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A STATISTICAL MECHANICAL TREATMENT OF FATTY ACYL CHAIN ORDER IN PHOSPHOLIPID BILAYERS AND CORRELATION WITH EXPERIMENTAL DATA

A. THEORY

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A theoretical model has been developed in order to describe the organization of acyl chains in phospholipid bilayers. Since the model is intended to reproduce highly quantitative experimental results such as the deuterium magnetic resonance (NMR) data and to supplement the experimental information, all the rotameric degrees of freedom, the excluded volume interactions and the van der Waals interactions have been considered. The model is a direct extension of a generalized van der Waals theory of nematic liquid crystals to flexible molecules. In this picture, the anisotropy of the short-range repulsive forces which are treated by a hard core potential is introduced as the dominant factor governing intrinsic order among the chains. The anisotropy of the attractive forces, which are approximated by a molecular field, plays a somewhat secondary role. The dependence of the energy of interaction on the relative chain conformations is approximated by two order parameters reflecting respectively the 'average shape' of the molecules and the 'average shape' in a 'mean orientation'. The influence of the interactions in the polar region on the lateral chain area is accounted for by an effective lateral pressure. In certain aspects the model has features in common with the Marčelja theory.

Introduction

Pure phospholipid bilayers can be considered the simplest model of biological membranes, and for this reason they have been the object of many physical studies during the last few years. The most dramatic effect occurring in a membrane, the gel to liquid-crystal transition, has been widely documented by calorimetry [1], dilatometry [2] and various other methods (see references cited in Refs. 2 and 3). Many investigators using very different techniques have attempted to characterize the physical state of the acyl chains in both phases. Of all the methods, deuterium magnetic resonance (^{2}H-NMR) [4] using selectively deuterated phospholipid molecules has provided a unique and detailed picture of the acyl chains above the phase transition. A recent improvement of ^{2}H-NMR using the quadrupolar spin-echo technique [5] may provide similar data below the transition temperature [6]. ^{2}H-NMR, by using non-perturbing and specific probes, has the advantage that the signal can easily be assigned. Moreover, neutron diffraction [7] of selectively deuterated lamellar samples has been used to complement the NMR results. An exact description of the state of the chains in terms of the most probable spectrum of chain segment conformations or in terms of mean spatial occupancy is still not fully possible. This situation has lead to some controversy regarding the interpretation of ^{2}H-NMR data [4,8].

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In order to understand the dynamic and conformational meaning of these physical studies, several theoretical approaches have been devised. The large numer of theoretical models for a lipid bilayer [3,9–28] is symptomatic of the complexity of the physical situation. As an exact theoretical solution to this problem is not yet possible, two main lines of approximate solutions are considered. One approach is to simplify the actual physical situation into one that can be solved exactly or by controlled approximations [29]. It is considered the most rigorous and has the most physical meaning. Another approach is phenomenological in nature. It tries to solve the general model by resorting to approximations that may, in part, lose physical meaning. The success of the model along with the approximations cannot be used to justify the model itself. However, we have deliberately chosen the second alternative. The use of mathematical approximations can be applied directly to monitor the physical data as it provides a means to reproduce exactly these data. Presently, the more rigorous approach resulting from the simplification of the molecular situation does not share this capability. Therefore the choice here of a particular type of model results from the practical necessities of reproducing the experimental data and should not be regarded as a permanent alternative to other useful theoretical approaches. As will be shown in the accompanying paper [36], the analysis which follows does indeed reproduce the experimental results with accuracy and most of all, it is only because of this accuracy that the basis of specific experimental data can be understood.

The Marčelja theoretical approach, which involves the computation of each chain conformation, is a model which has been really directed toward the accurate reproduction of highly specific experimental data such as the ^{2}H-NMR results. A previous work [30] using ^{2}H-NMR data from bilayers of dipalmitoyl-3-*sn*-phosphatidylcholine illustrated the applicability of the Marčelja model. The model discussed in this report represents an attempt to improve the Marčelja theory with regard to the following points. (a) As an extension to flexible molecules of the Maier and Saupe theory for a nematic liquid crystal [31] Marčelja's theory suffers from the same lack of consideration of the effect of the short-range repulsions between the molecules on the order of the system [32]. (b) The molecular field expression, which for obvious reasons is not the same as for rod-like molecules, is successful only through the insertion of an ad hoc phenomenological parameter.

In recent works, Cotter [33,34] and Gelbart and Baron [35], in complementary but independent efforts, have described a generalized van der Waals theory of a nematic liquid crystal which considers both the strong short-range repulsions and weak longer-range attractions between rigid rod-like molecules. Interestingly, their theories introduce explicity the molecular shape dependence in the energy of interaction. In the present model we follow directly the derivations of these authors and adapt them to the complex case of flexible molecules by means of some approximations. The anisotropy of short-range intermolecular repulsion and the overall strength of the intermolecular attraction are introduced as the dominant factors governing intrinsic order among the chains, while the anisotropy of the attractive forces plays a somewhat secondary role.

Although our main goal is to produce a theoretical tool for the practical computation of all the statistical quantities describing the mean state of an acyl chain in a bilayer, we restrict ourselves in this paper to the theoretical derivation of the model. In a subsequent paper we shall present a thorough application of the theory to practical examples [36].

Theory

The organization of phospholipid molecules in bilayer leaflets exhibits a pattern which is closely analogous to the situation found in thermotropic liquid crystals of the smectic class [37]. Since our main concern is to describe the actual state of the acyl chains, we shall ignore the overall structure and limit ourselves to the approximation of a bilayer composed of two independent monolayers [3,9–28]. The important features are as follows: the interactions between polar head groups and the surrounding water and other polar head groups, which provide the energy responsible for maintaining the bilayer, modulate indirectly the interactions between the chains and will be accounted for by an effective lateral pressure, whereas the geometry of the monolayer gives rise to boundary conditions for the chain conformations. Hence the model system to be considered consists of N interacting chain molecules, each containing N_s segments subject to a lateral pressure and boundary conditions. The intermolecular interaction can be separated into two components: a repulsive force which will be treated as arising from core exclusions, and an attractive van der Waals force described by a mean field in which each molecule is assumed to interact with its neighbours.

When the average separation between the molecules due to the density of the system becomes comparable to the molecular dimensions, the internal molecular structure as well as the geometry of the molecules become important for the mutual interactions [38]. Hence, in a bilayer both the intermolecular and intramolecular energy are determined by the respective conformations of the chains. Therefore it is necessary to specify exactly the concept of conformation in this model.

The conformational states of the hydrocarbon chains can be adequately described by the arrangement of *trans* and *gauche* configurations along the chain [39]. For simplicity however, each CH_2 chain monomer is approximated by a chain segment which is represented by a vector connecting the midpoint of two consecutive C-C bonds [3,18,23]. In this segmental representation, the conformation of a chain is completely described by the polar angle with respect to the bilayer normal and the rotameric state of each segment. The all-*trans* conformation, which corresponds to all parallel segments is assumed to exist only parallel to the bilayer normal. Finally, the hydrocarbon chains can only occupy the space bounded on one side by the polar head groups and water.

In a nematic liquid crystal the anisotropy arises because of the tendency of the rod-like molecules in the fluid to align their long axis parallel to the director defined here as d. At finite temperatures, the thermal motion of the molecules prevent perfect alignment with d. The angular orientations of the molecules varies but the most probable direction is parallel to d. Clearly, ordering in the polar angle distinguishes the nematic structure from the isotropic liquid [40]. Thus in analogy to the orientational order exhibited by nematic liquid crystals, it appears quite natural to characterize the most probable state of the acyl chains by a tendency for the chains to assume the all-*trans* conformation and to get aligned parallel to the normal to the bilayer. Considering the definition of the all-*trans* conformation in this model the last condition is fulfilled by definition. Formally this implies the use of a variable conformation to describe the state of a chain and is analogous to the use of a polar angle to describe the orientational state of rod-like molecule.

On the practical side, however, such a conformational variable is rather cumbersome and has to be approximated by one or several other variables which can be handled more easily mathematically. Moreover, these new variables should (a) lead to order parameters sensitive to the thermal phase transition and (b) be appropriate for mean field treatment reflecting the symmetry as well as typical forces felt by a molecule, actually two aspects of the same phenomenom [40]. The afore-mentioned order parameters are quantities which characterize the system unequivocally both above and below the phase transition. Finally, to be exact in the formulation of this model, we note that any correlation between the rotational and translational states and the conformational states of a chain will be ignored.

First the theory of a nematic liquid crystal by Cotter [33,34] and by Gelbart and Baron [35] is extended to the case of flexible chains. Apart from the kinetic energy contribution, the configuration partition function for the system of N chains in a volume V is:

$$Q(N, V, T) = \frac{1}{N!} \sum_{c_1 \ldots c_N} \int \mathrm{d}r^{3N} \exp[-\beta U_N] \tag{1}$$

where $\beta = 1/kt$. Since a conformation is made of a particular sequence of the *trans*, *gauche*$^+$ and *gauche*$^-$ orientations along the chain [39], in our case the conformation c_i appears naturally as a discrete variable in contrast to the orientation of hard rods. The interaction potential, U_N, depends on both the conformations $c_1 \ldots c_N$ and the positions $r_1 \ldots r_N$ of the chains. If we assume that a chain can take n different conformations, we can perform the sum over the conformations in Eqn. 1 in close analogy with a method first developed by Onsager [41]. We introduce the number N_c of chains in the conformation c and rewrite Eqn. 1

$$Q = \frac{1}{N!} \sum_{\{N_c\}} \frac{N!}{N_1! \ldots N_n!} \int \mathrm{d}r^{3N} \exp[-\beta U_N(r^{3N}, \{N_c\})] \tag{2}$$

The sum is over all conformational distributions $\{N_c\} = \{N_1, \ldots N_n\}$ with $\Sigma_c N_c = N$. In the maximum term approximation one retains only the term with the distribution $\{N_c\}_{\max}$ giving the maximal contribution

$$Q = \left(\prod_{c=1}^{n} N_c!\right)^{-1} \int \mathrm{d}r^{3N} \exp[-\beta U_N(r^{3N}, \{N_c\}_{\max})] \tag{3}$$

Taking the natural logarithm and introducing the quantities

$$p_c = N_c/N \qquad \text{and} \qquad \rho = N/V \tag{4}$$

we get the result

$$N^{-1} \ln Q = 1 - \ln \rho - \sum_{c=1}^{n} p_c \ln p_c + N^{-1} \ln[V^{-N} \int \mathrm{d}r^{3N} \exp[-\beta U_N(r^{3N}, \{p_c\})] \tag{5}$$

The configurational free energy F then takes the form

$$\beta N^{-1} F = -N^{-1} \ln Q = -1 + \ln \rho + \sum_{c=1}^{n} p_c \ln p_c - \beta N^{-1} \phi_N(\{p_c\}) \tag{6}$$

Here $-Nk\Sigma_{c=1}^{n} p_c \ln p_c$ is the mixing entropy of the chains which are distinguishable because of their different conformations. ϕ_N is the excess free energy due to the elasticity of the chains and to their mutual interactions and is considered to be the sum of two terms. The first contribution, the intramolecular energy is given by

$$\phi_{\text{intra}} = N \sum_{c=1}^{n} p_c E_{\text{int}}(c) = N\langle E_{\text{int}}\rangle \tag{7}$$

where $E_{\text{int.}}(c)$ is the internal energy of a chain with the conformation c.

The intermolecular energy will be considered as a pair potential split into a long-range attractive and a short-range repulsive part, both depending on the distance vector r_{ij} and the conformations c_i and c_j.

$$U_N(r^{3N}, c_1 \ldots c_N) = U_N^{(a)}(r^{3N}, c_1 \ldots c_N) + U_N^{(r)}(r^{3N}, c_1 \ldots c_N) = \sum_{1 \leqslant i < j \leqslant N} U_2^{(a)}(\boldsymbol{r}_{ij}, c_i, c_j) + \sum_{1 \leqslant i < j \leqslant N} U_2^{(r)}(\boldsymbol{r}_{ij}, c_i, c_j) \tag{8}$$

Following the generalized van der Waals method [34,35] we extract for the given distribution p_c, the attractive part ${U_N}^{(a)}$ $(r^{3N}, C, \ldots, C_N)$ of the total interaction U_N $(r^{3N}, C, \ldots C_N)$ by approximating ${U_N}^{(a)}$ by its average value and obtain from the last term of Eqn. 5

$$\phi_{\text{inter}} = -\boldsymbol{k}T \ln \{e^{-\beta \bar{U}_N^{(a)}(\{p_c\})} V^{-N} \int \mathrm{d}r^{3N} \times \exp[-\beta U_N^{(r)}(r^{3N}, \{p_c\})]\} \tag{9}$$

${\bar{U}_N}^{(a)}$ is approximated by means of a pair distribution function [42] where the pair correlation function itself for two chains of fixed conformations is approximated by the repulsive interaction alone, so that

$$\phi_{\text{inter}} = \frac{N^2}{2V} \sum_{c,c'} p_c p_{c'} \int \mathrm{d}^3 r U_2^{(a)}(\boldsymbol{r}, c, c') \times \exp[-\beta U_2^{(r)}(\boldsymbol{r}, c, c')] - \boldsymbol{k}T \ln V^{-N} \int \mathrm{d}r^{3N} \times \exp[-\beta U_N^{(r)}(r^{3N}, \{p_c\})] = \phi^{(a)} + \phi^{(r)} \tag{10}$$

At this point, the free energy is a sum of the mixing entropy, the internal energy ϕ_{intra}, the attractive potential energy $\phi^{(a)}$ and a contribution from the repulsion, $\phi^{(r)}$. If ${U_N}^{(r)}$ is expressed by a hard core potential, then $\phi^{(r)}$ corresponds to a packing entropy, which is of importance for flexible chains packed in a bilayer.

$\phi^{(r)}$ has been investigated for liquid crystals of rod-like molecules by a scaled particle theory [33,43] and for liquid crystals of slightly non-spherical molecules by a perturbation theory [44]. Both methods actually are designed to overcome the difficult problem of the evaluation of the pair correlation function. The common result is that $\phi^{(r)}$ has the general form

$$\phi^{(r)}_{\text{h.r.}} \propto a_1(\rho, D) - a_2(\rho, D)\langle P_2(\cos\theta)\rangle^2 \tag{11}$$

where h.r. stands for hard rod. In the case of hard rod molecules the term $\langle P_2(\cos\theta)\rangle^2$ is obtained [33] by approximating the following integral

$$\langle\langle|\sin(\Omega\cdot\Omega')|\rangle\rangle = \iint|\sin(\Omega\cdot\Omega')|f(\Omega)\cdot f(\Omega')\,\mathrm{d}\Omega\,\mathrm{d}\Omega'$$

by (11a)

$$\langle\langle|\sin\langle(\Omega\cdot\Omega')|\rangle\rangle \cong \frac{\pi}{4} - (5\pi/32)\langle P_2\cos\theta\rangle^2$$

where Ω and Ω' are the sets of Euler angles and $f(\Omega)$ the orientational distribution function. In Eqn. 11a, $(\Omega - \Omega')$ is the angle between the long axis of two molecules with orientations and respectively. The set of variables (θ, ρ, D) for $\phi^{(r)}_{\mathrm{h.r.}}$ illustrates the dependence upon the polar angle with the director axis θ, the macroscopic density ρ, and the dimensions and shape of the molecules globally expressed here by D. The symbols a_1 and a_2 are two functions of the density and molecular shapes and $\langle P_2(\cos\theta)\rangle$ is the thermodynamic average of the second Legendre polynomial. The main problem, and it will become the central assumption of the theory, is to 'translate' the general form of Eqn. 11 into a semi-empirical expression for flexible chains. First of all we note in Eqn. 11 the dependence upon the shape and upon the orientation relative to the director axis. Hence we assume that the conformational dependence of the interaction between chain molecules may be reduced to a shape and an orientational problem. To obtain an expression similar to Eqn. 11, it is important to note that, in contrast to rigid molecules, the molecular shapes must be thermodynamically distributed. Observing in addition that a_2 (ρ, D) has to vanish for hard spheres, we replace the shape function with a stretching factor $\sigma(c)$. Parenthetically de Gennes [45] and Jähnig [3,2] also used a similar concept to describe the state of a chain, but with a different physical meaning. Here σ describes, in the interchain interactions, the ordering effect due only to the shape anisotropy of the respective conformations. It does not account for a unidirectional constraint as in Refs. 3, 23, 45. $\sigma(c)$ in 0 for coiled conformations (to be defined later on) and 1 for fully extended chains. For the orientation of the overall chain, the mean value

$$\overline{P_2(\cos\theta_c)} = \frac{1}{N_s}\sum_{j=1}^{N_s} P_2(\cos\theta_c^j)$$

(first introduced by Marčelja [18]) is summed over all segments. Here θ_c^j denotes the angle between the bilayer normal and the normal to the plane spanned by the C^j-H bonds. This expression implies that an effective axial symmetry is assumed for the molecules as a whole. In analogy to Eqn. 11 we make the assumption that $\phi^{(r)}$ has the form

$$\phi^{(r)} = NkT(R_0\langle\sigma\rangle^2 - R_2N_s\langle\sigma\overline{P}_2\rangle^2) \tag{12}$$

It is quite clear that in comparison to Eqn. 11. Eqn. 12 is less informative since now the constants R_0 and R_2 are free parameters to be fitted from the experimental data, whereas $a_1(\rho, D)$ and $a_2(\rho, D)$ are analytical functions [33].

To specify more closely the stretching factor for any conformation we proceed by some heuristic considerations. For example, the fully extended chain (all-*trans* conformation) gives the largest repulsive contribution in stabilizing conformational order. We can, therefore, correlate the shape contribution to the anisotropy of the short-range intermolecular repulsion with the degree of extension of the chain. A chain conformation having no two adjacent segments with the same orientation is assumed not to stabilize conformational order. With this definition the stretching factor becomes

$$\sigma(c) = \frac{N_{\mathrm{tr}}}{N_s} \tag{13}$$

where N_{tr} is the number of segments in the *trans* state. The factor N_{tr}/N_s was first introduced by Marčelja [18]. The analytical expression proposed for $\sigma(c)$ is based on rather empirical considerations and other forms are conceivable. For example, we could assume that the hydrocarbon chains in the bilayer fill cylindrical volumes $V(c)$ = constant, with a height L_c, which becomes $L_{\max}$ for the fully extended conformation. For a critical height $L_D = \sqrt[3]{4V/\pi}$ the shape becomes more sphere-like, since the height and diameter are equal. This leads to the alternative definition.

$$\sigma(c) = \frac{L_c - L_D}{L_{\max} - L_D} \tag{14}$$

Actually a decision on favouring one expression over another can be based on a comparison with the experimental data. The important point, however, is to note that an unequivocal description of the interaction between two chains may not be reduced to a simple orientational formulation. Similar considerations apply to the attractive interaction. Let us first reconsider the free energy pertaining to this interaction (Eqn. 10)

$$\phi^{(a)} = \tfrac{1}{2}N\rho \sum_{c,c'} p_c p_{c'} \int d^3 r\, U_2^{(a)}(\boldsymbol{r}, c, c') \times \exp[-\beta U_2^{(r)}(\boldsymbol{r}, c, c')] \qquad (15)$$

It is clear from Eqn. 15 that the average of the attractive interaction introduced in Eqn. 9 is equivalent to considering that the chains are moving in a spatially uniform one particle potential (mean) field as described by Cotter [34]

$$\begin{aligned} U_1^{(a)}(c) &= \rho \sum_{c'} p_{c'} \int d^3 r\, U_2^{(a)}(\boldsymbol{r}, c, c') \times \exp[-\beta U_2^{(r)}(\boldsymbol{r}, c, c')] \\ &= \rho \sum_{c'} p_{c'} \int_{S(c,c')} d^3 r\, U_2^{(a)}(\boldsymbol{r}, c, c') \end{aligned} \qquad (16)$$

where the hard core repulsion confines the range of the separation vector $\boldsymbol{r}$ to lie outside the surface S (c, c') of the excluded volume and hence depends on the conformation of both molecules. For hard rods a simplified form for the dispersion interaction yields [35]

$$\begin{aligned} U_{1,\text{h.r.}}^{(a)}(\theta_i) &= \rho \int d\Omega_j f(\Omega_j) \int_{S(i,j)} d^3 r_{ij} \times \left[-\frac{C_{\text{iso}}}{r_{ij}^6} - \frac{C_{\text{aniso}}}{r_{ij}^6} \cos\theta_{ij} \right] \\ &= -\rho[b_0 + b_2 P_2(\cos\theta_i)\langle P_2\rangle + \ldots] \end{aligned} \qquad (17)$$

θ_{ij} is the angle between the two molecular axes, $f(\Omega_j)$ the singlet orientational distribution function, and θ_i the angle between the molecular long axis and the nematic director, The striking result is that b_2 and b_0 are dominated by the shape-dependent integral over the isotropic term, C_{iso}/r_{ij}^6 [35,46]. Thus, even if the intermolecular attractions were completely isotropic, the effective potential felt by a single molecule would be orientation dependent. This is because the hard rod repulsions 'build in' short-range correlation between r_{ij} and the angle θ_{ij}. We cannot explicity perform the integration for flexible chains, but we argue that the mean attractive field must, to lowest order and apart from a constant, be proportional to our stretching factor $\sigma(c)$. Replacing again $P_2(\cos\theta_i)$ by the average $\overline{P_2(\cos\theta_c)}$ over all segments, we obtain

$$U_1^{(a)}(c) = -\rho\left[B_0 \sigma(c)\langle\sigma\rangle + B_2 \sigma(c) \sum_{j=1}^{N_s} P_2(\cos\theta_c^j)\langle\sigma\bar{P}_2\rangle\right] \qquad (18)$$

One might argue that for sphere-like molecules, where $(c) \rightarrow 0$, and which are still optically anisotropic, the one particle potential must reduce to a term proportional to $\Sigma_{j=1}^{N_s} \bar{P}_2(\cos\theta_c^j) \cdot \langle P_2\rangle$ as known from the Maier and Saupe theory. It is, however, an important result of Gelbart and Baron [35] and Gelbart and Gelbart [46] that the term $C_{\text{aniso}} \cos\theta_{ij}$ in the interaction (Eqn. 17) which produces the Maier-Saupe field, contributes only negligibly to the anisotropy of the total mean field for rod-like molecules. The same conclusion is also reached by Wulf [32.47}. It may play a certain role for extremely coiled chains. However, these are not likely to occur for several energetic considerations.

Moreover, we can hope that the mean attractive field (Eqn. 18) describes additional effects such as a possible loss of polarizability for non-parallel neighbouring segments inside the chain. The loss of polarizability may cause a deviation from the additivity [48] and decrease the mean field. The additivity is implicitly assumed for the segment-segment interaction when the arithmetic mean $P_2(\cos\theta_c)$ is used.

The attractive free energy using Eqns. 16, 18 and 19 is

$$\phi^{(a)} = -\tfrac{1}{2}N\rho[B_0\langle\sigma\rangle^2 + N_s B_2 \langle\sigma P_2\rangle^2] \qquad (19)$$

The energy is determined by the same order parameters $\langle\sigma\rangle$ and $\langle\sigma P_2\rangle$ as the repulsion term $\phi^{(r)}$, Eqn. 12. Again the positive constants B_0 and B_2 must be determined from experimental data. The total free energy, Eqn. 6, now can be written as

$$\beta N^{-1}F = \sum_c p_c \ln p_c + \beta\langle E_{\text{int}}\rangle$$
$$+ (R_0\langle\sigma\rangle^2 - N_s R_2\langle\sigma\bar{P}_2\rangle^2)$$
$$- \beta\rho\tfrac{1}{2}(B_0\langle\sigma\rangle^2 + N_s B_2\langle\sigma\bar{P}_2\rangle^2) + \ln\rho - 1 \qquad (20)$$

So far we have not considered the fact that the electrostatic interactions between the polar head groups and between the head groups and water are stabilizing the layer structure. They tend to minimize the effective area, $A(c)$, occupied by the flexible molecule in the conformation c. Therefore we introduce an effective lateral pressure π and add to F a 'volume' energy shown by the product $\pi\langle A\rangle$. Eqn. 20 then becomes the free enthalpy G by the relationship

$$\beta N^{-1}G = \sum_c p_c \ln p_c + \beta\langle E_{\text{int}}\rangle$$
$$+ (R_0\langle\sigma\rangle^2 - N_s R_2\langle\sigma\bar{P}_2\rangle^2)$$
$$- \tfrac{1}{2}\rho\beta(B_0\langle\sigma\rangle^2 + N_s B_2\langle\sigma\bar{P}_2\rangle^2)$$
$$+ \beta\pi\langle A\rangle + \ln\rho - 1 \qquad (21)$$

To be complete we should include in Eqn. 21 a head group free energy term. However, interactions between head groups, and head groups and water are rather complex and would require the introduction of further assumptions and free parameters. Since the head groups at the phase transition contribute little to the transition enthalpy [2] any additional free energy terms can be neglected in a first approximation.

In order to extract p_c, the conformational one-particle distribution function, we have to minimize $N^{-1}\beta G(p_c)$ subject to the normalization condition $\Sigma_c p_c = 1$. This is done by solving the variational equation

$$\frac{\partial\left[G/NkT - \lambda\sum_c p_c\right]}{\partial p_c} = 0 \qquad (22)$$

where λ is a Lagrange parameter. This yields

$$p_c = \frac{1}{Z}\exp\left\{-\left[(2R_0 - \beta\rho B_0)\,\sigma(c)\langle\sigma\rangle - (2R_2 + \beta\rho B_2)\right.\right.$$
$$\left.\left.\times\,\sigma(c)\sum_{j=1}^{N_s} P_2(\cos\theta_c^j)\langle\sigma\bar{P}_2\rangle + \beta E_{\text{int}}(c) + \beta\pi A(c)\right]\right\} \qquad (23)$$

where Z is the normalization constant. The self-consistent equations for the order parameters, i.e., for the molecular fields, are

$$\langle\sigma\rangle = \sum_c p_c\sigma(c)$$
$$\langle\sigma\bar{P}_2\rangle = \sum_c p_c\sigma(c)\,\overline{P_2(\cos\theta_2)} \qquad (24)$$

Introducing Eqn. 23 into Eqn. 21 the free enthalpy is explicitly

$$\beta N^{-1}G = -\ln Z - (R_0 - \tfrac{1}{2}\beta\rho B_0)\langle\sigma\rangle^2$$
$$+ (R_2 + \tfrac{1}{2}\beta\rho B_2)\,N_s\langle\sigma\bar{P}_2\rangle^2 + \ln\rho \qquad (25)$$

In Eqn. 25 the factor -1 of Eqn. 21 has been dropped as it is of no consequence. Eqns. 24 can be solved simultaneously and iteratively. Since the functions exhibits continuous loops in the neighbourhood of the phase transition, three solutions are found; two stable and one unstable. The first-order phase transition or the so-called order-to-disorder transition is obtained for two stable solutions, fulfilling the thermodynamic requirement G(gel) = G(liquid crystalline). For the purpose of simplicity we shall always set the density equal to 1 and ignore any changes at the phase transition. This assumption is largely supported by recent density measurements on lipid bilayers [2]. Using the same property and in agreement with Marčelja [18], we shall defined $\langle A\rangle = A_0L_0/\langle L(c)\rangle$, where $\langle L(c)\rangle$ and L_0 are respectively the mean and maximal chain length projections on the bilayer normal and A_0 is the cross-sectional area of the fully extended chain. Again for simplicity no PV term in Eqn. 21 will be considered. With these simplifications we can compute the following thermodynamical functions: the internal energy U, the mixing entropy S_{M}, the packing entropy S_{P} and the total entropy S.

$$N^{-1}U = -\tfrac{1}{2}(B_0\langle\sigma\rangle^2 + N_s B_2\langle\sigma\bar{P}_2\rangle^2) + \langle E_{\text{int}}\rangle$$

$$N^{-1}S_{\mathrm{M}} = \boldsymbol{k} \ln Z + \left(2\boldsymbol{k}R_0 - \frac{1}{T}B_0\right)\langle\sigma\rangle^2 - \left(2\boldsymbol{k}R_2 + \frac{1}{T}B_2\right)N_{\mathrm{s}}\langle\sigma\bar{P}_2\rangle^2 + \frac{1}{T}\langle E_{\mathrm{int}}\rangle + \frac{1}{T}\pi\langle A\rangle$$

$$N^{-1}S_{\mathrm{P}} = -\boldsymbol{k}(R_0\langle\sigma\rangle^2 - N_{\mathrm{s}}R_2\langle\sigma\bar{P}_2\rangle^2)$$

$$N^{-1}S = \boldsymbol{k} \ln Z + \left(\boldsymbol{k}R_0 - \frac{1}{T}B_0\right)\langle\sigma\rangle^2 - \left(\boldsymbol{k}R_2 + \frac{1}{T}B_2\right)N_{\mathrm{s}}\langle\sigma\bar{P}_2\rangle^2 + \frac{1}{T}\langle E_{\mathrm{int}}\rangle + \frac{1}{T}\pi\langle A\rangle \qquad (26)$$

Discussion

As noted earlier, the application of the model to experimental data as well as a comparison with the Marčelja theory will be presented in a separate communication [36].

Most of the models on lipid monolayers or bilayers emphasize the predominance of the repulsive interactions in governing order among the acyl chains. The attractive energy of interactions is regarded as providing an overall cohesive component. This view arises from the current picture of a simple liquid where the structure is thought to be determined primarily by the hard core part of the potential and that the main effect of the non-hard core contribution is to provide a uniform background potential in which the molecules move [49]. On the other hand, Marčelja's theory assumes that only the anisotropy of the attractive interactions governs order among the chains. Indeed, this fact motivated our reassessment of the Marčelja model. The present model consists of an adaptation of a generalized van der Waals theory of nematic liquid crystals as applied to flexible molecules. It is in agreement with the current views of liquids and with most of the other models for lipid bilayers, but there are some important distinctions.

First of all, in the same way as Marčelja's theory and in the limits of the boundary conditions, we are not making any arbitrary choices in accepting or rejecting a particular chain conformation. This is most important in order to obtain an accurate fit to the ^{2}H order profile as determined by NMR. This was the essential goal of the model analysis [36]. The model-data correlation is even more important in the consideration of bilayers containing cholesterol or proteins. Such membrane components impose additional boundary conditions on the system, and therefore tend to modify the ^{2}H order profile.

Secondly, most of the models include an attractive component in the potential in order to account for an enthalpy change at the phase transition. This is done rather than modulating specifically order within the acyl chains. With the present approach, an anisotropic attractive energy of interaction can be introduced and from this, it is possible to gain insight into the origin of the anisotropy.

A second objection to the Marčelja theory is directed towards the ad hoc introduction of the one particle potential expression. The detailed analysis of the influence of one chain conformation on the other chains outlined above, although performed in a heuristic way, led to the introduction of the factor $\sigma(c)$. If $\sigma(c)$ is set equal to the factor $N_{\mathrm{tr}}/N_{\mathrm{s}}$ in the Marčelja theory we can now better understand the physical basis for the Marčelja's expression and complement it. Our derivation, however, led to the introduction of the free parameters R_0, R_2 (Eqn. 12) and B_0, B_2 (Eqn. 18) and they should be considered more closely. From Cotter [33] it obvious that R_0 and R_2 are related in an unknown way to the geometrical mean of chain conformation which has not been considered explicitly in $\sigma(c)$. No direct physical meaning to these parameters can presently be assigned in the simulations. For the same reason R_0/N_{s} and R_2 may vary for hydrocarbon chains of different lengths. According to others, [35,46,50] B_0 and B_2 should reflect the geometry of the mean conformation and the strenght of the dispersion constants. It is therefore not possible to define more accurately the value of the sum $B_0/N_{\mathrm{s}} + B_2$ other than it should not exceed the heat of sublimation of one monomer (1.84 kcal/mol CH_2) [48,51]. It is also to be expected that B_0/N_{s} and B_2 will vary with the chain length. These parameters are therefore one shortcoming of this model and others in the literature [18,19,23,26].

The concept of reducing the conformational dependence of the energy of interaction between the chains to a set of two order parameters (i.e., the shape and the orientation of the chain) has already been used in the literature [3,22,23], but only with serious variance from our model. At this point we would like to emphasize the importance of choosing order parameters describing unequivocally the conformational state of a chain. Let us consider the effect of setting $\sigma(c) = 1$ in this or Marčelja's model. This would be the same as describing the state of a chain by a pure orientational order parameter. The direct consequence is that neither model predicts a phase transition. The reason is easy to understand if we remember that the entropy of mixing arises (eqns. 2 to 6) from the mixing of chains with different conformations. Approximating a chain conformation by a purely orientational factor results in an underestimation of the entropy of mixing in the liquid crystalline state and leads to the absence in the liquid crystalline state and leads to the absence of a phase transition. Consider now the factors which may influence the entropy of mixing and thus the phase transition. Clearly an increase in the effective lateral pressure will result in a decrease of the mixing entropy possibly leading first to a continuous phase transition and then to no phase transition at all. More selective boundary conditions are also a means of reducing the entropy of mixing. If we consider for example the presence of cholesterol or proteins in the membrane as further boundary conditions and assuming no finite size effects, we can expect a parallel decrease of the enthalpy change at the phase transition. Theoretically, at certain concentrations we may expect the occurrence of critical phenomena in full analogy with the nematic-isotropic transition in surface-aligned nematic films [52,53]. Finally concerning the entropy of mixing we can foresee that less than a certain chain length, if bilayers could be formed [54], then no phase transition would occur [2].

To summarize we wish to reiterate the goals and the motivations which have led to the derivation of the present model. Although we have attempted to describe in the best possible way, the phenomena associated with the gel-to-liquid thermotropic transition, the specific goal of this work is of a more practical nature. Our specific intention has been to create a theoretical tool closely associated with experimental techniques and to extend the present understanding of lamellar lipids beyond the limitations associated with the use of experimental methods alone. Specific examples of phospholipid bilayers analyzed using this model is the subject of the accompanying paper.

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