

Stereochemistry of Nucleic Acids and Their Constituents. XIV.* Crystal and Molecular Structure of 2,4-Dithiouridine Monohydrate

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The crystal structure of 2,4-dithiouridine monohydrate has been determined by X-ray diffraction, using 1297 three-dimensional intensity data. The crystals belong to the orthorhombic space group $P2_12_12_1$ with four molecules in a unit cell of dimensions $a=27.992 \pm 0.002$, $b=8.430 \pm 0.001$, $c=5.254 \pm 0.001$ Å. The observed and calculated densities of the crystal are 1.569 g.cm^{-3} . The position of the sulfur atom S(2) was located in a sharpened Patterson synthesis. The structure was determined by refinement of the phases of S(2) by the tangent method. Full-matrix least-squares with anisotropic temperature factors for non-hydrogen atoms and isotropic temperature factors for hydrogen atoms gave a final R value of 0.039. The estimated errors in the bond distances are 0.005–0.008 Å and in bond angles 0.3–0.4°. The base is found to be non-planar and is folded along the virtual bond C(2)···C(5). The glycosidic torsional angle is *anti*, $\chi = 19.5^\circ$; the sugar conformation is C(3') *endo*. The conformation about the C(4')–C(5') bond is *gauche-gauche*. There is practically no overlap of the parallel bases in adjacent molecules; the closest contact between the base and an adjacent sugar is C(4)···O(1') = 3.14 Å. Twofold screw related water molecules are hydrogen bonded to one another to form an infinite twofold helix parallel to the c axis. The only inter-base hydrogen bond is between N(3)–H and S(4) of an adjacent molecule.

Introduction

The structure of 2,4-dithiouridine monohydrate (Fig. 1), a synthetic nucleoside (Ueda, Iida, Ikeda & Mizuno, 1966), has been determined as part of a program of research in these laboratories to provide accurate bond distances, bond angles, base stacking, conformation and hydrogen bonding information on the modified constituents of transfer ribonucleic acids (tRNAs). The thio derivatives 4-thiouridine (Lipsett & Doctor, 1961; Lipsett, 1965; Cory, Marcker, Dube & Clark, 1968), 5-methylaminomethyl-2-thiouracil (Carbon, David & Studier, 1968), 2-thiocytidine (Carbon *et al.*, 1968), and N^6 -(3-methyl-2-butenyl)-2-methylthioadenosine (Burrows, Armstrong, Skoog, Hecht, Boyle, Leonard & Occolowitz, 1968) are found in several *E. coli* tRNAs, while 6-thioinosine and 6-thioguanosine are effective as enzyme inhibitors and antitumor agents (Elion & Hitchings, 1965). The function of the thio analogues of the pyrimidine and purine systems in tRNAs is not clear. The crystal structure of 4-thiouridine monohydrate (Saenger & Scheit, 1969) has recently been reported. The molecule was found to exhibit a most unusual *syn* conformation about the glycosidic bond, whereas in the crystal structure of 4-thiouridine disulfide (Shefter & Kalman, 1968) both the pyrimidine nucleosides were found to be in the normal *anti* conformation. The conformation about the glycosidic bond was found to be *anti* in the present work. In the crystal structure of 6-thioinosine (Shefter, 1968) the

two independent molecules in the asymmetric unit were found to display the *syn* conformation. Among these structures the present structure of 2,4-dithiouridine is by far the most precisely determined.*

Experimental

2,4-Dithiouridine monohydrate was provided by Professor Tohru Ueda of Hokkaido University, Japan. A large yellow prismatic crystal elongated along one of the major axes was crystallized from aqueous ethanol solution. A piece with dimensions $0.1 \times 0.05 \times 0.3$ mm was cut from the above crystal for X-ray analysis, and was mounted in a capillary tube with the crystallographic c axis coaxial to the φ axis of the

Table 1. *Crystal data for 2,4-dithiouridine monohydrate*

Formula	$C_9H_{12}N_2O_4S_2 \cdot H_2O$
Crystal dimensions	$0.1 \times 0.05 \times 0.3$ mm
Space group	$P2_12_12_1$
Lattice constants	$a = 27.992 \pm 0.002$ Å $b = 8.430 \pm 0.001$ Å $c = 5.254 \pm 0.001$ Å
Volume of unit cell	1240 Å ³
Molecular weight	294.327
Density	$D_{calc} = 1.569 \text{ g.cm}^{-3}$ $D_{obs} = 1.569 \text{ g.cm}^{-3}$
Z	4
μ	183 cm^{-1}

* The crystal structure of 2-thiocytidine dihydrate has now been completed in these laboratories (Lin, Sundaralingam & Arora, 1971). This structure also exhibits the *anti* conformation about the glycosidic bond. In addition the rare base of 2-methylthioisopentenyladenine has been completed (McMullan & Sundaralingam, 1971).

* Part XIII: Rao & Sundaralingam (1970). *J. Amer. Chem. Soc.* **92**, 4963

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Table 2. Observed and calculated structure amplitudes

Columns 1, 2 and 3 represent h, 10|F_o| and 10|F_c| respectively. Unobserved reflections are marked with * and reflections suffering from secondary extinction are marked with E.

A complex table with multiple columns of numerical data representing structure amplitudes. The data is organized in a grid-like format with various sub-headers (e.g., M_1, M_2, M_3) and labels (e.g., H, K, L) indicating different reflection conditions or phases. The table contains thousands of rows and columns of numbers, including observed values and calculated values, with some cells marked as unobserved or extinct.

goniostat. Oscillation and Weissenberg photographs showed orthorhombic symmetry with absent reflections $h00$ with $h=2n+1$ and $0k0$ with $k=2n+1$, and $00l$ (from diffractometer data) with $l=2n+1$, consistent with the space group $P2_12_12_1$.

The unit-cell dimensions are summarized in Table 1. There are four molecules in a unit cell. The density calculated for $C_9H_{12}N_2O_4S_2 \cdot H_2O$ is 1.569 g.cm^{-3} , while that measured by flotation method, using a mixture of chloroform and carbon tetrachloride, was also 1.569 g.cm^{-3} . This indicates that the crystals are of the monohydrate.

Intensity data were collected on a Picker four-circle automatic diffractometer, using Ni-filtered $Cu K\alpha$ radi-

ation ($\lambda=1.5418 \text{ \AA}$). Reflections with 2θ values up to 133° were collected by the $2\theta-\theta$ scan technique at a scan rate of 2° per min. A constant scan range of 3.40° in 2θ , from -1.70 to $+1.70^\circ$ from the calculated 2θ value, was used. Stationary-counter background counts of 20 seconds were taken at each end of the scan. Attenuators were inserted automatically when the intensity of the diffracted beam exceeded 10^4 counts per second during the scan. As a check on electronic and crystal stability during the time period of data collection, the intensity of a standard reflection was measured after about every 40 reflections. This showed a deviation of only 1% from the mean. Altogether, 1297 reflections were collected. Reflections with I greater than $1.2 \sigma(I)$ were considered as observed, where $\sigma(I) = [I_{\text{scan}} + I_{\text{background}}]^{1/2}$. The data were then corrected for Lorentz-polarization effects.

Determination of the structure

From the three Harker sections at $u=\frac{1}{2}$, $v=\frac{1}{2}$ and $w=\frac{1}{2}$ of a sharpened three-dimensional Patterson map, it was possible to locate the position of only one of the sulfur atoms. This atom was used as an initial input for the calculation of a three-dimensional electron density map. In this map other atoms of the molecule could not be clearly identified. Therefore, the phases derived from the sulfur atoms were refined by using the tangent formula (Karle & Hauptman, 1956). In the initial tangent phase refinement, about 50 reflections with normalized structure factors $E > 1.7$ were chosen. Subsequently, 282 reflections with $E > 1.2$ were used for the refinement. The refinement was computed for 10 cycles. At the end of refinement, the R value was 0.212, where R is defined by $R = \sum ||E_o| - |E_c|| / \sum |E_o|$. A three-dimensional E map was then computed. All atoms except hydrogen were clearly identified from the map.

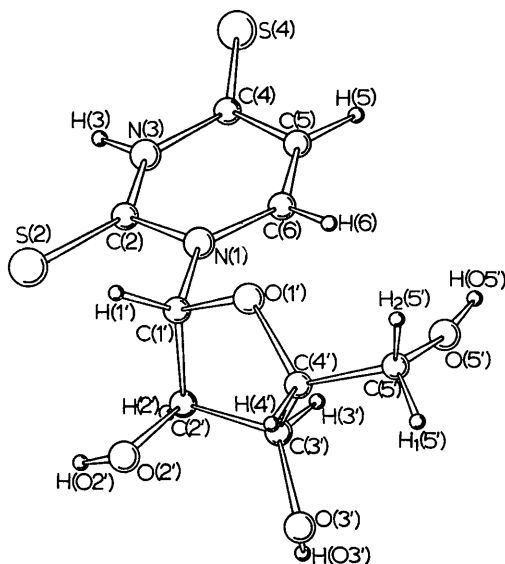


Fig. 1. Numbering of atoms in 2,4-dithiuridine

Table 3. Positional parameters* and anisotropic thermal parameters† of the nonhydrogen atoms

	x	y	z	β_{11}	β_{22}	β_{33}	β_{12}	β_{13}	β_{23}
S(2)	10940 (5)	1402 (2)	1843 (3)	16 (0)	90 (2)	545 (8)	95 (1)	-37 (1)	-26 (3)
S(4)	-2125 (4)	4890 (2)	-2508 (3)	10 (0)	133 (2)	468 (7)	-6 (1)	-21 (1)	41 (4)
N(1)	9205 (11)	4377 (4)	3361 (8)	7 (0)	97 (5)	291 (18)	30 (1)	-3 (2)	7 (9)
C(2)	8120 (14)	3123 (5)	1792 (11)	7 (1)	100 (7)	402 (26)	-20 (1)	-8 (3)	32 (12)
N(3)	4527 (12)	3400 (5)	118 (9)	8 (1)	93 (6)	381 (22)	-19 (1)	-8 (3)	18 (11)
C(4)	2090 (14)	4799 (5)	-291 (10)	6 (1)	117 (7)	387 (23)	10 (2)	0 (3)	40 (13)
C(5)	3598 (16)	6052 (6)	1304 (12)	8 (1)	107 (8)	406 (26)	67 (2)	6 (3)	12 (12)
C(6)	6961 (15)	5827 (6)	3074 (12)	8 (1)	100 (7)	376 (26)	54 (2)	4 (4)	10 (13)
C(1')	12833 (15)	4113 (6)	5396 (11)	10 (1)	87 (7)	324 (24)	1 (2)	-3 (3)	15 (11)
C(2')	18030 (16)	4220 (6)	4454 (12)	8 (1)	117 (8)	276 (24)	69 (2)	-6 (2)	-14 (12)
C(3')	19190 (16)	5941 (6)	5032 (10)	8 (1)	121 (8)	217 (22)	16 (2)	1 (3)	5 (11)
C(4')	16630 (15)	6233 (6)	7518 (11)	9 (1)	92 (7)	299 (21)	17 (2)	1 (3)	-19 (13)
O(1')	12248 (9)	5296 (3)	7303 (7)	9 (0)	103 (5)	325 (15)	-24 (1)	11 (2)	-11 (8)
O(2')	20945 (14)	3232 (5)	5973 (9)	13 (1)	115 (6)	443 (21)	130 (16)	-25 (3)	-27 (10)
O(3')	24060 (10)	6321 (5)	5334 (8)	7 (0)	183 (7)	385 (19)	-53 (1)	6 (2)	-43 (11)
C(5')	15332 (19)	7918 (6)	8160 (13)	11 (1)	112 (8)	432 (30)	13 (2)	3 (4)	-15 (14)
O(5')	13654 (15)	8711 (5)	5942 (9)	17 (1)	128 (7)	502 (22)	172 (2)	5 (3)	48 (11)
O(W)	22530 (17)	222 (5)	4196 (11)	16 (1)	134 (7)	638 (28)	13 (2)	-3 (4)	2 (12)

* Positional parameters and their standard deviations in parentheses, x values have been multiplied by 10^5 , while y and z values have been multiplied by 10^4 .

† Anisotropic thermal parameters and their standard deviations in parentheses have been multiplied by 10^4 . The temperature factor is of the form $\exp [-(\beta_{11}h^2 + \dots + 2\beta_{12}hk + \dots)]$.

Refinement of the structure

Four cycles of isotropic full-matrix least-squares refinement on all the 18 non-hydrogen atoms followed by one cycle of anisotropic refinement reduced the R value, which is defined by $R = \sum ||F_o| - |F_c|| / \sum |F_o|$, from 0.208 to 0.064. A difference electron density map computed at this stage gave the positions of all except the O(5') and water hydrogen atoms. The atomic parameters were subjected to two more cycles of refinement with anisotropic temperature factors for non-hydrogen atoms and isotropic temperature factors for hydrogen atoms. A difference Fourier synthesis calculated at this stage revealed the positions of the remaining hydrogen atoms. The refinement was terminated after two more cycles. The final R value is 0.039 for the observed reflections, and the final shift/ σ ratio is less than 0.096 for all parameters refined.

The scattering factors for S, O, N and C atoms used throughout the analysis were those of Cromer & Waber (1965). The scattering factors for H were those of Stewart, Davidson & Simpson (1965). The weighting scheme used in the refinement was that of Evans (1961).

Results and discussion

A list of the observed and calculated structure amplitudes is given in Table 2. The final positional parameters and

thermal parameters for non-hydrogen and hydrogen atoms are shown in Tables 3 and 4, respectively. The intramolecular bond lengths and bond angles involving non-hydrogen atoms are given in Table 5. All distances and angles involving hydrogen atoms are normal.

Table 4. *Positional parameters and isotropic thermal parameters of hydrogen atoms**

	$x(\times 10^4)$	$y(\times 10^3)$	$z(\times 10^3)$	$B(\text{\AA}^2)$
H(3)	365 (15)	256 (5)	-103 (9)	3 (1)
H(5)	207 (19)	705 (7)	137 (12)	6 (2)
H(6)	802 (15)	666 (6)	441 (10)	4 (1)
H(1')	1214 (16)	299 (6)	622 (10)	4 (2)
H(2')	1820 (15)	396 (5)	273 (10)	3 (1)
H(3')	1756 (15)	655 (6)	360 (10)	4 (1)
H(4')	1867 (16)	586 (6)	895 (10)	4 (1)
H ₁ (5')	1300 (17)	792 (6)	960 (10)	4 (1)
H ₂ (5')	1825 (14)	837 (5)	884 (9)	3 (1)
H(02')	2113 (21)	247 (7)	544 (14)	5 (2)
H(03')	2532 (19)	645 (8)	364 (13)	6 (2)
H(05')	1172 (23)	927 (9)	696 (16)	12 (2)
H ₁ (W)	2505 (31)	-14 (12)	520 (15)	13 (3)
H ₂ (W)	2009 (22)	-29 (8)	383 (16)	10 (2)

* Standard deviations are given in parentheses.

Bond distances and bond angles

The base

Again, as found in other thio bases (Lin, Sundaralingam & Arora, 1971) the thione form of the base is preferred to the mercapto form. Thus, at least in the

Table 5. *Bond lengths (Å) and bond angles (°)* not involving hydrogen atoms in 2,4-dithiouridine and 2,4-dithiouracil*

Bond lengths			Bond angle				
	2,4-Dithio- uridine	2,4-Dithio- uracil	$ d /\sigma_{12}$		2,4-Dithio- uridine	2,4-Dithio- uracil	$ d /\sigma_{12}$
C(2)—S(2)	1.653 (5)	1.645 (6)	0.9	C(2)—N(1)—C(6)	120.9 (0.3)	126.6 (0.5)	9.8
C(4)—S(4)	1.660 (5)	1.684 (6)	3.1	C(2)—N(1)—C(1')	117.8 (0.3)		
N(1)—C(2)	1.374 (6)	1.342 (8)	3.2	C(6)—N(1)—C(1')	121.3 (0.3)		
C(2)—N(3)	1.356 (6)	1.406 (7)	5.4	N(1)—C(2)—N(3)	114.9 (0.3)	112.4 (0.5)	4.3
N(3)—C(4)	1.379 (6)	1.358 (7)	2.3	N(1)—C(2)—S(2)	124.1 (0.3)	125.9 (0.5)	3.1
C(4)—C(5)	1.411 (7)	1.414 (8)	0.3	N(3)—C(2)—S(2)	121.1 (0.3)	121.7 (0.4)	1.2
C(5)—C(6)	1.340 (8)	1.365 (9)	2.1	C(2)—N(3)—C(4)	128.0 (0.3)	125.5 (0.5)	4.3
C(6)—N(1)	1.383 (6)	1.360 (8)	2.3	N(3)—C(4)—C(5)	113.7 (0.3)	117.4 (0.5)	6.3
N(1)—C(1')	1.492 (6)			N(3)—C(4)—S(4)	120.0 (0.3)	119.3 (0.4)	1.4
O(1')—C(1')	1.424 (6)			C(5)—C(4)—S(4)	126.3 (0.3)	123.2 (0.4)	6.2
C(1')—C(2')	1.539 (7)			C(4)—C(5)—C(6)	121.0 (0.4)	118.9 (0.5)	3.2
C(2')—C(3')	1.518 (7)			C(5)—C(6)—N(1)	121.4 (0.4)	119.2 (0.5)	3.4
C(3')—C(4')	1.510 (7)			N(1)—C(1')—C(2')	113.8 (0.4)		
C(4')—O(1')	1.462 (5)			N(1)—C(1')—O(1')	108.8 (0.3)		
C(2')—O(2')	1.413 (7)			O(1')—C(1')—C(2')	107.1 (0.3)		
C(3')—O(3')	1.410 (5)			C(1')—C(2')—C(3')	101.1 (0.4)		
C(4')—C(5')	1.505 (7)			C(1')—C(2')—O(2')	109.2 (0.4)		
C(5')—O(5')	1.423 (8)			C(3')—C(2')—O(2')	109.1 (0.4)		
				C(2')—C(3')—C(4')	103.1 (0.4)		
				C(2')—C(3')—O(3')	116.5 (0.4)		
				C(4')—C(3')—O(3')	108.9 (0.3)		
				C(3')—C(4')—O(1')	104.1 (0.3)		
				C(3')—C(4')—C(5')	117.5 (0.4)		
				O(1')—C(4')—C(5')	108.9 (0.3)		
				C(4')—O(1')—C(1')	109.6 (0.3)		
				C(4')—C(5')—O(5')	109.6 (0.4)		

* Standard deviations of bond lengths and bond angles are given in parentheses; those for bond lengths have been multiplied by 10³.

† $|d| = |l_1 - l_2|$, $\sigma_{12} = (\sigma_1^2 + \sigma_2^2)^{1/2}$; l_1 and l_2 are bond lengths or bond angles and σ_1 and σ_2 are standard deviations for 2,4-dithiouridine and 2,4-dithiouracil, respectively.

pyrimidine derivatives, the keto (thione) tautomeric form is preferred.

The bond distances and bond angles in 2,4-dithiouracil (Shefter & Mautner, 1967) are also given for comparison with the base in the nucleoside, Table 5. There are significant differences ($> 3\sigma$) in the bonds C(4)–S(4), N(1)–C(2) and C(2)–N(3). The difference in the N(1)–C(2) bond is in part attributable to the steric and electronic influence of the ribose attached to the N(1) position. Most of the bond angles are significantly different in the two structures.

The C–S bond lengths of 1.653 and 1.660 Å have mainly double bond character since they are much shorter than a C–S single bond of 1.82 Å but closer to a C–S double bond of 1.56 Å. In 2,4-dithiouracil the C–S bond lengths differ markedly, about 0.04 Å; the C(4)–S(4) bond is again the longer bond. The C(4)–S(4) bond in 2,4-dithiouridine is significantly shorter than the values found in 4-thiouridine, 1.679 Å, and 2,4-dithiouracil, 1.684 Å.

The C(1')–N(1) glycosidic bond length of 1.492 Å is similar in magnitude to those found in other structures.

The bond angles in the base are quite similar to the values observed in 4-thiouridine (Saenger & Scheit, 1969), thymidine (Young, Tollin & Wilson, 1969) and the other 5-substituted bases such as 5-methyluridine (Hunt & Sabramanian, 1969), 5-fluoro-2'-deoxyuridine (Harris & MacIntyre, 1964) and 5-chlorouridine (Coulter, 1969). The exocyclic angles involving the C–S bond are remarkably close to the values found in the corresponding keto compounds.

The ribose

Bond lengths and bond angles in the ribose moiety of 2,4-dithiouridine are within the usual range (Sundaralingam, 1965; Sundaralingam & Jensen, 1965*b*). The

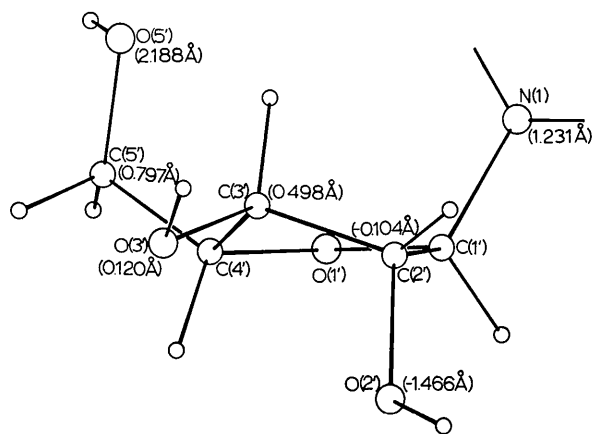


Fig. 2. A view of the ribose showing the twist of the C(2')–C(3') bond with respect to C(1')O(1')C(4') plane and deviations of atoms from plane. The puckering is C(3')-endo-C(2')-exo. Equation of plane: $-0.0123x + 0.0754y - 0.131z = -0.993$. For the best four atom plane, the equation of plane is $-0.0110x + 0.0743y - 0.136z = -0.990$ and the deviations from plane are: C(1') 0.022, C(2') -0.013, C(3') 0.578, C(4') 0.014 and O(1') -0.023 Å (average e.s.d. = 0.005 Å).

exocyclic C(4')–C(5') distance of 1.505 Å is significantly shorter than the normal C_{sp^3} – C_{sp^3} bond distance. This shortening is correlated with the widening of the exocyclic angle C(3')–C(4')–C(5'). The intra-ring C–O bonds are significantly (by $\sim 6\sigma$) different, C(4')–O(1') $>$ C(1')–O(1'), as found earlier (Sundaralingam, 1965).

Conformation of the nucleoside

Glycosidic torsional angle

The torsional angle χ about the glycosidic bond, defined as the clockwise rotation angle (Sundaralingam, 1969) of the C(6)–N(1) bond with reference to the C(1')–O(1') bond is 19.5°. Therefore, it belongs to the *anti* conformation which is typical of the pyrimidine nucleosides and nucleotides (Sundaralingam, 1969). 4-Thiouridine is the only known pyrimidine nucleoside in the *syn* conformation (Saenger & Scheit, 1969).

Planarity of the base

The glycosidic linkage was found to be β as expected. The least-squares plane through the six atoms comprising the pyrimidine ring shows that the ring atoms are generally displaced quite significantly from the plane (see Table 6). The largest displacements of 0.026 and 0.018 Å are exhibited by atoms C(2) and C(5) respectively. The remaining ring atoms are displaced on the opposite side of the plane, thus the ring is slightly folded along the C(2)–C(5) axis giving it a shallow boat shape. The dihedral angle between the two halves of the pyrimidine ring C(2)N(1)C(6)C(5) and C(2)N(3)C(4)C(5) is 3.2°. S(2) and S(4) are displaced on opposite sides of the ring, the former being displaced by about seven times as much as the latter. The C(2)–S(2) bond makes an angle of 5° with the plane of the pyrimidine ring. The sulfur atom S(4) is displaced by 0.178 Å from the pyrimidine plane in 4-thiouridine (Saenger & Scheit, 1969).

Table 6. Deviations of atoms from the least-squares plane of the base*†

	Deviation from plane	E.s.d.
N(1)	-0.017 Å	0.004 Å
C(2)	0.026	0.005
N(3)	-0.014	0.004
C(4)	-0.008	0.005
C(5)	0.018	0.005
C(6)	-0.005	0.005
S(2)	0.143	
S(4)	-0.021	
C(1')	-0.095	

* Equation of plane: $0.0246x + 0.0335y - 0.127z = 1.653$. The equation to the plane is in the form $lx + my + nz = d$ where l , m , n are the direction cosines of the normal to the plane; d is the distance of the plane from the origin in Å; and x , y , z are fractional coordinates.

† Atoms N(1), C(2), N(3), C(4), C(5) and C(6) were used in the calculation of the plane.

The ribose carbon atom C(1') is also significantly displaced from the base plane and on the opposite side of S(2) as found in other pyrimidine nucleosides.

It is interesting to note that the aromatic benzene ring and its derivatives, in the absence of overcrowded substituents, is always found to be planar, while the conjugated pyrimidine ring is often not planar.

Ribose conformation

All possible planes comprising four atoms of the furanose ring of the ribose moiety were computed. It was found that atoms C(1'), C(2'), C(4') and O(1') define the best least-squares plane (see Fig. 2). C(3') is displaced 0.578 Å out of this plane on the same side of C(5'). The conformation of the sugar is thus referred to as C(3') *endo*. This type of conformation is commonly observed in nucleosides (Spencer, 1959; Sundaralingam, 1965). The atoms in the best four-atom plane show significant deviations from the plane. The displacements of C(2') and C(3') from the plane defined by C(1'), O(1') and C(4') were also computed. With respect to this plane the ribose conformation is C(3')-*endo*-C(2')-*exo*. A view of the ribose showing the twist of the C(2')-C(3') bond with respect to C(1') O(1')C(4') plane and the deviations of atoms from the plane is shown in Fig. 2.

The conformation of the sugar ring is best described by the torsional angles τ_0 , τ_1 , τ_2 , τ_3 and τ_4 (Sundaralingam, 1969) about the ring bonds which are 4.1, -25.9, 37.2, -35.9 and 19.9°.

The dihedral angle between the least-squares plane of the pyrimidine ring and that of the ribose ring excluding C(3') is 63.6°. This angle is close to the value in cytidine-3'-phosphate (62°) (Sundaralingam & Jensen, 1965a). However, it is quite different from that in 5-fluoro-2'-deoxy- β -uridine (71.9°) (Harris & Macintyre, 1964), 5-methyluridine (71.9°) (Hunt & Sabramanian, 1969), 4-thiouridine (98°) (Saenger & Scheit, 1969) and cytidine (75°) (Furberg, 1950).

The torsional angles about the exocyclic C(4')-C(5') bond denoted by φ_{OO} [O(1')-C(4')-C(5')-O(5')] and φ_{CO} [C(3')-C(4')-C(5')-O(5')] (Shefter & Trueblood, 1965) are -77.2° and 40.7° respectively. Therefore, the conformation about C(4')-C(5') is *gauche-gauche* which is the most preferred conformation (Sundaralingam, 1965; Shefter & Trueblood, 1965).

The torsional angles for H(2')-C(2')-O(2')-H(O2') and H(3')-C(3')-O(3')-H(O3') are -31.5 and -38.5°, respectively. The *gauche* conformation of the H-C-O-H bond sequence appears to be a preferred conformation (Sundaralingam, 1968).

Base 'stacking' and sugar-base contacts

Projections perpendicular to the base plane and parallel to the base plane showing the base 'stacking'* and sugar-base contacts are given in Fig. 3(a) and (b),

* Here defined as the parallel arrangement of adjacent bases, not necessarily involving overlap of base rings.

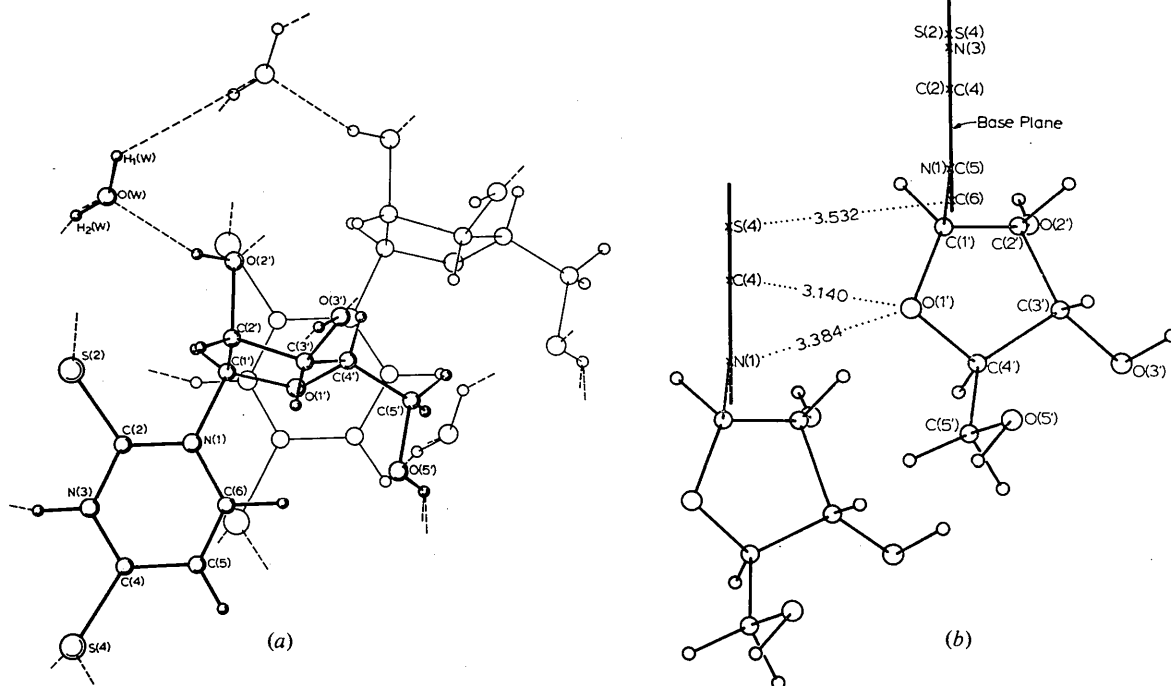


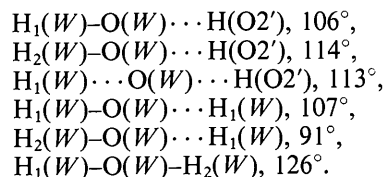
Fig. 3. Projections (a) perpendicular (b) parallel to the base plane showing the lack of base stacking and the shortest sugar-base distance and the hydrogen bonding environs. The oxygen of the ribose ring points into the pyrimidine ring of an adjacent molecule, a feature found in many of the known nucleoside and nucleotide structures (Sundaralingam, 1968; Shefter, Barlow, Sparks, & Trueblood, 1969).

respectively. There is little contact between adjacent bases. However, the contact between the base and the ribose ring is noteworthy. The vertical distance between the base planes is 3.52 Å. The shortest distance between parallel bases is 3.532 Å for S(4)···C(6). The base 'stacking' conformation observed here is quite different from those typically observed in the analogous keto bases (Sundaralingam, Rao & Bugg, 1969; Bugg, Thomas, Sundaralingam & Rao, 1971). The shortest distance between the sugar and base is O(1')···C(4) of 3.140 Å. Similar contacts involving the sugar ring oxygen O(1') and the base appear to compete with base stacking in the known nucleoside and nucleotide structures. The only known dinucleoside phosphate structure, adenosine-2',5'-uridine phosphoric acid (Shefter, Barlow, Sparks & Trueblood, 1969) also shows a similar approach distance. It would be interesting to know whether the hyperchromicity normally associated with base-base and hydrogen bonding interactions in nucleic acid constituents may also be partly due to the basesugar interaction.

Hydrogen bonding

The intermolecular hydrogen bond scheme is illustrated by broken lines in Fig. 4. The detailed hydrogen bond distances and angles are given in Table 7. The

2,4-dithiouridine molecule forms nine hydrogen bonds with its neighboring molecules and with water molecules. The twofold screw related water molecules form hydrogen bonds with one another, thus generating an infinite twofold helix of water molecules in the crystallographic *c* direction. The environment of the water oxygen is approximately tetrahedral. The angles involved are:



The atoms S(4), S(2), O(2') and O(5') act as acceptors and atoms N(3), O(2') and O(5') act as donors in hydrogen bonds. S(4) is hydrogen bonded to N(3), the distance S(4)···N(3)=3.33 Å and S(4)···H(3)=2.41 Å. S(4) is displaced by 0.4 Å from the hydrogen bonded base plane. The O(5')···S(4) and O(5')···S(2) distances are 3.476 Å and 3.218 Å respectively. Although the O(5')···S(4) distance is longer than the sum of the van der Waals radii of O and S (3.25 Å), the observed H(O5')···S(4) distance of 2.75 Å is shorter than the sum of van der Waals radii of H and S (3.05 Å). This could be an indication of a weak hydrogen bond

Table 7. *Hydrogen bonds and angles*

Atom A	Atom B	Symmetry code	Distance	Bond	Angle
S(2)	O(5')	IV	3.218 Å	C(2)-S(2)·····O(5')	137.9°
				C(5')-O(5')·····S(2)	163.1
S(2)	H(O5')	IV	3.24*	C(2)-S(2)·····H(O5')	122
				O(5')-H(O5')·····S(2)	81
S(4)	N(3)	VII	3.330	C(4)-S(4)·····N(3)	118.3
				C(2)-N(3)·····S(4)	105.3
				C(4)-N(3)·····S(4)	126.6
S(4)	H(3)	VII	2.41	C(4)-S(4)·····H(3)	113
				N(3)-H(3)·····S(4)	158
S(4)	O(5')	VI	3.476	C(4)-S(4)·····O(5')	118.7
				C(5')-O(5')·····S(4)	104.3
S(4)	H(O5')	VI	2.75	C(4)-S(4)·····H(O5')	128
				O(5')-H(O5')·····S(4)	139
O(2')	O(3')	V	2.710	C(2')-O(2')·····O(3')	133.9
				C(3')-O(3')·····O(2')	115.8
O(2')	H(O3')	V	1.77	C(2')-O(2')·····H(O3')	134
				O(3')-H(O3')·····O(2')	165
O(5')	O(W)	II	2.939	C(5')-O(5')·····O(W)	100.4
O(5')	H ₂ (W)	II	2.28	C(5')-O(5')·····H ₂ (W)	108
				O(W)-H ₂ (W)·····O(5')	137
O(W)	O(W)	III	2.992		
O(W)	H ₁ (W)	III	2.21	O(W)-H ₁ (W)·····O(W)	141
O(W)	O(2')	I	2.740	C(2')-O(2')·····O(W)	116.5
O(W)	H(O2')	I	2.04	see text	
Symmetry codes:		I	<i>x</i>	<i>y</i>	<i>z</i>
		II	<i>x</i>	<i>y</i> +1	<i>z</i>
		III	$\frac{1}{2}$ - <i>x</i>	\bar{y}	<i>z</i> - $\frac{1}{2}$
		IV	<i>x</i>	<i>y</i> -1	<i>z</i>
		V	$\frac{1}{2}$ - <i>x</i>	1- <i>y</i>	$\frac{1}{2}$ + <i>z</i>
		VI	\bar{x}	<i>y</i> - $\frac{1}{2}$	$\frac{1}{2}$ - <i>z</i>
		VII	\bar{x}	<i>y</i> + $\frac{1}{2}$	$-\frac{1}{2}$ - <i>z</i>

* This is for the observed position of H(O5'), see text for discussion.

between O(5') and S(4). Because of the errors in the hydrogen atom H(O5') position, it was of interest to calculate an assumed position for H(O5') using an O-H distance of 1.00 Å and a C(5')-O(5')-H(O5') angle of 109° along the direction of the observed torsional angle C(4')-C(5')-O(5')-H(O5') 147.2°. Using this position for H(O5'), the recalculated hydrogen bond distances and angles are:

- S(2)···H(O5'), 2.71 Å;
 S(4)···H(O5'), 2.64 Å;
 O(5')-H(O5')···S(2), 112°;
 O(5')-H(O5')···S(4), 141° (Fig. 5).

These values suggest that O(5') may be involved in a bifurcated hydrogen bond to S(2) and S(4) (Donohue, 1968). However, this conclusion should be considered tentative. O(5') and H(O5') are displaced 1.4 and 2.0 Å respectively from the base plane.

O(2') is an acceptor from O(3') of a screw-related molecule, and O(3') is a donor to O(2') of another screw-related molecule. A right-handed helix of hydrogen bonds about the screw axis in the *c* direction is formed by the ribose hydroxy groups. This helix is linked to the helix of water molecules through the hydrogen bond involving the hydrogen atom on O(2') and the non-bonded electron pair of the water, while the remaining hydrogen atom of the water is involved in a donor hydrogen bond to O(5') of the ribose.

Some van der Waals distances are shown by dotted lines in Fig. 4. The shortest intermolecular S···S distance involving S(4) and S(2) is 3.59 Å. The distance between S(4) and H(5) is 3.14 Å.

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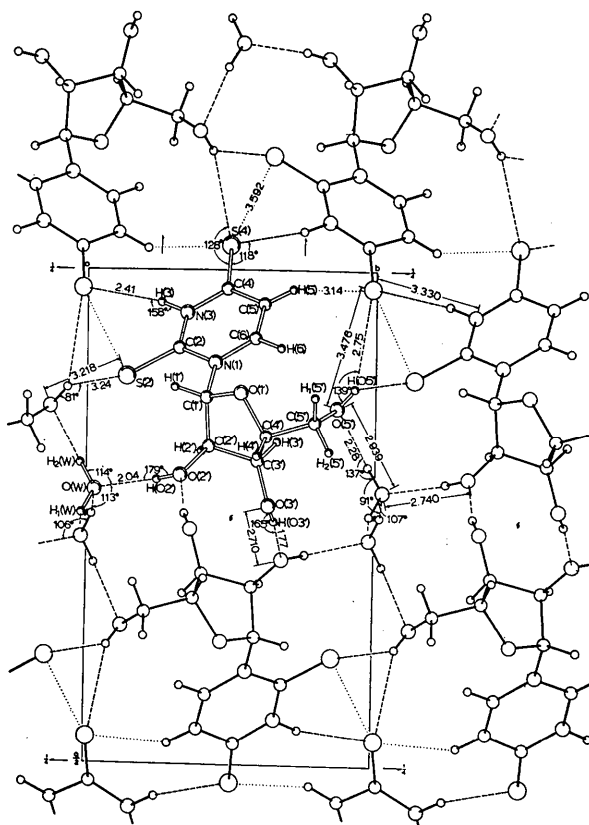


Fig. 4. Intermolecular hydrogen bonding scheme (dashed lines) and some van der Waals distances (dotted lines).

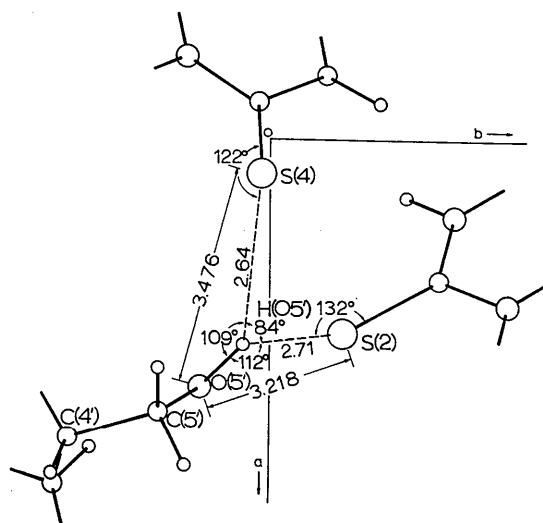


Fig. 5. Hydrogen bond distances and angles between O(5') and S(2) and between O(5') and S(4) using calculated coordinates for H(O5').

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The Crystal Structure of the 1:1 Solvate of 2-Propanol and 1,1',3,3'-Tetraethylimidazo[4,5-*b*]quinoxalinocyanine Iodide

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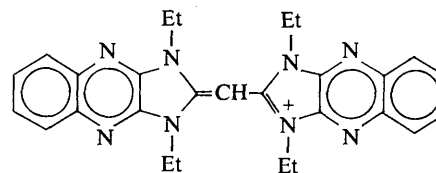
(Received 6 July 1970)

The crystal structure of the title compound, $C_{27}H_{29}N_8I \cdot C_3H_8O$, has been determined from single-crystal X-ray intensity data collected with an automated diffractometer. The crystals are orthorhombic, *Pcab*, with $a = 18.337$ (12), $b = 21.855$ (14), $c = 15.485$ (11) Å, and $D_x = 1.397$ g.cm⁻³ for $Z = 8$. The structure was solved by the heavy-atom technique and refined by least squares to $R = 0.082$. Because of steric interference between ethyl substituents, the cation is not planar but can be divided into a central and two end planes. The dihedral angles are 55.4° between the end planes and 35.3° and 21.0° between the central and end planes. Several angular distortions of about 5° help to relieve the strain but the crowded methylene carbon atoms are still only 3.28 Å apart. Bond lengths show that despite the distortions the cation is highly conjugated. The cations are packed end to end in chains with their end planes almost parallel and 3.4 Å apart.

Introduction

The influence of molecular overcrowding on the optical properties of cyanine dyes was recognized almost thirty years ago (Sheppard, Lambert & Walker, 1941) and has been investigated extensively since (Brooker, White, Heseltine, Keyes, Dent & Van Lare, 1953; Brooker, 1966; Dewar, 1958). In spite of this interest, few crystal structures have been reported. A partially refined structure of an unsymmetrical 'simple' cyanine with an overcrowded cation has been reported (Efinger, Germain, Meunier, Vanderauwera & Van Meerssche, 1960). The structures of two phosphocyanines analogous to the simple cyanines have been determined (Allmann, 1966; Kawada & Allmann, 1968). The other structures that have been published are four similar unsymmetric merocyanines (Germain, Piret, Van Meerssche & DeKerf, 1961, 1962; Germain, Patternotte, Piret & Van Meerssche, 1964; Bois

D'Enghien-Peteau, Meunier-Piret & Van Meerssche, 1968) and a symmetric carbocyanine and two of its solvates (Wheatley, 1959*a, b*; Vorontsova, Zvonkova & Zhdanov, 1963). To learn more about the conformation of overcrowded cyanines we have determined the structure of the dye 1,1',3,3'-tetraethylimidazo[4,5-*b*]quinoxalinocyanine iodide,



Crystal data

$(C_{27}H_{29}N_8)^+I^- \cdot C_3H_8O$ M.W. 652.59.
1,1',3,3'-Tetraethylimidazo[4,5-*b*]quinoxalinocyanine iodide-2-propanol.