propyl-9-methyl-1,3-dioxo-3a,4,5,6,7,8,9,9a-octahydronaphtho[2,3-*c*]-furan, 92013-80-4; (4α,4aβ,10aβ,11α)-4,11-dimethyl-5,10-dioxo-1,3,4,4a,10a,11-hexahydroanthra[2,3-*c*]furan, 92013-81-5; *N*-phenylmaleimide, 941-69-5; naphthoquinone, 130-15-4; maleic anhydride, 108-31-6; dicyclopentadienyltitanium dichloride, 1271-19-8; sodium amalgam, 11110-52-4.

Leucomitomycins

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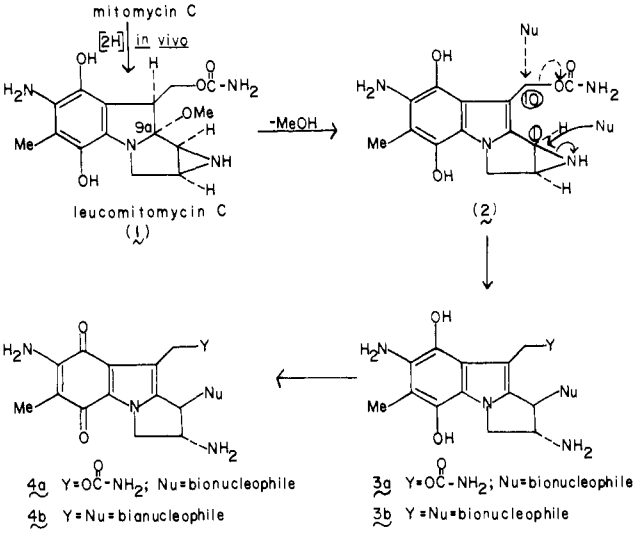
Mitomycin C, a member of a larger family of structurally related antibiotics, has become a clinically useful resource in cancer chemotherapy.[1,2] In view of its novel structure and its chemical fragility, a great deal of research has gone into establishing the mode of antitumor action of this compound.[3] The prevailing consensus is that mitomycin C undergoes in vivo reduction of its quinone ring and that the resultant hydroquinone, **1**, suffers very rapid loss of methanol to afford the indole, **2**.[4-6] In compound **2**, the benzylic aziridine carbon (C_1) becomes a potent electrophile toward various nucleic acid centered nucleophiles. Less securely supported is the proposition that the benzylic carbon (C_{10}) bearing the carbamoyloxy group can also alkylate a nucleophilic center of DNA. The alkylation product **3a** (or cross-linked product **3b**), upon reoxidation, is converted to the quinone form (**4a** or **4b**).



In a commendably rigorous investigation, Tomasz and Nakanishi[7,8] described the reduction of mitomycin C either by hydrogen over PtO_2 or by NADPH in the presence of d(GpC) and identified the site of apparent "bioelectrophilicity" to be C_1. The nucleophilic center was shown to be the "amidic" oxygen of the deoxyguanosyl system.

Pursuant to a synthetically oriented study, we became interested in an earlier step of the sequence, i.e., the loss of the 9a-oxygen function. As a consequence of the early work addressed to the structure determination of the mitomycins,[9,10] it was assumed that, upon reduction of the quinone ring to the hydroquinone level (cf. **1**), the loss of the C_{9a} hetero function, resulting in formation of the indolohydroquinone (cf. **2**), is inevitable.[11] It has been reasoned that only in the quinone series is the reactivity of the pyrrolidine nitrogen sufficiently attenuated to allow for survival of the 9a heterofunction. *The findings reported herein run sharply counter to this conventional wisdom and demonstrate that aromatization of compounds such as* ***1****, dihydro(leuco)mitomycin C and* ***6****, dihydro(leuco)mitomycin B is far from inevitable.*

We first investigated the reduction of the very rare mitomycin B (**5**).[12] The NMR spectrum of compound **5** in pyridine-d_5 at 250 MHz was recorded. The spectrum indicated the sample to be substantially homogeneous. To this solution was added 10% palladium on charcoal. Hydrogen gas was bubbled through the system. Within seconds, the purple color faded completely. The NMR spectrum of the "leuco" compound was recorded. The NMR spectrum rigorously confirms the conclusion, which would be gathered from the disappearance of the purple color, i.e., that compound **5** is no longer present.

The multiplicities in the NMR spectrum of the resultant leuco product indicate that it is not the expected "dihydro–anhydro" compound **2**. Most convincing in this regard is the fact that the proton at C_9 is clearly seen as a doublet of doublets δ 4.76, with couplings of 3.24 and 9.91 Hz to the diastereotopic protons at C_{10}. Each of these protons (H_{10} δ 5.40 and $H_{10'}$ 5.74) is, in turn, seen as a doublet of doublets: $J_{10,10'} = 10.37$ Hz; $J_{10,9} = 9.91$ Hz; $J_{10',9} = 3.24$ Hz.

The NMR-based conclusion is fully in keeping with the chemical events. When a stream of oxygen is passed through the solution of leuco compound, the purple color reemerges almost instantaneously. The high-field NMR spectrum of this material indicates it to be the starting compound **5**. Under these conditions, no aziridinomitosene (**8**) or degradation products thereof could be detected. In fact, the sequence of reduction followed by oxidation leads to virtually no erosion of the homogeneity of compound **5**.

The infrared spectra of compound **5** and of the leuco compound each exhibit stretching maxima (1724 cm^{-1} for **5**; 1721 cm^{-1} for the dihydro compound) which are attributable to the carbonyl group of the carbamate. In the case of compound **5**, quinone maxima at 1649 and 1628 cm^{-1} are also present. In the case of the leuco compound, the intensities of the quinonoid peaks are

(1) Remers, W. A. In "Anticancer Agents Based on Natural Product Models"; Cassady, J. M., Duoros, J. D., Eds.; Academic Press: New York, 1980; pp 131 ff.

(2) Carter, S. K.; Crooke, S. T. "Mitomycin C—Current Status and New Developments"; Academic Press: New York, 1979.

(3) For a recently revised assignment of absolute configurations of mitomycin C, see: Shirahata, K.; Hirayama, N. *J. Am. Chem. Soc.* **1983**, *105*, 7199. In this paper the previous absolute configuration of mitomycin B (**5**) was also questioned. For the moment, we continue to draw the absolute configuration of **5** as formulated by: Yahashi, R.; Matsubara, J. *J. Antibiot.* **1976**, *29*, 104; **1978**, *31*, 6.

(4) For recent reviews, see: (a) Lown, J. W. In "Molecular Aspects of Anticancer Drug Action"; Neidle, S., Waring, M. J., Eds.; Verlag Chemie; Deerfield Beach, FL, 1983. (b) Lown, J. W. *Acc. Chem. Res.* **1982**, *15*, 381. (c) Szybalski, W.; Iyer, V. N. In "Antibiotics: Mechanism of Action"; Gottlieb, D., Shaw, P. D., Eds.; Springer-Verlag: New York, 1967; pp 211-245.

(5) For earlier papers in the original literature, see: (a) Iyer, V. N.; Szybalski, W. *Science (Washington, D.C.)* **1964**, *145*, 55. (b) Lown, J. W.; Begleiter, A.; Johnson, D.; Morgan, A. R. *Can. J. Biochem.* **1976**, *54*, 110. (c) Tomasz, M.; Lipman, R. *J. Am. Chem. Soc.* **1979**, *101*, 6063.

(6) For a comprehensive treatment of the subject of bioreductive alkylating agents, see: Moore, H. W. *Science (Washington, D.C.)* **1977**, *197*, 527.

(7) Tomasz, M.; Lipman, R.; Snyder, J. K.; Nakanishi, K. *J. Am. Chem. Soc.* **1983**, *105*, 2059.

(8) Weaver, J.; Tomasz, M. *Biochim. Biophys. Acta* **1982**, *697*, 252.

(9) (a) Patrick, J. B.; Williams, R. P.; Meyer, W. E.; Fulmor, W.; Cosulich, D. B.; Broschard, R. W.; Webb, J. S. *J. Am. Chem. Soc.* **1964**, *86*, 1889. (b) Stevens, C. L.; Taylor, K. G.; Munk, M. F.; Marshall, W. S.; Noll, K.; Shah, G. D.; Uzu, K. *J. Med. Chem.* **1964**, *8*. (c) Kinoshita, K.; Uzu, K.; Nakano, K.; Shimizu, M.; Takahashi, T.; Matsui, M. *J. Med. Chem.* **1971**, *14*, 103. (d) Hornemann, U.; Ho, Y. K.; Mackey, J. K.; Srivastava, S. C. *J. Am. Chem. Soc.* **1976**, *98*, 7069.

(10) For excellent reviews of the voluminous chemistry of the mitomycins, see: (a) Remers, W. A. In "The Chemistry of Antitumor Antibiotics"; Wiley-Interscience: New York, 1979; pp 221 ff. (b) Franck, R. W. *Fortschr. Chem. Org. Naturst.* **1979**, 381.

(11) The possibility of trapping either through reduction or through reaction with other nucleophiles, imminium species intermediates which would otherwise aromatize is implicit in the work of Matsui[9c] and Hornemann,[9d] respectively. One apparent precedent suggesting the viability of systems such as **1** and **6** is a result quoted by Franck[10b] wherein it is claimed, without supporting data, that reduction of mitomycins A or C with lithium aluminum hydride followed by reoxidation (air) affords the 10-decarbamoyl compounds with the angular methoxy group intact. Also, Patrick et al.[9a] briefly indicate that in the catalytic hydrogenation of mitomycin B, inclusion of triethylamine retards the rate of indolization. However, previous to our work no one has characterized compounds such as **1** or **6**.

(12) We thank Dr. N. Shirahata of the Kyowa Institute of Japan for a sample of mitomycin B.

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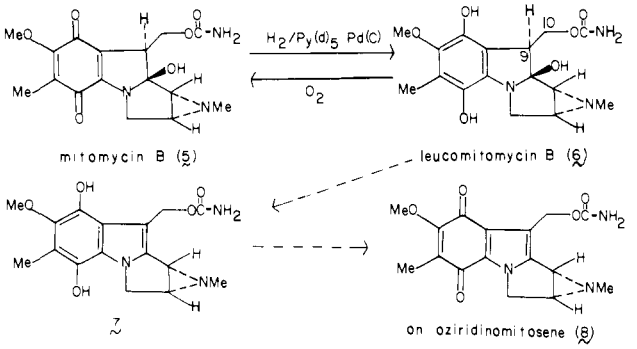
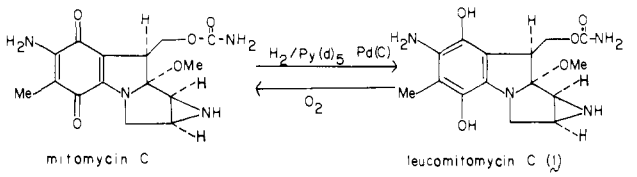
sharply reduced. (At the moment we are unable to carry out the transfer from the NMR tube to the IR cell without incurring some slight oxidation). The infrared spectrum of the material produced by air oxidation of the leuco compound is identical with that of the original **5**. *On the basis of its infrared and NMR spectra,*[13] *and on the basis of its clean reconversion to* **5**, *the leuco compound can be confidently formulated as* **6**.[14]

The leuco compound, **6**, can also be generated by reduction of **5**, in ethanol. Upon proper sensitivity to experimental detail, when an ethanolic solution of compound **6** is evaporated to dryness and the residue redissolved in pyridine-d_5, the resulting NMR spectrum is virtually identical with that obtained when the reduction is carried out in pyridine directly. However, the material handled in this way is of lower purity than that generated directly in pyridine **5**. Reoxidation of compound **6** generated in ethanol also restores essentially pure **5**.



An identical reduction in pyridine-d_5 was carried out with mitomycin C. Again, a leuco product was produced. The NMR spectrum (at 490 MHz) of this lecuo system is similar to, but clearly different from, that of mitomycin C. In this case, the NMR spectroscopic measurements were complicated both by serious line broadening (possibly due to the aminohydroquinone and secondary (NH) aziridine functionalities) and by the presence of a large H_2O–HOD resonance.[15] Though the multiplicities of the various signals are not sufficiently resolved to allow for full assignment of the NMR spectrum, its key features including the signals from the C_6 Me (δ 2.46, s, 3 H), the C_{9a} OMe (3.31, s, 3 H), and the six remaining carbon-bound protons are clearly discerned.[15] Once again, treatment of the leuco solution with oxygen results in the immediate regeneration of essentially pure mitomycin C, as evidenced by its virtually clean NMR spectrum. Adventitious oxidation of the leuco dihydro system **1** is even more facile than the corresponding process in the case of compound **6**. Thus, the act



of transferring compound **1** from the NMR tube to an IR cell resulted in substantial reoxidation back to mitomycin C thereby compromising the value of the infrared spectrum. However, as in the B series, elimination to the indolohydroquinone **2** is by no means automatic. In fact, from an experimental standpoint, avoidance of reoxidation back to the parent mitomycin poses a greater challenge than avoidance of elimination.

The dihydro (leuco) compounds constitute new access points for understanding the biological behavior of the fascinating family of mitomycin drugs. Experiments intended to further that understanding are planned.

Acknowledgment. This work was supported by PHS Grant CA28824. NMR spectra were obtained through the auspices of the Northeast Regional NSF/NMR Facility at Yale University, which was supported by NSF Chemistry Division Grant CHE7916210. We also thank Dr. K. Shirahata of the Kyowa Chemical Co. for a gift of mitomycin B.

Registry No. **1**, 92056-69-4; **5**, 4055-40-7; **6**, 92056-68-3; mitomycin C, 50-07-7.

Supplementary Material Available: Experimental procedures for the preparations of dihydro(leuco)mitomycins **1** and **6**, and NMR spectral tabulations (4 pages). Ordering information is given on any current masthead page.

(13) The fully assigned NMR spectrum of compound **6** in conjunction with that of compound **5** is provided in the supplementary material.

(14) Our NMR data[13] do not decisively deal with the question to whether the leuco compound might exist as its azocinone (secondary amino keto valance isomer). The FT IR spectrum does not suggest the presence of an additional ketone.

(15) The chemical shifts of these protons are provided in the supplementary material. However, due to serious line broadening in the NMR spectrum of the leuco compound, a definitive assignment of protons via decoupling is not possible. By running the 490-MHz spectrum at 40 °C, the H_2O–HOD absorption is moved upfield and out of the range of the critical methine and methylene protons.

Synthesis of Prostaglandins via a 2,3-Dioxabicyclo[2.2.1]heptane (Endoperoxide) Intermediate. Stereochemical Divergence of Enzymatic and Biomimetic Chemical Cyclization Reactions

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The view that prostaglandin (PG) biosynthesis from C_{20} polyunsaturated fatty acids occurs by a free radical process through intermediates such as **1**–**3**[1] is supported by the actual isolation of the endoperoxides PGG_2 and PGH_2.[2] In this paper we report the first realization of a purely chemical synthesis of prostaglandins by a free radical pathway through an endoperoxide intermediate. Evidence is also provided that the enzymatic and purely chemical pathways differ with regard to stereochemical preference in the step that determines the relative orientation of the two appendages on the 2,3-dioxabicyclo[2.2.1]heptane nucleus. A rational explanation is offered for the selective formation of cis oriented appendages (exo,exo and endo,endo) in the nonenzymatic ring closure to the endoperoxide system.

For this study we utilized as substrates the racemic[3] isomeric hydroperoxides **4** and **5** which differ with regard to geometry of the homoallylic 3,4 double bond. These hydroperoxides were obtained in ca. 45% overall yield from the corresponding alcohols by (a) conversion to mesylate using mesyl chloride–triethylamine in CH_2Cl_2 at −78 °C for 20 min, (b) reaction of the mesylate with a dry (20%) solution of hydrogen peroxide in ether initially at −110 °C (30 min) then at −110 to −78 °C (30 min) and finally at −78 °C (1 h) and (c) quenching the reaction mixture at −78 °C with deionized water, extraction with ether, and preparative sg TLC[4] at 5 °C using 1:1 ether–hexane containing 1% of triethylamine

(1) Nugteren, D. H.; Beerthuis, R. K.; van Dorn, D. A. *Recl. Trav. Chim. Pays-Bas* **1966**, *85*, 405.

(2) Hamberg, M.; Samuelsson, B. *Proc. Natl. Acad. Sci. U.S.A.* **1973**, *70*, 899.

(3) All intermediates and products described herein are racemic. Satisfactory ^1H NMR, infrared, ultraviolet and (in the case of thermally stable substances) mass spectral data were obtained for each new compound.

(4) Thin-layer chromtography on silica gel.

(5) The hydroperoxides **4** and **5** were obtained in 90% yield as a 1:1 mixture with the isomeric hydroperoxide having the (*E*,*E*)-diene unit and hydroperoxide function at C(10). The byproduct, apparently formed as a result of a competing S_Ni pathway, was not separated since it did not interfere with either the formation or purification of **6** or **7** in the next step.

(6) For previous use of this methodology, see: Corey, E. J.; Marfat, A.; Falck, J. R.; Albright, J. O. *J. Am. Chem. Soc.* **1980**, *102*, 1433.

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