Cytidylic Acid "a" Trihydrate: Structure and Conformation

Abstract. *Cytidylic acid "a" (cytidine 2′-monophosphate) crystallizes in the triclinic space group* P1 *with two molecules of cytidine monophosphate ($C_9H_{14}O_8N_3P$) and six molecules of water in the unit cell. X-ray analysis of this crystal shows that in both molecules, the base is in the* anti *conformation, the ribose ring is C(2′)-*endo *puckered, and the hydroxyl O(5′) is* gauche-gauche. *The two molecules are linked by a short hydrogen bond through the phosphate oxygens.*

The two forms of cytidylic acid, cytidine 2′-monophosphate (2′-CMP) and cytidine 3′-monophosphate (3′-CMP), are well known for their inhibitory action of the enzyme ribonuclease. While the modes in which 2′-CMP and 3′-CMP bind to the enzyme are believed to be similar, the effect of binding on the conformation of both the enzyme and the inhibitor are considered to be significantly different (*1*). Moreover, the presence of 2′-CMP has been found to suppress the temperature-induced optical rotatory change characteristic of ribonuclease A (*2*). While data have been reported for two crystalline modifications of 3′-CMP (*3, 4*), no structural data are available for 2′-CMP. In this report we note the results of the crystal structure analysis of a hydrated form of 2′-CMP.

Hydrated cytidylic acid "a" crystallizes in the triclinic system with unit cell parameters $a = 9.64$, $b = 13.67$, $c = 7.46$ Å; and $\alpha = 106.0°$, $\beta = 98.5°$, $\gamma = 118.4°$. The space group is $P1$ with two molecules of 2′-CMP and six molecules of water in the unit cell. The intensity data, consisting of 3472 independent reflections, were collected manually with a General Electric (model XRD-3) diffractometer by the stationary crystal, stationary counter method. The positions of the two phosphate groups were established from sharpened Patterson maps, and the complete structure was obtained by an iterative structure factor calculation, a least squares refinement, and an electron density calculation. All atoms except the phosphorus atoms were assigned isotropic thermal parameters in the final stages of the least squares cycles, the final reliability index R being 13 percent. The intramolecular distances and angles at this stage compared well with those for the orthorhombic (*5*) and monoclinic forms of 3′-CMP. The orthogonal projection of the *ab* plane showing the two molecules (A and B) of 2′-CMP and the six water molecules is shown in Fig. 1. Both A and B have very similar molecular features, the only exception being the orientation of the phosphate groups about the C(2′)-O(2′) bond. The pyrimidine rings are essentially planar with the O(2) and N(4) atoms on opposite sides of the mean plane. In molecule A, atom C(1′) is also found to be out of the plane by about 0.1 Å and on the same side as O(2). The bond angles suggest that the nitrogen atoms N(3) in both pyrimidine rings are protonated (*6*).

The torsion angles describing the conformation of molecules A and B as well as the corresponding values reported for the orthorhombic and monoclinic forms of 3′-CMP are given in Table 1. The two molecules are linked by a strong hydrogen bond (2.48

Table 1. Comparison of the torsion angles in 2′-CMP (molecules A and B) and orthorhombic (*5*) and monoclinic (*4*) 3′-CMP. Values are in degrees.

Torsion angle	2′-CMP Molecule A	2′-CMP Molecule B	3′-CMP Orthorhombic	3′-CMP Monoclinic
C(4′)-O(1′)-C(1′)-C(2′)	−20	−23	−19.8	−17.6
O(1′)-C(1′)-C(2′)-C(3′)	33	33	36.6	34.6
C(1′)-C(2′)-C(3′)-C(4′)	−30	−30	−38.8	−37.5
C(2′)-C(3′)-C(4′)-O(1′)	19	18	28.2	27.9
C(3′)-C(4′)-O(1′)-C(1′)	−1	−3	−6.7	−7.1
O(1′)-C(1′)-N(1)-C(6)	−45	−51	−41.8	−39.3
C(2′)-C(1′)-N(1)-C(6)	73	66	74.7	76.9
C(1′)-C(2′)-O(2′)-P	109	103		
C(2′)-C(3′)-O(3′)-P			91.5	111.0
C(3′)-C(4′)-C(5′)-O(5′)	55	52	43.8	45.5
O(1′)-C(4′)-C(5′)-O(5′)	−66	−67	−74.7	−73.7
O(2′)-C(2′)-C(3′)-O(3′)	−37	−33	−42.3	−48.3
O(7)-P-O(2′)-C(2′)	165	76		
O(7)-P-O(3′)-C(3′)			68.0	170.7

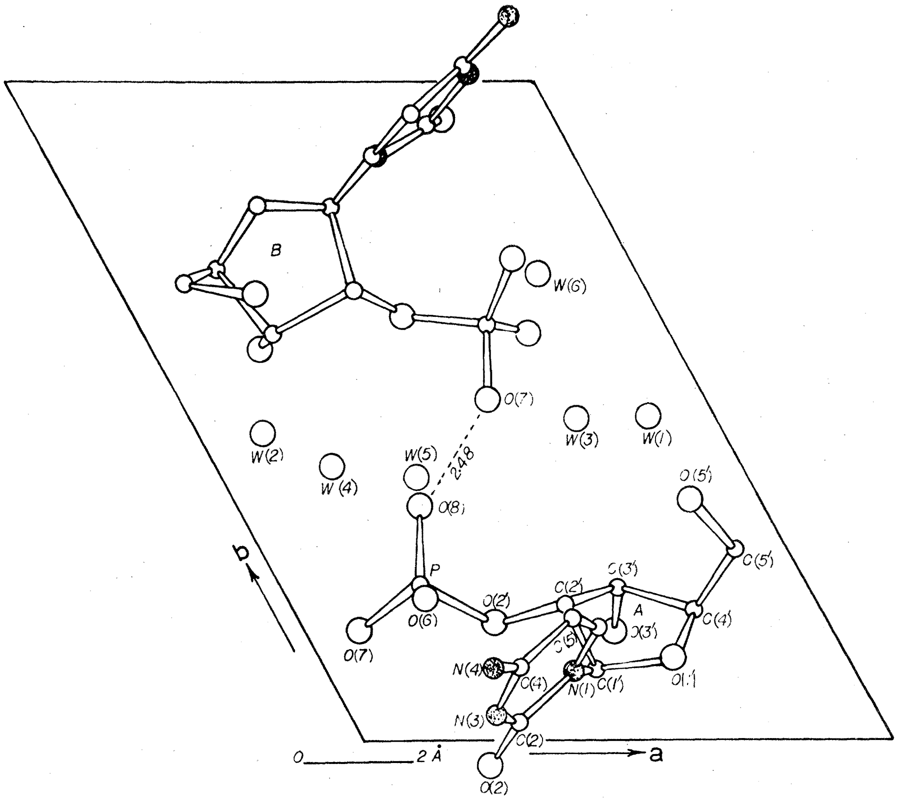

Fig. 1. Orthogonal projection on the *ab* plane, showing two molecules of 2′-CMP and six molecules of water.

Å) through the phosphate oxygens, a situation also found in the orthorhombic form of 3′-CMP. The mean planes of the pyrimidine rings of the two molecules are inclined to each other by 13°. The structure is also stabilized by other hydrogen bonds formed through the water molecules.

A study of the torsion angles shows that the bases in both molecules are in the *anti* conformation, which is considered energetically more favorable (*7*). In both molecules the hydroxyl O(5′) orients itself in the *gauche-gauche* configuration. The puckering of the sugar ring is found to be C(2′)-*endo* in both molecules, the C(2′) atoms being displaced by 0.49 Å (molecule A) and 0.56 Å (molecule B) from the mean plane through the remaining four atoms in the respective sugar rings. In standard notation, the envelope (*E*) conformation of the sugar rings may be described as ${}^2E$ for both molecules (*8*). Earlier nuclear magnetic resonance studies have also strongly favored the C(2′)-*endo* puckering of the molecules in solution. Such a puckering results in the least energy barrier to rotation about the glycosidic bond (*9*). This may also result in the less energetically favored *syn* conformation of the pyrimidine ring becoming permissible under special conditions, such as binding of the molecule to the enzyme (*1*).

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Polar Desert Adaptations of a High Arctic Plant Species

Abstract. *Plants of* Saxifraga oppositifolia *(Saxifragaceae) possess metabolic adaptations that allow them to grow successfully in polar desert microenvironments. Net photosynthesis (net carbon uptake) continues to be positive during drought until the leaf water stress declines to the range of −21 to −29 bars, which is considerably below the nonstress range of 0 to −10 bars. The plants can survive leaf water stresses of at least −44 bars in the field and leaf water stresses of −55 bars in a growth chamber.*

The interacting factors of restricted moisture availability and a short low-temperature growing season make polar desert environments among the most severe for plants (*1*). The lack of a regularly recurring summer moist period has resulted in the exclusion of most vascular plants, including all annuals, from arctic polar desert sites. Only about two dozen species of perennial vascular plants occur in the hundreds of thousands of square kilometers of true arctic polar desert habitats (*2*). These plants complete their annual cycle of growth in an environment characterized by low soil moisture contents, droughts which are unpredictable in both occurrence and duration, and low total precipitation. The severity of drought in polar deserts suggests that those species which do occur should possess adaptations of growth and metabolism similar to those of at least some of the perennial plants found in deserts of more temperate regions.

Saxifraga oppositifolia L., a semiwoody cushion plant, is a long-lived perennial with a circumpolar distribution in the Arctic and in some alpine regions. The species occupies a broad range of habitats, including many polar desert sites (*3*). I studied several populations of this species on polar desert microsites in Truelove Lowland on the north coast of Devon Island (76°N) in the Canadian Arctic Archipelago. Most of the surface of the lowland is mesic to wet tundra meadow, but a series of raised beach ridges in the lowland provides well-drained polar desert microsites. The research sites were on beach ridges located in the middle of the lowland.

The beach ridge surfaces (*4*) are covered by a desert pavement of pebbles overlying several meters of coarse gravel. The plant cover is a sparse polar desert vegetation consisting primarily of low-growing plants of *Salix arctica* and *Saxifraga oppositifolia*, along with scattered individuals of about seven other species of vascular plants. The vascular plant density averages about 45 individuals per square meter, with a total cover of about 17 percent of the ground surface. Lichens cover many of the pebbles of the pavement, with windblown lichen fragments present in depressions. The total lichen cover frequently exceeds 50 percent on the dry beach ridges. Mosses generally have a maximum cover of 1 percent.

At this latitude, the sun remains above the horizon from late April until the third week of August. Flowering and leaf growth of *Saxifraga oppositifolia* on the exposed beach ridges begin by the end of the first week of June. Fruit and leaf development can be temporarily interrupted by drought or snow cover during the growing season, and then continue when conditions become more favorable.

The records of 1969 through 1971 indicate that the total precipitation of the "usual" growing season on this lowland (mostly scattered rain showers) is less than 50 mm. During all 3 years of this study there were periods of several weeks in the growing season with no precipitation. The research sites were free of snow from late June until late August or early September. The zone of thaw had progressed below a depth of 15 cm (below most roots) prior to 10 June. There was a drought in each of the three growing seasons. The most severe drought occurred during 1971, when no precipitation fell from the time of snowmelt until early July. During this drought, the soil water availability remained below −15 bars for about 4 weeks (Fig. 1). The drought coincided with relatively high levels of solar radiation resulting in leaf water stresses (ψ_{leaf}) of between −25 and −35 bars in plants of *Saxifraga oppositifolia* during midday (*5*). The maximum leaf water stress on a clear day was −44 bars; the plants survived this stress. The atmospheric vapor pressure deficits during this period ranged between 0.3 and 4.0 mm-Hg. In 1969 and 1971, the period of drought occurred early in the growing season. In 1970, however, the drought lasted from late July until mid-