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Viscoelastic deformation of lipid bilayer vesicles† (Supplementary Information)

Shao-Hua Wu,[a‡] Shalene Sankhagowit,[b‡] Roshni Biswas,[a] Shuyang Wu,[b] Michelle L. Povinelli,[a] and Noah Malmstadt*[b]

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Characterizing the laser response

The diode laser response was characterized using a photodetector (918D-SL-OD3, Newport Corp). Fig. S1 shows the measured power (blue curve) from both diode lasers upon ramping laser power from 100 to 500 mW, as in the GUV-stretching experiment. A single-exponential decay function was used to fit the measured laser power and yield the effective laser response time, 3.7 ms; orders of magnitude below the observed rate of membrane deformation.

Characterizing GUV bending energy

When stretching a giant unilamellar vesicle (GUV) at low tension, below 0.1 mN/m,[1] the area strain of the membrane is proportional to the log of the applied tension as a fraction of resting tension, $\ln(\sigma_h/\sigma_0)$;[2] the bending modulus is related to the constant of proportionality. To measure membrane bending modulus in our system and determine whether the deformation we observed was in the bending (rather than area dilation) regime, we increased the laser power in four steps from 100 mW to 500 mW, as in our previous work.[3] We measured a bending modulus of 11.47 ± 1.36 k_BT; similar to the results in our previous work.[3] The linearity of the data shown in Fig. S2 confirms that, at 500 mW laser power, the GUVs remained in the low-tension regime and that the increased area arose from stretching out stored, sub-optical membrane undulations.

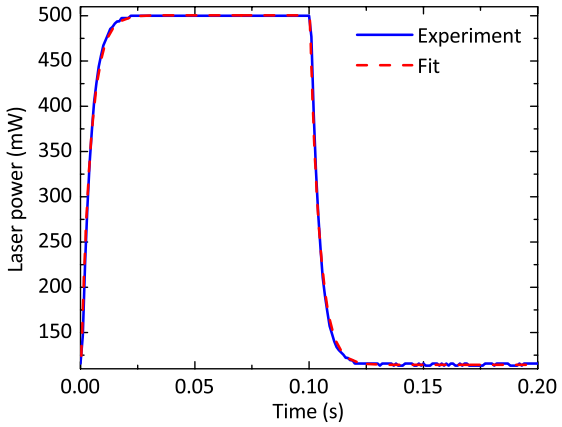

Fig. S1 Laser power intensity profile. The power of the diode lasers was switched between 100 and 500 mW in 5 Hz square waves; one period of the measured power is shown (blue) and used to fit to a single-exponential decay function (red) to yield the time delay of 3.7 ms for increasing or decreasing laser power.

Quantifying heating from laser absorbance during GUV stretching

Temperature-dependent rhodamine B fluorescence was used to map temperature changes in the microfluidic channel.[4] A solution of 0.1 mM rhodamine B in heavy water was flowed through the glass capillary while the laser power was turned on to 500 mW for 40 s. Note that the flow was maintained at ~90 μL/h to avoid photobleaching the dye. Fig. 3SA shows a single line scan of the rhodamine B fluorescence intensity profile as a function of time and position in the channel from the wall (0 μm) to the center (~50 μm) of the capillary cross-section. The purple (t = 2 s) and blue (t = 42 s) dashed lines indicate laser power ramp-up and ramp-down, respectively, between 100 to 500 mW. The fluorescence intensity (a.u.) was normalized to 1. There was no observable fluorescence intensity change during the 40 s period

[a] Ming Hsieh Department of Electrical Engineering, University of Southern California, 3737 Watt Way, PHE 614, Los Angeles, CA 90089-0271, USA.
[b] Mork Family Department of Chemical Engineering and Materials Science, University of Southern California, 925 Bloom Walk, HED 216, Los Angeles, CA 90089-1211, USA. Fax: +1 213 740 1056; Tel: +1 213 821 2034; E-mail: malmstad@usc.edu
† Electronic Supplementary Information (ESI) available: [details of any supplementary information available should be included here]. See DOI: 10.1039/b000000x/
‡ These authors contributed equally to this work.

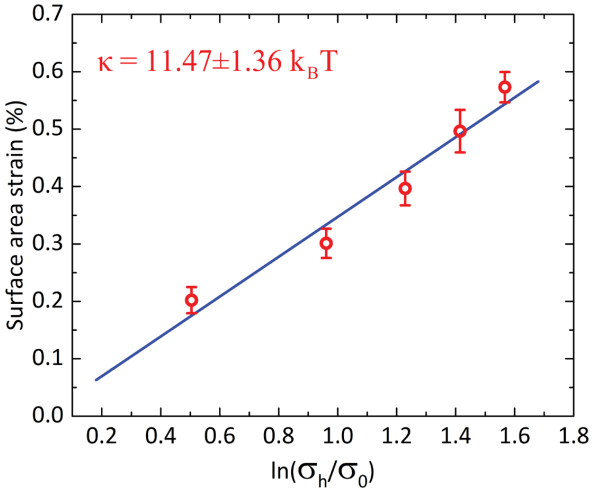

Fig. S2 Surface area strain versus the natural logarithm of the lateral tension over the resting tension. The error bar represents one standard deviation from the mean surface area strain.

at 500 mW laser power, and thus no observable temperature rise, induced by the diode lasers in our measurements.

To determine the limit of detection of this technique, an infrared heat lamp was aimed at the capillary from ~15 cm away to induce an observable temperature change; the temperature adjacent to the capillary was measured using a thermocouple connected to a multimeter (Fluke). The fluorescence intensity is shown as a function of time in Fig. S3B (black). A 3rd-order polynomial function was then used to fit to the intensity profile, Fig. S3B (blue). Twice of the mean square error (MSE) of fit, 0.0056, was then used as the minimum temperature change detectable with rhodamine B fluorescence. Temperature was concomitantly measured with fluorescence intensity, and its values are shown as red circles in Fig. S3C. Interpolation with these data points was then implemented to estimate the minimally observable temperature change at ~1.76 s (corresponding to the point equal to twice the MSE of the fit). The minimally observable temperature change by this method is estimated to be ~0.28 K.

To estimate the relative increase in the vesicle area strain due to the maximum possible temperature increase (ΔT) of 0.28 K at the experimental condition of 298 K (T_1) and under constant lateral tension, the expression for area strain ($\Delta A/A_0$) as a function of the bending modulus (κ) and applied tension relative to resting tension (σ_h/σ_0) is considered:[2]

$$\frac{\Delta A}{A_0} = \frac{k_B T}{8\pi\kappa} \ln\left(\frac{\sigma_h}{\sigma_0}\right) \tag{S1}$$

where k_B is the Boltzmann constant and T is temperature. The dependence of bending modulus on temperature is also considered:[5,6]

$$\ln(\kappa) = \frac{\varepsilon_K}{k_B T} + const \tag{S2}$$

where ε_K is the internal energy. Note that the value of ε_K was not available for POPC, so 7×10^{-21} J for 1,2-dioleoyl-sn-glycero-3-phosphocholine (DOPC) membranes[5] was used. At the starting temperature ($T = T_1 = 298$ K) and after temperature increase ($T = T_2 = T_1 + \Delta T = 298.28$ K), the area strains are equal to

$$\left(\frac{\Delta A}{A_0}\right)\Big|_1 = \frac{k_B}{8\pi} T_1 \exp\left(-\frac{\varepsilon_K}{k_B T_1}\right) \ln\left(\frac{\sigma_h}{\sigma_0}\right) \tag{S3}$$

$$\left(\frac{\Delta A}{A_0}\right)\Big|_2 = \frac{k_B}{8\pi} T_2 \exp\left(-\frac{\varepsilon_K}{k_B T_2}\right) \ln\left(\frac{\sigma_h}{\sigma_0}\right) \tag{S4}$$

respectively. The relative change of strain is then expressed as

$$\frac{\left(\frac{\Delta A}{A_0}\right)\Big|_2 - \left(\frac{\Delta A}{A_0}\right)\Big|_1}{\left(\frac{\Delta A}{A_0}\right)\Big|_1} = \frac{\left(\frac{\Delta A}{A_0}\right)\Big|_2}{\left(\frac{\Delta A}{A_0}\right)\Big|_1} - 1 = \frac{T_2}{T_1} \exp\left[\frac{\varepsilon_K}{k_B}\left(\frac{1}{T_2} - \frac{1}{T_1}\right)\right] - 1 \tag{S5}$$

Note that the first fraction and the natural logarithm from the right-hand side of Eq. S3 and S4 were divided out in Eq. S5. The right-hand side of Eq. S5 was calculated to be 0.00254 with 0.28 K temperature rise from 298 K.

Reproducibility of time constant measurements

We repeated dynamic stretching measurements on single GUVs to determine their reproducibility. Fig. S4 shows two stretching trials on the same GUV. The time constants are statistically indistinguishable. This was the case for all GUVs for which we repeated dynamic mechanical characterization.

References

1 J. R. Henriksen and J. H. Ipsen, *European Physical Journal E*, 2004, **14**, 149–167.

2 E. Evans and W. Rawicz, *Physical Review Letters*, 1990, **64**, 2094–2097.

3 M. E. Solmaz, S. Sankhagowit, R. Biswas, C. A. Mejia, M. L. Povinelli and N. Malmstadt, *RSC Advances*, 2013, **3**, 16632–16638.

4 J. J. Shah, M. Gaitan and J. Geist, *Analytical Chemistry*, 2009, **81**, 8260–8263.

5 J. Pan, S. Tristram-Nagle, N. Kucerka and J. F. Nagle, *Biophysical journal*, 2008, **94**, 117–124.

6 R. Podgornik and V. A. Parsegian, *Langmuir*, 1992, **8**, 557–562.

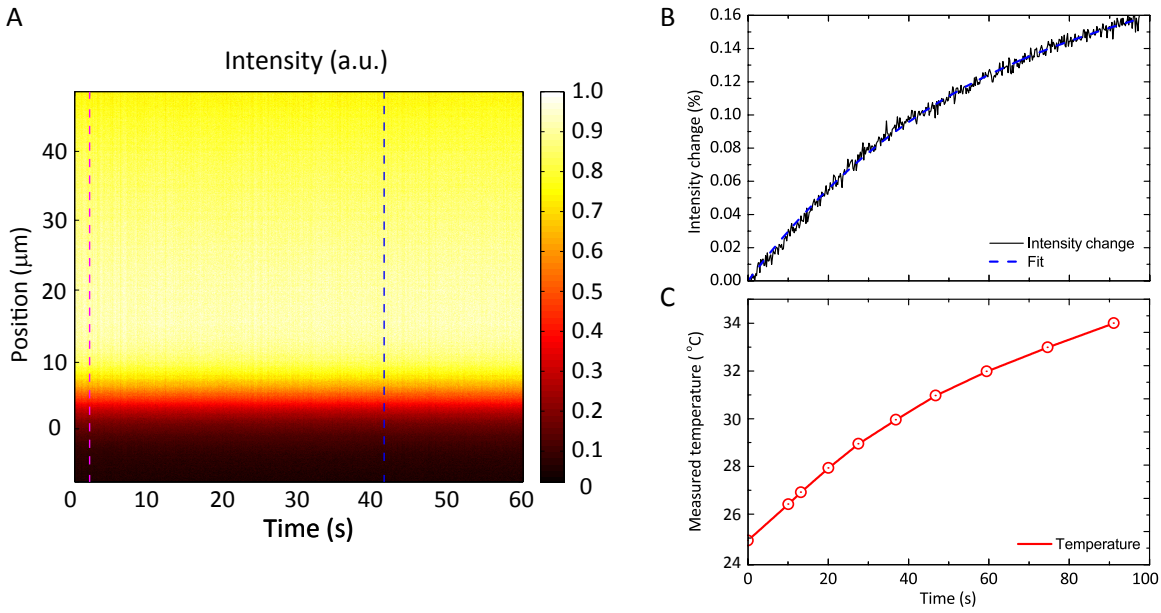

Fig. S3 Temperature profiles probed by rhodamine B fluorescence intensity. (A) Heating with lasers; the fluorescence intensity profile in half of the capillary cross-section with time. At t = 0 s, the laser power was at 100 mW. At t = 2 s (purple, dashed line), the power was increased to 500 mW and held for 40 s. At t = 42 s (blue, dashed line), the power was returned to 100 mW and held for 20 s. (B) Heating with the infrared lamp; fluorescence intensity at the center of the capillary cross-section (black, solid) over time, fitted to a 3rd-order polynomial function (blue, dashed). (C) Measure temperature change as a function of time.

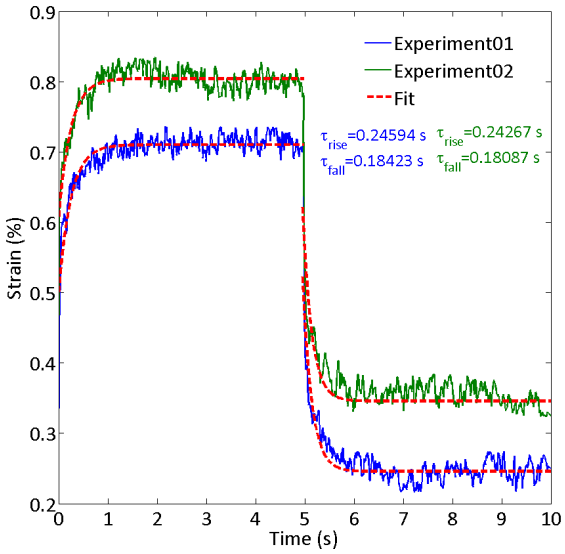

Fig. S4 Repeated dynamic mechanical analysis of the same GUV. The rise and fall time constants are statistically indistinguishable from trial to trial.