Supporting Information

Charge density as a molecular modulator of nanostructuration in intrinsically disordered protein polymers

*Sergio Acosta, Leander Poocza, Luis Quintanilla-Sierra, and José Carlos Rodríguez-Cabello**

Bioforge lab, CIBER-BBN, University of Valladolid, Paseo Belén 19, 47011, Valladolid, Spain

*roca@bioforge.uva.es

Materials: Glassware was soaked in acid water (1% hydrochloric acid), washed with Alconox®, rinsed with distilled water and dried at 80 °C overnight prior to use. All chemicals were purchased from Sigma-Aldrich (Merck Group, Germany) and used as received unless otherwise mentioned. Ultrapure water (15 MΩ·cm) was used from a Milli-Q® Integral water purification system (Merck Group, Germany).

Supporting Tables:

Table S1. Comparison of the theoretical (Theo.) and experimental (Exp.) molecular weights (MW) calculated by MALDI-TOF.

	Theo. MW (Da)	Exp. MW (Da)
E-I	46972.8	46916.3±7.1
S-I	46070.2	45996.3±16.9
$E_{1/2}$-I	36585.8	36609.8±8.9
oE-I	30593.4	30580.5±3.6
o$E_{1/2}$-I	28396.0	28380.3±4.4

Table S2. Amino acidic composition of the ELdcRs calculated by HPLC. It is represented the theoretical (Theo.) composition comparing to the experimental (Exp.) values obtained.

Amino acid	E-l		S-l		$E_{1/2}$-l		oE-l		$oE_{1/2}$-l	
	Theo.	Exp.	Theo.	Exp.	Theo.	Exp.	Theo.	Exp.	Theo.	Exp.
Asp	-	-	-	-	-	-	-	-	-	-
Glu	11	12.02	1	1.17	6	6.66	11	14.51	6	7.18
Asn	-	-	-	-	-	-	-	-	-	-
Ser	1	1.52	51	50.35	1	1.32	1	1.05	1	1.17
Gln	-	-	-	-	-	-	-	-	-	-
His	-	-	-	-	-	-	-	-	-	-
Gly	220	224.45	220	222.91	170	170.56	140	142.99	130	129.48
Thr	-	-	-	-	-	-	-	-	-	-
Arg	-	-	-	-	-	-	-	-	-	-
Ala	-	-	-	-	-	-	-	-	-	-
Tyr	-	-	-	-	-	-	-	-	-	-
Cys	-	-	-	-	-	-	-	-	-	-
Val	151	139.68	111	105.83	106	97.22	71	59.87	66	61.91
Met	1	1.26	1	1.15	1	1.22	1	1.09	1	1.19
Trp	-	-	-	-	-	-	-	-	-	-
Phe	-	-	-	-	-	-	-	-	-	-
Ile	60	61.05	60	61.87	60	62.56	60		60	61.49
Leu	2	2.73	2	2.46	2	2.61	2	2.79	2	2.74
Lys	-	-	-	-	-	-	-	-	-	-
Pro	111	115.48	111	111.89	86	89.52	71	72.18	66	66.04
Total aas	557	558.2	557	557.63	432	431.7	357	356.8	332	331.21

Supporting Figures:

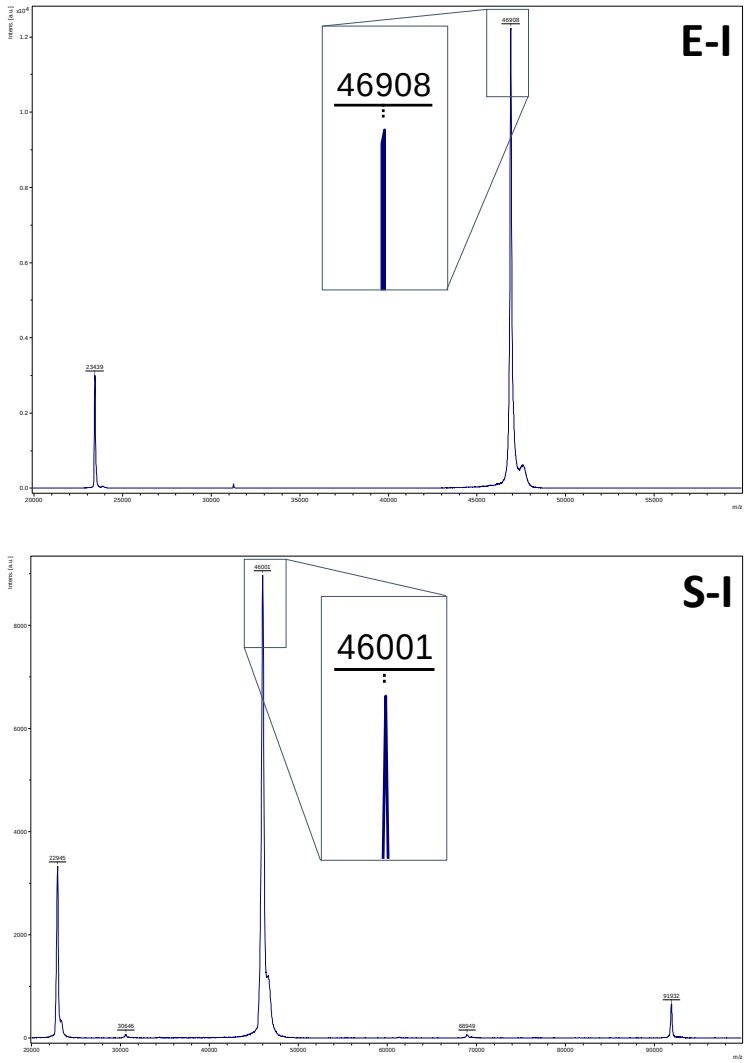

Figure S1. MALDI-TOF spectra of the ELdcRs E-I and S-I.

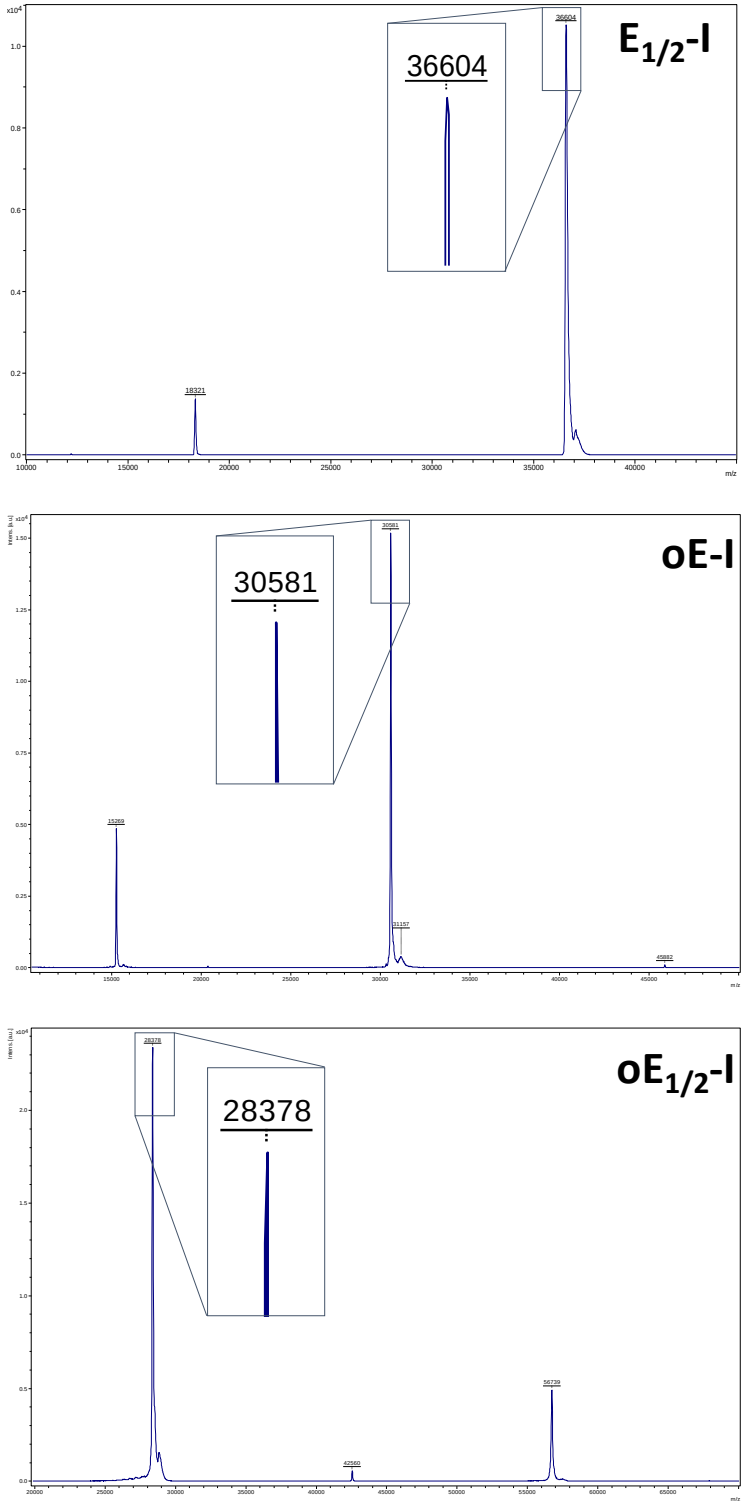

Figure S2. MALDI-TOF spectra of the anionic ELdcRs with unbalanced length ratio between the hydrophilic and hydrophobic blocks.

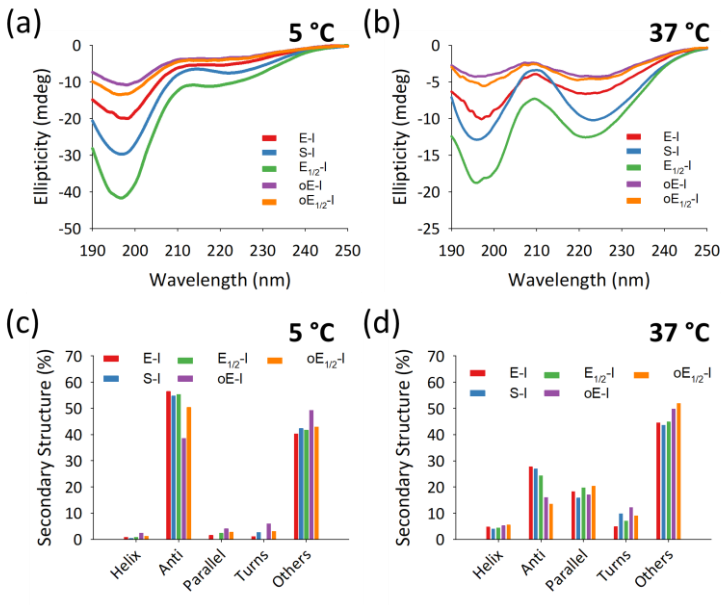

Figure S3. Secondary structure characterization of the ELdcRs above the LCST: CD spectra at (a) 5 °C and (b) 37 °C. Secondary structure quantification from CD spectra at (c) 5 °C and (d) 37 °C using the BeStSel algorithm for deconvolution of the CD data.

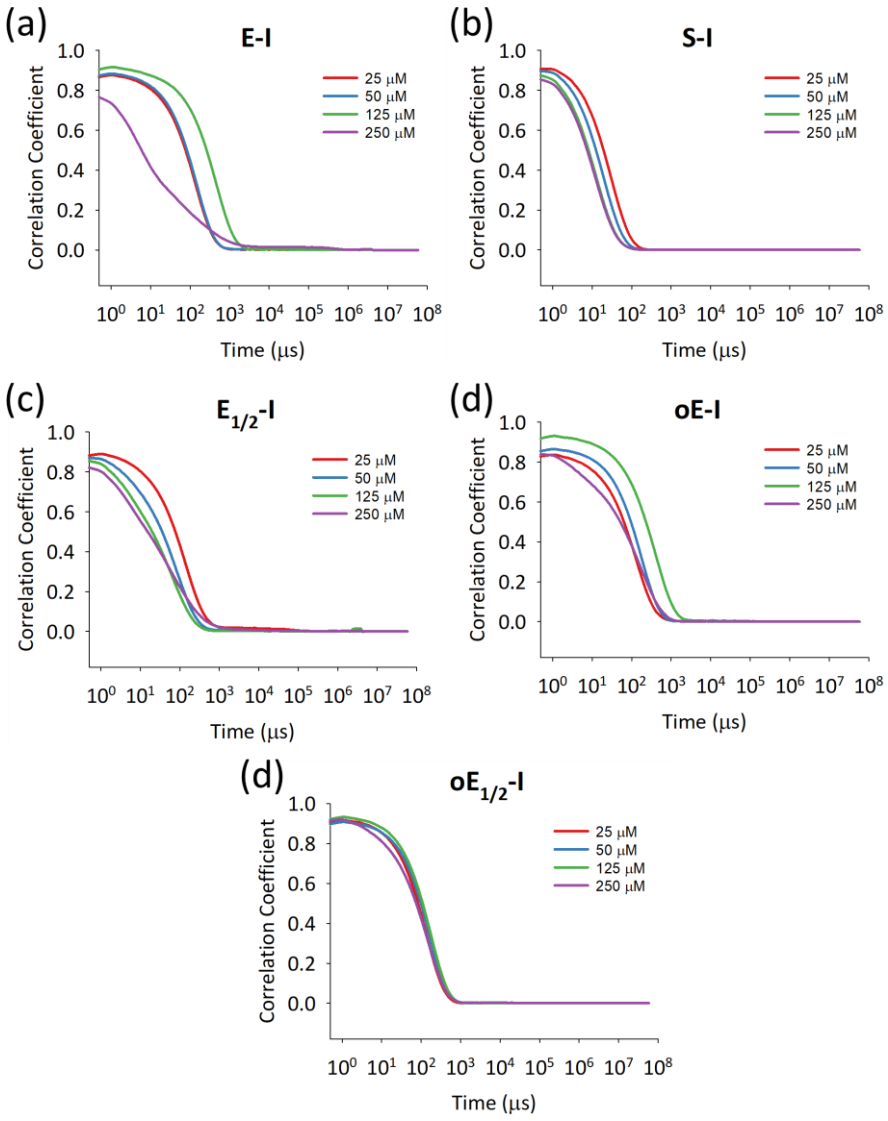

Figure S4. Raw correlation functions obtained by DLS for the diverse ELdcRs at different concentrations (25, 50, 125 and 250 µM) in ultrapure water.

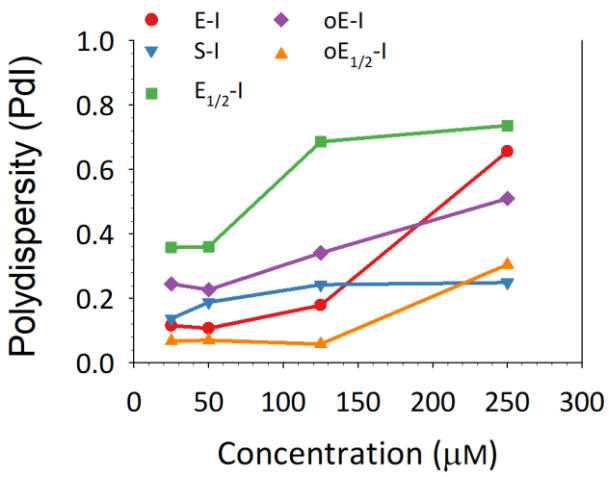

Figure S5. Additional DLS data. Effect of the concentration on the diameter of the main peak (Peak 1) of the intensity size distributions (a) and on the polydispersity of the sample (b).

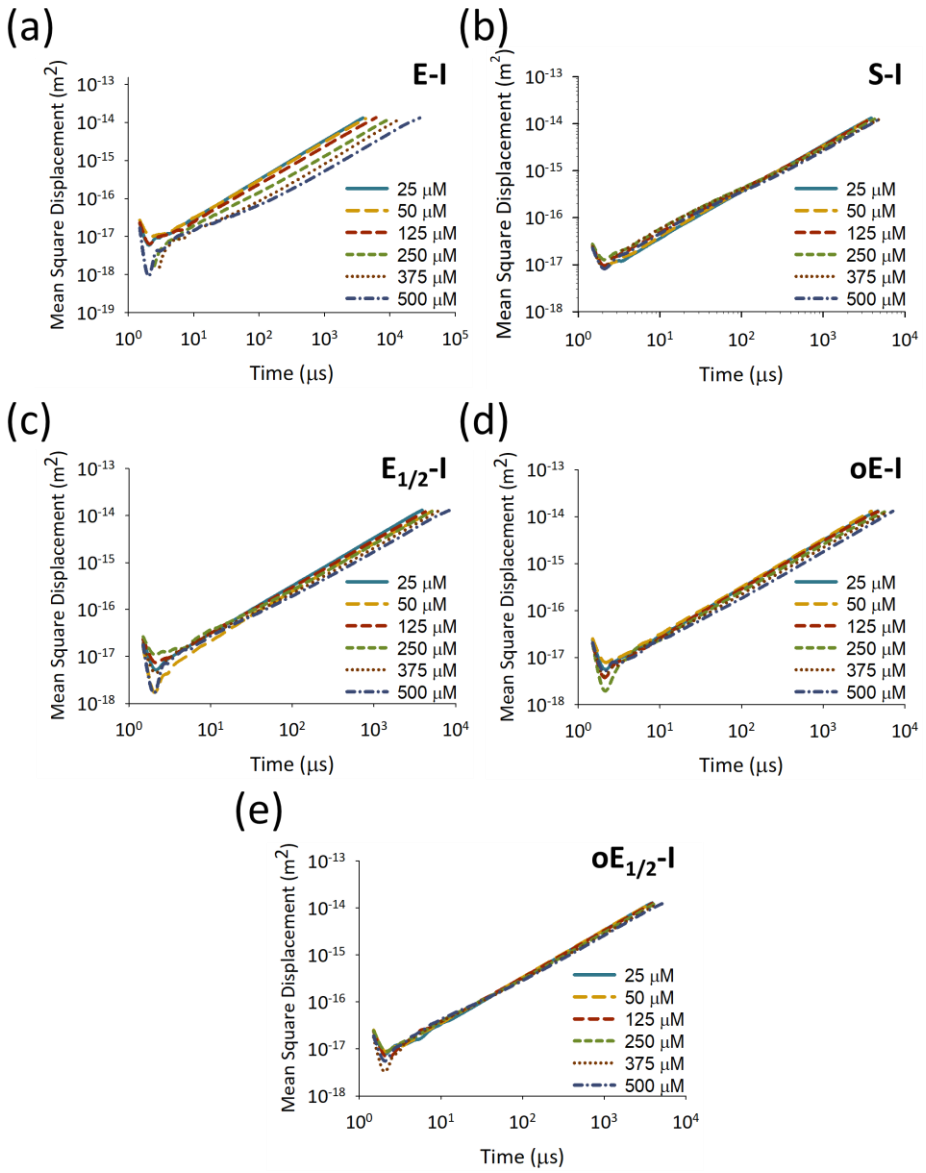

Figure S6. Mean square displacement (MSD) of the tracer particles in the ELdcR solutions: (a) E-I, (b) S-I, (c) $E_{1/2}$-I, (d) oE-I and (e) $oE_{1/2}$-I.

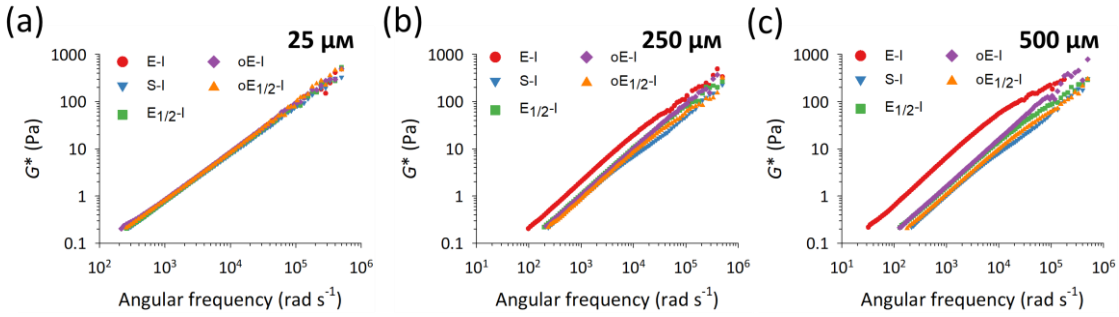

Figure S7. Shear complex modulus (G^*) as a function of the angular frequency of semidilute solutions of the ELdcRs at (a) 25 μM, (b) 250 μM and (c) 500 μM. The tendency to self-assemble into micellar strings and nanogels of the E-I, E½-I and oE-I was associated with an increase in G^*.

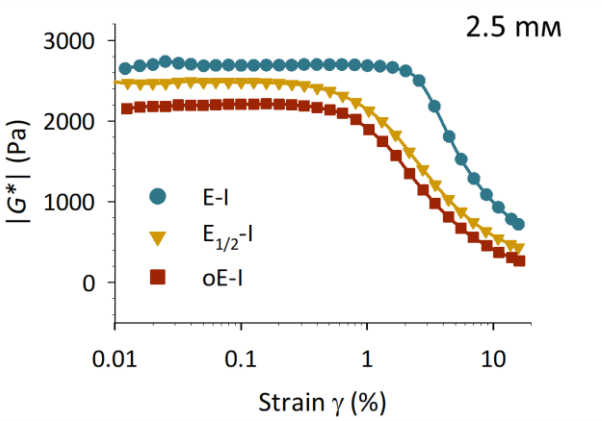

Figure S8. Strain sweep rheology of the physically crosslinked hydrogels at 37 °C, 1 Hz.

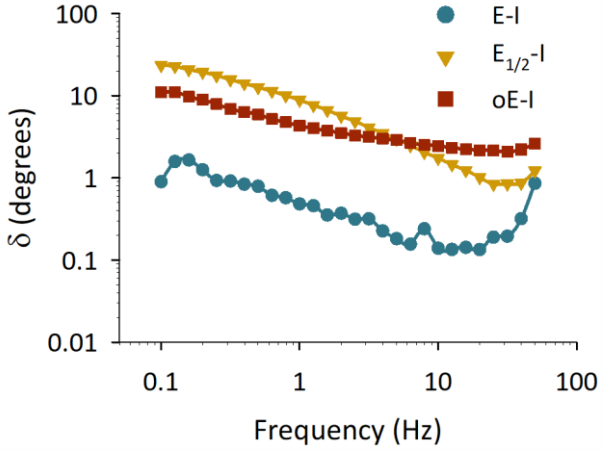

Figure S9. Evolution of phase angle (δ) as a function of frequency.

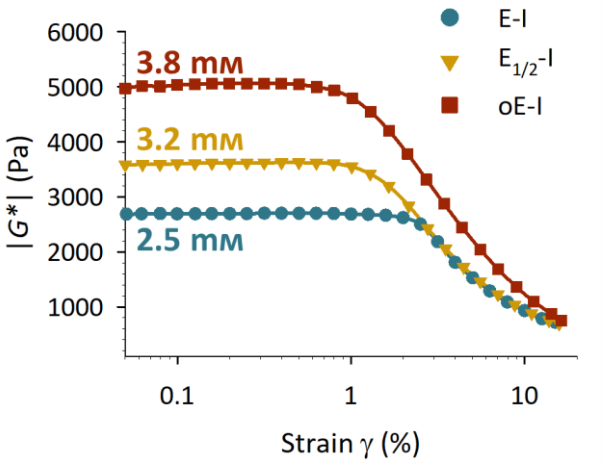

Figure S10. Strain sweep rheology of the physically crosslinked hydrogels (37 °C, 1 Hz) at 11.7% (w/v). The corresponding mass concentrations (mM) are noted on the graph (2.5 mM for E-I, 3.2 mM for $E_{1/2}$-I and 3.8 mM for oE-I).