Excitation of Fluorescent Aromatic Hydrocarbons in Chemiluminescence Reactions of Bis (2, 4, 6-trichlorophenyl) oxalate and Hydrogen Peroxide

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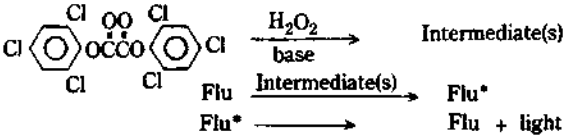
The excitation mechanism of fluorescers for the chemiluminescence arising from the reaction between *bis*(2, 4, 6-trichlorophenyl) oxalate (TCPO) and hydrogen peroxide in the presence of fluorescent polycyclic aromatic hydrocarbons has been studied. A key intermediate is generated from the reaction of TCPO with H_2O_2 and then the excitation of fluorescer involves the formation of a charge-transfer complex between the key intermediate and fluorescer.

Introduction

Peroxyoxlate chemiluminscence (CL) refers to the light emission resulting from a reaction of oxalic acid derivatives with hydrogen peroxide. It has gained increasing importance for its utilization such as to detect fluorescent compounds with better signal-to-noise ratio than direct excitation using a light source.[1-6] The CL produced by a reaction between aryl oxalate esters, such as bis (2,4,6-trichlorophenyl) oxalate (TCPO), and hydrogen peroxide in the presence of a base catalyst, e.g., sodium salicylate, and added fluorescers may be represented by Scheme I[3]



Scheme 1

where Flu stands for a fluorescer. As a key intermediate 1,2-dioxetanedione was proposed[7], but the nature of the intermediate has never been unequivocally established. Recently, however, at least two intermediates were proposed to generate CL based on two maxima[8] compared with single exponential decay[9] in the spectra of emitted light as a function of time.

Nevertheless, the intermediate(s) should be capable of transferring as much as 105 kcal/mol of energy[7] to a fluorescer. Fluorescers with lower excitation energies appear to give high CL efficiencies.[6] However, studies on several different fluorescers failed to indicate any correlation of excitation yield to the singlet energy of the fluorescer.[10] If a charge-transfer type interaction is important[11], the fluorescers of good electron donating ability should be highly efficient in the CL reaction. Although some evidences seem to support the charge-transfer type complex[4a] between intermediate and fluorescer, quantitative results are rare.

For these reasons a peroxyoxalate CL reaction of TCPO has been attempted with a variety of fluorescers to elucidate at least the excitation mechanism of the fluorescent compounds.

Experimental

Reagents. TCPO was prepared by following the method of Mohan and Turro[12] and recrystallized from benzene. It was identified with m.p. (192-194°C), IR, and NMR. H_2O_2 was purchased from Riedel-de Haēn and vacuum distilled to obtain 94% H_2O_2, whose concentration was determined by iodometrically. Solvents, dimethylphthalate and dibutylphthalate, and fluorescers were of reagent grade from Aldrich except 9-(2-phenylethenyl) anthracene and 1-(2-phenylethenyl) pyrene which were donated from the Polymer Laboratory of Korea University.

Table 1. Data for identification of energy transfer mechanism

fluorescer	ϕ_f	$\lambda I_m{}^a$	I_m	I_m/ϕ_f	$E_{1/2(ox)}{}^c$	$t_m{}^d$	$t_{1/e}{}^d$
anthracene	0.2	409	14	70	1.09	1.3	12.3
9-anthraldehyde	0.02	457	0.1	5	1.34	1.0	—
9,10-diphenylanthracene	0.83	435	160	193	1.07	1.3	11.8
9-(2-phenylethenyl)-anthracene	0.37	475	60	162	0.89	1.6	13.6
9-cyanoanthracene	0.75	444	1.3	1.7	1.45	1.5	8.7
rubrene	1.0[b]	562	992	992	0.82	1.3	10.2
perylene	1.0	474	668	668	0.85	1.3	9.3
1-(2-phenylethenyl)-pyrene	0.93	446	215	231	0.93	1.0	9.7
pyrene	0.56[b]	396	2.7	4.8	1.15	1.3	11.0

[a] in nm. [b] reference 13. [c] in volts vs. S.C.E. [d] in min.

Instrumentation. Fluorescence quantum yields were obtained by comparing fluorescers to that of perylene whose fluorescence quantum yield is set to 1.0[13]. For this, the absorbances of the compounds were measured by a Shimadzu UV-240 spectrophotometer and the fluorescence spectra by a Hitachi 650-60 spectrofluorimeter. All absorption and fluorescence spectra were obtained in an identical solvent as the CL intensities. A PAR 273 potentiostat/galvanostat was employed to record the cyclic voltammograms of the fluorescers from which the oxidation half-wave potentials were measured. The CL reaction was carried out in a 1.0-cm fluorescence cell inserted in a thermostatic housing of the spectrofluorimeter. A 1.5m*l* solution of TCPO and a fluorescer in dibutylphthalate was added to a cell. The solution was purged with nitrogen. The reaction was initiated by injecting 0.5m*l* solution of H_2O_2 and sodium salicylate in 4:1 dimethylphthalate /*t*-butylalcohol which had been purged with nitrogen. The decay of CL intensity with time was monitored by the spectrofluorimeter with the lamp turned off at wavelengths such that the reabsorption of emission was negligible.

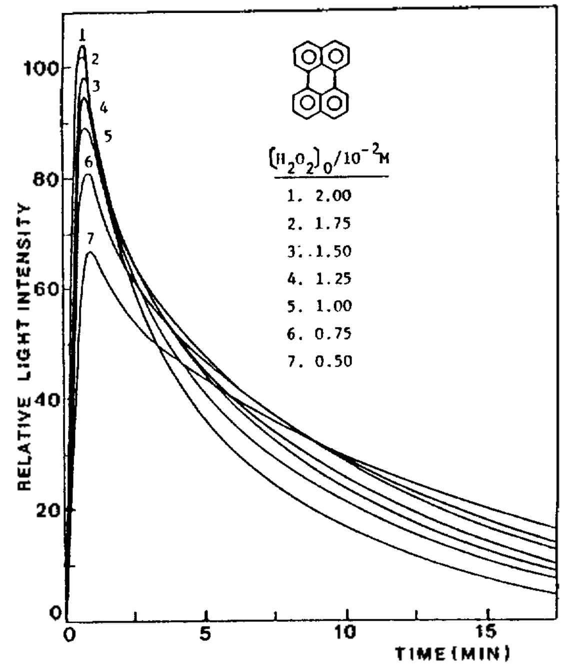


Figure 1. Relative intensity of the peroxyoxalate CL vs. time of 5.0×10^{-5}M perylene, 5.0×10^{-4}M TCPO, 1.2×10^{-4}M sodium salicylate, and various concentrations of H_2O_2.

Results and Discussion

Table 1 summarizes the experimental results of the relative maximum CL intensities (I_m) of polycyclic aromatic hydrocarbons studied under identical conditions together with the data for fluorescence quantum yield and oxidation half-wave potential. Also included are the time to reach the CL maxima (t_m) and that to decay to $1/e(t_{1/e})$ of the maximum intensity where e is the base of the natural logarithms. Fluorescers were confirmed to be emitting species by matching the CL spectra with the fluorescence spectra.

The initial rise up to the maximum CL intensity was rather rapid while the subsequent decay was slow. The decay for the light emitted at 474nm as typically shown in Figure 1 for perylene, exhibited apparent single exponential curves regardless of the concentrations of H_2O_2. Since under the conditions of excess H_2O_2 the total CL intensities should be dependent on the TCPO concentration, at later times the CL intensities became higher for lower H_2O_2 concentrations. The single exponential behavior was observed for all fluorescers as evidenced by nearly equal times of t_m and $t_{1/e}$ and agrees with the result of Catherall *et al.*,[9] although less viscous solvent and a more electronegative aryl oxalate ester were utilized than those used in this study. It is believed that a key intermediate is responsible for the single exponential decay. Recently, however, Givens and coworkers observed two maxima in the time courses for the CL intensity emitted by the reaction of TCPO with H_2O_2 and triethylamine in ethyl acetate.[8] They interpreted that the CL had been generated by at least two intermediates. At present, it is not certain whether the CL reactions here proceeded by a single or multiple intermediates. There may exist more than one intermediate, however, under present conditions probably one of them appeared to predominate the CL reaction.

For a given key intermediate the CL efficiency (ϕ_{CL}) may be expressed as the product of the excited state yield (ϕ_{ex}) and the fluorescence quantum yield (ϕ_f) of a fluorescer which correspond to the second and the third steps of Scheme I, respectively,

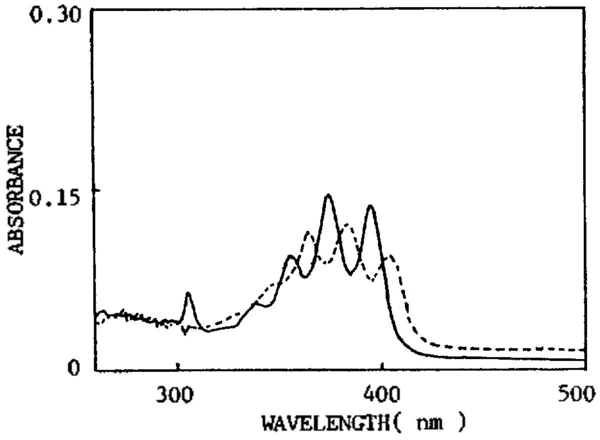


Figure 2. Absorption spectra of 1.0×10^{-5}M of 9,10-diphenylanthracene (solid line) and 9-cyanoanthracene (broken line).

$$\phi_{CL} = k\ \phi_{ex} \cdot \phi_f \tag{1}$$

where *k* is the yield of the key intermediate. Since the shape of the CL intensity vs. time remained nearly the same and the concentrations of the reagents were kept constant, the I_m should be proportional to ϕ_{CL}. Thus, the I_m corrected for ϕ_f is linearly related to ϕ_{ex},

$$I_m / \phi_f \propto \phi_{ex} \tag{2}$$

The calculated values of I_m/ϕ_f are included in Table 1.

When a non-radiative energy transfer of the Förster type[14] is operating on ϕ_{ex}, the more the absorption spectrum of a fluorescer overlaps with the emission spectrum of the key intermediate, the larger the CL intensity is expected. As shown in Figure 2, 9, 10-diphenylanthracene and 9-cyanoanthracene have similar absorption spectra. In addition, both compounds exhibit comparable fluorescence efficiencies (Table 1). Therefore, even if the emission spectrum of the key intermediate is unknown, I_m/ϕ_f of the two compounds would be close to each other. Contrary to our expectation, a large difference in $I_m/\ \phi_f$ observed. Also, the large differences in I_m/ϕ_f between 9-(2-phenylethenyl) anthracene or 1-(2-phenylethenyl) pyrene and 9-anthraldehyde cannot be explained even though the molar absorptivities are taken into account (Figure 3). Based on these results the non-radiative energy transfer appeared to be unimportant to the systems examined.

As can be noted in Table 1, the CL intensities were enhanced with the substitution of phenyl and phenylethenyl groups, but diminished with carbonyl and cyano substituents. Furthermore, the ability for a fluorescer to accept chemical energy and to emit light subsequently generally parallels with the decrease of its oxidation half-wave potential ($E_{1/2\,(ox)}$). When a charge-transfer mechanism is operating between key intermediate and fluorescer the rate constant for the excitation step may be written as

$$k_a = A \exp| - (IP - EA - e^2/\varepsilon r - \Delta E_{sol})/RT| \tag{3}$$

where IP is the ionization potential of the fluorescer, an electron donor, EA is the electron affinity of the key interme-

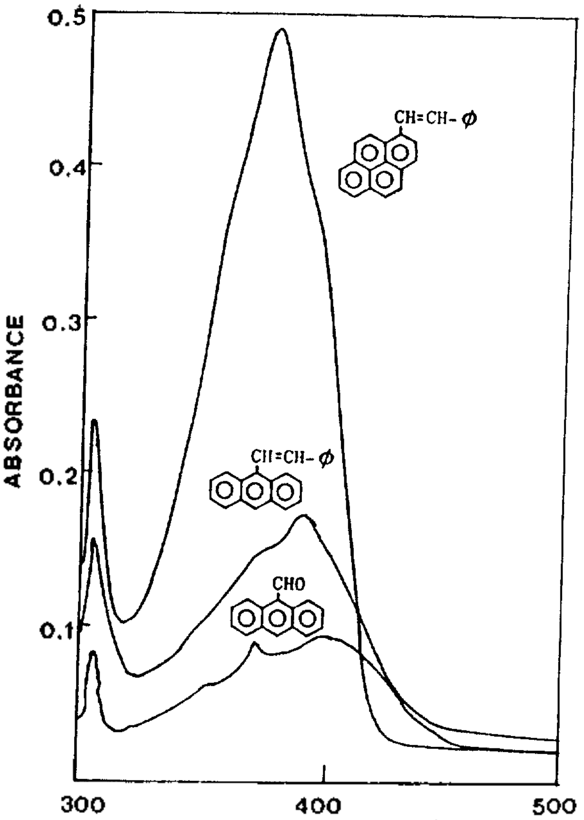


Figure 3. Absorption spectra of 1.0×10^{-5}M solution of 9-(2-phenylethenyl) anthracene, 1-(2-phenylethenyl) pyrene, and 9-anthraldehyde.

diate, an electron acceptor, $-e^2/\varepsilon r$ is the Coulombic energy between the radicals separated by r in a medium of dielectric constant ε, and ΔE_{sol} is the difference in solvation energy between the compounds and their ions.

Assuming that the latter two terms remain almost the same for varying the fluorescers studied, ϕ_{ex} of different fluorescers can be related to IP as

$$\phi_{ex} \propto k_a \propto \exp(-IP/RT) \tag{4}$$

since EA is a constant under the present condition. Furthermore, the ionization potentials for a number of aromatic hydrocarbons can be linearly correlated with the oxidation half-wave potentials[15] as

$$IP = a\ E_{1/2(ox)} + b \tag{5}$$

where a and b are solvent-dependent constants.

The combination of Eqs.(2) through (5) gives

$$I_m / \phi_f \propto \exp\{-\frac{a}{RT}E_{1/2(ox)}\} \tag{6}$$

Figure 4 illustrates that the relationship between the logarithm of the CL intensity at maximum corrected for ϕ_f and the oxidation half-wave potential agrees qualitatively well with the expectation from Eq. (6). The constant a determined from Figure 4 is about four times lower than that obtained for the ionization against the oxidation half-wave potential in acetonitrile.[15] A possible explanation of this discrepancy is that both the ionization potential and the solvation energy may be functions of solvent as well as the structure of the fluorescers. Nevertheless, it can be stated that the charge-transfer type of energy transfer appears to be suitable to fit the data obtained here because the CL efficiency corrected for fluorescence quantum yield increase as the oxidation half wave potential of a fluorescer decreases.

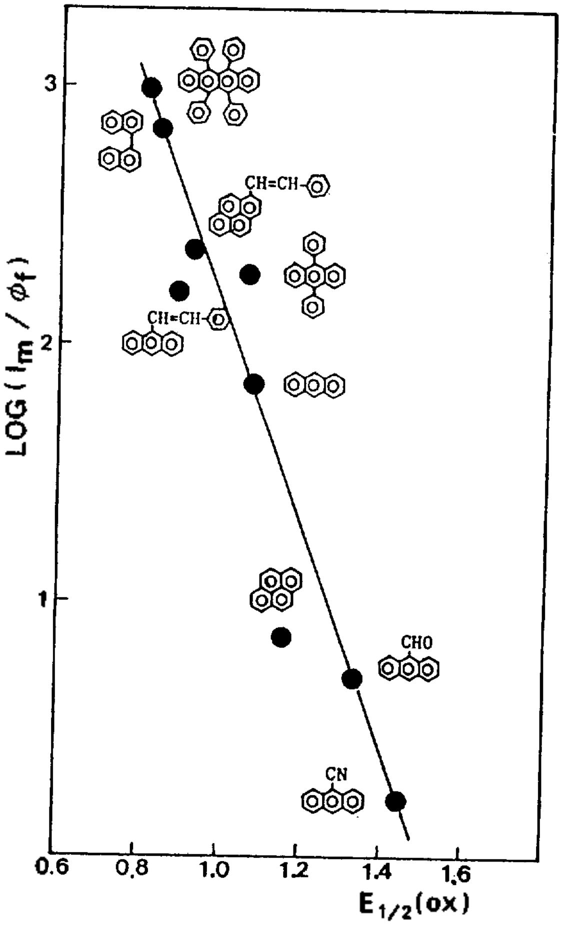


Figure 4. Relationship between the logarithm of the CL intensity at maximum (I_m) corrected for fluorescence quantum yield (ϕ_f) and the oxidation half-wave potential ($E_{1/2(ox)}$) of the fluorescers.

Finally, we have assumed implicitly that the fluorescer does not participate in the formation of the key intermediate. A preliminary absorption experiment showed that the CL decay was practically independent of fluorescers. A further study on the kinetics and mechanism of the peroxyoxalate CL is underway.

Acknowledgement. The financial support for this work from the Korea Science and Engineering Foundation is gratefully acknowledged.

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Selective Esterification of N-Benzyl-L-aspartic Acid. (I). Some Modified Methods for the Preparation of N-Benzylaspartic Anhydride Hydrobromide

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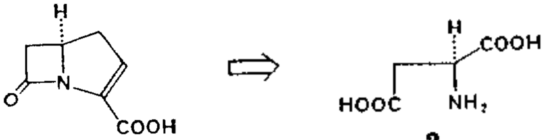
Racemic N-benzylaspartic acid (**4**) was prepared from maleic anhydride and used to modify and develop some efficient methods for the preparation of N-benzylaspartic anhydride hydrobromide (**5**). Thus, successive treatment of the compound **4** with 30% HBr in acetic acid and acetic anhydride afforded the title compound **5** in 75% yield. From this compound **5**, α-benzyl and α-methyl N-benzylaspartates were also prepared.

Introduction

The recently discovered carbapenems (**1**, derivatives of (5R)-7-oxo-1-azabicyclo [3.2.0] hept-2-ene-2-carboxylic acid)^1 possess the characteristic (5R)-configuration, which can be derived from naturally occurring L-aspartic acid (**2**). Thus far several reports^2 have described the elaboration of β-lactam rings from L-aspartic acid.



1　　**2**

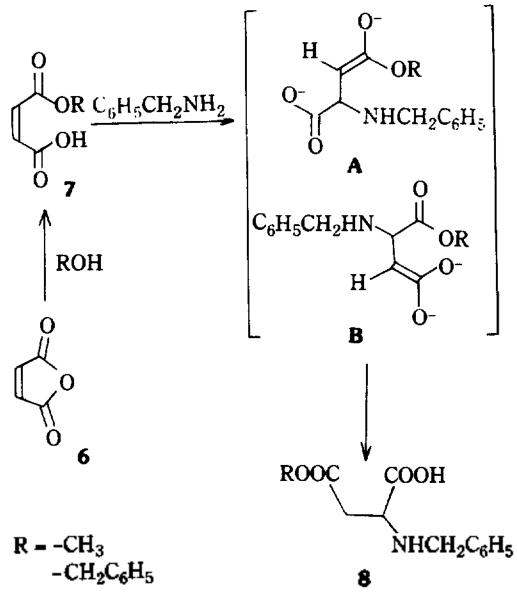
Intramolecular cyclization of α-alkyl L-aspartates, the β-amino acids, gives rise to 4-substituted β-lactams possessing the correct stereochemistry at 4-position, which corresponds to the (5R)-configuration of carbapenems.

These α-alkyl L-aspartates are readily available by the alcoholysis^3 of L-aspartic anhydride hydrobromide (**3**). The compound **3** has thus far been prepared by deprotection of N-benzyloxycarbonyl-L-aspartic anhydride with 30% HBr in acetic acid^3, by direct dehydration of L-aspartic acid with thionyl chloride in trifluoroacetic acid followed by the treatment with HBr in acetic anhydride^4, and by direct dehydration and salt formation of L-aspartic acid with PBr_3 in THF^5.

In this paper, we would like to report the modified methods for the preparation of N-benzylaspartic anhydride hydrobromide (**5**) from N-benzylaspartic acid (**4**) and the synthesis of α-and β-esters of N-benzylaspartic acid.

Results and Discussion

Alcoholysis of maleic anhydride (**6**) in methanol or benzyl alcohol produced in excellent yield methyl maleate (**7a**) or benzyl maleate (**7b**), which was treated with benzylamine to obtain the Michael-type addition products. The addition reaction proceeded by attacking β-position to the ester group rather than that to the free carboxylic acid, affording in excellent yield β-methyl N-benzylaspartate (**8a**)^6 or β-benzyl N-benzylaspartate (**8b**), respectively. This regioselectivity would probably be derived from the greater stability of the generated dianion **A** compared to that of the dianion **B**, in which the negative charges are accumulated (see Scheme 1). Hydrolysis of the ester groups of the compounds **8a** and **8b** gave rise to racemic N-benzylaspartic acid (**4**).



Scheme 1. Synthesis of β-Alkyl N-Benzylaspartates