

a measure of the effective electrode area, current density at the outset of a typical experiment amounted to 9.8 mA/cm². This decreased continuously during the experiment to a constant value of 9 μ A/cm². Product analyses for Pr₂NH, PrNH₂, and Et-CHO were performed by glpc using Dowfax 9N9 with NaOH and also Carbowax 20M as liquid phases. These were repeated at 75, 100, and 150°.

Identification of Propionaldehyde-2-d.—A 150-ml MeCN solution, 0.25 M in NaClO₄, 230 mM in D₂O, and 50 mM in dipropylamine, was electrolyzed at a Pt anode at +1.00 V vs. Ag-AgNO₃ (0.10 M). The product mixture was distilled through a spinning-band column, the first 12 ml of distillate being taken at 72–74°. This fraction was redistilled with the separation efficiency monitored by glpc. A 2-ml cut contained most of the aldehyde; it was further fractionated by glpc, using a Carbowax 20M column at 100°. The aldehyde fraction was trapped and taken up in CDCl₃ for nmr and ir examination. A sample of propionaldehyde in MeCN was subjected to the same separation to ascertain that the separation procedure was not causing exchange. There was indication that some of the aldehyde was lost because of condensation at various stages.

The mass spectra were obtained by distilling some of the concentrated solution produced from the spinning-band column into the spectrometer inlet system while the fraction was maintained below ice temperature. It was ascertained, using a sample of propionaldehyde in MeCN of the appropriate concentration, that the molecular ion of the aldehyde is observed by this procedure. However, the solvent peaks obscure the fragmentation pattern of the aldehyde.

Preparation of 2-Methyl-2-pentenalpropylamine.—A sample of this product was prepared to provide a comparison with the electrolytically generated material by the procedure previously described.¹

Registry No.—Di-*n*-propylamine, 142-84-7.

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Mechanism of the Ferricyanide-Catalyzed Chemiluminescence of Luminol

PHILIP B. SHEVLIN AND HAROLD A. NEUFELD

Biological Sciences Laboratory, Fort Detrick, Frederick, Maryland 21701

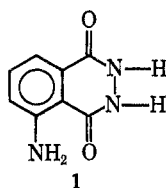
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A study of the mechanism of the potassium ferricyanide chemiluminescent oxidation of luminol (5-amino-2,3-dihydro-1,4-phthalazinedione) is reported. An important feature of the proposed mechanism is the one-electron oxidation of the luminol dianion by ferricyanide to 5-aminophthalazine-1,4-semidione. The semidione intermediate may react with oxygen to produce electronically excited 3-aminophthalic acid and nitrogen. Alternatively, the semidione may be further oxidized by ferricyanide in a nonluminescent reaction.

In chemiluminescent systems, the rate of formation of the excited state is given by

$$d[x]/dt = i/\phi \quad (1)$$

where *x* is the luminescing molecule, ϕ the quantum yield for fluorescence of *x*, and *i* the intensity of emitted light. Thus, in systems where ϕ does not change during the reaction, the intensity is a measure of the rate of reaction. We have used this relationship to study the mechanism of the potassium ferricyanide oxidation of luminol (5-amino-2,3-dihydro-1,4-phthalazinedione, 1) in aqueous base.



The chemiluminescence of luminol has been the subject of numerous investigations.¹ The mechanism of the reaction in aqueous dimethyl sulfoxide has been studied by White.² The role of ferricyanide as a catalyst has been explored;³ however, the exact mechanism of its action has not been elucidated.

We studied the kinetics of the reaction in order to clarify the role of ferricyanide in this oxidation. The chemiluminescence with ferricyanide as a catalyst has

generally been studied in the presence of hydrogen peroxide. In order to simplify analysis of the kinetics we have studied the reaction in the absence of peroxide.

Experimental Section

Materials.—Commercially available luminol was used without further purification. When the sodium salt of luminol,⁴ which had been recrystallized from water, was used, the results were unchanged. All other reagents were of analytical grade and were not further purified. All solutions used were freshly prepared.

Light Intensity Measurements.—The reaction vessel was a test tube (15 × 150 mm) placed in a light-tight compartment provided with a shutter opening to an RCA IP21 phototube. The output voltage from the phototube was displayed on a Varian recorder as a function of time. The apparatus was calibrated with a standard luminol reaction by the method of Lee and Seliger⁵ so that the intensity was reported in photons second⁻¹.

A solution of luminol (3.0 ml) in aqueous sodium hydroxide was placed in the test tube, and the reaction was initiated by injection of a potassium ferricyanide solution (0.3 ml). A stream of air was bubbled continuously through the reacting solution. This air stream ensured that the solution was kept saturated with oxygen and that mixing of the reactants was rapid. The rate of mixing appears to be fast compared to intensity decay; changing the rate of air flow into the solution did not alter the initial intensity of emitted light.

A Y-shaped reaction vessel equipped with a vacuum stopcock was used to measure intensity as a function of pressure. The luminol solution (3.0 ml) was placed in one arm of the vessel, and the ferricyanide solution (0.3 ml) in the other. The solutions were degassed, and air was introduced at known pressure. The vessel was placed in a light-tight box, and the reaction was

(1) E. H. White, "Light and Life," W. D. McElroy and B. Glass, Ed., Johns Hopkins Press, Baltimore, Md., 1961, p 183.

(2) E. H. White, O. Zafiriou, H. M. Kagi, and J. H. M. Hill, *J. Amer. Chem. Soc.*, **86**, 940 (1964); E. H. White and M. M. Bursley, *ibid.*, **86**, 941 (1964).

(3) F. H. Stross and G. K. Branch, *J. Org. Chem.*, **3**, 385 (1938).

(4) E. H. Huntress, L. N. Stanley, and A. S. Parker, *J. Chem. Educ.*, **11**, 241 (1934).

(5) J. Lee, A. S. Westley, J. F. Ferguson, III, and H. H. Seliger, in "Bioluminescence in Progress," F. H. Johnson and Y. Haneda, Ed., Princeton University Press, Princeton, N. J., 1966, p 35.

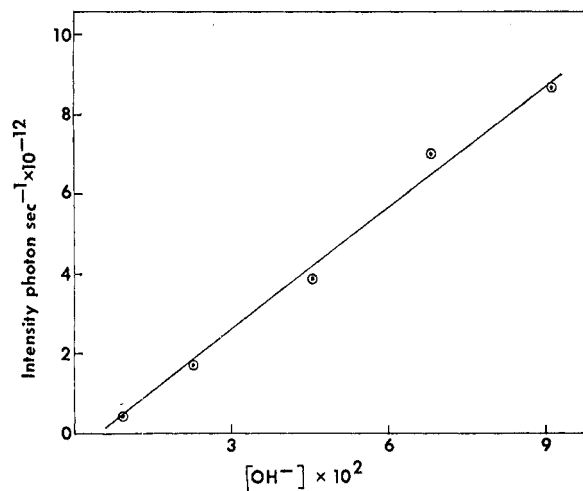


Figure 1.—Initial intensity as a function of base concentration: initial concentration of luminol and $K_3Fe(CN)_6$, 5.14×10^{-4} ml $^{-1}$; no $K_4Fe(CN)_6$ added initially.

initiated by a device that rocked the Y-tube back and forth, thus mixing the reactants. Emitted light was monitored with an RCA IP21 phototube.

Spectral Measurements.—The chemiluminescence spectra were measured in an Aminco-Bowman spectrophotofluorometer modified to permit injection of ferricyanide solution into the sample cell. The fluorescence spectra were measured on a Turner Associates Model 210 spectrophotofluorometer.

Results and Discussion

It has been established that 3-aminophthalic acid (**6**) is the emitting species when the reaction is carried out in aqueous dimethyl sulfoxide.² Similarly, we have compared the chemiluminescence spectra of the ferricyanide-catalyzed reaction with the fluorescence spectra of **6** in the presence of potassium ferricyanide. The spectra are identical, both exhibiting wavelengths of maximum emission at 450 $m\mu$. These data lead us to conclude that **6** is also the emitting species in the ferricyanide-catalyzed chemiluminescence. The fluorescence maximum of **6** is shifted from 425 to 450 $m\mu$ by the addition of an equimolar amount of potassium ferricyanide. We have shown that equimolar amounts of potassium ferricyanide or potassium ferrocyanide do not quench the fluorescence of 3-aminophthalate under the reaction conditions.

The rate of formation of **6** should be proportional to the intensity of emitted light. With this relationship in mind, we have measured the effect of reactant concentration on intensity. The reactants in question are hydroxide ion, luminol, potassium ferricyanide, and oxygen. The effect of varying the concentration of a given reactant on intensity was measured while holding the concentration of all other reactants constant. In all cases, the initial intensity, i_0 , was recorded as a function of concentration.

Figure 1 shows that the reaction is first order in hydroxide ion concentration over the pH range 12–13. In this pH range, complete formation of the monoanion of luminol ($pK_a = 6$)⁶ is expected. If the monoanion were the reactive species, an effect of hydroxide ion on intensity would not be anticipated in this pH range. However, because such an effect is observed, we postu-

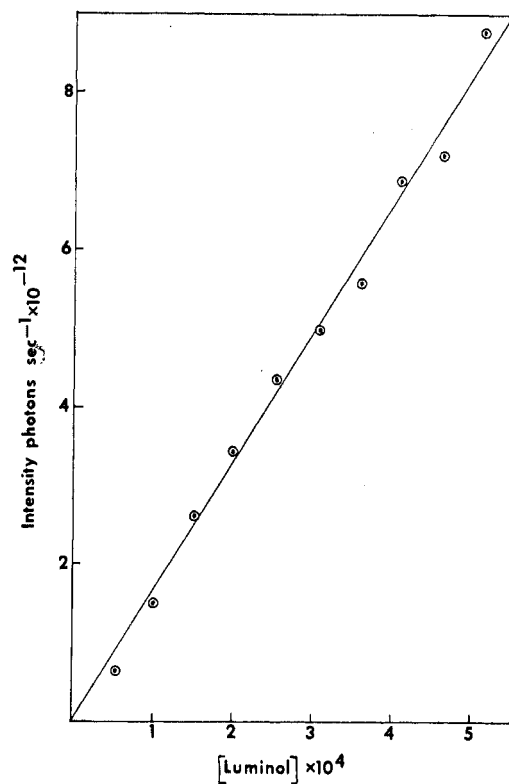


Figure 2.—Initial intensity as a function of luminol concentration: initial concentration of hydroxide, 9.1×10^{-2} ml $^{-1}$; of $K_3Fe(CN)_6$, 5.14×10^{-4} ml $^{-1}$; no $K_4Fe(CN)_6$ added initially.

late that the dianion is the reactive species in this system. White² has postulated that the dianion is autoxidized directly in potassium *t*-butoxide-dimethyl sulfoxide solution. However, such an autoxidation would not be expected to be rapid in aqueous solution since the concentration of the dianion is much lower.

The reaction is also first order in luminol concentration as shown in Figure 2.

The effect of potassium ferricyanide concentration on reaction rate is shown in Figure 3. The rate does not increase linearly with increasing ferricyanide but tends to level off at higher ferricyanide concentration.

Next, the effect of oxygen on the chemiluminescence was investigated. The light intensity was usually measured while bubbling a stream of air into the solution. When this air stream was replaced by a nitrogen stream, no light was emitted upon injection of ferricyanide solution. Furthermore, replacement of the nitrogen by air 10 min after injection of the ferricyanide gave no light. These results, along with those of Wilhelmson, *et al.*,⁷ show that oxygen is necessary for the production of light and that, in its absence, the luminol is consumed by ferricyanide in a nonluminescent reaction.

Figure 4 shows the effect of air pressure above the luminescing solution on intensity. Because air pressure is proportional to concentration of oxygen in solution, Figure 4 shows the dependence of reaction rate on oxygen concentration. The curve is similar to that obtained for the effect of ferricyanide concentration on intensity. The effect of oxygen on intensity cannot be due solely to quenching of fluorescence of aminophtha-

(6) A. K. Babko and L. I. Dufovenko, *Ukr. Khim. Zh.*, **29**, 479 (1963); *Chem. Abstr.*, **59**, 4594c (1963).

(7) P. C. Wilhelmson, R. Lumry, and H. Eyring, in "The Luminescence of Biological Systems," F. H. Johnson, Ed., American Association for the Advancement of Science, Washington, D. C., 1955, p 75.

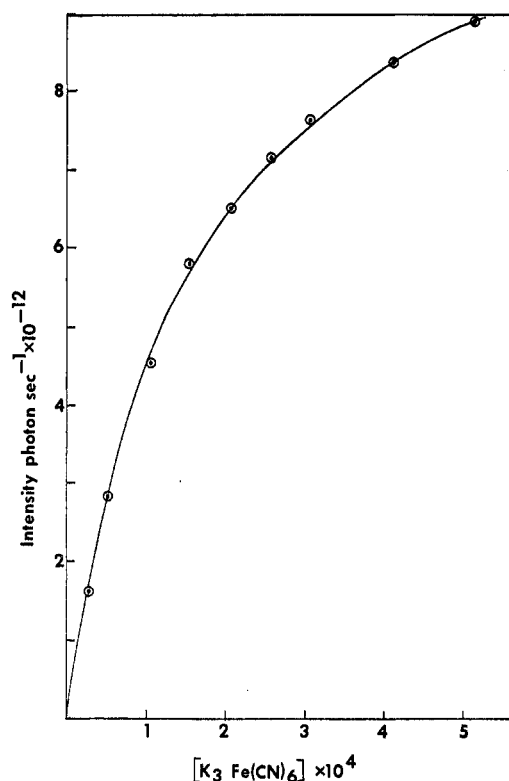


Figure 3.—Initial intensity as a function of $K_3Fe(CN)_6$ concentration: initial concentration of hydroxide, $9.1 \times 10^{-2} \text{ ml}^{-1}$; of luminol, 5.14 ml^{-1} ; no $K_4Fe(CN)_6$ added initially,

late by O_2 . The fluorescence of 3-aminophthalate is decreased by only 10% in the presence of pure oxygen as compared with its fluorescence under nitrogen.

To explain the effect of reactant concentration, the mechanism outlined in Scheme I is proposed.

SCHEME I

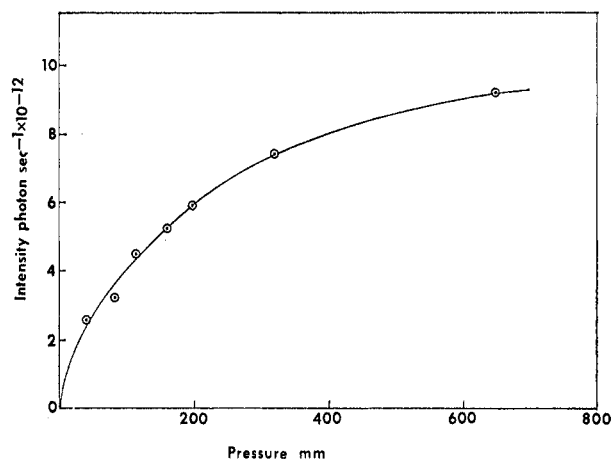
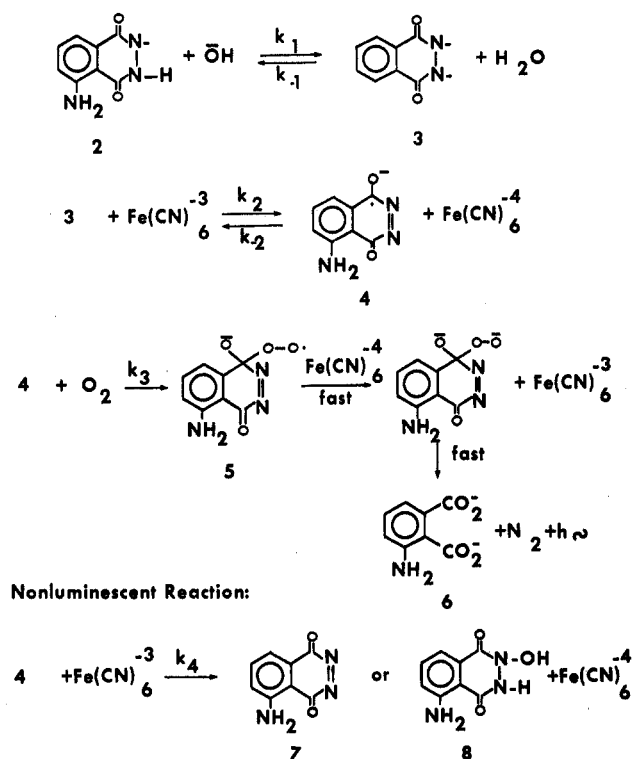
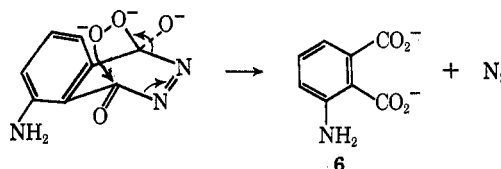


Figure 4.—Initial intensity as a function of initial pressure of air above solution: initial concentration of hydroxide, $9.1 \times 10^{-2} \text{ ml}^{-1}$; of luminol and $K_3Fe(CN)_6$, $5.14 \times 10^{-4} \text{ ml}^{-1}$; no $K_4Fe(CN)_6$ added initially.

The first step is simply the formation of the dianion **3** of luminol. The dianion is reversibly oxidized by ferricyanide to the semidione structure **4** shown in one of its resonance forms. The oxidation of **3** is written as reversible because addition of potassium ferrocyanide decreases the intensity of emitted light. Reversible one-electron transfers have been proposed for other ferricyanide oxidations.⁸ Alternatively, a sequence can be written involving oxidation of **2** followed by removal of a proton by hydroxide to give the semidione.

The semidione reacts with oxygen to give the peroxy radical **5**. The peroxy radical is subsequently reduced in a fast step to give peroxy anion. We have depicted the reducing agent as ferrocyanide produced during the reaction. We feel that in the absence of any other obvious reducing agent, ferrocyanide is the most logical choice. The fact that ferricyanide is only 50% consumed during chemiluminescence supports this contention. Reduction of **5** by ferrocyanide must occur after the rate-determining step to explain the inhibitory effect of ferrocyanide ion.

The peroxy anion decomposes to electronically excited **6** with loss of nitrogen. A concerted decomposition of peroxy anion to give aminophthalate and nitrogen, as depicted, would be expected to be rapid with loss of nitrogen as the driving force. In the light-pro-



ducing sequence proposed in Scheme I, the ferricyanide acts only as a catalyst and is not consumed. In this connection it should be noted that the potassium ferricyanide is not completely consumed during the chemiluminescence. A solution containing $2.82 \times 10^{-4} \text{ M}$ $K_3Fe(CN)_6$ and $2.82 \times 10^{-4} \text{ M}$ luminol initially, was found, by ultraviolet spectroscopy, to contain $1.24 \times 10^{-4} \text{ M}$ $K_3Fe(CN)_6$ at the termination of luminescence. However, both luminol and ferricyanide are destroyed

(8) C. G. Haynes, A. H. Turner, and W. A. Walters, *J. Chem. Soc.*, 2823 (1956).

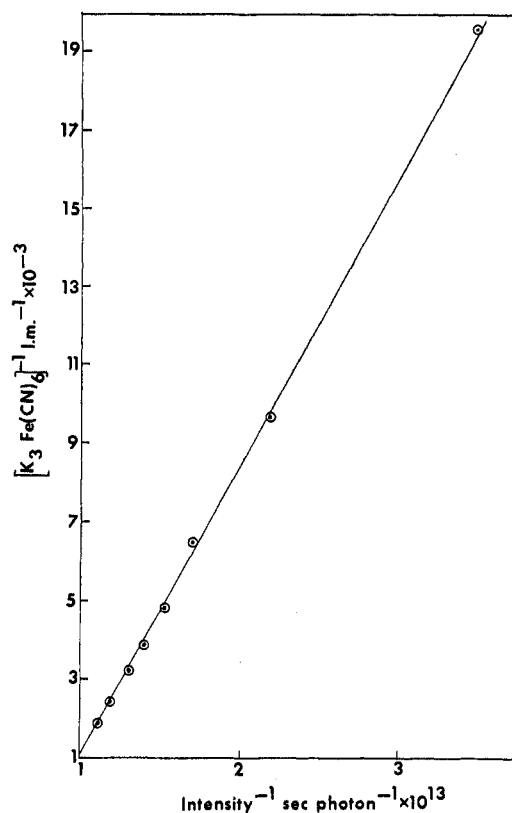


Figure 5.—Plot of reciprocal of initial intensity vs. reciprocal of $[K_3Fe(CN)_6]$.

in a competing nonluminescent reaction. This nonluminescent reaction is the two-electron oxidation of luminol by ferricyanide. Although the product of the two-electron oxidation has not been isolated, it is most likely 7 or 8. Both of these products would be extremely labile under the reaction conditions.⁹

Let us consider whether the proposed mechanism is consistent with the effects of concentration on intensity.

If a steady-state concentration of 4 is assumed,¹⁰ the rate of product formation is given by

$$d[6]/dt = \frac{k_a[-OH][2][Fe(CN)_6^{3-}]k_3[O_2]}{k_3[O_2] + k_{-2}[Fe(CN)_6^{4-}] + k_4[Fe(CN)_6^{3-}]} \quad (2)$$

where

$$k_a = k_1k_2/k_{-1}[H_2O]$$

From eq 1 and 2, an expression for intensity as a function of reactant concentration at any time is obtained (eq 3). This expression is consistent with the observed

$$i = \phi k_a \frac{[-OH][2][Fe(CN)_6^{3-}]k_3[O_2]}{k_3[O_2] + k_{-2}[Fe(CN)_6^{4-}] + k_4[Fe(CN)_6^{3-}]} \quad (3)$$

first-order dependence of luminol and hydroxide ion. Equation 3 may be written in a more useful form.

$$\frac{1}{[Fe(CN)_6^{3-}]} + \frac{k_{-2}[Fe(CN)_6^{4-}]}{k_3[O_2][Fe(CN)_6^{3-}]} + \frac{k_4}{k_3[O_2]} = \frac{\phi k_a[-OH][2]}{i} \quad (4)$$

(9) R. A. Clement, *J. Org. Chem.*, **25**, 1724 (1960); T. J. Kealy, *J. Amer. Chem. Soc.*, **84**, 966 (1962).

(10) A. A. Frost and R. G. Pearson "Kinetics and Mechanism," 2nd ed, John Wiley & Sons, Inc., New York, N. Y., 1961, p 237.

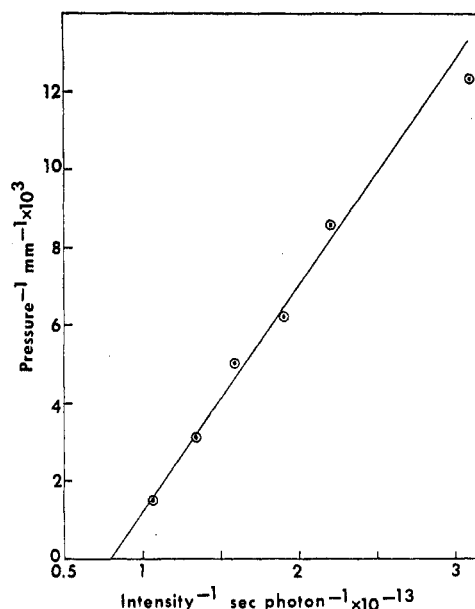


Figure 6.—Plot of reciprocal of initial intensity vs. reciprocal of initial pressure of air above solution.

If the initial intensity, i_0 , is measured, and no ferrocyanide is added, the second term on the left-hand side of eq 4 drops out.

$$\frac{1}{[Fe(CN)_6^{3-}]} + \frac{k_4}{k_3[O_2]} = \frac{\phi k_a[-OH][2]}{i_0} \quad (5)$$

A plot of $[K_3Fe(CN)_6]$ vs. i_0^{-1} should be linear with a slope equal to $\phi k_a[2][-OH]$ and an intercept of $k_4/k_3[O_2]$. Such a plot is shown in Figure 5; a least-squares treatment gives $\phi k_a = 1.6 \times 10^{21}$ l.³ photon mol⁻³ sec⁻¹ and $k_4/k_3[O_2] = 6.5 \times 10^3$ l. mol⁻¹.

Substitution of $k_4/k_3[O_2]$ into eq 3 enables calculation of ϕk_a from the data in Figures 1 and 2. Such calculations yield $\phi k_a = 1.7 \times 10^{21}$ l.³ photon mol⁻³ sec⁻¹ from Figure 1 and $\phi k_a = 1.5 \times 10^{21}$ l.³ photon mol⁻³ sec⁻¹ from Figure 2.

A plot of $[O_2]^{-1}$ vs. reciprocal of initial intensity should also be linear when the concentrations of all other reactants are held constant. This linearity is demonstrated in Figure 6 as the reciprocal of air pressure over the solution plotted against i_0^{-1} .

When potassium ferrocyanide is added to the reaction initially, the second term on the left-hand side of eq 5 does not drop out. If the concentrations of all other reactants are held constant, a plot of added ferrocyanide vs. i_0^{-1} should permit evaluation of $k_2/k_3[O_2]$. This plot, shown in Figure 7, gives $k_2/k_3[O_2] = 8.9 \times 10^3$ l. mol⁻¹.

A chain mechanism similar to that proposed for certain autoxidations may be envisioned.¹¹ In such a mechanism, the ferricyanide acts as an initiator, and the peroxy radical 5 is reduced in a chain-carrying step by the luminol dianion. If this mechanism were operative, the expression for intensity as a function of concentration would be

$$i = \frac{\phi k_a[-OH][Fe(CN)_6^{3-}]k_3[O_2][2]}{k_{-2}[Fe(CN)_6^{4-}] + k_4[Fe(CN)_6^{3-}]} \quad (6)$$

This relationship, however, is inconsistent with the observed effect of oxygen concentration on intensity. A

(11) G. A. Russel, E. G. Janzen, A. G. Bemis, E. J. Geels, A. J. Moye, S. Mak, and E. T. Strom, "Selective Oxidation Processes," R. F. Gould, Ed., American Chemical Society, Washington, D.C., 1965, p 121.

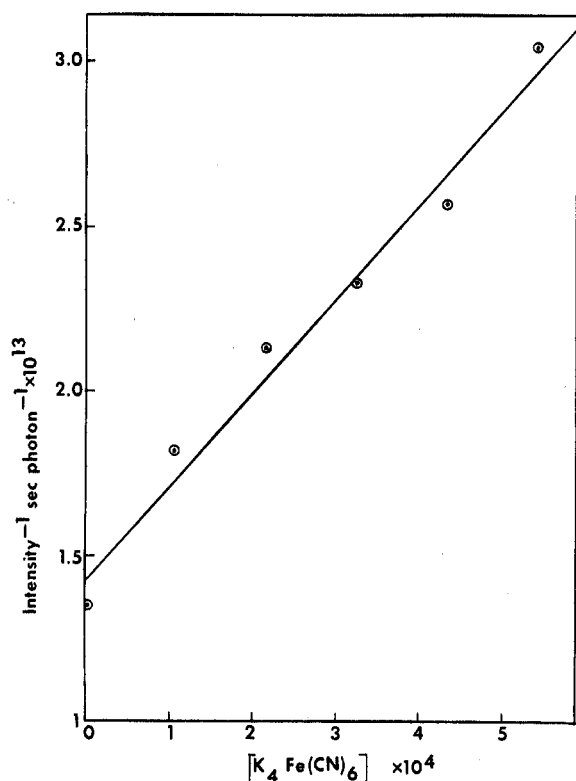


Figure 7.—Plot of concentration of $K_4Fe(CN)_6$ vs. reciprocal of initial intensity: initial concentration of hydroxide, $8.2 \times 10^{-2} \text{ ml}^{-1}$; of luminol, $4.99 \times 10^{-4} \text{ ml}^{-1}$; of $K_3Fe(CN)_6$, $5.06 \times 10^{-4} \text{ ml}^{-1}$.

mechanism in which the nonluminescent reaction is the oxidative dimerization of luminol is ruled out because the reaction would not be first order in luminol.

A key feature of the reaction is competition of oxygen and ferricyanide for the semidione. Systems which provide a direct source of hydroperoxy radical for reaction with the semidione will enhance light production. Thus hydrogen peroxide increases the intensity and quantum yield. A solution $4.62 \times 10^{-5} \text{ M}$ in luminol, $5.14 \times 10^{-4} \text{ M}$ in $K_3Fe(CN)_6$, and 0.08 M in hydroxide has a quantum yield of 3.8×10^{-7} . A similar solution $8 \times 10^{-3} \text{ M}$ in hydrogen peroxide has a quantum yield of 3.25×10^{-6} . In the presence of oxygen, the semidione is consumed by further oxidation with ferricyanide.

This competition between reaction with oxygen and further oxidation is probably a general feature of all chemiluminescent reactions of luminol in which a one-electron oxidant is employed. The fact that chemiluminescence occurs at all is due to the stability of the semidione to further oxidation. In this connection, note that electron-donating substituents on the aromatic ring enhance luminescence. Such substituents would be expected to increase the stability of the semidione.¹²

Registry No.—Potassium ferricyanide, 13746-66-2; 1, 521-31-3.

(12) E. H. White and M. M. Bursley, *J. Org. Chem.*, **31**, 1912 (1966). A referee has pointed out that electron-donating substituents also increase the fluorescence quantum yield of the phthalates.

Pyrolysis Studies. XIX.¹ Substituent Effect of 1-Aryl-3-buten-1-ols

GRANT GILL SMITH^{2a} AND KENT J. VOORHEES^{2b}

Department of Chemistry, Utah State University, Logan, Utah 84321

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The unimolecular homogeneous thermolysis of nine 1-aryl-3-buten-1-ols has been studied in a seasoned, constant-volume, stainless steel reactor. Arrhenius parameters have been evaluated in the temperature range of 610–644°K. The small ρ value (–0.26) from a Hammett $\rho\sigma$ plot indicates a minor substituent effect for the *meta* and *para* isomers with apparently little or no charge development at the 1 position in the proposed concerted six-membered ring transition state. An *o*-methoxy substituent showed a marked proximity effect with an activation energy 4–7 kcal/mol lower and an entropy of activation of 6–9 eu, more negative than the other compounds studied.

β -Hydroxy olefins have been reported to pyrolyze to olefins and carbonyl compounds by a unimolecular homogeneous reaction, likely through a six-membered-ring transition state.^{3,4} The influence of 3- and 4-phenyl and 1-alkyl substituents on the ease of thermolysis of 3-buten-1-ol has been reported by Smith and Yates.⁵

They found that π contribution increased the rate of pyrolysis at the 3 position more than at the 4 position and that the rate of pyrolysis followed the sequence tertiary > secondary > primary for alkyl substitution at the carbinol position. They presented a qualitative picture consisting of a positive charge forming at the 3

position and a slight negative charge developing at the 4 position in the transition state.

No direct study of π contribution has been reported at the carbinol position; only competitive^{6,7} type reactions have been studied.

The gas-phase thermolysis of 1-aryl-3-buten-ols reported in this study, when compared with the result reported by Smith and Yates,⁵ further substantiates the nature of this reaction and gives additional insight concerning the transition state.

Results

1-Aryl-3-buten-1-ols were pyrolyzed in a deactivated stainless steel reactor⁸ over a temperature range of

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(2) (a) To whom all communications should be sent; (b) National Defense Education Act Predoctoral Fellow, 1968–1970.

(3) R. T. Arnold and G. Smolinsky, *J. Org. Chem.*, **25**, 129 (1960).

(4) G. G. Smith and R. Taylor, *Chem. Ind. (London)*, **35**, 949 (1961).

(5) G. G. Smith and B. L. Yates, *J. Chem. Soc.*, 7242 (1965).

(6) A. Viola, E. J. Iorio, K. K. Chen, G. M. Glover, U. Nayak, and P. J. Kocienski, *J. Amer. Chem. Soc.*, **89**, 3462 (1967).

(7) K. J. Voorhees, G. G. Smith, R. T. Arnold, D. G. Mikolasek, and R. R. Covington, *Tetrahedron Lett.*, 205 (1969).

(8) G. G. Smith and J. A. Kirby, *Analyst (London)*, **94**, 242 (1969).