Effective Area-Elasticity and Tension of Micro-manipulated Membranes

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We evaluate the effective Hamiltonian governing, at the optically resolved scale, the elastic properties of micro-manipulated membranes. We identify floppy, entropic-tense and stretched-tense regimes, representing different behaviors of the effective area-elasticity of the membrane. The corresponding effective tension depends on the microscopic parameters (total area, bending rigidity) and on the optically visible area, which is controlled by the imposed external constraints. We successfully compare our predictions with recent data on micropipette experiments.

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Micro-manipulation techniques, where, e.g., magnetic or optical traps pull on micrometric beads attached to a material, are increasingly used to probe the elastic response of various soft-matter systems, such as biological polymers (e.g., DNA \cite{1} or proteins \cite{2}), phase boundaries of Langmuir monolayers \cite{3}, surfactant vesicles \cite{4} and even living cells \cite{5}. Combined with optical observations, these techniques should allow in the future to finely test the elastic theories of complex systems, e.g., by monitoring the shape and fluctuations of a system while imposing inhomogeneous boundary conditions. Although surfactant membranes and vesicles \cite{4,5} are apparently one of the simplest system, there is still some confusion regarding the appropriate Hamiltonian describing their elasticity, and in particular about the role and value of an effective tension $\sigma$. The latter is not a well-identified microscopic quantity, contrary to the bending rigidity $\kappa$. In different thermodynamic ensembles \cite{8,13–16}, various approaches have been proposed: phenomenological self-consistent theories \cite{9–11}, approximations involving Lagrange multipliers \cite{14,15}, or formal renormalization schemes \cite{8,5,21}. In this Letter, we propose an approach based on a large but finite coarse-graining \cite{14,15}, devised for the interpretation of measurements combining micro-manipulation and optical observations.

Fluid membranes in aqueous solutions often consist of a fixed number of highly insoluble lipid or surfactant molecules. Since stretching a flat membrane involves very high energies, while macroscopically bending it involves energies of order $k_B T$, membranes are commonly modeled as fluctuating two-dimensional sheets with a prescribed microscopic area $A$ and a curvature elasticity $\kappa$. At the macroscopic scale, membranes actually appear very different: their optically visible area $A$ fluctuates about some value depending on the temperature and on the external constraints. Part of the total area $\bar{A}$ is stored in short scale fluctuations that are optically unresolved \cite{8,14}. For such a critically fluctuating system, a coarse-grained effective Hamiltonian $\mathcal{H}_{\text{eff}}$, integrating all sub-optical details, is clearly more adequate than the microscopic Hamiltonian.

Our goal is to calculate this effective macroscopic Hamiltonian $\mathcal{H}_{\text{eff}}$ and to investigate the associated area-elasticity and tension. We start by considering a quasi-planar membrane with a fixed microscopic area $\bar{A}$, which is attached to a fixed frame of area $L^2$. We choose the simplest microscopic curvature Hamiltonian: the lowest-order, quadratic approximation of the Canham-Helfrich Hamiltonian \cite{13,14}:

$$\mathcal{H}_c[h_m] = \int d^2 x \frac{\kappa}{2} (\nabla^2 h_m)^2 . \tag{1}$$

Here $h_m(x)$ is the height of the membrane above a reference plane (Monge gauge), as resolved microscopically. We then determine the coarse-grained Hamiltonian $\mathcal{H}_{\text{eff}}[h]$, where $h(x)$ is the height of the membrane as resolved optically. This Hamiltonian is such that $\exp(-\beta \mathcal{H}_{\text{eff}}[h])$ gives the probability for the occurrence of any optically visible membrane shape $h$, whatever its fluctuating microscopic detail. Technically, this is a one-step renormalization of the fixed area constraint.

We find that $\mathcal{H}_{\text{eff}}$ involves a non-linear area-elasticity energy $\mathcal{H}_a(A)$ for the coarse-grained, optically visible, area $A$. This is the effective potential which is probed by pulling a membrane in an optically resolved micro-manipulation. Depending on the microscopic excess area $\alpha_m = (\bar{A} - L^2)/L^2$ and on the constraints exerted on the membrane, we find three distinct regimes: a floppy regime, an entropic-tense regime, and a stretched-tense regime. We provide explicit formulae for the effective tension $\sigma(A) \equiv d\mathcal{H}_a/dA$ in these three regimes. To better describe the tense regime, we further incorporate a microscopic stretching elasticity. We then contrast our approach and the resulting picture with the common use of a heuristic tension proposed by Helfrich and Servuss \cite{9}.

Eventually, we point out that our results can be applied to giant vesicles with a fixed volume $V_0 = \frac{4}{3} \pi R_0^3$, by taking $L^2 = 4 \pi R_0^2$. This allows us to perform a first test of our theory: re-analyzing the micro-pipette experiments of Evans and Rawicz \cite{10,22}, we find an excellent fit for the cross-over between the entropic-tense and the stretched-tense regimes. Finally, we point out the ap-
Fourier space, we can rewrite Eq. (2) as

$$\lambda \lesssim q < \Lambda$$

where \(\Lambda\) corresponds to the experimental limit of resolution, e.g., optical, and \(h\) has wavevectors in the range \(0 < q < \Lambda\). Similarly, an initially “floppy” membrane \((a_1)\) can be externally led into a tense regime \((a_2)\).

proximations involved in our calculations and we propose several possible experiments.

The thermal fluctuations of a membrane of fixed microscopic area \(A \approx \int d^2 x \left(1 + \frac{1}{2} (\nabla h_m)^2\right)\) are described by the partition function

$$Z = \int \mathcal{D}[h_m] \exp \left\{ -\beta [\mathcal{H}_c[h] + \mathcal{H}_s(A)] \right\}$$

where \(A[h] = L^2 + \int d^2 x \frac{1}{2} (\nabla h)^2\) is the optically visible area, and

$$\mathcal{H}_s(A) = -\frac{1}{\beta} \int_{-\infty}^{\infty} d\lambda \exp \left\{ -\lambda (A - A) - \frac{L^2}{2} \int q^2 \ln (\beta \kappa q^4 - \lambda q^2) \right\}.$$ 

This defines the effective Hamiltonian at the optical scale \(\mathcal{H}_e[h] = \mathcal{H}_c[h] + \mathcal{H}_s(A[h])\). In the thermodynamic limit, the above integral can be evaluated at the saddle point:

$$\mathcal{H}_s(A) \approx \frac{\lambda}{\beta} (A - A) + \frac{L^2}{2\beta} \int q^2 \ln (\beta \kappa q^4 - \lambda q^2),$$

with \(\lambda\) the solution of \(A - A = \frac{1}{2} L^2 \int q^2 (\beta \kappa q^4 - \lambda q^2)^{-1}\). The effective tension for an optical area \(A\), \(\sigma(A) = d\mathcal{H}_s/dA = -\lambda/\beta\), is thus related to the area stored in the sub-optical modes by:

$$\frac{A - A}{L^2} \approx \frac{1}{8\pi \beta \kappa} \ln \frac{\kappa \Lambda^2 + \sigma}{\kappa \Lambda^2 + \sigma}. $$

Integrating, we obtain an explicit formula for the surface potential (see Fig. 3):

$$\mathcal{H}_s(A) = \frac{L^2 \Lambda_0^2}{8\pi \beta} \left[ \left( \frac{\Lambda^2}{\Lambda_0^2} - 1 \right) \ln (e^X - 1) + X \right],$$

where \(X = 8\pi \beta \kappa (A - A)/L^2\). \(\mathcal{H}_s(A)\) has a minimum for

$$A^* = A - \frac{L^2}{4\pi \beta \kappa} \ln \frac{\Lambda_0}{\Lambda},$$

at which the tension \(\sigma\) vanishes. For \(A < A^*\), the tension saturates to the negative value \(\sigma \approx -\kappa \Lambda^2\), while for \(A \rightarrow A^-\) it diverges as \(\sigma \approx (\Lambda_0^2/L^2)/[8\pi \beta (A - A)]\), as a consequence of the prescribed microscopic area \(A\).

Floppy and tense regimes. – A crucial remark is that the optical area \(A\) cannot be smaller than the frame area \(L^2\), which results in two possibilities (Fig. 3). If the microscopic excess area \(\alpha_m = (A - L^2)/L^2\) is larger than

$$\alpha_m = \frac{1}{4\pi \beta \kappa} \ln \frac{\Lambda_0}{\Lambda},$$

we have \(A^* > L^2\) and the minimum of \(\mathcal{H}_s(A)\) is indeed physically realizable [Fig. 2 (a1)]. The coarse-grained unperturbed membrane is then in a floppy state [Fig. 1 (a1)], in which the tensions has a vanishing optimal value.
Conversely, if the attachment to the frame results in \( \alpha_m < \alpha_m^* \), the unphysical hatched region of Fig. 2 comes to the right of the minimum, and the zero-tension state is no longer accessible. The unperturbed coarse-grained membrane finds then its minimum energy in a tense, optically flat state, with \( A = L^2 \) [see Figs. 2(b1) and 1(b1)]. Two tense regimes are however possible: (i) when \( \kappa L^2 \ll \sigma \ll \kappa A_0^2 \), the membrane is in an entropic-tense state with (still in the flat state):

\[
\sigma_0 \simeq \kappa A_0^2 \exp (-8\pi\beta\kappa\alpha_m),
\]

according to Eq. (3). While \( \kappa A_0^2 \) compares with ordinary liquids tensions, the exponential reduction factor in Eq. (11) leads to extraordinary small values of membrane tensions, in line with many observations [22] (ii). When \( \sigma \) compares with \( \kappa A_0^2 \), due to the smallness of \( \alpha_m \), the membrane is in a stretched-tense regime, where the tension describes the divergence of \( \mathcal{H}_s \) for \( A \to A^* \), i.e., \( \sigma \simeq A_0^2/(8\pi\beta\kappa\alpha_m) \). (This behavior will be corrected next by the introduction of a microscopic stretching elasticity.)

A remarkable point is that in the tense states, \( \sigma \) is independent of the coarse-graining scale, \( \Lambda \). Besides, since \( \alpha_m < \alpha_m^* \), the characteristic length \( \lambda = (\kappa/\sigma)^{1/2} \) is always smaller than the optical cutoff, \( \Lambda^{-1} \), thus the bending rigidity is masked by the effective tension.

If now the membrane is stretched by external means, the typical value of \( A \) is increased compared to its free value (\( A^* \) or \( L^2 \)), and the system can be brought into tenser regimes. The response to weak perturbations of an initially floppy membrane [\( \alpha_m > \alpha_m^* \), Figs. 1(a1) and 2(a1)] is described, from Eq. (3), by a quadratic elasticity around the minimum \( A^* \):

\[
\mathcal{H}_s(A) \simeq \frac{1}{2} k_{\text{eff}} L^2 \left( \frac{A - A^*}{L^2} \right)^2, \quad k_{\text{eff}} = 8\pi\beta\kappa L^2,
\]

Under stronger stretching, the membrane reaches an entropic-tense state [Figs. 1(a2) and 2(a2)]. Its effective elasticity is then similar to that of an initially tense membrane. According to Eq. (12), the tension is given by

\[
\sigma \simeq \sigma_0 \exp (8\pi\beta\kappa\alpha_m),
\]

where \( \alpha = (A - L^2)/L^2 \) is the apparent excess area. Eventually, further pulling brings the membrane into the stretched-tense regime with \( \sigma \simeq \kappa A_0^2/[8\pi\beta(\alpha_m - \alpha)] \).

Including Stretching Elasticity. – A better description of a strongly stretched membrane can actually be achieved by taking into account the small but finite extensibility at microscopic scales. We remove the delta-function in Eq. (3), and replace the Hamiltonian by

\[
\mathcal{H}_c + \frac{k_m}{2\Lambda} \left[ L^2 + \int d^2x \left( \frac{1}{2} (\nabla h)^2 - \bar{A} \right)^2 \right].
\]

Applying a Hubbard-Stratonovich transformation [7], we obtain Eq. (4) with an additive correction \( \frac{1}{2}(\bar{A}/\beta k_m)^2 \) in the exponential. Thus, instead of Eq. (3), the saddle-point equation becomes (with \( \sigma = -\lambda_\lambda/\beta \)):

\[
\bar{A} \left( 1 + \frac{\sigma}{k_m} \right) - A = \frac{L^2}{8\pi\beta\kappa} \ln \frac{\kappa A_0^2 + \sigma}{\kappa\Lambda^2 + \sigma}.
\]

Integrating this equation leads to an improved form for \( \mathcal{H}_s(A) \). Clearly, as long as \( \sigma \ll k_m \), Eqs. (3) and (14) are equivalent. The present correction is useful only to describe the cross-over to and the tense-stretched regime \( \sigma \) comparable to or larger than \( \min(k_m, \kappa A_0^2) \), when it modifies the divergence of \( \mathcal{H}_s \) for large values of \( A \).

Comparison with the model of Helfrich and Servuss. – In Ref. [8], the membrane is macroscopically depicted as a flat surface (\( A \equiv L^2 \)), and \( \sigma \) is introduced as a microscopic surface tension, which is self-consistently determined by prescribing the average value of the fluctuating microscopic excess-area (in the ensemble in which the frame is fixed). Then the contribution of the stretching elasticity is added by hand. In our theory, we take into account the actual microscopic membrane elasticity, and we define the effective tension \( \sigma(A) \) as the derivative of the area elasticity \( \mathcal{H}_s(A) \) associated with the coarse-grained membrane area \( A \). Although Eqs. (3) and (14) are very similar to those derived in Ref. [8], our clear distinction between \( L^2 \), \( A \) and \( \bar{A} \), naturally allows to describe the area elasticity of a membrane deformed by external actions (while the description of Ref. [8] allows no distinction between \( A \) and \( L^2 \)).

Quasi-spherical vesicles. – Let us now briefly discuss the case of a quasi-spherical vesicle with a fixed volume \( V_0 = \frac{4}{3}\pi R_0^3 \). Parameterizing its shape by \( R(\theta, \phi) = R_0 [1 + \sum_{\ell m} u_{\ell m} Y_{\ell m}(\theta, \phi)] \) and taking into account the volume constraint \( 4\pi R_0 u_{0,0} = -\sum_{\ell \geq 2, m} u_{\ell m}^2 \), the microscopic area is fixed, in the partition function, by the factor (see, e.g., [1]):

\[
\delta \left( 4\pi R_0^3 + R_0^2 \sum_{\ell \geq 2, m} \frac{1}{2}(\ell - 1)(\ell + 2) |u_{\ell m}|^2 - \bar{A} \right).
\]

Comparing with the delta-function in Eq. (3), we see that the area \( 4\pi R_0^2 \) plays the role of the frame area \( L^2 \). It will be shown in detail elsewhere that a coarse-graining procedure yields the same results as in the flat case, provided the vesicle is large enough for the optical wavelength to corresponds to quasi-planar modes. At the optical resolution, the vesicle still has a prescribed volume \( V \sim V_0 \) (the volume is almost unaffected by quasi-planar modes), and is described by the effective Hamiltonian \( \mathcal{H}_\text{eff} \) obtained previously, upon replacement of \( L^2 \) by \( 4\pi R_0^2 \).

To check our model, we have re-analyzed the most recent micropipette experiments [21]. A certain amount of membrane area, initially invisible at the optical resolution, is aspirated from a vesicle my means of a pressurized micropipette. The reduced tension \( \sigma \), obtained from
Laplace’s law, is measured as a function of the variation of the apparent area. Our Eq. (14) can be rewritten in the tense regime as

\[ \alpha = C_1 + (8\pi\beta\kappa)^{-1}\ln \sigma + C_3 \frac{\sigma}{k_m}, \tag{16} \]

with \( C_1 = \alpha_m - (8\pi\beta\kappa)^{-1}\ln(\kappa A_0^2) \) and \( C_3 = 1 + \alpha_m \). As shown in Fig. 3, this yields a nice fit with \( C_1 - \alpha_0 = 0.0258 \pm 0.0003 \) (with \( \alpha_0 \) the unknown expansion at lowest pressure \[23\]), \( \beta\kappa = 10.7 \pm 0.3 \), and \( k_m/C_3 = 183 \pm 6 \text{ erg/cm}^2 \). The slope in the linear low tension regime precisely determines \( \beta\kappa \). The value of \( C_1 \) is hard to interpret, since \( \alpha_0 \) and \( \alpha_m \) are unknown. An estimate of \( k_m \) can be obtained by assuming \( \alpha_m \simeq 0.05 \) (the range of the area expansion variation); this yields \( k_m \simeq 192 \text{ erg/cm}^2 \). In Ref. [21], the data was also nicely fitted with Helfrich’s formula [9], since it equally leads to Eq. (16) in the tense regime, however with different definitions for \( C_1 \) and \( C_3 \). These differences affect the determination of \( k_m \), since \( C_3 = 1 \) in Helfrich’s theory.

**Conclusion.** — Starting from a clearly defined picture at the microscopic scale (an almost incompressible membrane built of a fixed number of lipids, attached to a fixed frame, or enclosing a quasi-spherical volume), we have derived an explicit Hamiltonian \( \mathcal{H}_{\text{eff}}[h] \) describing the elasticity of a membrane, as gauged by its optically resolved shape \( h \). This effective description allows, in theory, to determine the response to an external perturbation described by a Hamiltonian \( \mathcal{H}_{\text{ext}}[h] \), e.g., to a set of pulling micron-size beads. Our formulae for the effective tension in the stretched states resemble those of Ref. [9], but our formalism offers a clearer definition, a wider applicability, and basically a justification of the concept of effective tension. This will allow us to emphasize elsewhere the distinction between the effective membrane tension and the mechanical frame tension \( \Sigma = \beta^{-1}d(\ln Z)/dL^2 \).

We have kept throughout the description at the Gaussian, quadratic level (neglecting renormalization of the bending rigidities and associated effects), however we believe that our results in their present form offer a reasonably simple frame for the analysis of experiments, including new micro-manipulation studies.

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\[ \alpha - \alpha_0 \]

FIG. 3. Fit by Eq. (16) of the data obtained in a recent micropipette experiment by the group of Evans [21] (see text).