EFFECTS OF HALOGEN SUBSTITUENTS ON BASE STACKING IN NUCLEIC ACID COMPONENTS: THE CRYSTAL STRUCTURE OF 8-BROMOGUANOSINE

Charles E. Bugg*
Institute of Dental Research
and Department of Chemistry
University of Alabama Medical Center
Birmingham, Alabama 35233

Ulf Thewalt**
A. A. Noyes Laboratories
of Chemical Physics***
California Institute of Technology
Pasadena, California

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The crystal structure of 8-Bromoguanosine dihydrate was determined from three-dimensional diffractometer data. Base stacking in the structure involves close contacts between the bromine atom and the purine moiety of a crystallographically related nucleoside. Comparison with the crystal structure of guanosine suggests that halogen substituents may modify solid state base stacking patterns.

The physical, chemical and biological properties of polynucleotides are modified considerably when halogenated pyrimidines are substituted for the naturally occurring bases (Grunberg-Manago and Michelson, 1964 a, b; Howard et.al., 1969; Howard and Tessman, 1964; Inman and Baldwin, 1964; Massoulié et.al., 1966; Michelson and Monny, 1967; Tessman et.al., 1964). In view of extensive data supporting the importance of vertical stacking of purine and pyrimidine bases as a stabilizing force in nucleic acids (Ts'o, 1968), the effects of halogenated bases have been attributed to the ability of halogen

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Present Address: Mineralog. Institut of the University, Marburg/Lahn
Deutschhausstr. 10, West Germany

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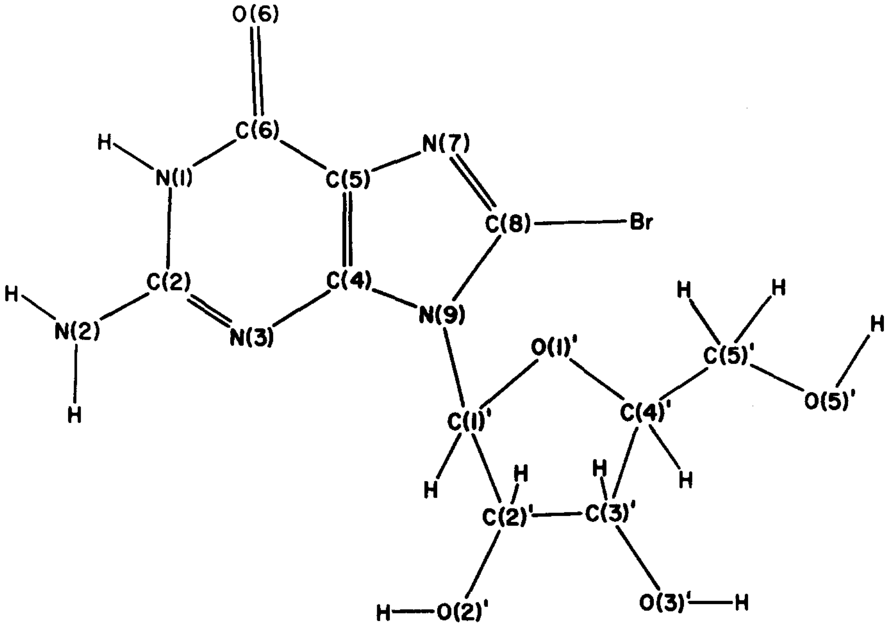
atoms to alter base-stacking interactions (Howard et.al., 1969; Massoulié et.al., 1966); evidence supporting this possibility has been provided by studies which demonstrate that base stacking in aqueous solution occurs to a greater extent for 5-bromouridine than for thymidine or uridine (Ts'o, 1968). We report here the crystal structure of 8-bromoguanosine (I) which suggests that halogen substituents also affect base stacking interactions in the solid state.



(I)

METHODS

Orthorhombic crystals of 8-bromoguanosine dihydrate were obtained by slowly cooling a hot, saturated aqueous solution. The space group is $P2_12_12_1$ and the unit cell dimensions are a = 24.541, b = 12.008 and c = 4.948 Å. Intensity data were collected on a Datex-automated General Electric XRD-5 diffractometer using iron-filtered cobalt radiation and a $\theta - 2\theta$ scan technique. Each of the 1208 unique reflections in the range $7° \leq 2\theta \leq 154°$ was measured.

The structure was solved by the heavy-atom method and was refined by three-dimensional, block-diagonal least squares. The nine hydrogen atoms which could be positioned on stereochemical grounds were included in the refinement; the remaining hydrogen atoms were ignored. The positional parameters, along with anisotropic temperature factors for the heavy atoms and isotropic temperature factors for the hydrogen atoms, were refined to give a final R index of 0.052.

RESULTS AND DISCUSSION

The nucleoside is in the syn conformation with ϕcn, the torsion angle around the glycosidic linkage (Donohue and Trueblood, 1960; Sundaralingam and Jensen, 1965), equal to 127°. This conformation is similar to that found for deoxyguanosine in the crystal structure of deoxyguanosine-5-bromodeoxycytidine (Haschemeyer and Sobell, 1965). Also similar to deoxyguanosine, 8-bromoguanosine has an intramolecular hydrogen bond between atoms O(5)' of the sugar ring and N(3) of the base, and the ribose ring is puckered with atom C(2)' displaced to the same side of the plane of the other four ring atoms as atom C(5)'.

An interesting feature of the crystal structure is the manner in which parallel purine rings are stacked. This stacking, as viewed perpendicular to the base plane, is shown in figure 1. Planes of adjacent purine rings are separated by 3.48 Å. There is very little overlap of the bases; the major contact between stacked bases involves interactions between the bromine substituent and the ring system of an adjacent purine. The distance between the bromine atom and atom C(5) of a parallel base is 3.464 Å which is somewhat shorter than the expected sum of the atomic van der Waals radii (3.65 Å) (Pauling, 1960).

When the crystal structure of 8-bromoguanosine is compared with that of guanosine (Bugg et.al., 1968), it appears that the bromine substituent has a marked influence on the solid state base stacking. The two different modes of stacking found in the crystal structure of guanosine are shown in figures 2(a) and 2(b); in both modes, parallel guanine moieties are essentially superimposed with a great deal of base overlap. The fact that crystals of inosine and guanosine are isostructural (Bugg et.al., 1968), in spite of differences in hydrogen bonding,

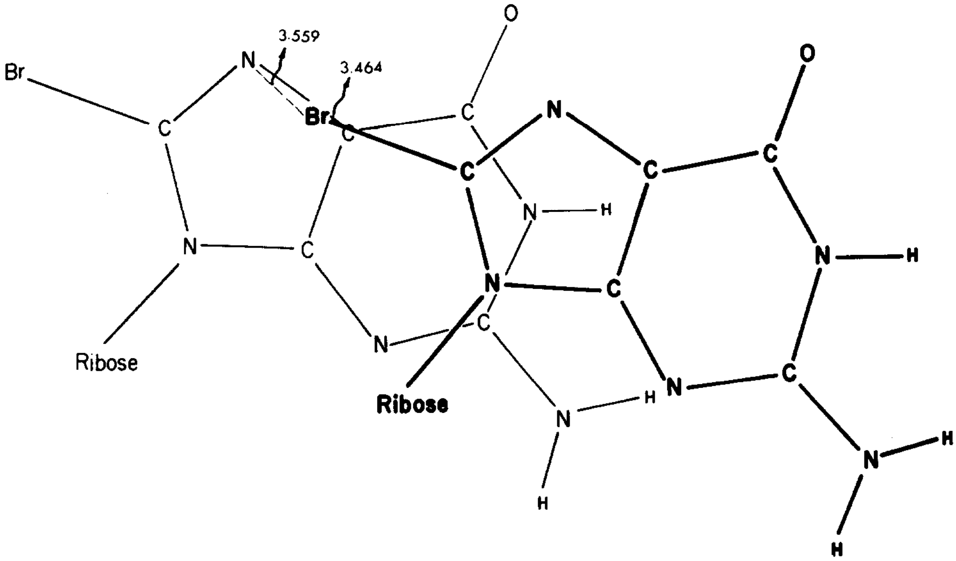


FIGURE 1. Base stacking in 8-Bromoguanosine as viewed perpendicular to the purine plane.

implies that intimate base overlap is an important stabilizing feature in these structures. Therefore, it is interesting that a different type of stacking, characterized by close bromine-purine contacts, is found in the crystal structure of 8-bromoguanosine.

As in the crystal structures of most other nucleic acid constituents (Sundaralingam et.al., 1969; Bugg et.al., 1969), dipole-induced dipole and London dispersion forces appear to be primarily responsible for the base stacking association in the crystal structure of 8-bromoguanosine. Since carbon and bromine differ in electronegativity (Pauling, 1960), the C(8)-Br bond should possess a considerable moment which could interact with the polarizable pi electron system of a neighboring base by inductive effects as described by Wallwork (1961) and Briegleb (1932, 1949). London dispersion forces, taking advantage of the polarizability of the bromine atom, would be expected to further stabilize this stacked configuration. Similar interactions, involving the positioning of halogen atoms over aromatic rings, occur in a

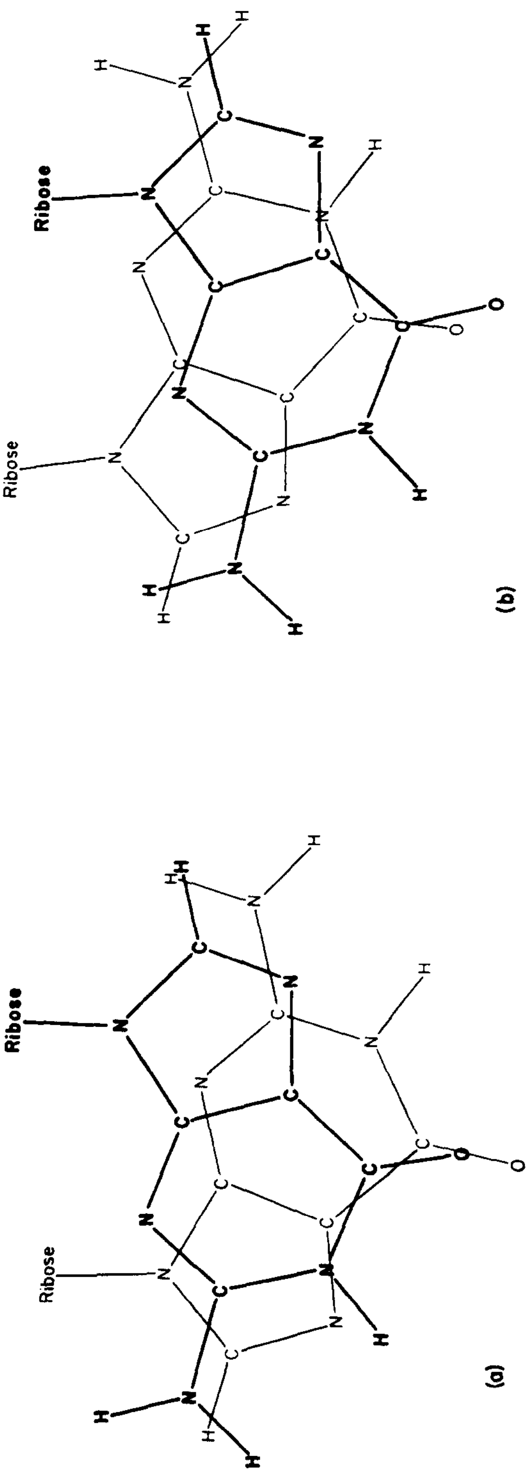


FIGURE 2. Two types of base stacking in guanosine as viewed perpendicular to the purine plane.

number of other crystal structures of halogenated purines and pyrimidines (Bugg et.al., 1969) and also seem to be an important structural feature in some donor-acceptor complexes (Hassel and Stromme, 1958; Strieter and Templeton, 1962). It seems likely that interactions between halogen substituents and purines and pyrimidines may also account for the effects which halogenated bases have on the physical, chemical and biological properties of polynucleotides and nucleic acids.

We have recently learned that Dr. S. S. Tavale and Dr. Henry M. Sobell have also determined the crystal structure of 8-bromoguanosine dihydrate. They have kindly supplied us with their unpublished results which are not significantly different from ours. Since they have submitted their complete structural data for publication we do not intend to publish any additional information about this structure. We shall gladly furnish complete structural data upon request.

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