

Bending of Membranes by Anchored Polymers.

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Abstract. - Polymers which are attached to a fluid membrane by a single anchor exhibit a dilute mushroom regime and a semi-dilute brush regime. Scaling arguments and explicit calculations for ideal polymers show that the membrane is bent by the anchored polymer: For *smooth* bends, an anchored mushroom with linear size R_{po} induces a spontaneous curvature of order $\approx T/\kappa R_{po}$, where T and κ are the temperature and the bending rigidity, respectively; for *sharp* cone-like bends, it induces a bending angle $\approx T/2\pi\kappa$. A brush with mean anchor separation ξ_{an} leads to a curvature of order $\approx (R_{po}/\xi_{an})^{2/\nu} T/\kappa\xi_{an}$

Polymers at surfaces and interfaces have been studied for a long time both experimentally and theoretically. In these studies, the surface was usually governed by its interfacial tension and thus was rather rigid. Essentially tensionless and very flexible surfaces are provided by membranes such as lipid bilayers in their fluid state [1]. Indeed, several experiments have been reported recently which indicate that anchored polymers can have a strong effect on the shape of such membranes [2,3].

Polymers can be anchored to membranes in several ways: i) by lipid anchors, *i.e.* the polymer is covalently bound to the head group of a lipid molecule [4,5]; ii) by hydrophobic sidegroups of the polymers which are inserted into the bilayer [2,3]; and iii) by membrane-spanning hydrophobic domains of the polymer, which is the typical situation for membrane-bound proteins.

In order to be specific, I will focus on linear polymers with a *single* lipid anchor at one end. Apart from this anchor, the monomers of the polymer are repelled from the membrane surface. Such a situation must be distinguished i) from polymer adsorption where the whole polymer is attracted towards the membrane [6] and ii) from polymers embedded within the membrane [7]. Furthermore, in contrast to polymers grafted to solid surfaces, polymers anchored at a *fluid* membrane can diffuse laterally along the membrane surface.

Concentration regimes. – The polymer coverage Γ of the membrane surface (*i.e.* the number of monomers of anchored polymers per unit area) may be controlled by varying the time over which this surface is exposed to the polymer solution. In the dilute regime, *i.e.* as

long as the mean separation ξ_{an} of the anchored polymers is large compared to their linear size R_{po} , one has well-separated polymer mushrooms.

The linear size R_{po} of an anchored polymer consisting of N monomers can be estimated by the mean end-to-end distance of its freely suspended state which behaves as $R_{\text{po}} \approx aN^\nu$ for large N . The length scale a is the persistence length which is taken to be of the order of the monomer size (thus, the polymers are assumed to be quite flexible and effects of their bending rigidity will be ignored). The exponent $\nu = 1/3$, $1/2$ and $\approx 3/5$ for collapsed, ideal, and self-avoiding chains, corresponding to bad, θ and good solvent conditions, respectively, see, *e.g.*, ref. [8].

The anchoring free energy of a single mushroom consists i) of the energy gained by inserting the anchor into the bilayer membrane and ii) of the entropy lost by constraining the chain by the membrane surface. A self-avoiding chain anchored at a *flat* surface suffers the entropy loss $-T \Delta \mathcal{S}_{\text{po}} \approx T(\gamma - \gamma_1) \ln(N)$, with $\gamma - \gamma_1 \approx 0.5$ where the critical exponents γ and γ_1 refer to the infinite and the semi-infinite space, respectively, see, *e.g.*, ref. [9] (here and below, the temperature is measured in energy units, *i.e.* the Boltzmann constant k_B is contained in T).

The entropy loss must be overcome by the anchoring energy ΔE_{an} . For lipid anchors with two hydrocarbon chains containing n_c carbon atoms, the anchoring energy $\Delta E_{\text{an}} \approx -1.7n_c T$ as follows from the critical monomer concentration of phospholipids [10]. For dipalmitoyl phosphatidyl choline (DPPC) with $n_c = 16$, for example, one has $\Delta E_{\text{an}} \approx -27 T$. On the other hand, a rather long chain with $N = 10^5$ monomers leads to the entropy loss $-T \Delta \mathcal{S}_{\text{po}} \approx 5.8 T$. Thus, the free energy $\Delta E_{\text{an}} - T \Delta \mathcal{S}_{\text{po}}$ is still large compared to T , and one may ignore the chemical equilibrium between freely suspended and anchored polymers.

The mushroom regime applies up to the overlap coverage $\Gamma^* \approx N/R_{\text{po}}^2$ at which the membrane becomes completely covered by anchored polymers. For $\Gamma > \Gamma^*$ one enters the semi-dilute brush regime which has been studied in the context of polymers grafted at rigid surfaces [11,12]. In this regime, each polymer experiences an additional loss of entropy arising from the confinement by the neighbouring chains. This brush regime extends up to another limiting concentration Γ_{max} at which the overall loss of entropy of the chain becomes comparable with the anchoring energy, and the polymers are frequently exchanged between the membrane and the solution (where the polymers may aggregate into micelles).

Mushroom regime: smooth bends. – Now consider a polymer mushroom on a curved membrane segment ⁽¹⁾ with mean curvature $M \equiv (1/2)(1/R_1 + 1/R_2)$ and Gaussian curvature $G = 1/R_1 R_2$, where R_1 and R_2 are the principal curvature radii. Compared to the flat surface with $M = G = 0$, the configurational entropy of the polymer will change. In general, this excess entropy will depend on the polymer size R_{po} and on the length scale l_{an} which describes the distance of the anchored polymer end from the surface. However, in the limit of small $l_{\text{an}}/R_{\text{po}}$, dimensional analysis implies that the excess entropy $\Delta \mathcal{S}_{\text{po}}(M, G) \equiv \mathcal{S}_{\text{po}}(M, G) - \mathcal{S}_{\text{po}}(0, 0)$ depends only on $R_{\text{po}} M$ and $R_{\text{po}}^2 G$ for self-avoiding and ideal chains. Because of the asymmetry of the decorated membrane, one must then have

$$\Delta \mathcal{S}_{\text{po}}(M, G) = \Omega(R_{\text{po}} M, R_{\text{po}}^2 G) \approx c_s R_{\text{po}} M \quad (1)$$

⁽¹⁾ In order to estimate the curvature of the membrane segment as induced by the polymer, it is assumed that this segment can adapt freely to the forces arising from the polymer. In general, the segment belongs to a larger membrane which experiences additional forces or constraints; one example are polymers attached to a vesicle with constraints on membrane area and vesicle volume as will be considered in future work.

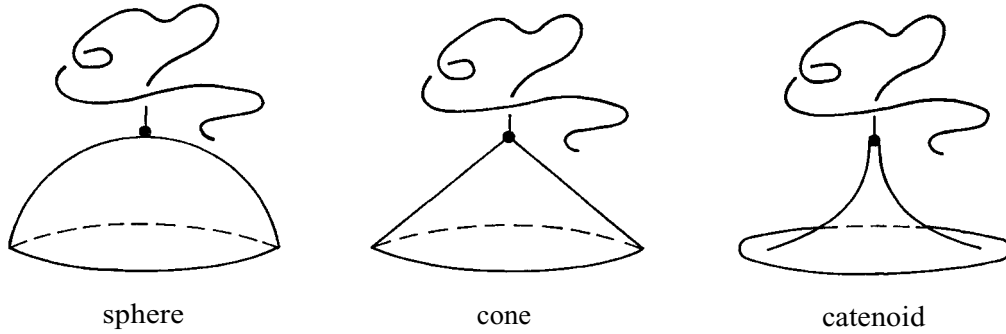


Fig. 1. - Polymer mushroom anchored at membrane segments of different shapes.

to first order in the curvature radii. The coefficient c_s is positive if the membrane curves away from the polymer, see fig. 1.

The entropy gain induced by the curvature of the surface is balanced by the increase in bending energy $\Delta\mathcal{E}_{\text{me}}$ of the membrane. For a symmetric bilayer segment which forms a spherical cap of area $\mathcal{A} = \pi R_{\text{po}}^2$, the bending energy is given by

$$\Delta\mathcal{E}_{\text{me}}(M) = \int d\mathcal{A} \frac{1}{2} \kappa (2M)^2 = 2\pi\kappa R_{\text{po}}^2 M^2, \quad (2)$$

where κ denotes the bending rigidity of the membrane. Minimizing the free energy $\Delta\mathcal{F} \equiv \Delta\mathcal{E}_{\text{me}}(M) - T\Delta\mathcal{S}_{\text{po}}(M)$, one obtains the spontaneous mean curvature

$$M = M_{\text{sp}} = c_s T / 4\pi\kappa R_{\text{po}} \sim T / \kappa a N^\nu. \quad (3)$$

Thus, the induced curvature is larger for smaller polymer size R_{po} : if one starts with good-solvent conditions with $\nu \approx 3/5$ and changes the temperature towards θ -solvent conditions with $\nu = 1/2$, the curvature will *increase*. Further changes in the temperature will then lead to bad-solvent conditions in which the polymer size becomes even smaller and the spontaneous curvature should continue to grow.

The scaling arguments leading to (3) can be confirmed by explicit calculations for ideal chains. Thus, consider the partition function $q^N Z(\mathbf{r}_{\text{an}} | \mathbf{r}, N)$ of all chain configurations for which the two ends of the chain are at the positions \mathbf{r}_{an} and \mathbf{r} . The parameter q describes the number of ways in which one may add a single monomer to a given chain segment ($q = 6$ for a simple cubic lattice). The total statistical weight is defined by

$$\mathcal{Z} \equiv \int d\mathbf{r} Z(\mathbf{r}_{\text{an}} | \mathbf{r}, N). \quad (4)$$

In the continuum limit, the reduced partition function Z satisfies a Schrödinger-type equation [13,8]. In the present context, the external potential experienced by the monomers of the chain is given by repulsive hard-wall interactions with the membrane surface. As a consequence, the Schrödinger-type equation becomes equivalent to the diffusion equation with absorbing boundaries which has been studied in much detail [14].

Let us first consider an ideal chain anchored at distance l_{an} from a *flat* surface. In this case, the total statistical weight within the half-space accessible to the polymer is given by [15]

$$\mathcal{Z}_{\text{hs}} = \text{erf}(\sqrt{q} l_{\text{an}} / 2 R_{\text{po}}) \approx \sqrt{q/\pi} l_{\text{an}} / R_{\text{po}} \sim N^{-1/2}, \quad (5)$$

where $\text{erf}(x)$ denotes the error function [16], and the asymptotic equality holds for small $l_{\text{an}}/R_{\text{po}}$

Next, consider a *spherical* membrane surface with mean curvature $M = 1/R$. The polymer sits outside of this sphere and is anchored at distance l_{an} from its surface. Extending the results of ref. [14], one finds after some computation the total statistical weight

$$\mathcal{Z} = [\text{erf}(\sqrt{q}l_{\text{an}}/2R_{\text{po}}) + l_{\text{an}}M]/[1 + l_{\text{an}}M]. \quad (6)$$

If one combines (6) and (5), the excess entropy $\Delta\mathcal{S}_{\text{po}}(M) = \ln(\mathcal{Z}/\mathcal{Z}_{\text{hs}})$ for an ideal chain is obtained as

$$\Delta\mathcal{S}_{\text{po}}(M) \approx c_s R_{\text{po}} M - \frac{1}{2} c_s^2 (R_{\text{po}} M)^2 \quad \text{with} \quad c_s = \sqrt{\pi/q} \quad (7)$$

for small M . Minimizing the excess free energy $\Delta\mathcal{F} = \Delta\mathcal{E}_{\text{me}}(M) - T\Delta\mathcal{S}_{\text{po}}(M)$, with $\Delta\mathcal{E}_{\text{me}}$ as given by (2) then leads to the spontaneous mean curvature $M_{\text{sp}} = c_s T/4\pi\kappa_{\text{eff}}R_{\text{po}}$ as in (3) with the increased bending rigidity $\kappa_{\text{eff}} = \kappa + c_s^2 T/4\pi$. The latter expression for κ_{eff} will be changed if one includes the bending modulus κ_G of the Gaussian curvature [17] which has been ignored here.

Mushroom regime: sharp bends. – So far, I have tacitly assumed that the membrane segment is bent in a *smooth* way. However, the anchor represents a point-like defect in the bilayer, and it is thus reasonable to consider other shapes of the curved membrane which involve *sharp* bends. Thus, let us assume that the membrane segment forms a *cone* with the polymer anchor at distance l_{an} from its apex, see fig. 1. In polar coordinates, the surface of the cone is given by $\theta = \theta_{\text{co}} \equiv \pi/2 + \psi$, where ψ is the bending angle with respect to the flat surface.

The corresponding diffusion problem has also been studied [14]. Expanding the reduced partition function in powers of $l_{\text{an}}/R_{\text{po}}$, one finds $\mathcal{Z} \approx \bar{c}(m)(l_{\text{an}}/R_{\text{po}})^m$, with $m = m_{\text{min}}$. The parameter m_{min} is the smallest solution of the implicit equation $P_m(\cos\theta_{\text{co}}) = 0$, where $P_m(x)$ is a Legendre polynomial [16]. For small bending angle ψ , $m_{\text{min}} \approx 1 - \psi$ and $\bar{c}(m_{\text{min}}) \approx \sqrt{q/\pi}(2/\sqrt{q})^\psi$, and the excess entropy

$$\Delta\mathcal{S}_{\text{po}} = \ln(\mathcal{Z}/\mathcal{Z}_{\text{hs}}) \approx \psi \ln(R_{\text{po}}/l_{\text{an}}). \quad (8)$$

For a cone with area $\mathcal{A} = \pi R_{\text{po}}^2$, the bending energy $\Delta\mathcal{E}_{\text{me}} = \pi\kappa[\sin^2\psi/\cos\psi] \ln(R_{\text{po}}/a_{\text{an}}) \approx \pi\kappa\psi^2 \ln(R_{\text{po}}/a_{\text{an}})$ for small ψ , where a_{an} denotes the lateral size of the anchor. For a lipid anchor with $a_{\text{an}} \approx l_{\text{an}}$, the excess free energy $\Delta\mathcal{F} \approx \ln(R_{\text{po}}/l_{\text{an}})[\pi\kappa\psi^2 - T\psi]$ leads to the spontaneous bending angle

$$\psi_{\text{sp}} \approx T/2\pi\kappa. \quad (9)$$

The corresponding decrease of the free energy of the membrane segment is given by $\Delta\mathcal{F} \approx -(T^2/4\pi\kappa) \ln(R_{\text{po}}/l_{\text{an}})$

For self-avoiding chains, no calculation for a cone seems to be available. One may, however, use the analogy between polymer statistics and critical phenomena [8] in order to conclude that the total statistical weight \mathcal{Z}_{co} of the cone behaves as $\mathcal{Z}_{\text{co}} \sim N^{\gamma_1 - 1}$, where i) $\gamma_1 = \gamma_1(\theta_{\text{co}})$ is a smooth function of the cone angle θ_{co} and ii) $\gamma_1(\theta_{\text{co}} = \pi/2) \approx 0.7$ as appropriate for the planar surface. This implies the excess entropy $\Delta\mathcal{S}_{\text{po}} \approx \psi(\partial\gamma_1/\partial\psi)_0 \ln(N)$ for small bending angles $\psi = \theta_{\text{co}} - \pi/2$. It seems feasible to obtain the function $\gamma_1(\theta_{\text{co}})$ from an expansion around $d = 4$ dimensions (as has been explicitly performed for a wedge geometry, see [18]).

If the anchor forms a defect, one may also consider a catenoid deformation as shown in fig. 1, which does not cost any bending energy. For a membrane segment of area $\mathcal{A} = \pi R_{\text{po}}^2$, the height of the catenoid surface is $\approx a_{\text{an}} \ln(R_{\text{po}}/a_{\text{an}})$. Using the total statistical weight \mathcal{Z}_{hs} for the half-space as given by (5) with $l_{\text{an}} = a_{\text{an}} \ln(R_{\text{po}}/a_{\text{an}})$, one obtains the bound $\Delta\mathcal{F} = -T\Delta\mathcal{S}_{\text{po}} > -T\ln[\ln(R_{\text{po}}/a_{\text{an}})]$ for the excess free energy of the catenoid. Therefore, for large R_{po} , the free energy of the catenoid is larger than the free energy of the cone, and a cone-like shape is more favourable.

Brush regime. – As soon as the polymer coverage (= monomers per area) exceeds the overlap coverage $\Gamma_* \approx N/R_{\text{po}}^2$, the polymers start to squeeze one another and thus to further reduce their entropy. For good-solvent conditions, each anchored polymer within such a brush can be viewed as a string of blobs: for a flat surface, the extension of these blobs is given by the mean separation ξ_{an} of the anchors [11]; for a curved surface, this extension grows with the distance from the surface [12].

For a spherical membrane segment with mean curvature $M = 1/R$, scaling arguments lead, in the limit of small M , to the brush height $h \sim N(a/\xi_{\text{an}})^{1/\nu} \xi_{\text{an}}$ and to the excess free energy $\Delta\mathcal{F}_{\text{po}} \sim T(h/\xi_{\text{an}})[(1/hM)\ln(1+hM) - 1] \sim -Th^2M/\xi_{\text{an}}$. If this excess free energy is balanced against the bending energy of the curved membrane, one now obtains the spontaneous mean curvature

$$M_{\text{sp}} \sim (R_{\text{po}}/\xi_{\text{an}})^{2/\nu} T/\kappa\xi_{\text{an}}, \quad \text{with} \quad \nu \approx 3/5 \quad (10)$$

For sharp cone-like bends, the free-energy balance leads to the spontaneous bending angle $\psi_{\text{sp}} \sim (T/\kappa)(R_{\text{po}}/\xi_{\text{an}})^{2/\nu}$ (where the logarithmic factor $\ln(\xi_{\text{an}}/a)$ has been ignored). The average curvature $\langle M \rangle \approx \psi_{\text{sp}}/\xi_{\text{an}}$ arising from these sharp bends is of the same order as the spontaneous mean curvature (10).

As the coverage Γ is increased, the excess free energy $\Delta\mathcal{F}_{\text{po}}$ of each chain becomes eventually comparable to the anchoring energy $\Delta\mathcal{E}_{\text{an}} \equiv -\Delta e_{\text{an}}T$. For a flat membrane, one has $\Delta\mathcal{F}_{\text{po}} \sim TN(a/\xi_{\text{an}})^{1/\nu}$ which leads to the minimal value $\xi_{\text{an}} \approx R_{\text{po}}/\Delta e_{\text{an}}^\nu$ of the anchor separation and to the maximal value $\Gamma_{\text{max}} \approx \Gamma_* \Delta e_{\text{an}}^{2\nu}$ of the polymer coverage. For DPPC anchors with $\Delta e_{\text{an}} \approx 27$, the minimal anchor separation is $\approx R_{\text{po}}/7$ and the maximal coverage is $\approx 52 \Gamma_*$. If the dissolved and the anchored polymers are in chemical equilibrium, the decorated membranes should exhibit the spontaneous curvature induced by this maximal coverage.

Open ends. – The work described above may be extended in several ways. First, the polymeric architecture within the dilute mushroom regime may be varied: the chains may be attached by two or several anchors as in ref. [2,3]; they may be comb-like, star-like or randomly branched (a star polymer with b ideal arms, for example, which is anchored at its centre has an excess entropy which is again given by (3) and (9) but with T replaced by bT); they may be crosslinked and thus form small gel balls. Secondly, the shape fluctuations or undulations of the membranes are also affected by the anchored polymers: in the brush regime, the effective bending rigidity of the membrane should be *increased* by the presence of the polymer brush; in the mushroom regime, on the other hand, the anchored polymers act like local curvature defects [19] which tend to *decrease* the bending rigidity^(?). Finally, the effective interaction between the mushrooms need not be repulsive but can be attractive.

^(?) If the polymers are attached to both sides of the membrane with equal probability, this reduction of the bending rigidity is $\sim pT$, where p is the area fraction covered by the polymer.

This happens, *e.g.*, in bad-solvent conditions or in the presence of crosslinkers. The anchored mushrooms then tend to form clusters or domains even below the overlap coverage. The formation of intramembrane domains generically leads to domain-induced budding [20] which could explain some of the experimental observations in ref. [2,3].

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