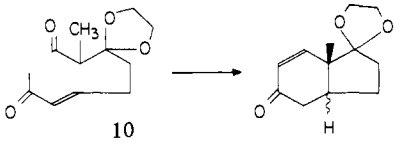
were in complete agreement with those obtained in the cyclization of **1**, using the same base systems already mentioned. It is further remarkable that examination of the results obtained with the additional systems $Mg(OCH_3)_2$, $Ca(OCH_3)_2$, and $Ba(OH)_2$, all in methanol, again showed that the ratio of *trans*- to *cis*-hydrindenones eventually formed follows the order expected on the assumption that the metals forming the tighter bond to oxygen would lead to more *trans* product. The observed trans/cis ratios (determined by NMR integration) were as follows: LiOH, 4/1; $Mg(OCH_3)_2$, 12/1; $Ca(OCH_3)_2$, 10/1; $Ba(OH)_2$, 3/1; $Zr(OnPr)_4$, 25/1 (determined by VPC analysis, 3% FFAP, 180 °C).[13]



10

It is clear that the internal Michael addition is a useful route to angularly methylated *trans*-hydrindanes and an important method in the control of vicinal stereochemistry.

**Acknowledgment.** We thank the National Institutes of Health and the National Science Foundation for their support of this work.

**Registry No.** (*E*)-**1**, 79971-12-3; (*Z*)-**1**, 79971-13-4; **2**, 79971-14-5; *cis*-**3**, 79971-15-6; *trans*-**3**, 17429-25-3; **4**, 19980-33-7; **5**, 52456-90-3; **6**, 76712-36-2; **7**, 79971-16-7; **8**, 79971-17-8; *cis*-**9**, 79971-18-9; *trans*-**9**, 62719-12-4; **10**, 79971-19-0; *cis*-2′,3′,3′a,7′a-tetrahydro-7′a-methylspiro[1,3-dioxolane-2,1′-[1*H*]inden]-5′(4′H)-one, 79971-20-3; *trans*-2′,3′,3′a,7′a-tetrahydro-7′a-methylspiro[1,3-dioxolane-2,1′-[1*H*]inden]-5′(4′H)-one, 79971-21-4; dimethyl (2-oxopropyl)phosphonate, 4202-14-6.

(13) The stereochemistry of the hydrindenones was easily established by hydrogenation followed by ketal hydrolysis to give the well-known *cis*- (Boyce, C. B. C.; Whitehurst, J. S. *J. Chem. Soc.* **1960**, 4547) and *trans*-hydrindanediones (Baggaley, K. H.; et al. *J. Chem. Soc. C* 1971, 2671; Hajos, Z. G., Parrish, D. R. *J. Org. Chem.* **1973**, *38*, 3239).

## Thioacrolein[1]

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The preparation of reactive organic intermediates by means of thermal decomposition in the gas phase can be optimized advantageously by following the PE spectroscopic "molecular fingerprints" in the respective flow system:[2] those of the starting materials vanish and those of the products emerge. Applying this technique, many thiocarbonyl derivates like $H_2C{=}S$[3a], or $H_2C{=}C{=}S$,[3b] or alkyl derivatives thereof[3a] have been detected and characterized. Still missing is the PE spectroscopic proof for monomeric $H_2C{=}CH{-}HC{=}S$,[4] the thio analogue to well-known acrolein, although MNDO calculations[5] predict it to be the most

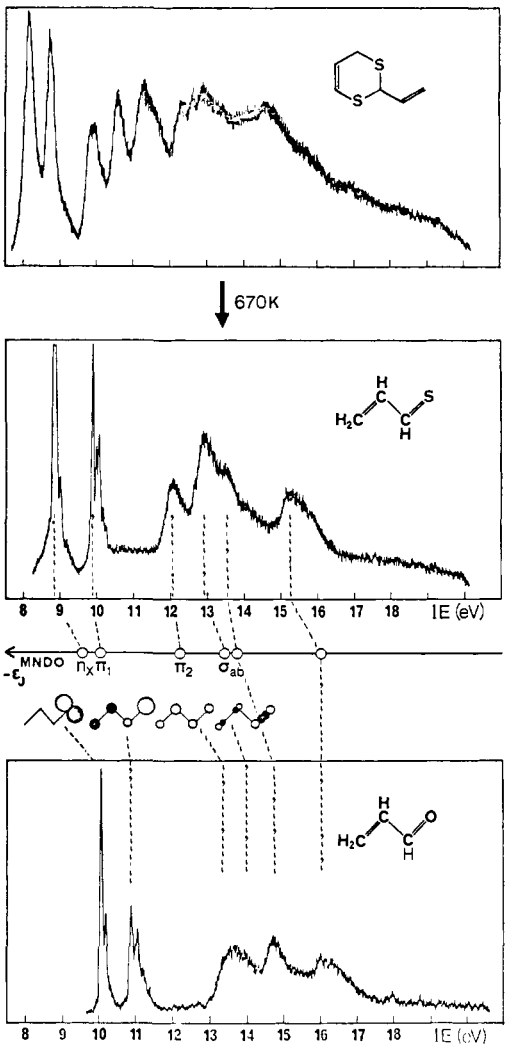
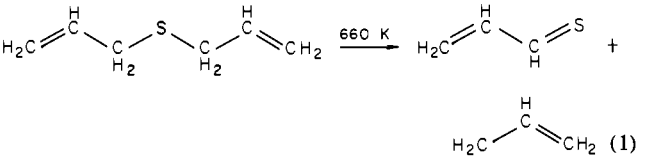



**Figure 1.** PE spectra of thioacrolein.

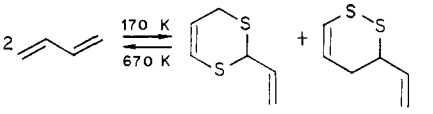
stable isomer out of the set of 12 which can be constructed for the ensemble $C_3H_4S$ by using normal valency rules.

One of the most favorable leaving groups in thermal decompositions from allylic compounds is propene,[2,6] and expectedly, diallyl sulfide[4] chosen as precursor yields quantitatively thioacrolein at the PE spectroscopically optimized temperature of 660 K.




(1)

The PE spectra recorded on heating the flow system show the developing 8.87 eV ionization peak of thioacrolein (Figure 1), but all other bands at higher energies overlap with those of propene (reaction 1). The PE spectrum of pure thioacrolein (Figure 1), however, could be obtained after cool trapping the Diels–Alder dimer mixtures




(2)

and evaporating the slowly polymerizing liquid again while heating

(1) Gas-phase Reactions. 29. Part 28: H. Bock. *Chem. Rundsch.*, **34** (29), 3 (1981).

(2) For a recent review, see H. Bock and B. Solouki, *Angew. Chem.*, *93*, 425 (1981); *Angew. Chem., Int. Ed. Engl.*, **20**, 427 (1981).

(3) (a) Cf. E. Block, E. R. Corey, R. E. Penn, T. L. Renken, P. F. Sherwin, H. Bock. T. Hirabayashi, S. Mohmand, and B. Solouki, *J. Am. Chem.Soc.*, in press; H. Bock, T. Hirabayashi, and S. Mohmand, Chem. Ber., in press; see the literature reviewed, e.g., H. W. Kroto and R. J. Suffolk, *Chem. Phys. Lett.*, **15**, 545 (1972);B. Solouki, P. Rosmus, and H. Bock, *J. Am. Chem. Soc.*, **98**, 6054 (1976); H. Bock, B. Solouki, S. Mohmand, E. Block, and L. K. Revelle, *J. Chem. Soc., Chem. Commun.* **1977**, 287; (b) H. Bock, B. Solouki, G. Bert, and P. Rosmus, *J. Am. Chem. Soc.*, **99**, 1663 (1977). See also P. Rosmus, B. Solouki, and H. Bock, *Chem. Phys. Letts.*, *22*, 453 (1977).

(4) Cf. W. J. Bailey and M. Isogawa, *Am. Chem. Soc., Div. Polymer Chem.*, **14** (1), 300 (1973); G. Giles, R. A. Marty, and P. de Mayo, *J. Chem. Soc., Chem. Commun. 1974*, 409; *Can. J. Chem.*, **54**, 537 (1976).

(5) H. Bock, S. Mohmand, T. Hirabayashi, and A. Semkow, *Chem. Ber.*, in press.

(6) Cf., e.g., R. F. C. Brown "Pyrolytic Methods in Organic Chemistry", Academic Press, New York, 1980 and literature cited. See also F. A. Houle and J. J. Beauchamp, *J. Am. Chem. Soc.*, **100**, 329o (1978); H. Bock, A. Bowling, B. Solouki, T. J. Barton, and G. T. Burns, *ibid.*, **102**, 429 (1980).

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the vapor to 670 K oven temperature.

The PE spectroscopic assignment is easily accomplished by either the Koopmans' correlation $IE_n = -\epsilon_j^{MNDO}$ with MNDO eigenvalues—the MO sequence (Figure 1) starts with the lone pair $n_s$ and $2\pi$ orbitals—or by $M^{+\cdot}$ state comparison with the iso (valence) electronic acrolein, the PE spectrum[8] of which (Figure 1) displays a similar ionization pattern shifted to higher energy due to the increased effective nuclear charge of oxygen relative to sulfur.

Thus, by thermal retrodiene splitting of the Diels–Alder dimer mixtures, which can be isolated and steam-distilled, thioacrolein becomes available for preparative use[6] and further spectroscopic investigation.

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**Registry No.** Thioacroleine, 53439-64-8; diallylsulfide, 592-88-1; 2-vinyl-4*H*-1,3-dithiine, 80028-57-5; 3-vinyl-4*H*-1,2-dithiine, 62488-53-3.

(7) Anal. Calcd for $(C_3H_4S)_2$: C, 49.96; H, 5.59; S, 44.45; $M_r$ 144.25. Found: C, 49.60; H, 5.80; S, 45.0. The structure of the dimers and their relative concentrations have been established by $^1H$ NMR[4] spectroscopy: 90% 2-vinyl-4*H*-1,3-dithiine and 10% 3-vinyl-4*H*-1,2-dithiine. MNDO calculations predict $\Delta H_f^\circ$ = 111 kJ/mol for the 1,3 isomer and 126 kJ/mol for the 1,2 isomer with a disulfide link, in good agreement with the product ratio found.

(8) Cf., e.g., D. W. Turner, C. Baker, A. D. Baker, and C. R. Brundle, "Molecular Photoelectron Spectroscopy", Wiley-Interscience, London, 1970; A. Katrib and J. W. Rabalais, *J. Phys. Chem.*, 77, 2358 (1973). The vibrational fine structures in our record amount to $\nu^+ = 1200 \pm 100$ cm$^{-1}$ for $IE_1$ to $\nu^+ = 1450 \pm 100$ cm$^{-1}$ for $IE_2$, further confirming the assignment ($\nu_{C=S} \sim 1200$ cm$^{-1}$ and $\nu_{C=C} \sim 1650$ cm$^{-1}$.

## Cleavage of Double Helical DNA by (Methidiumpropyl-EDTA)iron(II)

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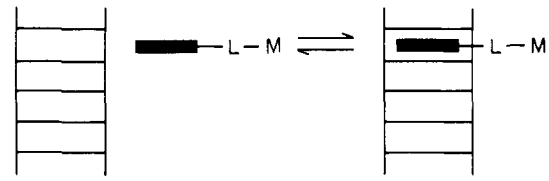
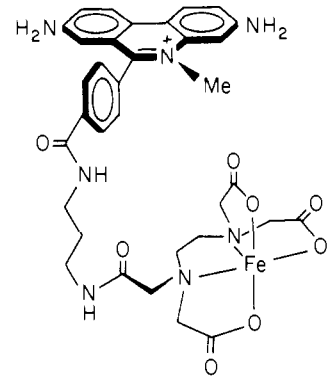
Metal ions have been implicated as cofactors in the strand scission of DNA for a number of antitumor antibiotics. Bleomycin, a glycopeptide antibiotic, binds to and cleaves DNA in a reaction that depends on ferrous ion and molecular oxygen.[2-4] The antitumor agent streptonigrin is capable of causing single-strand breaks in DNA by using oxygen and cuprous ions.[5] Recently, the 1,10 phenanthroline–cuprous complex has been shown to cleave DNA in the presence of oxygen.[6,7] These examples involve the concept of using a DNA-binding molecule to deliver a metal ion to the site of the DNA helix where activation of molecular oxygen results in cleavage of the DNA.

We report the synthesis of a simple bifunctional molecule, methidiumpropyl-EDTA (MPE) (**1**), which contains the DNA intercalator methidium[8] covalently bound by a short hydrocarbon




tether to the metal chelator EDTA.[11] In the presence of ferrous ion and oxygen this reagent *efficiently* produces single-strand breaks and some double-strand breaks in double helical DNA.




The acylimidazole ester of *p*-carboxymethidium chloride (**2**)[12,13] was allowed to react with an excess of 1,3-diaminopropane in $Me_2SO$ at 25 °C, affording a maroon solid product, methidiumpropylamine (**3**) (eq 1). Condensation of **3** with excess EDTA in dry DMF at 120 °C yielded (methidiumpropyl-EDTA) (MPE), in an overall yield of 59% after chromatography on silica gel 60 (230–400 mesh ASTM). MPE was rendered metal free by treatment of an acidic aqueous solution with $Na_2EDTA$ followed by purification on Amberlite XAD-2 polystyrene resin.[14]

The cleavage of DNA was followed by monitoring the conversion of supercoiled (form I) pBR-322 plasmid DNA, $10^{-5}$ M in base pairs (bp), to open circular and linear forms (forms II and III, respectively). The introduction of one single-strand break converts form I to form II. EDTA–$Fe^{II}$ at $>10^{-4}$ M concentrations will cleave plasmid DNA; however, at concentrations $\leq 10^{-4}$ M little or no cleavage takes place. The addition of intercalator ethidium bromide (EB) to Fe(II) or EDTA–$Fe^{II}$ does not promote the cleavage reaction. We find that MPE–$Fe^{II}$ at two orders of magnitude lower concentration ($10^{-6}$ M) cleaves plasmid DNA (Table I). MPE alone or MPE–$Fe^{III}$ is inactive at these con-

(1) Camille and Henry Dreyfus Teacher Scholar, 1978–1983.

(2) "Bleomycin: Chemical, Biochemical and Biological Aspects"; Hecht, S. M., Ed.; Springer-Verlag: New York, 1979.

(3) Sausville, E. A.; Peisach, J.; Horwitz, S. B. *Biochemistry* **1978**, *17*, 2740. For a recent review, see: Burger, R. M.; Peisach, J.; Horwitz, S. B. *Life Sci.* **1981**, *28*, 715 and references cited therein.

(4) Lown, J. W.; Sim, S. K. *Biochem. Biophys. Res. Commun.* **1977**, *77*, 1150.

(5) Cone, R.; Hasan, S. K.; Lown, J. W.; Morgan, A. R. *Can. J. Biochem.* **1976**, *54*, 219 and references cited there.

(6) (a) Sigman, D. S.; Graham, D. R.; D'Aurora, V.; Stern, A. M. *J. Biol. Chem.* **1979**, *254*, 12269. (b) Graham, D. R.; Marshall, L. E.; Reich, K. A.; Sigman, D. S. *J. Am. Chem. Soc.* **1980**, *102*, 5419. (c) Marshall, L. E.; Graham, D. R.; Reich, K. A.; Sigman, D. S. *Biochemistry* **1981**, *20*, 244.

(7) Que, B. G.; Downey, K. M.; So, A. G. *Biochemistry* **1980**, *19*, 5987.

(8) Previous studies[9] have shown that substitution of a methyl for an ethyl group or addition of a *p*-carboxyl group to ethidium bromide[10] has little effect on the unwinding angle.

(9) Waring, M. J.; Wakelin, L. P. *Mol. Pharmacol.* **1974**, *9*, 544.

(10) LePecq, J. B.; Paoletti, C. *J. Mol. Biol.* **1967**, *27*, 87.

(11) For a crystal structure of EDTA–$Fe^{III}$, see: Kennard, C. H. L. *Inorg. Chim. Acta* **1967**, *1*, 347.

(12) May and Baker, Ltd., Nottingham, England.

(13) Dervan, P. B.; Becker, M. M. *J. Am. Chem. Soc.* **1978**, *100*, 1968.

(14) MPE was ≥99% pure by HPLC in two solvent systems (ALTEX Ultrasphere ODS; 86:14 $H_2O$–$CH_3CN$, retention time 8.4 min, and 70:30 $H_2O$–MeOH, retention time 19.2 min). The NMR and IR spectra data were consistent with the assigned structure. MPE was isolated from methanol/water as the hexahydrate. Anal. Calcd for $C_{34}H_{51}N_7O_{14}$: C, 52.23; H, 6.57; N, 12.54. Found: C, 52.48; H, 6.12; N, 12.50.

(15) pBR-322 plasmid was 95% form I and 5% form II. The data in Tables I, II, and III are corrected for the 5% form II and the decreased stainability of form I.[16]

(16) The correction factor for form I pBR-322 DNA was determined to be 1.22 by the method of Haidle et al.[17]

(17) Haidle, C. W.; Lloyd, R. S.; Robberson, D. L. In "Bleomycin: Chemical Biochemical and Biological Aspects"; Hecht, S. M., Ed.; Springer Verlag: New York, 1979; p 222.

(18) The average number of single-strand scissions per DNA molecule, $S$, was calculated from the Poisson distribution, $P_n = [S^n/n!]e^{-s}$ where $P_n$ is the fraction of molecules that have $n$ nicks each.[19] This equation assumes that the nicks are distributed at random among the DNA population.[20] When only forms I and II of the DNA are present, this simplifies to $S = -\ln f_I$ where $f_I$ is the fraction of form I molecules. When all three forms of DNA are present, $S$ can be calculated from the following equation:[21] $f_I + f_{II} = [1 - S(2h + 1)/2L]^{S/2}$ where $h$ is the distance between nicks on opposite strands needed to produce a linear molecule (16 bp)[21] and $L$ is the total number of bp's in pBR-322 (4361).

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