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Phase Transition Autocatalysis of the Hydrolysis of Some Esters of Azo Dyes

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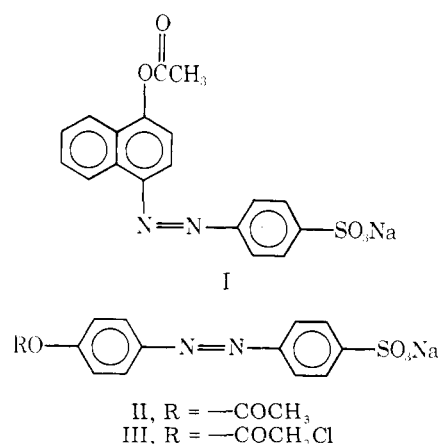
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Abstract: Acetate and chloroacetate esters of some water-soluble azonaphthol and azophenol dyes form supersaturated solutions at concentrations as low as 10^{-5} M when aqueous solutions are mixed with buffers to give final ionic strengths as low as 0.01. Nucleation occurs slowly in the presence of atmospheric dust to give dilute colloidal dispersions of particles having Stokes radii of 0.3–0.4 μm . In the absence of dust, nucleation does not occur for periods up to 2 weeks. At neutral pH values, where hydrolysis of the acetates in homogeneous solution is very slow, formation of the colloidal particles is accompanied by accelerations by factors of 80–100. When nucleation takes place before appreciable hydrolysis has occurred in the supersaturated solution, the result is autocatalysis. Under conditions of higher pH or with more reactive chloroacetate esters, all the hydrolysis occurs in the homogeneous solution before nucleation can take place and autocatalysis is not observed. Ultrafiltration, seeding, and light-scattering experiments are described which establish that the autocatalysis results from formation of the colloidal phase.

A number of studies have shown that many amphiphilic azo dyes form association colloids in aqueous solution.¹⁻⁶ Those that form such colloids most readily are usually complex molecules such as Congo red or benzopurpurin with molecular weights of around 700 or greater. The presence of electrolytes facilitates colloid formation. On the other hand, many studies of simpler dyes with molecular weights around 400, such as the sulfonated phenylazonaphthols, have shown that in the absence of high electrolyte concentrations, the degree of aggregation is quite low.^{1,2,6-10} Many properties of the solutions of the low molecular weight dyes have been interpreted in terms of a monomer-dimer equilibrium to relatively high dye concentrations.⁷⁻¹⁰ What has apparently gone unnoticed, however, is the fact that a number of the simpler dyes readily form metastable solutions in water or dilute buffers and yield colloidal aggregates on aging. Light scattering results from these laboratories have shown that a number of simple dyes form colloidal dispersions from supersaturated solutions at surprisingly low concentrations.¹¹ Nucleation to particle sizes of less than 1 μm may occur quite slowly and is promoted by the presence of atmospheric dust or some other suitable surface. The simplest dyes such as methyl orange or orange II may nucleate to colloidal dispersions in the vicinity of a roughened Wilhelmy plate at concentrations where the dyes have been thought to be soluble.¹²

We now wish to report that these phase transitions occur in buffered aqueous solutions of esters of simple dyes. The formation of the colloids can accelerate by factors of 80–100 hydrolysis reactions that are quite slow in homogeneous solution. Since the phase transition involves the reactant and occurs during the hydrolysis, the result is an autocatalysis of the hy-

drolysis. To our knowledge, there have been no previous reports of either nucleation from supersaturated ester solutions or of autocatalysis of ester hydrolysis by any mechanism. We report kinetic results for the hydrolysis of esters I–III which are re-



spectively the acetates or chloroacetates of the dyes orange I (IV) and 4'-hydroxyphenylazobenzene-4-sulfonic acid (V).

Experimental Section

Orange I acetate was material used in an earlier study.¹³ Dye V was esterified in *N,N*-dimethylformamide solution by treatment with an excess of the appropriate acid anhydride, using triethylamine as catalyst. The dye was purified beforehand by recrystallization of the sodium salt until a single band was obtained by a high-resolution chromatographic method.¹⁴

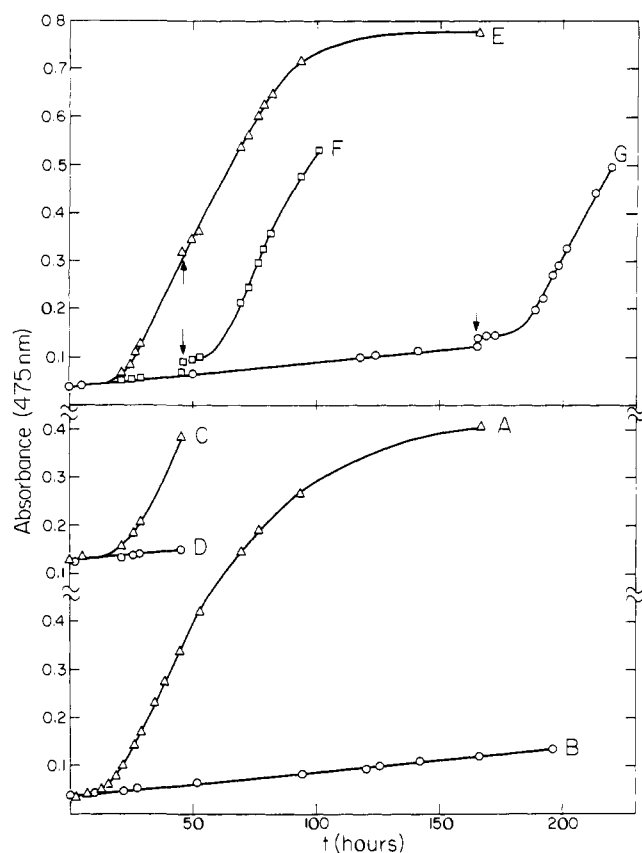


Figure 1. Plots of absorbance of IV as a function of time from hydrolysis of I in MES buffer, pH 6.2, $\mu = 0.01$, 25 °C. Curve A, formation of product in an unfiltered solution; curve B, formation of product in a dust-free solution filtered through an ultrafilter of 0.2 μm pore size; curves C and D, the effect of 10% added product on the initial rates of an unfiltered (curve C) and a filtered solution (curve D); curves E, F, and G, the effects of seeding previously filtered solutions (curves F and G) with solution taken from an unfiltered solution (curve E). Vertical arrows indicate times at which the seed solution was removed from E and added to F and G.

Where the rates were slow enough the hydrolyses were followed on a Cary 118C recording spectrophotometer. Complete absorption curves of reactants and products were recorded when convenient. Rapid rates were monitored on a Durrum stopped-flow spectrophotometer. In all cases, reaction was begun by mixing equal volumes of a solution of ester in water with buffer or sodium hydroxide solution to give a final ionic strength of 0.01.

The light-scattering intensities and the correlation function of inelastically scattered light were measured on a prototype photometer of conventional design which utilizes photon counting and digital signal processing. The scattering solutions were irradiated with the 632.8-nm line from a helium-neon laser. Neither the ester nor the dye absorbs at this wavelength. The intensity of scattered light was measured at an angle of 22°. Solutions for light-scattering measurements were first mixed in buffer and were then filtered through Nuclepore ultrafiltration membranes of 0.2- μm pore size. The presence of electrolyte facilitated dust removal. The solutions were filtered directly into dust-free scattering cells through seals of aluminum foil.

Results and Discussion

Good pseudo-first-order kinetics were observed for the hydrolysis of III over the entire pH range studied (5.0–12.3). Rate constants for the base-catalyzed (k_{OH}) and uncatalyzed (k_0) reactions are $1.26 \times 10^3 \text{ mol L}^{-1} \text{ s}^{-1}$ and $1.2 \times 10^{-4} \text{ s}^{-1}$, respectively. The longest half-time measured for III was 2.4 h. Pseudo-first-order kinetics were observed for I only in the pH range of 9.0–14.0; $k_{\text{OH}} = 3.80 \text{ mol L}^{-1} \text{ s}^{-1}$. Where complete absorption curves could be measured during hydrolysis, excellent isosbestic points were observed for I at pH >9.0 and for III at all pH values. Pseudo-first-order rate constants ob-

tained at pH 9.2 ($\mu = 0.01$) at five concentrations of I in the range 3×10^6 to $5 \times 10^5 \text{ M}$ showed only small random variation (standard deviation = $\pm 2\%$). There is thus no kinetic evidence for significant aggregation of I in freshly prepared buffered solutions.

At concentrations of I and II around $2 \times 10^{-5} \text{ M}$ and in neutral or slightly acid buffers of low ionic strength ($\mu = 0.01$) plots of absorbance of product as a function of time were S-shaped. A typical plot for the hydrolysis of I in a buffer of 2-*N*-morpholinoethanesulfonic acid (MES) is shown in Figure 1, curve A. The initial rates were slow; the induction periods were usually 10–20 h but were variable and unreproducible. Sigmoid plots were obtained at neutral pH in MES, phosphate, and *N*-methylmorpholine buffers. Complete absorption curves taken during the course of hydrolysis in the neutral pH region failed to give sharp isosbestic points.

Attempts were made to model the hydrolysis of I by computer simulation according to eq 1, since this mechanism will give S-shaped plots of product concentration vs. time if the intermediate X is allowed to accumulate. Complete digitized absorption curves were used for the analysis.



Even when the three rate constants and the unknown absorption curve of X were allowed to vary as adjustable parameters, the mechanism given by eq 1 was inadequate to reproduce the experimental absorption curves as a function of time.

The solutions remained clear to transmitted light following the induction period but the acceleration was accompanied by an increase in light scattering at low angles. If the solutions were filtered immediately after mixing the stock solution of I in water with buffer, all atmospheric dust was removed and autocatalysis was eliminated. The rates of hydrolysis of the filtered solutions were then equal to the initial rates of the unfiltered solutions in the same buffer (Figure 1, plot B). No light scattering developed in the filtered solutions.

Addition of hydrolysis product (IV) to the extent of 10% of the ester concentration at the beginning of a run failed to change the initial rate. Curve D of Figure 1 shows that in the presence of added product the initial rate of a filtered solution is the same as that of a filtered solution without added product. The plot of absorbance as a function of time for an unfiltered solution shows the same induction period and initial rate (curve C) as was found for an unfiltered solution containing no product. Thus the autocatalysis is not the result of an interaction between reactant and product.

Seeding experiments were carried out to establish that the autocatalysis results from nucleation of the esters to colloidal dispersions, catalyzed by atmospheric dust. Two hydrolysis runs of I were started at different times in filtered MES buffer, pH 6.2. After approximately 1 and 6 days of hydrolysis, respectively, a third run was begun in an unfiltered MES buffer to provide seeds. The results are shown in plots E, F, and G of Figure 1. At the times indicated by vertical arrows, 0.3 mL was removed from the unfiltered solution showing autocatalysis and was injected into 3.0 mL of the filtered solutions through aluminum foil seals. Autocatalysis was induced in the two filtered solutions. Both of the seeded solutions ultimately achieved the same maximum rate as the unfiltered solution. It is significant, however, that the accelerations were not seen immediately upon adding the seeds. Induction periods still follow the seeding, indicating that additional processes must occur before the onset of autocatalysis. These processes may involve adsorption or further nucleation and particle growth to give more surfaces.

Parallel light-scattering and absorbance measurements were made on seeded solutions to show that particle growth must accompany or precede the acceleration of hydrolysis. The

hydrolysis was monitored by following the absorbance increase at the wavelength of maximum absorbance of the dye product, IV (475 nm). Portions of a filtered solution of I (2.5×10^{-5} M) in MES buffer, pH 6.2, were introduced into separate dust-free absorption and scattering cells and the uncatalyzed rate and background scattering were monitored for 40 h. The background scattering remained constant. Both solutions were then seeded with 0.3 mL from an unfiltered solution showing autocatalysis and light scattering. The results are shown in Figure 2, plots A and B. The scattering intensity is in units of photon counts per second (cps); the ordinates of the two plots bear no quantitative relationship to each other. Both plots show induction periods following seeding. If one assumes that identical solutions seeded in identical ways will nucleate at about the same rate, the experiment indicates that nucleation and particle growth precede autocatalysis. Until simultaneous scattering and absorbance measurements can be made on the same solution, we are reluctant to attach quantitative significance to the time lag between particle growth and hydrolysis shown in Figure 2. We are not presently equipped to make such simultaneous measurements.

The colloidal particles probably consist of both ester and product, depending upon the extent of hydrolysis. We have shown that both the ester and pure orange I dye give supersaturated solutions and nucleate slowly in neutral pH buffers of low ionic strength, at concentrations similar to those used for the kinetic measurements. By means of inelastic light-scattering measurements, an average diffusion coefficient of $6.1 \times 10^7 \text{ cm}^2 \text{ s}^{-1}$ was determined for the colloidal particles present 4 days after seeding. At this time, the scattering intensity had become constant and the hydrolysis was 75% complete (Figure 2). From the value of the diffusion coefficient an average Stokes radius of $0.35 \mu\text{m}$ was computed for the particles.

To determine whether autocatalysis could be suppressed, once initiated, by removal of the catalytic particles, an initially unfiltered solution was allowed to achieve its maximum autocatalytic rate and was filtered through an ultrafilter after 45% reaction. No low-angle scattering was detected after filtration. The results are shown in Figure 2, plot C. Removal of the colloidal particles suppressed autocatalysis temporarily, but after an induction period the autocatalytic acceleration resumed. Assuming that no catalytic particles were introduced inadvertently, the experiment shows that nuclei small enough to pass through the $0.2\text{-}\mu\text{m}$ pores are sufficient to promote further growth of particles to catalytic size.

The results show that the autocatalysis in neutral and slightly acidic buffers arises from heterogeneous catalysis of the hydrolysis on the surface of colloidal particles of the reactant and/or product which form from initially supersaturated solutions. Autocatalysis is not observed with ester III at any pH or with I and II in alkaline solution because the nucleation is slow and hydrolysis is complete during times when the esters remain in homogeneous supersaturated solutions. The autocatalysis is evident only when hydrolysis is slow enough that nucleation can occur and the heterogeneous catalysis can be superimposed onto the homogeneous reaction.

Whereas there are several possible mechanisms for the heterogeneous catalysis, we would like to speculate on an intriguing possibility to explain why the presence of the colloidal particles causes the marked rate accelerations. A number of ionic dyes are known to align themselves on the surfaces of charged colloids with the flat planes of the molecules somewhat parallel. When such molecular alignment also results in

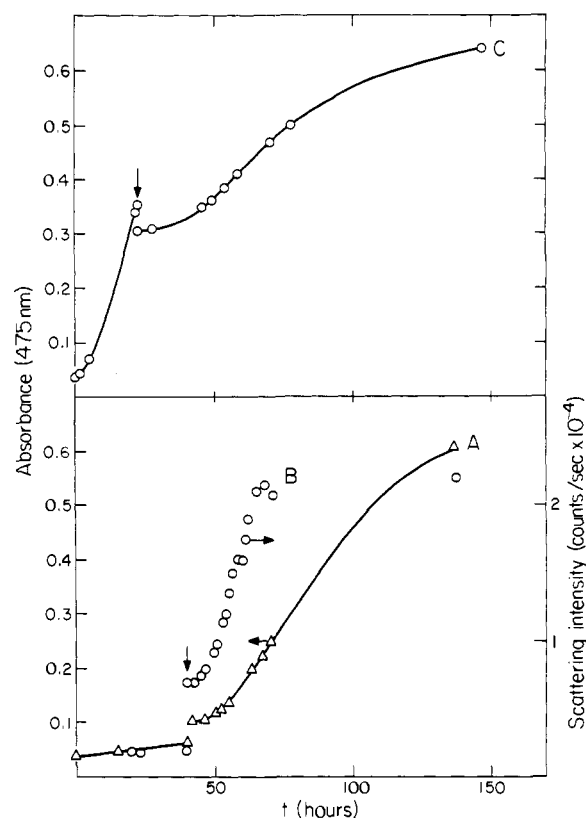


Figure 2. Hydrolysis of I in MES buffer, pH 6.2, $\mu = 0.01$, 25°C . Curves A and B, comparison of hydrolysis rate (curve A) with the rate of increase in light-scattering intensity (curve B) following the seeding of previously filtered solutions; curve C, the effect of filtering an initially unfiltered solution on the course of hydrolysis. The vertical arrows indicate the points at which solutions were seeded or filtered.

alignment of the transition dipoles of closely spaced dyes, spectral shifts known as metachromasy result.¹⁵ If the esters are aligned in a similar way at the surface of the colloidal particles, the opportunity exists for catalysis from a proximity effect. The hydrolysis of an ester in the organized array produces a phenol which might then act as a general acid for catalyzing hydrolysis of its near neighbor ester, etc. The particles might serve as templates to hold the substrate molecules in close proximity and proper alignment for the "zipper effect" to operate.

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