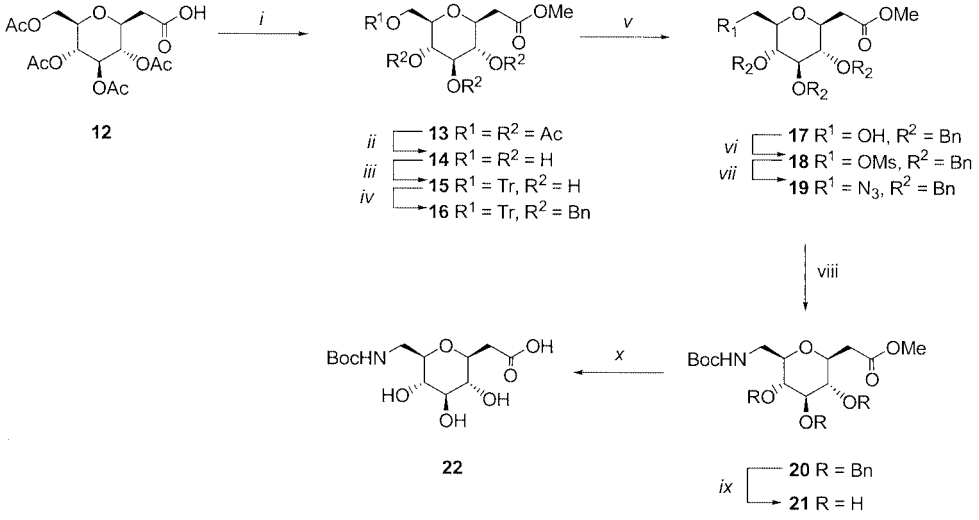
The synthesis of pyranoid ε-SAA **22** (Scheme 2) commences with condensation of the known[12] peracetylated C-glycoside **12** with methanol to furnish ester **13** in 83% yield. Subjection of **13** to Zemplén deacetylation conditions, followed by protection of the primary hydroxy group as the trityl ether, gave compound **15** in 85% yield over the two steps. Treatment of **15** with sodium hydride and benzyl bromide and subsequent removal of the trityl function with *p*TsOH furnished compound **17** in 88% yield. The free primary alcohol was now transformed into the azide as described above in the synthesis of the furanoid SAA (i.e., **17** to **19**) in 77% yield. The azide **19** was then converted into the Boc-protected amine **20** by means of a modified Staudinger reaction, with trimethylphosphane and 2-(*tert*-butoxycarbonyl)oxyimino-2-phenylacetonitrile (Boc-ON), in an excellent yield of 95%.[13] Palladium-catalysed hydrogenation of **20** and saponification of the intermediate methyl ester **21** yielded the desired pyranoid ε-SAA **22** in 20% overall yield based on **12**.

Solid-Support Synthesis of Cyclic SAA Oligomers 1−4

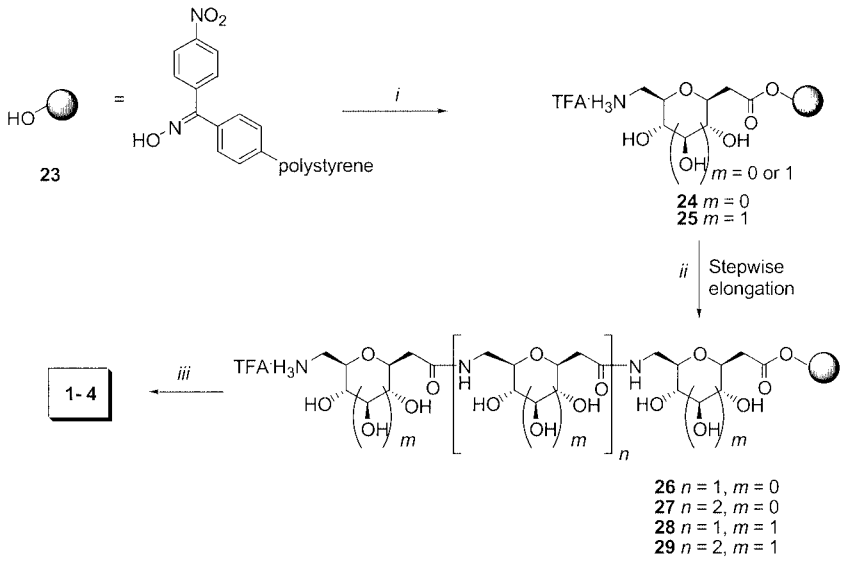
Previous work from our laboratory[7] demonstrated the use of a cyclization/cleavage strategy for the synthesis of



Scheme 1. Reagents and reaction conditions: (*i*) a) methyl (triphenylphosphoranylidene)acetate, CH_3CN, Δ; b) NaOMe (0.1 equiv.), MeOH (81%); (*ii*) MsCl, pyridine (93%); (*iii*) NaN_3, DMF, 75 °C, 1 h (94%); (*iv*) H_2, Pd/C, HCl/EtOH; (*v*) Boc_2O, $NaHCO_3$, Na_2CO_3, H_2O/dioxane (82%); (*vi*) NaOH, H_2O/dioxane (89%)



Scheme 2. Reagents and reaction conditions: (*i*) MeOH, DIC, DMAP, DCM, 16 h (83%); (*ii*) NaOMe, MeOH (quant.); (*iii*) TrCl, pyridine (85%); (*iv*) BnBr, NaH, DMF (88%); (*v*) *p*TsOH, DCM/MeOH (9:1); (*vi*) MsCl, pyridine (quant.); (*vii*) NaN_3, DMF, 75 °C, 2 h (77%); (*viii*) Me_3P, Boc-ON (95%); (*ix*) H_2/Pd-C, EtOH/EtOAc (1:1); (*x*) NaOH, dioxane (70%).



Scheme 3. Reagents and reaction conditions: (*i*) a) SAA **11** or **22** (5 equiv.), BOP (5 equiv.), DIPEA (6.5 equiv.), NMP/DCM (1:1, v/v), 2 h, 2 × ; b) 25% TFA, 1% TiPS, DCM; (*ii*) a) SAA **11** or **22** (5 equiv.), BOP (5 equiv.), DIPEA (6.5 equiv.), NMP/DCM (1:1, v/v), 45 min, 2 × ; b) 25% TFA, 1% TiPS, DCM; (*iii*) DIPEA (2 equiv.), AcOH (2 equiv.), DMF, 36 h

related artificial receptor molecules based on a combination of furanoid SAAs and naturally occurring amino acids.[14] The solid-phase synthesis of the cyclic oligomers **1**−**4** by the same strategy is presented in Scheme 3. In the case of cyclic trimer **1**, for instance, Kaiser's *p*-nitrobenzophenone oxime resin[15] **23** was loaded with SAA building block **11** under the agency of benzotriazol-1-yloxytris(dimethylamino)phosphonium hexafluorophosphate (BOP) and diisopropylethylamine (DIPEA).[16] The condensation step was performed twice in order to ensure optimal loading. The immobilised SAA **24** was treated with 25% TFA in DCM to remove the Boc group and subsequently condensed with the second SAA building block **11** (BOP/DIPEA).

By repetition of the deprotection/coupling/deprotection steps the linear immobilised precursor trimer **26** was obtained. Cyclization was accomplished by treatment of the resin with a 1:1 mixture of DIPEA and acetic acid for 36 h, furnishing the cyclic trimer **1** in 20% yield after RP-HPLC. Elongation of the immobilised linear trimer **26** to the corresponding tetramer **27**, followed by the described cyclisation/cleavage procedure, provided compound **3** with comparable efficiency. The immobilised linear precursors **28**−**29** were prepared from **25** in a similar fashion, following a sequential elongation using the corresponding pyranoid SAA building block **22**. Subsequent acid-catalysed cyclization gave compounds **2** and **4** in 5−10% yields after purification.

Conformational Analysis

NMR spectroscopy (in 10% D_2O in H_2O as solvent) in combination with molecular dynamics calculations was used to determine the three-dimensional structures of the cyclic oligomers **1** and **2**. On the NMR timescale, both compounds were shown to have C_n symmetry, such that only one set of signals for the SAA building blocks is observed. Because the observed ROEs can be caused by intra- and/or interresidual interactions the ROESY data should be interpreted with the utmost care. A three-dimensional model was used in order to correlate the ROE-derived distances.

Cyclic Trimer 1

The signals of three of the four protons of the furanoid rings in compound **1** overlap in the 1D spectrum (see Figure 2), making it impossible to calculate the conformations of the furan rings on the basis of the coupling constants.[17] The signals belonging to the protons of the two methylene side chains (i.e., H7a, H7b, H2a and H2b) have well resolved chemical shifts; however, diastereotopic assignment[18] of the individual protons was not possible at this point. The geminal proton at C2 resonating at higher field (arbitrarily assigned as H2b) displayed a large coupling constant with H3 ($J_{2b,3}$ = 9.3 Hz). This observation implies a rotamer population with one of the H2 protons in an *anti* relationship relative to H3.[19] The protons attached to C7 have small coupling constants with H6 ($J_{7a,6}$ = 2.9 and $J_{7b,6}$ = 5.5 Hz), giving no direct insight in the rotamer populations.

The ROESY spectrum of **1** showed medium cross-peaks between the amide proton and both neighbouring methyl-

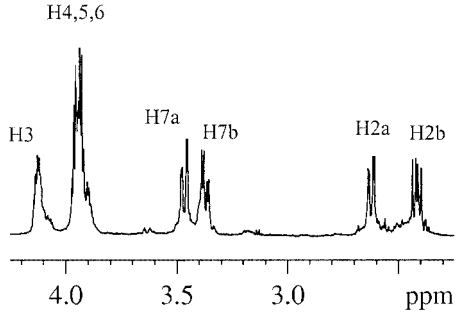


Figure 2. Part of the ^1H spectrum of the cyclic furanoid SAA trimer **1** (for the numbering see Figure 1) recorded at 600 MHz in 10% D_2O in H_2O

ene protons H7a and H7b. A strong cross-peak was observed between the amide proton and H2b, while H2a and H3 gave weak ROEs. The H2/HN cross-peaks most probably arise from the interaction between two sequential residues, indicating the energetically favourable *trans* conformation of the amide bonds. Because of our inability to assign the ROESY spectrum further it was decided not to use distance restraints for the molecular dynamics simulations. Instead, simulated annealing calculations[20] were carried out in order to search the entire conformational space accessible in solution for compound **1**, the outcome of which was correlated with the obtained NMR spectroscopic data in a later stage. To this end, one hundred and fifty low-energy conformers were obtained by gradual heating of a randomly build structure, starting from 300 and increasing to 800 K, and subsequent cooling down to 300 K. Each resulting structure was then minimised by use of steepest descent followed by conjugate gradient algorithms.

Analysis of the conformations of the furanoid rings in the 150 structures revealed that only two conformations occur. In one conformation, the five-membered rings adopt a twist conformation with C5 *endo* and C4 in the *exo* position (adopted nucleic acid nomenclature).[21] According to the concept of pseudorotation,[17] this is a north conformation [pseudorotational phase angle (P) = 0°]. The other conformation found for the furan ring is an envelope with C4 *endo*, corresponding to a south conformation (P = 167°). The occurrence of these two conformations during the simulation is illustrated by plotting the dihedral angle C3−C4−C5−C6 as a function of the frame number (Figure 3).

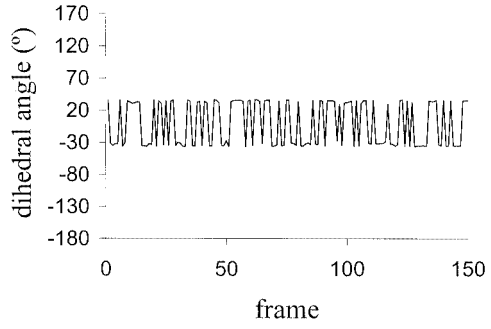


Figure 3. Graph of C3−C4−C5−C6 dihedral angle of one SAA residue during molecular simulations. A positive angle (35°) denotes the north conformation, and a negative angle (−35°) the south conformation.

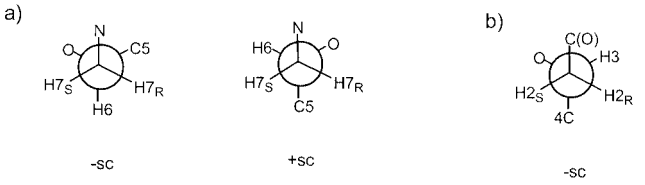


Figure 4. Newman projections of: a) the possible conformations around C6−C7, and b) the conformation around C2−C3 in **1**; the large coupling constant found for H2b in the NMR experiments corresponds to this *anti* relationship and H2b can therefore be assigned as $H2_S$.[18]

The resulting simulated structures were evaluated on the basis of their NMR spectroscopic data. All the conformations that did not have a *trans* amide bond and an *anti* relationship between H3 and one of the neighbouring protons H2, as indicated by the observed large coupling constant, were omitted. The geometries of the side chains at C6−C7 of the SAA residues in the selected structures were found to be either −synclinal (−sc)[22] or +sc (see Figure 4, a). There seems to be a small preference for the +sc conformation around C6−C7, which is corroborated by the difference in coupling constants for the two geminal protons at C7 (J = 2.9 and J = 5.5 Hz). The C2−C3 bond only adopts a −sc conformation in which $H2_S$ [17] has a *trans* relationship with H3 (Figure 4, b). The two sugar puckers, in combination with the geometries of the side chains, result in four different SAA conformations. These four different SAA conformations are represented in Figure 5, and a low-energy conformer of the cyclic trimer **1** is depicted in Figure 6.

Cyclic Trimer 2

In the 1D NMR spectrum of trimer **2** (Figure 7), all the ring protons displayed large coupling constants (J = 9 Hz), indicating a chair conformation of the pyranose rings with equatorial hydroxy groups (corresponding to the $^4C_1$ conformation of glucose).[23] Again, diastereotopic assignment of the methylene protons at C2 and C8 proved impossible. A large coupling constant ($J_{2b,3}$ = 9.7 Hz) was observed for one of the geminal protons attached to C2 (H2b), implying an *anti* relationship with H3. Because of the extensive overlapping with other signals, the coupling constants for the methylene protons at C8 could not be determined. Strong ROEs were observed between the amide proton and the neighbouring methylene protons (H8a and H8b), as well as between the amide proton and H2b. Furthermore, medium

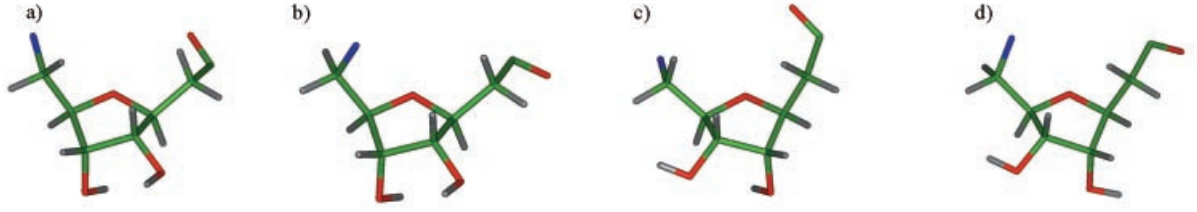


Figure 5. The four different monomer conformations resulting from simulated annealing calculations: a) ^{4}E and +sc; b) ^{4}E and −sc; c) $^5_4$T and +sc; d) $^5_4$T and −sc conformations for the furan ring and C6−C7 rotamer, respectively

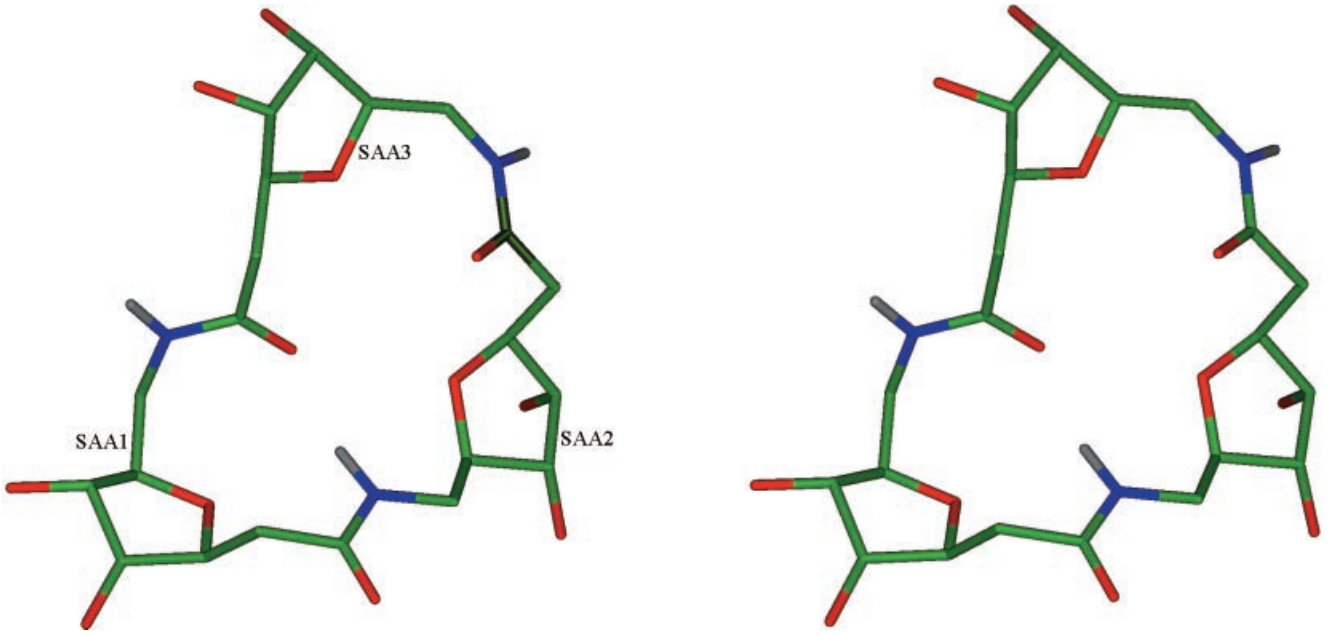


Figure 6. Stereoplot of a low-energy conformer of **1** resulting from simulated annealing calculations with SAA1: $^{5}_{4}$T and −sc; SAA2: $^{5}_{4}$T and +sc; SAA3: ^{4}E and +sc

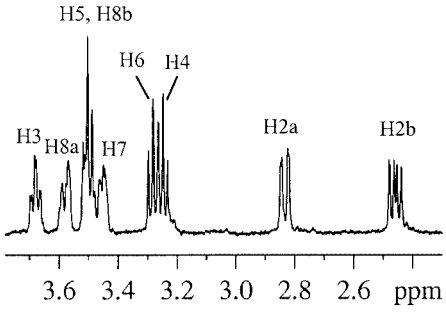


Figure 7. Part of the ^{1}H NMR spectrum of **2** (for the numbering see Figure 1) recorded in 10% D_2O in H_2O

ROEs were observed between the amide proton and H2a and H6.

As the strong H2b/HN and medium H2a/HN ROEs are judged to be sequential, the amide bonds of compound **2** have to be *trans* (as in **1**).

The same simulated annealing technique as used for the furanoid SAA trimer was performed on the pyranoid-containing trimer **2**. The chair conformations of the six-membered rings remain stable during the simulations. Furthermore, as also observed for the C2−C3 side chain of the furanoid SAA, $H2_S$ adopts an *anti* conformation in relation to H3 (*i.e.* −sc, Figure 8). All three possible staggered conformations around C7−C8 were found in the simulated structures. However, the −sc rotamer, with both methylene protons in a *gauche* position relative to H7, is the most populated (see Figure 8). This conformation has the amide proton positioned above the sugar ring, explaining the observed HN/H6 ROE cross-peak. An example of one low-energy structure from the simulated annealing calculations is displayed in Figure 9.

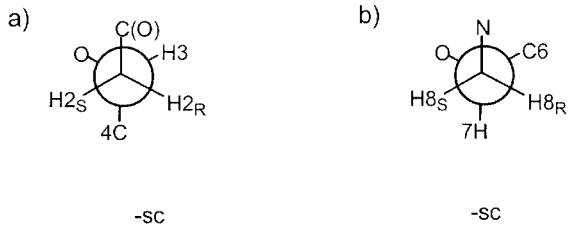


Figure 8. a) Rotamer conformation for C2−C3 bonds; b) most populated rotamer for C7−C8 of the pyranoid SAA in **2**; the signal of the methylene proton resonating at higher field (H2b), with a coupling constant $J_{2b,3}$ of 9.7 Hz, could therefore be assigned as $H2_S$

Discussion

In this paper we present the synthesis of a new series of cyclic SAA-containing molecules composed of furanoid ε- (i.e., **1**, **3**) and pyranoid ε-SAAs (i.e., **2**, **4**). The latter compounds were prepared on oxime resin by a cyclization/cleavage strategy, thus avoiding the difficulties associated with cyclization in solution. Because of the apparent C_n symmetry of the trimers **1** and **2** in their NMR spectra, an unrestrained simulated annealing technique was used to search the entire conformational space in order to compare their conformational behaviour. Analysis of the cyclic trimer **1** revealed that the five-membered rings of the SAA residues flip between twist (north, $P = 0°$) and envelope (south, $P = 167°$) conformations. As the pyranoid rings adopt chair conformations stabilised by the equatorial positions of all

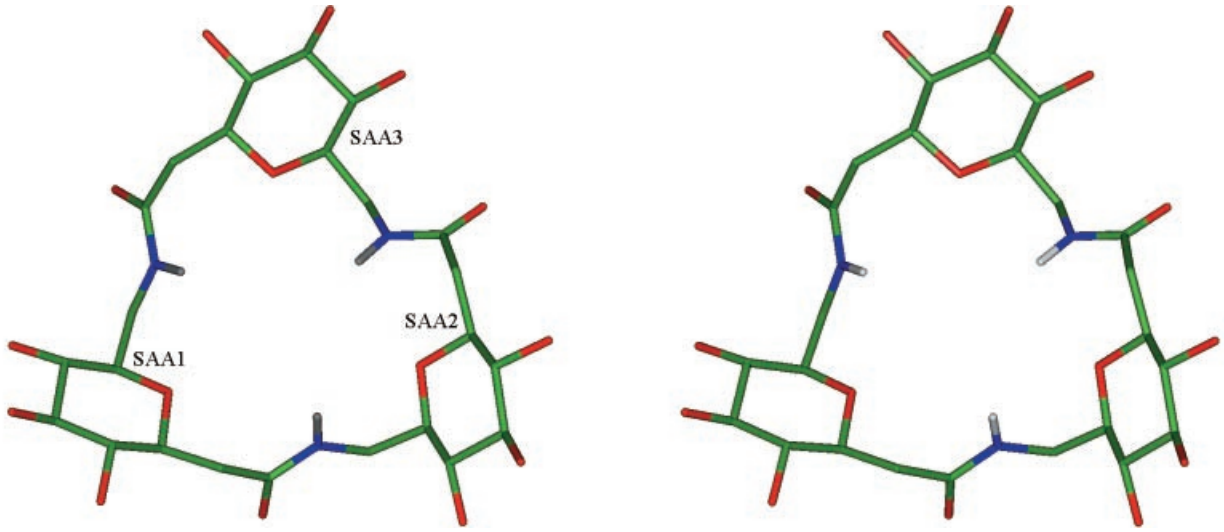


Figure 9. Stereoplot of a low-energy conformation of **2** with all C7−C8 rotamers as −sc

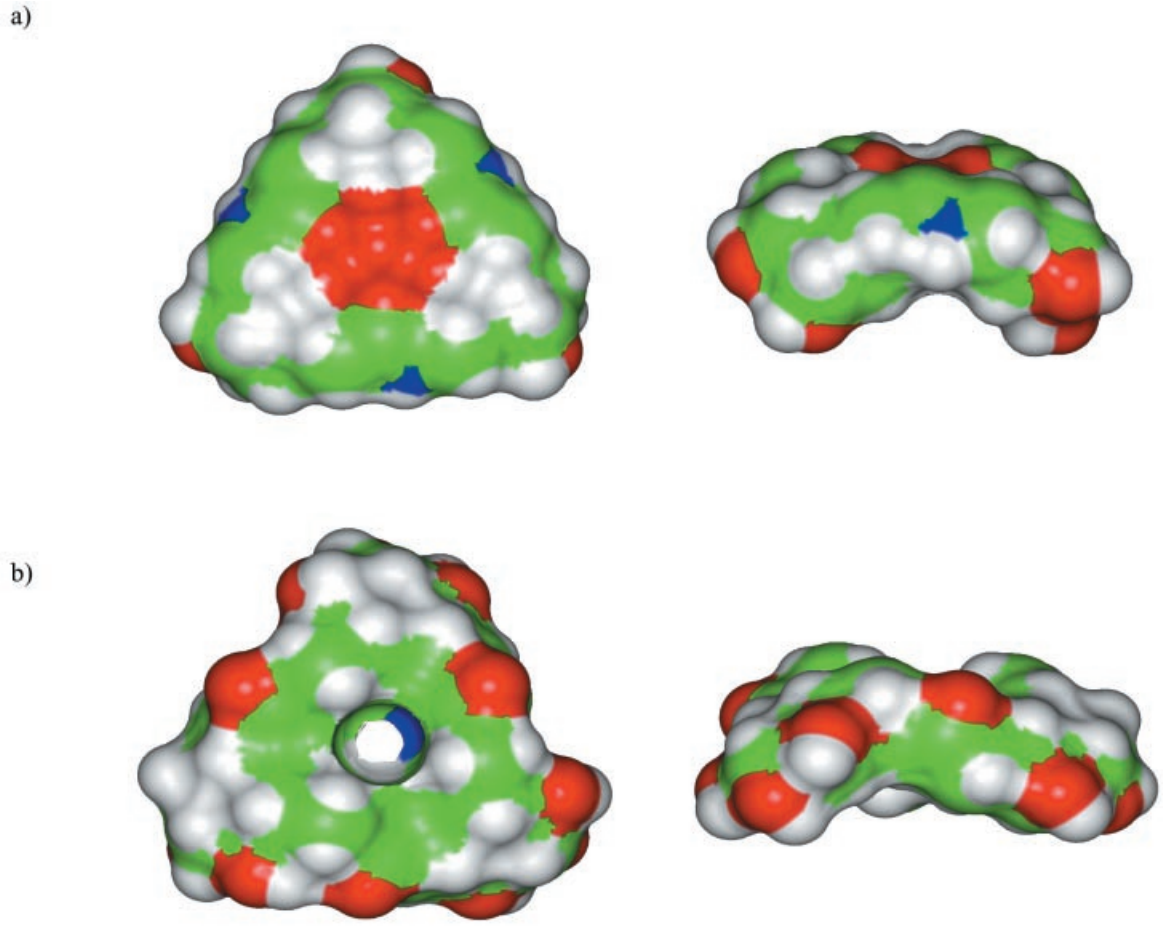


Figure 10. Side and top view of Connolly surface area plots of compounds **1** (a) and **2** (b)

the hydroxy groups, the trimer **2**, composed of pyranoid SAAs, is less flexible than its furanoid counterpart. The methylene functionalities attached to the amine groups of the SAA residues in the two trimers **1** and **2** are flexible, with varying rotamer conformations around the C6−C7 bond in **1** and C7−C8 bond in **2**, respectively. Surprisingly, the carbonyl side-chain methylene groups (i.e., C2) of the SAAs in both compounds **1** and **2** are restrained, as this region exclusively adopts the −sc conformation. In both cyclic trimers the oxygen atoms in the sugar ring are located on the interior and the secondary hydroxy groups are oriented outwards.

The possibility of cavity formation by the cyclic trimers is illustrated in the surface area plots of symmetrical conformers of **1** and **2** (Figure 10). It can clearly be seen that the structure of the furanoid SAA trimer is compact and does not contain a water-accessible cavity. On the other hand, the pyranoid SAA trimer does posses a small cavity. Both the furanoid SAA trimer and the pyranoid one have flat structures. The findings described in this paper provide important conformational data relating to the behaviour of furanoid and pyranoid ε-SAAs in small cyclic structures. By similar synthetic strategies, larger cyclic oligomers as well as structures containing more restrained SAA units should be constructed in the near future, as potential novel artificial host molecules.

Experimental Section

General Procedures and Materials: Mass spectra were recorded with PE/SCIEX API 165 with electronspray interface. Column chromatography was performed on Fluka 60 silica gel (0.04−0.063 mm). TLC analysis was conducted on "DC Plastikfolien" (Merck 60 F_{254} silica gel) with detection by UV absorption (254 nm) where applicable and by spraying with 20% H_2SO_4 in ethanol, a ninhydrin solution or ammonium molybdate (25 g/L) and ceric ammonium sulfate (10 g/L), followed by charring at ≈ 150 °C. Reactions were run at ambient temperature, unless stated otherwise. Reactions requiring anhydrous conditions were stirred under argon or nitrogen. Acetone (Acros, p.a.), DCE (Biosolve, HPLC-grade), DMF (Baker, p.a.), toluene (Biosolve, p.a.), 1,4-dioxane (Baker, p.a.), pyridine (Baker, p.a.) and DCM (Baker, p.a.) were stored over molecular sieves (4 Å). Acetonitrile (Biosolve, p.a.) and MeOH (Biosolve, p.a.) were stored over molecular sieves (3 Å). Triethylamine (Acros) was boiled under reflux for 3 h with CaH_2, distilled and stored over KOH. Methyl (triphenylphosphoranylidene)acetate (Aldrich), 2,2-dimethyl-1,3-dioxan-4,6-dione (Acros), Boc-ON (Janssen chimica), oxime resin (Nova Biochem), BOP (Senn chemicals) and DIPEA (Biosolve) were used as received. Solid-phase chemistry was performed in DCM (Biosolve, peptide synthesis grade) and NMP (Biosolve, peptide synthesis grade). Preparative RP HPLC was performed on an Altima C18 reversed-phase column-TFA 10.00 mmd installed on BioCad "Vision" workstation (Perseptive Biosystems). The products were eluted at 5 mL/min with CH_3CN/H_2O containing 0.1% TFA; detection was performed at 214 nm.

NMR Measurements: ^1H and ^{13}C NMR spectra were recorded with Jeol JNM-FX-200 (200/50.1 MHz), Bruker WM 300 (300/75.1 MHz) or Bruker DMX 600 (600/150 MHz) spectrometers. Chemical shifts are given in ppm (δ) relative to tetramethylsilane as internal standard. All ^{13}C spectra given are proton-decoupled. All spectra of the cyclic SAA homooligomers were recorded in H_2O/D_2O (9:1, v/v) at 278 K with a Bruker DMX 600 or a Bruker DMX 500 spectrometer equipped with a pulsed field gradient accessory. Standard DQF-COSY (512c × 2084c) spectra were recorded with use of presaturation for solvent suppression. NOESY and ROESY spectra (400c × 2048c, τ_{mix} = 180 ms) were recorded with use of the Watergate solvent suppression.[24] All spectra were recorded in phase-sensitive mode, by using either the TPPI or States-TPPI for quadrature detection in the indirect dimension. Homonuclear coupling constants were determined from the corresponding ^1H spectra.

Computer Simulations: The structure calculations were performed with a Silicon Graphics Origin R10000 computer. Energy minimisation (EM) and molecular dynamics (MD) calculations were carried out with the DISCOVER program by use of the CVFF force field[25] and a dielectric constant of 80. After EM using steepest descent and conjugate gradient, the system was heated gradually, starting from 300 K and increasing to 800 K, and subsequently cooled to 300 K with 5 ps steps at every temperature, each by direct scaling of velocities. All the structures originating from the MD simulations were minimised, again by use of steepest descent and conjugate gradient algorithms. During the MD calculations no restraints were taken into account. The resulting structures were finally evaluated on the basis of the NOEs and ^3J coupling constants.

Methyl 3,6-Anhydro-2-deoxy-4,5-*O*-isopropylidene-D-*allo*-heptulonate (6): Compound **5** (29 g, 154 mmol) was coevaporated with toluene (3 × 50 mL) and dissolved in acetonitrile (500 mL), and methyl (triphenylphosphoranylidene)acetate (56.5 g, 169 mmol) was added. The reaction mixture was heated under reflux for 2 h and concentrated, and the residue was dissolved in MeOH (300 mL). Sodium methoxide (0.83 g, 15.4 mmol) was added, and the reaction mixture was stirred for 10 min. The reaction mixture was neutralised with Dowex-H^+, the ion-exchange resin was removed by filtration, and the filtrate was concentrated. The residue was purified by silica gel column chromatography (eluent: EtOAc/light petroleum, 2:8 → 8:2, v/v) to give compound **6** in 81% yield (30.6 g) as a colourless syrup. The spectroscopic data were in full agreement with those reported in the literature.[13]

Methyl 3,6-Anhydro-2-deoxy-4,5-*O*-isopropylidene-7-*O*-mesyl-D-*allo*-heptulonate (7): Compound **6** (14.76 g, 60 mmol) was coevaporated with pyridine (3 × 50 mL) and dissolved in pyridine (300 mL). Methanesulfonyl chloride (5.6 mL, 72 mmol) was added, and the reaction mixture was stirred for 16 h. The solvent was removed under reduced pressure and the residue was dissolved in EtOAc (300 mL). The organic phase was washed with H_2O (150 mL), aq. $NaHCO_3$ (3 × 150 mL) and brine (100 mL), dried ($MgSO_4$) and concentrated. The crude product was applied to a silica gel column and eluted with EtOAc/light petroleum (3:7 → 7:3, v/v) to afford compound **7** in 93% (18.2 g) as a colourless oil. ^1H NMR ($CDCl_3$): δ = 4.65 (dd, H5, $J_{4,5}$ = 6.6, $J_{5,6}$ = 4.4 Hz, 1 H), 4.55 (dd, $J_{3,4}$ = 3.7, $J_{4,5}$ = 6.6 Hz 1 H, H4), 4.35 (m, 3 H, H3, H7a, H7b), 4.18 (m, 1 H, H6), 3.71 (s, 3 H, OMe), 3.05 (s, 3 H, Ms), 2.73 (dd, $J_{2a,3}$ = 5.1 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2a), 2.60 (dd, $J_{2b,3}$ = 6.9 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2b), 1.55 (s, 3 H, CH_3 isoprop), 1.35 (s, 3 H, CH_3 isoprop) ppm. ^{13}C NMR ($CDCl_3$): δ = 170.3 (C1), 114.5 (Cq isoprop), 83.7, 81.6, 80.8 (C3, C4, C5, C6), 68.9 (C7), 51.5 (CH_3 OMe), 37.7 (C2), 37.1 (CH_3 Ms), 27.0 (CH_3 isoprop), 25.1 (CH_3 isoprop) ppm. ES-MS: *m/z* = 324.9$[M + H]^+$, 346.9 $[M + Na]^+$, 363 $[M + K]^+$.

Methyl 3,6-Anhydro-7-azido-2,7-dideoxy-4,5-*O*-isopropylidene-D-*allo*-heptulonate (8): Compound **7** (18.2 g, 56 mmol) was coevaporated with toluene (3 × 50 mL) and dissolved in DMF (200 mL). Sodium azide (9.1 g, 140 mmol) was added and the resulting mixture was stirred at 75 °C for 1.5 h. The reaction mixture was allowed to cool down to room temperature, after which it was diluted with H_2O (100 mL). Extraction with EtOAc (3 × 200 mL) and subsequent drying ($MgSO_4$) of the combined organic phases gave compound **8**, after purification by column chromatography (eluent: EtOAc/light petroleum, 2:8 → 7:3, v/v), in a yield of 94% (14.3 g) as a colourless syrup. ^1H NMR ($CDCl_3$): δ = 4.51 (m, 2 H, H5, H6), 4.32 (dd, 1 H, H4), 4.14 (m, 1 H, H3), 3.71 (s, 3 H, OMe), 3.55 (dd, 1 H, H7a, $J_{7a,6}$ = 3.7 Hz, $J_{7a,7b}$ = 13.1 Hz), 3.34 (dd, 1 H, H7b, $J_{7b,6}$ = 5.1 Hz, $J_{7a,7b}$ = 13.2 Hz), 2.75 (dd, 1 H, H2a, $J_{2a,3}$ = 5.1 Hz, $J_{2a,2b}$ = 15.3 Hz), 2.64 (dd, 1 H, H2b, $J_{2b,3}$ = 6.9 Hz, $J_{2a,2b}$ = 15.3 Hz)., 1.55 (s, 3 H, CH_3 isoprop), 1.35 (s, 3 H,

CH_3 isoprop) ppm. ^{13}C NMR ($CDCl_3$): δ = 170.5 (C1), 114.7 (Cq isoprop), 83.9, 82.8, 81.8, 80.6 (C3, C4, C5, C6), 51.9 (C7), 51.6 (CH_3 OMe), 37.8 (C2), 27.2 (CH_3 isoprop), 25.2 (CH_3 isoprop) ppm. ES-MS: *m*/*z* = 293.9 $[M + Na]^+$.

Methyl 3,6-Anhydro-7-[(*tert*-butoxycarbonyl)amino]-2,7-dideoxy-D-*allo*-heptulonate (10): Compound **8** (12.85 g, 50 mmol) was dissolved in ethanol (200 mL) containing HCl (3 M, 20 mL). After degassing of the solution, Pd/C (1.4 g) was added and the solution was degassed again. The reaction mixture was stirred for 18 h under H_2, after which only baseline material was visible by TLC. The solution was filtered through Glass Fibre (GF/2A, Whatman) and the filtrate was concentrated under reduced pressure. Crude **9** was dissolved in H_2O (100 mL), and Na_2CO_3 (10.6 g, 100 mmol) and $NaHCO_3$ (16.8 g, 200 mmol) were added. After the reaction mixture had been cooled to 0 °C, a solution of Boc_2O (11.99 g, 55 mmol, 1.1 equiv.) in dioxane (100 mL) was added. After stirring for 18 h the solution was acidified with HCl (1 M) and extracted with EtOAc (3 × 150 mL). The combined organic phases were dried ($MgSO_4$) and concentrated. The residue was purified by column chromatography (eluent: EtOAc/light petroleum, 2:8 → 8:2, v/v) to give compound **10** in a yield of 82% (12.9 g), over the two steps, as a slightly yellow syrup. ^1H NMR ($CDCl_3$): δ = 4.87 (t, *J* = 6.2 Hz, 1 H, HN), 4.12 (m, 1 H, H6), 3.95−3.82 (m, 3 H, H3, H4, H5), 3.73 (s, 3 H, OMe), 3.35 (dd, *J* = 4.4 Hz, *J* = 5.8 Hz, 2 H, H7a, H7b), 2.72 (dd, $J_{2a,3}$ = 5.1 Hz, $J_{2a,2b}$ = 15.3 Hz, 1 H, H2a), 2.63 (dd, $J_{2b,3}$ = 6.6 Hz, $J_{2a,2b}$ = 15.7 Hz, 1 H, H2b), 1.45 (s, 9 H, *tert*-Bu Boc) ppm. ^{13}C NMR ($CDCl_3$): δ = = 171.4 (C1), 156.2 [C(O) Boc), 82.3, 79.0, 73.9, 71.5 (C3, C4, C5, C6), 51.5 (CH_3 OMe), 41.8 (C7), 39.2 (C2), 27.2 (3 × CH_3 Boc) ppm. ES-MS: *m*/*z* = 306 $[M + H]^+$, 328.2 $[M + Na]^+$, 633.5 $[2M + Na]^+$

3,6-Anhydro-7-[(*tert*-butoxycarbonyl)amino]-2,7-dideoxy-D-*allo*-heptulonic Acid (11): NaOH (1 M, 10 mL) was added to a mixture of compound **9** (2.57 g, 8.4 mmol) in dioxane (20 mL). After stirring for 2.5 h, the reaction mixture was acidified to pH 2 with HCl (1 N) and extracted with EtOAc (6 × 60 mL). The combined organic phases were dried ($MgSO_4$) and concentrated. The residue was applied to a silica gel column and eluted with EtOAc/MeOH (1:0 → 9:1, v/v) containing 1% AcOH to give compound **11** in 89% yield (2.25 g) as an amorphous white solid. ^1H NMR (MeOD): δ = 4.94 (s, 1 H, HN), 4.12 (m, 1 H, H6), 3.86−3.73 (m, 3 H, H3, H4, H5), 3.25 (dd, $J_{6,7a}$ = 2.9 Hz, 1 H, H7a), 3.17(dd, $J_{6,7b}$ = 4.0 Hz, 1 H, H7b), 2.65 (dd, $J_{2a,3}$ = 4.4 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2a), 2.45 (dd, $J_{2b,3}$ = 8.2 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2b), 1.44 (s, 9 H, *tert*-Bu Boc) ppm. ^{13}C NMR (MeOD): δ = 174.8 (C1), 158.3 [C(O) Boc), 84.1, 80.3, 75.5, 73.2 (C3, C4, C5, C6), 43.4 (C7), 37.7 (C2), 27.9 (3 × CH_3 Boc) ppm. ES-MS: *m*/*z* = 313.9$[M + Na]^+$, 605.4 $[2M + Na]^+$, 896.5 $[3M + Na]^+$.

Methyl 4,5,6,8-Tetra-*O*-acetyl-3,7-anhydro-2-deoxy- D-*glycero*-D-*gulo*-octulonate (13): Compound **12**[13] (14.43 g, 37 mmol) was coevaporated with DCE (3 × 50 mL) and dissolved in DCM (200 mL). MeOH (2.23 mL, 55 mmol), DIC (6.29 mL, 40.7 mmol) and DMAP (0.45 g, 3.7 mmol) were added, and the reaction mixture was stirred at room temperature. After 3 the reaction mixture was filtered through a short pad of Hyflo and concentrated. Purification by column chromatography (eluent: EtOAc/light petroleum, 0:1 → 1:1, v/v) afforded compound **13** in 83% (12.5 g) as a colourless syrup. ^1H NMR ($CDCl_3$): δ = 5.20 (t, *J* = 9.5 Hz, 1 H, H6), 5.07 (t, *J* = 9.5 Hz, 1 H, H5), 4.93 (t, *J* = 9.5 Hz, 1 H, H4), 4.25 (dd, $J_{7,8a}$ = 5.1 Hz, $J_{8a,8b}$ = 12.4 Hz, 1 H, H8a), 4.06 (dd, $J_{7,8b}$ = 2.2 Hz, $J_{8a,8b}$ = 12.4 Hz, 1 H, H8b), 3.99−3.90 (m, 1 H, H7), 3.74−3.65 (m, 1 H, H3), 3.70 (s, 3 H, OMe), 2.53 (m, 2 H, 2 × H2), 2.07, 2.05, 2.03, 2.00 (4 × s, 12 H, 4 × CH_3 Ac) ppm. ^{13}C NMR ($CDCl_3$): δ = 170.1, 169.8, 169.3, 169.1 [C1, 4 × C(O) Ac], 75.4, 74.1, 73.7, 71.1, 68.0 (C3, C4, C5, C6, C7), 61.7 (C8), 51.5 (OMe), 36.6 (C2), 20.2 (4 × CH_3 Ac) ppm.

Methyl 3,7-Anhydro-2-deoxy-8-*O*-trityl-D-*gulo*-D-*glycero*-octulonate (15): Compound **13** (12.5 g, 30.9 mmol) was coevaporated with toluene (3 × 50 mL) and dissolved in MeOH (200 mL). NaOMe (0.17 g, 3.1 mmol) was added, and the resulting mixture was stirred for 2 h. The reaction mixture was neutralised with Dowex-H^+, the ion-exchange resin was removed by filtration, and the filtrate was concentrated. The residue was coevaporated with pyridine (3 × 50 mL) and dissolved in pyridine (150 mL). TrCl (10.3 g, 37.1 mmol) was added and the reaction mixture was stirred for 16 h. After concentration of the solution, the residue was taken up in EtOAc (150 mL), washed with H_2O(100 mL), aq. $NaHCO_3$ (3 × 100 mL) and brine (100 mL), dried ($MgSO_4$) and concentrated. Crude **15** was purified by column chromatography (eluent: EtOAc/light petroleum, 1:9 → 1:1) to give a yield of 85% (12.55 g) of a colourless oil. ^1H NMR ($CDCl_3$): δ = 7.44−7.12 (m, 15 H, Tr), 4.88 (br. s, 1 H, OH), 4.43 (br. s, 1 H, OH), 3.88 (br. s, 1 H, OH), 3.65 (m, 1 H, H3), 3.58 (s, 3 H, OMe), 3.37, 3.20 (2 × m, 6 H, H4, H5, H6, H7, 2 × H8), 2.79 (dd, $J_{2a,3}$ = 2.9 Hz, $J_{2a,2b}$ = 15.0 Hz, 1 H, H2a), 2.45 (dd, $J_{2b,3}$ = 8.8 Hz, $J_{2a,2b}$ = 15.3 Hz, 1 H, H2b) ppm. ^{13}C NMR ($CDCl_3$): δ = 172.1 (C1), 143.8 (Cq Tr), 128.4, 127.5, 126.7 (CH_{arom} Tr), 86.2 (Cq Tr), 78.3, 75.9, 73.2, 70.8 (C3, C4, C5, C6, C7), 60.1 (C8), 51.6 (OMe), 37.6 (C2) ppm.

Methyl 3,7-Anhydro-4,5,6-tri-*O*-benzyl-2-deoxy-8-*O*-trityl-D-*gulo*-D-*glycero*-octulonate (16): After coevaporation of compound **15** (8.61 g, 18 mmol) with toluene (3 × 30 mL), DMF (100 mL), BnBr (7.7 mL, 64.8 mmol) and NaH (2.38 g, 59.4 mmol) were added and the resulting mixture was stirred for 16 h. MeOH (10 mL) was added to quench the excess of NaH, and the solution was concentrated. The residue was dissolved in EtOAc (150 mL), washed with H_2O(100 mL), aq. $NaHCO_3$ (3 × 100 mL) and brine (100 mL), dried ($MgSO_4$) and applied to a silica gel column. Compound **16** was obtained as an amorphous white solid in 88% (11.84 g) by elution with EtOAc/light petroleum (0:1 → 3:7, v/v). ^1H NMR ($CDCl_3$): δ = 7.51−7.16 (m, 30 H, Tr, 3 × Bn), 4.89 (m, 3 H, CH_2 Bn), 4.69 (2 × d, *J* = 4.8 Hz, *J* = 11.0 Hz, 2 H, CH_2 Bn), 4.37 (d, *J* = 10.2 Hz),1 H, CH_2 Bn, 3.85−3.39 (m, 6 H, H3, H4, H5, H6, H7, H8a), 3.65 (s, 3 H, OMe), 3.16 (dd, $J_{7,8b}$ = 3.7 Hz, $J_{8a,8b}$ = 10.2 Hz, 1 H, H8b), 2.86 (dd, $J_{2a,3}$ = 3.7 Hz, $J_{2a,2b}$ = 14.6 Hz, 1 H, H2a), 2.58 (dd, $J_{2b,3}$ = 8.0 Hz, $J_{2a,2b}$ = 14.6 Hz, 1 H, H2a) ppm. ^{13}C NMR ($CDCl_3$): δ = 171.2 (C1), 143.8 (Cq Tr), 138.2−137.6 (3 × Cq Bn), 128.6−126.7 (CH_{arom} Tr, Bn), 86.0 (Cq Tr), 86.9, 81.1, 78.5, 78.3, 75.7 (C3, C4, C5, C6, C7), 75.6−74.9 (3 × CH_2 Bn), 62.2 (C8), 51.6 (OMe), 37.5 (C2) ppm.

Methyl 3,7-Anhydro-4,5,6-tri-*O*-benzyl-2-deoxy-D-*gulo*-D-*glycero*-octulonate (17): Compound **16** (11.84 g, 15.9 mmol) was treated with a mixture of 3% *p*TsOH in DCM/MeOH (1:1, v/v, 100 mL). When TLC analysis revealed complete conversion (≈ 3 h) into a lower running product, the reaction mixture was neutralised with aq. $NaHCO_3$ and concentrated. The residue was taken up in EtOAc (150 mL), washed with H_2O (100 mL), aq. $NaHCO_3$ (3 × 100 mL) and brine (100 mL) and dried ($MgSO_4$). Purification by column chromatography (eluent: EtOAc/light petroleum, 0:1 → 4:6, v/v) afforded **17** in quantitative yield (7.90 g) as a colourless syrup. ^1H NMR ($CDCl_3$): δ = 7.31 (m, 15 H, H_{arom} 3 × Bn), 4.92 (m, 4 H, CH_2 Bn), 4.64 (2 × d, *J* = 8.4 Hz, *J* = 11.0 Hz, 2 H, CH_2 Bn), 3.85−3.39 (m, 7 H, H3, H4, H5, H6, H7, 2 × H8), 3.63 (s, 3 H, OMe), 2.74 (dd, $J_{2a,3}$ = 3.7 Hz, $J_{2a,2b}$ = 15.7 Hz, 1 H, H2a), 2.40 (dd, $J_{2b,3}$ = 8.4 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2b) ppm. ^{13}C NMR ($CDCl_3$): δ = 171.3 (C1), 138.3−137.9 (3 × Cq Bn), 128.4−127.7

(CH_{arom} Bn), 86.9, 81.2, 79.2, 78.1, 75.6 (C3, C4, C5, C6, C7), 75.5−75.0 (3 × CH_2 Bn), 61.7 (C8), 51.7 (OMe), 37.3 (C2) ppm. ES-MS: m/z = 507.2 $[M + H]^+$, 329.2 $[M + Na]^+$.

Methyl 3,7-Anhydro-4,5,6-tri-*O*-benzyl-2-deoxy-8-*O*-mesyl -D-*gulo*-D-*glycero*-octulonate (18): Compound **17** (2.5 g, 5 mmol) was coevaporated with pyridine (20 mL) and then redissolved in pyridine (30 mL), and MsCl (0.46 mL, 6 mmol) was added. The reaction mixture was stirred for 16 h, after which TLC analysis showed the reaction to be complete. After concentration of the solution, the residue was dissolved in EtOAc (50 mL) and washed with H_2O (25 mL), aq. $NaHCO_3$ (3 × 25 mL) and brine (25 mL). Drying over $MgSO_4$ and purification by column chromatography (eluent: EtOAc/light petroleum, 0:1 → 4:6, v/v) afforded **18** in quantitative yield (2.94 g) as colourless oil. ^1H NMR ($CDCl_3$): δ = 7.31 (m, 15 H, H_{arom} 3 × Bn), 4.90 (m, 4 H, CH_2 Bn), 4.63 (dd, 2 H, 2 × CH_2 Bn), 3.82−3.51 (2 m, 6 H, H3, H4, H5, H6, H7, H8a), 3.63 (s, 3 H, OMe), 3.35 (t, J = 9.5 Hz, 1 H, H8b), 2.97 (s, 3 H, Ms), 2.74 (dd, $J_{2a,3}$ = 3.7 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2a), 2.38 (dd, $J_{2b,3}$ = 8.4 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2b) ppm. ^{13}C NMR ($CDCl_3$): δ = 170.9 (C1), 137.9, 137.5, 137.3 (3 × Cq Bn), 128.4−127.7 (CH_{arom} Bn), 86.6, 80.6, 77.3, 76.5, 75.7 (C3, C4, C5, C6, C7), 75.4−74.9 (3 × CH_2 Bn), 68.6 (C8), 51.6 (OMe), 37.3 (Ms), 37.0 (C2) ppm.

Methyl 3,7-Anhydro-8-azido-4,5,6-tri-*O*-benzyl-2,8-dideoxy-D-*gulo*-D-*glycero*-octulonate (19): Traces of water were removed from compound **18** (2.94 g, 5 mmol) by coevaporation with toluene (3 × 20 mL). DMF (25 mL) and NaN_3 (0.72 g, 11 mmol) were added, and the resulting mixture was stirred for 1.5 h at 75 °C. The solution was diluted with H_2O (20 mL) and extracted with EtOAc (3 × 40 mL) and the combined organic phases were dried ($MgSO_4$). The crude product azide was purified by column chromatography (eluent: EtOAc/light petroleum, 0:1 → 1:1, v/v) to give product **19** in a yield of 77% (2.05 g) as a colourless syrup. ^1H NMR ($CDCl_3$): δ = 7.32 (m, 15 H, H_{arom} 3 × Bn), 4.91 (m, 4 H, CH_2 Bn), 4.62 (dd, J = 9.5 Hz, J = 10.6 Hz, 2 H, CH_2 Bn) 3.84−3.33 (2 × m, 6 H, H3, H4, H5, H6, H7, H8a), 3.63 (s, 3 H, OMe), 3.24 (dd, $J_{7,8b}$ = 4.4 Hz, $J_{8a,8b}$ = 12.8 Hz, 1 H, H8b), 2.74 (dd, $J_{2a,3}$ = 3.7 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2a), 2.42 (dd, $J_{2b,3}$ = 8.4 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2b) ppm. ^{13}C NMR ($CDCl_3$): δ = 170.8 (C1), 137.9, 137.6, 137.5 (3 × Cq Bn), 128.2−127.4 (CH_{arom} Bn), 86.6, 80.8, 78.6, 78.3, 75.5 (C3, C4, C5, C6, C7), 75.2−74.8 (3 × CH_2 Bn), 51.6 (OMe), 50.8 (C8), 37.0 (C2) ppm.

Methyl 3,7-Anhydro-4,5,6-tri-*O*-benzyl-8-[(*tert*-butoxycarbonyl)-amino]-2,8-dideoxy-D-*gulo*-D-*glycero*-octulonate (20): Compound **19** (2.05 g, 3.9 mmol) was coevaporated with toluene (10 mL) and redissolved in toluene (20 mL). Me_3P (4.1 mL of a 1 M solution in toluene) was added and the reaction mixture was stirred for 1 h. The solution was cooled (−20 °C) and Boc-ON (2.24 g, 3.7 mmol), dissolved in toluene (2.5 mL), was added slowly. The reaction mixture was allowed to warm to room temperature and stirring was continued for 5 h. The reaction mixture was concentrated and redissolved in EtOAc (25 mL), washed with H_2O, aq (15 mL). $NaHCO_3$ (3 × 15 mL) and brine (15 mL) and dried ($MgSO_4$). Purification by column chromatography (eluent: EtOAc/light petroleum, 0:1 → 4:6, v/v) afforded compound **20** in 95% yield (2.24 g) as a colourless oil. ^1H NMR ($CDCl_3$): δ = 7.32 (s, 15 H, H_{arom} 3 × Bn), 4.97−4.79 (m, 4 H, CH_2 Bn), 4.66 (d, J = 5.9 Hz, 1 H, CH_2 Bn), 4.60 (d, J = 6.6 Hz, 1 H, CH_2 Bn), 3.76−3.27 (2 × m, 7 H, H3, H4, H5, H6, H7, 2 × H8), 3.64 (s, 3 H, OMe), 2.73 (dd, $J_{2a,3}$ = 3.7 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2a), 2.37 (dd, $J_{2b,3}$ = 8.8 Hz, $J_{2a,2b}$ = 15.4 Hz, 1 H, H2b), 1.44 (s, 9 H, *tert*-Bu Boc) ppm. ^{13}C NMR ($CDCl_3$): δ = 171.6 (C1), 156.1 [C(O) Boc), 138.7−138.3 (3 × Cq Bn), 128.9, 128.3, 128.1 (15 × CH_{arom} Bn), 87.3, 81.6, 79.6, 78.1 (C3, C4, C5, C6, C7), 75.6, 75.5 (3 × CH_2 Bn), 52.1 (OMe), 41.5 (C8), 37.8 (C2), 28.8 (*tert*-Bu Boc) ppm.

Methyl 3,7-Anhydro-8-[(*tert*-butoxycarbonyl)amino]-2,8-dideoxy-D-*gulo*-D-*glycero*-octulonate (21): A solution of compound **20** (0.215 g, 0.36 mmol) in EtOAc (3 mL) was degassed and Pd/C (20 mg) was added, and the solution was degassed for a second time. The reaction mixture was stirred under H_2 for 16 h. Filtering of the solution through Glass Fibre (GF/2A, Whatman) and concentration of the filtrate afforded the crude product, which was applied to a silica gel column. Elution with EtOAc/light petroleum (3:7 → 1:0, v/v) afforded **21** in quantitative yield (0.12 g) as a pale green syrup. ^1H NMR ($CDCl_3$): δ = 5.17 (br. s, 1 H, HN), 3.68 (s, 3 H, OMe), 3.53−3.26 (m, 7 H, H3, H4, H5, H6, H7, 2 × H8), 2.88 (bd, $J_{2a,2b}$ = 15.3 Hz, 1 H, H2a), 2.44 (dd, $J_{2b,3}$ = 9.5 Hz, $J_{2a,2b}$ = 13.5 Hz, 1 H, H2b), 1.42 (s, 9 H, *tert*-Bu Boc) ppm. ^{13}C NMR ($CDCl_3$): δ = 171.9 (C1), 156.5 [C(O) Boc), 79.3 (Cq Boc), 77.9, 77.2, 75.7, 73.1, 71.1 (C3, C4, C5, C6, C7), 51.4 (OMe), 41.2 (C8), 37.0 (C2), 27.9 (*tert*-Bu Boc) ppm. ES-MS: m/z = 358.2.2 $[M + Na]^+$, 693.4 $[2M + Na]^+$.

3,7-Anhydro-8-[(*tert*-butoxycarbonyl)amino]-2,8-dideoxy-D-*gulo*-D-*glycero*-octulonic Acid (22): A mixture of compound **21** (0.12 g, 0.36 mmol) and aq. NaOH (0.4 mL of a 1 M solution) in dioxane (1 mL) was stirred for 4 h. The solution was acidified to pH 2 with HCl (1 M) and extracted thoroughly with EtOAc (6 × 5 mL). The combined organic phases were dried ($MgSO_4$), concentrated and purified by column chromatography (eluent: MeOH/EtOAc, 0:1 → 1:9, v/v, containing 0.5% AcOH). Compound **22** was obtained in a 70% yield (0.081 g) as an amorphous white solid. ^1H NMR (MeOD): δ = 3.60 (dt, J = 2.6 Hz, J = 9.5 Hz, 1 H, H3), 3.51−3.04 (6 H, H4, H5, H6, H7, 2 × H8), 2.84 (dd, $J_{2a,3}$ = 2.6 Hz, $J_{2a,2b}$ = 15.7 Hz, 1 H, H2a), 2.34 (dd, $J_{2b,3}$ = 9.1 Hz, $J_{2a.2b}$ 15.7 Hz, 1 H, H2b), 1.43 (s, 9 H, *tert*-Bu Boc) ppm. ^{13}C NMR (MeOD): δ = 175.6 (C1), 158.2 [C(O) Boc), 80.1 (Cq Boc), 79.4, 78.3, 77.1, 74.2, 72.1 (C3, C4, C5, C6, C7), 42.4 (C8), 38.7 (C2), 29.3 (*tert*-Bu Boc) ppm. ES-MS: m/z = 322.2 $[M + H]^+$, 344.2 $[M + Na]^+$, 643.7 $[2M + H]^+$, 665.3 $[2M + Na]^+$.

General Procedure for the Synthesis of Cyclic SAA Molecules: The assembly of the cyclic molecules was performed on an ABI 433A (Applied Biosystems, division of Perkin−Elmer) peptide synthesiser by use of a Boc strategy. The synthesis was performed on Kaiser oxime resin (0.53 mmol/g) on a 50 μmol scale. Stock solutions of BOP (0.5 M), and DIPEA (0.65 M) in NMP were prepared. The building blocks were dissolved in 1:1 mixtures of NMP and DCM at concentrations of 0.25 M. For the Boc cleavage reactions, a solution of 25% TFA in DCM containing 1% TIPS was prepared.

The resin was placed in the reaction vessel and was swollen in DCM (5 × 2.5 mL) and NMP (5 × 2.5 mL). The stock solutions of BOP (0.5 mL, 5 equiv.) and DIPEA (0.5 mL, 6.5 equiv.) were added to the building block solution (1 mL, 5 equiv.), and the resulting mixture was added to the resin. The reaction vessel was shaken for 2 h, after which it was drained. Without rinsing of the resin, a second coupling reaction was performed under the same conditions. The resin was washed with NMP (5 × 2.5 mL) and DCM (5 × 2.5 mL). The Boc group was removed by several 5 min treatments of the resin with the TFA solution (5 × 2 mL). The resin was washed with DCM and NMP (5 × 2.5 mL each). Successive SAA building blocks were coupled as described above, with reaction times of 45 min. Each coupling was followed by the same washing and Boc removal procedure.

For the cyclization reaction, AcOH (0.1 M, 1 mL, 2 equiv.) and DIPEA (0.1 M, 1 mL, 2 equiv.) in DMF (2 mL) were added and

the reaction vessel was shaken for 36 h. The filtrate was collected, the resin was rinsed with DMF (3 × 2 mL), and the combined DMF fractions were concentrated.

The crude cyclic molecules were analysed by LCMS and purified by RP-HPLC with H_2O/CH_3CN as eluent. The pure fractions were lyophilised and analysed by LCMS and NMR spectroscopy.

Cyclic Furanoid SAA Trimer 1: ^{1}H NMR, COSY, NOESY, ROESY (10% D_2O/H_2O, 600 MHz): δ = 8.41 (s, 3 H, 3 × HN), 4.12 (m, 3 H, 3 × H3), 3.97−3.89 (m, 9 H, 3 × H4, 3 × H5, 3 × H6), 3.46 (dd, $J_{6,7a}$ = 2.9 Hz, $J_{7a,7b}$ = 14.7 Hz, 3 H, 3 × H7a), 3.37 (dd, $J_{6,7b}$ = 5.5 Hz, $J_{7a,7b}$ = 14.7 Hz, 3 H, 3 × H7b), 2.62 (dd, $J_{2a,3}$ = 3.7 Hz, $J_{2a,2b}$ = 14.4 Hz, 3 H, 3 × H2a), 2.41 (dd, $J_{2b,3}$ = 9.3 Hz, $J_{2a,2b}$ = 14.5 Hz, 3 H, 3 × H2b) ppm. LCMS: R_t = 7.35 (ACN/H_2O, 0 → 25%), *m/z* = 520.4 $[M + H]^+$. HRMS: calcd. for $C_{21}H_{33}N_3O_{12}$ [M + H] 520.2143; found 520.2254.

Cyclic Furanoid SAA Tetramer 3: ^{1}H NMR, COSY, NOESY (10% D_2O/H_2O, 600 MHz): δ = 7.81 (s, 4 H, 4 × HN), 4.15 (t, $J_{3,4}$ = $J_{4,5}$ = 5.6 Hz, 4 H, 4 × H4), 4.04 (dt, $J_{2a,3}$ = 4.0 Hz, $J_{2b,3}$ = 6.4 Hz $J_{3,4}$ = 5.6 Hz, 4 H, 4 × H3), 3.96 (t, $J_{4,5}$ = $J_{5,6}$ = 5.9 Hz, 4 H, 4 × H5), 3.89 (dt, $J_{5,6}$ = 6.1, $J_{6,7a}$ = 6.2 Hz, $J_{6,7b}$ = 3.3 Hz, 4 H, 4 × H6), 3.53 (dt, *J* = 6.9 Hz, *J* = 14.3 Hz, 4 H, 4 × H7a), 3.34 (dt, *J* = 3.3 Hz, *J* = 14.4 Hz, 4 H, 4 × H7b), 2.67 (dd, $J_{2a,3}$ = 3.8 Hz, $J_{2a,2b}$ = 15.1 Hz, 4 H, 4 × H2a), 2.57 (dd, $J_{2b,3}$ = 6.7 Hz, $J_{2a,2b}$ = 15.1 Hz, 4 H, 4 × H2b) ppm. LCMS: R_t = 6.93 (ACN/H_2O, 0 → 25%), *m/z* = 693.4 $[M + H]^+$. HRMS: calcd. for $C_{28}H_{44}N_4O_{16}$ [M + H] 693.2831; found 693.2369.

Table 1. List of cross-peaks observed in a ROESY spectrum of **1** in 10% D_2O in H_2O with a mixing time of 180 ms

Atom 1	Atom 2	Intensity[a]
SAA^i:HN	SAA^{i-1}:H2a	m
SAA^i:HN	SAA^{i-1}:H2b	w
SAA^i:HN	SAA^i:H7a	w
SAA^i:HN	SAA^i:H7b	w
SAA^i:HN	SAA^{i-1}:H3	w
SAA^i:H2a	SAA^i:H3	s
SAA^i:H2b	SAA^i:H3	s
SAA^i:H2a	SAA^i:H2b	s
SAA^i:H7a	SAA^i:H7b	s

[a] The intensities of the cross-peaks are given as weak (w), medium (m) or strong (s).

Table 2. List of cross-peaks observed in a ROESY spectrum of **2** in 10% D_2O in H_2O with a mixing time of 180 ms

Atom 1	Atom 2	Intensity[a]
SAA^i:HN	SAA^{i-1}:H2a	m
SAA^i:HN	SAA^{i-1}:H2b	w
SAA^i:HN	SAA^i:H8a	m
SAA^i:HN	SAA^i:H8b	m
SAA^i:HN	SAA^{i-1}:H6	w
SAA^i:H2a	SAA^i:H3	s
SAA^i:H2b	SAA^i:H3	m
SAA^i:H2a	SAA^i:H4	m
SAA^i:H2b	SAA^i:H4	m
SAA^i:H2a	SAA^i:H2b	s
SAA^i:H7a	SAA^i:H7b	s

[a] The intensities of the cross-peaks are given in weak (w) medium (m) or strong (s).

Cyclic Pyranoid SAA Trimer 2: ^{1}H NMR, COSY, ROESY(10% D_2O/H_2O, 600 MHz): δ = 7.88 (t, $J_{8a,N}$ = $J_{8b,N}$ 5.1 Hz, 3 H, 3 × HN), 3.68 (dt, $J_{2a,3}$ = 2.9 Hz, $J_{2b,3}$ = $J_{3,4}$ = 9.2 Hz, 3 H, 3 × H3), 3.58 (m, 3 H, 3 × H8a), 3.51 (m, 3 H, 3 × H5), 3.49 (m, 3 H, 3 × H8b), 3.45 (m, 3 H, 3 × H7), 3.28 (t, $J_{5,6}$ = $J_{6,7}$ = 9.3 Hz, 3 H, 3 × H6), 3.25 (t, $J_{3,4}$ = $J_{4,5}$ = 9.3 Hz, 3 H, 3 × H4), 2.83 (dd, $J_{2a,3}$ = 2.5 Hz, $J_{2a,2b}$ = 14.7 Hz, 3 H, 3 × H2a), 2.46 (dd, $J_{2b,3}$ = 9.1 Hz, $J_{2a,2b}$ = 14.7 Hz, 3 H, 3 × H2b) ppm. LCMS: R_t = 10.48 (ACN/H_2O, 0 → 25%), *m/z* = 610.5 $[M + H]^+$. HRMS: calcd. for $C_{24}H_{39}N_3O_{15}$ [M + H] 610.2460; found 610.2568.

Cyclic Pyranoid SAA Tetramer 4: ^{1}H NMR, COSY(10% D_2O/H_2O, 600 MHz): δ = 7.89 (t, $J_{8a,N}$ = $J_{8b,N}$ 5.3 Hz, 4 H, 4 × HN), 3.64 (d, $J_{8a,8b}$ = 14.6 Hz, 4 H, 4 × H8a), 3.60 (dt, $J_{2a,3}$ = 1.5 Hz, $J_{2b,3}$ = $J_{3,4}$ = 9.5 Hz, 4 H, 4 × H3), 3.42 (t, $J_{4,5}$ = $J_{5,6}$ = 9.0 Hz, 4 H, 4 × H5), 3.35 (t, $J_{6,7}$ = $J_{7,8b}$ = 8.3 Hz, 4 H, 4 × H7), 3.21 (m, 4 H, 4 × H8b), 3.17 (t, $J_{5,6}$ = $J_{6,7}$ = 9.5 Hz, 4 H, 4 × H6), 3.15 (t, $J_{3,4}$ = $J_{4,5}$ = 9.4 Hz, 4 H, 4 × H4), 2.77 (dd, $J_{2a,2b}$ = 14.9 Hz, 4 H, 4 × H2a), 2.39 (dd, $J_{2b,3}$ = 9.8 Hz, $J_{2a,2b}$ = 15.3 Hz, 4 H, 4 × H2b). LCMS: R_t = 18.74 (ACN/H_2O, 0→25%), *m/z* = 813.4 $[M + H]^+$. HRMS: calcd. for $C_{32}H_{52}N4O_{20}$ [M + H] 813.3253; found 813.3482.

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