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The Crystal and Molecular Structures of Two Polymorphic Crystalline Forms of Virazole (1-β-D-Ribofuranosyl-1,2,4-triazole-3-carboxamide). A New Synthetic Broad Spectrum Antiviral Agent

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The crystal structures of two polymorphic forms of virazole, a new synthetic broad spectrum antiviral agent, have been determined. The five-membered triazole base ring of virazole makes this compound unique among the nucleoside antibiotics. Both forms crystallize in the orthorhombic system, space group $P2_12_12_1$ with $a=14{\cdot}863$, $b=7{\cdot}512$, $c=8{\cdot}788$ Å and $a=25{\cdot}034$, $b=7{\cdot}719$, $c=5{\cdot}289$ Å for crystal form V1 and crystal form V2 respectively. The structures were solved by direct methods and refined to R indices of 0·050 (V1) and 0·036 (V2) using respectively 940 and 915 intensities measured on a diffractometer. The conformations of the molecules are different in the two crystals. In V1 the glycosyl torsion, the sugar pucker (pseudorotation phase angle **P**) and the exocyclic C(4′)–C(5′) bond torsion are respectively *anti* ($\chi=10{\cdot}4°$), ${}^3T_2$ ($\mathbf{P}=11{\cdot}7°$) and *gauche*$^+$ (g^+) while in V2 they are 'high *anti*' ($\chi=119{\cdot}0°$), ${}_2T^1$ ($\mathbf{P}=335{\cdot}8$ or $-24{\cdot}2°$) and *trans* (t). The 2′-*exo* puckering observed for V2 is uncommon for β-nucleosides. The carboxamide group in V2 is engaged in hydrogen bonding to the base ring of a symmetry-related molecule whereas there is no interbase hydrogen bonding in V1. The usual hydrogen-bonding sites of the base ring in V1 are involved in hydrogen bonding, while the N(2) site of the base in V2 is not hydrogen bonded. Interestingly, in both structures one of the amino hydrogen atoms is not engaged in hydrogen bonding.

Introduction

Virazole (Fig. 1) is a new synthetic broad spectrum antiviral agent and belongs to a class of nucleoside antibiotic structures containing a five-membered triazole ring (Sidwell, Huffman, Khare, Allen, Witowski & Robins, 1972). Virazole crystallizes in two polymorphic modifications which exhibit differences in their KBr infrared spectra and melting points (Robins, 1972) suggesting differences in their hydrogen bonding and crystal packing schemes. X-ray structure analysis of the two polymorphic forms of virazole was undertaken to unequivocally establish the molecular structure and conformation and to elucidate the differences in the properties of the polymorphs in terms of differences in their packing patterns. It is also of interest to compare the structure of virazole with showdomycin (Tsukuda & Koyama, 1970) and pyrazomycin (Jones & Chaney, 1972), the two other known five-membered base nucleoside antiviral agents and antibiotics. A preliminary communication of this work has been presented elsewhere (Prusiner & Sundaralingam, 1973).

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Experimental section

Crystals of both polymorphic forms of virazole (V1 and V2) were kindly supplied by Dr Roland K. Robins of the Nucleic Acid Research Center, Irvine, California. The pertinent crystal data are given in Table 1. The cell constants were determined by a least-squares refinement of the goniostat angles 2θ, ω, and χ of 12 reflections measured in the 2θ range of 40–60° on a diffractometer.

Three-dimensional monochromatized X-ray intensity data were collected for V1 on an automatic FACS-1 Picker diffractometer using Cu radiation whereas for V2 no monochromator was used. The θ–2θ scan mode was employed with a scan rate of 2° min^{-1}. The data were corrected for Lorentz and polarization effects, but no absorption corrections

Table 1. *Crystal data for virazole 1 (V1) and virazole 2 (V2)*

	V1	V2
Formula	$C_8N_4O_5H_{12}$	$C_8N_4O_5H_{12}$
a	14·863 (4) Å	25·034 (8) Å
b	7·512 (1)	7·719 (2)
c	8·788 (2)	5·289 (1)
Space group	$P2_12_12_1$	$P2_12_12_1$
D_{obs}	1·65 g cm^{-3} (by flotation in $CHCl_3$–$CHBr_3$ mixture)	1·58 g cm^{-3} (by flotation in CCl_4–$CHBr_3$ mixture)
D_{calc}	1·652 g cm^{-3}	1·585 g cm^{-3}
Z	4	4
μ	12·09 cm^{-1}	11·58 cm^{-1}

were made. Reflections with intensities greater than $1{\cdot}5\sigma$ were considered as observed and there were 940 reflections for V1 and 915 for V2 out of totals of 958 and 974, respectively.

Structure determination

The structure of V1 was solved using direct methods (Karle & Karle, 1966). The phases of 137 reflections with $|E|$'s > 1·40 were refined using the tangent formula in the program X-RAY 70 (Stewart, Kundell & Baldwin, 1970). The $R(E)$ value was 0·17, where $R(E)=\sum||E_{obs}|-|E_{calc}||/\sum|E_{obs}|$, and $|E_{obs}|$ and $|E_{calc}|$ are the observed and calculated normalized structure factors, respectively. The three-dimensional E map based on the above phases revealed all the 17 nonhydrogen atoms of the nucleoside among the 17 largest peaks.

V2 was also solved by direct methods using the multiple solution tangent formula method (Main, Woolfson & Germain, 1971). Four reflections served to define both the origin and enantiomorph and the phase of another reflection was derived from $\sum_1$ relationships. The tangent formula was then used to extend this phase information to 231 $|E|$ values greater than 1·3. Altogether, four phase angle sets were obtained, one of which, according to consistency criteria, contained the correct solution (figure of merit = 1·17 and residual index = 34·2). An E map generated with the latter set clearly revealed the 17 nonhydrogen atoms among the 17 strongest peaks.

Refinement of the structure

The refinement of the structures was carried out on a Univac 1108 computer using the full-matrix least-squares program of Busing, Martin & Levy (1962) modified by Rao (1968). Weights from counting statistics were applied at first in both instances. V1 was first subjected to two cycles of refinement with iso-

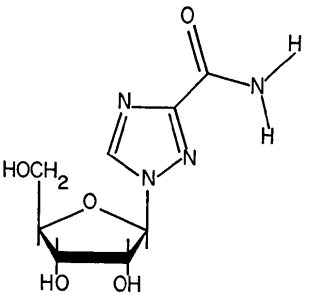

Fig. 1. Chemical structure for virazole.

Table 2. *Positional and thermal parameters of atoms in virazole (V1)*

Positional parameters of nonhydrogen atoms have been multiplied by 10^4. Positional parameters of hydrogen atoms have been multiplied by 10^3. Anisotropic thermal parameters have been multiplied by 10^4. Anisotropic temperature factor is of the form $\exp[-(\beta_{11}h^2+\ldots+2\beta_{12}hk+\ldots)]$. Standard deviations refer to the least significant digits.

	x	y	z	β_{11}	β_{22}	β_{33}	β_{12}	β_{13}	β_{23}
N(1)	5935 (2)	9890 (5)	3297 (4)	12 (1)	51 (6)	56 (4)	8 (2)	2 (2)	−5 (5)
N(2)	5758 (2)	11525 (5)	3915 (4)	19 (2)	49 (6)	53 (4)	−3 (2)	4 (2)	5 (5)
C(3)	5132 (3)	11129 (6)	4929 (5)	12 (2)	75 (7)	52 (6)	0 (3)	−4 (3)	4 (6)
N(4)	4889 (2)	9385 (5)	4990 (5)	20 (2)	53 (6)	71 (5)	4 (3)	10 (3)	9 (5)
C(5)	5421 (3)	8657 (6)	3941 (6)	17 (2)	63 (7)	76 (6)	−4 (3)	7 (3)	−16 (7)
C(6)	4720 (3)	12496 (6)	5934 (6)	13 (2)	92 (8)	62 (6)	10 (3)	−5 (3)	1 (6)
O(7)	4149 (2)	12041 (5)	6878 (4)	32 (2)	120 (6)	83 (5)	19 (3)	28 (2)	20 (5)
N(8)	4996 (3)	14163 (5)	5746 (5)	31 (2)	69 (7)	98 (6)	8 (3)	9 (3)	−16 (6)
C(1′)	6630 (3)	9742 (6)	2106 (5)	16 (2)	48 (7)	45 (5)	0 (3)	4 (3)	−1 (5)
O(1′)	6684 (2)	8033 (4)	1494 (3)	19 (1)	72 (5)	57 (4)	10 (2)	−9 (2)	−20 (4)
C(2′)	7574 (3)	9993 (7)	2763 (5)	11 (2)	65 (7)	41 (4)	−1 (3)	7 (2)	3 (6)
O(2′)	8109 (2)	10686 (4)	1552 (3)	19 (1)	67 (5)	57 (3)	−12 (2)	7 (2)	4 (4)
C(3′)	7829 (3)	8081 (6)	3144 (5)	15 (2)	51 (7)	40 (5)	−4 (3)	−2 (3)	−2 (5)
O(3′)	8779 (2)	7927 (4)	3231 (4)	15 (1)	80 (5)	119 (5)	0 (3)	−7 (2)	20 (5)
C(4′)	7420 (3)	7073 (6)	1815 (5)	12 (2)	52 (7)	56 (6)	8 (3)	−2 (3)	−11 (6)
C(5′)	7194 (3)	5133 (6)	2051 (6)	27 (2)	61 (8)	85 (6)	−2 (4)	9 (3)	−19 (7)
O(5′)	6613 (3)	4928 (5)	3343 (4)	49 (2)	87 (6)	103 (5)	−28 (3)	22 (3)	−16 (5)

	x	y	z	B		x	y	z	B
H(5)	542	750	373	2·0	H(3′)	749	790	386	2·0
H(81)	540	1457	550	2·5	H(O3′)	933	675	361	3·0
H(82)	462	1488	630	2·5	H(4′)	792	742	133	2·0
H(1′)	642	1076	158	2·0	H(51′)	692	469	116	2·0
H(2′)	760	1064	373	2·0	H(52′)	781	421	220	2·0
H(O2′)	842	1142	177	3·0	H(O5′)	644	411	309	3·0

tropic temperature factors for the atoms followed by two cycles with anisotropic temperature factors. This lowered the residual index R $(=\sum||F_{obs}|-|F_{calc}||/\sum|F_{obs}|$, where $|F_{obs}|$ and $|F_{calc}|$ are the observed and calculated structure factors, respectively) from an initial value of 0·23 to 0·086. A difference electron density map at this stage revealed four of the 12 hydrogen atoms. The weighting scheme at this point was replaced by the empirical scheme $1/\sqrt{w}=0{\cdot}035|F_{obs}|+18{\cdot}30$ which was based on the error curve $||F_{obs}|-|F_{calc}||$ *versus* $|F_{obs}|$. Two more cycles of anisotropic refinement reduced the R value to 0·05 for 940 observed reflections. The final shift/σ was less than 0·3 for all nonhydrogen parameters. A difference Fourier map clearly revealed the positional parameters of all the remaining hydrogen atoms. The hydrogen atoms were not refined in this structure.

For V2, two cycles of isotropic followed by two cycles of anisotropic refinement of the nonhydrogen atoms reduced R from 0·19 to 0·07. All 12 hydrogen atoms were determined from a difference electron density map. At this stage an empirical weighting scheme where $1/\sqrt{w}=-0{\cdot}069|F_{obs}|+4{\cdot}05$ for $|F_{obs}|<20{\cdot}0$, $1/\sqrt{w}=2{\cdot}57$ for $20{\cdot}0<|F_{obs}|<51{\cdot}0$, and $1/\sqrt{w}=0{\cdot}013|F_{obs}|+1{\cdot}93$ for $|F_{obs}|>51{\cdot}0$. Two additional least-squares cycles with anisotropic temperature factors for nonhydrogen atoms and isotropic temperature factors for hydrogen atoms lowered R to 0·036 for the 916 observed reflections. The final shift/σ was less than 0·30 for the nonhydrogen atom parameters and less than 0·50 for hydrogen atoms.

The scattering factors used for O, N and C atoms were from Cromer & Waber (1965) and for H atoms from Stewart, Davidson & Simpson (1965).

Results and discussion

The final positional and thermal parameters for nonhydrogen and hydrogen atoms are given in Tables 2 and 3 for V1 and V2, respectively.* The bond distances and bond angles involving nonhydrogen atoms are shown in Figs. 2 and 3 for V1 and V2 respectively. The distances and angles involving hydrogen atoms are in the usual ranges.

Molecular conformation and geometry

The glycosyl bond

Fig. 4(*a*) and (*b*) are *ORTEP* drawings (Johnson, 1965) of V1 and V2 respectively, showing the thermal ellipsoids of the atoms with 50% probability surfaces. Their molecular conformations are different in some important respects. V1 exhibits a glycosyl torsion angle χ[O(1′)–C(1′)–N(1)–C(5)] of 10·4° which falls within the range of angles observed for the common pyrimidine and purine nucleosides and nucleotides. In contrast, V2 exhibits a χ value of 119·0° which strictly is in the *syn* region according to the definition (Sundaralingam, 1969, 1973), although it is much lower

* A list of structure factors has been deposited with the British Library Lending Division as Supplementary Publication No. SUP 31210 (9 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 13 White Friars, Chester CH1 1NZ, England.

Table 3. *Positional and thermal parameters of atoms in virazole (V2)*

Positional parameters of nonhydrogen atoms have been multiplied by 10^4. Positional parameters of hydrogen atoms have been multiplied by 10^3. Anisotropic thermal parameters have been multiplied by 10^4. Anisotropic temperature factor is of the form $\exp[-(\beta_{11}h^2+\ldots+2\beta_{12}hk+\ldots)]$. Standard deviations refer to the least significant digits.

	x	y	z	β_{11}	β_{22}	β_{33}	β_{12}	β_{13}	β_{23}
N(1)	3230 (1)	3836 (4)	5180 (5)	6 (0)	105 (5)	149 (10)	−4 (1)	−4 (2)	−1 (7)
N(2)	2828 (1)	3394 (4)	3578 (5)	7 (0)	129 (6)	141 (9)	−5 (1)	−2 (2)	−2 (7)
C(3)	2496 (1)	2575 (5)	5067 (7)	7 (0)	104 (6)	165 (12)	0 (1)	1 (2)	−11 (8)
N(4)	2648 (1)	2473 (4)	7537 (6)	9 (0)	164 (6)	158 (11)	−9 (1)	0 (2)	18 (8)
C(5)	3113 (1)	3276 (5)	7514 (6)	9 (1)	157 (7)	112 (12)	−4 (2)	−1 (2)	14 (9)
C(6)	1974 (1)	1862 (5)	4199 (7)	10 (1)	111 (7)	182 (13)	−4 (2)	5 (3)	4 (8)
O(7)	1627 (1)	1474 (4)	5754 (5)	11 (0)	279 (7)	210 (9)	−26 (1)	10 (2)	−18 (8)
N(8)	1909 (1)	1746 (5)	1724 (6)	9 (1)	159 (6)	176 (11)	−11 (2)	−4 (2)	7 (8)
C(1′)	3688 (1)	4817 (4)	4116 (6)	7 (1)	91 (6)	139 (12)	−2 (1)	−1 (2)	10 (8)
O(1′)	3942 (1)	3794 (3)	2265 (4)	7 (0)	128 (5)	135 (8)	6 (1)	−6 (2)	−14 (6)
C(2′)	4115 (1)	5234 (4)	6096 (6)	7 (1)	97 (6)	120 (11)	−2 (1)	−1 (2)	−6 (8)
O(2′)	4406 (1)	6633 (3)	5038 (5)	12 (0)	95 (4)	242 (9)	−12 (1)	−21 (2)	21 (6)
C(3′)	4439 (1)	3544 (5)	6105 (6)	6 (1)	96 (6)	146 (12)	−3 (2)	−9 (2)	11 (8)
O(3′)	4968 (1)	3738 (3)	7010 (4)	8 (0)	126 (5)	204 (10)	−3 (1)	−16 (2)	44 (6)
C(4′)	4398 (1)	2901 (4)	3363 (6)	6 (1)	94 (6)	129 (12)	0 (1)	−5 (2)	8 (7)
C(5′)	4317 (2)	974 (5)	3233 (8)	15 (1)	98 (6)	175 (13)	−2 (2)	2 (3)	−11 (8)
O(5′)	4280 (1)	392 (3)	684 (5)	10 (0)	115 (4)	220 (9)	−7 (1)	6 (2)	−43 (6)

	x	y	z	B		x	y	z	B
H(5)	339 (2)	343 (8)	924 (11)	7·2 (1·4)	H(3′)	424 (2)	274 (6)	732 (10)	2·7 (1·0)
H(81)	219 (2)	191 (8)	52 (12)	4·8 (1·4)	H(O3′)	515 (2)	440 (9)	596 (13)	8·8 (1·8)
H(82)	159 (2)	136 (7)	98 (11)	5·6 (1·3)	H(4′)	473 (2)	324 (6)	217 (9)	1·5 (0·9)
H(1′)	351 (2)	584 (6)	322 (10)	2·8 (1·0)	H(51)	465 (2)	36 (8)	415 (12)	3·9 (1·4)
H(2′)	396 (2)	554 (5)	793 (9)	1·7 (0·9)	H(52)	399 (2)	68 (8)	425 (12)	4·8 (1·4)
H(O2′)	466 (2)	714 (8)	637 (12)	6·1 (1·5)	H(O5′)	403 (2)	−37 (8)	40 (12)	5·0 (1·4)

than the conventional *syn* range ($\chi \simeq 208$–$256°$). Consequently, this intermediate region has also been referred to as the *syn B* to distinguish it from the more familiar *syn* region (Rao & Sundaralingam, 1970). However, there is actually a continuity of χ angles extending from about $\chi = 0°$ to 130° (Yathindra & Sundaralingam, 1973) and the extended region ($\chi > 90°$) has also been referred to as the 'high anti' conformation (Prusiner, Brennan & Sundaralingam, 1973). This conformation is commonly observed in nucleoside analogs (Prusiner *et al.*, 1973; Singh & Hodgson, 1974).

The ribose geometry

The bond distances and bond angles in the ribose moieties are generally in agreement with those found in nucleosides and nucleotides (Sundaralingam, 1965, 1973; Sundaralingam & Jensen, 1965; Prusiner, 1974). However, there are some significant differences between V1 and V2 due to the variation in the sugar ring pucker. In V1 the endocyclic angles C(1′)–C(2′)–C(3′) and C(2′)–C(3′)–C(4′) are nearly equal while in V2 the angle at C(3′) is larger by 2·5° than that at C(2′). In V1 the endocyclic angle O(1′)–C(1′)–C(2′) is larger than C(3′)–C(4′)–O(1′) by 3·7°, while in V2 they are equal. The exocyclic angle O(3′)–C(3′)–C(2′) in V1 is 4·2° smaller than in V2 while the exocyclic angle C(3′)–C(4′)–C(5′) in V1 is 5·9° larger than in V2. This difference is perhaps also augmented by the variation in the C(4′)–C(5′) bond conformations. The bond distances C(2′)–C(3′) and C(3′)–C(4′) are smaller in V1 than in V2. Thus, the main differences in the two structures involve the atoms C(3′) and C(4′).

The ribose conformation

The torsion angles (τ_0 to τ_4) for the ribose bonds and the pseudorotation parameters **P** and τ_m (phase angle of pseudorotation and maximum amplitude of pseudorotation) (Altona & Sundaralingam, 1972) for the two structures are given in Table 4. The **P** value of V1, 11·7°, differs from that of V2, 335·8°, by 35·9°. [In the preliminary communication (Prusiner & Sundaralingam, 1973) the **P** value of V1 was erroneously quoted as 36·4° instead of 11·7°.] The furanose ring conformation observed in V2 is unusual for β-nucleo-

Table 4. *Deviations* (Å) *of the atoms from the least-squares planes for the ribose, the ring torsion angles, and the pseudorotation parameters*

Atoms used in fitting the least-squares planes are denoted by asterisks.

Plane I: $0{\cdot}522X + 0{\cdot}267Y - 0{\cdot}809Z = -2{\cdot}058$
Plane II: $-0{\cdot}630X - 0{\cdot}776Y - 0{\cdot}022Z = -4{\cdot}265$

Atom	Virazole 1 Plane I	Virazole 2 Plane II
C(1′)	−0·103*	0·202
O(1′)	−0·046*	−0·095*
C(2′)	0·212*	−0·219*
O(2′)	1·628	0·449
C(3′)	−0·244*	0·159*
O(3′)	0·400	1·544
C(4′)	0·181*	−0·046*
C(5′)	−0·459	−1·298
O(5′)	−1·962	−1·355
r.m.s. deviation of fitted atoms	0·173	0·158

Ring torsion angles	V1	V2
τ_0 C(4′)–O(1′)–C(1′)–C(2′)	4·6°	25·2°
τ_1 O(1′)–C(1′)–C(2′)–C(3′)	−27·1	−35·7
τ_2 C(1′)–C(2′)–C(3′)–C(4′)	37·8	32·3
τ_3 C(2′)–C(3′)–C(4′)–O(1′)	−36·1	−18·9
τ_4 C(3′)–C(4′)–O(1′)–C(1′)	20·1	−3·8

Pseudoration parameters	Virazole 1	Virazole 2
τ_m	39·4°	36·4°
P	11·7	335·8

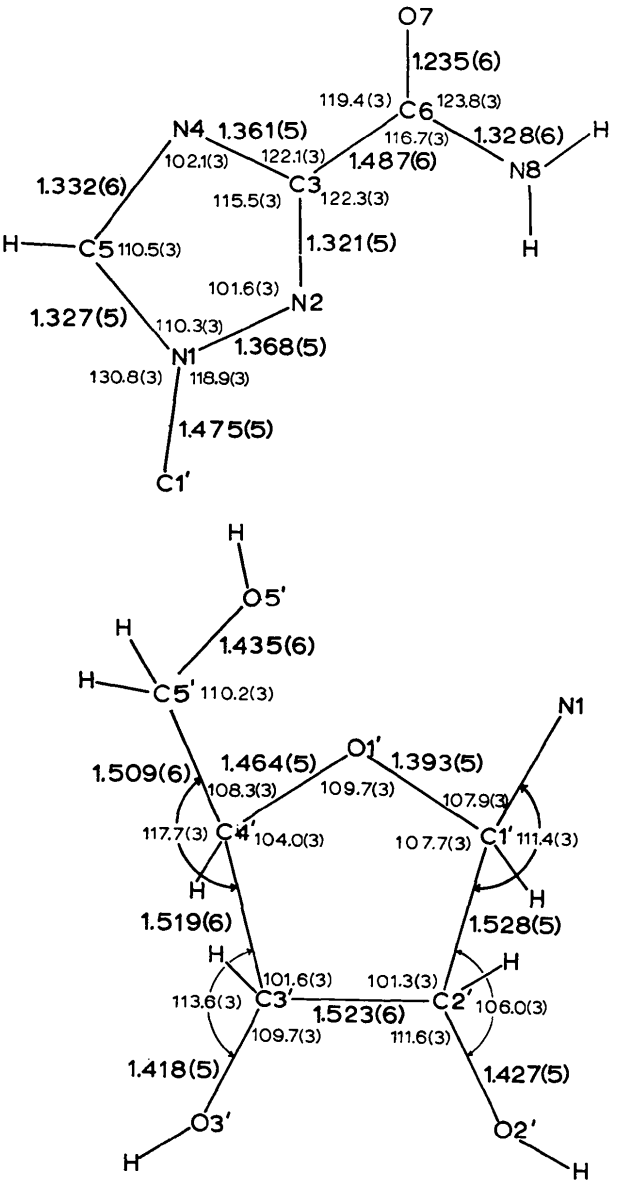

Fig. 2. The bond distances and bond angles in V1.

sides, although quite common in the α-nucleosides (Sundaralingam, 1971). 2′-*O*-Methyladenosine is the only other example of a β-nucleoside possessing the **P** value (349·9°) in this unusual range (Prusiner & Sundaralingam, 1976). In terms of the least-squares plane notations (Sundaralingam, 1965) the ribose conformation in V1 is 3′-*endo*–2′-*exo* ($^3T_2$), and 2′-*exo*–1′-*endo* ($_2T^1$) in V2. The conformation about the exocyclic C(4′)–C(5′) bond is *gauche*$^+$ in V1 and *trans* in V2.

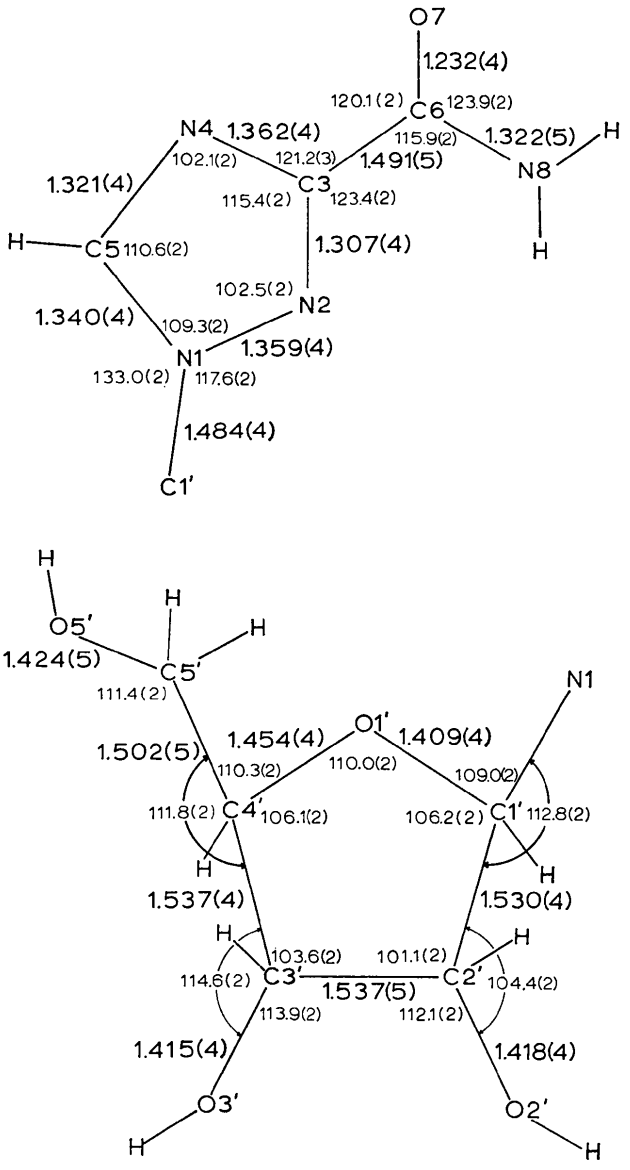

Fig. 3. The bond distances and bond angles in V2.

The base

The bond distances and angles are generally in good agreement between the two structures with the exception of the exocyclic angle C(1′)–N(1)–C(5) which may be due to the differences in χ values. The increase in the valence angle C(1′)–N(1)–C(5) in V2 is probably due to the steric interaction between the base C(5)–H and the ribose C(2′)–H(2′) groups. A survey of the known pyrimidine and purine crystal structures in fact reveals that the exocyclic angles at the glycosyl nitrogen are correlated with χ (Rao & Sundaralingam, 1970; Saenger, 1971; Sundaralingam, unpublished results).

Table 5 shows the comparison of the bond distances and bond angles in the triazole rings of V1 and V2 with the imidazole ring of *N*-(β-D-ribofuranosyl)-imidazole (James & Matsushima, 1973), the pyrrole ring of showdomycin (Tsukuda & Koyama, 1970) and the diazole ring of pyrazomycin (Jones & Chaney, 1973). It is seen that the geometry of the five-membered rings is markedly affected by the positional and numeric variation of nitrogen atoms. The valency angles appear to be especially sensitive to the number of nitrogen atoms in the ring. In showdomycin (pyrrole ring) the difference between the maximum and minimum endocyclic angles is 4·4°, in *N*-(β-D-ribofuranosyl)imidazole (imidazole ring), 6·8°, in pyrazomycin (diazole ring), 5·1°, and in V1 and V2 (triazole rings), 13·9 and 13·2°, respectively. The N–N bond distances in V1 and V2 are similar to those found for pyrazomycin while the C–N bond lengths vary from 1·307 (V2) to 1·388 Å (showdomycin). In general it appears that shorter C–N bonds are associated with heterocyclic rings containing more nitrogen atoms and longer C–N bonds with a smaller number of nitrogen atoms.

The N(1)–N(2) bond distances of 1·368 Å in the base in V1 and 1·359 Å in V2 are similar to those found in the purine analogs formycin, N(7)–N(8) = 1·363 Å (Prusiner

Table 5. *Comparison of the bond distances* (Å) *and angles* (°) *in the heterocyclic five-membered base moieties of the triazole rings of V*1(**1**) *and V*2(**2**), *the neutral imidazole ring of N-*(β-D-*ribofuranosyl)imidazole*(**3**), *the diazole ring of pyrazomycin*(**4**), *and the pyrrole ring of showdomycin*(**5**)

Compound	X(1)	R1	X(2)–R2	X(3)–R3	X(4)–R4
1	N	-H	N	C–$CONH_2$	N
2	N	-H	N	C–$CONH_2$	N
3	N	-H	C–H	N	C–H
4	C	-OH	C–$CONH_2$	N–H	N
5	C	=O	N–H	C=O	C–H

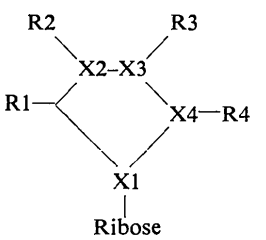

Table 5 (*cont.*)

	1	2	3	4	5
X(1)–C	1·327	1·340	1·376	1·409	1·510
C—X(2)	1·332	1·321	1·360	1·385	1·368
X(2)–X(3)	1·361	1·362	1·378	1·359	1·388
X(3)–X(4)	1·321	1·307	1·316	1·375	1·498
X(1)–X(4)	1·368	1·359	1·351	1·358	1·318
σ bond	0·005	0·004	0·003	0·006	0·007
	1	**2**	**3**	**4**	**5**
X(1)–C—X(2)	110·5	110·6	105·7	106·0	106·3
C—X(1)–X(4)	110·3	109·3	107·0	109·8	108·4
C—X(2)–X(3)	102·1	102·1	110·3	107·4	110·6
X(2)–X(3)–X(4)	115·5	115·4	105·1	110·9	106·2
X(1)–X(4)–X(3)	101·6	102·5	111·9	105·8	108·5
σ angle	0·3	0·2	0·2	0·4	0·4

et al., 1973) and allopurinol, N(8)–N(9) = 1·374 Å (Prusiner & Sundaralingam, 1972).

The planarity of the base and carboxamide groups

The carboxamide group is coplanar with the base ring in V1, whereas it is rotated approximately 15° out of the base plane in V2. The dispositions of the carboxamide groups in the two structures are similar.

Molecular packing and hydrogen bonding

The hydrogen bond distances and angles in V1 and V2 are given in Table 6. All potential sites on the base in V1 are involved in hydrogen bonding with the riboses of screw and translation related molecules (see Fig. 5). O(7) and N(8) form a hydrogen-bonded 'pair' to O(2′) and O(3′) of an adjacent ribose. In V2 N(2) of the base is not involved in hydrogen bonding (Fig. 6). But like V1 there is hydrogen bonding between the base and the ribose. Only one of the amino

Table 6. *Hydrogen-bond lengths* (Å) *and angles* (°)

Estimated standard deviations are given in parentheses and refer to the least significant digit.
Symmetry operations (1) X,Y,Z; (2) $-X,\frac{1}{2}+Y,\frac{1}{2}-Z$; (3) $\frac{1}{2}-X,-Y,\frac{1}{2}+Z$; (4) $\frac{1}{2}+X,\frac{1}{2}-Y,-Z$.

Symmetry operation	Translation X	Y	Z		Angle	Length	Length from hydrogen
Virazole 1							
4	0	1	1	O(3′)–H(O3′)···N(4)	149 (1)	2·860 (5)	1·71 (5)
1	0	−1	0	O(5′)–H(O5′)···N(2)	143 (1)	2·899 (5)	2·30 (5)
4	0	2	1	O(2′)–H(O2′)···O(7)	158 (1)	2·684 (5)	1·98 (5)
4	−1	2	1	N(8)–H(82)···O(3′)	148 (1)	2·976 (5)	2·11 (5)
Virazole 2							
1	0	0	−1	N(8)–H(81)···N(4)	166 (1)	2·939 (4)	1·99 (5)
2	1	0	1	O(2′)–HO(2′)···O(3′)	158 (1)	2·745 (4)	1·78 (5)
2	1	0	0	O(3′)–HO(3′)···O(5′)	159 (1)	2·683 (4)	1·85 (5)
3	0	0	−1	O(5′)–HO(5′)···O(7)	158 (1)	2·690 (4)	1·85 (5)

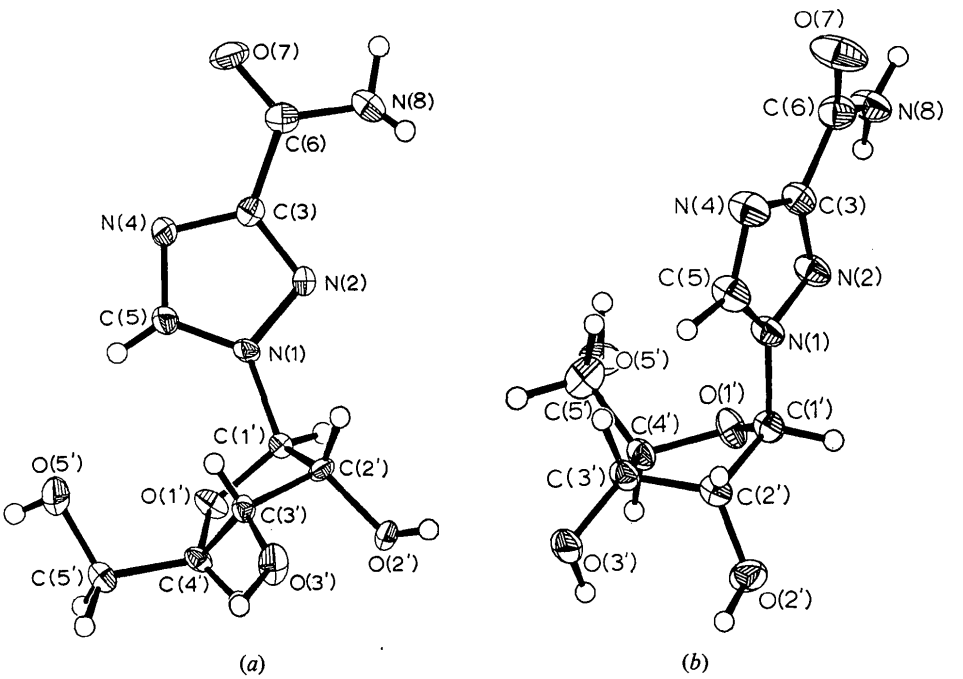

Fig. 4. The ellipsoids of thermal vibrations of the nonhydrogen atoms and molecular conformations of (*a*)V1 and (*b*)V2.

hydrogen atoms is engaged in hydrogen bonding in V1 and V2. In V1, H(81) is hydrogen bonded and H(82) is free while the reverse is the case in V2. Further there is an interbase hydrogen bond between N(4) and N(8) of translation related molecules in V2 which is not present in V1. In addition, V2 shows a short C–H···O contact between C(5) and the ribose O(1′) [see also Sprang & Sundaralingam (1973)]. In both crystal

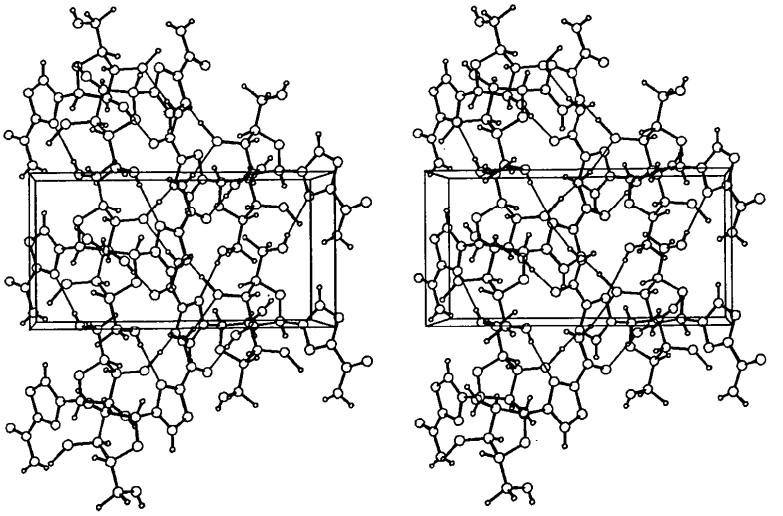

Fig. 5. A stereoscopic view of the crystal packing in V1 along the *c* axis.

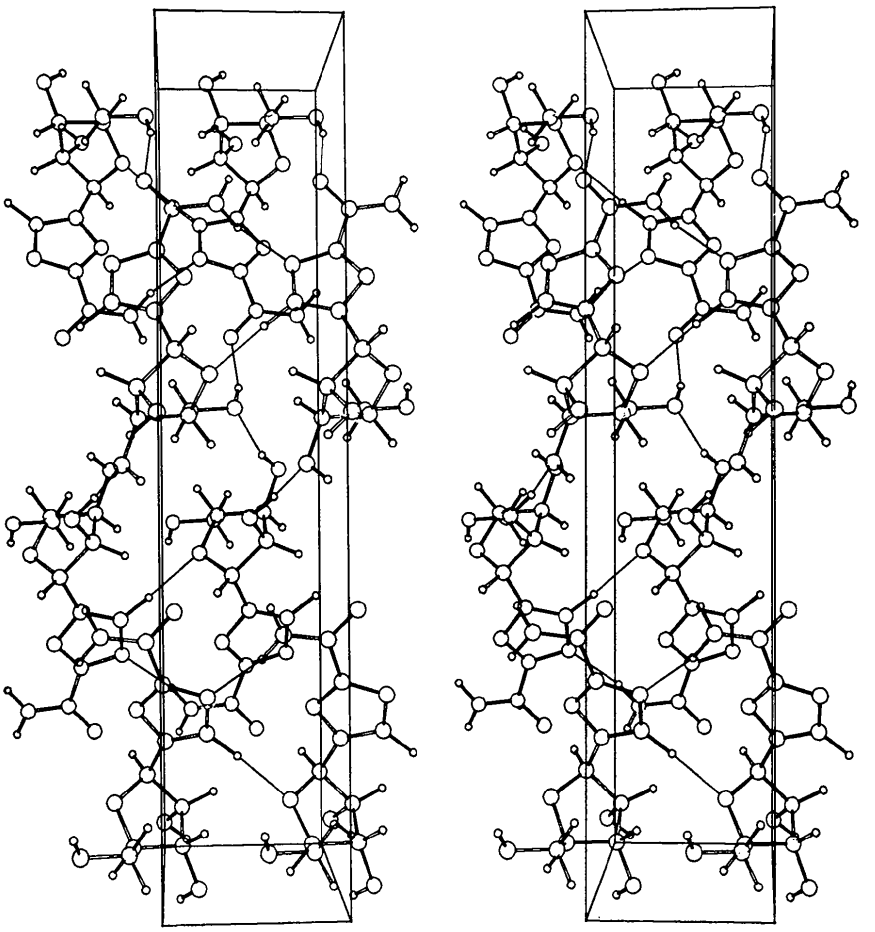

Fig. 6. A stereoscopic view of the crystal packing in V2 along the *b* axis.

structures the crystal packing may be described as constituting alternating ribbons of bases and riboses, a commonly occurring feature in nucleoside crystal structures. Neither structure exhibits intramolecular hydrogen bonding.

The melting point of V1 (174–176°C) is 8° higher than for V2 (166–168°) although V1 contains only four hydrogen bonds compared to five in V2. Apparently, the melting point is dependent on the molecular packing forces besides the intermolecular hydrogen bonding interactions. It may also be noted that the unit cell volume of V1 is smaller than V2 by about 40 Å^3.

Conclusion

Virazole inhibits the synthesis of new viral RNA and DNA but does not stimulate the production of interferon, a protein which counteracts a wide spectrum of viruses (Sidwell *et al.*, 1972). An explanation for the mode of action of virazole may be found in its similarity to inosine or guanosine. An examination of the X-ray structures of virazole reveals that the O(7) and N(8) atoms are spatially similar to the O(6) and N(1) atoms of inosine and guanosine. The notion that the biological activity of virazole is due to its structural similarity to guanosine or inosine is consistent with the work of Streeter, Witkowski, Khare, Sidwell, Bauer, Robins & Simon (1973) who have shown that virazole 5′-phosphate inhibits inosine 5′-phosphate dehydrogenase, thus blocking the synthesis of guanosine 5′-phosphate, a vital precursor in the synthesis of both deoxy- and ribonucleic acids. These workers have further suggested that the binding of virazole 5′-phosphate to the enzyme involves the hydrogen bonding accepting and donating capabilities of O(7) and N(8), respectively. Interestingly, the analogous positions [O(6) and N(1)] in guanosine and inosine are believed to be involved in hydrogen bonding to the active site of IMP dehydrogenase from *A. aerogenes* (Hampton, 1973). It is noteworthy that V1 and V2 were found in slightly different conformations about the glycosyl bond, V1 in normal *anti* and V2 in 'high *anti*'. These characteristics have also been observed for the common substrates guanosine and inosine [Thewalt, Bugg & Marsh (1970); see also Rao & Sundaralingam (1970) and Prusiner *et al.* (1973)].

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