The crystal structure features an intermolecular O(2)···HO(3^{vi}) hydrogen bond, 2·669 (7) Å. The O···H–O angle is 175°, whereas that in the intramolecular hydrogen bond is 135°. Though the positions of the H atoms have not been defined with great accuracy, the latter angle appears to deviate significantly from 180°.

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References

BRAIN, E. G., CASSIDY, F., CONSTANTINE, M. F., HANSON, J. C. & TIDY, D. J. D. (1971). *J. Chem. Soc. C*, pp. 3846–3851.

CRUICKSHANK, D. W. J. (1957). *Acta Cryst.* **10**, 504–508.

HALL, S. R. & MASLEN, E. N. (1965). *Acta Cryst.* **18**, 265–279.

KARLE, J. & KARLE, I. L. (1966). *Acta Cryst.* **21**, 849–859.

KERR, K. A. (1966). PhD Thesis, Univ. of Glasgow.

MUIR, K. W. & SIM, G. A. (1968). *J. Chem. Soc. B*, pp. 667–673.

WEEKS, C. M., POKRYWIECKI, S. & DUAX, W. L. (1973). *Acta Cryst.* B**29**, 1729–1731.

Acta Cryst. (1978). B**34**, 2204–2208

Crystal Structure and Conformation of 5-Aminouridine

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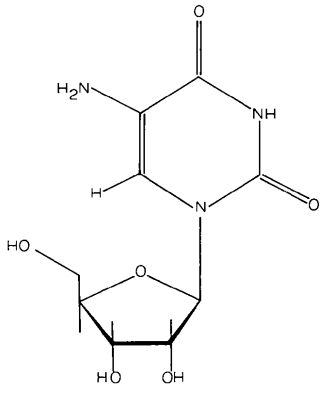
$C_9H_{13}N_3O_6$ is orthorhombic, space group $P2_12_12_1$, with a = 16·565 (8), b = 12·341 (6), c = 5·268 (5) Å, Z = 4. The structure, which was refined to R = 0·076, reveals the influence of a π-electron-donating 5-substituent, not only on the base geometry but also on the N(1)–C(1′) and C(1′)–O(1′) lengths, which is discussed in comparison with other 5-substituted uridines. The orientation of the base at the glycosidic bond is *anti* (χ = 61·0°); the ribosyl moiety shows a C(2′)-*endo* conformation and a *gauche–gauche* arrangement of C(4′)–C(5′). These conformational features are stabilized by the characteristic intramolecular interaction C(6)–H···O(5′). The bases are stacked along c with a short base–base distance of 3·30 Å. Two base stacks are nearly perpendicular to each other and are connected by interbase hydrogen bonds N(5)–H···O(4) leading to a helical arrangement.

5-Aminouridine (a^5Urd) has a wide range of biological effects and inhibits the growth of tumours and viruses. It is incorporated into the DNA of Ehrlich ascites cells (Werkheiser, Winzler & Visser, 1955). The homopolynucleotide poly(a^5U), comparable to polyuridylate, stimulates the binding of Phe-tRNA to bacterial ribosomes and promotes polyphenylalanine synthesis in the same system. Trinucleoside diphosphates, however, like U-U-a^5U containing the 5-aminouridine in the 3′-terminal position (the wobble position) do not stimulate the binding of their cognate tRNA in a Nirenberg–Leder assay system (Hillen & Gassen, 1975). This may be taken as evidence that no aminouridine–guanosine base pair is formed in the triplet coded binding of a tRNA. Furthermore, oligonucleotides terminating in 5-aminouridine have been used as affinity labels with iodoacetyl chloride as bridging agent to identify the proteins of the aminoacyl site on ribosomes (Luehrmann & Gassen, 1976).

In our attempt to clarify the function of 5-substitution on the structure of nucleosides and oligonucleotides, we now report the structure of 5-aminouridine. The comparison with 5-nitrouridine (–M), whose structure has been reported (Egert, Lindner, Hillen & Gassen, 1977*a*), demonstrates the (+M) effect of a π-electron-donating substituent.



Experimental

1-β-D-Ribofuranosyl-5-aminouracil was synthesized by a known procedure (Visser, 1968). Crystals were grown from water as clusters of thin, light-yellow needles. Weissenberg photographs showed them to be orthorhombic with the systematic absences $h00$, $0k0$ and $00l$ for odd indices determining the space group as $P2_12_12_1$. Crystal data are summarized in Table 1. Intensities were collected on a Stoe two-circle diffractometer (Cu $K\alpha$ radiation) equipped with a graphite monochromator; the crystal was orientated along c. 849 symmetry-independent reflections $hk0$ to $hk3$ with $\theta < 60°$ were measured in the θ–2θ scan mode. The data were corrected for background and for Lorentz and polarization factors, but not for absorption.

Structure determination and refinement

The structure was solved with *SHELX* 76 (Sheldrick, 1976) which uses scattering factors from *International Tables for X-ray Crystallography* (1974).

Isotropic refinement with unit weights including 749 reflections having $|F| > 3\sigma_F$ reduced R to 0·123, which then dropped to 0·103 with inclusion of anisotropic temperature factors. At this stage seven of the thirteen H atoms were located by a difference synthesis. Weights $w = 1/\sigma(|F|)^2$ were introduced, and further refinement with fixed H atom parameters yielded the positions of the six remaining H atoms and finally converged at an R of 0·076. None of the positional parameters (Table 2) shifted more than $0{\cdot}03\sigma$ in the last cycle.*

Table 1. *Crystal data*

Molecular formula: $C_9H_{13}N_3O_6$	$V = 1076{\cdot}9$ Å^3
$M_r = 259$	$Z = 4$
Space group: $P2_12_12_1$	$\rho_0 = 1{\cdot}59$ g cm^{-3} (flotation)
$a = 16{\cdot}565$ (8) Å	$\rho_c = 1{\cdot}597$
$b = 12{\cdot}341$ (6)	$\mu = 10{\cdot}6$ cm^{-1}
$c = 5{\cdot}268$ (5)	

Results and discussion

Structure of the base

The base geometry (Fig. 1) agrees well with that of 5-hydroxyuridine (Thewalt & Bugg, 1973) which has a 5-substituent with similar electronic properties. The bond lengths are nearly identical, with the exception that N(1)–C(6) is longer by 0·026 Å in 5-aminouridine. The bond angles also show good agreement.

The pyrimidine ring is, within experimental error, planar with a standard deviation of the ring atoms of $\sigma = 0{\cdot}011$ Å. Of all substituents, only O(4) lies in the plane whereas N(5) (0·127 Å), O(2) (0·052 Å) and C(1′) (−0·046 Å) show perceptible deviations. Table 3 shows a comparison of the bond lengths of 5-aminouridine with those of uridine (Green, Rosenstein,

* Lists of structure factors and anisotropic thermal parameters have been deposited with the British Library Lending Division as Supplementary Publication No. SUP 33419 (7 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

Table 2. *Positional parameters and their e.s.d.'s*

	x	y	z
N(1)	0·5559 (3)	0·3528 (4)	0·725 (2)
C(2)	0·6259 (4)	0·2994 (5)	0·669 (2)
O(2)	0·6326 (3)	0·2340 (4)	0·491 (1)
N(3)	0·6900 (3)	0·3222 (5)	0·828 (2)
C(4)	0·6922 (4)	0·3944 (5)	1·029 (2)
O(4)	0·7537 (3)	0·4078 (4)	1·147 (1)
C(5)	0·6173 (4)	0·4513 (6)	1·071 (2)
N(5)	0·6172 (4)	0·5307 (5)	1·251 (1)
C(6)	0·5539 (4)	0·4294 (6)	0·925 (2)
C(1′)	0·4843 (4)	0·3336 (6)	0·581 (2)
C(2′)	0·4103 (4)	0·2958 (5)	0·707 (2)
C(3′)	0·3439 (4)	0·3297 (5)	0·537 (2)
C(4′)	0·3739 (5)	0·4392 (6)	0·442 (2)
O(1′)	0·4617 (3)	0·4333 (4)	0·461 (1)
O(2′)	0·4154 (3)	0·1805 (4)	0·752 (1)
O(3′)	0·3392 (3)	0·2583 (4)	0·324 (1)
C(5′)	0·3439 (5)	0·5380 (7)	0·576 (2)
O(5′)	0·3543 (4)	0·5247 (5)	0·838 (2)
H(N3)	0·740	0·296	0·78
H(N5)	0·664	0·540	1·38
H′(N5)	0·565	0·542	1·31
	0·504	0·464	0·96
H(C1′)	0·503	0·267	0·46
H(C2′)	0·400	0·337	0·92
H(C3′)	0·287	0·338	0·60
H(C4′)	0·362	0·452	0·28
H(O2′)	0·353	0·168	0·80
H(O3′)	0·389	0·236	0·35
H(C5′)	0·277	0·547	0·51
H′(C5′)	0·376	0·611	0·53
H(O5′)	0·359	0·604	0·90

Shiono, Abraham, Trus & Marsh, 1975), 5-nitrouridine the 5-substituent of which has opposite electronic properties, and other 5-substituted uridines (Hawkinson & Coulter, 1971; Hunt & Subramanian, 1969; Hillen, Egert, Lindner & Gassen, 1978). It appears that the π-electron-accepting nitro group shortens N(1)–C(6) and lengthens C(5)–C(6) because of a push–pull interaction between N(1) and the nitro substituent. On the other hand, this delocalization of the lone pair of N(1) pushes the keto–enol tautomerism of the 2-carbonyl group back, so that N(1)–C(2) is longer and C(2)–O(2) shorter than in uridine. For the π-donating 5-amino group the influence on the bond lengths is converse, but less pronounced because of the more limited π-electron delocalization. The bond lengths of the other 5-substituted uridines are between these two extremes.

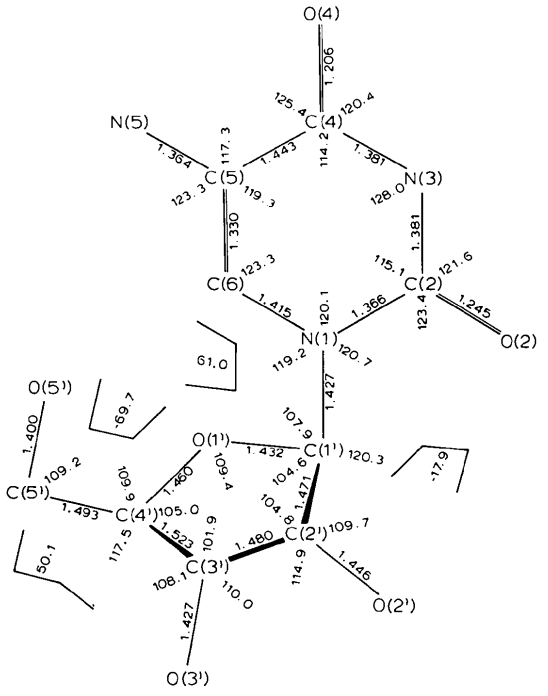


Fig. 1. The bond lengths, and bond and dihedral angles of 5-aminouridine ($\sigma_{xx} = 0{\cdot}009$ Å, $\sigma_{xxx} = 0{\cdot}7°$, x = C, O, N).

Structure of ribose

The ribose has a C(2')-*endo* conformation (Fig. 2) with C(1'), C(3'), C(4'), and O(1') in the plane (σ = 0·025 Å) while C(2') deviates by 0·54 Å.

The orientation at N(1)–C(1') is *anti* with a dihedral angle C(6)–N(1)–C(1')–O(1') of χ = 61·0°. This value supports the common relation between the puckering of the ribose and the orientation of the base (Sundaralingam, 1969; Egert, Lindner, Hillen & Gassen, 1977*b*). From the data listed in Table 4 (Rahman & Wilson, 1970; Hawkinson, 1977; Lin & Sundaralingam, 1971; Schwalbe & Saenger, 1973; Egert, Lindner, Hillen & Gassen, 1977*b*) it appears that this relation could be the result of the preferred formation of the intramolecular hydrogen bond C(6)–H···O(5') (Saenger, 1973), which requires a greater dihedral angle χ in C(2')-*endo* than in C(3')-*endo* puckering. Moreover, the C(5')–O(5') bond is fixed by this interaction *gauche* to C(3')–C(4') and to O(1')–C(4') with dihedral angles of 50·1 and −69·7° (Fig. 1).

In all published nucleoside structures, C(1')–O(1') is shorter than C(4')–O(1'), but the difference between these two bond lengths depends on the electronic properties of the 5-substituents (Table 3) and drops to only 0·028 Å in 5-aminouridine, compared with 0·041 Å in uridine and 0·071 Å in 5-nitrouridine. Also, the

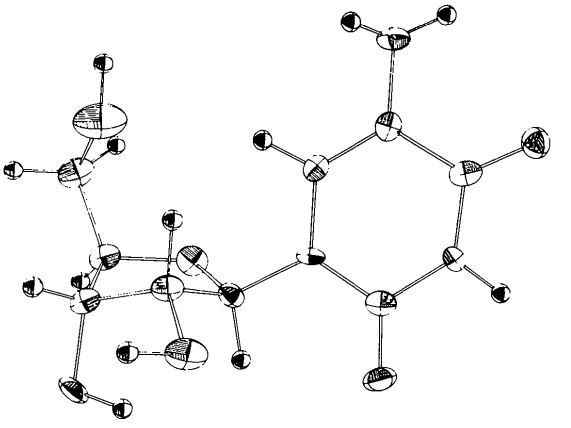


Fig. 2. The structural representation of 5-aminouridine with the thermal ellipsoids of the heavy atoms.

Table 3. *Influence of 5-substitution on the bond lengths* (Å) *of uridine*

Compound	N(1)–C(2)	C(2)–O(2)	N(1)–C(6)	C(5)–C(6)	N(1)–C(1')	C(1')–O(1')
5-Nitrouridine	1·414	1·193	1·288	1·388	1·504	1·390
5-Chlorouridine	1·375	1·215	1·373	1·335	1·474	1·410
Uridine*	1·371	1·222	1·369	1·334	1·490	1·413
5-Methyluridine	1·377	1·196	1·361	1·345	1·481	1·410
5-Methoxyuridine	1·367	1·225	1·379	1·355	1·490	1·395
5-Hydroxyuridine	1·358	1·230	1·389	1·337	1·467	1·416
5-Aminouridine	1·366	1·245	1·415	1·330	1·427	1·432

* Mean values of the two independent molecules found in the asymmetric unit.

Table 4. *Structural parameters of some uridine derivatives listed in the order of increasing dihedral angle* χ *(Egert, Lindner, Hillen & Gassen,* 1978*b; Morikawa, Torii, Iitaka & Tsuboi,* 1975)

Compound	Conformation of the ribose	χ (°)	C(6)–O(5′)(Å)	∠C(6)–H···O(5′) (°)
2-Thio-5-carboxymethyluridine	C(3′)-*endo*	3·3	C(4′)–C(5′) → *gauche–trans*	
5-Iodouridine I	C(3′)-*endo*	13·2	3·353	No data available
2-Thiouridine	C(3′)-*endo*	17·0	C(4′)–C(5′) → *gauche–trans*	
Uridine I	C(3′)-*endo*	18·3	3·112	147·8
2,4-Dithiouridine	C(3′)-*endo*	19·5	3·419	151·1
Uridine II	C(3′)-*endo*	24·3	3·548	169·3
5-Methoxyuridine	C(3′)-*endo*	25·4	3·242	158·0
5-Nitrouridine	C(3′)-*endo*	25·6	3·333	159·7
5-Methyluridine	C(3′)-*endo*	29·4	3·429	170·6
Uridine-5-oxyacetic acid methyl ester	C(3′)-*endo*	34·3	3·128	157·4
5-Hydroxyuridine	C(2′)-*endo*	42·1	Intermolecular C(6)–H···O(2) interaction	
5-Chlorouridine	C(2′)-*endo*	51·4	3·288	164·8
3-Deazauridine	C(2′)-*endo*	52·3	3·406	162·0
3-Deaza-4-deoxyuridine	C(2′)-*endo*	53·5	3·297	165·8
5-Iodouridine II	C(2′)-*endo*	58·7	C(4′)–C(5′) → *trans–gauche*	
5-Aminouridine	C(2′)-*endo*	61·0	3·539	158·0

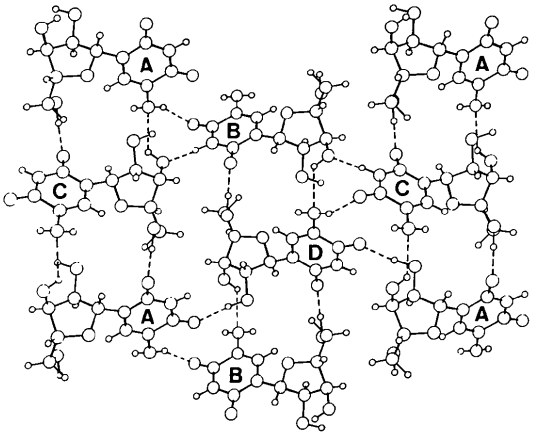


Fig. 3. A view down *c* with *a* horizontal and *b* vertical. The individual base stacks are designated with letters. They are interrelated through the following symmetry operations: $A \leftrightarrow B$: screw axis parallel to *c*; $A \leftrightarrow C$: screw axis parallel to *b*; $A \leftrightarrow D$: screw axis parallel to *a*. The hydrogen bonds are dashed.

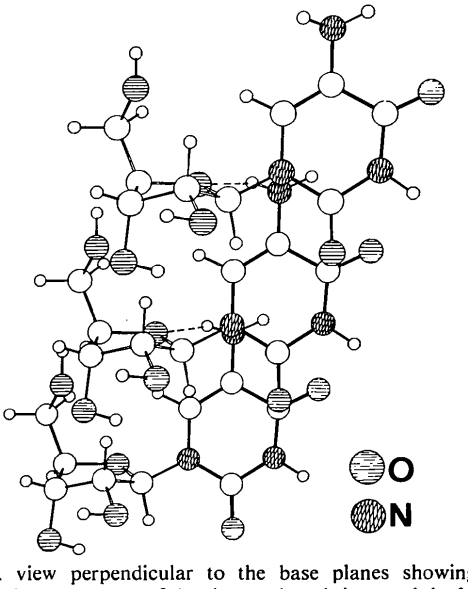


Fig. 4. A view perpendicular to the base planes showing the staggered arrangement of the six-membered rings and the N(5)–H′···O(1′) interaction (dashed).

N(1)–C(1′) distance varies with the 5-substitution, but shows the converse tendency. Thus electron-donating groups in the 5-position of uridine shorten N(1)–C(1′) and lengthen C(1′)–O(1′), contrary to electron-withdrawing substituents. MINDO/3 calculations of the structure and conformation of 5-substituted uridines confirm these facts (Egert, Lindner, Hillen & Gassen, 1978*a*).

Packing of the molecules

A view down *c* (Fig. 3) illustrates the crystal structure, which is determined by three screw axes parallel to the edges of the cell. The packing of the molecules differs from that of other nucleoside structures, because there is no definite separation of base and ribose regions. Intermolecular hydrogen-bonding between base and ribosyl moieties contributes to this arrangement whereas interactions between riboses do not occur. The bases are stacked along *c* at the short distance of 3·30 Å (Fig. 4). They are not located directly one upon another but are staggered with only two overlaps: N(1)···N(5) (3·47 Å) and O(2)···C(4) (3·29 Å).

Table 5. *List of intermolecular hydrogen bonds including the characteristic intramolecular* C(6)–H···O(5′) *interaction*

X–H···*Y*	*X*–H (Å)	H···*Y* (Å)	*X*···*Y* (Å)	∠*X*–H–*Y* (°)	Symmetry operation		
N(3)–H···O(3′)	0·98	1·81	2·783	171	0·5 + *x*,	0·5 − *y*,	1 − *z*
N(5)–H···O(4)	0·92	2·22	3·081	156	1·5 − *x*,	1 − *y*,	0·5 + *z*
N(5)–H′···O(1′)	0·80	2·37	3·050	143	*x*,	*y*,	1 + *z*
O(2′)–H···O(4)	1·09	1·89	2·941	161	−0·5 + *x*,	0·5 − *y*,	2 − *z*
O(3′)–H···N(5)	0·89	2·39	2·928	119	1 − *x*,	−0·5 + *y*,	1·5 − *z*
O(5′)–H···O(2)	1·04	1·71	2·744	172	1 − *x*,	0·5 + *y*,	1·5 − *z*
C(6)–H···O(5′)	0·96	2·63	3·539	158	*x*,	*y*,	*z*

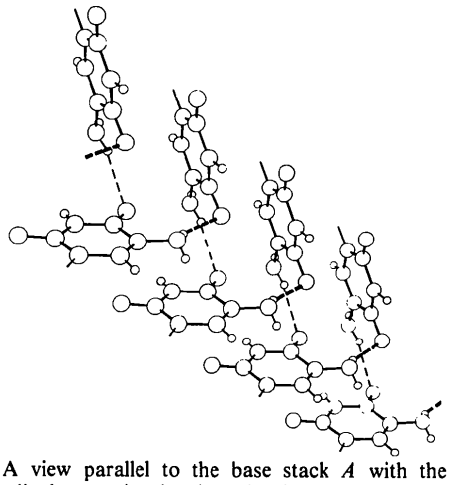


Fig. 5. A view parallel to the base stack *A* with the bases *B* perpendicular to it showing the interbase hydrogen bonds (dashed) which lead to a helical arrangement.

The stacked bases *A* are nearly perpendicular to the six-membered rings of the adjacent base stack *B*, and the same is true for the bases *C* and *D* (Fig. 3). *A* and *B* (also *C* and *D*) are connected by interbase hydrogen bonds between the 5-amino and the 4-keto groups (Table 5). This arrangement leads to a hydrogen-bonded double stack such that a helix is formed involving $O(4)_1\cdots NH(5)_2–C(5)_2–C(4)_2–O(4)_2\cdots NH(5)_3–C(5)_3\cdots$ (Fig. 5).

Conclusion

Our crystallographic data strongly support the conclusion that, as in other nucleosides, the *anti*-conformation of the base over the ribosyl moiety is stabilized by the hydrogen bond C(6)–H···O(5′) and that through this intramolecular interaction the dihedral angle C(6)–N(1)–C(1′)–O(1′) is related to the puckering of the ribose. The effect of the 5-amino group is not restricted to the base moiety but also influences the N(1)–C(1′) and the C(1′)–O(1′) lengths and the conformation of the ribose *via* the glycosidic bond.

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References

EGERT, E., LINDNER, H. J., HILLEN, W. & GASSEN, H. G. (1977*a*). *Nucleic Acids Res.* **4**, 929–937.

EGERT, E., LINDNER, H. J., HILLEN, W. & GASSEN, H. G. (1977*b*). *Acta Cryst.* B**33**, 3704–3707.

EGERT, E., LINDNER, H. J., HILLEN, W. & GASSEN, H. G. (1978*a*). In preparation.

EGERT, E., LINDNER, H. J., HILLEN, W. & GASSEN, H. G. (1978*b*). In preparation.

GREEN, E. A., ROSENSTEIN, R. D., SHIONO, R., ABRAHAM, D. J., TRUS, B. L. & MARSH, R. E. (1975). *Acta Cryst.* B**31**, 102–107.

HAWKINSON, S. W. (1977). *Acta Cryst.* B**33**, 80–85.

HAWKINSON, S. W. & COULTER, C. L. (1971). *Acta Cryst.* B**27**, 34–42.

HILLEN, W., EGERT, E., LINDNER, H. J. & GASSEN, H. G. (1978). Submitted for publication.

HILLEN, W. & GASSEN, H. G. (1975). *Biochim. Biophys. Acta*, **407**, 347–356.

HUNT, D. J. & SUBRAMANIAN, E. (1969). *Acta Cryst.* B**25**, 2144–2152.

International Tables for X-ray Crystallography (1974). Vol. IV, pp. 99–102. Birmingham: Kynoch Press.

LIN, G. H.-Y. & SUNDARALINGAM, M. (1971). *Acta Cryst.* B**27**, 961–969.

LUEHRMANN, R. & GASSEN, H. G. (1976). *Eur. J. Biochem.* **66**, 1–9.

MORIKAWA, K., TORII, K., IITAKA, Y. & TSUBOI, M. (1975). *Acta Cryst.* B**31**, 1004–1007.

RAHMAN, A. & WILSON, H. R. (1970). *Acta Cryst.* B**26**, 1765–1775.

SAENGER, W. (1973). *Angew. Chem.* **85**, 680–690.

SCHWALBE, C. H. & SAENGER, W. (1973). *Acta Cryst.* B**29**, 61–69.

SHELDRICK, G. M. (1976). *SHELX* 76. Program for crystal structure determination. Univ. of Cambridge, England.

SUNDARALINGAM, M. (1969). *Biopolymers*, **7**, 821–860.

THEWALT, U. & BUGG, C. E. (1973). *Acta Cryst.* B**29**, 1393–1398.

VISSER, D. W. (1968). *Synthetic Procedures in Nucleic Acid Chemistry*, edited by W. W. ZORBACH & R. S. TIPSON, Vol. 1, pp. 407–408. New York: Interscience.

WERKHEISER, W. C., WINZLER, R. J. & VISSER, D. W. (1955). *Cancer Res.* **15**, 641–648.