Electronic Supplementary Material (ESI) for Soft Matter.

Formation and Properties of Liposome-Stabilized All-Aqueous Emulsions Based on PEG/Dextran, PEG/Ficoll, and PEG/Sulfate Aqueous Biphasic Systems

Andrew T. Rowland and Christine D. Keating*

**Department of Chemistry, Pennsylvania State University, University Park, Pennsylvania 16802, USA. E-mail: keating@chem.psu.edu*

Figures in Supporting information

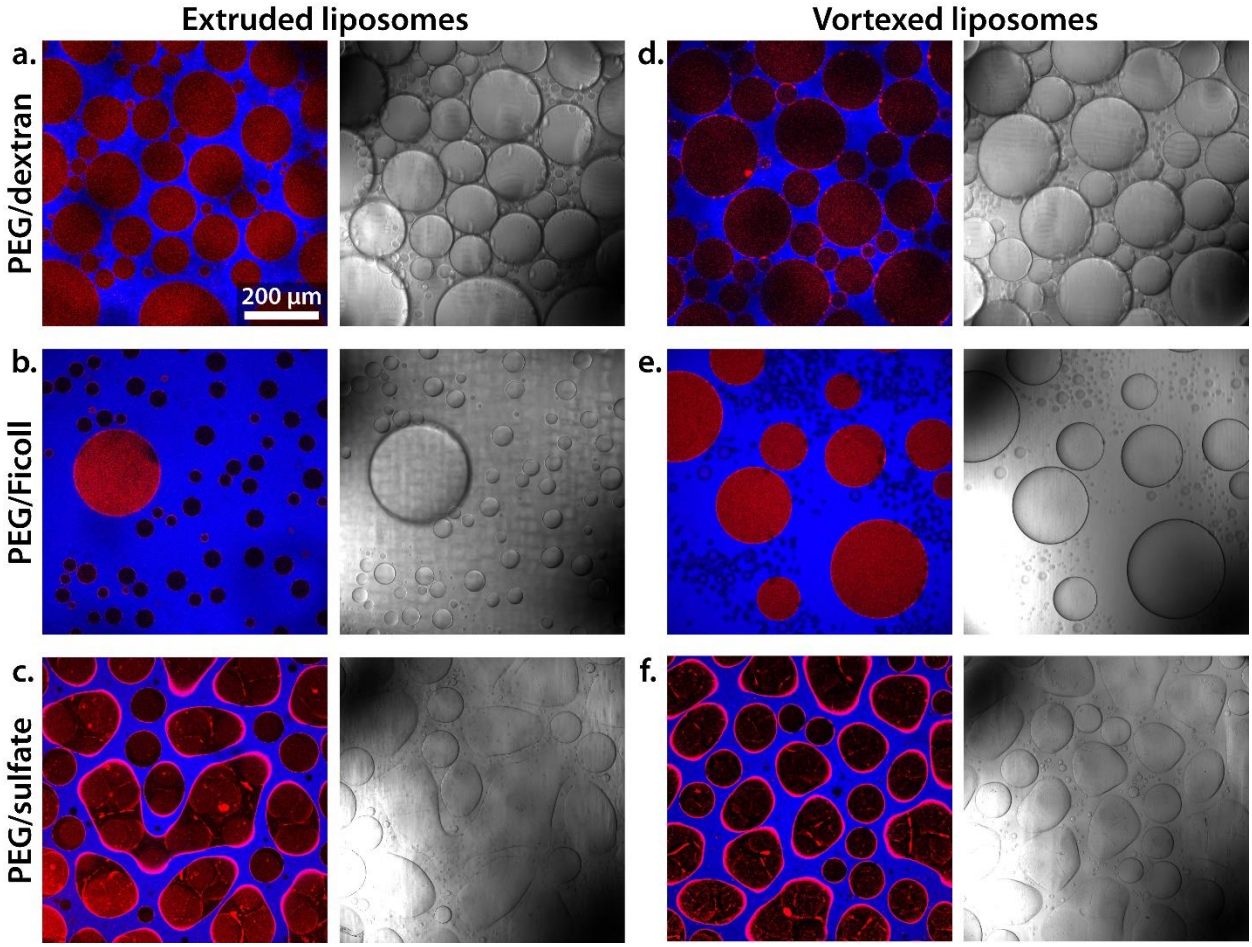

Figure S1: Comparison of different vesicle-stabilized all-aqueous emulsions at 5 days. fluorescent confocal microscopy images of various aqueous two-phase separation (ATPS) emulsions stabilized with extruded **(a, b, c)** and vortexed **(d, e, f)** liposomes. 5 days after initial sample preparation. Vortexed liposomes were able to effectively stabilize droplets of one phase within a continuous PEG-rich phase in the same manner as extruded liposomes with identical lipid composition. **(a,d)** PEG/dextran emulsions; **(b,e)** PEG/Ficoll emulsions; **(c,f)** PEG/sulfate emulsions. Left panels = overlaid fluorescence; right panels = brightfield. Rhodamine-tagged liposomes have been false colored red, Alexa 647-tagged PEG has been false colored blue.

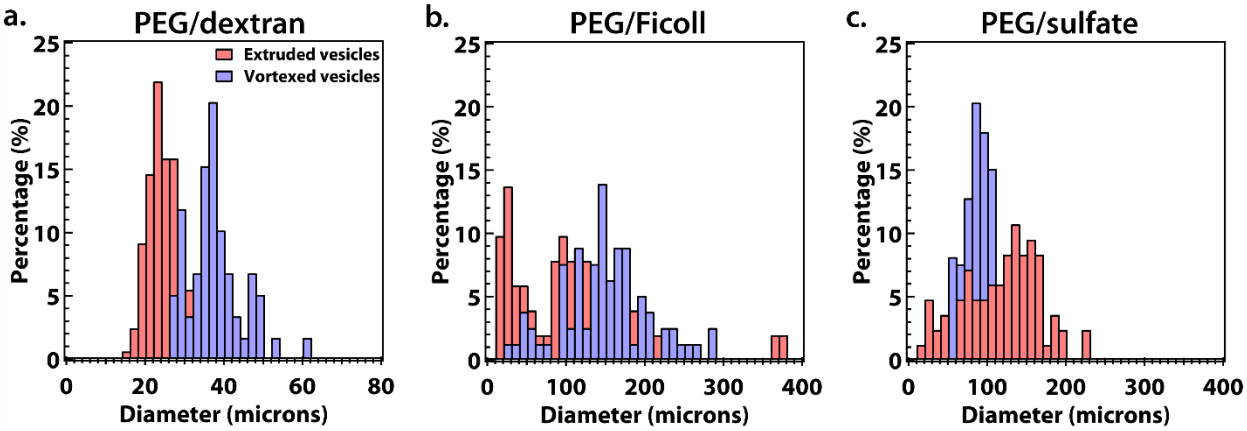

Figure S2: Effect of ATPS identity and lipid vesicle preparation method on size distributions of vesicle-stabilized all-aqueous emulsions. Size histograms of droplets in extruded and vortexed vesicle-stabilized ATPS emulsions, 24 hours after preparation. In general, vesicle extrusion results in a decrease in average droplet diameter and distribution spread. **(a)** Measurements of vesicle (extruded and vortexed)-stabilized dextran-rich droplets; **(b)** measurements of vesicle-stabilized Ficoll-rich droplets; **(c)** measurements of vesicle-stabilized sulfate-rich droplets. Red bars indicate emulsions with extruded vesicles, blue indicate vortexed. Approximately 100 droplets were measured across 4 images for each data set.

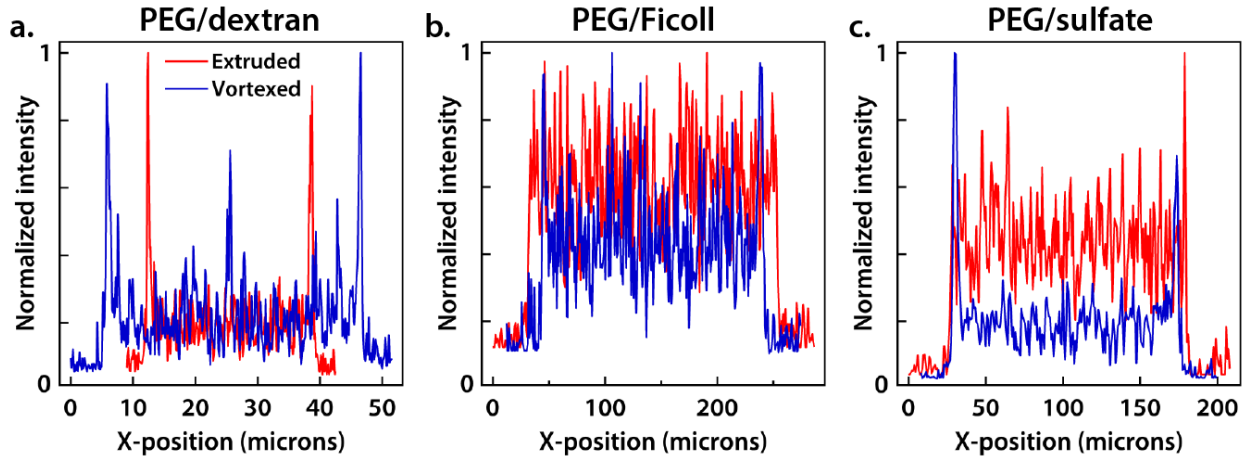

Figure S3: Fluorescent 2D line scans of Rhodamine fluorescence in vesicle-stabilized ATPS emulsions 24 hours after initial preparation. Line scans were performed across the full diameter of individual droplets in **(a)** PEG/dextran, **(b)** PEG/Ficoll, or **(c)** PEG/sulfate samples. Raw fluorescence data were extracted from fluorescent confocal microscopy images.

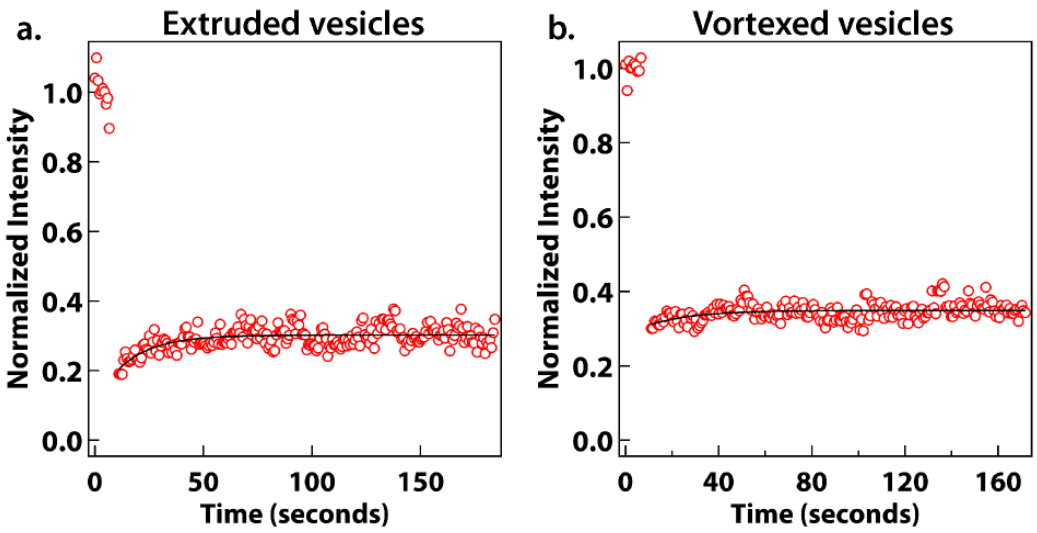

Figure S4: Examining the mobility of vesicles at interface of stabilized emulsions using FRAP. Vesicles assembled at the interface are similarly immobile for extruded and vortexed vesicles. Fluorescence recovery after photobleaching (FRAP) curves for vesicle diffusion in vesicle stabilized PEG/dextran ATPS emulsions. Curves depict the fluorescent bleaching and recovery of Rhodamine-tagged **(a)** extruded and **(b)** vortexed vesicles at the PEG/Dx interface. A fraction of the interface was bleached and monitored for fluorescence recovery, which would indicate exchange with unbleached labeled lipid vesicles.

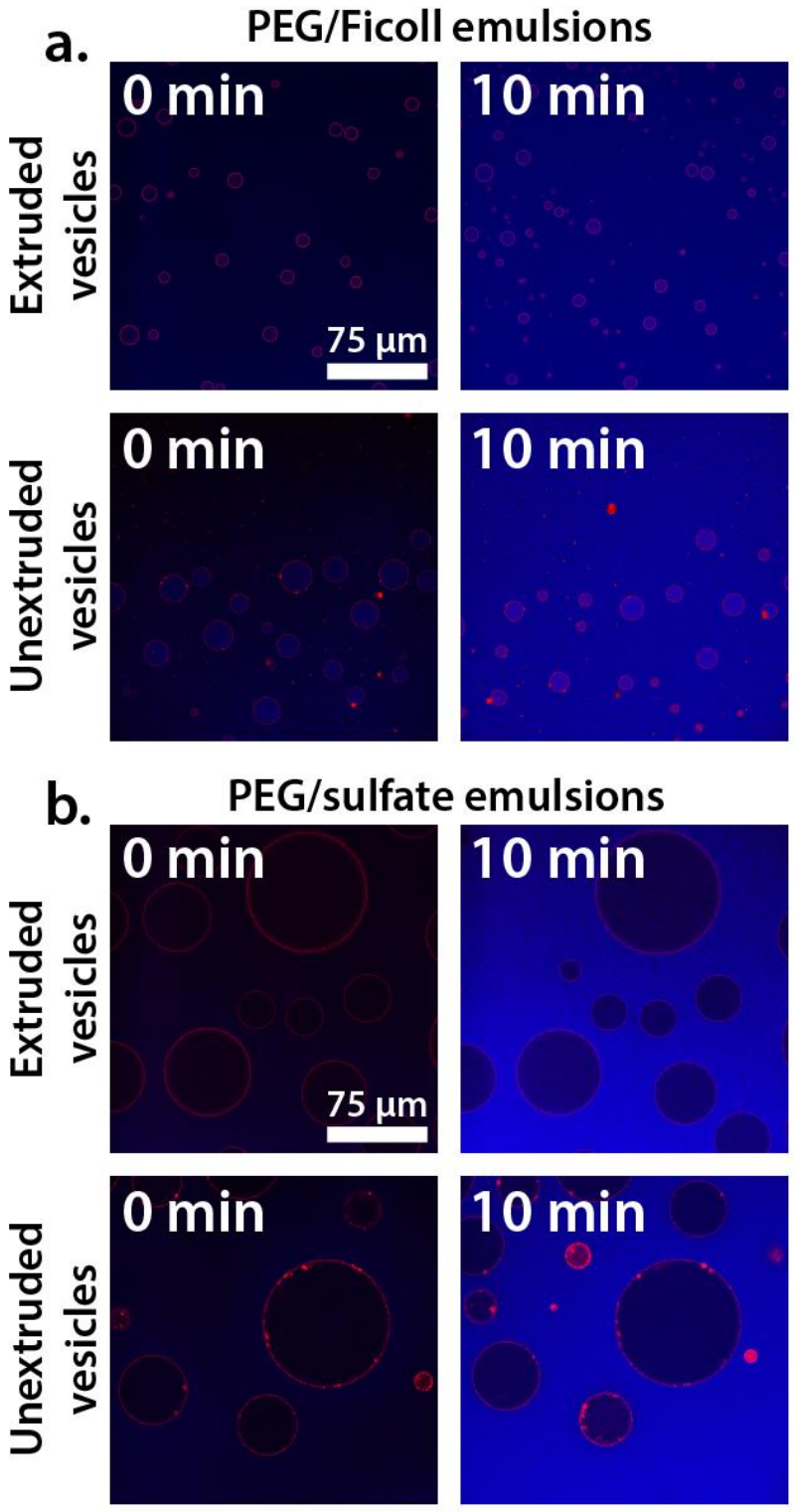

Figure S5: Tracking the diffusion of fluorescently tagged U15 oligoRNA throughout stabilized emulsions. Fluorescent confocal microscopy images depicting diffusion of labeled-RNA (U15) through vesicle-stabilized **(a)** PEG/dextran and **(b)** PEG/sulfate ATPS emulsions. Experimental setup depicted in Figure 5a was used for all samples. Inset refers to time after diffusion initiation. Rhodamine-tagged vesicles have been false-colored red, Alexa 647-tagged U15 blue.

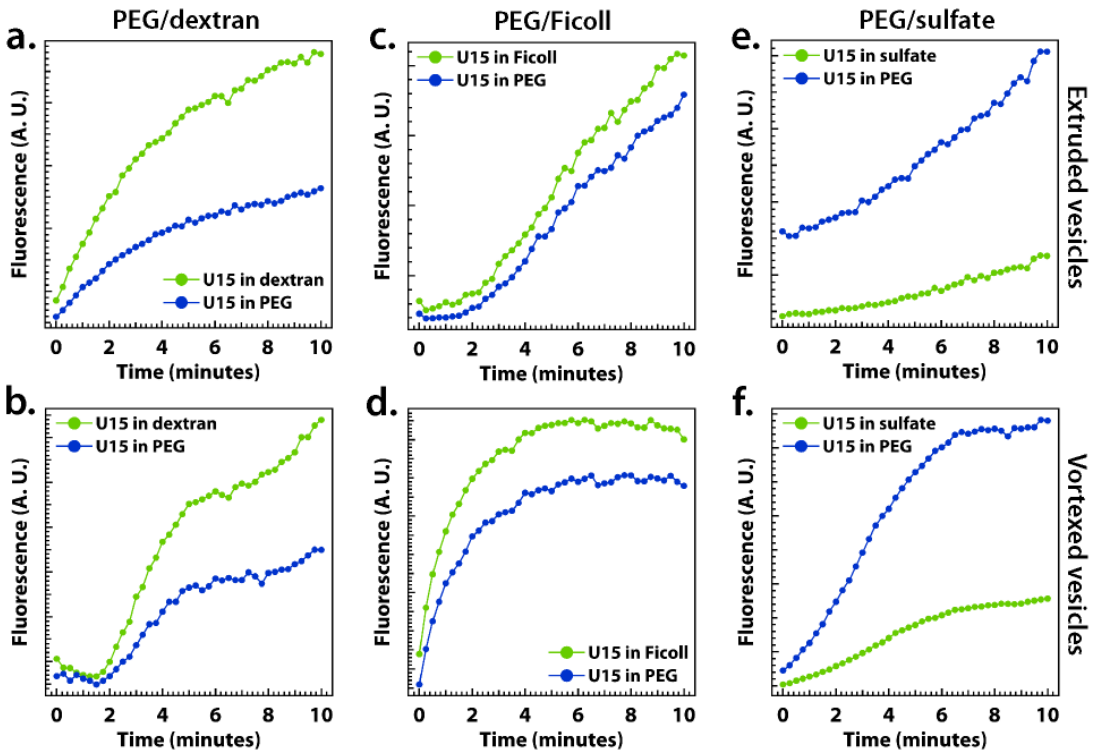

Figure S6: Tracking the diffusion of fluorescently tagged U15 oligoRNA inside stabilized emulsions. Relative Alexa-647 fluorescence in liposome-stabilized **(a,d)** PEG/Dx, **(b,e)** PEG/Ficoll, and **(c,f)** PEG/sulfate ATPS prepared with **(a-c)** extruded or **(d-f)** vortexed liposomes during U15 diffusion. Experimental setup depicted in Figure 5a was used for all samples. Time refers to time after addition of the cover slip and fusion of droplets.