Reading for Lectures 30-32: PKT Chapter 11 (skip Ch. 10)

Final result for membrane bending energy/unit area : $\varepsilon_s = \frac{\kappa_b}{2}\left(\frac{1}{R_1}+\frac{1}{R_2}\right)^2$

For entire membrane S (usually a closed surface): $E_b = \frac{\kappa_b}{2}\int_S dA\left(\frac{1}{R_1}+\frac{1}{R_2}\right)^2$

Compare polymer energy/unit length: $\varepsilon_p = \frac{\kappa}{2}\left(\frac{1}{R}\right)^2$ and $E_b = \frac{\kappa}{2}\int_0^L ds\left(\frac{1}{R_1}\right)^2$

Comments:

1. Scale invariance.

Bending energy depends only on the shape of the surface and not on its size.

Consider, e.g., a spherical surface, so R_1=R_2=R (sphere radius), so $E_b = \frac{\kappa_b}{2}\left(4\pi R^2\right)\left(\frac{2}{R}\right)^2 = 8\pi\kappa_b$.

This is, in fact, very general (not just for spheres):

Suppose you magnify all dimensions by a factor b but without changing the shape, then

$E_b = \frac{\kappa_b}{2}\int_{S_b} b^2 dA\left(\frac{1}{bR_1}+\frac{1}{bR_2}\right)^2$. All the factors b cancel.

2. Significance of $\kappa_b >> k_BT$ (for typical biomembranes).

Membrane shape is a low-temperature problem! (Unlike polymers.)

Membrane shape fluctuations of scale R leading to radii of curvature of scale R are strongly suppressed, so entropic effects are much less important for membranes than they are for polymers.

Comments:

- You might think that this would prevent flat, open membrane sheets from closing to form vesicles. But, this is incorrect. There is an additional large energy, λ, the edge energy. The combination will close sheets larger than nm scale (HW).
- And, the shapes of closed vesicles are largely determined by bending energy (see below).

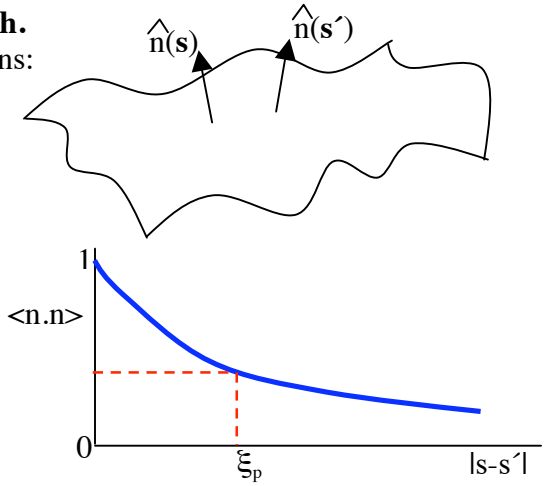
3. The de Gennes-Taupin persistence length.

Consider a large sheet with thermal fluctuations:

The normal-normal correlations function, $\langle \hat{n}(s)\cdot\hat{n}(s')\rangle$=1 when $\vec{s}=\vec{s}'$ but dies off at large separations.

The die off is NOT exponential (cf. 1D).

The persistence length is the characteristic separation for die-off to, say, 1/e.



It can be shown that $\xi_p \sim d\,e^{\frac{4\pi}{3}\cdot\frac{\kappa_b}{k_BT}}$,

where d is the membrane thickness, i.e.,

$\xi_p \sim d\,e^{\frac{4\pi}{3}\cdot\frac{\kappa_b}{k_BT}} \sim 4\,e^{100}\ nm$ (exponentially large)

Compare polymers $\xi_p \sim \frac{\kappa}{k_BT}$.

Branched-polymer instability.

This is another example of how small thermal fluctuation effects are for membranes.

We have seen in the case of E_b how bending energy/unit area goes as $\left(\frac{d}{R}\right)^2$. There are higher-order curvature effects involving higher powers of $\left(\frac{d}{R}\right)$ but these are correspondingly small for objects like the plasma membrane with μm dimensions compared to the membrane thickness. But,..

4. Gaussian curvature energy.

There is in general another contribution at order $\left(\frac{d}{R}\right)^2$: $E_g = \kappa_g \int_S dA \frac{1}{R_1 R_2}$

The quantity $K \equiv \frac{1}{R_1 R_2}$ is called the "Gaussian curvature"; the modulus κ_g can have either sign and is of the same generic magnitude as the bending modulus.

Amusingly, this term does not affect the shapes of closed objects:

The reason is that the integral of this geometrical object over a closed surface is a "topological invariant," i.e., its value depends on topology only!

Gauss-Bonnet Theorem: $\int_{\substack{S \\ closed \\ surface}} dA\, K = 4\pi(1-g)$, where g is the "genus" of the surface, i.e., the number of "handles".

Sphere: g=0; torus: g=1;… Check for sphere: $\int_{sphere} dA\, K = 4\pi R^2 \cdot \frac{1}{R^2} = 4\pi$.

5. Curvature of membranes in relaxed state.

I have argued that *symmetrical* membranes are naturally flat. What happens when the leaves are different (as is normally the case biologically)? Different numbers of different molecules.

The upshot is that the membrane develops a preferred curvature which is not zero (flat). There are two effects:

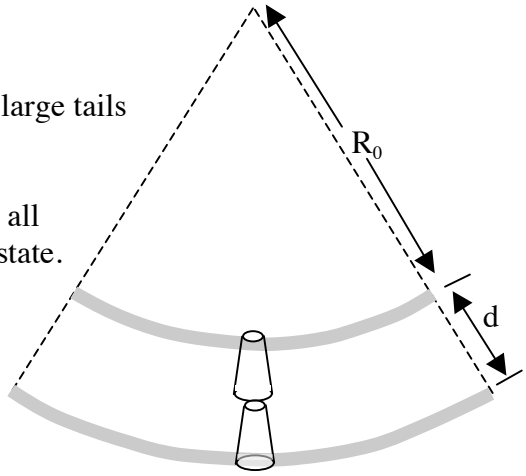
A. Spontaneous curvature, the effect of molecular shape.

Suppose on average lipids in one layer have small heads and large tails but, in the other layer, they have large heads and small tails.

Naturally, not as an effect of strain deformations (Lect. 32).

In this case, there will be a natural radius of curvature R_0 and all the bending energy costs will now be relative to this relaxed state.

This leads to a modified form of the bending energy due to Helfrich: $E_{Helfrich} = \frac{\kappa_b}{2} \int_S dA \left(\frac{1}{R_1} + \frac{1}{R_2} - C_0\right)^2$.



$C_0 = \frac{2}{R_0}$ is called the "spontaneous curvature."

Note

- $C_0>0$ allows both the R's to be positive, i.e., locally convex surfaces.
- $C_0<0$ allows both the R's to be negative, i.e., locally concave surfaces.
- $C_0=0$ allows flat surfaces at no energy cost but also allows saddle-shaped surfaces for which $\frac{1}{R_1} = -\frac{1}{R_2}$. Such surfaces are called "minimal surfaces" and can have quite complex shapes.

B. Area-difference curvature, different numbers of lipids molecules in inner and outer leaves. 33.3

For closed membranes shapes, like vesicles, the two leaves of the bilayer have no way of exchanging lipids, since thermal flip-flop is slow. This means that, once set, the number of lipids in the two leaves, N_{in} and N_{out}, do not change (at least, over short time intervals).

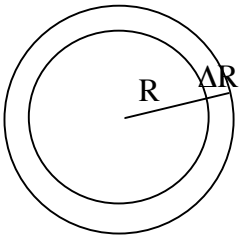
I assume here for simplicity all lipids are of one kind.

Since the relaxed areas of the leaflets are $A_{in} = N_{in}a_0, A_{out} = N_{out}a_0$, the relaxed area difference $\Delta A_0 = (N_{out} - N_{in})a_0$ remains fixed. If $\Delta A_0 = (N_{out} - N_{in})a_0 \neq 0$, the relaxed shape of the membrane cannot be flat, since the flat state would require the more-dense leaflet to be a little compressed and the less dense leaflet to be a little expanded.

Upshot: The leaflet with more area will always want to be on the outside of the curve.

Analog: Bimetallic strip.

More generally, there is always an area difference between the two leaflets of a curved surface with ΔR separation. Consider the sphere:



$$A + \Delta A = 4\pi(R + \Delta R)^2$$

$$A = 4\pi R^2$$

$$\boxed{\Delta A = 8\pi R \Delta R}$$

The general expression is $\Delta A[S] = \Delta R \int_S dA\, 2H = \Delta R \int_S dA \left(\frac{1}{R_1} + \frac{1}{R_2}\right)$, where H is the mean curvature.

Check that this works for the sphere: $\Delta A = \Delta R\left(4\pi R^2\right)\cdot\left(\frac{1}{R} + \frac{1}{R}\right) = 8\pi R \Delta R$.

The membrane is in a "relaxed" state iff the preferred area difference ΔA_0 matches the actual (shape-dependent) area difference $\Delta A[S]$. If this is not so, then there is an additional elastic energy cost in forming the membrane into the shape S. This cost is, again, related the stretching modulus K_s.

Upshot: The is another contribution to the shape energy,

$E_{area-difference} = \frac{\bar{\kappa}}{2}\frac{\pi}{A_0 d^2}\left(\Delta A[S] - \Delta A_0\right)^2$, with $\bar{\kappa}$ comparable in magnitude to κ_b.

The upshot of a calculation which I will not show you is that this effect can be completely described as an addition to the spontaneous curvature: $C_0 \rightarrow C_0^{eff} = C_0 - \frac{2\pi}{dA_0}\frac{\bar{\kappa}}{\kappa_b}\left(\Delta A[S] - \Delta A_0\right)$.

Message: Adding extra lipids to one of the two leaves of the bilayer is a way of manipulating the effective spontaneous curvature.

Shapes of Vesicles and Red Blood Cells:

Both vesicles produced in the lab and red cells adopt a shape which minimizes the membrane energy.

That shape depends on:

- the area of the membrane (set by the number of molecules it contains)
- the enclosed volume (set osmotically)
- the moduli κ_b and $\bar{\kappa}$
- the spontaneous curvature C_0 and the preferred area ΔA_0

What kinds of shapes do you find?

Let's imagine starting with the maximum volume for given area.

That's a sphere and there is only one shape possible.

Imagine reducing the volume a bit.

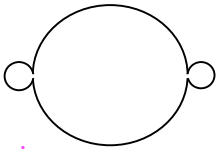
Now it makes a difference whether the spontaneous curvature favors convex or concave surfaces.

- If $C_0^{eff}<0$, then, as soon as there is sufficient free surface, the minimum-energy shape will begin to show a cup-shaped or invaginated form. **33.4**
- If $C_0^{eff}>0$, then, as soon as there is sufficient free surface, the minimum-energy shape will begin to try to bulge out and achieve a radius of curvature R_0.

Interestingly, for R_0 sufficiently small, the minimum energy shape becomes vesiculated/budded, with two (or more) small buds connected by a narrow neck (or necks) to a more-or-less spherical central region.

You might think that this could not be a low-energy shape because the radii of curvature in the neck region are very small (so 1/R is very large).

But, notice that the two radii of curvature are opposite in sign.



Note narrow necks!

In fact, the neck shape adjusts to be very close to $\left(\frac{1}{R_1}+\frac{1}{R_2}-C_0^{eff}\right)=0$.

In fact for very narrow necks, the contribution of the neck region to the bending energy of the entire shape goes to zero.

So far I have been talking about artificial vesicles, which can be made in the lab.

What does this have to do with red blood cell shapes? (see figure on next page)

You never see red-blood-cell shapes with buds!

Red blood cells have exactly this kind of elastic energy.

But, and in addition, they have a stretching and shear energy associated with the membrane skeleton. This cytoskeletal is very soft (elastic moduli small) and for shapes that are smooth, the cytoskeletal contributions are small and do not play an important roll.

But, there is one important exception: When buds try to form, there is a lot of shear deformation in the narrow-neck region, so the effect of the cytoskeletal contribution is to turn buds into "spicules."

The stomatocyte-discocyte-echinocyte sequence of the human red blood cell:

Normal red cells are biconcave discs.

However, if change the relative area of the inner and outer leaflets of the plasma membrane, you can adjust the value of $C_0^{eff}=C_0-\frac{2\pi}{dA_0}\frac{\bar{\kappa}}{\kappa_b}\left(\Delta A[S]-\Delta A_0\right)$ and, thereby, change the shape. A number of different agents affect this relative area. Since the effect of these agents on the shape takes place only through C_0^{eff}, any such agent drives the red cell through the same sequence of shapes, either decreasing C_0^{eff} (stomatocytic) or increasing it (echinocytic).

What are the different agents?

Stomatocytic:	**Echinocytic:**
positively charged amphiphilic drugs	negatively charged amphiphilic drugs
Cholesterol depletion	Cholesterol addition
low salt (hypotonic saline)	high salt (hyprtonic saline)
Low pH	High pH
	Proximity to glass

Some of these mechanisms are understood; others remain conjectural.

Example:

Amphiphile effect: Inner leaflet of pm has preponderance of lipids which carry negative charge, while outer-leaf lipids are neutral. Positively charged drugs are attracted to the inner leaflet and

increase its area, thereby favoring invagination. Negatively charged drugs are repelled and end up in the outer leaflet, thereby favoring echinocytes.

33.5

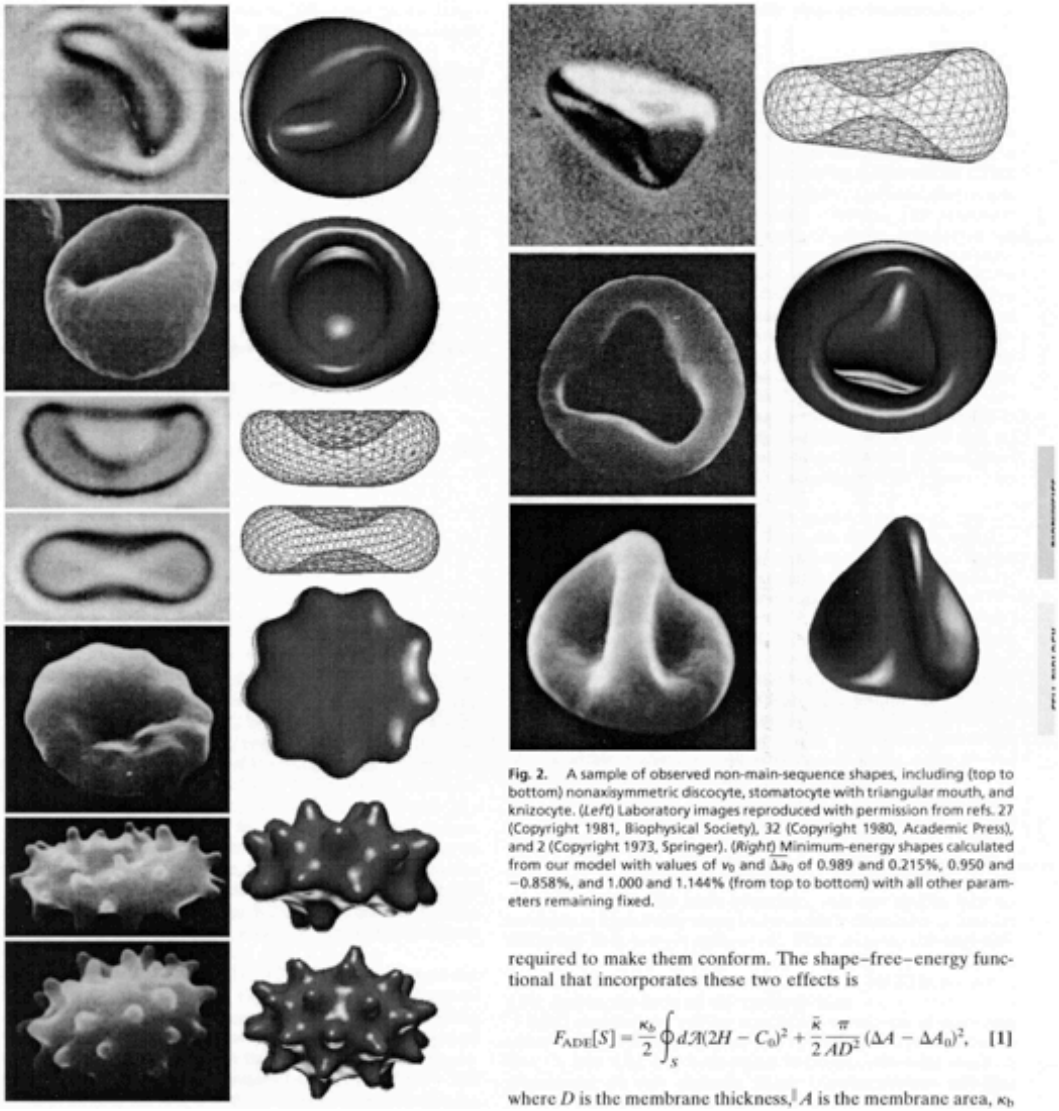


Fig. 2. A sample of observed non-main-sequence shapes, including (top to bottom) nonaxisymmetric discocyte, stomatocyte with triangular mouth, and knizocyte. (*Left*) Laboratory images reproduced with permission from refs. 27 (Copyright 1981, Biophysical Society), 32 (Copyright 1980, Academic Press), and 2 (Copyright 1973, Springer). (*Right*) Minimum-energy shapes calculated from our model with values of v_0 and $\overline{\Delta a_0}$ of 0.989 and 0.215%, 0.950 and −0.858%, and 1.000 and 1.144% (from top to bottom) with all other parameters remaining fixed.

required to make them conform. The shape–free–energy functional that incorporates these two effects is

$$F_{\mathrm{ADE}}[S] = \frac{\kappa_b}{2}\oint_S dA(2H - C_0)^2 + \frac{\bar{\kappa}}{2}\frac{\pi}{AD^2}(\Delta A - \Delta A_0)^2, \quad [1]$$

where D is the membrane thickness,‖ A is the membrane area, κ_b

Lim, Wortis, Mukhopadhyay, PNAS 99, 16766 (2002)

Problems:
derive 2D PB from “geometry” and solve: DNA connection? Manning condensation?
Compare 1D PB exact solution from class with linearized form
linearize PB to Guoy-Chapman
solve PB for isolated ion or charged rod (DNA)?
Osmotic lysis of rbc
look at PKT problems in both chapters
give problem with motion reversal invariance and not?
sheet closing using edge energy