Density fluctuations in saturated phospholipid bilayers increase as the acyl-chain length decreases

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ABSTRACT A systematic computer simulation study is conducted for a model of the main phase transition of fully hydrated saturated diacyl phosphatidylcholine bilayers (DMPC, DPPC, and DSPC). With particular focus on the fluctuation effects on the thermal properties in the transition region, the study yields data for the specific heat, the lateral compressibility, and the lipid-domain size distribution. Via a simple model assumption the transmembrane passive ion permeability is derived from the lipid-domain interfacial measure. A comparative analysis of the various data shows, in agreement with a number of experiments, that the lateral density fluctuations and hence the response functions increase as the acyl-chain length is decreased.

1. INTRODUCTION

Fully hydrated pure lipid bilayers undergo a first-order acyl-chain melting transition, the so-called main transition, from the low-temperature gel phase to the high-temperature fluid phase (Cevc and March, 1987). The transitional properties, specifically the transition temperature, T_m, and the transition enthalpy, depend on the type of lipid species in question (Silvius, 1982), e.g., the length of the acyl chains in the case of fully saturated diacyl phosphatidylcholine bilayers. In the case of mixed bilayers, e.g., binary lipid mixtures, the main transition manifests itself in macroscopic phase equilibria between gel and fluid phases in certain temperature and composition regimes. This provides a mechanism for lateral organization of the membrane in terms of *static* heterogeneity, as now well characterized experimentally (Lee, 1977) and theoretically in the case of binary systems (Ipsen and Mouritsen, 1988). However, there also exists a mechanism for providing *dynamic* membrane heterogeneity for a one-component lipid bilayer due to the strong lateral density fluctuations which accompany the main transition in many lipid systems (Mitaku et al., 1983; Mouritsen and Zuckermann, 1985; Ruggiero and Hudson, 1989).

In this paper we present a theoretical model study of this type of dynamically induced membrane heterogeneity by performing a comparative and systematic analysis of the microscopic as well as macroscopic manifestations of the lateral density fluctuations in a series of saturated diacyl phosphatidylcholine bilayers, specifically dimyristoyl phosphatidylcholine (DMPC), dipalmitoyl phosphatidylcholine (DPPC), and distearoyl phosphatidylcholine (DSPC) bilayers. The theoretical analysis is carried out by means of a microscopic statistical mechanical interaction model which gives a faithful representation of the cooperative interactions between the different conformational states of the acyl chains. The properties of the model are calculated by Monte Carlo computer-simulation techniques (Mouritsen, 1984) which give direct access to the typical microscopic system configurations which underlie the phase transition and the fluctuation phenomena.

The nature of the main transition is still a topic of considerable controversy. It is generally assumed that the transition is of first order involving a considerable transition enthalpy (Albon and Sturtevant, 1978) although every experimental quantity measured varies continuously throughout the transition region. Based on careful equilibrium calorimetric measurements, Biltonen (1990) has recently argued that the transition is effectively continuous (or weakly first order) and only a vanishing small amount of heat is involved. The effectively continuous character of the transition is reflected in the behavior of the thermal response functions, such as specific heat (Albon and Sturtevant, 1978) and lateral compressibility (Evans and Kwok, 1982), which display apparent divergencies as the transition temperature is approached. These signals enhanced density fluctuations at length scales much larger than the intermolecular distances. A number of other well-studied condensed-matter systems are associated with similar phenomena, notably the isotropic-nematic (Stinson and Lister, 1973) and the smectic A—nematic (McMillan, 1972) transitions of liquid crystals, which display so-called pretransitional phenomena.

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The biophysical motivation for studying the weak-first-order nature of the main transition in lipid bilayers is formed by the observation that the accompanying density fluctuations may trigger a number of membrane processes, such as passive permeation of e.g., ions (Papahadjopoulos et al., 1973; Kanehisa and Tsong, 1978), exchange of molecules between different membrane systems (Bar et al., 1987), and activity of certain membrane-bound enzymes (Op den Kamp et al., 1975; Menashe et al., 1986). Fluctuation phenomena in lipid membranes have been studied experimentally by a number of techniques, including calorimetry (Hatta et al., 1983; Freire and Biltonen, 1978; Imaizumi and Garland, 1987), fluorescence anisotropy (Ruggiero and Hudson, 1989), permeability measurements (Papahadjopoulos et al., 1973; Georgallas et al., 1987), ultrasonic relaxation (Mitaku et al., 1978; Mitaku and Date, 1982), and nuclear magnetic resonance (Hawton and Doane, 1987).

The fluctuation phenomena in lipid bilayers have been considered theoretically on the basis of phenomenological theories (Nagle and Scott, 1978; Freire and Biltonen, 1978), by analytical treatment of Ising-like model systems (Doniach, 1978; Kanehisa and Tsong, 1978), and by computer simulations of the ten-state Pink model (Mouritsen, 1983; Mouritsen et al., 1983; Mouritsen and Zuckermann, 1985; Cruzeiro-Hansson and Mouritsen, 1988). The work presented in this paper belongs to the last category. It may be considered as a supplementary study to our recent work on fluctuation-induced interface phenomena in lipid bilayers (Cruzeiro-Hansson and Mouritsen, 1988) and how the interfaces may be modulated by the presence of interfacially active molecules such as cholesterol (Cruzeiro-Hansson et al., 1989). Here we investigate how another system parameter, the acyl chain length of diacyl phosphatidylcholine lipids, influences the fluctuation phenomena in bilayers near their main phase transition temperature.

MODEL AND CALCULATIONAL METHOD

We adopt the ten-state model of Pink (Pink et al., 1980) as an appropriate model for the description of the main transition of diacyl phosphatidylcholine bilayers in terms of the acyl-chain conformational degrees of freedom. This model accounts in a detailed manner for the conformational energy due to *gauche* excitations, the conformational statistics, and the anisotropic dispersion forces between neighboring acyl chains. In the Pink model, the excluded-volume interactions are taken into account in an approximate way by assigning each acyl chain to a site on a triangular lattice. The most likely chain conformations, selected on the basis of optimal packing and low conformational energy, are grouped into ten states. Each of the ten states α, ($\alpha = 1, 2, \ldots, 10$), is described by an internal energy, E_α, a cross-sectional area of the acyl chain, A_α, and an internal degeneracy, D_α. One of the states is highly excited, carrying a large internal energy and a cross-sectional area which is characteristic of the fluid phase. The nine remaining states represent gel-like conformations with smaller excitational energies. One of these states corresponds to the all-*trans* conformation of the acyl chain. The Hamiltonian takes the form

$$\mathcal{H} = \sum_i \sum_{\alpha=1}^{10} (E_\alpha + \Pi A_\alpha)\mathcal{L}_{i\alpha} - \frac{J_0}{2} \sum_{\langle ij \rangle} \sum_{\alpha,\beta}^{10} I_\alpha I_\beta \mathcal{L}_{i\alpha}\mathcal{L}_{j\beta}. \quad (1)$$

$\mathcal{L}_{i\alpha}$ is the occupation variable ($=0,1$) for state α of the acyl-chain at site i, $\Sigma_\alpha \mathcal{L}_{i\alpha} = 1$. The nearest-neighbor interaction constants are given by an overall van der Waals interaction strength J_0 and conformationally dependent parameters I_α. For rod-like molecules $I_\alpha \propto S_\alpha A_\alpha{}^{-5/2}$, where S_α is the orientational order parameter of the acyl chain (Caillé et al., 1980). Π ($=30$ dyn/cm) is an internal pressure applied to assure bilayer stability.

Most of the applications of the Pink model to lipid membranes are built on mean-field approximation schemes (Caillé et al., 1980). Within this framework the ten-state model has demonstrated its ability to provide an overall accurate description of a variety of experimental data for membrane systems, including spectroscopic, thermodynamic, and thermomechanic properties. However, in the transition region the accordance with experimental data is much less good due to the fact that thermal fluctuations are suppressed in the mean-field approximation. This is a particularly cumbersome problem for pure lipid bilayers which are known to exhibit strong fluctuations at the main transition in a manner which is reminiscent of critical fluctuations: the transition is pseudo-critical (Mitaku et al., 1983; Mouritsen and Zuckermann, 1985; Ruggiero and Hudson, 1989).

Therefore, more accurate calculation methods have to be drawn upon, e.g., the Monte Carlo simulation technique (Mouritsen, 1990) which in principle provides a numerically exact solution to the statistical mechanical problem posed by the Hamiltonian of Eq. 1. Due to demands of excessive computer time, Monte Carlo simulations on lipid membrane models, specifically Eq. 1, have been restricted to a few limited cases, notably that of DPPC bilayers (Mouritsen, 1990). Hence, systematic studies of Eq. 1 for different acyl-chain lengths have until now only been conducted within the mean-field approximation (Caillé et al., 1980). A result of these mean-field

TABLE 1 Parameters used for the ten-state Pink model and experimental transition temperatures for the main transition in diacyl phosphatidylcholine bilayers

	M	E_{10} 10^{-13} erg	D_{10}	J_o 10^{-13} erg	T_mK
DMPC	14	1.94	6×3^8	0.618	296.9
DPPC	16	2.78	6×3^{10}	0.709	314.0
DSPC	18	3.62	6×3^{12}	0.815	327.9

The transition temperature values are from Silvius (1982).

calculations is the finding, for varying acyl-chain lengths, that a simple linear relationship between the free model parameters leads to estimates of the transition temperature and transition enthalpy which are in good agreement with experimental results (Caillé et al, 1980).

The acyl-chain length is expected to affect only a few of the model parameters characterizing the ten-state Pink model, specifically the conformational degeneracy, $D_{10} \propto 3^M$, the conformational energy, $E_{10} \propto M$, of the highly excited tenth state, and the overall van der Waals interaction energy parameters, $J_o \sim M$, where M is the number of carbons in the acyl chain (Caillé et al., 1980). We have adopted the above relationships for D_{10} and E_{10}, whereas J_o is determined by fitting experimental transition temperatures, $T_m(M)$, to the results of a mean-field calculation of the model for each of the three lipids DMPC, DPPC, and DSPC. The resulting model parameters are listed in Table 1. The linear dependence between J_o and M is approximately fulfilled for the fitted values of T_m. In Fig. 1 is shown, as an example, the results of the mean-field calculation in the case of the average cross-sectional area per lipid molecule, $A(T)$. The parameters of Table 1 have been used for this calculation. The sharp drop of A at some temperature signals the first-order phase transition.

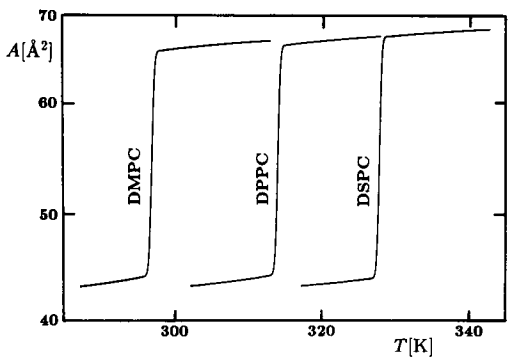

FIGURE 1 Mean-field results for the average cross-sectional area per lipid molecule as a function of temperature for DMPC, DPPC, and DSPC lipid bilayers.

To provide an accurate description of the properties in the transition region we have employed standard Monte Carlo simulation techniques (Mouritsen, 1984, 1990). The simulations are performed on lattices composed of 100×100 sites corresponding to 5,000 lipid molecules. This system size represents the thermodynamic limit according to previous finite-size analyses carried out with regard to the fluctuation properties in the transition region (Cruzeiro-Hansson and Mouritsen, 1988). The equilibrium properties are obtained by averaging over a series of system configurations generated by Glauber dynamics.

RESULTS

In this section we present the results from the numerical Monte Carlo simulations on the ten-state Pink model of the main transition in DMPC ($M = 14$), DPPC ($M = 16$), and DSPC ($M = 18$) bilayers. The simulation data are presented on a reduced temperature scale, $T/T_m(M)$, which facilitates a direct comparison of the different systems.

For all three lipid systems, the main transition is found to be accompanied by strong density fluctuations in the transition region. These fluctuations manifest themselves in terms of a lipid-cluster (or domain) formation by which the equilibrium bulk phase is invaded by clusters of the opposite phase, i.e. fluid clusters in the gel phase for $T < T_m$, and gel clusters in the fluid phase for $T > T_m$. The cluster formation leads to the creation of an interfacial environment which is defined by the borders between the clusters and the bulk (Cruzeiro-Hansson and Mouritsen, 1988). This phenomenon is a dynamic one: clusters are continuously created and annihilated and they persistently fluctuate in size. However, they are characterized by an equilibrium size-distribution function. Hence, the fluctuation phenomena lead to dynamically heterogeneous membrane configurations. Examples of such configurations are illustrated in Fig. 2 which shows snapshots of instantaneous interfacial regions formed in the transition region for each of the three lipid systems studied. These snapshots indicate the persistence of strong lateral density fluctuations in the transition region for all three types of bilayers and moreover suggest qualitatively that the fluctuations are more strongly manifested the shorter the acyl-chain length is.

A quantitative analysis of the cluster-formation phenomena is given in Fig. 3 in terms of the average cluster

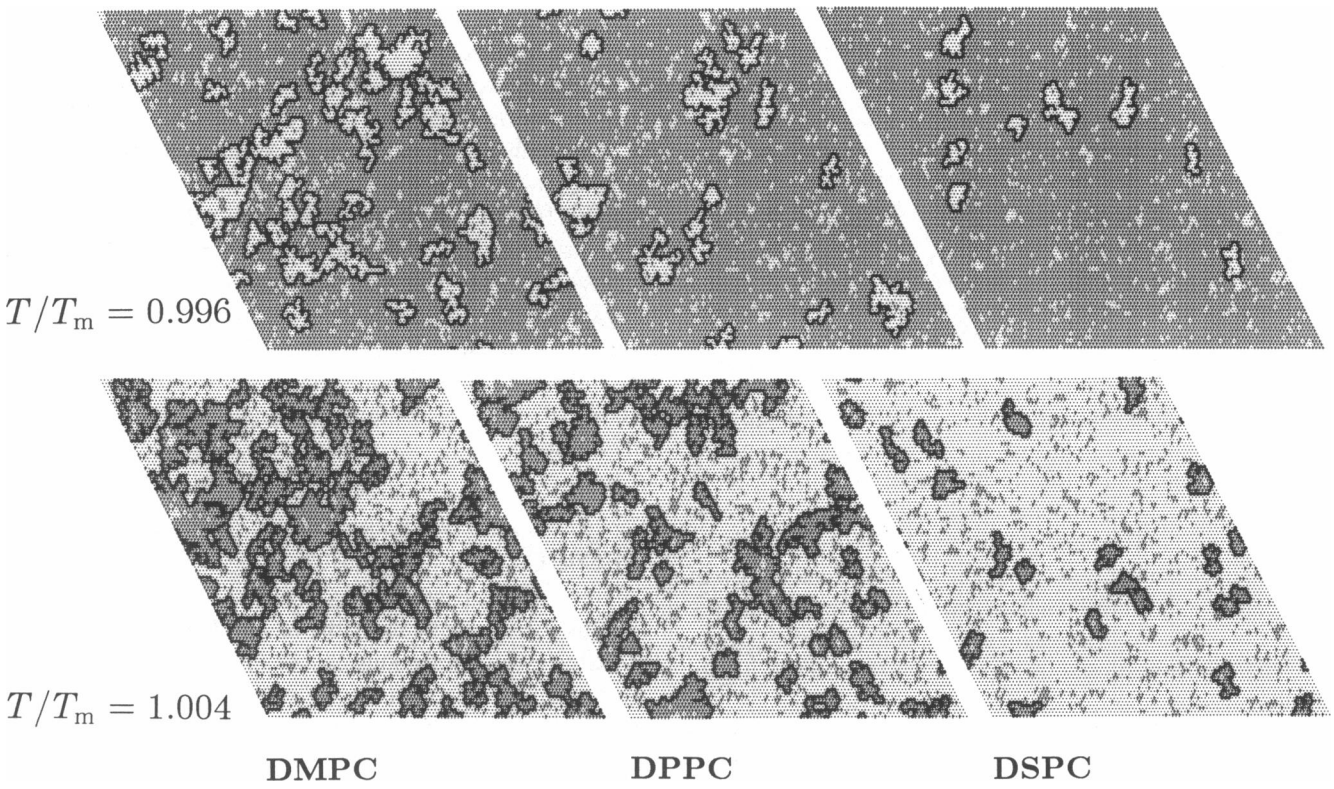

FIGURE 2 Snapshots of instantaneous microconfigurations typical of equilibrium at reduced temperatures, T/T_m, below and above the main transition of pure DMPC, DPPC, and DSPC bilayers. The light-grey regions denote the fluid phase, the dark-grey regions denote the gel phase, and the interface between the gel and fluid regions are highlighted in black.

size, $\mathcal{N}(T)$, as a function of temperature. The figure shows unequivocally that the density fluctuations are increased when the acyl-chain length is decreased. This effect is particularly pronounced in the wings of the transition point.

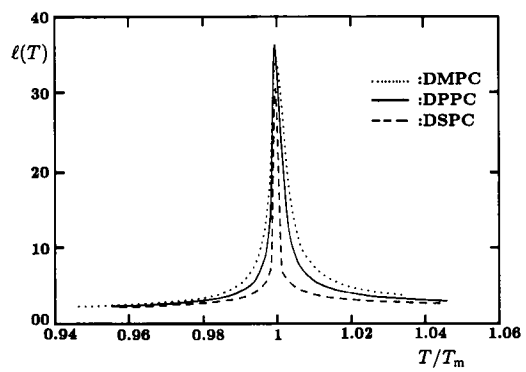

FIGURE 3 Monte Carlo data for the average lipid cluster size, $\mathcal{N}(T)$, (in units of numbers of molecules) in the transition region for DMPC, DPPC, and DSPC bilayers.

A macroscopic consequence of the density fluctuations at the main transition is a smearing out of the variation of the cross-sectional membrane area as the temperature is varied through the transition region. In Fig. 4 *a* is shown how this leads to an apparently continuous variation of the area per lipid molecule, $A(T)$, for all three lipid species. This behavior should be contrasted to that of the mean-field prediction shown in Fig. 1. It is noticed that the broadening of the transition becomes more pronounced for decreasing chain length. In Fig. 4 *b* is shown the temperature variation of that part, $A^{gel}(T)$, of the membrane area which corresponds to gel-like regions. This quantity may be of interest to those spectroscopic measurements on lipid bilayers which use certain fluorescent probes (Ruggiero and Hudson, 1989). $A^{gel}(T)$ is not a uniquely defined quantity in a microscopic model because it requires a criterion for how large a gel cluster needs to be to contribute to A^{gel}. This criterion would have to be chosen according to which phenomenon one wants to relate to the amount of gel domains. The data of Fig. 4 *b* correspond to no lower cut-off for the gel clusters and the figure is hence consistent with the data in Fig. 3 for the

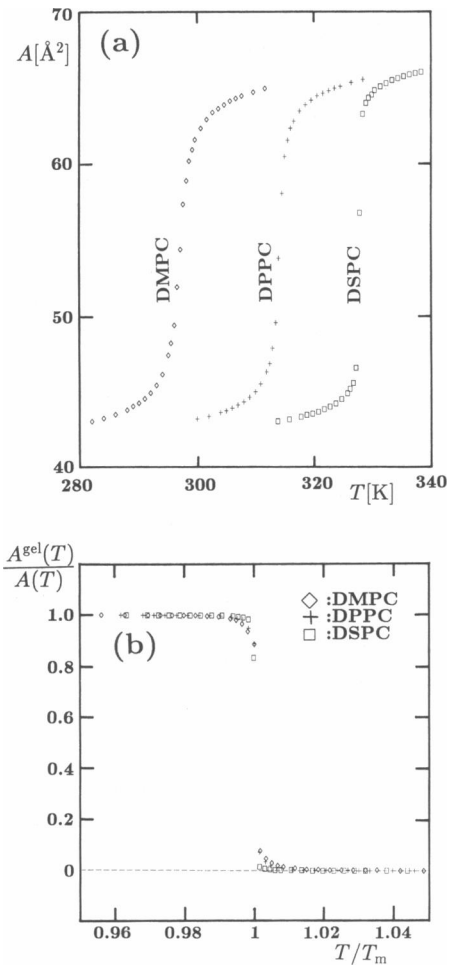

FIGURE 4 (*a*) Monte Carlo results for the average cross-sectional area per lipid molecule as a function of temperature for DMPC, DPPC, and DSPC lipid bilayers. (*b*) The part of the lipid membrane area which is associated with gel clusters.

average cluster size which did not introduce a cut-off on the cluster-size distribution function.

The macroscopic manifestations of the density fluctuations are most dramatically mirrored in the response functions, i.e., the lateral compressibility

$$\chi(T) = \frac{1}{Nk_BT}(\langle A^2\rangle - \langle A\rangle^2) \qquad (2)$$

and the specific heat

$$C_P(T) = \frac{1}{Nk_BT^2}(\langle \mathcal{H}^2\rangle - \langle \mathcal{H}\rangle^2), \qquad (3)$$

which are shown in Figs. 5 and 6, respectively. N is the number of molecules. Both $\chi(T)$ and $C_P(T)$ exhibit a

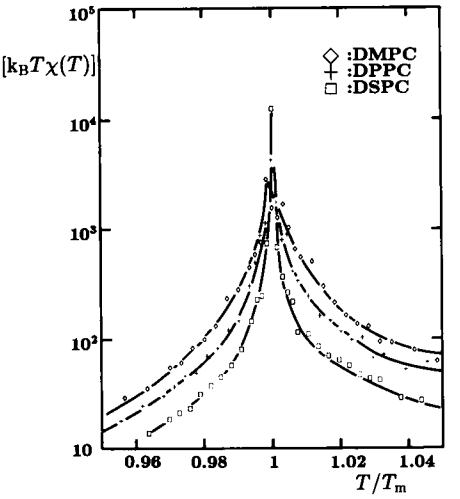

FIGURE 5 Semi-logarithmic plot of the Monte Carlo data for the lateral compressibility, $\chi(T)$ in Eq. 2, in the transition region for DMPC, DPPC, and DSPC lipid bilayers. The ordinate is in units of Å⁴.

singular behavior at the transition point. The effect of the acyl-chain length is more pronounced in the case of $\chi(T)$. The peak in $\chi(T)$ and $C_P(T)$ at the transition temperature is decreased in intensity as the acyl-chain length is reduced. However, at temperatures away from T_m a pronounced enhancement of the response functions is obtained for decreasing acyl-chain length. The overall peak in the transition therefore appears to be broadened as the chain length decreases. The behavior in $\chi(T)$ is very similar to that of the average cluster size in Fig. 3.

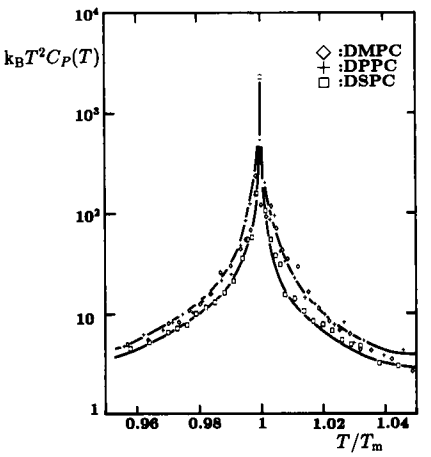

FIGURE 6 Semi-logarithmic plot of the Monte Carlo data for the specific heat, $C_P(T)$ in Eq. 3, in the transition region for DMPC, DPPC, and DSPC lipid bilayers. The ordinate is in units of 10^{-26}erg².

The peak in the response functions at T_m indicates that the coherence length, $\xi(T)$, is getting very large as T_m is approached. The thermal response functions can be expressed by $\xi(T)$. To the extent the fluctuations in some order parameter, e.g., the membrane area (Jähnig, 1981), can be described by a harmonic theory, we have $\chi(T) \sim \xi^2(T)$ away from T_m. In Fig. 7, $\chi^{-1}(T)$ is plotted vs. T. The approximate linear relation away from T_m indicates that

$$\xi^{-2}(T) \sim (T - T^*), \tag{4}$$

i.e., the classical (mean-field) critical exponent for $\xi(T)$ is found to apply for temperatures close to T_m for all three lipid species. The (pseudo-) critical temperature, T^*, is very close to T_m, $|T_m - T^*|/T_m \sim 10^{-4} - 10^{-3}$ (Mouritsen and Zuckermann, 1985). At temperatures very close to T_m correlation effects may modify the classical behavior to $\xi(T) \sim (T - T^*)^{-\nu}$, with $\nu < 1/2$, but the data are too imprecise to draw any conclusions regarding a nonclassical exponent value. In any case, the range of validity of the classical expression is very large and goes very close to T_m. The very large coherence length at $T = T_m$ leads to approximate scaling laws for the contributions to thermal quantities at characteristic length scales larger than the intermolecular distances but smaller than the maximum coherence length. Specifically we have found an effective scaling law to be operative for the cluster-size distribution function, $n(\ell) \sim \ell^{-\tau}$ for $T \sim T_m$, in the observed range of values of ℓ. The exponent value is $\tau \simeq 1.84$. This scaling law with the same exponent value holds for all three lipid species which demonstrates, together with the effective singular behavior of $\xi(T)$, an apparent universality in the pseudo-critical behavior of lipid bilayers.

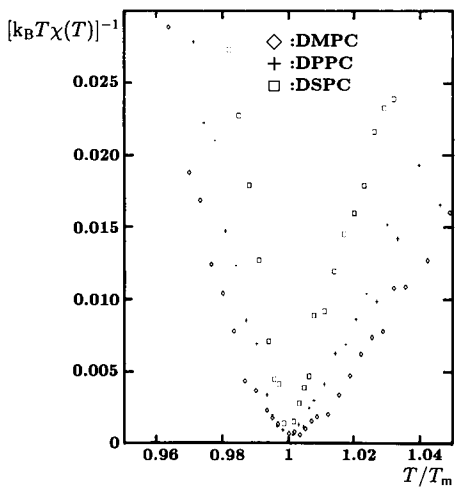

FIGURE 7 Inverse lateral compressibility, $\chi^{-1}(T)$ (cf. Fig. 6), plotted vs. temperature for DMPC, DPPC, and DSPC lipid bilayers.

The dynamically heterogeneous membrane configurations shown in Fig. 2 suggest that it is appropriate to divide the membrane area into three fractional contributions, the fractional bulk area, $a_b(T)$, the fractional cluster area, $a_c(T)$, and the fractional interfacial area, $a_i(T)$ (Cruzeiro-Hansson and Mouritsen, 1988), i.e., $a_b + a_c + a_i = 1$. $a_b(T)$ displays a minimum at the transition whereas both $a_c(T)$ and $a_i(T)$ have a pronounced peaked maximum. Building upon some early ideas of Papahadjopoulos et al. (1973), it was suggested in a minimal model by Cruzeiro-Hansson and Mouritsen (1988) that the thermal peak in $a_i(T)$ contains the main contribution to the dramatic thermal anomaly observed in the passive trans-membrane permeability of pure lipid bilayers (Papahadjopoulos et al., 1973; for a recent list of references to experimental and theoretical work on passive permeability, see e.g., Cruzeiro-Hansson and Mouritsen, 1988) for example for small cations such as Na^+. By assigning temperature-independent transmission coefficients, p_b, p_c, and p_i, to the three different membrane regions, the probability of an ion crossing the membrane may be written as

$$P(T) = a_b(T)p_b + a_c(T)p_c + a_i(T)p_i. \tag{5}$$

In the minimal model it was assumed that the interface gives the major transmission rate, i.e., $p_i \gg p_b, p_c$. The prediction for the relative passive ion permeability, $R(T)$, then becomes (Cruzeiro-Hansson and Mouritsen, 1988)

$$R(T) = A(T)^{-1/2}T^{1/2}P(T). \tag{6}$$

If we use the transmission coefficient values found in the earlier work on DPPC bilayer permeability (Cruzeiro-Hansson and Mouritsen, 1988) and hence assume that the chain-length dependence is to be found solely in the fractional areas, we obtain the relative passive ion permeabilities in the three different lipid bilayers as shown in Fig. 8 in the case of Na^+ permeation. It is straightforward to derive similar predictions in the case of other ionic species. The data clearly demonstrates that there is a significant enhancement of the permeability when the acyl-chain length is decreased.

RELATION TO EXPERIMENTS

The most convincing test of a theory of fluctuation phenomena in lipid membranes is to compare the calculated thermal response functions with corresponding experimental data. However, this procedure turns out to be difficult for a number of reasons. The pseudo-critical behavior of the main transition implies that an ideal response function is characterized by a delta function at $T = T_m$ and an algebraic relation $(T - T^*)^{-\delta}$ for

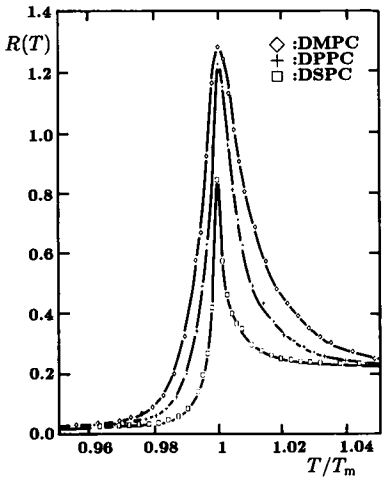

FIGURE 8 Relative passive ion permeability, $R(T)$ in Eq. 6, in the case of Na^+ permeation across DMPC, DPPC, and DSPC bilayers. The data assumes a lower cut-off of the cluster-size distribution function corresponding to seven lipid molecules (cf. Cruzeiro-Hansson and Mouritsen, 1988).

temperatures in the vicinity of T_m. T^* represents a critical temperature close to T_m (Mitaku et al., 1983) and δ is some nonnegative exponent. For temperatures away from T_m, the contribution from intramolecular excitations dominates the response functions. In an experimental measurement the finite instrumental resolution will tend to smear the delta-function singularity. This will in particular be true of the excess specific heat function which, when measured in a differential-scanning calorimetric experiment close to T_m, will not fulfill the condition of thermal equilibrium. The response functions will therefore appear broader for lipid systems with increasing acyl-chain length, because the jump-discontinuities of the thermal densities are increasing, e.g., the area change and the transition enthalpy. This effect appears to be in conflict with the predictions of the present model in the case of C_P and χ shown in Figs. 5 and 6. However, a critical comparison with experiments is particularly difficult in the case of the specific heat where the predicted effect on C_P away from T_m for varying acyl-chain length is relatively small. Another difficulty is related to the fact that, whereas the ten-state Pink model gives a good description of the main transition itself, it only gives an approximate description of the gel phase and the fluid phase in particular (Caillé et al., 1980). The possibility exists of comparing our equilibrium specific heat data with experimental specific heats obtained by alternating-current calorimetry (Hatta et al., 1983). This technique is only sensitive to the fluctuation part of C_P and neglects the delta-function contribution at T_m. However, the available data (Hatta et al., 1983) does not permit a direct comparison between results obtained for different lipid species.

From the model calculations presented in this paper we find that the effect on the thermal fluctuations due to varying the acyl-chain length is most strongly reflected in the isothermal compressibility. This effect should be observable in measurements of $\chi(T)$. However, detailed measurements of the temperature dependence of $\chi(T)$ do not exist in the literature for DPPC and DSPC, while the isothermal compressibility has been obtained for DMPC by pressure aspiration techniques applied to giant single-walled vesicles (Evans and Kwok, 1982). These data show a strong peak of $\chi(T)$ at the transition temperature.

Hence it appears that at present a comparison of the results of our systematic theoretical study with experimental data will have to be made with regard to phenomena which more indirectly are related to and controlled by the formation of lipid domains and interfaces in the transition region. The passive transmembrane permeability is such a phenomenon. The effect of acyl-chain length on the transmembrane permeability illustrated in Fig. 8 in the case of Na^+ permeation is in agreement with numerous experiments with ionic and nonionic agents (Singer and Finegold, 1985; Blok et al., 1975; Georgallas et al., 1987; de Gier et al., 1968). The permeability of different molecules can be numerically very different, but they all show the same general temperature-dependence with a peak at T_m. It should be noted that the increasing permeability for decreasing acyl-chain length has alternatively been explained by a decrease in the diffusion-barrier across the membrane (Georgallas et al., 1987). Another phenomenon related to membrane permeation and controlled by membrane fluctuations is cholesterol exchange between membranes (Bar et al., 1987). In a series of experiments on small unilamellar vesicles it was indeed found that the exchange rate in the transition region increased as the acyl-chain length was decreased (Bar et al., 1987), in accordance with the present model results. Yet another phenomenon closely related to membrane fluctuations is the partitioning of insecticides, such as lindane (Antunes-Madeira and Madeira, 1985), into membranes where it is found that the partition coefficient displays a maximum at the phase transition and that the partition coefficient increases with decreasing acyl-chain length.

In the absence of a more extensive set of experimental data obtained from systematic studies of lipid bilayers with different chain length we return to a comparison of our findings of universal classical pseudo-critical behavior with appropriate experiments. Our model calculations predict that the global thermal quantities exhibit pseudo-critical singularities, $(T - T^*)^{-\delta}$, where the exponents are well described by classical Landau theory, independent of acyl-chain length. Such a universal relationship

has been observed in various measurements of thermal quantities in the fluid phase. For example, the long-lifetime component obtained from time-resolved fluorescence anisotropy experiments on lipid bilayers suggests an approximate algebraic dependence as $T - T_c$, i.e., $\delta = 1$ (Ruggiero and Hudson, 1989). A similar exponent value has been obtained from measurements of the ultrasonic velocity in ultrasonic relaxation measurements on DPPC bilayers (Mitaku et al., 1983), in measurement of density fluctuations of mixed vesicles by small-angle neutron scattering techniques (Knoll et al., 1983), and in nuclear magnetic resonance experiments on DMPC, DPPC, and DMPE bilayers (Hawton and Doane, 1987). Our model calculations suggest that these exponents are universal in the sense that they are independent of the lipid species.

CONCLUSIONS

In this work we have presented the results of a theoretical model study systematically carried out to characterize how the lipid bilayer thermal density fluctuations vary with temperature and acyl-chain length. The microscopic interaction model on which the results are based takes careful account of the acyl-chain conformational degrees of freedom and their interactions and it is a reliable model of the main chain-melting phase transition of saturated diacyl phosphatidylcholine bilayers. Our main result, for the homologous lipid series DMPC, DPPC, and DSPC, is that the density fluctuations are strongly enhanced in the transition region and the response functions, such as specific heat and lateral compressibility, display pseudo-critical behavior. As the chain length is decreased the fluctuations get stronger. These general findings are in accordance with available experimental data. However, we wish to point out that there is at present a lack of systematically obtained data for different lipid bilayers, in particular with respect to equilibrium specific heat and compressibility. We hope that our work will stimulate further experimental work to provide such data.

It has for a long time been an important working model among experimenters in membrane science that a heterogeneous structure of the membrane and the associated lipid-domain interfaces with misfits and packing defects may facilitate a number of biological functions of the membrane. In several theoretical investigations the presence of such domains has been related to the presence of a hidden critical point close to the main transition temperature. Recently it has been suggested on the basis of a Landau theory that the source of the pseudo-critical behavior of lamellar membranes is the coupling of the lipid-chain order parameter and the intermembrane forces (Goldstein and Leibler, 1989). However, pseudocritical behavior of the main transition persists in single membrane systems such as giant vesicles (Evans and Kwok, 1982). It thus seems likely that pseudo-criticality is related to in-membrane properties. The ten-state model of Pink, which is a realistic model of the lipid bilayer main transition, does display pronounced psuedo-critical behavior at the transition.

In our previous work (Cruzeiro-Hansson et al., 1989) it was shown, on the basis of the same microscopic interaction model, that small amounts of substitutional impurities, e.g., cholesterol, enhance the lipid-domain formation away from the transition point. We wish to conclude the present paper by putting all these results into context with a general principle which concerns the conditions for producing dynamically heterogeneous membrane states. It appears that such states may be enhanced by manipulating the lipid bilayer so it effectively approaches a critical point. Because lipid bilayer heterogeneity is important for membrane function, it is clearly very important to come to grips with which factors couple to the density fluctuations and the formation of lipid domains and interfaces. The present work demonstrates theoretically that the acyl-chain length is such a factor.

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