Supporting Information

Unusual Salt Stability in Highly Charged Diblock Copolypeptide Hydrogels

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Materials and Methods

Samples All block copolypeptides were synthesized, purified and characterized as previously described.[1] Isolated yields of the final copolymers ranged between 75% and 90%. Amino acid compositions of the copolymers were found to be within 3% of predicted values. Chain lengths of the copolymers were found to be within 8% of predicted lengths with chain length distributions (weight average length/number average length) ranging between 1.1 and 1.3. Similar lysine-leucine block copolymers have been prepared in the past using different methods, however these materials contained both large chain length distributions and homopolymer contaminants.[2] We have found that block copolymers with chain length distributions greater than *ca*. 1.5 are unable to form hydrogels.[1]

Rheology Block copolypeptide solutions were prepared by dissolving freeze-dried samples in deionized water. Samples in ionic media were prepared either by direct dissolution of the sample in the media, or by dissolution of the sample at a higher than target concentration in deionized water, followed by dilution with a concentrated solution of the ionic media. The dissolution process of the samples was enhanced through vortex mixing. Solution properties were identical within experimental limits regardless of sample preparation method. The rheological properties were also not affected by the agitation; identical mechanical properties were obtained by letting the copolypeptides dissolve overnight.

Rheological measurements were performed on an ARES-LS1 controlled strain rheometer from Rheometrics Scientific (now TA Instruments). Additional studies of the dynamic moduli were also carried out on an MCR300 controlled stress rheometer from Paar-Physica. Both instruments were equipped with cone-plate configurations. Two different geometries were used on the ARES: a 50 mm diameter cone with a 2º angle and a 25 mm cone with 4º angle. The latter geometry is less stress-sensitive due to the smaller diameter, but has the benefit of a smaller sample volume. The choice between geometries was based on a qualitative judgment of the sample properties and the amount of material available: concentrated, gel-like samples could be measured with the 25 mm

cone, whereas dilute solutions required the sensitivity of the 50 mm cone. For a number of samples both geometries were employed to ensure reproducibility and excellent agreement was always observed. The MCR300 was operated with a 50 mm diameter 1º cone and a 25 mm 2º cone.

The rheological properties of all samples were determined through the following measurement protocol: oscillatory strain amplitude sweep (amplitude $\gamma_0 = 0.001 - 10$) at fixed frequency (6 rad/s) to establish the linear regime, oscillatory frequency sweep (frequency $\omega = 0.01 - 100$ rad/s) to measure linear viscoelasticity, recovery experiments and steady-shear flow curves. Between strain amplitude sweeps and frequency sweeps a waiting period of several minutes was applied to facilitate sample recovery from the non-linear oscillatory deformations.

References:

1) Nowak, A. P.; Breedveld, V.; Pakstis, L.; Ozbas, B.; Pine, D. J.; Pochan, D.; Deming, T. J. (2002) *Nature* **417**, 424-428.

2) Ostroy, S. E.; Lotan, N.; Ingwall, R. T.; Scheraga, H. A. (1970) *Biopolymers* **9**, 749-764.