Properties of Water Solubilized in Reversed Micellar Systems of Dodecylammonium Propionate in Nonpolar Solvents

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The properties of the reversed micelles of dodecylammonium propionate(DAP) and of solubilized water were examined for systems with hexane, chloroform and benzene as a nonpolar component. The aggregation number of DAP, the conductivity, the heat of solution of water, IR and NMR spectra, and near infrared spectra of solubilized water were measured. From the results the properties of the reversed micellar system were analyzed, the following conclusion being obtained. The aggregation state of DAP is scarcely affected by the addition of water. Water is first dispersed in the nonpolar organic phase, and then solubilized in the cores of the reversed micelles. The former is free or weakly bonded water, while the latter is bonded water like that in the bulk phase. The catalytic effect of DAP reversed micellar system is discussed.

Surfactant micelle catalyzes various reactions and serves as a model for the enzyme action. Fendler *et al.* observed that reversed micelles in nonpolar solvents significantly catalyze several reactions, and suggested that this could provide a favorable model for a microenvironment of enzyme active sites.[1] They also suggested that the water solubilized in the polar cavities of reversed micelles is less polar than the water in the bulk phase, which is closely related to the catalytic effect of the reversed micellar systems.[2,3]

We investigated the hydrolysis of adenosine 5′-triphosphate (ATP) in a reversed micellar system where an aqueous ATP phase is solubilized in hexane by dodecylammonium propionate (DAP), and found that the hydrolysis rate in this system is enhanced several times as compared with that in an aqueous system.[4] This reversed micellar system shows an enzyme-like behavior such that the addition of Mg^{2+} or Ca^{2+} enhances markedly the hydrolysis rate, suggesting that the rate enhancement is closely related to the polarity of the water solubilized in the reversed micelles. It is of interest to investigate by more direct methods the properties of the DAP reversed micelles and the solubilized water in relation to the catalytic activities of these systems.

Experimental

Materials. The preparation and purification of dodecylammonium propionate (DAP) were described previously;[4] mp 55—56 °C (Lit, 54—56 °C[5]); Found (Calcd), C, 69.68(69.44); H, 12.38 (12.82); N, 5.32 (5.40)%. Dodecyltrimethylammonium acetate (DTAA) was prepared by the anion exchange reaction of dodecyltrimethylammonium chloride with silver acetate in a 1-propanol-water mixed solvent, and was purified and dried three times by freeze-drying from its benzene solution. The structure and purity were confirmed by IR and NMR spectra and elementary analysis; Found (Calcd): C, 70.24 (71.02); H, 13.10 (12.97), N, 4.75 (4.87)%. Distilled water with conductivity below 10^{-7} mho·cm^{-1}, was used.

Measurements. Conductivity of solutions was measured with a Toa conductmeter model CM-2A equipped with a CG-7001PL cell, the sensitivity being 10^{-7} mho·cm^{-1}. The sample solution was stirred and maintained at 35.0 °C. Vapor pressure osmometric measurements (VPO) were made at 37.0 and 50.0 °C with a Hewlett-Packard vapor pressure osmometer model 302B.

Infrared spectra were recorded with a Jasco IRA-2 spectrometer at room temperature using a pair of cells of 0.05 mm in path length. The reference cell was filled with solvent in order to record spectra of DAP micellar solutions. Near infrared spectra were recorded with a Hitachi spectrophotometer model EPS-3T. The absorption cells were 10 mm in light path and thermostatted at 35.0 °C. A solution containing DAP of the same concentration as that of the sample solution was used as a reference, in order to record the spectra of water solubilized in the reversed micellar system.

The 90 MHz proton nuclear magnetic resonance spectra were taken with a Hitachi R-22 high-resolution spectrometer at 35 °C. The chemical shifts were recorded by using TMS as an internal standard. A coaxial double tube was used for measurements of spectra of water in DTAA reversed micellar system; pure water was contained in the inner tube as an external standard, and the sample solution in the outer tube.

The heat of solution was measured with a Tokyo Riko twin-isothermal calorimeter at 35.0 °C. The water was sealed in a glass ampoule, and after attaining thermal equilibrium the ampoule was crushed into the sample solution. The temperature difference between the sample cell and reference cell was detected by a thermocouple, and calibrated against a standard electric heater.

Results and Discussion

Conductivity of the Systems. Conductivity of DAP reversed micellar systems with hexane, chloroform, and benzene as a nonpolar component was measured at 35.0 °C. The results are shown in Fig. 1 as a function of water concentration. A break point observed in each conductivity curve coincides with

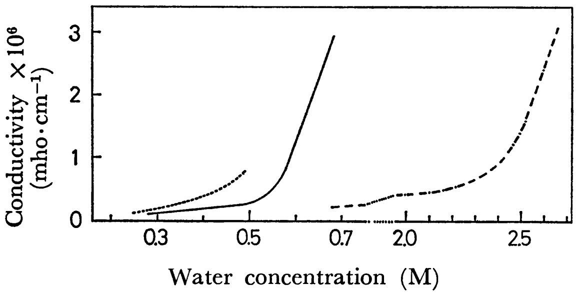

Fig. 1. Conductivities of the DAP (0.2 M) reversed micellar systems at 35.0 °C in hexane (——), chloroform (----), and benzene (-·-·).

the water concentration where a homogeneous solution becomes heterogeneous. At water concentrations below the break point this curve gives the conductivity of homogeneous DAP reversed micellar solution, the values lying within *ca.* 10^{-7}—10^{-6} mho·cm^{-1}. When an aqueous electrolyte solution (0.1 M sodium acetate) was used instead of pure water, the conductivity of the system remained nearly the same. It is thus evident that the conductivity in this concentration range is due to DAP in the system. Increase in water content scarcely affects the association state of DAP. However, the conductivity increases abruptly at water concentrations above the break point. By considering the large values of conductivity, this increase is thought to be caused by the dissociation of DAP salt in water phase which is not solubilized.

The amount of water that could be solubilized in each solution determined from the conductivity measurement is given in Table 1. The value for the benzene solution is almost the same as that reported by Kitahara[5] and Kon-no and Kitahara.[6]

Aggregation Numbers of DAP. Aggregation numbers of DAP in the reversed micellar systems were estimated from the average molecular weights of micelles determined by VPO. In these measurements, it was assumed that the vaporization of water solubilized in DAP reversed micelles is neglected in the range where the amount of water is comparable to that of DAP. The results are shown as a function of water concentration (Fig. 2). The values obtained are nearly the same as those determined by Seoud *et al.* through the NMR method for the systems in benzene, chloroform and carbon tetrachloride in the absence of water.[7] The aggregation number of DAP in the reversed micellar system is much smaller than that in aqueous micelles, the amount of water added scarcely affecting the value. This would mean that the addition of water does not alter significantly the aggregation state of DAP.

Table 1. Solubility of water in DAP (0.2 M) reversed micellar solutions (5 ml) at 35.0 °C

	Water	
	Amount (μl)	Concentration (M)
Hexane	54.1	0.599
Chloroform	42.5	0.472
Benzene	230	2.55

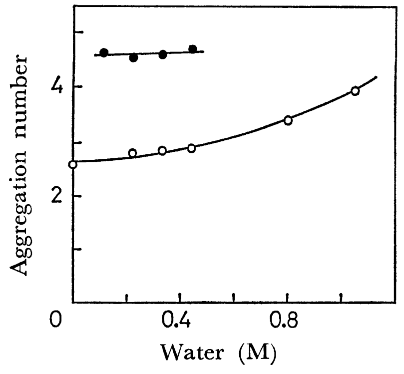

Fig. 2. Aggregation numbers of DAP (0.2 M) in hexane at 37.0 °C (-●-), and in benzene at 50.0 °C (-O-).

Infrared Spectra and Near Infrared Spectra. The effect of water concentration on IR spectra of DAP in the reversed micellar system was examined. It was found that the absorption band of asymmetric stretching of carboxylate anion shifts to a lower wave number side with an increase in water concentration (Table 2). This would indicate that the solubilized water interacts with the carboxylate group of DAP. No further information could be obtained as to the ammonium group of DAP and water, since the absorption bands of these groups are broad.

The near infrared spectra of water in various systems were recorded in the range 1800—2000 nm (5000—5556 cm^{-1}), where water exhibits a strong absorption band due to the $\nu_2+\nu_3$ combination mode. The spectra of the systems with various solvents and various concentrations of water and DAP are shown in Figs. 3, 4, and 5. The water solubilized in the reversed micellar systems in chloroform and benzene has a sharp band at about 1890 nm and a broad band at 1920—1950 nm, while in hexane the former band is observed as a shoulder of the broad band. These bands could be separated into two parts on figures by assuming that the sharp band is of a Gaussian type, and the areas of

Table 2. IR spectra of carboxylate anion of DAP in reversed micellar systems

Water concentration (M)	Wave number (cm^{-1})	
	In chloroform	In hexane
0	1569	—
0.111	1565	1566
0.167	1560	—
0.222	1554	1562
0.278	1553	—
0.333	1551	1560
0.389	1551	—
0.444	—	1557

DAP: 0.2 M.

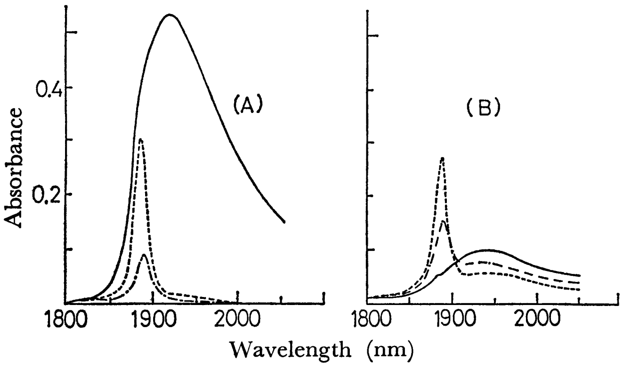

Fig. 3. Near infrared spectra of water at 35.0 °C: (A) water in the following solvents in the absence of DAP; water (——), chloroform (----), and benzene (-·-·); (B) water solubilized in the DAP (0.2 M) reversed micellar systems in hexane (——), chloroform (----), and benzene (-·-·).

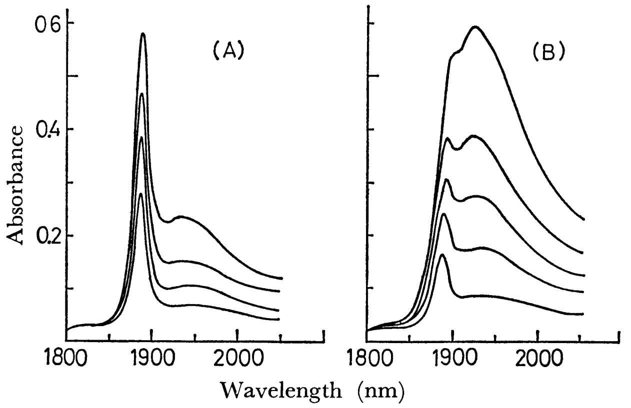

Fig. 4. Effect of water concentration on the near infrared spectra of solubilized water at 35.0 °C in chloroform (A), and benzene (B). DAP concentration is kept constant (0.2 M), and water concentration is varied as follows; (A): 0.111 M, 0.167 M 0.278 M, and 0.389 M; (B): 0.111 M, 0.222 M, 0.333 M, 0.444 M, and 0.666 M.

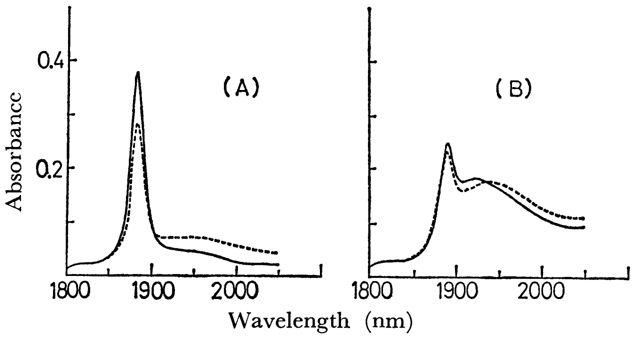

Fig. 5. Effect of DAP concentration on the near infrared spectra of solubilized water at 35.0 °C in chloroform (A), and benzene (B). Water concentration is kept constant ((A): 0.111 M, (B): 0.167 M), and DAP concentration is varied as follows; (A): 0.05 M (——), and 0.2 M (----); (B): 0.05 M (——), and 0.2 M (----).

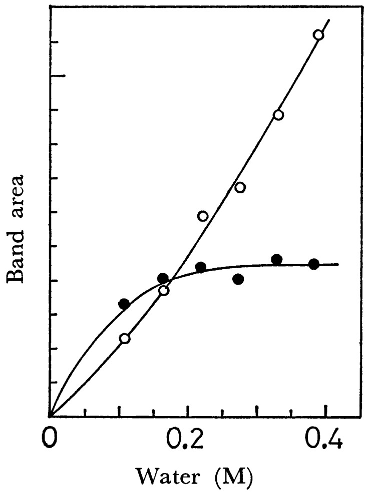

Fig. 6. Band areas of near infrared spectra of water in chloroform at 35.0 °C; one is the sharp band at 1890 nm (●); and the other is the broad band above 1900 nm (○).

these two parts is given in Fig. 6 as a function of water concentration in chloroform. The sharp band at 1890—1900 nm is ascribed to free or extremely weakly bonded water.[8] This assignment suggests that in the present system the sharp band at about 1890 nm is not due to the water solubilized in the polar region of reversed micelles, but to the water dispersed in the nonpolar organic phase. The interpretation is based as follows: 1) This band coincides in position and shape with that of water dissolved in pure solvents in the absence of surfactants (Fig. 3); 2) The increase in DAP concentration does not increase the intensity of this band (Fig. 5); 3) The increasing order of the intensity of this band, chloroform>benzene≫hexane, agrees with that of the solubility of water in these solvents.

The band occurring at wave lengths greater than 1900 nm is thought to be due to the bonded water, usually hydrogen bonded water.[8] The broad band observed at 1920—1950 nm in Fig. 3 is thus ascribed to the water solubilized in the reversed micelles because of the following reasons: 1) The band resembles that of pure water in shape, and water solubilized in micelles could be supposed to form a water phase, while it is hard to suppose that water dispersed in the organic phase forms a water phase; 2) The increase in DAP concentration increases the intensity of this band (Fig. 5); 3) The increase in the amount of water beyond the solubility of water in the organic solvent results exclusively in the increase in intensity of this absorption band. Furthermore, the position of this band ($\lambda_{\max}$: 1921—1953 nm) shifts a little to a greater wavelength side as compared with that of pure water ($\lambda_{\max}$: 1919 nm), the increase in water concentration diminishing the extent of this shift. The greater wavelength shift could be caused by the interaction between the water solubilized in the reversed micelles and polar groups of DAP.

These results could be summarized as follows: 1) Two types of water exist in the reversed micellar solution; one is the water dispersed in the organic phase which shows a sharp band at about 1890 nm, and the other is the water solubilized in the reversed micelles which shows a broad band at 1920—1950 nm. 2) In chloroform a considerable amount of water is dispersed in the organic phase, while in hexane almost all the water is solubilized in the reversed micelles. 3) As seen from Fig. 6, water added is at first dispersed in the organic phase and then solubilized in the reversed micelles. 4) The water solubilized in the reversed micelles is bonded to the head groups of DAP or to each other a little more strongly as compared with the case of pure water.

NMR Spectra. The effect of water concentration on chemical shifts of DAP protons in the reversed micellar system in deuterochloroform at 35 °C is shown in Table 3. As the ammonium protons of DAP and water protons gave only a single peak because of the rapid proton exchange between them, we could not observe the dependence of the chemical shift of ammonium protons on water concentration. The dependence of the chemical shift of these protons on solubilizate

Table 3. Chemical shifts, $\delta_{(\mathrm{PPM})}$, of DAP in deuterochloroform at 35 °C

	Water concentration (M)					in D_2O
	0	0.07	0.14	0.28	0.30	
$CH_3\text{-}\underline{(CH_2)_{10}}\text{-}CH_2\overset{+}{N}H_3$	1.23	1.24	1.24	1.24	1.24	1.26
$CH_3(CH_2)_{10}\text{-}\underline{CH_2}\text{-}\overset{+}{N}H_3$	2.73	2.74	2.76	2.77	2.78	2.89
$CH_3\text{-}\underline{CH_2}\text{-}COO^-$	2.16	2.15	2.15	2.15	2.15	2.14
$C_{12}H_{25}\text{-}\underline{\overset{+}{N}H_3+H_2O}$	7.97	6.94	6.54	5.99	5.97	—

DAP: 0.2 M.

Table 4. Chemical shifts, $\delta_{(\mathrm{PPM})}$, of DTAA in deuterochloroform at 35 °C

	Water concentration (M)			
	0	0.12	0.24	0.36
$\underline{CH_3}\text{-}(CH_2)_{11}\overset{+}{N}(CH_3)_3$	0.86	0.86	0.87	0.87
$CH_3\text{-}\underline{(CH_2)_{10}}\text{-}CH_2\overset{+}{N}(CH_3)_3$	1.24	1.24	1.24	1.24
$CH_3(CH_2)_{11}\overset{+}{N}\text{-}\underline{(CH_3)_3}$	3.36	3.31	3.26	3.23
$\underline{CH_3}\text{-}COO^-$	1.88	1.87	1.85	1.83
$\underline{H_2O}$	—	4.36	4.57	4.73

DTAA: 0.2 M.

concentration is reported for other solubilizates such as dimethyl sulfoxide or methanol.[9] We have carried out a similar measurement on the reversed micellar system of DTAA which has no ammonium protons exchangeable with water protons. The result is given in Table 4. Both the NMR spectra of DAP and DTAA in the reversed micellar system show a similar feature; the extent of chemical shifts of methylene and methyl protons adjacent to the cationic or anionic group depends somewhat on water concentration, while those of the other protons are not dependent on water content. A similar behavior was observed for other solubilizates.[9] This indicates that the water solubilized in reversed micelles exists near the polar head groups and, in other words, in the cores of reversed micelles.

The water protons give only a sharp single peak, and not a broad one or two peaks as in the case of near infrared spectra. This shows that the lifetime of water of each type is below the NMR time scale. The δ value of water protons in the DTAA reversed micellar system is smaller than that of pure water (δ=4.66) at lower water concentration, but a little greater than that of pure water at higher water concentration. Water dissolved in deuterochloroform without any surfactant gives a peak at δ=1.54. The observed peak position is said to be an average of the two peak positions, one of which is due to the water dispersed in the nonpolar organic phase which gives a high-field shift as compared with that of pure water, and the other to the water solubilized in the core of reversed micelles which gives a small low-field shift.

Calorimetric Measurements. The heat of solution of water in a DAP (0.05 M) reversed micellar system in hexane (100 ml) at 35.0 °C was measured. The results given as an integral heat of solution, ΔH_{soln} (cal/100 ml of hexane), and a differential heat of solution, Δh_{soln} (kcal/mol of water), are shown in Fig. 7. Since the addition of water does not cause a pronounced change in the state of micelles, the values show the enthalpy change between pure water and the water solubilized in the reversed micellar system. The initial step of addition of water is endothermic, while the subsequent steps are exothermic. By the addition of more than 0.03 M water, the differential heat of solution becomes nearly constant, $\Delta h_{\mathrm{soln}}=-450$ cal/mol of water. When water is dispersed in the nonpolar organic phase, the enthalpy change is thought to be positive because of destruction of water structure in the bulk phase. On the other hand, when water is solubilized in the reversed micelles the enthalpy change is thought to be negative, since water interacts with the polar groups of DAP as elucidated from IR and NMR spectra. Thus the calorimetric measurement provides results which coincide with other results.

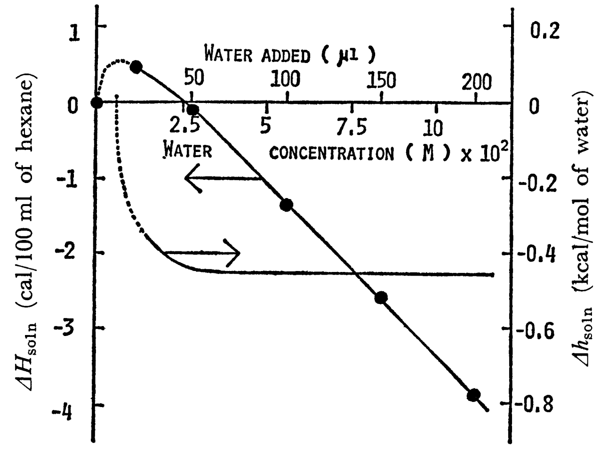

Fig. 7. Integral (ΔH_{soln}) and differential (Δh_{soln}) heats of solution of water in DAP (0.05 M) reversed micellar system in hexane (100 ml) at 35.0 °C.

Catalytic Effect of the DAP Reversed Micellar System. The hydrolysis rate of ATP in the DAP reversed micellar system in hexane is proportional to the DAP concentration and inversely proportional to the concentration of solubilized water.[4] The polarity of the core of DAP micelles in hexane, which was measured by the absorption band shift of pyridine 1-oxide solubilized in the reversed micelles, depends on the [water]/[DAP] ratio, and is less polar than pure water. The catalytic effect of this system would be closely related to this polarity.

The effect of water content on this system could be discussed from the following three related aspects: (a) the effect on the aggregation state of DAP, (b) the effect on the property of solubilized water, and (c) the effect on the substrate in the reversed micelles. In this paper we have discussed the system from (a) and (b), and in the preceding paper from (c).[4] Since the water content scarcely affects the aggregation states of reversed micelles, the micro-environment of the substrate is closely related to the property of water solubilized in the reversed micellar system. As almost all portions of the water added exist in the cores of DAP reversed micelles in hexane, it could be said that the catalytic effect of this system depends greatly on the property of the water solubilized in the reversed micelles, which is bonded a little more strongly than the water in the bulk phase.

References

1) E. J. Fendler, S. A. Chang, J. H. Fendler, R. T. Medary, O. A. El Seoud, and V. A. Woods, "Reaction Kinetics in Micelles," ed. by E. H. Cordes, Plenum Press, New York, N. Y. (1973), p. 127.

2) J. H. Fendler, F. Nome, and H. C. Van Woert, *J. Am. Chem. Soc.*, **96**, 6745 (1974).

3) J. H. Fendler, and L. Liu, *J. Am. Chem. Soc.*, **97**, 999 (1975).

4) M. Senō, S. Shiraishi, K. Araki, and H. Kise, *Bull. Chem. Soc. Jpn.*, **48**, 3678 (1975).

5) A. Kitahara, *Bull. Chem. Soc. Jpn.*, **28**, 234 (1955).

6) K. Kon-no and A. Kitahara, *J. Colloid Interface Sci.*, **33**, 124 (1970).

7) O. A. El Seoud, E. J. Fendler, J. H. Fendler, and R. T. Medary, *J. Phys. Chem.*, **77**, 1876 (1973).

8) O. D. Bonner, and Y. S. Choi, *J. Phys. Chem.*, **78**, 1723 (1974).

9) O. A. El Seoud, E. J. Fendler, and J. H. Fendler, *J. Chem. Soc.*, *Faraday I*, **70**, 450 (1974).