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Structural Investigations of Mode of Action of Drugs. II. Molecular Structure of Anthramycin Methyl Ether Monohydrate

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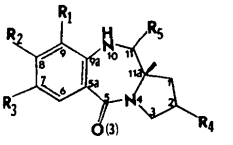
Abstract

The crystal and molecular structure of $C_{17}H_{19}N_3O_4.H_2O$, an antitumor agent, has been determined using X-ray diffraction. The space group is orthorhombic, $P2_12_12_1$. The cell dimensions, measured on a diffractometer, are $a = 7{\cdot}984$ (1), $b = 13{\cdot}495$ (3), $c = 16{\cdot}200$ (3) Å, and $Z = 4$. The structure was solved by direct methods and refined by anisotropic least squares to an R index of 0·050. The 1,4-diazepine ring has a boat conformation while the five-membered pyrrole ring is a flattened envelope. The stereochemistry at C(12) and C(13) is *trans*. There are one intramolecular and five intermolecular hydrogen bonds. One interesting feature is the involvement of the methoxy O atom in hydrogen bonding. A Kendrew-models study indicates that the anthramycin molecule is most probably covalently bound at C(11) to DNA through N(2) of guanine in the wide groove and also O(1) of the anthramycin molecule may be involved in hydrogen bonding to sugar O atoms.

Introduction

Anthramycin, sibiromycin and tomaymycin are structurally related antibiotics produced by various *actinomycetes*. Anthramycin was isolated as a pure substance and its chemical structure was confirmed by synthesis (Leimgruber, Stefanovic, Schenker, Karr & Berger, 1965; Leimgruber, Batcho & Schenker, 1965). The antibiotic has antitumor and antiprotozoal activity. Tomaymycin was first isolated by Arima, Kohsaka, Tamura, Imanaka & Sakai (1972) and the structure was reported by Kariyone, Yazawa & Kohsaka (1971). Sibiromycin was isolated and partially characterized by Gause, Preobrazhenskaya, Ivanitskaya & Sveshnikova (1969), but the complete structure was determined by Mesentsev, Kuljaeva & Rubasheva (1974). The chemical structures of anthramycin, sibiromycin and tomaymycin are shown in Fig. 1. The pyrrole rings have different degrees of unsaturation.



	R_1	R_2	R_3	R_4	R_5
Anthramycin	OH	CH_3	H	$CONH_2$	OH
Anthramycin methyl ether	OH	CH_3	H	$CONH_2$	OCH_3
Sibiromycin	OH	CH_3		CH_3	OH
Tomaymycin	H	OH	OCH_3	$CH\text{-}CH_3$	OH

Fig. 1. The structures of anthramycin methyl ether and related compounds.

In addition to the above antibiotics, three other structurally related antibiotics, dextrochrysin (Aoki, Miyairi, Ajisaka & Sakai, 1969), neothramycin A and B (Takeuchi *et al.*, 1976), have been isolated but no structural information has appeared. Reviews on the mechanism of action of anthramycin (Horwitz, 1971; Kohn, 1975) have appeared. Recently an excellent review by Hurley (1977) on comparative aspects of anthramycin, sibiromycin and tomaymycin has been published. The relative configurations at C(11) and C(11a) for anthramycin (Leimgruber, Stefanovic, Schenker, Karr & Berger, 1965) and tomaymycin (Gause, 1975) have been assigned from their NMR spectra. Crystal structure studies of anthramycin methyl ether were undertaken to elucidate the stereochemistry of this class of antibiotics.

Experimental

Needle-shaped crystals of anthramycin methyl ether were obtained by slow evaporation of a methanol solution from a sample kindly provided by Dr W. E. Scott of Hoffman–La Roche Inc. Preliminary photographs indicated that the crystals belong to the ortho-

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rhombic system, space group $P2_12_12_1$; crystal data are given in Table 1. A needle of 0·1 × 0·1 × 0·3 mm mounted along the *a* axis was used for intensity-data collection on a Syntex $P2_1$ four-circle computer-controlled diffractometer with a graphite monochromator (Mo $K\alpha$, $\lambda = 0{\cdot}71069$ Å) and a pulse-height analyzer. The intensities of 1811 reflections with $2\theta \leq 50{\cdot}0°$ were scanned using the θ–2θ scan technique, a variable scan rate (0·5–29·3° min^{-1}), a scan range of 2·0° and a background to scan ratio of 0·8. 768 reflections $>3\sigma(I)$ were considered observed. The intensities were corrected for Lorentz and polarization effects.

The structure was solved using the direct-methods program *MULTAN* (Germain, Main & Woolfson, 1971) with *E*'s $\geq 1{\cdot}5$. The *E* map revealed all the 24 nonhydrogen atoms, except the water O which was located from a difference map. Isotropic refinement of parameters brought *R* down from 0·238 to 0·109. Further refining of positional and anisotropic thermal parameters reduced *R* to 0·059. At this stage a difference Fourier map revealed all the H atoms. Further refinement with anisotropic thermal parameters for nonhydrogen atoms and isotropic for H atoms brought *R* to 0·050. The refinement was based on F_o, the quantity minimized being $\sum w(F_o - F_c)^2$. The scattering factors used were those of Hanson, Herman, Lea & Skillman (1964).* The weighting scheme used was based on counter statistics as defined by Corfield, Doedens & Ibers (1967); the value of *p* was 0·04.

Results and discussion

The atomic coordinates are given in Table 2. Fig. 2 shows the thermal-ellipsoid plot of the molecule. The deviations of the substituents N(10), C(5), C(16) and O(1) from the least-squares plane through the aromatic ring atoms are −0·02, 0·09, −0·11, and 0·07 Å respectively. The seven-membered diazepine ring adopts a boat conformation with the prow at C(11), as evidenced by the torsion angles (Bucourt, 1974). Going clockwise around the diazepine ring, starting from the C(5a)–C(9a) bond, the angles are −4·0 (8), −9·1 (8), −30·4 (9), 76·5 (7), −69·5 (8), 9·4 (8) and 25·9 (9)° respectively. The boat form is predominant for seven-membered rings which are fused to an aromatic ring. The protons on N(10) and C(11) are equatorial while that on C(11a) is axial. The five-membered ring is an almost flat envelope; the torsion angles going clockwise and starting with the C(1)–C(2) bond are −5·2 (9), 0·3 (8), 5·4 (7), −8·3 (9), and 7·9 (8)° respectively. The side chain at C(2) is almost coplanar with the five-membered ring, the deviations of C(12), C(13) and C(14) being 0·09, −0·03 and 0·12 Å. The stereochemistry of the protons at C(12) and C(13) is *trans*. The exocyclic torsion angles C(9a)–N(10)–C(11)–O(2) and N(4)–C(11a)–C(11)–O(2) have values of 88·6 (7) and −48·1 (8)° respectively. The conformations of the benzodiazepine and pyrrole rings in tomaymycin and sibiromycin should be similar.

The bond lengths and angles in the molecule are given in Table 3. The average standard deviations in the bond lengths and angles are in parentheses. The average values of bond distances and bond angles in the aromatic ring are 1·403 (15) Å and 120·0 (8)°, which are normal. The C(11a)–C(1) length is 1·583 (15) Å, which is surprisingly long, while C(2)–C(12) [1·418 (15) Å] is short. Other bond distances and angles in the diazepine and pyrrole rings are normal.

* Lists of structure factors and thermal parameters have been deposited with the British Library Lending Division as Supplementary Publication No. SUP 34633 (10 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

Table 1. *Crystal data for anthramycin methyl ether*

Formula	$C_{17}H_{19}N_3O_4.H_2O$	V	1745·454 Å^3
M_r	347·34	ρ_c	1·320 Mg m^{-3}
Space group	$P2_12_12_1$	Z	4
		Crystal size	0·1 × 0·1 × 0·3 mm
a	7·984 (1) Å	R	5·0% (758 observed data; Mo $K\alpha$, λ = 0·71069 Å)
b	13·495 (3)		
c	16·200 (3)		

Table 2. *Fractional coordinates* (×10^4) *with standard deviations in parentheses*

	x	*y*	*z*
O(1)	345 (9)	5715 (6)	4331 (4)
O(2)	3731 (10)	5195 (6)	1932 (4)
O(3)	2571 (10)	2151 (5)	2457 (5)
O(4)	10417 (10)	3150 (6)	−82 (5)
O(5)	2700 (9)	1204 (5)	986 (4)
C(1)	6961 (14)	4339 (8)	2537 (7)
C(2)	6889 (14)	3560 (8)	1857 (7)
C(3)	5421 (14)	3058 (7)	1897 (7)
N(4)	4390 (11)	3402 (6)	2548 (6)
C(5)	2963 (14)	2941 (9)	2773 (6)
C(5a)	1874 (15)	3404 (9)	3405 (7)
C(6)	777 (16)	2734 (9)	3805 (7)
C(7)	−405 (16)	3022 (9)	4378 (8)
C(8)	−521 (16)	4035 (9)	4590 (6)
C(9)	467 (16)	4706 (10)	4177 (7)
C(9a)	1725 (14)	4419 (8)	3611 (7)
N(10)	2638 (12)	5211 (6)	3298 (6)
C(11)	4076 (13)	5206 (9)	2784 (7)
C(11a)	5157 (13)	4272 (9)	2941 (6)
C(12)	8141 (15)	3377 (8)	1255 (7)
C(13)	9629 (15)	3767 (8)	1244 (7)
C(14)	10831 (16)	3555 (10)	552 (8)
N(15)	12410 (12)	3825 (7)	668 (5)
C(16)	−1751 (15)	4331 (10)	5252 (7)
C(17)	3132 (19)	6147 (9)	1675 (8)

Table 3. *Bond lengths* (Å) *and angles* (°) *involving non-hydrogen atoms*

C(1)–C(2)	1·524 (14)	C(8)–C(9)	1·374 (17)
C(1)–C(11a)	1·583 (15)	C(8)–C(16)	1·508 (16)
C(2)–C(3)	1·356 (15)	C(9)–O(1)	1·388 (15)
C(2)–C(12)	1·418 (15)	C(9)–C(9a)	1·414 (16)
C(3)–N(4)	1·415 (14)	C(9a)–N(10)	1·389 (14)
N(4)–C(5)	1·349 (14)	N(10)–C(11)	1·417 (14)
N(4)–C(11a)	1·470 (14)	C(11)–O(2)	1·407 (13)
C(5)–O(3)	1·222 (13)	C(11)–C(11a)	1·548 (16)
C(5)–C(5a)	1·481 (15)	C)12)–C(13)	1·301 (16)
C(5a)–C(6)	1·416 (16)	C(13)–C(14)	1·503 (16)
C(5a)–C(9a)	1·415 (16)	C(14)–O(4)	1·212 (15)
C(6)–C(7)	1·379 (17)	C(14)–N(15)	1·326 (14)
C(7)–C(8)	1·414 (17)	C(17)–O(2)	1·433 (14)
C(2)–C(1)–C(11a)	102·9 (8)	C(8)–C(9)–O(1)	121·2 (9)
C(1)–C(2)–C(3)	110·0 (8)	C(8)–C(9)–C(9a)	122·9 (9)
C(1)–C(2)–C(12)	126·3 (9)	O(1)–C(9)–C(9a)	115·8 (8)
C(3)–C(2)–C(12)	123·7 (8)	C(9)–C(9a)–C(5a)	118·5 (8)
C(2)–C(3)–N(4)	112·0 (8)	C(9)–C(9a)–N(10)	113·5 (8)
C(3)–N(4)–C(5)	122·8 (7)	C(5a)–C(9a)–N(10)	127·9 (8)
C(3)–N(4)–C(11a)	110·0 (7)	C(9a)–N(10)–C(11)	129·4 (8)
C(5)–N(4)–C(11a)	127·2 (7)	N(10)–C(11)–O(2)	114·5 (7)
N(4)–C(5)–O(3)	120·3 (7)	N(10)–C(11)–C(11a)	111·1 (7)
N(4)–C(5)–C(5a)	119·2 (8)	O(2)–C(11)–C(11a)	105·1 (7)
O(3)–C(5)–C(5a)	120·5 (8)	C(1)–C(11a)–C(11)	113·2 (8)
C(5)–C(5a)–C(6)	114·2 (8)	C(1)–C(11a)–N(4)	104·3 (7)
C(5)–C(5a)–C(9a)	128·3 (9)	N(4)–C(11a)–C(11)	110·3 (7)
C(6)–C(5a)–C(9a)	117·3 (9)	C(2)–C(12)–C(13)	125·6 (9)
C(5a)–C(6)–C(7)	123·5 (9)	C(12)–C(13)–C(14)	121·1 (9)
C(6)–C(7)–C(8)	118·8 (9)	C(13)–C(14)–O(4)	123·1 (9)
C(7)–C(8)–C(9)	118·8 (9)	C(13)–C(14)–N(15)	116·7 (9)
C(7)–C(8)–C(15)	118·1 (9)	O(4)–C(14)–N(15)	120·2 (8)
C(9)–C(8)–C(16)	123·1 (9)	C(11)–O(2)–C(17)	110·0 (7)

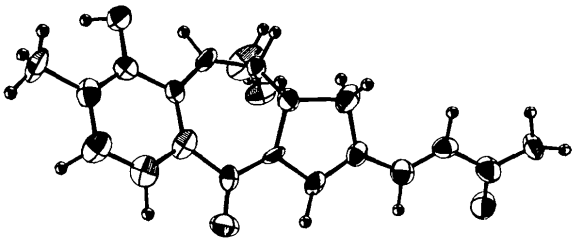

Fig. 2. Thermal-ellipsoid plot of the molecule.

Fig. 3 shows the packing of the molecules in the unit cell. The only intramolecular hydrogen bond is N(10)–H(1)···O(1), 2·57 (1) Å, and the N(10)–H(1)···O(1) angle is 113·3 (0·8)°. The water O(5) atom acts as donor to O(3)(x,y,z) and O(4)($-\frac{1}{2}+x, \frac{1}{2}-y, -z$) with distances of 2·71 (1) and 2·76 (1) Å. The other hydrogen bonds are O(1)–H···O(5)($x, \frac{1}{2}+y, \frac{1}{2}-z$) of 2·57 (1) Å, N(15)–H(1)···O(1)($\frac{3}{2}-x, 1-y, -\frac{1}{2}+z$) of 2·88 (2) Å and N(15)–H(2)···O(2)($1+x, y, z$) of 2·96 (2) Å. The hydrogen-bond distances are within the normal ranges. One interesting feature is that the methoxy O(2) atom acts as acceptor in the hydrogen bond with N(15). Hydrogen bonds involving methoxy O atoms have not been frequently observed. One case is in colchicine (Lessinger & Margulis, 1978; Koerntgen & Margulis, 1977). In view of the rapid exchange of the methoxy group at C(11) with the hydroxyl groups, this type of interaction may have an important bearing on the physiological activities of these substances.

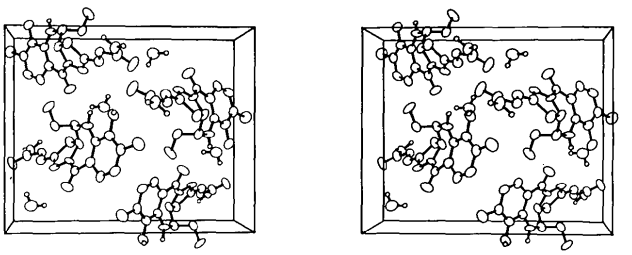

Fig. 3. Stereoscopic diagram of the molecule with the *b* axis vertical and the *c* axis horizontal.

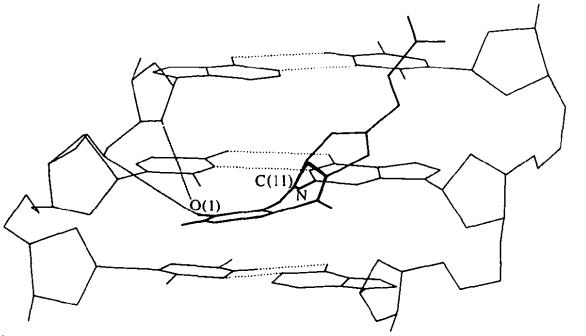

Fig. 4. Possible drug binding to DNA. Three base pairs (AT, CG, TA) are shown

Using the Kendrew models of antibiotics and DNA, an attempt was made to find the covalent binding site of the drug to DNA. The stereochemistry obtained in this study and also information obtained from electric-dichoism studies (Glaubiger *et al.*, 1974) regarding the angle between the chromophore of the drug and the base pairs in DNA were used. The results indicated that the most probable binding sites are C(11) of the drug and N(2) of guanine of DNA in the wide groove. Fig. 4 depicts the above results. The O(1) atom of the drug seems to be involved in bifurcated hydrogen bonding with the O atoms of the sugar and phosphate groups of the backbone. Attempts to crystallize a covalently bound complex of anthramycin and CpG are in progress.

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Détermination de la Structure Cristalline et Moléculaire de l'Octahydro-1,2,3,4,5,6,7,8 Diisopropano-1,4:5,8 Diméthyl-4,5 Phényl-9 Acridine

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Abstract

The crystal structure of 1,2,3,4,5,6,7,8-octahydro-1,4:5,8-diisopropano-4,5-dimethyl-9-phenylacridine, $C_{27}H_{33}N$, M_r = 371·5, has been determined from three-dimensional data collected with Cu $K\alpha$ radiation. The crystals, $[\alpha]_D$ = +269·5°, are monoclinic, space group $P2$, with a = 15·007 (4), b = 10·936 (4), c = 6·966 (2) Å, β = 100·01 (2)°, Z = 2, V = 1125·8 Å^3, d_m = 0·95, d_c = 1·09 Mg m^{-3}. The structure was solved by direct methods with *MULTAN* 77 (R = 8·2% for 955 observed reflexions). This structure shows that during the reaction of ammonia with dibornanonylphenylmethane, ring closure is followed by unexpected dehydrogenation and the formation of a pyridine derivative. On the other hand, a coplanar arrangement of the benzene and pyridine rings is impossible due to steric effects. A twist angle of 51° has been found and π-electronic interaction across the coannular bond is greatly hindered.

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Introduction

L'action de l'ammoniac en solution alcoolique, en présence de chlorure d'ammonium sur le (+)-dibornanonylphénylméthane (Sotiropoulos, 1968) (Fig. 1), préparé à partir du (+)-camphre, conduit par une réaction d'hétérocyclisation à un produit solide dont les

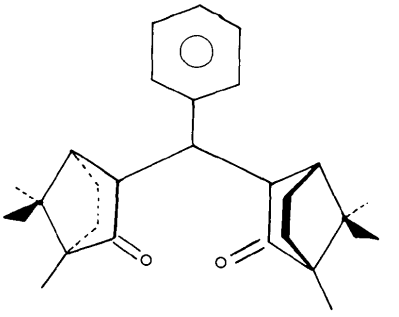

Fig. 1. Dibornanonylphénylméthane.