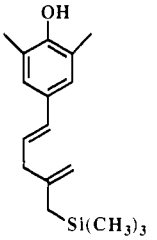
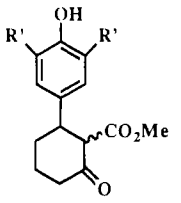
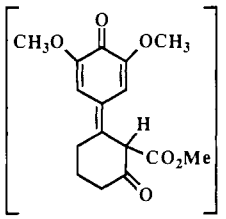
membered ring carbocycles. Entry 2 shows the tolerance of the cyclization to a protected alcohol (trimethylsilyl) adjacent to the allyl silane. Entries 4-6 show β-keto esters to be excellent cyclization terminators. In entries 4 and 5, the initially formed quinone methide **9** affords cyclohexenone **10**. Formation of **10** must occur via the oxidation of the initially formed product **14** to quinone methide **15** followed by loss of the acidic hydrogen, flanked by both a ketone and an ester. In support of this notion, cyclohexanone **14b** (1:1 mixture of diastereomers by $^1$H NMR) was isolated, characterized,[11] and resubmitted to the oxidation conditions to afford **10b**. Entry 5 in Table I shows that even with two oxygen substituents a quinone methide is still an excellent cyclization initiator.[12]




4




14a R' = $CH_3$
14b R' = $OCH_3$

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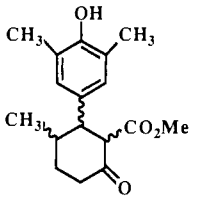


16

Entry 6 shows that in the presence of a Lewis acid ($ZnCl_2$) the addition of a β-keto ester to a quinone methide is clearly reversible. The initial cyclization of **12** gives **16** as a mixture of three diastereomers (1:1:1 ratio in $CDCl_3$ by $^1$H NMR spectroscopic analysis). Treating this mixture with $ZnCl_2$ (2.3 equiv, $CDCl_3$, 25 °C, 42 h) afforded cyclohexanone **13** as a 1.8:1 mixture of two diastereomers. This "epimerization", which undoubtedly occurs via a reversible addition of the β-keto ester to quinone methide **12**, allows *complete* control of the relative stereochemistry between the methyl and aryl substituents on the cyclohexanone. The 300 MHz $^1$H NMR spectrum of the major α epimer of **13** shows the benzylic hydrogen as a triplet ($\delta$ 2.81, $J$ = 12 Hz) indicative of the assigned stereochemistry.[11]

In conclusion, our work demonstrates the viability of quinone methide initiated cyclization reactions as a potentially general synthetic method. We are currently exploring the application of this methodology to the synthesis of natural products as well as investigating systems employing other terminators and highly reactive *o*- and *p*-quinone methides.[13]

**Acknowledgment.** This study was supported by grants from the UCR Committee on Research and National Institutes of Health (GM 39354). We thank Professors Tom Morton and Mark Midland for helpful comments on the manuscript.

**Supplementary Material Available:** A scheme showing the preparation of cyclization substrates and general experimental procedures for the preparation of **3**, **7**, **10**, and **13** along with full spectral data for all new compounds in Table I (8 pages). Ordering information is given on any current masthead page.

(12) The formation of **9b** could be observed by $^1$H NMR spectroscopy; however, we were unable to get a pure sample for characterization as **9b** forms **10b** upon standing in solution.

(13) Preliminary experiments show a furan to be an efficient cyclization terminator: Angle, S. R.; Yang, W.-J., unpublished results.

## Electron Spin Resonance Studies of Monoamine Oxidase B. First Direct Evidence for a Substrate Radical Intermediate

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*Received March 21, 1988*

Mitochondrial monoamine oxidase (MAO; E.C.1.4.3.4) is a flavoenzyme that has been known for 60 years to catalyze the oxidation of biogenic amines.[1] On the basis of the mechanism for chemical,[2] electrochemical,[3] and photochemical[4] amine oxidation, Silverman and co-workers[5] and Krantz and co-workers[6] proposed that MAO catalyzes amine oxidation by a single electron-transfer mechanism (Scheme I). Indirect evidence for radical intermediates has been obtained by Silverman and co-workers with the use of the *N*-cyclopropyl analogues, *N*-cyclopropylbenzylamine,[7] *N*-(1-methylcyclopropyl)benzylamine,[8] *N*-cyclopropyl-α-methylbenzylamine,[9] *trans*-2-phenylcyclopropylamine,[10] 1-phenylcyclopropylamine,[11] and 1-benzylcyclopropylamine.[12] All of these compounds are mechanism-based inactivators[13] of MAO and lead to the ring-opened adducts expected from cyclopropylaminyl radical intermediates.[14] 1-Phenylcyclobutylamine is metabolized by MAO to 2-phenyl-1-pyrroline,[15] the expected product of the amine radical cation-induced homolytic cleavage of the cyclobutyl ring, intramolecular radical capture by the incipient carbon-nitrogen double bond, and second-electron transfer.[16] All of these chemical probes for a radical intermediate support the single electron-transfer mechanism (Scheme I). Despite this strong indirect evidence, Tan et al.[17] found no ESR spectral support for radical intermediates in the MAO-catalyzed oxidation of benzylamine, even in the presence of spin traps. This inability to observe a radical intermediate may be based on kinetic or thermodynamic grounds. The oxidation of benzylamine may be so efficient that radical intermediates are processed rapidly

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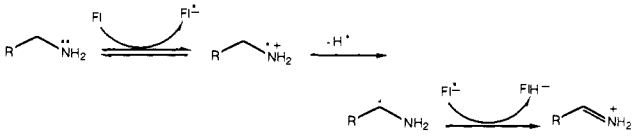
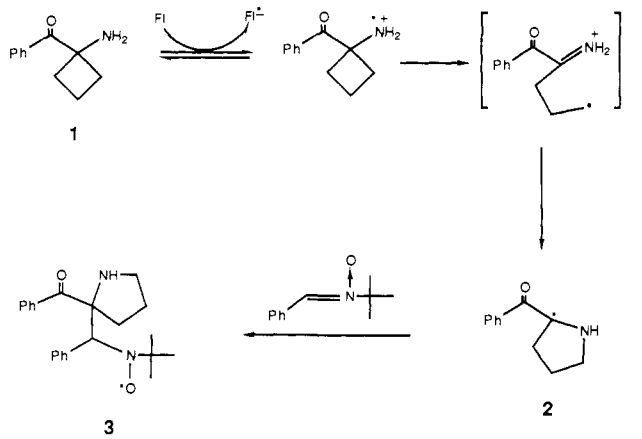
**Scheme I.** Proposed[5,6] Mechanism for Monoamine Oxidase-Catalyzed Amine Oxidation



**Scheme II.** Generation and Trapping of the Radical Intermediate from Monoamine Oxidase-Catalyzed Oxidation of 1-Amino-1-benzoylcyclobutane



with no leakage from the active site. Alternatively, the benzylamine radicals may not be sufficiently thermodynamically stable and are very short-lived. Spin-trapping techniques, however, have been effective in the identification of radicals generated by cytochrome P-450[18] and dopamine $\beta$-hydroxylase.[19] In this communication we report the first ESR spectral evidence for a spin trapped substrate radical intermediate generated by MAO.

Enhanced radical stabilization by the combined effect of donor and acceptor groups is known as capto-dative stabilization.[20] This is the basis for the design of a new substrate (**1**) for MAO which, by analogy to the mechanism of oxidation of 1-phenylcyclobutylamine by MAO,[15] is expected to produce the capto-dative radical **2** (Scheme II). This radical is very similar in structure to a capto-dative stabilized radical described by Koch et al.[21] which is stable at room temperature.

Compound **1**[22] did not inactivate beef liver MAO B at a concentration of 100 $\mu$M over a period of 25 h; at higher concentrations of **1** slow inactivation was observed with a $K_I$ = 16.7 mM and $k_{inact}$ = 0.016 min$^{-1}$. It also is a substrate, having a $K_m$ = 330 $\mu$M and a $k_{cat}$ = 0.2 min$^{-1}$ at pH 7.0. Benzylamine has a $K_m$ = 340 $\mu$M and $k_{cat}$ = 270 min$^{-1}$ at pH 7.0.[7b] Incubation of MAO B with **1** at room temperature, followed by ESR analysis at room temperature or at liquid nitrogen temperature, produced no signals above background in the ESR spectrum.[25] However,

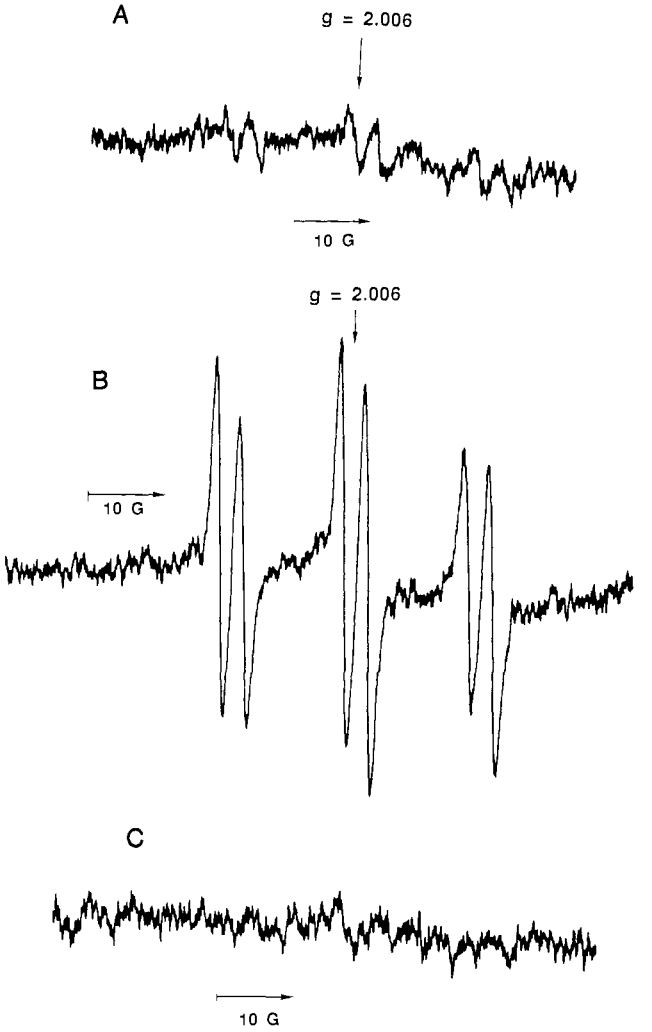



**Figure 1.** ESR spectra recorded after 2 (A) and 17 h (B) of incubation of MAO (500 $\mu$g), **1** (3 mM), and PBN (6 mM) in 20 mM potassium phosphate buffer, pH 7.0. A typical control spectrum (see text) also is shown (C).

when the spin trap, $\alpha$-phenyl *N-tert*-butylnitrone (PBN), was added at room temperature, the ESR spectrum obtained after 2 h was that shown in Figure 1A. After incubation at room temperature for 17 h, the ESR spectrum shown in Figure 1B was observed. A triplet of doublets ($a_N$ = 15.9 G; $a_\beta^H$ = 2.9 G) centered about a *g* value of 2.006 is exactly what is predicted for **3** (Scheme II), the adduct between PBN and **2**. Similar results were obtained when 1-phenylcyclobutylamine was the substrate. The slow rate of formation of **3** reflects several factors including the small rate constant of **1**, a small amount of leakage of **2** from the active site and/or the efficiency of radical trapping, and the fact that **1** also is an inactivator of MAO and PBN is a noncompetitive inhibitor of the enzyme. The following control experiments support the contention that the ESR signals arise from the spin-trapped intermediate of the reaction of **1** with MAO B: no ESR signals are observed after 17 h (1) in the absence of enzyme or in the presence of inactive enzyme; (2) if active enzyme is pretreated with the MAO inactivator pargyline; (3) if flavin mononucleotide or bovine serum albumin are substituted for MAO; (4) if **1** is omitted; (5) if the product of 1-phenylcyclobutylamine oxidation,

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(25) X-band ESR measurements were performed on a highly modified Varian E-4 spectrometer with 100-kHz field modulation. The microwave frequency was measured with a Hewlett-Packard Model X532B frequency meter, and the magnetic field was calibrated with 2,2-diphenyl-1-picrylhydrazyl as the external standard ($g$ = 2.0036). The sample was placed in either capillary tubes (50 $\mu$L) or a flat cell. A typical incubation mixture contained purified beef liver MAO B[26] (500 $\mu$g, specific activity 4 units/mg at pH 9.0) and substrate (3 mM) in a total of 260 $\mu$L of 20 mM potassium phosphate buffer, pH 7.0.

namely, 2-phenyl-1-pyrroline, is substituted for 1-phenylcyclobutylamine; (6) if benzylamine is substituted for **1**. The last control confirms the report of Tan et al.[17] who also observed no ESR signal with MAO B and benzylamine in the presence of PBN. It also indicates that the signals are not coming from hydrogen peroxide or any products of normal substrate turnover, although we have not yet identified the structure of the radical product. A typical control ESR spectrum is shown in Figure 1C. The possibility that a trace contaminant of cytochrome P-450, which has been shown to generate radical intermediates,[18] is responsible for the observed signals was negated on the basis of the following experiments: (1) pretreatment of the enzyme with acetylene, a known inactivator of cytochrome P-450,[27] had no effect on the generation of the ESR signal; (2) increasing amounts of PBN did not inhibit the generation of the ESR signal, as would be expected on the basis of the results of Augusto et al.[18] We have yet to be able to prepare the oxidation product of **1**, 2-benzoyl-1-pyrrolidine, presumably because of its instability.

These results provide the first direct evidence for a radical intermediate in the MAO-catalyzed oxidation of a substrate amine. The scope of this observation currently is under investigation.

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## Mechanism of the Photochemical Degradation of Poly(di-*n*-alkylsilanes) in Solution

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The photodegradation of polysilanes[2] $(RR'Si)_n$ with UV light in the neat solid[3-5] or in solution[6,7] is of considerable current interest. We now communicate the results of a mechanistic study of this process. Primary solution products are believed to be the silylenes RR'Si: identified by trapping with $Et_3SiH$ and polysilyl radicals -SiRR'-SiRR'•. The latter are implicated by the isolation of the silanes $H(SiRR')_2H$ and $H(SiRR')_3H$ after exhaustive irradiation at 254 nm. Irradiated solutions of polysilanes induce the polymerization of olefins providing further evidence for the homolytic cleavage route.[8] Since the photon energies are insufficient for simultaneous generation of two silyl radicals and a silylene by the cleavage of two adjacent SiSi bonds, we believe that silylene extrusion (eq 1) and homolytic cleavage (eq 2) are competing processes. Both processes are known in short-chain oligosilanes.[9,10]

$$-SiRR'-(SiRR')_2-SiRR'- \xrightarrow{h\nu} -SiRR'-SiRR'-SiRR'- + RR'Si: \quad (1)$$

$$-SiRR'-(SiRR')_2-SiRR'- \xrightarrow{h\nu} -SiRR'-SiRR'^{\bullet} + {}^{\bullet}SiRR'-SiRR'- \quad (2)$$

Recently, the structures of the persistent radicals present in irradiated solutions have been assigned as -SiRR'-•SiR-SiRR'- and -SiRR'-•SiR'-SiRR'-.[11] To account for their formation a minor third chain cleavage path was proposed starting from either (3A) or (3B); steps 4 and 5 produce the observed radical structures.

$$-SiRR'-(SiRR')_2-SiRR'- \longrightarrow \begin{cases} \longrightarrow -SiRR'-SiR_2R' + \\ \qquad R'\ddot{S}i-SiRR'- \\ \longrightarrow -SiRR'-SiRR_2' + \\ \qquad R\ddot{S}i-SiRR'- \end{cases} \quad (3A),\ (3B)$$

$$R\ddot{S}i-SiRR'-SiRR'- \longrightarrow -SiRR'-SiR{=}SiRR' \quad (4)$$

$$-SiRR'-SiR{=}SiRR' + {}^{\bullet}SiRR'-SiRR'- \longrightarrow -SiRR'-{}^{\bullet}SiR-(SiRR')_2-SiRR'- \quad (5)$$

Support for this proposal was provided by GC-MS observation of trialkylsilyl-terminated short chains upon exhaustive irradiation at 248 nm. We now report results that support the existence of separate paths (eq 1 and 2) which dovetail nicely with the previously proposed[12] interpretation of the photophysical behavior of alkylated polysilanes.

In cyclohexane-$Et_3SiH$ (1:1), exhaustive irradiation of poly(di-*n*-hexylsilane)[p-$(Hx_2Si)$] or poly(di-*n*-butylsilane)[p-$(Bu_2Si)$] at 248 nm (pulsed) or at 254 nm (cw)[6] produces the silylene trapping product $Et_3SiSiR_2H$ and the homolytic cleavage products $H(SiR_2)_nH$ ($n$ = 2, 3, R = Hx or Bu), respectively. Use of $Et_3SiD$ yields $Et_3SiSiR_2D$ and $H(SiR_2)_nH$ ($n$ = 2, 3, R = Hx or Bu). Previous work did not rule out the possibility that $R_2Si$: is only produced from very short photodegraded silicon chains and not from the high polymer. However, in the irradiation of p-$(Bu_2Si)$, the $Et_3SiSiBu_2H$ appears with no induction period and we thus conclude (i) that reaction 1 occurs from the high polymer. The quantum yield of $Et_3SiSiBu_2H$ decreases as the irradiation wavelength increases (GC-MS, internal standard) and falls to zero above 300 nm, while the polymer absorption band at 315 nm still disappears rapidly and persistent ESR signals are observed.

We conclude therefore that (ii) silylene production and radical formation indeed occur as two distinct processes, presumably reactions 1 and 2, and that (iii) the proposed[4] thermal fragmentation of polysilyl radicals with a sequential loss of single RR'Si: units (see below) does not occur at room temperature. This

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