Singlet Oxygen Production Photosensitized by Fluorescein in Reversed Micellar Solutions

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The quantum yield of singlet oxygen production was investigated in dodecylammonium propionate reversed micellar solutions containing 1,3-diphenylisobenzofuran and fluorescein as photosensitizer. The quantum yield was highly enhanced by the binding of fluorescein to reversed micelles, and increased with decreasing the solubilized water content in reversed micelles. Results obtained was discussed with the optical properties of fluorescein (absorption, fluorescence and phosphorescence) in reversed micellar solutions.

Introduction

Recently, chemical reactions in micellar systems attract increasingly various worker’s attention from the interest of bio-mimic chemistry. The DF oxidation by singlet oxygen produced by the photosensitization of pyrene has been studied by us in various micellar solutions [1–3]. The DAP reversed micellar solutions provide a hydrophilic environment in the interior of micelles in non-polar phase. Therefore, various interesting reaction systems can be constructed in these solutions different from aqueous micellar ones. It is known that the micellar size depends on the solubilized water content [4]. At the stoichiometric H2O concentrations below 0.37 M, all the water molecules are bound to the surfactant head group (bound water range). With increasing the stoichiometric H2O concentration above 0.37 M, the average aggregation number and the number of free water molecules entrapped in a reversed micelle increased rapidly (free water range) [4].

We found that F acted as considerably good photosensitizer for singlet oxygen production in DAP reversed micellar cyclohexanic solutions and the quantum yield for singlet oxygen production was highly enhanced by DAP reversed micelles depending on the solubilized water content.

In the present investigation, the DF photo-oxidation in DAP reversed micellar solutions containing F and DF, and the optical properties (absorption, fluorescence and phosphorescence) of F bound to DAP reversed micelles were dealt with changing the solubilized water content, and the information about the mechanism of the enhancement of the quantum yield for singlet oxygen production by DAP reversed micelles was sought for.

Materials and Methods

Cyclohexane, KCr(NH3)4(NCS)4 (Raineck’s salt), DAP and F purchased from Katayama Co. were guaranteed or extra pure reagents. DF obtained from Aldrich Chemical Co. was of special grade. Laboratory-distilled water was further distilled from alkaline KMnO4 solutions.

Reversed micellar solution was prepared by dissolving DAP in cyclohexanic solution containing DF at room temperature. The DAP cyclohexanic solution was stirred for ten minutes in the dark at 40 °C after adding powdered F or aqueous F solutions at various water concentrations.

The concentration of F and DF in reversed micellar solutions were determined by absorbances at 500 and 415 nm, respectively. The absorbances were measured with a Hitachi spectrophotometer type 356. The reaction mixture in a quartz vessel (1 x 1 x 4 cm3) was irradiated with the yellow light isolated from a 150 W xenon lamp through a cut filter (colour glass filter, type V–Y 48, Toshiba Electric Co.) at 40 °C.

The number of photons absorbed by F was determined by the chemical actinometry method (sensitive to 504 nm light) of Wegner and Adamson [5]. The actinometer cell containing a 5.0 · 10−3 M KCr(NH3)4(NCS)4 aqueous solution at pH 5.3 was placed just behind the target cell, and was illuminated for 20 min in the presence and absence of the target cell.

The fluorescence spectrum of F at room temperature was measured with a Hitachi type 203 spectrofluorimeter.

A glass tube with a diameter of 5 mm and a length of 20 cm was employed for the fluorescence
and phosphorescence measurements at 77 K. Anaerobic solutions were prepared by the freeze-pump-thaw cycles.

The phosphorescence spectrum and intensity of F were measured by the phosphoroscope method. The excitation light was provided from a 150 W tungsten lamp through an interference filter (peak, 479 nm; halfwidth, 16 nm). The phosphoroscope (rotating cylinder type) was used for separating the phosphorescence from the fluorescence. A cylinder rotated at the constant rate of 3.6 · 10³ rpm. The observation of the emission was delayed by 0.6 msec after excitation. The durations of the observation and excitation were designed to be equally 2 msec per one rotation of the cylinder. The phosphorescence intensity was detected with a photomultiplier (RCA 1C31034A) through a Nikon P-250 type monochromator, and its signal was fed to a homemade lock-in-amplifier. The phosphorescence decay was measured with a wave memory (NF model E-5001) and an oscilloscope (Hewlett Packard model 1725 A).

**Results and Discussion**

When the DAP cyclohexanic solution (air-bubbled) containing F and DF was irradiated with the yellow light (> 480 nm) absorbed by F, the DF concentration exhibited an initial, linear decrease with the irradiation time followed by a saturation.

The decrease in DF concentration was caused by the oxidation of DF by singlet oxygen produced by the F photosensitization. DF is known to be highly reactive with singlet oxygen.

The initial rate (R) of the DF oxidation increased linearly with increasing DF concentration in low DF concentration range as shown in Fig. 1. The initial slope in the curve of R vs DF concentration increased with decreasing the solubilized H₂O content.

Simplified diagram for the photochemical reaction in DAP reversed micellar solutions containing F and DF is shown in Fig. 2, assuming that singlet oxygen is consumed by the DF oxidation, the quenching by empty micelles and the physical decay to triplet oxygen. H₂O and F are bound to the interior of micelles (F, hydrophylic), and DF may be dispersed in cyclohexanic phase (DF, hydrophobic). The quantum yield (Φ₀₂) for the DF oxidation is represented as follows.

\[
\Phi_{DF} = \Phi O_2 \left( \frac{k_r [DF]}{k_d + k_r [DF] + k_q [M]} \right),
\]

where Φ₀₂, k₁, k₂ and k₃ are illustrated in the legend of Fig. 2, and [M] is the empty micellar concentration. The k₃ values were obtained by us at various H₂O concentrations in the previous paper [3], and the k₃[M] value was estimated to be 3.0 · 10⁴ to 5.1 · 10⁴ sec⁻¹ for the H₂O concentrations of 0 to 0.6 M. We assumed that one micelle bound one F molecule. The F concentration (3.6 · 10⁻⁶ M) used was much less than that of reversed micelles (order of 10⁻³ M).

The micellar concentration was determined using the aggregation number obtained by Correll et al. [4]. Since the k₁ and k₃ values were 5.3 · 10⁸ M⁻¹ · sec⁻¹ and 5.8 · 10⁴ sec⁻¹ [3], respectively, k₁[DF] (< 5.3 · 10³ sec⁻¹) could be neglected compared with k₂ + k₃[M] in eq. (1) in the DF concentration range below 1.0 · 10⁻⁵ M. Then, Φ₀₂ is proportional to the DF concentration, and the slope is represented as follows.

![Fig. 2. Simplified diagram for the photochemical reaction in DAP reversed micellar solutions containing F and DF.](image-url)
The experimental values of $\Phi_{DF}$ also exhibited a linear relation with the DF concentration below $1.0 \cdot 10^{-5}$ M. Accordingly, the quantum yield of singlet oxygen production ($\Phi^{1}O_{2}$) could be calculated at various $H_2O$ concentrations from eq. (2). The results obtained were shown in Fig. 3.

The quantum yield ($\Phi^{1}O_{2}$) decreased rapidly with increasing the $H_2O$ concentration below about 0.4 M $H_2O$ (bound water range) and showed a gradual decrease above this $H_2O$ concentration (free water range). The $\Phi^{1}O_{2}$ value at 0 M $H_2O$ was about 0.15. This value was 1.5 times higher than that at 0.6 M $H_2O$.

Usui [6] described that the quantum yield of singlet oxygen production by F photosensitization was 0.06 in aqueous solutions. The value obtained by Gollnick and Schenck [7] was 0.03 in water. It is notable that $\Phi^{1}O_{2}$ was highly enhanced compared with that in water phase and strongly dependent on the solubilized $H_2O$ content, when F was introduced into DAP reversed micelles.

To seek for the information about the effects of DAP micelles and solubilized $H_2O$ on $\Phi^{1}O_{2}$, the following experiments were carried out. The absorption and fluorescence spectra of F were measured in DAP reversed micellar solutions (Fig. 4). At room temperature, the absorption band at 490 nm in bulk aqueous F solutions red-shifted in DAP reversed micellar solutions. The band positions at 0 (Curve a) and 0.6 M $H_2O$ appeared at 500 nm and 499 nm, respectively. The red-shift of the absorption band in DAP micellar solutions may be caused by the binding interaction between F and DAP micelle.

The fluorescence band appeared at 526 and 525 nm in DAP reversed micellar solutions containing 0 M $H_2O$ at room temperature; Curve 1, fluorescence spectrum of F solubilized in DAP micellar solutions containing 0 M $H_2O$ at room temperature; Curve 2, fluorescence spectrum of F solubilized in DAP micellar solutions containing 0 M $H_2O$ at 77 K; Curve a, absorption spectrum of F solubilized in DAP micellar solutions containing 0 M $H_2O$ at room temperature.

The fluorescence intensity at room temperature are plotted against the $H_2O$ content in Fig. 5. The fluorescence intensity decreased slightly with increasing the $H_2O$ content, but it was highly quenched compared with that in bulk aqueous solutions. The phosphorescence could not be measured at room temperature.

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Fig. 6. Phosphorescence spectra of F at 77 K. Concentrations of F and DAP, 1.0 \times 10^{-5} and 8.0 \times 10^{-2} M, respectively; H_2O concentrations for Curves 1, 2, 3 and 4, 0, 0.2, 0.6 M in anaerobic samples and 0 M in O_2-bubbled sample, respectively.

Fig. 6 shows the phosphorescence spectra of F at various H_2O concentrations at 77 K. The phosphorescence band blue-shifted from 640 to 630 nm with the increase of the H_2O content from 0 to 0.6 M. The phosphorescence intensity depended on the H_2O content, and it decreased with increasing the H_2O content. Furthermore, the phosphorescence intensity was highly quenched by O_2. The fluorescence and phosphorescence intensities are shown in Fig. 7.

Further, the phosphorescence was highly enhanced in anaerobic micellar solutions compared with that in anaerobic water solutions as seen in Fig. 7. The fluorescence was not quenched by O_2.

As seen in Fig. 4, F bound to the interior of DAP reversed micelle had the absorption and fluorescence bands at longer wavelengths than those of free F. The fluorescence intensity was highly quenched by binding of F to micelle (Fig. 5). Moreover, the absorption and fluorescence band positions, and the fluorescence intensity changed by the solubilized H_2O content (Figs. 4 and 5). These indicate that the electronic state of F is highly affected by the adsorptive interaction between F and DAP micelle. Singlet oxygen is considered to be produced through the transfer of the triplet energy of F to DAP micelle. The lifetime of the triplet state of F has been obtained as 20 msec by Lidqvist using the T-T absorption method \[8\] in bulk aqueous solutions at room temperature. The lifetime of the phosphorescent state obtained by us in DAP micellar solutions at 77 K was about 450 msec at the H_2O content of 0\textendash0.6 M. We could not determine the lifetime of the F triplet state at room temperature owing to extreme weakness of the phosphorescence, but the lifetime of the F triplet state might be highly lengthened inside DAP micelle compared with that of free F. This is also another important factor responsible for the high value of \(\Phi^{O_2}\) in DAP micellar solutions.

At present, we can not go into further mechanism for the effects of DAP micelle and solubilized water on \(\Phi^{O_2}\). We must make clear up the structure of
DAP micelle depending on the solubilized water, the binding site and electronic state of F in DAP micelle, and the polarity and viscosity inside DAP micelle in future.