

Supporting Information for
**Stacking and Branching in Self-Aggregation of Caffeine in
Aqueous Solution: From the Supramolecular to Atomic Scale
Clustering**

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Dynamic Light Scattering Data Analysis

Stretched exponential functions are used to describe a distribution of decay rates and, in turns, a distribution of diffusion constants. The n^{th} order momentum of the τ distribution is:⁵

$$\langle \tau^n \rangle = \frac{\tau^n}{\beta} \Gamma\left(\frac{n}{\beta}\right). \quad (\text{SI.1})$$

The distribution polydispersity index σ is defined as the ratio $s/\langle \tau \rangle$ between the standard deviation s and the mean value $\langle \tau \rangle$.

The individual decay constant $\langle \tau(\theta) \rangle$ is directly related to the average apparent diffusion constant of the particles in solution:

$$D(\theta) = \frac{1}{\langle \tau(\theta) \rangle Q^2} \quad (\text{SI.2})$$

In general, in presence of large particles and polydispersed samples, the diffusion coefficient measured at a given θ is an apparent diffusion constant. The zero-angle diffusion constant D_0 was extracted from a linear extrapolation of $\frac{1}{\langle \tau(Q) \rangle Q^2}$ vs. Q^2 since

$$\frac{1}{\langle \tau(Q) \rangle Q^2} \approx D_0 + \kappa Q^2 \quad (\text{SI.3})$$

where κ is a sample dependent constant. The average hydrodynamic particle radii, $\langle R_{h1} \rangle$ and $\langle R_{h2} \rangle$, were then obtained applying the well-known Stokes-Einstein relation as

$$\langle R_{h^i} \rangle = \frac{k_B T}{6\pi\eta(T)\langle D_0^i \rangle} \quad (\text{SI.4})$$

where k_B is the Boltzmann constant, T the absolute temperature and $\eta(T)$ the solvent viscosity.

Equations SI.1 to SI.4 were used for the calculation of the zero-scattering angle hydrodynamic radius. The last step of the extrapolation is reported in Figure SI.1.

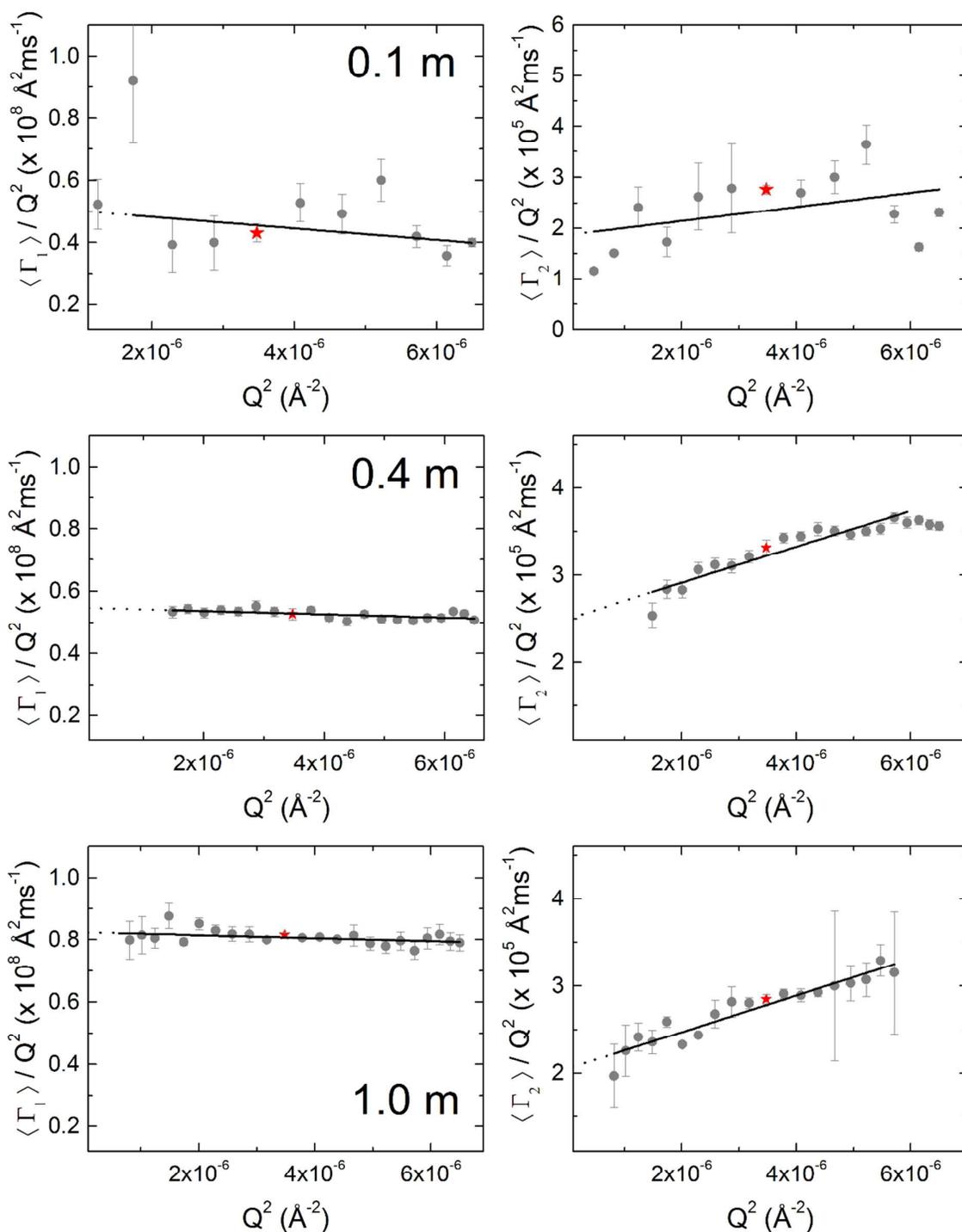


Figure SI.1. Angular dependence of the apparent diffusion constants $D_i(\theta) = \frac{\langle \Gamma_i(\theta) \rangle}{Q^2}$ obtained from the analysis of the intensity auto-correlation functions for the 0.1 m, 0.4 m and 1.0 m caffeine samples measured in D_2O at the 25°C , 43°C and 80°C respectively. The apparent diffusion constants measured at 90° scattering angle are indicated by red symbols (stars). Significant differences were present especially in the case of $D_2(90^\circ)$ vs. $D_2(0^\circ)$ values. Average hydrodynamic radii reported in the manuscript were always derived from

the extrapolated $D_i(0^\circ)$ values.

Guinier Analysis of SLS data

Static light scattering data were analyzed in terms of Equation (1). The fits, in the Q -range where the Guinier approximation is valid, are reported in Figure SI.2.

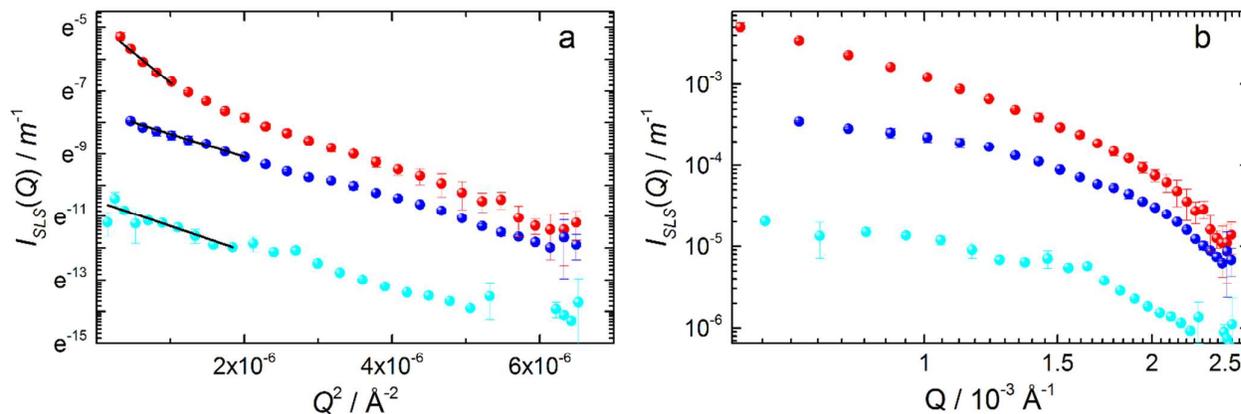


Figure SI.2. **a)** Guinier plots for the SLS data for 0.1 *m* (cyan), 0.4 *m* (blue) and 1.0 *m* (red) caffeine samples measured in D₂O. The more concentrated sample was characterized by the presence of larger aggregates (higher slope at low Q) as already indicated by DLS data. Fits according to Equation (1) of the manuscript are reported as solid lines. **b)** SLS data in the classical $I(Q)$ vs. Q representation. The Guinier region is clearly visible.

Isotopic Effect

In order to reduce the background incoherent scattering coming from ^1H atoms and to enhance the scattering intensity SANS measurements were performed in D₂O. Light scattering measurements were performed on the same solutions as used for SANS but they were also repeated in H₂O to investigate any possible effect induced by the isotopic substitution. As shown here by the intensity intensity auto-correlation functions reported in Figure SI.3, exchange of H₂O for D₂O does not affect the self-aggregation processes. The small differences visible could be induced by differences in the overall caffeine concentration.

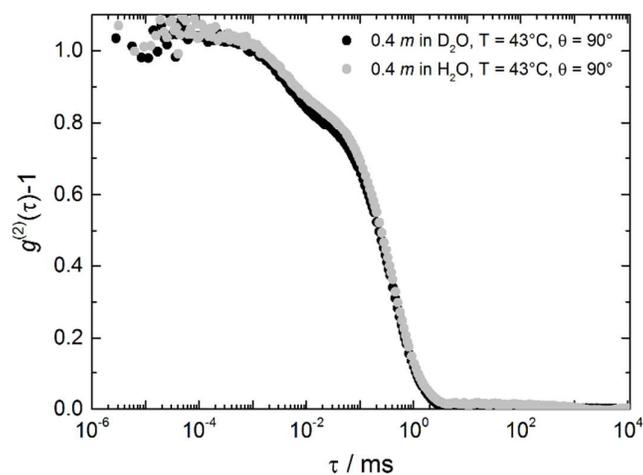


Figure SI.3. Comparison between the intensity auto-correlation functions for 0.4 *m* caffeine samples in H_2O (gray) and in D_2O (black). Time-axis values for the D_2O sample were corrected by a factor 1.118 in order to account for viscosity differences (at 45°C $\eta_{\text{D}_2\text{O}} = 0.671$ cP and $\eta_{\text{H}_2\text{O}} = 0.600$ cP). No substantial differences are visible.