by slightly more than two esd's, suggesting that the delocalization of the C–O bond at the 3 position has weakened the O–H bond there, thus making the 3 position hydroxyl hydrogen more labile. 8-Azaestradiol has only weak estrogenic activity. A private communication from R. E. Brown of Warner-Lambert Pharmaceuticals (Sept. 27, 1971) states..."Uterotrophic activity was assayed by the procedure of Rubin, *et al.* [*Endocrinology*, **49** (1951)] by Dr. R. Kroc and Mr. C. Rassaert of the Warner-Lambert Research Institute. At a dose of 150 mg/kg po, 8-azaestradiol was practically devoid (0.01% that of ethynyl estradiol) of estrogenic activity...." Yet this compound shows almost identical molecular parameters with estradiol. Therefore, its lack of estrogenic activity must be caused by the competition between the admittedly weak protonation at the phenolic oxygen (attached to position 3) and the potential protonation of the nitrogen at the 8 position during the initial glucuronidation. Without this acid-catalyzed protonation at the normal 3 position, the sequence of reactions necessary to stimulate estrogenic activity will be suppressed.

The two previous studies of azasteroids showed no hydrogen bonding between either the oxygen or nitrogen. This was attributed to the presence of the bromide ion. This study does show hydrogen bonding similar to that in estriol. The hydrogen bonded to O-3 is 1.78 Å from O-17 (in equivalent position $1 - x$, $^1/_2 + y$, $^3/_2 - z$) and forms the angle O-3–H···O-17, 172°, whereas in estriol the corresponding parameters are 1.80 Å and 177°, respectively. In addition, the hydrogen at O-17 is 1.92 Å from the nitrogen (equivalent position x, $^1/_2 - y$, $^1/_2 + z$) and forms the angle O-17–H···N, 176°.

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Stereochemistry of Nucleic Acids and Their Constituents. XXVI. The Crystal and Molecular Structure of a 1:1 Adduct of Ethanol and 3′-Deoxy-3′-(dihydroxyphosphinylmethyl)adenosine, an Analog of Adenosine 3′-Monophosphate[1]

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Abstract: The structure of 3′-deoxy-3′-(dihydroxyphosphinylmethyl)adenosine a synthetic analog of adenosine 3′-phosphate, has been determined using 1417 reflections measured on a diffractometer. The compound crystallized in the space group $P2_12_12_1$ with four molecules of the analog and four molecules of ethanol in a unit cell having $a = 5.592$, $b = 20.287$, and $c = 15.198$ Å; $d_{calcd} = 1.507$ g cm^{-3} and $d_{obsd} = 1.503$ g cm^{-3}. The observed and calculated densities are consistent with the presence of four molecules of ethanol per unit cell, which was subsequently confirmed by X-ray analysis. The molecule is in the anti conformation with respect to the glycosyl torsion angle, having $\chi = 28.1°$. The conformation about the C(4′)–C(5′) bond is gauche–gauche, where O(1′)–C(4′)–C(5′)–O(5′) = −69.0° and C(3′)–C(4′)–C(5′)–O(5′) = 49.2°. The conformation of the sugar ring is $^3T_2$, the torsion angles about the ring bonds being O(1′)–C(1′) = 8.9°, C(1′)–C(2′) = −29.5°, C(2′)–C(3′) = 37.4°, C(3′)–C(4′) = −33.1°, C(4′)–O(1′) = 15.4°. These conformational parameters are similar to those found in adenosine 3′-phosphate dihydrate. The molecule is a zwitterion N(1) of the base being protonated by an adjacent phosphonate hydrogen. The same phosphonate group is also hydrogen bonded to N(6) of the base. Similarly, the second hydrogen on N(6) and the site N(7) are involved in a hydrogen-bonded pair to a symmetry related phosphonate group. The base–phosphonate (or phosphate) hydrogen bonding is a characteristic feature of the crystal chemistry of adenine and cytidine nucleotides. The alcohol of solvation is hydrogen bonded to an adjacent ribose O(5′) atom. The remaining potential hydrogen bonding sites are also involved in hydrogen bonding.

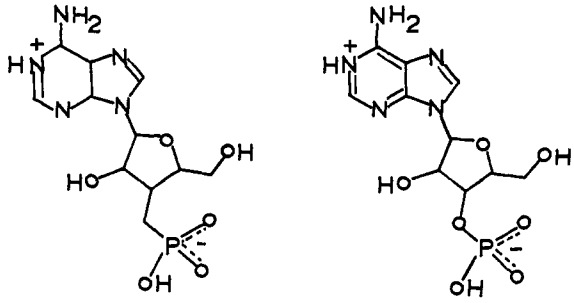
The syntheses of phosphonate analogs of nucleoside phosphates have been of considerable interest since the compounds are of use in the study of the mechanisms of enzyme and hormone action.[2] This, in turn, makes a description of the three-dimensional structures of these analogs, relative to the structures of the naturally occurring phosphates, of particular interest in terms of the physical basis of their biological activities. The structure of adenosine 3′-phosphate dihydrate (3′-AMP) has been described in detail previously.[3] We now describe the structure of 3′-deoxy-

(1) For part XXV of this series, see P. Pyusiner and M. Sundaralingam, *Acta Crystallogr.*, in press.

(2) H. P. Albrecht, G. H. Jones, and J. G. Moffatt, *J. Amer. Chem. Soc.*, 92, 5511 (1970).

(3) M. Sundaralingam, *Acta Crystallogr.*, **21**, 495 (1966).

3′-(dihydroxyphosphinylmethyl)adenosine (**1**)[2] (Figure 1), a methylene analog of 3′-AMP (**2**) with particular emphasis on the structural similarities and differences between this compound and the natural metabolite. In the following paper, the structure of the cyclic 3′,5′-phosphonate analog of 3′,5′-cyclic adenosine monophosphate is described.[4] Some of the methylene analogs of the nucleotides have been shown to possess biological activity.[5]



1 2

Experimental Section

A crystalline sample of **1** ($C_{11}H_{16}N_5O_6P$, mol wt 346) was obtained as the free acid from Dr. J. G. Moffatt, Institute of Molecular Biology, Syntex Research, Palo Alto, Calif. The compound had been crystallized from aqueous ethanol as needles. A single crystal was chosen for X-ray analysis and mounted on a glass fiber along the needle axis *a*. Analysis of the oscillation and Weissenberg photographs indicated that the crystal system was orthorhombic; the systematic absences *h*00, 0*k*0, 00*l*, *h*,*k*,*l* = 2*n* + 1 indicated the space group to be $P2_12_12_1$. The cell constants, determined from measurements on medium angle reflections on a Picker automatic diffractometer with Cu Kα (λ = 1.5418 Å) radiation, were found to be $a = 5.592 \pm 0.001$, $b = 20.287 \pm 0.002$, $c = 15.198 \pm 0.002$ Å. The density of the crystal, determined by the method of flotation in CCl_4–$CHCl_3$, is 1.503 g cm^{-3}, which is in good agreement with the value of 1.507 g cm^{-3}, based on four molecules of the nucleotide and four molecules of ethanol in the unit cell. Complete three-dimensional intensity data up to 2θ = 127° were collected on a Picker automatic diffractometer with Ni-filtered Cu radiation employing the θ–2θ scan technique. The crystal was mounted along the *a* axis and parallel to the ϕ axis of the goniostat. A total of 1769 reflections was recorded. An analysis of the intensities of the systematically absent reflections indicated that a reflection could be considered observed if $I > 1.5\sigma(I)$. Based on this criterion, 1417 reflections were considered observed and were used in the analysis of the structure after correction for the usual Lorentz and polarization factors.

Structure Determination. An initial, unsuccessful attempt was made to solve the structure by using the coordinates of the phosphorus atom, determined from a Patterson function, to generate a three-dimensional electron density map computed with the phosphorus phases. The structure was solved by the tangent formula[6] by a refinement utilizing the phosphorus phases for the 194 strongest reflections with $E > 1.5$. The tangent formula was used to determine new phases as well as to further refine the phases already determined. This procedure afforded all of the 23 nonhydrogen atoms associated with the nucleotide analog.

Structure Refinement. The coordinates of the 23 atoms obtained from the *E* map were subjected to two cycles of isotropic refinement using the full matrix least-squares program[7] modified for use on the Univac 1108 computer.[8] A difference electron density map calculated at this point indicated the position of one of the nonhydrogen atoms of the solvating ethanol molecule. An additional two cycles of isotropic refinement on 24 atoms reduced the agreement index $R = \Sigma||F_o| - |F_c||/\Sigma|F_o|$, where F_o and F_c are observed and calculated structure factors, respectively) to 0.144 and indicated the presence of the remaining two nonhydrogen atoms of ethanol. One cycle of isotropic refinement on these three atoms dropped the *R* value to 0.108. This was followed by one cycle of anisotropic refinement on all of the 26 nonhydrogen atoms, *R* = 0.070, and then two additional cycles which reduced the *R* value to 0.058. A difference electron density map calculated at this point indicated the position of all 22 hydrogen atoms in the structure. These 22 atoms were subjected to two cycles of isotropic refinement which lowered the *R* value to 0.045. A final round of anisotropic refinement on the nonhydrogen atoms, with the hydrogen atoms fixed, resulted in a *R* value of 0.044 for the observed 1417 reflections. At this point the analysis was terminated. The *R* value for all reflections, both observed and unobserved (total: 1769 reflections), is 0.05. The ratios of the shifts to estimated standard deviations of the parameters were in the range 0.00 to 0.97 with an average value of 0.24.

The weighting scheme employed was essentially that of Hughes.[9] The scattering values for C, N, P, and O were those of Cromer and Waber,[10] while the values for H were taken from Stewart, *et al.*[11]

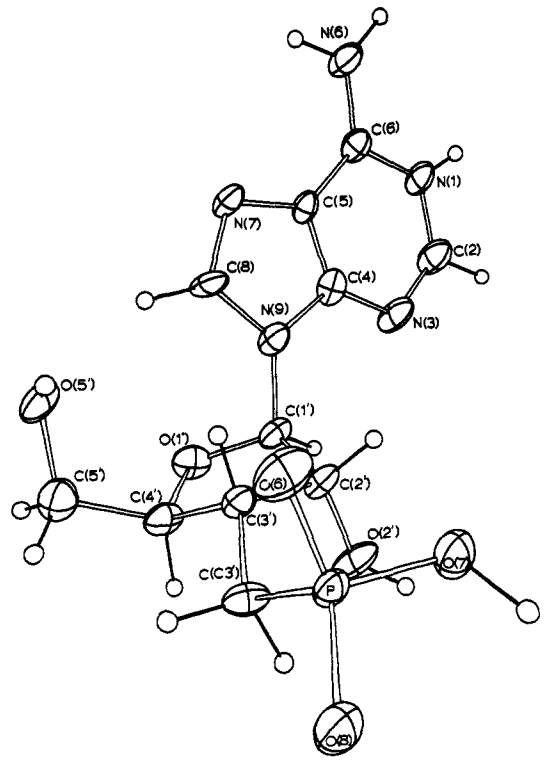


Figure 1. The thermal ellipsoids and atom numbering.

Results

The atomic parameters are listed in Table I. The observed and calculated structure factors have been deposited with the NAPS-ASIS agency.[12] The thermal

(4) M. Sundaralingam and J. Abola, *J. Amer. Chem. Soc.*, in press; see also M. Sundaralingam and J. Abola, *Nature (London), New Biol.*, **235**, 244 (1972).
(5) J. G. Moffatt, private communication.
(6) J. Karle and H. Hauptman, *Acta Crystallogr.*, **9**, 635 (1956).
(7) W. R. Busing, K. A. Martin, and H. A. Levy, Oak Ridge National Laboratory, Report ORNL-TM-305, Oak Ridge, Tenn., 1962.
(8) S. T. Rao, unpublished results.
(9) E. Hughes, *J. Amer. Chem. Soc.*, **63**, 1737 (1941).
(10) D. T. Cromer and J. T. Waber, *Acta Crystallogr.*, **18**, 104 (1965).
(11) R. F. Stewart, E. R. Davidson, and W. T. Simpson, *J. Chem. Phys.*, **42**, 3175 (1965).
(12) This table has been deposited as document No. NAPS-01813 with the ASIS National Auxiliary Publication Service, c/o CCM Informational Corp., 909 Third Ave., New York, N. Y. 10022. A copy may be secured by citing the document number and by remitting $1.00 for microfilms or $3.00 for photocopies. Advance payment is required. Make checks or money orders payable to: ASIS-NAPS. The structure factor table will also appear following these pages in the microfilm edition of this volume of the journal. Single copies may be obtained from the Business Operations Office, Books and Journals Division, American Chemical Society, 1155 Sixteenth St., N.W., Washington, D. C. 20036, by referring to code number JACS 72-4314. Remit check or money order for $3.00 for photocopy or $2.00 for microfiche.

Table I. Positional and Thermal Parameters of the Atoms[a]

Atom	$X \times 10^4$	$Y \times 10^4$	$Z \times 10^4$	$\beta_{11} \times 10^4$	$\beta_{22} \times 10^4$	$\beta_{33} \times 10^4$	$\beta_{12} \times 10^4$	$\beta_{13} \times 10^4$	$\beta_{23} \times 10^4$
N(1)	4871 (10)	−2422 (2)	2127 (3)	248 (21)	12 (1)	15 (2)	−7 (4)	2 (6)	4 (1)
C(2)	6335 (14)	−2484 (3)	2859 (4)	339 (30)	15 (1)	19 (2)	13 (6)	−5 (8)	3 (1)
N(3)	8118 (11)	−2895 (2)	2926 (3)	348 (24)	14 (1)	16 (2)	−4 (5)	−14 (6)	6 (1)
C(4)	8396 (12)	−3260 (2)	2178 (3)	230 (22)	6 (1)	21 (2)	3 (4)	14 (7)	−0 (1)
C(5)	7148 (11)	−3226 (2)	1416 (3)	202 (20)	7 (1)	13 (2)	−4 (4)	9 (6)	−0 (1)
C(6)	5183 (11)	−2790 (2)	1379 (3)	221 (22)	9 (1)	16 (2)	−2 (4)	−0 (7)	0 (1)
N(6)	3651 (10)	−2711 (2)	716 (3)	256 (20)	16 (1)	16 (2)	−6 (4)	−13 (6)	3 (1)
N(7)	8011 (10)	−3674 (2)	816 (2)	242 (19)	11 (1)	11 (1)	−3 (4)	−3 (5)	−0 (1)
C(8)	9798 (12)	−3976 (2)	1229 (3)	232 (24)	15 (1)	15 (2)	−6 (5)	−24 (6)	−4 (1)
N(9)	10075 (9)	−3750 (2)	2076 (3)	239 (19)	8 (1)	16 (1)	−6 (4)	−4 (5)	1 (1)
C(1′)	11856 (11)	−3974 (2)	2722 (3)	189 (21)	14 (1)	14 (2)	8 (5)	−3 (6)	−3 (1)
C(2′)	11023 (11)	−4538 (2)	3314 (3)	239 (23)	11 (1)	10 (2)	4 (5)	−2 (6)	−0 (1)
O(2′)	12278 (10)	−4458 (2)	4109 (2)	396 (21)	18 (1)	14 (1)	11 (4)	−31 (6)	0 (1)
C(3′)	11815 (11)	−5136 (2)	2789 (3)	161 (20)	12 (1)	14 (2)	−0 (4)	1 (6)	−3 (1)
C(C3′)	12152 (9)	−5781 (2)	3299 (2)	202 (15)	11 (1)	21 (1)	−0 (3)	−12 (5)	−6 (1)
P	9530 (16)	−6247 (3)	3569 (4)	145 (36)	8 (2)	12 (3)	−0 (8)	5 (10)	−2 (2)
O(6)	8114 (8)	−6378 (1)	2744 (2)	261 (15)	15 (1)	22 (1)	14 (3)	−3 (5)	−6 (1)
O(7)	8044 (6)	−5779 (1)	4189 (2)	273 (12)	15 (1)	25 (1)	−20 (3)	29 (4)	−6 (1)
O(8)	10244 (8)	−6856 (1)	4062 (2)	217 (15)	13 (1)	18 (1)	−1 (3)	6 (4)	−3 (1)
C(4′)	14191 (11)	−4901 (3)	2407 (3)	146 (19)	16 (1)	20 (2)	10 (4)	2 (6)	−8 (1)
C(5′)	15012 (12)	−5212 (3)	1555 (4)	214 (25)	21 (1)	24 (2)	−2 (5)	7 (7)	0 (1)
O(5′)	13185 (9)	−5211 (2)	887 (2)	323 (19)	21 (1)	11 (1)	−14 (4)	9 (5)	−3 (1)
O(1′)	13863 (7)	−4198 (2)	2265 (2)	192 (15)	11 (1)	27 (2)	10 (3)	−1 (5)	−6 (1)
C(9)	12337 (18)	−1395 (4)	4928 (5)	604 (40)	54 (3)	60 (4)	30 (10)	−54 (12)	−12 (3)
C(10)	10258 (16)	−1697 (3)	4503 (4)	569 (36)	34 (2)	48 (3)	−1 (8)	−23 (10)	−8 (2)
O(9)	9352 (8)	−1318 (1)	3806 (2)	419 (16)	29 (1)	27 (1)	−40 (4)	−10 (5)	−1 (1)

Atom	$X \times 10^3$	$Y \times 10^3$	$Z \times 10^3$	B, Å^2
H(1)	367 (17)	−229 (3)	225 (5)[b]	10.68
H(2)	602 (17)	−220 (4)	344 (5)	4.25
H1(6)	223 (19)	−235 (4)	67 (6)	11.25
H2(6)	410 (13)	−285 (3)	19 (4)	4.61
H(8)	1060 (19)	−446 (4)	92 (6)	9.29
H(1′)	1218 (13)	−351 (3)	319 (4)	4.59
H(2′)	913 (14)	−455 (3)	338 (4)	3.88
H(02′)	1157 (16)	−460 (4)	458 (5)	8.53
H(3′)	1077 (11)	−516 (2)	226 (3)	3.57
H1(C3′)	1315 (13)	−608 (3)	291 (4)	3.77
H2(C3′)	1302 (15)	−568 (3)	392 (4)	5.73
H(07)	757 (18)	−598 (4)	477 (5)	9.35
H(4′)	1545 (19)	−493 (4)	292 (5)	8.50
H1(5′)	1535 (13)	−566 (3)	174 (4)	5.28
H2(5′)	1656 (14)	−500 (3)	132 (4)	5.19
H(05′)	1197 (20)	−553 (4)	91 (6)	14.99
H1(9)	1364 (17)	−156 (4)	444 (5)	11.11
H2(9)	1138 (23)	−83 (5)	511 (6)	12.74
H3(9)	1303 (20)	−166 (4)	553 (6)	14.02
H1(10)	889 (18)	189 (4)	505 (5)	8.59
H2(10)	1078 (19)	−225 (4)	460 (5)	7.49
H(09)	1058 (22)	−142 (5)	338 (6)	13.43

[a] Standard deviations are given in parentheses and refer to the least significant digit. [b] The anisotropic temperature factors are of the form $\exp[-(\beta_{11}h^2 + \ldots + 2\beta_{12}hk + \ldots)]$.

ellipsoids of the atoms projected on the plane containing the purine base are represented by the drawing in Figure 1. The bond lengths and bond angles involving the nonhydrogen atoms are shown in Figure 2. The average estimated standard deviations in the bond distances and bond angles are 0.008 Å and 0.4°, respectively. The bond distances and bond angles are generally in agreement with those found in **2**.[3] In Table II are listed the average values and the range of values of the bond distances and bond angles involving the hydrogen atoms.

Table II. Bond Lengths and Angles Involving Hydrogen Atoms[a]

Bonds	Range, Å	Mean, Å	Esd, Å
C–H bonds (15)	0.99–1.29	1.11	0.08
N–H bonds (3)	0.74–1.07	0.90	0.08
O–H bonds (4)	0.88–1.03	0.96	0.09

Angles	Range, deg	Mean, deg	Esd, deg
C–C–H angle (tetrahedral) (17)	96–116	107	3
O–C–H angle (tetrahedral) (7)	105–135	116	3
C–O–H angle (tetrahedral) (3)	97–118	110	4
N–C–H angle (5)	105–127	117	3
C–N–H angle (4)	111–127	119	4
H–C–H angle (tetrahedral) (6)	76–135	107	4
H–N–H angle (trigonal) (1)		112	4
P–O–H angle (1)		114	3
P–C–H angle (2)		105	3

[a] The number of bonds or angles in each group is indicated in parentheses in column 1.

Discussion

Bond Distances and Bond Angles. The N(1) position of the base in **1** is protonated by a phosphonate proton as in **2**; thus the nucleotide is a zwitterion. The bond distances and bond angles of the common parts

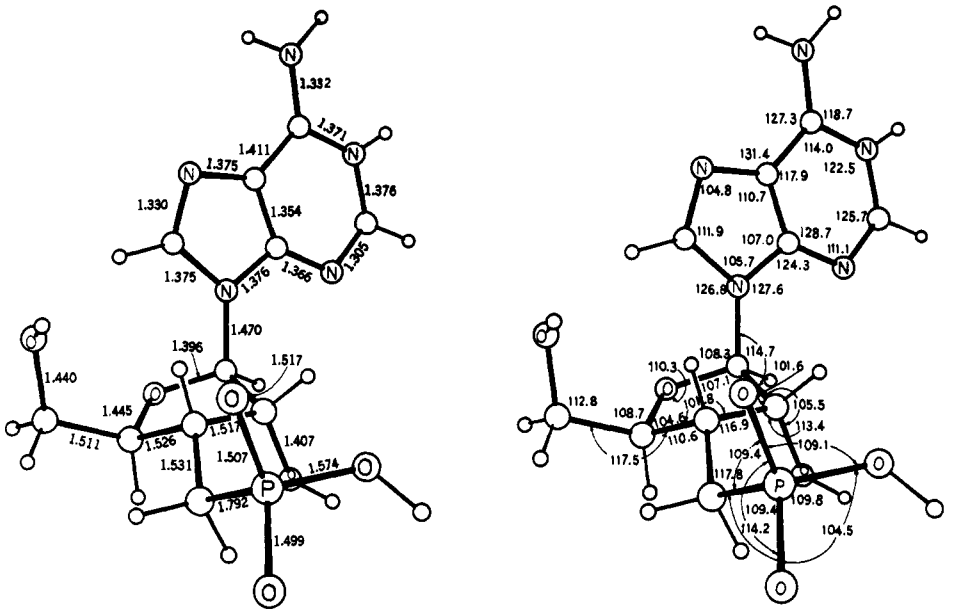


Figure 2. Bond distances and bond angles.

of the analog compare well with those of **2**. The greatest differences, as expected, occur in the phosphonate group. The C(3′)–C(C3′), and P–C(C3′) bond distances are 1.531 Å and 1.792 Å, respectively, which may be compared with the corresponding values in **2**, C(3′)–O(3′), 1.440 Å, and P–O(3′), 1.612 Å. The latter difference is evident in spite of the fact that the ribose groups in both molecules are C(3′)-endo. The resulting difference in the distance of the phosphorus atom from the rest of the molecule is at least partially compensated by the decreased bond angle C(3′)–C(C3′)–P in **1** (117.8°) compared with the angle of C(3′)–O(3′)–P in **2** (119.1°). The other large differences in the bond angles also involve the C(C3′) atom, *viz.* **1** (**2**) C(C3′)–P–O(6), 109.4° (103.4°); C(2′)–C(3′)–C(C3′), 116.9° (112.4°). In addition the O(6)–P–O(7) and O(6)–P–O–(8) angles are about 3° smaller in **1** than in **2**.

The C–C and C–O bond distances in the ethanol of solvation are considerably shorter than the normal values for such bonds. This is probably attributable to the thermal vibration executed by the ethanol. The bond lengths and angles involving the hydrogen atoms (Table II) are within the usual range.

Planarity of the Base. The least-squares plane through the nine ring atoms (plane I) was calculated, as were the planes through the atoms comprising the pyrimidine (plane II) and imidazole (plane III) rings. The displacements of the individual atoms from these planes are given in Table III, where the atoms were fitted to the planes with the same weights being given to each atom. The substituent atoms C(1′) and N(6) show significant displacements from plane I. The dihedral angle between the planes of the pyrimidine and imidazole rings is 2.3°. Similar values have been found in other purine derivatives.

Ribose Puckering. The least-squares planes which illustrate the puckering of the ribose moiety and the displacement of each of the atoms from these planes are presented in Table IV. The best four-atom plane is plane IV in which C(3′) is displaced endo, while C(2′) is displaced exo with respect to plane V. The displacements are with reference to the displacement of C(5′). Hence the sugar is in the preferred twist

Table III. Least-Squares Planes for the Base and Deviation of the Atoms from the Planes[a,b]

Atoms	Plane I, Å	Plane II, Å	Plane III, Å
N(1)	*−0.021*	*−0.005*	0.024
C(2)	*0.015*	*0.013*	0.096
N(3)	*0.019*	*0.002*	0.102
C(4)	*0.005*	*−0.016*	0.049
C(5)	*0.028*	*0.022*	*0.034*
C(6)	*−0.026*	*−0.011*	*−0.020*
N(6)	−0.094	−0.062	−0.120
N(7)	*0.023*	0.010	−0.002
C(8)	*−0.001*	−0.034	*−0.007*
N(9)	*−0.042*	−0.081	*−0.006*
C(1′)	−0.083	−0.143	−0.016
σ_{RMS}	0.023	0.013	0.018

[a] Atoms used in fitting the least-squares planes are shown in italics. [b] Equations of the planes: plane I, $0.626X + 0.695Y - 0.355Z = -2.836$; plane II, $0.614X + 0.702Y - 0.361Z = -2.941$; plane III, $0.632X + 0.703Y - 0.326Z = -2.814$.

Table IV. Least-Squares Planes for the Sugar Moiety[a,b]

Atoms	Plane IV	Plane V	Plane VI
O(1′)	*−0.050*	*0.085*	*0.000*
C(1′)	*0.048*	*−0.054*	*0.000*
C(2′)	*−0.028*	−0.551	−0.230
C(3′)	0.571	0.047	0.387
C(4′)	*0.030*	*−0.079*	*0.000*
C(5′)	0.837	0.909	0.892
σ_{RMS}	0.040	0.068	0.000

[a] Atoms used in fitting the planes are shown in italics. [b] Equations of the planes: plane IV, $-0.638X - 0.248Y - 0.730Z = -5.298$; plane V, $-0.456X - 0.077Y - 0.887Z = -6.015$; plane VI, $-0.571X - 0.194Y - 0.798Z = -5.527$.

form and the puckering with respect to the three-atom plane C(1′)–O(1′)–C(4′) is C(3′)-endo–C(2′)-exo or $^3T_2$.[13] In the case of **2** the sugar puckering with respect

(13) M. Sundaralingam, *J. Amer. Chem. Soc.*, in press.

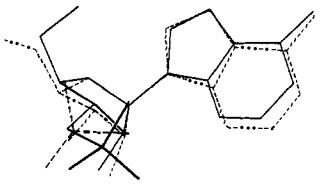


Figure 3. An overlay of the isosteric phosphonate analog of 3′-AMP (solid line) and 3′-AMP itself (broken line), the common points being O(1′), C(1′) and N(9).

to the best four-atom plane is also C(3′)-endo, but the puckering is C(4′)-exo with respect to the next best four-atom plane. Therefore, the ribofuranose ring in **2** is puckered, C(3′)-endo–C(4′)-exo, $^{3}T_{4}$, with respect to the three-atom plane O(1′)–C(1′)–C(2′).[14]

Table V. A Comparison of the Conformations of **1** and **2**

	1, deg	**2**, deg
C(4′)-O(1′)-C(1′)-C(2′)	9.0	−3.4
O(1′)-C(1′)-C(2′)-C(3′)	−29.5	−20.1
C(1′)-C(2′)-C(3′)-C(4′)	37.1	34.6
C(2′)-C(3′)-C(4′)-O(1′)	−32.9	−37.6
C(3′)-C(4′)-O(1′)-C(1′)	15.2	25.6
χ	28.1	3.8
O(1′)-C(4′)-C(5′)-O(5′)	−68.5	57
C(3′)-C(4′)-C(5′)-O(5′)	49.8	171
C(4′)-C(3′)-C(C3′)-P	−164.9	−123.4
C(2′)-C(3′)-C(C3′)-P	79.1	121.3
C(3′)-C(C3′)-P-O(6)	54.1	51.3
C(3′)-C(C3′)-P-O(7)	−62.6	−64.2
C(3′)-C(C3′)-P-O(8)	179.9	177.7

The exact difference in the puckering of the sugars in **1** and **2** can also be seen when the torsion angles about the furanose ring bonds are considered (Table V).

Conformation of 1. The glycosyl torsion angle[15] χ is 28.1°; the conformation, therefore, is anti.[15,16] This value may be compared with the χ value of 3.8° in the anti conformation of **2**. The hydroxymethyl group of the ribose is in the preferred gauche–gauche conformation, the torsion angles O(1′)–C(4′)–C(5′)–O(5′) and C(3′)–C(4′)–C(5′)–O(5′) being −68.5° and 49.8°, respectively. In contrast, in **2** the conformation was gauche–trans, the corresponding torsion angles being 57° and 171°.

The stereochemistry of the phosphonate with respect to the ribose is given by the torsion angles C(4′)–C(3′)–C(C3′)–P and C(2′)–C(3′)–C(C3′)–P, and the three torsion angles about the bond C(C3′)–P. These angles are compared for **1** and **2** in Table VI.

The differences in the conformations of **1** and **2** are shown by the composite drawing, Figure 3. The conformational differences about the individual bonds are listed in Table VI. The largest differences in the torsion angles occur about the bonds C(4′)–C(5′) and C(3′)–C(C3′) (C(3′)–O(3′)).

Molecular Packing and Hydrogen Bonding. A packing diagram viewed from the *a* axis is presented in Figure 4. The hydrogen bond distances and angles are given in Table VI. The six hydrogen atoms that are involved in hydrogen bonding are H1(6), H(1), H(O5′), H(O7), H(O2′), and H(O9), the latter belonging to the ethanol molecule of solvation. The nucleotide itself is involved in 12 hydrogen bonds, while the ethanol is involved in two. Perhaps the most interesting aspect of the hydrogen bonding scheme is the pairing of the phosphonate and the base *via* the hydrogen bonds $^{-}$O(6)···H–N^{+}(1) and O(8)···H–N(6). A similar phosphate–base pairing scheme involving the Watson–Crick base pairing sites in the adenine base was found in **2**.[3] Since the phosphonate hydrogen has protonated the base site N(1), both the phos-

Table VI. Hydrogen Bond Lengths and Angles

Sym no.	Translation X	Translation Y	Translation Z	Atoms	Angle, deg	Length, Å	Length from hydrogen, Å
3	−1	−1	0	O(8)···H1(6)–N(6)	161.8	2.806 (7)	1.76 (10)
				P–O(8)···N(6)	111.3		
				C(6)–N(6)···O(8)	118.9		
3	−1	−1	0	N(1)–H(1)···O(6)	137.0	2.705 (6)	2.12 (8)
				P–O(6)···N(1)	113.7		
				C(2)–N(1)···O(6)	112.4		
				C(6)–N(1)···O(6)	124.4		
3	0	0	0	O(9)–H(O9)···O(6)	151.0	2.752 (6)	1.87 (11)
				C(10)–O(9)···O(6)	115.9		
				P–O(6)···O(9)	115.8		
3	0	0	0	O(5′)–H(05′)···O(9)	157.4	2.698 (6)	1.80 (9)
				C(10)–O(9)···O(5′)	121.1		
				C(5′)–O(5′)···O(9)	104.5		
4	−1	−1	0	O(7)–H(O7)···N(7)	175.4	2.773 (5)	1.75 (8)
				P–O(7)···N(7)	113.9		
				C(5)–N(7)···O(7)	141.4		
				C(8)–N(7)···O(7)	113.3		
4	0	−1	0	O(2′)–H(O2′)···O(5′)	147.4	2.796 (6)	2.01 (8)
				C(2′)–O(2′)···O(5′)	139.2		
				C(5′)–O(5′)···O(2′)	138.3		

(14) It should be noted that in earlier work the three-atom plane was erroneously chosen to be C(1′)–O(1′)–C(4′) and consequently the puckering from **2** was called $^{3}T^{2}$. However, a more proper definition of the puckering of the furanose ring should take into consideration the pseudorotation executed by such rings (see K. S. Pitzer and W. E. Donath, *J. Amer. Chem. Soc.*, **81**, 3213 (1959). Therefore, the three-atom plane is selected on the basis that these atoms show the least deviations from the least-squares plane of the five ring atoms. (See M. Sundaralingam, S. T. Rao, and J. Abola, *Science*, **172**, 725 (1971).) In **2** the three-atom plane turns out to be O(1′)-C(1′)-C(2′), and the puckering is $^{3}T_{4}$. A description of the furanoside ring conformations using the concept of pseudorotation has been presented by C. Altona and M. Sundaralingam, *J. Amer. Chem. Soc.*, submitted for publication.

(15) M. Sundaralingam, *Biopolymers*, **7**, 821 (1969).

(16) J. Donohue and K. N. Trueblood, *J. Mol. Biol.*, **2**, 363 (1960).

phonate oxygens, O(6) and O(8), act as acceptors in the hydrogen bonding. O(6) of the phosphonate is simultaneously involved in an acceptor hydrogen bond involving the solvent. The second hydrogen atom of the amino group and site N(7) are engaged in a base–phosphonate pairing scheme to a symmetry related phosphonate group. In this pairing, therefore, the Hoogsteen sites in the base, N(6) and N(7), are involved. The ribose hydroxyl O(5′)H donates a hydrogen bond to the solvent and accepts a hydrogen bond from the O(2′)H group of another ribose. Thus the hydrogen bonding involving the following five nonhydrogen atoms forms a closed loop, –O(2′)–H → O(5′)–H → O(9)–H → O(6) ← H–N(1)– (Figure 3).

The only potential hydrogen-bonding site on the base that is not involved in hydrogen bonding is N(3). As seen here and in other structures, position N(7) in the adenine and guanine bases shows a strong tendency for hydrogen bonding. Therefore, in double-helical nucleic acids, although this site is not engaged in the Watson–Crick base pairing scheme, it probably is involved in hydrogen bonding to solvent molecules. To a lesser extent this site shows metal coordination properties, and may be bonded to monovalent metal ions such as Na^+ or divalent metal ions such as Zn^{2+} (for a review, see ref 17).

The hydrogen bonding in this structure varies from approximately linear to markedly nonlinear, the angles being in the range 137.0–175.4° (Table VI).

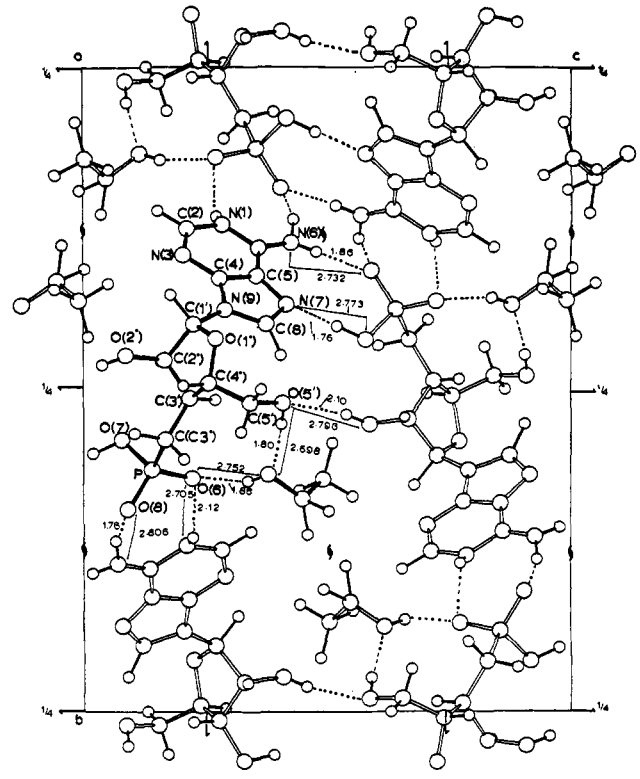


Figure 4. Hydrogen bondings cheme projected down the *a* axis and the hydrogen bond distances. The molecule in solid bonds depicts the reference molecule.

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(17) M. Sundaralingam and J. A. Carrabine, *J. Mol. Biol.*, **61**, 287 (1971).

Spectroscopic Characterization of Poly(Ala-Gly-Gly)

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Abstract: Poly(Ala-Gly-Gly) was synthesized in our laboratory as part of a study concerning the role of tripeptide sequences, omitting proline, in the structure of collagen. Three forms of the polymer, two ordered and one disordered, have been isolated depending on solvent and temperature. These have been characterized using ultraviolet absorption and circular dichroism spectroscopy in conjunction with infrared-absorption and linear dichroism spectroscopy. Form I, with a cross β conformation, was isolated from dichloroacetic acid or acetic acid. Form II, with a 3_1-helical conformation, was found in dilute aqueous solution or could be film cast from this solvent, whereas form III, a disordered form, is found in hot aqueous or 6 *M* calcium chloride solutions, as well as in films cast from trifluoroacetic acid or hot water. This polytripeptide is therefore apparently the first which is known to have a stable 3_1 helix in aqueous solution and which does not contain proline. Furthermore the thermal melting is the first observation of a polyglycine II helix–random coil transition.

Spectroscopic characterization of synthetic polyamino acids has yielded much available information which has subsequently been applied to conformational aspects of both fibrous and globular proteins. More recently sequential polypeptides have been synthesized and studied as models for fibrous proteins such as silk and collagen. Whereas much of the early work concentrated on the β sheet and α helix (usually right handed) and transitions of the latter (helix–coil), much of the recent work has concentrated on the left-hand 3_1 helix of poly-L-proline and its derivatives and the distorted supercoiled 3_1 helix found in proline-containing