

Binary Mixtures of Asymmetric Phosphatidylcholines with One Acyl Chain Twice as Long as the Other[†]

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ABSTRACT: High-resolution differential scanning calorimetry and ³¹P NMR spectroscopy have been used to study aqueous phosphatidylcholine (PC) dispersions prepared from colyophilized mixtures of C(10):C-(22)PC/C(22):C(12)PC of various molar ratios. These two lipid species are highly asymmetric but have a common structural feature; namely, one acyl chain in the fully extended conformation is about twice as long as the other. Our experimental results support two conclusions: (1) These two component lipids are miscible in all proportions in both gel and liquid-crystalline states. This type of system behaves as a nearly ideal mixture. Its calorimetric parameters are those expected on the basis of the mole fraction weighted average of the corresponding parameters for the pure components. (2) The component lipids appear to self-assemble, at $T < T_m$, into a mixed interdigitated bilayer in excess water. In a mixed interdigitated bilayer, the short acyl chain of one asymmetric phosphatidylcholine on one side of the bilayer leaflet is apposed with the short acyl chain of another lipid molecule on the other side of the bilayer leaflet, while the longer acyl chain from each of the two leaflets crosses the entire hydrocarbon width of the bilayer. The fundamental packing unit, as well as the dynamic unit describing the axial rotator motion about the bilayer normal for this mixed interdigitated bilayer, is thus a dimer, whereas the packing unit assigned for the noninterdigitated bilayer such as C(16):C(16)PC lamellae is a monomer.

The thermotropic behavior of binary mixtures of pure symmetric phospholipids in excess water has been extensively studied by differential scanning calorimetry (DSC)¹ [for reviews, see Mabrey and Sturtevant (1978), McElhaney (1982), and Bach (1984)]. Much valuable information on lipid-lipid interactions in the bilayer has been obtained from these DSC studies. For instance, if the component lipids have the same head group but differ in chain lengths by only two methylene units, the two components are found to be miscible in all proportions in both gel and liquid-crystalline states. A binary mixture of C(14):C(14)PC and C(16):C(16)PC serves as an example of a nearly ideal mixture (Mabrey & Sturtevant, 1976; van Dijk et al., 1977). By contrast, mixtures of two component lipids differing by six methylene units in both of their acyl chains usually show two major endotherms. These appear at approximately the transition temperatures detected for the bilayers that consist of only one component lipid. A specific example of this type of binary mixture with monotectic behavior is exhibited by multilamellar dispersions formed from C(12):C(12)PC and C(18):C(18)PC (Mabrey & Sturtevant, 1976).

In recent years, calorimetric studies of a wide range of asymmetric mixed-chain phosphatidylcholines in excess water have been reported (Boggs & Mason, 1986; Chen & Sturtevant, 1981; Keough & Davis, 1979; Mason et al., 1981; Serrallach et al., 1984; Stümpel et al., 1981; Xu & Huang, 1987). In general, it has been shown that asymmetric phosphatidylcholines, in which the *sn*-1 acyl chain is longer than the *sn*-2 acyl chain by more than four methylene units, can form various interdigitated bilayers at $T < T_m$ while symmetric phosphatidylcholines form noninterdigitated bilayers [for reviews, see Huang and Mason (1986) and Keough (1986)]. However, the phase behavior and thermotropic properties of

any binary mixture composed of highly asymmetric mixed-chain phosphatidylcholines in the bilayer are yet to be established.

In this paper, we present experimental data from high-resolution differential scanning calorimetric and ³¹P NMR studies of aqueous lipid dispersions prepared from mixtures of C(10):C(22)PC and C(22):C(12)PC in excess water. We have chosen these two lipid species for our binary mixture because they have the following properties: (1) They are highly asymmetric phosphatidylcholines with a common structural feature; namely, one acyl chain is almost twice as long as the other. (2) They are distinctively different in their acyl chain lengths esterified at the *sn*-1 and *sn*-2 positions of the glycerol backbone, respectively. In fact, the acyl chain length difference between these two species at either the *sn*-1 or the *sn*-2 position is 10 or more methylene units. (3) If one compares either the long or the short acyl chains of these two species irrespective of their *sn* positions, the difference in acyl chain length is, however, only about two methylene units. (4) The transition temperatures of these two fully hydrated lipid species are sufficiently different that the formation of an ideal or of a nonideal mixture in the same bilayer in excess water can be detected calorimetrically.

MATERIALS AND METHODS

C(10):C(22)PC and C(22):C(12)PC were synthesized by reacylation of an appropriate 1-acyllysophosphatidylcholine obtained from Avanti Polar Lipids, Inc. (Birmingham, AL), with an anhydride of the fatty acid (Sigma) desired to be in the 2-acyl position of the mixed-chain product. The reacylation was catalyzed by 4-pyrrolidinopyridine, and the product was then purified by silicic acid column chromatography employing

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¹ Abbreviations: C(X):C(Y)PC, saturated L- α -phosphatidylcholine having X carbons in the *sn*-1 acyl chain and Y carbons in the *sn*-2 acyl chain; DSC, differential scanning calorimetry; $\Delta\sigma$, chemical shift anisotropy; EDTA, ethylenediaminetetraacetic acid; Pipes, piperazine-*N,N'*-bis(2-ethanesulfonic acid).

a gradient of chloroform and methanol. Further details of all these procedures as well as the method for determining phospholipid concentration may be found elsewhere (Mason et al., 1981). Each synthesized mixed-chain phosphatidylcholine with purity ≥ 98 mol % exhibited only a single spot on thin-layer chromatography when tested with $\text{CHCl}_3/\text{CH}_3\text{OH}/11.6\% \text{ NH}_4\text{OH}$ (65:35:5). The purified mixed-chain lipids were stored at -20°C in chloroform.

Sample Preparation. In general, aliquots of known amounts of C(10):C(22)PC and C(22):C(12)PC were dried separately from solvent chloroform and then redissolved in anhydrous benzene. The lipid/benzene solutions were then mixed, and the mixture of C(10):C(22)PC and C(22):C(12)PC of a known molar ratio was lyophilized. The colyophilized lipid mixture was suspended in 50 mM NaCl aqueous solution containing 5 mM phosphate buffer and 1 mM EDTA at pH 7.4 to give a total lipid concentration in the range of 2–4 mM. The lipid dispersion was heated to 55°C , which is about 12°C above the gel to liquid-crystalline phase transition temperature of the higher melting component of the lipid sample [C(22):C(12)PC]. The dispersion was immediately vortexed at the elevated temperature for 5 min and then cooled slowly from 55 to 10°C at a rate of $0.3^\circ\text{C}/\text{min}$. This heating/cooling cycle was repeated 2 more times, and the colyophilized lipid sample was then stored at 0°C for 2 days. The resulting colyophilized lipid sample was degassed under reduced pressure and loaded into the sample cell of the calorimeter which had been thermally equilibrated at 2°C .

A mixture of pure C(10):C(22)PC and C(22):C(12)PC dispersions at a fixed 1:1 molar ratio was also prepared. First, pure C(10):C(22)PC and C(22):C(12)PC were suspended separately in NaCl (50 mM) aqueous solution containing 5 mM phosphate buffer and 1 mM EDTA (pH 7.4) to give each lipid suspension a concentration of 3 mM. Next, each lipid sample was subjected to the heating/cooling cycle between 50 and 10°C 3 times, followed by a prolonged incubation at 0°C for more than 3 days. Then, equal volumes of the resulting C(10):C(22)PC and C(22):C(12)PC dispersions were degassed and mixed at 0°C . Finally, the mixture was loaded into the sample cell of the calorimeter, and the mixed sample was equilibrated thermally in the calorimeter cell for 60 min at 2°C prior to the DSC scan.

Calorimetry. DSC experiments were carried out by using a high-resolution MC-2 differential scanning microcalorimeter (Microcal Co., Amherst, MA) equipped with the DA-2 digital data acquisition system as described previously (Xu & Huang, 1987). Unless indicated otherwise, all samples were scanned between 2 and 50°C at a constant speed of $10^\circ\text{C}/\text{h}$ in an ascending temperature mode. In all experiments, the samples were rescanned at least once. Transition temperatures (T_m) were taken from the transition peaks at the maximum peak height positions, and calorimetric enthalpies (ΔH) were calculated from the peak areas by using software provided by Microcal Co. The onset (T_o) and completion (T_c) temperatures of the thermal transition were extrapolated from the intercepts of maximal slopes of the onset and completion boundary of the transition curve with the base line, respectively.

Phosphorus-31 NMR. All ^{31}P NMR measurements were performed at 24.15 MHz under conditions of continuous broad-band proton noise decoupling at a power of 5 W using a JEOL-FX60Q spectrometer equipped with quadrature detection as described previously (Huang et al., 1986). Prior to the NMR measurements, the phospholipid sample (100 mM lipid in 50 mM KCl aqueous solution containing 2 mM EDTA and 10 mM Pipes at pH 7.0) in a 10-mm NMR tube was first

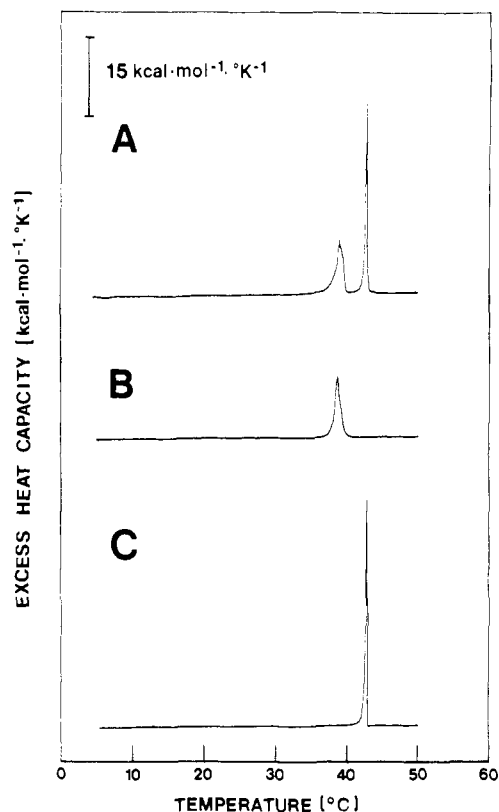


FIGURE 1: DSC heating thermograms of dispersions prepared from pure C(10):C(22)PC and/or pure C(22):C(12)PC. (A) Scan of a 1:1 mixture of C(10):C(22)PC and C(22):C(12)PC dispersions. (B) Scan of a C(10):C(22)PC sample. (C) Scan of a C(22):C(12)PC sample.

heated to 10 – 15°C above the phase transition temperature and was vortexed immediately at the elevated temperature for about 2–4 min. The sample was then cooled to 0°C . After the heating/cooling cycle had been repeated 2 more times, the sample was incubated at 0°C overnight. The sample tube was then put into the spectrometer's sample cavity, which had been preequilibrated at the desired temperature. A thermal equilibration time of at least 1 h was allowed for the lipid sample before data acquisition. Succeeding experiments at ascending temperatures were performed by leaving the sample in the spectrometer for a minimum of 30-min equilibration at each temperature.

RESULTS

Mixture of Pure C(10):C(22)PC and C(22):C(12)PC Dispersions at a 1:1 Molar Ratio. Figure 1A shows the first heating thermogram for the mixture of pure C(10):C(22)PC and C(22):C(12)PC dispersions. The DSC curve shows two clearly discernible phase transitions. The lower temperature transition is characterized by a phase transition temperature (T_m) of 39.0°C , an enthalpy change (ΔH) of 12.2 kcal/mol, and a half-height width ($\Delta T_{1/2}$) of 0.90°C . This transition behavior is in accord with that exhibited by dispersions of pure C(10):C(22)PC ($T_m = 38.7^\circ\text{C}$, $\Delta T_{1/2} = 0.82^\circ\text{C}$, and $\Delta H = 12.5$ kcal/mol) as shown in Figure 1B. The higher temperature transition is centered at 42.7°C with a half-height width of 0.34°C and a ΔH of 13.0 kcal/mol; this characteristic transition corresponds to that recorded for pure C(22):C(12)PC dispersions ($T_m = 42.8^\circ\text{C}$, $\Delta T_{1/2} = 0.25^\circ\text{C}$, and $\Delta H = 13.1$ kcal/mol) as shown in Figure 1C. It is worthwhile to mention that although the half-height width for C(10):C(22)PC dispersions is more than triple the width of the endothermic transition for C(22):C(12)PC dispersions, the phase

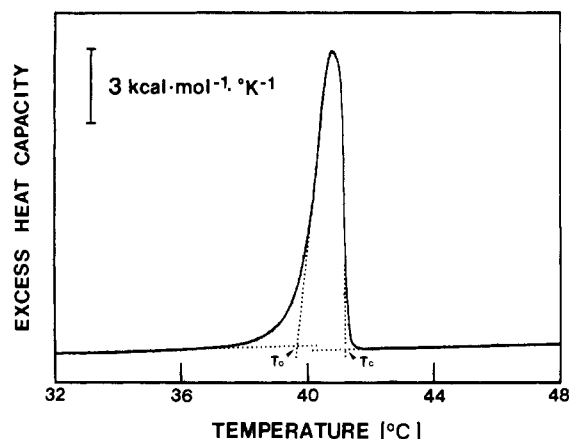


FIGURE 2: DSC heating thermogram of a sample prepared from an equimolar mixture of colyophilized C(10):C(22)PC and C(22):C(12)PC. Extrapolated dotted lines are drawn to show how the onset (T_0) and the completion (T_c) of the phase transition are obtained.

transition is still highly cooperative with a half-height width of less than 1 °C for C(10):C(22)PC dispersions. The broader transition may reflect that the C(10):C(22)PC preparation contains some trace amounts of positional isomer.

After being cooled from 55 to 2 °C in the calorimeter, the same sample was immediately rescanned. The second heating run exhibits two endotherms which are indistinguishable from those observed in the first heating thermogram. These results show that the chain melting of each pure lipid bilayer in the mixture is independent of the other and that the phase transition characteristics do not depend on the thermal history of the mixed dispersion.

Equimolar Mixture of Colyophilized C(10):C(22)PC and C(22):C(12)PC. Figure 2 shows the heating thermogram of dispersions prepared from an equimolar mixture of colyophilized C(10):C(22)PC and C(22):C(12)PC. In contrast to Figure 1A, only a single endothermic peak is detected for the colyophilized binary lipid dispersions. The phase transition is characterized by a T_m of 40.74 °C with a $\Delta T_{1/2}$ of 0.97 °C and a ΔH of 13.1 kcal/mol. The value of T_m is in between those of the pure components. The onset (T_0) and the completion (T_c) temperatures of the thermal transition as indicated in Figure 2 are 39.64 and 41.27 °C, respectively.

To demonstrate that the lipid dispersions prepared from the colyophilized binary lipid mixture are at equilibrium throughout the calorimetric run, the effect of scanning rate on the thermodynamic parameters was examined. Figure 3 shows the plot of T_m , T_0 , and T_c vs. the scanning rate. These experimental data clearly establish that the thermodynamic parameters do not depend appreciably on scan rate and that the values obtained at 10 °C/h, a scan rate being used throughout the present investigation, are virtually identical with the extrapolated values at zero scanning rate. Consequently, the onset and completion temperatures of the experimental curves obtained from high-resolution differential scanning calorimetric experiments performed at a scan rate of 10 °C/h can be used for the construction of a phase diagram for the binary mixtures of C(10):C(22)PC/C(22):C(12)PC in excess of water.

Phase Diagram. DSC thermograms were obtained for lipid dispersions prepared from colyophilized lipid mixtures of C(10):C(22)PC and C(22):C(12)PC with various molar ratios. Figure 4 shows typical DSC traces for samples of C(10):C(22)PC containing 0, 10, 20, 40, 50, 60, 80, 90, and 100 mol % of C(22):C(12)PC. These traces are characterized by single

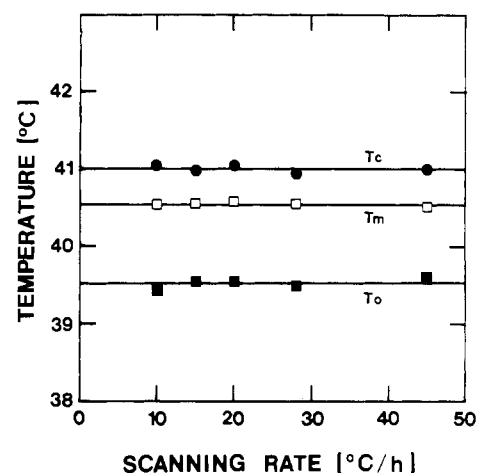


FIGURE 3: Plot of T_0 (closed squares), T_m (open squares), and T_c (closed circles) vs. the scanning rate. The data are obtained from DSC heating curves of an equimolar mixture of colyophilized C(10):C(22)PC and C(22):C(12)PC in excess water.

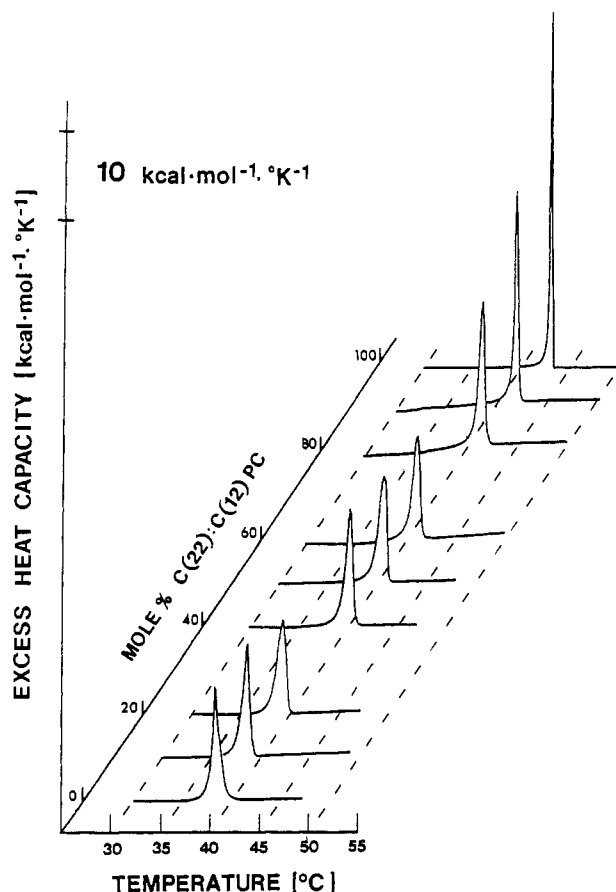


FIGURE 4: DSC heating thermograms for samples of C(10):C(22)PC containing various concentrations of C(22):C(12)PC. The curves are plotted relative to each other according to their mole percent of C(22):C(12)PC as indicated on the slanted axis.

endothermic transitions with nearly symmetric shapes. The presence of an increasing amount of C(22):C(12)PC results in a progressively linear increase in both T_m and ΔH as depicted in Figure 5. A temperature–composition phase diagram for C(10):C(22)PC/C(22):C(12)PC can be constructed based on the values of T_0 and T_c obtained from DSC traces after correction for the finite width of the phase transitions of the pure components (Mabrey & Sturtevant, 1976).

As shown in Figure 6, a phase diagram with solidus and liquidus curves of different slope is obtained for binary mix-

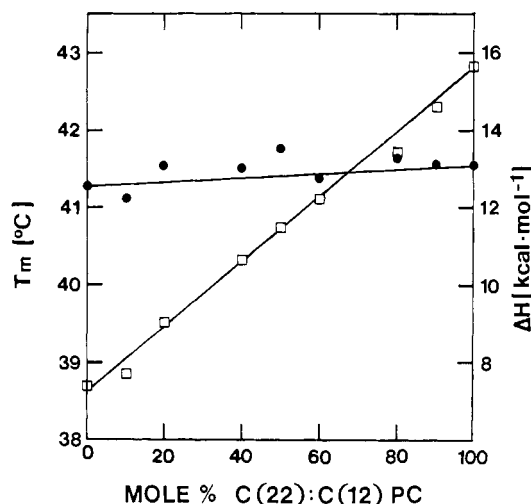


FIGURE 5: Plot of T_m (open squares) and ΔH (closed circles) of the phase transition vs. mole percent of C(22):C(12)PC in the binary mixture of C(10):C(22)PC and C(22):C(12)PC. The solid line is calculated on the basis of the values of T_m (or ΔH) for the pure components and the mole fraction (X) as follows: $T_m(\text{mixture}) = T_m^1 X^1 + T_m^2 X^2$, where the superscripts 1 and 2 stand for the pure component C(10):C(22)PC and C(22):C(12)PC, respectively.

tures of C(10):C(22)PC and C(22):C(12)PC in the presence of excess water. The cigar-shaped phase diagram for C(10):C(22)PC/C(22):C(12)PC mixtures is very similar to that observed for C(14):C(14)PC/C(16):C(16)PC mixtures (Mabrey & Sturtevant, 1976; van Dijck et al., 1977; Schmidt & Knoll, 1985), demonstrating complete miscibility of the two component lipids with almost ideal mixing in both gel and liquid-crystalline states over the entire composition range.

Phosphorus-31 NMR Studies. Representative ^{31}P NMR spectra of C(10):C(22)PC, C(22):C(12)PC, and an equimolar mixture of colyophilized C(10):C(22)PC and C(22):C(12)PC dispersions recorded at about 10 °C above and below their respective T_m values are shown in Figure 7. All ^{31}P NMR spectra exhibit an axially symmetric pattern with a "bilayer" line shape, which is characterized by a high-field peak with a broad low-field shoulder. This indicates that these asymmetric lipids and their equimolar mixture adopt a lamellar organization in excess water (Tilcock et al., 1986) and that the lipid molecules in the bilayer undergo axially rotational motions with an averaging of the components of the chemical shift tensor (Cullis & de Kruijff, 1978; Seelig, 1978; Griffin et al., 1978). The negative chemical shift anisotropy ($\Delta\sigma$) determined from the separation between points of maximum slope of the low-field shoulder and of the high-field peak is plotted in Figure 8 as a function of the reduced temperature (T_r). The reduced temperature is defined as $T_r = (T - T_m)/(273.15 + T_m)$, where T , in Celsius, is the experimental temperature and T_m is the main phase transition temperature, also in Celsius, determined by DSC. The value of $\Delta\sigma$ is observed to decrease linearly with increasing T_r up to $T_r \approx 0$. Within a very narrow temperature range (<1 °C) centered at $T_r = 0$, $\Delta\sigma$ decreases abruptly to a lower value; thereafter, $\Delta\sigma$ remains relatively constant at higher T_r . Interestingly, all ^{31}P NMR data obtained from dispersions prepared from C(10):C(22)PC, C(22):C(12)PC, and an equimolar mixture of C(10):C(22)PC/C(22):C(12)PC fall on a single transition curve as seen in Figure 8.

DISCUSSION

Mabrey and Sturtevant (1976) were the first to apply high-resolution DSC to investigate the mixing and the phase

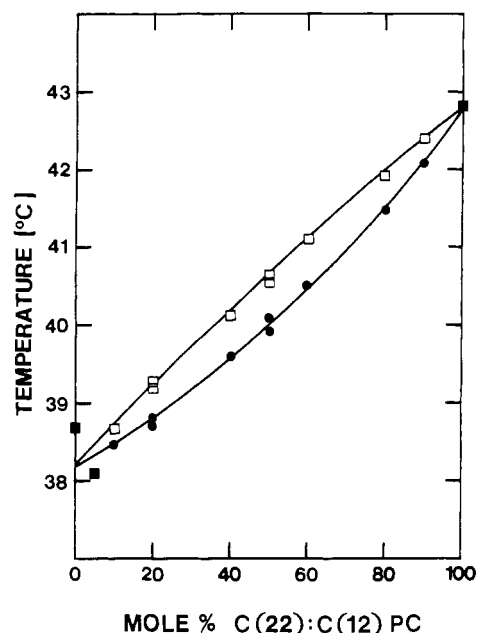


FIGURE 6: Phase diagram for the C(10):C(22)PC/C(22):C(12)PC system. The phase diagram is constructed on the basis of the onset and completion temperatures of the phase transition curves for C(10):C(22)PC/C(22):C(12)PC mixtures including those shown in Figure 5, after correction for the finite width of the transition curves of the pure components (Mabrey & Sturtevant, 1976). The line through the onset temperatures (closed circles) is the solidus line, while the one through the completion temperatures (open squares) is the liquidus curve.

separation of various binary mixtures of saturated phosphatidylcholines containing two identical fatty acyl groups. Curatolo et al. (1985) further extended the investigation of the mixing and phase separation of binary phosphatidylcholines in bilayers to include mixtures of a mixed-chain phosphatidylcholine (1-palmitoyl-2-oleylphosphatidylcholine) and various symmetric lipids. On the basis of 21 pairs of different binary mixtures, Curatolo et al. drew a general conclusion that phosphatidylcholines behave similarly in binary mixtures regardless of whether they are saturated, unsaturated, or mixed-chain varieties. Furthermore, binary mixtures of phosphatidylcholines exhibit gel-state miscibility, if the difference between the T_m values of the two component lipids (ΔT_m) is less than 33 °C, while those for which $\Delta T_m > 33$ °C exhibit gel-state immiscibility (Curatolo et al., 1985).

The present study represents the first high-resolution DSC investigation of binary mixtures comprised of two types of highly asymmetric phosphatidylcholines. Our DSC results thus further extend the data compiled by Curatolo et al. (1985). For the binary mixture of C(10):C(22)PC and C(22):C(12)PC, the values of T_m and ΔH are observed, within experimental error, to increase linearly with increasing C(22):C(12)PC content (Figure 5). In fact, the thermodynamic parameters (T_m and ΔH) of the binary mixture can be linearly related to the corresponding parameters of the pure components and the mole fraction (X) as follows: $T_m(\text{mixture}) = T_m^1 X^1 + T_m^2 X^2$, where the superscript denotes the pure component 1 or 2. Our DSC results suggest that C(10):C(22)PC and C(22):C(12)PC mix nearly ideally over the entire composition range in both gel and liquid-crystalline states. The cigar-shaped phase diagram shown in Figure 6 is an excellent documentation of our conclusion that binary C(10):C(22)PC/C(22):C(12)PC mixtures exhibit complete miscibility both above and below the phase transition temperature. The gel-phase miscibility of C(10):C(22)PC/C(22):C(12)PC mixtures can be attributed to the ability of the component lipids to form

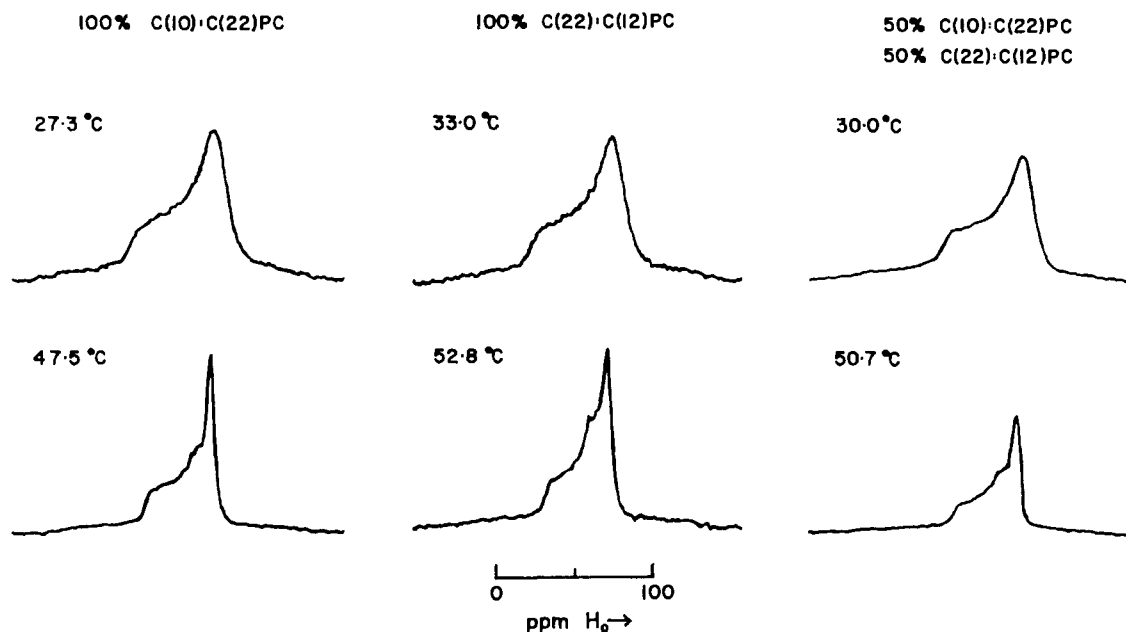


FIGURE 7: ^{31}P NMR spectra recorded for dispersions of C(10):C(22)PC, C(22):C(12)PC, and an equimolar mixture of colyophilized C(10):C(22)PC/C(22):C(12)PC at the indicated temperatures.

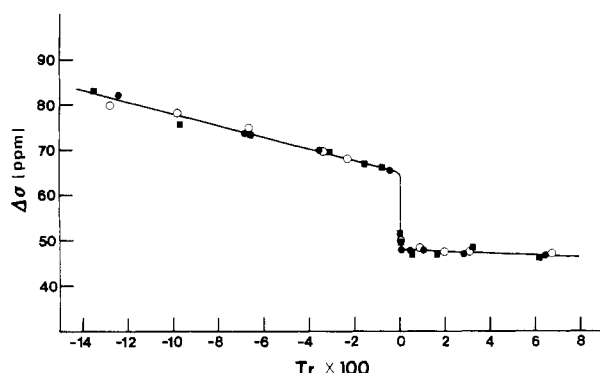


FIGURE 8: Reduced temperature dependence of ^{31}P chemical shift anisotropy (in ppm) for dispersions of C(10):C(22)PC (closed circles), C(22):C(12)PC (closed squares), and an equimolar mixture of colyophilized C(10):C(22)PC/C(22):C(12)PC (open circles). The reduced temperature (T_r) is defined as $T_r = (T - T_m)/(273.15 + T_m)$ as discussed in the text.

mixed interdigitated bilayers at $T < T_m$ (vide infra). In a mixed interdigitated bilayer, the acyl chains of C(10):C(22)PC and C(22):C(12)PC adopt a nearly fully extended conformation. This conformation permits van der Waals contacts between lipid species and allows these highly asymmetric lipid molecules to pack in such a manner that the sum of two short acyl chains packs laterally with the longer acyl chain.

An interesting finding resulting from this study is the demonstration that the measured chemical shift anisotropies for dispersions prepared from C(10):C(22)PC, C(22):C(12)PC, and an equimolar mixture of C(10):C(22)PC/C(22):C(12)PC are, within experimental error, identical at the same reduced temperature both above and below the phase transition temperature (Figure 8). A change in $\Delta\sigma$ can be attributed to conformational and/or motional differences of the lipid molecules in bilayers with respect to the phosphate moiety of the head group (Büldt & Wohlgemuth, 1981; Smith & Ekiel, 1984). Since the head-group conformation of phosphatidylcholine molecules is invariant in different bilayer states in the absence of di- or polyvalent metal ions (Seelig & Seelig, 1980), the observed constant value of $\Delta\sigma$ at the same reduced temperature shown in Figure 8 can thus be ascribed

primarily to the identical motional properties of these different asymmetric phosphatidylcholine molecules in both gel and liquid-crystalline states. On the basis of the observed values of $\Delta\sigma$, it is possible to compare the axially rotational motions of the asymmetric phosphatidylcholines under study with those of the well-known symmetric ones such as C(16):C(16)PC in the gel-state bilayer. Prior to our discussion of the comparison, it is pertinent to point out that there is experimental evidence to suggest that pure C(10):C(22)PC or C(22):C(12)PC molecules form mixed interdigitated bilayers at $T < T_m$ (Xu & Huang, 1987). In this packing model, two lipid molecules from opposing bilayer leaflets are coupled noncovalently to constitute the fundamental packing unit in the mixed interdigitated bilayer (Xu & Huang, 1987). The motional properties of the coupled dimeric unit in the interdigitated bilayer can thus be expected to be different from those of the monomeric packing unit in the noninterdigitated bilayer such as C(16):C(16)PC bilayers.

Axially symmetric ^{31}P NMR spectra with $\Delta\sigma = 52$ –69 ppm have been observed for C(16):C(16)PC molecules in the gel-state bilayer at various temperatures (Büldt & Wohlgemuth, 1981; Herzfeld et al., 1978); the limiting value of $\Delta\sigma = 69$ ppm is recorded for C(16):C(16)PC bilayers at $T_r = -0.16$. In contrast, the values of $\Delta\sigma$ observed for C(10):C(22)PC, C(22):C(12)PC, and an equimolar mixture of C(10):C(22)PC/C(22):C(12)PC in the gel state are 65–83 ppm, and the limiting value of 83 ppm is seen at $T_r = -0.14$ as shown in Figure 8. Clearly, the chemical shift anisotropy of C(16):C(16)PC dispersions is considerably smaller than that of dispersions prepared from C(10):C(22)PC, C(22):C(12)PC, and an equimolar mixture of C(10):C(22)PC/C(22):C(12)PC recorded at a common reduced temperature in the gel state. The difference in $\Delta\sigma$ can be attributed primarily to a reduced motional averaging due to a decrease in the rate of axial rotator motions of the asymmetric phosphatidylcholines in the gel-state bilayer. This reduced motional averaging is, in fact, expected, since the fundamental packing unit for mixed interdigitated lamellae is a dimer (vide supra) which must undergo the axially rotational motion at a slower rate in comparison with that of a monomeric unit such as C(16):C(16)PC in the gel-state bilayer. Our ^{31}P NMR results, although not defi-

nitive, nevertheless can be taken to support the concept that C(10):C(22)PC, C(22):C(12)PC, and the C(10):C(22)PC/C(22):C(12)PC mixture in excess water adopt an organization of mixed interdigitated lamellae, at $T < T_m$, with a fundamental dimeric packing unit. Moreover, the dimeric packing unit undergoes axial rotator motions about the bilayer normal as a dynamic unit. Finally, it should be emphasized that an equimolar mixture of C(10):C(22)PC/C(22):C(12)PC exhibits axially symmetric ^{31}P NMR spectra with values of $\Delta\sigma$ similar to those exhibited by its pure components at a common reduced temperature both above and below the T_m . This result supports the conclusion drawn from DSC data that C(10):C(22)PC and C(22):C(12)PC must mix almost ideally in the bilayer in both gel and liquid-crystalline states.

ACKNOWLEDGMENTS

It is with gratitude and pleasure that we dedicate this paper to Professor Robert G. Langdon on the advent of his 65th birthday, the occasion of his retirement. We thank F. Wilcoxen for secretarial assistance.

Registry No. C(10):C(22)PC, 106268-93-3; C(22):C(12)PC, 106268-90-0.

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