

## Supporting Information

### Responsive hybrid block co-polymer conjugates of proteins – controlled architecture to modulate substrate specificity and solution behaviour.

Gökçen Yaşayan,<sup>a</sup> Aram O. Saeed,<sup>a</sup> Francisco Fernández-Trillo,<sup>a</sup> Stephanie Allen,<sup>a</sup> Martyn C Davies,<sup>a</sup> Abdulhakim Jangher,<sup>c</sup> Alison Paul,<sup>c</sup> Kristofer J. Thurecht,<sup>b</sup> Stephen M. King,<sup>c</sup> Ralf Schweins,<sup>d</sup> Peter C Griffiths,<sup>c</sup> Johannes P. Magnusson,<sup>\*a</sup> and Cameron Alexander<sup>\*\*a</sup>

Figure S1: PGSE NMR showing temperature dependence of the hydrodynamic radii for trypsin (triangles), precursor (squares) and hybrid I (circles) (left panel) and hybrid II (right panel) in D<sub>2</sub>O.

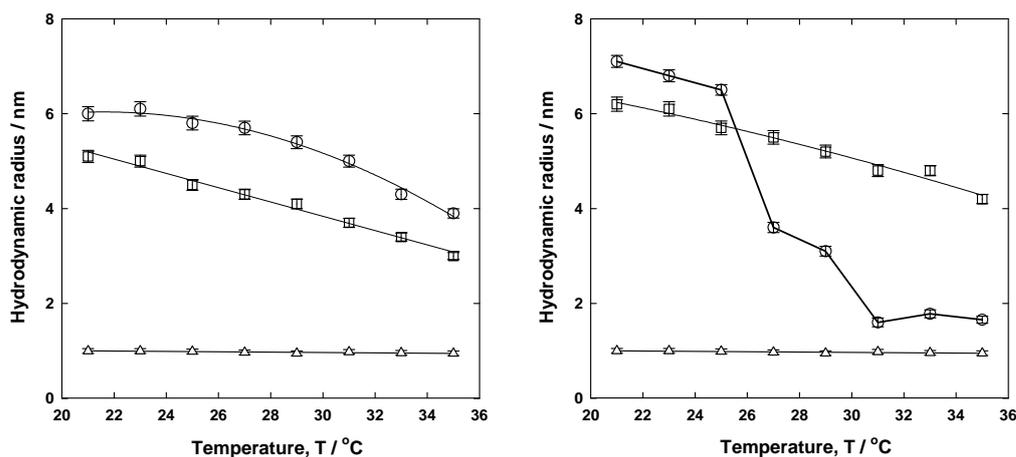


Figure S2: SANS graphs for (PEGMA-EE-246)<sub>85</sub>-stat-PEGMA-ME-475)<sub>15</sub> (Co-Polymer 1) (top row) and [(PEGMA-EE-246)<sub>85</sub>-stat-PEGMA-ME-475)<sub>15</sub>]<sub>92</sub>-graft-(PEGMA-ME-475)<sub>50</sub> (Co-Polymer 2) (bottom row). Left column D2O. Right column 0.3 M Na2SO4. Blue trace 20°C, Green trace 35°C and Red trace 50°C.

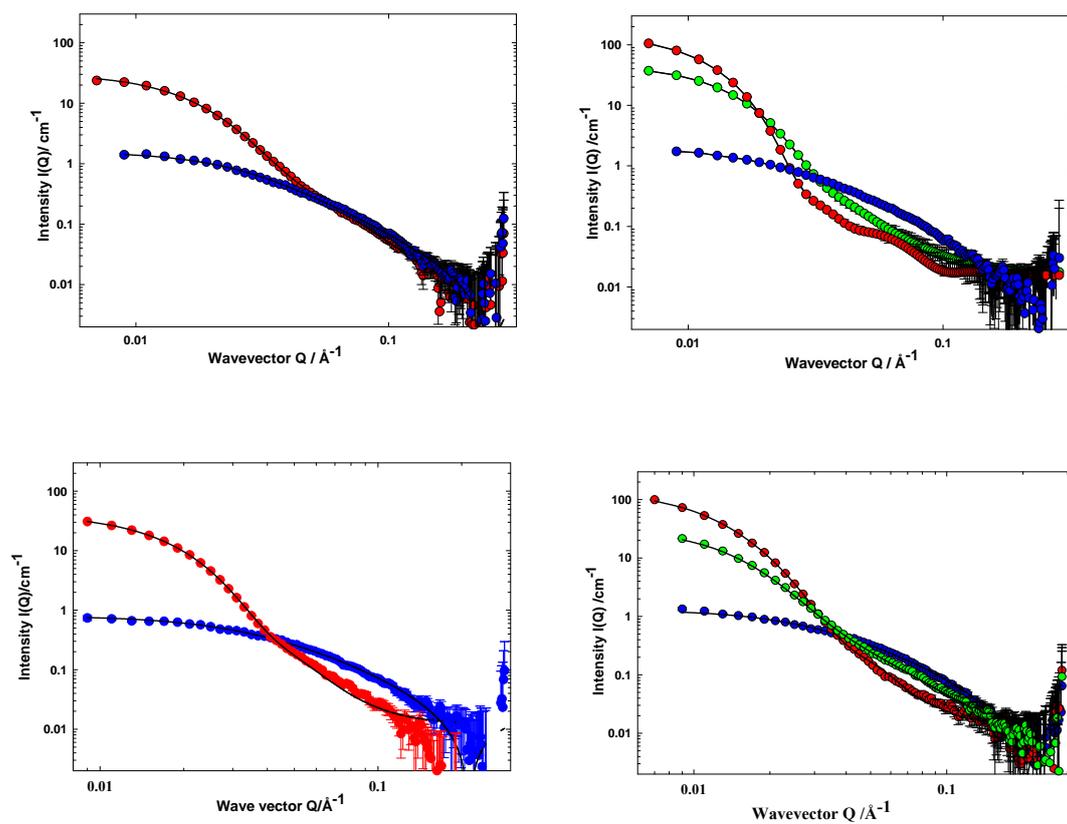


Figure S3: SANS graphs for Hybrid I (top row) and Hybrid II (bottom row) in D2O (left column) and PBS (right column) at stated temperatures.

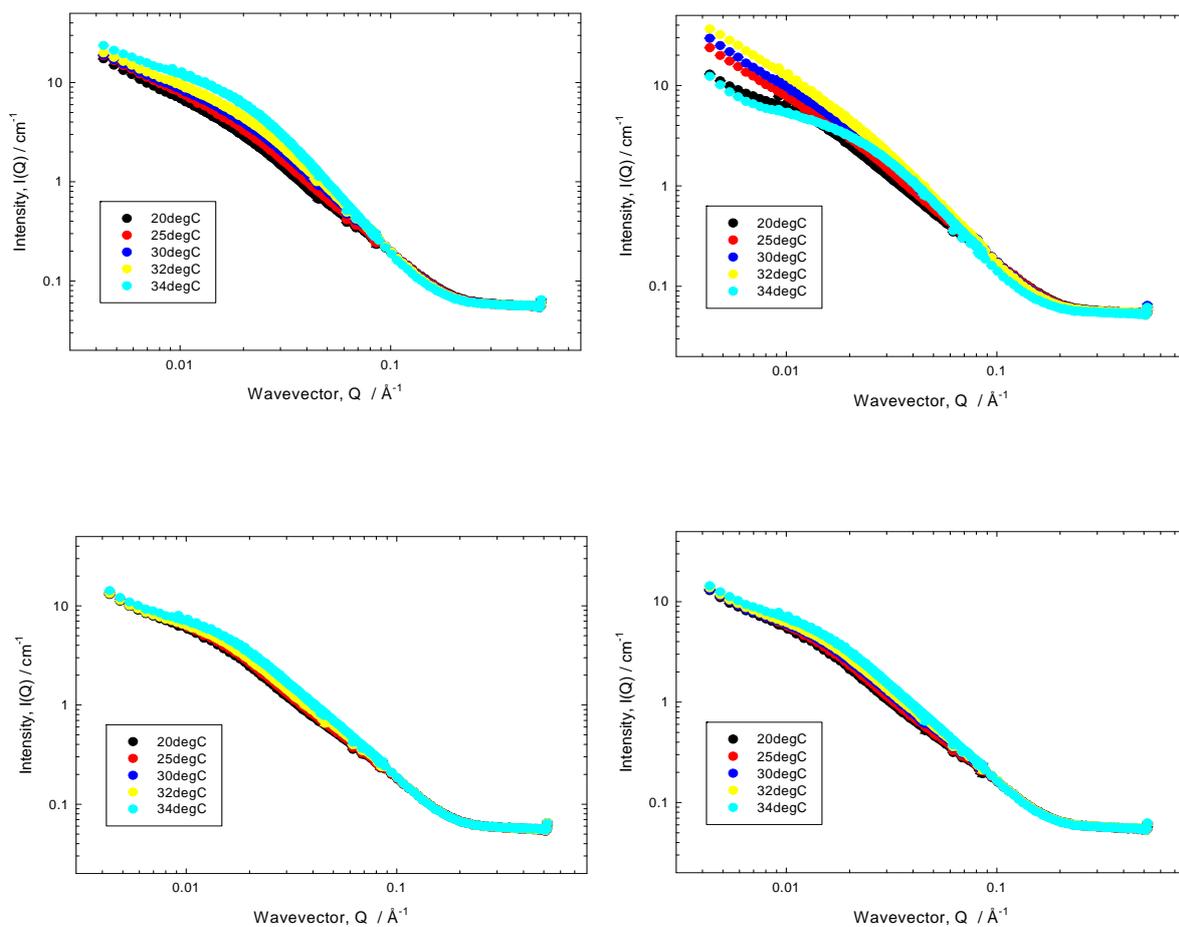
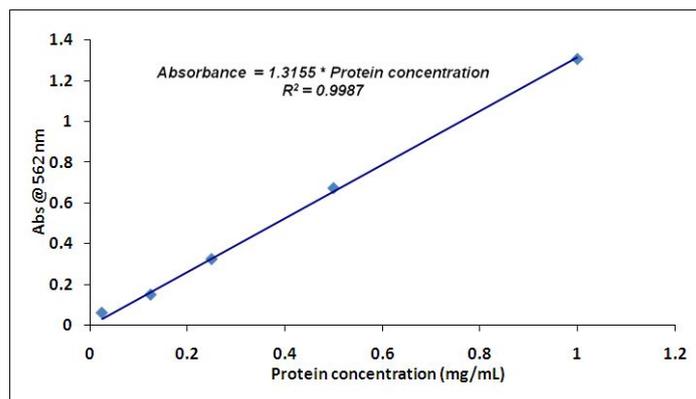


Figure S4 – BCA calibration curve made from bovine serum albumin standards.



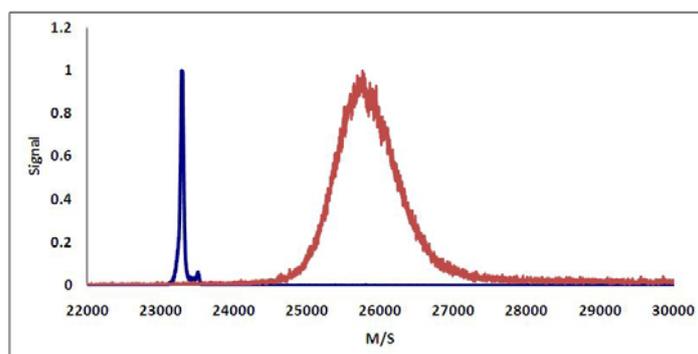
Absorbance of solution **A** : 0.5847, Protein concentration in sample = 0.444 mg/mL.

Percentage of protein in A = 14.82 %.

Absorbance of solution **B** : 0.5847, Protein concentration in sample = 0.274 mg/mL.

Percentage of protein in B = 9.14 %.

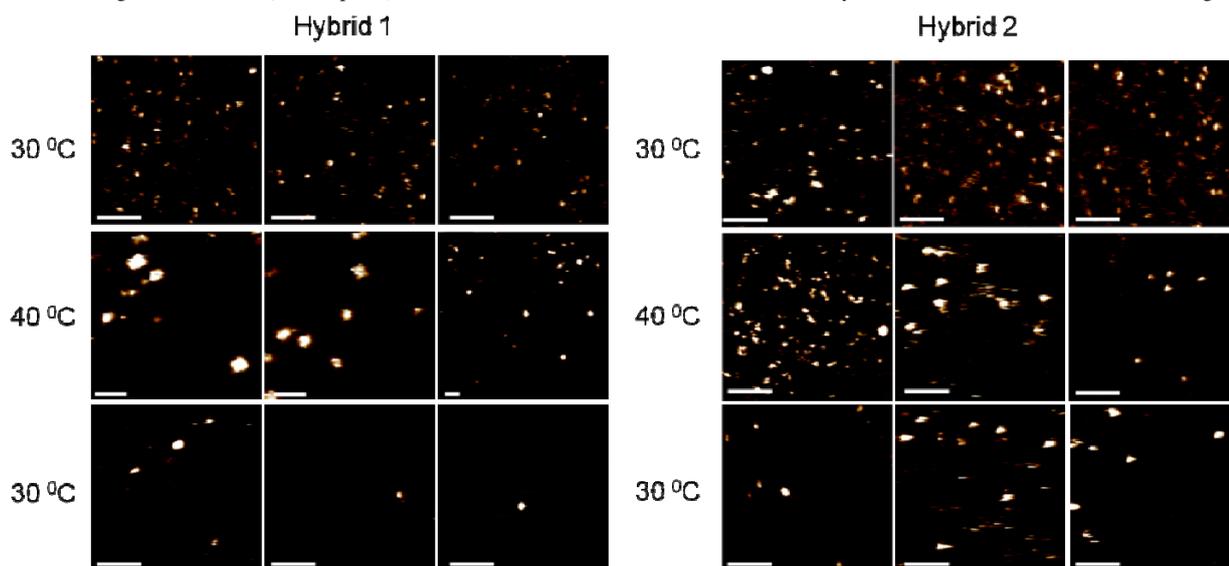
Figure S5 – MALDI TOF (Matrix-assisted laser desorption/ionization – time of flight)



Native trypsin (Blue trace). ATRP initiator functionalised trypsin (Red trace).

$M_p$  molecular weight difference = 2460 Da. 5.12 initiators per trypsin molecule on average (Range 3-8).

Figure S6 – Selected AFM topography images of Hybrid I and Hybrid II. The system was heated to 30°C (top panel), then to 40°C (middle panel) before being cooled to 30°C (bottom panel). Scale bars are 100 nm. Vertical scale is 8 nm for Hybrid I at 40°C and 2 nm for the other images.



#### Dynamic light scattering (DLS)

Figure S7 - DLS analysis of Hybrid I in PBS a) PBS buffer at 25°C (below LCST) – Intensity model. b) PBS buffer at 25°C (below LCST) – number model. c) PBS buffer at 40°C (above LCST) – Intensity model. d) PBS buffer at 40°C (above LCST) – number model.

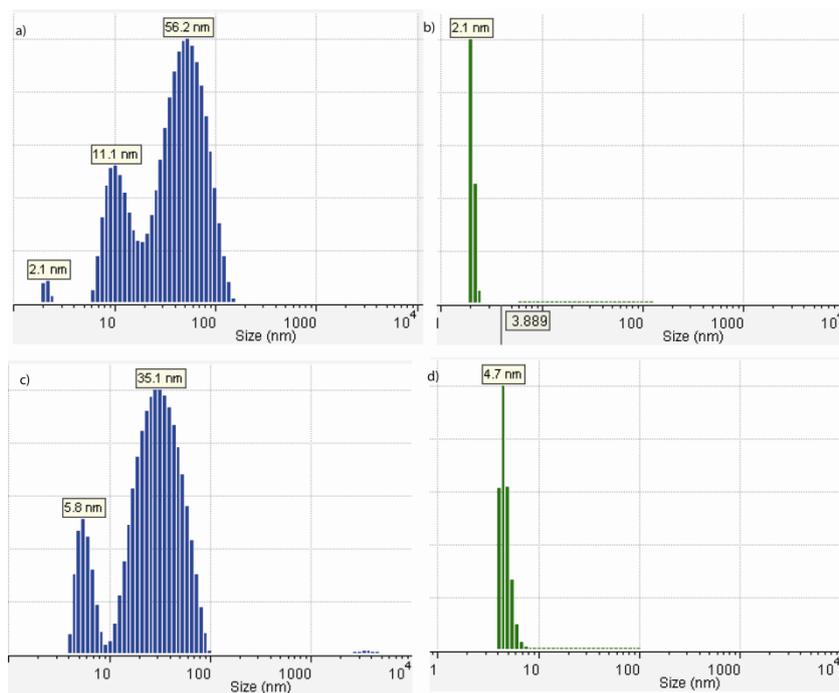


Figure S8 - DLS analysis of Hybrid I in Tris a) Tris buffer at 25°C (below LCST) – Intensity model. b) Tris buffer at 25°C (below LCST) – number model. c) Tris buffer at 40°C (above LCST) – Intensity model. d) Tris buffer at 40°C (above LCST) – number model.

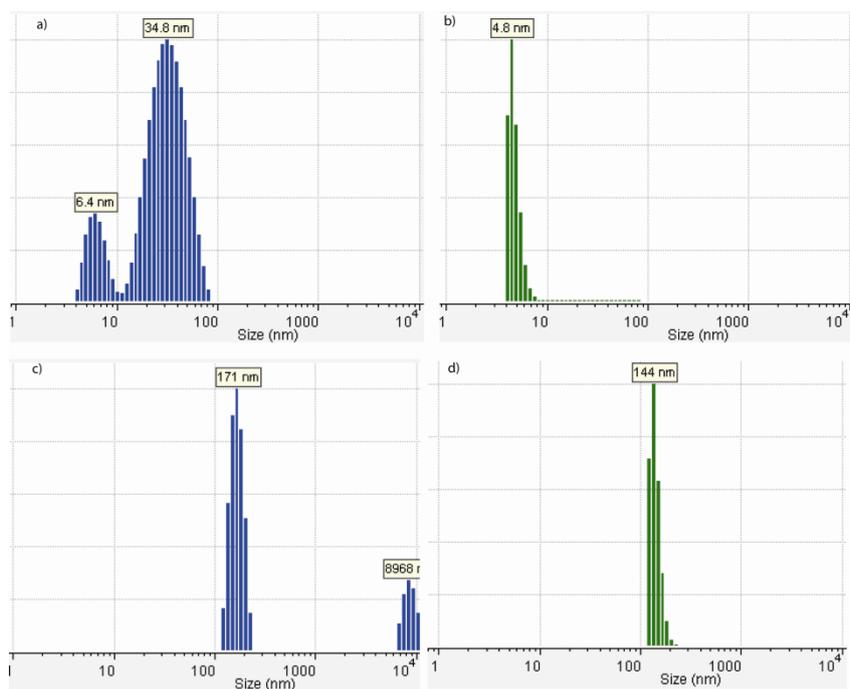


Figure S9 - DLS analysis of Hybrid II in PBS a) PBS buffer at 25°C (below LCST) – Intensity model. b) PBS buffer at 25°C (below LCST) – number model. c) PBS buffer at 40°C (above LCST) – Intensity model. d) PBS buffer at 40°C (above LCST) – number model.

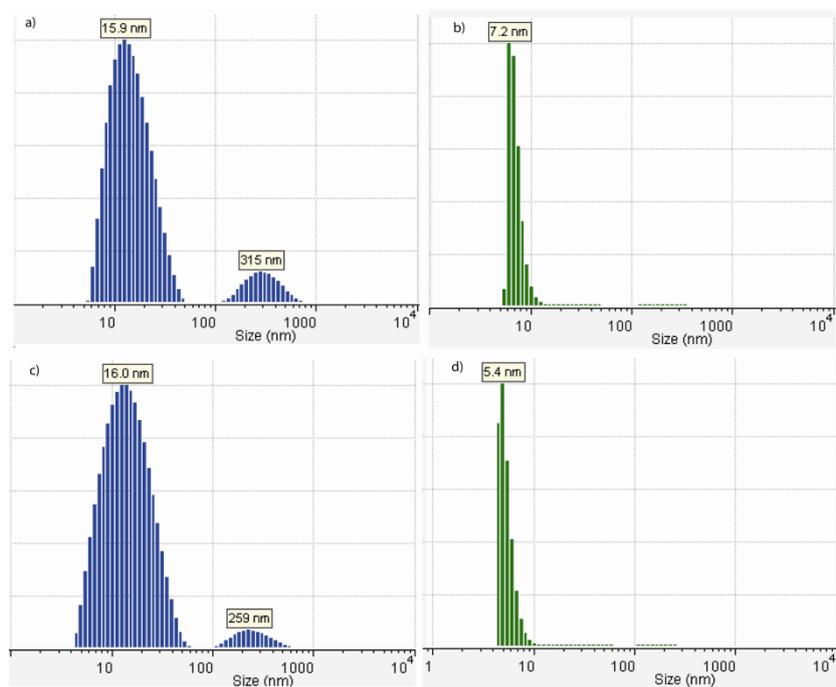


Figure S10- DLS analysis of Hybrid II in Tris a) Tris buffer at 25°C (below LCST) – Intensity model. b) Tris buffer at 25°C (below LCST) – number model. c) Tris buffer at 40°C (above LCST) – Intensity model. d) Tris buffer at 40°C (above LCST) – number model.

