

- (13) A. Nickon, G. P. Pandit, and R. O. Williams, *Tetrahedron Lett.*, 2851 (1967).
 (14) R. S. Bly, R. K. Bly, A. O. Bedenbaugh, and O. R. Vail, *J. Am. Chem. Soc.*, **89**, 880 (1967).
 (15) We also generated the brexyl-5 cation by acetolysis of exo and endo brexyl-5 brosylate to produce exo-endo acetates **14** and brexene-4 (<6%). Separately we established that cation **13** does not convert to **15** or **16** (and vice versa) even under equilibrating conditions.
 (16) R. R. Sauers and J. A. Whittle, *J. Org. Chem.*, **34**, 3579 (1969).
 (17) A. Nickon, F. Huang, R. C. Weglein, K. Matsuo, and H. Yagi, *J. Am. Chem. Soc.*, **96**, 5264 (1974).
 (18) All new compounds (**10a**, **10c**, exo-**14**, endo-**14**) were characterized spectroscopically and gave satisfactory elemental analyses.
 (19) An additional view that emerges from this study is that changes in migration ratio (e.g., as a function of solvent, departing group, etc.) might be used to probe the subtleties of timing in Wagner-Meerwein rearrangements for systems where the sp² and sp³ alignment factors favor different bonds.
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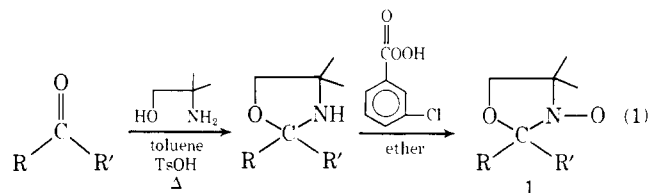
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A Versatile Synthesis of Doxyl Spin Labels Bypassing the Usual Ketone Precursors

Sir:

Doxyl (4,4-dimethyloxazolidine-*N*-oxyl) spin labels, since their inception in 1967,¹ have proven highly productive in studies of anisotropic motion, orientation, polarity, and diffusion within biological systems.² Heretofore, the doxyl group was prepared by condensation of an appropriate ketone with 2-amino-2-methyl-1-propanol followed by oxidation of the resulting oxazolidine with *m*-chloroperoxybenzoic acid (MCPA) (eq 1).¹ We now describe a new versatile method for assembling doxyl spin labels which does not depend on the availability of the ketone and which permits facile synthesis of doxyl derivatives containing unsaturation in the carbon chain, for example.



In this method advantage is taken of the availability of an impressive variety of 4,4-dimethyloxazoline derivatives **2-4** (eq 2) stemming from the elegant work of Meyers.³⁻⁵ It occurred to us that if these oxazolines could be converted into the corresponding nitrones, the latter might be susceptible to attack by an organometallic reagent⁶ leading to an *N*-hydroxyoxazolidine, mild oxidation of which would produce the corresponding doxyl derivative.

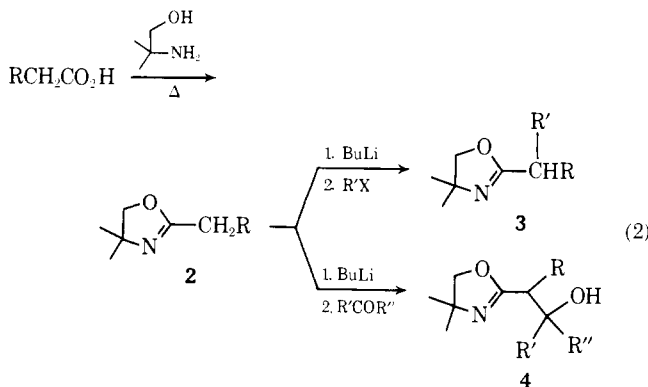
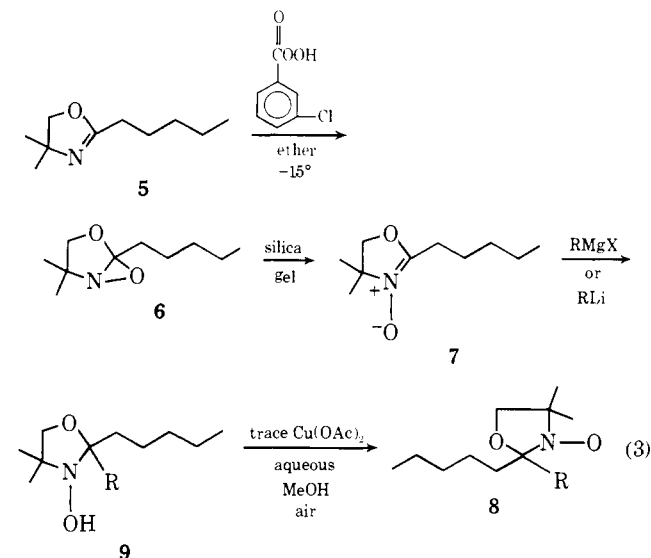


Table I. Summary of Experimental Conditions and Results

Doxyl derivative 8	Organometallic reagent	Temp, °C	Reaction time	% yield ^d
R = CH ₃ - <i>a</i>	CH ₃ MgI	-15	5 min	30
R = CH ₃ CH ₂ - <i>b</i>	CH ₃ CH ₂ MgBr	-15	5 min	27 ^e
R = CH ₃ (CH ₂) ₆ - <i>b</i>	CH ₃ (CH ₂) ₆ MgBr	-15	5 min	27 ^e
R = CH ₂ =CH- <i>b</i>	CH ₂ =CHLi ^c	-78	2 hr	29 ^e

^a Identical by ir with sample prepared by eq 1. ^b A satisfactory C, H, and N elemental analysis was obtained. All doxyl derivatives showed the expected mass spectral fragmentation patterns¹² and each (10⁻⁴ M in EtOAc) showed the typical three-line nitroxide ESR spectrum. ^c Alfa Chemical Co. ^d Isolated yield, based on starting nitron. ^e Analytical sample obtained by preparative VPC on a 2 ft 5% SE 30/Firebrick column.

To this end 2-pentyloxazoline (**5**) (eq 3) was prepared in 80% yield from hexanoic acid and 2-amino-2-methyl-1-propanol by the method of Allen and Ginos.⁷ Dropwise addition of an ether solution of 1.0 equiv of MCPA to an ether solution of **5** at -15° followed by a 24-hr period at 8° produced, after washing the ether solution with cold 10% Na₂CO₃ and drying (K₂CO₃), oxaziridine **6** as an oil (stable when stored at -20°) (90% yield, >97% pure by NMR).⁸ A CHCl₃ solution (0.5 ml) of crude **6** (150 mg) was adsorbed onto a dry silica gel column (1 × 15 cm) and CHCl₃ was added to the top of the column such that after 1 hr the CHCl₃ eluant containing the impurities present with **6** arrived at the bottom. Subsequent elution with MeOH afforded nitron **7**⁹ (140 mg (95%), solvent removal <25°) as an oil which is best used immediately as it is slowly converted into a hydrate upon standing at 25°.



Reaction of nitron **7** (100 mg) with a two-three-fold excess of the Grignard or alkyllithium reagent in ether (10 ml) at low temperatures gave, after cold aqueous workup, the corresponding *N*-hydroxyoxazolidine **9** (by NMR).¹⁰ Crude **9** was dissolved in MeOH-H₂O (9:1) (10 ml) containing ~2 mg of Cu(OAc)₂·H₂O¹¹ and the solution was stirred under air for 2 hr at 25°. Dilution with H₂O and extraction of the product with ether afforded, after chromatography over silica gel, the pure nitroxide **8** (see Table I). The isolated yield of nitroxide **8** (R = CH₃) compares favorably to that obtained via the route outlined in eq 1.¹ Vinyl nitroxide **8** (R = vinyl) is the first doxyl molecule incorporating an olefinic linkage in the doxyl chain. Unsaturated doxyl molecules will undoubtedly become important in assessing the role of unsaturation in biological systems using the spin labeling technique.²

Note Added in Proof. The "hydrate" of nitron 7 has been shown to be 2-methyl-2-(hydroxyamine)propyl hexanoate.

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References and Notes

- (1) J. F. W. Keana, S. B. Keana, and D. Beetham, *J. Am. Chem. Soc.*, **89**, 3055 (1967).
- (2) For reviews, see O. H. Griffith and A. S. Waggoner, *Acc. Chem. Res.*, **2**, 17 (1969); H. M. McConnell and B. G. McFarland, *Q. Rev. Biophys.*, **3**, 91 (1970); P. Jost, A. S. Waggoner, and O. H. Griffith, "Structure and Function of Biological Membranes", L. J. Rothfield, Ed., Academic Press, New York, N.Y., 1971, p 83; P. Jost and O. H. Griffith, "Methods in Pharmacology", Vol. 2, C. F. Chignell, Ed., Appleton-Century-Crofts, New York, N.Y., 1972, pp 223-276; I. C. P. Smith, "Biological Applications of Electron Spin Resonance Spectroscopy," J. R. Bolton, D. Borg, and H. Schwartz, Ed., Wiley-Interscience, New York, N.Y., 1972, pp 483-539; R. J. Melhorn and A. D. Keith, "The Molecular Biology of Membranes," F. C. Fox and A. D. Keith, Ed., Sinauer Associates, Stamford, Conn., 1973, pp 192-227.
- (3) A. I. Meyers, D. L. Temple, R. L. Nolen, and E. D. Mihelich, *J. Org. Chem.*, **39**, 2778 (1974).
- (4) A. I. Meyers, E. D. Mihelich, and R. L. Nolen, *J. Org. Chem.*, 2783 (1974).
- (5) A. I. Meyers, D. L. Temple, D. Haidukewych, and E. D. Mihelich, *J. Org. Chem.*, 2787 (1974).
- (6) This reaction is analogous to the addition of methyl magnesium iodide to 2,5,5-trimethyl- Δ^1 -pyrroline *N*-oxide: E. Lunt, *Nitro Compounds, Proc. Int. Symp., Warsaw*, 291 (1963); *Chem. Abstr.*, **64**, 676 (1966); see also, J. Hamer and A. Macaluso, *Chem. Rev.*, **64**, 473 (1964).
- (7) P. Allen and J. Ginos, *J. Org. Chem.*, **28**, 2759 (1963).
- (8) Several oxaziridines have been prepared from simple imines using MCPA. See, R. G. Pews, *J. Org. Chem.*, **32**, 1628 (1967).
- (9) The 100-MHz NMR spectrum (CDCl_3) of 7 δ 1.50 (s, geminal Me); 2.61 (t, CH_2); 4.29 (s, $-\text{CH}_2-\text{O}$); uv spectrum (EtOH) 244 nm (ϵ , 4540); mass spectrum *m/e* 185.139 (molecular ion) (calcd 185.142).
- (10) The main impurity in crude 9 was the starting nitron and its hydrate. These compounds undoubtedly arose by simple formation of the anion α to the nitron by the organometallic reagent.
- (11) See, R. Bonnett, R. F. C. Brown, V. M. Clark, I. O. Sutherland, and A. Todd, *J. Chem. Soc.*, 2094 (1959).
- (12) S. Chou, J. A. Nelson and T. A. Spencer, *J. Org. Chem.*, **39**, 2356 (1974).
- (13) Fellow of the Alfred P. Sloan Foundation; recipient of a National Institutes of Health Research Career Development Award.

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On the "Variation of Fluorescence Quantum Efficiency with Selective Population of Vibrational Levels" in Chemiluminescent Electron-Transfer Reactions

Sir:

A recent paper by Keszthelyi,¹ which deals with a supposed comparison between the fluorescence quantum yields of thianthrene when populated by optical excitation or cation-anion annihilation, contains serious errors which cast considerable doubt upon his conclusions.

Basically, the author is talking about what is normally referred to as a wavelength effect in which the photochemistry and/or photophysics of an excited molecule may depend on the initial level of excitation. This is certainly known and understandable when one is dealing with different electronic states but is not expected in the lower vibrational levels of the lowest excited singlet state. In general, vibrational relaxation in S_1 (ca. 10^{12} sec⁻¹) is fast compared to intersystem crossing for molecules which fluoresce and this is to be expected for a molecule such as thianthrene which has τ_F of ca. 30 nsec. A significant variation in Φ_F across the lowest energy absorption band would be unusual. On the other hand, wavelength effects can occur when the excited state lifetime is comparable to vibrational state lifetimes.²

The author has not distinguished between the onset of absorption (the 0-0 band of the first transition) and the

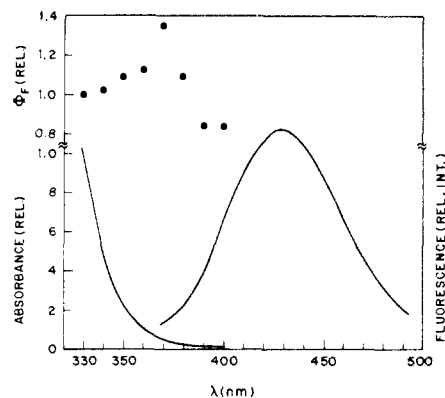


Figure 1. The absorption tail of thianthrene (ca. 10^{-3} M) in acetonitrile and the fluorescence spectrum (corrected) excited at 320 nm. The upper left part of the figure gives the relative quantum yields of fluorescence, measured at the maximum (428 nm), for solutions with optical density <0.1 (to avoid errors caused by differing absorption depths).

wavelength of maximum absorption intensity which frequently involves absorption to a higher lying state. This is the case for thianthrene where the strong band, peaking at 4.8 eV, has a long tail extending out nearly to the visible (see Figure 1). Thus the first transition is only weakly allowed, necessitating a long-lived excited state and accounting for the low fluorescence yield. This point has been discussed briefly by Bonnier and Jardon.³ Their results ($\Phi_F = 3.6\%$, $\tau_F = 3 \times 10^{-8}$ sec) suggest a natural lifetime of about 1 μ sec, indicating a very weak transition. The absorption band related to the fluorescence appears to be hidden under the tail of the intense band and thus there is really no large Stokes shift. Further, there is confusion regarding the 0-0 band of the fluorescence (or its onset) and λ_{max} ; the former, not the latter, represents the energy of the excited state in question.

The question of a wavelength effect on the fluorescence of thianthrene can be decided by simply measuring the relative values of Φ_F as a function of excitation wavelength. We have done this experiment and found no evidence for any meaningful variation in Φ_F in the absorption tail. We believe that our experimental error is certainly less than 50% and this is sufficiently accurate for comparison with the enormous factors of 2, 10, 50, and 1000 reported by Keszthelyi. The absorption and fluorescence (corrected) spectra are shown in Figure 1 which also includes Φ_F . The errors are as large as they are because of the very weak absorbance of the solution in the region of the long tail into the visible. It is also quite possible that a trace impurity could be responsible for the observed deviation of the quantum yield.

The experimental results given by Keszthelyi do not support his conclusions in any substantive manner and the following points are noteworthy. (1) The luminescence intensity observed when DPA is present is enormously greater than that for thianthrene itself. The two fluorescence spectra overlap, and the measurement does not and, indeed, cannot distinguish which species is the emitter. Note that the fluorescence yield of DPA (~ 1) is more than 25 times that of TH. (2) The oxidation potential of DPA in CH_3CN (1.3 V vs. SCE) is so little above TH that it would not be possible to oxidize the latter without involving the former. Thus the reaction of DPA^- with DPA^+ can compete with the $\text{DPA}^- \text{TH}^+$ reaction. The same conclusion results from a consideration of data for DMF solutions given recently by Keszthelyi, Tachikawa, and Bard.⁴ (3) The triplet level of TH (~ 480 nm, 2.6 eV)⁴ is considerably above that of DPA