Communications to the Editor

Fredericamycin A, an Antitumor Antibiotic of a Novel Skeletal Type

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We report here the structure of fredericamycin A (NSC-305263), an antitumor antibiotic produced by *Streptomyces griseus* (FCRC-48). The compound has an entirely novel spiro ring system, not previously found in the antibiotic structures. The isolation and some of the physicochemical and biological properties of fredericamycin A, B, and C have been reported earlier.[1,2]

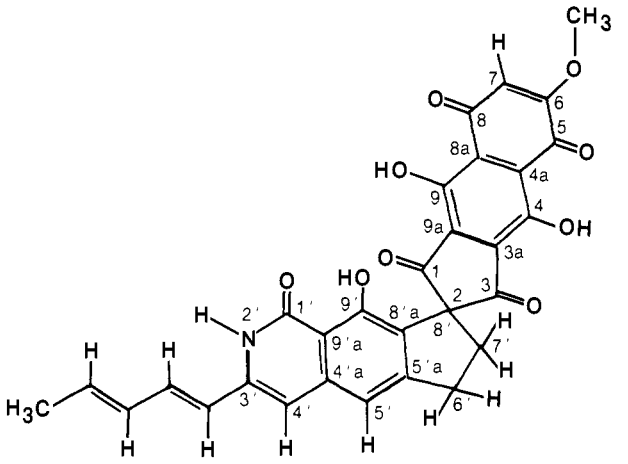
Fredericamycin A, the major active component of the fermentation broth, is sparingly soluble in most polar solvents with the exception of *N,N*-dimethylformamide, dimethyl sulfoxide, and pyridine.

After a number of attempts, we were able to crystallize fredericamycin A into thin, plateletlike crystals from acetonitrile–water. The structure elucidation was carried out by single-crystal X-ray analysis and by consideration of various types of mass spectral and spectroscopic data.

The crystalline material, mp >350 °C dec, showed the molecular ion at m/z 539.1218, analyzed by high-resolution electron impact mass spectrometry (HREIMS), which corresponded to the molecular formula $C_{30}H_{21}NO_9$.[3] In the field-desorption mass spectrum (FDMS),[4] it showed two ions, one for the oxidized (quinone) form (m/z 539) and the other for the reduced (hydroquinone) form (m/z 541),[5] varying in relative intensities. The fast-atom bombardment mass spectrum (FABMS)[6] showed ions for the hydroquinone form at m/z 542.1441[7] [$C_{30}H_{24}NO_9$ $(M + H)^+$] in the positive ion mode and m/z 540 $(M - H)^-$ in the negative ion mode.

The ^1H NMR spectra[8] of fredericamycin A (300 MHz) in DMF-d_7 and in $CDCl_3$ allowed the identification of certain structural characteristics, especially the unsaturated side chain and some of the ring substitution patterns.

The X-ray crystallographic determination[9] yielded the complete structure as (*E,E*)-6′,7′-dihydro-4,9,9′-trihydroxy-6-methoxy-3′-(1,3-pentadienyl)spiro[2*H*-benz[*f*]indene-2,8′-[8*H*]cyclopent-[*g*]isoquinoline]-1,1′,3,5,8(2′*H*)-pentone (**1**). The molecular



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conformation is shown in Figure 1. Although the X-ray intensity data were not of extremely high quality, all hydrogen atoms were found except for those attached to the terminal methyl groups. The bond lengths and hydrogen positions allowed an unequivocal assignment of the structure.

The operation relating the two independent molecules is not a simple 2-fold rotation as a cursory examination of Figure 1 might suggest. The corresponding aromatic portions of the two molecules are approximately parallel, but the moieties with the pentadienyl side chains point in opposite directions. Although in each case the two aromatic portions are nearly at right angles to each other, the conformations of the two molecules are not identical as betokened by the O(9′)–O(1) and O(9′)–O(3) distances, which are

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(7) Analyzed by high-resolution (HR) FABMS.

(8) The ^1H NMR spectrum of fredericamycin A (300 MHz, DMF-d_7) showed signals at δ 1.83 (dd, J = 6.3, 1.2 Hz, =CHCH$_3$), 2.58 (t, J = 7.5 Hz, -CH$_2$-), 3.29 (t, J = 7.5 Hz, -CH$_2$-), 6.04 (dq, J = 15.0, 6.3 Hz, =CH—), 6.29 (ddq, J = 15.0, 10.5, 1.2 Hz, =CH—), 6.35 (d, J = 15.9 Hz, =CH—), 6.75 (d, J = 0.9 Hz, =CH—), 7.06 (s, =CH—), 7.31 (dd, J = 15.9, 10.5, =CH—), 11.61 (d, J = 0.9 Hz, NH), 13.30 (s, OH). A dilute solution of fredericamycin A in $CDCl_3$ showed all the ^1H NMR signals at δ 1.84 (d, J = 6 Hz, =CHCH$_3$), 2.55 (t, J = 6 Hz, -CH$_2$-), 3.32 (t, J = 6 Hz, -CH$_2$-), 4.00 (s, OCH$_3$), 5.98 (m, =CH—), 6.14 (m, =CH—), 6.20 (m, =CH—), 6.30 (s, =CH—), 6.39 (s, =CH—), 6.70 (m, =CH—), 6.90 (s, =CH—), 9.47 (NH), 12.12 (OH), 12.56 (OH), 13.19 (OH).

(9) The crystal used was a red plate obtained from acetonitrile–water, 0.4 × 0.25 × 0.05 mm^3. Crystal data: $C_{30}H_{21}NO_9$; M 539.50; space group $P2_12_12_1$, a = 10.226 (1) Å, b = 20.202 (1) Å, c = 23.736 (2) Å, V 4903.52 Å^3, λ (Cu Kα) = 1.5418 Å, Z = 8; d = 1.46 g mm^{-3}. The phase problem was solved by using negative quartet techniques (Silverton, J. V.; Kabuto, C.; Akiyama, T. **1978**, *Acta Crystallogr., Sect. B. B34*, 588–593) and MULTAN (Main, P.; Hull, S. E.; Lessinger, L.; Germain, G.; Declercq, J. P.; Woolfson, M. M. MULTAN78 **1978**, A system of computer programmes for the automatic solution of crystal structures from X-ray diffraction data, Universities of York and Louvain). The structure was refined, with anisotropic thermal parameters for the heavier atoms and isotropic parameters for the hydrogen atoms, to an R factor of 5.2% by using 2407 reflections with $1 > \sigma(I)$. Refinement programs were from the XRAY72 system (Stewart, J. M.; Kruger, G. J.; Ammon, H. L.; Dickinson, C.; Hall, S. R., XRAY system, version of June 1972, Tech. Report TR-192, University of Maryland).

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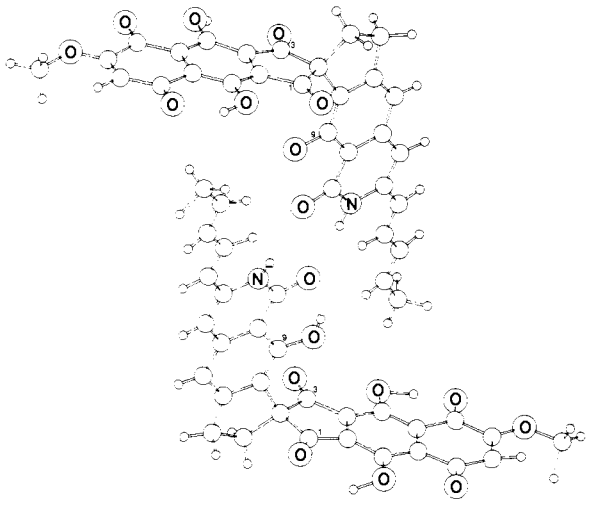

Figure 1. Computer-generated perspective view of the two independent molecules in the X-ray structure of fredericamycin A (**1**).

4.08 and 3.37 Å in one molecule and 3.37 and 3.93 Å in the other. The differences are probably due to packing forces and the quite extensive hydrogen bonding.

The spiro[4,4]nonane system found in fredericamycin A has not been observed in any other types of antibiotics. It imposes certain interesting spacial characteristics on the molecule, which may have an important role in determining its biological activity. The spiro ring system is also very interesting from a biogenetic point of view.

Fredericamycin A has been shown to be a potent antitumor agent. Its activity against glioblastoma cells is comparable to that of 1,3-bis(2-chloroethyl)-1-nitrosourea (BCNU),[2] one of the most potent clinically useful agents. It is also highly cytotoxic[2] against murine leukemias KB, P388, and L1210 cell lines, with ED_{50} values of 7×10^{-1}, 5×10^{-4}, and 2×10^{-4} μg/mL, respectively. It has also been shown to be a powerful inhibitor against ovarian tumor growing in a human tumor cloning system.[10]

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Supplementary Material Available: A diagram of the crystallographic nomenclature of fredericamycin A and tables of interatomic distances, bond angles, atomic parameters, and hydrogen atom positions (6 pages). Ordering information is given on any current masthead page.

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Concerning the Mechanism of Ziegler-Natta Polymerization: Isotope Effects on Propagation Rates

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Ziegler-Natta polymerization, a major industrial organometallic process, is poorly understood at the molecular level. Several sets of molecular descriptions have been proposed for this reaction which differ fundamentally from one another.[1-5] The source of this disagreement is the very mode of carbon-carbon bond formation. Before the more subtle distinctions between and within these sets of mechanisms can be elucidated, this most crucial and elementary aspect of the polymerization process must be understood.

Two of the most clearly defined proposals among the many sets suggested are the carbene-to-metallacycle mechanism of Green and Rooney[3] (Scheme I, a) and the direct four-center olefin insertion mechanism of Cossee and Arlman (Scheme I, b).[1] Neither of these schemes is inconsistent with the known kinetic and stereochemical aspects of the process,[6] and known reactions have been cited as models in justifying each step of both proposals.[3,7-10] The important difference between the two suggestions is the involvement of hydrogen migration in a (Scheme I). This mobility implies a large primary kinetic isotope effect on chain propagation in a and related reactions but no such effect in b. In this paper we report our efforts to determine this isotope effect and conclude that if such an effect exists it is quite small.

Earlier workers have examined[11] the rates of polymerization of C_2D_4 and C_2H_4 and concluded these rates are the same. However, this work allows for k_H/k_D of between 0.7 and 1.4. Since isotope effects on the rate of catalyst generation were also observed even wider variations can not be ruled out.

Recent studies provide values expected for titanocene systems that involve carbenoid intermediates. The abstraction of an α hydrogen by an aluminum alkyl is modeled by the formation of $Cp_2TiCH_2Al(CH_3)_2Cl$ from Cp_2TiCl_2 and $(CH_3)_3Al$.[12] The isotope effect for this reaction is 3. Other related α abstractions fall between 3 and 3.5.[13] Even if the α-hydrogen migration is not a part of the rate-determining step, the reverse of eq 3 provides an expected secondary isotope effect for reactions involving titanium carbene intermediates. The secondary isotope effect determined in these systems is large, ranging from 1.2 to 1.4.[14] Since few models exist for direct insertion into a metal-carbon bond, good values are not available. However, since this reaction does not involve hydrogen migration or major hybridization

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