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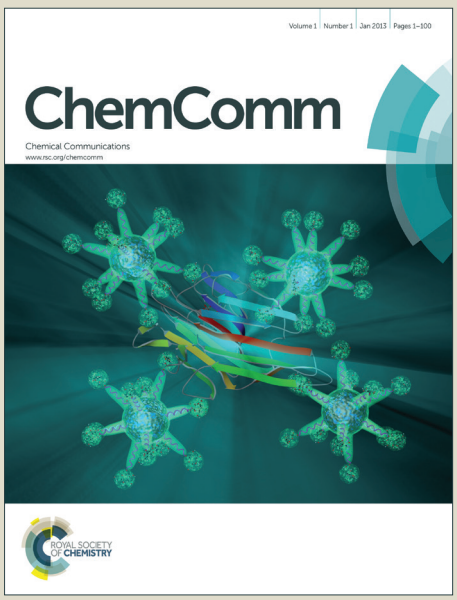

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ARTICLE TYPE

Macrocyclic arylopeptoids – a novel type of cyclic *N*-alkylated aromatic oligoamides forming nanotubular assemblies

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The head-to-tail conversion of linear arylopeptoids (oligomeric *N*-substituted aminomethyl benzamides) into the derived novel macrocycles has enabled the first x-ray structures of arylopeptoid constructs and the identification of well-defined architectures in solution.

Conformational order is a typical characteristic of biologically active oligomers and macrocyclisation is a method often utilized by Nature for bringing conformational order to otherwise flexible oligomeric systems.[1] Indeed, cyclisation of amino acid-based molecules is a strategy used by many organisms to produce compounds for defence, signalling and microbial competition. The study of synthetic macrocycles with well-defined architectures is therefore attracting an ever increasing interest.[2] A family of synthetic macrocycles that has received particular attention since the early 1970's is macrocyclic *N*-alkylated aromatic oligoamides including *N*-alkylated *ortho*-,[3] *meta*-,[4] or *para*[5]-benzanilides (Fig. 1, top left), and *N*-alkylated *para*-cyclophanamides[6] (Fig. 1, top right).

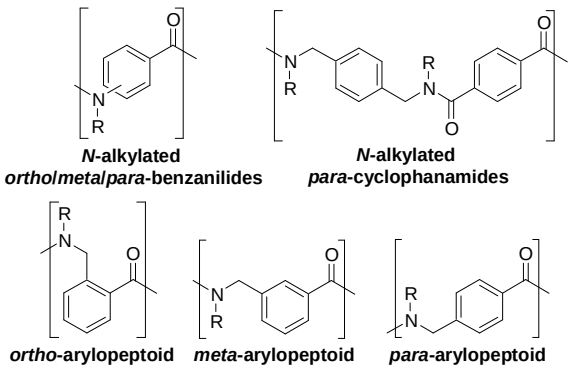
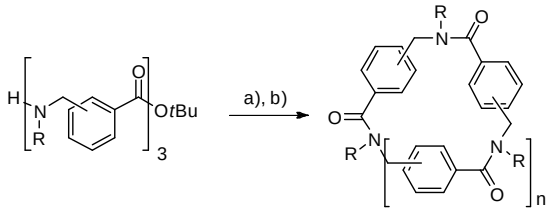


Fig. 1 Repeating units in selected macrocyclic *N*-alkylated aromatic oligoamides.

Notably, the macrocyclic *N*-alkylated *para*-cyclophanamides have been shown to act as hosts for various guests.[6] By installing long hydrophobic side chains, a so called "octopus cyclophane" with a hydrophobic cavity was formed which can be regarded as an apoenzyme model.[6a, b] The *N*-alkylated *para*-cyclophanamides have thus shown promise within applications as selective hosts and artificial enzymes,[7] but they have exclusively been obtained by one-pot polymerisation which severely restricts the available ring sizes, backbone types and side chain diversity. With the aim to develop highly tailorable *N*-alkylated aromatic cyclooligoamides, we have recently begun to unveil the potential of a class of oligoamides with repeating units that are closely related to the *N*-alkylated *para*-cyclophanamides: *N*-alkylated aminomethyl benzamides termed "arylopeptoids" (Fig. 1, bottom).[8] These oligoamides were originally conceived as a subclass of peptoids[9] (*N*-substituted glycines) where each backbone residue is "extended" with a phenyl ring.[10, 11] Since then, we have reported highly efficient synthetic methods to access *ortho*- *meta*-, and *para*-arylopeptoids, all based on "submonomer" approaches.[8] Although these methods allow for access to immense diversity at low cost, our conformational studies have demonstrated that well-defined structures in linear arylopeptoids are only formed under certain limited circumstances due to *cis*-*trans* isomerism of *N*,*N*-disubstituted amide bonds.[8a,b,d] Herein we present that this drawback may be turned into an advantage since it facilitates efficient head-to-tail macrocyclisation,[12] whereby constructs with well-defined structures may be formed.

Six trimeric arylopeptoids ***p*/*m*/*o*-1a-b** with either *para*-backbone (*para*-series), *meta*-backbone (*meta*-series), or *ortho*-backbone (*ortho*-series), carrying either ethyl side chains (a-series) or the more bulky isopropyl side chains (b-series) were used in this preliminary study (Scheme 1). These trimers were obtained as previously described[8a,d] and were then subjected to the deprotection-macrocyclisation procedure we have developed before for β- and α,β-peptoids (Scheme 1 and Table 1).[12c,13] Thus, after *tert*-butyl group removal using TFA/CH_2Cl_2, the crude intermediates were cyclised in the presence of HATU and DIPEA as 5.0 mM solutions in CH_2Cl_2/DMF 4:1.



Scheme 1 Synthesis of macrocyclic arylopeptoids. Key: (a) TFA/CH_2Cl_2 1:1, 0 °C, 3 h (***p*/*m*-1a-b**) or 0 °C, 3 h and rt, 1 h (***o*-1a-b**). (b) HATU (1.2 equiv.), DIPEA (approx. 5.0 equiv.), CH_2Cl_2/DMF 4:1, 0 °C to rt, 3 d. See Table 1 for yields.

Table 1 Yields of macrocyclic arylopeptoids from linear trimers.

Entry	Linear precursor 1		Macrocyclic products 2 and 3	
	Comp.	Backbone	Cyclotrimer (%)	Cyclohexamer (%)
1	***p*-1a**	*para*/Et	***p*-2a**: -[a]	***p*-3a**: 57[b]
2	***p*-1b**	*para*/*i*Pr	***p*-2b**: -[a]	***p*-3b**: 61[b]
3	***m*-1a**	*meta*/Et	***m*-2a**: 83[b]	***m*-3a**: -[a]
4	***m*-1b**	*meta*/*i*Pr	***m*-2b**: 88[b]	***m*-3b**: -[a]
5	***o*-1a**	*ortho*/Et	***o*-2a**: 48[c]	***o*-3a**: 45[c]
6	***o*-1b**[d]	*ortho*/*i*Pr	***o*-2b**: 4[c]	***o*-3b**: 33[c]

[a] Not quantifiable (< 5%). [b] Purified by flash chromatography (see SI for HPLC purity). [c] Purified by preparative HPLC (>99% HPLC purity). [d] conversion ratio: 65%.

For *para*-trimers ***p*-1a** and ***p*-1b** the relatively inflexible backbone combined with the *para*-substitution pattern disfavour direct macrocyclisation as none of the derived cyclotrimers ***p*-2a** and ***p*-2b** were isolated (Table 1, entries 1 and 2).[14] Instead, the corresponding cyclohexamers ***p*-3a** and ***p*-3b** formed by cyclodimerisation were isolated in good yields (57 % and 61 % respectively). On the contrary, the *meta*-substitution pattern in trimers ***m*-1a** and ***m*-1b** strongly favour direct macrocyclisation since the derived cyclotrimers ***m*-2a** and ***m*-2b** were obtained in excellent yields: 83% and 88%, respectively (Table 1, entries 3 and 4). The *ortho*-trimers represented an intermediate case (Table 1, entries 5 and 6). For trimer ***o*-1a** with ethyl side chains, the cyclic trimer ***o*-2a** and the cyclic hexamer ***o*-3a** were obtained in similar amounts (48% and 45% yield respectively). The lowest number of connective bonds between the *C*- and *N*-termini are found in the *ortho*-series but the increased "congestion" of the backbone thus to some degree disfavours direct ring closure. This was underlined by the result obtained for trimer ***o*-1b** which carries the more bulky isopropyl side chains. The reaction produced a complex crude mixture from which we isolated 33% of the cyclohexamer ***o*-3b** and only 4% of the cyclotrimer ***o*-2b**.

Cyclotrimers ***m*-2a** and ***o*-2a** produced ^{1}H NMR spectra with broad signals indicating the presence of several conformers in equilibrium at the NMR time scale. However, decreasing the acquisition temperature to 268 K produced well-resolved spectra with an AB system signal for each backbone methylene indicating a preferred conformation in solution (> 90%). As expected, the cyclotrimers ***m*-2b** and ***o*-2b** carrying more bulky isopropyl side chains furnished well-resolved spectra even at room temperature. Nevertheless, our attempts to elucidate backbone conformation by 2D-NMR experiments were unsuccessful. Fortunately, colourless needle-like crystals formed from EtOAc enabled the crystal structure determination of ***m*-2b,** the first to be solved for an arylopeptoid construct (Fig. 2). The compound crystallises in the $P2_1/c$ space group with four molecules in the unit cell. The backbone of the macrocycle is slightly curved with *trans* amide bonds and the three isopropyl side chains roughly perpendicular to the mean plane of the ring. Two of them are oriented towards the convex face of the structure, together with one carbonyl amide. The third isopropyl group and the remaining two C=O are projected towards the other face. In the crystal lattice, the molecules stack on top of each other along the *a* axis to form tubular assemblies as a result of van der Waals interactions (Fig. 2b and ESI).[15] A second level of organisation based notably on aromatic π-π stacking produces a nanoscale hollow tube (inner 15 x 6 Å) consisting of six parallel, slightly interdigitated, tube-like structures (see ESI).

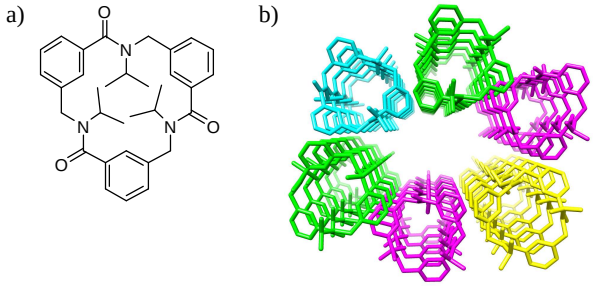


Fig. 2 X-ray structure of ***m*-2b**: a) structure of ***m*-2b** b) Crystal packing: 6-membered columnar assembly.

In the *ortho*-series, single crystals of cyclotrimer ***o*-2a**, suitable for X-ray diffraction were obtained from slow evaporation of MeOH. The crystal structure of ***o*-2a** belongs to space group P1 with two molecules in the unit cell.

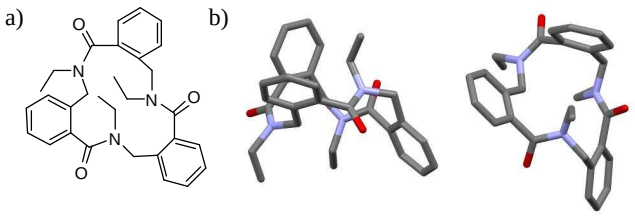


Fig. 3 X-ray structure of ***o*-2a**: a) structure of ***o*-2a**, b) side view (left) and top view (right) of the crystal structure (H-atoms omitted for clarity).

The backbone of ***o*-2a** is characterised by two *trans* and one *cis* amide bonds (Fig. 3). The *cis* amide bond deviates by over 20° from the planarity (ω 21.5°), revealing the strain present in the macrocycle which explains the difficulty encountered in forming cyclic trimers in the *ortho* series. However, according to molecular modelling, the *cis-trans-trans* arrangement (*ctt*) is the lowest in energy among all the possible amide configuration combinations (*ccc*, *ttt*, *ctt*, *cct*) in solution (see ESI for details).

Cyclohexamers ***o*-3a** and ***p*-3a-b** resulting from cyclodimerisation produced ^{1}H NMR spectra reflecting high conformational heterogeneity in solution. By contrast, the single set of signals observed for cyclohexamer ***o*-3b** is indicative of a discrete conformation with a 6-fold rotational symmetry (Fig. 4a). NOESY experiments revealed a *trans* geometry of the amide bonds (see ESI). Starting from energetically optimised dimeric models, we propose a predicted conformation fulfilling criteria arising from the NMR studies i.e. *trans* amide bonds and symmetry (see ESI). This model shows a S_6-symmetry with the *N*-substituents oriented perpendicular to the ring and alternating between the two faces of the macrocycle. This well-ordered conformation of a 30-membered ring in solution lends promise for the development of cyclic arylopeptoids as scaffolds. Moreover a similar conformation was obtained in solid state. Single crystal X-ray diffraction revealed that cyclohexamer ***o*-3b** adopts a quasi planar ring conformation with a large cavity containing one acetonitrile molecule (crystallisation solvent). Despite the absence of NH-O type hydrogen bonding ability, the macrocycles stack along the *b* axis to form a tubular array (Fig. 4b). Consecutive cycles in the column are symmetry related by the b-glide plane perpendicular to a axis and interact through weak CH-O type hydrogen bonds and CH-π type interactions. Besides, a water molecule bridging two consecutive rings through

hydrogen bonds with backbone carbonyl oxygen atoms (D_{O1W-O3} = 2.81 Å and D_{O1W-O6} = 2.79 Å , O-H-O angles of 165° and 160°, respectively) may stabilize this tubular assembly.

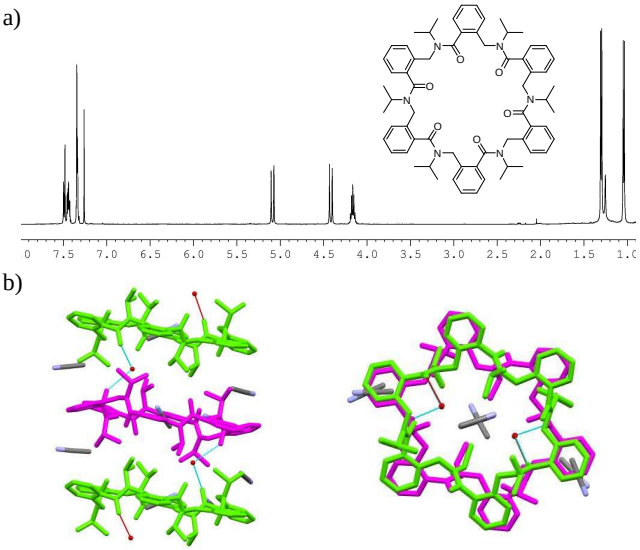


Fig. 4 a) ^{1}H NMR spectra (5 mM in $CDCl_3$ at 298K) and structure of ***o*-3b**; b) Crystal packing of ***o*-3b**: (left) side view (right) top view (H-atoms omitted for clarity); solvent molecules (acetonitrile) are represented in grey-blue sticks and water molecules as red balls, hydrogen bonding between water and backbone carbonyl groups are highlighted in turquoise blue.

Conclusions

Macrocyclic arylopeptoids represent an entirely new and intriguing class of macrocycles with well-defined backbones. The conformational constraint imposed on these oligoamides by cyclisation has resulted in the first X-ray structures of arylopeptoid architectures and the identification of arylopeptoid constructs with well-defined structures in solution. Notably a *ortho*-arylopeptoid cyclohexamer was found to adopt a unique symmetric conformation with an interior cavity sufficiently large to accommodate guest molecules. Linear arylopeptoid precursors of designable lengths with variable side chains are readily available which lends promise of access to a wide diversity of precisely tailored macrocycles. Thus, depending on the side chain decorations, cavity sizes and conformations of the macrocycles, they may be used as hosts for a range of biologically important guest ions and molecules.

Acknowledgements

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Notes and references

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† Electronic Supplementary Information (ESI) available: [Experimental procedures and characterisation data; HPLC and NMR spectra; x-ray structures; molecular modelling]. See DOI: 10.1039/b000000x/

‡ CCDC 967255 (***m*-2b**) - CCDC 967256 (***o*-2a**) and CCDC 982403 (***o*-3b**) contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

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