

Chapter 6

Effect of Salts on the Lamellar-Lamellar Coexistence in the DDAB-Water System

6.1 Introduction

Charged amphiphilic systems are often found to exhibit complicated phase behaviour. Different counterions of same valency are found to lead to different kinds of phase behaviour [1, 2, 3]. Even neutral lipid bilayers are found to show varying affinity to adsorb different monovalent counterions [4]. These ion specific effects force us to recognize additional physical interactions beyond double-layer electrostatics. However, a proper theoretical understanding of these effects is still lacking.

In this chapter, we study the effect of different monovalent counterions on the lamellar-lamellar coexistence seen in the ionic surfactant, didodecyldimethylammonium bromide (DDAB). Inorganic salts, sodium bromide (NaBr) and sodium chloride (NaCl) and organic salts, 3-sodium-2-hydroxy naphthoate (SHN) and sodium salicylate (SS) were used to introduce the different counterions. Earlier studies on DDAB have been briefly described in section 6.2. This lipid is found to exhibit a lamellar-lamellar coexistence over a range of concentration. Interestingly, the analogous surfactant with Cl^- counterion instead of Br^- does not show such a two phase region. There have been many theoretical attempts to explain the lamellar-lamellar coexistence seen in the presence of Br^- counterions; these are discussed in section 6.3. A brief description of the experimental techniques is given in the next section. The results of these studies are presented in section 6.4. It is found that all the salts shift the coexistence region to much lower concentrations of the surfactant. Interestingly all the salts are found to be equally efficient in inducing the coexistence, in contrast

to some recent theoretical predictions. In the case of the organic salts, the collapsed lamellar phase is found to transform into a bicontinuous cubic phase corresponding to the space group $Pn3m$ at much higher salt concentrations. Such a transition is also found with increasing temperature at slightly lower concentrations of these salts. In section 6.6, our results are discussed in terms of the theoretical expectations on the behaviour of these systems. The last section contains conclusions that can be drawn from the present studies.

6.2 Earlier studies

The synthetic cationic double-chain surfactant DDAB in water shows a phase behaviour strikingly different from those of other double-chain lipid systems (Fig. 6.1) [1, 5]. At very dilute aqueous solution it forms a phase of single layer vesicle ($^A L_3$) coexisting with random open bilayer structures. The swollen lamellar phase (L_α) is found at higher concentration which is made up of multilamellar vesicles with an onion-like structure. The most interesting part of the phase diagram is the coexistence region of a swollen L_α phase and a collapsed lamellar phase (L'_α) with very little water between the bilayers. The region starts from ~ 40 wt% of DDAB and spreads over upto 85 wt%. At still higher concentrations only the collapsed phase is seen. On heating the coexistence region disappears and a single lamellar phase is seen. The transition temperature increases with the DDAB wt% and reaches to a critical temperature ($\sim 80^\circ$ C). At still higher temperatures an optically isotropic phase (I) is seen at these concentrations. Small angle neutron scattering (SANS) experiments on this phase show only a broad correlation peak. Some authors have reported a flow birefringent phase (L_3) at very low surfactant concentrations [6]. Later on this phase has been shown to be a metastable phase [5]. Didodecyldimethylammonium chloride (DDAC) in water shows a different phase behaviour from DDAB. It shows a single lamellar phase over the whole concentration range [7]. This difference due to the presence of Cl^- ion instead of Br^- ion has led to some detailed experiments on similar systems. The effect of other counterions, such as sulphate, acetate and hydroxide, has also been studied. Interestingly, none of these systems shows the coexistence of two lamellar phases.

Osmotic pressure experiment has been carried out on DDAB-water and DDAC-water systems

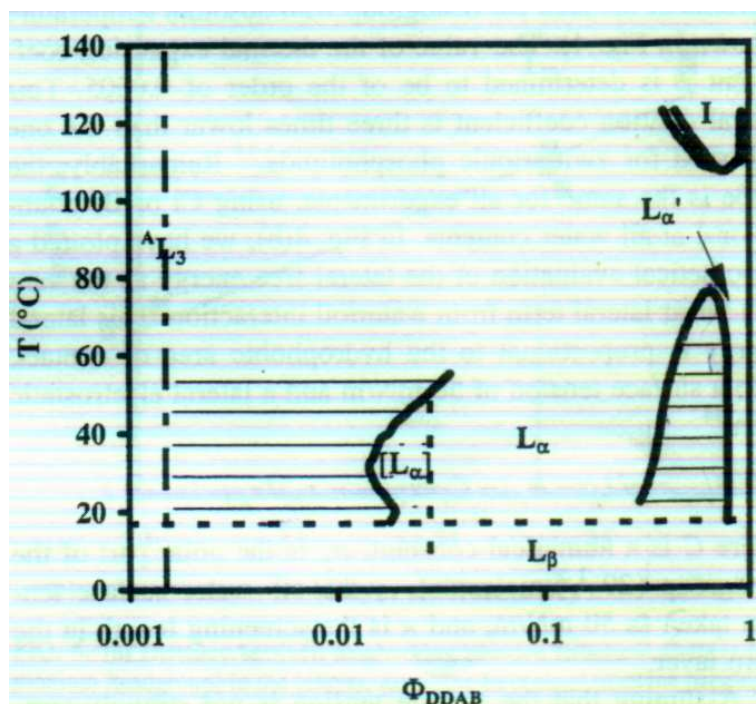


Figure 6.1: Phase diagram of DDAB-water binary system. $[L_\alpha]$, L_α , L'_α and I denote the vesicles, swollen lamellar, collapsed lamellar and isotropic phases respectively [1].

using polyethylene glycol solutions as the stressing medium [1]. The dependence of the lamellar periodicity (d) on the osmotic pressure (Π) is found to be very different for the two systems. d decreases monotonically with increasing Π in the case of DDAC. On the other hand, the d vs Π curve in DDAB shows a discontinuity, with two values of d coexisting over a range of Π .

There are very few reports in the literature of systems which show such a coexistence of two lamellar phases with different water content in between the bilayers. Sodium dodecyl-5-*p*-benzenesulfonate-water binary system shows such a behaviour characterized by an upper critical temperature of $\sim 39^\circ\text{C}$ [8]. The authors attribute the coexistence of two lamellar phases to the presence of an additional interbilayer attractive potential caused by specific Na^+ ion-head group interaction. Coexistence of two L_α phases has also been observed in some systems in the presence of certain polymers [9]. In this case the two phases differ in the amount of polymer they contain.

A detailed characterization of cubic phases observed in DDAB system in the presence of some hydrophobic molecules has been reported in references [10] and [11]. Cyclohexane is found to induce both body-centred and primitive cubic phases separated by a coexistence region containing

cubic phases belonging to the space groups $Im3m$ and $Pn3m$, respectively. In case of styrene the existence of five distinct bicontinuous cubic phases has been reported. The structural sequence of the four cubic phases on increasing the water content has been identified as $Ia3d \rightarrow Pn3m \rightarrow Im3m(P) \rightarrow Im3m(C(P))$. The structure of the fifth cubic phase is still unknown. The ternary phase diagram of DDAB-lecithin-water also contains a bicontinuous $Ia3d$ cubic phase over a narrow composition range in the water-poor part [12]. Cubic phases of lipids have potential medical and industrial applications. This phase has been used as a substrate for the crystallization of membrane proteins, which are very difficult to crystallize using conventional protocols [13]. Recently this phase has also been used as a vehicle for drug delivery. This phase is also used in nanotechnology as a template for synthesizing nanoparticles [14, 15].

6.3 Theoretical studies on lamellar-lamellar coexistence in amphiphilic systems

There have been some theoretical attempts to explain the lamellar-lamellar coexistence in some neutral and ionic amphiphilic systems. Leibler and Lipowsky showed the possible existence of a continuous “unbinding transition” where a system goes continuously from a bound state with finite bilayer separation to an unbound state of infinite swelling [16, 17]. In several papers Wennerstrom and his co-workers proposed this transition to be a first order one with a lamellar-lamellar coexistence region [18, 19]. The effective interaction potential between fluctuating membranes of zwitterionic lipids can be expressed as [20],

$$V_{eff} = A_H e^{-L/\lambda_H} - \frac{W}{12\pi} \left[\frac{1}{L^2} - \frac{2}{(L+\delta)^2} + \frac{1}{(L+2\delta)^2} \right] + \frac{(k_B T)^2}{\kappa} \frac{1}{L^2}, \quad (6.1)$$

where δ is the bilayer thickness, λ_H the decay length of short-range hydration force and A_H its amplitude. L and W are the bilayer separation and Hamaker constant respectively. Here $k_B T$ is the thermal energy and κ is the membrane bending rigidity. The three terms in the above expression represent contributions from hydration repulsion, van der Waals attraction and the Helfrich repulsion, respectively. The short range hydration force originates from the thin layer of ordered water molecules at the bilayer-water interface. This repulsive interaction dominates when the interven-

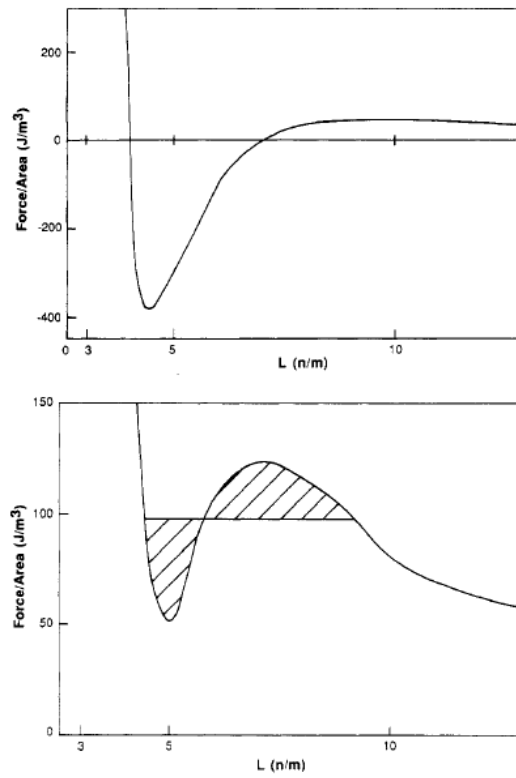


Figure 6.2: Plotting of effective force $-dV_{eff}/dL$ versus membrane separation. Parameters: $A_H = 0.2 \text{ J/m}^2$, $\lambda_H = 0.3 \text{ nm}$, $W = 6 \times 10^{-21} \text{ J}$, $k_B = 1.5 \times 10^{-19} \text{ J}$. $T = 270 \text{ K}$ (top) and 300 K (bottom) [20].

ing water layer is very small (≤ 1 nm). The van der Waals or dispersion force can be calculated from the dielectric permittivities of the interacting media. The steric repulsion originates from the thermal undulations of the bilayers. This force along with the electrostatic repulsion (in case of a charged system) balances the van der Waals attraction at intermediate to large bilayer spacings. By taking the spacial derivative of the above potential one obtains the pressure as shown in figure 6.2. The pressure versus distance curve is analogous to the pressure (P)-volume (V) curve used in elementary treatment of liquid-gas transition. The Maxwell equal area construction can also be applied here. On the basis of this simple approach, for certain values of parameters in equation (6.1) (see Fig. 6.2), one can obtain a first order transition between a strongly bound state with a small bilayer separation to a weakly bound state with a large separation at a particular value of osmotic pressure.

The above treatment has been extended to charged bilayers by Ricoul et al. [21]. Along with the van der Waals attractive term they have introduced another shorter-range adhesion force contributed by the polar head group of the lipid molecules,

$$\Pi_{adh} = -\Pi_a e^{-(L-e_a)/\lambda_a}, \quad (6.2)$$

where the notations are similar to those of hydration force. e_a can be chosen arbitrarily. The electrostatic repulsive force at large distance ($L > 10$ nm) was expressed as a modified Poisson-Boltzmann equation as,

$$\Pi_{elec} \approx 64k_B T c_s \gamma^2 e^{k_d(L-e_e)}, \quad (6.3)$$

where $\gamma = 1$ for highly charged membrane and $1/k_d$ is the Debye length. c_s is the salinity of the reservoir and $(L - e_e)$ is the water thickness accessible to the counterions. Two attractive forces; van der Waals and adhesion are balanced by two repulsive forces hydration and electrostatics to give rise the equation of state,

$$\Pi_{total} = \Pi_{vdw} + \Pi_{adh} + \Pi_{hyd} + \Pi_{elec} \quad (6.4)$$

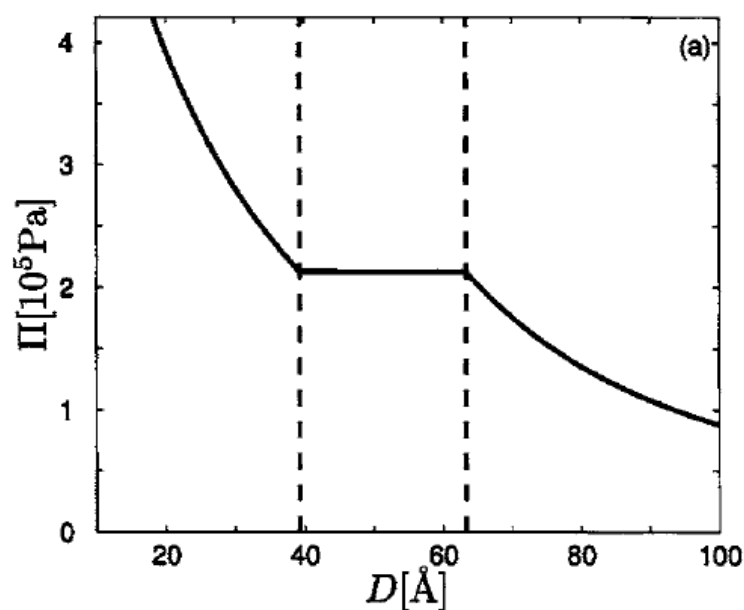


Figure 6.3: The osmotic pressure as a function of inter lamellar spacings from equation (6.5) using parameters $\alpha = -6$, $a = 8 \text{ \AA}$, and $\chi = 12$ [22].

The undulation interaction was neglected since the bilayers are assumed to be rather rigid, as suggest by the diffraction data from some of the systems. Using certain reasonable values of parameters in the above equations, the pressure (Π_{total}) was calculated as a function of the bilayer separation (L). The Maxwell construction allows the determination of the equilibrium pressure plateau and the periodicities of the collapsed and swollen lamellar phases. Although this approach explains the coexistence of two lamellar phases, it suffers from the shortcoming that the origin of the short range adhesion force is not specified; it remains an unsubstantiated assumption.

A theoretical approach has been proposed recently to explain the lamellar-lamellar coexistence specifically in the DDAB-water system [22]. From experiments it is evident that different ions associate differently with the amphiphile-water interface and that their binding can give rise to conformational changes at the interface [23]. In this model the authors assume that only a fraction of Br^- ions are dissociated from the surfactant bilayer. They then assume nonideal mixing of the charged and neutral amphiphiles in the plane of the bilayer, which leads to a phase separation of these two species so as to minimize the line tension between them. There are two phenomenological parameters in this model; α describes the degree of dissociation of the ions and χ describes the

lateral phase separation of the neutral and charged amphiphiles. Negative values of α correspond to a specific nonelectrostatic attraction between the counterions and the surface and results in more counterions to be associated at the surface. On the other hand, positive values of χ correspond to the tendency of surfactants to phase separate into domains of neutral and charged species.

The total free energy (F_{tot}) as a function of bilayer separation (D) and surface area fraction of charged surfactants (η_s), which is related to α , is expressed as,

$$\frac{a^2}{k_B T} F_{tot} = -\frac{1}{2\pi l_B} [4\pi l_B \eta_s - \frac{1}{2}(Ka)^2 D] + \frac{1}{2}\chi\eta_s^2 + \ln(1 - \eta_s) \quad (6.5)$$

where l_B is the Bjerrum length at which the electrostatic interaction between two elementary charges is equal in magnitude to the thermal energy ($\approx 7 \text{ \AA}$ at room temperature for aqueous solutions). a is the dimension of surfactant head group and the constant K is expressed as,

$$KD \tan(KD/2) = \frac{2\pi l_b}{a^2} \eta_s D, \quad (6.6)$$

This expression for the total free energy comes from an extended Poisson-Boltzmann theory, which takes into account volume contributions coming from the electrolyte solution including ions and surface contributions having their origin at the interface. It does not include the contributions from hydration force.

The osmotic pressure (Π) can be obtained by taking the spatial derivative of the free energy and a typical isotherm has been shown in figure 6.3. For $\alpha = -6$, $a = 8 \text{ \AA}$, and $\chi = 12$, the Maxwell construction gives a coexistence between a phase with $D \approx 39 \text{ \AA}$, and low $\eta_s \leq 0.1$, and another with $D \approx 64 \text{ \AA}$, and $\eta_s \approx 1$. For large bilayer separation most of the counterions are dissociated ($\eta_s \approx 1$), whereas for the collapsed lamellar phase, only a small fraction of the counterions are dissociated ($\eta_s \leq 0.1$). The plateau in Π indicates the condensed to swollen lamellar transition to be a first order one. In the present scenario, the contribution of hydration force is needed only to explain the experimental value of osmotic pressure at very low layer separation. The absence of lamellar-lamellar coexistence in case of DDAC can be attributed to the unfavourable value of the phenomenological parameter α . For fully dissociated surface ($\eta_s = 1$) where all Cl^- ions are in solution, $\alpha \rightarrow \infty$ giving rise to standard Poisson-Boltzmann isotherm (Fig. 6.4). This is

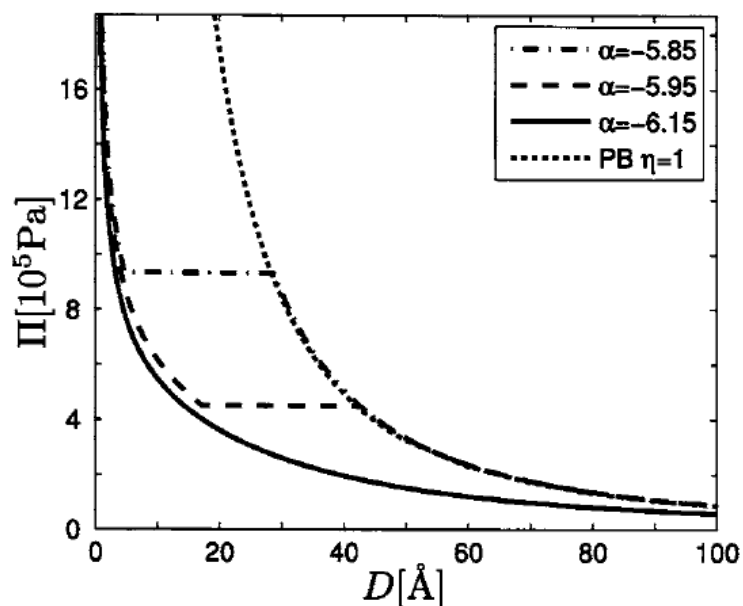


Figure 6.4: The osmotic pressure isotherm for different binding strengths: $\alpha = -5.85$, -5.95 , and -6.15 . The other parameters, are $\chi = 12$ and $a = 8 \text{ \AA}$. The usual PB isotherm is also shown with $\eta_s = 1$ [22].

reasonable since experiments show the larger Br^- ion to have much more affinity to get adsorbed at the bilayer [24, 25]. By comparing the predicted behaviour with the experimentally determined osmotic pressure data, the authors have estimated the values of the two parameters for Br^- and Cl^- counterions. The value of α is found to be -7.4 and -3.4 , respectively, for $\chi = 14.75$ and $a = 8 \text{ \AA}$.

The influence of added NaBr was considered within the same framework by these authors, using the appropriate values of χ and α . With increasing concentration of NaBr, the Debye-Huckel screening length decreases and the phase transition which is observed in the absence of salt is found to become gradually less prominent and finally it is wiped out at higher salt concentrations and only the collapsed phase is obtained (Fig. 6.5). Although the authors have not performed the corresponding calculations for NaCl, the less favourable value of α obtained for Cl^- suggests that this salt should be less effective in the formation of the collapsed phase.

6.4 Experimental

DDAB-SHN and DDAB-SS samples were prepared by first weighing out appropriate amounts of DDAB and SHN or SS into a glass tube. Then deionized water was added to it to get the required

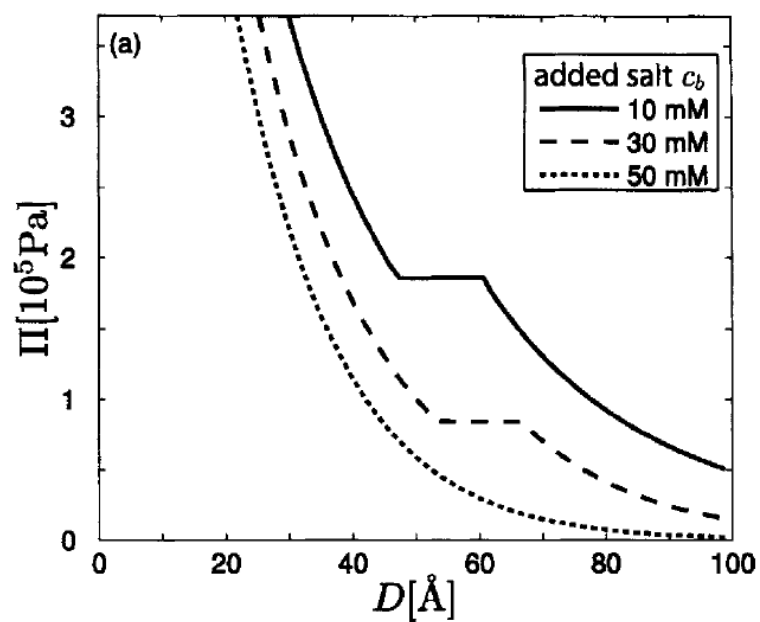


Figure 6.5: Effect of added NaBr on the lamellar - lamellar phase transition. Note that the transition vanishes at higher salt concentrations. The parameters are, $\alpha = -5$, $a = 8 \text{ \AA}$, and $\chi = 10.19$ [22].

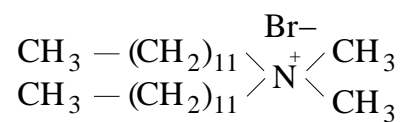


Figure 6.6: Chemical structure of didodecyldimethylammonium bromide (DDAB).

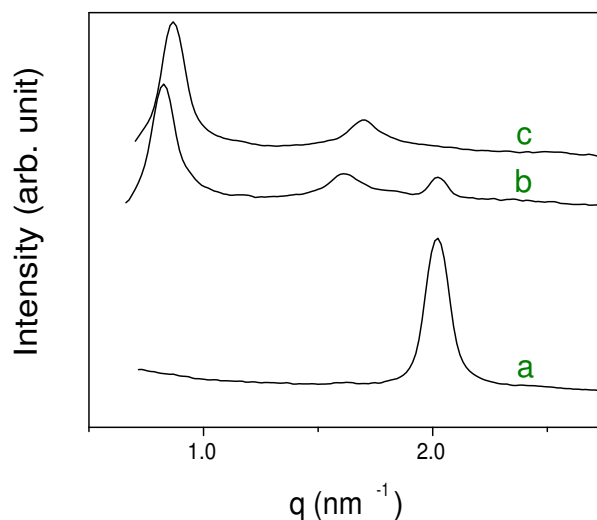


Figure 6.7: Diffraction patterns of the DDAB-NaBr system at $\phi_s = 30$ and $[\text{NaBr}] = 45\text{mM}$; (a) L'_α phase at 30°C , (b) coexistence of L'_α and L_α phases at 50°C and (c) L_α at 70°C .

value of ϕ_s ($= \frac{\text{DDAB} + \text{SHN}}{\text{DDAB} + \text{SHN} + \text{water}}$). In case of inorganic salts, NaBr and NaCl, first stock salt solutions were prepared and they were added to DDAB to get the required ϕ_s . The chemical structure of DDAB has been shown in figure 6.6 and the structures of other chemicals have already been shown in previous chapters. Details of the x-ray diffraction and polarizing microscopy techniques used to characterize the phases have been described in chapter 2.

6.5 Results

6.5.1 Effect of NaBr

The influence of NaBr on the swollen lamellar phase (L_α) of DDAB was studied in detail. The boundary of the two-phase region ($\phi_s \sim 45$ wt%) was far from the concentrations at which these experiments were performed ($\phi_s \sim 20$ and $30\text{wt}\%$). With the gradual addition of NaBr the bilayer separation is initially found to decrease by a few Å and then the collapsed lamellar phase is induced. The diffraction patterns of both the swollen and collapsed lamellar phases consist of few peaks with their q values in the ratio 1:2:3. The coexistence of the swollen (L_α) and collapsed (L'_α) lamellar phases persists over a range of NaBr concentration. When the L'_α phase first appears, the L_α phase swells taking in the water released from the former (Table 6.1). On further increase of salt content

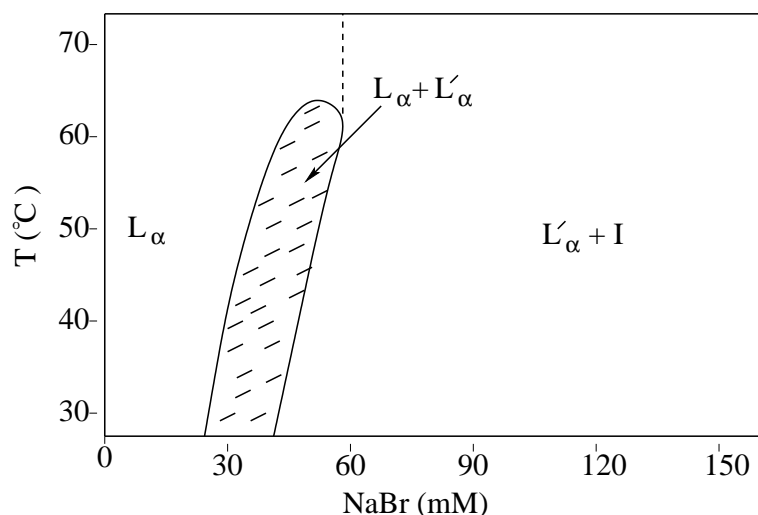


Figure 6.8: Phase diagram of DDAB-NaBr system at $\phi_s = 30$. L_α and L'_α denote the swollen and collapsed lamellar phases respectively. The shaded regions in all the phase diagrams denote the coexistence of two phases. The dashed line refers to the phase boundary which is not determined precisely.

the collapsed phase exists along with a very dilute phase which looks like water, which we denote as an isotropic phase. We are, however, unable to precisely determine the boundary between the L_α - L'_α coexistence region and the L'_α -I coexistence region, due to the large periodicity of the swollen lamellar phase and its weak birefringence. Addition of more salt does not affect the L'_α phase upto 1M NaBr concentration. The L'_α phase observed at medium salt concentrations is found to transform to the L_α phase through a two phase region on heating. Diffraction experiments show a gradual decrease of the L'_α peak intensity and a gradual increase of the L_α peaks on heating (Fig. 6.7). The transition temperature increases with increasing NaBr concentration (Fig. 6.8). It is also seen from figures 6.8 and 6.9 that more dilute the DDAB solution, more the concentration of NaBr at which the collapsed lamellar phase is first seen. This results in the shift of the boundary of the two-phase region towards higher salt concentrations for lower ϕ_s .

6.5.2 Effect of NaCl

The effect of NaCl on the phase behaviour of dilute DDAB solutions is found to be very similar to that of NaBr. NaCl is also able to induce the collapsed lamellar phase (L'_α). The diffraction pattern of a sample in the coexistence region is shown in figure 6.10, which shows the presence of

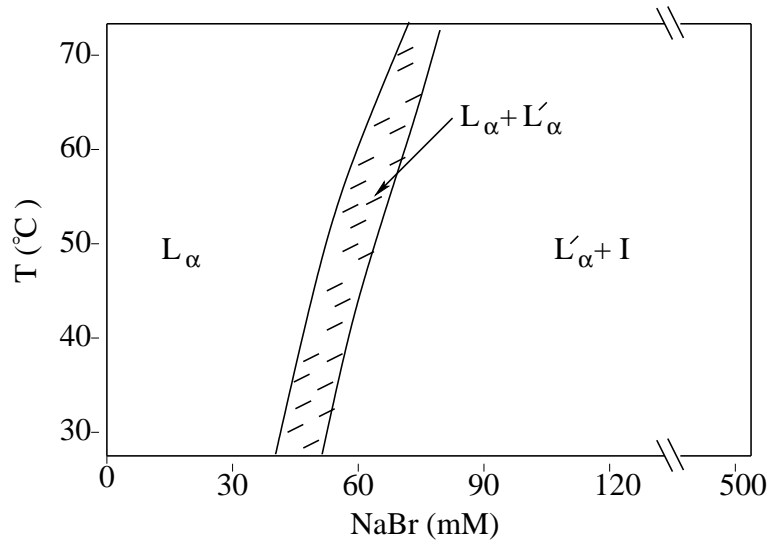


Figure 6.9: Phase diagram of DDAB-NaBr system at $\phi_s = 20$. The symbols are the same as those in figure 6.8. Note that more salt needs to be added to induce the collapsed phase in this case compared to $\phi_s = 30$.

Table 6.1: Diffraction data of the DDAB-water system at 30°C in presence of NaBr. d_1 and d_2 denote the lamellar periodicity in L_α and L'_α phases respectively.

ϕ_s	NaBr(mM)	d_1 (nm)	d_2 (nm)	phase
30	0	8.32	-	L_α
	15	7.86	-	L_α
	20	7.56	-	L_α
	30	8.44	3.08	L_α, L'_α
	45	-	3.08	L'_α
	75	-	3.08	L'_α
	150	-	3.08	L'_α
	1000	-	3.04	L'_α
20	0	11.8	-	L_α
	15.5	10.24	-	L_α
	30	10.24	-	L_α
	40	10.10	-	L_α
	50	-	3.08	L'_α
	60	-	3.11	L'_α
	143	-	3.10	L'_α

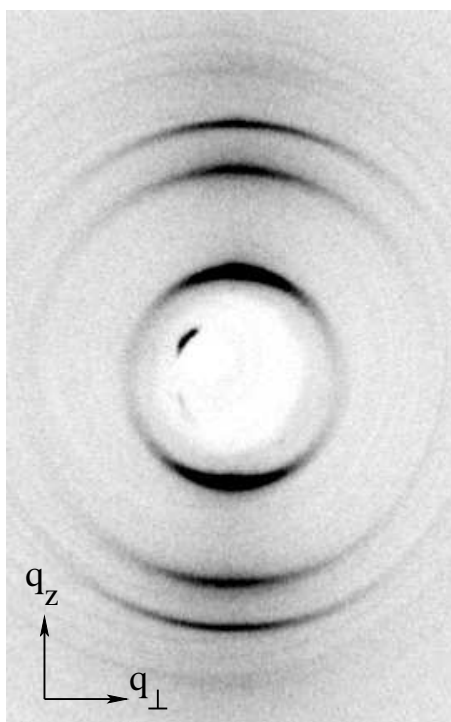


Figure 6.10: Diffraction pattern of a partially aligned sample showing the coexistence of two lamellar phases, L_α and L'_α in DDAB-NaCl system at $\phi_s = 30$, $[\text{NaCl}] = 20\text{mM}$ and $T = 30^\circ\text{C}$.

two sets of lamellar reflections. The surprising observation is that the salt concentration at which the coexistence regime is first observed is very similar for NaBr and NaCl (Table 6.1 & 6.2). As discussed later this observation is unexpected on the basis of the theoretical predictions described in section 6.3. The phase diagrams at $\phi_s = 30$ and 20 in the presence of NaCl are very similar to the respective phase diagrams with NaBr (Figs. 6.8 and 6.9). As in the case of NaBr the addition of more NaCl does not alter the structure of the collapsed lamellar phase.

6.5.3 Effect of SHN

At low concentrations SHN behaves identical to NaBr and NaCl. The critical salt concentration for inducing coexistence is also similar to the other two inorganic salts. The L'_α phase is found to coexist with a dilute isotropic phase over a wide range of salt concentration (Fig. 6.11). Again more SHN is required to induce the collapsed phase in more dilute DDAB solutions. However, at high concentrations SHN has strikingly different effect on the phase behaviour of DDAB compared to NaBr and NaCl. Above $[\text{SHN}] \sim 0.5\text{M}$, the collapsed lamellar phase is transformed into a viscous

Table 6.2: Diffraction data of the DDAB-water system at 30°C in presence of NaCl. d_1 and d_2 denote the lamellar periodicity in L_α and L'_α phases, respectively.

ϕ_s	NaCl(mM)	d_1 (nm)	d_2 (nm)	phase
30	0	8.32	-	L_α
	15	7.24	-	L_α
	20	7.58	3.11	L_α, L'_α
	31	8.32	3.11	L_α, L'_α
	45	8.46	3.10	L_α, L'_α
	60	-	3.11	L'_α
	150	-	3.11	L'_α
20	0	11.8	-	L_α
	15.8	10.59	-	L_α
	60	9.70	-	L_α
	75.7	-	3.13	L'_α
	149	-	3.13	L'_α

isotropic phase for $\phi_s = 20$ (Fig. 6.12). Samples at these compositions show the coexistence of two isotropic phases. X-ray studies show a number of sharp reflections from the viscous isotropic phase, indicating a high degree of positional ordering. These peaks are in the ratio $\sqrt{2} : \sqrt{3} : \sqrt{4} : \sqrt{6} : \sqrt{8} : \sqrt{9} : \sqrt{10} : \sqrt{12}$ (Table 6.3). The absence of the $\sqrt{7}$ reflection indicates the lattice to be a primitive cubic lattice corresponding to the space group $Pn3m$ [26]. The indexing scheme and the diffraction profile are very similar to those reported in the literature for the $Pn3m$ cubic phase of DDAB induced by cyclohexane and styrene [10, 11]. The phase boundary of this cubic phase shifts towards lower salt concentrations on decreasing the DDAB concentration; the opposite trend is found for the appearance of the collapsed lamellar phase. Interestingly, the critical molar ratio of the two charged components ($\beta = [\text{SHN}]/[\text{DDAB}]$) at the onset of the cubic phase is found to be the same at all DDAB concentrations. On the other hand, such a trend is not observed at the onset of the collapsed lamellar phase.

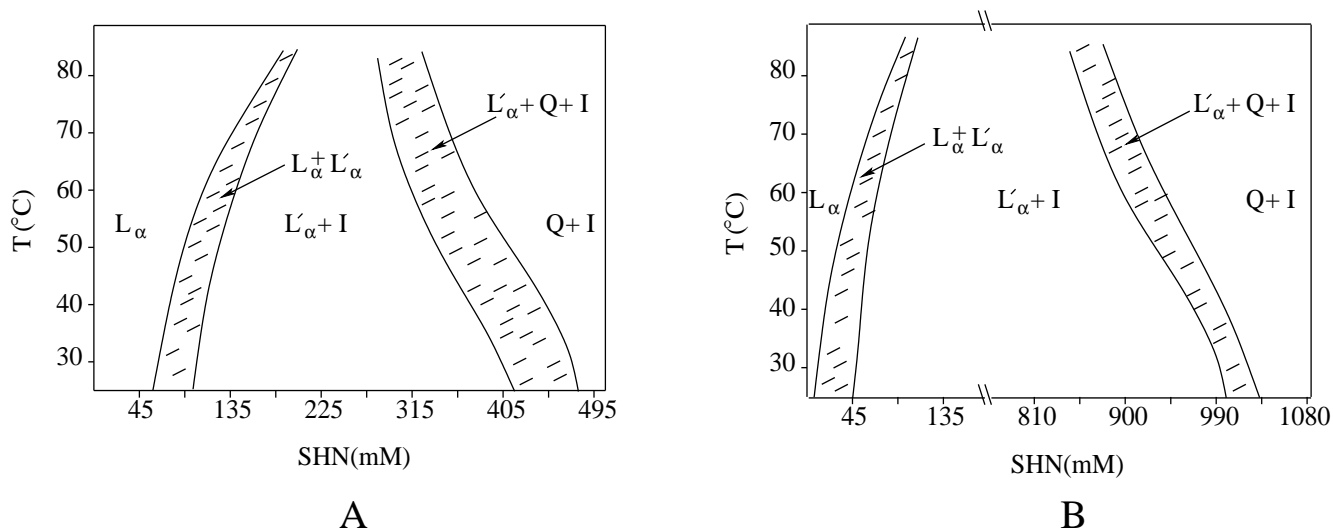


Figure 6.11: Phase diagrams of DDAB-SHN systems for $\phi_s = 20$ (A) and 40 (B). Q denotes the cubic phase corresponding to the space group $Pn3m$.

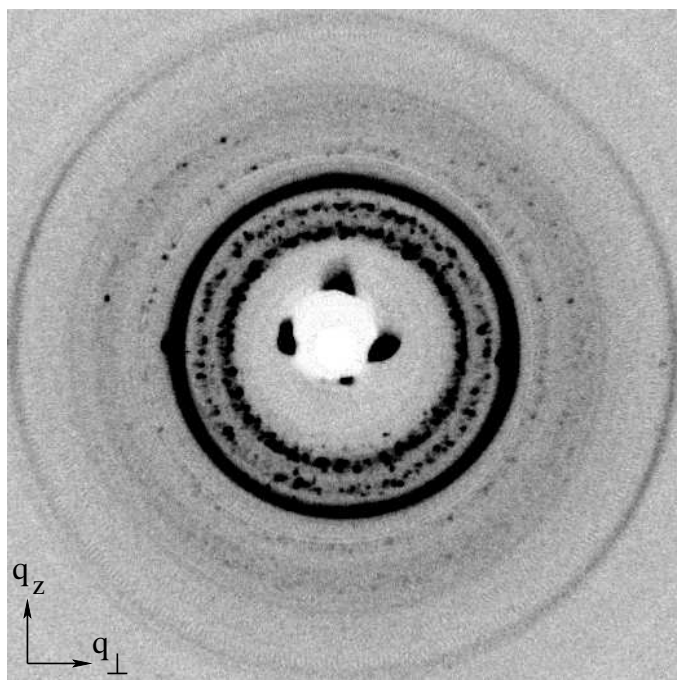


Figure 6.12: Diffraction pattern of coexistence of collapsed lamellar (L'_{α}) and cubic (Q) phases in DDAB-SHN system at $\phi_s = 30$, $[\text{SHN}] = 0.62 \text{ M}$ and $T = 50^{\circ}\text{C}$. The L'_{α} phase shows smooth rings whereas the Q phase always shows rings of granular spots.

Table 6.3: X-ray diffraction data of DDAB-SHN-water system at $\phi_s = 40$, $[\text{SHN}] = 1.37 \text{ M}$ and $T = 30^\circ\text{C}$ indexed on a primitive cubic lattice corresponding to the space group $Pn3m$ with the lattice parameter $a=6.35 \text{ nm}$.

$d_{exp}(\text{nm})$	$d_{calc}(\text{nm})$	plane	$h^2 + k^2 + l^2$	intensity
4.49	4.49	(110)	2	vs
3.66	3.62	(111)	3	s
3.17	3.18	(200)	4	m
2.59	2.593	(211)	6	m
2.25	2.26	(220)	8	m
2.11	2.09	(221)	9	m
2.01	2.02	(301)	10	w
1.84	1.82	(222)	12	w

6.5.4 Effect of SS

The effect of SS on the phase behaviour of DDAB is found to be very similar to that of SHN. It also induces the collapsed lamellar phase at around similar concentration (Fig. 6.13). On further increasing the salt concentration, the collapsed lamellar phase transforms to a cubic (Q) phase with similar diffraction pattern as seen in DDAB-SHN system. A typical diffraction pattern of this phase is shown in figure 6.14 and can be indexed as in table 6.3. Even the molar ratio (β) of SS and DDAB to induce this Q phase is similar. Only the lattice parameter is found to be few Å lower than that of SHN system shown in table 6.4.

6.6 Discussion

The origin of the lamellar-lamellar coexistence seen in some ionic amphiphilic system is still not well understood. Specially in the case of DDA^+ surfactant, many approaches have been proposed to explain such a coexistence observed with Br^- counterion and not with Cl^- . All of them based on the selective binding of bromide counterions on the amphiphilic bilayer. The theoretical model of Harries et. al [22], which has been discussed in section 6.3, also predicts that the addition of NaBr to a dilute DDAB solution in the swollen lamellar phase can induce the collapsed phase.

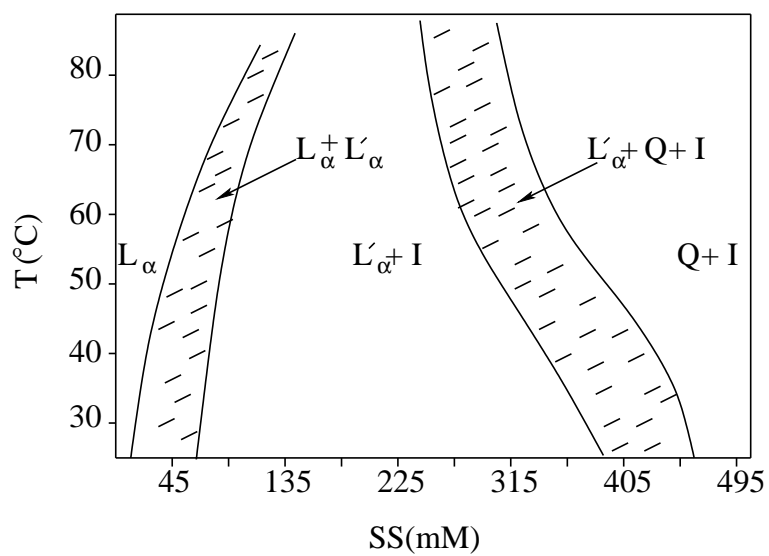


Figure 6.13: Phase diagrams of DDAB-SS system for $\phi_s = 30$. The symbols are same as those in figures 6.8 and 6.11

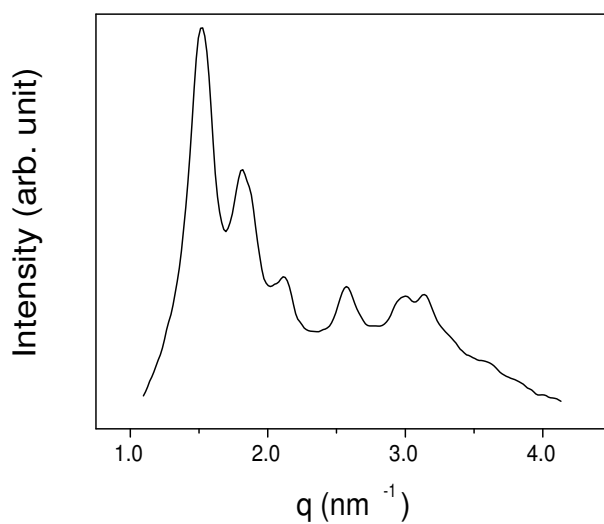


Figure 6.14: Diffraction pattern of the cubic phase corresponding to space group $Pn3m$ in DDAB-SS system at $\phi_s = 20$, $[SS] = 0.40 M$ and $T = 30^\circ C$.

Table 6.4: Variation of the lattice parameter a of the cubic phase in DDAB system with ϕ_s and concentration of organic salts at 30 °C.

ϕ_s	salt	β	conc.(M)	a (nm)
50	SHN	1	-	6.37
40	SHN	1	1.37	6.35
30	SHN	1	0.88	6.36
	SS	1	0.89	6.05
		0.7	0.63	6.77
20	SS	1	0.40	5.88
10	SHN	1	0.23	5.97

The values of phenomenological ion dissociation parameter (α) and the non ideal lipid mixing parameter (χ) were estimated for the Br^- and Cl^- counterions using the osmotic pressure data on these systems. Although the effect of added NaCl on the DDAB-water system was not explicitly calculated by these authors, the fact that the DDAC-water system does not show the collapsed lamellar phase suggests that the Cl^- ion should be less efficient in inducing the collapsed phase. But our observations show such a lamellar-lamellar coexistence even with the addition of NaCl, with critical salt concentration almost identical for both NaBr and NaCl. Hence, the efficiency to induce such a coexistence by these salts is found to be very similar (Table 6.1 and 6.2). The other organic salts, SS and SHN are also able to give rise to the same phenomenon, and at similar salt concentrations. These observations are clearly in conflict with the theoretical predictions of reference [22], and point to shortcomings in their model.

In the DDAB-water binary system an isotropic phase is found at higher temperatures for high DDAB concentrations [1]. This isotropic phase is most probably a sponge phase where the bilayer forms a random network. Because of high thermal energy, the bilayer becomes very flexible and its persistence length becomes comparable to the bilayer separation resulting in the sponge phase. As mentioned earlier, many hydrophobic molecules induce different kinds of bicontinuous cubic phases in DDAB, where there are two water channels separated by a continuous bilayer. All these observations suggest that DDAB bilayers have a tendency to form interbilayer connections.

The observation of tubule formation in DDAB vesicles in very dilute solutions supports this possibility [27]. As described in chapters 3 and 4 and also in [28], SHN can bridge the cylindrical micelles to form a 2-D network and give rise to mesh phases in systems which form rod-like micelles. One might expect a similar behaviour in a bilayer forming system. In that case it would form connections across bilayers and form a network. A gradual increase in the number of such interconnections might ultimately result in the lamellar to cubic transition. There are reports in the literature of 'stalk' formation in lipid bilayers, which are such interconnections [29, 30, 31]. Now the question arises as to whether the collapsed lamellar phase results from the formation of interconnections across bilayers. This will provide a new mechanism for the formation of the collapsed phase, which cannot be understood on the basis of present theories of these systems. One way to test this proposal would be to probe regions of high membrane curvature using techniques such as nuclear magnetic resonance (NMR). Clearly further experiments are needed to test these ideas.

X-ray diffraction data show that the optically isotropic phase formed at high SS and SHN concentrations is a cubic phase belonging to the space group $Pn3m$. A schematic of the structure of this phase is shown in figure 6.15 [32]. To a good approximation the mid-plane of the bilayer in these phases describe a minimal surface, which has vanishing mean curvature at every point [33]. As discussed in section 6.2 cubic phases belonging to different space groups have been reported in the literature in the DDAB system in the presence of different hydrophobic molecules [10, 11, 12]. In the present study, the organic salts SHN and SS also contain a hydrophobic part which is possibly responsible for inducing the cubic phase. Inorganic salts NaBr and NaCl do not induce such a phase. From table 6.4, it is seen that the lattice parameter is almost independent of the surfactant concentration (ϕ_s). This can be attributed to the fact that these systems contain very little water. The phase boundary of this cubic phase is found to depend upon ϕ_s . But at each ϕ_s , the value of β at which the cubic phase forms is similar. It shows that the bilayer has the same composition at the onset of the cubic phase in all the samples. This suggests that some properties of the bilayer are being varied gradually by the addition of SHN and SS, and results in destabilizing the flat bilayers beyond a critical composition. Experiments to measure the elastic constants associated with mean and Gaussian curvature of the bilayer might shed some light on the

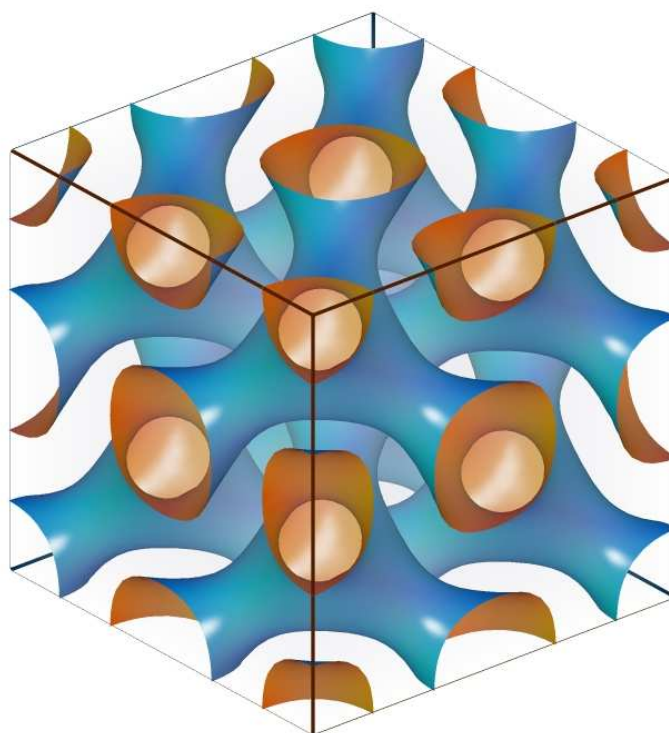


Figure 6.15: The structure of D-surface belonging to $Pn3m$ space group [32].

situation.

6.7 Conclusion

The influence of some inorganic and organic salts on the phase behaviour of the DDAB-water system has been studied. With the addition of NaBr and NaCl, the coexistence region of two lamellar phases found in the DDAB-water system shifts to lower surfactant concentrations. The concentration necessary to induce the collapsed lamellar phase at a given value of the surfactant concentration is found to be almost identical for all the salts used. This critical concentration was found to increase with decreasing DDAB concentration. Organic salts SHN and SS, in addition, form a cubic phase at much higher concentrations. This cubic phase is found to be of the bicontinuous type belonging to the space group $Pn3m$. The observation that similar concentrations of the different salts are required to induce the collapsed phase is in conflict with recent theoretical predictions. These studies suggest an alternative mechanism for the formation of the collapsed lamellar phase, which needs to be tested with additional experiments.

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