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The Crystal Structure of Formycin Hydrobromide Monohydrate

BY GUNJI KOYAMA AND HAMAO UMEZAWA
Institute of Microbial Chemistry, Kamiosaki, Shinagawa-ku, Tokyo 141, *Japan*

AND YOICHI IITAKA
Faculty of Pharmaceutical Sciences, University of Tokyo, Hongo, Tokyo 113, *Japan*

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The title compound, $C_{10}H_{13}N_5O_4.HBr.H_2O$, crystallizes in $P1$ with $a=6{\cdot}67$ (3), $b=11{\cdot}00$ (5), $c=4{\cdot}96$ (2) Å, $\alpha=103{\cdot}4$ (5), $\beta=101{\cdot}2$ (5), $\gamma=96{\cdot}0$ (5)°, $Z=1$. The structure was solved by the heavy-atom method and refined by the full-matrix least-squares method to an R value of 0·083 for 1475 observed structure factors obtained by the photographic method. The molecule is in the *syn* conformation with a χ angle of $-149{\cdot}3°$ and a hydrogen bond O(5′)–H···N(3) of 2·883 Å is formed within the molecule. The conformation of the sugar is C(2′)-*endo*–C(3′)-*exo* and that about the C(4′)–C(5′) bond is *gauche–trans*. It was found that on formation of a salt with hydrobromic acid, the molecule is protonated at N(1), with the migration of a hydrogen atom from N(7) to N(8).

Introduction

Formycin, $C_{10}H_{13}N_5O_4$, was first isolated from a culture filtrate of *Nocardia interforma* and was found to have an inhibitory effect on the growth of Yoshida rat sarcoma 180, *Mycobacterium* 607 and *Xanthomonas oryzae* (Umezawa, Hori, Ito, Takita, Koyama & Takeuchi, 1964). On the basis of the physical and chemical properties of formycin and formycin B (a deaminated product of formycin), it was suggested that these antibiotics are a kind of nucleoside. Usual procedures of degradation of common nucleosides, however, were unsuccessful in determining the total structure. An X-ray analysis was therefore undertaken in order to elucidate the molecular structure including its absolute configuration and to compare the structure with those of the nucleosides and nucleotides studied to date. A preliminary report of the present study has already been published (Koyama, Maeda, Umezawa & Iitaka, 1966). Recently, a detailed study of the crystal structure of formycin monohydrate has been reported (Prusiner, Brennan & Sundaralingam, 1973). We therefore describe the details of our structure determination of formycin hydrobromide monohydrate and compare the result with that obtained for formycin monohydrate.

Experimental

Slow cooling of an aqueous hydrobromic acid solution of formycin free base yielded crystals of formycin hydrobromide monohydrate which were recrystallized in the form of colourless prisms from an aqueous solution. Specimens used in the present work had the dimensions $0{\cdot}7\times0{\cdot}3\times0{\cdot}5$ and $0{\cdot}5\times0{\cdot}3\times0{\cdot}3$ mm. The former was used for the intensity measurements around the c axis and the latter around the b axis. The density was measured by the flotation method using a mixture of carbon tetrachloride and bromoform. The lattice constants were determined from $hk0$, $h0l$ and $0kl$ precession photographs taken with Cu $K\alpha$ radiation ($\lambda=1{\cdot}5418$ Å). Equi-inclination integrated Weissenberg photographs of the layers $hk0$–$hk4$ and $h0l$–$h7l$ were taken with Cu $K\alpha$ radiation using the multiple-film technique. The intensities were measured by a Narumi microphotometer. Lorentz and polarization corrections were applied but no correction was made for absorption effects. The structure factors were put on the same relative scale by correlating the values on various layers. 1475 structure factors were obtained, which correspond to about 98% of the total independent reflexions for $\sin\theta\leq0{\cdot}98$.

Crystal data

Formycin hydrobromide monohydrate, $C_{10}H_{13}N_5O_4.HBr.H_2O$, F.W. 366·18, m.p. 180–185°C. Triclinic, $a=6{\cdot}67$ (3), $b=11{\cdot}00$ (5), $c=4{\cdot}96$ (2) Å, $\alpha=103{\cdot}4$ (5), $\beta=101{\cdot}2$ (5), $\gamma=96{\cdot}0$ (5)°, $U=343$ Å^3, $D_m=1{\cdot}80$, $D_x=1{\cdot}78$ g cm^{-3}, $Z=1$, space group $P1$.

The atomic scattering factors for the bromide ion, oxygen, nitrogen and carbon atoms were taken from *International Tables for X-ray Crystallography* (1962).

Determination and refinement of the structure

In the space group $P1$, the choice of the origin is arbitrary and the position of the bromide ion was assumed to be the origin of the unit cell. A three-dimensional Fourier synthesis phased by the bromide ion revealed 40 well resolved prominent electron-density peaks in a cell distributed around a false centre of symmetry. This number was just twice that expected for one structure unit. The distances of all the possible pairs of peaks within 2·2 Å were then taken into account and trials were made to reject unreasonable peaks lying too far away or too near to form chemical bonds. This process was repeated until all the peaks were

interpreted as constituting a plausible molecular model. One cycle of full-matrix least-squares refinement was carried out assuming all the light atoms were oxygen. The R value dropped to 0·16. Inspection of the individual thermal parameters allowed us to assign conclusively the five oxygen atoms of a ribose moiety and a water of crystallization. At this stage, a difference Fourier synthesis was computed and the exocyclic nitrogen atom of the amino group was located. A further two cycles of full-matrix least-squares refinement (isotropic) and difference Fourier syntheses yielded the final structure of formycin. Up to this stage of the analysis, all calculations were carried out using 1130 visually estimated independent structure factors and the R value was reduced to 0·11. The result was reported in the preliminary paper. Further refinement was carried out on the basis of data recollected by a microphotometer. Seven cycles of block-diagonal least-squares refinement in which individual anisotropic thermal parameters were applied for all atoms reduced the R value to 0·083 for 1475 reflexions. The final atomic parameters and their estimated standard deviations are listed in Table 1.

The weighting system used for the calculations was:

$$\sqrt{w} = 30/F_o, \text{ when } 30 < F_o$$
$$\sqrt{w} = 1, \text{ when } 8 \leq F_o \leq 30$$
$$\sqrt{w} = 0{\cdot}3, \text{ when } F_o < 8 .$$

A comparison of the observed and calculated structure factors is given in Table 2.* The bond distances and angles are shown in Fig. 1 and also listed in Tables 3 and 4, compared with those found in formycin monohydrate.

* Table 2 (F_o and F_c) has been deposited with the British Library Lending Division as Supplementary Publication No. SUP 30350 (7pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 13 White Friars, Chester CH1 1NZ, England.

Determination of the absolute configuration

The absolute configuration was determined by the use of the anomalous dispersion method (Bijvoet, Peerdeman & van Bommel, 1951). The anomalous dispersion corrections used for the scattering factor of the bromine atom were $\Delta f' = -0{\cdot}9$ and $\Delta f'' = 1{\cdot}5$ (Dauben & Templeton, 1955). Out of 53 pairs of reflexions in $hk0$ and

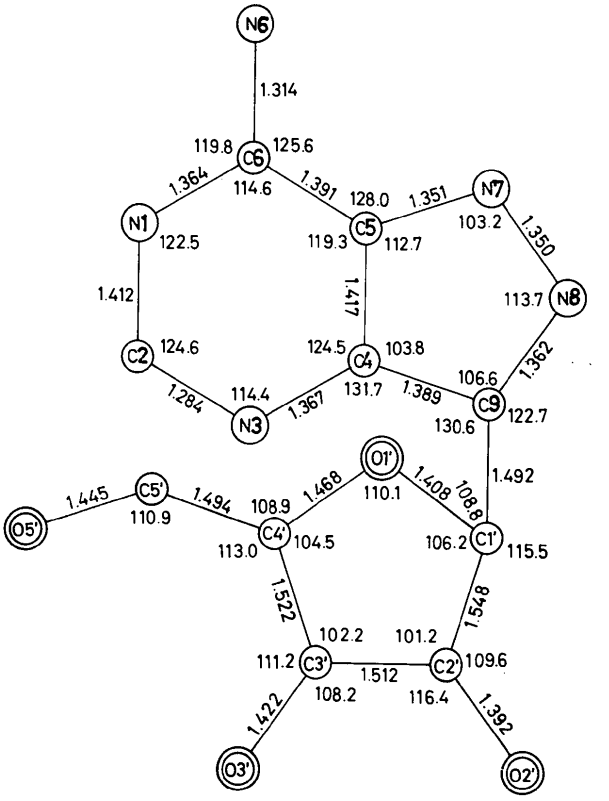

Fig. 1. Bond lengths (Å) and angles (°).

Table 1. *Final atomic parameters with estimated standard deviations in parentheses*

The temperature factors are of the form $T = \exp[-(\beta_{11}h^2 + \beta_{22}k^2 + \beta_{33}l^2 + 2\beta_{12}hk + 2\beta_{13}hl + 2\beta_{23}kl)]$. Values are $\times 10^4$.

	x	y	z	β_{11}	β_{22}	β_{33}	β_{12}	β_{13}	β_{23}
Br	0 (0)	0 (0)	0 (0)	65 (2)	43 (1)	125 (3)	11 (1)	5 (2)	−19 (1)
N(1)	2574 (17)	1703 (10)	6018 (21)	89 (22)	39 (8)	94 (37)	9 (10)	−54 (22)	−21 (14)
C(2)	1159 (19)	2541 (12)	6636 (26)	95 (26)	39 (10)	129 (43)	18 (12)	−14 (27)	−25 (17)
N(3)	1398 (17)	3386 (10)	8990 (22)	96 (22)	39 (8)	117 (37)	23 (11)	2 (24)	−13 (14)
C(4)	3225 (17)	3465 (11)	10889 (25)	46 (21)	32 (9)	162 (44)	19 (10)	−3 (25)	18 (16)
C(5)	4688 (17)	2632 (10)	10485 (22)	57 (21)	27 (8)	82 (38)	2 (10)	−22 (23)	1 (14)
C(6)	4369 (16)	1723 (11)	7910 (23)	33 (20)	44 (9)	93 (40)	0 (11)	3 (22)	−8 (16)
N(6)	5626 (19)	918 (12)	7223 (24)	133 (27)	53 (10)	143 (43)	41 (13)	12 (27)	−35 (16)
N(7)	6334 (17)	2894 (10)	12719 (23)	79 (22)	42 (9)	159 (42)	3 (11)	−67 (24)	−23 (15)
N(8)	5926 (16)	3912 (10)	14521 (19)	92 (22)	39 (8)	43 (33)	18 (10)	−29 (21)	−16 (13)
C(9)	4090 (19)	4281 (12)	13554 (25)	81 (24)	38 (9)	114 (42)	11 (11)	−17 (25)	15 (16)
C(1′)	3363 (17)	5407 (10)	15165 (23)	80 (23)	19 (8)	110 (41)	1 (10)	11 (24)	−13 (14)
C(2′)	3986 (18)	6667 (11)	14438 (24)	72 (23)	33 (9)	131 (43)	4 (11)	7 (26)	19 (16)
C(3′)	2238 (17)	7371 (10)	15108 (25)	68 (23)	23 (8)	172 (46)	5 (11)	14 (26)	−4 (16)
C(4′)	369 (18)	6325 (11)	14040 (23)	77 (23)	44 (10)	86 (39)	26 (12)	10 (25)	5 (16)
O(1′)	1182 (13)	5192 (8)	14575 (19)	58 (16)	34 (7)	173 (34)	2 (8)	22 (19)	10 (12)
O(2′)	5971 (14)	7220 (10)	16012 (23)	58 (19)	54 (8)	292 (44)	−4 (10)	9 (23)	11 (16)
O(3′)	2626 (15)	7864 (9)	18107 (21)	102 (20)	51 (8)	195 (40)	42 (11)	−1 (23)	−49 (15)
C(5′)	−544 (21)	6086 (13)	10931 (26)	117 (29)	43 (10)	123 (45)	−17 (13)	−14 (29)	−22 (17)
O(5′)	−1858 (15)	4869 (9)	9841 (21)	115 (22)	49 (8)	180 (39)	−13 (10)	−69 (23)	12 (15)
O(W)	6047 (18)	8988 (12)	12553 (22)	156 (27)	78 (11)	160 (40)	−27 (13)	8 (26)	−19 (17)

h0l for wich the intensity differences are expected to be measurable, 33 pairs showed significant differences in the *c* and *b* axes Weissenberg photographs. The results, given in Table 5, indicate that the absolute configuration is expressed by taking the atomic coordinates of Table 1 referred to the right-handed set of axes.

Table 3. *Bond distances with their estimated standard deviations in parentheses*

	FM.HBr.H_2O (Present study)	FM.H_2O (Prusiner, Brennan & Sundaralingam, 1973)
N(1)–C(2)	1·412 (18) Å	1·355 (6) Å
N(1)–C(6)	1·364 (15)	1·336 (6)
C(2)–N(3)	1·284 (15)	1·313 (5)
N(3)–C(4)	1·367 (15)	1·374 (5)
C(4)–C(5)	1·417 (17)	1·379 (6)
C(4)–C(9)	1·389 (14)	1·426 (6)
C(5)–C(6)	1·391 (14)	1·420 (6)
C(5)–N(7)	1·351 (14)	1·359 (5)
C(6)–N(6)	1·314 (18)	1·334 (5)
N(7)–N(8)	1·350 (15)	1·363 (5)
N(8)–C(9)	1·362 (17)	1·324 (5)
C(9)–C(1′)	1·492 (17)	1·501 (5)
C(1′)–C(2′)	1·548 (17)	1·535 (5)
C(1′)–O(1′)	1·408 (14)	1·438 (5)
C(2′)–C(3′)	1·512 (17)	1·533 (5)
C(2′)–O(2′)	1·392 (13)	1·407 (5)
C(3′)–C(4′)	1·522 (15)	1·538 (5)
C(3′)–O(3′)	1·422 (15)	1·431 (5)
C(4′)–O(1′)	1·468 (16)	1·453 (5)
C(4′)–C(5′)	1·494 (16)	1·499 (5)
C(5′)–O(5′)	1·445 (15)	1·428 (5)

Table 4. *Bond angles and their estimated standard deviations in parentheses*

	FM.HBr.H_2O (Present study)	FM.H_2O (Prusiner, Brennan & Sundaralingam, 1973)
C(2)–N(1)–C(6)	122·5 (1·1)°	119·1 (0·3)°
N(3)–C(2)–N(1)	124·6 (1·2)	129·0 (0·3)
C(4)–N(3)–C(2)	114·4 (1·1)	112·5 (0·3)
C(5)–C(4)–N(3)	124·5 (1·1)	123·0 (0·3)
C(5)–C(4)–C(9)	103·8 (1·0)	105·5 (0·3)
N(3)–C(4)–C(9)	131·7 (1·1)	131·5 (0·3)
C(6)–C(5)–C(4)	119·3 (1·0)	119·9 (0·3)
C(6)–C(5)–N(7)	128·0 (1·1)	133·1 (0·3)
C(4)–C(5)–N(7)	112·7 (1·0)	107·0 (0·3)
N(6)–C(6)–N(1)	119·8 (1·1)	120·5 (0·3)
N(6)–C(6)–C(5)	125·6 (1·1)	123·1 (0·3)
N(1)–C(6)–C(5)	114·6 (1·0)	116·4 (0·3)
N(8)–N(7)–C(5)	103·2 (1·0)	110·9 (0·3)
C(9)–N(8)–N(7)	113·7 (1·0)	106·8 (0·3)
C(1′)–C(9)–C(4)	130·6 (1·1)	128·2 (0·3)
C(1′)–C(9)–N(8)	122·7 (1·1)	121·9 (0·3)
C(4)–C(9)–N(8)	106·6 (1·1)	109·8 (0·3)
C(2′)–C(1′)–C(9)	115·5 (1·0)	115·2 (0·3)
C(2′)–C(1′)–O(1′)	106·2 (0·9)	103·4 (0·3)
C(9)–C(1′)–O(1′)	108·8 (0·9)	109·4 (0·3)
C(3′)–C(2′)–C(1′)	101·2 (0·9)	102·3 (0·3)
C(3′)–C(2′)–O(2′)	116·4 (1·0)	114·7 (0·3)
C(1′)–C(2′)–O(2′)	109·6 (1·0)	114·6 (0·3)
C(4′)–C(3′)–C(2′)	102·2 (0·9)	102·7 (0·3)
C(4′)–C(3′)–O(3′)	111·2 (1·0)	108·9 (0·3)
C(2′)–C(3′)–O(3′)	108·2 (1·0)	111·6 (0·3)
O(1′)–C(4′)–C(3′)	104·5 (0·9)	106·9 (0·3)
O(1′)–C(4′)–C(5′)	108·9 (1·0)	108·9 (0·3)
C(3′)–C(4′)–C(5′)	113·0 (1·0)	112·3 (0·3)
O(5′)–C(5′)–C(4′)	110·9 (1·1)	111·6 (0·3)
C(1′)–O(1′)–C(4′)	110·1 (0·9)	109·3 (0·3)

Table 5. *Comparison of the observed and calculated intensity differences used for the establishment of the absolute configuration*

h	k	l	$F_o^2(hkl)/F_o^2(\bar{h}\bar{k}\bar{l})$	$I_o(hkl)/I_o(\bar{h}\bar{k}\bar{l})$
$\bar{1}$	0	0*	1·258	>1
0	0	$\bar{1}$	0·858	<1
$\bar{3}$	0	$\bar{1}$	1·284	>1
3	0	$\bar{2}$	0·507	<1
$\bar{1}$	0	$\bar{3}$	0·819	<1
$\bar{3}$	0	$\bar{3}$	1·405	>1
4	0	$\bar{4}$	1·269	>1
3	0	$\bar{4}$	1·394	>1
2	0	$\bar{4}$	0·682	<1
$\bar{1}$	0	$\bar{4}$	0·699	<1
2	0	$\bar{6}$	1·525	>1
0	0	$\bar{6}$	0·689	<1
0	13	0	0·710	<1
0	$\bar{5}$	0	0·781	<1
0	$\bar{9}$	0	0·860	<1
$\bar{1}$	11	0	2·487	>1
$\bar{1}$	10	0	0·847	<1
$\bar{1}$	7	0	0·731	<1
$\bar{1}$	0	0*	1·258	>1
$\bar{1}$	$\bar{1}$	0	0·819	<1
$\bar{1}$	$\bar{10}$	0	0·826	<1
$\bar{1}$	$\bar{12}$	0	1·700	>1
$\bar{2}$	5	0	0·859	<1
$\bar{2}$	2	0	0·862	<1
$\bar{2}$	$\bar{4}$	0	0·747	<1
$\bar{2}$	$\bar{6}$	0	0·627	<1
$\bar{3}$	$\bar{1}$	0	0·816	<1
$\bar{3}$	$\bar{10}$	0	0·856	<1
$\bar{4}$	$\bar{6}$	0	0·820	<1
$\bar{5}$	$\bar{2}$	0	0·859	<1
$\bar{5}$	$\bar{5}$	0	1·278	>1
$\bar{5}$	$\bar{8}$	0	0·838	<1
$\bar{5}$	$\bar{9}$	0	0·770	<1
$\bar{7}$	$\bar{4}$	0	0·719	<1

* This reflexion was observed in both *b*- and *c*-axis data.

Discussion

The structural formulae and nomenclature of formycin are shown in Fig. 2.

It is seen that the molecule consists of an amino-substituted pyrazole[4,3-*d*]pyrimidine base and a D-ribofuranose moiety forming a nucleoside-like molecule. Both the groups are linked through a carbon–carbon single bond in place of the nitrogen–carbon glycosyl bond in the usual nucleosides. This unusual feature of the attachment of the sugar moiety to the base has also been found in some natural products, such as pseudouridine (Rohrer & Sundaralingam, 1970), showdomycin (Tsukuda & Koyama, 1970) and pyrazomycin (Jones & Chaney, 1972). We shall discuss the structure in comparison with the published data for formycin monohydrate, adenosine and its analogues.

The pyrazolopyrimidine base residue

The base moiety including the exocyclic amino nitrogen is nearly planar. The least-squares plane through

the ten non-hydrogen atoms is represented by the equation

$$-0{\cdot}496X-0{\cdot}714Y+0{\cdot}673Z=-0{\cdot}154$$

where X, Y, Z are taken parallel to the a, b, c axes and measured in Å. The maximum deviation from the plane is observed for C(5), 0·039 Å, and the root-mean-square deviation is 0·026 Å. The exocyclic N(6) is coplanar with a deviation of 0·015 Å. The displacement of the atom C(1′) from the plane is 0·140 Å and the glycosyl bond C(1′)–C(9) makes an angle of about 5·4° with the base plane.

As pointed out by Rao & Sundaralingam (1970) there are significant differences in the bond distances and angles between the neutral and the protonated adenine base. Some characteristic features of the N(1)-protonated adenine are also seen in the present structure: the two C–N bonds around N(1) are longer, while the C(2)–N(3) and C(6)–N(6) bonds are shorter, than those found in the neutral base and the bond angles N(1)–C(6)–C(5) and N(1)–C(2)–N(3) are significantly smaller, while C(6)–N(1)–C(2) is larger than those found in the neutral base. These points, together with the consideration of the hydrogen bonding, suggest that the protonated site in the present structure is also on the N(1) atom. However, another drastic difference in the structures of the protonated bases in adenosine and formycin has drawn our attention. A comparison of the bond lengths and angles between the protonated and unprotonated formycin bases (Tables 3 and 4) clearly shows a significant increase in lengths C(4)–C(5) 0·038 Å, N(8)–C(9) 0·038 Å, and in angles N(7)–N(8)–C(9) 6·9°, N(7)–C(5)–C(4) 5·7° on the one hand, and a decrease in lengths C(5)–C(6) −0·029 Å, C(9)–C(4) −0·037 Å, and in angles N(7)–C(5)–C(6) −5·1°, C(4)–C(9)–N(8) −3·2°, N(8)–N(7)–C(5) −7·7° on the other. These differences, together with the fact that N(8) acts as a hydrogen donor in the N(8)–H···O(5′) hydrogen bond indicate that the protonation of formycin base at N(1) gives rise to a migration of a hydrogen atom from N(7) to N(8), thus forming a different resonance structure, as shown in Fig. 2.

The ribofuranose residue

It is well known that the furanose ring of the ribose derivatives is puckered at either C(2′) or C(3′) which are displaced from the plane formed by the remaining four ring atoms by about 0·5–0·6 Å. The least-squares planes were calculated for all combinations of four ring atoms. Table 6 shows the deviations of the atoms from the plane and the root-mean-square deviations of the in-plane atoms. In every case, the root-mean-square deviations are relatively large, implying that there is no such combination of four atoms that best fits a plane. However, the values for the planes I and II are much smaller than those for III, IV and V. This suggests that the puckering of the ribofuranose ring in formycin involves the displacement of both C(2′) and C(3′). Therefore the deviations of C(2′) and C(3′) from the plane, $-0{\cdot}208X+0{\cdot}110Y+0{\cdot}941Z=7{\cdot}271$, formed by the remaining three atoms were calculated (VI); this showed that the mode of puckering is best described by the term 2′-*endo*–3′-*exo*. It is noteworthy that such an intermediate mode of puckering in the furanose ring has not been found so frequently (Arnott & Hukins, 1972). This unusual situation is also reflected in the values of the endocyclic torsion angles as shown in Table 7.

The *gauche–trans* conformation about the C(4′)–C(5′) bond, which is less commonly observed but is also found in formycin monohydrate, is one of the most striking features in its relation to the conformation about the glycosyl bond. This point will be discussed below.

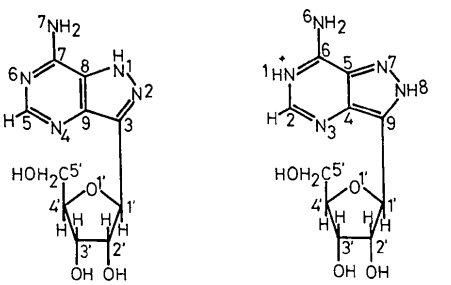

Fig. 2. Structural formulae of formycin. (*a*) Free form numbered on the basis of the formal nomenclature, 7-amino-3-(β-D-ribofuranosyl)pyrazolo[4,3-*d*]pyrimidine, put forward in the previous paper (Koyama *et al.*, 1966). (*b*) Protonated form in hydrobromide with conventional numbering. (All atoms in the tables and figures are designated by the conventional numbering for adenosine.)

Table 6. *Displacement of atoms from the least-squares plane*

The value is positive when the displacement is measured on the same side as the C(5′) atom, which is *endo*. Values marked with an asterisk are for atoms not involved in the least-squares calculation.

	I	II	III	IV	V	VI
C(1′)	+0·051 Å	−0·057 Å	−0·184 Å	−0·340 Å*	−0·142 Å	0 Å
C(2′)	+0·577*	+0·033	+0·171	+0·119	+0·233	+0·273*
C(3′)	−0·046	−0·584*	−0·107	−0·192	−0·236	−0·367*
C(4′)	+0·076	−0·036	+0·402*	+0·198	+0·146	0
O(1′)	−0·081	+0·059	+0·120	−0·125	−0·027*	0
R.m.s. dev. of in-plane atoms	0·065	0·048	0·149	0·163	0·195	0

Table 7. *Torsional angles for the ribofuranose residue*

	FM.HBr.H_2O (Present study)	FM.H_2O (Prusiner, Brennan & Sundaralingam, 1973)
Glycosyl angle, χ	−149·3°	109·8°
Sugar ring angles		
τ_0[O(1′)–C(1′)]	−10·6	−30·6
τ_1[C(1′)–C(2′)]	31·3	39·4
τ_2[C(2′)–C(3′)]	−38·9	−33·1
τ_3[C(3′)–C(4′)]	33·7	15·7
τ_4[C(4′)–O(1′)]	−14·4	9·4
Puckering	C(2′)-*endo*–C(3′)-*exo*	C(2′)-*endo*
Conformation about C(4′)–C(5′)	*gauche–trans*	*gauche–trans*
φ_{OO}O(1′)–C(4′)–C(5′)–O(5′)	46·6	57·6
φ_{OC}C(3′)–C(4′)–C(5′)–O(5′)	162·4	175·8

The plane formed by the three ribose ring atoms C(1′), C(4′) and O(1′) is at a dihedral angle of 64·8° to the base plane and this value is very similar to the known values for the usual nucleosides and nucleotides. The bond lengths and angles in the ribose moiety are somewhat different from those found in formycin monohydrate but are still in the usual range.

Conformation of the molecule

For the description of the conformation about the glycosyl C–N bond in the structures of nucleosides and nucleotides, the torsion angle φ_{C-N} or χ (Donohue & Trueblood, 1960; Sundaralingam & Jensen, 1965) is usually used. This angle χ[O(1′)–C(1′)–C(9)–N(8)] is −149·3° and falls in the *syn* conformational range.

Table 8. *Hydrogen-bond distances*

Estimated standard deviations are in parentheses.

Atom (1)	Atom (2)	Translation for atom(2) *x*	*y*	*z*	Distance	Possible mode
Br	N(1)	0	0	0	3·179 (9) Å	Br·····H—N(1)
O(2′)	O(*W*)	0	0	0	2·880 (18)	O(2′)—H···O(*W*)
O(5′)	N(3)	0	0	0	2·883 (16)	O(5′)—H···N(3)
O(3′)	O(*W*)	0	0	1	2·789 (13)	O(3′)···H—O(*W*)
N(8)	O(5′)	1	0	1	2·669 (12)	N(8)—H···O(5′)
N(6)	Br	1	0	1	3·332 (13)	N(6)—H···Br
O(*W*)	N(6)	0	1	1	2·838 (16)	O(*W*)···H—N(6)
O(*W*)	Br	1	1	1	3·334 (13)	O(*W*)—H···Br
O(3′)	Br	0	1	2	3·152 (10)	O(3′)—H···Br

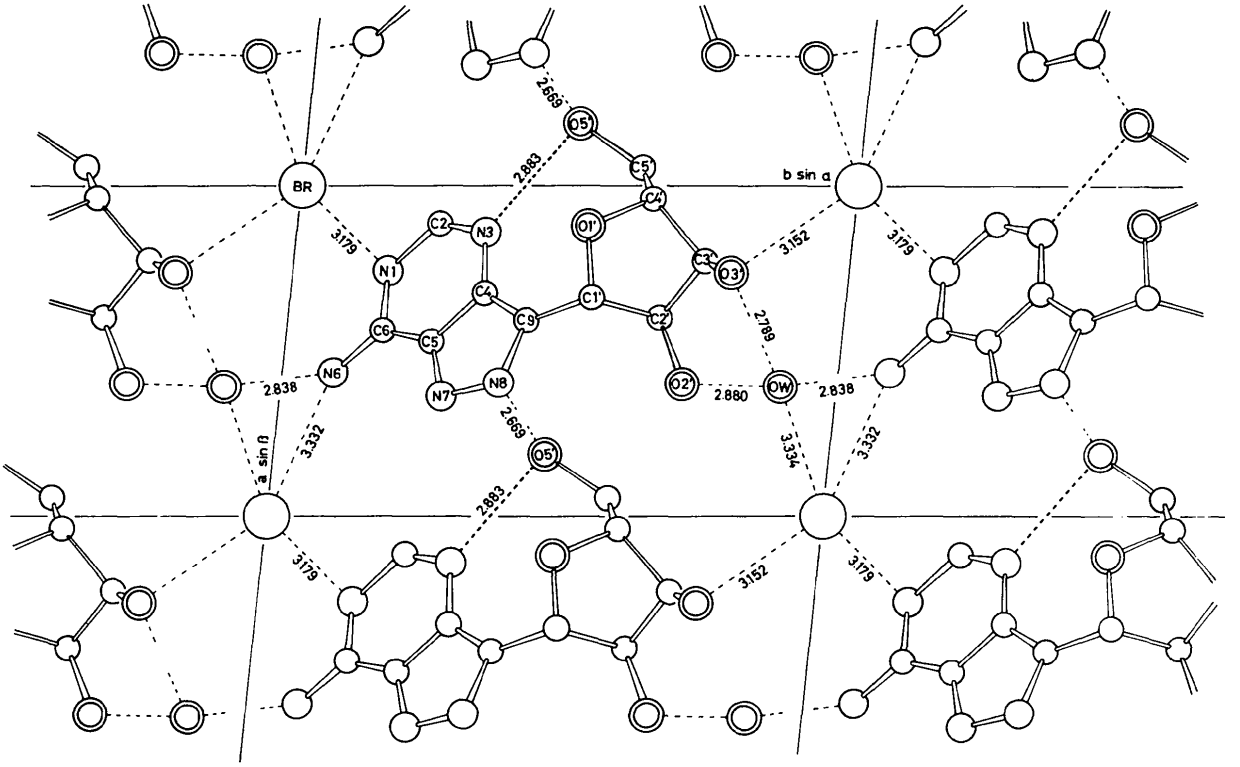

Fig. 3. Projection of the crystal structure along the *c* axis. Hydrogen bonds are shown by dashed lines.

The *syn* conformation has been found in several structures, such as deoxyguanosine in its crystalline complex with 5-bromodeoxycytidine (Haschemeyer & Sobell, 1965), 6-thioinosine (Shefter, 1968), 8-bromoadenosine (Tavale & Sobell, 1970), 8-bromoguanosine (Bugg & Thewalt, 1969; Tavale & Sobell, 1970) and 3-*O*-acetyladenosine (Rao & Sundaralingam, 1970) and these are summarized by Rao & Sundaralingam (1970). According to the general conclusion drawn from the results of the calculation of the interactive energy between the non-bonded atoms (Haschemeyer & Rich, 1967; Wilson & Rahman, 1971) it seems to be possible for the purine nucleosides involving C(2′)-*endo* puckering in the sugar moiety to take both *anti* and *syn* conformations. For this reason it may not be surprising that formycin takes the *syn* conformation under the condition of the C(2′)-*endo*–C(3′)-*exo* puckering of ribose. The *syn* conformation observed in the present study is also of great interest in connexion with the conformation about the C(4′)–C(5′) bond, which has already been mentioned. As seen in the summary table of Rao & Sundaralingam (1970), most of the structures of the purine system with the *syn* conformation prefer the *gauche-gauche* conformation about this bond and an intramolecular hydrogen bond of O(5′)–H···N(3) type is observed at the same time. The conformations of formycin in the present study are *syn* and *gauche–trans* with the puckering of the sugar in the C(2′)-*endo*–C(3′)-*exo* mode, while the intramolecular hydrogen bond in question still seems possible, and thus the O(5′)–N(3) distance is calculated to be 2·883 Å. It may be concluded that all these less common structural features can endow this antibiotic with interesting biochemical activities.

Hydrogen bonding and crystal packing

The crystal structure viewed down along the *c* axis is shown in Fig. 3. Nine different kinds of hydrogen bond are found in the crystal and given as dashed lines in the Figure. The distances between non-hydrogen atoms of these bonds are listed in Table 8. The bromide ion forms four hydrogen bonds nearly tetrahedrally, accepting hydrogens from O(3′), O(*W*), N(1) and N(6). The possible mode of the hydrogen bond $Br^- \cdots H^+$–N(1) again suggests that the protonated site in formycin hydrobromide is on N(1) of the base ring. The water molecule is also involved in four hydrogen bonds, donating its hydrogen atoms to the bromide ion and the O(3′) atom of the molecule in the adjacent cell and accepting two hydrogens from atoms O(2′) and N(6). The remaining two hydrogen bonds are formed between the base and the sugar moieties, the one being intermolecular, O(5′)···N(8) (2·669 Å), and the other intramolecular O(5′)···N(3) (2·883 Å). Because the locations of the hydrogen atoms were not determined in the present study, the mode of these two hydrogen bonds is to be assigned tentatively. There can be two possible modes considered for these bonds: one is that involving O(5′), as O(5′)···H–N(8) and O(5′)–H···N(3), and the other as O(5′)–H···N(8) and O(5′)–H···N(3), The former possibility arises from the assumption that the hydrogen atom on the pyrazole ring is not located at a definite position but rather is mobile and moves from N(7) to N(8), accompanied by the protonation at N(1). In the latter case the hydrogen atom on O(5′) is involved in two bonds, being oriented to both N(3) and N(8) atoms. As mentioned previously, the bond lengths and angles in the pyrazolopyrimidine base strongly support the former possibility.

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