

LETTERS

Crystalline Glycylglycine Bolaamphiphile Tubules and Their pH-Sensitive Structural Transformation

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The assembled structure of the heptane bolaamphiphile, bis(*N*- α -amido-glycylglycine)-1,7-heptane dicarboxylate, displays a sensitivity to the acidity of a solution. At pH 4, the heptane bolaamphiphile grows to a crystalline tubule in two weeks. At pH 8, a helical ribbon structure is formed in one week. The degree of carboxylic acid protonation was used to control the final assembled structures since the structures are determined by the strengths of the amide–amide and carboxylic acid dimer hydrogen bonds. Direct structural transformation between tubules and helical ribbons was also confirmed as a function of pH using optical microscopy and Raman microscopy. Conversion from the helical ribbons to the tubules occurs within one day, while the reverse conversion, from the tubules to the helical ribbons, is ten times slower.

In order to design nanometer-scale devices, understanding the rules of molecular self-organization has been an important challenge.^{1,2} We have been able to learn assembly mechanisms from a variety of biological systems that build functional nanoscale structures.³ To match the ability of self-organization in biological systems, peptide assemblies have been synthesized that mimic natural systems.^{4–6} Peptide assemblies may also have potential application as delivery vehicles into living cells.

It has been observed that crystalline microsphere assemblies are triggered by protonation and that the microspheres are formed instantaneously.^{7,8} These results indicate that protonation of carboxylic groups within molecular assemblies can change their structures dramatically.^{9,10} Recently, a family of bolaamphiphiles was described whose protonation induced tubule and vesicle formations.¹¹ Our bolaamphiphiles have two carboxylic acid headgroups and the assembled structures are expected to be sensitive to solution pH.

In this work, one of the bolaamphiphiles, bis(*N*- α -amido-glycylglycine)-1,7-heptane dicarboxylate (Figure 1), was dispersed in solutions of various pH. Synthesis of the heptane bolaamphiphile is described elsewhere.¹¹ Assemblies in citric

acid solutions appeared after one to three weeks at room temperature. At pH 8, the heptane bolaamphiphiles assembled into a helical ribbon in one week (Figure 1a). At pH 4, the bolaamphiphiles assembled into a tubule in two weeks (Figure 1b). The tubule diameter is much smaller than the width of the helical structure. Figure 1b suggests that the tubules are hollow structures.

Raman spectra of the helical ribbon, tubule, and heptane bolaamphiphile crystal phases (Figure 2) were obtained using a Raman microscope (Jobin Yuon/Horiba, LabRam). The crystalline spectrum (c) and the tubule spectrum (b) are almost identical. Thus, the assembled bolaamphiphile tubules have a crystalline structure. Discrepancies of peak positions between the helical ribbon spectrum (a) and tubule spectrum (b) occur in the region from 1600 to 1700 cm^{-1} . This region represents C=O stretches in amide groups. Observed C=O stretch frequency shifts depend on the strengths of the intermolecular hydrogen bonds between C=O and HN. Raman spectra show that the tubule and crystal forms have two C=O peaks at 1637 and 1660 cm^{-1} while the helical ribbon phase has a peak for the C=O stretch at 1644 cm^{-1} . The C=O stretch with the

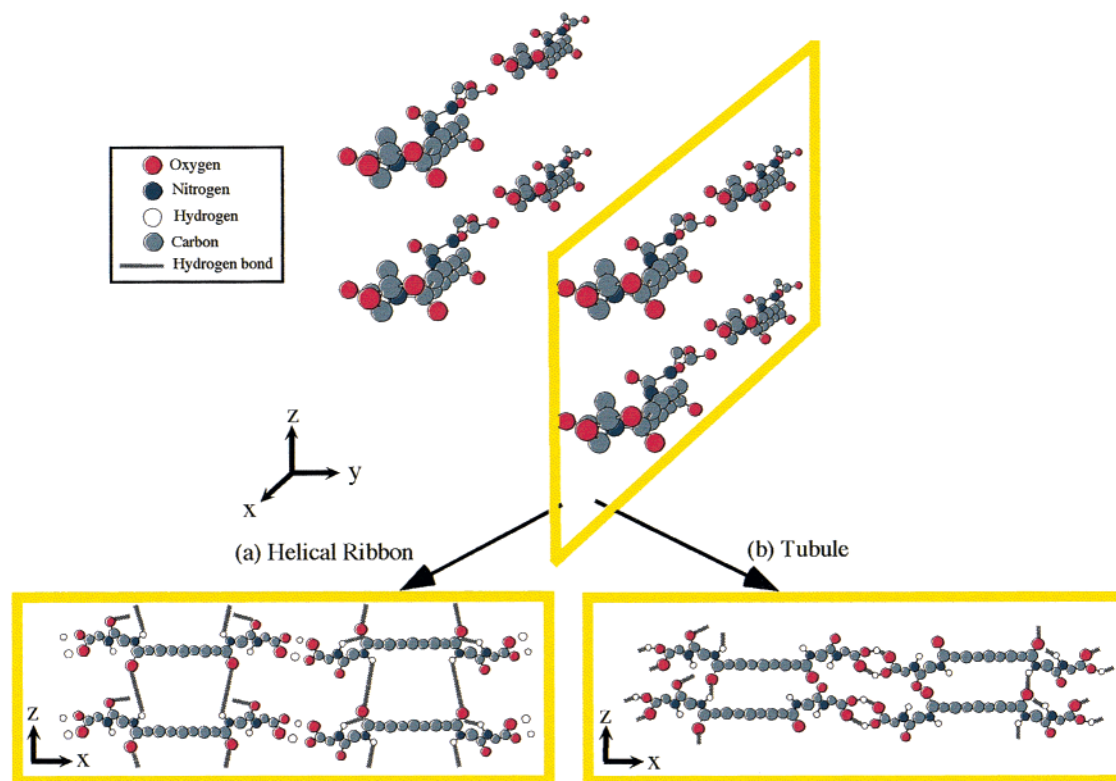


Figure 3. Assembled structures of the heptane bolaamphiphile in (a) the helical ribbon: A pair of the heptane bolaamphiphiles is connected by two amide–amide hydrogen bonds along the z direction and along the $1/2y + 1/2z$ direction, respectively, while a pair of heptane bolaamphiphiles is connected via acid–anion interactions ($\text{COO}^- - \text{H}^+ - \text{OOC}$) along the x direction. (b) The tubule: A pair of heptane bolaamphiphiles is connected by hydrogen bonds between two COOH groups via acid–acid dimer interactions in the x direction. An intermolecular amide–amide hydrogen bond is formed along the z direction and along the $1/2y + 1/2z$ directions, respectively.

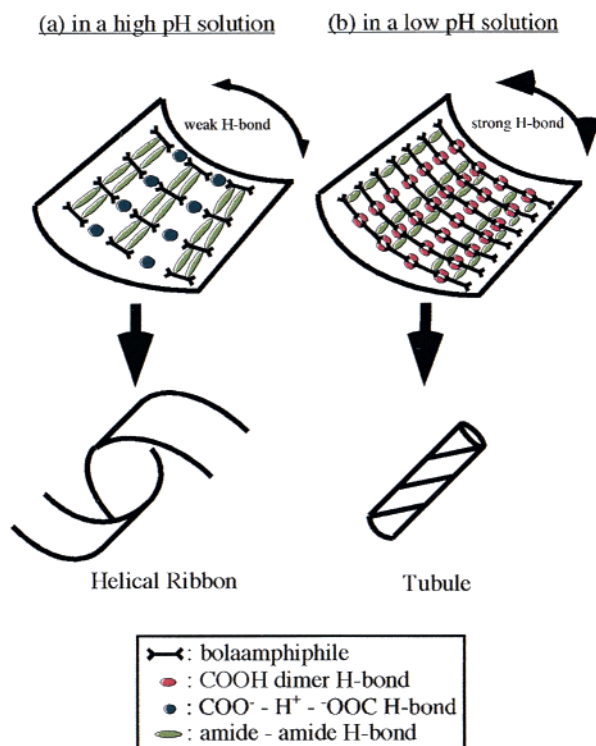


Figure 4. Proposed heptane bolaamphiphile assembly mechanism: (a) the helical ribbon (b) the tubule.

amide and two weak anion–acid hydrogen bonds (helical ribbon), or four strong amide–amide and two acid–acid hydrogen bonds (tubule). The helical ribbons have the same number of hydrogen bonds as the hexane bolaamphiphile assembly, but

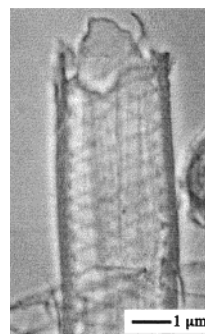


Figure 5. Tubule assembly of the heptane bolaamphiphile in a pH = 5 solution by light microscopy.

the hydrogen bonds in the helical ribbons are weaker. Although the hydrogen bonds in the tubules are stronger, more hydrogen bonds are formed in the hexane bolaamphiphile assembly. Therefore, the heptane bolaamphiphiles seem to have more freedom in the assemblies for molecular tilt to develop. The degree of freedom in self-assemblies, therefore, may play an important role in giving structural transformations with pH change.

Direct structural transformation between the tubules and helical ribbons was also confirmed as a function of pH. Conversion from the helical ribbons to the tubules occurs within one day, while the reverse conversion, from the tubules to the helical ribbons, is ten times slower. Yield of the helical ribbons from tubules under basic conditions is much lower than the yield of tubules from the helical ribbons under acidic conditions. This observation is reasonable since the crystallinity of the tubule is higher than the helical ribbon probed by Raman microscopy.

More flexible helical ribbon structures seem to undergo structural transformations relatively easily.

The heptane bolaamphiphile results presented here may have potential in controlled release applications. Carriers with simple release mechanisms may be more effective if they can be activated at the desired target area. Releasing via structural transformation is one of the simplest mechanisms. The rigid heptane bolaamphiphile tubule structure may also have an advantage for holding molecules until they reach the target location. Optimization of the bolaamphiphile configurations and experimental conditions is proceeding to increase the yield of tubules from the helical ribbon for these applications.

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