Electronic Supplementary Information (ESI)

Self-assembly of collagen peptides into hollow microtubule

Armando E. Reimer, Katie M. Feher, Daniel Hernandez, Katarzyna Slowinska*

Department of Chemistry and Biochemistry, California State University, Long Beach, 1250 Bellflower Blvd.,
Long Beach, CA 90840
To whom correspondence should be addressed. E-mail: katarzyna.slowinska@csulb.edu

Contents:
1. Experimental: Materials – S1
2. Experimental: Circular Dichroism – S1
3. Experimental: Light Microscopy – S1
4. Experimental: Thermogravimetric Analysis (TGA) – S1
5. Supplementary Figure S1: CD of F-POG4 unassembled – S2
6. Supplementary Figure S2: Histogram of microtube size distribution – S2
7. Supplementary Figure S3: microtube stability in pH 2 and 10 – S3
8. Enlarged Figure 1 from the text – S4
9. Enlarged Figure 2 from the text – S5

Materials

F-PGO4 [FITC-(β)Ala-Gly-Gly-(Por-Hyp-Gly)₄-COONH₂ (EX=494 nm, EM= 521 nm) was purchased from
tufts Core Facility (solid support synthesis and HPLC purification). Poly-HEMA [Poly(2-hydroxyethyl metha-
crylate, 1.88% cross-linked] and PAM [polyacrylamide, 1% cross-linked] were received as a 3mm-thick slab
from Dr. Helton (University of Washington). Rhodamine B cyclohexanolamine (EX=540 nm, EM= 625 nm)
was received from Dr. Schramm (CSULB). The buffers were purchased as standards from Fisher Scientific: car-
bonic acid, pH=10.01, 0.01M; hydrochloric acid, pH=2.00, 0.01M. Distilled water was purified by Milli-Q (Mil-
lipore) deionizing system.

Circular Dichroism

The spectra were recorded with Jasco J-810 spectropolarimeter. A scan speed of 50 nm/min and 0.2 cm cell were
used. A reference spectrum containing DI water was subtracted from the peptide containing samples. The con-
centration of peptide solution was 1.1×10⁻⁷ M.

Light Microscopy

The images of microtubes were recorded with Nikon TE2000 equipped with mercury lamp (bright field and fluo-
rescent) with 10x and 40x objectives. The more detailed images were recorded with Olympus Confocal Micro-
scope (FluoView 1000) with 20x, 40x and oil 100x. The z-scans were performed every 0.1 μm. Before imaging
the samples were washed with distill water, and placed on the microscope slide (single well, Fisher Scientific),
covered with water and cover slip.

Thermogravimetric Analysis (TGA)

The 0.5 mg of dehydrated microtubes (desiccator) or lyophilized peptide was placed in the TA Instruments ana-
lyzer (SDT Q600). The samples were equilibrated for 10 min in 100 ml/min nitrogen flow. The temperature ramp
was 10°C/min form ambient temperature.
Supplementary Figures:

**Figure S1.** Circular Dichroism spectrum of unassembled F-POG4. The lack of positive peak at 220 indicates coil conformation and lack of triple helix.

**Figure S2.** The distribution of microtube outer diameter. The distribution was generated from measurements of 60 different fiber diameters observes with confocal microscopy. Average = 2.0, SD = 0.4 μm.
Figure S3. Bright field images of F-POG4 microtubes after incubation in (a) pH = 2.00, (b) pH = 10.01, for 65h at room temperature. The image is taken with 40x objective and microtubes are immersed in the buffer solution.
**Figure 1.** Fluorescence (a) 10x obj, (b) 20x obj and bright field (b) 20x obj , (c,d,e) 40x obj images of F-POG4 microtubes. The black arrows mark the closed end of the tubes. The white arrows mark the “buds” during the growth process.
Figure 2. Confocal fluorescence images of F-POG4 micro-tubes at 100x obj (a) tubes filled with F-POG4 “growth” solution, (b) empty tubes after 30 min in boiling water, (c, d) tubes field with rhodamine-BC after 36h of incubation; (c) rhodamine filter, (d) FITC filter. The arrows point to the cross-section of a tube: inside filled with rhodamine-BC (c) and wall outlined with FITC (d). Scale bar = 10μm for all images.