

Photoreduction of Indigo Dyes by Electron Donors. One- and Two-Electron-Transfer Reactions as a Consequence of Excited-State Quenching¹

Kirk S. Schanze,² Lester Y. C. Lee, Charles Giannotti,³ and David G. Whitten*

Contribution from the Department of Chemistry, University of Rochester, Rochester, New York 14627. Received May 6, 1985

Abstract: The indigoid dyes, thioindigo (TI), *N,N'*-diacetylindigo, (NDI) and oxalyindigo (OI), all undergo reduction upon irradiation of the dyes in the presence of electron donors such as triethylamine (TEA) or *N*-benzyl-1,4-dihydropyridinone (BNAH). Product analysis by NMR and high-resolution mass spectrometry has shown that the products for TI and NDI are the formal H₂ adducts TIH₂ and NDIH₂; the product for OI has been shown to be the semireduced radical OIH• which is readily detected by its characteristic ESR spectrum. Mechanistic studies have been carried out for the visible-light-induced reduction of the three dyes. For TI it has been found that both a direct two-electron (electron-proton-electron transfer sequence) transfer within the complex from which singlet quenching occurs and a disproportionation of the semireduced species TIH• formed by one-electron transfer contribute to the product formation in paths whose relative importance is solvent dependent. For NDI the same net reaction occurs but there is little evidence for any important free radical component; evidently in this case quenching of singlet NDI is followed by a rapid sequence leading to net two-electron or hydride transfer to generate directly the conjugate base of the product. For OI reduction occurs from the excited triplet and involves exclusively a single electron-transfer process. Although the "direct" two-electron reduction occurs on a rapid time scale and should not be as susceptible to quenching processes as the slower radical disproportionation path, it occurs with generally low efficiencies in the cases studied due to competition with various paths leading back to starting materials. For the system TI/BNAH flash spectroscopic and ESR investigations indicate the presence of a radical chain process in the photoreduction.

Photoreduction of various chromophores (e.g., ketones, transition-metal complexes, and dyes) absorbing in the near-UV or visible by electron donors has been a very well studied process.⁴⁻¹⁸ In a number of cases excited-state quenching by electron transfer from the reductant leads to the production of radical ions as transient products, detectable by fast spectroscopic investigations, but no permanent chemical change occurs due to back electron transfer unless reagents are added which "scavenge" one of the initial products. For several compounds electron-transfer quenching of the excited chromophore generates a reduced species, usually a radical anion, which is quite basic and thus readily protonated such that a "semireduced" free radical accumulates;¹⁹⁻²²

in these cases disproportionation of the product of the electron-proton transfer sequence can lead to production of the two-electron reduction product or dihydro compound (among other products) as a metastable or permanent photoproduct. Predominant among chromophores undergoing this type of photoreduction are ketones,¹⁶⁻¹⁸ quinone derivatives,^{23,24} and other "Wertz-type" reagents such as thionine dyes.²⁵⁻²⁷

In a recent communication¹ we reported that irradiation of thioindigo (TI) in the presence of several potential reductants leads to accumulation of the two-electron reduction product *leuco*- or dihydrothioindigo (TIH₂). Although part of the product would be demonstrated to arise from disproportionation of the semireduced radical, TIH•, a major portion of the product was found to be formed by a net two-electron transfer occurring within the donor-acceptor pair formed directly in the quenching process. The reaction could be attributed to cooperativity between the donor and acceptor involved in the process. Thus in this case reduction of excited thioindigo leads to a basic radical anion which, once protonated, is more easily reduced than TI itself.^{28,29} At the same time reductants—in this case tertiary amines—are oxidized to acidic radical cations which, after proton loss, are more powerful electron donors than the starting amine. The net result is a sequence of electron, proton, electron transfer or a net hydride transfer within the quenching complex.

The present paper contains a detailed account of the photoreduction of thioindigo as well as two other structurally related

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(3) C.N.R.S., Institut de Chimie des Substances Naturelles, Gif-sur-Yvette, France.

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indigo dyes, *N,N'*-diacetylintdigo (NDI) and oxalyindigo (OI). Both of these dyes undergo photoreduction with reagents capable of photoreducing TI. A mechanistic study of the photoreduction of these three dyes by the amine reductants triethylamine and *N*-benzyl-1,4-dihydronicotinamide (BNAH) has been carried out; the results of this study indicate that each of the three dyes reacts by a different overall mechanism although certain steps occur in common. The pathways observed for the three dyes appear to be characteristic and thus indicative of mechanisms which may be generally anticipated for specific donor-acceptor combinations.

Experimental Section

Materials. Benzene and methylene chloride were reagent grade and were used without further purification. Carbon tetrachloride was distilled from KMnO_4 . Chloroform was purified by washing with water, dried over CaCl_2 , and distilled from CaSO_4 . Tetrahydrofuran was distilled from LiAlH_4 after refluxing. Acetonitrile was distilled from CaH_2 after refluxing. Dimethylformamide was distilled from calcium oxide. Methylene-*d*₂ chloride was Norrell NMR grade, 99+% atom *d*. Chloroform-*d* was Merck, Sharpe and Dohme NMR grade, 99+% atom *d*. Triethylamine, diethylamine, diethylaniline, and diethylisopropylamine were distilled from KOH. 1,3-Cyclohexadiene and 1,4-cyclohexadiene were reagent grade.

Thioindigo was a gift generously provided by the Ciba-Geigy Corporation of Basel, Switzerland. *N,N'*-Diacetylintdigo was prepared by acylation of indigo (acetyl chloride).³⁰ Oxalyindigo was a gift from Dr. G. M. Wyman of the U.S. Army Research Office. The sample was recrystallized twice from nitrobenzene, yielding small yellow crystals which were dried by heating under vacuum for several hours.

1-Benzyl-1,4-dihydronicotinamide (BNAH) was prepared via benzylation of nicotinamide followed by reduction of the cationic *N*-benzylpyridinium salt.³¹ Three grams (25 mmol) of nicotinamide (Aldrich) and 4.25 g (25 mmol) of benzyl bromide (Fischer) were refluxed in 50 mL of toluene overnight. The pyridinium salt precipitated out as a chalky-white solid; the product was filtered, washed with CHCl_3 , and dried under vacuum. Seven grams (24 mmol) of the bromide salt of 1-benzylnicotinamide (from the previous step) was dissolved in 50 mL of H_2O . The solution was stirred and purged with N_2 for 20 min. The 5.4 g (37 mmol) of $\text{Na}_2\text{S}_2\text{O}_4$ and 4.2 g (40 mmol) of Na_2CO_3 were added as a mixed solid. The solution immediately turned orange-yellow and a reddish oil separated from the aqueous phase. The aqueous suspension was then extracted twice with CHCl_3 ; the organic layer was collected and dried over Na_2SO_4 . The CHCl_3 was subsequently removed by vacuum evaporation, leaving a red-orange oil. The oil was dissolved in 50 mL of ethanol with gentle heating. When the oil had dissolved H_2O was added to induce crystallization; the solution was then cooled overnight and red-yellow crystals formed. The crystals were collected and recrystallized from 50% ethanol- H_2O with 1.4 g (6 mmol) of BNAH obtained as small yellow crystals in 25% yield. The product was dried under vacuum; the sample had mp 119° dec (lit. mp 120–122 °C dec)³² and its purity was further established by FT-NMR.

Proton NMR Analysis of Indigo Reduction Products. Analysis of the photoproduct from the reaction of NDI or TI with an electron donor was complicated by the fact that the product is air sensitive. Further problems were encountered because the dyes are practically insoluble in most organic solvents. As a result, experiments were designed whereby small amounts of the photochemical reaction product could be generated and handled under deaerated conditions. Preparation of the dye/quencher photoproduct was carried out under conditions identical with those of the spectroscopic experiments (at low dye concentration); the product was then isolated by distilling off the solvent and excess quencher. Subsequently an NMR solvent was added via vacuum distillation and the NMR spectrum was obtained on the crude photoproduct. During the entire process the photoproduct was under a vacuum. Eighty to one hundred milliliters of a benzene solution which was 10^{-4} M in dye and 0.02 M in TEA was degassed on a vacuum line through 3 freeze-pump-thaw cycles and then the system was closed while the pressure was $<10^{-4}$ torr. A UV-vis spectrum was obtained on the sample prior to irradiation. (A Pyrex test tube had been fused onto the main cell to allow for spectrophotometric monitoring of the solution during photolysis.) The solution was then irradiated with use of a Rayonet photochemical reactor which was fitted with gold fluorescent tubes ($\lambda > 500$ nm). During exposure the solution was cooled by immersion in an ice bath. Photolysis

was continued until the visible spectrum showed less than 5% of the starting absorbance at 550 nm (2–3 h). The cell was reconnected to the vacuum line; the benzene solvent along with excess TEA was distilled from the solid. Upon completion of the distillation the solid was pumped by the vacuum line to remove traces of solvent. After the product was dried, 0.7 mL of CDCl_3 was connected to the vacuum line and degassed through 3 freeze-pump-thaw cycles. Then the CDCl_3 was distilled onto the solid and the cell containing the CDCl_3 and sample was sealed and removed from the vacuum line. The cell was swirled to dissolve the photoproduct in the NMR solvent and the resulting solution was carefully poured into an NMR tube sidearm. Finally, the NMR tube containing the photoproduct was sealed and removed. An NMR spectrum was run on the sample immediately after completion of the operations described.

Product Analysis Experiments by Mass Spectroscopy. Photoproduct samples for MS analysis were prepared by using a similar procedure as described for the NMR experiment above. The photoproduct sample was prepared as described above for the NMR experiment except that after transferring the CHCl_3 solution (CHCl_3 was used in this case instead of CDCl_3) of the photoproduct into a capillary manifold, the solvent was distilled to leave the photoproduct as a residue inside the capillary tubes. After removal of the solvent the capillary tubes were sealed with a torch. The samples were maintained under a vacuum until immediately prior to insertion into the mass spectrometer injection port.

Kinetics and Quantum Yield Determinations. Solutions containing 5×10^{-5} M TI, 1.5×10^{-4} M NDI, or 1.0×10^{-4} M OI to give optical densities of approximately 1.0 at the wavelength of maximum absorbance were prepared. Quenchers were added from stock solutions or neat with volumetric pipets. All solutions were degassed on a vacuum line through 5 freeze-pump-thaw cycles and sealed at $<10^{-5}$ torr with a torch. All solution preparations and degassing were carried out in a darkroom under a dim red light.

The solutions were irradiated on a merry-go-round apparatus in parallel with a Hanovia-type 450-W medium-pressure Hg lamp; Corning band-pass (440–580 nm) and cutoff (500 nm) filters were used in combination to isolate the 546-nm line for TI and NDI, and a cutoff filter (420 nm) was used for OI. Ten percent conversion occurred within 0.25 to 1.0 h, depending upon quencher concentration. The reaction was monitored by measuring the dye absorbance with either a Perkin-Elmer 576 or a Hewlett-Packard 8451A spectrometer. Plots of absorbance (λ_{max}) vs. time provided straight lines for up to 10% conversion; the slopes of the plots are proportional to the quantum yields for the reaction after correction for incomplete light absorption. Quantum yields were determined relative to a secondary actinometer which was irradiated in parallel with the solutions. The secondary actinometer was a deaerated solution of benzene containing 5×10^{-5} M TI and 0.02 M TEA; the quantum yield for bleaching of TI in this solution (ϕ_{ac}) was measured relative to Reineckes salt.³³

Fluorescence quenching experiments were carried out with solutions 5×10^{-6} – 1×10^{-5} M in dye (optical density 0.1–0.3 at $\lambda_{\text{excitation}}$). Fluorescence spectra were recorded on either an SLM 8000 or a Hitachi MPF-2A fluorimeter. Quenching constants were measured through the Stern-Volmer relationship.

Conventional flash spectroscopy experiments were performed on an apparatus described elsewhere.³⁴ Laser flash photolysis experiments were performed with a Q-switched ND:YAG laser (Quanta Ray DCR 2(10)A) as the excitation source and a 150-W Xe lamp (Osram XBO 150 W/S) as the monitoring light beam. The lamp was mounted in a Photochemical Research Associates (PRA housing ALH 215) system and powered by a PRA Model 301 power supply and arced with a PRA Model 301S starter. Operating lamp intensity was at 6–7 amps for continuous monitoring and 50 amps when pulsed. The light beam was passed through a water filter before impinging on the entrance aperture of the cell holder. An electronic shutter (Ealing Electronic Shutter, Model No. 22-8411, Ealing Co., South Natick, MA) was placed between the water filter and the cell holder which was opened for 30 ms during each flash. The electronic shutter was controlled by a slightly modified shutter power supply (Ealing Model No. 22-8411); the modifications enabled the shutter to be controlled by externally supplied driver pulses. Two lenses were used to collimate and focus the monitoring light beam from the cell holder exit onto the entrance slit of the monochromator (Bausch and Lomb 0.25-m grating monochromator, blazed at 300 nm with 50-Å/mm dispersion). A photomultiplier detector mounted inside a housing (Ealing PMT housing No. 28-4646) was fitted to the monochromator exit slit port. Either a RCA 1P28 or a RCA 4840 tube was used (red sensitive to 700 nm). The PMT was wired specifically to allow for high output currents which are caused by the high monitoring lamp

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intensity. The voltage divider circuit which was used is described in the literature.³⁵

PMT signals were monitored with a Tektronix 7912 AD transient digitizer equipped with a Tektronix 7A26 vertical amplifier and a delaying time base (Tektronix 7B92A). For short-lived transient signals ($\tau < 1 \mu\text{s}$), the PMT was AC coupled to allow offset of the large, low-frequency signal due to I_0 .

Triggering and proper timing of firing for the various components of the laser flash photolysis system was achieved by an external pulse generator (PG). The PG provided for firing of the DCR 2A excitation flashlamps continuously at 10 Hz (which is optimal for stability of the laser) while allowing selection of the laser Q-switch, shutter, pulsed monitoring lamp, and the transient digitizer triggering rates up to 1 Hz or with a single shot capability. This allowed flash photolysis experiments to be carried out in a signal average mode at rates up to 1 Hz or in a single shot mode. The transient digitizer was interfaced with a PDP 11/34 computer (floppy disk system) equipped with a Tektronix 4010-1 terminal, a Tektronix 4662 digital plotter, a Digital Decwriter IV printer, and Tektronix SPS Basic Version 2 software. Data acquisition, manipulation, storage, and plotting were carried out by Basic programs written with use of Tek SPC Basic.

The signal obtained from a flash photolysis experiment was the change in intensity of the monitoring light, ΔI , as a function of time at a given wavelength. Conversion of transient signals to changes in absorbance (ΔA) is obtained by eq 1, where $\Delta I(t)$ is the observed signal (mV) and

$$\Delta A(t) = \log \left(\frac{I_0}{I_0 - \Delta I(t)} \right) \quad (1)$$

I_0 is the monitoring lamp intensity measured prior to the flash (mV). First-order rate constants were obtained from $\ln(\Delta A(t))$ vs. time plots. Transient spectra at delay time t_d after the laser pulse were constructed by obtaining several decay traces over the wavelength region of interest and subsequently plotting $\Delta A(t_d)$ vs. λ .

Optimal results were obtained with use of solutions with optical density of 1.0 at the wavelength of excitation. Optical densities < 0.5 at monitoring wavelengths were used whenever possible. Indigo dye concentrations were generally $5\text{--}8 \times 10^{-5}$ M. The solutions were contained in standard 1×1 cm quartz fluorescence cells. For signal-averaged experiments the cells had a reservoir fused on top of the quartz cell which would hold 15 mL of solution and a stirring fin was placed in the bottom of the cell. The solutions were outgassed with Ar and then sealed; the experiment was immediately carried out with vigorous stirring of the solution during signal acquisition. Typically no more than 10 pulses were averaged for solutions which were photolabile. For single-shot experiments a cell which would hold 100 mL of solution and could be vacuum degassed was used; this cell has been described in the literature.³⁶

The photochemical-ESR experiments were carried out at the Institut de Chimie des Substances Naturelles, CNRS, Gif-sur-Yvette, France. The experimental procedures have been reported in the literature.³⁴

Electrochemical Experiments. Electrochemical experiments were performed on a set up consisting of a modified wave form generator,³⁷ a Princeton Applied Research Model 174A Polarographic Analyzer, and a Hewlett-Packard 7015 B recorder. For cyclic voltammetry, 20 mL scintillation vials were used as one-compartment cells. The reference electrode was a saturated sodium chloride electrode (SSCE) with a platinum wire as the counter electrode. The working electrode was a Teflon-shrouded Pt disk for C-V and a Pt gauge for electrolysis. Tetraethylammonium (TEAP) (0.1 M) was used as supporting electrolyte. Typically, dye concentrations used for C-V were 0.01 and 10^{-4} M for electrolysis in DMF.

Results

The three dyes investigated most thoroughly in this study, thioindigo (**1**), *N,N'*-diacetylindigo (**2**), and oxalyindigo (**3**), have all been found to undergo "permanent" photochemical reaction when irradiated in deaerated solution in the presence of potential electron donors such as triethylamine (TEA) or 1-benzyl-1,4-dihydropyridinamide. In most cases the dye photoproduct reverts back to give the absorption spectrum of starting dye upon admission of air or oxygen to the sample. Typical absorption spectral changes upon irradiation of the indigo dyes with triethylamine

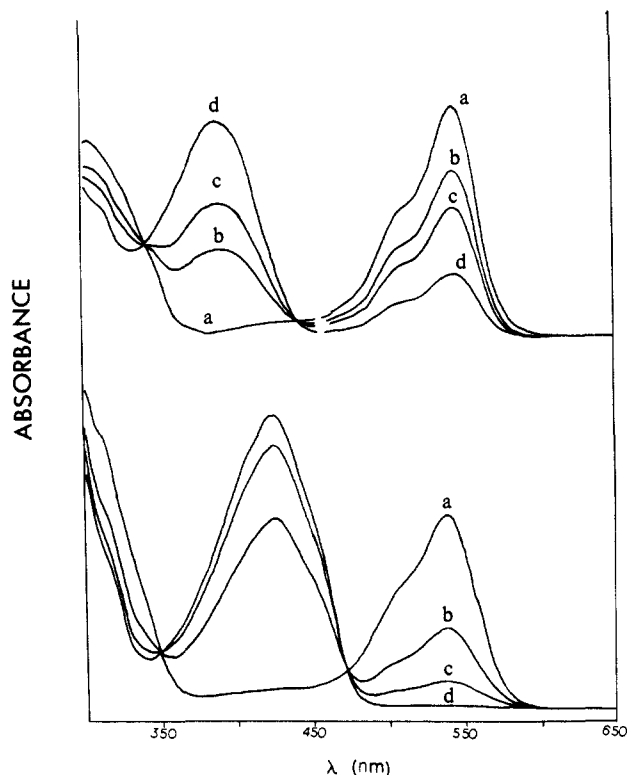


Figure 1. Photoreduction of thioindigo by triethylamine; spectral changes observed on irradiation in benzene (upper trace) (a, 0 min; b, 15 min; c, 25 min; d, 50 min) and in acetonitrile (lower trace) (a, 0 min; b, 5 min; c, 10 min; d, 20 min).

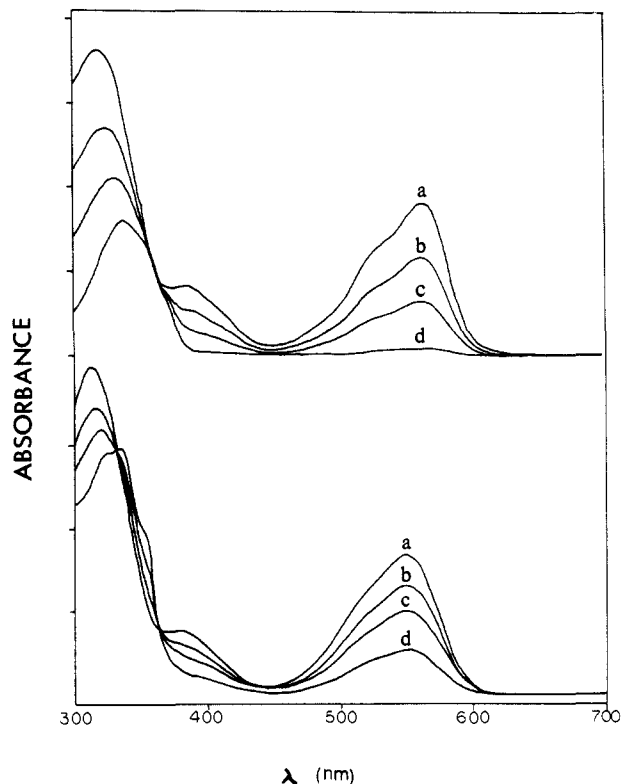


Figure 2. Photoreduction of *N,N'*-diacetylindigo by triethylamine; spectral changes in benzene (upper plot) (a, 0 min; b, 30 min; c, 65 min; d, 180 min) and in acetonitrile (lower plot) (a, 0 h; b, 5.5 h; c, 11.0 h; d, 19.5 h).

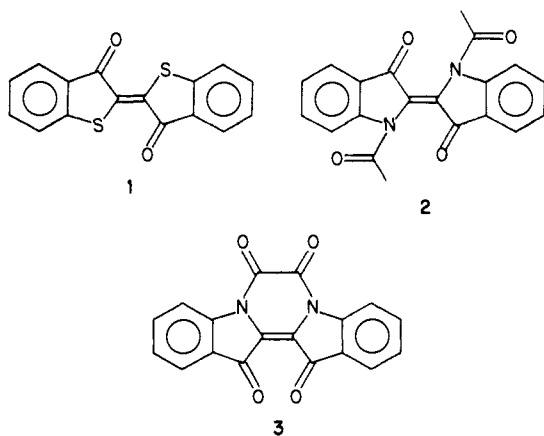
are shown in Figures 1–3. The products of these reactions were determined in each case as outlined in the following section.

Thioindigo. Photolysis (546 nm) of degassed solutions of TI in the presence of various reducing agents was found to lead to

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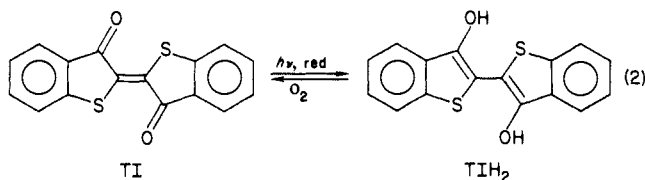
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disappearance of the "indigoid" absorption at 545 nm concurrent with appearance of a strong absorbance in the blue region (Figure 1). For both benzene and acetonitrile, as well as several other solvents, two isosbestic points are maintained in each case within the spectral region examined (330–650 nm). These results suggest that the photoreaction proceeds cleanly to produce a single product or a mixture of products in a constant ratio. Similar changes were observed as a result of irradiation of the dye in the presence of other tertiary amines, BNAH, diethylamine, and *N*-methylacridan (NMA). Since the reaction occurs with reductive quenchers and under degassed conditions it is reasonable to assume that the photoproduct results from reduction of TI. This hypothesis is further substantiated by the observation that exposure of solutions of the photoproduct to air leads to rapid, quantitative regeneration of the absorption characteristic of the starting material, TI, in each case.

A sample of the photoproduct from photoreduction of TI by TEA was generated; the crude product was sealed in several capillary tubes and submitted for mass spectral analysis. The base peak in the MS corresponds to the molecular ion of TIH_2 , occurring at m/e 298; the second most intense peak occurs at m/e 149 which could correspond either to a doubly charged molecular ion or to a fragmentation into "half-molecule" cations and radicals. A mass spectrum was also obtained on isolated photoproduct which had been exposed to air for several hours. This spectrum was identical with that published for TI;³⁸ the base peak here corresponds to the molecular ion at m/e 296.

Reductive electrolysis of a *N,N*-dimethylformamide solution of TI and excess benzoic acid at $E = -0.5$ V (vs. SSCE) gives a product having an absorption spectrum nearly identical with that of the TI/TEA photoproduct. This evidence together with the mass spectral results suggests strongly that the photoreaction involves the $2e^-$, 2H^+ reduction of TI to yield "leuco" or dihydrothioindigo (TIH_2) (eq 2).



An experiment was carried out whereby a sample of the TI/TEA photoproduct was generated and transferred to a sealed NMR tube under a vacuum. The proton NMR of the photoproduct which was obtained showed broad, unresolved peaks as follows: δ 4.3–5.4 (broad and unresolved, rel area 1), δ 7.0–7.4 (unresolved, sharp peak, rel area 2), δ 7.5–8.0 (unresolved, some fine structure, rel area 2). Since the product, TIH_2 , even if not highly purified, would be expected to give a more resolved spectrum, it seems reasonable that TIH_2 probably consists of several equilibrating structures as shown in Scheme I.³⁹ Structures

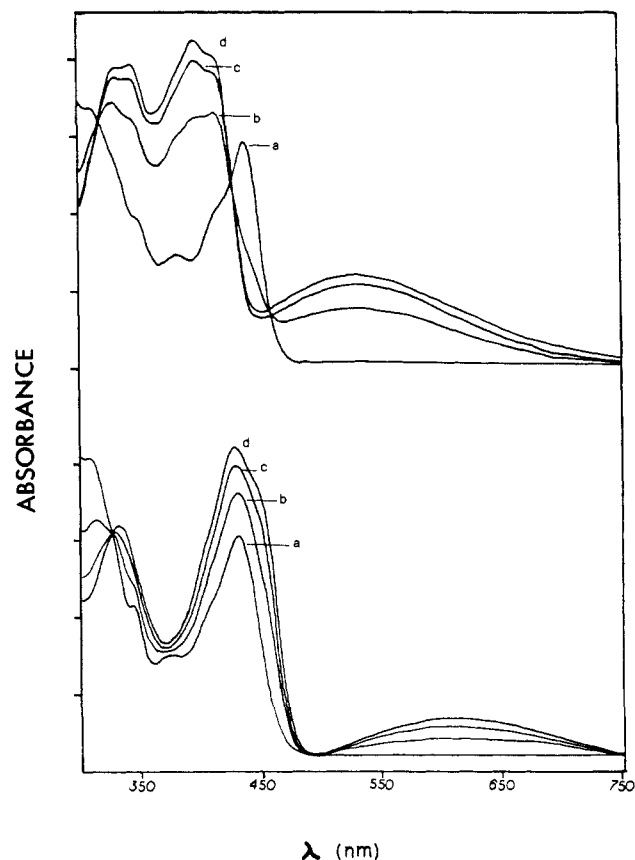
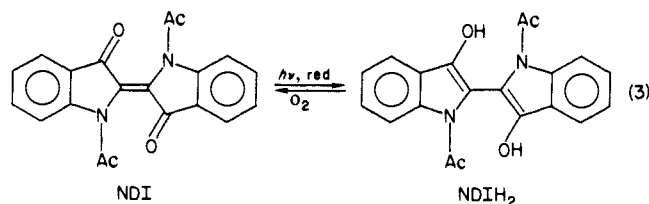


Figure 3. Changes observed upon irradiation of oxalyindigo with triethylamine; upper trace, benzene (a, 0 min; b, 2 min; c, 6 min; d, 25 min); lower trace, acetonitrile (a, 0 min; b, 2 min; c, 6 min; d, 25 min).

a–d can equilibrate either by tautomerism or via anion e; the rather pronounced changes of photoproduct spectra from TI with solvent compared to the starting dyes can probably be attributed to changes in equilibrium distributions of the various products.

***N,N'*-Diacetylindigo.** Photolysis of NDI (546 nm) in degassed solution with various reducing agents causes bleaching of the visible absorption band of the dye with concurrent changes in the UV region. Figure 2 shows the changes in the electronic spectrum which were observed during photolysis of NDI in benzene and acetonitrile with 0.02 M TEA. Photolysis in acetonitrile leads to a decrease in absorption for $\lambda > 365$ nm; the NDI absorption at 315 nm decreases and is replaced by a photoproduct absorption band which is centered at 337 nm. Two isosbestic points are maintained during the irradiation; these occur at 342 and 362 nm. The resulting photoproduct absorption spectrum consists of a band centered at 337 nm with shoulders at 328 and 352 nm; the product does not absorb at $\lambda > 365$ nm and is therefore colorless. Irradiation of NDI in degassed solution with BNAH and NMA as reductants also caused bleaching of the visible absorption band; however, due to the strong UV absorption of BNAH and NMA this region could not be monitored for spectral changes. Interestingly, exposure of the bleached solutions to air was observed to cause slow but quantitative regeneration of the absorption spectrum of NDI.

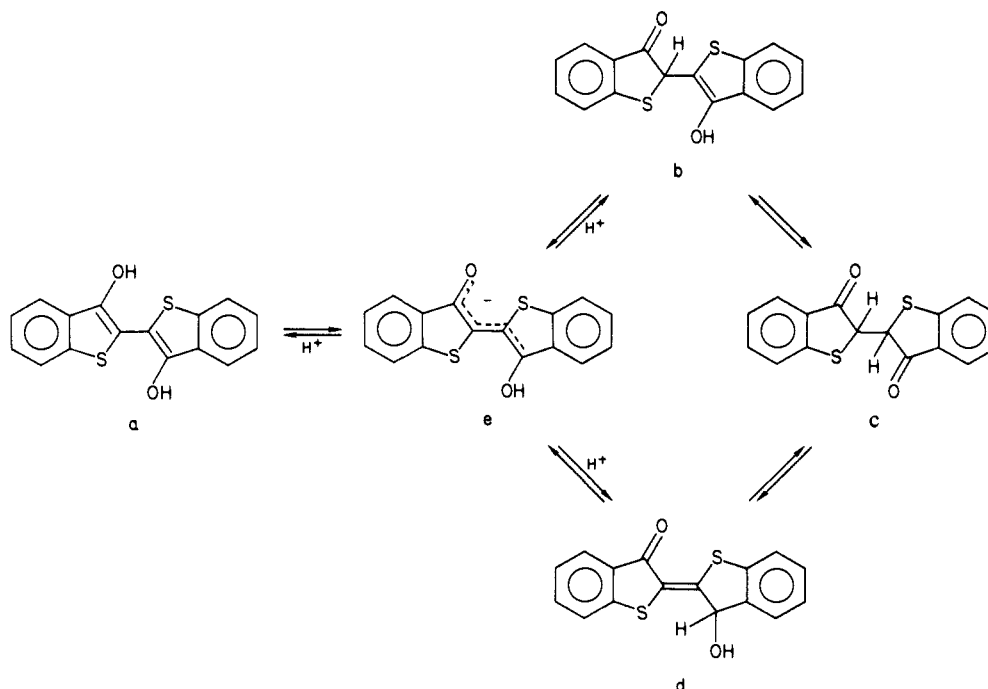
The spectral changes resulting from photolysis of NDI in degassed solution with tertiary amines and other reductants suggest that the dye is photoreduced to a leuco form, eq 3, in a reaction which is analogous to the photoreduction described above for TI.



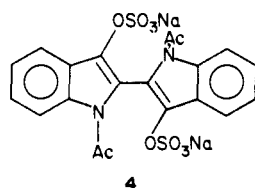
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(39) In accord with expectations for an "exchangeable" proton, the peak at δ 4.3–5.4 disappears upon treatment of the photoproduct with CD_3OD .

Scheme I



That NDIH_2 is formed is further evidenced by an experiment described by Dokunikhin et al.⁴⁰ These authors chemically reduced indigo and subsequently esterified the *leuco* form with chlorosulfonic acid; neutralization of the sulfonic acid ester resulted in formation of the H_2O soluble salt (4). They reported the



absorption spectrum for 4 as follows: $\lambda_{\text{sh}} = 325 \text{ nm}$; $\lambda_{\text{max}} = 340 \text{ nm}$; $\lambda_{\text{sh}} = 355 \text{ nm}$. Significantly, this spectrum is remarkably similar to the spectrum of photoreduced NDI, a fact which suggests that NDI has a similar structure.

Electrochemical reduction of NDI (at -0.5 V) in DMF solution with benzoic acid as a proton donor was carried out; it was found that the absorption spectrum of the electrolytically reduced dye is similar to the photoproduct absorption. The reduced dye was found to be slowly reoxidized by air back to NDI. This indicated that NDI can also be electrochemically reduced to form NDIH_2 as observed for TI.^{28,29}

Mass spectra were obtained for samples of NDI and the NDI/TEA photoproduct. The spectrum for NDI shows a small molecular ion at m/e 346; the base peak occurs at m/e 262 which is the molecular weight of indigo. The photoproduct gives a markedly different mass spectrum; a strong peak for the parent ion is at m/e 348. Relatively intense fragments of m/e 306, 175, and 133 correspond to loss of an acetyl group, fragmentation with capture of H^\cdot , and deacylation of this fragment, respectively.

These results indicate that a leuco structure as shown in eq 3 is formed upon photoreduction of NDI; however, the precise structure of NDH_2 remains somewhat elusive. Analogous tautomers to those shown in Scheme I for TIH could equilibrate; while the similarity of spectra of NDIH_2 and 4 suggests the structure shown in eq 3, the lack of intense absorption in the near-UV such as observed with TIH_2 may indicate that in this case conjugation between the two halves of the molecule is broken due to a sterically induced deviation from planarity.

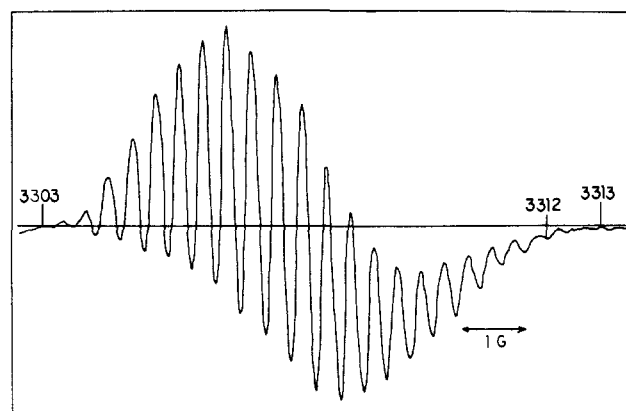
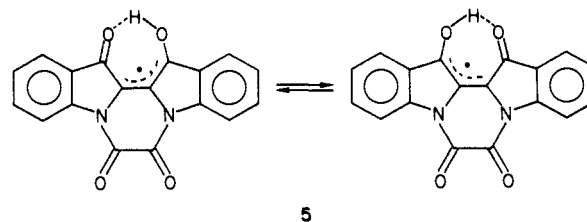


Figure 4. ESR spectrum of the oxalyindigo/triethylamine photoproduct.

Oxalyindigo. Photolysis of OI (436 nm) in degassed solution with 0.02 M TEA leads to changes in the absorption spectrum as shown in Figure 3. The photoproduct spectrum is quite variable with solvent. Although the photoproduct spectrum is indefinitely stable in deaerated solution, introduction of air causes *rapid* (<1 min) regeneration of the spectrum characteristic of OI. Photochemical ESR experiments with OI/TEA give results quite different from those with TI/TEA (vide infra); here it is found that irradiation of a CH_2Cl_2 solution of OI/TEA leads to the appearance of a *strong* 18-line ESR signal (Figure 4). This signal builds up during the irradiation and persists, undiminished, after the photolysis stops. The ESR signal can be attributed to the semireduced radical OIH^\cdot (5) generated by formal addition of a hydrogen atom to OI. Thus, in contrast to TI and NDI, OI appears to undergo only a one-electron reduction.



5

Excited-State Quenching and Reactivity. Previous studies have shown that the fluorescence of TI is quenched by electron donors;

(40) Dokunikhin, N.; Levin, E. *Compt. Rend. (Dokl.) Acad. Sci. USSR* 1942, 35, 110.

Table I. Fluorescence Quenching of TI by Various Electron Donors

quencher	$k_q, M^{-1} s^{-1}$	$E_{1/2}(ox), V$
TEA ^a	8.2×10^9	0.96 ^d
TEA ^b	1.3×10^{10}	0.96 ^d
BNAH ^a	1.6×10^{10}	0.76 ^e
1,3-CHD ^{a,c}	4.8×10^9	1.8 ^f
1,4-CHD ^{a,c}	4.2×10^7	1.3 ^f

^aSolvent: benzene. ^bSolvent: acetonitrile. ^cCHD: cyclohexadiene. ^dFrom ref 48. ^eReference 32. ^fBard, A. J.; Lund, H., Eds. "Encyclopedia of Electrochemistry of the Elements"; Marcel Dekker: New York, 1980; Vol. XI.

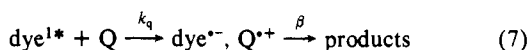
Table II. Stern-Volmer Constants and Limiting Quantum Yields (β) for Photoreduction of TI and NDI by TEA^a

dye	benzene		acetonitrile	
	$k_q \bar{C}^0, M^{-1}$	β^b	$k_q \bar{C}^0, M^{-1}$	β
TI	92 (110)	0.0041	136 (119)	0.034
NDI	34 (31)	0.01		

^aValues in parentheses were obtained from fluorescence quenching experiments. ^b β values extrapolated from double-reciprocal plot.

work of Memming and co-workers⁴¹ has shown that quenching rates show correlations between $\log k_q$ and donor oxidation potentials very similar to those observed by Rehm and Weller⁴² and many other investigators in now-classic examples of quenching by one-electron transfer.^{43,44} The reductants used in this study show quenching rate constants generally in accord (Table I) with the correlations observed by Memming and co-workers;⁴¹ in general similar rate constants were found for quenching of fluorescence of TI and NDI for the same reductants although an extensive investigation for NDI was not conducted. Quenching of the trans \rightarrow cis isomerization, which has been shown for TI and NDI to be a triplet process,⁴⁵⁻⁴⁷ is also observed concurrent with fluorescence quenching.

The correlations of $\log k_q$ with donor oxidation potential and the observation that the donors TEA and BNAH quench TI and NDI fluorescence at near-diffusion-controlled rates suggests that the observed photoreduction could be initiated by one-electron-transfer quenching of the excited singlet in each case. The simplest mechanism for such a process would be given by eq 4-7. The



quantum yield for product formation is then given by eq 8, which, upon taking reciprocals leads to eq 9 which in turn predicts a linear

$$\phi_p = \frac{\beta k_q [Q]}{k_f + k_{isc} + k_d + k_q [Q]} \quad (8)$$

$$\frac{1}{\phi_p} = \frac{1}{\beta} + \frac{1}{\beta k_q \tau_0 [Q]} \quad (9)$$

relationship between $1/\phi_p$ and $1/[Q]$ with an intercept/slope ratio = $k_q \tau_0$. Linear double-reciprocal plots were obtained for both

(41) Memming, R.; Kobs, K. *J. Phys. Chem.* **1981**, *85*, 2771.

(42) (a) Rehm, D.; Weller, A. *Isr. J. Chem.* **1970**, *8*, 259. (b) Rehm, D.; Weller, A. *Ber. Bunsenges. Phys. Chem.* **1969**, *73*, 834.

(43) Bock, C. R.; Meyer, T. J.; Whitten, D. G. *J. Am. Chem. Soc.* **1975**, *97*, 2909.

(44) Balzani, V.; Bolletta, F.; Scandola, F. *J. Am. Chem. Soc.* **1980**, *102*, 2152 and references therein.

(45) Kirsch, A. D.; Wyman, G. M. *J. Phys. Chem.* **1977**, *81*, 413.

(46) Grellmann, K. H.; Hentschel, P. *Chem. Phys. Lett.* **1978**, *53*, 545.

(47) Görner, H.; Schulte-Grohlinde, D. *Chem. Phys. Lett.* **1979**, *66*, 363.

Table III. Quantum Yields of Photoreduction of TI by TEA^a

$\phi \times 10^3$ ^b	solvents	regeneration of TI upon exposure to O ₂
2.7	benzene	yes
0.50	CHCl ₃	yes
1.9	CH ₂ Cl ₂	yes
3.8	CCl ₄	no
2.1	THF	yes
17.8	DMF	no
1.5	Me ₂ SO	yes
31.3	CH ₃ CN	yes

^a[TI] = 1×10^{-4} M, [TEA] = 0.02 M. ^bQuantum yields in solvents other than benzene were determined relative to benzene.

Table IV. Quantum Yields for Photoreduction of NDI by TEA^a

solvent	$\phi \times 10^3$	regeneration of NDI upon exposure to O ₂
benzene	4.9	very slow (days)
CHCl ₃	0.54	no
CH ₂ Cl ₂	0.39	yes
CH ₃ CN	0.34	yes

^a[TEA] = 0.02 M, [NDI] = 7×10^{-5} M.

Table V. Quantum Yields of Photoreduction of Indigo Dyes by BNAH

ϕ	indigo dye	solvent
0.13	TI ^a	benzene
0.06	TI ^a	CH ₃ CN
0.00053	NDI ^b	benzene
0.00011	NDI ^b	CH ₃ CN

^a[TI] = 5×10^{-5} M. [BNAH] = 1.0 mM. ^b[NDI] = 7×10^{-5} M. [BNAH] = 1.3 mM.

Table VI. Quantum Yields of Photoreduction of OI^a by TEA^b

ϕ	solvent
0.054	CH ₂ Cl ₂
0.082	CHCl ₃
0.022	CH ₃ CN
0.087	benzene

^a[OI] = 1×10^{-4} M. ^b[TEA] = 0.02 M.

NDI and TI and the intercept/slope ratios, as listed in Table II, were found equal, within experimental error, to the Stern-Volmer constants for fluorescence quenching. The intercept values yield β , the fraction of quenching events which lead to product; these values, listed in Table II, are quite small and indicate that for benzene and acetonitrile solutions more than 95% of the quenching leads to regeneration of the starting materials with no net reaction.

Actual quantum yields for TEA photoreduction of TI and NDI at [TEA] = 0.02 M, where quenching of the fluorescence is nearly complete, are listed in Tables III and IV. Although no quantitative correlations have been drawn, the reduction of TI by TEA appears to generally increase in efficiency with increase in solvent polarity while the reverse is observed with NDI. Considerably higher quantum efficiencies for reduction of TI were observed with BNAH; here it is not possible to obtain a double-reciprocal plot due to the presence of a thermal reaction in several solvents. Values for ϕ_{red} for BNAH photoreduction are listed in Table V.

Although OI undergoes photoreduction with several of the same reagents reactive with TI and NDI, its reaction is clearly different. OI is nonfluorescent; however, a transient having a lifetime of 42 μ s can be observed by laser flash photolysis (vide infra) in benzene; from its lifetime and the observation that it is quenched by O₂ it can be inferred that this transient is a triplet. The triplet of OI is quenched by TEA and other reductants. Quantum yields for photoreduction of OI are given in Table VI.

Photochemical ESR Experiments. Photolysis with high-intensity visible light ($\lambda > 400$ nm) of an Ar degassed solution of TI with 1.0 M TEA in benzene leads to the appearance of the ESR signal shown in Figure 5 which can be attributed to TIH \cdot ; the same ESR signal is also observed during photolysis of benzene solutions of

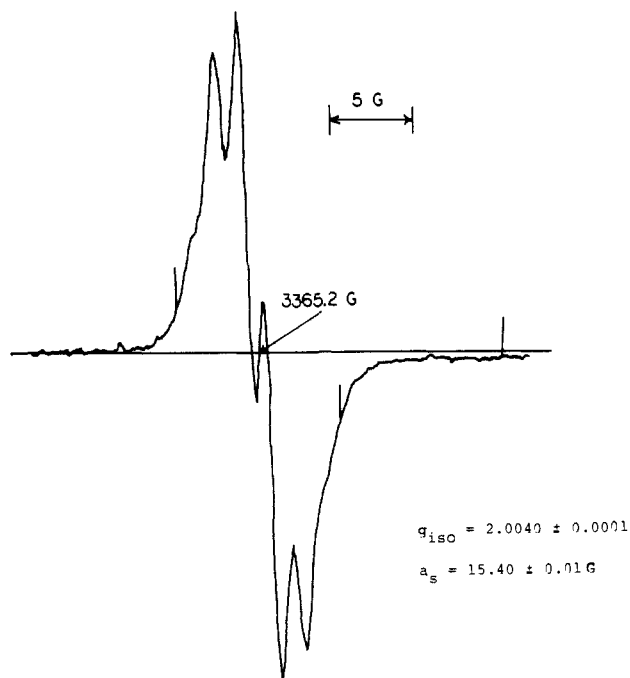
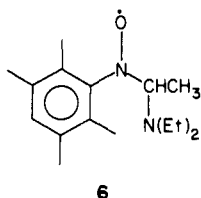


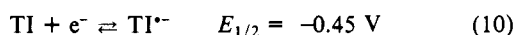
Figure 5. ESR spectrum observed during irradiation of thioindigo/triethylamine in benzene. The same spectrum is also observed during irradiation of thioindigo in the presence of other reductants.

TI with diisopropylethylamine, hydroquinone, and phenol. The $\text{TIH}\cdot$ radical is observed to decay immediately upon discontinuing irradiation and also upon addition of air to the solutions. Quantitative comparison of the intensity of the ESR signal due to $\text{TIH}\cdot$ in benzene/0.5 M TEA and acetonitrile/0.5 M TEA shows that the signal is stronger by a factor of 6 in acetonitrile. The alkylamino radical, $\text{CH}_3\text{CHNET}_2$, has not been directly observed in photo-ESR experiments on the TI/TEA system; however, addition of the spin-trap, nitrosodurene, to the solution leads to the appearance of a weak 13-line signal due to **6**; the identity of this radical is clear as it has been observed in spin-trap studies



with other photosystems involving TEA as a reducing agent.³⁴ Photo-ESR experiments in which NDI is irradiated in solution with TEA or other reducing agents *does not* lead to observation of an ESR signal. Further, spin-trap experiments with added nitrosodurene lead to the appearance of only a small signal for **6**. As noted above, photo-ESR experiments with solutions of OI and TEA have detected very strong signals for the semireduced species, **5** (Figure 4). This radical persists after irradiation, in contrast to that observed with TI/TEA. Spin-trap experiments have also detected strong signals for **6**, further supporting the hypothesis that photoinduced net hydrogen atom transfer occurs as noted earlier.

Electrochemical Studies Yeh and Bard have presented a detailed account of the electrochemical behavior of TI on both qualitative and quantitative levels;^{28,29} we have repeated and extended some of these experiments for TI. Cyclic voltammetry for TI in DMF with reductive scanning to -0.65 V (all potentials vs. SSCE) shows one reversible wave with $E_{1/2} = -0.45$ V. This wave is attributed to reduction of TI forming an anion radical (eq 10). Upon scanning to more cathodic potentials the CV



becomes more complex; in addition to the first wave a second mostly

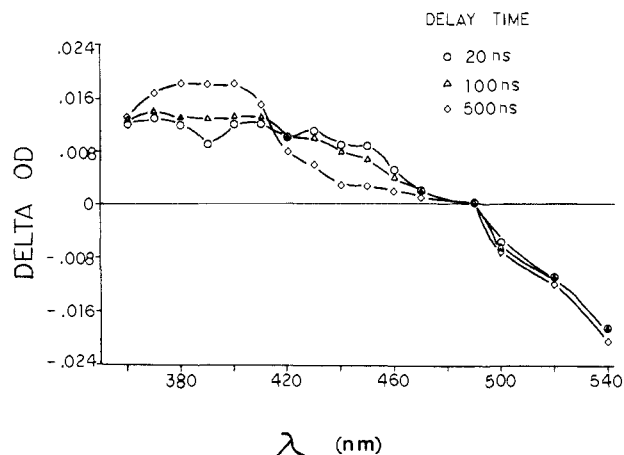
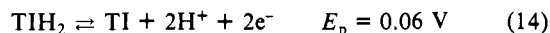
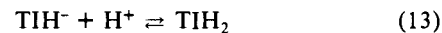
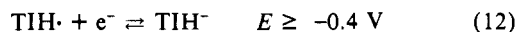


Figure 6. Difference spectra observed in laser-flash photolysis of thioindigo/triethylamine in benzene at different time intervals after the pulse.

irreversible reduction is noted at $E_{1/2} = -1.1$ V and a new oxidative wave is found at 0.03 V. The second cathodic wave is attributed to reduction of $\text{TI}^{\cdot -}$, to the dianion TI^{2-} ; the wave is irreversible due to the fact that TI^{2-} conproportionates with TI to form $\text{TI}^{\cdot -}$. Yeh and Bard have proposed that the cathodic wave at 0.05 V is due to oxidation of a dimer dianion, TI_2^{2-} , which is in equilibrium with $\text{TI}^{\cdot -}$.²⁸

Addition of benzoic acid to the DMF solution of TI causes pronounced changes in the electrochemical behavior of TI. A quasireversible reduction is observed; a cathodic wave appears at -0.42 V and the corresponding anodic wave occurs at 0.06 V. Yeh and Bard have shown that the cathodic wave has $n_{\text{eff}} = 2$, indicating that the reduction involves transfer of two electrons.²⁸ The CV is accounted for by eq 10–14. Electrochemical reduction



under these conditions leads to spectrally detectable TIH_2 ; the CV suggests that $\text{TIH}\cdot$ is spontaneously reduced to TIH^- : at an applied potential of -0.4 V. Alternatively TIH_2 could be formed by disproportionation (eq 15); this could also account for formation of TIH_2 electrochemically with $n_{\text{eff}} = 2$.



NDI shows similar electrochemical behavior; in DMF there are two reversible waves at $E_{1/2} = -0.4$ and -0.95 V. Addition of benzoic acid results in only a single irreversible wave at -0.48 V for a cathodic scan to -1.0 V; a small oxidative wave on return scan is observed at 0.1 V. As noted above, electrolysis of NDI with added benzoic acid results in formation of NDIH_2 ; a reaction sequence similar to that proposed for TI (eq 10–14) can account for these results. The excited singlet reduction potentials for TI and NDI are estimated to be $+1.7$ and $+1.6$ V, respectively.

Laser Flash Photolysis Experiments. Figure 6 shows the time resolved absorbance spectrum which was obtained for a benzene solution of TI with 1.0 M TEA. Immediately after the laser pulse (20-ns delay), a broad transient absorbance is observed which extends from 360 to 500 nm. With increased time delay the transient absorbance evolves to a more resolved absorption centered at 390 nm and extending to 450 nm. The spectrum which is shown for a 500-ns delay persists out to the maximum time delay obtainable (50 μs). Concurrent with the appearance of the transient absorbance, a bleaching is noted for $\lambda > 500$ nm due to disappearance of TI. Time evolution of the absorbance at 390 and 460 nm is shown in Figure 7. At 460 nm a transient absorbance is noted; the transient decays to 20% of the initial value with a first-order rate constant $k = 3.6 \times 10^6 \text{ s}^{-1}$ ($\tau = 280$ ns) and remains constant out to long times. At 390 nm an initial step is

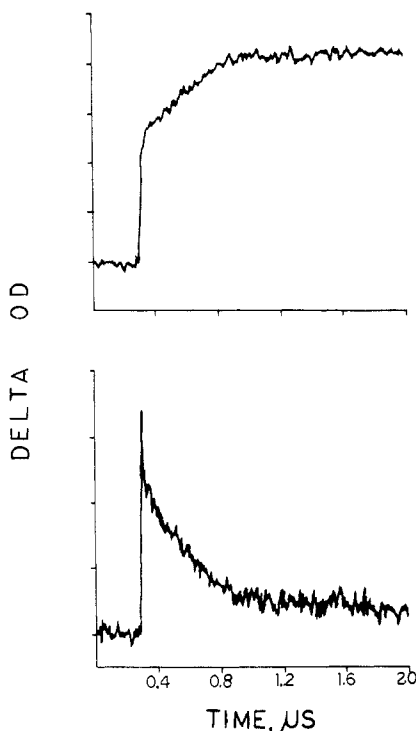


Figure 7. Time evolution of transients from thioindigo/triethylamine in benzene at different wavelengths: upper trace, $\lambda = 390$ nm; lower trace, $\lambda = 460$ nm.

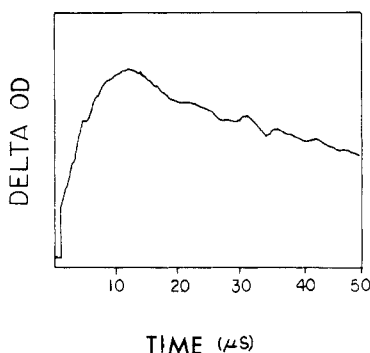


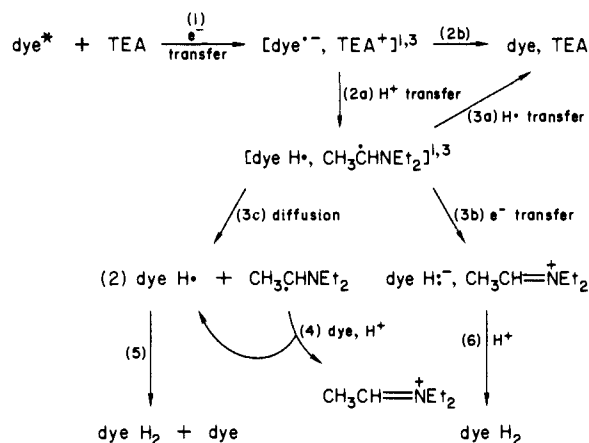
Figure 8. Conventional flash photolysis of thioindigo/triethylamine in acetonitrile; transient formation and decay at 420 nm.

noted with the laser pulse followed by a grow-in concurrent with the above decay. The 460-nm transient is attributed to the species $\text{TIH}\cdot$ as first a radical pair ($\text{TIH}\cdot\cdots\text{CH}_3\text{CHNEt}_2$) and subsequently the free radical. The 390-nm absorbance appears due to both $\text{TIH}\cdot$ and TIH_2 , but primarily to the latter at long times.

The laser flash experiments suggest that two pathways exist for formation of TIH_2 from the radical pair formed via photoinduced electron transfer–proton transfer as will be discussed later. The residual absorbance at 460 nm can be attributed to the free radical, $\text{TIH}\cdot$, which forms TIH_2 by disproportionation. These radicals have been detected in the photo-ESR experiments described above. However, the bulk of the product formed in benzene is generated too rapidly to occur via this second-order process. The relatively rapid reaction of $\text{TIH}\cdot$ from the radical pair which competes with free radical formation is evidently a second electron transfer occurring within the radical pair (hence first order with $k = 3.6 \times 10^6 \text{ s}^{-1}$), as outlined below. This path provides a two-electron reduction or net *hydride transfer* from the tertiary amine to TI before the radicals diffuse apart.

Laser flash photolysis experiments with TI/1.0 M TEA in acetonitrile gave quite different results from those which have been described with benzene. A strong transient absorbance attributed to $\text{TIH}\cdot$ was observed with the temporal profile shown in Figure 8. A small step is noted concurrent with the laser pulse which is followed by a slow increase in the signal over 10 μs ; the transient

Scheme II



subsequently decays over a 50- μs time scale.

As indicated above BNAH photoreduces TI to TIH_2 . Here conventional flash photolysis reveals a relatively longer persisting photoreaction; following flash excitation of TI a bleaching of ground-state TI is observed at 546 nm. A return to a new baseline (due to “permanent” reaction of TI) occurs with a $t_{1/2} = 1$ ms, considerably slower than the reactions observed for TI/TEA in either benzene or acetonitrile.

Laser-flash experiments with NDI and OI and TEA lead to quite different results. With the apparatus used for TI we detect no transients for NDI/TEA; attempts to observe the time scale of product formation are not very conclusive due to the relatively weak absorption of the product in accessible regions of the spectrum. With OI and TEA laser flash experiments show (a) that the triplet is completely quenched and (b) the product formation appears to be a virtual step-function with only a small increase subsequent to the laser-flash.

Discussion

Photoreduction of TI by TEA. The results presented above for reduction of TI lead to a fairly clear picture of the mechanisms leading to its photoreduction by TEA. It is clear that quenching of the excited singlet, $^1\text{TI}^*$, by the amine leads ultimately to the two-electron reduction product TIH_2 (or its conjugate base, $\text{TIH}^{\bullet-}$). The most reasonable quenching process is one-electron transfer to produce the radical ion pair $\text{TI}^{\bullet-}, \text{TEA}^+$; although this pair is generated from two singlets, intersystem crossing could occur so that the occurrence of two radical ion pairs, one singlet and one triplet, should be considered. If we assume proton transfer rapidly converts most of the initially formed ion pairs into a caged radical pair, again both singlet and triplet pairs are likely involved, the overall reactions occurring are outlined in Scheme II.

Steps 1 and 2 should occur as outlined above; both the radical ion pair and the neutral radical pair could return to starting materials (steps 2b and 3a, respectively) by electron- or atom-transfer processes which account for the relatively low overall efficiencies observed. Diffusion apart of the radical pair (step 3c) should result in product formation via the energetically favored⁴⁸ dark reduction of TI by the amine radical (step 4) and by disproportionation of the semireduced radical, $\text{TIH}\cdot$ (step 5). Alternatively, product formation can occur by a second electron transfer (step 3b) within the geminate pair.⁴⁹ The sequence culminating in step 3b, electron–proton–electron transfer, amounts to a net hydride ion transfer process.

For thioindigo in nonpolar solvents the laser flash photolysis results give a clear indication that most (up to ca 80% in benzene) of the product is formed by a process much too fast (and first order) to be consistent with radical disproportionation (step 5).¹

(48) Monserrat, K.; Foreman, T. K.; Grätzel, M.; Whitten, D. G. *J. Am. Chem. Soc.* **1981**, *103*, 6667.

(49) As suggested by a reviewer, an alternate path for the second in-cage reduction could be disproportionation to generate Dye H_2 and $\text{CH}_2=\text{CH}-\text{NEt}_2$.

The rapid-product formation appears to consist of two steps, one unresolvable by the flash and a second on the 100-ns time scale. A reasonable interpretation of the results for thioindigo is that, even though dye singlets are quenched, the reaction by paths 2 and 3 is much slower if a triplet pair is involved where processes 2b, 3a, and 3b should be spin-retarded. The slower bimolecular decay of dye H \cdot for thioindigo (concurrent with product formation) is clearly due to the semireduced radical disproportionation (step 5). This path to product, although readily quenched by radical scavengers such as O $_2$, is efficient since the free species dye H \cdot , in the absence of scavengers, can only lead to product. For thioindigo the quantum yields are higher in polar solvents where it is clear by ESR studies that formation of free dye H \cdot is more efficient. This is reasonable in view of the irreversibility of step 5.

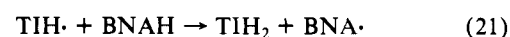
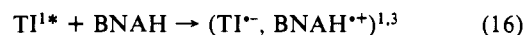
The formation of the two-electron reduction product, TIH $_2$ (or dye H $_2$), by the direct sequence (1), (2a), (3b), and (6) is interesting in that it represents a coupling of congruent electrochemical processes. For the partners in the photoinduced electron-transfer reaction between TI and TEA both species undergo "ece" reactions⁵⁰ due to the reactivity of the radical ions formed by the one-electron redox processes. Thus TI $^{\cdot-}$ is a strong base and its protonation converts it to a semireduced species, TIH \cdot , which is more easily reduced than TI itself. Conversely, TEA $^{+\cdot}$ is a strong acid whose deprotonation product, CH $_3$ CHNET $_2$, is a stronger reductant than TEA. The coupling of these processes in the initially formed radical ion pair allows for a direct two-electron redox process to occur by what amounts to a net hydride ion transfer. This novel result should, in principal, be fairly general for other donor-acceptor pairs⁵¹ and could lead to highly efficient redox processes involving net conversion and storage of energy. However, the low efficiencies observed for TI-TEA indicate that there are several ways of regenerating the starting material in net energy wasting processes, even though the overall reaction is clean and efficient as to chemical conversion. Key "energy-wasting" steps likely include (2b) and (3a). The relative importance of back electron transfer vs. hydrogen atom transfer is hard to assess but it appears likely that (2b) should be extremely important in a number of cases, particularly when singlet pairs are involved or when proton transfer (TEA $^{+\cdot}$ is a carbon acid) is slow. The question of proton transfer rates for TEA $^{+\cdot}$ and similar radical ions appears at this point not fully resolved. Although there are indications that it may be very rapid by some picosecond studies,¹⁹⁻²² we have recently found that some radical cations derived from tertiary amines can undergo C-C bond cleavage in competition with deprotonation.⁵²

The comparative photoreductions of NDI and OI by TEA offer interesting contrasts to that of TI. Since NDI and TI have comparable excited singlet energies and electrochemical behavior and are both quenched by TEA with similar rates to give the two-electron products dye H $_2$, it would be reasonable to infer that they react by similar mechanisms.⁵³ In fact several features of the reaction of NDI are quite different. First, in ESR studies no direct production of a semireduced dye, dye H \cdot , can be observed. Experiments with the spin-trap nitrosodurene lead to a weak radical signal having hyperfine splitting consistent with the adduct of ND to CH $_3$ CHNET $_2$ which has been easily observed in other photoredox processes involving TEA.⁵⁴ Laser flash photolysis experiments with NDI-TEA also reveal no long-lived transients. Although it is difficult to follow the appearance of product due both to the low efficiency of its formation and its relatively weak absorption (compared to TIH $_2$), it appears that most of the product is produced in a time too short (~ 10 ns) to be resolved by our facilities. These observations can be most consistently explained

by assuming that all of the reaction between NDI and TEA occurs by the net two-electron transfer sequence described by (1), (2a), (3b), and (6). The low efficiency and lack of a pronounced solvent effect are in accord with the radical pair sequence and considerable inefficiency via (2b) and (3a). One possible explanation for the different behavior of TI and NDI is that for the former the sulfur induces some intersystem crossing so that both singlet and triplet pairs are involved while for NDI quenching of the singlet leads to only short-lived singlet pairs which rapidly form product but even more rapidly and efficiently return to the starting materials.

The reaction of OI is quite different; the chief difference is that the photoredox process with OI originates from triplet dye and the net result is a relatively more efficient 1-electron-1-proton transfer which culminates with the accumulation of dye H \cdot ($\phi_{\text{red}} = 0.144$). The accumulation of only a single radical can be explained by the occurrence of steps 1, 2, 3c, and 4 for OI as outlined in Scheme I. The key point here is that neither step 3b, the second electron transfer, or radical disproportionation (step 5) occur. That OI should behave in such a manner can be readily understood in view of its electrochemistry. In anhydrous DMF OI shows one-electron reduction waves at -0.56 and -1.56 V vs. SSCE; addition of benzoic acid shifts these waves to -0.33 and -1.29 V so that although protonation renders both reductions more facile, the second reduction is still relatively unfavorable and no process analogous to the "ece" reactions for TI and NDI can occur. The potentials given above also indicate that equilibrium should not favor disproportionation and hence although the amine radical rapidly promotes the dark reduction of OI (step 4), the radical accumulates and undergoes slow decomposition under prolonged photolysis.

The reaction of the three dyes with BNAH, a much stronger and potentially reversible reductant, also shows some interesting contrasts. For TI, photolysis in the presence of BNAH leads to TIH $_2$ with a quantum efficiency of 0.15 in benzene; in comparison NDI undergoes photoreduction with BNAH but the quantum efficiencies are extremely low ($\sim 10^{-4}$) and OI does not give OIH \cdot as with TEA but undergoes dark reaction with BNAH to yield a product whose structure has not been determined. BNAH has generally been found to act as a net hydride donor in other photoreductions⁵⁴⁻⁵⁹ and it has been suggested that in some cases it can participate in a radical chain process.^{60,61} A reasonable sequence of reactions in the photoreduction of TI by BNAH is given by eq 16-22. Thus although the "direct" two-electron



reduction according to Scheme II or eq 16-18 is possible, any radicals escaping after one-electron reduction (eq 19) should be

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able to promote a chain reaction promoting reduction of TI in the dark until termination via eq 22 ends the process. Evidence for such a process is found by following the bleaching of TI absorption at 546 nm by flash photolysis. In contrast to the experiments with TI-TEA, the bleaching is much slower and persists up to 2 ms following the flash. However, in contrast to TI-TEA no ESR signal for $\text{TIH}\cdot$ is observed when solutions of TI and BNAH are irradiated; in fact steady photolysis in the ESR spectrometer reveals no readily detectable radical signals. However, photochemical ESR experiments with TI-BNAH in acetonitrile to which the spin trap nitrosodurene has been added lead to a complex spectrum which can be shown to consist of the spectrum of $\text{TIH}\cdot$ superimposed on a strong six-line spectrum attributable to the adduct of $\text{BNA}\cdot$ to ND. Addition of ND also quenches appreciably the conversion of TI to TIH_2 by BNAH.

The much lower quantum efficiency for reduction of NDI by BNAH can probably be attributed to the lack of production of kinetically "free" radical by a step analogous to eq 19. Thus for NDI with BNAH all reaction probably proceeds via a singlet radical pair which efficiently decays to starting material or forms products but in either case does not survive long enough to yield radicals. The contrast in reaction efficiencies between TI and NDI is especially remarkable in that the two dyes both have apparently the same potential for the chain reaction (e.g., similar quenching efficiencies, excited state energies, and redox potentials) but the

evident lack of leakage from quenched singlets to long-lived triplet radical pairs for NDI prevents this path from occurring.

The overall patterns of reactivity observed in this study with potentially electron accepting indigo dyes and donors such as TEA and BNAH are ones which should be fairly general. The individual reactivity of both the electron acceptors and donors insofar as the various electron and proton transfer steps are concerned is not unusual and indeed several similar reaction sequences have been observed with other chromophores. What is more unique about the present results is the coupled sequence of electron-proton-electron transfer resulting from encounter of an excited acceptor with a donor. The rapid occurrence of such a process under conditions not especially sensitive to free radical scavengers or other reagents could offer an attractive route for accomplishing chemically useful conversions. Although relatively low quantum efficiencies are associated with the "concerted" process in the present cases, it is reasonable to anticipate that future investigations can lead to reactive combinations that can result in net energy storage with relatively high efficiencies.

Acknowledgment. We are grateful to the U.S. Department of Energy (contract DEAC02-84ER13151) for support of this research.

Registry No. 1, 522-75-8; 2, 33934-64-4; 3, 2533-00-8; TEA, 121-44-8; 1-benzyl-1,4-dihydropyridinamide, 952-92-1.

Sequential General Base-Acid Catalysis in the Hydrolysis of RNA by Imidazole¹

Ronald Breslow* and Marc Labelle²

Contribution from the Department of Chemistry, Columbia University, New York, New York 10027. Received October 4, 1985

Abstract: The cleavage of poly-U by imidazole buffer was studied by using a recently described kinetic assay technique. A bell-shaped profile is seen for rate vs. buffer protonation state, but the reaction is kinetically first order in buffer. It is proposed that a sequential base-acid mechanism is involved: the base catalyzes formation of a 5-coordinate phosphorus isomer of the starting material by transferring a proton from nucleophilic hydroxyl to equatorial oxygen, and the acid then catalyzes the forward decomposition of that intermediate to form the products. Some kinetically equivalent alternative mechanisms have been excluded. The proton transfer catalysis mechanism is suggested for the enzyme ribonuclease as well.

The hydrolysis of RNA by bovine pancreatic ribonuclease involves catalysis by an imidazole group acting as a general base and an imidazolium group acting as a general acid.³ Additional binding interactions are present, and a lysine cation may act as a third catalytic group by coordinating to the phosphate anion of RNA. The hydrolytic sequence involves two steps. In the first one there is a transesterification in which the 2'-hydroxyl group of a ribose unit attacks the phosphate ester, with the formation of a 2'-3' cyclic phosphate and the cleavage of the RNA chain. In a second step the enzyme catalyzes the hydrolysis of this cyclic phosphate intermediate.

Both steps apparently have similar mechanisms, involving the general base catalysis by imidazole and general acid catalysis by imidazolium ion. A principal piece of evidence for this is the bell-shaped pH vs. rate profile, with a rate maximum when the

base group is unprotonated and the acid group carries its proton. However, there is also evidence about these mechanisms from X-ray studies of complexes of the enzyme with substrate and with transition-state analogues.⁴

We have described the bifunctional catalysis of a phosphate ester hydrolysis related to that performed by the enzyme ribonuclease.⁵ We mounted two imidazole groups on the primary face of β -cyclodextrin and showed that this catalyst would hydrolyze a cyclic phosphate ester of 4-*tert*-butylcatechol. The selectivity of the hydrolysis could be understood in terms of the geometry expected for general base delivery of a water molecule to the phosphate group.⁶ The pH vs. rate profile for this reaction also was bell-shaped as in the enzyme, indicating that the catalysis

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(1) Support of this work by the NIH is gratefully acknowledged.

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