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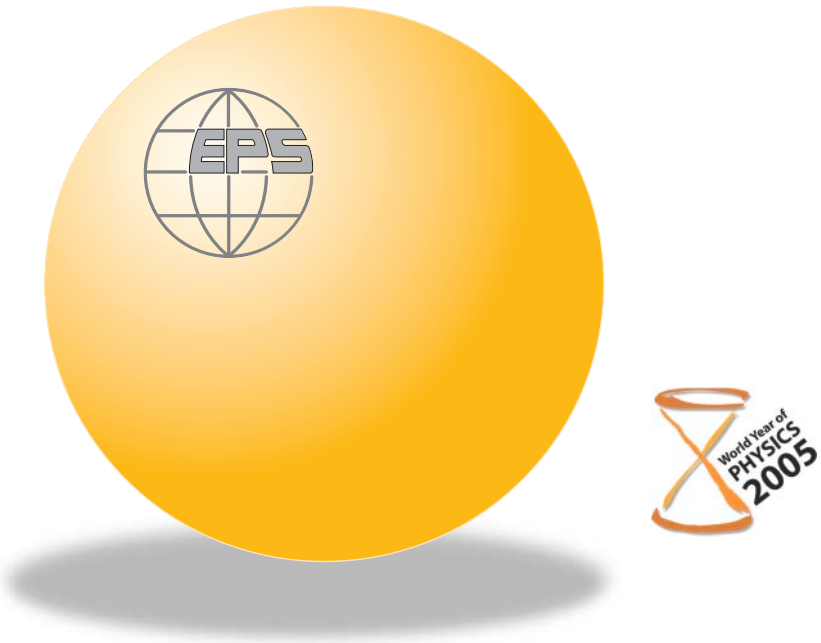

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Self-assembly of linear aggregates on curved membranes

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Abstract. – Recent experiments evidence the strong influence that linear aggregation of beta-sheet peptides can have on the conformation and stability of lipid membranes. We have therefore studied in the dilute limit the self-assembly of one-dimensional aggregates bound to a fluid-like membrane of prescribed curvatures. Each aggregate is characterized by its persistence length and an interaction potential with the membrane. For strong adsorption of flexible aggregates the bending stiffness of the composite membrane grows linearly with the total concentration of aggregate-forming units. For weak adsorption of rigid aggregates the dependence is cubic for which we show —based on a hydrophobic matching hypothesis— that the aggregates can significantly contribute to or even dominate the bending stiffness. We also show how the membrane curvature affects the orientational and size distribution of the aggregates.

Introduction. – The reversible self-assembly into one-dimensional (1D) aggregates is an ubiquitous phenomenon, frequently encountered in surfactant solutions as "worm-like" micelles and in living cells for cytoskeletal components such as actin and tubulin. Another class of linearly aggregating biomolecules, which has recently attracted much interest, are β-sheet forming peptides. The self-assembly of these peptides can be pathological, which is related to the formation of amyloid fibrils found in various diseases including Alzheimer. It appears that β-sheet peptides often interact with lipid membranes [1]. The peptide's tendency to self-assemble is then influenced by the presence of the membrane and, conversely, may affect the conformation of the membrane. For example, peptide B18, which consists of a highly conserved sequence of the sea urchin protein bindin is known to self-assemble under appropriate conditions into β-sheet amyloid fibrils. The fibrils interact strongly with lipid membranes, transforming unilamellar vesicles into extended lamellar sheets that are covered by long parallel peptide fibrils [2]. The formation of amyloid fibrils in the membrane bound state has also been reported for other amyloidogenic proteins such as the amyloid β (Aβ) peptide [3], the human islet amyloid polypeptide [4], the prion protein PrP^{Sc} [5], and for certain viral fusion peptides. There is accumulating evidence that linearly self-assembling, membrane-active peptides can strongly affect the structural stability of the host bilayer, ranging from mere changes of vesicle shape [5] over the initiation of fusion events to the solubilization of entire membrane patches [6]. While these modes of interaction depend on experimental

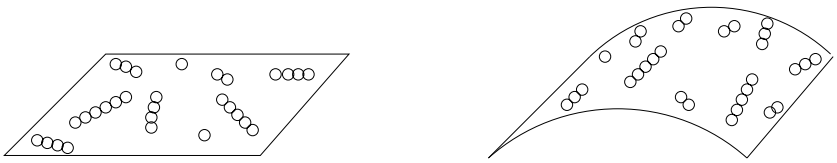

Fig. 1 – Self-assembly of linear aggregates on a rigorously flat (left) and on a homogeneously curved membrane (right). The membrane curvature will affect the size distribution of the aggregates and their average orientation. The aggregates contribute to the elastic properties of the composite membrane.

conditions such as membrane composition, pH, peptide conformation, and kinetic aspects, they all should be influenced by the general physical principles that underlie the self-assembly of 1D objects on a curved membrane.

In the present work we suggest a phenomenological model for the linear self-assembly on a curved membrane. We shall focus on the dilute limit where the membrane-associated aggregates do not interact with each other. As another major approximation of this work we shall investigate the limit of prescribed, uniform curvatures; see fig. 1. That is, the presence and self-assembly of the aggregates on the membrane is assumed not to affect the local membrane curvatures. This assumption is appropriate if either the interaction strength between the aggregates and the membrane is weak or if the membrane is conformationally confined as can be the case in small spherical, unilamellar vesicles, tense liposomes, on a substrate-supported membrane, or in a membrane stack. For strong adsorption and if the membrane is free to locally adjust its curvatures, our calculated free energy expression, see eq. (13) below, still provides an upper bound. We note that taking the local adjustment of curvatures into account would also give rise to membrane-mediated interactions between rod-like inclusions [7–9]. These are not an issue in the present work. For the linear self-assembly on a rigorously flat membrane, see fig. 1 (left scheme), equilibrium thermodynamic properties, particularly the aggregate's size distribution, are well known [10] and will be recalled shortly below. Uniformly bending the membrane affects both the size and orientational distribution of the 1D aggregates as is schematically illustrated in fig. 1 (right scheme). At the same time, the energetic coupling of the linear aggregates to the membrane will influence the elastic properties of the composed system. We calculate the relevant thermodynamic properties; specifically, we show that for sufficiently stiff aggregates their contribution to the bending rigidity of the membrane, $\Delta\kappa \sim x_{tot}^3$, grows cubically with the total concentration of membrane-adsorbed particles. By estimating the prefactor we specify conditions for which this increase starts to dominate the bending rigidity and thus can potentially affect the structural stability of the composite membrane.

Self-assembly in dilute solution. – We consider the self-assembly of initially N identical monomeric units, such as associating peptides or proteins, on a lipid membrane of overall lateral area A. The N monomers are adsorbed onto the membrane surface, being able to freely diffuse laterally. We denote the cross-sectional area of each monomer by $a_0 = l_0 \times l_0$ implying a maximal number $M = A/l_0^2$ of adsorbed molecules. Upon self-association, the molecules form N_i aggregates of aggregation number i; $i = 1$ denoting the monomeric state, $i = 2$ the dimeric state, etc. It is convenient to express the aggregate size distribution on the scale of mole fractions through $x_i = iN_i/M$. The distribution x_i then fulfills the particle conservation condition $x_{tot} = \sum_{i=1}^{\infty} x_i$, where $x_{tot} = N/M$ denotes the total mole fraction of

membrane-adsorbed molecules. In the dilute limit, where $x_{tot} \ll 1$, the Helmholtz free energy F of the associating molecules can be written as

$$\frac{F}{M} = \sum_{i=1}^{\infty} \left[x_i \tilde{\mu}_i^0 + kT \frac{x_i}{i} \left(\ln \frac{x_i}{i} - 1 \right) \right] + \mu \left(x_{tot} - \sum_{i=1}^{\infty} x_i \right), \tag{1}$$

where kT is the thermal energy and $\tilde{\mu}_i^0$ is the standard chemical potential per monomer in an aggregate of size i. The chemical potential μ ensures the particle conservation condition to be fulfilled. The free energy, eq. (1), implies the equilibrium distribution $x_i = i \exp[-i(\tilde{\mu}_i^0 - \mu)/kT]$ which upon insertion back into eq. (1) yields

$$\frac{F}{M} = \mu x_{tot} - kT \sum_{i=1}^{\infty} \frac{x_i}{i} \tag{2}$$

or, equivalently, $F = G - \Pi A$, where $G = \mu N$ is the Gibbs free energy and $\Pi = -\partial F/\partial A = kT \sum_{i=1}^{\infty} N_i/A$ is the lateral osmotic pressure in the dilute limit.

One-dimensional growth. – To calculate the distribution x_i we need to specify the internal energy of the aggregates as a function of their aggregation number. This piece of information is contained in the standard chemical potential $\tilde{\mu}_i^0$ per monomer. Subject of the present work is the growth into linear aggregates. In this case, a reasonable model is based on the *linear superposition assumption* [10] where each monomeric unit delivers the same contribution, μ_b, into the standard chemical potential of an i-mer, $\mu_i^0 = i\tilde{\mu}_i^0$, independent of its position along the one-dimensional aggregate. Only each of the two terminal segments does add an additional *edge energy* $\delta/2$. The corresponding model for one-dimensional growth can thus be written as $\mu_i^0 = i\mu_b + \delta$. For sufficiently large edge energy, $\delta \gg 1$, the aggregates grow long, and we can replace the summation over all aggregation sizes by an integration, $\sum_{i=1}^{\infty} \to \int_0^{\infty} \mathrm{d}i$, as well as all discrete variables by continuous ones, *i.e.* $x_i \to x(i)$, etc. In this case, the chemical potential is given by $\mu = \mu_b - kT/\sqrt{e^{\delta/kT} x_{tot}}$, and the aggregate size distribution reads

$$x(i) = i \exp\left[-\frac{\delta}{kT} - \frac{i}{\sqrt{e^{\delta/kT} x_{tot}}} \right]. \tag{3}$$

We remark that the function $x(i)$ adopts a maximum at aggregation number $i_m = \sqrt{e^{\delta/kT} x_{tot}}$. Weight-averaged quantities of any given physical quantity $Q = Q(i)$ are calculated via $\langle Q \rangle = \int_0^{\infty} x(i)\, Q(i)\, \mathrm{d}i / \int_0^{\infty} x(i)\, \mathrm{d}i$. For example, the weight-averaged size distribution is $\langle i \rangle = 2i_m$, and for the standard deviation of the size distribution we obtain $\sigma = \sqrt{\langle (i - \langle i \rangle)^2 \rangle} = \sqrt{2}\, i_m$. Finally, the Helmholtz free energy given in eq. (2) becomes

$$\frac{F}{M} = x_{tot} \left[\mu_b - \frac{2kT}{\sqrt{e^{\delta/kT} x_{tot}}} \right]. \tag{4}$$

Note that for small T all molecules are incorporated into one single aggregate, and the free energy is —as expected— $F(N, T \to 0) = N\mu_b$.

Internal energy of an individual membrane-adsorbed aggregate. – The standard chemical potential, $\mu^0(i)$, of a membrane-adsorbed linear aggregate does generally not satisfy the linear superposition assumption. Here, we suggest a simple model that describes how the function $\mu^0(i)$ is modified by the prescribed *curvature* of the membrane. The starting point of our model

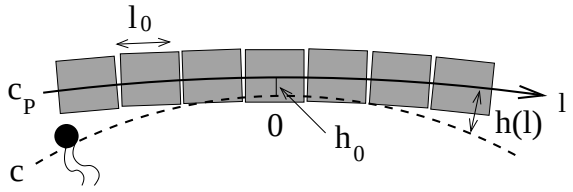

Fig. 2 – An aggregate of aggregation number $i = 7$ adsorbed on a bent lipid layer of curvature c. The shaded regions mark the individual monomers of which the aggregate consists. The contour length of the aggregate is $L = il_0$, where l_0 is the linear dimension of each monomeric unit. The curvature of the aggregate, measured along its mid-axis (solid line), is c_P, and the local distance between the aggregate and the lipid layer is $h(l)$ with $h_0 = h(l = 0)$. A single lipid is illustrated schematically.

is the membrane adsorption energy, $u(h)$, for each individual monomeric unit; $u(h)$ depends only on the distance to the membrane but not explicitly on the size of the aggregate into which the monomeric unit is incorporated nor on the membrane curvature [11]. (However, as above we shall assume that the two terminal segments of any given aggregate each contribute an edge energy $\delta/2$.) A single membrane-adsorbed monomer ($i = 1$) is able to optimize h and thus $\mu_1^0 = \mu_b + \delta$, where μ_b is identified with the minimum of the function $u(h)$. Consider now a linear self-assembled aggregate with $i > 1$, adsorbed on a bent membrane of given curvature, c, measured along its contour length. Here, owing to the flexural rigidity of the polymeric aggregate, its actual curvature c_P may deviate from the curvature c of the lipid layer. That is, not all individual units of the aggregate may be able to optimize their distance to the membrane, as illustrated in fig. 2. For small deviations h of the actual distance from the optimal one, we can expand the adsorption potential

$$u(h) = \mu_b + kT\frac{K_s}{2}\left(\frac{h}{l_0}\right)^2, \tag{5}$$

where K_s is a dimensionless spring constant. We write the internal energy of a membrane-adsorbed polymeric aggregate as the sum of three contributions:

$$f = \frac{1}{l_0}\int_{-L/2}^{L/2} u[h(l)]\,\mathrm{d}l + \frac{\kappa_P}{2}\int_{-L/2}^{L/2}\left[c_p(l) - c_p^0\right]^2\mathrm{d}l + \delta. \tag{6}$$

The first is a linear superposition [11] of the individual monomer-membrane interaction energies where the integration extends over the contour length $L = il_0$ of the polymeric aggregate. Up to linear order in curvature the distance $h(l)$ is given as

$$h(l) = h_0 + \frac{c - c_p}{2}\,l^2, \tag{7}$$

where the (yet unknown) constant h_0 is the distance adopted for $l = 0$ at the mid-segment of the polymer; see fig. 2. The second contribution in eq. (6) accounts for the intrinsic energy to bend the polymer; $\kappa_P = kTl_0\xi$ is the flexural rigidity of the polymeric aggregate which we express in terms of the (dimensionless) persistence length, ξ, measured in units of l_0. In absence of axial symmetry around its mid-axis, the polymeric aggregate is expected to possess a spontaneous curvature, c_p^0, which we can easily include into our theoretical description. Note that for a sufficiently stiff aggregate the curvature $c_P = c_P(l)$ can be treated as a constant.

Finally, the last term in eq. (6) accounts for twice the edge energy, $\delta/2$, contributed by the terminal units. Upon insertion of eq. (7) into eq. (5) and that into eq. (6), we can carry out the integrations in eq. (6). Note that the membrane-adsorbed polymer is able to optimize both its curvature c_P and its distance h_0 to the lipid layer; calculation of the minimum of $f(c_P, h_0)$ leads to the optimal values

$$h_0 = -\frac{L^2}{24}\,\frac{c - c_P^0}{1 + \frac{L^4 K_s}{720\xi l_0^4}}\,, \quad c_P = c - \frac{c - c_P^0}{1 + \frac{L^4 K_s}{720\xi l_0^4}}\,. \tag{8}$$

Inserting h_0 and c_P into eq. (7) gives rise to the local deviation $h(l) \sim (c - c_P^0)(12l^2 - L^2)$ of the polymer's segments from their individual equilibrium position. Solving for $h = 0$ results in either $c = c_P^0$ or $l = \pm L/\sqrt{12}$. The former is merely the special case of choosing the membrane curvature to equal the preferred one of the polymer —in this case, all polymer segments are located at their optimal distance $h = 0$. The latter case tells us that for any given membrane curvature c, segments at the two positions $l \approx \pm 0.29L$ reside at $h = 0$. Using the optimal values for h_0 and c_P we finally obtain for the internal energy of the aggregate $f = i\chi(i)kTl_0^2(c - c_P^0)^2 + i\mu_b + \delta$, where we have defined the dimensionless function

$$\chi(i) = \frac{i^4 K_s}{1440}\,\frac{1}{1 + \frac{i^4 K_s}{720\xi}}\,. \tag{9}$$

Self-assembly on a curved membrane. – The expression for the internal energy f, indeed suggests that bending the lipid layer modifies the linear behavior $f \sim i$. To write down an explicit expression for the standard chemical potential $\mu^0(i)$ we take the rotational in-plane degree of freedom of the adsorbed aggregate into account. Upon rotating the polymeric aggregate laterally on the membrane surface by an angle ϕ, the local curvature seen by the i-mer changes according to $c = H + D\cos(2\phi)$, where $H = (c_1 + c_2)/2$ and $D = (c_1 - c_2)/2$ are the mean and deviatoric curvatures of the lipid layer, respectively, with c_1 and c_2 being the two local principal curvatures of the membrane. Accounting for statistical averaging in the small curvature limit (that is, for small deviations away from the optimal curvature $H = c_P^0$ and $D = 0$) we can write for the standard chemical potential

$$\mu^0(i) = \frac{1}{2\pi}\int_0^{2\pi} f\,\mathrm{d}\phi = i\mu_b + \delta + kTil_0^2\chi(i)\,\left[\frac{D^2}{2} + \left(H - c_P^0\right)^2\right]. \tag{10}$$

Note that for optimal membrane curvature, $D = 0$ and $H = c_P^0$, we recover the linear superposition assumption, $\mu^0(i) = i\mu_b + \delta$. The model for $\mu^0(i) = i\tilde{\mu}^0(i)$ in eq. (10) allows us to calculate the chemical potential, μ, in the small curvature limit. Recall that μ must be determined from the condition $x_{tot} = \int_0^\infty x(i)\,\mathrm{d}i$ with $x(i) = i\exp[-i[\tilde{\mu}^0(i) - \mu]/kT]$. The result is up to second order in curvature

$$\mu = \mu_{ref} + kT\frac{\langle i\chi(i)\rangle_{ref}}{\langle i\rangle_{ref}}l_0^2\left[\frac{D^2}{2} + \left(H - c_P^0\right)^2\right], \tag{11}$$

where here and in the following the index "*ref*" refers to the reference state $H = c_P^0$ and $D = 0$, around which the membrane is deformed. In fact, for $c_P^0 = 0$ the reference state is the planar membrane. Recall that $\mu_{ref} = \mu_b - kT/i_m$ and $\langle i\rangle_{ref} = 2i_m$ with $i_m = \sqrt{e^{\delta/kT}x_{tot}}$. Using μ in eq. (11) we can write for the distribution $x(i)$,

$$\frac{x(i) - x_{ref}(i)}{x_{ref}(i)} = l_0^2\left[\frac{D^2}{2} + \left(H - c_P^0\right)^2\right] i\left[\frac{\langle i\chi(i)\rangle_{ref}}{\langle i\rangle_{ref}} - \chi(i)\right], \tag{12}$$

where $x_{ref}(i) = x(i, D = 0, H = c_P^0)$ is the size distribution for the reference state $D = 0$ and $H = c_P^0$, given in eq. (3). Because $\chi(i)$ is a monotonously increasing function, it must be $x(i) > x_{ref}(i)$ for small i, and $x(i) < x_{ref}(i)$ for large i. That is, bending the membrane generally favors the formation of smaller aggregates and suppresses larger ones. The aggregation number $i = i^\star$ for which the mole fraction $x(i)$ remains unaffected can be found from solving the equation $\langle i\chi(i)\rangle_{ref} = \langle i\rangle_{ref}\chi(i)$. The Helmholtz free energy F can be calculated from eq. (2), using μ from eq. (11) and $x(i)$ from eq. (12). The result is

$$\frac{F}{M} = \frac{F_{ref}}{M} + x_{tot}kTl_0^2\left[\frac{D^2}{2} + \left(H - c_P^0\right)^2\right]\langle\chi(i)\rangle_{ref}\,, \tag{13}$$

where F_{ref} is the Helmholtz free energy for the reference state $D = 0$ and $H = c_P^0$; see eq. (4).

Curvature elastic moduli. – The dependence of the free energy on the membrane curvature is manifested in the magnitude of the bending rigidity κ and the Gaussian modulus $\bar{\kappa}$ which appear in the familiar bending energy per unit area of a fluid-like membrane [12], $F/A = (\kappa/2)(c_1 + c_2 - c_0)^2 + \bar{\kappa}c_1c_2$. Comparison with eq. (13) allows us to calculate the contributions, $\Delta\kappa$ and $\Delta\bar{\kappa}$, of the self-assembled membrane-adsorbed aggregates to the bending rigidity and the Gaussian modulus, respectively. The results are $\Delta\kappa = x_{tot}kT(3/4)\langle\chi(i)\rangle_{ref}$ and $\Delta\bar{\kappa}/\Delta\kappa = -2/3$. Two limits are easily accessible. For *strong adsorption of flexible aggregates*, implying $K_s \gg \xi$, we have $\chi(i) = \xi/2$ independent of i. In this case, the growth characteristics is not affected by the membrane curvature, the linear superposition assumption is valid, and the bending stiffness $\Delta\kappa(K_s \gg \xi) = 3kTx_{tot}\xi/8$ grows linearly with x_{tot}. Below, we argue that in this limit, the self-assembled aggregates are not expected to notably affect the elastic properties of the membrane. To investigate the other limit, *weak adsorption of rigid aggregates*, where $K_s \gg \xi$, we formally rewrite the general result for the bending stiffness into an infinite sum

$$\Delta\kappa = x_{tot}kT\frac{3}{4}\langle\chi(i)\rangle_{ref} = -kT\frac{3}{8}x_{tot}\xi\sum_{j=1}^{\infty}\left(-\frac{K_sx_{tot}^2e^{2\delta/kT}}{720\xi}\right)^j(1+4j)! \tag{14}$$

The limit $K_s \ll \xi$ is then given by the first term of this sum,

$$\Delta\kappa(K_s \ll \xi) = \frac{kT}{16}K_sx_{tot}^3e^{2\delta/kT}, \tag{15}$$

revealing a $\Delta\kappa \sim x_{tot}^3$ dependence on concentration. Note also that for $K_s \ll \xi$ we obtain $i^\star = (360)^{(1/4)}i_m$. That is, the population of aggregates with $i < 4.36\,i_m$ grows upon bending the membrane whereas for $i > 4.36\,i_m$ it shrinks. Note that the limit of *strong adsorption of rigid aggregates* ($K_s \sim \xi \gg 1$) is not captured by our model since local membrane deformations are supposed to dramatically lower the free energy in this case.

We finally remark that we obtain the same elastic behavior for a *frozen* distribution, $x(i) = x_{ref}(i)$ which cannot adjust during bending of the membrane. This can easily be seen by inserting $x(i)$ from eq. (3) into the free energy F given in eq. (1) (with $\mu = 0$), using the curvature-dependent standard chemical potential $\mu_i^0 = i\tilde{\mu}_i^0$ in eq. (10).

Discussion. – To access the experimental relevance of the calculated bending stiffness and the relevant regime (that is $K_s \gg \xi$ or $K_s \ll \xi$), we need to estimate the (dimensionless) spring constant K_s introduced in eq. (5). Obviously, the magnitude of K_s will depend first and foremost on the nature of the interaction between the self-assembling molecules and the

membrane. For example, electrostatic attraction is generally expected to contribute to the adsorption of charged particles onto oppositely charged membranes. Another type are hydrophobic interactions, relevant for membrane-penetrating peptides or proteins. In this case, pulling the particle out of the membrane induces an elastic membrane response that can be described using the concept of *hydrophobic mismatch* [13]. Here, the spring constant can be calculated based on membrane elasticity theory [13,14], leading to $K_s = 2l_0 d_0 (\kappa/4Kd_0^2)^{(1/4)} K/kT$, where $\kappa \approx 10kT$ is the bending stiffness of a bare lipid monolayer, $K \approx 0.2kT/\,\text{Å}^2$ is the area stretching modulus, and $d_0 \approx 15\,\text{Å}$ is the monolayer thickness. The factor of two in the expression for K_s results from the two sides of each aggregate that face the membrane. Assuming $l_0 \approx d_0$ (as will be reasonable for a typical membrane-adsorbed peptide) we obtain $K_s \approx 40$. Based on this estimate we see that for a (dimensionless; scaled by l_0) persistence length $\xi \ll 40$ the linear superposition approximation is valid. To investigate the other limit, $\xi \gg K_s$, we re-express eq. (15) as $\Delta\kappa/kT = K_s x_{tot} i_m^4/16$, where i_m is the most abundant aggregation number. Clearly, for sufficiently long aggregates, $\Delta\kappa$ can be larger —even much larger— than kT. For example, $K_s = 40$, $x_{tot} = 0.001$ and $i_m = 10$ leads to $\Delta\kappa = 27kT$, on the same order as the bending stiffness of a bare lipid layer. For $i_m = 20$ we obtain $\Delta\kappa = 400kT$. This should be contrasted to the regime $K_s \gg \xi$ for which the contribution of the self-assembled aggregates to the bending stiffness, $\Delta\kappa = 3kTx_{tot}\xi/8$, is always negligible; even for $\xi = 30$ and $x_{tot} = 0.01$ it is $\Delta\kappa \ll kT$. In summary, we have calculated how the self-assembly of linear aggregates bound to a lipid membrane affects the membrane elastic properties. The effect is expected to be significant if the 1D aggregates are relatively stiff, with $\xi \gg K_s$.

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REFERENCES

[1] Critchley P., Kazlauskaite J., Eason R. and Pinheiro T. J. T., *Biochem. Biophys. Res. Commun.*, **313** (2004) 559.
[2] Ulrich A. S., Tichelaar W., Forster G., Zschornig O., Weinkauf S. and Meyer H. W., *Biophys. J.*, **77** (1999) 829.
[3] Yip C. M., Darabie A. A. and Mclaurin J., *J. Mol. Biol.*, **318** (2002) 97.
[4] Porat Y., Kolusheva S., Jelinek R. and Gazit E., *Biochemistry*, **42** (2003) 10971.
[5] J. Kazlauskaite N., Sanghera I. S. C. V.-B. and Pinheiro T. J. T., *Biochemistry*, **42** (2003) 3295.
[6] Tashima Y., Oe R., Lee S., Sugihara G., Chambers E., Takahashi M. and Yamada T., *J. Biol. Chem.*, **279** (2004) 17587.
[7] Holzlohner R. and Schoen M., *Eur. Phys. J. B*, **12** (1999) 413.
[8] Dommersnes P. G. and Fournier J. B., *Eur. Phys. J. B*, **12** (1999) 9.
[9] Golestanian R., Goulian M. and Kardar M., *Phys. Rev. E*, **54** (1996) 6725.
[10] Ben-Shaul A. and Gelbart W. M., in *Micelles, Membranes, Microemulsions, and Monolayers*, edited by Gelbart W. M., Ben-Shaul A. and Roux D., first edition (Springer, New York) 1994, sect. 1, pp. 359-402.
[11] This assumption will not be fulfilled if the membrane is allowed to locally adjust its curvatures. Neither will it be valid for electrostatically charged aggregate-membrane complexes in the limit of low salt content where the Debye length $l_D \gg h$.
[12] Helfrich W. Z., *Naturforsch.*, **28** (1973) 693.
[13] Huang H. W., *Biophys. J.*, **50** (1986) 1061.
[14] May S., *Langmuir*, **18** (2002) 6356.