UNIVERSITY OF TORONTO
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Cryo-TEM Imaging Report of Liposomal Vitamin C Effervescent Powder provided by Aurora Nutrascience Inc.

Sample: Liposomal Vitamin C Effervescent Powder

Method: Before analysis, open a sealed packet and add content into a bottle containing 500 mL of reverse osmosis water, shake the bottle, wait until foaming and fizzing complete to avoid interference from micro bubbles created by effervescence.

The liquid sample was then vitrified in liquid ethane by pipetting onto Quantifoil R2/2 copper grids (EMS) that had been glow discharged in air (Pelco easiGlow, Ted Pella, Inc.) and blotting using a Vitrobot Mark IV (Thermo Scientific). Grids were transferred into a single tilt cryo TEM holder (Gatan). Grids were imaged with a Talos L120C TEM (Thermo Scientific) using a high tension of 120 kV with a 4k x 4k BM-Ceta CMOS camera. Images were taken at magnifications 57,000x yielding a pixel size of 249 pm.

Results: The morphology of the liposomes varied, with both individual single layer formation, and agglomerates of multiple sphere structures in agglomerates and bilayers. The liposomal sphere formations show signs of internal materials coacervated in the structure which demonstrated dappling and signs of an encapsulated vitamin compound, captured in the liposomal spheres.
Overall, the majority of the liposomes organize in clusters that show stacks of amorphous, alternating lipid bilayers that form liposomal spheres.

Liposomal Size: The size range of liposomes found in our analysis was between 20nm and 500nm.

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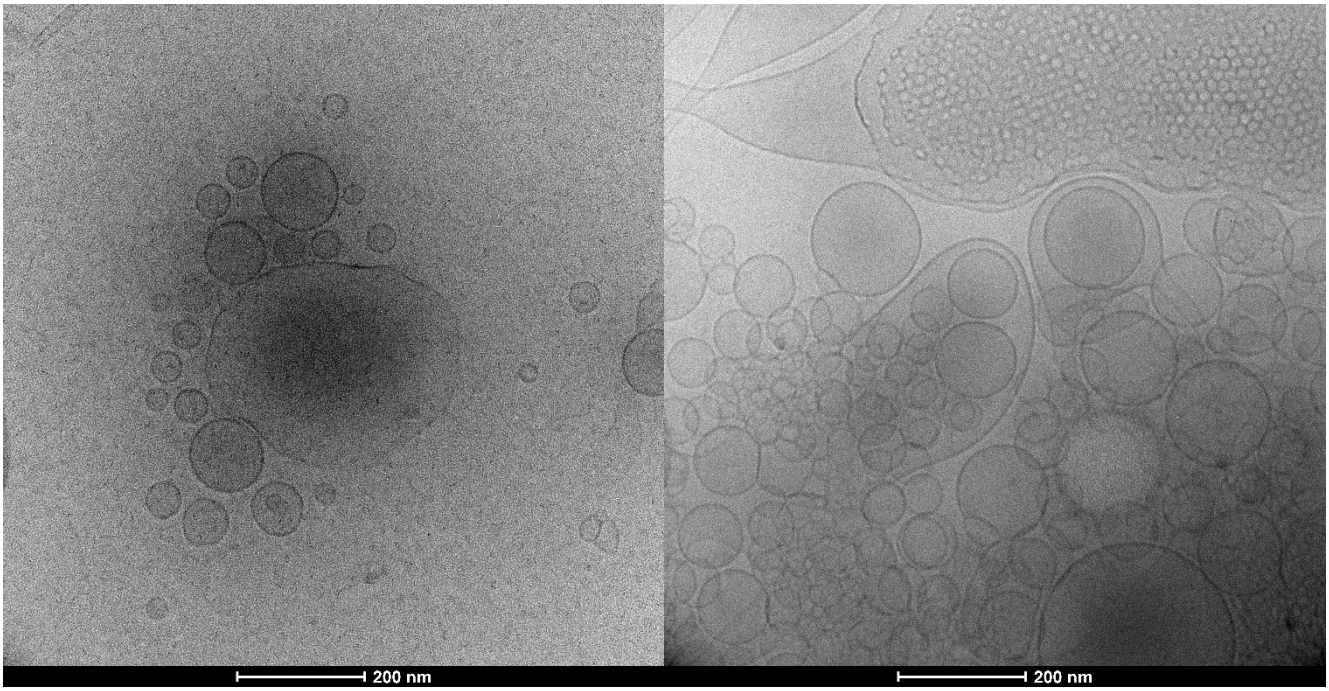

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