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**Supporting Material**

**Lipid Bilayer Domain Fluctuations as a Probe of Membrane Viscosity**

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# Supplementary material for “Lipid Bilayer Domain Fluctuations as a Probe of Membrane Viscosity”

## Supplement A. DETAILS ON THE DERIVATION OF THE SHAPE RELAXATION TIMES

Our calculation of the relaxation times is based on the work of both Stone and McConnell [1] and Lubensky and Goldstein [2]; indeed, our results follow immediately from the hydrodynamic analysis presented in these works with only slight modifications. The mathematical details are summarized here, elaborating upon the treatment in Appendix C of [2]. Within the Saffman-Delbrück picture of a fluid membrane sheet [3], it is possible to predict the in-plane velocity of all points on the membrane surface for an arbitrary distribution of in-plane forces acting on the membrane [2, 3]. Given a force density  $\mathbf{F}(\mathbf{r})$  (per unit area), the membrane velocity is calculated from the membrane Green’s function tensor  $T_{ij}(\mathbf{r})$  for velocity response to an applied point force,

$$v_i(\mathbf{r}) = \int d\mathbf{r}' T_{ij}(\mathbf{r} - \mathbf{r}') F_j(\mathbf{r}'). \quad (\text{A1})$$

Here, and in all that follows, the indices  $i$  and  $j$  refer to in-plane cartesian directions ( $x$  or  $y$ ), with summation implied in expressions with repeated indices. Although there is no simple closed-form expression for  $T_{ij}$ , it may be expressed as the integral [2]

$$\begin{aligned} T_{ij}(\mathbf{r}) &= \frac{-1}{2\pi\eta_m} \int_0^\infty \frac{dq}{q^2(q+1/L_{sd})} \left[ \frac{r_i r_j}{r^3} q J_0'(qr) + \left( \delta_{ij} - \frac{r_i r_j}{r^2} \right) q^2 J_0''(qr) \right] \\ &= \frac{-1}{2\pi\eta_m} \int_0^\infty \frac{dq}{q^2(q+1/L_{sd})} \left[ - \left( \delta_{ij} - \frac{r_i r_j}{r^2} \right) J_0(qr) q^2 + \left( \delta_{ij} - 2 \frac{r_i r_j}{r^2} \right) J_1(qr) \frac{q}{r} \right] \end{aligned} \quad (\text{A2})$$

where  $L_{sd} = \eta_m/(2\eta_f)$ ,  $\eta_m$  is the membrane surface viscosity,  $\eta_f$  is the bulk fluid viscosity, and  $J_n$  are Bessel functions of the first kind (the primes indicate differentiation). We emphasize that this result differs slightly from the form used in [2]. The bilayer geometry considered in this work requires  $2\eta_f$  to appear in the denominator of  $L_{sd}$  whereas the monolayer geometry at the air-water interface considered in [2] places  $\eta_f$  (without the factor of 2) in this constant. The bilayer is subject to dissipation from the bulk fluid both above and below the bilayer, which accounts for the factor of 2 [4, 5].

Our result for domain fluctuation dynamics is calculated in the limit of linear response, considering only small fluctuations of domain shape away from the minimum energy configuration of a perfect circle of radius  $R$ . These small fluctuations in domain shape give rise to restoring forces, which are explicitly written in Eq. 3 of the main paper. The important point about this expression, is that the force vanishes for the undeformed circle - only linear (and, in principle, higher order) contributions are present. In order for the velocity of Eq. A1 to be linearly dependent on the shape deformations, the Green’s function must be evaluated for the undeformed domain geometry of the perfect circle. Any deviations from the zeroth order geometry in  $T_{ij}$  would necessarily lead to second (and higher) order contributions in the velocity when multiplied against the forces. We are thus led to a less general form of expression A1, which considers the velocity of the domain boundary at points  $(R, \theta)$  as driven by restoring forces at points  $(R, \theta')$  in polar coordinates.

$$v_i(R, \theta) = \int_0^\infty dr' r' \int_0^{2\pi} d\theta' T_{ij}(R\hat{\mathbf{r}}(\theta) - \mathbf{r}') \delta(r' - R) f_r(\theta') \hat{r}'_j(\theta') = R \int_0^{2\pi} d\theta' T_{ij}(R\hat{\mathbf{r}}(\theta) - R\hat{\mathbf{r}}'(\theta')) f_r(\theta') \hat{r}'_j(\theta'). \quad (\text{A3})$$

The unit vectors  $\hat{\mathbf{r}}(\theta) = (\hat{r}_x(\theta), \hat{r}_y(\theta)) = (\cos \theta, \sin \theta)$  and  $\hat{\mathbf{r}}'(\theta') = (\hat{r}_x(\theta'), \hat{r}_y(\theta')) = (\cos \theta', \sin \theta')$  point along the outward radial direction for the indicated polar angles. The radially directed velocity is then given by  $v_r(\theta) = \hat{\mathbf{r}}(\theta) \cdot \mathbf{v}(R, \theta)$  so that

$$\begin{aligned} v_r(\theta) &= R \int_0^{2\pi} d\theta' \hat{r}_i(\theta) (T_{ij}(R\hat{\mathbf{r}}(\theta) - R\hat{\mathbf{r}}'(\theta')) f_r'(\theta') \hat{r}'_j(\theta')) \\ &\equiv \int_0^{2\pi} d\theta' R T_{\hat{\mathbf{r}}\hat{\mathbf{r}}'}(R\hat{\mathbf{r}}(\theta) - R\hat{\mathbf{r}}'(\theta')) f_r'(\theta') \end{aligned} \quad (\text{A4})$$

The velocities and forces are defined explicitly in the main paper. If the radial velocity is measured at angle  $\theta$  on the circle, as driven by a radially directed force at angle  $\theta'$ , the cartesian vector separating these two points is  $\mathbf{r} - \mathbf{r}' = R(\cos \theta - \cos \theta', \sin \theta - \sin \theta')$  with a separation of  $\mathcal{R} = |\mathbf{r} - \mathbf{r}'| = 2R \sin(\frac{\theta - \theta'}{2})$ . It is clear by symmetry that the Green’s function for radially directed forces and velocities  $T_{\hat{\mathbf{r}}\hat{\mathbf{r}}'}(R\hat{\mathbf{r}}(\theta) - R\hat{\mathbf{r}}'(\theta'))$  can only depend on the angles  $\theta$  and  $\theta'$  via their difference  $\theta - \theta'$ ; a rotation of both

points around the origin will not affect value of the radially directed Green's function on the circle. Carrying out the calculation explicitly, starting from Eq. A2 leads to

$$\begin{aligned}
T_{\hat{\mathbf{r}}\hat{\mathbf{r}}'}(\theta - \theta') &\equiv T_{\hat{\mathbf{r}}\hat{\mathbf{r}}'}(R\hat{\mathbf{r}}(\theta) - R\hat{\mathbf{r}}'(\theta')) = \frac{-1}{2\pi\eta_m} \int_0^\infty \frac{dq}{q^2(q + 1/L_{sd})} \left[ -\cos^2\left(\frac{\theta - \theta'}{2}\right) J_0(q\mathcal{R})q^2 + \frac{J_1(q\mathcal{R})q}{\mathcal{R}} \right] \\
&= \frac{-1}{2\pi\eta_m} \int_0^\infty \frac{dq}{q^2(q + 1/L_{sd})} \left\{ -\cos^2\left(\frac{\theta - \theta'}{2}\right) J_0\left[2qR \sin\left(\frac{\theta - \theta'}{2}\right)\right] q^2 + \frac{J_1\left[2qR \sin\left(\frac{\theta - \theta'}{2}\right)\right] q}{2R \sin\left(\frac{\theta - \theta'}{2}\right)} \right\} \\
&= \frac{-1}{2\pi\eta_m} \int_0^\infty \frac{dx}{x^2(x + R/L_{sd})} \left\{ -\cos^2\left(\frac{\theta - \theta'}{2}\right) J_0\left[2x \sin\left(\frac{\theta - \theta'}{2}\right)\right] x^2 + \frac{J_1\left[2x \sin\left(\frac{\theta - \theta'}{2}\right)\right] x}{2 \sin\left(\frac{\theta - \theta'}{2}\right)} \right\}. \\
&= \frac{-1}{2\pi\eta_m} \int_0^\infty \frac{dx}{x^2(x + R/L_{sd})} \left\{ \frac{\partial^2}{\partial(\theta - \theta')^2} J_0\left[2x \sin\left(\frac{\theta - \theta'}{2}\right)\right] \right\}. \tag{A5}
\end{aligned}$$

This expression may be substituted into Eq. A4 to yield

$$v_r(\theta) = \int_0^{2\pi} d\theta' RT_{\hat{\mathbf{r}}\hat{\mathbf{r}}'}(\theta - \theta') f_{r'}(\theta') \tag{A6}$$

which has the form of a simple convolution in the polar angle around the domain perimeter, so that

$$v_n(t) = \pi \mathcal{T}_n f_n(t) \tag{A7}$$

$$\mathcal{T}_n \equiv \frac{R}{\pi} \int_0^{2\pi} d\theta e^{-in\theta} T_{\hat{\mathbf{r}}\hat{\mathbf{r}}'}(\theta) = \frac{R}{\pi} \int_0^{2\pi} d\theta \cos(n\theta) T_{\hat{\mathbf{r}}\hat{\mathbf{r}}'}(\theta) \tag{A8}$$

and the final equality originates from the fact that  $T_{\hat{\mathbf{r}}\hat{\mathbf{r}}'}(\theta)$  is even around the point  $\theta = \pi$ , which is clear from symmetry considerations as well as the explicit mathematical expressions. The integral over  $\theta$  is taken using the last line of Eq. A5 and applying integration by parts twice to move the derivatives off the Bessel function and on to  $\cos(n\theta)$ . The boundary terms vanish.

$$\begin{aligned}
\mathcal{T}_n &= \frac{n^2 R}{2\pi^2 \eta_m} \int_0^\infty \frac{dx}{x^2(x + R/L_{sd})} \int_0^{2\pi} d\theta \cos(n\theta) J_0\left[2x \sin\left(\frac{\theta}{2}\right)\right] \\
&= \frac{n^2 R}{\pi^2 \eta_m} \int_0^\infty \frac{dx}{x^2(x + R/L_{sd})} \int_0^\pi d\phi \cos(2n\phi) J_0[2x \sin \phi] \\
&= \frac{n^2 R}{\pi \eta_m} \int_0^\infty \frac{dx J_n(x)^2}{x^2(x + R/L_{sd})}. \tag{A9}
\end{aligned}$$

The final integral over  $\theta$  may be found in standard tables [6]. Eq. A9 and Eq. A7 lead immediately to Eq. 5 of the main paper.

### Supplement B. THE MEANING OF $\eta_m$ WHEN DOMAIN AND SURROUNDINGS DO NOT SHARE THE SAME VISCOSITY

For future reference, we restate Eq. 5 from the main paper

$$\tau_n = \frac{\eta_m R}{\sigma} \frac{1}{n^2(n^2 - 1)} \left[ \int_0^\infty dx \frac{J_n^2(x)}{x^2(x + R/L_{SD})} \right]^{-1} = \frac{\eta_m R}{\sigma} \frac{1}{I_n(2R\eta_f/\eta_m)n^2(n^2 - 1)}. \tag{B1}$$

This expression and its derivation above are restricted to the case that both the domain and its surroundings have identical surface viscosity,  $\eta_m$ . Though we are not able to present a fully general theory for the case where domain viscosity  $\eta_d$  differs from the surrounding viscosity  $\eta_s$ , we can argue that that the measured quantity  $\eta_m$  (obtained via fitting data to Eq. B1) is approximately equal to the mean viscosity  $(\eta_d + \eta_s)/2$ , when the experimental data is observed to fit the form of Eq. B1. We can also place bounds on the accuracy of this approximation.

In the limiting case where dynamics are governed solely by the behavior within the membrane, it is known that Eq. 8 of the main paper may be generalized to the case of distinct  $\eta_d$  and  $\eta_s$  [7]

$$\tau_n^{\text{membrane}} = \frac{2(\eta_d + \eta_s)R}{n\sigma}. \tag{B2}$$

In this limit, the correspondence mentioned above is exact. Measuring mode relaxation times and fitting to paper Eq. 8 yields  $\eta_m = (\eta_d + \eta_s)/2 \equiv \bar{\eta}$ . More generally, for any finite mode number  $n$ , the experimental relaxation times will always be longer than  $\tau_n^{\text{membrane}}$  of eq. B2 owing to the additional dissipation afforded by the surrounding bulk solvent. Since we assume that the available experimental data is well fit by eq. B1 this means that

$$\frac{\eta_m R}{\sigma} \frac{1}{I_n(2R\eta_f/\eta_m)n^2(n^2 - 1)} > \frac{2(\eta_d + \eta_s)R}{n\sigma} \quad (\text{B3})$$

where  $\eta_m$  must now be interpreted as a fitting parameter or effective viscosity that incorporates the influence of both  $\eta_d$  and  $\eta_s$ . The influence of solvent decreases with increasing  $n$  and hence approaches equality most closely for the largest mode number measured in a given experiment  $n_{max}$ . This inequality may be rearranged and considering only  $n_{max}$  yields the tightest bound on  $\bar{\eta}$  possible from the experimental measurements

$$\begin{aligned} \bar{\eta} &< \alpha\eta_m \\ \alpha &\equiv \frac{n_{max}}{4I_n(2R\eta_f/\eta_m)n_{max}^2(n_{max}^2 - 1)}. \end{aligned} \quad (\text{B4})$$

The dimensionless quantity  $\alpha > 1$  provides a measure for how closely mode  $n_{max}$  approaches the idealized ‘‘membrane only’’ limit. Values of  $\alpha$  close to one are nearly in the limiting regime whereas larger values correspond to systems more strongly influenced by the bulk solvent. The domains analyzed in this work span the range of  $\alpha = 1.1 - 2.1$ .

We also note that the measured relaxation time for mode  $n$  will always be shorter than that for a hypothetical membrane with homogeneous viscosity  $\eta_{lim} = \max\{\eta_s, \eta_d\}$ , because such a membrane is subject to additional dissipation over the region of the membrane that has been replaced by higher viscosity. For a given value of  $\bar{\eta}$ , the largest value that  $\eta_{lim}$  can possibly assume is  $2\bar{\eta}$ , which would correspond to the membrane having one region (either domain or surroundings) with vanishing viscosity. If we assume Eq. B1 provides a good fit to the data (with  $\eta_m$  the measured fit constant) then the preceding argument implies that  $\tau_n(\eta_m) < \tau_n(2\bar{\eta})$ , by which we mean

$$\frac{\eta_m R}{\sigma} \frac{1}{I_n(2R\eta_f/\eta_m)n^2(n^2 - 1)} < \frac{2\bar{\eta}R}{\sigma} \frac{1}{I_n(R\eta_f/\bar{\eta})n^2(n^2 - 1)} \quad (\text{B5})$$

Since  $\tau_n(\eta)$  increases monotonically with  $\eta$ , this expression implies

$$\eta_m < 2\bar{\eta} \quad (\text{B6})$$

Combing Eq. B4 and Eq. B6 leads to bounds on the value of the mean bilayer viscosity  $\bar{\eta}$  in terms of the measured effective viscosity  $\eta_m$

$$\frac{\eta_m}{2} < \bar{\eta} < \alpha\eta_m. \quad (\text{B7})$$

This expression quantifies the meaning of our prior assertion that  $\eta_m \sim \bar{\eta}$ . As noted above, the experimental data considered in this work involves domains with  $\alpha$  values on the order of two or smaller and so  $\frac{\eta_m}{2} < \bar{\eta} < 2\eta_m$  provides a conservative estimate of the uncertainty in our measurement of  $\bar{\eta}$ . To within a factor of two,  $\eta_m = \bar{\eta}$ .

We stress that Eq. B7 does not preclude the possibility that  $\eta_m = \bar{\eta}$ , since  $\alpha > 1$  for measurement of any finite  $n$ . The equality does hold in the limit of large  $n$  and it is possible that this equality extends over all  $n$ . Indeed, if perfect experimental data extending over all  $n$  values were found to fit Eq. B1, it would necessarily be the case that  $\eta_m = \bar{\eta}$  since the equality must hold at high  $n$ , thus setting the value for the entire set of modes. The preceding analysis accounts for the possibility that the agreement between Eq. B1 and experiment may only be apparent/approximate due to uncertainty in the data. Without data extending down to the  $\alpha = 1$  limit, it is not possible to surmise that the functional form of B1 with a constant  $\eta_m$  value holds over all  $n$  values.

### Supplement C. DETAILS OF THE COMPARISON BETWEEN THEORY AND EXPERIMENT

Eq. B1 predicts the  $R$  dependence of relaxation times for mode  $n$ . The form of this expression suggests a natural collapse of the data onto a single dimensionless curve for each mode

$$\frac{\sigma\tau_n}{\eta_m L_{sd}} = g_n(R/L_{sd}) \quad (\text{C1})$$

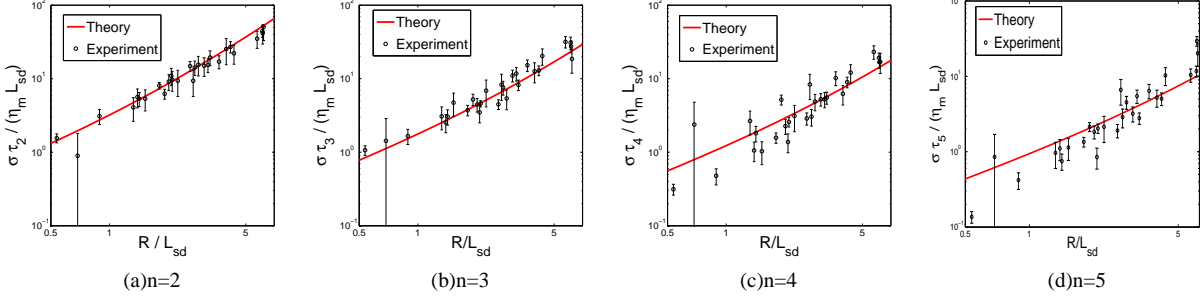


FIG. 1: Rescaling relaxation times collapses the data to the form predicted by the extended theory, Eq. C1. Error bars are propagated from the uncertainty in the measured quantities  $\tau_n$  and  $\sigma$ . The experimental data closely matches the theoretical predictions.

where  $g_n(y) = \frac{1}{n^2(n^2-1)}y \left[ \int_0^\infty dx \frac{J_n^2(x)}{x^2(x+y)} \right]^{-1}$ . Since the experimental data is collected from domains with a continuous range of  $R$ , but only a few discrete  $n$  values, Eq. C1 provides an appealing theoretical prediction to assess the validity of the proposed theoretical model over the full range of collected experimental data.

We note that Eq. C1 contains four distinct physical parameters that contribute to  $\tau_n$ :  $\sigma$ ,  $\eta_m$ ,  $\eta_f$  and  $R$ . We assume three of these quantities to be known from measurements other than the relaxation time. The viscosity of water,  $\eta_f$ , is well known. The line tension of the domain is known from the equilibrium fluctuations as described in the main paper and the domain radius is known from direct observation. The values of  $R$  and  $\sigma$  do vary from domain to domain and so analysis of the decay times is necessarily carried out individually for each domain. After determining the domain boundary  $r(\theta, t)$  as in the paper and extracting the deviations from circular shape,  $u_n(t)$ , we fit the autocorrelation functions  $\langle u_n(t)u_{-n}(0) \rangle$  to the form  $\langle |u_n|^2 \rangle e^{-t/\tau}$  to determine the relaxation times for each observable mode ( $n=2,3,4,5$ ). (Here, and in all subsequent fits, we use MATLAB's implementation of the Levenberg-Marquardt algorithm for nonlinear least-squares.) We then fit these four modes simultaneously to Eq. B1 to determine the single best fit  $\eta_m$  for each domain. Since  $\eta_m$  represents the best fit to the behavior of the entire domain, and not a single mode, the measured quantities  $\tau_n^{experimental}$  will generally differ from the  $\tau_n^{predicted}$  obtained by inserting  $\sigma$ ,  $\eta_m$ ,  $\eta_f$  and  $R$  into Eq. B1; the magnitude and nature of the deviations provide an estimate of the fit quality afforded by Eq. B1 to the experimental data. In Figs. 1 and 2 we present two different visualizations of the quality-of-fit obtained by the theoretical model proposed in the present work (Eq. B1). Fig. 1 plots the measured relaxation times, rescaled to the dimensionless form suggested by Eq. C1. The experimental data is in very good agreement with the theoretical predictions. The deviations between experiment and theory that do exist for individual points appear to be non-systematic, i.e. they are equally apparent over all mode numbers and for all domain radii and the scatter falls on both sides of the theoretical predictions. (It could be argued that the theory for mode  $n = 5$  does show some systematic deviations from the data, however the  $n = 5$  data is right at the limit of experimental resolution for many domains and this data may not be fully reliable. The analysis discussed in this section was also carried out using only modes  $n = 2, 3, 4$  and no significant changes were observed.)

Alternatively, we can simply plot the ratio between the measured relaxation times and the relaxation times predicted by Eq. B1

$$\tau_n^{experimental} / \tau_n^{predicted} = \frac{\tau_n^{experimental}}{\eta_m R} \sigma n^2 (n^2 - 1) \int_0^\infty dx \frac{J_n^2(x)}{x^2(x + \Lambda)}. \quad (C2)$$

If the experimental data carried no uncertainties and Eq. B1 served as a perfect description of physical reality, this quantity would equal unity for all  $n$  and for all domains. The data plotted in this form is given in Fig. 2. Although we see scatter just as in the representation of Fig. 1, the agreement between theory and experiment is perhaps more striking here - the data points straddle the theoretical predictions and show no systematic trends in the deviations that are observed.

By contrast to the above, if we attempt to explain the experimental measurements by fitting to the Stone-McConnell form with a similar procedure, using the bulk solvent viscosity as the independent fit parameter, we see much worse results. In this case we assume Eq. 7 of the main paper is the correct model for the dynamics

$$\tau_n^{fluid} = \frac{2\pi R^2 \eta_f}{\sigma} \frac{n^2 - 1/4}{n^2(n^2 - 1)}. \quad (C3)$$

We use the same procedure outlined above to determine the  $\tau_n$ 's from experiment, and find the best fitting value of the *bulk viscosity*  $\eta_f$  for each domain (as previously, the domain radius  $R$  and line tension  $\sigma$  are known from the thermal fluctuations) by fitting to modes  $n = 2, 3, 4, 5$  simultaneously. If Eq. C3 were a good model for the system's dynamics, the ratio

$$\frac{\sigma \tau_n^{experimental}}{2\pi R^2 \eta_f} \frac{n^2(n^2 - 1)}{n^2 - 1/4} = \tau_n^{experimental} / \tau_n^{fluid} \quad (C4)$$

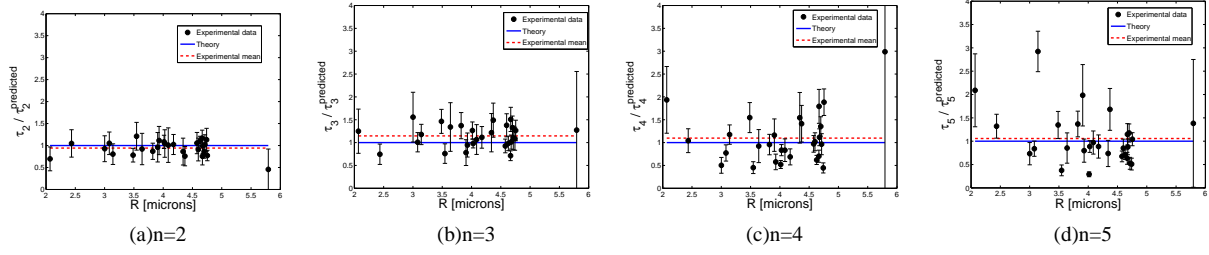


FIG. 2: No strong systematic error is seen in using Eq. B1 to “predict” observed data from the best fit viscosity. As in Fig. 1, error bars are propagated from the uncertainty in the measured quantities  $\tau_n$  and  $\sigma$ . The dashed red line simply measures the average taken over all the experimental points in each plot. The proximity of this line to one for all  $n$  indicates there is very little, if any, systematic error associated with the predicted expression (Eq. B1).

should be close to one for all domain radii and all mode numbers  $n$ . In fact, what we find is that the  $n = 2$  mode relaxation times are systematically low, while the times for  $n = 3$ ,  $n = 4$ , and  $n = 5$  are systematically long (Fig. 3), with the disagreement becoming more pronounced as  $n$  gets larger. This systematic deviation is predicted by Eq. B1. To demonstrate this, we generate

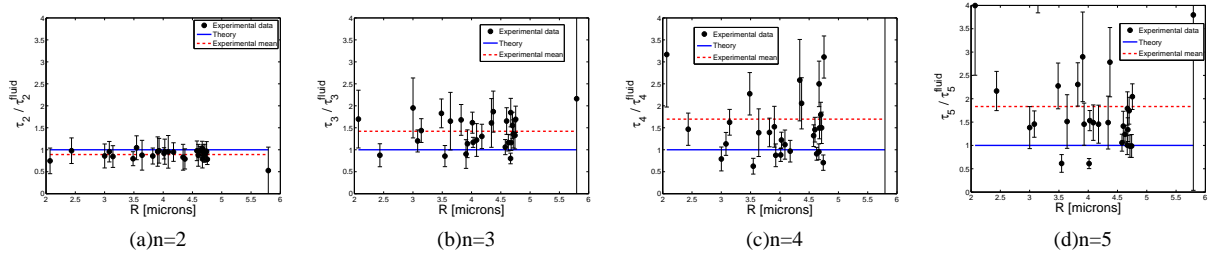


FIG. 3: Attempting to fit experimental data to the Stone-McConnell theory, using a different bulk viscosity for each domain, causes systematic problems;  $n = 2$  relaxation times are systematically low compared to the best fit, and  $n = 3, 4, 5$  are increasingly large compared to the best fit.

relaxation times from Eq. B1 for domains in the range  $R = 2 - 5$  microns, with a membrane surface viscosity  $\eta_m = 3 \times 10^{-6}$  s.P., bulk fluid viscosity of  $\eta_f = 1$  cP and line tension  $\sigma = 0.2$  pN. This manufactured “data” was fit to the Stone-McConnell form using the above procedure (Fig 4). The results are in striking agreement with Fig. 3 - the best fit  $n = 2$  times are below theoretical predictions, whereas modes  $n = 3, 4, 5$  show the opposite behavior. Even the magnitudes of the average deviations are close to the experimental plot. The Stone-McConnell theory fails in a manner completely consistent with the fact that the experimental data agrees with the predictions of Eq. B1.

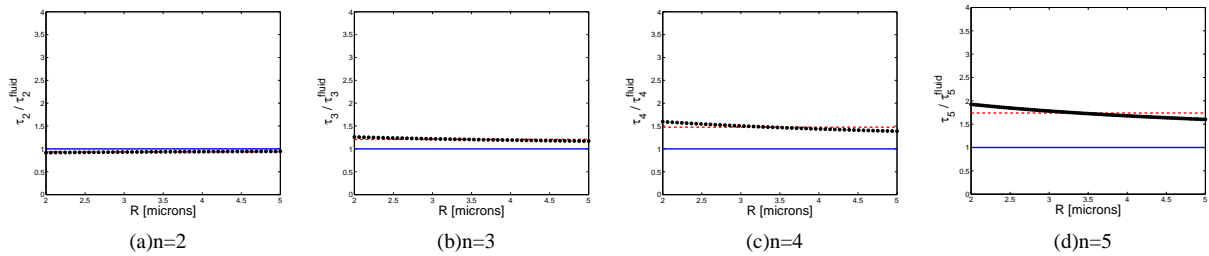


FIG. 4: Attempting to fit relaxation times given by Eq. B1 to the Stone-McConnell form (using  $\eta_f$  as the fit parameter) generates systematic deviations similar to those observed experimentally (Fig. 3).

In summary, we have shown that our prediction (Eq. B1) provides a globally satisfactory fit to the experimental data. The theory does a good job reproducing experiment over a range of domain sizes and all observable mode numbers with a single fit parameter ( $\eta_m$ ) used to describe each domain. By contrast, the Stone-McConnell theory provides an unsatisfactory fit to the

experimental results. Furthermore, the errors seen in the SM fits are predicted by our theoretical analysis and are fully consistent with an experimental system that behaves in accord with Eq. B1. Although we have ruled out the Stone-McConnell theory as an adequate description of the experiments solely on the basis of the data, we feel an even stronger case against this model is the fact that it requires one to assume bulk solvent viscosities that vary from domain to domain with values as high as 3 cP or three times higher than the known viscosity of water at the experimental temperatures (see the next section). It is sensible that individual domains sampled from different vesicles may have different membrane surface viscosities and line tensions since the lipid stoichiometry is variable from vesicle to vesicle and each domain thus represents a distinct physical system from the perspective of the bilayer. The bulk solvent surrounding the bilayer however is constant from domain to domain. It is impossible to reconcile the experimental data with the Stone-McConnell form (Eq. C3), either from the physical or statistical perspectives.

#### Supplement D. ADDITIONAL DATA

We present twelve additional data traces, to indicate the quality of fits involved (Fig. 5). Each of the plots is similar to Fig. 3 of the main paper, which is seen to be in no way extraordinary in comparison to the behavior of other domains. In each of these, the solid blue line indicates the Stone-McConnell result with  $\eta_f = 1$  centipoise (cP), and line tension as determined from the equilibrium measurement. The experimental relaxation times are almost universally significantly longer than the Stone-McConnell prediction. The red line indicates the best fit to our general form, Eq. B1, with the line tension given by the equilibrium measurements. The best fit value of the membrane surface viscosity, as well as the equilibrium line tension, are given in each figure. Plotted in green is the best fit to the Stone-McConnell form by adjusting the bulk solvent viscosity; the required bulk viscosities range from 130% to 300% of the known value of 1 cP at 20° C. Also, the deviations observed in Fig. 3 can be seen; the Stone-McConnell result does not have the correct slope to fit the observed data, though this is more apparent in aggregate (as in the preceding section) than in any individual trace.

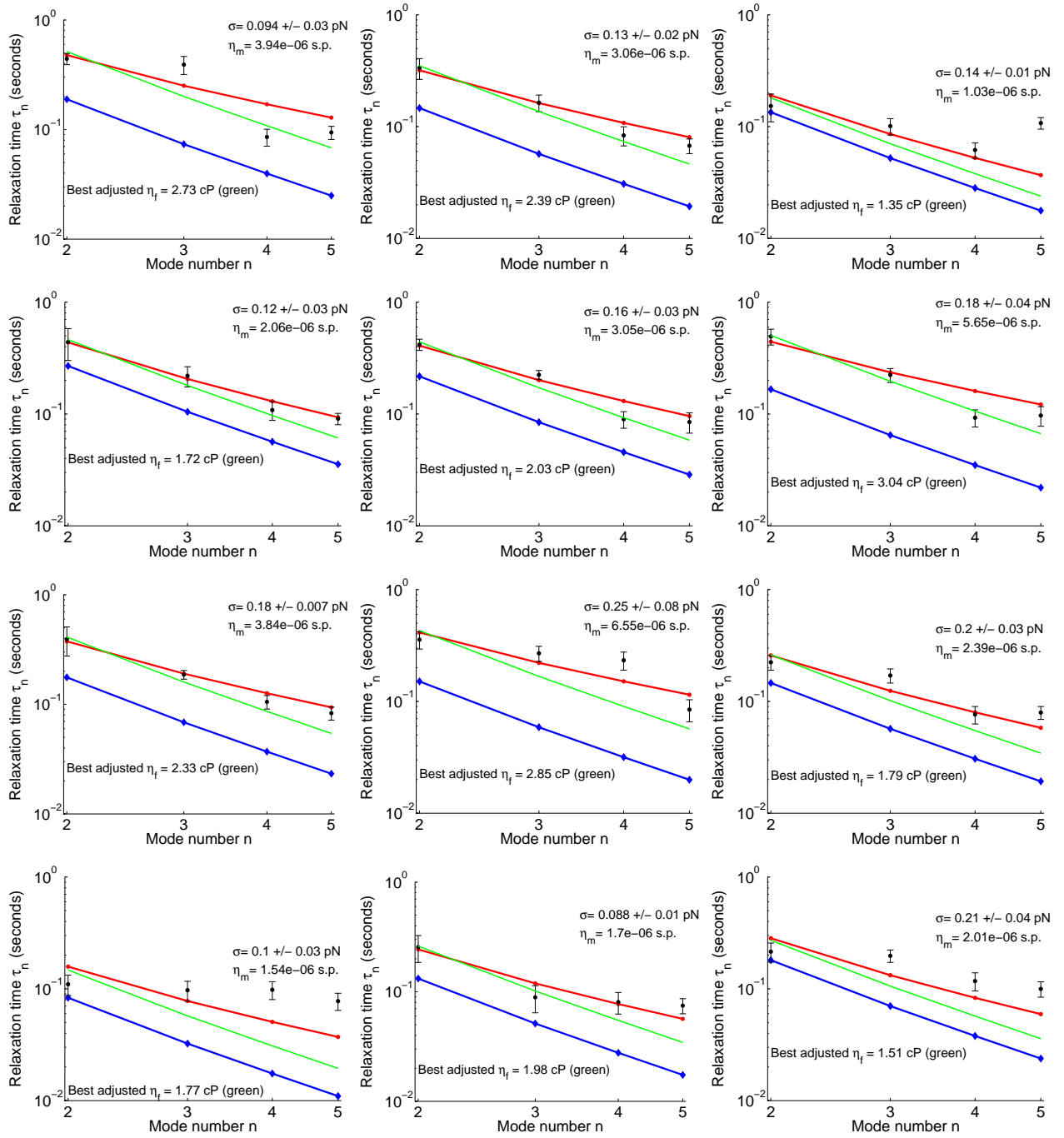


FIG. 5: Data from additional domains.

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