SUPPORTING INFORMATION

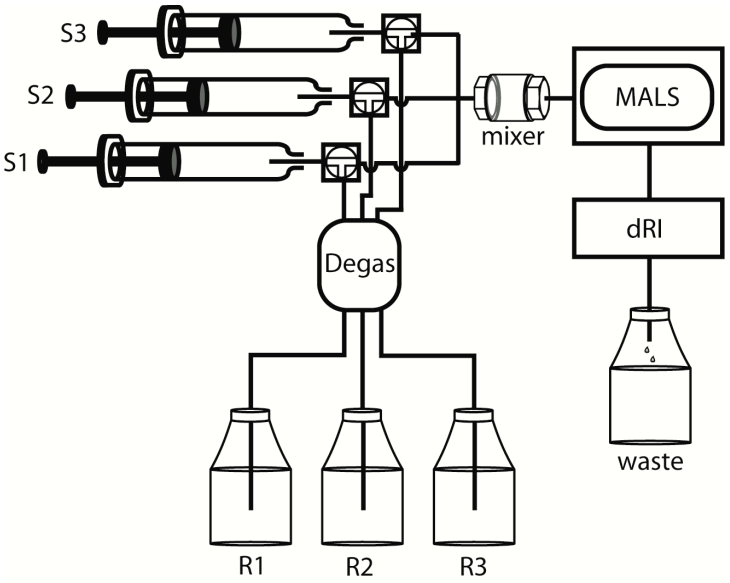
for

Tunable Encapsulation of Proteins within Charged Microgels

*Michael H. Smith, L. Andrew Lyon**

School of Chemistry & Biochemistry and the Petit Institute for Bioengineering & Bioscience, Georgia Institute of Technology, Atlanta, GA 30332-0400, USA

I. Supporting Figures and Schemes



Scheme S1. Schematic of flow system employed for particle characterization via MALS. Buffers, microgel and protein solutions were placed in reservoirs R1-R3 and loaded into syringes S1-S3. Scheme adapted from Ref 1.

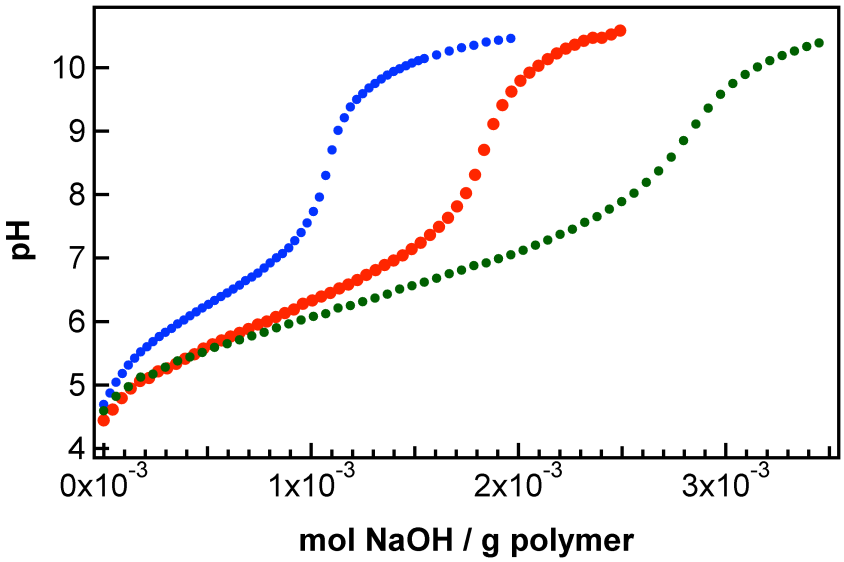


Figure S1. Potentiometric titrations of microgels for the measurement of total acid content. Particles were synthesized to contain 30 mol% (green), 20 mol% (red), and 10 mol%(blue) AAc comonomer.

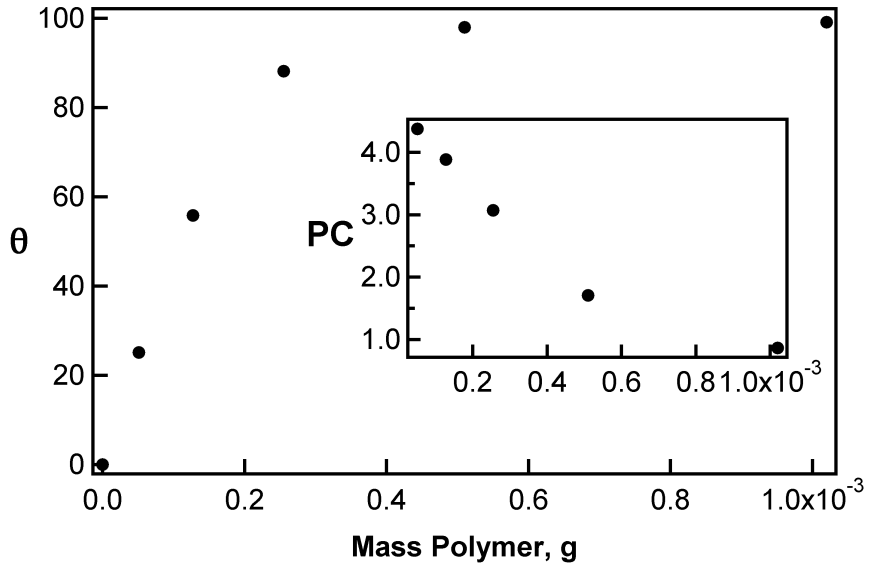


Figure S2. Traditional centrifugation/supernatant analysis methods to estimate the fraction of bound protein (θ) for 30 mol% AAc microgels at varying polymer concentrations in pH 7.0 and in the presence of 24 μM cyt c. Polyelectrolyte capacity values (inset) were calculated from the concentration of bound protein per mass of polymer.

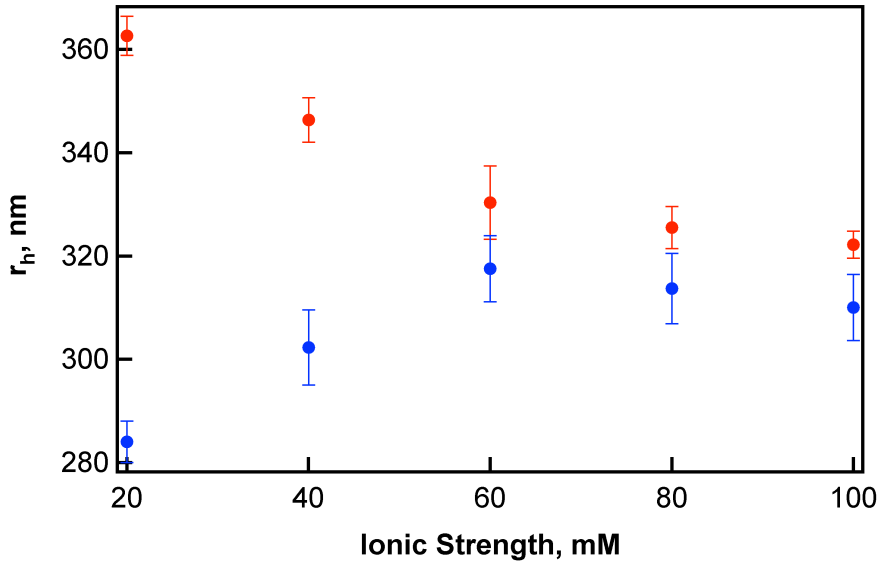


Figure S3. Influence of ionic strength on the hydrodynamic radius of loaded (blue, [cyt c] = 24 μM) versus unloaded (red) 30 mol% AAc microgels. All r_h values for were measured at identical particle concentrations in pH 7.0 phosphate buffer (10 mM) of varying salinity. Error bars represent one standard deviation about the means of several replicated measurements (n = 5).

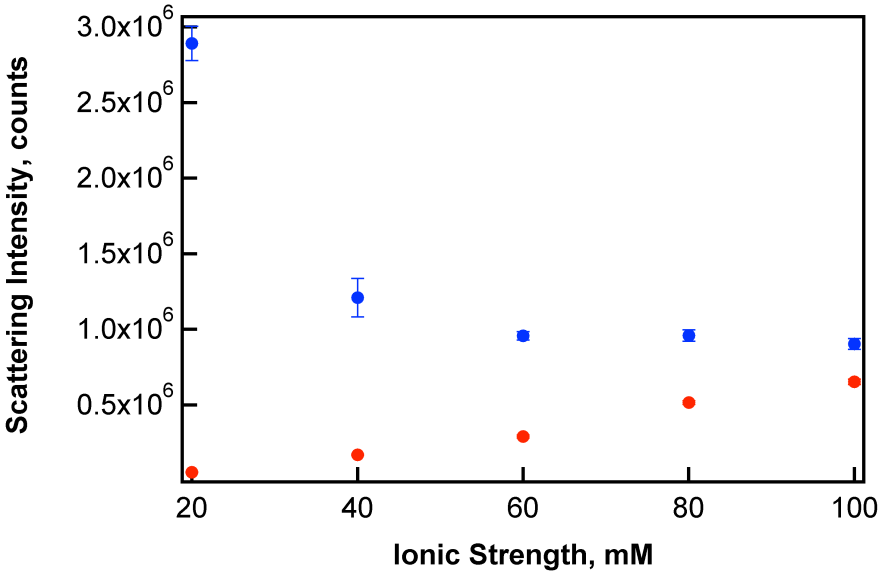


Figure S4. Influence of loading on overall microgel scattering intensity, as measured using the Dynapro DLS instrument at 90º scattering angle. Measurements were performed under identical conditions as reported in Figure S3 for bound (blue) and unbound (red) microgels. Error bars represent one standard deviation about the means of several replicated measurements (n = 5).

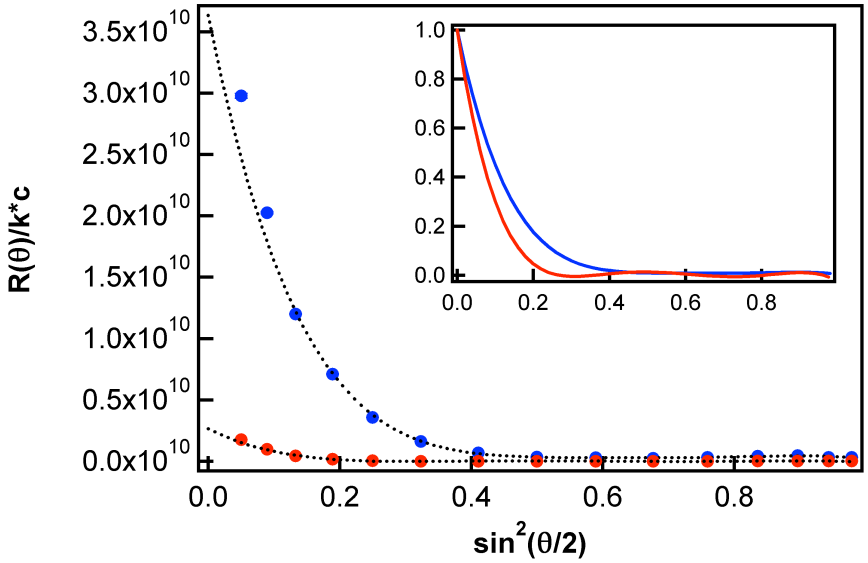


Figure S5. Example Debye plots for 30 mol% microgels (red) and for those microgels in the presence of 24 μM cytochrome c (blue) with polynomial fitting of the angle-dependent data shown (dotted black line). Inset: normalized polynomial fits for the particles in the bound (blue) and unbound (red) state.

II. Derivation of Eq. 3 from text

As described in the main text, the polyelectrolyte capacity is often measured for macroscopic hydrogels via weighing techniques, where

$$PC = \frac{\text{g of protein}}{\text{1 g of polymer}} \tag{S1}$$

Using MALS and measurements of particle Mw before and after binding, the PC was derived where

$$PC_{MALS} = \frac{\text{mass of protein loaded}}{\text{mass of a single microgel}} = \frac{M_{tot,cytc}}{Mw_{unbound} * (1/N_A)} \tag{S2}$$

$M_{tot,cytc}$ is the total mass of protein per microgel, $Mw_{unloaded}$ is the molar mass of native microgels (without protein), and N_A is Avagadro's number. The $M_{tot,cytc}$ was determined through

$$M_{tot,cytc} = \frac{dMw}{Mw_{cytc}} * \frac{1}{N_A} * Mw_{cytc} = \frac{dMw}{N_A} \tag{S3}$$

where dMw is the overall change in molar mass as described by equation 2 of the main text. Combining Eq. S2 with Eq. S3 and simplifying yields Eq. S4, reported in the main text as Eq. 4.

$$PC_{MALS} = \frac{dMw}{Mw_{unloaded}} \tag{S4}$$

References

1. Smith, M. H.; Herman, E. S.; Lyon, L. A. *The Journal of Physical Chemistry B* **2011,** *115* (14), 3761-3764.