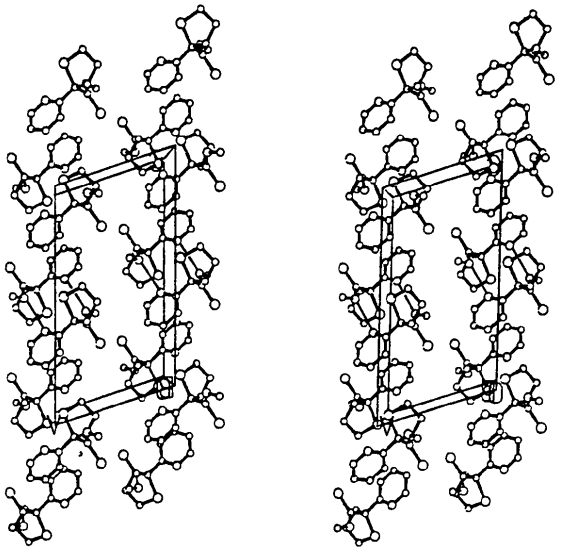

Fig. 2. A stereoscopic view of the crystal packing, viewed along the *b* axis. The hydrogen bonds forming cyclic dimers are shown by thin lines.

judging from its bonding parameters: N(14)···S(11) = 2·921 (2), H(14)···S(11) = 2·31 (3) Å and ∠N(14)—H(14)···S(11) = 117 (2)°. This interaction restricts the rotation around the C(7)—C(12) bond. Thus, the relative orientation of the thioamide group with respect to the pyridine ring appears to be highly fixed.

A stereoscopic view of the crystal packing is shown in Fig. 2. The molecules related by *c*-glide symmetry are arranged along the *c* axis and are stably held by normal van der Waals contacts among the neighboring molecules. N(14) participates in a hydrogen bond with the centrosymmetrically related N(1) [N(14)(x,y,z)···N(1)($-x$, $-y$, $2-z$) = 3·075 (3), H(14)···N(1) = 2·28 (3) Å and ∠N(14)—H(14)···N(1) = 133 (2)°], thus forming a cyclic dimer.

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Vernamycin B_α

BY ISABELLA L. KARLE AND JUDITH L. FLIPPEN-ANDERSON

Laboratory of the Structure of Matter, Naval Research Laboratory, Washington, DC 20375–5000, *USA*

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Abstract. 5-[4-(Dimethylamino)-*N*-methyl-L-phenylalanine]-virginiamycin S_1 monohydrate, 3-HyPic-Thr-D-Abu-Pro-MePheN$(CH_3)_2$-4-oxoPip-PhGly.-O—— H_2O, $C_{45}H_{54}N_8O_{10}.H_2O$, $M_r = 866{\cdot}94 + 18{\cdot}02$, orthorhombic, $C222_1$, $a = 22{\cdot}426\ (6)$, $b = 24{\cdot}043\ (6)$, $c = 19{\cdot}647\ (5)$ Å, $V = 10593{\cdot}4$ Å^3, $Z = 8$, $D_x = 1{\cdot}110$ g cm^{-3}, λ(Cu $K\alpha$) = 1·54178 Å, $\mu = 0{\cdot}63$ mm^{-1}, $F(000) = 3760$, room temperature, $R(F) = 0{\cdot}050$ for 3631 reflections with $|F_o| > 3\sigma$ and 605 parameters refined. The peptide contains both a linear portion and a 19-atom depsipeptide ring with a junction at the threonine residue. The 19-atom backbone ring assumes a cup-like conformation folded around the 3-HyPic residue to form a globular entity with a predominantly hydrophobic surface. The conformation of the molecule is similar to that of virginiamycin (factor S) [Declercq, Germain, Van Meerssche, Hull & Irwin (1978). *Acta Cryst.* B**34**, 3644–3648].

Introduction. An unusual class of antibiotic peptides contains both a cyclic backbone and a linear peptide chain. Although these peptides occur naturally in diverse sources such as a Caribbean tunicate and various fungi, their common feature is a threonine

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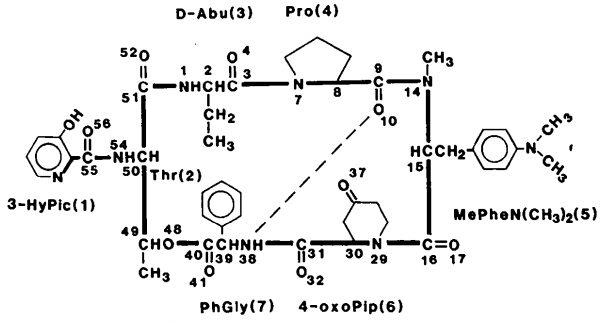
residue, the C^α atom of which acts as the junction between the linear peptide chain and the cyclic depsipeptide ring. The NH of Thr(2) is part of the linear peptide, whereas the OH in the Thr(2) side chain forms an ester bond with the C-terminus carboxyl [PhGly(7)] to form the cyclic part. Vernamycin B_α, an antimicrobial produced by *Streptomyces loidensis* (Bodanszky & Ondetti, 1963), has the following formula:



Five of the seven amino acids do not occur in proteins. Unusual amino acids such as these are often found in peptides produced by microorganisms. In addition, the Abu(3) has the D hand.

Crystal-structure analyses have been reported for virginiamycin (factor S) (Declercq, Germain, van Meerssche, Hull & Irwin, 1978), an antibiotic produced by *Streptomyces virginiae*, having a formula differing from vernamycin B_α only in the lack of the $-N(CH_3)_2$ group on the phenyl ring in residue (5); and for didemnin B (Hossain, van der Helm, Antel, Sheldrick, Sanduja & Weinheimer, 1988), an antiviral and cytotoxic agent, with a sequence quite different from virginiamycin and vermamycin, *R*-MeLeu-Thr-isoSta-Hip-Leu-Pro-Me_2Tyr, but having a similar junction between the linear and cyclic parts of the peptide mediated by the Thr residue.

X-ray data for vernamycin B_α were collected 10 years ago. With the publication of the virginiamycin (factor S) paper (Declercq *et al.*, 1978), the vernamycin B_α analysis was shelved, partly for the lack of appropriate computing facilities. The structure analysis and refinement have now been completed. The conformations of the virginiamycin (factor S) and vernamycin B_α molecules are very similar, although the space group and the packing in the cell are different.

Experimental. Diffraction data were collected from a single crystal sealed in a thin-walled glass capillary with a drop of mother liquor with an automated four-circle Picker diffractometer using Cu $K\alpha$ radiation, a Ni filter, 2° scan, 2° min^{-1} scan speed, θ–2θ mode, $2\theta_{max} = 126°$, and *hkl* ranges with $0 \le h \le 26$, $0 \le k \le 28$, $0 \le l \le 22$. Lorentz and polarization corrections were applied, but not absorption. A total of 3631 unique reflections with $|F_o| \ge 3{\cdot}0\sigma(F)$ were observed.* Excursions in the final difference map were $+0{\cdot}25$ and $-0{\cdot}19$ e $Å^{-3}$, 50 H atoms in calculated positions were riding on C atoms to which they are bonded, four H atoms bonded to O or N were refined isotropically, $R = 0{\cdot}0505$ and $wR = 0{\cdot}0708$ for 3631 reflections where $w = 1/[\sigma^2(|F_o|) + 0{\cdot}00025(F_o)^2]$, $S = 3{\cdot}25$ and maximum shift/e.s.d. = 0·02.

The structure was solved by direct phase determination using the *SHELXTL* (Sheldrick, 1978) package of programs and refined by full-matrix least squares with anisotropic thermal parameters for all C, N and O atoms. Coordinates for the non-hydrogen atoms are listed in Table 1.† There are no unusual bond lengths or bond angles. Torsion angles are shown in Table 2.

Discussion. Two views of vernamycin B_α are shown in Figs. 1 and 2. The 19-membered depsipeptide backbone ring is folded at C(8) and C(39), the C^α atoms of Pro(4) and PhGly(7), into an *L*-shape, as can be seen in Fig. 2. Backbone atoms C(8)C(9)N(14)C(15) and C(30)C(31)N(38)C(39) form one plane with an r.m.s. deviation of 0·17 Å; backbone atoms C(8)N(7)C(3)C(2)N(1)C(51)C(50) C(49)O(48)C(40)C(39) form another plane with an r.m.s. deviation of 0·31 Å. The angle between these two planes defining the fold is 101°. The bent ring is cupped around the linear portion of the peptide, 3-HyPic(1), that joins the ring at C(50), the C^α atom of Thr(2). Six of the amide bonds are in the *trans* conformation. The amide bond for MePheN$(CH_3)_2$(5) is in the *cis* conformation; see C(15)C(16)N(29)C(30) in Fig. 1. There is one hydrogen bond across the 19-membered ring, N(38)H···O(10). This hydrogen bond forms a 10-membered loop as in 4→1 type β-bonds, but includes a *cis* residue. Similar *cis*-containing 4→1 bonds have been observed infrequently, as, for example, in ilamycin B_1 (Iitaka, Nakamura, Takada & Takita, 1974), in cyclic(Gly-Pro-Pro)$_2$ (Czugler, Saśvari & Hollośi, 1982) and in the 1:2 complex of Mg^+ and cyclic(Gly-Pro-Pro)$_2$ (Karle & Karle, 1981). The distinguishing feature of each *cis* loop for

* In the 10 year interim since the data were collected, a number of details concerning crystallization and data collection, and the intensity data $|F_o| < 3{\cdot}0\sigma(F)$ were lost due to several major changes of computer, complete relocation of laboratory and departure of personnel.

† Lists of structure factors, bond lengths and angles, anisotropic thermal parameters and calculated hydrogen positions have been deposited with the British Library Document Supply Centre as Supplementary Publication No. SUP 52304 (19 pp.). Copies may be obtained through The Technical Editor, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

Table 1. *Atomic coordinates and equivalent isotropic displacement parameters* (Å^2)

	x	y	z	U_{eq}
N(1)	0·6618 (2)	0·6559 (2)	0·8888 (2)	0·072 (1)
C(2)	0·6693 (2)	0·6703 (2)	0·8169 (2)	0·071 (1)
C(3)	0·7346 (2)	0·6864 (2)	0·8087 (2)	0·070 (2)
O(4)	0·7727 (2)	0·6495 (2)	0·8050 (2)	0·089 (1)
C(5)	0·6526 (3)	0·6221 (2)	0·7712 (3)	0·090 (2)
C(6)	0·5891 (3)	0·6038 (3)	0·7794 (5)	0·123 (3)
N(7)	0·7502 (2)	0·7400 (2)	0·8099 (2)	0·065 (1)
C(8)	0·8132 (2)	0·7542 (2)	0·8095 (2)	0·073 (2)
C(9)	0·8426 (2)	0·7325 (2)	0·8751 (2)	0·074 (2)
O(10)	0·8122 (1)	0·7280 (2)	0·9275 (1)	0·080 (1)
C(11)	0·8127 (3)	0·8178 (3)	0·8074 (3)	0·096 (2)
C(12)	0·7522 (3)	0·8335 (3)	0·7795 (3)	0·102 (2)
C(13)	0·7124 (2)	0·7891 (2)	0·8111 (3)	0·083 (2)
N(14)	0·9007 (2)	0·7207 (2)	0·8748 (2)	0·080 (1)
C(15)	0·9265 (2)	0·7005 (2)	0·9396 (2)	0·070 (2)
C(16)	0·9426 (2)	0·7504 (2)	0·9827 (2)	0·067 (1)
O(17)	0·9804 (2)	0·7825 (2)	0·9604 (2)	0·089 (1)
C(18)	0·9408 (3)	0·7342 (4)	0·8176 (3)	0·115 (3)
C(19)	0·9810 (2)	0·6629 (3)	0·9295 (2)	0·086 (2)
C(20)	0·9999 (2)	0·6349 (2)	0·9945 (3)	0·074 (2)
C(21)	1·0350 (2)	0·6606 (2)	1·0430 (3)	0·074 (2)
C(22)	1·0523 (2)	0·6346 (2)	1·1018 (3)	0·076 (2)
C(23)	1·0361 (2)	0·5808 (2)	1·1148 (3)	0·088 (2)
C(24)	0·9998 (4)	0·5546 (2)	1·0674 (4)	0·120 (3)
C(25)	0·9822 (3)	0·5826 (3)	1·0083 (4)	0·111 (3)
N(26)	1·0555 (3)	0·5515 (2)	1·1726 (3)	0·121 (2)
C(27)	1·0535 (5)	0·4937 (3)	1·1739 (6)	0·162 (5)
C(28)	1·0859 (5)	0·5811 (4)	1·2256 (5)	0·147 (4)
N(29)	0·9181 (2)	0·7582 (2)	1·0452 (2)	0·065 (1)
C(30)	0·8720 (2)	0·7240 (2)	1·0781 (2)	0·061 (1)
C(31)	0·8211 (2)	0·7611 (2)	1·1015 (2)	0·065 (1)
O(32)	0·8116 (2)	0·7697 (2)	1·1615 (2)	0·097 (1)
C(33)	0·9419 (2)	0·8015 (2)	1·0897 (2)	0·074 (2)
C(34)	0·9688 (2)	0·7754 (2)	1·1536 (2)	0·075 (2)
C(35)	0·9314 (2)	0·7326 (2)	1·1860 (2)	0·069 (2)
C(36)	0·8994 (2)	0·6934 (2)	1·1389 (2)	0·066 (1)
O(37)	0·9289 (2)	0·7270 (2)	1·2476 (2)	0·092 (1)
N(38)	0·7896 (2)	0·7854 (2)	1·0530 (2)	0·078 (1)
C(39)	0·7426 (2)	0·8243 (3)	1·0671 (3)	0·086 (2)
C(40)	0·6824 (2)	0·7968 (3)	1·0769 (3)	0·086 (2)
O(41)	0·6402 (2)	0·8201 (2)	1·1030 (3)	0·119 (2)
C(42)	0·7423 (3)	0·8717 (3)	1·0139 (3)	0·096 (2)
C(43)	0·7946 (4)	0·8931 (4)	0·9927 (5)	0·140 (4)
C(44)	0·7980 (6)	0·9361 (5)	0·9477 (7)	0·173 (6)
C(45)	0·7482 (7)	0·9569 (4)	0·9193 (6)	0·161 (5)
C(46)	0·6967 (6)	0·9354 (5)	0·9382 (7)	0·186 (6)
C(47)	0·6934 (4)	0·8921 (4)	0·9852 (6)	0·150 (4)
O(48)	0·6810 (1)	0·7451 (2)	1·0531 (2)	0·078 (1)
C(49)	0·6251 (2)	0·7153 (2)	1·0574 (2)	0·077 (2)
C(50)	0·6324 (2)	0·6670 (2)	1·0078 (2)	0·074 (2)
C(51)	0·6346 (2)	0·6888 (2)	0·9334 (2)	0·070 (2)
O(52)	0·6106 (2)	0·7320 (2)	0·9184 (2)	0·103 (2)
C(53)	0·6139 (3)	0·6947 (4)	1·1294 (3)	0·109 (3)
N(54)	0·6828 (2)	0·6325 (2)	1·0246 (2)	0·074 (1)
C(55)	0·6779 (3)	0·5819 (2)	1·0501 (3)	0·081 (2)
O(56)	0·6297 (2)	0·5571 (2)	1·0555 (2)	0·109 (2)
C(57)	0·7344 (3)	0·5542 (2)	1·0706 (3)	0·082 (2)
N(58)	0·7849 (2)	0·5845 (2)	1·0666 (3)	0·093 (2)
C(59)	0·8345 (3)	0·5597 (3)	1·0857 (5)	0·118 (3)
C(60)	0·8371 (3)	0·5060 (3)	1·1095 (5)	0·129 (3)
C(61)	0·7872 (3)	0·4750 (3)	1·1135 (4)	0·114 (3)
C(62)	0·7335 (3)	0·4992 (3)	1·0939 (3)	0·094 (2)
O(63)	0·6832 (2)	0·4693 (2)	1·0968 (3)	0·121 (2)
W(1)	0·8790 (5)	0·6197 (4)	0·7396 (7)	0·263 (6)

Equivalent isotropic U is defined as one-third of the trace of the orthogonalized U_{ij} tensor.

Table 2. *Torsional angles* (°)

	3-HyPic 1	Thr 2	D-Abu 3	Pro 4	MePheN-$(CH_3)_2$ 5	4-oxoPip 6	PhGly 7
φ(N—C$^\alpha$)	179	−127	105	−64	−85	−128	−88
ψ(C$^\alpha$—C′)	5	30	−97	154	120	67	−17*
ω(C′—N)	−174	−178	173	179	−4	−176	−178†
							−68‡
χ^1(C$^\alpha$—C$^\beta$)	−179	58§	60	−22	−170	52	−41
		−62¶					138

* With O$^\gamma$ of Thr(2).
† With C$^\beta$O$^\gamma$ of Thr(2).
‡ C′(2)C$^\alpha$(2)C$^\beta$(2)O$^\gamma$(2) or C(51)C(50)C(49)O(48).
§ N(54)C(50)C(49)O(48).
¶ N(54)C(50)C(49)C(53).

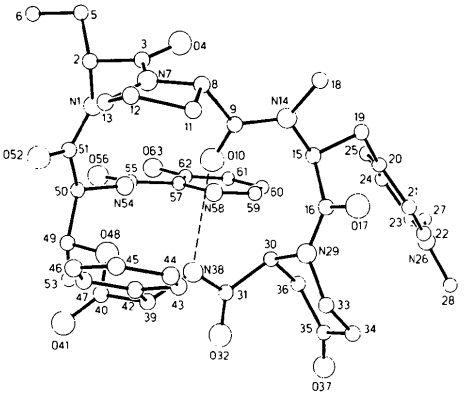


Fig. 1. A view of vernamycin B_α with all the C, N and O atoms numbered. H atoms are omitted. The dashed line indicates the intra-ring hydrogen bond.

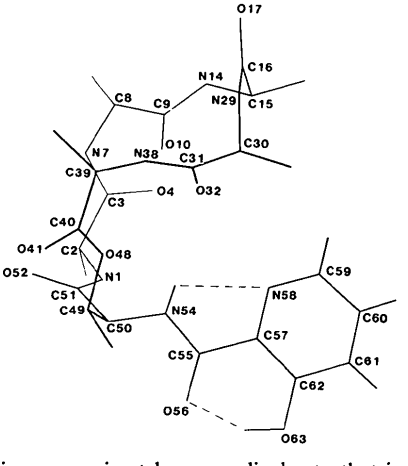


Fig. 2. A view approximately perpendicular to that in Fig. 1. In the cyclic portion, only the backbone atoms and the C$^\beta$ atoms of the side chains are shown. The view is rotated away somewhat from a projection showing the *L*-shaped bend in the backbone ring since too many atoms would be superimposed. Dashed lines indicate possible hydrogen bonds.

the hydrogen bond is the occurrence of one or two amide nitrogens with substituents such as methyl, prolyl or 4-oxopipecolyl, in virginiamycin B_1 as well as the present case.

The N(1) moiety, the only other N atom with a proton in the depsipeptide ring, participates in an intermolecular hydrogen bond to O(63) in a neighboring molecule. The other intramolecular hydrogen bonds occur in the 3-HyPic(1) residue and the adjacent Thr(2) residue. They are O(63)H···O(56) and possibly N(54)H···N(58); see Fig. 2 and Table 3. The 'linear' portion of the peptide, atoms C(50) and N(54) to O(63), is planar to within ±0·020 Å, except

Table 3. *Hydrogen bonds*

	Donor(*D*)	Acceptor(*A*)	*D*—*A*(Å)	H—*A*(Å)	Angle *D*—H—*A*(°)
Intramolecular	N(38)	O(10)	2·87	2·14	176
	N(54)	N(58)	2·69	2·27	116
	O(63)	O(56)	2·56	1·56	148
Intermolecular	N(1)	O(63) $(x, 1-y, 2-z)$	3·06	2·30	147
	W(1)	O(4)	2·80		
	W(1)	O(41) $(1\frac{1}{2}-x, 1\frac{1}{2}-y, -\frac{1}{2}+z)$	3·08		

for N(54), O(56) and O(63) with deviations from the plane of −0·079, +0·050 and −0·038 Å, respectively. The cocrystallized molecule of H_2O provides protons for hydrogen bonds to O(4) and O(41), carbonyl oxygens in neighboring peptide molecules (see Figs. 3 and 4).

The overall shape of the molecule is globular, as shown by the space-filling diagram in Fig. 3 in which the parallel stacking of the pyrrolidine ring in Pro(4) and the phenyl ring in PhGly(7) is visible at the left. However, the closest distances between *C*-ring atoms in Pro(4) and *C*-ring atoms in PhGly(7) are near 4·0 Å. The surface is mainly hydrophobic except for the carbonyl oxygens which, with one exception, are directed outward and constitute the polar areas.

The conformation of the vernamycin molecule is very similar to that of virginiamycin (Declercq *et al.*, 1978) which lacks the $-N(CH_3)_2$ moiety on the MePhe(5) residue. The values for the φ torsional angles (C′—N—C^α—C′) in the two molecules are within 5°, except for the Pro(4) residue where the difference is 9°; and the values for the ψ angles are also very similar with the largest difference being 14° for N(1)—C(2)—C(3)—N(7). The largest torsional differences occur in the residues that interact with the H_2O molecule present in the vernamycin crystal but absent in the virginiamycin crystal. The signs for all the torsion angles in the virginiamycin paper should be reversed to conform with the convention adopted by the IUPAC–IUB Commission on Biochemical Nomenclature (1970).

The didemnin B molecule (Hossain *et al.*, 1988) is similar to vernamycin in the sense that a Thr residue occurs at the junction between the linear peptide chain and the cyclic depsipeptide portion. The residues are mostly quite different in didemnin B and vernamycin B_α. The large depsipeptide rings in each have a cup-like fold with a mostly hydrophobic exterior except for the carbonyl oxygens which are directed outward in each. There are significant differences in the torsion angles of the Thr residue. For didemnin, φ, ψ and χ' are −69, 156 and −155°, whereas for vernamycin they are −126, 30 and 58°, respectively. The differences in these values are correlated with the compact globular shape of vernamycin in which the linear peptide portion is folded against the cupped cyclic portion, whereas in didemnin the linear peptide portion extends away from the large ring for a less compact structure.

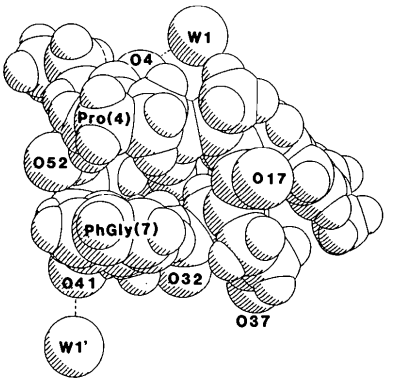


Fig. 3. A space-filling diagram of the molecule as oriented in Fig. 1. The overall shape is globular with a predominantly hydrophobic surface. Carbonyl oxygens directed outward provide the scattered polar areas on the surface. Hydrogen bonding to the water molecule and its symmetry equivalent is indicated.

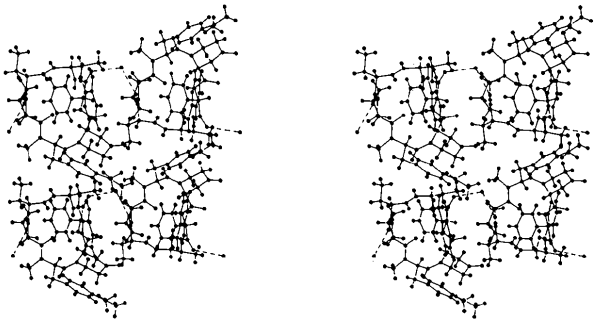


Fig. 4. Stereodiagram of four of the eight molecules in the unit cell. The peptide molecules are joined head-to-tail by means of hydrogen bonds to an intervening water molecule (dashed lines) along a horizontal screw axis. The axial directions are **a**↑, **c**→ and **b** directed out of the page.

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