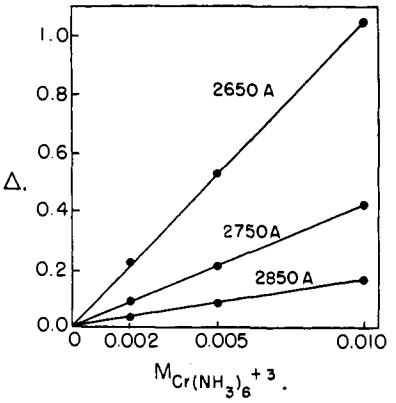

Fig. 4.—The interaction of 1.0 M SCN^- with various concentrations of $Cr(NH_3)_6^{+3}$ at 3°.

studied, only N_3^- shows a similar entropy loss on association.[8] The unique characteristic of N_3^- and SCN^- is that each is a linear ion which might possess rotational entropy which would be lost on association. Unfortunately, no data exist on the entropy of either of these anions from which it would be possible to decide whether the ions in aqueous solution possess rotational entropy. However, for gas phase association, Evans and Nancollas estimate rotational entropy loss for N_3^- of 11 cal. degree^{-1} mole^{-1}.[8] A similar calculation for SCN^- shows its gas phase rotational entropy loss to be 16 cal. degree^{-1} mole^{-1}. This value is sufficiently large to change the total entropy of association from a positive value, such as is found for other anions, to a negative value defined by the present experiments, and, hence, can account for the very slight tendency toward association noted for SCN^-.

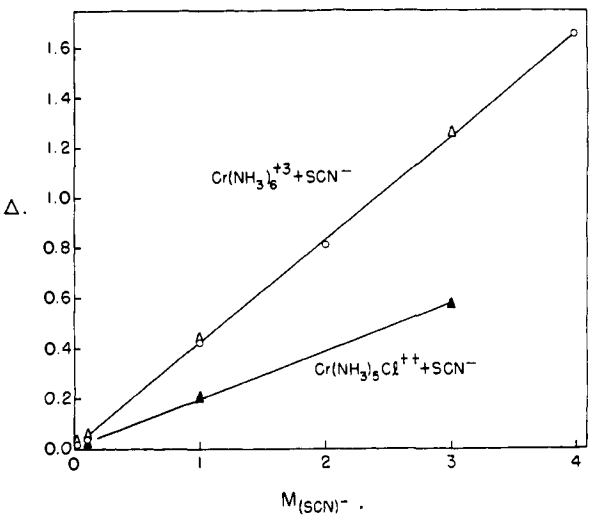

Fig. 5.—The interaction of 0.01 M $Cr(NH_3)_6^{+3}$ and 0.005 M $Cr(NH_3)_5Cl^{+2}$ with various concentrations of SCN^-. Data obtained from optical density measurements at 2750 Å. Circles represent points obtained at 3°; triangles, at 26°.

ITHACA, N. Y.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, POLYTECHNIC INSTITUTE OF BROOKLYN]

Photoreduction of Eosin in the Bound State[1a,b]

BY JUDITH S. BELLIN AND GERALD OSTER

RECEIVED NOVEMBER 30, 1956

The spectral and photochemical properties of eosin Y bound to polyvinylpyrrolidone differ considerably from those of the free dye. The bound dye exhibits self-quenching of fluorescence at much lower concentrations than does the free dye, whereas for self-quenching of phosphorescence (in rigid media) the opposite is true. Furthermore, *p*-phenylenediamine which quenches the phosphorescence of the free dye is without effect on the bound dye. Nitrobenzene retards the photoreduction of free eosin but inhibits the photoreduction of bound eosin, the duration of the inhibition period being proportional to the concentration of the inhibitor. *p*-Phenylenediamine both inhibits and retards the photoreduction of bound eosin while it only retards the reaction for the free dye. For small dye concentrations the quantum yield of photoreduction of bound eosin increases with increasing dye concentration while for free dye the opposite is the case. These rate studies suggest that a bound dye molecule in the first electronically excited state rapidly exchanges energy with a bound dye molecule in the ground state to produce a long-lived excited species which reacts with the reducing agent (here ascorbic acid). A mechanism is proposed for dye-sensitization in silver halide photography based on photoreduction of bound dye.

Introduction

The photochemical properties of dyestuffs when bound to substrates are considerably different from those of the free dye in solution. For example, rose bengal (2′,4′,5′,7′-tetraiodo-3,4,5,6-tetrachlorofluorescein) is readily photoöxidized in solution whereas the dye when bound to polyvinylpyrrolidone (PVP) resists photoöxidation even when exposed to sunlight for one month.[2] On the other hand, dyes which are bound to high polymeric substrates are more readily photoreduced than are the free dyes.[3-5]

The present paper is concerned with the photoreduction of eosin Y (2′,4′,5′,7′-tetrabromofluorescein) when bound to PVP in solution. The kinetics of photoreduction of free eosin Y have been studied.[6] It has been shown that the kinetics of photoreduction of free fluorescein and its halo-

(1) (a) This paper represents a part of the dissertation submitted by Judith S. Bellin to the faculty of the Graduate School of the Polytechnic Institute of Brooklyn in partial fulfillment of the requirements for the degree of Doctor of Philosophy. (b) This research was supported by the United States Air Force through the Air Force Office of Scientific Research of the Air Research and Development Command under Contract No. AF18(600)1182.

(2) G. Oster, *J. Polymer Sci.*, **9**, 553 (1952).
(3) G. Oster, *Trans. Faraday Soc.*, **47**, 660 (1951).
(4) G. Oster, *J. Polymer Sci.*, **9**, 553 (1952).
(5) G. Oster and J. Bellin, THIS JOURNAL, **78**, 294 (1957).
(6) G. Oster and A. H. Adelman, *ibid.*, **78**, 913 (1956).

genated derivatives are qualitatively similar.[7] Likewise, we have found that the photochemical properties of the fluorescein-type dyes, erythrosin, phloxine and rose bengal, when bound to PVP are similar to those of bound eosin which are described in detail in the present paper.

The results of the present studies have direct bearing on the phenomena underlying dye-sensitization in silver halide photography.

Experimental

Materials.—Eosin Y as obtained from Eastman Kodak Co. was used without further purification. Polyvinylpyrrolidone (PVP) of molecular weight 5.6×10^5 was obtained from Schenley Laboratories. All the other reagents were obtained from Fisher Scientific Co. as C.P. grade. Helium (Airco) was used to flush oxygen from the solutions.

Procedures.—The methods used for determining the rate of photoreduction of eosin Y in the presence of ascorbic acid as well as the methods used in the measurements of absorption spectra, fluorescence spectra and fluorescence and phosphorescence intensities are identical with those described in our previous paper.[5] The wave length of the actinic light employed was that maximally absorbed by eosin Y, namely, 518 mμ.

Precautions were necessary in measuring the fluorescence quenching by *p*-phenylenediamine (PPD) since the oxidation of this substance is sensitized by eosin.[8] Freshly prepared mixtures were used for each measurement. A buffered stock solution of PPD was kept under helium in order to prevent its oxidation by air.

Results

Spectra.—A dilute solution of eosin Y is orange (absorption maximum at 518 mμ) and exhibits a yellow-green fluorescence when excited by blue light. With excess PVP the color changes to red (absorption maximum at 528 mμ with excess PVP) and the fluorescence is yellow and is of greater intensity. In contrast to our observations with triphenylmethane dyes,[5] the shape of the absorption spectra of eosin Y is unchanged on binding to the high polymer and is merely shifted to longer wave lengths.

By following the change in optical density at the wave length where the spectral changes are the largest (518 mμ for eosin Y) as a function of the amount of PVP added, we obtained a binding curve.[9] Employing the customary analysis of the binding data, we find at *p*H 5.0 that one gram of PVP binds 1.0×10^{-5} mole of eosin (see Fig. 1).

Fluorescence and Phosphorescence.—Solutions of free eosin show evidence of self quenching of fluorescence at a dye concentration of about 10^{-4} *M* (Fig. 2). An actual decrease in fluorescence intensity occurs at concentrations exceeding 10^{-3} *M*. On the other hand, in presence of PVP self-quenching becomes evident at concentrations of dye as low as 10^{-6} *M* and the fluorescence intensity is a maximum at 10^{-5} *M* (Fig. 2).

PPD quenches the fluorescence of eosin Y in the bound as well as in the free state. In all cases the quenching obeys the Stern–Volmer equation. At *p*H 5.0 the quenching constants of PPD for the dye in the free and in the bound states are, respectively, 4.37 and 3.14 liters/mole.

Oxygen-free solutions of eosin Y in glycerol at temperatures below −90° exhibit a yellow phosphorescence when the system is excited by blue light. Addition of PVP enhances the intensity of the phosphorescence and makes the phenomenon observable at temperatures as high as −60°.

Self quenching of the phosphorescence of eosin Y in glycerol at −90° is evident at dye concentrations of 10^{-4} *M*. In the presence of PVP, however, this self quenching does not occur. PPD up to a concentration of at least 10^{-4} *M* is not a quencher of the phosphorescence of bound eosin Y, whereas it is a powerful quencher of the phosphorescence of free eosin.

Photoreduction.—The quantum yield of photobleaching with ascorbic acid at *p*H 5.0 of eosin Y, rose bengal, phloxine and fluorescein is greater in the bound than in the free state. The resulting colorless solutions exhibit a weak yellow fluorescence when excited by near ultraviolet light. Both bound and free fluorescein-type dyes reduce tetrazolium salts to insoluble formazans under the conditions of the photobleaching reaction, while in the dark no formazan is produced. This shows that in the photobleaching process a reduced form of the dye is produced which in turn is capable of reducing the tetrazolium salts. As is the case with free fluorescein-type dyes,[6] the quantum yield ϕ of the photoreduction of bound eosin Y as a function of reductant (A), here ascorbic acid, follows the relation $\phi = (A)/\alpha + \beta(A)$ where α and β are constants, independent of incident light intensity; there is, however, a striking difference between the free and the bound dye as regards their dependency of quantum yield on dye concentration. Whereas, with free dye the quantum yield of photoreduction decreases with increasing dye concentration, in the case of bound dye the quantum yield increases with increasing dye concentration up to about 6×10^{-6} *M* in eosin and a limiting value is attained at higher dye concentrations (Fig. 3). The photoreduction of bound triphenylmethane dyes[5] is similar in all these respects to those of bound eosin.

Photoreduction of bound eosin Y is inhibited by small amounts of nitrobenzene. For example, the addition of 10^{-6} *M* nitrobenzene causes an induction period of one minute during which the nitrobenzene is destroyed and after which the reaction proceeds at a rate equal to that of the inhibitor-free system. The induction period is directly proportional to the amount of nitrobenzene added. This phenomenon is similar to that observed for the case of bound triphenylmethane dyes.[5] In the case of free eosin, however, nitrobenzene acts as a retarder rather than as an inhibitor.

The effect of PPD on the rate of photoreduction of bound eosin Y is illustrated in Fig. 4. There is a period of inhibition, after which the photoreduction takes place at a reduced rate compared with that of the inhibitor-free system.

Discussion

The increased fluorescence yield of eosin on binding to PVP may be due to the increased planarity of the dye when bound.[10] The data of Fig. 1 show that the rate of photoreduction of bound eosin is not directly proportional to the degree of

(7) A. H. Adelman and G. Oster, THIS JOURNAL, **78**, 3977 (1956).
(8) G. Oster and M. Schrader, to be published.
(9) G. Oster, *J. Polymer Sci.*, **16**, 235 (1955).
(10) Compare G. Oster and Y. Nishijima, THIS JOURNAL, **78**, 1581 (1956).

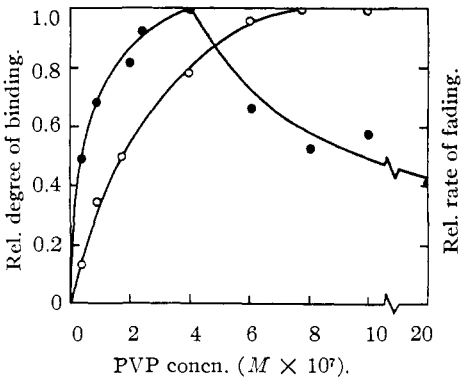

Fig. 1.—Open circles, degree of binding of eosin Y to polyvinylpyrrolidone (PVP) by changes in optical density at 518 mμ. Solid circles, rate of fading at *p*H 5.0 in the presence of 1.6×10^{-3} *M* ascorbic acid. Dye concentration 5×10^{-6} *M*.

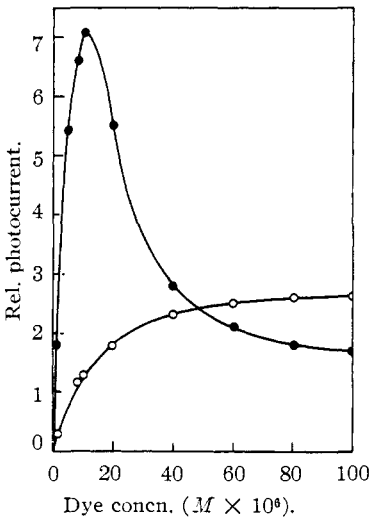

Fig. 2.—Fluorescence as a function of concentration of eosin Y for free dye (open circles) and for dye bound to 0.6% PVP (closed circles). A yellow Corning 7-30 filter was interposed between the sample and the phototube (RCA 931 A.).

binding; a maximum rate of fading is attained at a ratio of dye to polymer at which not all the dye molecules are considered by the absorption criterion to be bound. This suggests a redistribution of dye molecules on the polymer chain resulting in further changes in optical density but with a decreased rate of fading due to decreased possibilities of dye–dye interaction (see below).

As in the case of the bound triphenylmethane dyes,[5] the following kinetic scheme is consistent with the fading data of bound eosin: (1) D + $h\nu$ → D*, (2) D* → D + heat (and/or $h\nu_f$), (3) D* → D′, (4) D* + D → D′ + D, (5) D′ → D + $h\nu_p$, (6) D′ + A → D + A, (7) D′ + A → M, (8) M → colorless products (I), (9) M + × → colorless products (II) + D. In our terminology, D, D* and D′ refer to bound dye molecules in the ground, first electronically excited, and long-lived electronically excited states, respectively, the unbound dye being photochemically inert under the conditions employed in these studies. The

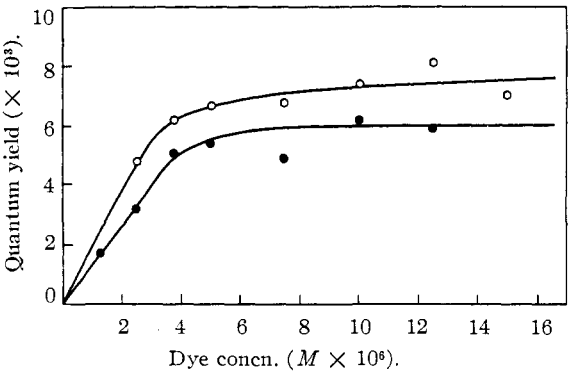

Fig. 3.—Quantum yield of fading of eosin Y as a function of dye concentration at *p*H 5.0. Throughout 1.1×10^5 g. of PVP per mole of dye. Open circles, 3.2×10^{-3} *M* ascorbic acid, filled circles, 1.6×10^{-3} *M* ascorbic acid.

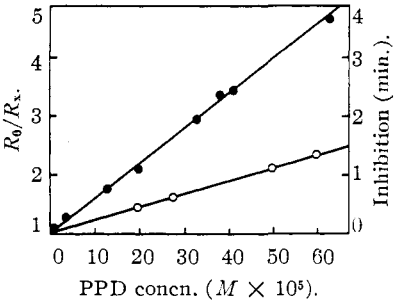

Fig. 4.—Time of inhibition (open circles) and retardation (closed circles) of fading of eosin Y as a function of concentration of paraphenylenediamine (PPD). Dye concentration, 5×10^{-6} *M*, PVP concentration, 0.06%, ascorbic acid concentration, 1.6×10^{-3} *M*, in acetate buffer at *p*H 5.0.

reductant A reacts only with D′ either to quench the phosphorescence (step 6) or to form an intermediate complex M (step 7). This complex in turn can decompose to give reduced dye products or, in the presence of inhibitor X, can cause the destruction of X with the regeneration of dye. The justification for this formulation is treated in detail in our previous paper.[5] Using steady-approximations for the transient species (D*, D′ and M) these reaction steps lead to the following equation for the quantum yield of photoreduction

$$\Phi = \left[\frac{k_8}{k_8 + k_9(\mathrm{X})}\right]\left[\frac{k_7(\mathrm{A})}{k_5 + (k_6 + k_7)(\mathrm{A})}\right]\left[\frac{k_3 + k_4(\mathrm{D})}{k_9 + k_3 + k_4(\mathrm{D})}\right]$$

where the *k*'s are the rate constants for the reactions indicated by the subscripts. Since the quantum yield is zero when extrapolated to zero dye concentration (Fig. 3), k_4(D) is much larger than k_3 and hence, as with bound triphenylmethane dyes, step 3 is negligible compared with step 4. In contrast to the behavior of bound triphenylmethane dyes, however, k_4(D) for bound eosin must be comparable to k_2, since a plateau in the rate of fading is reached at higher dye concentrations. As in the case of bound triphenylmethane dyes we evaluate the ratio of the constants[5] which for eosin are $k_4/k_2 = 2.24 \times 10^5$, $k_6/k_5 = 450$, $k_7/k_5 = 211$, liters per mole, respectively. We estimate that k_2, the reciprocal of the lifetime of bound

eosin, is about 3×10^8 sec.$^{-1}$ [11] so that k_4 would be 6×10^{13} l./mole/sec. As might be expected, this value of k_4 cannot be accounted for on the basis of diffusion-controlled bimolecular encounters (6.6×10^9 encounters/sec./mole/liter in aqueous systems at 25°) but more likely represents the rate constant for energy transfer between dye molecules fixed on the polymer chain to yield dye molecules in the long-lived excited state. The quenching of this long-lived state by reductant (step 6) is twice as efficient as step 7 which leads to the intermediate complex.

Small amounts of PPD retard the rate of photoreduction of free eosin[6] whereas with bound eosin inhibition as well as retardation occurs (Fig. 4). Since there is no appreciable fluorescence quenching at these low PPD concentrations, the step D* + X → D + X is unimportant but reaction occurs as in step 9 to account for the observed inhibition (compare ref. 5). The retardation effect of PPD requires that products formed in step 9 quench the long-lived excited dye molecules, *i.e.*, (10) D′ + products (II) → D + products (II). Assuming steady-state concentrations for D*, D′ and M we derive the following expression for the ratio R_0/R_x of the rates of photoreduction in the absence and in the presence of substance X

$$\frac{R_0}{R_x} = \left[1 + \frac{k_9}{k_8}(\mathrm{X})\right]\left[1 + \frac{k_{10}(\text{Products (II)})}{k_5(k_6 + k_7)(\mathrm{A})}\right]$$

Since no photofading occurs until the inhibition period is ended k_9 must be much larger than k_{10}. Step 9 requires that the concentration of Product II be proportional to the amount of PPD added (X_0). We may express the amount of retardation which occurs after the inhibition period is completed as

$$\frac{R_0}{R_x} = 1 + \frac{k_{10}'(X)_0}{k_5 + (k_6 + k_7)(\mathrm{A})}$$

Thus, as shown in Fig. 4, the retardation is directly proportional to the amount of PPD originally added. From the slope of this line and using the ratios of the rate constants enumerated above we obtain $k'_{10}/k_6 = 1.35 \times 10^6$. If the concentration of Products II is comparable in magnitude to that of X_0 then quenching by this product is much more efficient than quenching by reductant. These results are consistent with the observation that PPD itself does not quench the phosphorescence of bound eosin although it strongly quenches the phosphorescence of free eosin.

The purely inhibiting action of nitrobenzene on the photoreduction of bound eosin is explainable in exactly the same way as that for bound triphenylmethane dyes.[5] In this case, step 10 is not important since no retardation occurs.

It has been known for many years that eosin applied to silver halide photographic emulsions will render the emulsion sensitive to green light.[12] Silver bromide emulsions sensitized with eosin have been studied in our laboratory.[13] The sensitivity of the emulsion is a maximum at the wave length where the bound dye absorbs light maximally. The binding curve for the dye as a function of silver bromide concentration is similar to that of Fig. 1. The emulsion exhibits sensitivities as a function of silver halide concentration and eosin concentration in a manner analogous to the fading curves of Figs. 1 and 3. Small amounts of nitrobenzene suppress the green sensitivity while the blue sensitivity (a property of the silver halide itself) is unchanged. It would appear that in this system, at least, the bound dye is photoreduced and the reduced dye donates electrons to the silver halide to form the latent image. This is an alternative suggestion to those contained in current theories of dye-sensitization in photography which postulate photoelectric phenomena.[14]

(11) Compare, P. Pringsheim, "Fluorescence and Phosphorescence," Interscience Publishers, Inc., New York, N. Y., 1949, p. 316.

(12) J. Waterhouse, *Brit. J. Photo.*, **23**, 233 (1876).

(13) G. Oster, to be published.

(14) For review, see W. West and B. H. Carroll in "The Theory of the Photographic Process," Ed. by C. E. K. Mees, Revised Edition, The Macmillan Co., New York, N. Y., 1954.

BROOKLYN, N. C.

[CONTRIBUTION FROM THE QUARTERMASTER RESEARCH AND DEVELOPMENT CENTER]

Spectroscopic Studies on Dyes. IV. The Fluorescence Spectra of Thioindigo Dyes[1]

BY DELBERT A. ROGERS, J. DAVID MARGERUM AND GEORGE M. WYMAN[1a]

RECEIVED JANUARY 28, 1957

The fluorescence spectra of eight thioindigo dyes in benzene solution were determined, using the 546 mμ Hg-line for excitation. Each dye has a fluorescence band at a wave length somewhat longer (usually by *ca.* 35 mμ) than its first absorption maximum. Exposure of the dye solutions to yellow light prior to the measurement results in decreased fluorescence. The intensity of fluorescence was found to be proportional to the concentration of *trans* isomer present. This behavior, which is similar to that observed for stilbene,[2] indicates that *trans*-thioindigo possesses a tightly-held coplanar structure, while its *cis* isomer is non-coplanar. These conclusions are shown to be consistent with the visible and infrared absorption data. The addition of ethanol quenches the fluorescence of these dyes. The possible implications of this phenomenon with regard to the photochemistry of thioindigo-dyed cellulosic materials are discussed.

Introduction

In their study of the fluorescence spectra of *cis*- and *trans*-stilbene, Lewis and his co-workers have found that only the *trans* isomer exhibits fluorescence.[2] While a large number of articles dealing with fluorescence spectra have appeared in the lit-

(1) Presented before the Division of Physical and Inorganic Chemistry at the 130th Meeting of the American Chemical Society, Atlantic City, New Jersey, September 16–21, 1956. (1a) U. S. Army Res. and Dev. Liaison Group, (D671 DU), Rheingau Allee 2, Frankfurt-am-Main, Germany.

(2) G. N. Lewis, T. T. Magel and D. Lipkin, THIS JOURNAL, **62**, 2973 (1940).