

# Self-Assembling Nanotubes Consisting of Rigid Cyclic $\gamma$ -Peptides

Liangchun Li, Hongmei Zhan, Pengfei Duan, Jian Liao, Junming Quan, Yu Hu, Zhongzhu Chen, Jin Zhu, Minghua Liu, Yun-Dong Wu,\* and Jingen Deng\*

Self-assembling cyclic peptide nanotubes (SPNs) have been extensively studied due to their potential applications in biology and material sciences. Cyclic  $\gamma$ -peptides, which have a larger conformational space, have received less attention than the cyclic  $\alpha$ - and  $\beta$ -peptides. The self-assembly of cyclic homo- $\gamma$ -tetrapeptide based on *cis*-3-aminocyclohexanecarboxylic acid ( $\gamma$ Ach) residues, which can be easily synthesized by a one-pot process is investigated. Fourier transform infrared (FTIR) and NMR analysis along with density functional theory (DFT) calculations indicate that the cyclic homo- $\gamma$ -tetrapeptide, with a non-planar conformation, can self-assemble into nanotubes through hydrogen-bond-mediated parallel stacking. Atomic force microscopy (AFM) and transmission electron microscopy (TEM) experiments reveal the formation of bundles of nanotubes in  $\text{CH}_2\text{Cl}_2$ /hexane, but individual nanotubes and bundles of only two nanotubes are obtained in water. The integration of TEG (triethylene glycol) monomethyl ether chains and cyclopeptide backbones may allow the control of width of single nanotubes.

## 1. Introduction

Novel functional nanomaterials based on self-organizing systems are of great current interest.<sup>[1,2]</sup> Self-assembling peptide nanotubes (SPNs), formed from cyclic oligopeptides consisting of various amino acids, are widely studied for their potential applications

in areas such as biology and materials science.<sup>[3–6]</sup> In contrast to the morphologies found in inorganic nanostructures, the nanotubes formed by cyclopeptide aggregate into bundles<sup>[3–9]</sup> that may have applications in the preparation of nanosized wires.<sup>[7,10]</sup> Recently, it was reported that the self-assembling morphologies could be controlled by polymer-conjugated peptides.<sup>[11,12]</sup>

The majority of cyclic peptide nanotubes so far have hydrophilic inner surfaces.<sup>[3]</sup> Granja et. al. designed and synthesized cyclic  $\alpha$ , $\gamma$ -peptides, which formed tubular structures via antiparallel  $\beta$ -sheet-like hydrogen bonds.<sup>[7,13–15]</sup> In such structures, the  $\beta$ -methylene moiety of *cis*-3-aminocyclohexanecarboxylic acid ( $\gamma$ Ach) residues is projected into the lumen of the cylinder, creating a partially hydrophobic cavity, which has different transport/carrier properties in polar/apolar solvents

with the modified pore interior.<sup>[13–15]</sup> However, the investigation of the self-assembling morphologies of cyclic homo- $\gamma$ -peptides has not been reported. Herein, we report the first preparation of cyclic  $\gamma$ -peptide **2** (Scheme 1) by one-pot cyclodimerization of dipeptide **1**, which has a hydrophobic inner surface that is

Dr. L. C. Li, Prof. J. G. Deng  
Key Laboratory of Drug-Targeting of Education  
Ministry and Department of Medicinal Chemistry  
West China School of Pharmacy  
Sichuan University  
Chengdu 610041, China  
E-mail: jgdeng@cioc.ac.cn

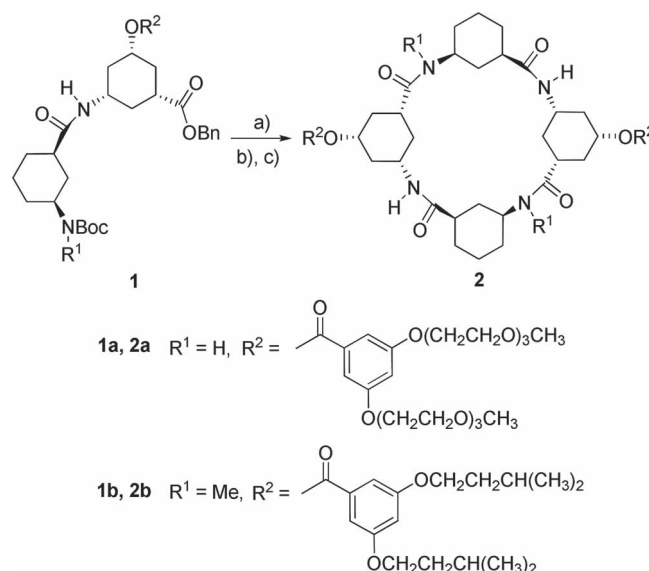


H. M. Zhan, Prof. J. Liao, Y. Hu, Z. Chen, Prof. J. Zhu  
Key Laboratory of Asymmetric Synthesis & Chirotechnology  
of Sichuan Province  
Chengdu Institute of Organic Chemistry the Chinese Academy of Sciences  
Chengdu 610041, China

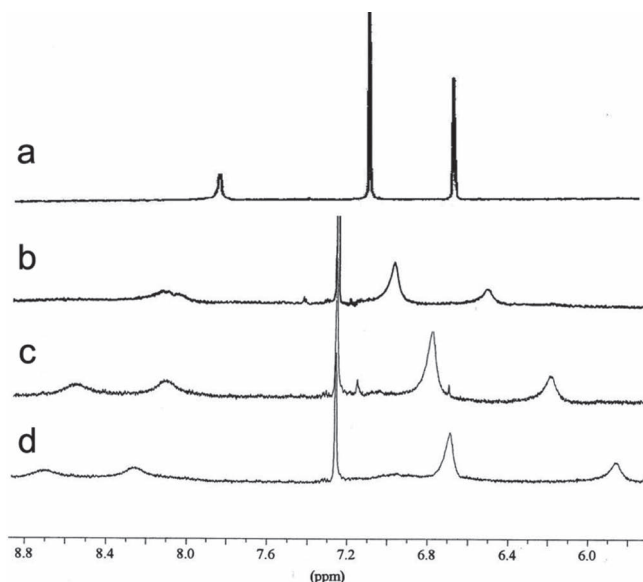
P. F. Duan, Prof. M. H. Liu  
Institute of Chemistry  
the Chinese Academy of Sciences  
Beijing, 100080, China

Dr. J. M. Quan, Prof. Y.-D. Wu  
School of Chemical Biology and Biotechnology  
Peking University Shenzhen Graduate School  
Shenzhen 518055, China  
E-mail: chydwu@ust.hk

DOI: 10.1002/adfm.201200488



**Scheme 1.** Synthesis of **2**: a)  $\text{H}_2$ , Pd-C (10%), DCM; b) TFA, DCM; c) HATU, HOAt, DIPEA, DCM.



**Figure 1.** Selected region of  $^1\text{H}$  NMR spectrum of a solution of **2a**: a) in DMSO, b) 10 mM, c) 20 mM, and d) 40 mM in  $\text{CDCl}_3$ , showing the downfield shifts of two Ph-H and N-H.

decorated by four methylene groups contributed by the cycloalkane rings. Moreover, we demonstrate that well dispersed individual nanotubes can be formed from the cyclic peptide bearing dendritic PEG side chains through parallel  $\beta$ -sheet-like stacking.

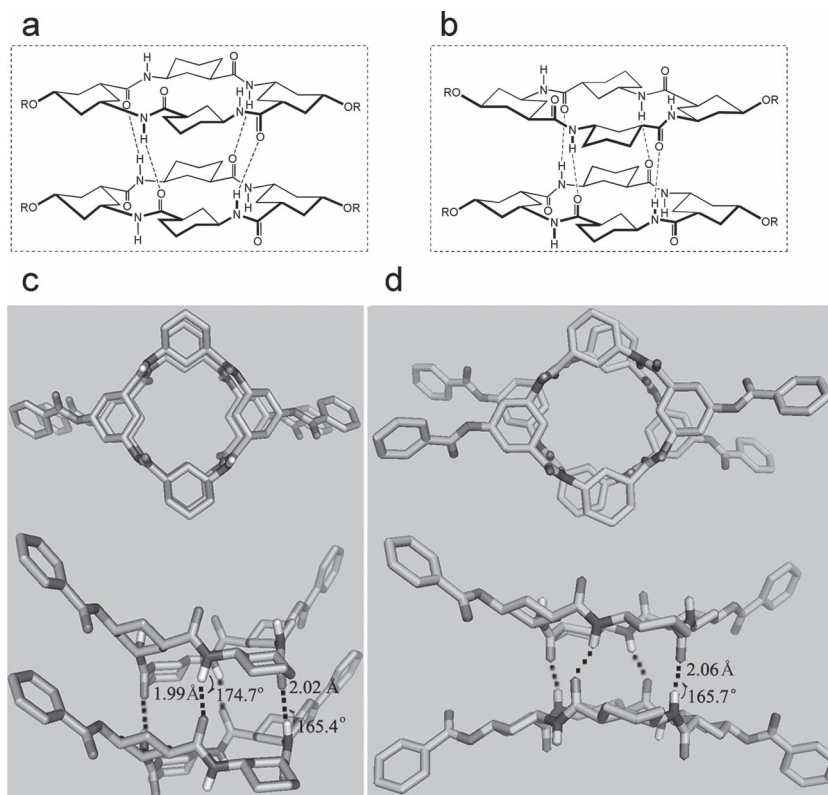
Cyclic  $\gamma$ -peptides, which have a larger conformational space, have received less attention than the cyclic  $\alpha$ - and  $\beta$ -peptides.<sup>[3]</sup> For the design of cyclic  $\gamma$ -peptide nanotubes,  $\gamma$ -Ach was chosen as the basic residue, because the cyclohexane ring of  $\gamma$ -Ach is expected to stabilize the desired flat conformation of cyclic  $\gamma$ -peptide. Such a flat conformation should favor self-assembly of the cyclic  $\gamma$ -peptide into tubular structures through face-to-face molecular stacking mediated by multiple hydrogen bonds.<sup>[13–16]</sup> Thus, cyclic homo- $\gamma$ -peptide **2** was designed to consist of alternating chirality of (1*R*,3*S*)- $\gamma$ -Ach and (1*S*,3*R*)- $\gamma$ -Ach residues. The latter residue carries a hydroxyl group that allows the attachment of various functional groups, which not only increases the solubility of the peptide, but also allows to modify the property of the nanomaterials formed by the peptide for applications in areas such as biosensing, pharmaceuticals, catalysis and electronics, etc.<sup>[1–6]</sup> It is well known that PEGylation leads to improved biocompatibility of proteins and reduced biodegradation rates.<sup>[17]</sup> Based on our own experience,<sup>[18]</sup> 3,5-bis(triethylene glycol monomethyl ether)benzoate, containing two tri-EG (TEG) monomethyl ethers, was used to modify (1*S*,3*R*)- $\gamma$ -Ach. To the best of our

knowledge, this is the first example of cyclic peptides bearing EG side chains.<sup>[19,20]</sup>

## 2. Results and Discussion

Cyclic tetrapeptide **2a** was conveniently prepared from the dimerization of deprotected dipeptide that was resulted from hetero-dipeptide **1a** and obtained in 42% yield by column chromatography with  $\text{CHCl}_3$  and MeOH (30:1, v/v) as mobile phase (Scheme 1). The structure of **2a** was confirmed by NMR and ESI-HRMS analyses. Moreover, the clean formation of **2a** was indicated by examining the crude product using MALDI-TOF mass, which revealed  $[\text{M}+\text{Na}]^+$  peak with a mass/charge ratio of 1412. The fact that no other cyclopeptides and oligopeptides were detected by MALDI-TOF suggested that the rigid conformation of  $\gamma$ -Ach residues had a high propensity for forming the cyclic tetrapeptide.

$^1\text{H}$  NMR spectroscopy was then employed to probe the aggregation of **2a** in solution. In  $\text{DMSO}-d_6$ ,  $^1\text{H}$  NMR spectrum of **2a** displayed sharp and well-resolved signals (Figure 1a). NOESY spectrum of **2a** in  $\text{DMSO}-d_6$  (Figure S10, Supporting Information) shows that the N-H and  $\text{C}_\gamma\text{-H}$  bonds were *trans* to each other while the N-H and the  $\text{C}_\alpha\text{-H}$  of adjacent cyclohexane rings were in a *cis* relationship, as shown in Figure 2a,b. While in  $\text{CDCl}_3$  solution, poor-resolved peaks with significant line-broadening that showed concentration-dependence were



**Figure 2.** Theoretical and calculated dimer resulting from a,c) parallel and b,d) antiparallel  $\beta$ -sheet-like hydrogen bonding of cyclic peptides. The side chains are shown as R for simplified model and the geometry of dimers (all nonpolar hydrogen atoms were merged for clarity) were optimized with B3LYP/6-31G(d) and shown as a top view (top) and side view (bottom).

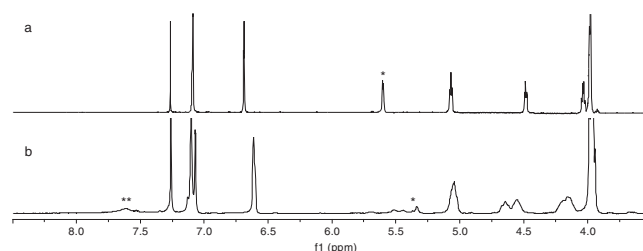
observed (Figure 1b–d, Figure S6 (Supporting Information)). The NH signals lying at  $>8.00$  ppm (10–40 mM in  $\text{CDCl}_3$ ) suggest that these protons are involved in hydrogen-bonding. Thus, these observations are consistent with hydrogen-bond-mediated aggregation of **2a** in  $\text{CDCl}_3$ , giving rise to multiple supra-molecular species in fast exchange on the NMR time scale.<sup>[21]</sup> The aromatic ring proton (Ph-H) signals change from 6.58 and 6.97 ppm (in Figure 1b) to 5.85 and 6.69 ppm (Figure 1d) upon the increase of concentration from 10 mM to 40 mM. The significant upfield shifts of these signals indicate that there is  $\pi$ – $\pi$  stacking interactions between adjacent monomers upon aggregation. Furthermore, an NOE cross peak between the Ph-H and the  $\text{C}_\alpha\text{-H}$  of modified  $\gamma\text{Ach}$  residues from adjacent cyclopeptide molecule exists in the NOESY in  $\text{CDCl}_3$  (Figure S11 vs S10 (Supporting Information)). This suggests that the adjacent cyclopeptide molecules stack in a parallel fashion.

Moreover, FT-IR studies of films deposited on KBr pellets as well as in solution showed substantial evidence for the self-assembly of the peptide by  $\beta$ -sheet like hydrogen bonding (see Supporting Information). The FT-IR spectrum of **2a** in  $\text{CHCl}_3$  showed an amide I absorption ( $\text{C}=\text{O}$  stretching mode), an amide II absorption (mainly N-H bending and C-H stretching modes) and an amide A absorption (N-H stretching mode) at  $1633\text{ cm}^{-1}$ ,  $1552\text{ cm}^{-1}$  and  $3286\text{ cm}^{-1}$ , respectively. The observed amide A band indicates a tight intermolecular hydrogen bonding, and the amide I and II absorptions are characteristic for nanotubes formed from  $\beta$ -sheet-like networks.<sup>[13–15,22,23]</sup> In the meantime, the absence of signal near  $1690\text{ cm}^{-1}$  corresponding to antiparallel  $\beta$ -sheet in **2a** further suggests parallel  $\beta$ -sheet structures in this compound.<sup>[22,23]</sup>

Parallel and antiparallel  $\beta$ -sheet-like hydrogen-bonding of cyclic peptides can both explain the self-assembling behavior (Figure 2a, 2b). To better understand the conformation of this cyclic peptide, the structural and self-assembling features of **2a** were explored by density functional theory (DFT)<sup>[24]</sup> calculations. Because of the conformational rigidity of the  $\gamma\text{Ach}$  residues, only one low-energy conformation was found for the backbone of **2a** and it is noteworthy that the four cyclohexanes form a non-planar (boat-shaped with side view) conformation with an  $S_4$  symmetry (Figure S17, S18, Supporting Information). Further calculations showed that **2a** could form a reasonable dimer via parallel or antiparallel  $\beta$ -sheet-like hydrogen bonding (Figure 2c, 2d and Figure S19, S20 (Supporting Information)). When the optimized structures of the dimers are viewed along the dimer axes (top view in Figures 2c and 2d), it is apparent that one of the monomers is slightly rotated with respect to the other, and the monomers of the antiparallel stacking are rotated more than the parallel stacking. This feature may be attributed to the geometrical characteristic of  $\text{C}=\text{O}\cdots\text{HN}$  hydrogen bonds (the best alignment would be  $120^\circ$  for  $\text{C}=\text{O}\cdots\text{H}$  angles due to the directionality of the oxygen lone pairs) and steric repulsions. Furthermore, the non-planar conformation of **2a** is more fitted for parallel stacking because of the better alignment of the structures. The distance between Ph-H and  $\text{C}_\alpha\text{-H}$  of modified  $\gamma\text{Ach}$  residue from adjacent cyclopeptide molecules in the parallel stacking is shorter ( $3.23\text{ \AA}$ ) than that in the antiparallel stacking ( $7.00\text{ \AA}$ ) (Figure S12, Supporting Information), which is in agreement with the NOESY experiment.

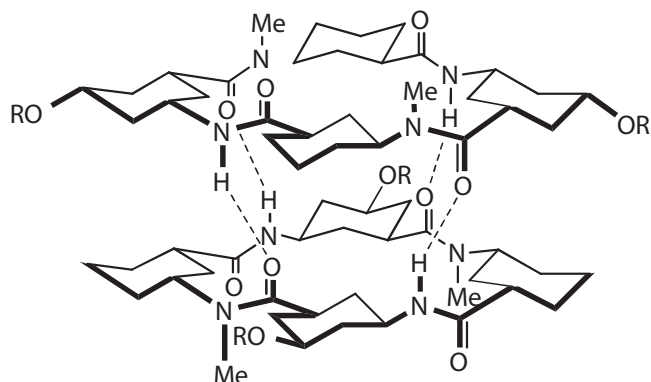
In the gas phase the BSSE-corrected binding energies calculated with the M05-2X/6-31G+(d,p) method<sup>[25–27]</sup> are  $-34.4$  and  $-23.0\text{ kcal/mol}$  for the parallel and antiparallel dimers, respectively. The parallel dimer is more stable than the antiparallel one by about  $11.4\text{ kcal/mol}$ , because the latter forms weaker hydrogen bonds judging from hydrogen-bonding distances and angles and also suffers from a geometrical deformation. The solvent effect on the binding energy was estimated by the IEFPCM (integral equation formalism of the polarizable continuum model)<sup>[28,29]</sup> method at the M05-2X/6-31G+(d,p) level using the gas phase optimized structures. The dimerization energy of the parallel dimer in chloroform is reduced to  $-12.4\text{ kcal/mol}$ , while that of the antiparallel one becomes  $0.1\text{ kcal/mol}$ . These results suggest that the parallel stacking of **2a** is energetically much more favourable than the antiparallel one, and the latter may not be stably formed.

Thus, the above NMR, FT-IR experiments and DFT calculations all suggest the formation of tightly hydrogen-bonded parallel  $\beta$ -sheet structures for cyclopeptide **2a** in  $\text{CDCl}_3$ . We have also obtained more direct evidence for the conformational insights. Previous experiments of cyclopeptides indicated that partial N-methylation of the NH groups of one face in this cyclic  $\gamma$ -peptide may be an excellent strategy for the study of stacking mode.<sup>[6,13–15,21]</sup> Unfortunately, the preparation of N-methyl cyclopeptide of **2a** is unsuccessful. Thus, N-methyl cyclopeptide **2b** was designed and synthesized to study the self-assembly properties (Scheme 1). As shown in Figure 3a, the  $^1\text{H}$  NMR spectrum of **2b** in  $\text{CDCl}_3$  is well defined, which reflects a  $C_2$  symmetry of the monomeric species, and the NH signal at 5.60 ppm shows that it is not involved in hydrogen-bonding. These indicate that **2b** cannot dimerize through antiparallel stacking (parallel stacking is prohibited by the methylation), which is in qualitative agreement with the previously mentioned DFT calculations. When equimolecular **2b** and its enantiomer were mixed in  $\text{CDCl}_3$ , a broad NH signal at 7.50–7.80 ppm of  $^1\text{H}$  NMR spectrum appeared (Figure 3b), indicating the formation of hydrogen-bonds between the two enantiomers of **2b**. In this case, parallel hydrogen-bonding (Figure 4) is possible. As shown in Figure 5, an NOE cross peak between the  $\gamma\text{-H}$  of N-methylated amino acid ( $^{\text{Me}}\text{N-}\gamma\text{Ach}$ ) and the NH of the other  $\gamma\text{Ach}$  exists in the NOESY (can only be observed in the assembly, Figure 4), which strongly suggest the parallel dimer structure. This is in an excellent agreement with the previous observations for the aggregation of **2a**. It can be concluded that cyclotetrapeptide **2a** self-assemble via parallel stacking instead of antiparallel stacking to form nanotubes.



**Figure 3.** Selected region of  $^1\text{H}$  NMR spectrum of a) enantiomerically pure peptide **2b** in  $\text{CDCl}_3$  (30 mM) and b) a mixture of peptides **2b** and its enantiomer (20 mM). \* denotes the NH signal in the monomer (5.60 ppm), \*\* denotes the NH signal in the dimer (7.60 ppm).

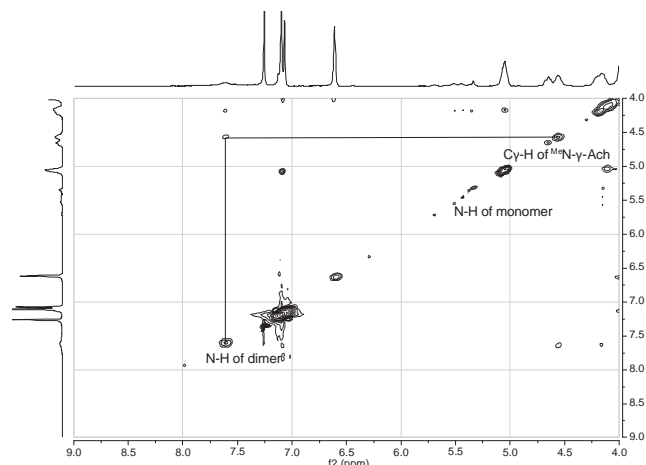




**Figure 4.** Theoretical heterodimer. The side chains are shown as R for simplified model. The structure results from parallel  $\beta$ -sheet-like hydrogen bonding of cyclic peptides **2b** and its enantiomer.

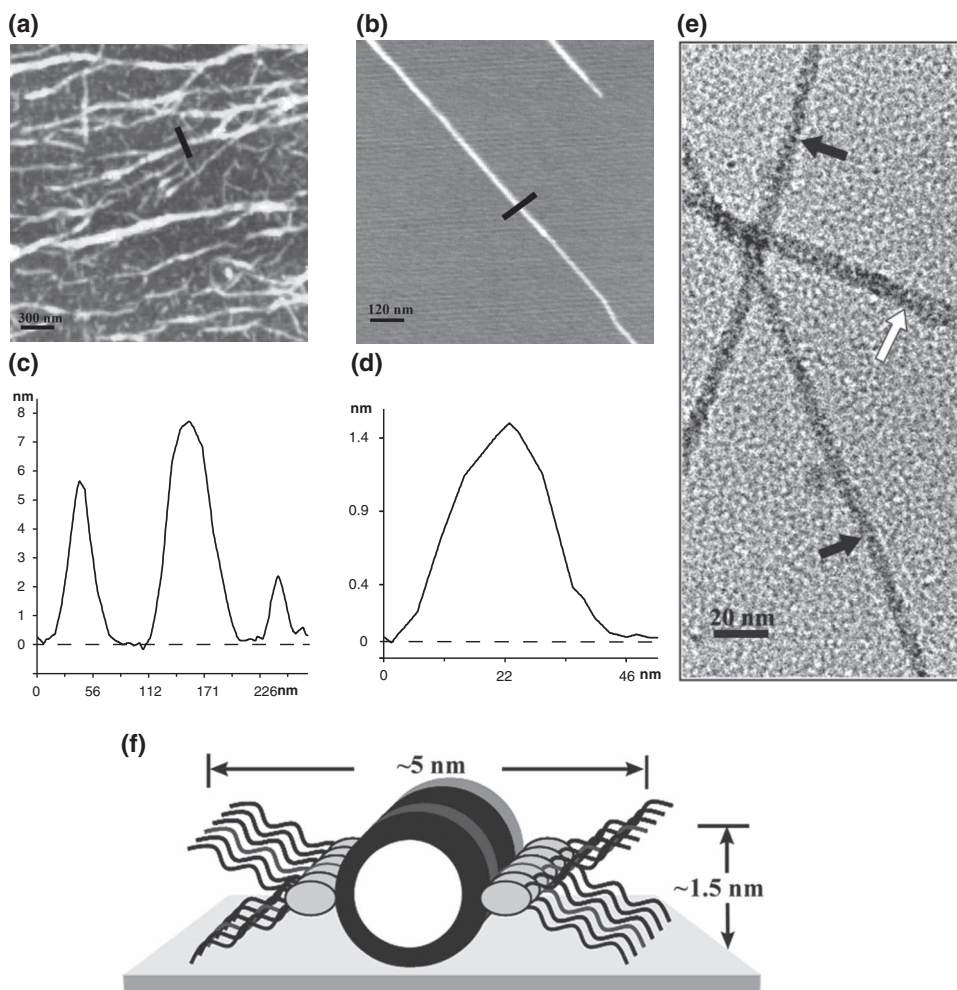
A solution of **2a** in  $\text{CH}_2\text{Cl}_2$  (2 mM) was equilibrated against *n*-hexane via vapor-phase diffusion. After incubating at the room temperature for three days it led to the formation of a colorless gel. The organogel was then dispersed in *n*-hexane by sonication. The resulting sample was examined using atom force microscopy (AFM). A high resolution AFM image of the gel dispersed in *n*-hexane showed a fiber with a height of 2.1–7.8 nm and a length over several hundred nanometres (Figure 6a,c).

Self-assembling in aqueous media is of particular interest. Recently, dendritic aromatic amphiphiles bearing branched EG chains were shown interesting property of enhanced self-assembly into a variety of nanostructures in aqueous media. One-dimensional tubular aggregates were also formed by the self-assembly of an amphiphilic rigid macrocycle in aqueous solution.<sup>[30]</sup> The self-assembling behaviour of **2a**, which has a rigid hydrophobic interior and flexible hydrophilic TEG exterior sidechains, was investigated in aqueous media. A direct investigation on the aggregation of **2a** in aqueous solution was difficult because of its poor solubility in water. Thus, the organogel of **2a** formed in  $\text{CH}_2\text{Cl}_2$ /*n*-hexane was dispersed in water by sonication. A drop of the dispersed suspension was then placed on a mica surface and dried in vacuum. The AFM image of the material after being treated with water revealed long filaments (>1  $\mu\text{m}$ )



**Figure 5.** Selected region of the NOESY spectrum of equimolecular **2b** and its enantiomer in  $\text{CDCl}_3$ .

with height of 1.4–1.6 nm (Figure 6b,d), same as the molecular dimension of the cyclic peptide **2a**. More detailed information of the sample was obtained from transmission electron



**Figure 6.** a,b) AFM images of the gels dispersed in *n*-hexane and in water; c,d) profile obtained along the black line shown in (a,b), respectively. e) TEM field of negatively stained filaments. f) Schematic illustration of branched TEG-peptide nanotubes adsorbed onto a mica surface.

microscopy (TEM), which revealed the presence of long filaments (>400 nm) with widths of ~5 nm and ~9 nm (Figure 6e, shown by black and white arrows, respectively). Thus the AFM and TEM images reveal that the nanomaterial of **2a** dispersed in water exists in both individual nanotubes as shown in Figure 6f (height of about 1.5 nm and width of about 5 nm) and bundles of only two nanotubes (width of about 9 nm). These results also suggest that the TEG side chains of a nanotube extended onto the surface of mica.<sup>[11,12]</sup>

It is quite interesting that the bundled nanotubes of **2a** formed in CH<sub>2</sub>Cl<sub>2</sub>/*n*-hexane deassemble into mono nanotubes and bundles of only two nanotubes upon dispersing into water. It is likely that the hydrophilic TEG chains play a key role. When they are sufficiently hydrated, they suppress lateral aggregation.<sup>[11,12,19,20,30]</sup> Meanwhile, the hydrophobic interactions among the cyclohexyl groups,  $\pi$ - $\pi$  stacking interactions between the benzene rings, and the intermolecular hydrogen bonds are strong enough to maintain the nanotubes in aqueous media. It is worth noting that, in Granja's mixed  $\alpha,\gamma$ -cyclic peptide system the 1D nanotubes<sup>[7]</sup> formed via antiparallel-sheet hydrogen-bonding interaction and the interstrand salt-bridge interactions, while in our system the 1D nanotubes formed via parallel-sheet hydrogen-bonding interaction.

### 3. Conclusions

In summary, we have investigated the self-assembly of cyclic  $\gamma$ -peptide **2a**, which can be easily synthesized by a one-pot process. FT-IR and NMR analysis along with DFT calculations indicate that **2a**, with a non-planar conformation, can self-assemble into nanotubes through hydrogen-bond-mediated parallel stacking. AFM and TEM experiments reveal the formation of bundles of nanotubes in CH<sub>2</sub>Cl<sub>2</sub>/*n*-hexane; but individual nanotubes and bundles of only two nanotubes are obtained in water. The integration of TEG chains and cyclopeptide backbones may allow the control of width of single nanotubes. Further investigation on these issues is currently being carried out to better understand the conformation of analogous  $\gamma$ -peptides and potential transport/carrier applications.

### 4. Experimental Section

**General:**  $\gamma$ -Ach and modified  $\gamma$ -Ach were prepared according to previous reports.<sup>[31,32]</sup> O-(7-azabenzotriazol-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate (HATU), 1-Hydroxy-7-azabenzotriazole (HOAt) were used as obtained from GL Biochem (Shanghai) Ltd. All other reagents obtained from commercial suppliers were used without further purification unless otherwise noted. Dichloromethane (DCM) was dried and distilled over calcium hydride. Analytical thin-layer chromatography was performed on silica gel GF254 plates from Qingdao Haiyang Chemical Co. Ltd. Silica gel flash chromatography was performed using silica gel (200–300 mesh) from Qingdao Haiyang Chemical Co. Ltd. Proton nuclear magnetic resonance (<sup>1</sup>H NMR) spectra were recorded on Bruker WM-600 MHz or WM-300 MHz spectrometers. Chemical shifts were reported in parts per million (ppm,  $\delta$ ) from TMS or solvent resonance as the internal standard. <sup>1</sup>H NMR splitting patterns are designated as singlet (s), doublet (d), triplet (t), quartet (q). All first-order splitting patterns were assigned on the basis of the appearance of the multiplet. Splitting patterns that could not be easily interpreted are designated as

multiplet (m) or broad (br). Carbon nuclear magnetic resonance (<sup>13</sup>C NMR) spectra were recorded on Bruker WM-600 MHz or WM-300 MHz spectrometers. Electrospray (ESI) mass spectra were recorded on a Bruker BioTOF Q mass spectrum. FT-IR measurements were made on a Nicolet MX-1E spectrometer by the KBr method or the 40 mm CHCl<sub>3</sub> solution FT-IR measurements placed in a 0.1 mm KBr solution IR cell were made on a ThermoFisher Nicolet 6700 spectrometer.

**Synthesis of CP 2a:** After two vacuum/H<sub>2</sub> cycles to replace air inside the reaction tube with hydrogen, the mixture of **1a** (50 mg, 0.06 mmol), 10% Pd/C (10 wt% of the substrate) in DCM (2 mL) was vigorously stirred at room temperature (ca. 20 °C) under 1 atm of hydrogen for 24 h. The reaction mixture was filtered using silica gel, and the filtrate was concentrated. Then the BOC group of the residue was removed by trifluoroacetic acid (TFA)/DCM (1:1, v/v), and the product was dissolved in dry 30 mL of DCM. To the solution, HOAt (9 mg, 0.07 mmol), HATU (28 mg, 0.07 mmol), and *N,N*-Diisopropylethylamine (DIPEA) (40  $\mu$ L, 0.25 mmol) were added at room temperature. After stirring the resulting solution until completion of the reaction, the solution was concentrated in vacuum. 50 mL of CHCl<sub>3</sub> was added to the residue and stirred, filtered, and then the combined organic filtrates were washed with 10% KHSO<sub>4</sub> and 10% Na<sub>2</sub>CO<sub>3</sub>. After the organic layer was dried over anhydrous Na<sub>2</sub>SO<sub>4</sub> and concentrated in vacuum, the crude cyclopeptide was purified by column chromatography with CHCl<sub>3</sub> and MeOH (30:1, v/v) as mobile phase to provide a pure **2a** (35 mg) in 42% yield. <sup>1</sup>H NMR (600 MHz, CDCl<sub>3</sub>+CD<sub>3</sub>OD,  $\delta$ ): 0.89–0.95 (m, 4H), 1.11–1.26 (m, 6H), 1.35–1.40 (q, *J* = 11.8 Hz, 2H), 1.44–1.46 (m, 2H), 1.61 (d, *J* = 12.0 Hz, 2H), 1.67–1.76 (m, 8H), 1.81–1.83 (m, 2H), 2.02 (d, *J* = 12.0 Hz, 2H), 2.11–2.19 (m, 4H), 2.27 (t, *J* = 12.0 Hz, 2H), 3.30 (s, 12H), 3.48–3.49 (m, 8H), 3.58–3.62 (m, 18H), 3.65–3.67 (m, 8H), 3.78–3.83 (m, 10H), 4.07 (t, *J* = 4.4 Hz, 8H), 4.87–4.92 (m, 2H), 6.61 (t, *J* = 2.2 Hz, 2H), 7.10 (d, *J* = 2.1 Hz, 4H), 7.47 (d, *J* = 7.8 Hz, 4H); <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>+CD<sub>3</sub>OD,  $\delta$ ): 23.4, 25.4, 29.5, 30.4, 31.3, 35.9, 37.4, 38.2, 39.8, 42.9, 45.2, 47.2, 58.7, 67.6, 69.4, 70.3, 70.4, 70.6, 71.2, 71.7, 106.5, 108.0, 131.8, 159.6, 165.5, 174.5, 176.5; FT-IR (KBr):  $\nu$  = 3283.3 (amide A), 2926.2, 1717.7, 1640.2 (amide I), 1596.4, 1547.6 (amide II), 1447.0, 1296.7, 1176.7, 1103.7, 851.8, 762.0; FT-IR (40 mm, 293 K, CHCl<sub>3</sub>):  $\nu$  = 3286.0 (amide A), 2935.3, 1633.2 (amide I), 1600.1, 1552.2 (amide II), 1446.1, 1299.5, 1241.5, 1217.2, 1175.0, 1104.0; HRMS (ESI, *m/z*): calcd. for C<sub>70</sub>H<sub>108</sub>N<sub>4</sub>Na<sub>2</sub>O<sub>24</sub> ([M + 2Na]<sup>2+</sup>): 717.3569, found: 717.3589; MALDI-TOF (*m/z*): 1412.0 [M+Na]<sup>+</sup>.

**Synthesis of CP 2b:** **2b** was synthesized using the same method like **2a** but starting from **1b** and yield in 30%. <sup>1</sup>H NMR (600 MHz, CDCl<sub>3</sub>,  $\delta$ ): 0.84–0.97 (m, 2H), 1.25–1.35 (m, 4H), 1.37–1.48 (m, 6H), 1.52–1.58 (m, 2H), 1.66–1.73 (m, 12H), 1.80–1.89 (m, 8H), 1.91–1.95 (m, 4H), 2.08 (d, *J* = 12.5 Hz, 2H), 2.18 (t, *J* = 12.1 Hz, 2H), 2.32 (d, *J* = 11.0 Hz, 2H), 2.79–2.83 (m, 2H), 2.87 (s, 6H), 4.01 (t, *J* = 6.6 Hz, 8H), 4.05–4.08 (m, 2H), 4.44 (t, *J* = 11.8 Hz, 2H), 5.01–5.05 (m, 2H), 5.60 (d, *J* = 9.2 Hz, 2H), 6.64 (s, 2H), 7.13 (d, *J* = 2.2 Hz, 4H); <sup>13</sup>C NMR (150 MHz, CDCl<sub>3</sub>,  $\delta$ ): 22.6, 23.9, 25.0, 25.9, 28.4, 29.7, 32.4, 34.6, 35.7, 36.4, 37.0, 37.9, 44.4, 45.2, 66.7, 51.9, 70.9, 106.4, 107.8, 132.0, 160.2, 165.6, 173.9, 175.0; HRMS (ESI, *m/z*): calcd. for C<sub>64</sub>H<sub>96</sub>N<sub>4</sub>O<sub>12</sub>Na ([M + Na]<sup>+</sup>): 1135.6922, found: 1135.6973.

**Preparation of the Gels:** A solution of 2 mm of **2a** in CH<sub>2</sub>Cl<sub>2</sub> equilibrated against *n*-hexane via vapor-phase diffusion, and then incubated at room temperature for three days, the colorless gel was formed.

**AFM Measurements:** After the gels were dispersed in *n*-hexane by sonication, a piece of clean hydrophobic silicon slide was dipped into the hot *n*-hexane solution of the gels and cooled to room temperature, then the slide was dried under vacuum for AFM measurement. Similarly, after the gels were dispersed in water by sonication, a drop of dispersed suspension was placed on a mica surface and dried in vacuum for AFM measurement. All AFM pictures were measured with a Digital Instrument Nanoscope III Multimode system (Santa Barbara, CA) with a silicon cantilever using the tapping mode. All the AFM images are shown in the height mode without any image processing except flattening.

**TEM Measurements:** The gels were dispersed in water by sonication and droplets of 10  $\mu$ L of the suspension were placed onto the specimen

holder, a 400 mesh copper grid. After 2 minutes, the specimens were stained with 2% (w/v) uranyl acetate by incubation of 2 min. Excess of solution was removed by suction with a filter paper attached to the edge of the grid, leaving behind a thin film of solution, which dried rapidly. Specimens were studied with Tecnai G2 F20 U-TWIN electron microscope operating at 200 kv. Images were analyzed with an Analysis 3.2 software package (Soft Imaging System GmbH, Münster, Germany).

## Supporting Information

Supporting Information is available from the Wiley Online Library or from the author.

## Acknowledgements

The authors thank the National Nature Science Foundation of China (grant No. 91027022, 21002099 and 20428202) and the National Basic Research Program of China (973 Program, No. 2010CB833300) for financial support of the research.

Received: February 17, 2012  
Published online: April 12, 2012

- [1] R. V. Ulijn, A. M. Smith, *Chem. Soc. Rev.* **2008**, 37, 664.
- [2] B. A. Grzybowski, C. E. Wilmer, J. Kim, K. P. Browne, K. J. M. Bishop, *Soft Matter* **2009**, 5, 1110.
- [3] R. J. Brea, C. Reiriz, J. R. Granja, *Chem. Soc. Rev.* **2010**, 39, 1448.
- [4] D. T. Bong, T. D. Clark, J. R. Granja, M. R. Ghadiri, *Angew. Chem. Int. Ed.* **2001**, 40, 988.
- [5] X. Gao, H. Matsui, *Adv. Mater.* **2005**, 17, 2037.
- [6] D. Seebach, J. Gardiner, *Acc. Chem. Res.* **2008**, 41, 1366.
- [7] C. Reiriz, R. J. Brea, R. Arranz, J. L. Carrascosa, A. Garibotti, B. Manning, J. M. Valpuesta, R. Eritja, L. Castedo, J. R. Granja, *J. Am. Chem. Soc.* **2009**, 131, 11335.
- [8] T. Nakanishi, H. Okamoto, Y. Nagai, K. Takeda, I. Obataya, H. Mihara, H. Azebara, Y. Suzuki, W. Mizutani, K. Furukawa, K. Torimitsu, *Phys. Rev. B* **2002**, 66, 165417.
- [9] R. Hourani, C. Zhang, R. van der Weegen, L. Ruiz, C. Li, S. Keten, B. A. Helms, T. Xu, *J. Am. Chem. Soc.* **2011**, 133, 15296.
- [10] M. Reches, E. Gazit, *Science* **2003**, 300, 625.
- [11] J. Couet, J. D. J. S. Samuel, A. Kopyshev, S. Santer, M. Biesalski, *Angew. Chem. Int. Ed.* **2005**, 44, 3297.
- [12] M. G. J. ten Cate, N. Severin, H. G. Börner, *Macromolecules* **2006**, 39, 7831.
- [13] M. Amorín, L. Castedo, J. R. Granja, *J. Am. Chem. Soc.* **2003**, 125, 2844.
- [14] M. Amorín, L. Castedo, J. R. Granja, *Chem. Eur. J.* **2005**, 11, 6543.
- [15] R. García-Fandiño, J. R. Granja, M. D'Abramo, M. Orozco, *J. Am. Chem. Soc.* **2009**, 131, 15678.
- [16] F. Fujimura, M. Fukuda, J. Sugiyama, T. Morita, S. Kimura, *Org. Biomol. Chem.* **2006**, 4, 1896.
- [17] F. M. Veronese, *Biomaterials* **2001**, 22, 405.
- [18] P. N. Liu, Y. C. Chen, X. Q. Li, Y. Q. Tu, J. G. Deng, *Tetrahedron: Asymmetry* **2003**, 14, 2481.
- [19] H. M. König, A. F. M. Kilbinger, *Angew. Chem. Int. Ed.* **2007**, 46, 8334.
- [20] T. Xu, N. Zhao, F. Ren, R. Hourani, M. T. Lee, J. Y. Shu, S. Mao, B. A. Helms, *ACS Nano* **2011**, 5, 1376.
- [21] W. S. Horne, C. D. Stout, M. R. Ghadiri, *J. Am. Chem. Soc.* **2003**, 125, 9372.
- [22] T. D. Clark, J. M. Buriak, K. Kobayashi, M. P. Isler, D. E. McRee, M. R. Ghadiri, *J. Am. Chem. Soc.* **1998**, 120, 8949.
- [23] K. Rosenthal-Aizman, G. Svensson, A. Undén, *J. Am. Chem. Soc.* **2004**, 126, 3372.
- [24] Gaussian 03, Revision E.01, M. J. Frisch, G. W. Trucks, H. B. Schlegel, G. E. Scuseria, M. A. Robb, J. R. Cheeseman, G. Scalmani, V. Barone, B. Mennucci, G. A. Petersson, H. Nakatsuji, M. Caricato, X. Li, H. P. Hratchian, A. F. Izmaylov, J. Bloino, G. Zheng, J. L. Sonnenberg, M. Hada, M. Ehara, K. Toyota, R. Fukuda, J. Hasegawa, M. Ishida, T. Nakajima, Y. Honda, O. Kitao, H. Nakai, T. Vreven, J. A. Montgomery Jr., J. E. Peralta, F. Ogliaro, M. Bearpark, J. J. Heyd, E. Brothers, K. N. Kudin, V. N. Staroverov, R. Kobayashi, J. Normand, K. Raghavachari, A. Rendell, J. C. Burant, S. S. Iyengar, J. Tomasi, M. Cossi, N. Rega, J. M. Millam, M. Klene, J. E. Knox, J. B. Cross, V. Bakken, C. Adamo, J. Jaramillo, R. Gomperts, R. E. Stratmann, O. Yazyev, A. J. Austin, R. Cammi, C. Pomelli, J. W. Ochterski, R. L. Martin, K. Morokuma, V. G. Zakrzewski, G. A. Voth, P. Salvador, J. J. Dannenberg, S. Dapprich, A. D. Daniels, Ö. Farkas, J. B. Foresman, J. V. Ortiz, J. Cioslowski, D. J. Fox, Gaussian, Inc., Wallingford CT, **2009**.
- [25] Y. Zhao, N. E. Schultz, D. G. Truhlar, *J. Chem. Theory Comput.* **2006**, 2, 364.
- [26] Y. Zhao, D. G. Truhlar, *J. Chem. Theory Comput.* **2007**, 3, 289.
- [27] R. García-Fandiño, L. Castedo, J. R. Granja, S. A. Vázquez, *J. Phys. Chem. B* **2010**, 114, 4973.
- [28] V. Barone, M. Cossi, *J. Phys. Chem. A* **1998**, 102, 1995.
- [29] V. Baribe, M. Cossi, J. Tomasi, *J. Comput. Chem.* **1998**, 19, 404.
- [30] J.-H. Ryu, D.-J. Hong, M. Lee, *Chem. Commun.* **2008**, 1043.
- [31] Y. Hu, S. L. Yu, Y. J. Yang, J. Zhu, J. G. Deng, *Chin. J. Chem.* **2006**, 24, 795.
- [32] H. M. Zhan, Y. Hu, J. Liao, J. Zhu, J. G. Deng, *Chin. J. Synth. Chem.* **2007**, 15, 588.