time" on Fig. 2. Clearly, no cloud about the spacecraft can be brighter than the difference (1.1×10^{-12} B⊙) between the smaller day values and those of night. But the experiment is capable of determining a limit half this large.

To confirm the interpretation that the array of data points represents light scattered from a certain antenna with a known position, the situation was simulated in the laboratory with an identical photometer and light shield. The result is the solid curve (Fig. 2) which was normalized to pass thru the orbital data at 68°. The orbital data points appear to be approaching the brightness of the night sky at an antenna angle of 90° in the same manner as the laboratory data do. We may show an upper limit to cloud brightness by assuming that there is such a cloud and by showing that this causes disagreement between the orbital data and the laboratory simulation: If there had been around the spacecraft a cloud whose sunlit brightness in the antisolar direction was 5×10^{-13} B⊙, then we should add this to the ground simulation before a comparison is made with the orbital data. The result (dashed curve, Fig. 2) is obtained by doing this and by normalizing the ground data again to fit the orbital data at an antenna angle of 68°. Since the functional forms are no longer within experimental error of each other, one concludes that there was no cloud as bright as 5×10^{-13} B⊙. This upper limit determined experimentally is equivalent to 0.01 erg cm^{-2} sec^{-1} $sterad^{-1}$, or 12 fifth-visual-magnitude solar-type stars per square degree.

From the last form of the upper limit, there is evidently very little interference in measuring bright stars with instruments which, typically, have fields of view less than several minutes of arc. But the situation is different for observing extended sources of white light, in that the above limit is brighter than the Milky Way and much of the zodiacal light. In the antisolar direction, the true sky brightness is about 1×10^{-13} B⊙, whereas the measured upper limit to the cloud brightness in this direction is five times larger. If the particle size distribution in the cloud were similar to that of the dust which causes the zodiacal light, then this brightness ratio of about 5 would be maintained over all portions of the zodiacal light to within 30° of the sun. Closer to the sun than 30°, scattering from free electrons enhances the zodiacal light and solar corona so that the ratio would fall, perhaps to 1. It is important to study the amount of solid debris in the vicinity of a spacecraft and to investigate the rates of formation and release of micron-size particles. Otherwise, observations of dim extended sources might be compromised whenever a large satellite is in sunlight.

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20 September 1967

Crystal Structure of the 1 : 1 Complex of 5-Fluorouracil and 9-Ethylhypoxanthine

Abstract. *9-Ethylhypoxanthine and 5-fluorouracil form a 1 : 1 crystalline complex. The structure of this complex has been solved by x-ray diffraction analysis. The molecules crystallize in a monoclinic lattice and form a sheet structure in which pairs of fluorouracil molecules are held together by two hydrogen bonds. The 9-ethylhypoxanthine residues fill up the rest of the molecular sheet by forming single hydrogen bonds with each uracil pair.*

The cocrystallization of two different molecules into the same lattice is usually a reflection of interactions which occur between these molecules in solution. A particularly important type of cocrystallization is that which has been observed with purine and pyrimidine derivatives of nucleic acids. It has been shown in a number of crystallographic studies that adenine derivatives cocrystallize with uracil (or thymine) derivatives (*1*, *2*), and guanine derivatives with those of cytosine (*3*). In these crystal structures, the purine-pyrimidine pairs are held together by two or three hydrogen bonds. The molecular packing and hydrogen bonding between these residues is of interest since many of them have a direct bearing on the molecular organization of the macromolecular nucleic acids. In these latter molecules, the specificity of hydrogen bonding plays a central role in the transmission of genetic information, in the production of messenger RNA as well as in the polymerization of amino acids to form proteins. Thus there is considerable interest in the way purines and pyrimidines associate with each other.

One of the characteristics of the crystallographic studies carried out in the past is the fact that only adenine derivatives have crystallized with uracil derivatives and likewise guanine derivatives with cytosine derivatives. No cases have been reported in which there has been cocrystallization of members of the adenine-uracil family with members of the guanine-cytosine family. Infrared and nuclear magnetic resonance studies (*4*) carried out on the association of these molecules in solution shows that there is a selective hydrogen bonding affinity between adenine and uracil derivatives and between guanine and cytosine derivatives. However, there is little or no hydrogen bonding association between members of these two families. Accordingly, it was of considerable interest when we found that it was possible to cocrystallize 5-fluorouracil with 9-ethylhypoxanthine. This appeared initially to be inconsistent with the specificity described above, since hypoxanthine is considered a derivative of guanine. Here we report the results of the crystal structure analysis of this 1 : 1 complex. Even though these molecules have crystallized together, they do not form pairs held together by two hydrogen bonds. Instead, there is a pairing of the uracil derivatives with each other, and the hypoxanthine derivatives are connected to these pairs with single hydrogen bonds.

5-Fluorouracil and 9-ethylhypoxanthine (Cyclo Chemical Co., Los Angeles) were dissolved in equimolar amounts in water and allowed to evaporate at room temperature. Well-developed prismatic crystals were formed, some of which had a diameter as great as 1 mm. Single crystals were isolated and redissolved and were

shown to have a 1 : 1 composition by spectrophotometric and chromatographic analyses. The crystals were found to be monoclinic, and the crystallographic data are presented in Table 1. The asymmetric unit contains one molecule each of 5-fluorouracil and 9-ethylhypoxanthine, and the unit cell contains four such pairs. The diffraction data were collected on a four-circle Picker automatic diffractometer out to a 2θ maximum value of 125°. This included a total of 1632 independent reflections, excluding systematic absences. Of these, 1464 reflections were above the background. The crystal measured 0.3 by 0.3 by 0.5 mm and no absorption correction was applied to the data.

The structure was solved by the reiterative application of the Sayre equations (*5*). The structure factors were reduced to normalized structure factors (E), and the method was applied with a program written by Long (*6*). This program produces various possible combinations of signs, and the correct one was among the first four which were examined. The first Fourier synthesis (E map) carried out with these phases revealed 20 out of the 21 atoms in the structure, excluding the hydrogen atoms. These 20 atoms were then refined isotropically by using a least squares program (*7*). After two cycles of refinement, the residual factor was reduced to 23.4 percent using all the data. A difference Fourier was calculated at that stage and the terminal carbon atom of the ethyl group was revealed. Anisotropic temperature factors were then introduced, and further refinement reduced the residual to 8.9 percent. Eight very strong reflections and the very weak reflections were then excluded and a difference Fourier was calculated from the remaining data. All of the hydrogen atoms were revealed at this stage except for two hydrogens found at the end of the ethyl group. These appeared in the difference Fourier after further refinement. Positional and thermal parameters for the hydrogen atoms were not refined. The final residual factor was 5.5 percent for the 1464 observed reflections.

The molecules form a layer structure in which they are organized into a hydrogen-bonded network, and these sheets are stacked, held together by van der Waal's interactions. The sheets are tilted off the *bc* plane at an angle of 46°42′, separated by a spacing of 3.24 Å. All of the atoms are in the plane of the sheet with the exception of the terminal carbon atom of the ethyl group, which lies halfway between the sheets. Figure 1 shows a section of the electron density map in the plane of the molecules. All of the atoms are seen in this plane except the terminal

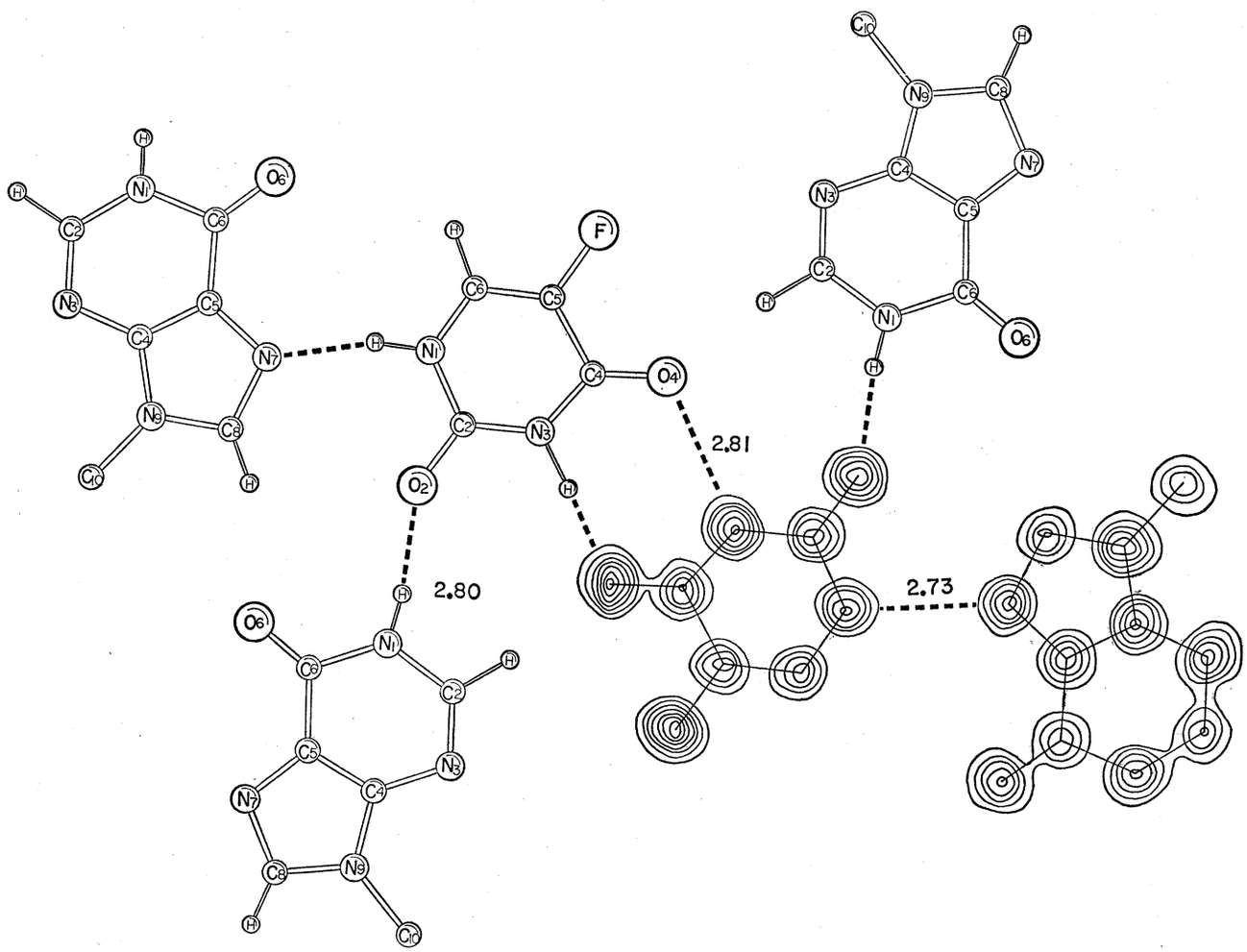

Fig. 1. The electron density map in the plane of the molecular sheet of the 5-fluorouracil : 9-ethylhypoxanthine structure. The contours occur at arbitrary levels of electron density in this section. The terminal carbon atom (C-11) on the ethyl group of 9-ethylhypoxanthine does not lie on the plane. The hydrogen bond lengths are indicated in angstrom units. The center of symmetry lies between the paired fluorouracil molecules.

Table 1. Crystallographic data for the 1 : 1 complex of 5-fluorouracil and 9-ethylhypoxanthine.

Space group	$P2_{1/c}$ $Z = 4$
Unit cell	$a = 4.656$ Å $b = 15.276$ Å $c = 17.807$ Å $\beta = 90°48'$
Calculated density	1.54 g/cm³
Observed density	1.55 g/cm³

carbon atom of the ethyl group. The 5-fluorouracil molecules are paired, held together by two hydrogen bonds of length 2.81 Å between N-3 and O-4. They are organized around a center of symmetry. This is the same pairing of uracil residues which is seen in the crystal structure of N-1-methyl uracil (*8*). The 9-ethylhypoxanthine molecules are connected to each fluorouracil pair by single hydrogen bonds. The hydrogen atom attached to the N-1 of hypoxanthine is bonded with O-2 of fluorouracil, while the hydrogen atom on N-1 of uracil is bonded to N-7 of a different hypoxanthine residue. The latter hydrogen bond is short (2.73 Å), as is frequently found in association with the purine imidazole group (*2*). The packing is quite compact and there are no open areas in the plane of the molecular sheet.

Although it is not shown in Fig. 1, the fluorine atom is in contact with the terminal carbon atoms of the ethyl side chain of hypoxanthine. However, it does not participate in any hydrogen bonding. The fluorine atoms are located in a column parallel to the *a*-axis, surrounded by ethyl groups from neighboring hypoxanthine molecules. There is very little overlap of the purine and pyrimidine rings of one layer with those of the layers on either side. A small amount of overlap occurs with the imidazole rings of hypoxanthine and N-1 and C-2 of the uracil rings immediately below. With this exception, the π-electrons of the unsaturated rings are not in contact with other π-electrons. Thus this crystal does not show a stacking of purine residues which is seen prominently in several layer structures involving cocrystals of adenine and uracil derivatives (*2*). Where overlapping of unsaturated rings does occur, the spacing between the adjacent layers is near 3.4 Å, as is found in the stacked bases of the nucleic acids. Partial overlap in the present structure is associated with a spacing of 3.24 Å. In the crystal of N-1-methyl uracil, the pyrimidine rings form a planar net with no overlap and there the spacing between the layers is 3.14 Å (*8*). The bond angles and distances of the purine and pyrimidine rings are similar to those which have been reported earlier in other crystallographic studies of similar compounds (*9*). Table 2 lists the atomic coordinates of all the atoms in this structure except hydrogens.

Table 2. Fractional atomic coordinates.

Position	*x/a*	*y/b*	*z/c*
	9-Ethylhypoxanthine		
N-1	0.8514	0.3024	0.3389
C-2	.0290	.2533	.3843
N-3	.0404	.1687	.3844
C-4	.8516	.1338	.3331
C-5	.6667	.1759	.2857
C-6	.6583	.2690	.2855
N-7	.5131	.1169	.2427
C-8	.6085	.0395	.2650
N-9	.8149	.0457	.3201
C-10	.0388	.4744	.1405
C-11	.2126	.4417	.0757
O-6	.5088	.3184	.2460
	5-Fluorouracil		
N-1	0.0812	0.1305	0.1380
C-2	.9950	.0508	.1143
N-3	.7709	.0493	.0622
C-4	.6333	.1216	.0333
C-5	.7440	.2034	.0616
C-6	.9565	.2060	.1124
O-2	.8930	.4831	.3635
O-4	.4323	.3855	.4874
F	.6178	.2762	.0353

This crystal structure is of interest since it contains a guanine derivative and a uracil derivative. As such it might seem to violate the type of complementary pairing relationship which has been seen in the other crystal structures of base pairs. Both of these molecules have hydrogen donor and acceptor sites. However, an infrared study of hypoxanthine derivatives and uracil derivatives shows that there is no selective hydrogen bonding between these molecules in solution (*10*). The hydrogen-bonded pair involving two fluorouracil derivatives is similar to that found in N-1-methyl uracil (*8*) and this may also be related to the helical double-stranded hydrogen-bonded form of polyuridylic acid which is stable at lower temperatures (*11*). However, it is likely that the self-dimerization of fluorouracil is energetically less favorable than its association with adenine derivatives. The 1 : 1 crystalline complex of 9-ethyladenine and N-1-methyl-5-fluorouracil shows that these molecules form dimers held together with two hydrogen bonds (*12*).

In the present structure, the hypoxanthine molecules appear to fill space fully in the crystal lattice, and they are connected to individual uracil residues by single hydrogen bonds rather than a pair. In solution studies it has been shown that hydrogen bonding association between two molecules connected with a single hydrogen bond is much less stable than with a pair of bonds (*4*). In the present structure, the single hydrogen bond connecting the hypoxanthine with the fluorouracil pair undoubtedly stabilizes the crystal lattice, but it is unlikely that this could confer specificity to the interaction in a biological system. The existence of this 1 : 1 complex is probably determined more by the crystal lattice energy and the excellent packing and stacking than by a strong association of its component molecules. Thus the analysis of the present complex does not violate our understanding of the complementary nature of the hydrogen bonded pairs found between the purines and pyrimidines in the nucleic acids.

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18 August 1967