

## The unbinding transition of mixed fluid membranes

S. KOMURA<sup>1</sup>(\*) and D. ANDELMAN<sup>2</sup>(\*\*)

<sup>1</sup> *Department of Chemistry, Faculty of Science, Tokyo Metropolitan University  
Tokyo 192-0397, Japan*

<sup>2</sup> *School of Physics and Astronomy, Raymond and Beverly Sackler Faculty  
of Exact Sciences, Tel Aviv University - Ramat Aviv 69978, Tel Aviv, Israel*

(received 5 August 2003; accepted in final form 8 October 2003)

PACS. 87.16.Dg – Membranes, bilayers, and vesicles.

PACS. 68.05.-n – Liquid-liquid interfaces.

PACS. 64.60.-i – General studies of phase transitions.

**Abstract.** – A phenomenological model for the unbinding transition of multicomponent fluid membranes is proposed, where the unbinding transition is described using a theory analogous to the Flory-Huggins theory for polymers. The coupling between the lateral phase separation of inclusion molecules and the membrane-substrate potential provides a rich phase behavior. Our model describes the first-order nature of the unbinding transition in multicomponent membranes as was observed in a recent experiment. In particular, we predict different scenarios of phase coexistence between bound and unbound membrane states.

*Introduction.* – Adhesion of membranes and vesicles is responsible for cell-cell adhesion which plays an important role in all multicellular organisms. In general, bio-adhesion is governed by the interplay of various generic and specific interactions [1]. Specific interactions act between complementary pairs of proteins such as ligand and receptor, or antibody and antigen. Well-studied examples of such coupled systems are biotin-avidin complexes [2], or selectins and their sugar ligands [3].

The problem of adhesion of multicomponent membranes is intimately related to that of domain formation. Experimentally, adhesion-induced lateral phase separation has been observed for various systems [4–6]. It was reported that adhesion molecules aggregate spontaneously and form tight-adhering domains. From the theoretical point of view, this problem has been considered in [7] using a phenomenological model, where the inter-membrane distance is coupled to the concentration of sticker molecules on the two adhering membranes. In a different approach, a lattice model for a multicomponent membrane in contact with another substrate was proposed [8], and studied via detailed Monte Carlo simulations [9, 10]. More recently, a work combining these two approaches has been published [11].

In a recent experiment by Marx *et al.* [12], the role of long-range repulsions due to thermal fluctuations (Helfrich repulsion) of adhering membranes has been addressed. By using Reflection Interference Contrast Microscopy [4], a multicomponent bilayer membrane with added

---

(\*) E-mail: komura@comp.metro-u.ac.jp

(\*\*) E-mail: andelman@post.tau.ac.il

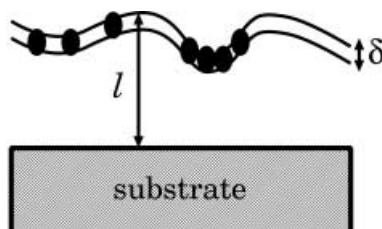


Fig. 1 – A mixed fluid membrane adhering to a substrate. Black filled ovals indicate inclusions such as proteins or lipopolymers. The height of the upper membrane leaflet from the substrate is  $\ell$ , whereas the thickness of the bilayer membrane is  $\delta$ .

lipopolymers (modified DOPE lipid with polyethylenoxide) and cholesterol is examined in the vicinity of an attractive substrate. Analyzing the probability distribution of the membrane-substrate spacing for various multicomponent membranes, a phase separation between two distinct lipopolymer-poor and lipopolymer-rich states having two different spacings from the substrate was suggested, where both states are *unbound* from the substrate. Although this phenomenon is dynamic in nature, it was interpreted as a lateral phase segregation induced by the Helfrich repulsion. However, a clear physical description for the appearance of such a phase coexistence has not been given. Moreover, a *first-order* unbinding transition scenario is required to account for the multiple time scales in the time series of the membrane fluctuations [12].

In this letter, we propose a simple phenomenological model for a multicomponent (mixed) fluid membrane which can undergo simultaneously a lateral phase separation and an unbinding transition. The model is motivated by the experiment [12] and relies on the coupling between the inclusion concentration and the membrane-substrate spacing. The lateral phase separation of the inclusion affects the second virial coefficient of the unbinding transition which is taken into account in analogy to the Flory-Huggins theory for polymers [13]. Our model exhibits various types of phase coexistence, including a phase separation between bound and unbound states as well as between two unbound ones. The former phase coexistence indicates the first-order nature of the unbinding transition, as was anticipated in the experiment [12].

*Unbinding transition.* – Fluid membranes in a lamellar stack or close to a substrate experience steric repulsion arising from their reduced undulation entropy due to the confinement effect. The corresponding interaction energy per unit area has been given by Helfrich [14] as

$$v_s(\ell) = \frac{b(k_B T)^2}{\kappa(\ell - \delta)^2}, \quad (1)$$

where  $k_B$  is the Boltzmann constant,  $T$  the temperature,  $\kappa$  the bending rigidity of the membrane having thickness  $\delta$ , and  $\ell$  the average height of the upper membrane lipid leaflet from the substrate (see fig. 1). The numerical prefactor  $b$  was calculated as  $b = 3\pi^2/128$  in [14], but its value is still debatable in the literature [15]. The combination of the above steric repulsion and other direct microscopic (van der Waals, electrostatic, hydration) interactions determines whether the membrane binds to the substrate or unbinds. Using functional renormalization group techniques, Lipowsky and Leibler have shown that this unbinding transition is of *second order* [16, 17]. The average spacing  $\ell$  diverges as the strength of the attractive van der Waals interaction  $W$  (Hamaker constant) approaches a critical value from above, *i.e.*,  $\ell \sim (W - W_c)^{-\psi}$  with  $\psi \approx 1.0$ . Here  $W_c$  is the critical strength of the Hamaker constant.

While a simple superposition of the Helfrich repulsion, eq. (1), and other direct interactions gives an incorrect (first-order) description of the unbinding transition, a simple theory for the

unbinding transition in a bulk of lamellar phase was considered by Milner and Roux [18], and is briefly reviewed below. Following the spirit of the Flory-Huggins theory for polymers [13], the Helfrich estimate of the entropy is taken into account accurately, whereas the other interactions are approximately incorporated via a second virial term. This approach is justified since the perturbative effect of the direct interactions is included through a virial expansion rather than the simple superposition of the interactions. The resulting free energy *per unit volume* of a lamellar stack is

$$g_{\text{MR}}(w) = -k_{\text{B}}T\bar{\chi}w^2 + \frac{b(k_{\text{B}}T)^2}{\kappa\delta^3}w^3 - \mu_w w, \quad (2)$$

where the membrane volume fraction  $w = \delta/\ell \geq 0$  cannot be negative. Note that  $\ell$  represents the lamellar repeat distance,  $\bar{\chi}$  is the second virial coefficient, and  $\mu_w$  the chemical potential which accounts for the conservation of the membrane volume fraction  $w$ . The second virial coefficient is predicted to behave as  $\bar{\chi} \sim (W - W_c)$ . For  $\bar{\chi} > 0$  the free energy (2) describes a first-order unbinding transition as  $\mu_w$  is lowered.

To consider the unbinding behavior of a single membrane (see fig. 1) as in the experiment [12], eq. (2) is multiplied by the lamellar repeat distance  $\ell$ . We also note that the membrane average distance  $\ell$  from the substrate is not a conserved quantity. Then the free energy *per unit area* of a single membrane can be expressed as

$$f(w) = -k_{\text{B}}T\delta\bar{\chi}w + \frac{b(k_{\text{B}}T)^2}{\kappa\delta^2}w^2. \quad (3)$$

Minimization of  $f(w)$  with respect to  $w$  shows that this free energy has a continuous second-order transition at  $\bar{\chi} = 0$  between a bound state ( $w > 0$ ) for  $\bar{\chi} > 0$  and an unbound state ( $w = 0$ ) for  $\bar{\chi} < 0$ . A similar free energy for a single membrane undergoing the unbinding transition was proposed by Helfrich [19]. Equation (3) is the form of the free energy which will be used in our model.

*Unbinding transitions of mixed membranes.* – So far, the unbinding transition of a single-component membrane has been discussed. In the case of multicomponent membranes, the lateral phase separation affects the direct interactions between the membrane and the substrate, and hence their unbinding behavior. For simplicity, we consider a two-component membrane adhering to a substrate as in fig. 1. The overall membrane state is characterized by its average distance  $\ell$  from the substrate. The internal degree of freedom, on the other hand, corresponds to the membrane composition. In addition to the lipid component, that is the main building block of the membrane, we introduce a second component or an “inclusion” representing additional proteins, cholesterol, or lipopolymers residing on the membrane. Let the concentration of these inclusions be denoted by  $\Phi$  ( $0 \leq \Phi \leq 1$ ). Here we discuss the case in which the interaction between two inclusions is attractive leading to a condensation transition. Then, below a certain temperature  $T_c$ , the mixed membrane undergoes a first-order transition, where an inclusion-poor phase coexists with an inclusion-rich phase as observed experimentally [20]. The first-order transition terminates at a critical point, having a critical concentration  $\Phi_c$ . The concentration difference is then defined as  $\phi = \Phi - \Phi_c$  [21].

We now combine the unbinding transition and the lateral phase separation of the inclusion close to the critical point. The proposed free energy *per unit area* of a single mixed membrane undergoing the unbinding transition is

$$f(\phi, w) = -\mu\phi + \frac{1}{2}t\phi^2 + \frac{1}{4}\phi^4 - \chi w + \frac{1}{2}w^2 + \gamma\phi w, \quad (4)$$

with the constraint  $w \geq 0$ . Here all energy terms have been scaled by  $2b(k_{\text{B}}T)^2/\kappa\delta^2$ , and are now dimensionless. The first three terms in eq. (4) depend only on  $\phi$ , and correspond to the

Landau free energy of a two-component membrane undergoing a lateral inclusion-lipid phase separation [22],  $\mu$  is the chemical potential conjugate to  $\phi$ , and  $t \sim (T - T_c)$  the reduced temperature. The quartic coefficient has been chosen to be positive and is arbitrary set to  $\frac{1}{4}$  without loss of generality. The next two terms depend only on  $w$ , and represent the unbinding transition of a single membrane as have been described by eq. (3). In the above,  $\chi$  is the scaled virial coefficient.

The last term is the lowest-order coupling term between  $\phi$  and  $w$  with a dimensionless coupling coefficient  $\gamma > 0$ . The physical meaning of this bilinear term is as follows. When the mixed membrane is quenched into the two-phase region, an inclusion-poor phase ( $\phi < 0$ ) coexists with an inclusion-rich phase ( $\phi > 0$ ). This can lead to different direct interactions and hence different second virial coefficients  $\chi$  for each of the domains. We model this situation by considering an effective virial term in eq. (4) as  $-\chi_{\text{eff}}w = -(\chi - \gamma\phi)w$ , which leads to the coupling term  $\phi w$ .

In the above model, we have assumed that the phase separation of the inclusion molecules does not affect the homogeneous bending rigidity of the membrane. In other words, only the direct microscopic (van der Waals, electrostatic, hydration) interactions are modified by the phase separation, whereas the steric interaction remains unchanged. This is valid as long as the size of the inclusion molecule is close to the membrane thickness  $\delta$ . We note that a different scenario was suggested in [21, 23], where fluid membranes with spatially varying bending rigidity have been considered. In these models, the coupling between membrane curvature and inclusion concentration leads to a lateral phase separation.

Minimizing the free energy (4) with respect to  $w$  gives  $w = 0$  (unbound) for  $\phi > \chi/\gamma$  and  $w = \chi - \gamma\phi > 0$  (bound) for  $\phi < \chi/\gamma$ . By substituting back the value of  $w$  into eq. (4), the free energy as a function of  $\phi$  only becomes

$$f(\phi) = -\mu\phi + \frac{1}{2}t\phi^2 + \frac{1}{4}\phi^4, \quad \text{for } \phi > \chi/\gamma, \quad (5)$$

$$f(\phi) = -\frac{1}{2}\chi^2 + (\gamma\chi - \mu)\phi + \frac{1}{2}(t - \gamma^2)\phi^2 + \frac{1}{4}\phi^4, \quad \text{for } \phi < \chi/\gamma, \quad (6)$$

depending on the value of  $\chi/\gamma$ . Next, this free energy is minimized with respect to  $\phi$  to find the equation of state. The two-phase region is obtained by the Maxwell construction, and the whole phase diagram is calculated numerically.

*Phase behavior.* – Hereafter we set  $\gamma = 1$  without loss of generality. Then the phase behavior of the above model depends only on the value of  $\chi$ . For  $\chi > 0$ , a second-order transition line (critical line where  $\phi = \chi$ ) separates the unbound phase from the bound one, as shown in fig. 2 for  $\chi = 1$ . The critical line ends at a critical end-point (CEP —denoted by a filled square in fig. 2) on the first-order transition line. This first-order line itself ends at an ordinary critical point (CP —denoted by a filled circle in the figures) corresponding to a liquid/vapor-type CP between the two bound phases (B1+B2). Below the CEP temperature, the first-order line separates the bound phase from the unbound phase (B1+U).

The CP between the two bound phases is located at  $(\phi_c, t_c) = (0, 1)$ . Hence, the critical temperature is increased from  $t_c = 0$  to 1 due to the coupling between the inclusion concentration  $\phi$  and the membrane-substrate distance  $\ell = \delta/w$ . In other words, the phase separation is *enhanced* by the adhesion for  $\chi > 0$ . This result is in accordance with previous theoretical models [7, 9, 10]. Between the CEP and the CP of the two-phase region (B1+B2), the two coexisting values of  $\phi$  lead to different membrane-substrate distances given by  $\ell = \delta(\chi - \gamma\phi)^{-1}$ . Since the  $\ell$  value for B1 is smaller than that for B2, the phases B1 and B2 correspond to the

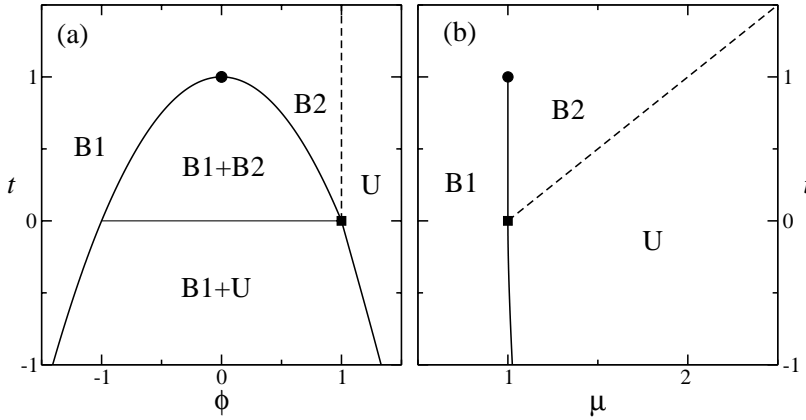


Fig. 2 – The phase diagrams for  $\chi = 1$  and  $\gamma = 1$  as a function of (a) inclusion concentration  $\phi$  and reduced temperature  $t$ , and (b) inclusion chemical potential  $\mu$  and reduced temperature  $t$ . The continuous line is a first-order line, whereas the dashed line is a second-order one. The critical point and the critical end-point are indicated by a filled circle and a square, respectively. The bound ( $w > 0$ ) and the unbound ( $w = 0$ ) phases are denoted by B and U, respectively. Below the critical point, there is a coexistence region either of two bound phases (B1+B2), or bound and unbound phases (B1+U).

“tight” and the “loose” bound phases, respectively, as discussed in [7]. At  $\chi = 0$  the CEP meets with the CP and becomes the double critical end-point.

When  $\chi$  becomes negative, the second- and first-order lines join up at a tricritical point (TCP —denoted by a filled triangle in fig. 3). In addition to the coexistence between the bound and the unbound phases (B+U1), a new two-phase region between the two unbound phases (U1+U2) appears for  $-1/\sqrt{2} < \chi < -2/3\sqrt{3}$ . As an example, fig. 3 gives the phase diagrams for  $\chi = -0.5$ . In this case, the TCP is located at  $(\phi_{tcp}, t_{tcp}) = (-0.5, 0.25)$ , and the

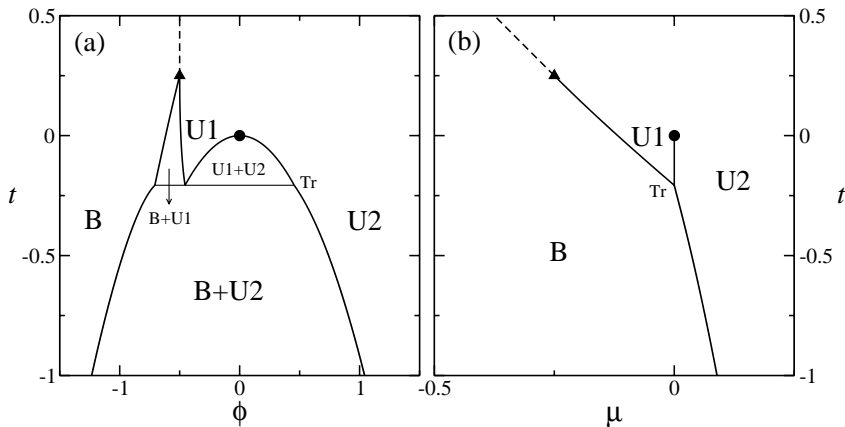


Fig. 3 – The phase diagram for  $\chi = -0.5$  and  $\gamma = 1$  as a function of (a) inclusion concentration  $\phi$  and reduced temperature  $t$ , and (b) inclusion chemical potential  $\mu$  and reduced temperature  $t$ . The same notation of different lines and symbols is used as in fig. 2. The critical point is indicated by a filled circle, the tricritical point by a filled triangle, and the triple point by Tr.

CP at  $(\phi_c, t_c) = (0, 0)$ . Notice that once the membrane unbinds from the substrate due to the Helfrich repulsion, the lateral phase separation between the two unbound phases occurs simply because of the temperature quench ( $t < 0$ ). There is also a triple point (Tr) at which the bound phase and the two unbound phases coexist (B+U1+U2). Below this triple point temperature, there is a region of two-phase coexistence between the bound and the unbound phases (B+U2).

For  $\chi < -1/\sqrt{2}$ , a similar  $(\phi, t)$ -phase diagram as in fig. 2 can be obtained (not shown here) with the critical line being located at  $\phi = \chi < 0$ . Below the CP at  $(\phi_c, t_c) = (0, 0)$ , the two unbound phases coexist until  $t$  reaches the CEP temperature. Then a region of two-phase coexistence between the bound and the unbound phase appears below the CEP. For  $\chi < 0$  the interaction is repulsive on average, but can become attractive when the phase separation becomes sufficiently strong. In this case, the effective second virial coefficient  $\chi_{\text{eff}} = \chi - \gamma\phi$  turns out to be positive for negative enough values of  $\phi$ .

In the model, the unbinding transition becomes first order below CEP or TCP for any value of  $\chi$ . This result is in accordance with the experimental prediction [12]. Our theory also describes the coexistence between the two unbound states where the membrane has an infinite separation from the substrate. In the experiment, the finite separation of the membrane was attributed to the gravitational effect [12]. Within our model, the gravitational energy per unit area should enter as a  $g/w$  term in eq. (4) with  $g > 0$ . In the presence of such a term, we obtain a coexistence between the two bound states with finite separations, which is in agreement with the experiment. However, the bound membrane goes continuously away to infinity and unbinds for  $g \rightarrow 0$  when  $\phi$  is large enough. This is the limit we have examined in this paper.

The present results can be compared with those obtained in other theoretical works. In a previous phenomenological model for the adhesion-induced lateral phase separation [7], we have considered only the phase coexistence between two bound phases. Hence a possible unbound state of the membrane was not taken into account. A phase coexistence between bound and unbound states has been predicted independently in [9, 10] by a mean-field treatment of a lattice model and Monte Carlo simulations. In those works, the topology of the phase diagram depends on several quantities such as the sticker binding energy, the potential range, or the strength of the *cis*-interaction between the stickers. However, the coexistence between the two unbound phases has not been predicted within the lattice model. A final remark is that the sequence of the phase behavior obtained from our model is similar to that for sponge phases in surfactant solutions when the symmetry-breaking parameter is varied [24].

In summary, we have proposed a phenomenological model for the unbinding transition of a two-component fluid membrane. The coupling between lateral phase separation and the membrane-substrate potential explains some of the recent experimental results. Further extensions of the present study, such as the effect of finite tension on the membrane, or adhesion between two fluctuating mixed membranes are currently under investigation.

\* \* \*

We thank H. DIAMANT, S. MARX, D. ROUX, E. SACKMANN, H. SHIROTORI, and T. WEIKL for useful discussions and comments. Support from the Ministry of Education, Culture, Sports, Science and Technology, Japan (Grant-in-Aid for Scientific Research No. 15540395) for SK, and from the Israel Science Foundation under grant No. 210/02 and the Alexander von Humboldt Foundation for DA is gratefully acknowledged. DA also would like to thank the Japan Society for the Promotion of Science (JSPS) for a travel grant.

## REFERENCES

- [1] LIPOWSKY R. and SACKMANN E. (Editors), *Structure and Dynamics of Membranes*, Vol. **1B** (Elsevier, Amsterdam) 1995.
- [2] LECKBAND D. E., ISRAELACHVILI J. N., SCHMITT F.-J. and KNOLL W., *Science*, **255** (1992) 1419.
- [3] ZUCKERMAN D. and BRUINSMA R., *Phys. Rev. Lett.*, **74** (1995) 3900.
- [4] ALBERSDÖRFER A., FEDER T. and SACKMANN E., *Biophys. J.*, **73** (1997) 245.
- [5] NARDI J., FEDER T., BRUINSMA R. and SACKMANN E., *Europhys. Lett.*, **37** (1997) 371.
- [6] BRUINSMA R., BEHRISCH A. and SACKMANN E., *Phys. Rev. E*, **61** (2000) 4253.
- [7] KOMURA S. and ANDELMAN D., *Eur. Phys. J. E*, **3** (2000) 259.
- [8] LIPOWSKY R., *Phys. Rev. Lett.*, **77** (1996) 1652.
- [9] WEIKL T. R., NETZ R. R. and LIPOWSKY R., *Phys. Rev. E*, **62** (2000) R45.
- [10] WEIKL T. R. and LIPOWSKY R., *Phys. Rev. E*, **64** (2001) 011903.
- [11] WEIKL T. R., ANDELMAN D., KOMURA S. and LIPOWSKY R., *Eur. Phys. J. E*, **8** (2002) 59.
- [12] MARX S., SCHILLING J., SACKMANN E. and BRUINSMA R., *Phys. Rev. Lett.*, **88** (2002) 138102.
- [13] DE GENNES P.-G., *Scaling Concepts in Polymer Physics* (Cornell University, Ithaca) 1979, p. 69.
- [14] HELFRICH W., *Z. Naturforsch.*, **33a** (1978) 305.
- [15] SORNETTE D. and OSTROWSKY N., in *Micelles, Membranes, Microemulsions, and Monolayers*, edited by GELBART W. M., BEN-SHAUL A. and ROUX D. (Springer-Verlag, New York) 1994, p. 251.
- [16] LIPOWSKY R. and LEIBLER S., *Phys. Rev. Lett.*, **56** (1986) 2541.
- [17] LEIBLER S. and LIPOWSKY R., *Phys. Rev. B*, **35** (1987) 7004.
- [18] MILNER S. T. and ROUX D., *J. Phys. I*, **2** (1992) 1741.
- [19] HELFRICH W., in *Phase Transitions in Soft Condensed Matter*, edited by RISTE T. (Plenum Press) 1989, p. 271.
- [20] KELLER S. L., PITCHER III W. H., HUESTIS W. H. and MCCONNELL H. M., *Phys. Rev. Lett.*, **81** (1998) 5019.
- [21] In the continuum treatment of the membrane, we do not distinguish between the lower and the upper leaflets of the membrane. However, for a mixture of two different lipids, it is of interest to consider the phase separation between the two leaflets. See, *e.g.*, NORO M. G. and GELBART W. M., *J. Chem. Phys.*, **11** (1999) 3733.
- [22] A gradient term in  $\phi$  results in line tension associated with interfaces between coexisting phases. Such a term is not important for the present treatment since only homogeneous membrane states are considered.
- [23] NETZ R. and PINCUS P., *Phys. Rev. E*, **52** (1995) 4114.
- [24] ROUX D., COULON C. and CATES M. E., *J. Phys. Chem.*, **96** (1992) 4174.