

Biphasic Effect of Alkyltrimethylammonium Bromides on Gel-to-liquid-crystalline Phase Transition Temperature of Dilauroylphosphatidic Acid (DLPA) Vesicle Membrane

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The effect of alkyltrimethylammonium bromides on the gel-to-liquid-crystalline phase transition temperature of DLPA vesicle membrane was studied. In the measured concentration range, the surfactants with C_{14}/C_8 chain lengths elevated/depressed the transition temperature monotonously, whereas those with C_{10} and C_{12} elevated it at low concentrations and depressed it at high concentrations.

The interaction of surfactants with model membranes has become to be investigated related to the solubilization or fusion of biological membranes and to the preparation of lipid vesicles by detergent removal methods. In addition to these rather practical interests, surfactants may serve as a model of amphiphilic drugs to elucidate their action mechanism; e.g., alkyltrimethylammonium salts are regarded as a model of tertiary amine local anesthetic. From the latter point of view, we have been studying the effect of various surfactants on the membrane properties of the lipid vesicle paying a special attention to the gel-to-liquid-crystalline phase transition.¹⁾ In this communication, we report an interesting behavior of the phase transition temperature of the negatively charged vesicle membrane induced by the addition of alkyltrimethylammonium bromides.

Synthetic dilauroylphosphatidic acid (DLPA) was obtained from Sigma (98%, sodium salt). Octyl-, decyl-, dodecyl-, and tetradecyltrimethylammonium bromide (OTABr, DeTABr, DTABr, and TTABr, respectively) were recrystallized twice from

acetone-ethanol mixture. The vesicle in water was prepared by sonication in the cup-horn of a Branson Sonifier Model 185 at 35 °C for 15 min. The sample was prepared by mixing the DLPA suspension and the surfactant solution to give desired concentrations, and sonicated briefly at 35 °C. The pH of the sample suspension was 7.5 ± 0.2 , where DLPA has an electrical charge of -1 .²⁾ The DLPA concentration was about 0.5 mM^\dagger throughout the experiments. The gel-to-liquid-crystalline phase transition of DLPA vesicle membrane was monitored by the scattered light intensity of the sample.³⁾

Typical scattered light intensity profiles with rising temperature are shown in Fig. 1. Drastic change corresponding to the phase transition is observed. In contrast to dipalmitoylphosphatidylcholine (DPPC) vesicle, the transition is fairly asymmetric; this seems to be general for vesicle membranes of charged lipids.⁴⁾ The transition temperature, T_m , was determined by drawing straight lines as indicated by dotted lines in Fig. 1. T_m of pure DLPA obtained for 18 runs was 32.0 °C with standard deviation of 0.2 °C, which is in agreement

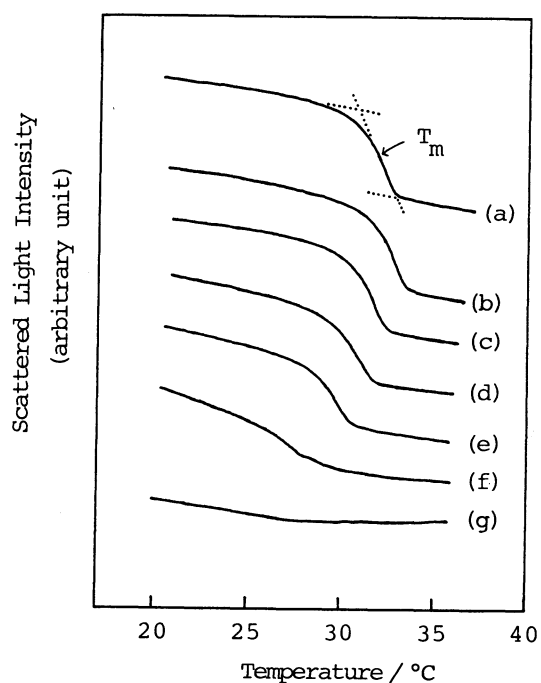


Fig. 1. Typical traces of the change in scattered light intensity with the temperature rise observed with DLPA-DeTAbR system. Wave length is 400 nm. Heating rate is 0.5 °C/min. DLPA concentration is 0.506 mM. DeTAbR concentrations (mM) are (a) 0, (b) 0.026, (c) 0.052, (d) 0.078, (e) 0.104, (f) 0.15, and (g) 1.0.

[†] 1 mM = $1 \times 10^{-3} \text{ mol dm}^{-3}$.

with literature.⁵⁾ As shown in Fig. 1, T_m is affected by the addition of DeTABr. With the increased surfactant concentration, the change in scattered light intensity at the transition region became small accompanied with deformation of the transition curve (curve (f) in Fig. 1), and further increase of the concentration eliminated the transition (curve (g) in Fig. 1). Thus, surfactant concentrations were limited to the range where normal phase transition behavior was observed.

Figure 2 illustrates the change in the transition temperature, ΔT , with the added surfactant concentration. ΔT shows a complex behavior depending on the alkyl chain length of the surfactants. Short-chained OTABr decreases T_m and long-chained TTABr increases it monotonously, whereas DeTABr and DTABr having an intermediate chain length exhibit a biphasic effect on T_m . To alkyltrimethylammonium bromides, DLPA behaves quite differently from DPPC, where T_m is decreased almost linearly with the concentration regardless of the alkyl chain length of the surfactants.¹⁾ This may be attributed to the difference in the net electrical charge on the lipid head group; negative charge (DLPA) vs 0 (DPPC). Similar biphasic behavior of the phase transition temperature has also been reported with phosphatidylcholine - 1-alkanol systems,⁶⁾ although the effect is reverse; 1-alkanols with $C_9 - C_{13}$ chain length depress the transition

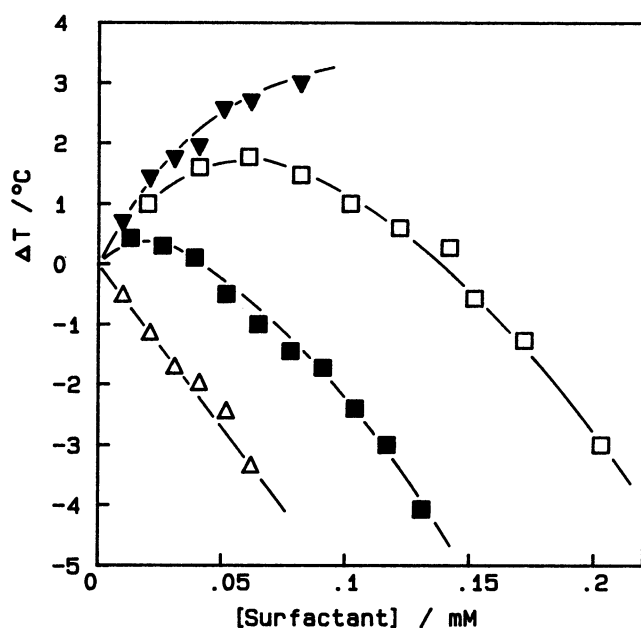


Fig. 2. Effect of alkyltrimethylammonium bromides on the phase transition temperature of DLPA vesicle membrane. $\Delta T = T_m - T_{m,0}$, where T_m and $T_{m,0}$ are the transition temperature in the presence of and in the absence of the surfactant, respectively. Δ ; OTABr, \blacksquare ; DeTABr, \square ; DTABr, and \blacktriangledown ; TTABr.

temperature at lower concentrations and elevate it at higher concentrations.

Another remarkable difference between DLPA and DPPC is the required concentration ranges of the surfactants for the perturbation. For DPPC, the range strongly depends on the alkyl chain length of the surfactants; the shorter chain, the higher concentration.¹⁾ This is interpreted as the interaction between DPPC vesicle membrane and the surfactants is mainly due to the hydrophobic effect. On the other hand, the perturbing effect on DLPA vesicle membrane appears in a similar surfactant concentration range irrespective of the alkyl chain length. The change in the transition temperature is considered to be caused by the incorporation of the surfactant molecule into the membrane. Thus, it is suggested that the electrostatic interaction plays a dominant role for the binding of alkyltrimethylammonium ion to DLPA vesicle membrane.

The monotonic variation of ΔT with the additive concentration may be readily interpreted based on the van't Hoff model for the freezing point depression. The biphasic behavior of ΔT observed in this study seems to need some other considerations. Although no detailed interpretation can be given in the present stage, the observed biphasic phenomenon may deserve further attention in relation to the interaction between negatively charged lipid bilayer and cationic long-chain amphiphiles.

This work was supported in part by funds from the Central Research Institute of Fukuoka University.

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(Received November 11, 1987)