Supporting Information

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Calculation of Measurable Properties

The free energy of solution A is in our approach given by the sum of three contributions:

$$A = A^{\mathrm{id}} + A^{\mathrm{hs}} + A^{\mathrm{ass}}, \qquad \textbf{[S1]}$$

where A^{id} is the ideal part, A^{hs} the hard-sphere contribution (1), and A^{ass} the protein–protein associative part (2–4). The ideal and the hard-sphere contributions are given by the next two expressions:

$$\frac{\beta A^{\mathrm{id}}}{N} = \ln\left(\Lambda^3 \rho\right) - 1, \qquad \textbf{[S2]}$$

$$\frac{\beta A^{\mathrm{hs}}}{N} = \frac{\eta(4-3\eta)}{(1-\eta)^2}, \qquad \textbf{[S3]}$$

where Λ is the de Broglie thermal wavelength (5) and η is the packing fraction, related to the density, $\eta = \pi\rho\sigma^3/6$. Note that A^{ass} is defined by Eq. **6** of the main text.

Once the free energy is known other thermodynamic quantities can easily be calculated (5). Chemical potential μ of the protein molecule and pressure P needed in calculations of the coexistence lines are obtained via these equations:

$$\mu = \left[\frac{\partial(A/V)}{\partial\rho}\right]_{T,V}, \qquad \textbf{[S4]}$$

$$P = \rho\mu - \frac{A}{V}. \qquad \textbf{[S5]}$$

Note that the ideal and the hard-sphere free-energy terms were differentiated analytically, whereas the associating term was evaluated numerically.

In the one-component system of two coexisting phases with equilibrium densities ρ_{a} and ρ_{b}, additional restrictions must be fulfilled. For every temperature T the equilibrium conditions read

$$\mu(T, \rho_{\mathrm{a}}) = \mu(T, \rho_{\mathrm{b}}), \qquad \textbf{[S6]}$$

$$P(T, \rho_{\mathrm{a}}) = P(T, \rho_{\mathrm{b}}). \qquad \textbf{[S7]}$$

To compute the number concentrations of the coexisting phases ρ_{a} and ρ_{b} efficiently, the iterative Newton–Ralphson method was used. The convergence criterium, requesting for the sum of relative errors in ρ_{a} and ρ_{b} to be less than 10^{-4}, was typically reached in fewer than five iterations.

The results for the reduced chemical potential $\beta\mu$ and dimensionless pressure $\beta P\sigma^3$ showing the coexisting points for lysozyme at $T = 270$ K and γIIIa-crystallin at $T = 300$ K are shown in Fig. S1. Dashed horizontal lines connect the number densities ρ_{a} and ρ_{b} of the two coexisting phases. The differences between densities of the coexisting phases, $\rho_{\mathrm{b}} - \rho_{\mathrm{a}}$, observed in Fig. S1 increase with the number of binding sites M. It is clear from our computation that larger number of sites M (solid line, γIIIa-crystallin, $M = 14$) yields a broader liquid–liquid phase diagram. For a lower number of sites (dotted and dashed line, lysozyme, $M = 10$) a somewhat deeper potential well would be needed to obtain the same critical temperature, determined by the $\beta\mu$ and $\beta P\sigma^3$ curves.

Among thermodynamic properties, the second virial coefficient B_2 is very important. In the framework of Wertheim's theory it is given by the next expression (6):

$$B_2 = B_2^{(\mathrm{hs})} - 2\pi M^2 \int_{\sigma}^{2d+a_{\mathrm{W}}} e_{\mathrm{R}}(r)\bar{f}_{\mathrm{ass}}(r) r^2 dr, \qquad \textbf{[S8]}$$

where $e_{\mathrm{R}}(r) = \exp[-\beta u_{\mathrm{R}}(r)]$. $\bar{f}_{\mathrm{ass}}(r)$ is the solid-angle average of the Mayer function defined as

$$\bar{f}_{\mathrm{ass}}(r) = \int f_{\mathrm{AB}}(\boldsymbol{x}_{\mathrm{AB}}(\boldsymbol{r}))d\Omega_{\mathrm{A}}d\Omega_{\mathrm{B}}, \qquad \textbf{[S9]}$$

$f_{\mathrm{AB}}(\boldsymbol{x}_{\mathrm{AB}}(\boldsymbol{r})) = \exp[-\beta u_{\mathrm{AB}}(\boldsymbol{x}_{\mathrm{AB}}(\boldsymbol{r}))] - 1$ is the site–site Mayer function, and Ω_{A} and Ω_{B} are the solid angles. Wertheim (7) obtained the result in analytical form

$$\bar{f}_{\mathrm{ass}}(r) = \frac{\exp(\beta\epsilon_{\mathrm{W}}) - 1}{24 d^2 r}(a_{\mathrm{W}} + 2d - r)^2. \times (2a_{\mathrm{W}} - 2d + r) \qquad \textbf{[S10]}$$

Another measurable quantity, closely related to B_2, is the osmotic compressibility χ_{osm} (8):

$$\chi_{\mathrm{osm}} = \left(\frac{\partial\beta P}{\partial\rho}\right)_{N,T} = 1 + 2B_2\rho + \ldots. \qquad \textbf{[S11]}$$

χ_{osm} was obtained by numerical differentiation of the pressure as suggested in the first part of Eq. **S11**.

1. Mansoori GA, Carnahan NF, Starling KE, Leland TW (1971) Equilibrium thermodynamic properties of the mixture of hard spheres. *J Chem Phys* 54(4):1523–1525.
2. Wertheim MS (1986) Fluids with highly directional attractive forces. III. Multiple attraction sites. *J Stat Phys* 42(3-4):459–476.
3. Wertheim MS (1986) Fluids with highly directional attractive forces. IV. Equilibrium polymerization. *J Stat Phys* 42(3-4):477–492.
4. Chapman WG, Jackson G, Gubbins K (1988) Phase equilibria of associating fluids chain molecules with multiple bonding sites. *Mol Phys* 65(5):1057–1079.
5. Hansen JP, McDonald IR (2006) *Theory of Simple Liquids* (Elsevier, Amsterdam).
6. Bianchi E, Tartaglia P, Zaccarelli E, Sciortino F (2008) Theoretical and numerical study of the phase diagram of patchy colloids: Ordered and disordered patch arrangements. *J Chem Phys* 128(14):144504.
7. Wertheim MS (1986) Fluids of dimerizing hard spheres, and fluid mixtures of hard spheres and dispheres. *J Chem Phys* 85(5):2929–2936.
8. Rosenbaum DF, Kulkarni A, Ramakrishnan S, Zukoski CF (1999) Protein interactions and phase behavior: Sensitivity to the form of the pair potential. *J Chem Phys* 111(21):9882–9890.

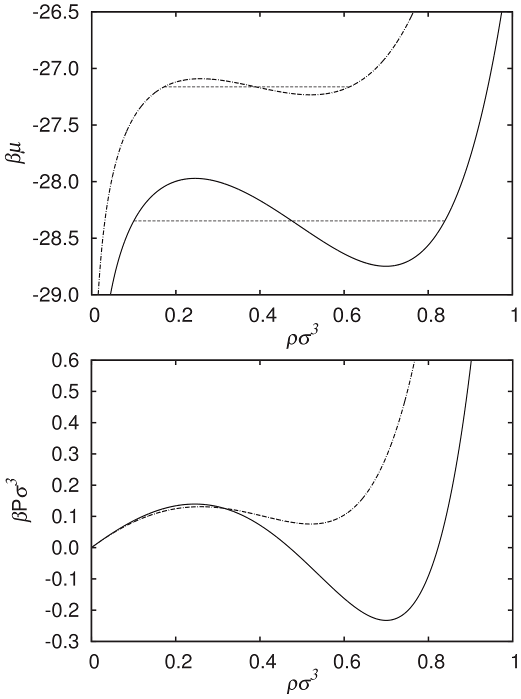

Fig. S1. Calculated reduced chemical potential $\beta\mu$ and dimensionless pressure $\beta P\sigma^3$ for lysozyme (dotted and dashed line, $T = 270$ K) and γIIIa-crystallin (solid line, $T = 300$ K), based on parameters from Table 1 of the main text.