sity* to be 2·893 g.cm.$^{-3}$ the number of molecules per unit cell is found to be $Z = 4·092$ i.e. 4. Absent spectra were found to be $(0kl)$ for k odd and $(h0l)$ for $h+l$ odd. Hence the space group is $Pbn2_1$. This can be converted to space group $Pna2_1$ by interchanging the a and b axes. The complete structure analysis is in progress.

* Handbook of chemistry and physics, 39th edition, p.630. Ohio: Chemical Rubber Publishing Co.

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The structure of crystals containing a hydrogen-bonded complex of 1-methylthymine and 9-methyladenine.*

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Introduction

A fundamental structural feature of the two-strand helical configuration proposed by Watson & Crick (1953) for deoxyribose nucleic acid (DNA) is the arrangement of the purine and pyrimidine bases in hydrogen-bonded pairs of adenine–thymine and guanine–cytosine. As part of a program of research on the structure of the nucleic acids in progress at this Institute, we have been interested in the possibility of preparing and determining the structure of crystals containing hydrogen-bonded pairs of these bases as a means of establishing the existence of such arrangements and of providing a direct experimental determination of the dimensions of the molecules and the manner of hydrogen bonding. Even if crystals could be obtained containing the nucleosides thymidine and adenosine in hydrogen-bonded pairs, they would undoubtedly be so complicated that a satisfactory determination of their structure would not be feasible. Use of the simple bases, thymine and adenine, would not be satisfactory because in them the nitrogen atoms 1 and 9, respectively, which in the nucleosides are attached to the sugar deoxyribose, are free for the formation of other hydrogen bonds that might lead to structures very different from the particular hydrogen-bonded structure that may be present in DNA. The most desirable crystal appeared to be one composed of derivatives of thymine and adenine in which the respective 1 and 9 nitrogen positions are blocked by the simplest possible organic radical, namely, methyl. We have now prepared crystals containing hydrogen-bonded pairs of the two compounds, 1-methylthymine and 9-methyladenine, and have definitely established the molecular arrangement and the manner of the hydrogen bonding.

A preparation of 1-methylthymine was made available to us by Prof. James English of the Department of Chemistry, Yale University; through the courtesy of Dr C. P. Rhodes and Dr G. B. Brown of the Sloan-Kettering Institute for Cancer Research and Dr G. H. Hutchings of the Welcome Research Laboratories, we were supplied with crystals of 9-methyladenine.

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1-Methylthymine and 9-methyladenine

Crystals of 1-methylthymine were obtained by evaporation of an aqueous solution at room temperature. The crystals were prismatic in shape with forms {100} and {111} predominating. Crystals of 9-methyladenine grown under similar conditions appeared as needles with forms {010}, {110}, and {001} well developed, the c axis being parallel to the needle axis of the crystal. The space groups and unit-cell dimensions of both compounds were determined from rotation and Weissenberg photographs taken with Cu $K\alpha$ ($\lambda = 1·5418$) radiation. The crystallographic data are tabulated below.

1-Methylthymine	9-Methyladenine
$a = 7·11 \pm 0·03$ Å	$a = 7·67 \pm 0·03$ Å
$b = 11·96 \pm 0·04$	$b = 12·24 \pm 0·04$
$c = 7·52 \pm 0·03$	$c = 8·47 \pm 0·03$
$\beta = 90° 0' \pm 10'$	$\beta = 123° 26' \pm 10'$
Space group: $P2_1/c$	Space group: $P2_1/c$
Density: 1·415 g.cm.$^{-3}$ (meas.)	Density: 1·471 g.cm.$^{-3}$ (meas.)
$Z = 4$	$Z = 4$

Systematic absences (both crystals):

$h0l$ absent for $l = 2n+1$, $0k0$ absent for $k = 2n+1$.

1-Methylthymine-9-methyladenine complex

Equimolecular quantities of 1-methylthymine and 9-methyladenine were dissolved in hot water. Upon cooling and evaporating to dryness at room temperature, good crystals were obtained in the form of monoclinic needles with the needle axis parallel to the b crystallographic axis. Forms {001} and {100} predominated. The crystallographic data as determined from rotation and Weissenberg photographs are:

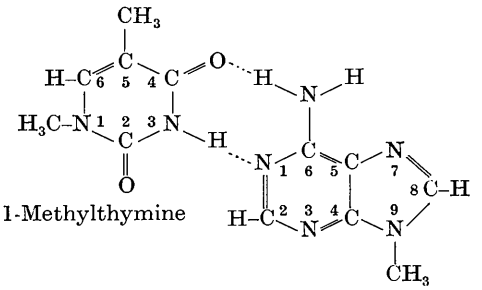
Systematic absences: $0k0$ absent for $k = 2n+1$

$a = 8·28 \pm 0·03$, $b = 6·51 \pm 0·03$, $c = 12·75 \pm 0·05$ Å;
$\beta = 106° 48' \pm 10'$.

Space group: $P2_1$ or $P2_1/m$.
Density: 1·433 g.cm.$^{-3}$ (meas.).

Measurement of the ultraviolet absorption spectrum of the crystals dissolved in 0·1N HCl indicated that they contained both 1-methylthymine and 9-methyladenine. The intensity distributions on the zero, second, and fourth layer Weissenberg photographs taken around the *b* axis were very similar to one another, as were also the intensity distributions on the first and third layer photographs. In addition, the 0*k*0 reflections, as recorded on the zero layer Weissenberg photograph taken around the *c* axis, were all very strong. These observations indicated that the structure is a layer structure and that the atoms lie very close to the (010) plane. Calculations based on the crystallographic data lead to a molecular weight of 286 for each of the two asymmetric units in the unit cell. This is in excellent agreement with the combined molecular weights of 1-methylthymine and 9-methyladenine (289).

The space group was assumed to be $P2_1/m$ with all carbon, nitrogen, and oxygen atoms lying in the mirror plane. The *h*0*l* and *hll* intensity data were used for the calculation of a generalized sharpened vector-density map. This vector-density map was interpreted in terms of an asymmetric unit composed of one molecule of 1-methylthymine and one molecule of 9-methyladenine held together by hydrogen bonds in the manner postulated by Watson & Crick in their proposed structure for DNA.



1-Methylthymine

9-Methyladenine

The first trial structure was characterized by good hydrogen bonding and molecular packing throughout the crystal. Its structural features and its good agreement with the vector-density map indicated strongly that this first trial structure was the correct one. Nevertheless, repeated attempts to refine the atomic coordinates of this structure were unsuccessful.

The fact that the first trial structure could not be refined led to a more critical scrutiny of the generalized projection and a greater emphasis on the significance of certain spurious peaks and on relatively large variations in the heights of peaks that were assumed to represent atoms. The correct structure was finally discovered by changing the positions of a few atoms in the 9-methyladenine portion of the asymmetric unit. The generalized electron-density projection of the correct structure is shown in Fig. 1. It differs fundamentally from the first trial structure in the orientation of the 9-methyladenine with respect to the 1-methylthymine. The amino group of the 9-methyladenine is still connected by means of a hydrogen bond to the oxygen atom attached to C_4 of 1-methylthymine, but the nitrogen atom N_3 of 1-methylthymine is bonded to N_7 rather than to N_1 of 9-methyladenine. The new structure was refined without difficulty by means of least-squares treatment of the *h*0*l* and *hll* data; the present disagreement index R is 16·4%. In the molecular plane, adjacent base pairs are held together by additional hydrogen bonds between the amino group of 9-methyladenine and the oxygen atom attached to C_2 of 1-methylthymine. Further refinement of this structure is in progress.

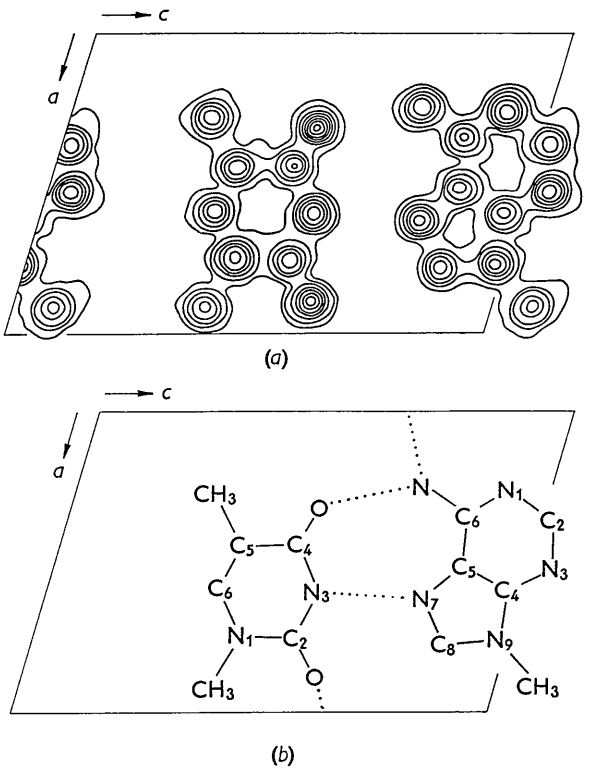


Fig. 1. Generalized electron-density projection of the structure on the (010) plane. The contour lines in (*a*) are drawn at arbitrary equidistant levels. In (*b*) the dotted lines represent the hydrogen bonds described in the text.

Some comments on the possible implication of this structure concerning the arrangements of polynucleotide chains in nucleic acids are being submitted for publication elsewhere.

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