Microfluidic Directed Self-Assembly of Soft Matter Nanoparticles

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Abstract:
We investigate the directed self-assembly of nanoscale lipid vesicles by microfluidic mixing [1], the application of these nanoscale vesicles as templates for liposome-hydrogel hybrid nanoparticles [2], and the characterization of single molecule encapsulation within these nanoscale vesicles [3].

Summary of Research:
We investigate the formation of unilamellar lipid vesicles (liposomes) with diameters of tens of nanometers by COntrolled Microfluidic Mixing And Nanoparticle Determination (COMMAND) [1]. Our study includes liposome synthesis experiments and numerical modeling of our microfluidic implementation of the batch solvent injection method. We consider microfluidic liposome formation from the perspective of fluid interfaces and convective-diffusive mixing, as we find that bulk fluid flow parameters including hydrodynamically focused alcohol stream width, final alcohol concentration, and shear stress do not primarily determine the vesicle formation process.

Microfluidic device geometry in conjunction with hydrodynamic flow focusing strongly influences vesicle size distributions, providing a coarse method to control liposome size, while total flow rate allows fine-tuning the vesicle size in certain focusing regimes. Although microfluidic liposome synthesis is relatively simple to implement experimentally, numerical simulations of the mixing process reveal a complex system of fluid flow and mass transfer determining the formation of non-equilibrium vesicles. These results expand our understanding of the microfluidic environment that controls liposome self-assembly and yield several technological advances for the on-chip synthesis of nanoscale lipid vesicles.

We present a microfluidic method to direct the self-assembly of temperature-sensitive liposome-hydrogel hybrid nanoparticles [2]. Our approach yields nanoparticles with structural properties and highly monodisperse size distributions precisely controlled across a broad range relevant to the targeted delivery and controlled release of encapsulated therapeutic agents. We used microfluidic hydrodynamic focusing to control the convective-diffusive mixing of two miscible nanoparticle precursor solutions (a DPPC:cholesterol:DCP phospholipid formulation in isopropanol, and a photopolymerizable N-isopropylacrylamide mixture in aqueous buffer) to form nanoscale lipid vesicles with encapsulated hydrogel precursors.

These precursor nanoparticles were collected off-chip and were irradiated with ultraviolet (UV) light in bulk to polymerize the nanoparticle interiors into hydrogel cores. Multi-angle laser light scattering in conjunction with asymmetric flow field-flow fractionation was used to...
characterize nanoparticle size distributions, which spanned the ≈ 150 nm to ≈ 300 nm diameter range as controlled by microfluidic mixing conditions, with a polydispersity of ≈ 3% to ≈ 5% (relative standard deviation).

Transmission electron microscopy was then used to confirm the spherical shape and core-shell composition of the hybrid nanoparticles. This method may be extended to the directed self-assembly of other similar cross-linked hybrid nanoparticle systems with engineered size/structure-function relationships for practical use in healthcare and life science applications.

We present a non-destructive method to accurately characterize low analyte concentrations (0-10 molecules) in nanometer-scale lipid vesicles [3]. Our approach is based on the application of fluorescence fluctuation analysis (FFA) and multi-angle laser light scattering (MALLS) in conjunction with asymmetric field flow fractionation (AFFF) to measure the entrapment efficiency (the ratio of the concentration of encapsulated dye to the initial bulk concentration) of an ensemble of liposomes with an average diameter less than 100 nm.

Water-soluble sulforhodamine B (SRB) was loaded into the aqueous interior of nanoscale liposomes synthesized in a microfluidic device. A confocal microscope was used to detect a laser-induced fluorescence signal resulting from both encapsulated and unencapsulated SRB molecules. The first two cumulants of this signal along with the autocorrelation function (ACF) were used to quantify liposome entrapment efficiency.

Our analysis moves beyond typical, non-physical assumptions of equal liposome size and brightness. These advances are essential for characterizing liposomes in the single molecule encapsulation regime. Our work has further analytical impact because it could increase the interrogation time of free solution molecular analysis by an order of magnitude and form the basis for the development of liposome standard reference materials.

References: