

Liquid Phase Transmission Electron Microscopy Visualization of Surface Pattern Formation during Chemical Reaction Driven Assembly of Nanoparticles

Thilini U. Dissanayake¹ and Taylor J. Woehl¹

¹Department of Chemical and Biomolecular Engineering, University of Maryland, College Park, MD, USA

Liquid phase transmission electron microscopy (LPTEM) has emerged as an important tool for visualizing nanoscale objects ranging from inorganic nanoparticles to biomolecules in the liquid phase in real time with nanometer spatial resolution. In particular, LPTEM imaging of the dynamics involved in nanoparticle self-assembly has revealed a number of surprising insights into assembly mechanisms. Prior LPTEM experiments have observed interesting nanoparticle structures and their assembly dynamics, such as chain-like assemblies [1], branched nanoparticle networks [2], nanoparticle superlattices [3], and nanoprism hexagonal lattices [4]. However, these prior studies have utilized conventional colloidal forces to drive self-assembly, such as Van der Waals and electrostatic forces, and have relied on assembling nanoparticles near equilibrium. On the other hand, cells utilize chemical reactions to maintain assembly processes far from equilibrium, which gives them unprecedented spatial and temporal control over the assembly of nanoscale biomolecular structures [5]. Use of chemical reactions to drive assembly of synthetic nanoparticles will open new avenues to assembling more complex and dynamic patterns and nanostructures, but first the complex nanoscale dynamics of these processes must be understood.

In this study we used conjugation chemistry to anchor gold nanoparticles to silicon nitride membrane as well as to modulate the surface hydrophobicity of particles during assembly, which was initiated and observed with LPTEM. Gold nanoparticles were deposited on silicon nitride membrane and imaged in STEM mode after introducing a carbodiimide chemical fuel to the liquid cell that rendered nanoparticle surfaces hydrophobic. For dose rates over 150 MGy/s, nanoparticles assembled into small (~10-15 particles) 2D clusters in less than 1 minute (Figure 1a). Interestingly, at a magnification of 500,000 x and decreased dose rate of 80 MGy/s, time-lapsed LPTEM imaging showed that the nanoparticles first assembled into small clusters, followed by formation of horizontal strings of nanoparticles, evenly spaced by ~100 nm and parallel to the STEM scan direction (Figure 1b). Rotation of the STEM scan by 90° produced the same result. Systematic variation of dose rate by changing beam current and magnification revealed a narrow range of electron dose rates for making these nanoparticle strings and no dependence on beam current. In the absence of the carbodiimide agent, the particles underwent rapid assembly into chain-like structures (< 30 s) followed by ejection from the image region, emphasizing the importance of the carbodiimide agent to anchor the nanoparticles to silicon nitride membrane and allow pattern formation. At 80 MGy/s, 500,000 x and half the carbodiimide fuel concentration, elongated, hexagonally ordered 2D nanoparticle clusters formed after 5 minutes of continuous imaging due to attractive interparticle hydrophobic forces (Figure 1c). Prolonged imaging didn't lead to string formation, but the ends of the 2D aggregates folded into 3D structures due to hydrophobic interactions. Surprisingly, at a comparatively low dose rate and magnification (23.4 MGy/s, ×300,000 x) we observed distinct spot patterns of nanoparticle clusters, similar to Turing patterns seen on leopards and fish (Figure 1d). Under these conditions, we observed distinct changes in the background intensity occurred concurrent with nanoparticle assembly (Figure 1 b,d). In the first case with stripes (Figure 1c), a set of ~50 nm thick horizontal bands parallel to nanoparticle stripes were observed. In the second case, we observed diffuse,

segregated dark spots with sizes of 50 – 100 nm on which nanoparticles congregated. The background intensity patterns reflected the local surface concentration of siloxane polymer, which reacts with oxidizing hydroxyl radicals in water produced by electron beam irradiation [6]. We posit that competing crosslinking and chain scission reactions led to fluctuations in the local siloxane concentration, which caused a reaction-diffusion instability leading to stable geometric pattern formation. Due to the presence of a chemical coupling agent between the polymer and nanoparticles, the nanoparticles adopted a similar pattern as the underlying polymer layer. Further evidence for the formation of siloxane rich domains was provided by HAADF-STEM images of the patterned area after removing the chips from the liquid cell and drying (Figure 2a). Nanoparticle patterns did not form when a hydroxyl radical scavenger, tert-butanol, was introduced, suggesting hydroxyl radical induced polymerization and depolymerization play important roles in forming the nanoparticle and polymer surface patterns (Figure 2b). This study has uncovered a new direction for using LPTEM as a tool for creation of nanoscale patterns without the need for surface templated or templating of the electron beam [7].

References:

- [1] Woehl, T. J. and Prozorov, T., *J. Phys. Chem. C*, **119** (36) (2015), p. 21261–21269.
 [2] Lin, G., Chee, S. W., Raj, S., *et al.*, *ACS Nano*, **10** (8) (2016), p. 7443–7450.
 [3] Park, J., Zheng, H., Lee, W. L., *et al.*, *ACS Nano*, **6** (3) (2012), p. 2078 – 2085.
 [4] Ou, Z., Wang, Z., Luo, B., *et al.*, *Nat. Mat.*, **19** (2020), p. 450 – 455.
 [5] Hess, H. & Ross, J.L., *Chem. Soc. Rev.*, **46** (2017), p. 5570 – 5587.
 [6] Woehl, T. J. and Abellan, P., *J. Microscopy*, **265**(2) (2016), p. 135–147.
 [7] We acknowledge funding from the Army Research Office (Grant #W911NF-20-1-0169).

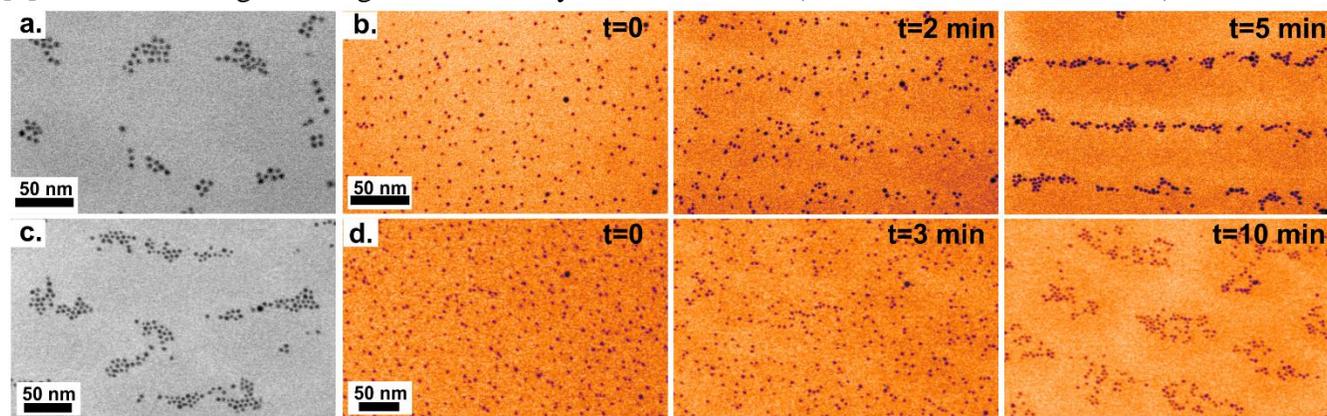


Figure 1. (a) LPTEM image of small clusters formed at carbodiimide concentration of 5 mM ($M = 800,000 \times$, $D = 150 \text{ MGy/s}$) and (b) Time-lapsed LPTEM images of NP stripe formation at $M = 500,000 \times$, $D = 80 \text{ MGy/s}$. (c) Aggregates formed at reduced carbodiimide concentration of 2.5 mM ($M = 500,000 \times$, $D = 80 \text{ MGy/s}$) and (d) Nanoparticle cluster spot patterns formed at $M = 300,000 \times$, $D = 23.4 \text{ MGy/s}$. (b) and (d) are false colored to show underlying polymer layers in darker orange color.

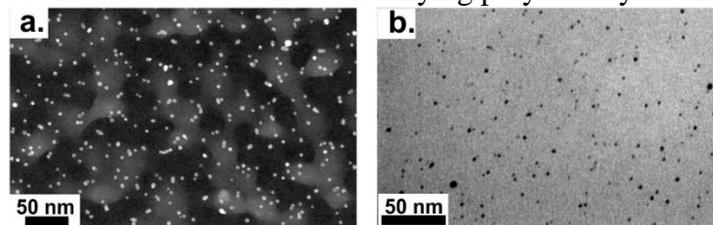


Figure 2. (a) HAADF-STEM image of an irradiated area in LPTEM showing regions of siloxane polymers formed by electron beam polymerization and chain scission. (b) Nanoparticle patterns did not form after 10 minutes of irradiation at $M = 500,000 \times$, $D = 80 \text{ MGy/s}$ in the presence of 20 vol% tert-butanol.