Electronic Supplementary Material (ESI) for Nanoscale. This journal is © The Royal Society of Chemistry 2017

Supplemental Material for:

Self-Assembled Hybrid Elastin-like Polypeptide / Silica Nanoparticles Enable Triggered Drug Release

Wei Han, ^{1,2} Ashutosh Chilkoti^{1,2,3} and Gabriel P. López^{1,2,3,4*}

- 1. Research Triangle Materials Science and Engineering Center, Durham, North Carolina 27708
- 2. Department of Biomedical Engineering, Duke University, Durham, North Carolina 27708
- 3. Department of Mechanical Engineering & Materials Science, Duke University, Durham, North Carolina 27708
- 4. Department of Chemical & Biological Engineering, The University of New Mexico, Albuquerque, New Mexico 87131

*Correspondence: gplopez@unm.edu

Experimental

Expression and purification of sp-ELP and con-ELP

Expression of sp-ELP and con-ELP was carried out in *E. coli* containing the assembled plasmids as described previously. ⁴⁰ Fresh terrific broth (TB) culture media was prepared at 55 g/L, autoclaved and supplemented with 45 mg/mL kanamycin. A hyperexpression protocol was used for ELP expression that relies on the inherent leakiness of the T7 promoter. ⁶² A single colony of *E. coli* harboring an expression plasmid that encodes for an ELP of interest was used to inoculate a 50 mL starter culture in TB. After one day of culture at 37 °C at 200 rpm, cells were centrifuged the following day and resuspended in 10 mL of fresh TB and transferred into 1 liter TB and cultured at 37 °C and 200 rpm rotation for 24 h. Cells were then centrifuged, resuspended in phosphate buffered saline (PBS; pH 7.4) and lysed by sonication. Purification of polypeptide was carried out by inverse transition cycling (ITC), as previously described. ⁴⁵ Briefly, one cycle of ITC consists of addition of NaCl and increase of temperature until solution becomes turbid, indicating that the ELP has transitioned, followed by centrifugation at 25 °C and 15,000 x g for

10 minutes to pellet the ELP, and resuspension of the pellet in fresh PBS or water supplemented by 20 mM TCEP, pH 7. This step is followed by centrifugation at 4 °C and 15,000 x g for 10 minutes to remove irreversibly aggregated material, the supernatant is transferred to a new tube. Three cycles of ITC were sufficient to produce pure polypeptides, as characterized by SDS-PAGE. The resulting yield of the sp-ELP construct is ~125 mg ELP per L culture and that of con-ELP is ~80 mg ELP per L culture.

Cell culture method

Subculturing of HT-29 cells followed the instructions provided by the ATCC website and in T-75 flasks. Briefly, used culture medium was removed and cells were rinsed using DPBS. 3 mL of 0.25% (w/v) Trypsin/EDTA solution was added to the flask and allowed to incubate for 15 minutes at 37° C. Subsequently, 6 mL 10% FBS supplemented McCoy's 5A Medium was added to the flask and the entire solution was transferred to a 50 mL conical tube, centrifuged at 120 x g for 10 minutes to pellet the cells, and the supernatant was removed. Finally, 5 mL fresh 10% FBS supplemented McCoy's 5A Medium was added to the tube, pipetted up and down to resuspend cells, and 1 mL of cells were transferred to a new T-75 flask containing 11 mL of the aforementioned growth medium (*i.e.*, subcultivation ratio of 1:5).

```
Met Ser Ser Lys Lys Ser Gly Ser Tyr Ser Gly Ser Lys Gly Ser Lys Arg Arg Gly Gly

T ATG AGC TCT AAA AAG AGT GGG TCC TAT TCG GGC AGC AAA GGT AGT AAG CGT CGC GGC GG AC TCG AGA TTT TTC TCA CCC AGG ATA AGC CCG TCG TTT CCA TCA TTC GCA GCG CCG

Ndel

B

Met Ser Gly Pro Gly Gly

T ATG AGC GGC CCA GGC GG
AC GCG CCG GGT CCG

Ndel

BseRI
```

Figure S1. Oligonucleotide inserts for creation of sp-ELP (A) and control ELP constructs (B). For each A and B, top: amino acid sequence; bottom: sense (5' -> 3') and antisense (3' -> 5') oligonucleotide sequence. Restriction enzyme recognition sites and base pair overhangs indicated with blue lines.

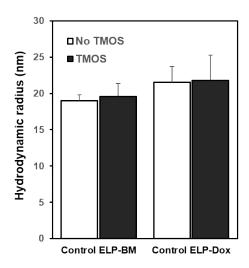


Figure S2. Hydrodynamic radii obtained from control silicification experiments. Control ELP-BM and Control ELP-Dox without TMOS (white) and post silicification (black) with 100 mM TMOS for 4 hours.