solution was allowed to gradually warm to room temperature overnight before 5 mL of 2% HCl in saturated $NH_4Cl$ and 5 mL of ether were added. The layers were separated, and the aqueous layer was washed twice with 5-mL portions of ether. The combined ether layers were dried over $MgSO_4$, filtered, and concentrated in vacuo to afford a crude oil. The extent of reaction was determined to be 0.20 by analytical HPLC techniques. The excess PhSSPh was removed by MPLC separation using 5% EtOAc/hexane as the solvent to afford 0.073 g (90% recovery) of a mixture of **6**, **4**, **21** and **22**. The recovered starting materials **6** and **4** were found to contain 47.0% $d_2$, 3.5% $d_1$, and 49.5% $d_0$ material, and the substituted products **21** and **22** were found to contain 39.3% $d_2$, 5.6% $d_1$, and 55.1% $d_0$ material.

**$^{13}$C NMR Spectrum of 20.** To a solution of 0.147 g (0.595 mmol) of **4** in 2 mL of THF-$d_8$ in a 10-mm NMR tube at −78 °C under a nitrogen atmosphere was added 0.51 mL (0.71 mmol) of *s*-BuLi. The solution was vigorously shaken by hand to insure complete mixing. The yellow anion solution was placed into the spectrometer, which had been previously cooled to −70 °C and locked on the resonance at 3.6 ppm of THF-$d_8$. The sample was allowed to equilibrate to the temperature of the probe over 15 min before the $^1$H decoupled, $^{13}$C NMR spectrum was obtained. The center peak of the downfield quintet of the THF-$d_8$ was used as the reference peak and was set to be 67.5 ppm: $^{13}$C NMR of **20** in the region from 40 to 200 ppm (75 MHz) $\delta$ 42.5, 46.7, 49.2, 55.9 (br), 103.1, 108.4, 116.3, 127.9, 129.6, 154.2, 185.4. The anion solution was quenched with excess $CH_3OD$ to give the $\beta$-deuterated product, which was found to contain 95% $d_1$ material at the $\beta$-position as determined by $^1$H NMR integration. The $^{13}$C NMR spectrum of **4** at −70 °C in THF-$d_8$ was obtained in a similar manner: $^{13}$C NMR of **4** (75 MHz) $\delta$ 19.0, 20.6, 20.9, 21.1, 21.2, 39.6, 41.6, 46.2, 49.2, 126.8, 128.9, 130.2, 141.8, 174.3.

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**Supplementary Material Available:** For **7**, crystal and experimental details, solution and refinement summary, ORTEP figures of the two independent molecules, atomic coordinates, thermal parameters, and bond lengths and angles; experimental procedures for the synthesis of **7**, **8**, **13–16**, and **23**; and the metalation of amides **8–12** and mixtures **2** and **3** (35 pages). Ordering information is given on any current masthead page.

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# Amine–Flavin Electron Transfer Photochemistry. Potential Models for Monoamine Oxidase Catalysis and Inhibition

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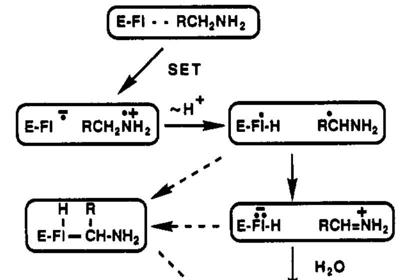
The photoreactions of 3-methyllumiflavin (3MLF) and a variety of amines have been explored. These studies have demonstrated that 3MLF undergoes efficient photoreactions with $\alpha$-silyl tertiary benzylamines to generate 4a-adducts by pathways involving sequential SET and desilylation followed by radical coupling. These adducts are unstable substances that react rapidly with nucleophiles (e.g., MeOH, $H_2O$, and $NaBH_4$) and oxygen. They are also photolabile, providing the corresponding 4a-benzyldihydroflavin upon irradiation. Non-silicon-containing primary and secondary amines also participate in SET-promoted photoreactions with 3MLF. The amine cation radicals formed in these processes undergo further transformations to produce radical intermediates by either $\alpha$-CH or NH deprotonation pathways. The potential relevance of these findings to the area of monoamine oxidase chemistry is considered.

## Introduction

**Monoamine Oxidase Biochemistry.** Monoamine oxidases (MAO) are a class of flavin-containing, membrane enzymes whose members function to control the levels of a number of biogenic amines.[1] These enzymes catalyze the oxidative deamination of their primary amine substrates (e.g. norepinephrine and serotonine) to produce aldehydes and ammonia. In recent years much attention has been given to studies of the mechanism for both catalysis by and inhibition of these enzymes. This intense interest has been stimulated by observations which show that inhibitors of these enzymes display important pharmacological properties related to their use as medicinal agents in the treatment of depression[1a,2] and Parkinson's disease.[3]

Scheme I



Recently, reasonable radical mechanisms have been proposed for both the catalytic and inhibition reactions of the monoamine oxidases. Krantz and Lewis, for example, have formulated a single electron transfer (SET) mechanism for the pharmacologically relevant inhibition reactions of propargylic (e.g. pargyline) and related alle-

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[1] (a) Kapeller-Adler, R. *Amine Oxides and Methods for their Study*; Wiley: New York, 1970. (b) *Monoamine Oxidases*; Singer, T. P., von-Korff, R. W., Murphy, D. L., Eds.; Academic Press: New York, 1979. (c) Kearney, E. B.; Salach, J. J.; Walker, W. H.; Seng, R.; Singer, T. P. *Biochem. Biophys. Res. Commun.* **1971**, *42*, 490. (d) Singer, T. P. *J. Neural Transm. Suppl.* **1987**, *23*, 1.

[2] (a) Palfreyman, M. G.; McDonald, I. A.; Bey, P.; Schechter, P. J.; Sjoerdsma, A. *Prog. Neuro-Psychopharmacol. Biol. Psychiatry* **1988**. (b) Squires, R. F. *Adv. Biochem. Psychopharmacol.* **1972**, *5*, 355. Neft, N. H.; Goridis, C. *Ibid.* **1972**, *5*, 307. Selikoff, I. J.; Robitzek, E. H.; Ornstein, G. G. *Q. Bull. Sea View Hosp.* **1972**, *13*, 17. Ho, B. T. *J. Pharm. Sci.* **1972**, *61*, 821. Sadler, M. *Proc. R. Soc. Med.* **1973**, *66*, 947.

[3] Birkmayer, W.; Knoll, J.; Riederer, P.; Youdin, M. B. H. *Monoamine Oxidase and Its Selective Inhibitors*; Beckmann, H., Riederer, P., Eds.; Karger, Busel: Rinne, U.K., pp 170–1766. *Acta Neurol. Scand. Suppl.* **1983**, *95*, 1.

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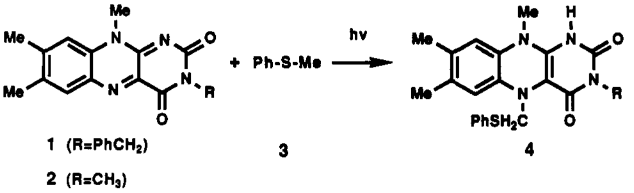
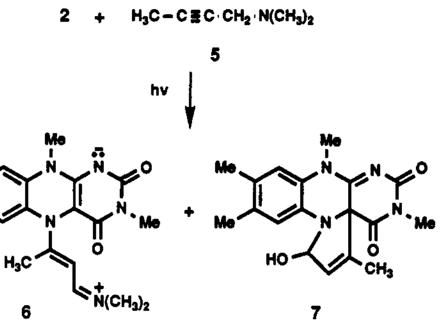
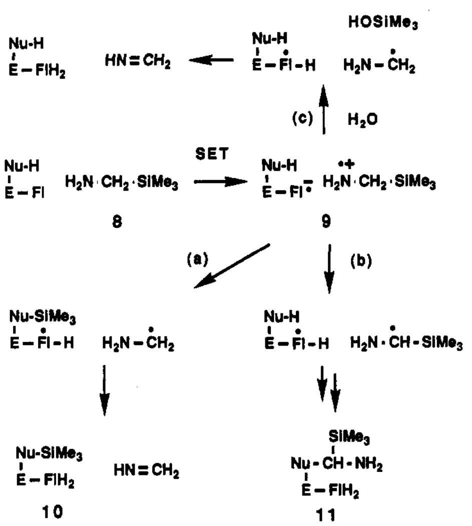
Scheme II



Scheme III



Scheme IV



nylmethylamines with MAO based upon the close similarity between the adducts formed in the enzyme inhibition reactions[4] and model flavin–amine photochemical processes.[4a,5] Silverman's[6] detailed studies of monoamine oxidase inhibition by a variety of cyclopropyl- and cyclobutylamines have provided results that also appear to implicate SET mechanisms in the biochemistry of this enzyme.

Based on these investigations, Silverman[6a] has proposed that the MAO-catalyzed oxidative deamination of amine substrates proceeds via a pathway initiated by SET from the amine donors to the covalently linked flavin grouping of the enzyme. This is followed by proton transfer from the amine cation radical to the flavin anion radical, yielding a radical pair that undergoes a second SET step to produce an imine precursor of the carbonyl product and ammonia (solid arrows in Scheme I). Reoxidation by triplet dioxygen of the dihydroflavin grouping in the reduced enzyme occurs following this sequence either before or after product release.[7] Alternatively, product formation could occur through a flavin substrate, C-4a or N-5 covalent intermediate formed by bond formation between radical pair or ion pair partners (dashed arrows in Scheme I). To our knowledge no evidence has yet been gained to allow inclusion or dismissal of the covalent intermediate mechanism for MAO catalysis. In contrast, inhibition of MAO by suicide substrates such as the allenylmethyl and propargylic amines does involve covalent bond formation at the flavin C-4a and N-5 positions (see above).[4]

**Flavin Photochemistry.** The relationship that may exist between MAO biochemistry and flavin photochemistry is interesting. A wide variety of flavin photoaddition reactions were uncovered in the past two decades principally through the intense efforst of Hemmerich and his co-workers.[8] These processes (e.g., Scheme II) that result in covalent bond formation at the flavin C-4a or N-5 sites are now recognized[9] to operate by SET mechanisms which mimic the initial steps in the proposed mechanism for MAO-catalyzed reactions of amine substrates. Likewise, Krantz[4a] has shown that the propargylic amine **5** and 3-methyllumiflavin (**2**) are transformed to the flavocyanine containing N-5 adduct **6** and the N-5, C-4a bridged adduct **7** upon irradiation (Scheme III), again demonstrating that covalent bond formation occurs in a process initiated by flavin excited state SET.

**MAO Inhibition by an α-Silyl Amine.** Of particular importance to the studies reported below are observations made recently by Silverman in his investigations of MAO inactivation by the α-amino silane **8**[10] (and a related α-aminogermane).[11] Inhibition observed in this instance has been attributed to the operation of an SET mechanism analogous to those followed in α-silyl amine enone, SET-induced photochemical reactions probed earlier by Mariano and his co-workers.[12] Silverman initially[10a] proposed that MAO inactivation by **8** is caused by the formation of a silylated enzyme **10** arising by transfer of the trimethylsilyl group from an intermediate cation radical to an active site nucleophile (pathway a in Scheme IV). More recent[10b] labeling experiments, however, have led Silverman to refine this original proposal. It is now believed that **8** undergoes MAO-induced oxidation by two competing routes. The first involves proton transfer in the

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(4) (a) Krantz, A.; Kokel, B.; Sachdeva, Y. P.; Salach, J.; Claesson, A.; Sahlberg, L. *Drug Action and Design. Mechanism Based Enzyme Inhibition*; Kalman, T., Ed.; Elsevier-North Holland: New York, 1979. (b) Krantz, A.; Lipkowitz, G. *J. Am. Chem. Soc.* **1977**, *99*, 4156. (c) Maycock, A. L.; Ables, R. H.; Salach, J. J.; Singer, T. P. *Biochemistry* **1976**, *15*, 14.

(5) (a) Simpson, J. T.; Krantz, A.; Lewis, F. D.; Kokel, B. *J. Am. Chem. Soc.* **1982**, *104*, 7155. (b) Krantz, A.; Kokel, B. *J. Org. Chem.* **1980**, *45*, 4245. (c) Gartner, B.; Hemmerich, P.; Zeller, F. A. *Eur. J. Biochem.* **1976**, *63*, 211. (d) Zeller, E. A.; Gartner, B.; Hemmerich, P. *Z. Naturforsch.* **1972**, *276*, 1050. (e) Franke, J.; Ott, U.; Kramer, H. E. A.; Traber, R. H. *Photochem. Photobiol.* **1989**, *49*, 131.

(6) (a) Silverman, R. B.; Hoffman, S. J.; Catus, W. B. *J. Am. Chem. Soc.* **1980**, *102*, 7126. Silverman, R. B.; Hoffman, S. J. *Biochem. Biophys. Res. Commun.* **1981**, *101*, 1396. (b) Vazquez, M. L.; Silverman, R. B. *Biochemistry* **1985**, *24*, 6538. Silverman, R. B.; Zieske, P. A. *Ibid.* **1985**, *24*, 2128. (c) Silverman, R. B.; Zieske, P. A. *Biochemistry* **1986**, *25*, 341. Yelecki, K.; Lu, X.; Silverman, R. B. *J. Am. Chem. Soc.* **1989**, *111*, 1138.

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(9) (a) Novak, M.; Miller, A.; Bruice, T. C.; Tollin, G. *J. Am. Chem. Soc.* **1980**, *102*, 1465. (b) Traber, R.; Vogelmann, E.; Schreiner, S.; Werner, T.; Kramer, H. E. A. *Photochem. Photobiol.* **1981**, *33*, 41.

(10) (a) Silverman, R. B.; Banik, G. M. *J. Am. Chem. Soc.* **1987**, *109*, 2219. (b) Banik, G. M.; Silverman, R. B. *Ibid.* **1990**, *112*, 4499.

(11) Silverman, R. B.; Vadnere, M. K. *Bioorg. Chem.* **1987**, *15*, 328.

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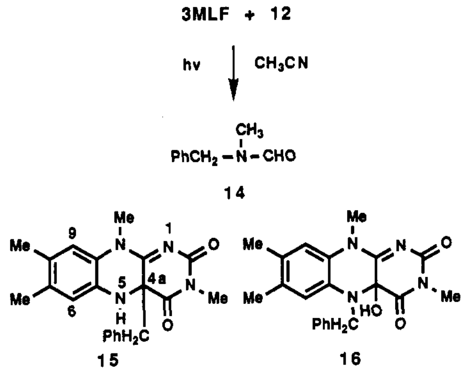
intermediate ion radical pair 9 (pathway b in Scheme IV) followed by a second SET step to form the intermediate silyliminium cation which is transformed to the covalent adduct 11 by addition of an active site nucleophile. In the second route, desilylation of the $\alpha$-silyl amine cation radical occurs (pathway c, Scheme IV) to yield, after a second SET step and hydrolysis, the normal formaldehyde and ammonia products. Importantly, this work contained no observation to suggest that inactivation is associated with covalent bond formation between the inhibitor and the flavin moiety of the enzyme. The unprecedented reactivity of the $\alpha$-silyl amine cation radical (i.e., deprotonation rather than desilylation in the absence of strong bases)[12] and the unusual formation and stability of the $\alpha$-silyl amine adduct 11 (compared to the reactivity of the related substrates benzylamine and neopentylamine) stand as particularly intriguing features of the Silverman mechanism for MAO inactivation by the $\alpha$-silyl amine 8.

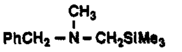
**Flavin–Amine Photochemical Studies.** The potential relationships that exist between the SET photochemistry of amines[13] and the pathways proposed for MAO catalysis and inhibition as of yet have not been subjected to experimental scrutiny except in the case of propargylic amine systems. Although the Lewis and Krantz study[5a] provided preliminary information about the interactions of saturated amines with the excited states of 3-methyllumiflavin, little information was gained about the detailed nature of the chemical reaction pathways followed.[14] Our interests in the area of SET photochemistry and, more specifically, in the photoaddition and photocyclization reactions of $\alpha$-silyl amines has stimulated the current investigation of flavin–amine excited state processes. The aims of this effort were to (1) explore the gross nature of these excited state reactions and (2) investigate their mechanistic features. The results of this effor could shed light on both the photochemical and MAO reactions and provide information about the relevance of SET mechanisms in the pathways for MAO catalysis and inhibition. In these investigations we have used 3-methyllumiflavin (3MLF, 2) as the prototype flavin since its key excited state and electrochemical properties as well as those of its derived radical (i.e., protonated semiquinone) are known.[5a] The substrates included a series of saturated primary, secondary, and tertiary amines, $\alpha$-trimethylsilyl analogues, and a carbamate derivative. The products of preparative photochemical reactions have been characterized and the nature of air- and/or water-sensitive intermediates formed have been elucidated by $^1$H NMR spectroscopic methods. Finally, the mechanistic sequences involved in these photochemical reactions are compared to those proposed for the MAO-catalyzed biochemical processes.

## Results

**Photoreactions of 3MLF with Selected Tertiary Amines.** Our initial efforts concentrated on the photoreactions of 3MLF with $N$-methyl-$N$-benzyl-$N$-[(trimethylsilyl)methyl]amine (12) and the non-silicon-containing analogue 13. The known[15b] $\alpha$-silyl amine 12 used in this study was prepared by silylmethylation of $N$-methyl-$N$-benzylamine following the general method of Noll.[15a] 3MLF was synthesized by the procedure described by Hemmerich.[15c] Preparative irradiations were conducted under the conditions described in the Experimental Section.

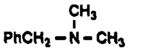
Scheme V





$$\mathrm{PhCH_2-\overset{CH_3}{\overset{|}{N}}-CH_2SiMe_3}$$

12



$$\mathrm{PhCH_2-\overset{CH_3}{\overset{|}{N}}-CH_3}$$

13

Irradiation of an MeCN solution of 3MLF and the $\alpha$-silyl amine 12 for 2 h (>75% conversion of 3MLF) led to production of three products characterized as $N$-methyl-$N$-benzylformamide (14) (57%), the 4a-benzyl-4a,5-dihydroflavin 15 (18%), and the 5-benzyl-4a-hydroxydihydroflavin 16 (<1%) along with recovered 3MLF (73%) (Scheme V). Spectroscopic data for the formamide 14 matched those of the independently synthesized material.[16] Likewise the structures of the modestly stable benzyldihydroflavins 15 and 16 were assigned by comparisons of their physical and spectroscopic properties with those of known substances prepared by the previously reported method of Hemmerich.[17] These adducts were generated for comparison purposes by photoaddition of tetraethylammonium phenylacetate to 3MLF in $H_2O$.

Formation of the benzyldihydroflavins 15 and 16 in this reaction was not expected. Consequently, additional experiments to gain information about the mechanistic origin(s) of these substances were conducted. Irradiation of 3MLF in the presence of silyl amine 12 in either 18% $D_2O$–MeCN or 18% $H_2O$–$CD_3CN$ gave the 4a-benzyl adduct 15 (in reduced yields; see below). $^1$H NMR analysis showed that in neither cases were deuteria incorporated at the benzylic position in this substance. Likewise, photoaddition of the dideuteriobenzylamine 12-$d_2$ in MeCN gave only benzyl-$d_2$ analogues of 14, 15, and 16. The combined results demonstrate that the benzyl groups in adducts 15 and 16 (and in amide 14) derive from the amine precursor 12 by pathways in which the benzylic C–H bonds are unaltered (see below).

Additional observations have provided further information about the nature and scope of flavin–amine photoreactions. For example, irradiation of a deoxygenated solution of 3MLF and the silyl amine 12 in a $H_2O$–MeCN mixture leads to generation of $N$-methyl-$N$-benzylamine along with the 4a-benzyl adduct 15. Furthermore, when the photolysate, derived by irradiation of an MeCN solution of 3MLF and silyl amine 12, is quenched by the addition of $NaBH_4$ prior to introduction of oxygen, $N$,$N$-

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(13) Pienta, N. J. In *Photoinduced Electron Transfer*; Fox, M. A., Chanon, M., Eds.; Elsevier: Amsterdam, 1988; Part C.

(14) Lewis and co-workers (ref 5a) have reported that while amine quenching of the 3MLF singlet excited state occurs at a near diffusion controlled rate it is nonproductive and that quenching of the 3MLF triplet is responsible for product formation. Product analysis was only reported for the 3MLF photoreaction with tri-$n$-propylamine. In this case propanal and di-$n$-propylamine were the major products detected.

(15) (a) Noll, J. *J. Am. Chem. Soc.* **1951**, *73*, 3867. (b) Koizumi, T.; Fuchigami, T.; Nanaka, T. *Bull. Chem. Soc. Jpn.* **1989**, *62*, 219. (c) Hemmerich, P.; Fallob, S.; Erlenmeyer, H. *Helv. Chim. Acta* **1956**, *39*, 1242.

(16) Freundenrich, C.; Samana, J. P.; Biellman, J. F. *J. Am. Chem. Soc.* **1984**, *106*, 3344.

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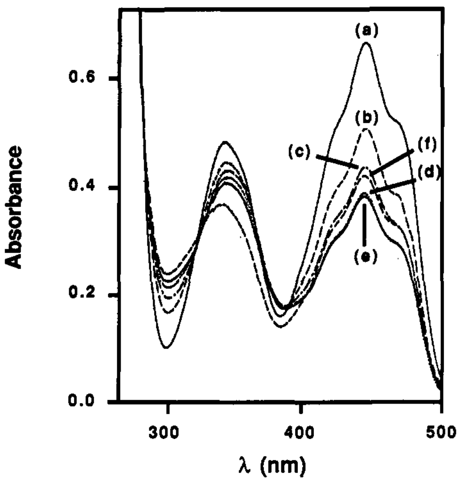


**Figure 1.** Time course revealed by UV spectroscopic monitoring of the reaction induced by irradiation (λ = 444 nm) of $4.9 \times 10^{-5}$ M 3MLF and $4.1 \times 10^{-4}$ M silyl amine 12 in $CH_3CN$. Spectra were recorded at (a) 0 min, (b) 15 min, (c) 30 min, (d) 45 min, (e) 60 min, and (f) 90 min time periods.

dimethyl-*N*-benzylamine 13 is produced. Finally, *N*,*N*-dimethyl-*N*-benzylamine 13 serves as a substrate in photoreaction with 3MLF. Accordingly, irradiation of an MeCN solution of 3MLF and 13 gives the same products and in the same distribution as is obtained from the 3MLF reaction with 12.

**The Progress of the 3MLF Photoreaction with Silyl Amine 12.** In order to determine how the benzyldihydroflavin and other products are formed in these photoreactions, the progress of the 3MLF–silyl amine 12 reaction was followed by UV spectroscopy. As displayed in Figure 1, absorptions at the wavelength maxima for 3MLF (343 and 440 nm) decrease upon irradiation. The 440 band decreases more rapidly because the photoadducts formed in this process have maxima at ca. 340–360 nm. The absence of isosbestic point(s) in the time course plot and the dependence of the changes on irradiation time suggest that the conversion of 3MLF and 12 to 4a-benzyldihydroflavin 15 involves initial formation of at least one intermediate and, as a result, more than one photochemical step. The presence of a long-lived yet unstable intermediate in this photoreaction is further evidenced by the observation that 3MLF is recovered from the preparative reactions in high yield (ca. 73%) even though UV analysis prior to workup indicates that ca. 75% of 3MLF is consumed. Moreover, the major product generated is the formamide 14 even though the irradiation solutions were oxygen-free. The combined results suggest that an initial intermediate is produced in the primary photochemical step in the reaction of 3MLF with 12 and that this species undergoes (1) a secondary photoreaction to generate the benzyldihydroflavin 15 under the irradiation conditions (λ > 320nm) and (2) a dark reaction occurs on work-up of the photolysate in air to form the amide 14 and 3MLF.

**Analysis of the 3MLF Photoreaction with Silyl Amine 12 by $^1$H NMR Spectroscopy.** Pertinent information about the characteristics of the 3MLF–silyl amine 12 photoreaction has come from a study in which the progress of this process is monitored by $^1$H NMR spectroscopy. The general procedures used are described in the Experimental Section. This technique made it possible to identify the unstable, primary photoadduct produced in reaction of 3MLF with 12 and to elucidate the course of its reaction with light, oxygen, and nucleophiles.

The $^1$H NMR spectroscopic data accumulated are displayed in Figures 2 and 3. As can be seen by inspection of these data, short period (10 min) irradiation (λ > 320nm) of a solution of 3MLF and the silyl amine 12 in $CD_3CN$ results in disappearance of the starting flavin and the amine with concomitant formation of a new substance 17 (Scheme VI) having $^1$H NMR characteristics of a 4a,5-dihydroflavin (compare Figures 2a and 2b). Diagnostic in this regard are the singlets for the H-6 and H-9 aromatic, the N-10 and N-3 methyl, and the C-8 and C-7 benzylic methyl proton resonances. The identity of the primary photoproduct 17 as a C-4a flavin adduct is further

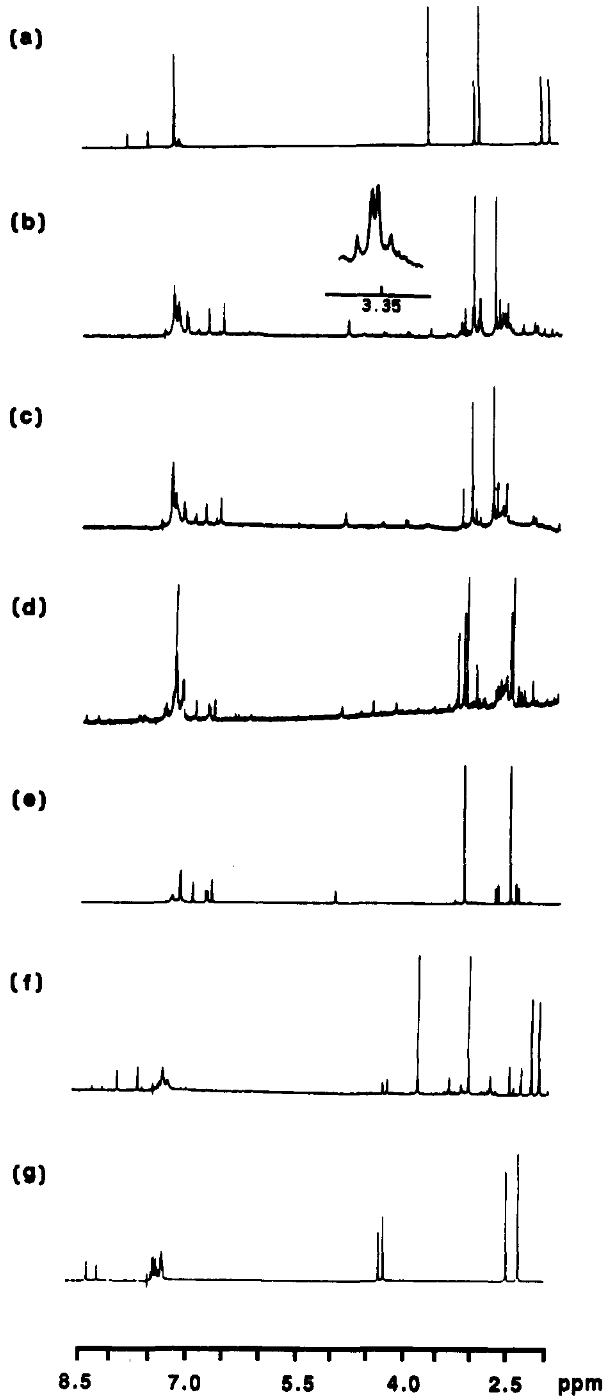
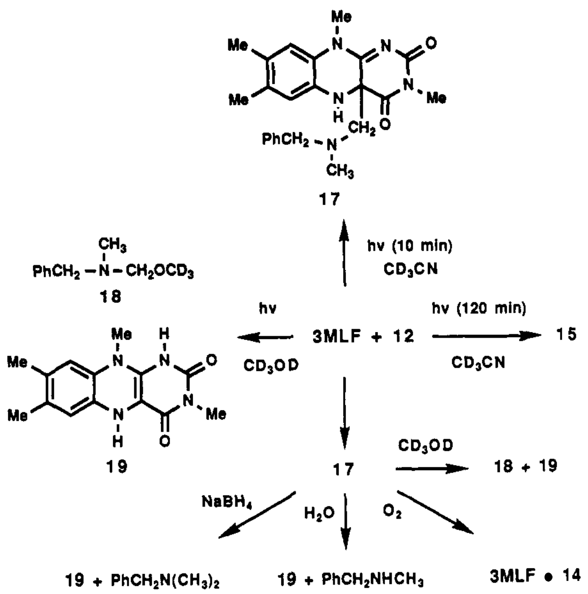


**Figure 2.** Key portions of the $^1$H NMR spectra (400 MHz) of a (a) degassed solution of 3MLF and silyl amine 12 in $CD_3CN$, (b) 10-min irradiated, degassed solution of 3MLF and 12 in $CD_3CN$, (c) 10-min irradiated, degassed solution of 3MLF and silylamine-$d_2$ 12-$d_2$ in $CD_3CN$, (d) 120-min irradiated, degassed solution of 3MLF and 12 in $CD_3CN$, (e) solution of 4a-benzyldihydroflavin 15 in $CD_3CN$, (f) 10-min irradiated, degassed solution of 3MLF and 12 followed by exposure to air, and (g) solution of formamide 14 in $CD_3CN$.

**Scheme VI**



revealed by the presence in its $^1$H NMR spectrum of an *N*-methyl singlet at 1.98 ppm (almost completely obscured by solvent resonances, Figure 2b), an N-H singlet at 5.06 ppm, and importantly an AB quartet (3.35 and 3.37 ppm) corresponding to the benzylic methylene protons, which are rendered diastereotopic by the presence of the C-4a chiral center. The latter assignment is aided by analysis of the $^1$H NMR spectrum of the primary adduct arising from reaction of the d$_2$-substituted silyl amine **12-d$_2$** with 3MLF (Figure 2c). Unfortunately, the $\alpha$-amino methylene protons in **17** resonate in regions obscured by solvent. This $^1$H NMR method has shown that adduct **17** is also formed initially in photoreaction of 3MLF with the non-silicon-containing, tertiary amine **13**.

This study has further demonstrated that **17** only slowly (24 h, 25 °C) decomposes in the dark and in the absence of oxygen or nucleophiles and that is serves as the precursor of the secondary photoproduct, 4a-benzyldihydroflavin **15**. $^1$H NMR analysis (Figure 2d) of a degassed solution of 3MLF and silyl amine **12** in $CD_3CN$ following irradiation for a long time period (120 min) clearly shows the presence of the benzyl adduct **15** (compare Figures 2c and 2e) and no primary adduct **17**. In addition, exposure of a $CD_3CN$ solution of **17**, formed by short-period irradiation, to air results in its rapid conversion to the formamide **14** and 3MLF (compare Figures 2f and 2g).

More information about the reactivity of the primary flavin photoadduct **17** has been gained through its trapping by methanol. Accordingly, $^1$H NMR monitoring of an irradiated (10 min) $CD_3OD$ solution containing 3MLF and silyl amine **12** shows that clean (ca. 100%) formation of the $\alpha$-amino ether **18** (Scheme VI) occurs (compare Figures 3a and 3b). The spectrum of this photolysate matches that of an authentic sample of **18** prepared by reaction of *N*-methyl-*N*-benzylamine with formaldehyde in $CD_3OD$ (Figure 3d). Clear evidence for the fact that amino ether **18** derives from rapid secondary reaction of the primary photoadduct **17** is found in the observation that **18** is produced (ca. 68%) when a $CD_3CN$ solution of **17** is diluted with $CD_3OD$ (Figure 3c). It is difficult to completely exclude oxygen during the $CD_3OD$ addition and, consequently, less than quantitative conversion to **18** is obtained and 3MLF is formed by oxidation of 1,5-dihydrolumiflavin **19**, generated in the reaction of $CD_3OD$ with **17**. The dihydroflavin **19** is obtained in the photoreaction of 3MLF with **12** in degassed $CD_3OD$ but its low solubility (a precipitate is formed) prevents its $^1$H NMR detection. Lastly, $^1$H NMR monitoring has also demonstrated that (1) irradiation of 3MLF in a 1.5% $H_2O$–98.5% $CD_3CN$ solution containing the silyl amine **12** produces significant quantities (ca. 36%) of the oxidative dealkylation product, *N*-methyl-*N*-benzylamine, and (2) the intermediate **17** reacts with $H_2O$ to form this secondary amine.

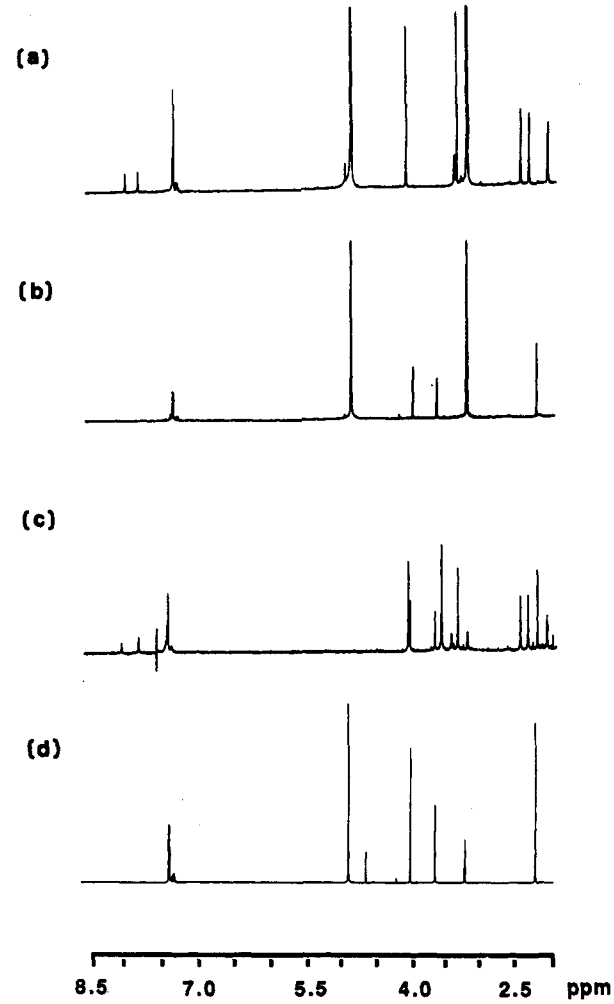
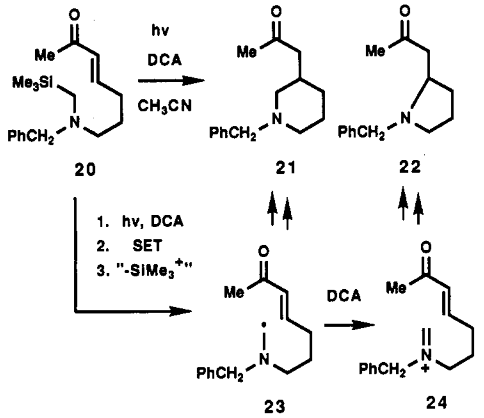


**Figure 3.** Key portions of the $^1$H NMR spectra (400 MHz) of a (a) degassed solution of 3MLF and silyl amine **12** in $CD_3OD$, (b) 10-min irradiated, degassed solution of 3MLF and **12** in $CD_3OD$, (c) 10-min irradiated, degassed solution of 3MLF and **12** in $CD_3CN$ followed by addition of $CD_3OD$, and (d) solution of amino ether **18** in $CD_3OD$ formed by reaction of *N*-methyl-*N*-benzylamine with formaldehyde and $CD_3OD$.

**Photochemistry of 3MLF with the Silyl Amino Enone 20.** In order to gain more insight into the detailed mechanistic nature of flavin–amine photoreactions, we have explored the excited state chemistry of 3MLF and the $\alpha$-silyl amino enone **20**. In previous studies,[12c] we have shown that the amino enone **20** participates in an SET-induced photocyclization reaction that produces the acetonylpiperidine **21** (Scheme VII). This process, photosensitized by 9,10-dicyanoanthracene (DCA), operates via a mechanism in which an amine cation radical, generated by SET from **20** to the singlet excited state of DCA, undergoes desilylation to form an $\alpha$-amino radical **23**. Cyclization of **23** then occurs to yield an $\alpha$-keto radical, which is transformed to **21** by sequential SET from the anion radical of DCA and protonation of the formed enolate anion. In studies with related ester systems, we have also observed[12c] that oxidation of $\alpha$-amino radicals related to **23** by thermodynamically favorable SET to DCA competes with radical cyclization and forms formaldiminium cations

Scheme VII



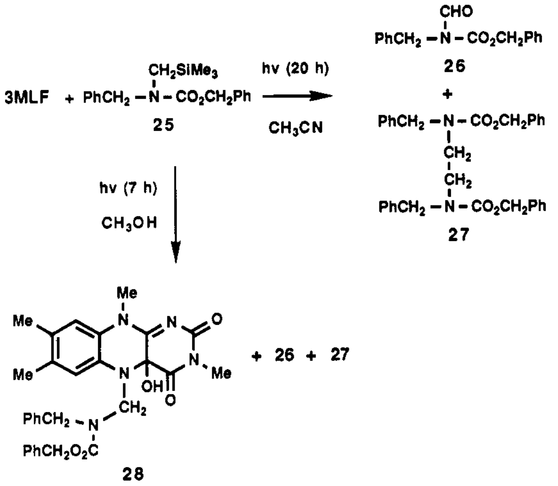
Scheme VIII

$$20 + 3MLF \xrightarrow{hv} 15 + 21 + 22$$

related to **24**. These serve as intermediates in oxidative desilylalkylation pathways yielding pyrrolidine products related to **22**. In view of these earlier observations, we anticipated that a study of the photoreaction of 3MLF with **20** would reveal pertinent information about the nature of SET-induced chemistry of flavins and, in particular, about the existence and fate of radical intermediates.

Preparative irradiation of a deoxygenated, $CH_3CN$ solution of 3MLF and **20**[18] (both at 3.1 mM) for 1 h (52% 3MLF conversion) results in generation of a product mixture containing the piperidine **21**[18] (40%), pyrrolidine **22** (44%), and a trace quantity (ca. 1%) of the 4a-benzyldihydroflavin **15** (Scheme VIII). To elucidate the mechanism for formation of pyrrolidine **22**, the **22**:**21** product ratio was determined as a function of the concentration of 3MLF in reaction with the silyl amino enone **20** (48 mM) in $CH_3CN$. This experiment clearly showed that the **22**:**21** ratio decreases as the 3MLF concentration decreases (e.g. **22**:**21** = 1.4 at [3MLF] = 2.4 mM, 0.3 at 0.49 mM, and 0.1 at 0.01 mM). Lastly, $^1H$ NMR monitoring of the progress of the photoreaction between 3MLF and **20** failed to reveal the formation of detectable quantities of an intermediate related to photoadduct **17**, which presumably serves as a precursor for the minor photoproduct, **15**. Moreover, a formamide related to **14** is not produced in this photoreaction even when irradiation is conducted on air saturated solutions. Thus, these results suggest that a primary photoadduct related to **17** is formed at most in only minute quantities in the photoreaction of 3MLF with **20**. Moreover, the pyrrolidine **22** must arise by 3MLF-induced oxidation of an intermediate α-amino radical and not by a pathway involving the intermediacy of a primary adduct (see below).

Scheme IX



**3MLF Photoreaction with the Silyl Carbamate 25.** The instability toward oxygen and nucleophiles of the primary photoadduct **17** could be due to the ability of this adduct to undergo (perhaps reversible) heterolytic cleavage of the exocyclic C-4a α-aminomethylene carbon–carbon bond. This fragmentation would give rise to a dihydroflavin anion and an iminium cation. If this route were operable, substituents on the side-chain amine nitrogen which destabilize cations should slow fragmentation and increase the stability of an adduct. Based on this proposal, we have probed reaction of the α-silyl carbamate **25** and 3MLF.

Irradiation of a deoxygenated-nitrogen, purged-$CH_3CN$ solution of 3MLF (2 mM) containing the silyl carbamate **25** led to slow (compared to reaction of 3MLF with silyl amine **12**) disappearance of the flavin. Workup of the photolysate after 20-h irradiation (31% conversion of 3MLF) followed by chromatographic separation gave recovered 3MLF (97%) and silyl carbamate **25** (55%) along with the (benzyloxy)formimide **26** (31%) and bis-carbamate **27** (ca. 1%). When the reaction solvent was changed to $CH_3OH$, the reaction time shortened to 7.5 h, and the period used for workup and chromatographic separation made shorter, the modestly stable, 5-(carbamoylmethyl)-4a-hydroxydihydroflavin **28** (12%) was isolated together with formimide **26**, bis-carbamate **27**, and recovered 3MLF (83%). These results are summarized in Scheme IX.

Identification of adduct **28** as an N-5 substituted dihydroflavin is made possible by its modest stability, an analysis of its characteristic spectroscopic properties, and by comparison of these with those accumulated for the known[17] 5-benzyl-4a-hydroxy analogue **16**. Indicative of its gross molecular composition is the mass spectrum of **28**, which contains a molecular ion at *m/e* 541 ($C_{30}H_{31}N_5O_5$) and peaks at *m/e* 270 and 254 corresponding to the 3MLF and *N*-methyl-*N*-[(benzyloxy)carbonyl]iminium cations, respectively. Both of these fragments arise by rupture of the exocyclic C–N5 bond. In addition, the presence of a methylene proton singlet at 6.97 ppm in the $^1H$ NMR spectrum associated with the N–$CH_2$–N group

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(18) For procedures to prepare the silyl amino enone **20** and characterization of the acetonylpiperidine **21**, see: Jeon, Y. T. Ph.D. Dissertation, University of Maryland, 1989. Spectroscopic data for the acetonylpyrrolidine **22** are reported in the Experimental Section.

(19) Rehm, D.; Weller, A. *Isr. J. Chem.* **1970**, *8*, 259.

(20) Das, S.; vonSonntag, C. *Z. Naturforsch.* **1986**, *416*, 505.

(21) Lewis, F. D. *Acc. Chem. Res.* **1986**, *19*, 401.

(22) Xu, W.; Mariano, P. S. *J. Am. Chem. Soc.* **1991**, *113*, 1431.

(23) (a) Lewis, F. D.; Zebrowski, B. E.; Correa, P. E. *J. Am. Chem. Soc.* **1984**, *106*, 187. (b) Lewis, F. D.; Reddy, G. D. *Ibid.* **1989**, *111*, 6465.

(24) Sugimoto, A.; Sumida, R.; Tamai, N.; Inoue, H.; Otsugi, Y. *Bull. Chem. Soc. Jpn.* **1981**, *54*, 3500.

(25) Clerin, D.; Bruice, T. C. *J. Am. Chem. Soc.* **1974**, *96*, 5571.

(26) An estimate based upon the competition between oxidation and cyclization suggests that the rate of α-amino radical **23** cyclization is in the range of $>10^7$ $s^{-1}$ (ref 12c).

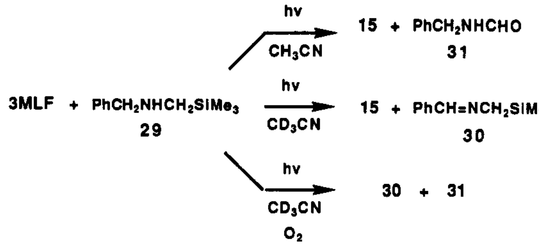
(27) Wayner, D. D. M.; McPhee, D. J.; Griller, D. *J. Am. Chem. Soc.* **1988**, *110*, 132.

(28) Anderson, R. F. *Biochem. Biophys. Acta* **1983**, *722*, 158.

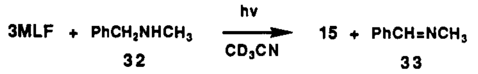
(29) The excited state oxidation potentials of dihydroflavins can be approximated based upon estimated singlet excited state energies gained from fluorescence spectroscopic data (ref 30) and ground-state oxidation potentials (ref 28).

(30) Ghisla, S.; Massey, V.; Lhoste, J. M.; Mayhew, S. G. *Biochemistry* **1974**, *13*, 589.

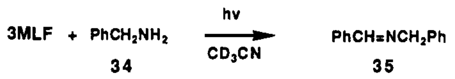
Scheme X



Scheme XI



Scheme XII



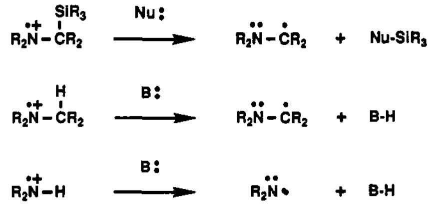
and in the $^{13}C$ NMR spectrum of methylene and quaternary carbon resonances at 67.9 (N–$CH_2$–N) and 73.4 ppm (C-4a), respectively, support the identity of **28**.

**3MFL Photoreactions with Other Primary and Secondary Amines.** Additional information pertaining to the SET behavior of flavin–amine systems has come from efforts exploring the photoreactions of 3MLF with the secondary amines, *N*-benzyl-*N*-[(trimethylsilyl)-methyl]amine (**29**) and *N*-methyl-*N*-benzylamine (**32**), and the primary amine, *N*-benzylamine (**34**). Preparative irradiation of 3MLF and the known[31] secondary silyl amine **29** in deoxygenated $CH_3CN$ for 45 min led to production of the 4a-benzyldihydroflavin **15** and known[16] *N*-benzylformamide **31**. $^1H$ NMR monitoring of the progress of this reaction revealed that irradiation in degassed $CD_3CN$ for 20 min (78% conversion of 3MLF) produced adduct **15** (10%) and the silyl benzaldimine **30** (65%). In contrast, irradiation of an air-saturated $CD_3CN$ solution of 3MLF and **29** produced the formamide **31** (42%) and imine **30** (35%). These results, outlined in Scheme X, suggest that the formamide **31** arises by oxidation of an amine–flavin adduct related to **17** during workup. Also, **15** would be produced by secondary photoreaction of this same intermediate. It should be noted that we were unable to isolate imine **30** from the preparative photoreaction mixture. This could be a result of its hydrolytic decomposition during chromatographic separation and the volatility of the derived degradation products.

Photoreaction of 3MLF with the non-silicon-containing secondary amine **32** follows the same course as with silyl amine **29**. Accordingly, $^1H$ NMR analysis of the photolysate arising by irradiation of a deoxygenated $CD_3CN$ solution of 3MLF and **32** prior to exposure to air showed the presence of *N*-methylbenzaldimine **33** (44%) and the 4a-adduct **15** (3%) (Scheme XI).

Finally, preparative irradiation of a deoxygenated $CH_3CN$ solution of 3MLF and *N*-benzylamine (**34**) leads to production of *N*-benzylbenzaldimine **35**. While the isolated (silica gel chromatography) yield of **35** is only 40%, a near quantitative yield is indicated by NMR analysis of the crude photolysate. In accord with this is the observation that **35** is the only product detected when the progress of the photoreaction of 3MLF with **34** was monitored by $^1H$ NMR. The imine **35** is formed quantitatively in sealed NMR tube reactions of 3MLF and **34** occurring in either deoxygenated or air-saturated $CD_3CN$ solutions (Scheme XII).

Scheme XIII



## Discussion

**Mechanistic Considerations.** The photochemical reactions described above are promoted by irradiation of 3MLF and involve pathways that are initiated by SET from the amine donors to the triplet excited state of the flavin. This proposal is fully supported by free energy considerations, which demonstrate that $\Delta G°_{SET}$ for electron donation from the amines ($E_{1/2}(+)$ = ca. 1–1.5 V)[15] to $3MLF^{T_1}$ ($E_{1/2}(-)$ = ca. +2.1 V)[5a] should be greatly negative and, thus, that the rate constant for SET in these systems should be near the diffusion controlled limit of ca. $1 \times 10^{10}$ $M^{-1}$ $s^{-1}$ (based on the Weller treatment).[19] Experimental evidence for this conclusion is found in earlier work by a number of groups. The flash photolysis and radical trapping studies by Bruice[9a] were the first to document the operation of an SET pathway in the excited state chemistry of flavins. Later efforts by Kramer and his co-workers[9b] confirmed this proposal. In addition, Lewis and Krantz[5a] showed clearly that propargyl and allenylamine quenching of $3MLF^{T_1}$ by SET leads to product formation while interaction of these donors with $3MLF^{S_1}$ is unproductive in terms adduct generation. This is presumably due to rapid back-electron transfer in the singlet ion radical pair produced by SET in the singlet manifold. Similar conclusions were reached in a later study by Kramer and his co-workers[5f] of the 3MLF photoaddition reaction with a tertiary, propargylic amine.

Several reaction pathways are available to triplet ion radical pair produced by amine–$3MLF^{T_1}$ SET and the relative efficiencies of these are dependent on the nature of the amine cation radical component. In our earlier investigations,[12a,b,d] we have shown that tertiary $\alpha$-silyl amine cation radicals undergo selective desilylation when paired with weakly basic anion radicals. For example, desilylation is the exclusive route followed in 9,10-dicyanoanthracene (DCA) sensitized reactions of tertiary $\alpha$-silyl amines in MeCN. In these processes the intermediate DCA anion radical is weakly basic ($pK_a$ of protonated form estimated to be <0).[12f] Thus, competitive $\alpha$ C-H deprotonation is not observed owing to the comparably unfavorable energetics of this reaction ($pK_a$ of tertiary amine cation radicals are ca. 8–10 in $H_2O$).[20] The anion radical of 3MLF is a weak base; the $pK_a$ of the 3MLF-H radical has been estimated to be ca. 8 in $H_2O$.[9b]

Cation radicals of tertiary amines that lack $\alpha$-silyl substituents react via proton-transfer processes with even weakly basic anion radicals.[13,21] The regioselectivity for $\alpha$-deprotonation in these systems is governed by statistics and kinetic acidities.[21,22] Lewis[21] has shown, for example, that the statistically corrected rates of deprotonation at benzylic and methylamine cation radical centers are nearly equivalent. The situation differs for cation radicals derived from secondary and primary amines where the presence of acidic N-H protons opens yet another route for ensuing reaction. A number of observations made by Lewis[23] and Otsuji[24] suggest that N-H deprotonation of these cation

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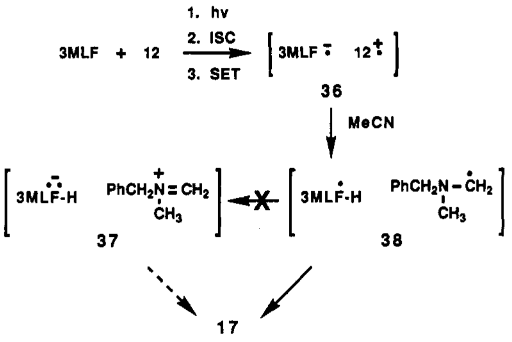
[31] Padwa, A.; Dent, W. In *Org. Synth.* **1988**, *67*, 133.

Scheme XIV

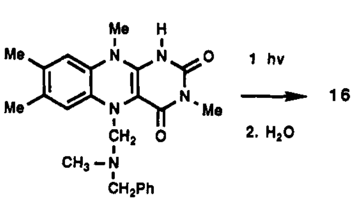


radicals is often favored over loss of an $\alpha$ C-H proton.

In summary, three pathways involving $\alpha$-desilylation, $\alpha$-deprotonation, and N-H deprotonation (Scheme XIII) are available to cation radicals formed by SET from amine donors to 3MLF.$^{T_1}$ The relative rates of these processes should determine the nature of products as well as the mechanisms for their formation.

**Adduct Formation in the Photoreaction of 3MLF with the Tertiary Silyl Amine 12.** Photoreaction of 3MLF with the tertiary $\alpha$-silyl amine 12 leading to production of the 4a-adduct 17 is easily rationalized in light of the above considerations. Accordingly, SET from 12 to 3MLF$^{T_1}$ generates the ion radical pair 36, which should undergo selective desilylation of the amine cation radical component (with MeCN as a nucleophile). This leads to formation of the $\alpha$-amino 3MLF-H radical pair 38 from which adduct 17 would arise by radical coupling at the flavin 4a-center (Scheme XIV). An alternative pathway involving SET between the $\alpha$-amino and 3MLF-H radicals followed by ion pair 37 coupling might also be a contributor to this process. However, based upon observations made in studies with the silyl amino enone 20, this redox step must be slow (see below) and perhaps noncompetitive with C-C bond formation. It is important to mention that a penultimate SET step has been proposed[5a] for propargylic amine additions to 3MLF to account for the selective nature of covalent bond formation at the amine $\gamma$ (expected for unsaturated iminium cation reactions with nucleophiles) rather than the $\alpha$ (expected for radical coupling) carbon.

Photoaddition of 12 to 3MLF favors formation of the 4a-adduct 17 over the N-5 adduct 39. The N-5 adduct presumably serves as the precursor of the minor secondary photoproduct 16. In contrast, the N-5 adduct 28 predominates in photoreaction of the carbamate analogue 25 with 3MLF. This behavior, while not easily explained, is observed in other flavin SET-photoaddition reaction (e.g. with tertiary propargyl vs allenylmethylamines).[5a] The different regiochemistries in reactions of 12 and 25 could have a kinetic origin, i.e. governed by the relative rates of radical coupling in a rapidly converting 3MLF-H radical. However, it would be difficult to see how an N-substituent in the $\alpha$-amino radical would offer regiocontrol in this event. Another possibility is that adduct formation is under thermodynamic control. Thus, an N-substituent, such as (benzyloxy)carbonyl, could slow the rate of interconversion of an initially formed N-5 adduct to a possibly more stable C-4a adduct by destabilizing an iminium cation, which could be involved as an intermediate in the equilibration process. Observations made by Bruice and Clerin[25] in their study of N-5 and C-4a indolylmethyl-substituted dihydrolumiflavins are relevant to this issue. These workers noted that equilibration of these adducts occurs in competititon with solvolysis in aqueous solution, and they offered the general proposal that initially formed N-5 adducts may be precursors of C-4a adducts depending on the carbocation stability of the migrating group. However, in the system studied by Bruice and Clerin the N-5 adduct is the more stable, predominating at equilibrium by a factor of 4-6. Furthermore, we have been unable to detect an initially formed N-5 adduct like 39 while monitoring the 3MLF photoreaction with 12 by $^1$H NMR methods. Thus, if 17 forms via the initial N-5 adduct 39, the interconversion in MeCN would have to be contrathermodynamic and very rapid.



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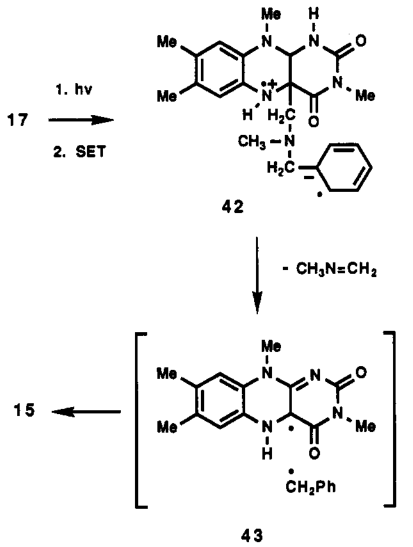
**Photoreaction of 3MLF with the $\alpha$-Silyl Amino Enone 20.** The results of studies of the photoreaction of the $\alpha$-silyl amino enone 20 with 3MLF provides further information about the detailed mechanism for the amine-flavin photo-SET processes. In particular, the observation that acetonylpiperidine 21 is the major, if not exclusive, product formed in the reaction of 20 with low concentrations of 3MLF serves as compelling evidence for the radical nature of amine-flavin photochemistry. This substance forms by the radical cyclization pathway outlined in Scheme VII. The minor amount of the 4a-benzyldihydroflavin 15, a secondary product derived from an initially formed adduct related to 17, reveals that radical coupling does occur in this system but that it occurs at a slower rate than radical cyclization,[26] which forms the $\alpha$-keto radical precursor of 21. The final step in the sequence yielding 21 could involve either H-atom abstraction or sequential electron-proton transfer from the 3MLF-H radical to the $\alpha$-keto radical. At high 3MLF concentrations, oxidation of the intermediate $\alpha$-amino radical 23 (Scheme VII) occurs to give the iminium cation 24, a precursor of the acetonylpyrrolidine 22. This occurs by SET from 23 ($E_{1/2}(+)$ = ca. -1 V)[27] to the easily reduced 3MLF ($E_{1/2}(-)$ = ca. -0.1 V).[5a] It should be noted that the ion radical pair formed in the redox process between radical 23 and 3MLF does not react to give a covalent adduct and, as a result, it suggests that in general adducts in this system arise by bond formation between radical rather than ionic intermediates. Furthermore, these observations indicate that the 3MLF-H radical does not oxidize $\alpha$-amino radical 23 at a rate comparable to its cyclization even though its reduction potential ($E_{1/2}(-)$ = ca. -0.12 V)[28] is sufficient to make this process modestly exothermic ($\Delta G_{SET}$ = ca. -0.9 eV).

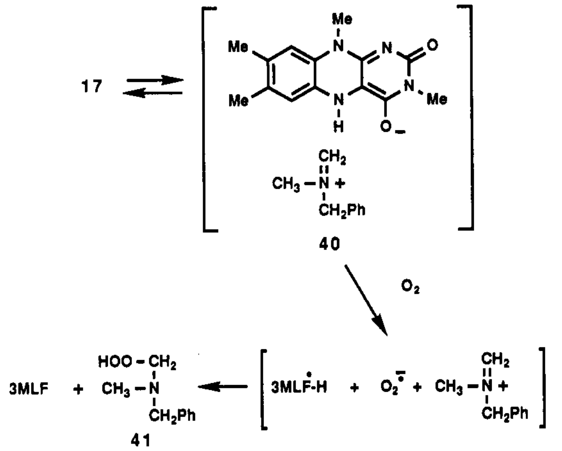
**Chemistry of the 4a-Adduct 17.** The accumulated observations made in this study demonstrate that the 4a-adduct 17 is a highly reactive substance when exposed to molecular oxygen, nucleophiles ($NaBH_4$, $H_2O$, MeOH), or light. The reactivity of 17 with MeOH and $H_2O$ to yield the respective amino ether 18 and the carbinolamine precursor of *N*-methyl-*N*-benzylamine is reminiscent of the behavior of the closely related (vinylogue) 4a-(indolylmethyl)dihydrolumiflavin studied earlier by Bruice.[25] Bruice has shown that this material is converted to (indolylmethyl)carbinol and dihydrolumiflavin (in addition to equilibrating with its N-5 analogue) via the corresponding indolylmethyl carbocation in aqueous media.

The related chemistry of 17 with nucleophiles and oxygen likewise can be understood in terms of the existence

**Scheme XV**



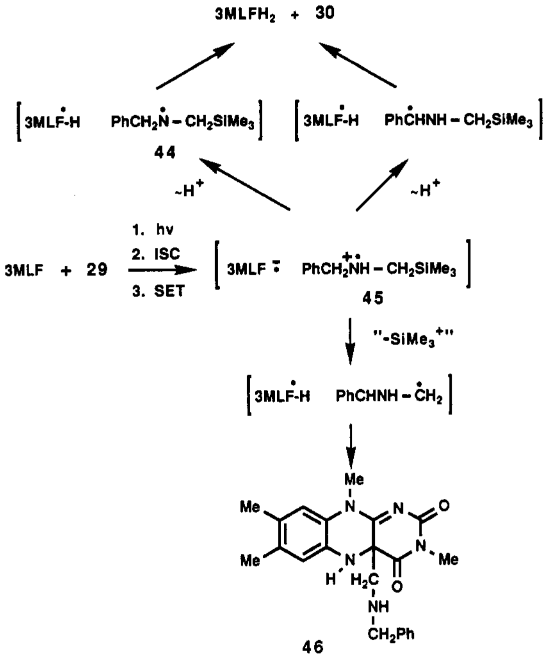
of a rapid equilibrium between the covalent 17 (favored in nonpolar media-like $CH_3CN$) and ionic structures **40**. The iminium cation fragment of **40** is the species captured by $NaBH_4$, MeOH, and $H_2O$ to give the tertiary amine, amino ether, and carbinolamine, respectively. Furthermore, formamide 14, produced by exposure of 17 to $O_2$, most likely arises via the ion pair **40** by SET from the hydroflavin anion to oxygen followed by addition of the resulting superoxide anion to the iminium cation and subsequent dehydration of the hydroperoxide 41.



The most significant feature of these observations concerns the unusual instability of adduct 17. The data accumulated in this study has allowed identification of 17 as the major primary adduct formed in photoreaction of 3MLF with amines 12 and 13. Thus is seems unlikely that the photoreactions conducted in $H_2O$ or MeOH involve direct formation of an iminium cation by a sequential two electron transfer route or that formamide formation from irradiations conducted on nondegassed solutions involves $O_2$ trapping of an initially generated $\alpha$-amino radical. Instead, the results suggest that the adduct 17 forms in a primary photochemical event and that reactions of this intermediate with nucleophiles such as MeOH and HOH or with $O_2$ are rapid secondary processes.

The photochemical transformation of 17 and related dihydroflavin adducts to the 4a-benzylflavin 15 is worthy of comment. This modestly clean and unanticipated photoreaction is difficult to fully explain. The deuterium labeling results clearly show that the benzyl group in 15 is derived by intact transfer (no C–H bond cleavages) of the benzyl group in 17. The most likely mechanism for the 17 to 15 phototransformation involves initial SET from the excited dihydroflavin chromophore to the arene ring of the benzyl grouping (Scheme XV). This intramolecular redox step appears reasonable in light of the exceptionally high singlet excited state oxidation potential of dihydroflavins ($E_{1/2}(+)$ = ca. −2.6 V).[29] Heterolytic fragmentation of the resulting zwitterionic diradical 42 liberates the undetected *N*-methylformaldimine and provides the dihydroflavin–benzylic radical pair 43. Radical coupling would then give the 4a-benzyl adduct 15.

**Scheme XVI**



**Photoreactions of 3MLF with Secondary and Primary Amines.** The results of studies of 3MLF photoreactions with the secondary silyl amine **29** suggest that two competing pathways are followed. Production of the 4a-benzylflavin **15** and formamide **31** in this system is indicative of the operation of a route involving desilylation of the intermediate amine cation radical **45** with ensuing formation of the primary adduct **46** (Scheme XVI). Respective photoreaction and oxidation of the initially formed adduct **46** yield the observed products **15** and **31**. Deprotonation of the amine cation radical **45** ether from nitrogen or the $\alpha$-benzylic position is competitive with desilylation as suggested by the formation of benzaldimine **30**. It is difficult to distinguish unambiguously between these two proton transfer pathways since the high regioselectivity (benzaldimine rather than formaldimine) observed for imine formation can be easily explained using both processes. Accordingly, the benzyl $\alpha$-CH bonds are known to be kinetically more acidic than the TMS-substituted $\alpha$-CH bonds,[22] and H-atom abstraction on the aminyl radical **44** arising by N-H deprotonation should strongly favor the benzylic center.

However, if $\alpha$-CH deprotonation is involved in aldimine **30** formation, it would be difficult to understand why a process of this type is not competitive with desilylation in the closely related tertiary amine cation radical arising from silyl amine **12**. On this basis, we propose that in SET reactions of 3MLF with primary and secondary amines, NH deprotonation of intermediate cation radicals is the major pathway used for aldimine formation.

Reaction of the non-TMS secondary amine **32** with 3MLF results in formation of only minor quantities of an adduct which serves as the precursor for the 4a-benzylflavin 15. In this case, fast desilylation of the intermediate cation radical is not an option and, as a result, NH deprotonation leading to aldimine 33 becomes the dominant pathway. The primary amine, benzylamine, reacts with 3MLF in a similar fashion generating the imine precursor of 35. In this instance the initially formed imine, $PhCH{=}NH_2$, is highly unstable and reacts with benzylamine to form *N*-benzylbenzaldimine 35 as a stable isolated product.

**Potential Relationships between Amine–Flavin Photochemistry and MAO-Catalyzed Oxidative Dealkylation of Amines.** Several features of the photochemical studies described above are potentially relevant to the oxidative dealkylations of amines catalyzed by the monoamine oxidases. The current working hypothesis for the mechanism of the enzymatic processes involves an initial SET step in which a bound amine serves as the electron donor and the MAO–flavin residue plays the role of electron acceptor. It should be emphasized that this SET process would be highly endergonic (ca. 40 kcal/mol) in the absence of participation by catalytic groupings within the enzyme in altering the donor and acceptor redox potentials. In contrast, the model flavin–amine photochemical reactions are promoted by thermodynamically favorable SET. Consequently, the photochemical models are capable of providing potentially useful information about the chemistry of key intermediates in the MAO-catalyzed reactions only in the event that SET pathways truly intervene in the enzymatic processes. Important differences could exist between the photochemical and enzymatic reactions even if both are promoted by SET. For example, the ground-state flavin is capable of interacting with transient radical intermediates formed in the photochemical reactions while in the MAO process this would be highy unlikely owing to localization of each reaction cycle in the protein active site. Likewise, the enzyme could influence the redox potentials (e.g. the reduction potential of the flavin anion radical or of its protonated form) and reactivity (e.g. deprotonation rates and regioselectivities) of reactive intermediates. Even with these provisos, pertinent information about the characteristics of a possible MAO SET mechanism can still come from studies of bonafide SET processes of closely related model systems.

The first potentially relevant results obtained in our photochemical investigations concerns the nature of the step immediately following the SET event in reactions of primary and secondary amines. The cation radicals derived from these substrates appear to show a preference for N-H rather than $\alpha$ C-H deprotonation by the paired flavin anion radical or another base (e.g. the amine in the excited state process or a basic protein residue in the enzymatic chemistry). This proposal is suggested by a comparison of the outcomes of reactions of the tertiary and secondary $\alpha$-silyl amines 12 and 13 and the precedence found in observations made by Lewis[23] and Otsugi[24] in their studies of primary and secondary amine SET photochemistry. Thus, if MAO-catalyzed oxidative dealkylation reactions of primary and secondary amines are indeed promoted by SET, one cannot dismiss the operation of a route involving loss of an N-H proton in the derived cation radical followed by H-atom transfer from the resulting aminyl radical. As far as we know, this option has not been considered previously.

A second point relates to the possible intermediacy of covalent amine–flavin adducts in the MAO-catalyzed reactions of tertiary amines. The photochemical studies have conclusively demonstrated that $\alpha$-amino radicals formed in reactions of 3MLF with the tertiary amines 12 and 13 by respective cation radical desilylation and $\alpha$ C-H deprotonation efficiently couple to the flavin radical anion (or its protonated form) to generate covalent adducts. Analogous transients in a putative MAO–amine SET pathway could show a similar propensity for covalent bond formation. The resulting enzyme substrate adducts could be competent intermediates in the oxidative dealkylation process since, as the efforts described above have demonstrated, they should react rapidly with water even in the absence of enzyme catalysis to give the carbinolamine precursors of the dealkylated amine products along with the reduced-flavin form of the enzyme.

We repeat that any parallel drawn between the mechanistic characteristics of the amine–flavin photochemical and MAO catalytic or inhibition processes must be considered speculative at this point since the evidence supporting the operation of SET pathways for the enzyme chemistry is not yet compelling. In spite of this, the photochemical efforts have suggested alternative views of the MAO catalytic mechanism and have provided insight into the design of substrates and/or inhibitors (e.g. the silyl aminoe enone 20) that might be useful in probing for the radical nature of MAO biochemistry.

## Experimental Section

**General.** $^1H$ NMR (200 or 400 MHz) and $^{13}C$ NMR (50 MHz) were recorded on $CDCl_3$ solutions unless otherwise noted. IR spectra were recorded on $CHCl_3$ solutions. Column chromatography was performed with either Merck-EM type 60 (230–400 mesh) or Alcoa type F-20 alumina (neutral, 80–200 mesh) absorbants. Preparative TLC was performed on 20 × 20 cm plates coated with Merck-EM type 60 GH-254 silica gel. Gas chromatographic analyses and separations were conducted on a Varian-940 chromatograph with flame ionization detection and a 10% SE-30 on chromosorb, 8 ft × $^1/_8$ in. column. All reactions were run under a dry $N_2$ atmosphere unless otherwise specified. All new compounds isolated in the course of this study were characterized by spectroscopic methods and were shown to be greater than 90% pure by $^{13}C$ and $^1H$ NMR analysis. The exception this is adduct 28 whose slow decomposition to form 3MLF leads to an ca. 85% purity.

Preparative photochemical reactions were run in an apparatus consisting of a 450-W Hanovia medium-pressure, mercury lamp (ACE) surrounded by a Uranium glass filter ($\lambda > 320$ nm) in a water-cooled quartz immersion well surrounded by Pyrex tubes or a well containing the solution being irradiated. The photolysis solutions were purged with Ar or deoxygenated $N_2$ both before and during irradiations. The progress of each preparative photochemical reaction was monitored by UV absorption spectrometry. The solvents used in the photoreactions were spectrograde $CH_3CN$ (Baker) or $CH_3OH$ (Baker) unless otherwise specified. Photoreactions in sealed NMR tubes were conducted with this same apparatus.

***N*-Methyl-*N*-benzyl-*N*-[(trimethylsilyl)methyl]amine (12).** This substance was prepared by the general method of Noll.[15a] Although this compound has been reported before,[15b] the details of preparation and characterization of this substance have not been described. A solution of 2.00 g (16.5 mmol) of *N*-methyl-*N*-benzylamine (Aldrich) in 20 mL of MeCN containing 4.20 g (19.6 mmol) of $Me_3SiCH_2I$ (Aldrich) was stirred at reflux for 15 h, cooled to 25 °C, and concentrated in vacuo. The residue was diluted with saturated $NaHCO_3$ and extracted with $CHCl_3$. The $CHCl_3$ extracts were dried ($Na_2SO_4$) and concentrated in vacuo, giving a residue which was subjected to silica gel column chromatography ($CHCl_3$ to 2.5% $MeOH-CHCl_3$) to yield 3.00 g (88%) of the tertiary silyl amine 12: $^1H$ NMR 0.04 (s, 9 H, $SiCH_3$), 1.89 (s, 2 H, $SiCH_2$), 2.17 (s, 3 H, $NCH_3$), 3.42 (s, 2 H, benzylic), 7.30 (m, 5 H, aromatic); $^{13}C$ NMR −1.33 ($SiCH_3$), 46.1 ($SiCH_2$), 49.9 (N-$CH_3$), 66.4 (benzylic), 126.8, 128.1, 128.9 and 140.0 (aromatic); IR 2960, 2780, 1455, 1365, 1250, 855, 740; mass spectrum, $m/e$ (relative intensity) 207 ($M^+$, 3), 194 (3), 192 (5), 134 (49), 116 (18), 91 (100), 73 (10), 65 (10); high-resolution mass spectrum, *m/e* 207.1477 ($C_{12}H_{21}NSi$ requires 207.1443).

**Preparative Irradiation of 3-Methyllumiflavin and the Silyl Amine 12 in MeCN.** A prepurged (deoxygenated $N_2$) solution of 90 mg (0.33 mmol) of 3-methyllumiflavin and 69 mg (0.33 mg) of the silyl amine **12** in 165 mL of MeCN was irradiated in a preparative apparatus for 2 h under a $N_2$ atmosphere. The irradiation was terminated when ca. 75% of 3-methyllumiflavin has been consumed (by UV monitoring) and the photolysate was exposed to air and concentrated in vacuo. The residue was subjected to preparative TLC (ether) to give 22 mg (18%) of 4a-benzyl-4a,5-dihydro-3-methyllumiflavin (**15**), 28 mg (57%) of *N*-benzyl-*N*-methylformamide (**14**), a trace quantity of 4a-hydroxy-5-benzyl-4a,5-dihydro-3-methyllumiflavin (**16**), and 66 mg (73%) recovered 3-methyllumiflavin.

Spectroscopic data for **14** matched those reported by Freudenreich[16] and independently synthesized material: $^1$H NMR 2.79 and 2.85 (s, 3 H, N-$CH_3$), 4.40 and 4.53 (s 2 H, benzylic), 7.26 (m, 5 H, aromatic), 8.16 and 8.29 (s, 1 H, aldehyde).

Spectroscopic data for **15** matched those reported by Hemmerich[17] and independently synthesized material (see below): $^1$H NMR 2.25 (s, 3 H, C-8), 227 (s, 3 H, C-7), 2.93 and 3.13 (AB quartet, $J$ = 13.0 Hz, 2 H, benzylic), 3.10 (s, 3 H, N-3), 3.65 (s, 3 H, N-10), 4.65 (s, 1 H, N-H), 6.65 (s, 1 H, C-9), 6.89 (s, 1 H, C-6), 6.80 and 7.20 (m, 5 H, aromatic); $^{13}$C NMR 19.4 (C-7 $CH_3$ and C-8 $CH_3$), 27.6 (N-10 $CH_3$), 32.0 (N-3 $CH_3$), 43.4 ($PhCH_2$), 58.9 (C-4a), 117.0 (C-6), 117.4 (C-9), 129.5 (C-7, C-8 and aromatic Ph), 132.2 (C9a), 134.5 (C-5a), 155.4 (C-10a), 161.5 (C-2), 168.9 (C-4); IR 1720, 1670, 1625, 1570, 1405; mass spectrum *m/e* (relative intensity) 362 ($M^+$, 11), 271 (100), 214 (12), 186 (4), 171 (3), 134 (5), 120 (3), 91 (19), 65 (5); high-resolution mass spectrum *m/e* 362.1743 ($C_{21}H_{22}O_2N_4$ requires 362.1743).

Spectroscopic data for **16** matched those reported by Hemmerich[17] and independently synthesized material: $^1$H NMR 2.15 (s, 3 H, C-7 $CH_3$), 2.20 (s, 3 H, C-8 $CH_3$), 3.22 (s, 3 H, N-3 $CH_3$), 3.50 (s, 3 H, N-10 $CH_3$), 4.65 (s, 2 H, benzylic), 7.00 (s, 1 H, H-9), 7.05 (s, 1 H, H-6), 6.75 and 7.20 (m, 5 H, aromatic).

**Preparative Irradiation of 3-Methyllumiflavin and the Silyl Amine 12 in $D_2O$–$CH_3CN$ and $H_2O$–$CD_3CN$.** A prepurged (Ar) solution of 16 mg (5.7 × $10^{-2}$ mmol) of 3-methyllumiflavin and 19 mg (9.2 × $10^{-2}$ mmol) of the silyl amine **12**, in 2 mL of $D_2O$ and 9 mL of $CH_3CN$ was irradiated for 55 min under an Ar atmosphere. The photolysate was concentrated in vacuo to give a residue that was subjected to preparative TLC ($Et_2O$) to yield 1 mg (6%) of the 4a-benzylflavin **15**, which was shown by $^1$H NMR to contain no deuterium incorporation at the benzylic carbon.

A similar irradiation in 2 mL of $H_2O$ and 9 mL of $CD_3CN$ gave after workup and separation the 4a-benzylflavin **15** with no deuterium incorporation at the benzylic carbon.

**Synthesis of *N*-Methyl-*N*-(benzyl-$\alpha,\alpha$-$d_2$)-*N*-(silylmethyl)amine (12-$d_2$).** To a suspension of 1.40 g (34 mmol) of $LiAlD_4$ (Aldrich) in 40 mL of anhydrous ether at 0 °C was added a solution of 3.08 g (29 mmol) of benzonitrile (Aldrich) in 30 mL of dry ether. After addition was complete (20 min), the solution was stirred at 25 °C for 4.5 h. Excess hydride was quenched by addition of a 20% NaOH solution. The mixture was filtered, and the filtrate was washed with saturated NaCl and extracted with $Et_2O$. The ethereal extracts were dried ($Na_2SO_4$) and concentrated in vacuo to give 2.80 g (89%) of benzylamine-$d_2$, which was used without further purification: $^1$H NMR 1.44 (s, 2 H, $NH_2$), 7.29 (m, 5 H, aromatic); $^{13}$C NMR 126.4, 126.7, 128.1, and 143.0 (aromatic).

To a solution of 2.4 g (22 mmol) of benzylamine-$d_2$ in 50 mL of MeCN containing 15.00 g (109 mmol) of $K_2CO_3$ was added 3.75 g (34.5 mmol) of ethyl chloroformate (Aldrich). The mixture was stirred at 25 °C for 14 h and filtered. The filtrate was concentrated in vacuo, giving a residue which was crystallized (*n*-hexane) to give 2.34 g (60%) pure ethyl *N*-(benzyl-$\alpha,\alpha$-$d_2$)carbamate (mp 41–42 °C): $^1$H NMR 1.25 (t, $J$ = 7.1 Hz, 3 H, $CH_3$), 4.14 (q, $J$ = 7.1 Hz, 2 H, methylene), 4.97 (br s, 1 H, NH), 7.34 (m, 5 H, aromatic); $^{13}$C NMR 14.6 ($CH_3$), 60.9 ($CH_2$), 127.4, 127.5, 128.6, and 138.5 (aromatic), 156.6 (carbonyl).

To a suspension of 1.47 g (38.7 mmol) of $LiAlH_4$ in 30 mL of anhydrous ether at 0 °C was slowly added a solution of 2.34 g (12.9 mmol) of the above carbamate in 25 mL of $Et_2O$. After addition was complete, the mixture was stirred at reflux for 20 h, cooled to 0 °C, and quenched with 20 mL of wet $Et_2O$ and 18 mL of 20% NaOH. The mixture was filtered, and the filtrate was extracted with saturated NaCl solution. The ethereal layer was dried ($Na_2SO_4$) and concentrated in vacuo to give 1.60 g (98% of *N*-methyl-*N*-(benzyl-$\alpha,\alpha$-$d_2$)amine), which was used without purification: $^1$H NMR 1.41 (s, 1 H, NH), 2.35 (s, 3 H, $CH_3$), 7.24 (m, 5 H, aromatic); $^{13}$C NMR 35.3 (N-$CH_3$), 126.3, 127.6, 127.7, and 139.6 (aromatic).

A solution of 1.27 g (10.0 mmol) of the above secondary amine and 1.97 g (16.0 mmol) of $Me_3SiCH_2Cl$ (Aldrich) in 50 mL of $CH_3CN$ was stirred at reflux for 67 h, cooled to 25 °C, and concentrated in vacuo. The residue was subjected to chromatography on silica gel ($CHCl_3$ to 2.5% $MeOH/CHCl_3$) to give 0.70 g (45%) of the tertiary amine **12**-$d_2$: $^1$H NMR 0.00 (s, 9 H, $SiCH_3$), 1.85 (s, 2 H, $SiCH_2$), 2.12 (s, 3 H, $NCH_3$), 7.25 (m, 5 H, aroamtic); $^{13}$C NMR −1.3 ($SiCH_3$), 46.1 ($SiCH_3$), 49.7 (N-$CH_3$), 126.8, 128.1, 128.9, and 139.6 (aromatic).

**Irradiation of 3-Methyllumiflavin and *N*-Methyl-*N*-(benzyl-$\alpha,\alpha$-$d_2$)-*N*-(silylmethyl)amine (12-$d_2$) in MeCN.** An Ar-purged solution of 52 mg (0.2 mmol) of 3-methyllumiflavin and 63 mg (0.3 mmol) of the deuterated silyl amine **12-d$_2$** in 180 mL of MeCN was irradiated for 60 min. Concentration of the photolysate followed by preparative TLC (ether) gave 6 mg (8%) of 4a-(benzyl-$\alpha,\alpha$-$d_2$)-4a,5-dihydro-3-methyllumiflavin (**15-d$_2$**), 13 mg (33%) of *N*-methyl-*N*-(benzyl-$\alpha,\alpha$-$d_2$)formamide (**14-d$_2$**), a trace quantity of 4a-hydroxy-5-(benzyl-$\alpha,\alpha$-$d_2$)-4a,5-dihydro-3-methyllumiflavin (**16-d$_2$**), and 30 mg (58%) recovered 3-methyllumiflavin. Spectroscopic data for **14-d$_2$**, **15-d$_2$**, and **16-d$_2$** matched for those of their photio analogues except for obvious differences associated with the presence of deuterium at the benzylic positions.

**Irradiation of 3-Methyllumiflavin and *N*,*N*-Dimethyl-*N*-benzylamine (13) in MeCN.** An Ar-purged solution containing 42 mg (1.5 × $10^{-1}$ mmol) of 3-methyllumiflavin and 31 mg (2.3 × $10^{-1}$ mmol) of *N*,*N*-dimethyl-*N*-benzylamine **13** in 180 mL of $CH_3CN$ was irradiated for 65 min. Concentration in vacuo followed by preparative TLC on the residue (silica gel, ether) gave 3 mg (5%) of the 4a-benzylflavin **15**, 4 mg (12%) of *N*-benzyl-*N*-methylformamide (**14**),[16] and trace quantities of the 4-hydroxy-5-benzylflavin **16**.

**Irradiation of 3MLF and the Silyl Amino Enone 20.** An $N_2$ prepurged solution of 14 mg (5.2 × $10^{-2}$ mmol) 3MLF and 25 mg (8.3 × $10^{-2}$ mmol) of the silyl amine enone **20**[18] in 17 mL of MeCN was irradiated for 1 h. The photolysate was exposed to air and concentrated in vacuo, giving a residue which was analyzed by $^1$H NMR. This showed that the mixture contained 1-benzyl-3-acetonylpiperidine (**21**)[12c,18] (40%), 1-benzyl-2-acetonylpyrrolidine (**22**) (40%), and a trace quantity of the 4a-benzylidhydroflavone **15**.

Spectroscopic data for **22**: $^1$H NMR 1.44 (m, 1 H, H-3), 1.66 (m, 3 H, H-3, H-4), 2.10 (m, 1 H, H-5), 2.13 (s, 3 H, $CH_3$), 2.46 (dd, $J$ = 16.2, 8.2 Hz, 1 H, $CH_2CO$), 2.75 (dd, $J$ = 16.2, 3.9 Hz, 1 H, $CH_2CO$), 2.86 (m, 2H, H-2, H-5), 3.25 (AB q, $J$ = 13.0 Hz, 1 H, $PhCH_2$), 3.89 (AB q, $J$ = 13.0 Hz, 1 H, $PhCH_2$), 7.24 (m, 5 H, aromatic); $^{13}$C NMR 22.3 (C-4), 30.9 $HCH_3CO$), 31.0 (C-3), 49.0 ($CH_2CO$), 53.9 (C-5), 58.8 ($PhCH_2$), 60.1 (C-2), 126.9, 128.2, 128.8, 139.5 (aromatic); IR 1710 cm$^{-1}$, mass spectrum, *m/e* (relative intensity) 217 ($M^+$, 0.4), 160 (17), 159 (31), 158 (10), 92 (10), 91 (100), 82 (6), 68 (7), 65 (9); high-resolution mass spectrum, *m/e* 217.1455 ($C_{14}H_{19}NO$ requires 217.1467).

The following procedure was used for irradiations of silyl amino enone solutions containing varying concentrations of 3MLF. $N_2$ prepurged solutions of 13 mg (4.3 × $10^{-2}$ mmol) of the silyl amino enone **20** containing 59 mg (2.2 × $10^{-1}$ mmol), 12 mg (4.4 × $10^{-2}$ mmol), and 2 mg (8.5 × $10^{-3}$ mmol) of 3MLF in 90 mL of MeCN were irradiated for 45, 80, and 80 min, respectively. The photolysates were concentrated in vacuo, giving residues that were subjected to GLC analysis (170 °C, 6 ft $^1/_8$ in. 3% OV101). The following pyrrolidine **22**:piperidine **21** product ratios were recorded: 1.4 (2.40 mM 3MLF), 0.3 (0.49 mM 3 MLF), and 0.1 (0.01 mM 3MLF).

**Synthesis of Benzyl *N*-Benzyl-*N*-[(trimethylsilyl)methyl]carbamate (25).** A mixture of 1.60 g (8.3 mmol) of *N*-benzyl-*N*-[(trimethylsilyl)methyl]amine,[31] 5.70 g (41 mmol) of $K_2CO_3$, and 1.67 (9.8 mmol) of benzyl chloroformate (Aldrich) in 50 mL of MeCN was stirred at 25 °C for 12 h, cooled to 25 °C, and filtered. The filtrate was concentrated in vacuo, giving a residue that dissolved in ether. The ethereal solution was washed with $H_2O$, dried ($Na_2SO_4$), and concentrated in vacuo to give a residue that was subjected to column chromatography on silica gel (hexane to 20% $Et_2O$–hexane), yielding 1.54 g (57%) of the carbamate **25**: $^1H$ NMR 0.00 (d, $J$ = 17.5 Hz, 9 H, $SiCH_3$), 2.73 (d, $J$ = 9.1 Hz, 2 H, $SiCH_2$), 4.48 (s, 2 H, $CH_2N$), 5.16 (s, 2 H, $OCH_2$), 7.28 (m, 10 H, aroamtic); $^{13}C$ NMR −1.55 ($SiCH_3$), 37.3 and 38.6 ($SiCH_3$, isomers), 52.5 (benzylic) 67.2 ($OCH_2$), 127.3, 127.9, 128.2, 128.4, 128.5, 136.9 and 137.6 (aromatic), 156.0 and 157.0 (carbonyl, isomers); IR 3064, 3031, 2952, 1697, 1496, 1455, 1364, 1248, 1228, 1100, 855; mass spectrum (chemical ionization), *m/e* (relative intensity) 328 ($M^+ + 1$, 0.5), 312 (27), 236 (81), 192 (100), 176 (3), 100 (11), 91 (99), 73 (99), 65 (23); high-resolution mass spectrum, *m/e* 328.1730 ($M^+ + 1$, $C_{19}H_{26}O_2NSi$ requires 328.1733).

**Irradiation of 3-Methyllumiflavin with Benzyl *N*-Benzyl-*N*-[(trimethylsilyl)methyl]carbamate (25) in MeCN.** An $N_2$ (deoxygenated) purged solution of 90 mg (0.33 mmol) of 3-methyllumiflavin and 130 mg (0.40 mmol) of the silyl carbamate **25** in 165 mL of MeCN was irradiated for 20 h. Concentration of the photolysate in vacuo gave a residue, which was subjected to preparative TLC on silica gel (20% ether–pentane), yielding 33 mg (31%) of benzyl *N*-benzyl-*N*-formylcarbamate **26**, trace quantities of the bis carbamate **27**, and 87 mg (97%) of recovered 3-methyllumiflavin.

Spectroscopic data for **26**: $^1H$ NMR 4.80 (s 2 H, $CH_2N$), 5.20 (s, 2 H, $CH_2O$), 7.25 (m, 10 H, aromatic), 9.32 (s, 1 H, aldehyde); $^{13}C$ NMR 44.1 ($CH_2N$), 69.0 ($CH_2O$), 127.7, 128.3, 128.4, 128.5, 128.7, 128.8, 134.6, and 136.6 (aromatic), 154.0 (COO), 162.6 (COH); IR 1750, 1700, 1410, 1355–1320, 1220–1180; mass spectrum (chemical ionization), *m/e* (relative intensity) 270 ($M^+ + 1$, 5), 226 (4), 180 (14), 134 (43), 106 (25), 91 (100), 84 (5), 79 (14), 73 (4), 65 (5); high-resolution mass spectrum, *m/e* 270.1130 ($M^+ + 1$, $C_{16}H_{16}O_3N$ requires 270.1134).

Spectroscopic data for **27**: $^1H$ NMR 4.59 (s, 4 H, $NCH_2$), 4.81 (s, 4 H, benzylic), 5.18 (s, 4 H, $OCH_2$), 7.25 (m, 20 H, aromatic); $^{13}C$ NMR 50.3 ($NCH_2$), 67.6 ($NCH_2Ph$), 71.5, 73.0 ($OCH_2Ph$), 127.5, 127.9, 128.6, 136.0, 137.6 (aromatic), 156.0 ($CO_2$); IR 1700, 3400 $cm^{-1}$; mass spectrum, *m/e* (relative intensity) 300 (11), 289 (15), 288 (66), 255 (44), 254 (99), 210 (99), 181 (100), 167 (38), 150 (99), 136 (99), 118 (99); high-resolution mass spectrum, *m/e* 254.1169 ($M^+/2$, $C_{16}H_{16}O_2N$ requires 254.1181).

**Irradiation of 3-Methyllumiflavin with Benzyl *N*-Benzyl-*N*-[(trimethylsilyl)methyl]carbamate (25) in MeOH.** A $N_2$ (deoxygenated) purged solution of 90 mg (0.33 mmol) of 3-methyllumiflavin and 130 mg (0.40 mmol) of the silyl carbamate **25** in 165 mL of MeOH was irradiated for 7.5 h. Irradiation was terminated when 78% of 3-methyllumiflavin had been consumed (by UV monitoring) and the photalysate was concentrated in vacuo to give a residue, which was subjected to the same workup and separation procedures used for the MeCN reaction (see above). This yielded 25 mg (12%) of 4a-hydroxy-5-[(*N*-benzyl-*N*-[(benzyloxy)carbonyl]amino]methyl]-4a,5-dihydro-3-methyllumiflavin (**28**), trace quantities of benzyl *N*-benzyl*N*-formylcarbamate (**26**) and the bis-carbamate (**27**), and 75 mg (83%) of recovered 3-methyllumiflavin. Compound **28** is unstable and decomposes to give 3MLF. Thus all attempts to obtain a pure (>90%) sample of this substance were unsuccessful.

Spectroscopic data for **28** (obtained on a sample contaminated with ca. 5–10% of 3MLF and as amixture of carbamate rotamers): $^1H$ NMR 2.20 (s, 3 H, C-8 $CH_3$), 2.28 (s, 3 H, C-7 $CH_3$), 3.26 (s, 3 H, N-3 $CH_y$), 3.62 (s, 3 H, N-10 $CH_3$), 4.22 (broad d, 1 H, OH), 4.70–5.30 (broad m, 4 H, benzylic), 6.97 (s 2 H, N-5 $CH_2$), 6.79 and 7.23 (m, 12 H, C-6, C-9, and aromatic); $^{13}C$ NMR 19.4 (C-7 $CH_3$), 19.7 (C-8 $CH_3$), 28.2 (N-3 $CH_3$), 32.5 (N-10 $CH_3$), 49.8 (benzylic), 61.2 ($OCH_2$), 67.9 (N-5 $CH_2$), 73.4 (4a), 117.5 (C-6), 122.8 (C-9), 128.2 (m), 130.2 and 132.7 (aromatic), 133.1 (C-7), 134.1 (C-8), 135.9 (5a), 136.3 (9a), 155.1 (C-10a), 156.1 (COO), 158.1 (C-2), 166.9 (C-4); IR 3338 (broad), 2946, 1700, 1670, 1570, 1452; mass spectrum, *m/e* (relative intensity) 542 (9), 541 (19), 288 (19), 287 (23), 271 (12), 270 (38), 254 (5), 246 (100), 231 (12), 210 (29), 181 (18), 150 (37), 118 (96), high-resolution mass spectrum, *m/e* 541.2325 ($C_{30}H_{31}N_5O_5$ requires 541.2328).

**Irradiation of 3-Methyllumiflavin with *N*-Benzyl-*N*-[(trimethylsilyl)methyl]amine (29) in MeCN.** An $N_2$ (deoxygenated) prepurged solution of 42 mg ($1.6 \times 10^{-1}$ mmol) of 3-methyllumiflavin and 45 mg ($2.3 \times 10^{-1}$ mmol) of the known[31] secondary silyl amine **29** in 350 mL of MeCN was irradiated for 45 min (33% conversion of 3-methyllumiflavin by UV monitoring). Concentration of the phospalysate in vacuo followed by preparative TLC (ether) gave 1 mg (2%) of 4a-benzyl-4a,5-dihydro-3-methyllumiflavin (**15**), 3 mg (10%) of *N*-benzylformamide (**31**), and 21 mg (50%) recovered 3-methyllumiflavin. Spectroscopic data for *N*-benzylformamide (**31**) matched those reported by Freudenreich[16] and independently synthesized material. $^1H$ NMR data for **31**: 4.44 and 4.47 (s, 2 H, benzylic), 5.93 (broad s, 1 H, NH), 7.30 (m, 5 H, aromatic), 8.23 (s, 1 H, CH=O).

**Irradiation of 3-Methyllumiflavin with *N*-Benzylamine (34) in MeCN.** A $N_2$ (deoxygenated) purged solution of 60 mg (0.22 mmol) of 3-methyllumiflavin and 24 mg (0.22 mmol) of benzylamine (**34**) (Aldrich) in 165 mL of MeCN was irradiated for 2.5 h. The photolysate was concentrated in vacuo to give a residue, which was subjected to preparative TLC (ether) to yield 17 mg (40%) of *N*-benzylidenebenzylamine (**35**) (commercially available from Aldrich). $^1H$ NMR data for **35**: 4.80 (d, $J$ = 1.2 Hz, 2 H, benzylic), 7.34, 7.41, and 7.76 (m, 10 H, aromatic), 8.38 (t, $J$ = 1.2 Hz, 1 H, benzylidene).

**NMR-Tube Irradiations of 3-Methyllumiflavin and Amine Substrates.** The NMR tube samples for irradiation were prepared by the following procedure. A solution (0.4 mL) containing 3-methyllumiflavin and the amine substrate in $CD_3CN$ or $CD_3OD$ in a thick-walled NMR tube (Wilmad: no. 524-PP) was subjected to repeated freeze–thaw cycles before the tubes were sealed under vacuum. The solutions were then irradiated by using Uranium glass filtered light (λ > 320 nm). A 400-MHz spectrometer was used to record the $^1H$ NMR spectra. The results are given below for 3-MLF, amine, solvent, and irradiation time.

3-Methyllumiflavin ($3.3 \times 10^{-3}$ M), silyl amine **12** ($3.9 \times 10^{-3}$ M), $CD_3CN$, 10 min. $^1H$ NMR monitoring indicated that both 3-methyllumiflavin and **12** were consumed and that 4a-(*N*-benzyl-*N*-methylamino)methyl-4a,5-dihydro-3-methyllumiflain (**17**) was produced as a major product. This product was unstable in the presence of oxygen, and it decomposed to yield 3-methyllumiflavin and *N*-benzyl-*N*-methylformamide (**14**) as soon as the NMR tube was opened to air. $^1H$ NMR data for **17**: 1.98 (s, 3 H, side chain N-$CH_3$), 2.16 (s, 3 H, C-8 $CH_3$), 2.18 (s, 3 H, C-7 $CH_3$), 3.16 (s, 3 H, N-3 $CH_3$), 3.35 and 3.37 (AB quartet, $J$ = 6.7 Hz, 2 H, benzylic), 3.43 (s, 3 H N-10 $CH_3$), 5.06 (s, 1 H, N-5 H), 6.67 (s, 1 H, C-6), 6.86 (s, 1 H, C-9), 7.13 and 7.30 (m, 5 H, aromatic). Addition of $H_2O$ and $CD_3OD$ to the photolysate containing the adduct **17** gave after workup 24% of *N*-benzylmethylamine and 43% of *N*-benzyl-*N*-methyl-*N*-(methoxy-$d_3$-methyl)amine (**18**). The $^1H$ NMR spectrum of **18** matched that of independently prepared material: $^1H$ NMR ($CD_3OD$) 2.36 (s, 3 H, N-$CH_3$), 3.71 (s, 2 H, benzylic), 4.04 (s, 2 H, ($NCH_2O$), 7.28 (m, 5 H, aromatic).

3-Methyllumiflavin ($4.1 \times 10^{-3}$ M), silyl amine **12** ($3.9 \times 10^{-3}$ M), 1.5% $H_2O$–$CD_3CN$, 10 min. Under these conditions, 3-methyllumiflavin was reduced to 1,5-dihydro-3-methyllumiflavin (**19**), formed as a precipitate on the bottom of the tube. $^1H$ NMR analysis showed that 69% of the silyl amine **12** was consumed to give 25% of *N*-benzylmethylamine.

3-Methylumiflavin ($4.6 \times 10^{-3}$ M), silyl amine **12** ($4.8 \times 10^{-3}$ M), $CD_3OD$, 10 min. $^1H$ NMR analysis indicated that both 3-methyllumiflavin and the silyl amine **12** disappeared with simultaneous formation of *N*-benzyl-*N*-methyl-*N*-(methoxy-$d_3$-methyl)amine (**18**) as the exclusive product (100%).

3-Methyllumiflavin ($3.0 \times 10^{-3}$ M), silyl amine **12-d₂**, $CD_3CN$, 10 min. $^1H$ NMR analysis showed that 3-methyllumiflavin disappeared to give 4a-[[*N*-(benzyl-α,α-$d_2$)-*N*-methylamino]-methyl]-4a,5-dihydro-3-methyllumiflavin (**17-d₂**) as the sole product.

3-Methyllumiflavin ($3.0 \times 10^{-3}$ M), *N*,*N*-dimethyl-*N*-benzylamine (**13**) ($6.0 \times 10^{-3}$ M), $CD_3CN$, 10 min. $^1H$ NMR analysis showed that 3-methyllumiflavin disappeared, giving the same adduct **17** as that resulting from the irradiation of 3-methyllumiflavin with the silyl amine **12** in $CD_3CN$.

3-Methyllumiflavin ($2.2 \times 10^{-3}$ M), silyl carbamate **25**, $CD_3CN$, 5 h. $^1H$ NMR analysis indicated that 26% of the silyl carbamate **25** had disappeared but no evidence for adduct formation was detected. A similar NMR-tube irradiation on a nondegassed solution of 3-methyllumiflavin and **25** resulted in 34% conversion of the silyl carbamate **25**, givine 26% of benzyl *N*-benzyl-*N*-formylcarbamate (**26**).

3-Methyllumiflavin (3.1 × $10^{-3}$ M), silyl amine **29** (3.1 × $10^{-3}$ M), $CD_3CN$, 20 min. $^1H$ NMR analysis indicated that 3-methyllumiflavin disappeared to give a substance characterized by its $^1H$ NMR spectrum to be *N*-[(trimethylsilyl)methyl]benzaldimine (**30**) and 4a-benzyl-4a,5-dihydro-3-methyllumiflavin (**15**) as major and minor products, respectively, and that other products of unknown identity had formed. $^1H$ NMR for **30**: 0.03 (s, 9 H, TMS), 3.37 (d, *J* = 1.3 Hz, 2 H, N-$CH_2$), 7.42 and 7.68 (m, 5 H, aromatic), 8.17 (t, *J* = 1.3 Hz, 1 H, benzylidene). NMR-tube irradiation of a nondegassed solution of 3-methyllumiflavin and **29** gave *N*-benzylformamide **31** (42%) and *N*-[(trimethylsilyl)-methyl]benzaldimine (**30**) (35%) in a 1:1 ratio.

3-Methyllumiflavin (3.0 × $10^{-3}$ M), *N*-benzyl-*N*-methylamine (**32**) (3.8 × 101–$^{3}$ M), $CD_3CN$, 10 min. $^1H$ NMR analysis indicated that 3-methyllumiflavin disappeared to give a substance characterized by its $^1H$ NMR spectrum to be *N*-methylbenzaldimine (**33**) and 4a-benzyl-4a,5-dihydro-3-methyllumiflavin (**15**) as major and minor products, respectively. $^1H$ NMR for **33**: 3.43 (d, *J* = 1.6 Hz, 3 H, N-$CH_3$), 7.42 and 7.70 (m, 5 H, aromatic) 8.29 (q, *J* = 1.6 Hz, 1 H, benzylidene). NMR-tube irradiation of a non-degassed solution of 3-methyllumiflavin and *N*-benzyl-*N*-methylamine (**32**) gave 100% conversion to *N*-methylbenzaldimine (**33**).

3-Methylumiflavin (3.3 × $10^{-3}$ M), benzylamine **34** (4.0 × $10^{-3}$ M), $CD_3CN$, 15 min. $^1H$ NMR analysis indicated that the amount of 3-methyllumiflavin and benzylamine decreased, and that *N*-benzylbenzaldimine (**35**) was formed as the sole product. A similar NMR tube reaction on a nondegassed solution gave of 3-methyllumiflavin and benzylamine gave complete conversion to *N*-benzylbenzaldimine (**35**).

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**Supplementary Material Available:** $^{13}C$ and $^1H$ NMR spectra for compounds **12**, **12-d$_2$**, **22**, **25**, **26**, **27**, and **28** (7 pages). Ordering information is given on any current masthead page.

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# Conformations and Structures of Tetra-*O*-alkyl-*p*-*tert*-butylcalix[4]arenes. How Is the Conformation of Calix[4]arenes Immobilized?

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*p*-*tert*-Butylcalix[4]arene ($1H_4$) was tetra-*O*-alkylated with alkyl halogens (RX: R = Me, Et, *n*-Pr, and *n*-Bu) in the presence of NaH as base, and the products ($1R_4$) were analyzed by HPLC and $^1H$ NMR spectroscopy. It was found that (i) ring inversion is suppressed by R greater than Et, (ii) the final conformer distribution in $1Pr_4$ and $1Bu_4$ is governed by the kinetic control, the main products being "cone" and "partial cone" (approximately in a 1:1 ratio), (iii) $1Me_4$ mostly exists as a thermodynamically stable partial-cone conformer, and (iv) $1Et_4$ shows an intermediary behavior between $1Me_4$ and $1Pr_4$: it mostly exists as a partial-cone conformer but slowly isomerizes to a "1,2-alternate" conformer at high temperature. The X-ray crystallographic analysis of partial-cone-$1Et_4$ was investigated. To clarify where and how the conformation of $1R_4$ is immobilized, we alkylated $1H_4$ in a stepwise manner. It was shown that when NaH is used as base, the conformation of $1Et_4$ is determined at the fourth ethylation step ($1HEt_3 \rightarrow 1Et_4$), whereas the conformation of $1Pr_4$ is determined at the third propylation step ($1H_2Pr_2 \rightarrow 1HPr_3$). The conformer distribution was significantly affected by alkali and alkaline earth metal cations used as base; in particular, it is worthy of mentioning that (i) when $Cs_2CO_3$ is used as base, 1,2-alternate-$1Pr_4$ is formed in addition to partial-cone-$1Pr_4$ and (ii) when $Ba(OH)_2$ is used as base, cone-$1Pr_4$ is yielded in 100% selectivity. On the basis of these studies, we discuss how the conformation of calix[4]arenes is immobilized.

Calix[4]arenes are cyclic oligomers made up from benzene units just as cyclodextrins are made up from glucose units. Although these two macrocyclic compounds have a similar cavity-shaped architecture, there exists a basic difference: the cyclodextrin cavity is conformationally fixed, whereas the conformational freedom still remains in the calixarene cavity.[1-7] It is known that unmodified *p*-*tert*-butylcalix[4]arene ($1H_4$) adopts a cone conformation because of strong hydrogen-bonding interactions among the OH groups, whereas introduction of alkyl or acyl substituents into the OH groups suppresses the conformational freedom because of steric hindrance (i.e., inhibition of the oxygen-through-the-annulus rotation) and results in conformational isomers.[1-13] However, a relation (if any) between the substituent effect and the conformer distribution has never been studied systemat-

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