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Enhanced Energy Transfer between Chlorophyll b and Chlorophyll a in the Premicellar Region of Aqueous Surfactant Solution

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Energy transfer between chlorophyll b and chlorophyll a was enhanced irrespective of the kind of surfactants in the premicellar region below the critical micelle concentration. This effect can be explained in terms of the formation of premicellar aggregates induced by the non-ionic hydrophobic pigments.

In view of the wide interests in their properties and usages in such areas as photochemistry, photobiology, photoelectrochemistry, etc.,1) it is important to understand the characteristic aspects of the photophysical and photochemical behavior of chlorophyll a (Chl a) and its related compounds in organized assemblies such as surfactant micelles. Although a number of studies have been performed for surfactant systems containing such pigments as Chl a, most of them were concerned with the surfactant concentrations above the critical micelle concentration (CMC). We have paid special attention to the absorption, fluorescence, and Raman spectroscopic behavior of Chl a at concentrations not only above but below CMC in solutions of typical surfactants such as sodium dodecyl sulfate (SDS).2-6) In these systems, the formation of solute-rich induced micelle (SRIM) was strongly suggested at surfactant concentrations below CMC (i.e., in the premicellar region). It has been known that SRIM is formed between solute and surfactant ions of opposite charge.7) However, there have been few reports to suggest the formation of premicellar aggregates consisting of the non-ionic hydrophobic solutes.3,5,6,8) For example, the formation of Chl a-induced premicelles was suggested by the enhanced fluorescence quenching and the fluorescence decay behavior of the Chl a-methylviologen system in the premicellar region of aqueous surfactant solutions.5)

Chl b is known to sensitize the fluorescence of coexisting Chl a in homogeneous and heterogeneous systems. It can be expected, therefore, that the formation of Chl-rich induced micelle (composed of Chl a, Chl b, and surfactant) may enhance energy transfer between Chl b and Chl a even at concentrations substantially below the CMC of surfactant used. This new type of SRIM is also anticipated to be formed irrespective of the kind of surfactant head group. The present paper deals with enhanced energy transfer of the system which contains Chl b as a donor and Chl a as an acceptor in aqueous buffer solutions of three typical surfactants. A possible explanation based on the formation of SRIM is proposed for the observed enhancement of energy transfer in the premicellar region of surfactant solutions.

Commercially available Chl a (Wako, >99%) and Chl b (Sensyu Scientific, >99.7 %) were used. SDS and cetyltrimethylammonium chloride (CTAC) were purified by recrystallization from ethanol. Triton X-100 (TX-100) was used without further purification. The sample solutions were prepared by injecting an aliquot of each alcoholic solution of Chl a and Chl b to an aqueous buffer solution of surfactant. In order to avoid the degradation such as pheophytinization and allomerization of the pigments in surfactant media, the sample solutions containing TX-100, CTAC, and SDS were buffered at pH 7.0 (phosphate), 7.0 (phosphate), and 9.5 (TAPS$^{2)}$), respectively. Both concentrations of Chl a and Chl b were fixed at 4×10^{-6} mol dm^{-3} throughout the whole experiments. The CMCs of TX-100, CTAC, and SDS in buffered solutions of Chl a and/or Chl b were determined conductometrically and tensiometrically to be 0.2, 0.1, and 3.7 mmol dm^{-3}, respectively. The sample contained in a 1-mm quartz cell was excited with 463-nm light. This excitation wavelength was selected to excite Chl b and to minimize the direct excitation of Chl a. In order to avoid the effect of reabsorption and reemission of fluorescence, absorbance at the excitation wavelength was kept always below 0.05 and front emission was collected. All measurements were made at 25 °C for aerated solutions after an approximately 15-min equilibration period.

In order to determine the efficiency of energy transfer as a function of surfactant concentration ([surfactant]), we have measured the fluorescence spectra of each pigment solution and of equimolar mixed solutions of Chl a and Chl b at a fixed pigment concentration of 4×10^{-6} mol dm^{-3} in the presence of surfactant. Energy transfer is accompanied by quenching of the donor fluorescence, and by appearance of sensitized fluorescence of the acceptor. Therefore, energy transfer efficiency can be described by the quantity, e_{ET}, defined by$^{9,10)}$

$$e_{ET} = \frac{(I_A/I_D)(q_D/q_A)}{1 + (I_A/I_D)(q_D/q_A)} \qquad (1)$$

where I_D and I_A are the fluorescence intensities of the donor and acceptor observed in the mixed solution, and q_D and q_A are the relative fluorescence efficiencies of the donor and acceptor, respectively. We assumed $q_D/q_A = 0.36$.$^{11)}$ The results are shown in Fig. 1 as a function of [surfactant] for the three surfactant systems, where the results of independent measurements on at least three samples are averaged for each value of e_{ET} and e_{EL} (vide infra). The efficiency of energy transfer can also be described by the "energy loss," the fractional loss of donor fluorescence, defined by

$$e_{EL} = (I_0 - I_D)/I_0 \qquad (2)$$

where I_0 is the fluorescence intensity of the donor (Chl b)-surfactant solution which has the same concentration of Chl b as the Chl b-Chl a-surfactant solution in question. The estimated values of e_{EL} are also shown in Fig. 1. We can see in Fig. 1 that both maxima in e_{ET} and e_{EL} appear for [TX-100] and [CTAC] close to CMC while those for [SDS] below CMC. It should be noted that efficient energy transfer occurs even at TX-100 and CTAC concentrations below CMC. At lower surfactant concentrations than CMC, the enhanced energy transfer in such a dilute pigment

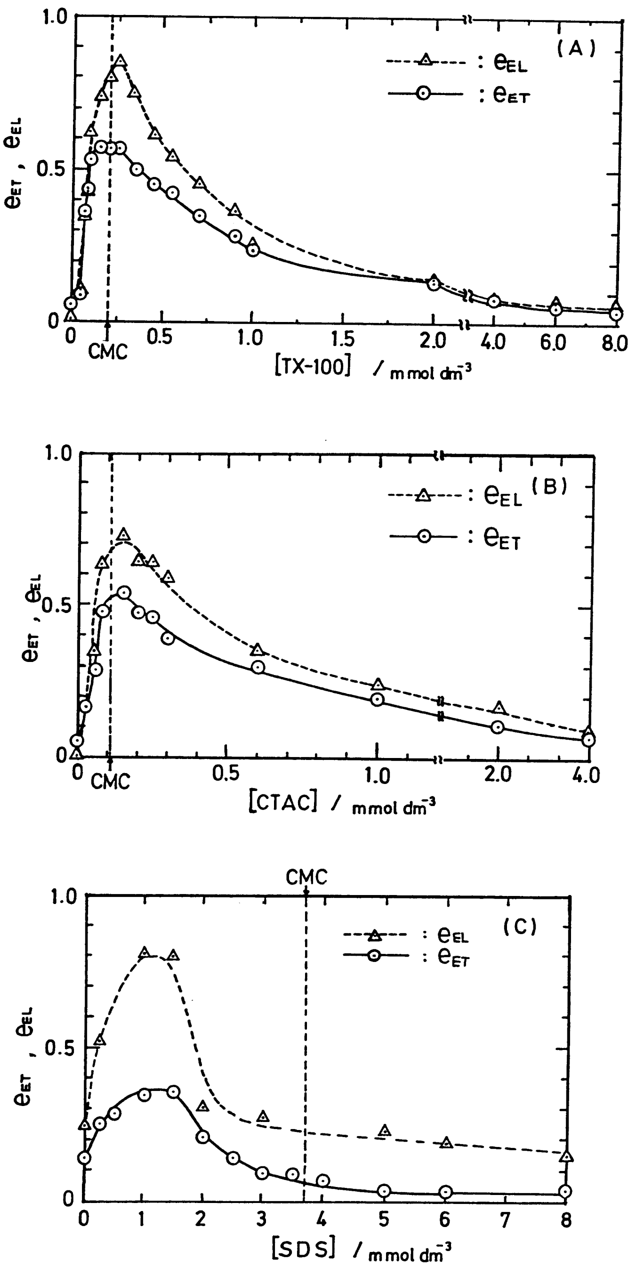

Fig. 1. Plots of e_{ET} and e_{EL} versus [surfactant]: A) TX-100, B) CTAC, C) SDS. The vertical broken lines refer to the critical micelle concentrations.

concentration as 4 X 10^{-6} mol dm^{-3} cannot be expected if the donor and acceptor molecules are uniformly distributed in the homogeneous solution.

It is known that charged solutes form water-insoluble complex salts with oppositely charged surfactants even at concentrations far below CMC.$^{7)}$ Furthermore, it has been postulated that the resulting water-insoluble salt induces the formation of solute-rich premicellar aggregates in the premicellar region just below CMC.$^{7)}$ In the present systems, therefore, it is also reasonable to assume the formation of Chl (Chl a and Chl b)-induced premicellar aggregates, because Chl is insoluble in water.$^{12)}$ In such an aggregate as SRIM, the donor-acceptor separations will become much smaller than that expected from the mean concentration and result necessarily in the enhanced energy transfer. Apart from the structural details of SRIM, the hydrophobic interaction among Chl a, Chl b, and surfactant molecules is supposed to play an important role in the formation of SRIM, since Chl a and Chl b are non-ionic but have long hydrophobic chains.

As is shown in Fig. 1, the maximum value of e_{ET} (e_{ET}^{max}) in the SDS system is smaller than those in the TX-100 and CTAC systems. It is known that hydrophobic colloids of Chl are negatively charged in aqueous solution.$^{13)}$ Thus, the observed decrease in e_{ET}^{max} in the SDS system will be ascribed partly to the depression of the formation of SRIM arising from the electrostatic repulsion between Chl and surfactant molecules.

It is easy to show that the quantity, e_{EL} - e_{ET}, approximately corresponds to e_q,$^{7,9)}$ the efficiency of energy transfer to the weakly- or nonfluorescent Chl aggregates (i.e., quenchers) which quench the fluorescence of the Chl-b monomer in competition with the Chl-a monomer. Our data (Fig. 1) show the existence of the quencher(s) in all systems studied, as seen in cases of dye-surfactant solutions.$^{9)}$

The efficiency of energy transfer, corresponding to e_{ET}, in acetone solution containing equal concentrations of Chl a and Chl b is known to have a maximum value of about 0.4-0.5 at a concentration of about 1 X 10^{-3} mol dm^{-3}.[14] On the basis of an assumption that e_{ET}^{max}(acetone) = e_{ET}^{max}(surfactant), we can roughly estimate an effective local concentration of the pigment in SRIM of 1 X 10^{-3} mol dm^{-3} which is larger two orders of magnitude than the analytical concentration (4 X 10^{-6} mol dm^{-3}). This implies that the pigment molecules are highly concentrated in SRIMs to give rise to the enhanced energy transfer.

Our systems thus indicate the enhancement in energy transfer between the non-ionic donor and acceptor pigments in the premicellar region of typical non-ionic and ionic surfactant solutions. It has been well-established that a conventional SRIM is formed only between solute and surfactant ions of opposite charge.[7] Our data conclusively show that SRIMs are formed irrespective of the type of surfactants even for the non-ionic hydrophobic pigments, though the mechanism of the enhanced energy transfer and the structural aspects of SRIM remain unsolved.

References

1) "The Chlorophylls," ed by L. P. Vernon and G. R. Seely, Academic Press, New York (1966); A. Harriman, "Photogeneration of Hydrogen," Academic Press, New York (1982); "Energy Resources through Photochemistry and Catalysis," ed by M. Grätzel, Academic Press, New York (1983); M. Lutz, "Resonance Raman Studies in Photosynthesis," in "Advances in Infrared and Raman Spectroscopy," ed by R. J. H. Clark and R. E. Hester, Wiley Heyden, London (1984), Vol. 11, Chap. 5, pp. 211-300; K. Kalyanasundaram, "Photochemistry in Microheterogeneous Systems," Academic Press, New York (1987).

2) Y. Kusumoto, J. Watanabe, and J. Kurawaki, Chem. Express, 2, 89 (1987).

3) Y. Kusumoto, J. Watanabe, and J. Kurawaki, Chem. Express, 2, 133 (1987).

4) Y. Kusumoto, J. Watanabe, J. Kurawaki, and I. Satake, Chem. Lett., 1987, 1417.

5) Y. Kusumoto, J. Watanabe, and J. Kurawaki, Chem. Express, 4, 153 (1989).

6) J. Kurawaki and Y. Kusumoto, Chem. Phys. Lett., 158, 495 (1989).

7) For a review on SRIM, see: Y. Kusumoto, Bunko Kenkyu, 37, 96 (1988).

8) K. Y. Law, Photochem. Photobiol., 33, 799 (1981).

9) H. Sato, M. Kawasaki, and K. Kasatani, J. Phys. Chem., 87, 3759 (1983) and the references cited therein.

10) A detailed spectrofluorometric method for determining the efficiency of energy transfer will be presented elsewhere.

11) P. Latimer, T. T. Bannister, and E. Rabinowitch, Science, 124, 585 (1956); G. Weber and F. W. J. Teale, Trans. Faraday Soc., 53, 646 (1957); D. Mauzerall, Isr. J. Chem., 21, 321 (1981).

12) Both Chl a and Chl b in aqueous solution of 4 X 10^{-6} mol dm^{-3} are considered to be in a colloidal state (see Ref. 13).

13) H. A. Bakker, Proc. R. Acad. Amsterdam, 37, 688 (1934).

14) W. F. Watson and R. Livingston, J. Chem. Phys., 18, 802 (1950); L. N. M. Duysens, Nature (London), 168, 548 (1951).

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