

Supplementary information

Supplementary Table 1. Characterization of pH-responsive ELP gene products.

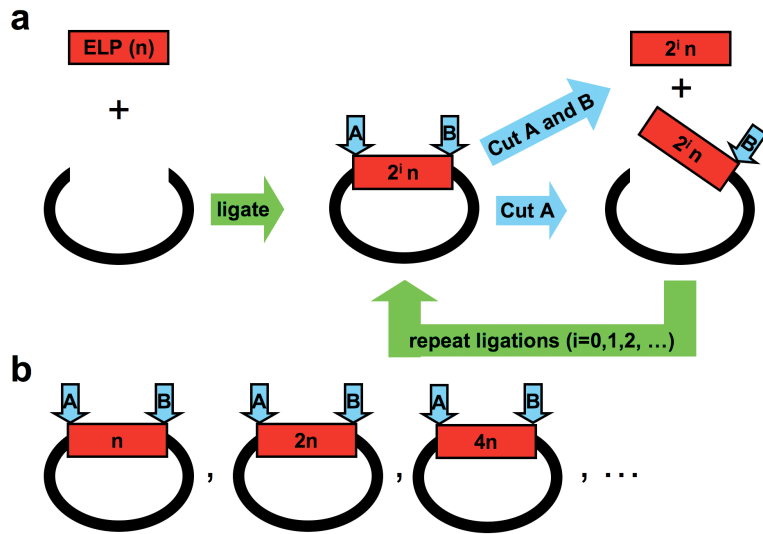
| ELP guest residues | Length, <i>L</i> (pentamers) | Amino acid sequence | ^a Expected MW (kD) | ^b Observed MW (kD) |
|--------------------|------------------------------|--|-------------------------------|-------------------------------|
| V:I:E | 40 | MSKGPG(VGVPGIGVPGIGVPGEGVPGIG) ₈ WPC | 17.9 | 17.6 |
| [1:3:1] | 80 | MSKGPG(VGVPGIGVPGIGVPGEGVPGIG) ₁₆ WPC | 34.9 | 34.6 |
| | 160 | MSKGPG(VGVPGIGVPGIGVPGEGVPGIG) ₃₂ WPC | 68.8 | 68.3 |
| V:H:G:A | 40 | MSKGPG(VGVPGHGVPGGGVPGHGVPGAG) ₈ WP | 16.4 | 17.2 |
| [1:2:1:1] | 60 | MSKGPG(VGVPGHGVPGGGVPGHGVPGAG) ₁₂ WP | 25.5 | 25.3 |
| | 100 | MSKGPG(VGVPGHGVPGGGVPGHGVPGAG) ₂₀ WP | 41.9 | 41.7 |
| | 120 | MSKGPG(VGVPGHGVPGGGVPGHGVPGAG) ₂₄ WP | 50.1 | 49.9 |

^aEstimated polypeptide masses for the gene products including the N-terminal methionine.

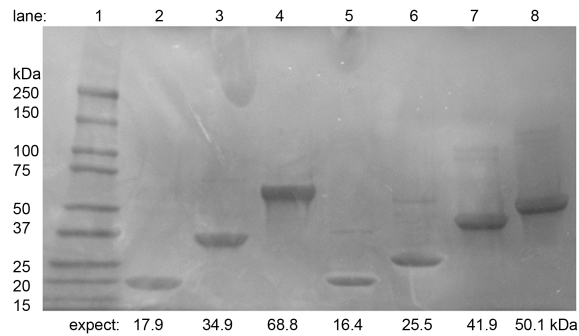
^bMasses determined using matrix assisted laser desorption ion mass spectrometry (MALDI).

Supplementary Table 2. List of variables and parameters in the manuscript.

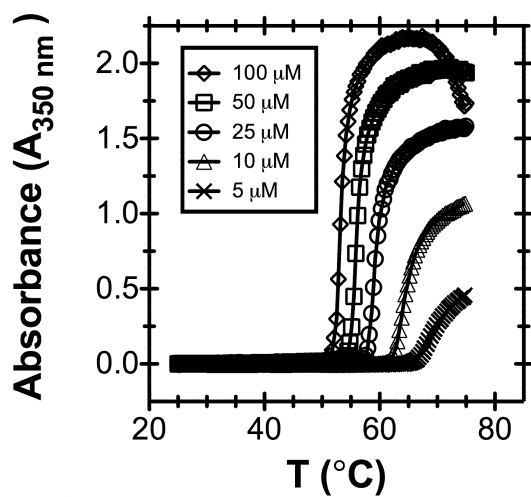
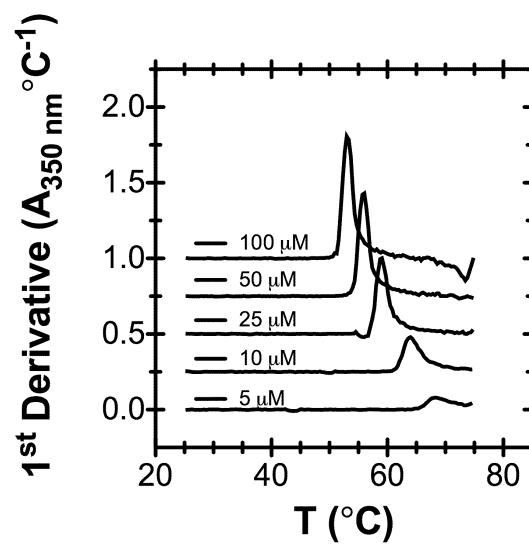
| Variables and parameters | Definition |
|---|--|
| b | Concentration dependence of transition temperature for ELP |
| b_{depro} | Concentration dependence of transition temperature for deprotonated ELP |
| b_{pro} | Concentration dependence of transition temperature for protonated ELP |
| C | ELP concentration |
| C_c | Critical ELP concentration |
| $C_{c,depro}$ | Critical deprotonated ELP concentration |
| $C_{c,pro}$ | Critical protonated ELP concentration |
| C_{depro} | Concentration of deprotonated guest residues |
| C_{pro} | Concentration of protonated guest residues |
| C_{total} | Total concentration of guest residues |
| f_{depro} | Fraction of guest residues that are deprotonated |
| k | Length-concentration dependence of ELP transition temperature |
| k_{depro} | Length-concentration dependence of deprotonated ELP transition temperature |
| k_{pro} | Length-concentration dependence of protonated ELP transition temperature |
| L | ELP length in pentamers |
| pH | pH |
| pH_t | ELP transition pH |
| pKa | pKa |
| T_c | Critical ELP transition temperature |
| $T_{c,depro}$ | Critical deprotonated ELP transition temperature |
| $T_{c,pro}$ | Critical protonated ELP transition temperature |
| T_{depro} | Transition temperature for deprotonated ELP |
| T_{pro} | Transition temperature for protonated ELP |
| T^{ref} | Transition temperature for ELP at reference concentration |
| T^{ref}_{depro} | Transition temperature for deprotonated ELP at reference concentration |
| T^{ref}_{pro} | Transition temperature for protonated ELP at reference concentration |
| T_t | ELP transition temperature |



Supplementary Figure 1. Steps of recursive directional ligation (RDL). **(a)** Starting with a chemically synthesized pair of oligonucleotides of length n , an ELP gene is inserted into a plasmid with restriction cut sites A (PflmI) and B (BglII). The synthetic gene, which can be ligated into other plasmids, is obtained by double digestion at sites A and B. Linear plasmid is obtained by single digestion at site A. The single and double digestion products are ligated together, doubling the size of the ELP gene. These products are then used as substrates to form successively longer gene products of length $2^i n$, where i represents the number of rounds of recursive ligation. **(b)** RDL is repeated to obtain the desired ELP lengths, which are multiples of the original length, n . Synthetic genes can be isolated by double digestion at sites A and B and then ligated into expression vectors with complementary restriction sites.



Supplementary Figure 2. SDS-PAGE demonstrating the identity and purity of pH-responsive ELPs. After confirming the identity of ELPs using DNA digestion and sequencing, the ELPs were purified from bacterial lysates. ELPs were characterized using SDS-PAGE by diluting 4 μ g of protein in 15 μ L of Laemmli loading buffer with betamercaptoethanol and heating at 95 $^{\circ}$ C for 5 min. The samples were run on a 4-20% Tris-HCl polyacrylamide gel and stained using 0.5 M copper chloride for 5 min. Lane 1: molecular weight ladder 250, 150, 100, 75, 50, 37, 25, 20, and 15 kDa. Lanes 2, 3, 4: ELP with $X = V:I:E$ [1:3:1] and $L = 40, 80, 160$, respectively. Lanes 5, 6, 7, 8: ELP with $X = V:H:G:A$ [1:2:1:1] and $L = 40, 60, 100, 120$, respectively.

a**b**

Supplementary Figure 3. Determination of the ELP inverse phase transition temperature using optical density at 350 nm under a temperature gradient of $1^{\circ}\text{C min}^{-1}$. **(a)** Optical density at 350 nm for an acidic ELP with V:I:E 1:3:1 with 80 pentamers at pH 6.5 with concentrations of 5, 10, 25, 50, and 100 μM . **(b)** First derivative of raw data with transition temperature defined at the maxima.