N. A. (1984) *Biochemistry 23*, 4038-4044.
Mena, P. L., & Djerassi, C. (1985) *Chem. Phys. Lipids 37*, 257-270.
Shah, J., Scripada, P. K., & Shipley, G. G. (1990) *Biochemistry 29*, 4254-4262.
Slater, J. L., & Huang, C. (1988) *Prog. Lipid Res. 27*, 325-359.
Stümpel, J., Eibl, H., & Nicksch, A. (1983) *Biochim. Biophys. Acta 727*, 246-254.
Wong, P. T. T., & Huang, C. (1989) *Biochemistry 28*, 1259-1263.
Wu, W., Chong, P. L., & Huang, C. (1985) *Biophys. J. 47*, 237-242.
Xu, H., & Huang, C. (1987) *Biochemistry 26*, 1036-1043.
Zaccai, G., Büldt, G., Seelig, A., & Seelig, J. (1979) *J. Mol. Biol. 134*, 693-706.

Differential Scanning Calorimetric Study of a Homologous Series of Fully Hydrated Saturated Mixed-Chain C(X):C(X+6) Phosphatidylcholines†

Zhao-qing Wang, Hai-nan Lin, and Ching-hsien Huang*
Department of Biochemistry, Health Sciences Center, University of Virginia, Charlottesville, Virginia 22908
Received February 20, 1990; Revised Manuscript Received April 20, 1990

ABSTRACT: The successive high-resolution differential scanning calorimetric (DSC) thermograms for aqueous dispersions of a homologous series of mixed-chain phosphatidylcholines, C(X):C(X+6)PC, have been recorded and analyzed. In this series of saturated mixed-chain phosphatidylcholines, the total number of carbon atoms in the *sn*-1 acyl chain increases from 11 to 20, and the *sn*-2 acyl chain is always 6 methylene units longer than the *sn*-1 acyl chain. In the initial heating DSC thermograms, two prominent endothermic transitions are detected for all the samples prepared from the various C(X):C(X+6)PCs except C(12):C(18)PC. In contrast, a single exothermic transition is observed on cooling for all the samples except C(13):C(19)PC. The temperature difference between the two endothermic transitions increases linearly as the acyl chain length of C(X):C(X+6)PC becomes progressively longer. Interestingly, the main phase transition occurs before the subtransition for C(11):C(17)PC dispersions. Our DSC data further demonstrate that the thermodynamic parameters (T_m, ΔH, and ΔS) associated with the main phase transition for fully hydrated C(13):C(19)PC and other identical MW phosphatidylcholines are inversely related to the corresponding values of the chain-length inequivalence ($\Delta C/CL$) for these lipids. This linear relationship can be employed to map the T_m values for aqueous dispersions prepared from a large number of mixed-chain phosphatidylcholines whose values of $\Delta C/CL$ are within the range of 0.1-0.4.

The systematic differential scanning calorimetric (DSC)[1] study of a homologous series of identical-chain phosphatidylcholines, C(X):C(X)PC, in excess water yields important information concerning the relative effect of chain ends on the chain packing order (Mason & Huang, 1981), the relative metastability of various lipid phases (Finegold & Singer, 1986), and the change in transition pathways as a function of chain length (Lewis et al., 1987). For instance, it has been demonstrated that fully hydrated short-chain phosphatidylcholines ($X \leq 8$) do not undergo the endothermic phase transition upon heating and that longer chain phosphatidylcholines ($X \geq 22$) do not self-assemble in excess water into the rippled gel phase ($P_{\beta'}$) at $T < T_m$. These interesting results may be of great importance in understanding the roles of various fatty acid compositions in biological membranes.

In order to extend the previous studies on identical-chain phosphatidylcholines, we have carried out the DSC study of a homologous series of mixed-chain phosphatidylcholines, C(X):C(X+6)PC, in which the *sn*-2 acyl chain is chosen to be 6 methylene units longer than the *sn*-1 acyl chain, and the total number of carbon atoms in the *sn*-1 acyl chain, X, increases systematically from 11 to 20. Although the *sn*-2 acyl chain in the fully extended conformation is always six methylene units longer than the *sn*-1 acyl chain in this homologous series of mixed-chain phosphatidylcholines, the normalized chain-length difference or inequivalence ($\Delta C/CL$) between the *sn*-1 and *sn*-2 acyl chains for the mixed-chain phospholipid in the gel-state bilayer decreases progressively as the value of X in C(X):C(X+6)PC increases systematically. Here, ΔC is the effective chain-length difference between the *sn*-1 and *sn*-2 acyl chains in terms of C–C bonds for phospholipid molecules in the gel-state bilayer, and it is a constant value of 4.5 for C(X):C(X+6)PC. The term CL is the effective chain length of the longer of the two acyl chains in C–C bonds, and it equals $X + 3.5$ for C(X):C(X+6)PC. In calculating the value of ΔC and CL, an inherent shortening of 1.5 C–C bond lengths for the *sn*-2 acyl chain has been taken into account; this shortening is caused by a sharp bend on the *sn*-2 acyl chain at the bond between C(2) and C(3) for phospholipids in the gel-state

†This research was supported in part by U.S. Public Health Service Grant GM-17452 from the NIH, Department of Health and Human Services.

[1] Abbreviations: C(X):C(X)PC, saturated identical-chain L-α-phosphatidylcholine having X carbons in the *sn*-1 chain as well as the *sn*-2 acyl chain; C(X):C(X+6)PC, saturated mixed-chain L-α-phosphatidylcholine having X carbons in the *sn*-1 acyl chain and $X + 6$ carbons in the *sn*-2 acyl chain; DSC, differential scanning calorimetry; MW, molecular weight.

0006-2960/90/0429-7072$02.50/0

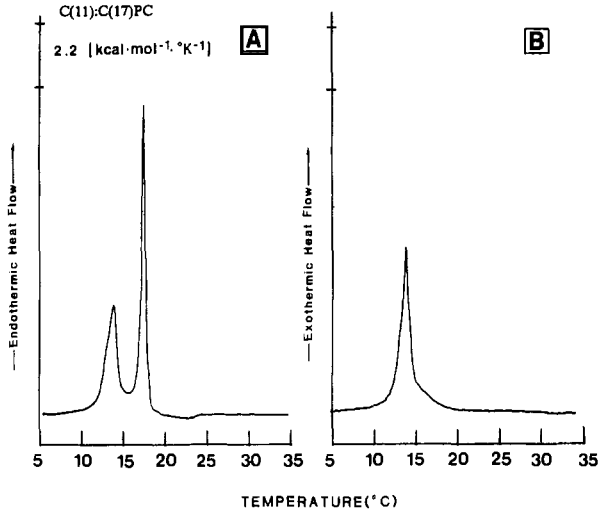

FIGURE 1: Successive DSC thermograms for a C(11):C(17)PC dispersion that has been preincubated at 0 °C for 10 days. Trace A is the initial heating thermogram, and trace B is the immediate cooling thermogram.

bilayer (Zaccai et al., 1979). The definition of $\Delta C/CL$ and its calculation have been reviewed recently (Slater & Huang, 1988). The values of $\Delta C/CL$ for C(11):C(17)PC, C(12):C(18)PC, C(13):C(19)PC, C(14):C(20)PC, C(15):C(21)PC, C(16):C(22)PC, C(17):C(23)PC, C(18):C(24)PC, and C(20):C(26)PC are 0.310, 0.290, 0.273, 0.257, 0.243, 0.231, 0.220, 0.209, and 0.191, respectively.

Experimental Procedures

Materials and Methods. Lysophosphatidylcholines were prepared enzymatically by hydrolyzing the identical-chain phosphatidylcholines according to the established procedure of Mason et al. (1981). Fatty acids were purchased from Sigma (St. Louis, MO). Isomerically pure (≥98 mol %) mixed-chain phosphatidylcholines, C(X):C(X+6)PC, were semisynthesized at room temperature by acylation of the $CdCl_2$ adduct of lysophosphatidylcholine with fatty acid anhydride in the presence of catalyst, 4-pyrrolidinopyridine (Mena & Djerassi, 1985). The mixed-chain phosphatidylcholines were purified by silica gel chromatography, and the purified lipid proved to be at least 98% pure by thin-layer chromatography. The lipid concentrations were determined from the lipid phosphorus by the method of Gomori (1942).

Sample Preparation. Aqueous dispersions of mixed-chain phosphatidylcholines used for DSC experiments were prepared from lyophilized lipids dispersed in 50 mM NaCl, 5 mM phosphate buffer, and 1 mM EDTA at pH 7.4. The final lipid concentrations of 4.0–6.0 mM are commonly used. The lipid dispersions were thoroughly mixed by subjecting to heating/cooling cyclies 3 times with constant vortexing as described elsewhere (Xu & Huang, 1987). The samples were then stored at 0 °C for a minimum of 4 days before use to ensure complete thermal equilibration at 0 °C.

DSC Measurements. All DSC runs were performed on a high-resolution MC-2 differential scanning calorimeter with cooling scan capability (Microcal Inc., Northampton, MA); this instrument was interfaced to an IBM-PC computer for data acquisition and analysis. Prior to DSC scans, samples were incubated in the calorimeter at the desired starting temperature for 90 min. Each sample was run at least 4 times: two heating (scan rate 10–15 °C/h) and two cooling (scan rate 5–15 °C/h). The transition temperatures and transition enthalpies were evaluated by the software package provided by Microcal Inc.

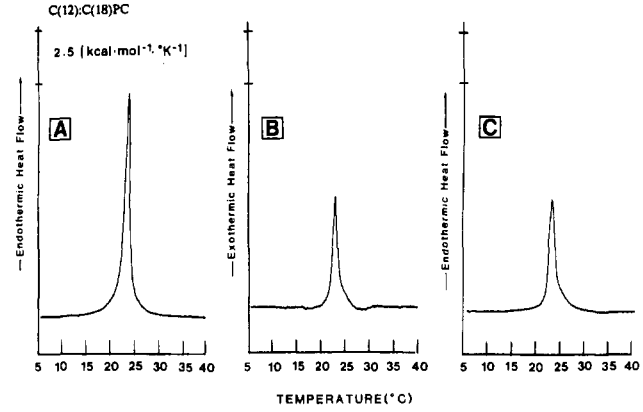

FIGURE 2: DSC thermograms for a C(12):C(18)PC dispersion that has been preincubated at 0 °C for 40 days. Traces A–C show the successive heating, cooling, and reheating curves of the same sample in the temperature interval of 5–40 °C.

Results

The successive DSC heating and cooling thermograms for an aqueous dispersion of C(11):C(17)PC which has been preincubated at 0 °C for 10 days are illustrated in Figure 1. The first heating scan is characterized by two partially overlapped endothermic transitions peaked at 13.9 and 17.3 °C with a total enthalpy of 9.3 kcal/mol. The low-temperature transition at 13.9 °C is also observed in its entirety in the cooling and reheating thermograms; hence, it corresponds to the main or gel/liquid-crystalline phase transition. The high-temperature transition at 17.3 °C ($\Delta H \simeq 5.0$ kcal/mol) disappears upon cooling or immediate reheating; hence, this irreversible transition appears to be a subtransition. On the basis of the large magnitude of ΔH, this subtransition is most probably the $L_c \rightarrow L_\alpha$ transition. Calorimetric detection of the subtransition at a temperature above the gel/liquid-crystalline phase transition temperature (T_m) is unusual, since the subtransition occurs most frequently at a temperature below the T_m (vide post). It should perhaps be noted here that identical-chain phosphatidylcholines with short acyl chains such as C(11):C(11)PC and C(12):C(12)PC exhibit a similarly unusual phase behavior (Finegold & Singer, 1986).

Figure 2 shows the successive heating, cooling, and reheating thermograms for an example of C(12):C(18)PC dispersions that has been preincubated at 0 °C for 40 days. A single sharp endotherm peaked at 23.8 °C is displayed in the first heating thermogram within the temperature interval of 5–40 °C; this phase transition exhibits a relatively large value of ΔH of 11.5 kcal/mol. In contrast, a single, reversible transition peaked at 23.3 ± 0.2 °C is observed on cooling or immediate reheating with a ΔH of 5.9 ± 0.5 kcal/mol. This reversible transition observed in the cooling and reheating thermograms can be assigned as the main or the gel/liquid-crystalline phase transition. The larger single transition observed in the initial heating thermogram appears to be a superposition of the subtransition and the main phase transition.

The initial heating and cooling thermograms for aqueous dispersions of C(13):C(19)PC, C(14):C(20)PC, C(15):C(21)PC, C(16):C(22)PC, C(17):C(23)PC, C(18):C(24)PC, and C(20):C(26)PC that have been preincubated at 0 °C for a minimum of 4 days are illustrated in Figure 3. This series of initial DSC heating thermograms are all characterized by two endothermic transitions. The transition temperatures of the two endotherms increase continually with increasing chain length; in addition, the relative peak height ratio of the high-temperature endotherm/low-temperature endotherm as well as the distance between the two endothermic peaks is

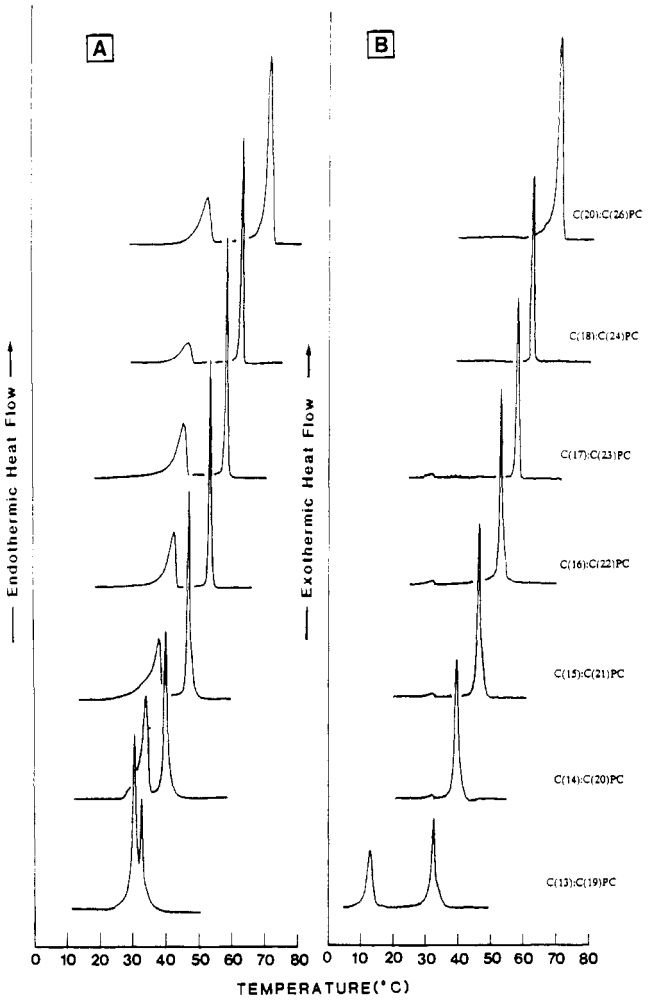

FIGURE 3: Initial heating and cooling thermograms for aqueous dispersions of C(13):C(19)PC, C(14):C(20)PC, C(15):C(21)PC, C(16):C(22)PC, C(17):C(23)PC, C(18):C(24)PC, and C(20):C(26)PC.

observed to increase steadily as the acyl chain length of this series of mixed-chain phospholipids increases progressively up to C(18):C(24)PC (Figure 3A). The DSC cooling thermograms recorded for aqueous dispersions of this family of phospholipids are illustrated in Figure 3B. Two exotherms are discernible for C(13):C(19)PC dispersions on cooling. The values of the transition temperature (32.5 °C) and the transition enthalpy (6.2 kcal/mol) of the high-temperature exotherm are virtually identical with those (32.6 °C and 6.7 kcal/mol) exhibited by the high-temperature endotherm observed in the initial heating thermogram; consequently, this high-temperature transition can be assigned as the reversible gel/liquid-crystalline phase transition. The low-temperature exotherm at 12.9 °C with ΔH = 5.9 kcal/mol appears to be a rather unique transition for C(13):C(19)PC, since all other mixed-chain phospholipids of this family do not exhibit such a prominent low-temperature exotherm upon cooling (Figure 3B). This low-temperature exotherm observed for C(13):C(19)PC is tentatively assigned as the transition corresponding to the formation of the subphase or crystalline phase. Despite the uncertainty of this assignment, the high-temperature endotherms observed for all the samples shown in Figure 3A can be assigned unambiguously as the gel/liquid-crystalline phase transitions, since these transitions are reversible as demonstrated by the cooling scans (Figure 3B). The low-temperature endotherms shown in Figure 3A, however, are all abolished at the corresponding temperatures upon cooling (Figure 3B);

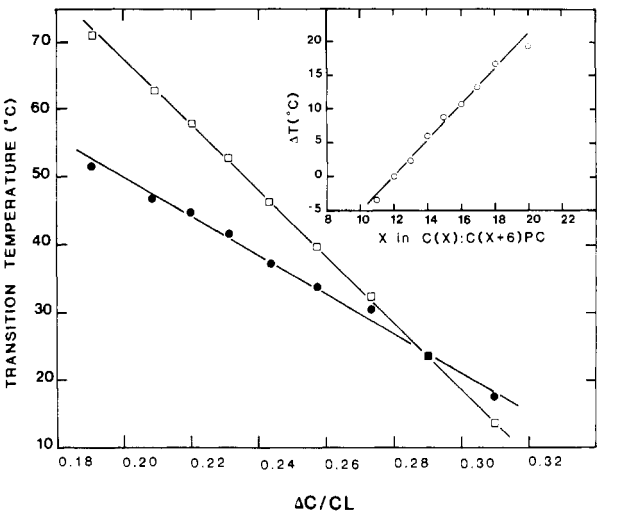

FIGURE 4: Plots of transition temperatures vs $\Delta C/CL$ for C(X):C(X+6)PC dispersions. The values of the main phase transition (T_m) are symbolized by open squares, and the values of the subtransition temperature (T_s) are represented by solid circles. The least-squares line for the T_s line is given by T_s (°C) = 106.6–284.2 ($\Delta C/CL$), and the least-squares line for the T_m line is given by T_m (°C) = 163.1–479.9 ($\Delta C/CL$). The insert represents the plot of ΔT or $T_m - T_s$ versus X or the total number of carbon atoms in the *sn*-1 acyl chain for C(X):C(X+6)PC.

consequently, they are assigned as the subtransitions. It should be noted that very small but discernible peaks near 30 °C are observed in the cooling scans for a number of mixed-chain phospholipids under study as shown in Figure 3B. These are the instrumentation artifacts.

Discussion

In Figure 4, the peak temperatures of the subtransition and the main phase transition detected in the first DSC heating scan are plotted against the value of $\Delta C/CL$ for all the mixed-chain phospholipids, C(X):C(X+6)PC, under study. This figure illustrates the inverse linear relationships between the phase transition temperatures (T_m and T_s) and the chain-length inequivalence ($\Delta C/CL$). It is also apparent that the line connecting the main phase transition temperatures intersects the subtransition line at the point of $\Delta C/CL$ = 0.29, corresponding to the single transition peak of C(12):C(18)PC. In addition, Figure 4 also shows that the temperature difference (ΔT) between the peaks of the main phase transition and the subtransition increases linearly as the acyl chain length of C(X):C(X+6)PC becomes progressively longer. This linear relationship provides additional evidence indicating that the subtransition of C(11):C(17)PC occurs at a temperature above the main phase transition temperature.

The enthalpy changes, ΔH, associated with the main phase transitions for aqueous dispersions of C(11):C(17)PC, C(12):C(18)PC, C(13):C(19)PC, C(14):C(20)PC, C(15):C(21)PC, C(16):C(22)PC, C(17):C(23)PC, C(18):C(24)PC, and C(20):C(26)PC are 3.1, 5.9, 6.7, 7.5, 11.0, 12.7, 14.0, 15.6, and 18.6 kcal/mol, respectively. These ΔH values were calculated from the areas of the main phase transition peaks detected in the initial DSC heating thermograms. In the case of C(12):C(18)PC, however, the value of ΔH was calculated from the transition curve obtained from the second DSC heating thermogram, since the transition peak for C(12):C(18)PC dispersions in the initial DSC heating thermogram appeared to be a superposition of two transitions. Clearly, the enthalpy change of the main phase transition is a linear function of X in C(X):C(X+6)PC. Interestingly, extrapolation of the linear function to ΔH = 0 gives a value of X = 9 for

Table I: Thermodynamic Parameters Associated with the Main Phase Transitions of Fully Hydrated Mixed-Chain Phosphatidylcholines with a Common Molecular Weight Identical with C(16):C(16)PC

phospholipid	$\Delta C/CL$	T_m (°C)	ΔH (kcal/mol)	ΔS (cal mol^{-1} K^{-1})
C(16):C(16)PC[a]	0.10	41.4	8.7_4	27.8
C(14):C(18)PC[b]	0.16	38.6	6.9	22.2
C(14):C(18)PC[c]	0.16	42.0	8.2	26.0
C(14):C(18)PC[d]	0.16	38.2	7.9	25.4
C(14):C(18)PC[e]	0.16	39.3	7.9	25.3
C(18):C(14)PC[b]	0.32	29.6	5.2	17.2
C(18):C(14)PC[d]	0.32	29.9	5.8	19.8
C(18):C(14)PC[f]	0.32	29.8	5.6	19.2
C(18):C(14)PC[c]	0.32	33.0	6.0	19.6
C(18):C(14)PC[g]	0.32	31.0	6.0	19.7
C(18):C(14)PC[e]	0.32	31.3_5	6.3	20.7

[a] Mabrey & Sturtevant (1976). [b] Chen & Sturtevant (1981). [c] Stümpel et al. (1983). [d] Mattai et al. (1987). [e] Unpublished work from this laboratory. [f] Huang & Mason (1986). [g] Boggs & Mason (1986).

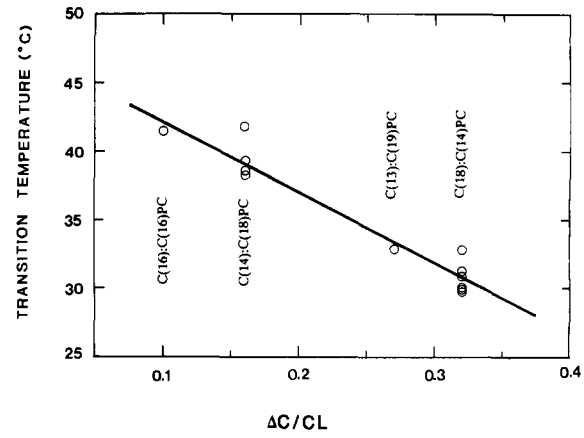

FIGURE 5: Transition temperatures associated with the main phase transitions for aqueous dispersions of C(16):C(16)PC, C(14):C(18)PC, C(13):C(19)PC, and C(18):C(14)PC are plotted against the values of $\Delta C/CL$ for these various lipids. The experimental data (open circles) are taken from Table I. The least-squares line has the expression $T_m = -52.56\ (\Delta C/CL) + 47.56$.

aqueous dispersions of C(X):C(X+6)PC. This suggests that fully hydrated C(X):C(X+6)PC with less than nine carbons in the *sn*-1 acyl chain will not exhibit a main phase transition. The same number of 9 has also been extrapolated for identical-chain phosphatidylcholines, C(X):C(X)PC, by Finegold and Singer (1986). It should be remarked that, in excess water, C(8):C(8)PC and C(9):C(9)PC form micelles and monodispersed unilamellar vesicles, respectively, whereas longer identical-chain phosphatidylcholines form multilamellar liposomes with a heterogeneous population of sizes (Racey et al., 1989). It remains to be shown whether the absence of the main phase transition ($\Delta H = 0$) for fully hydrated C(8):C(14)PC reflects the formation of highly disordered micellar structures by C(8):C(14)PC in excess water.

The molecular weight of C(13):C(19)PC is 734.1 which is identical with those of C(16):C(16)PC, C(14):C(18)PC, and C(18):C(14)PC, indicating that this group of phosphatidylcholines has the same total number of methylene units in their acyl chains. The main phase transition characteristics such as T_m, ΔH, and ΔS for C(16):C(16)PC, C(14):C(18)PC, and C(18):C(14)PC dispersions have been well studied by high-resolution DSC (Mabrey & Sturtevant, 1976; Chen & Sturtevant, 1981; Stümpel et al., 1983; Huang & Mason, 1986; Xu & Huang, 1987; Mattai et al., 1987). Their values (Table I) together with the T_m (32.6 °C), ΔH (6.7 kcal/mol), and ΔS (21.9 cal mol^{-1} K^{-1}) for C(13):C(19)PC dispersions determined in this work can be plotted against the values of $\Delta C/CL$ for these various phospholipids. A least-squares line with a correlation coefficient of 0.92 and a negative slope is obtained for the T_m vs $\Delta C/CL$ plot (Figure 5). Similar linear functions with negative slopes are evident for plots of the other two thermodynamic parameters (ΔH and ΔS).

The linear decrease in transition entropy (ΔS) with increasing value of $\Delta C/CL$ suggests strongly that the lateral chain–chain packing order of these various identical MW phospholipids in the bilayer becomes significantly more perturbed as the mismatch of the terminal methyl ends of the two acyl chains becomes progressively increased. On the basis of Raman spectroscopic data, it is known that acyl chains of identical MW phospholipids in the liquid-crystalline state usually exhibit similar disordering characteristics (Huang et al., 1982, 1983). The negative slope of the ΔS versus $\Delta C/CL$ plot can thus be attributed primarily to increased perturbations of the overall lipid acyl chain packing order in the *gel-state* bilayer by the progressively longer chain mismatches at the chain termini. Earlier comparative studies on the thermodynamic and spectroscopic properties of bilayers composed of identical-chain phosphatidylcholines arrived at the same conclusion that the disorder/order characteristics of gel-state bilayers are linearly correlated with the perturbations arising from inequivalent methyl regions at the acyl chain termini (Huang & Levin, 1983). It should be emphasized, however, that this type of linear relationship applies only to mixed-chain lipids whose values of $\Delta C/CL$ are within the range of 0.1–0.4 (Huang, 1990; Lin et al., 1990).

An immediate application of the linear plot of T_m (ΔH or ΔS) versus $\Delta C/CL$ as shown in Figure 5 lies in its predictions of the various values of thermodynamic parameters associated with the main phase transition for aqueous dispersions of other mixed-chain phosphatidylcholines with the same MW. For instance, C(17):C(15)PC has the same MW as C(16):C(16)PC or C(13):C(19)PC; the value of $\Delta C/CL$ for C(17):C(15)PC is 0.219 which is between 0.100 for C(16):C(16)PC and 0.273 for C(13):C(19)PC. The values of T_m, ΔH, and ΔS for C(17):C(15)PC dispersions can be calculated from the least-squares lines to be 36.1 °C, 7.1 kcal/mol, and 23.0 cal mol^{-1} K^{-1}, respectively.

Among the three thermodynamic parameters associated with the main phase transition for lipid dispersions, the value of T_m can be most accurately and reproducibly determined by high-resolution DSC. The values of T_m for identical-chain C(X):C(X)PC ranged from $X = 12$ to $X = 22$ are well documented in the literature (Lewis et al., 1987; Xu & Huang, 1987), and those for mixed-chain C(X):C(X+6)PC ranged from $X = 11$ to $X = 20$ are presented in this work. On the basis of these values and, in addition, on the basis of a reasonable assumption that the values of T_m for dispersions of phosphatidylcholines with the same MW are a linear function of their corresponding values of $\Delta C/CL$ within the range of $\Delta C/CL$ values from 0.1 to 0.4, a large number of unknown values of T_m for aqueous dispersions of mixed-chain phosphatidylcholines may be graphically and rapidly estimated. For instance, a straight line can be connected between the T_m of C(15):C(15)PC (34.7 °C) at $\Delta C/CL = 0.107$ and the T_m of C(12):C(18)PC (23.8 °C) at $\Delta C/CL = 0.29$ as shown in Figure 6. The values of T_m for dispersions of C(17):C(13)PC ($\Delta C/CL = 0.343$), C(16):C(14)PC ($\Delta C/CL = 0.233$), and C(13):C(17)PC ($\Delta C/CL = 0.172$) can then be graphically determined to be 20.6, 27.2, and 30.8 °C, respectively. Similarly, the T_m values for C(16):C(12)PC ($\Delta C/CL = 0.367$),

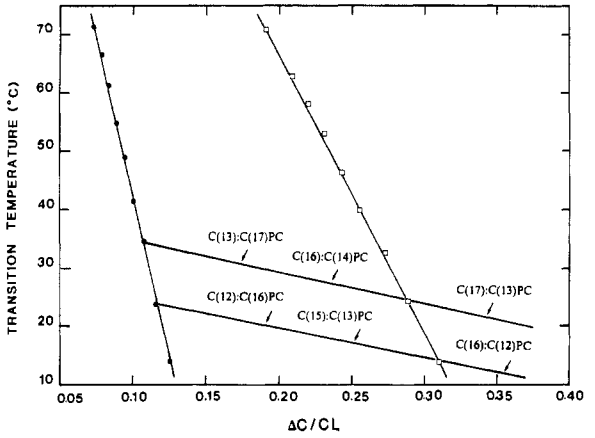

FIGURE 6: Mapping the T_m values for various mixed-chain phosphatidylcholines. The two nearly vertical lines are the least-squares lines connecting the known values of T_m for C(X):C(X)PC (solid circles) and C(X):C(X+6)PC (open squares), and the two nearly parallel lines intersecting the two vertical lines are used to estimate the value of T_m based on the calculated value of $\Delta C/CL$ for a given mixed-chain phosphatidylcholine. The arrows indicate the various points where the unknown values of T_m for some of the mixed-chain phosphatidylcholines have been estimated as discussed in the text.

C(15):C(13)PC ($\Delta C/CL$ = 0.25), and C(12):C(16)PC ($\Delta C/CL$ = 0.185) can also be graphically estimated to be 11.0, 17.0, and 20.3 °C, respectively, based on a different straight line which connects the T_m value of 23.9 °C for C(14):C(14)PC ($\Delta C/CL$ = 0.115) and the T_m of 13.9 °C for C(11):C(17)PC ($\Delta C/CL$ = 0.31) as shown in Figure 6. In fact, many more T_m values of mixed-chain phosphatidylcholines can be approximated by this graphical approach using the two lines of T_m values for C(X):C(X)PC and C(X):C(X+6)PC as illustrated in Figure 6. Of course, future DSC experiments will indicate the veracity of our estimations.

In summary, the thermotropic phase behavior for aqueous dispersions of a series of C(X):C(X+6)PCs has been determined in this study by high-resolution DSC. It is interesting to observe that the main phase transition occurs before the subtransition for C(11):C(17)PC dispersions. Our calorimetric data also show that fully hydrated short-chain mixed-chain C(X):C(X+6)PCs with $X < 9$ do not undergo the endothermic phase transition upon heating. In addition, the thermodynamic parameters associated with the main phase transition for aqueous dispersions of C(13):C(19)PC and other identical MW phosphatidylcholine are inversely related to $\Delta C/CL$, suggesting that the lateral chain–chain packing characteristics of these phosphatidylcholines are perturbed proportionally in the gel-state bilayer as the normalized inequivalence of the two acyl chains within each lipid molecule increases progressively. This linear relationship can be applied to map, at least to a first approximation, the values of T_m for a large number of mixed-chain phosphatidylcholines whose values of $\Delta C/CL$ are within the range of 0.1–0.4.

References

Boggs, J. M., & Mason, J. T. (1986) *Biochim. Biophys. Acta 863*, 231–242.
Chen, S. C., & Sturtevant, J. M. (1981) *Biochemistry 20*, 713–718.
Finegold, L., & Singer, M. A. (1986) *Biochim. Biophys. Acta 855*, 417–420.
Gomori, G. (1942) *J. Lab. Clin. Med. 27*, 955–958.
Huang, C. (1990) *Klin. Wochenschr. 68*, 149–165.
Huang, C., & Levin, I. W. (1983) *J. Phys. Chem. 87*, 1509–1513.
Huang, C., & Mason, J. T. (1986) *Biochim. Biophys. Acta 864*, 423–470.
Huang, C., Lapides, J. R., & Levin, I. W. (1982) *J. Am. Chem. Soc. 104*, 5926–5930.
Huang, C., Mason, J. T., & Levin, I. W. (1983) *Biochemistry 22*, 2775–2780.
Lewis, R. N. A. H., Mak, N., & McElhaney, R. N. (1987) *Biochemistry 26*, 6118–6126.
Lin, H., Wang, Z. & Huang, C. (1990) *Biochemistry* (preceding paper in this issue).
Mabrey, S., & Sturtvant, J. M. (1976) *Proc. Natl. Acad. Sci. U.S.A. 73*, 3862–3866.
Mason, J. T., & Huang, C. (1981) *Lipids 16*, 604–608.
Mason, J. T., Broccoli, A. V., & Huang, C. (1981) *Anal. Biochem. 113*, 96–101.
Mattai, J., Scripada, P. K., & Shipley, G. (1987) *Biochemistry 26*, 3287–3297.
Mena, P. L., & Djerassi, C. (1985) *Chem. Phys. Lipids 37*, 257–270.
Racey, T. J., Singer, M. A., Finegold, L., & Rochon, P. (1989) *Chem. Phys. Lipids 49*, 271–288.
Slater, J. L., & Huang, C. (1988) *Prog. Lipid Res. 27*, 325–359.
Stümpel, T., Eibl, H., & Nicksch, A. (1983) *Biochim. Biophys. Acta 727*, 246–254.
Xu, H., & Huang, C. (1987) *Biochemistry 26*, 1036–1043.
Zaccai, G., Büldt, G., Seeling, A., & Seelig, J. (1979) *J. Mol. Biol. 134*, 693–706.