Supporting information for:

Polysaccharide-based polyelectrolyte complex nanoparticles from chitosan, heparin, and hyaluronan

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The dynamic light scattering instrument measures the intensity autocorrelation function, $g^{(2)}(\tau)$, of the scattered radiation.

$$g^{(2)}(\tau) = \frac{\langle I(t)*I(t+\tau)\rangle}{\langle I(t)^2\rangle} \qquad \text{(SI.1)}$$

From the intensity autocorrelation function, the electric field autocorrelation function, $g^{(1)}(\tau)$, can be obtained according to the Seigert relationship.[1]

$$g^{(1)}(\tau) = \left|\frac{g^{(2)}(\tau)-1}{B}\right|^{0.5} \qquad \text{(SI.2)}$$

where B is a parameter that depends upon the number of coherence volumes in the sample and can be obtained from the limit of $g^{(2)}(\tau)$ as τ approaches 0. DLS correlation functions were analyzed by two separate methods that are commonly used to interpret light scattering of polymer solutions and colloids.

First, the regularization algorithm in the Dynamics software package (Wyatt, Version 6.10.1.2) was used to determine relaxation rates, Γ_i, corresponding to the diffusion of nanoparticles (and, potentially, other modes of relaxation in the samples). The regularization algorithm is a regression method for obtaining a distribution of relaxation rates that fit the field autocorrelation function to a superposition of multiple relaxations according to

$$g^{(1)}(\tau) = \sum_i w_i \exp(-\Gamma_i\tau)] \qquad \text{(SI.3)}$$

The relative intensity weight of the ith relaxation rate, Γ_i, is w_i. ($\Sigma_i w_i = 1$.) The regularization algorithm makes no assumptions regarding the shape of the distribution of relaxation rates. For quasi-elastic light scattering from single particles the Γ_i are related to an observed translational diffusion coefficient for particles, D_i, by the scattering vector, q ($q = (4\pi n_0/\lambda)\sin(\theta/2)$),

$$\Gamma_i = D_i q^2 \qquad \qquad \textbf{(SI.4)}$$

where n_0 is the refractive index of dispersant, λ is the wavelength of the scattered radiation in a vacuum, and θ is the scattering angle (90°). Finally the hydrodynamic radii (R_H) of the particles corresponding to each relaxation rate is calculated using the Stokes-Einstein relationship.

$$R_{H,i} = \frac{k_B T}{6\pi \eta D_i} \qquad \qquad \textbf{(SI.5)}$$

The use of equation SI.3 is particularly useful when samples are multimodal and meaningful information can be extracted from the presence of multiple relaxation rates. However, use of equation SI.3 is an ill-posed problem in that the number of relaxation rates required to accurately describe the system is not known, so the solution is only unique when there are no experimental uncertainties in the data and there are no rounding errors in the fitting algorithm.[1] This may result in peaks in the distribution of relaxation rates that have no physical significance. In our experiments, five replicate measurements were made for each sample. For each sample, the distribution of relaxation rates contained a primary peak (present in all five replicate measurements and generally accounting for > 90% of the total distribution) that was attributed to the diffusion of the PCN. Peaks in the relaxation rate distribution that represented faster or slower rates generally accounted for less than 10 % of the distribution of relaxation rates for all

samples. These secondary peaks were often present, but did not appear in all five replicate measurements for any of the formulations.

Second, field autocorrelation functions, $g^{(1)}(\tau)$, were also fit by regression using a modified Levenberg-Marquardt algorithm to a function containing a "fast" single exponential followed by a "slow" stretched exponential (Williams-Watts equation) of the form.

$$g^{(1)}(\tau) = A\exp(\Gamma_f \tau) + (1 - A)\exp\left[- (\Gamma_s \tau)^\beta\right]$$

(SI.6)

In equation SI.6 Γ_f and Γ_s represent the characteristic fast and slow relaxation rates, respectively, and the parameter β is a measure of the breadth of the distribution of relaxation rates for the stretched exponential component. The fitting was performed using the function fitting operation in the Igor Pro software package with the default convergence criterion (< 0.1 % change in χ^2 between successive iterations). Equation SI.6 has been found to be a good description of DLS for amphiphillic polymers in aqueous solutions in general, and for polysaccharides in particular, which are capable of interparticle interactions that result in a coupling of the individual particle dynamics to the surroundings.[1-6] In the semidilute regime, the values of A and β both decrease as the coupling among individual particles' motions becomes stronger, approaching 0 at the gel point, where the autocorrelation function is no longer described by equation SI.6.[4, 6] Thus, A and β are measures of the relative strength of particle-particle interactions.

There are important differences between these two methods for interpreting the DLS data which make these two models complementary. While the regularization method (equation SI.3) requires a large number of adjustable parameters (two parameters for each of an unknown number relaxation rates), equation SI.6 describes the field autocorrelation function with only four

adjustable parameters. Typically, each of the relaxation rates obtained from equation SI.3 is assigned to a diffusive mode, corresponding to a particle size, while equation SI.6 can be used to provide a different interpretation of slower relaxations (e.g., particle-particle interactions). If equation SI.6 reveals significant coupling among particles' relaxations, the slower relaxation modes observed using equation SI.3 can be interpreted differently. Finally, equation SI.6 is not capable of effectively dealing with multimodal distributions; it assumes that there is only one characteristic diffusive mode in the sample.

The results from the model fit to equation SI.6 for the chi-hep and chi-ha nanoparticle formulations are shown in Tables SI.1 and SI.2, respectively.

Table SI.1. Parameters from fits to equation SI.6 for chi-hep PCN at each charge mixing ratio.

Charge mixing ratio (+/-)	A^a	Γ_f $(s^{-1})^a$	β^a
17.55	0.68	216	0.41
10.53	0.62	205	0.45
4.68	0.66	192	0.57
2.73	0.65	163	0.46
1.75	0.62	152	0.43
0.78	-	-	-
0.50	0.43	191	0.64
0.29	0.58	251	0.58
0.13	0.60	272	0.47
0.08	0.73	287	0.45

[a]The uncertainties on the parameter Γ_f are all less than 2 % of the parameter value, and the uncertainties on the values of A and β are all less than 6 % of the parameter value.

Table SI.2. Parameters from fits to equation SI.6, for chi-ha PCN at each charge mixing ratio.

Charge mixing ratio (+/-)	A^a	Γ_f $(s^{-1})^a$	β^a
19.2	-	-	-
8.52	0.80	171	0.35
3.19	0.87	213	0.44
1.4	-	-	-
0.53	0.88	258	0.41
0.23	0.77	249	0.25

[a]The uncertainties on the parameter Γ_f are all less than 2 % of the parameter value, and the uncertainties on the values of A and β are all less than 6 % of the parameter value.

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