Characterization of the Nanostructure of Complexes Formed by a

Redox-Active Cationic Lipid and DNA

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Supporting Information

SANS of BFDMA and DNA Complexes

SANS spectra obtained using complexes formed between reduced BFDMA and DNA at charge ratios of 2.75:1 and 4:1 are shown in Figure S1. Similar to the SANS spectrum obtained at a charge ratio of 1.1:1 (Figure 4), the spectra obtained at charge ratios of 2.75:1 and 4:1 also show a Bragg peak at $1.2nm^{-1}$. The intensity of the peak seen in the SANS spectra varies with charge ratio. Because samples with a charge ratio of 1.1:1 precipitated more than those prepared with a charge ratio of 4:1, we attribute this variation to a local change in solution concentration of BFDMA due to precipitation.

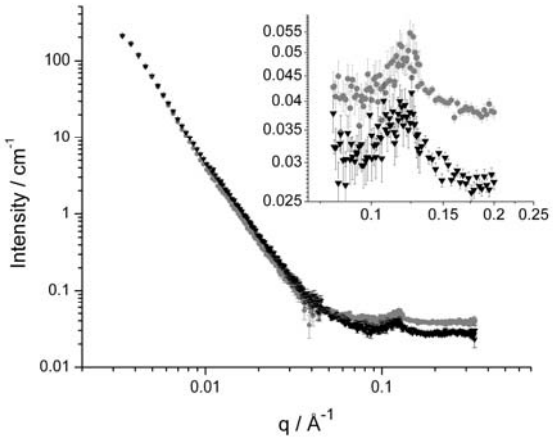

Figure S1. SANS spectra obtained using reduced BFDMA (1mM) in the presence of DNA at charge ratios of 2.75:1 (grey) and 4:1 (black). The inset shows the peak observed at 1.2nm^{-1}.

Cryogenic Transmission Electron Microscopy and Freeze-Fracture-Replication

Additional cryo-TEM images of complexes formed from reduced BFDMA and DNA at a charge ratio of 2.75:1 are shown in Figure S2. Consistent with the SANS measurements, the cryo-TEM images suggest that no significant change in the nanostructure of complexes formed between reduced BFDMA and DNA occurs when the charge ratio is varied from 1.1 to 2.75.

Figures S3, S4, and S5, show images of aggregates formed from oxidized BFDMA and DNA at charge ratios of 3.3, 8.25, and 12, respectively. To obtain panel C of Figure S3, we used freeze-fracture-replication (FFR) which allows the study (at relatively high resolution) of micron-size objects, or very viscous systems that cannot be made into thin liquid films. The technique involves freezing a small volume of the liquid, fracturing, and replication of the fracture surfaces by deposition at an angle ("shadowing") of a thin layer of a heavy metal, such as platinum, followed by deposition

of a carbon layer for mechanical stability. The specimen is melted or dissolved, the replica is cleaned, mounted on a TEM grid, dried, and examined by TEM at room temperature. A recent example of using direct-imaging cryo-TEM and FFR as complementary techniques is given by Ruthstein *et al.*[1]

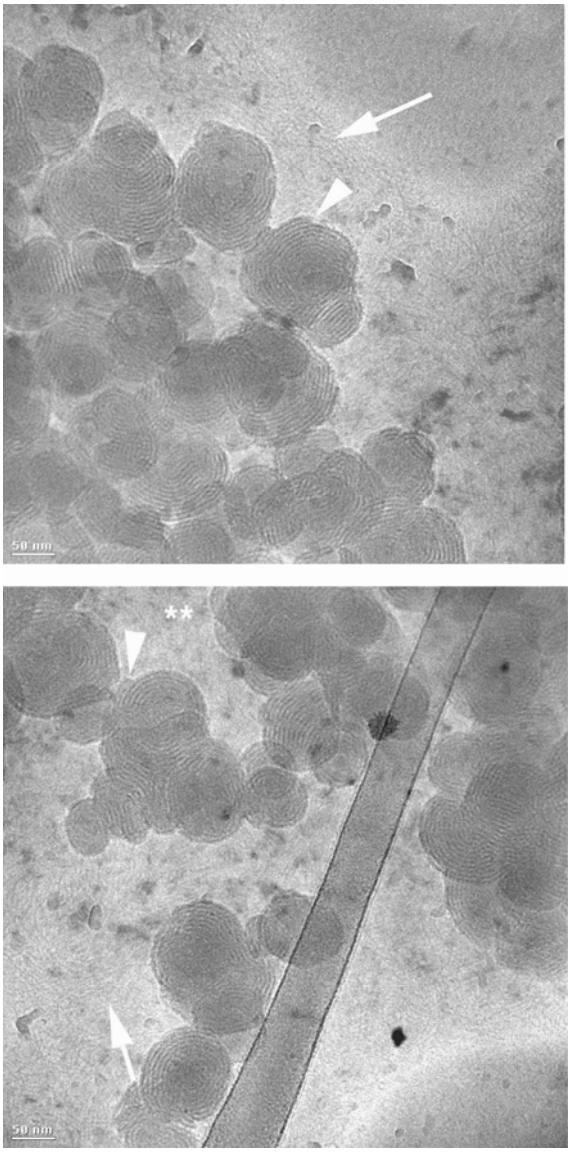

Figure S2. Cryo-TEM images of complexes formed by reduced BFDMA (20μM) in the presence of DNA (2.4ug/ml) at a charge ratio of 2.75 showing a multilamellar structure. The arrowhead shows a broken outer bilayer and the arrow indicates free DNA.

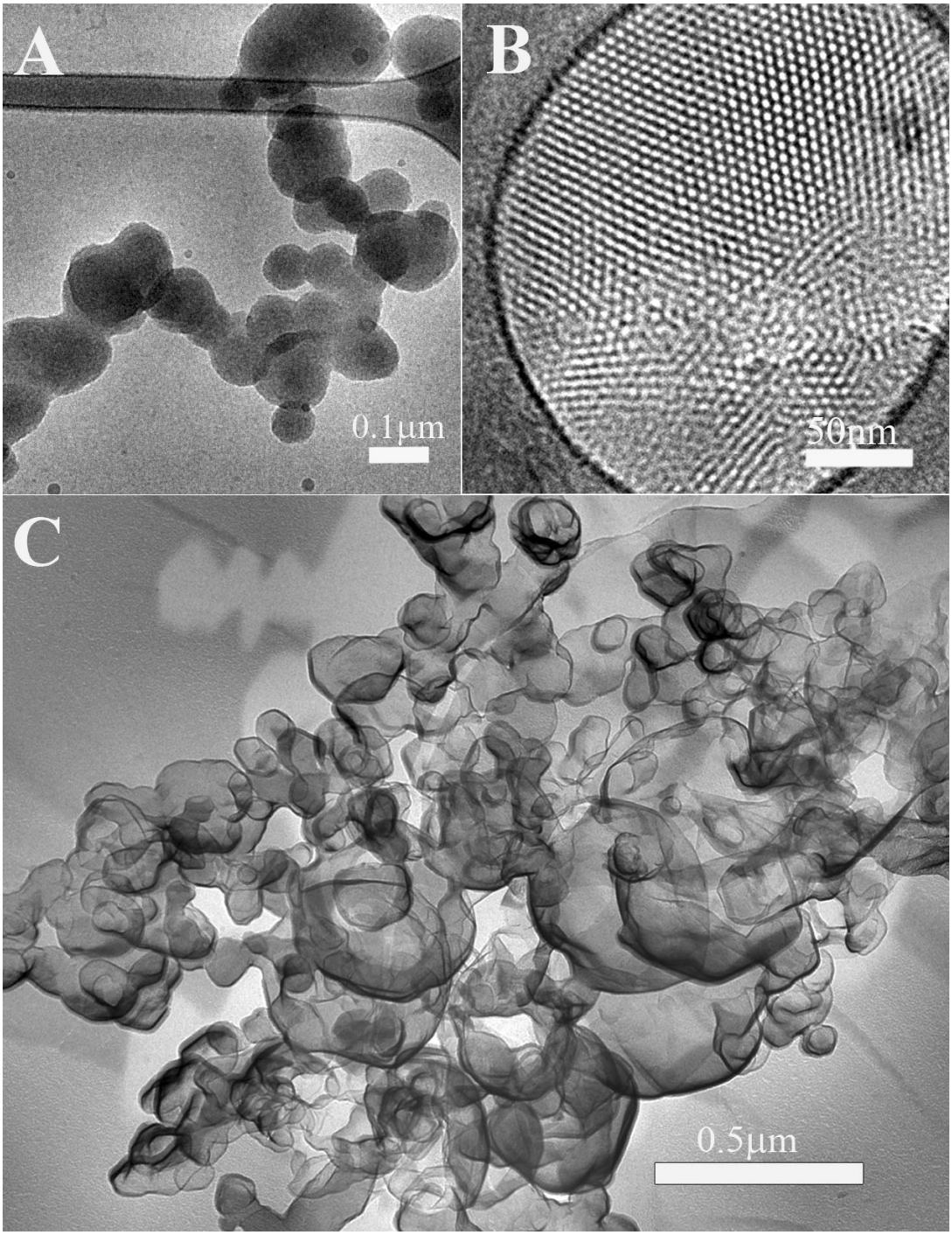

Figure S3. Cryo-TEM images of complexes formed between oxidized BFDMA (0.769 mM) and DNA (231 μg/mL) at a charge ratio of 3.3: A. Disordered aggregates; B. Ordered cubic phase; C. Freeze-fracture-replica of a large disordered aggregate that is too large to be observed by direct imaging cryo-TEM.

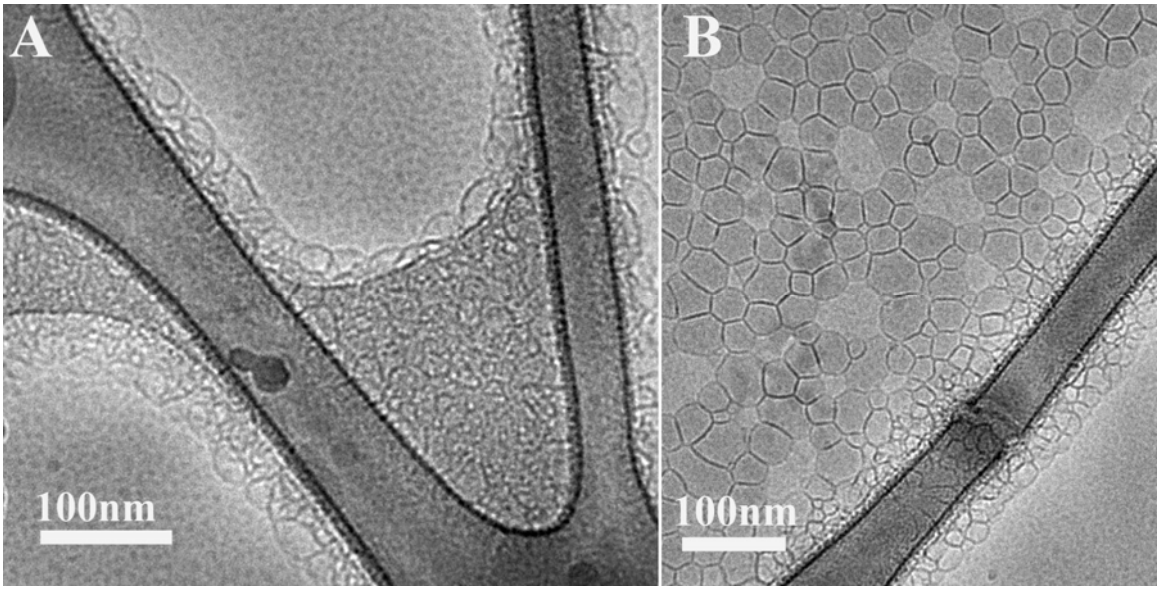

Figure S4. Cryo-TEM images of complexes formed between oxidized BFDMA (0.893 mM) and DNA (107 µg/mL) at a charge ratio of 8.25: A. Sponge-like phase adhering to the support film; B. A non-continuous cluster of fused vesicles.

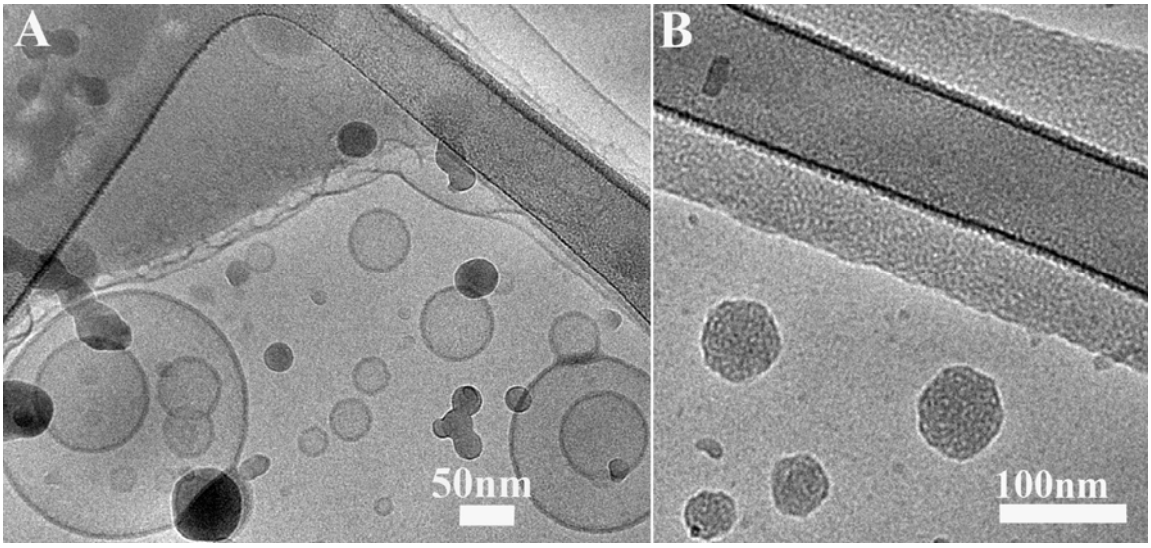

Figure S5. Cryo-TEM images of complexes formed between oxidized BFDMA (0.924 mM) and DNA (76 µg/mL) at a charge ratio of 12: A. Sponge-like phase domain adhering to the support film and vesicles; B. Small particles and larger domains of a sponge-like phase.

(1) Ruthstein, S.; Schmidt, J.; Kesselman, E.; Talmon, Y.; Goldfarb, D. *Journal of the American Chemical Society* **2006**, *128*, 3366.