methyl-^{14}C were radiochemically pure as evidenced by chromatography in systems C and D. The samples were mixed with known amounts of synthetic carrier indolmycenic acids and subjected to Kuhn–Roth oxidation.[16] The resulting acetic acid samples were further degraded by the Schmidt procedure.[11,17]

Degradation of Indolmycenic Acid to Indoleisopropionic Acid. Radiochemically pure indolmycenic acid-*3*-^{14}C (1.1×10^6 dpm), obtained by hydrolysis of indolmycin, and 5 mg of racemic synthetic indolmycenic acid were dissolved in 5 ml of dry ether. $LiAlH_4$ (10 mg) was added with stirring and the reaction was allowed to proceed for 15 min at room temperature. Excess $LiAlH_4$ was destroyed by the addition of 0.5 ml of water and the solution was acidified with 2 *N* H_2SO_4. The aqueous phase was extracted three times with ether, and any residual indolmycenic acid was removed from the ether solution by washing with a 10% Na_2CO_3 solution. The ether phase was dried over Na_2SO_4 and evaporated; yield 5.0×10^5 dpm of 3-(β-indolyl)butane-1,2-diol-*3*-^{14}C, 85% radiochemically pure as judged by tlc in system H. The diol dissolved in 1 ml of methanol was added to a stirred mixture of 0.5 ml of 0.1 *N* aqueous sodium periodate solution, 2 ml of petroleum ether, and 2 ml of ether at about 14° under nitrogen. After 12 min, the phases were separated and the aqueous layer was extracted twice with 5 ml of ether. The ether extract was dried over Na_2SO_4 and evaporated to give 3.88×10^5 dpm of 2-(β-indolyl)-propionaldehyde-*2*-^{14}C of 90% radiochemical purity (system H). Freshly prepared Ag_2O (20 μmol) and 1 ml of water were added to the aldehyde (3.5×10^5 dpm). The reaction mixture was allowed to stand at room temperature for 15 min with occasional shaking. After acidification to pH 3 with 1 *N* HCl the solution was extracted three times with ether and the ether phase was extracted with 10% Na_2CO_3 solution. The soda solution was acidified with tartaric acid and reextracted with ether. This ether extract was dried and evaporated to give 1.25×10^5 dpm of indoleisopropionic acid, which was shown by esterification with diazomethane and chromatography in system I to be 81% radiochemically pure. This material (3.69×10^4 dpm indoleisopropionic acid) was recrystallized repeatedly from isopropyl alcohol with 150 mg of (*R*)-indoleisopropionic acid cinchonine salt, and after each crystallization about 10 mg was used to measure the specific radioactivity of the salt. The recovery in each crystallization was about 60%.

Acknowledgments. We are indebted to Chas. Pfizer and Co. for providing us with an indolmycin-producing strain of *S. griseus* and a sample of indolmycin, to Dr. H. R. Snyder, Urbana, for a gift of the two isomers of 3-methyltryptophan, and to Dr. R. K. Hill, Athens, Georgia, for providing us with a copy of his manuscript prior to publication. H. G. Floss is the recipient of U. S. Public Health Service Research Career Development Award No. GM 42-389. Marilyn K. Speedie is the recipient of a Mead Johnson Undergraduate Research Award and a National Science Foundation Undergraduate Summer Research Participant. This work was supported by National Institutes of Health Research Grant No. NB 07646 and by grants from Eli Lilly and Company and the Purdue Research Foundation.

A Purine Nucleoside in Syn Conformation. Molecular and Crystal Structures of 5′-Methylammonium-5′-deoxyadenosine Iodide Monohydrate

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Abstract: 5′-Methylammonium-5′-deoxyadenosine (Figure 1), a poor substrate for adenosine desaminase, could be crystallized as its iodide monohydrate in space group $P2_1$. The structure was solved from three-dimensional X-ray data and refined to $R = 4.1\%$ for the 1415 significant data. The nucleoside exists in the unusual syn conformation, atom N-3 of the adenine residue being in intramolecular hydrogen bonding contact with the ammonium nitrogen N-5′. The conformation of the ribose is C-2′ endo, C-3′ exo. Atom N-5′ is in the trans,gauche position with respect to C-3′ and O-1′ and coplanar with the adenine heterocycle. The packing of the molecules is such that the heterocycles are not stacked but arranged in fishbone manner at 120° to each other. The iodide ions are located above and below the adenine heterocycles at only 3.82 Å distance. The water of hydration is fourfold disordered and forms a linear hydrophilic region within the crystal structure.

The deduction of the reaction mechanism of an enzyme is possible only if the structural properties of the substrates which are accepted by the enzyme are known. When 5′-methylammonium-5′-deoxyadenosine (Figure 1) was subjected to the reaction with the enzyme adenosine desaminase, it was observed that the conversion of this substrate to the corresponding inosine derivative was about two orders of magnitude slower than with adenosine itself.[1] Since 5′-methylammonium-5′-deoxyadenosine could be crystallized as its iodide, the protonated species being the same as the one present in the enzymatic assay, it became worthwhile to investigate its structural properties.

Experimental Section

5′-Methylammonium-5′-deoxyadenosine[2] was crystallized as its iodide from aqueous methanol at 4° by Dr. H. Hettler. One of the colorless rhomb-shaped crystals of approximate dimensions $0.1 \times 0.15 \times 0.1$ mm was used for all subsequent X-ray crystallographic investigations. The crystal lattice parameters (Table I) were derived from precession photographs and diffractometer measurements. The density of the crystals has been determined by the flotation technique and is in agreement with the calculated density but one water molecule of hydration per asymmetric unit must be included.

The intensity data were collected by Dr. H. A. Paulus, Darmstadt, on a Stoe four-circle automatic diffractometer equipped with a graphite monochromator in vertical position and a Philips microfocus Mo X-ray tube. The 1597 data were gathered up to a glancing

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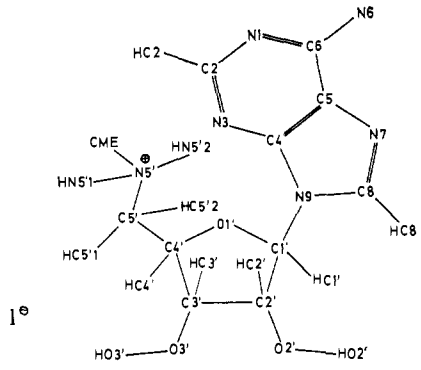

Figure 1. 5′-Methylammonium-5′-deoxyadenosine iodide.

angle $\theta = 25°$ in the $\omega,2\theta$ scan mode. The background was measured before and after each scan in the time ratio 1:2.5:1 for background, peak, background. The data were corrected for the usual Lorentz factor. In order to take into account the extra polarization caused by the monochromator the polarization factor used was[3] $p = [(1 + \cos^2 2\theta_M)\cos^2 2\theta]/(1 + \cos^2 2\theta_M)$. The glancing angle at the monochromator, θ_M, was 12°. In view of the low linear absorption coefficient and the small dimensions of the crystal an absorption correction was deemed unnecessary.

Table I. Crystal Lattice Parameters

Space group, $P2_1$ ($0k0$, $k = 2n + 1$ extinct)
$a = 8.740 \pm 0.002$ Å, $b = 13.288 \pm 0.004$ Å
$c = 8.476 \pm 0.002$ Å, $\beta = 118.30 \pm 0.04°$
Chem formula, $C_{11}H_{17}N_6O_3I$
Mol wt, 408.86
Density, obsd xylene–CH_3I, 1.716 g/cm³
calcd ($Z = 2$, mol wt = 408.86 + H_2O), 1.715 g/cm³
Linear absorption coefficient, $\mu = 20.8$ cm⁻¹

Since the background noise is rather low when monochromatized instead of filtered radiation is applied, the background counts often were unstatistical and unsymmetrical when high-order, weak intensities were measured. These unstatistical background counts were corrected using a plot of average background intensity *vs.* $\sin\theta$. From a Wilson[4] plot an absolute scale factor for the data and an overall isotropic temperature factor ($B = 3.97$ Å²) were evaluated.

Solution and Refinement of the Structure. The iodine atom was located from a sharpened Patterson section computed at $v = 1/2$. A Fourier synthesis calculated with the phases derived from the scattering of the iodine atom alone revealed the adenine heterocycle of the 5′-methylammonium-5′-deoxyadenosine molecule. Since a false inversion center was introduced into this computation, the ribose moiety was not clearly resolved but could be realized from a subsequent Fourier summation phased with the iodine atom and adenine heterocycle together. A further Fourier summation based on all so far located atoms revealed the molecule of water of hydration as a smeared-out region with four peaks of heights $1/3:1/3:1/3:1/3$ relative to the other oxygen atoms in this structure.

The refinement of the structure was accomplished by full-matrix least-squares methods[5] with a maximum of 132 parameters simultaneously varied.

A weighting scheme derived from counting statistics[6] was applied to the structure amplitudes. Amplitudes with $(F_o \leq 6)\sigma_{F_o}$ were treated as unobserved and were not included in the refinement of the structure.

The atomic scattering factors and anomalous dispersion correction for iodine ($\Delta f' = 0.6$, $\Delta f'' = 2.3$) were gathered from the International Tables for X-Ray Crystallography.[7] The 1415 significant data were used to refine the structure in three isotropic and four anisotropic cycles with minimization of the function $\Sigma w \cdot (|F_o| - |F_c|)^2$.

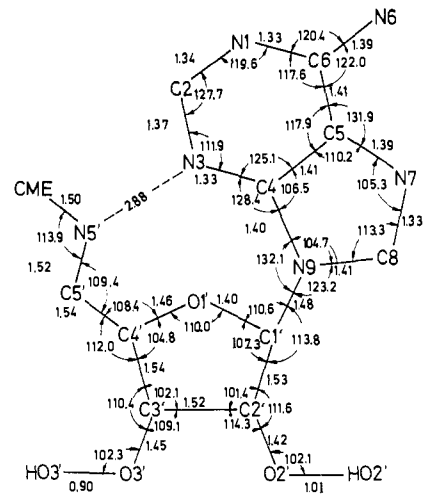

Figure 2. Bond angles and distances. The estimated standard deviations are about 0.8° and 0.01 Å, respectively.

The positions of the hydrogen atoms were picked from a difference Fourier synthesis computed after the third anisotropic refinement cycle. The occupational parameters of the four water sites and the positional and thermal parameters of all other nonhydrogen atoms were allowed to vary in the last cycle of refinement in which the average shifts were no more than one-third the estimated standard deviations. The final R factors, $\Sigma||F_o| - |F_c||/\Sigma|F_o|$, for the 1415 observed and all 1597 data are 4.1 and 5.3%, respectively.

Results

The final positional and thermal parameters of 5′-methylammonium-5′-deoxyadenosine iodide monohydrate and their standard deviations estimated from the least-squares normal equations are presented in Table II. In Figure 2 bond angles and distances are indicated. Their estimated standard deviations are about 0.01 Å for lengths and about 0.8° for angles, respectively. In Tables IV and V dihedral angles and least-squares planes through several parts of the molecule are listed, while in Figure 3 a stereographic plot for the thermal ellipsoids of the atoms is presented. The observed and calculated structure factors are collected in Table III. Projections of the structure along the c axis and on 010 are described in Figures 4 and 5.

Discussion

(a) Conformation of the Molecule. In a nucleoside the mutual rotation of sugar and heterocycle about the glycosidic C(1)′–N bond is not free but sterically hindered mainly by HC-2′.[8] The two preferred conformations are called anti or syn, respectively,[9] if in purine nucleosides, atom C-8 is pointing toward or away from the ribose moiety. A quantitative measure for this rotation is the dihedral angle C(2)′–C(1)′–N(9)–C(8) which is 0 to −180° in the case of anti and 0 to +180° in the case of syn conformation.[10] Most of the nucleosides investigated so far were found to exist in anti conformation in the crystalline state. However, 8-

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Table II. Atomic Positional and Thermal Parameters[a]

Atom	X	Y	Z	β_{11}	β_{22}	β_{33}	β_{12}	β_{13}	β_{23}
I	0.1869 (1)	0.2871 (0)	−0.1374 (1)	0.0164 (1)	0.0067 (0)	0.0140 (1)	−0.0016 (1)	0.0062 (1)	−0.0010 (1)
N-1	0.0957 (11)	0.0621 (7)	0.1973 (11)	0.0105 (13)	0.0047 (5)	0.0148 (16)	0.0017 (7)	0.0058 (12)	0.0021 (8)
C-2	0.0032 (17)	0.0434 (10)	0.0217 (12)	0.0136 (13)	0.0039 (5)	0.0096 (15)	0.0027 (7)	0.0036 (12)	0.0021 (7)
N-3	−0.1314 (10)	0.0987 (6)	−0.1043 (10)	0.0115 (13)	0.0032 (5)	0.0130 (15)	0.0007 (7)	0.0042 (12)	−0.0007 (7)
C-4	−0.1706 (10)	0.1765 (6)	−0.0322 (11)	0.0073 (13)	0.0017 (4)	0.0108 (15)	−0.0000 (6)	0.0028 (12)	0.0008 (7)
C-5	−0.0873 (11)	0.2006 (7)	0.1515 (11)	0.0091 (14)	0.0031 (5)	0.0097 (15)	−0.0011 (7)	0.0039 (12)	−0.0005 (7)
C-6	0.0499 (11)	0.1380 (7)	0.2690 (12)	0.0072 (13)	0.0044 (6)	0.0092 (14)	−0.0008 (8)	0.0020 (12)	0.0009 (8)
N-6	0.1402 (10)	0.1560 (7)	0.4526 (11)	0.0105 (13)	0.0049 (6)	0.0105 (14)	0.0003 (7)	0.0012 (12)	0.0027 (8)
N-7	−0.1637 (9)	0.2846 (11)	0.1816 (9)	0.0149 (13)	0.0050 (5)	0.0112 (12)	0.0015 (12)	0.0052 (11)	−0.0050 (12)
C-8	−0.2897 (12)	0.3111 (6)	0.0214 (12)	0.0134 (15)	0.0024 (7)	0.0134 (15)	−0.0001 (7)	0.0036 (13)	−0.0009 (7)
N-9	−0.2987 (9)	0.2499 (6)	−0.1181 (9)	0.0092 (12)	0.0039 (4)	0.0073 (12)	−0.0010 (6)	0.0036 (10)	−0.0014 (6)
C-1′	−0.4225 (10)	0.2679 (6)	−0.3092 (10)	0.0068 (12)	0.0020 (7)	0.0113 (13)	0.0003 (6)	0.0035 (11)	−0.0002 (6)
O-1′	−0.4650 (8)	0.1773 (5)	−0.4051 (8)	0.0082 (10)	0.0028 (4)	0.0111 (11)	−0.0016 (5)	0.0044 (9)	−0.0006 (5)
C-2′	−0.3539 (11)	0.3397 (7)	−0.4024 (12)	0.0079 (13)	0.0024 (5)	0.0124 (16)	0.0003 (7)	0.0032 (12)	0.0019 (8)
O-2′	−0.3820 (9)	0.4418 (5)	−0.3743 (9)	0.0103 (12)	0.0022 (4)	0.0158 (13)	0.0002 (5)	0.0010 (10)	0.0006 (6)
C-3′	−0.4554 (10)	0.3054 (6)	−0.5958 (10)	0.0124 (14)	0.0026 (7)	0.0110 (13)	0.0024 (8)	0.0066 (11)	0.0030 (8)
O-3′	−0.6293 (9)	0.3480 (6)	−0.6742 (9)	0.0135 (12)	0.0046 (4)	0.0106 (11)	0.0016 (6)	0.0018 (10)	−0.0005 (6)
C-4′	−0.4629 (12)	0.1910 (7)	−0.5751 (11)	0.0131 (16)	0.0026 (5)	0.0081 (14)	−0.0010 (7)	0.0063 (13)	0.0001 (7)
C-5′	−0.3010 (13)	0.1380 (7)	−0.5623 (14)	0.0168 (19)	0.0030 (5)	0.0159 (18)	0.0004 (8)	0.0107 (16)	0.0004 (9)
N-5′	−0.2798 (10)	0.0362 (6)	−0.4724 (10)	0.0092 (12)	0.0029 (4)	0.0121 (14)	0.0009 (6)	0.0050 (11)	0.0003 (7)
CME	−0.1352 (17)	−0.0263 (9)	−0.4683 (17)	0.0211 (25)	0.0035 (7)	0.0253 (30)	0.0027 (11)	0.0142 (23)	−0.0010 (11)
O-1	0.4338 (20)	0.0377 (12)	−0.1394 (22)	0.0199 (108)	0.0040 (31)	0.0334 (78)	0.0042 (46)	0.0186 (78)	0.0044 (41)
O-2	0.4068 (22)	0.0532 (13)	0.0159 (24)	0.0294 (62)	0.0043 (13)	0.0262 (58)	0.0015 (25)	0.0135 (48)	0.0003 (22)
O-3	0.4853 (20)	0.0313 (11)	−0.0754 (19)	0.0246 (102)	0.0060 (33)	0.0155 (88)	0.0035 (51)	0.0108 (91)	0.0084 (48)
O-4	0.5399 (22)	0.0137 (13)	0.0437 (21)	0.0260 (249)	0.0197 (128)	0.0193 (225)	−0.0143 (150)	0.0201 (207)	−0.0074 (141)
HC-2	0.0310	−0.0220	−0.0240	0.0115	0.0039	0.0123	0.0000	0.0056	−0.0000
HC-8	−0.3720	0.3670	0.0010	0.0127	0.0043	0.0136	−0.0000	0.0062	−0.0000
HC-1′	−0.5320	0.3000	−0.3200	0.0084	0.0028	0.0089	−0.0000	0.0041	−0.0000
HC-2′	−0.2250	0.3240	−0.3550	0.0090	0.0030	0.0095	−0.0000	0.0044	−0.0000
HC-3′	−0.3950	0.3200	−0.6700	0.0101	0.0034	0.0107	−0.0000	0.0049	−0.0000
HC-4′	−0.5650	0.1580	−0.6800	0.0078	0.0026	0.0083	−0.0000	0.0038	−0.0000
HC-5′1	−0.1940	0.1770	−0.4900	0.0114	0.0038	0.0121	−0.0000	0.0056	−0.0000
HC-5′2	−0.3130	0.1250	−0.6850	0.0114	0.0038	0.0121	−0.0000	0.0056	−0.0000
HN-5′1	−0.2560	0.0440	−0.3450	0.0111	0.0037	0.0118	0.0000	0.0054	0.0000
HN-5′2	−0.3900	−0.0050	−0.5400	0.0111	0.0037	0.0118	0.0000	0.0054	0.0000
HO-2′	−0.2620	0.4710	−0.3250	0.0117	0.0039	0.0125	0.0000	0.0057	0.0000
HO-3′	0.3300	0.3350	0.2100	0.0123	0.0041	0.0130	0.0000	0.0060	0.0000

[a] Expressed in fractions of the unit cell dimensions and in the form $T = \exp - (\beta_{11}h^2 + \beta_{22}k^2 + \beta_{33}l^2 + 2\beta_{12}hk + 2\beta_{13}hl + 2\beta_{23}kl)$. Estimated standard deviations are in parentheses. The occupancy factors for the disordered water atoms O-1, O-2, O-3, and O-4 are 0.3713, 0.3842, 0.2018, and 0.1095, respectively.

Table III. Geometrical Data for the Hydrogen Bonds

Atoms		Distances, Å			Angles, deg	
D	A	D···A	D–H	H···A	H–D···A	A···D–X
O-2′	N-1	2.74	1.01	1.81	17.2	117.6
N-5′	O-2′	2.89	1.00	1.90	10.3	107.8 (X = C-5′) 116.1 (X = CME)
N-5′	N-3	2.88	1.01	1.94	17.3	99.9 (X = C-5′) 101.1 (X = CME)
Angle A···D···A (O-2′···N-5′···N-3) = 116.9°						

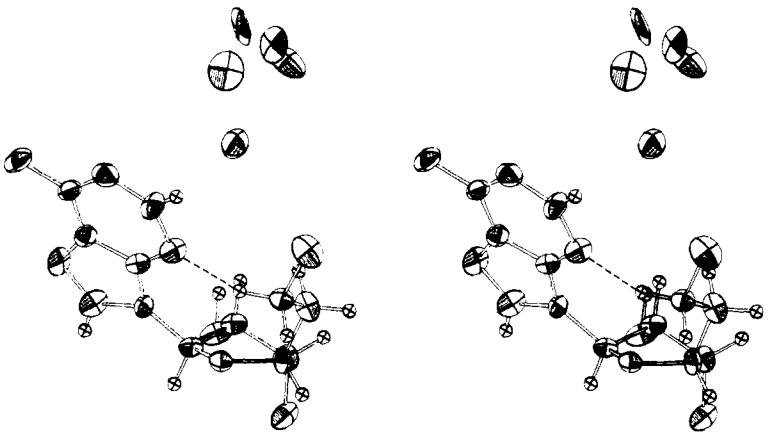

Figure 3. A stereoscopic representation of the structure viewed along the *b* axis plotted by ORTEP (C. K. Johnson, Oak Ridge Thermal Ellipsoid Plot Program, 1965; the plot was performed at Deutsches Rechenzentrum, Darmstadt; the probability of finding the atoms within the ellipsoids is 50%). The N(5)′–N(3) hydrogen bond is indicated by ----. The disordered water molecule is represented by four unconnected ellipsoids which belong to oxygen atoms O-3, O-4, O-2, and O-1 from left to right.

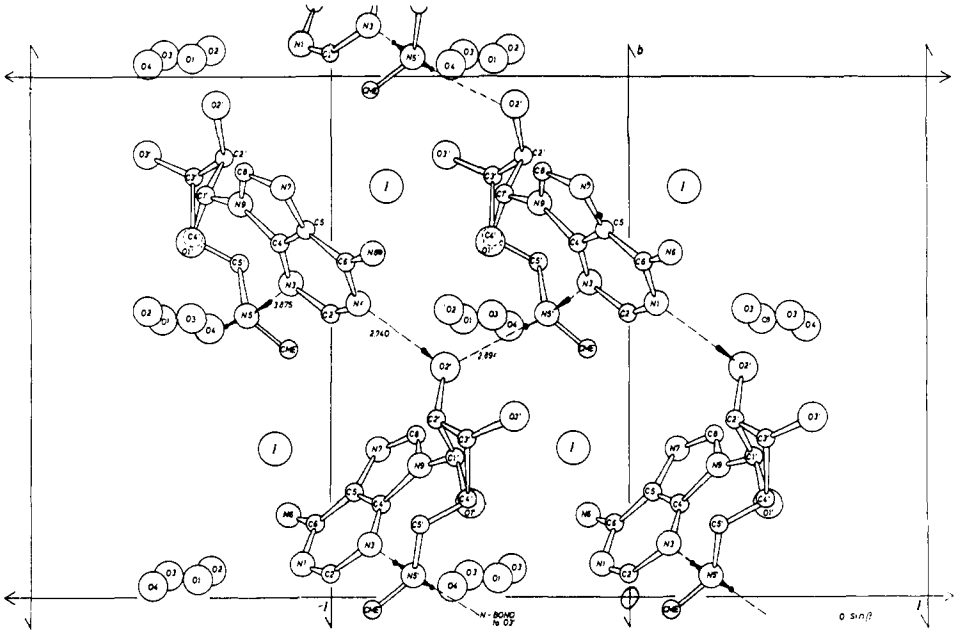

Figure 4. Projection of the structure along the *c* axis.

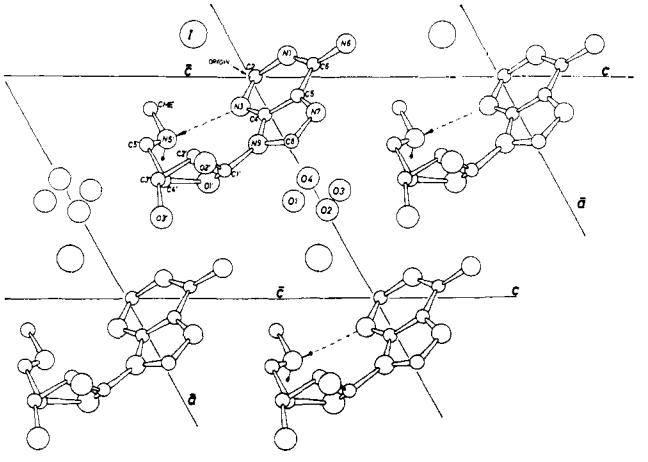

Figure 5. Projection of the structure on (010).

bromoadenosine,[11] 8-bromoguanosine,[11] adenosine 3′,5′-cyclophosphate,[12] deoxyguanosine,[13] 3′-*O*-acetyladenosine,[14] cytidine 2′,3′-*O*,*O*-cyclophosphate,[15] and 4-thiouridine,[10] were observed to occur in syn conformation.

Since in 5′-methylammonium-5′-deoxyadenosine, atom C-8 is pointing away from the ribose (Figure 3) and the dihedral angle C(2)′–C(1)′–N(9)–C(8) of 88.1° is in the range 0 to +180°, this nucleoside also exists in syn conformation, or, in the Klyne and Prelog[10] definition of conformational ranges, 5′-methylammonium-5′-deoxyadenosine exhibits the −syn-periplanar (−sp) conformation with $\tau_{CN-90°} = 1.9°$.

The cause for the occurrence of this nucleoside in the unusual syn conformation can be seen in the Coulomb attraction force between the positive charge

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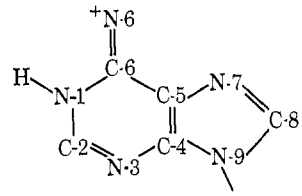
Table IV[a]

Angle	Deg
O(1)′-C(1)′-N(9)-C(4)	+29.2
C(2)′-C(1)′-N(9)-C(4)	−91.6
O(1)′-C(1)′-N(9)-C(8)	−151.2
C(2)′-C(1)′-N(9)-C(8)	+87.9
C(1)′-C(2)′-C(3)′-C(4)′	−36.9
C(2)′-C(3)′-C(4)′-O(1)′	+31.4
C(3) -C(4)′-O(1)′-C(1)′	−12.6
C(4)′-O(1)′-C(1)′-C(2)′	−11.5
O(1)′-C(1)′-C(2)′-C(3)′	+30.9
C(1)′-C(2)′-C(3)′-O(3)′	+79.7
O(2)′-C(2)′-C(3)′-C(4)′	−139.5
O(2)′-C(2)′-C(1)′-O(1)′	+152.9
C(2)′-C(3)′-C(4)′-C(5)′	−86.0
O(3)′-C(3)′-C(4)′-O(1)′	−84.4
O(3)′-C(3)′-C(4)′-C(5)′	+158.3
O(1)′-C(4)′-C(5)′-N(5)′	+43.0
C(3)′-C(4)′-C(5)′-N(5)′	+158.3
C(4)′-C(5)′-N(5)′-CME	+174.6
O(2)′-C(2)′-C(3)′-O(3)′	−40.5
N(9)-C(1)′-O(1)′-C(4)′	−136.2
N(9)-C(1)′-C(2)′-C(3)′	+153.6
N(9)-C(1)′-C(2)′-O(2)′	−84.4
C(5)′-C(4)′-O(1)′-C(1)′	+107.1

[a] Some dihedral angles, defined as zero when the two outer bonds are parallel and measured positive when the far bond is rotated clockwise.



is indicated in the data for the protonated form by the short C(6)–N(6) and long N(1)–C(6) and N(1)–C(2) distances, the bond distances are very similar in both cases with generally no more than twice the estimated standard deviation (0.02 Å) difference. However, the bond angles at N-1 and C-2 differ by about 5°, being 118.5 and 129.6° in the unprotonated and 123.0 and 125.6°, respectively, in the protonated adenine residue.

The geometry of the adenine heterocycle in the 5′-methylammonium-5′-deoxyadenosine cation is in overall agreement with the data for the unprotonated adenine (Figure 6B) except distances C(6)–N(6), C(8)–N(9), C(4)–C(5), and C(2)–N(3) which are longer in this structure than in the average adenine system.

Furthermore, the exocyclic angles at N-9, C(4)–N(9)–C(1)′ and C(8)–N(9)–C(1)′, are 132.1 and 123.2°, respectively, but the average angles in Figure 6B are

Table V. Some Least-Squares Planes within the 5′-Methylammonium-5′-deoxyadenosine Molecule[a]

	Coefficients for planes		Deviation of some atoms from planes[b]		
A. Adenine heterocycle	l = 0.804	N-1	0.031*	N-7	−0.011*
	m = 0.580	C-2	−0.025*	C-8	−0.018*
	n = −0.130	N-3	−0.015*	N-9	0.032
	d = −0.291	C-4	0.007*	C-1′	0.104
		C-5	0.006*	C-2′	1.532*
		C-6	−0.007*	I (x, y, z)	3.812
		N-6	−0.004	I (x, y − 1/2, $\bar{z}$)	−3.823
		N-5′		N-5′	0.007
Ribose atoms					
B. C-1′, C-3′, C-4′, O-1′	l = −0.916	C-1′	−0.045*	O-2′	−0.146
	m = 0.120	C-2′	−0.557	O-3′	1.435
	n = −0.382	C-3′	0.039*	N-9	−0.906
	d = −3.599	C-4′	−0.066*	C-5′	−1.437
		O-1′	0.071*		
C. C-1′, C-2′, C-4′, O-1′	l = −0.800	C-1′	0.061*	O-2′	0.659
	m = 0.373	C-2′	−0.035*	O-3′	2.009
	n = −0.469	C-3′	0.560	N-9	−0.948
	d = −4.311	C-4′	0.037*	C-5′	−1.361
		O-1′	−0.063*		
D. C-1′, O-1′, C-4′	l = −0.861	C-1′	0.0*	O-2′	0.267
	m = 0.252	C-2′	−0.291	O-3′	1.763
	n = −0.441	C-3′	0.324	N-9	−0.959
	d = −4.026	C-4′	0.0*	C-5′	−1.394
		O-1′	0.0*		

[a] The equations of the planes are of the form $lx + my + nz + d = 0$ and are based on an orthogonal system with x, y, z along a^*, b, c, respectively. [b] Atoms marked with an asterisk define the planes.

located at N-5′ and the lone electron pair at N-3. This is expressed by the short distance N(5)′–N(3) of only 2.88 Å, which together with the position of HN-5′1 relative to atoms N-5′ and N-3 (Table III) is indicative of a hydrogen bond between the two nitrogen atoms.

(b) Adenine Residue. In the literature, geometrical data for two different types of adenosine molecules have been reported: the unprotonated and protonated species, protonation taking place at N-1. These data are summarized in Figure 6. Although a major contribution of the structure

125.8 and 127.5°, respectively. Since the angles in 6B were derived from adenosine derivatives in anti conformation or 9-alkyladenine, the different distribution of the exocyclic angles at N-9 in 5′-methylammonium-5′-deoxyadenosine must be due to the unusual syn conformation of this molecule. A similar geometry was observed in *syn*-4-thiouridine[10] where the asymmetry in exocyclic angles at the glycosidic pyrimidine atom N-1 has been interpreted as a consequence of the proximity of ribose moiety and oxygen atom O-2 of the pyrimidine ring.

The adenine system in 5′-methylammonium-5′-

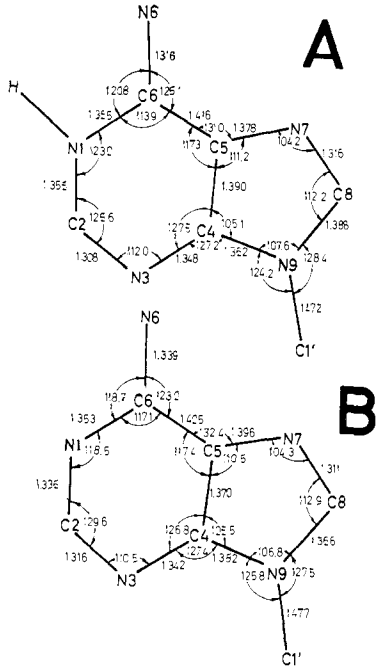


Figure 6. Comparison of averaged data for the protonated (A) and unprotonated (B) 9-substituted adenine. Data were taken from 9-methyladenine (K. Hoogsteen, *Acta Crystallogr.*, **16**, 907 (1963); W. Saenger and D. Suck, to be published), 9-ethyladenine (F. S. Mathews and A. Rich, *J. Mol. Biol.*, **8**, 89 (1964)), and deoxyadenosine,[20] and adenosine 3′-phosphate,[22] adenosine 5′-phosphate (D. G. Watson, D. J. Sutor, and P. Tollin, *Acta Crystallogr.*, **19**, 111 (1965)), and β-adenosine-2′-β-uridine-5′-phosphoric acid (E. Shefter, M. Barlow, R. A. Sparks, and K. N. Trueblood, *ibid.*, *Sect. B*, **25**, 895 (1969)), respectively.

deoxyadenosine is almost planar, the angle between the normal to least-squares pyrimidine and imidazole planes (Table V) being 1.3°. Atoms N-1 and N-9 deviate the most from the adenine plane by 0.03 Å. Atom C-1′ is 0.10 Å away on the same side of the plane as N-1, N-9, and C-2′.

(c) **Ribose.** Generally, ribose moieties in nucleosides occur in the envelope form. They are puckered such that atoms C-2′ or C-3′ are out of the plane through the other four ring atoms by about 0.5 Å.[16] The ribose conformation is called C-2′(C-3′) endo or exo if C-2′(C-3′) is on the same or opposite side of the plane as C-5′.

In 5′-methylammonium-5′-deoxyadenosine the ribose is not in an envelope form but puckered such that plane B with C-2′ endo pucker and plane C with C-3′ exo pucker (Table V) have similar planarity, the C-3′ exo pucker being a little more pronounced than the C-2′ endo pucker. This result is supported by the geometrical arrangement of atoms C-2′ and C-3′ with respect to plane D through atoms C(4)′–O(1)′–C(1)′. If the ribose would exist in an envelope conformation atoms C-2′ and C-3′ should be off this plane by about 0.1 and 0.5 Å on opposite sides of this plane, but they are 0.29 Å above (C-2′) and 0.32 Å below (C-3′) plane C(4)′–O(1)′–C(1)′ in this structure. Both the above-mentioned deviations from planarity would favor weakly a C-3′ exo conformation of the ribose.

However, since the exocyclic angles around C-2′ are about 3° greater than those at C-3′, a result which is indicative for a C-2′ endo pucker,[17,18] a decision

(16) M. Spencer, *Acta Crystallogr.*, **12**, 59 (1959).

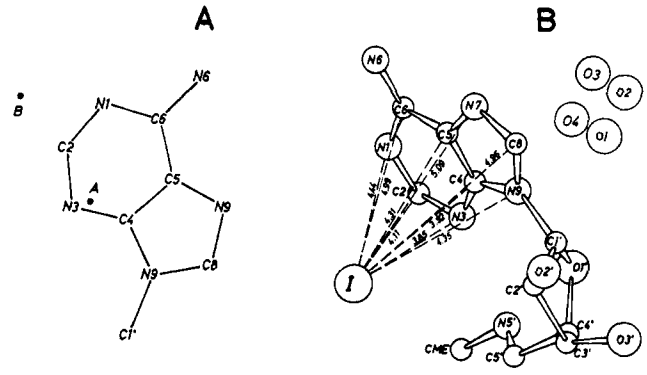


Figure 7. Projection of two symmetry-related iodine ions onto the adenine plane (left) and distances of adenine atoms from iodine ion A (right).

between C-2′ and C-3′ envelope conformation is not conclusive and the conformation of the ribose in 5′-methylammonium-5′-deoxyadenosine is best described as C-2′ endo, C-3′ exo. This pucker is quite unusual for the ribose of a nucleoside. It could be brought about by an intramolecular strain caused by the formation of the hydrogen bond between ribose atom N-5′ and adenine atom N-3′ within the same molecule.

The conformation about the C(4)′–C(5)′ bond which is defined by the angles C(3)′–C(4)′–C(5)′–N(5)′, 158.3°, and O(1)′–C(4)′–C(5)′–N(5)′, 43.0°, is trans,-gauche and not gauche,gauche, the form commonly observed.[19] So far only in deoxyadenosine,[20] uridine-2′,3′-*O*,*O*-cyclophosphorothioate,[18] thymidine,[21] and adenosine 3′-phosphate[22] has a trans,gauche conformation about the C(4)′–C(5)′ bond been observed.

The unusual position of atom N-5′ with respect to the ribose in 5′-methylammonium-5′-deoxyadenosine is a consequence of the intramolecular N(5)′–N(3) hydrogen bond which can exist only if atom N-5′ is nearly coplanar with the adenine heterocycle (Table V, plane A) and the arrangement of covalently and hydrogen bound atoms around atom N-5′ is tetrahedral (Figure 3).

The substituents CME and C-4′ at atoms N-5′ and C-5′ are as far away from each other as possible, rendering the dihedral angle C(4)′–C(5)′–N(5)′–N(5)′–CME 174.6°, *i.e.*, purely trans.

(d) **Iodide Ion.** The adenine heterocycle is located halfway between two iodide ions related to each other by the screw operation (Figure 4). The adenine plane is almost perpendicular to the I–I vector and the perpendicular distances of both iodide ions from the pyrimidine part of the heterocycle are equal, namely 3.82 Å (Table V, plane A). The projection of both iodide ions onto the adenine plane is demonstrated in Figure 7A. One iodide ion, A, is located above, the other, B, below and slightly outside the heterocyclic system. In Figure 7B the distances of atoms within the adenine

(17) M. Sundaralingam and L. H. Jensen, *J. Mol. Biol.*, **13**, 914 (1965).
(18) W. Saenger and F. Eckstein, *J. Amer. Chem. Soc.*, **92**, 4712 (1970).
(19) E. Shefter and K. N. Trueblood, *Acta Crystallogr.*, **18**, 1067 (1965).
(20) D. G. Watson, D. J. Sutor, and P. Tollin, *ibid.*, **19**, 111 (1965).
(21) D. W. Young, P. Tollin, and H. R. Wilson, *ibid.*, *Sect. B*, **25**, 1423 (1969).
(22) M. Sundaralingam, *J. Amer. Chem. Soc.*, **87**, 599 (1965).

group from iodide ion A are indicated. The heterocycle–iodide distance of 3.82 Å is significantly shorter than the sum of the van der Waals radii, 4.0 Å, for both partners, suggesting the formation of a charge-transfer type interaction between the π orbitals of the heterocycle and vacant 5d orbitals on the iodide ion.

(e) **Hydrogen Bonding and Packing Scheme.** The positions of hydrogen atoms and intermolecular nonbonded distances shorter than the sum of the van der Waals radii indicate only three inter- and intramolecular hydrogen bonds within this structure which are listed in Table III.

The ammonium nitrogen N-5′ is surrounded tetrahedrally by two covalently bound atoms C-5′ and CME and by two hydrogen-bonded atoms N-3 and O-2′. Atom N-3 is within the same molecule in 2.88 Å distance, whereas atom O-2′ belongs to a symmetry related molecule ($1 - x$, $y - {}^1/_2$, $1 - z$) 2.89 Å apart. Atom O-2′ is the acceptor for a hydrogen atom (HN-5′1) from atom N-5′ but the donor for a hydrogen bond from O-2′ (at x, y, z) to N-1 (at $\bar{x}$, $y - {}^1/_2$, $\bar{z}$) of 2.74 Å length (Figure 4).

Usually in crystal structures of nucleosides the molecules are packed such that the heterocyclic residues are parallel to each other. The parallel heterocycles then stack on top of each other as monomers or hydrogen bound dimers, thus forming endless bands. The packing arrangement in 5′-methylammonium-5′-deoxyadenosine is quite different from this scheme since no stacks or dimers are formed. The heterocycle planes are tilted about 30° to the b axis, and by action of the twofold screw they are rotated about a point near atom C-2 and translated by $b/2$, thus forming a zigzag pattern along b. The angle between adjacent heterocycles is approximately 120° (Figure 4).

The site of the disordered water molecule is not distributed within the crystal structure in the described zigzag fashion. Since the disordered water molecule is located approximately at $x = {}^1/_2$, $z = 0$ (Figure 5) the apparent action of the twofold screw axis is merely a translation by $b/2$. Thus at $x = {}^1/_2$, $z = 0$ a hydrophilic linear region extends parallel to b, bordered in the direction of the a axis by adenine heterocycles with iodide ions and in direction of the c axis by the CME–N(5)′–C(5)′–C(4)′–C(3)′–O(3)′ and O(1)′–C(1)′–C(2)′–O(2)′ parts of the ribose residues in alternating sequence.

Conclusion

5′-Methylammonium-5′-deoxyadenosine is a poor substrate for adenosine desaminase. A reason for this behavior could be the unusual stereochemistry of the nucleoside, *i.e.*, the syn conformation of the molecule and the substitution of the 5′-hydroxyl group by a methylamino group or the introduction of the positive charge in the 5′ position. In the enzymatic reaction process all three modifications of the natural adenosine structure could influence the binding of the substrate to the active site as well as the enzyme reaction. In order to clarify the situation it is intended to test enzymatic assays at different pH values, especially at higher pH where the 5′-methylammonium group becomes uncharged.

In light of previously published structures of nucleosides two reasons for the occurrence of the syn rather than the anti conformation in some of these structures can be deduced.

First, as in 5′-methylammonium-5′-deoxyadenosine, 8-bromoguanosine,[11] and 8-bromoadenosine,[11] molecular structural features such as the electron pair attracting the 5′-ammonium group or the bulky bromine atom are forcing the purine heterocycle from the usually preferred anti into the syn conformation to gain more electrostatic (hydrogen bonding) energy or to release steric strain.

Secondly, as in adenosine 3′,5′-cyclophosphate, deoxyguanosine, and 4-thiouridine, the crystal structural (packing) but not the molecular structural features are determining the relative positions of ribose and heterocycle. For instance, 4-thiouridine could be shown by nmr spectroscopy to exhibit the anti conformation in aqueous solution.[23] However, when this nucleoside was crystallized from aqueous solution, the molecules packed in syn conformation.[10] The crystal structure was governed by hydrophobic and hydrophilic regions formed by stacks of heterocycles on one side and riboses hydrogen bonded to interspersed water molecules on the other side. This packing scheme would not have been possible were the nucleoside in anti conformation.

From the above mentioned one should conclude that the conformation about the glycosidic C(1)′–N bond in nucleosides is determined by both intra- and intermolecular features. The final conformation of a nucleoside in solution will depend solely on intramolecular forces, but in polynucleotides and in crystal structures on the sum of intra- and intermolecular interactions.

Acknowledgment. The suggestion of Dr. H. Hettler and Dr. B. Jastorff to undertake this study and Dr. A. Murayama's gift of the material are gratefully acknowledged. The author is indebted to Professor F. Cramer for his interest in and support of this work, to Professor E. R. Wölfel for his consent to collect the data in the STOE factory at Darmstadt, to Dr. H. A. Paulus for his cooperation, and to Miss U. Wittenberg for her skillful assistance. All the computations except the ORTEP plot were performed on the IBM 7040 at Rechenzentrum Göttingen.

(23) K. H. Scheit and W. Saenger, *FEBS (Fed. Eur. Biochem. Soc.) Lett.*, **2**, 305 (1969).