Supporting Information

Materials and Methods.

Fetal calf serum(FCS) was purchased from Sigma Aldrich. FCS was sterile filtered through a 0.22 μm PES syringe filter prior to use.

Critical aggregation concentration (CAC) of LHRH-PEGMA CPT block copolymer:

Critical aggregation of the polymer was determined using Nile red as a fluorescent probe by adopting a previous protocol[1].

Various dilutions of the LHRH-PEGMA CPT block copolymer were prepared by dissolving and diluting them isotonic PBS buffer at pH 7.4, range 0.5-256μg/mL. 0.5 mL of solution for each dilution was prepared. A stock solution of Nile red was prepared in acetone at concentration $1 * 10^{-4}$ mol/L. 5 μL of the Nile red stock was added to each vial and the acetone allowed to evaporate overnight at room temperature. The samples were excited at 485 nm using a fluorescence spectrophotometer and the emission recorded at 525 nm. Critical aggregation concentration was obtained as the intersection of the two linear tangent in the graph.

Serum stability assay:

The stability of the polymer prodrug was examined in the presence of fetal calf serum (FCS). The polymer prodrug was dissolved in HPLC grade water at a concentration of 0.5 mg/mL. Aliquots of this polymer solution (150 μL) were diluted either with 1050 μL of FCS or PBS buffer and incubated at 37°C. Samples were taken at 0,1,2,4,6 and 24 hour time points. At each time point 100 μL of sample were taken and mixed with 900 μL of isopropanol. The resulting solution was placed in a freezer for 30 minutes and then subsequentlycentrifuged at 16kG for 10 minutes. A sample (500 μL) of the resulting supernatant was diluted with 500 μL of HPLC grade water. The solutions were subsequently analysed to determine free CPT content. Each experiment was carried out in triplicate. Free CPT content was determined by fluorescence (370 nm excitation/434 nm emission) detection following size exclusion chromatography (Column: Aquagel 30, Eluent: PBS/10% MeOH) on a Shimadzu lc-20ad/sil-20a HPLC system equipped with a RF-10AXL fluorescence detector. A control reading of 100% drug release was estimated by treating the polymer reference sample with TCEP and determining the amount of free CPT (+CPT-SH) present in the sample.

TEM analysis

TEM spectroscopy was carried out on a JEOL Ltd JEM-2100F microscope. 2 μL of sample (0.5 mg/mL polymer in HPLC water) was placed on a copper grid (Holey Carbon Film 300 Mesh Cu) for few minutes. The excess of sample was wicked away with the aid of filter paper. The sample was allowed to dry and then subsequently imaged in transmission electron mode without staining.

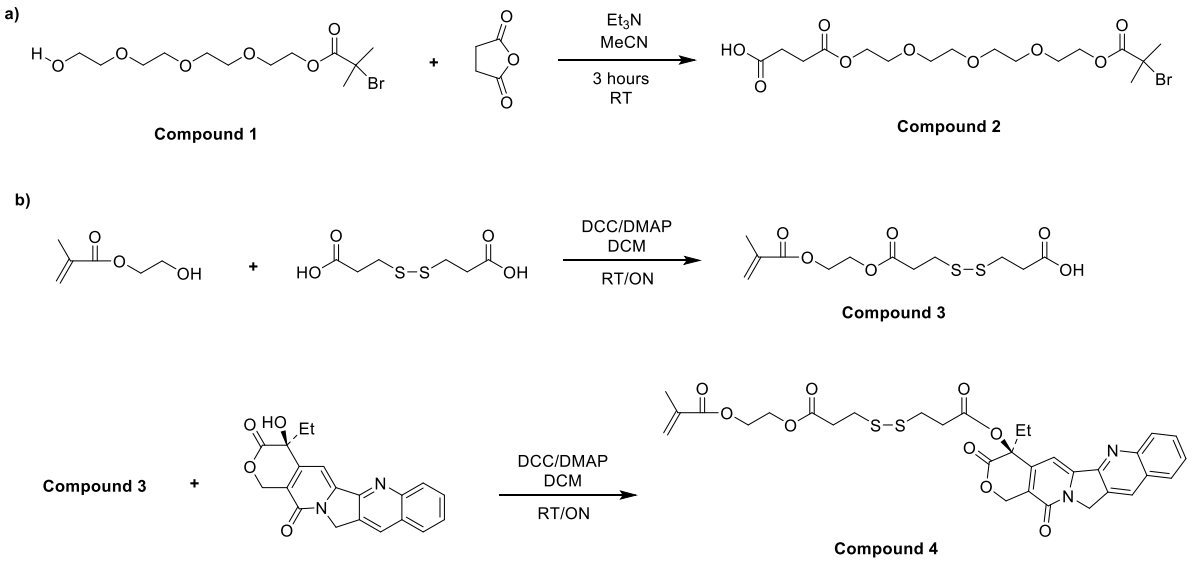

Figure S1. a) Synthesis of acid functional ATRP initiator (Compound 2). b) Synthesis of reducible Camptothecin monomer (Compound 4).

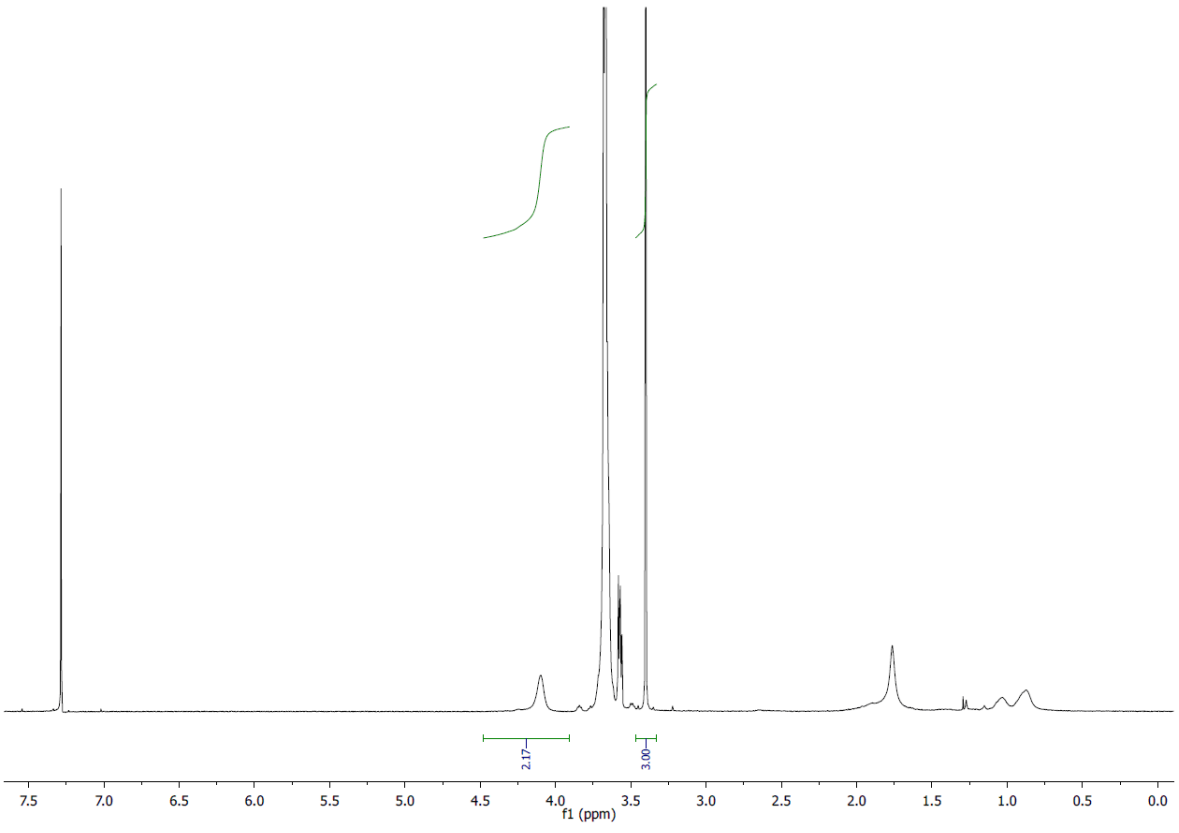

Figure S2. PEGMA block polymer H-NMR spectra. Mw calculation by NMR. (2n+4)/n = 2.17 => n = 24 units of PEGMA. Mw by NMR = 24*475 + 442 =12000.

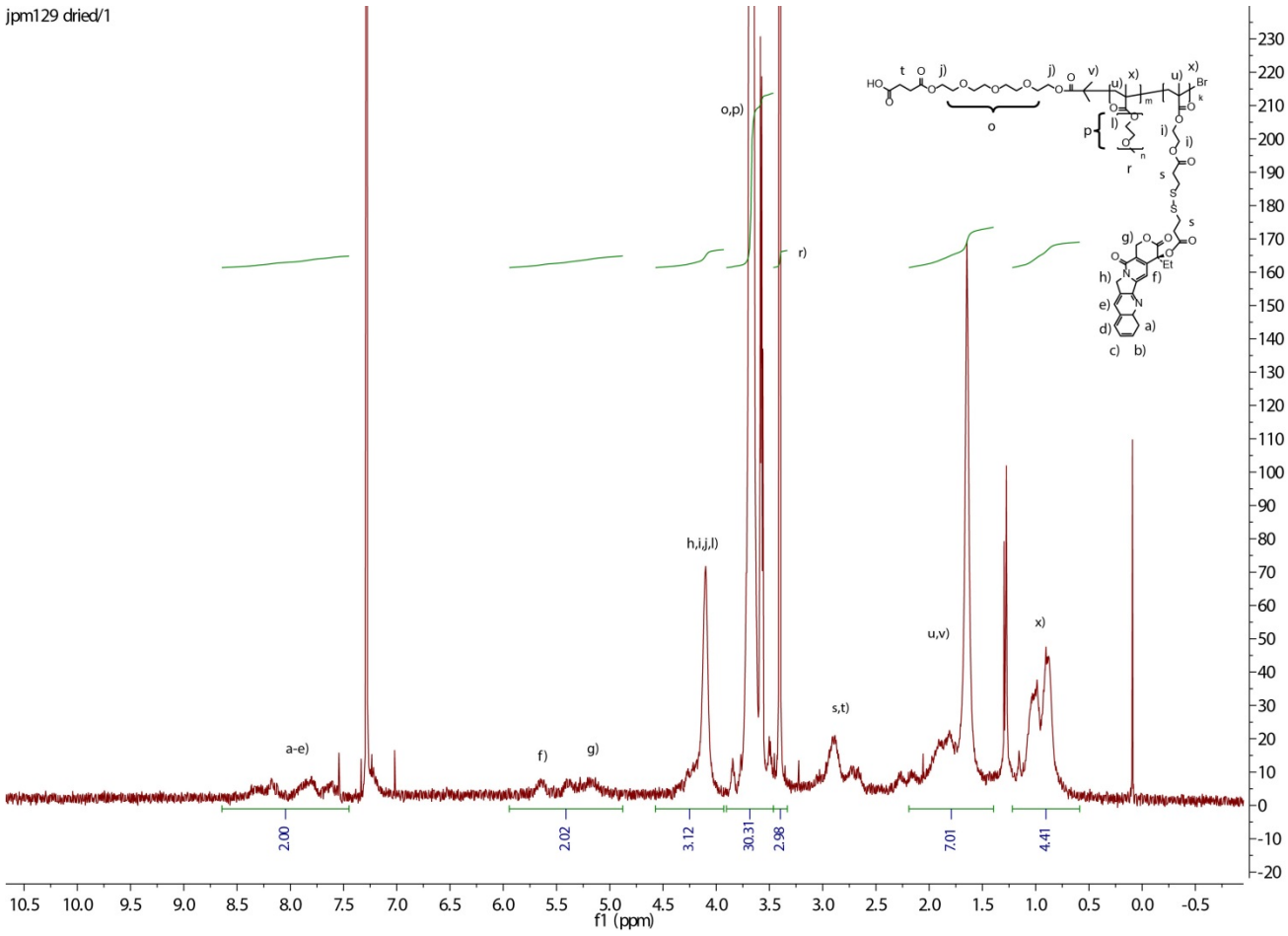

Figure S3. PEGMA-CPT block copolymer H-NMR spectra.

Composition by NMR: For 1 PEGMA units there are 0.4 units of CPT present. (2 aromatic protons(out of 5) per 1 unit of PEGMA (3 methoxy protons). 24 units of PEGMA in polymer => 10 units of CPT in polymer. Total Mw (by NMR) = 12000 + 10*653 =18500.

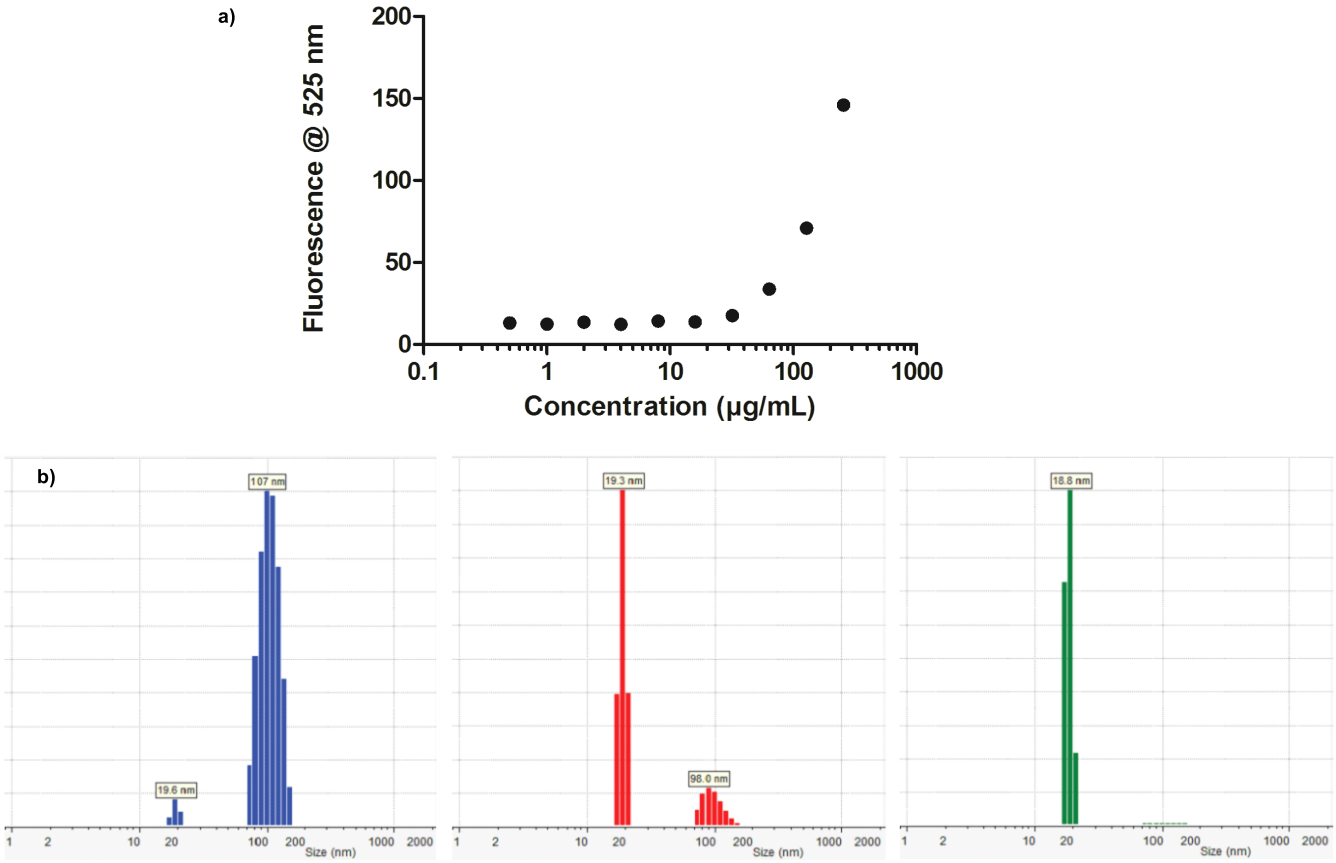

Figure S4 a) Critical aggregation concentration (CAC) of the LHRH-PEGMA CPT block copolymer. Critical aggregation concentration was obtained as the intersection of the two linear tangent in the graph. CAC was calculated to be 29.8 µg/mL. b) DLS of LHRH-PEGMA CPT block copolymer in PBS buffer pH 7.4. (Blue - intensity model. Red - mass model. Green - number model).

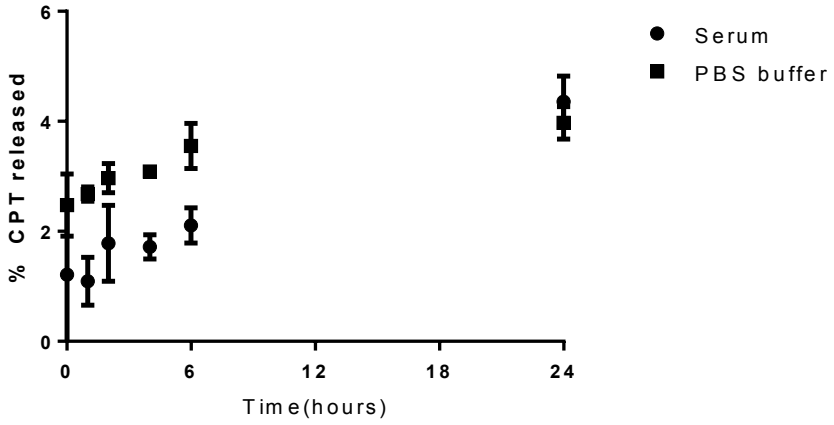

Figure S5 – Stability of polymer prodrug in PBS and serum.

The polymer prodrug displayed comparable stability in buffer and in serum, less than 5% of the free drug (+CPT-SH) was detected after 24 hour incubation. Although disulfide scrambling of the polymer was apparent in chromatograms the maximum amount of free drug (+CPT-SH) at any time did not exceed 5%.

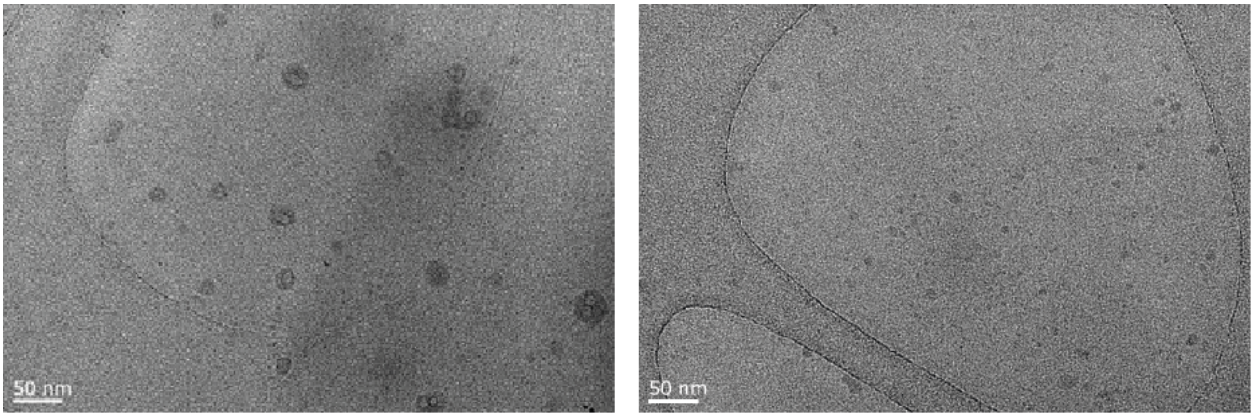

Figure S6- Transmission Electron Micrographs of Polymer 1 (CPT-containing block co-polymer) following dehydration.

Circular features in the image exhibit diameters in the range of 10-30 nm, in good agreement with number average DLS diameters of block co-polymers in aqueous solution of~ 40 nm.

References

1. Y. Shen, E. Jin, B. Zhang, C. J. Murphy, M. Sui, J. Zhao, J. Wang, J. Tang, M. Fan, E. Van Kirk, and W. J. Murdoch, *J. Am. Chem. Soc.*, 2010, **132**, 4259–4265.