

## Applications to Soft Matter: general discussion

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**Gourav Shrivastav** communicated:† The authors have shown the self assembly of the particles *via* oscillatory forces by periodical change in pH. 1. How does the rate of ionization of acids and bases affect the result?

2. Does slow oscillation break the dimer formed and therefore is a fast pH required to discriminate between the dimers and free particles ?

**Mario Tagliazucchi** communicated in reply: 1. We don't believe that the rate of ionization of weak acids or bases will impact the dynamics of our system. In our previous work<sup>1</sup> we have shown that the pH oscillations should have a timescale ranging from microseconds to seconds (depending on the size of the colloids) in order to obtain self-assembled structures. Acid–base neutralization is usually faster than this timescale.

2. Slow oscillations will indeed break the dimers into free particles. For that reason, fast oscillations (*i.e.* oscillations faster than the diffusional timescale of the particles) are required to obtain the dimers as well as the other self-assembled morphologies.

1 M. Tagliazucchi, E. A. Weiss and Igal Szleifer, *PNAS*, 2014, **111**, 9751–9756.

**Charusita Chakravarty** communicated: The authors use Brownian dynamics simulations of charged particles with weak acidic and basic character and oscillating pH to generate interesting self-assembled structures under far from

† The paper by I. Szleifer was not presented at the meeting, but questions and responses have been provided afterwards.

equilibrium conditions. What would be the possible experimental realizations of this simulation?

Could varying the salt concentration in the colloidal clay systems studied in the articles by Joshi and Bandhyopadhyay be used for this type of non-equilibrium self-assembly?

**Ranjini Bandyopadhyay** communicated in reply: There are, indeed, important similarities between the system discussed in paper 7909 and the clay suspensions studied experimentally in papers 8549 (by Ali and Bandyopadhyay) and 8570 (by Jataw and Joshi). For example, in both the clay suspensions (Sodium Montmorillonite, Na-MMT, and Laponite) discussed in 8549 and 8570, structures form spontaneously due to the details of the time-dependent interactions between the constituent particles/particle stacks (tactoids). The sensitivity of the assembled structures to the inter-particle potentials has been discussed in detail in paper 7909. There also exist important differences between the system studied in 7909 and the clay suspensions studied in 8549 and 8570. Clay tactoids (stacks), which invariably exist in aqueous suspensions, break up spontaneously and release, during this exfoliation process, sodium ions into the bulk medium. Since tactoid exfoliation is a largely uncontrolled process, controlling the details of the inter-particle potentials in clay suspensions is a difficult task. Furthermore, in the clay suspensions in papers 8549 and 8570, strong inter-particle interactions drive the systems toward kinetic arrest, with the suspensions forming attractive gels in the presence of salt in the bulk medium, or glassy suspensions that are dominated by inter-particle repulsive interactions in the absence of externally added salt. This unavoidable process of kinetic arrest is seen in both clay suspensions and is absent in the model system studied in paper 7909. It is also to be noted that the presence of strong charge polydispersity in the Laponite system studied in paper 8570 and the presence of charge, size and shape polydispersities in the Na-MMT system studied in paper 8549, rule out the possibility of the formation of ordered structures such as those reported in paper 7909.

**Mario Tagliazucchi** communicated in reply: The possible experimental realizations of this system were discussed in our recent published work.<sup>1</sup> Briefly, we first estimated the timescale of the pH oscillations required for dissipative self-assembly. This timescale is in the second–microsecond range for colloids in the nanometer–micrometer size range. We believe that pH oscillations can be achieved in this timescale using photoacids<sup>2</sup> or electrochemical pH control.<sup>3</sup> The main challenge in such experiments would be to achieve and maintain a sufficiently low ionic strength (in our system, the Debye length of the solution should be similar to the size of the particles in order to obtain the self-assembled structures). Oscillating the salt concentration of the clay systems of Joshi and Bandhyopadhyay may lead to dissipative structures if the timescale of the oscillations is faster than the diffusional timescale of the particles  $\sim d^{2/D}$ , where  $d$  is the size of the colloid and  $D$  is its diffusion coefficient. The structures that may form will be different from ours, since the system is different.

1 M. Tagliazucchi, E. A. Weiss and Igal Szleifer, *PNAS*, 2014, **111**, 9751–9756.

2 R. M. D. Nunes, M. Pineiro and L. G. Arnaut, *J. Am. Chem. Soc.*, 2009, **131**, 9456–9462.

3 R. Shacham, D. Avnir Daniel Mandler, *Adv. Mater.*, 1999, **11**, 384–388.

**Priyadarshi Roy Chowdhury** communicated: "The colloidal molecules are dissipative structures that cannot be obtained in the absence of pH oscillations". Could you discuss this comment in more detail.

**Mario Tagliazucchi** communicated in reply: In the conditions of composition and density where we observe colloidal molecules, well-defined colloidal molecules are absent in fixed pH simulations. Thus, the colloidal molecules only form in the presence of pH oscillations and they are dissipative structures. This result does not preclude, of course, that the same colloidal molecules can form for other systems (*i.e.* particles interacting through other type of potentials) or even in the same system for other conditions.

**Lynn Walker** opened the discussion on the paper by Madivala G. Basavaraj: The mechanism for adsorption of colloidal particles at fluid interfaces is one of pinning due to formation of a solid–fluid contact line. This should not decrease the interfacial tension at low coverages. So why is the drop in interfacial tension with time interpreted as adsorption of particles in one case (one pH condition) and not in another (another pH condition). I wonder if the interfacial tension is actually responding to the adsorption of some other species in the system?

**Madivala G. Basavaraj** replied: If we compare the dynamic surface tension data for a pure water–decane interface and pH=2 case, it can be argued that the ionic species in the suspension (at pH=2), even if they adsorb on to the interface, do not reduce the surface tension. In our experiments, the drop in surface tension is observed when the pendant hematite suspension drops at pH of 4.0 and 6.5 are present in decane. We argue that this drop is due to particle adsorption to the suspension drop–decane interface. Since there is no change in surface tension with time for pH=2, we infer no particle adsorption. We refer to our recent work,<sup>1</sup> where we observe similar results for the adsorption of silica nanoparticles, where the electrostatic double layer interactions are screened by the addition of sodium chloride. We have not looked at the adsorption of other ionic species present in the system at this point. However, in the silica nanoparticles system, if NaCl present in the suspension drop was to adsorb at the interface, then the interfacial tension should have increased.<sup>2</sup> As the decrease in surface tension is observed in two independent studies this is a result of adsorption of particles due to low surface charge density.

1 V. R. Dugyala, J. S. Muthukuru, E. Mani and M. G. Basavaraj, *Phys. Chem. Chem. Phys.*, 2016, **18**, 5499.

2 reference: Chapter 3, Arthur W. Adamson and Alice P. Gast, *Physical Chemistry of Surfaces*, Wiley-Blackwell.

**Vinothan N. Manoharan** addressed Madivala G. Basavaraj and Lynn Walker: Following up on Prof. Walker's question on the observed time-dependence of the dynamic surface tension: ellipsoids adsorbed at the interface must distort the shape of the interface, and as a result there are capillary interactions between adsorbed particles. When the particles are close enough, their capillary distortions overlap, reducing the surface energy. Could the change in the surface tension as a function of time reflect a transition between the adsorption of isolated ellipsoids (where their capillary distortions do not overlap) and the

adsorption of ellipsoids close to other particles, such that they interact with one another?

**Lynn Walker** replied: I wonder if this would be a way to estimate a surface concentration? Since the geometry of the particles is known, is a calculation of the surface concentration needed to have overlap of the capillary distortion possible? It would certainly be an estimate, but it could back up the dynamics.

**Madivala G. Basavaraj** responded: In the dynamic surface tension data, initially the decrease is relatively steep followed by a more gradual change. If the transition from the adsorption of isolated ellipsoids to multiple ellipsoids (close to each other and associated interface deformation overlap) were to cause the change in surface tension with time, then the surface tension should start to change after there are enough ellipsoids at the interface, which is not the case. Therefore throughout the dynamic surface tension measurements there is particle adsorption, however, the rate of adsorption depends on several factors which are discussed in the manuscript.

**Lynn Walker** remarked: As a follow up to my earlier question, I think that the dynamics of the interfacial tension might contain more information than is being considered. The dynamics are quite rich. If this is particle adsorption as Basa argues, then what can be learned from the timescales (the reaction-diffusion equation with an EOS like Volmer might be appropriate)? If this is adsorption of another species or even replacement of counterions near the interface, then that could also be interesting in understanding the phenomenon.

**Madivala G. Basavaraj** responded: I completely agree. It would be very interesting to look at the dynamics in more detail – especially – contact line dynamics, possible adsorption of ionic species, and accumulation of ions close to the interface. This would add to a much better and complete understanding of the adsorption of particles to fluid interfaces.

**Neena S. John** commented: In the case of isotropic particles, the effective radius is considered to calculate the interfacial free energy factor. For the present case of ellipsoids, do you consider the 'aspect ratio' for the calculations?

Also, will the ellipsoids assemble at a planar interface of decane–water in a similar manner?

**Madivala G. Basavaraj** answered: In the calculation of interfacial free energy, the area that the particle occupies at the interface is considered, that is how the radius of spherical particle figures in these calculations. The area is typically the area of intersection of the sphere and the interface plane. As long as this area is known, such calculations can be made for particles of any regular or arbitrary shape.

For ellipsoids, there are shape induced interface deformations, therefore, this area of intersection is not a perfect ellipse, but an undulated ellipse. Therefore, one has to consider this effect. Such deformations have been measured for ellipsoids and rod-shaped particles. Considering these effects in interface free energy calculations will involve more tedious calculations. Moreover, for complex shaped particles there are no analytical expressions for the calculation of the area

of intersection. The behaviour of ellipsoids at interfaces is known to be very different from that of spheres. Ellipsoids that are homogeneous (not Janus) are known to position themselves flat at the interface – with their major axis parallel to the interface plane. For ellipsoids, due to long range capillary forces, both the structure and interfacial properties are known to be aspect ratio dependent.

**Guruswamy Kumaraswamy** asked: Could you please comment on the change in wettability of your haematite particles with a change in pH? Could this be sufficient to account for their pH-dependent segregation at the oil–water interface, without having to resort to the image charge argument?

**Madivala G. Basavaraj** responded: The idea of a possible change in wettability of hematite particles with a change in pH is an interesting one. I wonder if there are any tools to measure such wettability changes in solution. We have measured the contact angle of micron sized hematite particles (of different shapes – such as spherocylinders, cubes, dumbbells) at the water–decane and water–air interface by considering an aqueous sub-phase maintained at pH=6.5. The contact angles are obtained from the analysis of microscopy images obtained *via* a gel trapping technique. The values of the three-phase contact angles obtained irrespective of particle shape are  $\sim 50$  and  $\sim 40$  degrees, respectively. This suggests that the particles are still partially hydrophilic. It would be interesting to carry out wetting studies by considering aqueous phases at different pH values to see if there is any significant change in particle wettability.

**B. L. V Prasad** asked two questions: 1. Are the particles that are left to equilibrate in the system for 1 hr assuming any Janus character? Can this phenomenon explain the observations? 2. Please comment on why 1 hr is needed? What could be happening in this 1 hr?

**Madivala G. Basavaraj** answered: In our drop compression experiments, 1 hr of equilibration time is used to provide enough time for the particles to diffuse and eventually adsorb to the interface. This time period helped us to clearly demonstrate the buckling phenomena in the suspension drops at pH=6.5. In principle, buckling is expected to occur even if the equilibration time is lower, however, at a higher compression ratio. There is no evidence to support the hypothesis that the particles would assume Janus character. Since there is no chemical or physical charge to the particle during our experiments, particles assuming Janus character is very much unlikely.

**Vinothan N. Manoharan** commented: Another reason that the surface tension might evolve in time is contact-line pinning. Experiments from my group<sup>1</sup> show that micrometer-scale particles can take months to years to reach their equilibrium position at a fluid–fluid interface. We attribute the slow relaxation to pinning of the contact line on defects on the particle surfaces. In a dense monolayer of such particles, the surface energy might gradually decrease over time.

1 D. M. Kaz, R. McGorty, M. Mani, M. P. Brenner and V. N. Manoharan, *Nature Mat.*, 2012, **11**, 138–142.

**Madivala G. Basavaraj** responded: In the present study we use ellipsoids that are about 250 nm in length (major axis) and 50 nm in diameter (minor axis). It is clear from our experiments and earlier reports that the orientation of homogeneous (in terms of surface chemistry) ellipsoids is such that their major axis is parallel to the water–decane interface. Thus, the relevant lengthscale is the length of the minor axis of ellipsoids and associated contact line. The lengthscale and timescale associated with contact-line pinning and equilibration period for the particles used in the present study are clearly much smaller than that for micron-sized particles. The timescale of the experiment which is of the order of hours seems much longer, however, the measurement of contact line dynamics for smaller particles will help conclude if surface tension evolution due to contact line pinning is indeed the reason for the decrease in surface tension observed. Since the adsorption of particles is energetically favored there is strong reason to believe that the adsorption of particles does decrease the interfacial tension. This in our study is confirmed from the buckling experiments and SEM micrographs.

**Ajeet Srivastav** remarked: How would the shape of the nanoparticles (from spherical to cylindrical), in particular the nanowires, affect the observations with the higher interfacial area available to interact with?

**Madivala G. Basavaraj** answered: In general, interfaces coated with anisotropic particles such as ellipsoids or graphene sheets are shown to be solid-like and show interesting buckling behavior. There are reports on the compression of planar monolayers of rod-like particles, however, we are not aware of any pendant drop buckling experiments.

**Ranjini Bandyopadhyay** opened the discussion on the paper by Lynn M. Walker: Since the amphiphilicity of the middle blocks of the triblock copolymers are temperature-dependent, have you looked at the temperature-dependence of nanoparticle transport in your experiments? Also, would you comment on the release of the nanoparticles at specified targets?

**Lynn Walker** answered: Temperature is an important variable in these systems. For this work, we chose to use concentration to vary the structure since temperature would alter the PEO/water environment and add another complication. Isothermal experiments allowed us to compare to polymer theories for diffusion in semidilute polymer solutions. We have done other work with temperature, but this adds in new micelle transitions (from spheres to cylinders) and phase boundaries. This would be the next stage of the work and we have not yet designed the appropriate study. To get the nanoparticulate phase out of the nanostructured material, we could either cool the system below the CMT ( $\sim 15^\circ\text{C}$ ) or dilute with solvent (water). This would result in a polymer/nanoparticle dispersion which could be separated with centrifugation or filtration. It is worth noting that the block polymer chosen is FDA approved for many uses inside the body, so separation might not be necessary.

**G V Pavan Kumar** asked: Can you arrest the nanoparticles and further transfer them on to a substrate ?

**Lynn Walker** replied: It should be possible. However, we have purposely focused on tools to allow for structuring nanoparticles in three dimensions. I think that there are probably better ways (lithography and directed assembly) to do things on a substrate. Here, we wanted to find ways to use self assembly in three dimensions.

**Alison Edwards** commented: It is self-evident that the materials you are discussing which give clear diffraction patterns that you have shown here are crystalline under the conditions of your experiment. Your patterns are reminiscent of those obtained both by experiment and in analysis of diffuse scattering from 'hard' materials – especially those published by Richard Welberry. I would note that it is the long range order which gives rise to the sharp diffraction spots definitive of what is considered to be crystallinity, whereas the 'disorder' at short lengthscales gives the intensity outside the Bragg peaks – of which you have a great deal. Intriguingly, diffuse scatter does not drop away with  $2\theta$  – do your patterns continue out in reciprocal space (or  $Q$  if that is your preferred measure)? The concepts developed by Prof. Welberry could be extended to make use of the data you have available in these patterns and there are some quite old modelling methods which can also be applied to such data to extract models for such materials.

**Lynn Walker** answered: We will follow up and thank you for the reference to Welberry's work. We come to this field from the structured fluid world and are realizing the importance of approaching these as crystalline materials (albeit 'soft' ones with significant form factor effects). Our patterns do continue in  $q$  space but only for 3–4 peaks. Using the proper conditions, we are able to align the power with shear to see 'single crystal' patterns. For some of these, we see higher order peaks. However, the peak height drops off quickly and the incoherent background comes up quickly. We usually have 3 orders of magnitude in intensity and that is often our limiting factor in peak resolution. As we have determined the conditions for 'annealing' the crystals, our scattering patterns are becoming even more crystalline and we will definitely approach these from the crystalline point of view!

**Yogesh M. Joshi** said: At 25°C how sharp is the liquid to crystalline transition for pluronic F127 when the concentration is increased? Is there any biphasic region where both crystal and liquid states coexist? Depending upon how fast the temperature of 25°C is attained, could there be a possibility of having a disordered glassy structure in Pluronic F127?

**Lynn Walker** replied: If you look at Fig. 1 in our manuscript, you will note that we avoided samples at the  $C_{gt}$  point (the concentration at 25°C of the transition). This is because the results are much more time dependent in this region. For all of our work, we were careful to slowly warm the samples from the liquid to the solid state and then allow for significant annealing time. So, we are confident that the results reported are for materials that are crystalline. It is possible to trap these materials in a glassy state by rapid heating from the liquid to the solid. It is also possible to create metastable structures by cooling/heating rapidly through the OOT seen in many of these systems. Our paper<sup>1</sup> discusses these issues with structural measurements. We have not yet addressed the issue of nanoparticle diffusion in a kinetically trapped system.

1 T. A. Lafollette and L. M. Walker, *Polymers*, 2011, 3, 281.

**Sudeep Punnathanam** asked: In cases where the mobility of the nanoparticles is severely limited, are you able to crystalize structures that are similar to those observed in substitutionally ordered alloy systems?

**Lynn Walker** responded: This is a good question and one for which I do not have an answer. There is no reason that this would not work and we are interested in investigating it. The nanoparticles in the hindered cases are held in the interstitial pockets by steric forces, so we lack the specific interactions in colloidal aggregates. I suspect that we would be able to form clusters of a specific size (set by the block copolymer geometry), but I am not sure that we would generate the highly ordered structures reported in oppositely charged nanoparticle or colloidal crystals and alloys.

**Siddharth Kulkarni** asked: By increasing the particle size above the critical micelle size (characteristic repeat distance), is it possible to see the particles at the grain boundaries? Further, is it possible that such particles partitioned at the boundaries can form a network and enhance the viscoelastic properties of the triblock copolymer matrix?

**Lynn Walker** replied: We did look at different particle sizes. If the particle size is significantly larger than the unit cell size (micelle size), then the particles sample the bulk behavior of the material. We see no templating or location of the particles coincident to the block copolymer matrix or any grain boundaries.<sup>1</sup> Our earlier templating work does not suggest partitioning to grain boundaries. We do see the spacing of the nanoparticles picking up the crystal structure of the block copolymer micelles. In this work, we sheared one sample to form a single crystal and saw no difference in diffusivity of the nanoparticles. This is indirect evidence, but adds to our body of work that suggests that particles do not partition to grain boundaries in this system. I should also note that we have not been able to determine a domain size for these materials; it requires very little shear to align these into near single crystals.

One difference to note is that this is a highly solvent swollen system. Most work on particles dispersed in block polymers has focused on melts. In a melt system, the energy difference between a grain boundary and the organized micelles is probably considerably more than in this solvent swollen system.

1 D. C. Pozzo, K. R. Hollabaugh and L. M. Walker, *J. Rheol.*, 2005, 49, 759–782.

**Guruswamy Kumaraswamy** queried: Could you please comment on the diffusivity of the fluorescent probes in your ordered micellar systems relative to semidilute PEO solutions where the mesh size is comparable to PEO in the micellar phase?

**Lynn Walker** answered: We thought about this comparison a great deal. Fig. 8 in the manuscript has a prediction for the diffusivity for PEO solutions equivalent to the coronas in the block copolymer solution. We used the molecular weight of the PEO block in the copolymer and calculated concentration using the volume



available without the PPO cores. What we see is that the diffusivity in the block copolymer is significantly slower than what would be observed in the equivalent PEO solution. We compared to experimental data (Fig. 8) and a theoretical model for diffusion of nonsticky spheres through unentangled polymer solutions (Fig. 9). The comparison differs slightly, but the overall results are similar in that the nanoscale structure of the block copolymer solution causes a decrease in transport beyond just transport through the corona.

**Priyadarshi Roy Chowdhury** communicated: Does the complexity of the self-assembly of block co-polymers cause any hindrance to the nanoparticle diffusivity?

**Lynn Walker** communicated in reply: Yes. This was the point of the work to show that the nanoscale structure is what controls mobility of the nanoparticulate phase dispersed in the block copolymer solution. The findings show that it is possible but that tuning the diffusivity requires understanding the details of both the static and dynamic nanoscale structure.

**Madivala G. Basavaraj** opened the discussion on the paper by Ranjini Bandyopadhyay: In your analysis of clay particles you mention that the particles are polydisperse and often exist as stacks. I am curious if particles in each stack are identical. If they are, would it be possible to separate them based on size and obtain monodisperse particles?

**Ranjini Bandyopadhyay** responded: It can be seen from the SEM image in Fig. 1 of our paper that the sodium montmorillonite (Na-MMT) clay particles are polydisperse, irregularly shaped and flexible. Na-MMT tactoids can therefore not be treated as monodisperse stacks. One of the aims of our work was to obtain a clearer understanding of the sedimentation of natural clay gels in the context of geophysical phenomena such as the formation of river deltas. Ordered clay stacks are unlikely in natural clay suspensions and therefore not desirable in our experiments. That said, it must be mentioned that Na-MMT tactoids exfoliate into smaller entities due to the hydration of the interlayer sodium ions which push the negatively-charged particles apart, ultimately resulting in an accelerated exfoliation process. Na-MMT can also be exfoliated by the intercalation of polymer chains which screen the electrostatic interaction between the clay particles, triggering exfoliation into smaller tactoids/stacks and ultimately to single entities. However, this process is extremely sensitive to the concentration of clay in the aqueous suspension.<sup>1</sup> Depending on the sample preparation protocol, there can also be a tactoid-reformation process which results in the tactoids growing in size.<sup>2</sup> Clay tactoids are more likely to eventually exfoliate into single clay particles if the suspension is diluted substantially. Once the tactoids have completely exfoliated, it is presumably possible to use some size segregation technique such as centrifugation to separate clay particles according to size. Complete exfoliation of all tactoids followed by size-separation may therefore be an easier way to size-segregate particles than by first making monodisperse stacks/tactoids followed by exfoliation.

1 S. Ali and R. Bandyopadhyay, *Langmuir*, 2013, **29**, 12663–12669.

2 S. Ali and R. Bandyopadhyay, *Appl. Clay Sci.*, 2015, **203**, 85.

**Yogesh M. Joshi** remarked: Where do the samples studied by you lie on the Na-Montmorillonite–water phase diagram. It is interesting to see that the system studied by you shows sedimentation. Such sedimentation is also observed for the Laponite suspension<sup>1</sup> wherein the average concentration of the clay rich region was independent of Laponite concentration. Do you also observe a similar phenomenon?

1 P. Mongondry, J. F. Tassin and Taco Nicolai, *J. Colloid Interface Sci.*, 2005, **283**, 397–405.

**Ranjini Bandyopadhyay** replied: The concentration of sodium montmorillonite (Na-MMT) in the samples studied in our paper is far below the concentration required to form a soft glassy sample in the absence of salt. Soft glassy behaviour in aqueous Na-MMT suspensions is first observed at 4 wt% in the absence of salt.<sup>1</sup> In our paper, the concentration of Na-MMT in the Na-MMT gels is fixed at 0.8 wt%. Without salt, therefore, these clay suspensions would have remained in a sol state. There are several similarities in the settling behaviour of Na-MMT gels (reported in our paper) and Laponite gels in the presence of salt.<sup>2</sup> In both cases, sedimentation is observed at low clay concentrations, with the settling speed depending on  $C_s$ , the concentration of salt. As mentioned earlier, we are in the low clay concentration ( $C$ ) regime, and explore the structures and sedimentation behaviours of Na-MMT gels over a range of  $C_s$ . We track the location of the settling interface and report that the time duration for the initial quiescent regime (the regime immediately before sedimentation starts) depends strongly on  $C_s$ . The sedimentation behaviour observed in our work is also very sensitive to the preparation protocol, *i.e.* whether salt is added before or after adding clay (SAC and SBC samples in our paper). In their work, Mongondry *et al.* speculate that their results do not rule out particle aggregation in the formation of Laponite gels. This picture agrees very well with our conclusions from the cryo-SEM images of Na-MMT gels (Fig. 11 in our paper). The authors have reported the possibility of gel formation even in samples with extremely low concentrations of clay in the absence of salt. Our paper, in contrast, deals exclusively with the structure and sedimentation of clay gels in the presence of substantial amounts of salt. Mongondry *et al.* plot the reduced scattered light intensity (Fig. 9 in their paper) vs the wavevector  $q$  and define a correlation length  $\xi$ , a characteristic lengthscale for structure formation of the clay suspensions studied by them. They find that for the highest clay concentrations investigated, there is no change in the reduced intensity. It is important to note here that these samples do not contain salt and the authors explain their results in terms of repelling unconnected aggregates or an ordered house-of cards (HoC) structure. In the Na-MMT gels studied by us, however, there is substantial salt in the medium and there are connections between tactoids/exfoliated particles in all the samples (Fig. 11). We systematically tracked the evolution of the clay network structure using these cryo-SEM images and explained the observed sedimentation behaviour in terms of the thermal fluctuation-induced particle rearrangements which modify the local yield stresses of the gel. It is clearly evident from Fig. 11 that in contrast to the work by Mongondry *et al.*, we do not have a well ordered house of cards structure. It is also important to note that the structures that form in our gels are of the order of several micrometers. It is therefore unlikely that if we were to perform systematic scattered intensity measurements for our clay gels, we would see a constant  $q$ -independent intensity as reported by those authors.

1 S. Ali and R. Bandyopadhyay, *Soft Matter*, 2016, **12**, 414–421.

2 P. Mongondry, J. F. Tassin and Taco Nicolai, *J. Colloid Interface Sci.*, 2005, **283**, 397–405.

**Daan Frenkel** commented: You mentioned the use of ultrasonic techniques to probe particle size distribution under conditions where light-scattering cannot be used due to the opacity of the sample. Could you explain how this ultra-sound technique works and indicate when it is (and when it is not) the preferred tool to determine particle-size distributions.

**Ranjini Bandyopadhyay** responded: Ultrasound attenuation spectroscopy (UAS) is a valuable tool to estimate particle size distributions (PSDs) for opaque and non-transparent samples.<sup>1</sup> The technique involves the measurement of attenuation of very low intensity ultrasound passing through a colloidal dispersion. The attenuation coefficient,  $\alpha$ , calculated using the Beer–Lambert law, is expressed in terms of a compression complex wave number,  $k$ . This compression complex wave number,  $k$ , is related to the PSD of the constituents of the dispersion by modelling the attenuation spectrum appropriately and with prior knowledge of the density of the medium, the density of the particulate phase in suspension, the viscosity of the medium at that temperature and the sound speed in the solid medium. In our fits to the acquired attenuation data, only viscous loss was considered. Complete details can be found in one of our publications.<sup>2</sup> We used UAS extensively to calculate the PSD of clay tactoids in aging clay suspensions at clay volume fractions very close to that required for dynamical arrest of the tactoids. In conventional techniques such as DLS, size estimation is performed by relating the decay of the intensity autocorrelation data to the diffusion coefficient which is then related to the particle size using the Stokes Einstein equation. In the volume fraction regime where the suspension is approaching non-ergodic behaviour, the Stokes Einstein equation ceases to be applicable, thereby making the above treatment using UAS extremely powerful in estimating PSDs. However, it is noted here that the PSD estimated using UAS is based on modelling the data appropriately. Some prior knowledge of the sample, as mentioned above, is therefore an essential requirement for successful analysis of the data.

1 A. S. Dukhin and P. J. Goetz, *Characterization of Liquids, Nano-and Microparticulates, and Porous Bodies using Ultrasound*, Elsevier, New York, 2nd edn., 2010.

2 S. Ali and R. Bandyopadhyay, *Langmuir*, 2013, **29**, 12663.

**G V Pavan Kumar** said: Your technique of using ultrasound is interesting. Adding to the discussion, I would like mention that one may possibly utilize light and sound simultaneously to probe the assembly in liquid. An example would be to use the Raman–Nath effect.

**Yogesh M. Joshi** remarked: In Fig. 11, the SEM for the suspension after 138 h shows continuous layers of clays. The lateral dimension of a clay particle is mentioned to be around 400 nm. Could such observation of continuous layers over several microns of lengthscale originate from sintering of the clay particles?

**Ranjini Bandyopadhyay** replied: As Prof. Guruswamy has pointed out, sintering of Laponite or montmorillonite plates is rather unlikely. These minerals are chemically very inert and are not expected to sinter.

**Yogesh M. Joshi** remarked: A suspension of clays such as bentonite, montmorillonite, Laponite, *etc.* when added to water shows a soft solid like consistency and is out of thermodynamic equilibrium. Such out of equilibrium structures are highly sensitive to preparation protocol. The work by Prof. Bandyopadhyay very clearly shows how an apparently trivial matter such as whether salt is added before or after mixing clay has a profound influence on the resultant structure and properties. It appears that being sensitive to preparation protocols is a characteristic feature of many out of equilibrium soft materials. Consequently, a product with soft solid-like consistency is influenced by not just the ingredients but the preparation protocol.

**Madivala G. Basavaraj** asked: What is the correct picture that emerges out of your study on the destabilization of clay dispersions – stable dispersions, then aggregation, followed by settling and then consolidation? How do the results of the sedimentation experiments compare to theories that have been developed for the setting of aggregates?

**Ranjini Bandyopadhyay** replied: The picture that emerges from our experiments can be summarized as follows: when clay is mixed in water with enough salt to partially screen the repulsion between the constituent particles, the clay particles aggregate in ‘overlapping coin’ or ‘house of cards’ configurations or in a combination of both configurations. The time required for stable gel formation is strongly dependent on the concentration of salt in the medium. The gel remains stable for a length of time that depends on the concentration of salt in the medium and the preparation protocol (salt added before clay (SBC) or salt added after clay (SAC)). This initial quiescent regime is followed by a transient/delayed collapse of the gel and a slow compaction of the clay particles into a dense sediment at the bottom of the sample cell. The results are understood in terms of thermal fluctuation-driven particle rearrangements that occur in the gel continuously, modifying the mechanical response of the gel and giving rise to the observed settling behaviour. This scenario has been summarized as a cartoon in Fig. 12 of our paper. Incidentally, the regimes of collapse discussed here have been observed previously in the sedimentation of depletion gels and in colloidal suspensions flocculated at their secondary minimum (references 17–23 of the paper). Using simple theoretical arguments, Manley *et al.* (reference 18 in this paper) showed that delayed sedimentation (or transient collapse) observed in colloidal Ludox gels depends on the comparative values of the gravitational stress on the gel and the network yield stress. The same arguments and physical picture apply to the colloidal sodium montmorillonite (Na-MMT) gels studied here. As speculated by us in the conclusions section of the paper, the observed delayed gel collapse and the subsequent densification could be, in principle, modelled theoretically by using a poroelastic model as a starting point with the clay particle/tactoid association rate  $K_A$  and the dissociation rate  $K_D$  of the clay platelets as input parameters. Unfortunately, given the complex nature of the exfoliation state of Na-MMT, measuring  $K_A$  and  $K_D$  will clearly not be easy. Therefore, even though general features such as the delayed collapse of gels have already been explained theoretically in the literature, modelling other observations such as the differences in settling behaviour in SAC and SBC gels remain a far greater challenge.

**Siddharth Kulkarni** addressed Ranjini Bandyopadhyay and Yogesh M. Joshi: Can the gel formation be predicted by looking at the frequency sweep spectra? Is direct visualization more reliable than the rheology data?

**Ranjini Bandyopadhyay** responded: The frequency sweep spectra of soft glassy materials are characterized by certain ubiquitous features such as the frequency-independent elastic modulus and a weakly frequency-dependent viscous modulus.<sup>1</sup> The rheological signatures of critical gel formation have been discussed by Professor Joshi in paper 8570. In our work, we correlated the honeycomb structures of our clay-in-salt suspensions and their stability behaviours by means of imaging using scanning electron microscopy (SEM) under cryogenic conditions and light transmission experiments. Cryo-SEM allows us to resolve the microscopic details of the structure at very short lengthscales (approximately 100 nanometers), something that bulk rheology measurements could not have achieved. In a previous work,<sup>2</sup> our group found that above a critical salt concentration, there is a microstructural change in clay gels and that the rheological responses of the suspension could be correlated to plate-plate associations of the Laponite particles probed using cryo-SEM. The nature of these plate-plate associations is clearly very important in determining the stability of the honeycomb structures. In the present paper, we systematically investigated the stability of these structures and correlated them with structural details (obtained using cryo-SEM and resolvable down to the scale of 100 nanometers) at a lower clay volume fraction.

1 H. M. Wyss, K. Miyazaki, J. Mattsson, Z. Hu, D. R. Reichman and D. A. Weitz, *Phys. Rev. Lett.*, 2007, **98**, 238303.

2 S. Ali and R. Bandyopadhyay, *Soft Matter*, 2016, **12**, 414.

**Yogesh M. Joshi** replied: The seminal work of Winter and Chambon<sup>1</sup> proposed that in a gel forming system,  $\tan \delta$  is independent of frequency at the critical gel state, such that  $G'$  and  $G''$  show power law dependence on frequency with identical exponent. Consequently, by monitoring evolution of  $G'$  and  $G''$  at different frequencies it is possible to infer the gel formation process. On the other hand scattering techniques such as light scattering and SAXS can also be used to obtain the fractal dimension associated with a gel. Interestingly, the fractal dimension obtained from frequency independent  $\tan \delta$  at the critical gel point matches very well with the one obtained from the scattering studies.

1 H. H. Winter and F. Chambon, *J. Rheol.*, 1986, **30**, 367–382.

**Guruswamy Kumaraswamy** said: Sintering of Laponite or montmorillonite plates is rather unlikely. These are 2 : 1 phyllosilicates with tetrahedral silicate layers that sandwich an octahedral middle layer. These are chemically very inert and are unlikely to sinter. One way to verify that there is no sintering in the aged gel would be to completely dry the gel, crush it into a powder and then redisperse to form a gel again. If the clay has sintered, then the gelation process and the nature of the gel formed after drying will be very different.

**Nirmalya Bachhar** opened the discussion on the paper by E. Eiser: How does the concentration of the NPs affect formation of the structure? Does it change the physical properties of the system which may affect the final application?

**Erika Eiser** replied: While the different ratios of colloids to rods did not lead to large changes in the homogenised structures (also apparent from the chord-distribution analysis) we did observe a much bigger change in structure for the rapidly cooled open structures. Indeed the characteristic lengthscales in the open structure shortened for higher gold-nanoparticle fractions. We argue that it depends what structure you aim for. In the slowly cooled homogenised structures we do not see significant changes.

**Yon Ju-Nam** asked: In paper 8600, the inclusion of thiol groups in the DNA molecules was mentioned as DNA–thiol is required for the ligand exchange carried out in the preparation of the functionalised gold nanoparticles. Could it be possible to know in more detail how these thiol groups are added to the DNA molecule?

**Erika Eiser** responded: While thiolated DNA can be bought readily from IDT technology, we also performed the thiolation of the 5'-phosphate end of the DNA using a scaled up version of a protocol by Zanocco *et al.*<sup>1</sup>.

The latter is advisable if large amounts of thiolated DNA are required.

1 G. T. Hermanson, *Bioconjugate Techniques*, Academic Press, 2nd edition, 2008.

**Sanat Kumar** asked: Do you get ordered phases as seen for cylinder–spherical mixtures in the limit of hard sphere (packing) interactions alone?

**Erika Eiser** replied: The phase diagram of the pure fd-virus solutions has been investigated in great detail by Seth Fraden and Zvonimir Dogic (Brandeis University). Fd-viruses are known to form a cholesteric phase. The Brandeis group has shown that the isotropic to cholesteric transition depends on the added-salt concentration, as the side-coat proteins are negatively charged. Hence, in the limit of high added-salt concentrations the viruses behave like hard rods. We also observe a transition to high-density liquid-crystalline phases. However, in the present work we only consider very low virus-concentrations.

**Charusita Chakravarty** asked: In your earlier work on DNA-colloids, you were able to generate finite aggregates. In this study, you generate open-structured gels. Could you provide some insight into the nature of effective interactions between colloids and mechanisms of self-assembly that lead to these differences?

**Erika Eiser** replied: The main difference in whether one would obtain finite size clusters or a percolating network is the length of the spacer between the colloid and the 'active' or 'sticky' part of the DNA. In our previous work<sup>1</sup> we coated 1 micrometer large polystyrene particles with lambda-DNA, which is about 4800 base pairs long and has a radius of gyration of 800 nm. While one end was attached to the colloids the main double-stranded part of the DNA served as steric stabiliser, and only the last 12 bases were in single stranded form for the binding. The lambda-DNA formed a mushroom-monolayer around the colloids. Because of the length of the dsDNA spacer all available ssDNA on one colloid could bind to all available complementary ssDNA on a different colloid. Hence, all bonds could be saturated, preventing the system from forming a percolating structure. We

showed that by shortening the polymeric spacer below a certain length two colloids with complementary DNA could no longer saturate all their available bonds thus forming percolating networks.<sup>2</sup> The same holds for nano-colloids. In our present study the DNA spacer was always a few nanometers shorter than the colloids used.

1 T. Schmatko, B. Bozorgui, N. Geerts, D. Frenkel, E. Eiser and W. C. K. Poon, *Soft Matter*, 2007, 3, 703.

2 N. Geerts, T. Schmatko and E. Eiser, *Langmuir*, 2008, 24, 5118.

**G V Pavan Kumar** remarked: Your structures may be harnessed for random lasing applications. Could you please comment on this further?

**Erika Eiser** replied: While my group focuses on designing disordered colloidal materials with photonic activity and as porous battery and electrode materials it has been shown that colloidal glasses and dry-pressed powders can act as random lasers. In a classical laser a weakly scattering material is placed in a cavity with two mirrors to provide gain for spontaneous emission. In such a system the cavity determines the frequency and thus the mode of the laser. In a disordered gel or glass made of strongly scattering colloids any incoming light will propagate through multiple scattering – in other words the wavelets undergo a diffusive motion. Letokhov<sup>1</sup> showed that in a diffusive process the gain is proportional to the colloidal volume while the losses are only proportional to the surface. Consequently, under certain circumstances the diffusing light can constructively interfere and produce higher order lasing modes that cannot be found in traditional lasers. How the light is diffusing through a random colloidal material can be influenced by the interaction strength between the colloids, which can be controlled, for instance, by the choice of the DNA attached to the colloids.

An excellent review on random lasers can be found in 'The physics and applications of random lasers' by D. S. Wiersma.<sup>2</sup>

1 V. S. Letokhov, *Zh. Eksp. Teor. Fiz.*, 1967, 53, 1442.

2 D. S. Wiersma, *Nat. Phys.*, 2008, 4, 359–367.

**Vinothan N. Manoharan** said: As I understand, your measurement of the structure factor is done optically. Do you think the gels might have some structure at higher  $q$  (that is, smaller lengthscales) than you are able to probe optically?

**Erika Eiser** replied: The structure factor is obtained *via* a Fourier transform of the microscope image. The maximum  $q$ -range that can be probed is determined by the (lateral) resolution of the microscope and the pixel size of the camera (whichever is largest). In our case, the pixel size corresponds to about 200 nm, which is somewhat smaller than the resolution of the microscope (approx. 500 nm). The observed decay of the structure factor for large  $q$  is therefore determined by the optical resolution of the microscope. With optical, single-photon imaging, it will not be possible to get a better resolution. Hence, although there may well be structure in  $S(q)$  at higher  $q$ , we are unable to probe it. With other techniques (*e.g.* SAXS) it should be possible to see structural features due to the correlation in the positions of the nano-particles.



**Vinothan N. Manoharan** asked further: In your paper, you compare your results for rod–sphere systems to sphere–sphere systems. How do the results compare to rod–rod systems – for example, either systems consisting solely of functionalized virus particles or systems with similar aspect ratios such as carbon nanotubes?

**Erika Eiser** answered: The big difference with rod-like hard particles without functionalisation is that these tend to form bundles. At high enough concentrations, fd-viruses in solution do show liquid crystalline order. Similarly DNA-functionalized viruses show an isotropic to cholesteric transition, when heated above the hybridisation temperature of the DNA attached. At volume fractions below that transition and below the DNA melt temperature, random aggregates of the viruses develop as they bind wherever they meet, displaying a network reminiscent of the cytoskeleton formed by actin filaments in cells. In an earlier publication<sup>1</sup> we imaged such aggregates with AFM, and measured the aggregation dynamics with dynamic light scattering.

1 R. R. Unwin, R. A. Cabanas, T. Yanagishima, T. R. Blower, H. Takahashi, G. P. C. Salmond, J. M. Edwardson, S. Fraden and E. Eiser, *PCCP*, 2015, 17, 8194–8202.

**Radhika Poojari** communicated: What does fd stand for? How can one calculate the number of virions attached to the gold nanoparticles? Can this technique be used in device based applications for biomolecular marker detection in diseases?

**Erika Eiser** communicated in reply: The name fd derives from the fact that it belongs to the filamentous phage fd. It can be genetically modified to produce viruses with different lengths and charge per unit length in a mono disperse fashion making them a great material for studying liquid crystal behaviour.<sup>1,2</sup> As the fd-viruses are only about 6 nm thick it is impossible to calculate the number of gold colloids per vision from micrographs. In our experiments we used ratios of 2.5, 5 and 10 colloids per virus. However, when allowing DNA hybridisation to take place, a gold-nanoparticle can bind several viruses. An important application of filamentous fd is phage display: phages are viruses that invade bacteria to reproduce. One can genetically insert any type of DNA segment into the virus DNA which is then expressed as a protein in the bacterial host. When the virus escapes the host it incorporates the ‘new protein’ in the coat protein, hence the word phage display.<sup>3</sup> But filamentous fd has also been used for piezoelectric energy generation.<sup>4</sup>

1 Z. Dogic and S. Fraden, *Curr. Opin. Colloid Interface Sci.*, 2006, 11, 47.

2 J. Sambrook and D. W. Russell, *Molecular cloning: a laboratory manual*, Cold Spring Harbor Laboratory Press, 3rd edn, 2001.

3 G. P. Smith and V. A. Petrenko, *Chem. Rev.*, 1997, 97, 391.

4 B. Yang Lee, J. Zhang, C. Zueger, W.-J. Chung, S. Y. Yoo, E. Wang, J. Meyer, R. Ramesh and S.-W. Lee, *Nat. Nanotechnol.*, 2012, 7, 351.

**Priyadarshi Roy Chowdhury** communicated: What would be the effect of very low temperature and moderately high temperature on the growth of inhomogeneities, as mentioned in this paper?



**Erika Eiser** communicated in reply: Colloidal gels and crystals driven by DNA binding are different to those formed due to depletion or other chemical interactions. Our structure factor analysis for different quench depths shows that the 'homogenised gels' all have a peak at roughly the same wave vector  $q$ . However, a system quenched into the spinodal decomposition region will show a structural peak position that depends on the depth of the quench. In other words, the peak shifts to smaller  $q$ -values for lower quenches. In view of this observation we can say that, if quenched not too fast, the DNA binding arrests the spinodally decomposing sample so rapidly that the resulting structure is fixed within the first few degrees. Consequently the depth does not change the system structure no matter how deep we quench. This is very different to for instance supercooled colloidal liquids.

However, if quenched much faster than needed for DNA hybridisation, it can happen that the DNA binding will be stuck in strongly out-of-equilibrium traps leading to a more open system similar to a cluster fluid.

**Zakiya Shireen** asked: What would happen if you used different shapes such as ellipsoids or cylinders and how would ellipsoid–ellipsoid or for that matter cylinder–cylinder interactions affect the binding of DNA? How would it be different from that of the rod and sphere interaction?

**Erika Eiser** responded: We have shown that when coating half of the virions with a given single-stranded DNA and the other half with the complementary ssDNA, we obtain random aggregates.<sup>1</sup> For high enough concentrations we expect a continuous, cross-lined network of these semi-flexible polymers similar to the cytoskeleton in cells. It is obvious that also ellipsoids and other shaped colloids can be used. Several techniques have been developed to coat many different materials, whether it is coat proteins for viruses, silica, polystyrene or others. However, different shapes such as ellipsoids may be much more rigid than the semi-flexible rod-viruses. Therefore the resulting disordered gel will have different mechanical properties for similar colloidal volume fractions. If the non-specific van der Waals and Coulomb interactions are suppressed, only the hard shape repulsions and short-ranged DNA attractions are present. So it is only the shape interactions that drive different structure formations.

1 R. R. Unwin, R. A. Cabanas, T. Yanagishima, T. R. Blower, H. Takahashi, G. P. C. Salmond, J. M. Edwardson, S. Fraden and E. Eiser, *PCCP*, 2015, 17, 8194–8202.

**Rajdip Bandyopadhyaya** said: This question is in two parts, and concerns how assemblies behave dynamically as a function of time, with implications in real-world applications:

1. During the formation of assembled structures of nanoparticles (either in a dispersion or in a polymer matrix), can it be deliberately made to happen as a function of time? In other words, can the structure be made to follow an arbitrary time-function, in the form of some premeditated timing?

2. Once formed, can such assemblies or structures be made to respond to changes in the environment? If so, how fast can these assemblies respond to imposed changes in the environment? How strong is the coupling between the assembly and the surrounding? Can such assemblies be made to be adaptive?

**Erika Eiser** responded: In DNA driven aggregation it is important to realise that the process is completely determined by the hybridisation of complementary DNA strands. This process is relatively slow, meaning the two complementary strands need to 'zip-up' correctly. In general only AT or GC base pairs can form in the presence of a monovalent salt. Hence when lowering the temperature of the sample these pairs can form spontaneously, but do not necessarily need to form along the correct position in the sequence. Typical cooling rates for forming correct hybridisation are required to be about 20 s per degree Centigrade. 1. Therefore, if cooled rapidly incorrect hybridisation can take place leading to metastable structures/bonds that will come apart slowly when well below the melt temperature of the sequence used. Thus, very slow arrangements can take place. If cooled along the equilibrium melt curves a much stronger gel can form. To conclude, yes, it is possible to build in a cooling protocol leading to a desired structure. 2. As shown in simulations<sup>1</sup> and in part in experiments (in preparation) DNA can act as a self-healing material because the strands are purely bound *via* hydrogen bonds, which are in essence weak. Consequently, DNA-bound colloidal gels can be sheared and then rejuvenated by heating the sample to around the DNA's melt temperature. Moreover, a gel can be set in any macroscopic shape required.

1 L. Di Michele, D. Fiocco, F. Varrato, S. Sastry, E. Eiser and G. Foffi, *Soft Matter*, 2014, **10**, 3633–3648.

**Nicholas Kotov** opened the discussion on the paper by Andrea R. Tao: In the last slide you showed interesting spectra of the gold dimers that appear to me exciting and intriguing. Could you go through what's going on with the dimers here?

**Andrea Tao** answered: What we observe is that our metal nanocube dimers are strongly coupled when assembled in the face-to-face configurations. However, if and only if light is polarized along the dimer axis do we see the appearance of the coupled mode (in the 700–800 nm window). At angles of incidence where the dimer is not 'visible', *i.e.* light only sees the cross-section of one nanocube, the optical response is almost identical to that of a single cube in a polymer.

**Oleg Gang** said: Since cubes have 3 distinctive plasmonic peaks (related to the length of the edges, and two diagonals), it might be interesting to consider different coupling of those plasmonic modes, depending on how the cubes are oriented relatively to each other.

**Andrea Tao** responded: I agree – we've only been able to achieve a large yield of dimer formation for the face-to-face assemblies but have looked at edge-to-edge arranged cubes for assemblies consisting of a broad distribution of cluster sizes. Near-field localization is definitely stronger in the edge-to-edge assemblies, but the far-field spectra are composed of largely broad features. Corner-to-corner assemblies would require 3D assembly or a controlled particle rotation, which we haven't been able to achieve just yet.

**Vinothan N. Manoharan** commented: An alternative approach to meta-materials that does not require long-range order is the metafluid,<sup>1</sup> which is

a disordered suspension of plasmonic *meta*-atoms. However, making such a material requires fabricating highly symmetric *meta*-atoms with isotropic optical response.

1 Y. A. Urzhumov, G. Shvets, J. Fan, F. Capasso, D. Brandl and P. Nordlander, *Optics Express*, 2007, **15**, 14129.

**Alamgir Karim** replied: Thanks! The concept of metafluids and the paper is indeed very interesting! It would be worthwhile trying to get more of those types of plasmonic *meta*-atoms and study more on their colligative properties in the nanopatterned polymeric matrices of the kind we are working on by a simple soft stamping method.

**Vimala Sridurai** asked: In the reflectance measurements of the *meta*-atoms, the scans are done at 4 degrees and 33 degrees for both horizontal and vertical arrangements.

Why were those two angles chosen and what do you expect in the reflectance peaks for the in between angles?

**Andrea Tao** answered: These two angles were chosen to be near-incidence and oblique angles to measure reflectance. The coupled mode of the dimers occurs in the 700–800 nm range; we see that for angles and polarizations that are incident with the dimer axis, the coupled mode is maximum. We expect to see the maximum reflectance intensity when the cross-section of the dimer is maximized.

**G V Pavan Kumar** commented: Your combination of vertical nanocubes is interesting, and may be harnessed as patch optical antennas. Have you tried performing SERS on them?

**Andrea Tao** responded: Yes, we have. The optical patch antenna works quite well as a SERS substrate, as long as your analyte is located within the optical gap. In some capabilities (*e.g.* if you have no way of binding your analyte to a metal surface), this can be limiting.

**Madivala G. Basavaraj** asked: During your talk you mentioned there is localisation of light in the tiny gap between the particles because of a linker for the case where cubes are attached face-to-face. If this is true, then can one use roughness and achieve similar localisation? That is – can we assemble two cubes attached face-to-face that have a similar gap now due to particle roughness. Would the effect be the same?

**Andrea Tao** responded: Surface roughness can certainly lead to smaller gap sizes, but it may not give you the desired effect of increasing the degree of electromagnetic field localization. The scale of your surface roughness will play a significant role in determining this. It is likely that surface roughness with RMS values.

**Madhura Som** asked: Are there alternate strategies we could use to prevent/slow down lateral diffusion of the nanocubes by engineering the polymer matrix,

perhaps something along the lines of Dr Lynn Walker's work on self-assembled block copolymer micelle solutions?

**Andrea Tao** answered: I think confinement strategies for dictating nanoparticle diffusion would be fascinating, if we could engineer particles to avoid getting pinned at interfaces and still undergo assembly.

**Vinothan N. Manoharan** addressed Andrea Tao and Erika Eiser: Prof. Tao and Dr Eiser, in both of your papers you describe methods to control the structure of nanoparticle assemblies, with applications toward optical materials. Are there applications in which it would be important to control the dynamics of the assemblies – that is, how the structure changes over time?

**Erika Eiser** responded: The structures that we present are intrinsically non-equilibrium structures. Hence the control of the kinetics is essential for the design of such materials for specific applications.

**Andrea Tao** replied: In the case of optical materials, potential applications could range from a simple color-based detection scheme where the material changes color upon the presence of a particular chemical species, to optical coatings that dynamically block out particular wavelengths of light. In more advanced optics, one could potentially think of designing a dynamic material for routing or switching capabilities. But such applications are inherently limited by the slow kinetics of nanoparticle assembly.

**Rajdip Bandyopadhyaya** commented: This is a follow-up on the earlier question of how nanoparticle assemblies behave as a function of time and how that couples the system (particle assembly) to the environment, when such assembly timescales are real-world timescales of minutes in an actual application scenario.

Now the question is that, real-world applications of such pre-programmed structures are supposed to realize some machine-like functions. The problem can come from two sources:

(a) Different components of such a machine (where each component can be a different kind of particle-assembly realizing different functionalities) can interact in an unpredictable manner because of undefined functions in some parameter-space (size, shape, composition *etc.*) of these assemblies.

(b) More importantly, a real-world environment will have many external random noise sources, which will impact how different assemblies interact with each other in an unpredictable way.

So is there a way to make each individual assembled structure in such a way to build-in many redundancies to make the overall collection of assemblies a robust system? All this could lead to a more deterministic response from an inherently mesoscopic-scale assembly of a few hundred nanoparticles.

**Lola Gonzalez-Garcia** questioned: Transparent conductive materials are gaining interest for the fabrication of devices such as solar cells, OLEDs or touch screens. The use of metal nanoparticle networks is a promising approach, however, a compromise between transparency and conductivity should be achieved. 1. Would it be realistic to use a self-assembly strategy to lower the

percolation threshold to get materials with percolating networks as their overall structure?

2. In this case, would it also be possible to address the problems that ligands cause, acting as insulating barriers, for electron transfer between the metal nanoparticles?

**Andrea Tao** replied: Self-assembly strategies could indeed be used to increase to degree of network formation in similar nanowire or nanoparticle films. One could imagine using this to design ultrathin metallic networks with a specific density of junctions or a specific transport pathway. There are a number of researchers and companies who are actively using colloidal metal nanostructures to address the fabrication of transparent conductive materials. Most of those that I am aware of use metal nanowires, where the nanowires are laid down in a mesh-like film and then welded together using heat or photoactivation. This certainly addresses the difficulties caused by ligands, which could indeed serve as potential barriers for electron conduction between particles or wires.

**Erika Eiser** answered: Our rod-sphere network demonstrates that one can achieve very low volume fraction samples to achieve percolating networks that might indeed form transparent electrodes. Although the fd-viruses are not conducting, and the DNA-bridges may present a resistance to electric conductivity it has been shown before that a DNA bridge can be metalized. The fd-viruses may be replaced by conducting carbon-nanotubes. Hence, it is possible to use these systems as transparent electrode materials. We are presently working on it.

Eventually, DNA may be exchanged by conducting host-guest molecules or ligands, which can be synthesised on a largescale.

**Daan Frenkel** opened the discussion on the concluding remarks by Oleg Gang: The possibilities of designing truly complex self-assembling structures is expanding rapidly. Undoubtedly, there will be important applications in the (not too distant future).

However, it is useful to make our aims more explicit. At this stage, we should formulate a few specific 'holy grails': the design of self-assembling materials or devices that can perform functions that cannot be carried out with any existing material. I am thinking of objectives that could inspire the field on a timescale of, say, five years.

**Nicholas Kotov** replied: My 'holy grail' would be self-replicating nanoparticles. The reason is that this is a system that would help us make quite a few fundamental discoveries. Technological applications of such nanoparticles in biomimetic self-healing catalytic systems, can be thought about as well. Enantioselective self-assembly of helices from nanoparticles under circularly polarized light indicates to me that the origin of homochirality on Earth might be related to self-organization of nanoscale inorganic matter. So, tracing potential mechanisms for how the self-assembly of inorganic nanoparticles can take place at the early age of Earth (*i.e.* Hadean eon) would be my second 'holy grail'.

**Nicholas Kotov** commented further: Replication of some functions of a microbe would be my other choice. Besides being enigmatic and, to some

degree, academically provocative, this Holy Grail should in nanoparticle assembly lead to multiple technological advances in catalysis and energy harvesting.

**Oleg Gang** responded: I fully agree that some kind of functional, autonomic and self-sufficient small machinery, like a microbe (but not a real one), would be an excellent demonstration of our capabilities in assembly. But we all know it is still far off. We should not try to just make a copy of what works in biology, but should exploit the new properties of nanoparticles. An airplane is inspired by a bird, but it does not duplicate it.

**Daan Frenkel** said: To give a rough indication of something to aim for: we should try to go beyond 'conventional' smart materials that exhibit one-to-one stimulus-response behaviour. Rather, we could consider materials that perform simple 'logical' operations that allow them to respond in a non-trivial (and certainly non-linear) way to the change in a number of independent external parameters. Mimicking living systems should not be the first objective.

**Nicholas Kotov** responded: It seems to me that non-trivial and non-linear system behavior is typical of living cells, isn't it?

**Madivala G. Basavaraj** commented: Over the years there have been exciting ways to assemble nanoparticles and colloids into a range of suprastructures. These assemblies have been put into use for example as gas sensors. One of the challenges in the future would be the sustainable reuse of these assemblies and their application in the real world where there may be dirt and other conditions that can either disrupt or hamper the functionality of these assemblies.

**Oleg Gang** responded: I agree, for any real-life application usually some "dirt" discrimination mechanism/process is needed.

**Daan Frenkel** remarked: Many potential applications of complex self-assembly are in chemistry. A nano-structure could be used to implement a multi-step reaction. There are already examples where DNA origami structures can assist in the sequential assembly of complex hetero-polymers (or oligomers). However, the future nano-scale chemical factories need not be DNA-based.

**Nirmalya Bachhar** questioned: Based on the entire discussion can we yet come to a conclusion on which lengthscale or timescale we should avoid using coarse grained models like DLVO theory? Can there be any rule of thumb for that beside the existing limitations it already has?

**Nicholas Kotov** answered: In my estimates 20 nm (particle diameter and the interparticle distance) would be a good rule of thumb when non-additivity of particle interactions starts to strongly affect their behavior.

**Oleg Gang** replied: I think it is highly system specific, moreover, it might be specific to the questions one asks about the systems.

**Guruswamy Kumaraswamy** addressed Daan Frenkel and Oleg Gang: What is the state-of-the-art in using self-assembly techniques to realize photonic meta-materials in the visible region? For example, is it currently possible to realize periodic assemblies of nanoscale horseshoe-type metal resonators (or metal nanoparticle assemblies with broken symmetry) over macroscopic distances of, say, 1 mm? If this hasn't yet been achieved, do you feel that this is a sufficiently challenging target for the next couple of years?

**Oleg Gang** answered: As far as I know, no assembly of nanoscale resonators has been achieved even for smaller areas. It does require not only resonator positioning, but also the fabrication and control of their relative orientations. It can be a very good targeted structure. However, it will be important to have optical calculations to support and guide the fabrication of such specific structures.

**Daan Frenkel** replied: As I am not an expert on photonic *meta*-materials, I do not feel that I can confidently assess the state of the art. I will therefore focus on what DNA-brick structures (or their successors) could enable. As our ability increases to control the growth of periodically repeated DNA-brick structures, we should hopefully be able to make millimetre-sized crystals (but crystal growth is an art). However, once such structures can be made, they can presumably be functionalized with resonators or other components, that can be placed with high precision in non-trivial, functional arrangements (not necessarily photonic).

**G V Pavan Kumar** questioned: The self assembly of nanoparticles has already made an impact on nano-optics and nano-spectroscopy by providing model systems such as plasmonic dimers with gaps less than 10 nm. The prospects of realizing optical metamaterials using self-assembly of nanoparticles is very interesting. I think it can open up new avenues in nanophotonics.

**Vinothan N. Manoharan** answered: I agree. In nanophotonics, self-assembly might actually be a more precise fabrication method than top-down techniques such as e-beam lithography. As an example, self-assembled monolayers can control the gap distance between particles to nanometer precision or better.

**Mukta Tripathy** addressed Vinothan N. Manoharan: During the introductory lecture by Steve Granick, Vinothan Manoharan suggested that we begin to think about time-anisotropy in order to build new functionalities. There are many systems where timescales can be separated, because of the occurrence of both fast and slow processes, such as glassy particles rattling in a cage at short times, and then 'hopping' out of a cage. However, the effects of entropy will dominate at long times, and perhaps to produce a functional material that switches between short-time and long-time behavior, we might have to think of energy-harvesting active matter.

I would like to know more about any specific directions of thought Vinothan has had, or any specific systems he has in mind.

**Vinothan N. Manoharan** responded: This is a good point. I had in mind materials made from nanoparticles whose interactions change in time. This can be done if the interactions are mediated by functional groups on the particles

such as DNA strands. I agree that to make time-anisotropic interactions that are different from what one might observe in a glassy dynamical process, one should focus on active systems (such as dynamic DNA nanotechnology), where there is an energy input.

**Sanat Kumar** asked **Oleg Gang**: You mentioned error correction and healing as being important parts of self-assembly but you didn't discuss them in detail in your talk. What do we know now? And beyond what we can think about at the moment do we need to achieve these goals?

**Nicholas Kotov** further commented: DNA assembly of gold nanoparticles impresses me with the diversity of structures that could be created and complexity of nanoparticle encoding that could be two or three levels deep. When the superstructure starts containing a very large number of particle errors, particle-particle attachment becomes a limiting factor. What is your thinking about this? How could we prevent or 'anneal' them?

**Priyadarshi Roy Chowdhury** communicated: Could you discuss the necessity of structural error correction associated with complex self assembly?

**Oleg Gang** answered: We can envision constructing those thermodynamically favorable pathways that allow for more efficient error correction, as for example demonstrated in Prof. Frenkel's talk. Also, depending on the system details, it is possible to consider more active error correction mechanisms. Again, they might just be thermodynamically driven, but might use particles or molecules that diffuse freely in the self-assembled matrix. We have certain examples of such realizations for nanoparticle systems, but they are only in the initial stages.

**Priyadarshi Roy Chowdhury** communicated: Could you discuss the contrasting properties of nanoparticle assembly, supramolecular assembly and colloids.

**Oleg Gang** communicated in reply: The most important differences are in the effects of diffusion, interactions, shapes and effect of temperature on the assembly behavior. For example, molecular shapes are significantly more complex than colloidal ones, as a result various unusual packings can be accommodated.

**Priyadarshi Roy Chowdhury** communicated: Could you discuss the criteria responsible for reorganization of assemblies.

**Oleg Gang** communicated in reply: One needs to establish the conditions for shape- or interaction-shifting

**Priyadarshi Roy Chowdhury** communicated: What are the effects of co-ordination number on nucleation in many component self assembly?

**Oleg Gang** communicated in reply: The answer is not clear at the present time since we know very little about those systems, in particularly from the experimental point of view.



**Priyadarshi Roy Chowdhury** communicated: What are the effects of colloids undergoing colored interactions?

**Oleg Gang** communicated in reply: We witness only the beginning of those studies. I believe there are many arrested states for those systems, and we will need to investigate to what degree they can hamper the structure formation, and what energy landscape can help to overcome them.