Chemiluminescence of *Renilla* (Sea Pansy) Luciferin and its Analogues

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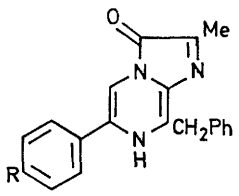
Summary *Renilla* (sea pansy) luciferin and certain of its synthetic analogues produce a brilliant blue chemiluminescence when dissolved in organic solvents such as dimethylformamide; we have examined the chemistry of this reaction and have identified the product excited state responsible for the luminescence.

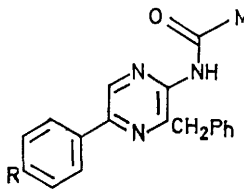
THE structure and chemical synthesis of a biologically active form of *Renilla* luciferin (Ia) has recently been accomplished.[1] We have studied the mechanism of the chemiluminescence of compounds (Ia) and (Ib) in order to provide information on the nature of the products produced during the reaction and to identify the electronic excited state responsible for the emission.

In the presence of dissolved oxygen (Ia) and (Ib) produce a brilliant blue luminescence (λ_{max} 480 and 470 nm, respectively) when dissolved in dimethylformamide (DMF). Depending upon whether (Ia) or (Ib) is used, the only detectable products are CO_2 and either (IIa) or (IIb) in high yield (80%). The structures given are in agreement with physical data (m.s., u.v., i.r., and n.m.r). Compound (IIa) was isolated as pale yellow needles, m.p. 199—200°,

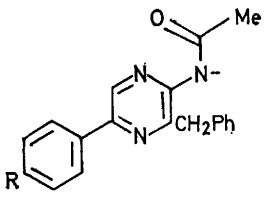
λ_{max} (DMF) 334 (ϵ_{mM} 13·6), 297 (14·7), 277 nm (13·5), δ (CD_3OD) 2·08 (3H, s), 4·25 (2H, s), 6·88 (2H, AB, J 9 Hz), 7·22 (5H, s), 7·86 (2H, AB, J 9 Hz), 8·62 (1H, s), m/e 319, 277, and 261. Compound (IIb), pale yellow needles, m.p. 212—213°, has λ_{max} (DMF) 330 (ϵ_{mM} 14·7), 294 nm (15·4), δ ($CDCl_3$) 2·18 (3H, s), 3·88 (3H, s), 4·31 (2H, s), 7·00 (2H,



(I) a; R = OH
b; R = OMe



(II) a; R = OH
b; R = OMe



(III) a; R = OH
b; R = OMe
c; R = O⁻

AB, J 9 Hz), 7·26 (5H, s), 7·96 (2H, AB, J 9 Hz), 8·61 (1H, s), m/e 333, 291, and 275. The structures of (IIa) and (IIb) were confirmed by acetylation of 2-amino-3-benzyl-5-(*p*-hydroxyphenyl)pyrazine and its methyl ether which were synthesized by previously described methods.[2]

The fluorescence emission of (IIb) (λ_{max} 470 nm), or of the spent chemiluminescence reaction of (Ib), in DMF plus potassium t-butoxide was identical to the chemiluminescence emission of (Ib) in DMF. The fluorescence emission of (IIa) (λ_{max} 530 nm), or of the spent chemiluminescence reaction of (Ia), in DMF plus potassium t-butoxide was identical to the chemiluminescence emission of (Ia) (λ_{max} 530 nm) in the same solvent system but not to the chemiluminescence emission of (Ib) (λ_{max} 470 nm) under the same conditions. Thus during chemiluminescence of (Ia) and (Ib) in DMF the electronic excited states can be represented as the monoanions (IIIa) and (IIIb), respectively. However, during chemiluminescence of (Ia) and (Ib) in DMF plus potassium t-butoxide the electronic excited state can be represented as the dianion (IIIc) and monoanion (IIIb), respectively.

In native luciferin the methyl group at the 2-position of (Ia) is replaced by an unknown group which does not interfere with the spectral properties of the native compound. Thus, during chemiluminescence the spectral observations made with native luciferin are the same as those found with (Ia) and therefore the same conclusions derived for (Ia) also appear to apply to it.

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[1] K. Hori and M. J. Cormier, *Proc. Nat. Acad. Sci., U.S.A.*, 1973, 70, 120.
[2] Y. Kishi, H. Tanino, and T. Goto, *Tetrahedron Letters*, 1972, 2747.