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Supplementary material

EXPLORING THE BINDING DYNAMICS OF BAR PROTEINS

DORON KABASO[1]*, EKATERINA GONGADZE[2], JERNEJ JORGAČEVSKI[3], MARKO KREFT[3], URSULA VAN RIENEN[2], ROBERT ZOREC[3] and ALEŠ IGLIČ[1]

[1]Laboratory of Physics, Faculty of Electrical Engineering, University of Ljubljana, Tržaška 25, SI-1000 Ljubljana, Slovenia, [2]Institute of General Electrical Engineering, University of Rostock, Albert-Einstein-Straße 2, 18051 Rostock, Germany, [3]Laboratory of Neuroendocrinology and Molecular Cell Physiology, University of Ljubljana, Zaloška 4, & Celica Biomedical Center, Tehnoloski park 24, Ljubljana, Slovenia

THE MODEL

The model hypotheses are that 1) the binding dynamics of a lipid bilayer to a BAR domain surface is driven by the coupling between the intrinsic curvature of negatively charged lipids and the membrane curvature, and that 2) the attraction between negatively charged lipids and positively charged amino acids of a BAR domain contributes a negative binding energy to the system free energy, further facilitating the bending of the membrane towards the BAR domain surface. We constructed a model to study the dynamic binding of a lipid bilayer to an I-BAR of positive intrinsic curvature or an F-BAR domain of negative intrinsic curvature. The model is a variation of previous theoretical work [1-3], and the present aim is to test whether the large intrinsic curvature of negatively charged lipids could facilitate the binding of a lipid bilayer to both I-BAR and F-BAR crescent-like shape surfaces.

Our model is a coarse-grained model, whereby we do not describe the details of the molecular scale level. The minimal length scale along the membrane that is relevant to this model is of the order 0.1 nm. The model is written as a set of equations of motion for the continuum fields that describe the membrane shape and density of negatively charged lipids, including the actual forces acting on the membrane, and the details of the membrane elasticity. For the sake of simplicity, the modeled membrane contour of a typical cell is considered to be nearly flat.

The bending energy part of the free energy of the model is based on the Helfrich energy form [4]. The free energy expression is:

$$F = \int (\frac{1}{2}\kappa(2H - \bar{H}n)^2 + \sigma_0 - \alpha n + kTn_s n(\ln(n) - 1))ds, \quad \text{(S1)}$$

where κ is the membrane bending rigidity, n is the area fraction density of negatively charged lipids, $\bar{H}$ is the effective intrinsic curvature of negatively charged lipids, H is the local mean membrane curvature, n_s is the saturation density of negatively charged lipids, σ_0 is the membrane surface tension, α is the binding energy, k is the Boltzmann constant, T is temperature, and $ds = d_m \cdot dl$ is an element of membrane area, where d_m is the dimension of the membrane perpendicular to the contour and dl is a line element along the contour.

The first term describes the bending energy due to the mismatch between the membrane curvature and the spontaneous curvature of negatively charged lipids. The second term describes the surface tension energy. The third term describes the negative binding energy of negatively charged lipids to positively charged regions along the BAR domain, which is estimated from the electrostatic attraction. The fourth term describes the entropic contribution due to the lateral thermal motion of the negatively charged lipids in the membrane in the limit of small n. The derivation leading to Eq. S1 is a one-dimensional version of the more general expressions derived in previous studies [5]. These expressions recover the familiar form for small undulations of a flat membrane in the Monge gauge form.

DERIVATION OF THE CURVATURE FORCE ON THE MEMBRANE

We derive the equations of motions of the membrane contour using the derivation of the free energy (Eq. S1) with respect to the membrane coordinate and the concentration of negatively charged lipids [6]. To take into account the drag due to viscous forces, we assume for simplicity only local friction forces [1,6], with coefficient ξ .

The equation of motion of the membrane is

$$\xi \frac{\partial \mathbf{r}}{\partial t} \cdot \mathbf{n} = -\frac{\delta F(s)}{\delta \mathbf{n}} \quad \text{(S2)}$$

where ξ is the coefficient of the local friction force due to viscous drag of the fluids surrounding the membrane, $\mathbf{r}$ is the radial vector in the (x, y) coordinate system, t is time, $\mathbf{n}$ is the normal direction, and $\frac{\delta F(s)}{\delta \mathbf{n}}$ is the derivation of Eq. 1 with respect to the x and y directions.

Here we consider only the changes along the y direction. The derivation of the free energy is projected to give the forces normal to the membrane contour [6]. The following is the list of parameter values incorporated in our model:

ξ = 1 s^{-1}g, D = 0.002 $\mu m^2 s^{-1}$, Λ = D/ $_{kT}$, α = 5 $_{kT}$, n_s = 1500 μm^{-2}, κ = 100 $_{kT}$, $\bar{H}$ = 100 μm^{-1}, σ 0.1 $_{kT}$, where $_{kT}$ is the thermal energy.

We next derive the forces at the membrane, by treating it as a "one-dimensional membrane", i.e. a thin strip of width *w*, with a bending modulus and tension coefficient. The free energy of this membrane is given in Eq. S1, and is used to derive the local restoring forces by the usual derivation method. Since the overall contour length is not constant in our system, the derivation of the coordinates has to be taken with respect to their absolute index *u* along the contour, which is constant. In these terms the curvature *H* appearing in the Helfrich part of the free energy [4] is written as (standard differential geometry)

$$H = \frac{\frac{dx}{du}\frac{dy^2}{du^2} - \frac{dx^2}{du^2}\frac{dy}{du}}{\sqrt{\frac{dx}{du}^2 + \frac{dy}{du}^2}}, \qquad \text{(S3)}$$

where the $\frac{d}{du}$ symbol denotes differentiation with respect to the index of the point along the contour, and the free energy is

$$F = w\int^{1}(\frac{1}{2}\kappa H^2 + \sigma)\sqrt{\frac{dx}{du}^2 + \frac{dy}{du}^2}\, du, \qquad \text{(S4)}$$

where $\frac{ds}{du} = \sqrt{\frac{dx}{du}^2 + \frac{dy}{du}^2}$. The derivation of this free energy gives the forces, for example in the *x* direction

$$F_x = -\frac{\delta F}{\delta x} = \frac{d}{du}\frac{\delta F}{\delta \frac{dx}{du}} - \frac{d^2}{du^2}\frac{\partial F}{\partial \frac{d^2x}{du^2}}. \qquad \text{(S5)}$$

The resulting equations of motion from this derivation give very long expressions, that are not amenable to easy analysis, although they can be used for the numerical simulations. In order to arrive at simpler expressions we will develop the terms in Eq. S5, and simplify at the end by assuming that the arc-length separation between the nodes along the contour are all the same. This is maintained as the simulation progresses by using the spline routine to re-discretize evenly the contour as its length evolves.

The first term on the r.h.s. of Eq. S5 is

$$\frac{\delta F}{\delta \frac{dx}{du}} = H^2\frac{\delta x}{\delta s} + 2H\frac{\partial H}{\partial \frac{dx}{du}}\frac{ds}{du}, \qquad \text{(S6)}$$

where $\frac{\delta \frac{dx}{du}}{\delta \frac{ds}{du}} = \frac{dx}{du} / \sqrt{\frac{dx}{du}^2 + \frac{dy}{du}^2}$.

$$\frac{d}{du}\frac{\delta F}{\delta \frac{dx}{du}} = 2\frac{ds}{du}HH'x' + \frac{ds}{du}H^2x'' + 2\frac{ds}{du}^2 H'\frac{\partial H}{\partial \frac{dx}{du}} + 2H\frac{\partial H}{\partial \frac{dx}{du}}\frac{d^2s}{du^2} + 2H\frac{ds}{du}^2\frac{\partial H'}{\partial \frac{dx}{du}} \quad \text{(S7)}$$

where $\frac{\partial H}{\partial \frac{dx}{du}} = \frac{d^2y}{du^2}\frac{ds}{du}^3 - 3x'H / \frac{ds}{du}$. We therefore need to find an expression for $\frac{d^2y}{du^2}$ (and $\frac{d^2x}{du^2}$), by using the definition of $H = (x'\frac{d^2y}{du^2} - y'\frac{d^2x}{du^2}) / \frac{ds}{du}^2$ and: $\frac{d^2s}{du^2} = x'\frac{d^2x}{du^2} + y'\frac{d^2y}{du^2}$. The final expressions that we get are: $\frac{d^2y}{du^2} = \frac{d^2s}{du^2}y' + \frac{ds}{du}^2 x'H$, and $\frac{d^2x}{du^2} = \frac{d^2s}{du^2}x' - \frac{ds}{du}^2 y'H$. We will now assume that $\frac{ds}{du}$ is independent of *u*, so that $\frac{d^2s}{du^2} = \frac{d^3s}{du^3} = \ldots = 0$. The last term in Eq. S7 becomes

$$\frac{\partial H'}{\partial \frac{dx}{du}} = -\frac{2}{\frac{ds}{du}}\frac{\partial}{\partial s}(x'H). \quad \text{(S8)}$$

The second term on the r.h.s. of Eq. S5 is

$$\frac{\partial F}{\partial \frac{d^2x}{du^2}} = \frac{-2Hy'}{\frac{ds}{du}} \quad \text{(S9)}$$

and we get

$$\frac{d^2}{du^2}\frac{\partial F}{\partial \frac{d^2x}{du^2}} = -2\frac{ds}{du}\frac{\partial^2}{\partial s^2}(Hy'). \quad \text{(S10)}$$

We now write *H* in terms of *s* as: $H = \vec{H}\cdot\hat{n} = -x''y' + y''x'$, where $\vec{H} = x''\hat{x} + y''\hat{y}$, and $\hat{n} = -y'\hat{x} + x'\hat{y}$ so that

$$H' = -y'x''' + x'y''' \quad H'' = -y''x''' + x''y''' - y'x^{(4)} + x'y^{(4)} \quad \text{(S11)}$$

There is another force contribution from the membrane tension, giving a term of the form: $F_x \propto \frac{ds}{du}x''$, so in the normal direction we get:

$F_n = \frac{ds}{du}\sigma(-y'x''+x'y'') = \frac{ds}{du}\sigma H$, where we used the identity:

$x'^2+y'^2=1$ and therefore: $2(x'x''+y'y'')=\partial(x'^2+y'^2)/\partial s=0$.

Putting everything together, the normal force acting on the membrane due to curvature and tension is

$$F=\vec{F}\cdot\hat{n}=-y'F_x+x'F_y \quad \text{(S12)}$$

$$=\frac{ds}{du}\frac{1}{2}\kappa\left(-2\nabla^2H-2H(y'y'''+x'x''')-3\left(\vec{H}\cdot\hat{n}\right)^3\right)+\frac{ds}{du}\sigma H$$

$$=\frac{ds}{du}\kappa\left(-\nabla^2H-\frac{1}{2}\left(\vec{H}\cdot\hat{n}\right)^3\right)+\frac{ds}{du}\sigma H \quad \text{(S13)}$$

$$=\frac{ds}{du}\frac{1}{2}\kappa\left(2(y'x^{(4)}-x'y^{(4)})-3\left(\vec{H}\cdot\hat{n}\right)^3\right)+\frac{ds}{du}\sigma H$$

where we used the identity: $x'^2+y'^2=1$, and therefore:

$2(x'x''+y'y'')=\partial(x'^2+y'^2)/\partial s=0$, and

$(y'y'''+x'x''')=-(x''^2+y''^2)=-H^2$.

Since we need forces per unit length, while we calculated above the forces per unit *u*, we divide by $\frac{ds}{du}$ and finally get $\kappa((y'x^{(4)}-x'y^{(4)})-\frac{3}{2}H^3)+\sigma H$. (S14)

NEGATIVELY CHARGED LIPIDS WITH SPONTANEOUS CURVATURE

When there are negatively charged lipids with spontaneous curvature, the free energy (Eq. S4) changes to

$$F=w\int^1(\kappa(H-\bar{H}n)^2+(\sigma-\alpha n))\sqrt{\frac{dx}{du}^2+\frac{dy}{du}^2}\,du, \quad \text{(S15)}$$

where *n* is the density of negatively charged lipids along the contour, which may not be uniform. Expanding the quadratic term we get: $H^2-2H\bar{H}n+(\bar{H}n)^2$. The derivation of the first term was done above (all the variations are of the integrand times the $\frac{ds}{du}$ factor).

The new contributions to the forces acting on the membrane are (normal force per unit length)

$$F_{spon,n}=\vec{F}_{spon}\cdot\hat{n}=-y'F_{spon,x}+x'F_{spon,y} \quad \text{(S16)}$$

$$=\frac{\kappa}{2}((\bar{H}n)^2H-2(\bar{H}n'')-n\bar{H}H^2$$

and

$$F_{tension,n}=\vec{F}_{spon}\cdot\hat{n}=-\alpha nH. \quad \text{(S17)}$$

FLUXES AND DIFFUSION OF NEGATIVELY CHARGED LIPIDS

The conservation equation for the negatively charged lipids along the contour becomes

$$\frac{1}{\frac{ds}{du}}\frac{\partial \frac{ds}{du}n}{\partial t}=\frac{D}{\frac{ds}{du}}\nabla^2 s(\frac{ds}{du}n)+\frac{1}{\frac{ds}{du}}\frac{\Lambda}{n_s}\nabla s(\frac{ds}{du}n\nabla s(\frac{1}{\frac{ds}{du}}\frac{\delta e}{\delta n}), \tag{S18}$$

where the number of negatively charged lipids in each unit contour length is $N=\frac{ds}{du}nn_s$ (n_s is the saturation concentration of the negatively charged lipids), F is the energy functional of Eq. S1, and the derivative along the contour is $\nabla s=\nabla u/\frac{ds}{du}$. We therefore get

$$\frac{\partial n}{\partial t}+\frac{n}{\frac{ds}{du}}\frac{\partial \frac{ds}{du}}{\partial t}=\frac{D}{\frac{ds}{du}}\nabla^2 s(\frac{ds}{du}n)+\frac{D}{kTn_s\frac{ds}{du}^2}\nabla u(n\nabla u\frac{\delta e}{\delta n}), \tag{S19}$$

where e is the energy per unit length, i.e. the integrand in Eq. S15 with respect to ds. If the number of negatively charged lipids is conserved, even though we allow the membrane overall length to change, then Eq. S19 is correct. If however there is a reservoir of membrane that allows it to change in length, then this membrane can include lipids and negatively charged lipids, so that the total number of negatively charged lipids is not conserved when the membrane length changes. In this case the change in the density due to length changes is removed, assumed to be balanced by the currents into/out of the reservoir. Eq. S19 is then modified by removing the second term on the left hand side.

NUMERICAL REALIZATION OF THE MODEL

Discretization of the model

Since the flat shape model represents a segment of the whole cell, we used periodic boundary conditions. Thus, the number of grid points N equals the number of discretizations. In our model, the density n of element i is given by:

$$n_i=\frac{N_i}{\Delta s_i}. \tag{S20}$$

The boundary conditions

We employed periodic boundary conditions. The calculation of the first and second derivatives of the function along the x direction was performed using the following explicit Euler method:

$$\frac{\partial x}{\partial s}=\frac{x_{n+1}-x_{n-1}}{2\Delta s_i}\quad \frac{\partial^2 x}{\partial s^2}=\frac{x_{n-1}-2x_n+x_{n+1}}{\Delta s_i^2}, \tag{S21}$$

where the subscripts *n*, *n+1*, *n-1* represent the current, next, and previous nodes, respectively. The derivatives of the function along the *y* direction were calculated in a similar manner. For the calculations of derivatives of the first point, the last point was added before it, while for the calculation of derivatives of the last point, the first point was added after it.

The derivation of the free energy equation

The derivation of the free energy is projected to give the forces normal to the membrane contour [6]. We now list the forces derived from the derivation of the free energy (Eq. S4) [6]

$$F_c = \kappa(-\nabla^2 H + \bar{H}\nabla^2 n + \frac{1}{2}n^2\bar{H}^2 H - \frac{1}{2}H^3) \quad \text{(S22)}$$

$$F_t = (\sigma - \alpha n)H \quad \text{(S23)}$$

$$F_e = kTn_s(n\ln(n) - 1)H \quad \text{(S24)}$$

where F_c is the force due to the curvature energy mismatch between the membrane curvature and the spontaneous curvature of the negatively charged lipids, and F_t is the membrane tension force. F_e arises from the entropy of the negatively charged lipids in the membrane, which acts to expand the length of the contour.

We now calculate the dynamics of the negatively charged lipid density, using the following conservation equation:

$$\frac{\partial n}{\partial t} = -\nabla\vec{J} = \frac{\Lambda}{n_s}\nabla s\left(n\nabla s\frac{\delta F}{\delta n}\right) - \frac{n}{\sqrt{g}}\frac{\partial\sqrt{g}}{\partial t}, \quad \text{(S25)}$$

where Λ is the mobility of negatively charged lipids and $\vec{J}$ is the total current of negatively charged lipids on the membrane, which includes the following terms:

$$J_{att} = \frac{\kappa\Lambda\bar{H}}{n_s}n\nabla H \quad \text{(S26)}$$

$$J_{disp} = -\frac{\kappa\Lambda\bar{H}^2}{n_s}n\nabla n \quad \text{(S27)}$$

$$J_{diff} = -D\nabla n \quad \text{(S28)}$$

where J_{att} is the attraction flux resulting from the interaction between the negatively charged lipids through the membrane curvature, J_{disp} is the dispersion flux due to the membrane resistance to negatively charged lipid aggregation due to their membrane bending effects, and J_{diff} is the usual thermal diffusion flux, which depends on the diffusion coefficient, $D = \Lambda$kT. The last term in Eq. S25 arises from the covariant derivative of the density with time on a contour whose length evolves with time [5]. In this term $\sqrt{g}$ is the matrix tensor, which in our one-dimensional contour is simply the line element *dl*. This term ensures that the total number of negatively charged lipids is conserved as the contour length changes.

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