Cluster Phases
in colloidal suspensions
and protein solutions

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Low-density: Cluster phases in dipolar colloids

- Amit Agarwal, post-doctoral researcher (2008 - 2009, now at DRDO, India)
- Ning Li, M.Sc. (2006 - 2008, now at Brandeis)

Simulation Collaborations: Marek Bromberek (2009 - ), Prof. I. Saika-Voivod, Ahmad Almudallal (Ph.D.), joint postdoctoral: Manuel Valera (now at SRU, PA).
Ultrahigh-density: phases in ultrasoft+di polar colloids

- Collaboration with Priti Mohanty and Peter Schurtenberger at Lund University.
Intermediate density: cluster phase in protein solutions

- Suliman Barhoum, Ph.D. (2008 - )
  Detection of Aggregate Structures in Protein and Micellar Solutions Using NMR Diffusometry

- Funding: NSERC, CFI, ACENet, MUCEP

Existence of liquid state requires attractive interactions. In fact, a sensitive balance between attractions and excluded volume repulsions.
Cluster Phases

Generalization of the liquid state

- Competition of short-range attractions and long-range repulsions can give rise to clusters in proteins and colloids (Stradner et al., Nature 2004).

Protein Clusters  Colloidal Clusters
Why would clusters have finite size in equilibrium?

- Micelles: Clustering arises from anisotropic interactions imposed by the hydrophobic-hydrophilic interface.
- Competition of interactions can lead to a free energy minimum at a finite, mesoscopic cluster size (Groenewold & Kegel, J. Phys. Chem. B., 2001).

Clustering in Complex Fluids

The tendency of molecules to associate with other molecules of the same species and to shun dissimilar molecules by bulk separation is well established. There is another kind of association, however, that is less understood in science: how do molecules or particles spontaneously form clusters that contain many particles or molecules and yet have a finite size? In the case of interparticle forces that are strongly attractive, like those between colloidal particles held together in suspensions by attractive forces, there is a universal tendency toward aggregation. In some cases, however, the interaction is purely repulsive, the precise shape of interparticle potential curve has critical, nonintuitive consequences. A hard-core repulsion with a softer repulsive shoulder (HCSS), for instance, forms a canonical sequence of phases with increasing particle density or pressure: a superlattice of spherical clusters, a 2D array of columnar clusters, multilamellar stacks,
Clusters in Protein Solutions

- How does one detect nanoscale clusters that are in constant motion?

Protein Clusters  Colloidal Clusters
An “Interference Peak” in SANS

- The “monomer” peak is at $q_m \approx 2 - 3 \text{ nm}^{-1}$
- There is an “interference peak”: $q_c \approx 1 \text{ nm}^{-1}$

Do Proteins Form “Equilibrium” Clusters?

- Stradner et al: YES! $q_c$ represents equilibrium protein clusters
- Shukla et al: NO! Peak position moves with concentration!
- We decided to test with a different technique: NMR.

Clusters in Protein Solutions

Neutron Spin Echo: Collective Diffusion at Short Times

- Commentary: “Different views from small angles”
- “Dynamic clusters at short times” (via $D_c$ at $q \to 0$)

“Macroscopic properties at the long time limit still determined by monomeric proteins”

Absence of equilibrium cluster phase in concentrated lysozyme solutions

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The different views from small angles

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The small-angle scattering of x-rays or neutrons from proteins in solution can provide important information about the structure of the protein and the nature of interactions or distance correlations among the protein molecules (1, 2). The temperature-dependent but does change with temperature and loses strength. It is the lack of concentration dependence in the positions of both of these peaks that is the basis for their proposal of equilibrium cluster formation. In their article, Structures of these aggregates with electron structure factor terms by taking into account the asymmetric shape of lysozyme, and the higher q concentration-independent peaks they obtain is attributed to the orientational coupling between the form and structure factors (11).
In a uniform magnetic field: Larmor frequency of a proton is not position dependent.

In a field gradient: Larmor frequency is position-encoded. Coherence of signal is lost due to “dephasing”

Stimulated Echo: “a pulse sequence” that refocuses signal loss due to static fields

Diffusion → irrecoverable signal loss → attenuation of echo.

\[ S'(g) = S(0) \exp (-kD), \text{ where } k \approx (\gamma g \delta)^2 \Delta, \Delta \text{ is the diffusion time.} \]
A two-site exchange model is known to work well in NMR of surfactant systems (Soderman & Stilbs, Prog. NMR Spec., 1994).

\[ D_{\text{observed}} = \frac{bD_{\text{mon}} + (1 - b)D_{\text{agg}}}{\text{Normalization}} \]  

Effects of NMR Relaxation

- Unique to NMR: ps - ns molecular correlation times give rise to ms - second NMR relaxation times.
- The relevant relaxation time in the stimulated echo is the longitudinal relaxation time $T_1$.

**Relaxation-weighted Diffusion**

- For single-species diffusion in the long-time limit: $D$ cannot be a function of $\Delta$.
- For two species the observed diffusion-coefficient is a relaxation-weighted average of $D$ for each species ("m" for monomer, "a" for aggregate):

$$D_{\text{observed}} = \frac{[bD_m \exp (-\Delta/T_{1,m}) + (1 - b)D_a \exp (-\Delta/T_{1,a})]}{\text{Normalization}} \quad (2)$$
Protein Solutions

![Graph showing D vs Δ (ms) for different concentrations C.](image)
• We measure diffusion coefficient $D$ in the long-time limit: $(\Delta \gg \tau_{\text{Brownian}})$

• $D_{\text{observed}}$ vs protein volume fraction.

• $D_{\text{observed}}$ increases with $\Delta$: this is consistent with relaxation weighting.
• We can fit different concentrations to obtain the monomer fraction \( b \) and aggregate fraction \( 1 - b \).

• Monomer diffusion \( D_m \) is consistent with simulations for crowded diffusion of model proteins (Han & Herzfeld, Biophys. J., 1993).

• **Main point:** short-range attractions and long-range repulsions can result in energetically favourable finite-size clusters.

Colloids with Dipolar Interactions
**Point Dipolar Approximation**

- Has inbuilt into it both repulsive and attractive interactions.
- External electric field $\vec{E}_{ext}$ induces dipoles.
- Dipoles interact:
  \[ U_{dip}(r)/k_B T \sim -\left[ \sigma^6 E_{ext}^2 / r^3 \right] (3 \cos^2 \theta - 1) / 2 \]
- Dipolar spheres self-organize into chains.
- Chains interact.

**Stacked chains: repulsive.**

**Staggered chains: attractive**
Colloids in Electric Fields

Experimental

- Laser-scanning confocal microscopy.
- Brownian microspheres:
  - silica $\sigma = 0.8\mu m$ or PNIPAM $\sigma = 1.45\mu m$ at 20°C
- AC electric fields: $f > 100$ kHz, oscillating too fast for double layer)
Experimental

- Laser-scanning confocal microscopy
- Brownian ($\sigma = 0.8\, \mu m$) microspheres:
  fluorescent-core–non-fluorescent-shell silica colloids in water-DMSO
- AC electric fields: $f > 100\, \text{kHz}$, oscillating too fast for double layer

Fluorescent 3D Confocal Microscopy
Dipolar colloids form body centred tetragonal (BCT) crystals.

- BCT is the theoretically minimum energy structure:

Microstructure at Lower Packing


- **High Densities:** Percolating particulate regions, non-percolating particle-poor regions.
- **Intermediate densities:** Particulate clusters, percolating particle-poor regions.


- Represent chains as particles with “stacked” or “staggered” effective interactions
- For $\phi > 5\%$, experiment and simulation agree.

- 3D: phase diagram at moderate to high densities Hynninen et al, PRE, 2005.
Coarsening of structures with time
Unusual cellular structures at low densities \((\Phi < 4\%)\)

- **Colloidal dipolar:**
**Mechanism:**

- Is likely an equilibrium structure: \( t_{\text{obs}} = 5000 \times \tau_{\text{Brownian}} \)
- Void phase (percolating non-crystalline particle networks) only exists at low densities.
- The lengthscale increases with sample thickness.
“Re-entrant” Percolation of Clusters

Experiment

• **High Densities:** Percolating particulate regions, non-percolating particle-poor regions.

• **Intermediate densities:** Particulate clusters, percolating particle-poor regions.

• **At very low densities:** “re-entrant” percolation of particulate regions, surrounding voids (particle-poor regions): **not seen in 2D simulation.**
Competing Interaction Forces

- A sensitive balance between intermediate-range attractive forces and shorter and longer range repulsions.

- Chain-chain undulations can produce fluctuation-induced attractions (Halsey & Toor, PRL 1990).

- Beyond a lengthscale $\ell > L \tan(54^\circ)$, repulsions dominate again.
Clusters in Dipolar Colloids

- Along the field direction, there are well-formed chains (a 1D crystal, albeit subject to strong fluctuations).
- **At large packings:** single-crystal or poly-crystalline BCT.
- **At lower packings:** crystalline cluster islands that do not merge to form one big blob.
- **At very low packing:** an unusual percolating cluster ("void") phase
- Void phase characterized by disordered microstructure in the plane perpendicular to the field.

As-yet unanswered question

What gives rise to the percolating networks at ultra-low densities?
• Interactions Yukawa-like for $a_s > \sigma$, much softer for $a_s < \sigma$.

• How does the introducing anisotropy affect phase behaviour?

• Graded dielectric spheres: what is the nature of the electromagnetic interactions? Can we make better electrorheological fluids?
Field-packing fraction phase diagram
Amorphous to crystalline: $\phi_{\text{eff}} = 0.85$

$\phi_{\text{eff}} = 0.85$, $E = 0.23 \text{ V/\mu m}$. 
Field-driven transitions

**Amorphous to amorphous:** $\phi_{\text{eff}} = 2.0$

- Field response at surprisingly low field strengths.
- Arrested phase separation at the highest packings: no sign of anisotropy!!
- Theoretical input welcome - probably should include the effects of mobile ions.
Conclusions

Dipolar colloids: percolating particle clusters at ultralow packing

- A 2D model captures physics at moderate and high densities.
- Intermediate-range attractions: dielectrophoresis?
- Unanswered: where does the void lengthscale come from?


Ultrasoft, dipolar colloids: new phases at ultrahigh packing

- Structure formation at low fields: mobile ions must be important
- Amorphous to crystal phase transition
- Amorphous to amorphous transition.


Equilibrium Clusters in Proteins

- Clusters exist in equilibrium.
References


Funding and Contacts

- NSERC, CFI, ACENet, MUCEP

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Can quantify this by measuring equations of state

In zero field:

As a function of field strength:

An intermediate-range attractive force?

- Dielectrophoretic particle segregation with patterned electrodes.
- The stable configuration is the region between the electrodes.
- The centre of the electrode is locally stable close to the bottom.
- With unpatterned electrodes, the particle-chains are the non-uniformity! S

Possible Origin of the Void Phase
Patterning in 3D

- The location AND shape of clusters can be controlled.
- The size of the clusters can be controlled with frequency and (low!) field strength.

**TOP**

**BOTTOM**