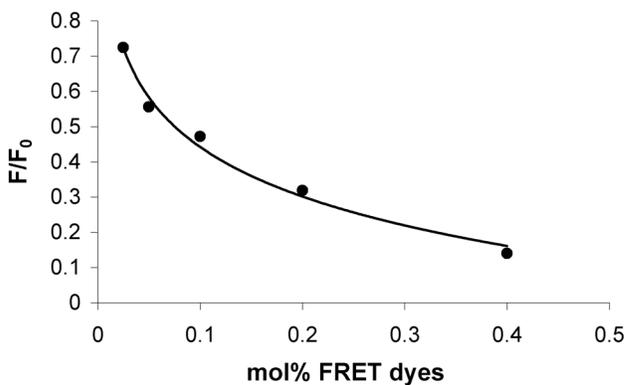


Supporting Information: Shrink-wrap Vesicles

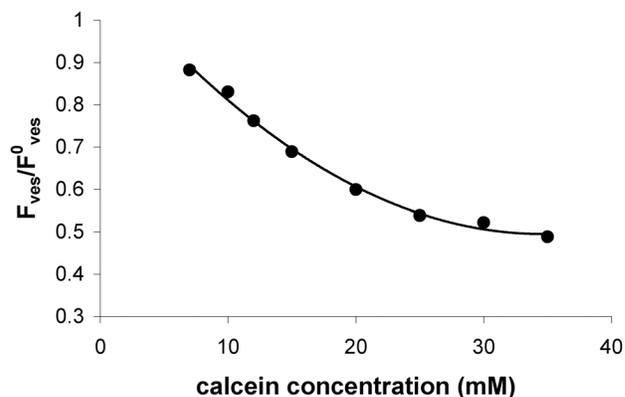
S.M. Fujikawa, I.A. Chen, J.W. Szostak

Figure S1.



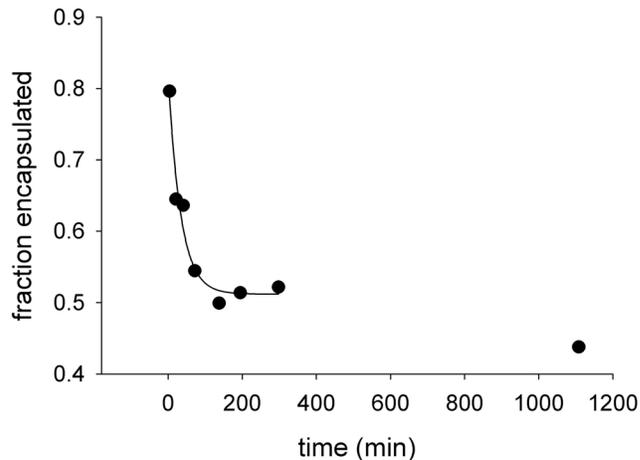
Standard curve of donor fluorescence vs. probe density for the FRET assay (F/F_0 = donor fluorescence/ donor fluorescence in 1% Triton X-100). The solid line shows a fitted curve ($y = -0.203*\ln(x) - 0.025$).

Figure S2.



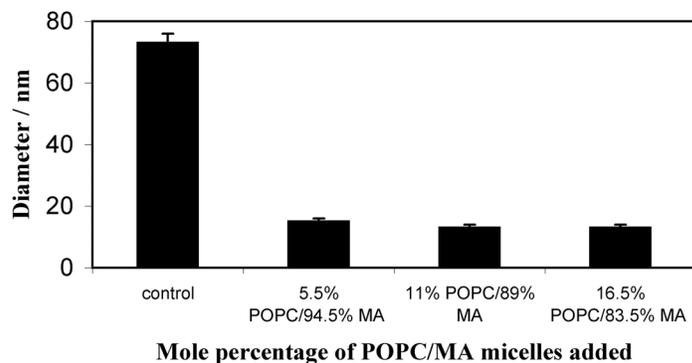
Standard curve of self-quenching of calcein vs. concentration (F_{ves}/F_{ves}^0 = calcein fluorescence/ calcein fluorescence in 1% Triton X-100). The solid line shows a fitted curve ($y = 0.00052*x^2 - 0.036*x + 1.12$).

Figure S3.



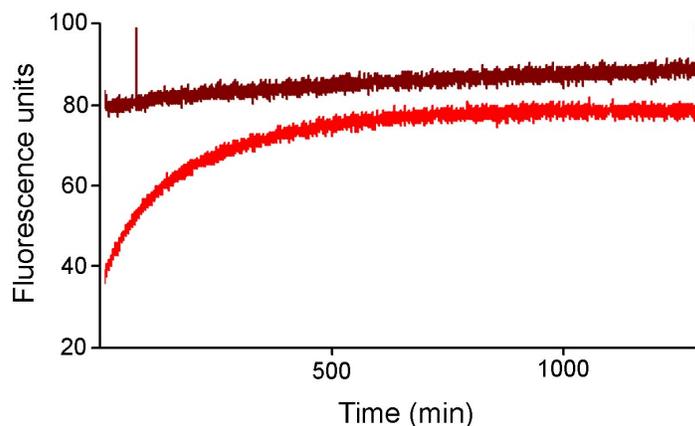
Release of calcein during and after shrinking of MA vesicles (a representative trial is shown). Although vesicle size has equilibrated by ~500 minutes (see Fig. 1A), calcein continues to be released slowly. The solid line shows a single exponential decay curve fit for the short time course ($k \sim 0.03 \text{ min}^{-1}$).

Figure S4.



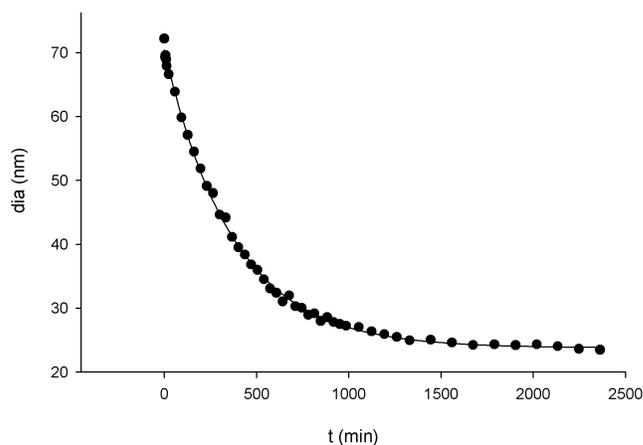
Final vesicle diameter after shrinking using micelles composed of varying mole percentages of POPC and MA, measured by DLS (~40 mM vesicles + 40 mM micelles).

Figure S5.



The fluorescence of calcein encapsulated in MA vesicles after dilution into an isotonic solution (0.2 M bicine, pH 8.5; brown line) or hypertonic solution (0.5 M bicine, pH 8.5; red line). In hypertonic solution, vesicles shrink osmotically, causing the internal concentration of calcein to increase and therefore the fluorescence to become self-quenched.^{1,2} The recovery of fluorescence within several hours indicates the return to a spherical volume, due to equilibration of bicine across the vesicle membrane.

Figure S6.



Shrinking of oleate vesicles (~3 mM) after adding MA/POPC (89% MA, 11% POPC) micelles (4 mM).

Supplementary Text

During vesicle growth, MA micelles that are not closely associated with a membrane (i.e., excess beyond 0.4 equivalents) aggregate into large metastable structures. However, during vesicle shrinking, because MA/POPC micelles do not aggregate at pH 8.5, excess MA/POPC micelles would increase the effective ratio of micelles to vesicles. Transfer of MA from vesicles to micelles may then occur in these complexes, a first-order process that would give rise to the observed exponential decay during shrinking.

Supplementary References

- (1) Sacerdote, M. G.; Szostak, J. W. *Proc Natl Acad Sci U S A* **2005**, *102*, 6004-6008.
- (2) Chen, P. Y.; Pearce, D.; Verkman, A. S. *Biochemistry* **1988**, *27*, 5713-5718.