Acta Cryst. (1983). C**39**, 295–297

Structure of Cytovaricin–Acetonitrile (1:1), $C_{47}H_{80}O_{16}.C_2H_3N$

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(*Received* 9 *August* 1982; *accepted* 19 *October* 1982)

Abstract. $M_r = 942{\cdot}17$, monoclinic, $P2_1$, $a = 13{\cdot}530\ (5)$, $b = 16{\cdot}901\ (4)$, $c = 11{\cdot}632\ (4)$ Å, $\beta = 93{\cdot}35\ (3)°$, $U = 2655\ (2)$ Å^3, $Z = 2$, $D_x = 1{\cdot}178$ Mg m^{-3}, λ(Mo $K\alpha$) $= 0{\cdot}71073$ Å, $\mu = 0{\cdot}081$ mm^{-1}, $T = 296$ K. Final $R = 4{\cdot}9\%$ for 2812 independent reflections. The unit cell contains one acetonitrile molecule per cytovaricin molecule. The skeleton of the molecule consists of a 22-membered macrolide ring, fused to two substituted tetrahydropyrans. One of them is linked to a third tetrahydropyran ring by a spiro junction.

Introduction. Cytovaricin is a novel neutral macrolide antibiotic recently isolated from cultures of *Streptomyces* sp. No. H–230, which resembles *Streptomyces diastatochromogenes* (Kihara, Kusakabe, Nakamura, Sakurai & Isono, 1981). It is extremely toxic to a variety of eukaryotic cells but not to prokaryotic cells. The antibiotic showed lethal effects towards Yoshida sarcoma cells in culture at a concentration as low as 0·005 μg cm^{-3}. Growth of *Chlorella vulgaris* and of some phytopathogenic fungi was also inhibited. Because of the presumed complex nature of the molecule, the single-crystal X-ray analysis was attempted.

Experimental. Single crystals obtained from acetonitrile solution, colourless, 0·3 × 0·4 × 0·4 mm; 12 reflections used for measuring lattice parameters, X-ray diffraction data collected on a Rigaku automated four-circle diffractometer with graphite-monochromatized Mo $K\alpha$ radiation, three standard reflections 400, 092 and $\bar{6}$,11,0 measured at every 150 reflections; within range $2\theta < 45°$ 3051 *hkl* reflections with $|F_o| \geq 3\sigma(F_o)$ measured, *h* $\bar{1}4$–14, *k* 0–18, *l* 0–12, intensities corrected for Lorentz and polarization factors and reduced to 2812 independent reflections, 1019 unobserved reflections in this range; structure solved by a Monte Carlo direct method (Furusaki, 1980), and refined by block-diagonal least squares based on $|F_o|$, unit weight given to all reflections; H(C33)2 and the H atoms of the hydroxyl groups and the solvent molecule not located, all other H atoms obtained by the difference Fourier syntheses, all coordinates and anisotropic temperature factors for non-H atoms and isotropic temperature factors for H atoms refined; $F(000) = 1024$, atomic scattering factors from *International Tables for X-ray Crystallography* (1974), $wR = 5{\cdot}1\%$; absolute configuration was determined by isolation of D-cymarose by acid hydrolysis and methyl β-D-cymaroside by methanolysis (Kihara & Isono, 1982); crystallographic calculations performed on a FACOM 230–75 computer of this Institute using *UNICS* III program system (Sakurai & Kobayashi, 1979).

Discussion. The atomic coordinates and thermal parameters are given in Table 1.*

A stereoscopic drawing of the molecule is shown in Fig. 1. Bond parameters and atom numbering are given in Fig. 2. A terminal methyl group C(34) has an unusually large temperature factor, and the coordinates may not be reliable. All other bond parameters are normal. The skeleton of the molecule consists of a 22-membered unsaturated macrolide ring fused to two substituted tetrahydropyrans. One of them is linked to a third tetrahydropyran ring by a spiro junction and another forms a hemiketal ring. All the six-membered rings have a chair conformation. The 22-membered ring consists of five linear-chain parts, that is C(1)···C(4), C(4)···C(8), C(7)···C(12), C(13)···C(16) and C(16)···C(21). Only one methyl group [C(20–1)] is directed toward the inside of the ring, all other substituents being situated outside. There are two possible intramolecular hydrogen bonds, that is O(5′)···O(7–5) [2·691 (9) Å] and O(17–9)···O(32–12) [2·919 (8) Å].

Among the neutral macrolide antibiotics, oligomycin B (Glehn, Norrestam, Kierkegaard & Maron, 1972) has some structural similarities. It has a 26-membered unsaturated lactone ring and a similar spiroketal ring system. However, oligomycin B lacks a hemiketal ring and a sugar moiety. Venturicidins (Brufani, Cerrini, Fedeli, Musu, Cellai & Keller-Schierlein, 1971) have a 20-membered macrolide ring fused with a hemiketal ring and a sugar side chain. However, they lack a spiroketal ring system. Avermectins (Albers-

* Lists of structure factors, anisotropic thermal parameters and H-atom parameters have been deposited with the British Library Lending Division as Supplementary Publication No. SUP 38194 (22 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

0108-2701/83/020295-03$01.50

Table 1. *Atomic parameters*

Positional parameters are multiplied by 10^4. The invariant parameter is presented without the standard deviation. The equivalent temperature factor is defined by $B_{eq} = \frac{4}{3}\sum_i\sum_j\beta_{ij}(a_i^*a_j^*)$.

	x	y	z	B_{eq}(Å^2)
C(1)	−207 (5)	0	4068 (7)	5·0 (0·3)
C(2)	−479 (6)	−454 (5)	5097 (8)	5·2 (0·3)
C(3)	149 (5)	−598 (5)	5984 (7)	4·5 (0·2)
C(4)	−91 (6)	−1089 (5)	7029 (7)	5·1 (0·3)
C(5)	758 (6)	−1693 (5)	7332 (7)	4·7 (0·2)
C(6)	1196 (5)	−2156 (5)	6320 (7)	4·2 (0·2)
C(7)	2113 (6)	−2594 (5)	6792 (6)	4·4 (0·2)
C(8)	2584 (5)	−3173 (4)	5954 (6)	3·8 (0·2)
C(9)	2956 (5)	−2781 (4)	4838 (6)	4·0 (0·2)
C(10)	3267 (6)	−3380 (5)	3910 (6)	4·2 (0·2)
C(11)	3521 (6)	−2915 (5)	2825 (7)	4·5 (0·2)
C(12)	3843 (7)	−3451 (5)	1830 (7)	5·5 (0·3)
C(13)	4426 (6)	−3007 (5)	916 (7)	5·1 (0·2)
C(14)	3782 (6)	−2435 (5)	228 (6)	4·5 (0·2)
C(15)	4038 (5)	−1691 (4)	18 (6)	3·9 (0·2)
C(16)	3428 (5)	−1133 (4)	−750 (6)	3·5 (0·2)
C(17)	3157 (5)	−372 (4)	−128 (6)	3·7 (0·2)
C(18)	2521 (6)	−572 (5)	897 (6)	4·1 (0·2)
C(19)	2123 (5)	132 (4)	1545 (6)	3·6 (0·2)
C(20)	1529 (5)	−164 (5)	2560 (6)	4·0 (0·2)
C(21)	1171 (5)	574 (5)	3159 (6)	3·8 (0·2)
O(1−1)	700 (3)	337 (3)	4232 (4)	4·1 (0·1)
C(22)	3957 (5)	−924 (4)	−1855 (6)	3·8 (0·2)
C(23)	3312 (5)	−378 (4)	−2603 (6)	3·9 (0·2)
C(24)	3049 (6)	354 (5)	−1934 (7)	4·6 (0·2)
O(19−10)	2947 (3)	614 (3)	1997 (4)	3·6 (0·1)
C(26)	2658 (5)	1318 (4)	2562 (6)	3·8 (0·2)
O(26−11)	2057 (3)	1808 (3)	1834 (4)	3·2 (0·1)
C(30)	2556 (5)	2115 (5)	837 (6)	3·9 (0·2)
C(31)	1764 (6)	2568 (5)	139 (6)	4·5 (0·2)
C(32)	935 (6)	2062 (6)	−381 (7)	5·1 (0·2)
C(33)	150 (7)	2578 (7)	−993 (9)	7·4 (0·3)
C(34)	−721 (10)	2182 (10)	−1462 (14)	14·0 (0·6)
C(1′)	3366 (6)	−4322 (4)	6806 (6)	4·2 (0·2)
C(2′)	4372 (6)	−4704 (6)	6860 (8)	5·8 (0·3)
C(3′)	4248 (6)	−5577 (6)	7201 (9)	6·4 (0·3)
C(4′)	3722 (6)	−5634 (5)	8325 (8)	5·6 (0·3)
C(5′)	2752 (6)	−5172 (5)	8241 (7)	4·6 (0·2)
O(4′)	3526 (4)	−6455 (3)	8548 (5)	5·9 (0·2)
O(1−2)	−714 (4)	50 (5)	3209 (6)	8·5 (0·3)
C(4−1)	−204 (8)	−534 (6)	8065 (8)	6·7 (0·3)
O(4−3)	−1000 (4)	−1510 (4)	6753 (5)	6·1 (0·2)
O(5−4)	332 (4)	−2216 (4)	8130 (5)	6·5 (0·2)
O(7−5)	1871 (5)	−3037 (4)	7787 (5)	6·4 (0·2)
O(9−6)	3756 (4)	−2260 (3)	5162 (5)	5·2 (0·2)
C(10−1)	2455 (7)	−3987 (5)	3644 (7)	5·8 (0·3)
O(10−7)	4138 (4)	−3799 (4)	4328 (5)	6·0 (0·2)
O(17−8)	4034 (3)	36 (3)	263 (4)	4·2 (0·1)
O(17−9)	2563 (3)	130 (3)	−876 (4)	4·1 (0·1)
C(23−1)	3836 (7)	−126 (5)	−3697 (7)	5·6 (0·3)
C(27)	3609 (5)	1723 (5)	3009 (6)	4·1 (0·2)
C(29)	3470 (6)	2601 (5)	1238 (6)	4·4 (0·2)
C(28)	4161 (5)	2073 (5)	1972 (7)	4·6 (0·2)
C(29−1)	3230 (7)	3381 (5)	1856 (8)	5·7 (0·3)
O(32−12)	1279 (4)	1508 (4)	−1214 (5)	6·2 (0·2)
C(6−1)	435 (6)	−2703 (6)	5687 (8)	5·7 (0·3)
C(20−1)	2118 (6)	−727 (5)	3376 (7)	5·2 (0·3)
C(25)	2018 (5)	1101 (4)	3572 (6)	3·8 (0·2)
O(1′)	3448 (4)	−3513 (3)	6561 (4)	4·2 (0·1)
O(3′)	3647 (5)	−5996 (4)	6352 (5)	6·7 (0·2)
C(7′)	4097 (11)	−6108 (7)	5291 (10)	11·0 (0·5)
C(6′)	2237 (9)	−5123 (6)	9369 (8)	8·3 (0·4)
O(5′)	2971 (4)	−4364 (3)	7921 (4)	5·1 (0·2)
N(*S*1)	6148 (6)	−3050 (6)	3798 (8)	8·3 (0·3)
C(*S*2)	6942 (8)	−2846 (6)	3743 (8)	6·6 (0·3)
C(*S*3)	7965 (9)	−2554 (7)	3628 (10)	8·8 (0·4)

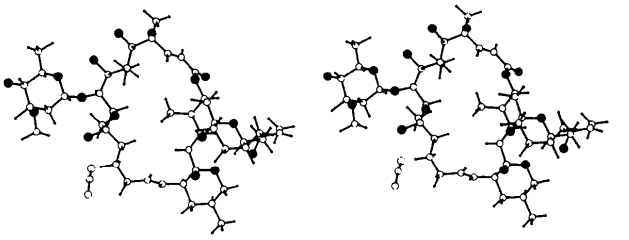

Fig. 1. A stereoscopic drawing of the molecule. The solvent molecule is included.

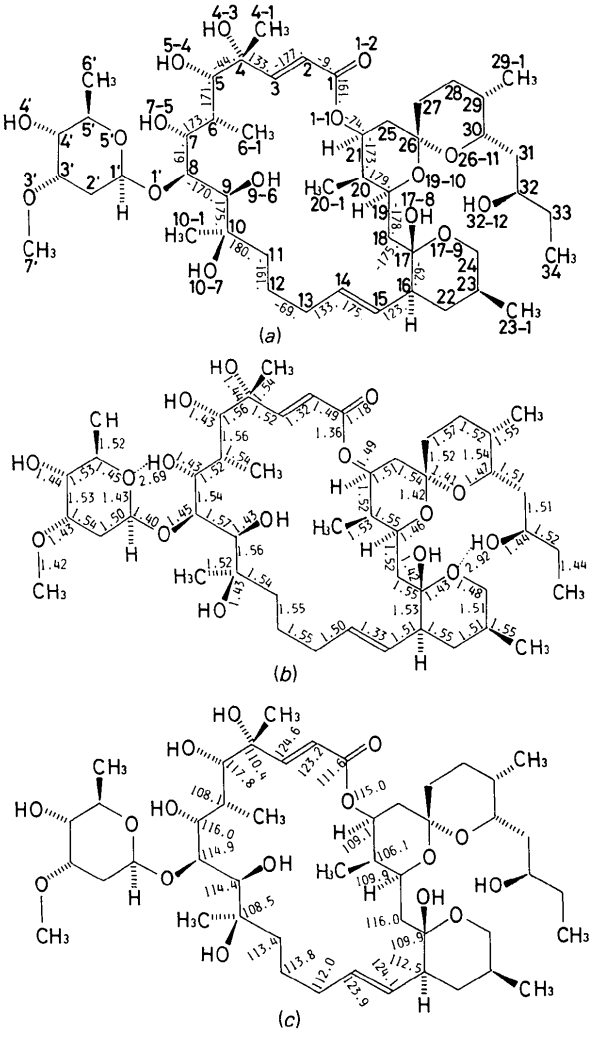

Fig. 2. Schematic structural formula and bond parameters of cytovaricin. (*a*) Torsion angles (°) around the 22-membered ring. The mean standard deviation is 0·7°. (*b*) Bond lengths (Å). The standard deviation is 0·01 Å. (*c*) Bond angles (°). The mean standard deviation is 0·6°.

Schönberg, Arison, Chabala, Douglas, Eskola, Fisher, Lusi, Mrozik, Smith & Tolman, 1981; Springer, Arison, Hirshfield & Hoogsteen, 1981) and milbemycins (Mishima, Kurabayashi, Tamura, Sato, Kuwano & Saito, 1975) are the 16-membered macrolide fused with a similar spiro ketal ring system. In addition, the

avermectins possess a disaccharide side chain. In contrast to L-oleandrose of avermectin, cytovaricin has a β-D-cymarosyl side chain.

The authors express their sincere thanks to Miss K. Kobayashi of this Institute for her technical help.

References

ALBERS-SCHÖNBERG, G., ARISON, B. H., CHABALA, J. C., DOUGLAS, A. W. ESKOLA, P., FISHER, M. H., LUSI, A. MROZIK, H., SMITH, J. L. & TOLMAN, R. L. (1981). *J. Am. Chem. Soc.* **103**, 4216–4221.

BRUFANI, M., CERRINI, S., FEDELI, W., MUSU, C., CELLAI, L. & KELLER-SCHIERLEIN, W. (1971). *Experientia*, **27**, 604–606.

FURUSAKI, A. (1980). *Acta Cryst.* A**35**, 220–224.

GLEHN, M., NORRESTAM, R., KIERKEGAARD, P. & MARON, L. (1972). *FEBS Lett.* **20**, 267–269.

International Tables for X-ray Crystallography (1974). Vol. IV. Birmingham: Kynoch Press.

KIHARA, T. & ISONO, K. (1982). In preparation.

KIHARA, T., KUSAKABE, H., NAKAMURA, G., SAKURAI, T. & ISONO, K. (1981). *J. Antibiot.* **34**, 1073–1074.

MISHIMA, H., KURABAYASHI, M., TAMURA, C., SATO, S., KUWANO, H. & SAITO, A. (1975). *Tetrahedron Lett.* pp. 711–714.

SAKURAI, T. & KOBAYASHI, K. (1979). *Rikagaku Kenkyusho Hokoku*, **55**, 69–77 (in Japanese).

SPRINGER, J. P., ARISON, B. H., HIRSHFIELD, J. M. & HOOGSTEEN, K. (1981). *J. Am. Chem. Soc.* **103**, 4221–4224.

Acta Cryst. (1983). C**39**, 297–300

The Structure of Tetrazole Steroid Analogues. III. Structure of 4,6-Diaza-*A*,*B*-bishomocholest-4a-eno[4,3-*d*][6,7-*d*]bistetrazole (HS-649), $C_{27}H_{42}N_8$*

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(*Received* 29 *June* 1982; *accepted* 25 *October* 1982)

Abstract. $M_r = 478{\cdot}7$, monoclinic, space group $C2$, $a = 35{\cdot}537\ (4)$, $b = 7{\cdot}494\ (1)$, $c = 10{\cdot}319\ (1)$ Å, $\beta = 101{\cdot}25\ (1)°$, $V = 2695{\cdot}2$ Å^3, $Z = 4$, $D_x = 1{\cdot}179$, $D_m = 1{\cdot}143\ (6)$ Mg m^{-3}, λ(Cu $K\alpha$) = 1·54178 Å. Final $R = 0{\cdot}054$ for 2318 observed reflexions. The molecule exhibits a bowing in the α-direction.

Introduction. The bistetrazolo steroids 3,6-diaza-*A*,*B*-bishomocholest-4a-eno[3,4-*d*][6,7-*d*] (HS-650) and 4,6-diaza-*A*,*B*-bishomocholest-4a-eno[4,3-*d*][6,7-*d*] (HS-649) are produced when cholest-4-ene-3,6-dione is treated with excess of hydrazoic acid and boron trifluoride in benzene (Singh & Bhutani, 1978). Our previous paper (Husain, Tickle, Palmer, Singh & Bhutani, 1982) described the crystal and molecular geometry of HS-650 and in the present paper a similar analysis of HS-649 is reported, together with a comparative study of the two.

Experimental. The synthesis has been described by Singh & Bhutani (1978). Good quality tabular, transparent crystals grown from a mixture of acetone and water at room temperature are monoclinic, b axis parallel to the needle axis. Consideration of preliminary X-ray photographs led to assignment of space group $C2$. Intensities and accurate cell parameters were measured on a Hilger & Watts Y290 four-circle diffractometer, Ni-filtered Cu $K\alpha$ radiation, $2\theta < 140°$. A floating window (Tickle, 1975) employing the $\omega/2\theta$ scanning mode was used to measure 5886 reflexions, including two symmetry equivalents. Absorption corrections (North, Phillips & Mathews, 1968; Tickle, 1979) were applied. Data set consisted of 2700 unique reflexions with a merging $R = 0{\cdot}0249$.

Many attempts were made to determine the crystal structure of HS-649 by direct methods using the programs *MULTAN* 78 (Main, Hull, Lessinger, Germain, Declercq & Woolfson, 1978), *YZARC* (Declercq, Germain & Woolfson, 1979) and *SHELX* 76 (Sheldrick, 1976), but all met with failure. The structure was

* Steroids and Related Studies. Part 59.

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0108-2701/83/020297-04$01.50