

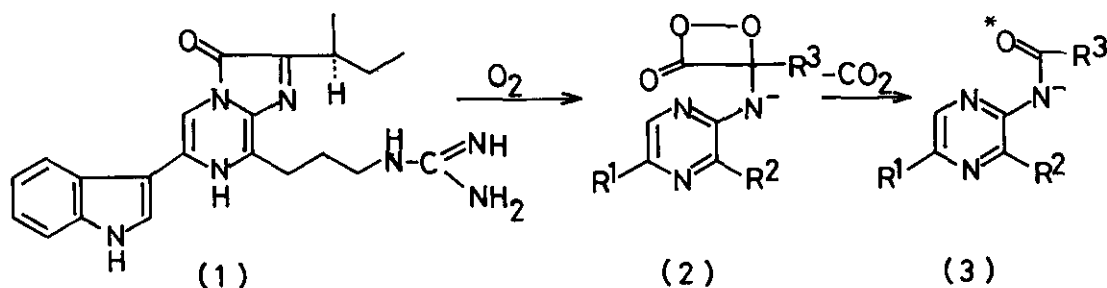
SYNTHESIS AND CHEMILUMINESCENCE OF 2-(INDOL-3-YL)-3-PHENYL-
DIHYDRO-1,4-DIOXIN 2,3-EPIDIOXIDE, A DIOXETANE HAVING
UNSUBSTITUTED INDOLE GROUP SIMILAR TO THE INTERMEDIATE
PROPOSED IN CYPRIDINA BIOLUMINESCENCE¹

Hideshi Nakamura and Toshio Goto*

Department of Agricultural Chemistry, Nagoya University
Chikusa, Nagoya 464

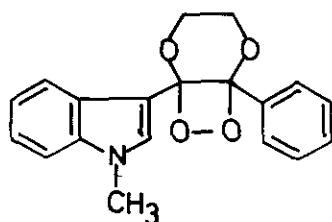
From 2-(indol-3-yl)-3-phenyl-1,4-dioxene was synthesized the title dioxetane, which gave strong chemiluminescence in aprotic solvents.

Cypridina luciferin (1) is one of the efficiently chemiluminescent substances² in addition to its bioluminescent activity. Extensive studies on the mechanism of the luminescence suggested the involvement of the very unstable dioxetane intermediate (2), which decomposes rapidly with production of the singlet excited state molecule (3) in high yields. The high efficiency of singlet excited state production and the instability (low activation energy) of the intermediate dioxetane in the luminescence may be explained in terms of the conjugation of the electron-donating and highly fluorescent chromophore with the excited state carbonyl group to be formed.³ Recently both Schuster et al.⁴ and McCapra⁵ have proposed an attractive mechanism (the CIEEL) for highly efficient dioxetane decomposition of this type.

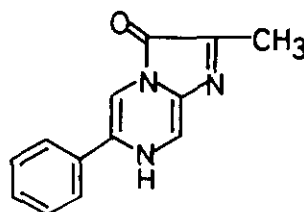


The β -indolyl group may be one of the best chromophores to fulfil the above requirements. We have prepared N-methyl- β -indolyldioxetanes, 2-(1-methylindol-3-yl)-3-phenyldihydro-1,4-dioxin 2,3-epidioxide (4) and its analogs, and found that these compounds are efficiently chemiluminescent as expected; the compound (4) gave 50% yield of singlet excited state.³

Cypridina luciferin (1) has an unsubstituted indole group, which has an important role for the production of efficient bioluminescence as well as chemiluminescence, since the phenyl analog (5) of the luciferin shows only 2 % of the chemiluminescence efficiency of Cypridina luciferin (1) (in diglyme containing acetate buffer pH 5.6) (the aliphatic substituents on the imidazopyrazine ring do not affect much on the chemiluminescence quantum yields). We have had much interest to see how effective the unsubstituted indole group is on the luminescence quantum yields when introduced into the dihydro-1,4-dioxin 2,3-epidioxide system, since the protection of NH group sometimes alters the character of the indole group completely.

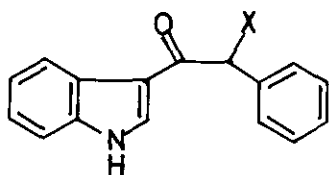


(4)



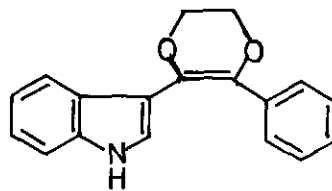
(5)

A solution of α -chlorophenylacetylindole (6)⁶ in dioxane containing sodium ethylene glycolate was heated at 100° C. Chromatography of the reaction products on a silica gel column using CH₂Cl₂-MeOH (95:5) as eluant gave 3-(2-(β -hydroxyethoxy)phenylacetyl)indole (7) as faintly reddish crystalline powder, mp153-153.5⁸ (yield 26%), m/z 295 (M^+). The β -hydroxyethyl ether (7) was refluxed in the presence of catalytic amount of camphorsulfonic acid for 1 h to give the dioxene (8)

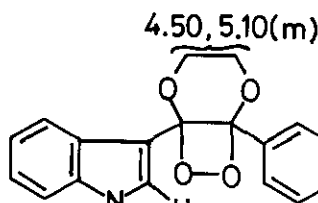


(6) X = -Cl

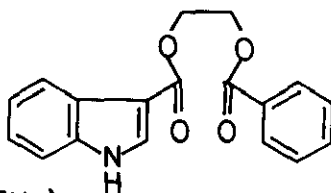
(7) X = -OCH₂CH₂OH



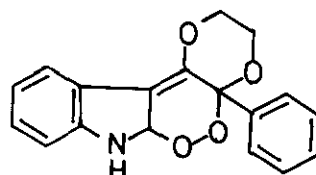
(8)



(9)



(10)



(11)

(62%), colorless solid, mp 156-157° C, m/z 277 (M^+), nmr ($CDCl_3$) 4.36 (4H, s), 6.8-7.5 (9H, m), 6.92 (1H, d, $J = 2.8$ Hz), 7.96 (1H, br.s, exchangeable). A solution of the dioxene (8) (27.7 mg) and polymer-bound Rose Bengal (40 mg) in dichloromethane (10 ml) was photooxygenated as previously reported⁷ at -78° C for 1.4 h. The reaction mixture containing the dioxetane (9) was warmed up (bright luminescence was observed) to room temperature and evaporated to dryness. The residue was chromatographed on preparative silica gel tlc using dichloromethane as eluant to give the diester (10) (21.5 mg, 71%) as colorless rods (from 80% ethanol), mp 138-139° C, m/z 309 (M^+); uv (CH_2Cl_2) 277 nm (ϵ 11100), 280 (11400), 293 (9000); nmr ($CDCl_3$) 4.69 (4H, s), 7.1-7.6 (6H, m), 7.50 (1H, br.s), 7.9-8.2 (3H, m), 7.95 (1H, d, $J = 3.2$ Hz).

The formation of the dioxetane (9) was confirmed by nmr analysis at -45° C of the photooxygenated product obtained from the dioxene (8) in $CDCl_3$ (in an nmr tube); in this experiment the 1,4-endo-peroxide (11) was not observed at -45° C. The chemiluminescence spectra of the dioxetane (9) are shown in Fig. 1. In dichloromethane and tetrahydrofuran the exciplex luminescence⁷ was observed around 390-400 nm.

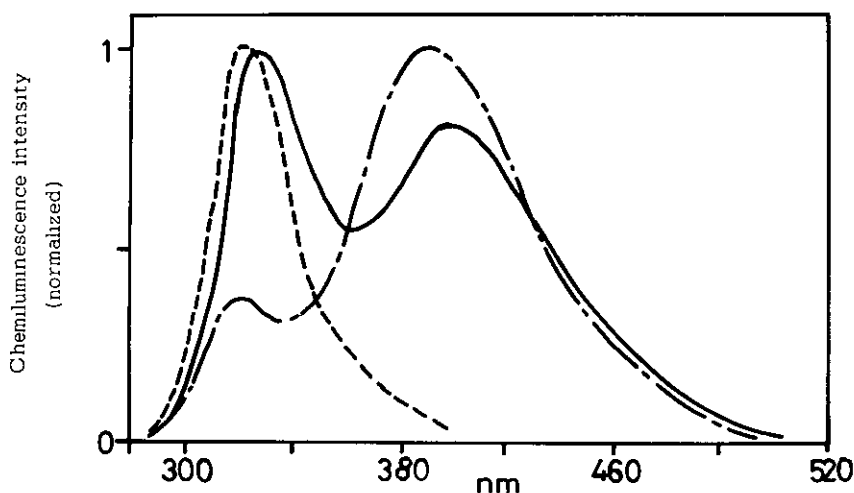


Fig. 1. Chemiluminescence spectra of the dioxetane (9)

----- in hexane, — in CH_2Cl_2 , — — in tetrahydrofuran

The chemiluminescence quantum yield and the activation parameters of the dioxetane (9) are listed in Table 1 in comparison with those of the N-methyl derivative (4). Contrary to the expectation the chemiluminescence quantum yield of the dioxetane (9) having the unsubstituted indole is rather lower than that of the dioxetane (4) having N-methylindole; the activation energy (E_a) of the former is also much higher than the latter (Table 1); thus, no superiority was observed with the former. Addition of bases such as triethylamine, *t*-BuOK, etc. did not improve chemiluminescence efficiency.

Table 1. Chemiluminescence of the dioxetanes (9) and (4)

Compound	Solvent	λ_{max}	Quantum yield of C. L. (%)	Ea (kcal/mole)	log A	S [‡] (298.2K)
(9)	Hexane	320	0.26			
	CH ₂ Cl ₂	325, 390	0.48	16.4	9.51	-17.0
(4)	Hexane	320	3.6			
	CH ₂ Cl ₂	330, 400	2.3	12.6	7.78	-25.0

Acknowledgement This work was partly supported by Grant-in-Aid for Scientific Research, The Ministry of Education, Science and Culture.

REFERENCES

1. Cypridina Bioluminescence X. Preceding paper: T. Goto, M. Isobe, Y. Kishi, S. Inoue, and S. Sugiura, Tetrahedron, 1975, 31, 939.
2. T. Goto, Pure Appl. Chem., 1968, 17, 421.
3. T. Goto and H. Nakamura, J.C.S. Chem. Comm., 1978, 781.
4. J.-Y. Koo and G. B. Schuster, J. Am. Chem. Soc., 1977, 99, 6107; J.-Y. Koo, S. P. Schmidt, and G. B. Schuster, Proc. Natl. Acad. Sci., 1978, 75, 30.
5. F. McCapra, J.C.S. Chem. Comm., 1977, 946.
6. J. Bergman, J. E. Bäckvall, and J. O. Lingström, Tetrahedron, 1973, 29, 971.
7. H. Nakamura and T. Goto, Photochem. Photobiol., 1979, 30, 27.
8. Satisfactory elementary analysis was obtained.

Received, 13th October, 1980