

of Health. We thank Professor R. Shiono for the use of computer programs, and the Gulf Research and Development Company, Harmorville, Pa. for the use of computing facilities (IBM 360/85) for the full-matrix least-squares calculation.

References

- BERKING, B. (1972). *Acta Cryst.* B28, 98–113.
 BERKING, B. & CRAVEN, B. M. (1971). *Acta Cryst.* B27, 1107–1115.
 BERTHOU, J., RÉRAT, B. & RÉRAT, C. (1965). *Acta Cryst.* 18, 768–777.
 BOLTON, W. (1963). *Acta Cryst.* 16, 950–956.
 BOLTON, W. (1965). *Acta Cryst.* 19, 1051–1055.
 CRAVEN, B. M. (1964). *Acta Cryst.* 17, 282–289.
 CRAVEN, B. M., CUSATIS, C., GARTLAND, G. L. & VIZZINI, E. A. (1973). *J. Mol. Struct.* 16, 331–342.
 CRAVEN, B. M. & SABINE, T. M. (1969). *Acta Cryst.* B25, 1970–1978.
 CRUICKSHANK, D. W. J. & ROBERTSON, A. P. (1953). *Acta Cryst.* 6, 698–705.
 GARTLAND, G. L. (1971). Ph. D. Dissertation, Univ. of Pittsburgh.
 GARTLAND, G. L. & CRAVEN, B. M. (1974). *Acta Cryst.* B30, 980–987.
 GATEHOUSE, B. M. & CRAVEN, B. M. (1971). *Acta Cryst.* B27, 1337–1344.
 GILLIER, H. (1965). *Bull. Soc. Chim. Fr.* pp. 2373–2393.
 GUTIERREZ, P. & BENSON, B. (1974). *J. Chem. Phys.* 60, 640–644.
International Tables for X-ray Crystallography (1962). Vol. III. Birmingham: Kynoch Press.
 KARLE, J. & KARLE, I. L. (1966). *Acta Cryst.* 21, 849–859.
 McCLURE, R. J. & CRAVEN, B. M. (1973). *Acta Cryst.* B29, 1860–1864.
 MATTHEWS, B. W. (1965). *Acta Cryst.* 18, 151–157.
 STEWART, R. F., DAVIDSON, E. R. & SIMPSON, W. T. (1965). *J. Chem. Phys.* 42, 3175–3187.

Acta Cryst. (1975). B31, 211

Structural Chemistry of N(7)-Substituted Purines: 7-Methyladenine Dihydrochloride*

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7-Methyladenine dihydrochloride, $C_6N_5H_7 \cdot 2HCl$, crystallizes in the orthorhombic system, space group *Pbma* or *Pb2₁a*, with crystal data $a = 15.534$ (8), $b = 6.589$ (2), $c = 9.403$ (4) Å, $Z = 4$, $D_m = 1.48$ (2), $D_c = 1.53$ g cm⁻³. Intensities for 1206 independent reflections were collected by counter methods. A structural solution in space group *Pbma* has been refined by full-matrix least-squares calculations, based on F , to an R value of 0.069. The final weighted R value and goodness-of-fit are 0.037 and 1.5, respectively. The analysis in *Pbma* requires: (1) exact planarity of the 7-methyladenine dication, (2) staggering of the methyl hydrogen atoms about the molecular plane, and (3) coplanarity of the dication and the chloride anions. The observed sites of protonation are N(3) and N(9). This study provides the first direct evidence of protonation at N(3) for an adenine derivative. The effect of the protonation at N(3) on the structural parameters is discussed. The crystal structure is dominated by a series of N–H···Cl⁻ hydrogen bonds and C–H···Cl⁻ interactions. The dications in adjacent layers ($y = \pm \frac{1}{2}$) show a slight overlap of exocyclic amine groups.

Introduction

We have recently studied the crystal and molecular structures of a series of transition-metal complexes containing purines as coordinated ligands (Kistenmacher, Marzilli & Chang, 1973; Marzilli, Kistenmacher & Chang, 1973; Marzilli, Kistenmacher, Darcy, Szalda & Beer, 1974; Kistenmacher & Szalda, 1974; Kistenmacher & Sorrell, 1974). One aspect of this work has centered on the effect on the structural parameters in the purine ligand due to the formation of the coordination bond. In pursuit of such information, we have undertaken a refinement of the structure of adenine hydrochloride hemihydrate (Kistenmacher

& Shigematsu, 1974a) and determined the structure of adenine dihydrochloride (Kistenmacher & Shigematsu, 1974b). Comparison of these results with the structural parameters for protonated adenine coordinated to a transition metal through N(7) (Taylor, 1973) gave an indication of the magnitude of the effects due to coordination (Kistenmacher & Shigematsu, 1974b).

In an attempt to take the analysis one step further, we decided to determine the structure of 7-methyladenine dihydrochloride in order to compare the effects of protonation, coordination and alkylation at N(7) on the protonated adenine nucleus. We fully anticipated at the onset of the project that 7-methyladenine would be protonated at N(1) and N(9), as was indicated to be the trend in adenine and N(9)-substituted adenine derivatives. However, our analysis shows (see below) that 7-methyladenine is protonated at N(3) and not

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N(1); this is in direct contrast with the experimental and theoretical (Bonaccorsi, Pullman, Scrocco & Tomasi, 1972) data and predictions for adenine and N(9)-substituted adenine derivatives. We have considered this alteration in the protonation pattern for N(7)-substituted adenine derivatives elsewhere (Kistenmacher, Shigematsu & Weinstein, 1974), and we attribute the preference for protonation at N(3) over N(1) to an enhancement of the electrostatic potential minima at N(3) due to the adjacent localized electronic charge distribution at N(9).

The protonation at N(3) and its effect on the structural parameters of the substituted adenine moiety are of particular interest. In this report we give a complete description of the crystal and molecular parameters in 7-methyladenine dihydrochloride, and compare the effects of protonation at N(3) with the well known geometric adjustments in N(1) protonated species.

Experimental

Crystalline 7-methyladenine dihydrochloride was obtained from a saturated solution of 7-methyladenine (kindly supplied by Professor S. I. Chan of the California Institute of Technology) in 6*N* hydrochloric acid. The crystals were orthorhombic prisms with [010] as the prism axis. The crystals are air-sensitive, and we found it necessary to coat them with a thin film of grease and mount them in thin-walled capillaries. The crystal system is orthorhombic with systematic absences $hk0$, $h=2n+1$; $0kl$, $k=2n+1$, which are consistent with the centrosymmetric space group *Pbma* or the non-centrosymmetric space group *Pb2₁a*. Unit-cell dimensions and standard deviations were derived from a least-squares fit to the 2θ , ω and χ settings for 15 reflections measured on a diffractometer; the density was measured by flotation. Complete crystal data are given in Table 1.

Table 1. *A comparison of crystal data for adenine dihydrochloride and 7-methyladenine dihydrochloride*

Adenine dihydrochloride C ₅ N ₅ H ₇ Cl ₂	7-Methyladenine dihydrochloride C ₆ N ₅ H ₉ Cl ₂
$a = 13.480$ (6) Å	$a = 15.534$ (8) Å
$b = 6.553$ (2)	$b = 6.589$ (2)
$c = 9.390$ (3)	$c = 9.403$ (4)
<i>Pnma</i>	<i>Pbma</i>
$Z = 4$	$Z = 4$
$D_m = 1.68$ (1) g cm ⁻³	$D_m = 1.48$ (2) g cm ⁻³
$D_c = 1.67$	$D_c = 1.53$

5474 reflections (the $+h$ -hemisphere to $2\theta \leq 55^\circ$) were measured on a Syntex *P*T computer-controlled diffractometer; molybdenum graphite-monochromatized radiation was employed. The crystal used in data collection was $0.15 \times 0.20 \times 0.35$ mm; the long axis of the crystal, b , was tilted relative to the ϕ axis of the diffractometer. Intensity data were collected by the θ - 2θ scan technique; individual scan speeds were deter-

mined by a rapid scan at the calculated Bragg peak, and the rate of scanning ranged from 2 to $24^\circ \text{ min}^{-1}$. Three standard reflections were monitored after every 100 data points, and their intensities showed no systematic variations during the course of the experiment. The 5474 measured intensities, which included standards and systematic absences, were then averaged to yield a set of 1206 symmetry independent values. All reflections were assigned observational variances based on the equation: $\sigma^2(I) = S + (B_1 + B_2) (T_S/2T_B)^2 + (pI)^2$, where S , B_1 and B_2 are the scan and background counts, T_S and T_B are the scan and individual background counting times ($T_B = \frac{1}{4}T_S$ for all reflections), and p was taken to be 0.03 and represents the error proportional to the diffracted intensity (Busing & Levy, 1957). Intensities and their standard deviations were corrected for Lorentz and polarization effects; the amplitudes of reflections with negative intensities were set equal to zero. Owing to the small size of the crystal and the low value for μ (6.4 cm^{-1}), no correction for absorption effects was deemed necessary (maximum error estimated to be about 4% in I). The squared structure factors were placed on an approximate absolute scale by the method of Wilson (1942).

Solution and refinement of the structure

7-Methyladenine dihydrochloride crystallizes with a unit cell which is very similar in size and symmetry to that found in adenine dihydrochloride (Kistenmacher & Shigematsu, 1974*b*) (Table 1). The cell lengths differ significantly only in the value for a ; this difference of about 2.05 Å is very similar to the magnitude of the van der Waals radius of 2.0 Å for a methyl group (Pauling, 1960). The presence of a very strong 020 reflection ($|F_o|$ about 311e) and a b -axis length of 6.589 (2) Å (leading to an interplanar separation of about 3.29 Å for the dications) gave further evidence that the two structures must be fairly similar (Table 1). We found in adenine dihydrochloride that a structural solution in space group *Pnma* (as opposed to *Pn2₁a*) provided a very satisfactory fit to the experimental data. The above considerations, plus the excellent agreement between reflections not required to be equivalent in the lower symmetry space group *Pb2₁a* [for example, the hkl and $h\bar{k}l$ octants are not required to be symmetry mates in the presence of significant anomalous dispersion in point group $m2m$, Ibers (1967)], prompted us to pursue a structural solution in the centrosymmetric space group *Pbma*. The analysis in *Pbma* places the following restrictions on molecular symmetry: (1) the 7-methyladenine dication must be perfectly planar; (2) the exocyclic methyl group is staggered about the adenine molecular plane with one of its hydrogen atoms lying in the plane and the two other methyl hydrogens symmetry related about the crystallographic (molecular) mirror plane.

A detailed examination of a three-dimensional Patterson map indicated that the chloride ions as well as

the 7-methyladenine dication lie in the mirror plane at $y = \frac{1}{4}$. The x and z coordinates of the two independent chloride ions were determined from the map, and a subsequent Fourier synthesis ($R = \sum ||F_o| - |F_c|| / \sum |F_o| = 0.49$) allowed the positioning of the remaining 11 heavy atoms. Five cycles of isotropic least-squares refinement, minimizing $\sum w(|F_o| - |F_c|)^2$ where $w = 1/\sigma^2(F_o)$, reduced R to 0.17. A difference Fourier was computed, and the 8 unique hydrogen atoms located.

Five further cycles of refinement were computed in which the following parameters were successively added: (1) an overall scale factor, (2) anisotropic temperature parameters for the 13 heavy atoms, (3) positional and isotropic temperature parameters for the 8 hydrogen atoms. The final unweighted and weighted residuals are: $R = 0.069$, and $R_w = [\sum w(F_o - F_c)^2 / \sum wF]^2 = 0.037$. The final value of the goodness-of-fit, $\{[\sum w(F_o - F_c)^2 / (n - p)]^{1/2}\}$, where $n = 1206$ and $p = 104$, was 1.5. In the final cycle of refinement no shift/error exceeded 0.7. Neutral scattering-factor curves for Cl, C and N were taken from the compilation of Hanson, Herman, Lea & Skillman (1964); the scattering curve for H was that of Stewart, Davidson & Simpson (1965). The real part of the scattering curve for Cl was corrected for anomalous dispersion effects (Cromer, 1965). Final atomic parameters for all atoms are given in Table 2.*

The structure factor and Fourier calculations were done with the X-RAY 67 series of programs (Stewart, 1967); the least-squares refinements were performed with an extensively modified version of ORFLS (Busing, Martin & Levy, 1962); the illustrations were pre-

Table 2. Final atomic parameters and their estimated standard deviations ($\times 10^4$ for the heavy atoms and $\times 10^3$ for the hydrogen atoms)

The form of the anisotropic ellipsoid is: $\exp[-(B_{11}h^2 + B_{22}k^2 + B_{33}l^2 + 2B_{12}hk + 2B_{23}kl)]$. For all the heavy atoms, the usual terms in the above expression $2B_{12}hk$ and $2B_{23}kl$ are equal to zero by symmetry. All the atoms have $y = \frac{1}{4}$, except H(6) where $y = 0.133$ (5). The isotropic thermal parameters for the hydrogen atoms have the form: $\exp[-B(\sin \theta/\lambda)^2]$.

	x	z	B_{11}	B_{22}	B_{33}	B_{13}
Cl(1)	8504 (1)	1683 (1)	36 (1)	306 (3)	57 (1)	2 (1)
Cl(2)	1202 (1)	2123 (1)	37 (1)	275 (3)	55 (1)	-5 (1)
N(1)	3702 (2)	3847 (3)	32 (1)	265 (10)	61 (3)	-2 (2)
N(3)	4040 (2)	1407 (3)	39 (2)	213 (10)	63 (3)	-4 (2)
N(7)	6059 (2)	3044 (3)	33 (1)	193 (8)	56 (3)	2 (2)
N(9)	5575 (2)	863 (3)	41 (2)	230 (10)	47 (3)	9 (2)
N(10)	4725 (3)	5589 (3)	35 (2)	289 (11)	55 (4)	4 (2)
C(2)	3486 (3)	2504 (4)	28 (2)	257 (13)	104 (5)	3 (2)
C(4)	4877 (2)	1725 (3)	37 (2)	138 (8)	57 (4)	4 (2)
C(5)	5162 (2)	3116 (3)	30 (1)	146 (8)	53 (4)	-2 (2)
C(6)	4558 (2)	4222 (3)	36 (2)	154 (10)	54 (4)	-1 (2)
C(7)	6679 (3)	4213 (4)	32 (2)	350 (17)	78 (5)	2 (3)
C(8)	6273 (3)	1675 (3)	38 (2)	219 (12)	61 (4)	9 (3)
H(2)	289 (2)	234 (4)	3.6 (8)			
H(3)	382 (2)	52 (4)	4.3 (9)			
H(4)	432 (2)	612 (4)	3.5 (10)			
H(5)	524 (3)	591 (4)	3.9 (10)			
H(6)	658 (2)	478 (3)	6.5 (8)			
H(7)	720 (2)	387 (4)	4.5 (11)			
H(8)	691 (2)	142 (3)	3.6 (9)			
H(9)	563 (2)	-4 (4)	4.0 (9)			

pared with the aid of the computer program ORTEP (Johnson, 1965).

Discussion

As noted above, we have solved and refined the structure of 7-methyladenine dihydrochloride in the centrosymmetric space group $Pbma$. The requirements on the crystal and molecular symmetry associated with placing the dication and the chloride ions in special

* A list of structure factors has been deposited with the British Library Lending Division as Supplementary Publication No. SUP 30645 (10 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 13 White Friars, Chester CH1 1NZ, England.

Table 3. A comparison of heavy-atom bond angles ($^\circ$) in several neutral and protonated purines

	T ^a	C ^b	Ado ^c	AdoHCl ^d	7MA2HCl ^e
C(2)—N(1)—C(6)	129.7 (4)	127.8 (3)	119.3 (2)	124.2 (3)	119.7 (4)
C(2)—N(3)—C(4)	118.6	119.9	110.4	111.6	116.9
C(5)—N(7)—C(7)	127.1	127.8	—	—	128.4
C(5)—N(7)—C(8)	105.0	105.4	103.5	103.8	107.3
C(7)—N(7)—C(8)	127.9	126.7	—	—	124.3
C(4)—N(9)—C(8)	103.2	103.4	105.7	105.8	108.1
N(1)—C(2)—N(3)	115.7	116.1	128.9	125.0	125.3
N(3)—C(4)—N(9)	126.4	126.5	126.7	127.0	130.3
N(3)—C(4)—C(5)	122.1	121.9	127.6	127.4	121.6
N(9)—C(4)—C(5)	115.5	111.6	105.7	105.6	108.1
N(7)—C(5)—C(4)	106.1	106.0	110.8	111.3	105.9
N(7)—C(5)—C(6)	130.7	130.5	132.8	130.5	134.8
C(4)—C(5)—C(6)	123.1	123.4	116.4	118.2	119.2
N(1)—C(6)—N(10)*	123.2	122.0	118.1	120.4	116.3
N(1)—C(6)—C(5)	110.6	110.6	117.4	113.5	117.2
N(10)*—C(6)—C(5)	126.2	127.4	124.5	126.1	126.5
N(7)—C(8)—N(9)	114.2	113.6	114.3	113.6	110.7

(a) Theobromine (Shefter, Brennan & Sackman, 1971). (b) Caffeine (Shefter, 1968). (c) Adenosine (Lai & Marsh, 1972). (d) Adenosine hydrochloride (Shikata, Ueki & Mitsui, 1973). (e) 7-Methyladenine dihydrochloride (this study).

* An exocyclic carbonyl oxygen in T^a and C^b. The numbering scheme is that used in Fig. 1.

positions in *Pbma* have also been discussed. There are several indications that the analysis in the centrosymmetric space group is a quite good approximation to the true structure, even if the space group is actually *Pb2₁a*: (1) the final weighted residual of 0.037 and the goodness-of-fit of 1.5 demonstrate that the final model and weights are in good agreement with the experimental data; (2) there are no unusually large root-mean-square displacements normal to the crystallographic mirror plane – the atom with the largest difference between in-plane and out-of-plane r.m.s.d.'s is Cl(1) with values: r.m.s.d._{min} = 0.159, r.m.s.d._{r0101} = 0.260 Å; (3) the bond lengths and angles derived from the final cycle of least-squares are in accord with prediction (see below).

We have not attempted any refinement in the lower symmetry space group *Pb2₁a* owing to the very probable ill-conditioned nature of the normal equations (Ermer & Dunitz, 1970; Donohue, 1971). The requirement that the purine dication be perfectly planar is possibly incorrect [see Voet & Rich (1970) for a summary of the non-planarity of purine bases in many crystal structures], but we anticipate (Kistenmacher & Shigematsu, 1974*b*) for the same reasons as discussed in the structure of adenine dihydrochloride that the deviations from planarity are small (≤ 0.01 Å), if they exist at all. These minor deviations from planarity, which are not accounted for in our model, should have a minimal effect on derived bond lengths and angles.

Bond distances and angles

An ORTEP (Johnson, 1965) drawing of the diprotonated 7-methyladenine cation is shown in Fig. 1; interatomic distances are also given in this figure. Bond angles involving the heavy atoms are presented in Table 3.

Estimated standard deviations are about 0.003–0.004 Å for the heavy-atom bond lengths and about 0.3–0.4° for heavy-atom bond angles. Analogous values for the bond lengths and angles involving hydrogen atoms are 0.04 Å and 4°.

Several crystallographic studies on N(1) protonated adenine derivatives have been published: adenine hydrochloride hemihydrate (Cochran, 1951; Kistenmacher & Shigematsu, 1974*a*), adenine dihydrochloride (Kistenmacher & Shigematsu, 1974*b*), 3'-AMP (Sundaralingam, 1966), 5'-AMP (Kraut & Jensen, 1963), adenosine hydrochloride (Shikata, Ueki & Mitsui, 1973), 9-methyladenine dihydrobromide (Bryan & Tomita, 1962), the dinucleoside phosphate adenosyl-2',5'-uridine (Shefter, Barlow, Sparks & Trueblood, 1964), polyriboadenylic acid (Rich, Davies, Crick & Watson, 1961) and bis(adeninium)[*trans*-bis(adenine) tetra-aquacobalt(II)]bis(sulfate) hexahydrate (deMeester & Skapski, 1973). Also, several transition metal complexes have been reported where N(1) protonated adenine is observed as a unidentate or bidentate ligand: deMeester, Goodgame, Price & Skapski (1970), Taylor (1973), deMeester, Goodgame, Price & Skapski (1971).

This first report of N(3) protonated adenine in 7-methyladenine dihydrochloride provides then an opportunity to test the various proposals developed on the basis of the N(1) protonated species.

(a) *Heavy-atom bond lengths.* Comparison, for example, of the pyrimidine ring bond lengths in adenosine (Lai & Marsh, 1972) and adenosine hydrochloride (Shikata, Ueki & Mitsui, 1973) reveals the following significant changes which can be attributed to the effect of the protonation at N(1): (1) the N(1)–C(2) bond length increases by 0.021 Å [1.340 (2) Å in adenosine, 1.361 (5) Å in the protonated cation]; (2) a decrease of 0.022 Å in the N(3)–C(2) bond length [1.330 (2) versus 1.308 (5) Å]; (3) the C(5)–C(6) bond length decreases by 0.018 Å [1.415 (2) versus 1.397 (5) Å]. Protonation at N(3) in 7-methyladenine has caused changes

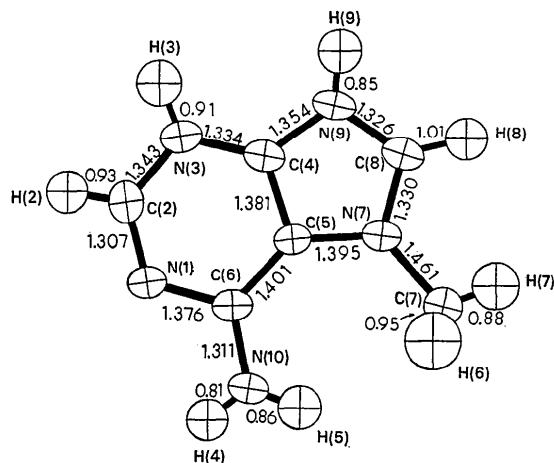


Fig. 1. Interatomic distances in the diprotonated 7-methyladenine dication. The estimated standard deviations in the bond lengths are 0.003–0.004 Å for those involving the heavy atoms, and 0.04 Å for those involving the hydrogen atoms.

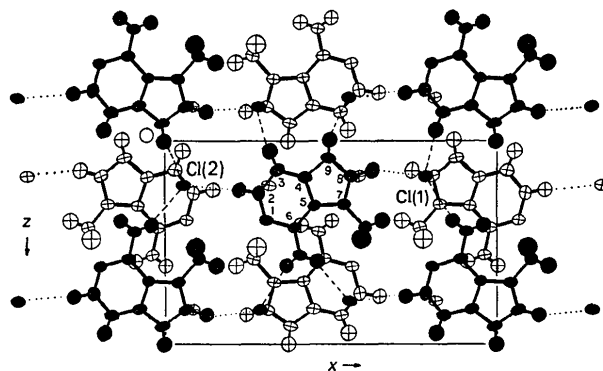


Fig. 2. A view normal to the *ac* plane of the crystal packing in 7-methyladenine dihydrochloride. The shaded atoms have their centroids at $y = \frac{1}{4}$; the unshaded atoms have their centroids at $y = -\frac{1}{4}$. Hydrogen bonds are indicated by dashed lines; close contacts are indicated by dotted lines.

of a similar magnitude, but as expected not always in the same bonds. In particular, the following changes are noted: (a) the N(1)–C(2) bond length *decreases* on N(3) protonation from 1.340 (2) Å in adenosine to 1.307 (3) Å in the diprotonated 7-methyladenine dication; (b) the N(1)–C(6) bond length increases by 0.025 Å [1.351 (2) to 1.376 (3) Å]; (c) the C(6)–N(10) bond is shortened by 0.021 Å [1.332 (2) *versus* 1.311 (3) Å].

The trends observed above are distinctly different for the N(1) and the N(3) protonated cases. Somewhat surprisingly, the protonation at N(3) has caused no major changes (greater than about 0.01 Å) in the two bonds directly involved with the heteroatom which is protonated, while the protonation at N(1) strongly affects at least one of the bonds to N(1), N(1)–C(2). Since the same general trend holds whether one compares adenosine to monoprotated adenine or diprotonated adenine (Kistenmacher & Shigematsu, 1974*a, b*), the charge on the purine base does not seem to be the discriminating factor. Part of the lack of change in the bonds adjacent to N(3) may lie in the fact that one of the bonds, N(3)–C(4), involves a carbon atom at the fused-ring junction between the pyrimidine and imidazole portions of the molecule.

The bond lengths in the imidazole ring are also given in Fig. 1. To our knowledge, two other N(7)-methylated purines have been studied: caffeine (Sutor, 1958; Shefter, 1968) and theobromine (Shefter, Brennan & Sackman, 1971). Neither caffeine nor theobromine contains a substituent at N(9). The addition of the proton to N(9) in 7-methyladenine dihydrochloride, however, has caused no major changes in the bond lengths in the imidazole ring: a result which is not unexpected considering our studies on the effect of the addition of a proton at N(7) in N(9)-substituted adenine derivatives (Kistenmacher & Shigematsu, 1974*b*).

The N(7)–C(7) bond length, 1.461 (5) Å, is in reasonable agreement with that found in caffeine [1.47 (2) Å, Sutor (1958); 1.470 (7) Å in a 1:1 complex between 5-chlorosalicylic acid and caffeine, Shefter (1968)] and slightly longer, 0.02 Å, than observed in theobromine [1.438 (7) Å in a 2:1 complex of 5-chlorosalicylic acid and theobromine, Shefter, Brennan & Sackman (1971)]. The effect of protonation on this bond is less than clear. For example, there is no discernible trend in the N(9)–C(1') bond length in N(9)-substituted adenosine derivatives: *neutral*-adenosine, 1.466 (3) Å (Lai & Marsh, 1972), 3'-acetyladenosine, 1.454 (5) Å (Rao & Sundaralingam, 1970); *protonated*-adenosine hydrochloride, 1.460 (4) Å (Shikata, Ueki & Mitsui, 1973); 3'-AMP, 1.477 (6) Å (Sundaralingam, 1966), 5'-AMP, 1.492 (12) Å (Kraut & Jensen, 1963).

(b) *Heavy-atom bond angles.* A point of considerable interest is what effect the protonation at N(3) has on the bond angles in the pyrimidine ring. Sundaralingam & Jensen (1965) and Singh (1965) have noted that addition of a substituent to a nitrogen atom in a hetero-

cyclic six-membered ring causes an increase of about 6–10° in the internal angle at the heteroatom in question. Voet & Rich (1970) have further noted that the bulk of this adjustment is accommodated at the interior angles adjacent to the site of addition. Using the pyrimidine ring parameters for adenosine (Lai & Marsh, 1972) as a basis, we can follow the changes in the interior bond angles in 7-methyladenine dihydrochloride (Table 3). The addition of the proton at N(3) has caused an increase of 6.5° in the bond angle C(2)–N(3)–C(4) in 7-methyladenine, 116.9 (3)°, *versus* adenosine, 110.4 (2)°. We should expect then, if Voet & Rich's (1970) observation holds for this case, about a 3° decrease in the bond angles at C(2) and C(4) and only small changes in the interior bond angles at N(1), C(6) and C(5). In general, these predictions hold: the N(3)–C(2)–N(1) angle is 3.6° smaller than observed in adenosine [125.3 (3)° *versus* 128.9 (2)°], and the angles C(2)–N(1)–C(6) and N(1)–C(6)–C(5) are essentially unchanged [119.7 (3)° and 117.2 (3)° in 7-methyladenine. 2HCl and 119.3 (2)° and 117.4 (2)° in adenosine].

There are, however, two anomalies: (1) the angle N(3)–C(4)–C(5), which we anticipated should decrease about 3°, has in fact decreased 6.0° [from 127.6 (2)° in adenosine to 121.6 (3)° in 7-methyladenine dihydrochloride], and (2) the C(6)–C(5)–C(4) angle, where little change was anticipated, has actually increased 2.8° [116.4 (2)° to 119.2 (3)°]. On the basis of these changes, we concluded that some of the 6.0° decrease in the bond angle N(3)–C(4)–C(5) is in response to the protonation at N(3) – about half, as anticipated from the conclusions of Voet & Rich (1970) – and that the remainder is probably a result of the same perturbation which causes the 2.8° increase in the C(6)–C(5)–C(4) bond angle. This second perturbation is probably involved with the steric crowding between the exocyclic amine and the methyl group at C(6) and N(7), respectively. That there is steric crowding between these groups seems evident from the exocyclic bond angles at N(7) and C(6), which are considerably more dissymmetric [particularly the angles off N(7)] than is found in theobromine and caffeine (Table 3). This steric crowding is not unexpected, since the carbonyl group at C(6) in caffeine and theobromine probably exhibits a more restricted van der Waals volume (Bondi, 1964) than the amine group in 7-methyladenine dihydrochloride. It should also be noted that the exocyclic bond angle at C(5), N(7)–C(5)–C(6), is about 4° larger in the 7-methyladenine dication than is observed in caffeine or theobromine (Table 3). The bond angle increase surely is a result of the steric interaction noted above.

The interior bond angles in the imidazole portion of the molecule are quite different from those found in caffeine or theobromine. We have noted elsewhere (Kistenmacher & Shigematsu, 1974*b*, and above) that, while the bond lengths in the imidazole ring of protonated adenine are not substantially altered owing to protonation of the ring, the bond angles are signifi-

cantly changed. In particular, the interior angles at C(5), C(8), N(7) and N(9) in theobromine and caffeine (Shefter, Brennan & Sackman, 1971; Shefter, 1968) are nearly identical, while the analogous angles in 7-methyladenine dihydrochloride show the following deviations from the averaged values for caffeine and theobromine: C(5), -0.2° ; C(8), -3.2° ; N(7), 2.1° ; N(9), 4.8° . The bond angle at C(4), N(9)-C(4)-C(5), shows a considerable variation over the three structures: $115.5(4)^\circ$ in theobromine, $111.6(3)^\circ$ in caffeine, and $108.1(4)^\circ$ in this study.

(c) *Bond lengths and angles involving the hydrogen atoms.* The hydrogen atoms have refined to positions which give reasonable bond distances and angles. While there seem to be discernible adjustments of the hydrogen atoms owing to molecular steric requirements and the formation of the hydrogen bonds, the estimated standard deviations preclude any detailed discussion.

Crystal packing and hydrogen bonding

A view of the crystal packing in 7-methyladenine dihydrochloride is illustrated in Fig. 2. The 7-methyladenine dication has four acidic hydrogens, those off the heterocyclic ring atoms N(3) and N(9) and those on the exocyclic amine nitrogen N(10). All four of these hydrogen atoms participate in hydrogen bonds, Fig. 2 and Table 4. Furthermore, the hydrogen atoms off C(2) and C(8), which have been shown in many instances to be involved in significant interactions with electronegative acceptor atoms [see for example, Kistenmacher & Shigematsu (1974*a, b*) and Rao & Sundaralingam (1970)], form close contacts with the chloride ions, Fig. 2 and Table 4.

Table 4. Distances and angles in the interatomic contacts of the type $D-H\cdots A$

D	H	A	$D\cdots A$	$H\cdots A$	$\angle D-H\cdots A$
Hydrogen bonds					
N(3)	H(3)	Cl(1) ^a	3.022 Å	2.12 Å	171°
N(10)	H(4)	Cl(1) ^b	3.190	2.42	160
N(10)	H(5)	Cl(2) ^c	3.145	2.38	149
N(9)	H(9)	Cl(2) ^d	2.972	2.15	162
C-H \cdots Cl ⁻ short contacts					
C(2)	H(2)	Cl(2) ^e	3.566	2.64	175
C(8)	H(8)	Cl(1) ^e	3.466	2.50	161
(a)	$-\frac{1}{2}+x, \frac{1}{2}-y, -z$		(d)	$\frac{1}{2}+x, \frac{1}{2}-y, -z$	
(b)	$-\frac{1}{2}+x, \frac{1}{2}-y, 1-z$		(e)	x, y, z	
(c)	$\frac{1}{2}+x, \frac{1}{2}-y, 1-z$				

Each of the lattice chloride ions has very similar in-plane and out-of-plane interactions. Cl(1) accepts two hydrogen bonds, from H(3)-N(3) and H(4)-N(10), and has a close contact with H(8)-C(8). Its closest out-of-plane contact is with C(8) of a 7-methyladenine dication. The second chloride ion, Cl(2), also accepts two

strong hydrogen bonds, from H(5)-N(10) and H(9)-N(9), and is involved in a C-H \cdots Cl⁻ interaction with H(2)-C(2). Cl(2) sits nearly over, and above, the C(2)-N(3) bond of a purine dication (Fig. 2 and Table 4).

There is only a minimal overlapping of the 7-methyladenine dications as observed in 9-methyladenine dihydrochloride (Bryan & Tomita, 1962) and adenine dihydrochloride (Kistenmacher & Shigematsu, 1974*b*). However, there may be some crystal stabilization due to the slight dipolar overlap of exocyclic amine groups [Fig. 2, see Bugg, Thomas, Sundaralingam & Rao (1971) for an extensive discussion of the overlap of purine bases in various crystal structures].

References

- BONACCORSI, R., PULLMAN, A., SCROCCO, E. & TOMASI, J. (1972). *Theoret. Chim. Acta*, **24**, 51-60.
 BONDI, A. (1964). *J. Phys. Chem.* **68**, 441-451.
 BRYAN, R. F. & TOMITA, K. (1962). *Acta Cryst.* **15**, 1179-1182.
 BUGG, C. E., THOMAS, J. M., SUNDARALINGAM, M. & RAO, S. T. (1971). *Biopolymers*, **10**, 175-219.
 BUSING, W. R. & LEVY, H. A. (1957). *J. Chem. Phys.* **26**, 563-568.
 BUSING, W. R., MARTIN, K. O. & LEVY, H. A. (1962). *ORFLS*. Oak Ridge National Laboratory Report ORNL-TM-305.
 COCHRAN, W. (1951). *Acta Cryst.* **4**, 81-92.
 CROMER, D. T. (1965). *Acta Cryst.* **18**, 17-23.
 DONOHUE, J. (1971). *Acta Cryst.* **B27**, 1071.
 ERMER, O. & DUNITZ, J. D. (1970). *Acta Cryst.* **A26**, 163.
 HANSON, H. P., HERMAN, F., LEA, J. D. & SKILLMAN, S. (1964). *Acta Cryst.* **17**, 1040-1044.
 IBERS, J. A. (1967). *Acta Cryst.* **22**, 604-605.
 JOHNSON, C. K. (1965). Oak Ridge National Laboratory Report ORNL-3794.
 KISTENMACHER, T. J., MARZILLI, L. G. & CHANG, C. H. (1973). *J. Amer. Chem. Soc.* **95**, 5817-5819.
 KISTENMACHER, T. J. & SHIGEMATSU, T. (1974*a*). *Acta Cryst.* **B30**, 166-168.
 KISTENMACHER, T. J. & SHIGEMATSU, T. (1974*b*). *Acta Cryst.* **B30**, 1528-1533.
 KISTENMACHER, T. J., SHIGEMATSU, T. & WEINSTEIN, H. (1974). *J. Mol. Struct.* In the press.
 KISTENMACHER, T. J. & SORRELL, T. (1974). *Acta Cryst.* To be submitted.
 KISTENMACHER, T. J. & SZALDA, D. J. (1974). *Acta Cryst.* To be submitted.
 KRAUT, J. & JENSEN, L. H. (1963). *Acta Cryst.* **16**, 79-88.
 LAI, T. F. & MARSH, R. E. (1972). *Acta Cryst.* **B28**, 1982-1989.
 MARZILLI, L. G., KISTENMACHER, T. J. & CHANG, C. H. (1973). *J. Amer. Chem. Soc.* **95**, 7507-7508.
 MARZILLI, L. G., KISTENMACHER, T. J., DARCY, P. E., SZALDA, D. J. & BEER, M. (1974). *J. Amer. Chem. Soc.* **96**, 4686-4688.
 MEESTER, P. DE, GOODGAME, D. M. L., PRICE, K. A. & SKAPSKI, A. C. (1970). *Chem. Commun.* pp. 1573-1574.
 MEESTER, P. DE, GOODGAME, D. M. L., PRICE, K. A. & SKAPSKI, A. C. (1971). *Biochem. Biophys. Res. Commun.* **44**, 510-512.
 MEESTER, P. DE & SKAPSKI, A. (1973). *J. Chem. Soc. Dalton*, pp. 1596-1601.

- PAULING, L. (1960). *The Nature of the Chemical Bond*, 3rd ed. Ithaca: Cornell Univ. Press.
- RAO, S. T. & SUNDARALINGAM, M. (1970). *J. Amer. Chem. Soc.* **92**, 4963–4970.
- RICH, A., DAVIES, D. R., CRICK, F. H. C. & WATSON, J. D. (1961). *J. Mol. Biol.* **3**, 71–86.
- SHEFTER, E. (1968). *J. Pharm. Sci.* **57**, 1163–1168.
- SHEFTER, E., BARLOW, M., SPARKS, R. & TRUEBLOOD, K. (1964). *J. Amer. Chem. Soc.* **86**, 1872–1874.
- SHEFTER, E., BRENNAN, T. F. & SACKMAN, P. (1971). *Chem. Pharm. Bull.* **19**, 746–752.
- SHIKATA, K., UEKI, T. & MITSUI, T. (1973). *Acta Cryst.* **B29**, 31–39.
- SINGH, C. (1965). *Acta Cryst.* **19**, 861–864.
- STEWART, J. M. (1967). X-RAY 67. Technical Report 67-68, Computer Science Center, Univ. of Maryland, College Park, Maryland, U.S.A.
- STEWART, R. F., DAVIDSON, E. R. & SIMPSON, W. T. (1965). *J. Chem. Phys.* **42**, 3175–3187.
- SUNDARALINGAM, M. (1966). *Acta Cryst.* **21**, 495–506.
- SUNDARALINGAM, M. & JENSEN, L. H. (1965). *J. Mol. Biol.* **13**, 930–943.
- SUTOR, D. J. (1958). *Acta Cryst.* **11**, 453–458.
- TAYLOR, M. R. (1973). *Acta Cryst.* **B29**, 884–890.
- VOET, D. & RICH, A. (1970). *Prog. Nucleic Acid Res. Mol. Biol.* **10**, 183–265.
- WILSON, A. J. C. (1942). *Nature, Lond.* **150**, 152.

Acta Cryst. (1975). **B31**, 217

Studies in Molecular Structure, Symmetry and Conformation. VIII. Crystal and Molecular Structure of L-Allothreonine*

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L-Allothreonine, $C_4H_9NO_3$, is orthorhombic, space group $P2_12_12_1$, with $a=13.69$ (1), $b=7.78$ (1), $c=5.17$ (1) Å, $Z=4$. The structure, which is closely isomorphous with that of L-threonine, was solved from this data and has been refined to an R of 10.7% for 597 visual reflexions. The bond lengths and angles are normal.

Introduction

A programme of studies on the structure, symmetry and conformation of organic and biomolecules is in progress in this laboratory. The major aim is to establish the molecular structure of these compounds in the solid state by X-ray methods and to correlate, if possible, the conformation and configuration through other techniques such as o.r.d., n.m.r., etc. Several simple amino acids have been X-ray analysed and their conformational and structural features studied. Recently the isomeric forms of amino acids containing more than one asymmetric carbon atom, namely threonine and isoleucine, have been studied by X-ray analysis (Srinivasan, Varughese & Swaminathan, 1973). This paper deals with the determination of the structure of L-allothreonine.

Experimental

Crystals were obtained by slow evaporation of an aqueous solution of DL-allothreonine (supplied by Nutritional Biochemicals Corporation, Cleveland, Ohio, U.S.A.) at room temperature. On crystallization,

a racemic mixture of D- and L-forms was obtained. The same phenomenon has been reported by Shoemaker, Donohue, Schomaker & Corey (1950) in the case of DL-threonine. The crystals were well defined needles. The cell dimensions and space group were determined by rotation, Weissenberg and precession methods.

Crystal data

$C_4H_9NO_3$, orthorhombic, $a=13.69$ (1), $b=7.78$ (1), $c=5.17$ (1) Å; space group $P2_12_12_1$; $h00$, $h=2n+1$ absent; $0k0$, $k=2n+1$ absent; $00l$, $l=2n+1$ absent. $Z=4$, $M=119.1$, $D_o=1.444$, $D_x=1.437$ g cm $^{-3}$, $\mu=10.68$ cm $^{-1}$, λ (for Cu $K\alpha$ radiation) = 1.5418 Å.

The density was determined by flotation in a mixture of benzene and bromoform.

Intensities were collected by the equi-inclination Weissenberg technique for hkL , $L=0$ to 4 and hkl , $K=0$ and 1. The crystal had dimensions $0.2 \times 0.1 \times 0.1$ mm. The intensities of the various reflexions were determined visually with a set of graded intensities prepared from the same crystal. The intensities were corrected for Lorentz and polarization factors, and for elongation of spots (Phillips, 1962). No absorption correction was applied ($\mu t < 2$). The two sets of data were then correlated by the method of Rollett & Sparks (1960). The discrepancy index between the two sets of

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