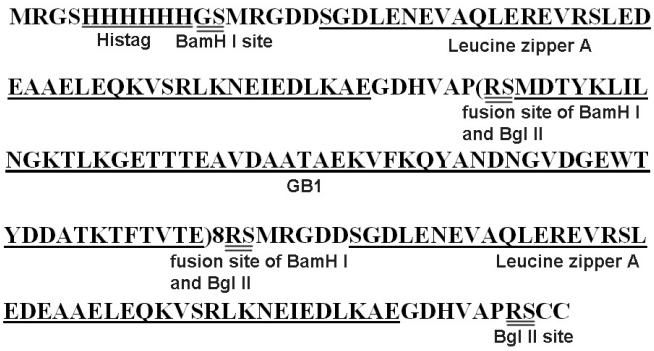
Electronic Supplementary Information

Experimental section

Protein Engineering and hydrogel formation

The gene encoding protein GB1 was a generous gift from David Baker of University of Washington. The gene that encodes G8 was constructed as previously reported[1, 2]. The DNA sequence of leucine zipper based coiled coil domain A, flanked with a 5' BamHI restriction site and 3' BglII and KpnI restriction sites, was synthesized by PCR (polymerase chain reaction) based oligonucleotides assembly. The expression vectorrs of pQE80L-A(G)8A was constructed by iterative cloning A, (G)8 and A genes into empty pQE80L vector, on the basis of the identity of the sticky ends generated by BamHI and BglII restriction enzymes. The resulting sequence of the entire A(G)8A protein is shown in Scheme S1.The expression vector was transformed into *Escherichia coli* strain DH5α. Cultures were grown at 37 ^{o}C in 2.5% LB containing 100mg/L ampicillin, and induced with 0.8mM isopropyl-1-β-D-thiogalactoside (IPTG) when its optical density reached ~1. Protein expression continued for 5 hours. The cells were harvested by centrifugation at 15,000g for 15min and lysised using French press. The soluble fraction was purified using Co^{2+} affinity chromatography. 10mM dithiothreitol (DTT) was added to both washing and elution buffers to avoid the oxidation of terminal cysteins of A(G)8A. The yield of the protein A(G)8A was in the range of 40mg to 80mg per liter of culture. The purity of the purified A(G)8A is around 90%, as estimated from the SDS-PAGE using AlphaEaseFC software (Version 4.0.0, Alpha Innotech Coporation, San Leandro, CA 94577) . The 10% "impurity" was likely to be truncated fragments of A(G)8A, which were frequently observed in the expression of polyproteins, such as (G)8.

The purified protein was then dialyzed against deionized water for 3 days to remove all the salt from elution buffer. During dialysis, the water was changed every 12 hours. The protein was then lyophilized after dialysis. The hydrogel was made by re-dissolve the protein sample into phosphate buffer (100mM, pH 7.6). Vigorously mixing helps dissolution of proteins. The trapped air bubbles can be removed by fast spinning.



Scheme S1. Protein sequence of A(G)8A. The sequence MRGSHHHHHH is resulted from the expression vector pQE80L; residues RS between leucine zipper A and GB1 domains in the protein sequence are resulted from either BglII site or the fusion site of BamHI and BglII.

Circular dichroism measurement (CD)

CD spectra were recorded on Jasco-J810 spectropolarimeter flushed with nitrogen gas. The spectra were recorded in a 0.2cm path length cuvette at a scan rate of 50nm/min. The protein samples were measured in 0.5×PBS at pH 7.4. Data have been corrected for buffer contributions. For each protein sample, an average of three scans is reported and the CD signal was converted into mean residue ellipticity (MRE) using following equation: $\theta_{MRE} = (100 \cdot \theta_{obs})/[dC(n-1)]$, where θ_{obs} is the observed ellipticity (in deg), d is path length (in cm), C is concentration of protein samples (in M), and n is total number of amino acids in the protein. For thermal melting measurement, the temperature increased from 15^{o}C to 90^{o}C with a rate of 3^{o}C/min.

Scanning electron microscopy (SEM)

SEM imaging of hydrogel was taken using Hitachi S4700 Scanning electron microscope. The 7% hydrogel sample was prepared in Eppendorf tubes and left at room temperature for 24 hours to allow the complete formation of hydrogels. Then the samples were shock-frozen in liquid nitrogen, quickly transferred to a freeze drier and lyophilized for 12 hours. The dry samples were then carefully fractured in liquid nitrogen and fixed on aluminum stubs. The surface of the sample was coated by 1nm of gold before SEM observation.

Erosion measurement

The erosion rate of 7% hydrogel A(G)8A was measured using similar method as reported by Shen et al[3]. 100mg of hydrogel was transferred into a cylindrical glass tube with a flat bottom (1.05cm diameter). The glass tube with the hydrogel was then centrifuged at 1700g for 10 minutes to completely flat down hydrogel sample to the bottom and smooth the surface of the hydrogel. The thin gel film together with the glass tube was then soaked in 5mL of 100mM phosphate buffer, pH 7.6, in a scintillation vial. The whole setup was placed on a compact rocker (FINEPCR) tilting at 50 rpm with amplitude of ± 9^{o}, at room temperature. The erosion profiles were determined by measuring the protein absorbance at 280 nm of the supernatant at successive time points using Nano-drop ultraviolet-visible spectrophotometer. Two different samples were measured and the average value was reported.

Supplementary Figures

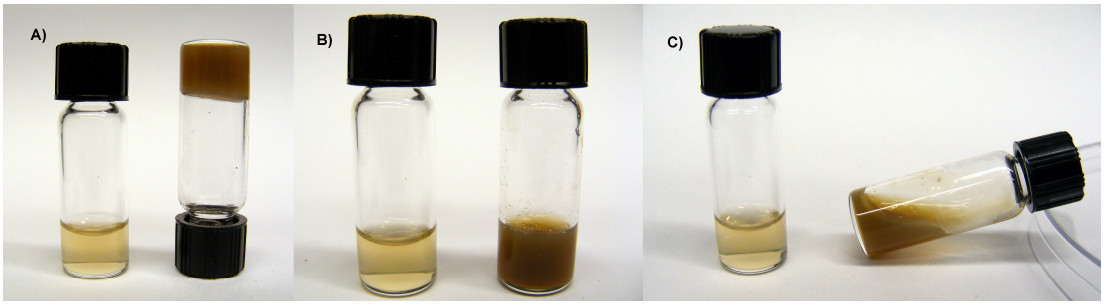


Fig. S1 A) hydrogel of A(G)8A at room temperature (the right one; the left, 7% (G)8 solution for comparison) B) and C) at 60^{0}C for 10 s.

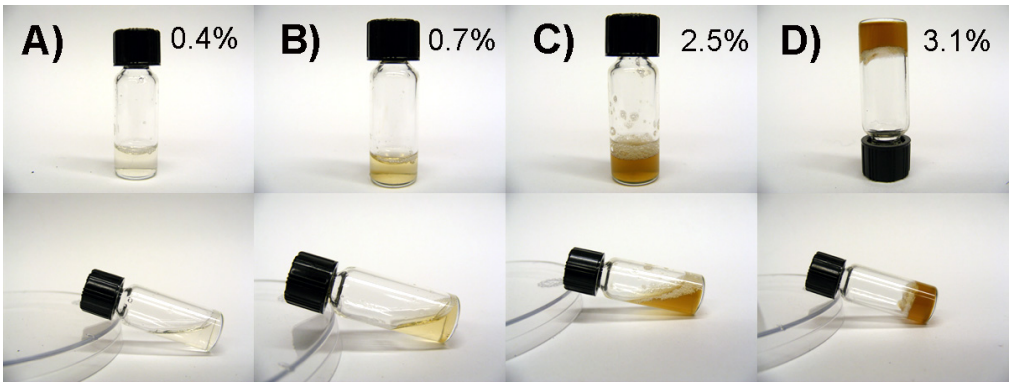


Fig. S2 Solutions of A(G)8A at different concentrations.

References

1. Y. Cao, C. Lam, M. Wang and H. Li, *Angew Chem Int Ed Engl*, 2006, **45**, 642-645.
2. Y. Cao and H. Li, *Nat Mater*, 2007, **6**, 109-114.
3. W. Shen, K. C. Zhang, J. A. Kornfield and D. A. Tirrell, *Nature Materials*, 2006, **5**, 153-158.