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Molecular Conformations of Carbon-Bridged Cyclodeoxyadenosines in the Solid State and in Solution. 2′-Deoxy-8,2′-methanoadenosine and 2′-Deoxy-8,2′-ethanoadenosine

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Two carbon-bridged cyclodeoxyadenosines, 2′-deoxy-8,2′-methanoadenosine (8,2′-MA) and 2′-deoxy-8,2′-ethanoadenosine (8,2′-EA), were investigated by X-ray and nuclear magnetic resonance (NMR) analyses. Both compounds crystallize in the orthorhombic system with space group $P2_12_12_1$. The unit-cell dimensions are $a=9.427$ (1), $b=17.287$ (2) and $c=6.908$ (1) Å for 8,2′-MA and $a=15.599$ (4), $b=19.468$ (5) and $c=9.955$ (3) Å for 8,2′-EA. Both structures were solved by direct methods and refined to R values of 0.037 for 8,2′-MA and 0.052 for 8,2′-EA. There are two independent cyclonucleoside molecules and one isopropanol molecule per asymmetric unit in the crystal of 8,2′-EA. The glycosidic torsion angles, χ, O(4′)–C(1′)–N(9)–C(4), are −68.7 (5)° and −77.2 (4)° for molecules (a) and (b) of 8,2′-EA, respectively, and −65.7(4)° for 8,2′-MA, and are in high-*anti* conformation (χ; −90°—−50°). The deoxyribose ring of 8,2′-MA takes the C(3′)-*exo*, C(4′)-*endo* twist pucker, and the conformation about the C(4′)–C(5′) bond is *gauche*$^+$. In contrast, both deoxyribose rings of 8,2′-EA exhibit C(1′)-*exo* envelope puckering, and the conformation about the C(4′)–C(5′) bond is *gauche*$^+$ for molecule (a) and *trans* for molecule (b). ^{1}H-NMR data indicate that both of the cyclonucleosides in solution exist in the equilibrium of several sugar and exocyclic conformations. 8,2′-EA adopts two different conformations, even around the glycosidic bond.

Keywords carbon-bridged cyclodeoxyadenosine; X-ray analysis; NMR; nucleoside conformation; conformational flexibility

Cyclonucleosides with a covalent bond between base and sugar moieties have served as useful model compounds for investigations of the conformational aspects of enzyme–substrate interactions[2)] and physicochemical properties of nucleosides and nucleotides in solution.[3)] In particular, carbon-bridged cyclonucleosides are much better model nucleosides for these studies because the bridging carbon atom has less effect on the physicochemical properties of the base or the sugar moiety than the oxygen or sulfur bridges, and because a variety of cyclonucleosides can be synthesized with the selection of the number of bridging carbon atoms.[4)] In this paper, we deal with the conformations of two carbon-bridged cyclodeoxyadenosines in the solid state and in solution. Both cyclonucleosides take the cyclization between C(8) in the base and C(2′) in the sugar: One through the methylene moiety, the other through the ethylene moiety.

Experimental

2′-Deoxy-8,2′-methanoadenosine (8,2′-MA)[4a)] and 2′-deoxy-8,2′-ethanoadenosine (8,2′-EA)[4b)] were synthesized as described in previous papers and crystallized from aqueous and isopropanol–water mixture solutions, respectively. The crystal data are as follows: 8,2′-MA, chemical formula $C_{11}H_{13}N_5O_3$; M_r 263.26; space group $P2_12_12_1$; $a=9.427$ (1), $b=17.287$ (2), $c=6.908$ (1) Å; $V=1125.8$ (2) Å^3; $Z=4$; $D_x=1.553$ Mg·m^{-3}; $F(000)=552$; $\mu(\mathrm{Cu}K\alpha)=0.94$ mm^{-1}. 8,2′-EA, chemical formula $C_{12}H_{15}N_5O_3 \cdot 1/2C_3H_7OH$; M_r 307.33; space group $P2_12_12_1$; $a=15.599$ (4), $b=19.468$ (5), $c=9.955$ (3) Å; $V=3023.1$ (15) Å^3; $Z=8$; $D_x=1.350$ Mg·m^{-3}; $F(000)=1304$; $\mu(\mathrm{Cu}K\alpha)=0.80$ mm^{-1}.

The intensity data were measured on a Rigaku automatic four-circle diffractometer with Cu$K\alpha$ radiation. By means of the ω–2θ scan technique, intensities of 990 (for 8,2′-MA) and 2517 (for 8,2′-EA) independent reflections with $\sin\theta/\lambda<0.562$ Å^{-1} were obtained. Intensities were corrected for Lorentz and polarization factors, but not for absorption, because of the small sizes of the crystals (0.3 × 0.2 × 0.07 mm for 8,2′-MA, 0.5 × 0.07 × 0.07 mm for 8,2′-EA).

Both structures were solved by direct methods using the program MULTAN78[5)] and refined by the block-diagonal least-squares method with anisotropic temperature factors for non-hydrogen atoms of nucleoside molecules. The non-hydrogen atoms of isopropanol solvent and all hydrogen atoms of nucleosides were located on the difference Fourier syntheses. The final refinements, including the non-hydrogen atoms of isopropanol and the hydrogen atoms of nucleosides with isotropic temperature factors, reduced the R value to 0.037 ($R_w=0.054$) for 981 reflections with $F_O \neq 0$ of 8,2′-MA and 0.052 ($R_w=0.073$) for 2468 reflections with $F_O \neq 0$ of 8,2′-EA. The quantity minimized was $\Sigma\omega(|F_O|-k|F_C|)^2$, where $\omega=(\sigma^2(F_O)+a|F_O|+b|F_O|^2)^{-1}$ ($a=0.096$, $b=0.003$ for 8,2′-MA, $a=0.062$, $b=0.003$ for 8,2′-EA) and k is the scale factor. The highest peak height in the final difference Fourier map is 0.14 eÅ^{-3} for 8,2′-MA and 0.50 eÅ^{-3} (about 1.0 Å from isopropanol molecule) for 8,2′-EA. All numerical calculations were carried out on an ACOS 850 computer at the Research Center for Protein Engineering, Institute for Protein Research, Osaka University, using the programs of the universal Crystallographic Computing System-Osaka (1979)[6)] and its modifications. The atomic scattering factors used were those cited in International Tables for X-Ray Crystallography (1974).[7)]

Proton nuclear magnetic resonance (^{1}H-NMR) spectra of 8,2′-MA and 8,2′-EA in D_2O were measured by a JEOL-GX 500 Spectrometer at 20 °C. Chemical shifts were recorded by reference to internal *tert*-BuOH (1.23 ppm). The resonance peaks were assigned by the method of homonuclear decoupling.

Results and Discussion

The final atomic parameters are given in Table I for 8,2′-MA and in Table II for 8,2′-EA. The bond distances and angles are listed in Tables III and IV, respectively. The relevant torsion angles are listed in Table V. ORTEP drawings[8)] of both cyclonucleosides are shown in Figs. 1 and 2. The molecular packings and hydrogen bondings are illustrated in Fig. 3 for 8,2′-MA and in Fig. 4 for 8,2′-EA.

Adenine Moiety For the most part, bond distances and angles in the adenine moiety for 8,2′-MA and the two independent molecules (a) and (b) of 8,2′-EA are in agreement with standard values found in crystal structures containing adenine bases.[9)] Greater variations occur in C(5)–C(6) [(a) of 8,2′-EA], N(7)–C(8) [(b) of 8,2′-EA] and C(8)–N(9) [(b) of 8,2′-EA] bond distances. The geometry and molecular dimensions of the adenine ring generally show little change by cyclization through the carbon atom(s) between C(8) and C(2′) atoms as well as the observation of the pyrimidine cyclonucleosides cyclized through a carbon atom between C(6) in the base and any atoms in the sugar

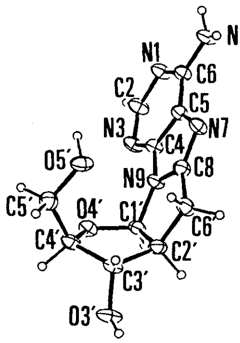

Fig. 1. Molecular Conformation of 8,2′-MA with Atomic Numberings

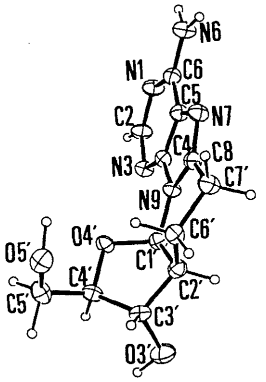

Fig. 2. Molecular Conformation of Molecule (a) of 8,2′-EA with Atomic Numberings

The molecular conformation of molecule (b) is similar to that of molecule (a) except for the conformation about the exocyclic C(4′)–C(5′) bond.

Table I. Atomic Coordinates and Isotropic Thermal Parameters (Å^2) for 8,2′-MA

Atom	x	y	z	B_{eq}/B_{iso}
N(1)	0.3288 (3)	−0.0368 (1)	0.3955 (4)	3.8 (1)
C(2)	0.1970 (3)	−0.0672 (2)	0.3973 (5)	4.0 (1)
N(3)	0.0744 (2)	−0.0313 (1)	0.3815 (4)	3.5 (1)
C(4)	0.0920 (3)	0.0449 (1)	0.3649 (4)	3.0 (1)
C(5)	0.2202 (3)	0.0852 (2)	0.3621 (4)	3.2 (1)
C(6)	0.3435 (3)	0.0403 (2)	0.3790 (4)	3.3 (1)
N(7)	0.1988 (2)	0.1647 (1)	0.3394 (4)	3.4 (1)
C(8)	0.0600 (3)	0.1702 (1)	0.3280 (5)	3.3 (1)
N(9)	−0.0089 (2)	0.1014 (1)	0.3414 (3)	3.1 (1)
N(6)	0.4744 (2)	0.0714 (1)	0.3798 (5)	4.2 (1)
C(1′)	−0.1622 (3)	0.1052 (2)	0.3271 (4)	3.2 (1)
C(2′)	−0.1892 (3)	0.1932 (2)	0.3100 (5)	3.4 (1)
C(3′)	−0.2823 (3)	0.2007 (2)	0.1269 (5)	3.1 (1)
C(4′)	−0.2630 (3)	0.1244 (1)	0.0178 (5)	3.3 (1)
C(5′)	−0.1641 (3)	0.1276 (2)	−0.1523 (4)	3.8 (1)
C(6′)	−0.0434 (3)	0.2346 (2)	0.3030 (5)	4.0 (1)
O(3′)	−0.4284 (2)	0.2069 (1)	0.1817 (4)	4.5 (1)
O(4′)	−0.2151 (2)	0.0685 (1)	0.1585 (3)	3.3 (0)
O(5′)	−0.0246 (2)	0.1515 (1)	−0.1025 (4)	4.1 (1)
H(2)	0.195 (4)	−0.124 (2)	0.417 (5)	2.0 (7)
H(N6)	0.484 (4)	0.122 (2)	0.339 (5)	3.3 (9)
H(N6′)	0.549 (4)	0.041 (2)	0.359 (6)	3.4 (8)
H(1′)	−0.211 (4)	0.076 (2)	0.437 (6)	3.0 (8)
H(2′)	−0.245 (3)	0.216 (2)	0.429 (5)	2.3 (7)
H(3′)	−0.255 (3)	0.249 (2)	0.050 (5)	2.0 (7)
H(4′)	−0.361 (4)	0.106 (2)	−0.043 (5)	2.4 (7)
H(5′)	−0.206 (4)	0.170 (2)	−0.251 (5)	2.6 (7)
H(5″)	−0.157 (4)	0.069 (2)	−0.211 (5)	2.1 (7)
H(6′)	−0.027 (4)	0.265 (2)	0.171 (5)	2.5 (7)
H(6″)	−0.031 (4)	0.272 (2)	0.407 (6)	3.0 (8)
H(O3′)	−0.461 (4)	0.255 (2)	0.175 (6)	3.4 (8)
H(O5′)	0.048 (4)	0.106 (2)	−0.099 (6)	3.6 (8)

moiety.[2d] All adenine rings are essentially coplanar. C(6′) of 8,2′-MA and C(7′) of molecules (a) and (b) of 8,2′-EA are almost on the planes and are displaced by 0.028 (4), 0.048 (6) and 0.125 (6) Å, respectively.

Glycosidic Bond and Newly Formed Cyclic Ring There are some characteristics in the bond angles of the newly formed cyclic ring for both cyclonucleosides. In 8,2′-EA, the bond angles around C(8) are similar to those found in 8-methyladenosine 3′-monophosphate.[10] And the bond angles around N(9) and C(1′) atoms also agree well with those of non-cyclonucleosides with adenine bases.[11] In contrast, those bond angles of 8,2′-MA show large distortions from those in 8-methyladenosine 3′-monophosphate and other adenine nucleosides. The C(4)–N(9)–C(1′) and N(9)–C(8)–C(6′) bond angles are larger by *ca.* 10°, while the N(9)–C(8)–C(6′), C(8)–N(9)–C(1′) and N(9)–C(1′)–C(2′) are smaller by *ca.* 10°. Such deviations are also observed in oxygen- and sulfur-bridged cyclonucleosides and cyclonucleotides such as 8,2′-anhydro-1-β-D-arabinofuranosyl-8-hydroxyadenosine (8,2′-OA),[12] 8,2′-anhydro-1-β-D-arabinofuranosyl-8-mercaptoadenosine 5′-monophosphate (8,2′-SAMP (5′))[13] and 8,2′-anhydro-1-β-D-arabinofuranosyl-8-mercaptoadenosine 3′-monophosphate (8,2′-SAMP (3′)).[14] The cyclization between the base and sugar moieties restrains the conformation about the glycosidic bond. Both cyclonucleosides show a high-*anti* conformation with the torsion angle χ, O(4′)–C(1′)–N(9)– C(4), of −65.7 (4)° for 8,2′-MA, −68.7 (5)° for molecule (a) and −77.2 (5)° for molecule (b) of 8,2′-EA. The model con-

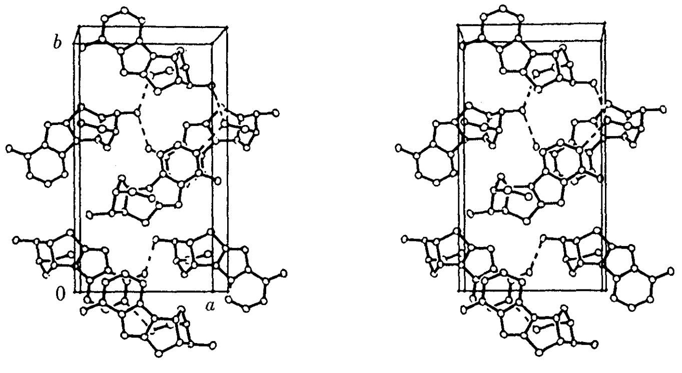

Fig. 3. Stereoscopic Drawing of the Crystal Structure of 8,2′-MA along the *c* Axis

Dashed lines indicate the hydrogen bonds.

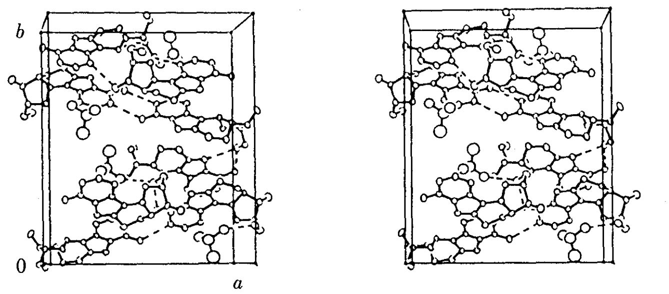

Fig. 4. Stereoscopic Drawing of the Crystal Structure of 8,2′-EA along the *c* Axis

Dashed lines indicate the hydrogen bonds.

TABLE II. Atomic Coordinates and Isotropic Thermal Parameters (Å^2) for 8,2′-EA

Atom	x	y	z	B_{eq}/B_{iso}
Molecule (a)				
N(1)	0.6667 (2)	0.1725 (2)	0.0894 (4)	3.8 (1)
C(2)	0.7447 (3)	0.1563 (2)	0.0427 (5)	3.7 (1)
N(3)	0.8208 (2)	0.1789 (2)	0.0828 (3)	3.1 (1)
C(4)	0.8131 (2)	0.2252 (2)	0.1829 (4)	2.8 (1)
C(5)	0.7365 (3)	0.2482 (2)	0.2372 (4)	3.1 (1)
C(6)	0.6615 (3)	0.2197 (2)	0.1900 (4)	3.1 (1)
N(7)	0.7515 (2)	0.2955 (2)	0.3411 (4)	3.5 (1)
C(8)	0.8352 (3)	0.2997 (2)	0.3465 (4)	3.2 (1)
N(9)	0.8762 (2)	0.2592 (2)	0.2534 (3)	2.8 (1)
N(6)	0.5844 (2)	0.2358 (2)	0.2380 (4)	4.1 (1)
C(1′)	0.9677 (3)	0.2494 (2)	0.2416 (4)	3.1 (1)
C(2′)	1.0198 (3)	0.2994 (2)	0.3277 (5)	3.4 (1)
C(3′)	1.1032 (3)	0.2602 (2)	0.3417 (5)	3.5 (1)
C(4′)	1.0780 (3)	0.1839 (2)	0.3229 (4)	3.3 (1)
C(5′)	1.0941 (3)	0.1371 (2)	0.4413 (5)	4.3 (1)
C(6′)	0.9748 (3)	0.3130 (2)	0.4615 (5)	3.3 (1)
C(7′)	0.8855 (3)	0.3437 (2)	0.4400 (5)	3.9 (1)
O(4′)	0.9879 (2)	0.1837 (1)	0.2935 (3)	3.1 (1)
O(3′)	1.1605 (2)	0.2820 (2)	0.2377 (4)	5.8 (1)
O(5′)	1.0504 (2)	0.1590 (2)	0.5587 (3)	4.5 (1)
H(2)	0.747 (3)	0.120 (2)	−0.028 (5)	1.4 (9)
H(N6)	0.575 (3)	0.269 (2)	0.310 (5)	2.1 (10)
H(N6′)	0.539 (4)	0.215 (3)	0.200 (6)	4.1 (13)
H(1′)	0.988 (3)	0.255 (2)	0.142 (6)	2.9 (11)
H(2′)	1.027 (3)	0.345 (2)	0.282 (5)	2.4 (11)
H(3′)	1.129 (3)	0.269 (2)	0.433 (5)	1.2 (9)
H(4′)	1.115 (3)	0.163 (2)	0.245 (5)	2.4 (10)
H(5′)	1.166 (3)	0.139 (2)	0.459 (5)	1.9 (10)
H(5″)	1.080 (3)	0.089 (3)	0.421 (5)	2.7 (11)
H(6′)	1.009 (3)	0.347 (2)	0.524 (5)	2.0 (10)
H(6″)	0.968 (3)	0.268 (2)	0.516 (5)	2.1 (10)
H(7′)	0.891 (3)	0.394 (2)	0.397 (5)	2.3 (11)
H(7″)	0.852 (4)	0.348 (3)	0.528 (6)	3.3 (12)
H(O3′)	1.197 (3)	0.241 (3)	0.194 (6)	3.6 (13)
H(O5′)	0.984 (5)	0.152 (4)	0.543 (9)	7.0 (19)
Molecule (b)				
N(1)	0.4275 (2)	0.1714 (2)	0.1256 (4)	3.6 (1)
C(2)	0.3502 (3)	0.1930 (2)	0.1633 (5)	3.7 (1)
N(3)	0.2739 (2)	0.1737 (2)	0.1194 (4)	3.4 (1)
C(4)	0.2815 (2)	0.1263 (2)	0.0208 (4)	2.7 (1)
C(5)	0.3560 (2)	0.1009 (2)	−0.0300 (4)	2.8 (1)
C(6)	0.4335 (2)	0.1233 (2)	0.0268 (4)	3.0 (1)
N(7)	0.3381 (2)	0.0544 (2)	−0.1344 (3)	3.4 (1)
C(8)	0.2562 (3)	0.0536 (2)	−0.1438 (4)	3.2 (1)
N(9)	0.2171 (2)	0.0968 (2)	−0.0509 (3)	2.9 (1)
N(6)	0.5107 (2)	0.1018 (2)	−0.0097 (4)	4.0 (1)
C(1′)	0.1259 (2)	0.1065 (2)	−0.0350 (4)	2.8 (1)
C(2′)	0.0743 (3)	0.0779 (2)	−0.1508 (4)	3.1 (1)
C(3′)	−0.0147 (3)	0.0733 (2)	−0.0853 (4)	3.1 (1)
C(4′)	0.0054 (2)	0.0661 (2)	0.0660 (4)	2.9 (1)
C(5′)	−0.0250 (3)	−0.0010 (2)	0.1263 (5)	3.9 (1)
C(6′)	0.1098 (3)	0.0089 (2)	−0.1977 (5)	3.9 (1)
C(7′)	0.2021 (3)	0.0165 (3)	−0.2447 (5)	4.5 (1)
O(4′)	0.0982 (2)	0.0681 (2)	0.0784 (3)	2.9 (1)
O(3′)	−0.0611 (2)	0.1337 (2)	−0.1125 (3)	4.4 (1)
O(5′)	0.0014 (2)	−0.0096 (2)	0.2603 (4)	4.4 (1)
H(2)	0.355 (3)	0.230 (2)	0.243 (5)	2.3 (10)
H(N6)	0.519 (3)	0.069 (3)	−0.080 (6)	3.6 (12)
H(N6′)	0.563 (4)	0.121 (3)	0.025 (7)	5.0 (15)
H(1′)	0.117 (3)	0.158 (2)	−0.020 (5)	1.3 (9)
H(2′)	0.081 (3)	0.112 (2)	−0.230 (5)	2.1 (10)
H(3′)	−0.049 (3)	0.032 (2)	−0.126 (5)	2.2 (10)
H(4′)	−0.023 (3)	0.105 (2)	0.118 (5)	1.7 (9)
H(5′)	−0.091 (3)	−0.001 (3)	0.125 (5)	2.3 (10)
H(5″)	0.003 (3)	−0.043 (2)	0.063 (5)	2.5 (11)
H(6′)	0.110 (2)	−0.026 (2)	−0.114 (4)	0.7 (8)
H(6″)	0.071 (3)	−0.010 (2)	−0.269 (5)	2.1 (10)
H(7′)	0.204 (4)	0.042 (3)	−0.328 (8)	7.0 (19)
H(7″)	0.232 (3)	−0.033 (2)	−0.261 (5)	2.4 (10)
H(O3′)	−0.102 (4)	0.146 (4)	−0.045 (7)	6.0 (17)
H(O5′)	0.058 (4)	−0.018 (4)	0.283 (7)	6.2 (18)
Isopropanol				
O(1)	−0.1175 (3)	0.1407 (3)	−0.3786 (5)	7.6 (1)
C(1)	−0.1898 (6)	0.0987 (5)	−0.4167 (1)	10.0 (2)
C(2)$^{a)}$	−0.171 (2)	0.027 (1)	−0.392 (3)	13.0 (7)
C(3)$^{a)}$	−0.178 (1)	0.079 (9)	−0.567 (2)	9.2 (4)
C(4)$^{a)}$	−0.222 (1)	0.113 (8)	−0.541 (2)	8.8 (4)
C(5)$^{a)}$	−0.222 (1)	0.050 (9)	−0.330 (2)	9.5 (4)

a) Occupancy factor is 0.5.

TABLE III. Bond Distances (Å)

	8,2′-MA	8,2′-EA	
		(a)	(b)
N(1)–C(2)	1.349 (4)	1.340 (6)	1.330 (6)
N(1)–C(6)	1.345 (4)	1.361 (6)	1.362 (5)
C(2)–N(3)	1.317 (4)	1.327 (6)	1.323 (6)
N(3)–C(4)	1.332 (4)	1.349 (5)	1.353 (5)
C(4)–C(5)	1.396 (4)	1.386 (6)	1.360 (5)
C(4)–N(9)	1.373 (3)	1.378 (5)	1.360 (5)
C(5)–C(6)	1.402 (4)	1.378 (6)	1.404 (6)
C(5)–N(7)	1.398 (4)	1.404 (5)	1.406 (5)
C(6)–N(6)	1.346 (4)	1.331 (6)	1.326 (6)
N(7)–C(8)	1.315 (4)	1.309 (5)	1.281 (5)
C(8)–N(9)	1.359 (4)	1.375 (5)	1.391 (5)
C(8)–C(6′)[C(7′)]	1.489 (4)	1.489 (6)	1.497 (6)
N(9)–C(1′)	1.449 (3)	1.444 (5)	1.444 (5)
C(1′)–C(2′)	1.548 (4)	1.530 (6)	1.512 (6)
C(1′)–O(4′)	1.417 (3)	1.414 (5)	1.421 (5)
C(2′)–C(3′)	1.545 (4)	1.515 (6)	1.538 (6)
C(2′)–C(6′)	1.550 (4)	1.528 (6)	1.525 (6)
C(3′)–C(4′)	1.530 (4)	1.546 (6)	1.545 (6)
C(3′)–O(3′)	1.433 (3)	1.433 (6)	1.408 (6)
C(4′)–C(5′)	1.501 (4)	1.510 (6)	1.514 (6)
C(4′)–O(4′)	1.443 (3)	1.436 (5)	1.454 (5)
C(5′)–O(5′)	1.420 (4)	1.419 (6)	1.406 (6)
C(6′)–C(7′)	—	1.531 (6)	1.521 (7)

sideration in 8,2′-EA indicates that the glycosidic conformation can adopt either of two forms: the *endo* form with C(7′) over the sugar ring, or the *exo* form with C(7′) on the opposite side. In this work, two 8,2′-EA molecules have the *exo* form. On the other hand, 2′-deoxy-6,2′-ethanouridine with the same six-membered cyclic ring system takes the *endo* form ($\chi = -91.9°$).15) In crystal structures of 8,2′-cyclonucleosides with a five-membered cyclic ring such as 8,2′-MA, the glycosidic torsion angles are evenly in the range of −52° to −75°.12−14,16) In 8,2′-MA, the newly formed five-membered ring is almost planar and the largest deviation from the best plane is 0.022 (4) Å for C(2′). In 8,2′-EA, the newly formed six-membered ring has a half-chair form with C(6′) and C(2′) displaced by 0.538 (8) and 0.166 (8) Å for molecule (a) and 0.466 (8) and 0.249 (8) Å for molecule (b), respectively, on opposite sides of the remaining four atoms.

Deoxyribose Moiety The bond distances and angles in the deoxyribose moiety are slightly different between those

TABLE IV. Bond Angles (°)

	8,2′-MA	8,2′-EA	
		(a)	(b)
C(2)–N(1)–C(6)	118.9 (3)	117.9 (4)	118.9 (4)
N(1)–C(2)–N(3)	128.6 (3)	129.0 (4)	129.3 (4)
C(2)–N(3)–C(4)	111.3 (2)	111.4 (3)	110.7 (3)
N(3)–C(4)–C(5)	127.1 (2)	125.5 (4)	126.4 (4)
N(3)–C(4)–N(9)	128.8 (2)	129.3 (3)	127.2 (3)
C(5)–C(4)–N(9)	104.1 (2)	105.2 (3)	106.4 (3)
C(4)–C(5)–C(6)	116.2 (2)	118.0 (4)	118.2 (4)
C(4)–C(5)–N(7)	111.5 (2)	110.9 (3)	109.9 (3)
C(6)–C(5)–N(7)	132.3 (3)	131.0 (4)	131.9 (4)
N(1)–C(6)–C(5)	118.0 (3)	118.2 (4)	116.4 (4)
N(1)–C(6)–N(6)	119.4 (3)	118.5 (4)	118.5 (4)
C(5)–C(6)–N(6)	122.6 (3)	123.3 (4)	125.1 (4)
C(5)–N(7)–C(8)	102.8 (2)	103.7 (3)	105.0 (3)
N(7)–C(8)–N(9)	114.1 (2)	113.6 (3)	112.4 (4)
N(7)–C(8)–C(6′)[C(7′)]	135.4 (3)	126.0 (4)	128.1 (4)
N(9)–C(8)–C(6′)[C(7′)]	110.5 (2)	120.4 (4)	119.3 (4)
C(4)–N(9)–C(8)	107.4 (2)	106.6 (3)	106.3 (3)
C(4)–N(9)–C(1′)	137.0 (2)	126.9 (3)	127.9 (3)
C(8)–N(9)–C(1′)	115.6 (2)	126.2 (3)	125.8 (3)
N(9)–C(1′)–C(2′)	102.3 (2)	113.3 (3)	113.1 (3)
N(9)–C(1′)–O(4′)	112.7 (2)	108.1 (3)	108.5 (3)
C(2′)–C(1′)–O(4′)	108.6 (2)	104.5 (3)	104.5 (3)
C(1′)–C(2′)–C(3′)	103.8 (2)	100.8 (3)	100.3 (3)
C(1′)–C(2′)–C(6′)	108.1 (2)	110.8 (3)	111.4 (3)
C(3′)–C(2′)–C(6′)	116.1 (2)	113.6 (3)	114.0 (3)
C(2′)–C(3′)–C(4′)	105.3 (2)	104.7 (3)	103.6 (3)
C(2′)–C(3′)–O(3′)	109.6 (2)	108.6 (4)	109.5 (3)
C(4′)–C(3′)–O(3′)	108.0 (2)	110.9 (3)	111.6 (3)
C(3′)–C(4′)–C(5′)	115.3 (2)	116.2 (4)	113.6 (3)
C(3′)–C(4′)–O(4′)	106.5 (2)	106.0 (3)	106.4 (3)
C(5′)–C(4′)–O(4′)	111.0 (2)	108.7 (3)	107.6 (3)
C(4′)–C(5′)–O(5′)	113.3 (2)	112.5 (4)	112.8 (4)
C(2′)–C(6′)–C(7′)	—	111.3 (3)	110.6 (4)
C(8)–C(7′)–C(6′)	—	110.1 (4)	112.0 (4)
C(1′)–O(4′)–C(4′)	111.3 (2)	106.9 (3)	104.4 (3)
C(2′)–C(6′)–C(8)	103.4 (2)	—	—

TABLE V. Selected Torsion Angles (°)

	8,2′-MA	8,2′-EA	
		(a)	(b)
C(4)–N(9)–C(1′)–O(4′) (χ)	−65.7 (4)	−68.7 (5)	−77.2 (4)
C(8)–N(9)–C(1′)–O(4′)	113.9 (3)	104.5 (4)	101.8 (4)
C(4′)–O(4′)–C(1′)–C(2′) (τ_0)	6.0 (3)	−40.1 (4)	−44.7 (3)
O(4′)–C(1′)–C(2′)–C(3′) (τ_1)	7.8 (3)	40.1 (4)	43.9 (4)
C(1′)–C(2′)–C(3′)–C(4′) (τ_2)	−17.7 (3)	−25.1 (4)	−25.9 (4)
C(2′)–C(3′)–C(4′)–O(4′) (τ_3)	21.7 (3)	2.9 (4)	1.0 (4)
C(3′)–C(4′)–O(4′)–C(1′) (τ_4)	−17.6 (3)	23.0 (4)	26.6 (4)
C(3′)–C(4′)–C(5′)–O(5′) (ψ)	59.0 (3)	58.4 (5)	174.0 (3)
O(4′)–C(4′)–C(5′)–O(5′)	−62.2 (3)	−61.0 (5)	56.4 (4)
C(1′)–N(9)–C(8)–C(7′)[C(6′)]	0.5 (3)	5.6 (6)	4.6 (6)
C(2′)–C(1′)–N(9)–C(8)	−2.6 (3)	−10.8 (5)	−13.6 (5)
C(6′)–C(2′)–C(1′)–N(9)	3.5 (3)	36.9 (5)	40.7 (4)
C(8)–C(6′)–C(2′)–C(1′)	−3.3 (3)	—	—
N(9)–C(8)–C(6′)–C(2′)	1.8 (3)	—	—
C(7′)–C(6′)–C(2′)–C(1′)	—	−59.5 (4)	−60.2 (5)
C(8)–C(7′)–C(6′)–C(2′)	—	52.9 (5)	50.1 (5)
N(9)–C(8)–C(7′)–C(6′)	—	−26.3 (5)	−23.0 (6)

TABLE VI. Hydrogen Bond Distances (Å) and Angles (°)

		D···A	H···A	D–H···A
8,2′-MA				
N(6)–H···O(3′)	$x+1, y, z$	2.863 (3)	2.01 (4)	154 (3)
O(3′)–H···O(5′)	$x-1/2, -y+1/2, -z$	2.668 (3)	1.80 (4)	167 (4)
O(5′)–H···N(1)	$-x+1/2, -y, z-1/2$	2.709 (3)	1.67 (4)	176 (4)
8,2′-EA				
N(6a)–H···N(1b)	x, y, z	2.970 (5)	2.07 (6)	175 (5)
N(6b)–H···N(1a)	x, y, z	2.964 (5)	2.00 (6)	173 (6)
O(I)–H···O(3′b)	x, y, z	2.794 (6)	—	—
N(6a)–H···O(5′a)	$x-1/2, -y+1/2, -z+1$	2.928 (5)	1.95 (5)	175 (4)
N(6b)–H···O(5′b)	$-x+1/2, -y, z-1/2$	2.915 (5)	1.99 (5)	162 (5)
O(3′a)–H···N(3b)	$x+1, y, z$	2.993 (5)	1.93 (5)	174 (5)
O(3′b)–H···N(3a)	$x-1, y, z$	2.819 (5)	1.87 (7)	174 (6)
O(5′a)–H···O(I)	$x+1, y, z+1$	2.716 (6)	1.78 (8)	145 (7)
O(5′b)–H···N(7b)	$-x+1/2, -y, z+1/2$	2.851 (5)	1.95 (7)	164 (6)

of three cyclonucleosides. The most interesting differences are observed in the C(1′)–O(4′)–C(4′) bond angles. This is probably due to the differences in the sugar conformation and the amplitudes of puckering as described below.[17] The deoxyribose conformation is C(3′)-*exo*, C(4′)-*endo* ($P=215°$, $\tau_m=22°$) for 8,2′-MA and C(1′)-*exo* ((a): $P=128°$, $\tau_m=41°$, (b): $P=125°$, $\tau_m=45°$) for both molecules of 8,2′-EA. All conformations deviate significantly from the usual pseudorotation range (N-type; $P=0$—$36°$, S-type; $P=144$—$180°$).[18] However, these unusual puckerings are observed in several cyclonucleosides and cyclonucleotides. For example, 8,2′-OA[12] and 8,2′-SAMP (3′)[14] have a similar C(4′)-*endo*, C(3′)-*exo* conformation and a C(1′)-*exo* puckering is observed in 7,2′-cycloorotidine.[18] The conformation of the exocyclic bond is *gauche*$^+$ for 8,2′-MA [$\psi=59.0(3)°$] and molecule (a) of 8,2′-EA [$\psi=58.4(5)°$], and *trans* for molecule (b) of 8,2′-EA [$\psi=174.0(3)°$]. In crystal structures of nucleosides and nucleotides, the *gauche*$^+$ conformation is most preferable, with the second being *trans*.[19] The amplitude (τ_m) of the sugar puckering in 8,2′-MA is considerably smaller than the mean value (38.7°) of nucleosides.[17] The smaller value of τ_m depends on the C(3′)-*exo*, C(4′)-*endo* puckering and the *gauche*$^+$ conformation, because these sugar and exocyclic bond conformations with the larger τ_m give rise to unallowed intramolecular short contacts between O(5′) and the base atoms. In 8,2′-MA, there are some close contacts with atomic distances of 3.096 (4) Å [O(5′)···C(8)] and 3.190 (3) Å [O(5′)···N(9)]. More close contacts were commonly found in 2,2′-cyclocytidine derivatives.[20]

Molecular Packing and Hydrogen Bonding In 8,2′-MA, molecules connect to neighboring molecules with three kinds of hydrogen bonds (Table VI). The base stacking between pyrimidine parts related by the 2-fold screw axis also contributes to the stabilization of the crystal packing. Extensive overlapping occurs between the pyrimidine rings. The dihedral angle and separation between the stacked bases are 12.5 (1)° and about 3.4 Å, respectively.

In 8,2′-EA, the two independent molecules (a) and (b) form the hydrogen bonded dimer with two N(6)–H···N(1) hydrogen bonds. The paired adenine rings are almost coplanar and the dihedral angle is 4.5 (1)°. Such a base pairing scheme is rarely observed in adenine nucleosides and nucleotides, although it is not so unusual in adenine bases.[21] All remaining hydrogen atoms attached to oxygen and nitrogen atoms also participate in hydrogen bonds as shown in Table VI. The hydroxyl group of isopropanol solvent is fixed by two hydrogen bonds with O(5′) of molecule (a) and O(3′) of molecule (b), but the isopropyl

Table VII. ^{1}H Chemical Shifts (δ in ppm) and Coupling Constants (J in Hz)

	δ			J	
	8,2′-MA	8,2′-EA		8,2′-MA	8,2′-EA
H2	8.002	7.997	H1′-H2′	6.1	6.1
H1′	6.364	6.026	H2′-H3′	3.7	5.5
H2′	3.696	2.793	H3′-H4′	3.7	5.5
H3′	4.286	4.176	H4′-H5′	3.7	3.0
H4′	4.236	4.026	H4′-H5″	5.5	5.2
H5′	3.519	3.768	H5′-H5″	−12.8	−12.7
H5″	3.371	3.610	H6′-H2′	10.4	6.4
H6′	3.333	2.067	H6″-H2′	3.7	6.4
H6″	3.093	2.067	H6′-H6″	−18.3	—
H7′	—	2.994	H6′-H7′	—	6.7
H7″	—	2.898	H6′-H7″	—	6.1
			H7′-H7″	—	−17.7

Table VIII. Observed and Calculated Coupling Constants (Hz) of 8,2′-MA

	J_{obs}	J_{calc}		
		X-Ray		Model
Glycosidic bond				
H2′-H6′	10.4	11.3		
H2′-H6″	2.8	3.7		
Sugar				
		$^4T_3$	$_4T^3$	$^4T_3:{}_4T^3=55:45$
H1′-H2′	6.4	9.0	8.7	8.9
H2′-H3′	3.7	0.2	5.5	2.6
H3′-H4′	3.7	0.2	9.5	4.4
Exocyclic bond				
		g^+	g^-/t	$g^+:g^-/t=45:55$
(H4′-H5′)+(H4′-H5″)	9.2	4.0	13.4	9.2

group is disordered with two orientations of which population ratio is 0.5:0.5. Base stackings are observed between the imidazole parts of the adjacent molecules (a) and (b) with the separation of about 3.5 Å and a dihedral angle of 0.5 (1)°.

^{1}H-NMR Analysis The observed chemical shifts and coupling constants of 8,2′-MA and 8,2′-EA are presented in Table VII. The conformation of the glycosidic bond, sugar ring and exocyclic bond can be estimated from the relationship between their coupling constants and HH torsion angles by applying the modified Karplus equation.[22] To investigate whether both cyclonucleosides in solution maintain the conformation found in the solid state, their vicinal coupling constants (J_{calc}, X-ray) calculated from observed HH torsion angles were compared to the observed ones (J_{obs}; Tables VIII and IX). The calculated values (J_{calc} for H2′–H6′ and H2′–H6″) around the C(2′)–C(6′) bond of 8,2′-MA are only in agreement with the observed ones. The others are significantly different from each other. This fact suggests that both of the nucleosides in solution take not only the conformers found in crystal but also other conformers, except for the glycosidic conformation of 8,2′-MA.

In 8,2′-cyclonucleosides with a five-membered cyclic ring such as 8,2′-MA, X-ray data indicate that the sugar puckerings are roughly grouped into two types of C(4′)-*exo*, C(3′)-*endo* ($_4T^3$) and C(4′)-*endo*, C(3′)-*exo* ($^4T_3$) conformations, although the C(4′)-*endo*, C(3′)-*exo* conformer (9 cases) is preferred to the C(4′)-*exo*, C(3′)-*endo* (1 case). Therefore, the sugar conformation may be assessed by making the assumption of C(4′)-*endo*, C(3′)-*exo*$\rightleftharpoons$C(4′)-*exo*, C(3′)-*endo* equilibrium. The calculated coupling constants in their two conformers are shown in Table VIII. The observed data fit best for 55% C(4′)-*endo*, C(3′)-*exo* and 45% C(4′)-*exo*, C(3′)-*endo* conformers. There is, however, a significant discrepancy between the calculated and observed coupling constants for the fragment H(1′)–C(1′)–C(2′)–H(2′). A similar observation has already been reported in 8,2′-SAMP (5′).[13] This is probably due to the strains in the bond angles around the C(1′) and C(2′) atoms of 8,2′-MA, as it was previously pointed out that the theoretically calculated coupling constants between protons attached to the carbon atoms with strains are smaller than those calculated from the modified Karplus equation, and that the modified Karplus relation can not be applied to the fragment around the carbon atoms with strains.[3e)] The conformation of the exocyclic bond in 8,2′-MA is estimated to be 45% *gauche*$^+$, with the other 55% from the $J_{4'5'}$ and $J_{4'5''}$ data by the usual method.[23]

Table IX. Observed and Calculated Coupling Constants (Hz) of 8,2′-EA

	J_{obs}	J_{calc}				
		X-Ray			Model	
Glycosidic bond						
		endo	*exo*		*endo* : *exo* = 50 : 50	
H2′-H6′	6.4	1.6	7.4		4.5	
H2′-H6′	6.4	11.3	0.2		5.8	
H6′-H7′	6.7	2.8	11.3		7.1	
H6′-H7″	6.1	11.3	2.8		7.1	
Sugar						
		$_1E$	$^0T_4$	$^3T_2$	$_1E:{}^0T_4$ 50 : 50	$_1E:{}^0T_4:{}^3T_2$ 40 : 40 : 20
H1′-H2′	6.1	4.7	8.4	6.7	6.6	6.6
H2′-H3′	5.5	0.2	4.3	11.2	2.3	4.0
H3′-H4′	5.5	2.7	8.2	10.1	5.5	6.3
Exocyclic bond						
		g^+	g^-/t		$g^+:g^-/t=55:45$	
(H4′-H5′)+(H4′-H5″)	8.2	4.0	13.4		8.2	

8,2′-EA is the first example of an 8,2′-cyclonucleoside with a six-membered cyclic ring determined by X-ray diffraction. Therefore, on consideration of the glycosidic and sugar conformations, we referred the data of pyrimidine 2′-cyclonucleosides with a six-membered cyclic ring. Their glycosidic bonds and sugar rings take either *endo* ($\chi \approx -90°$; 2 cases) or *exo* ($\chi \approx -70°$; 2 cases) form and either C(1′)-*exo* ($_1E$; 3 cases) or O(4′)-*endo*, C(4′)-*exo* ($^0T_4$; 1 case) conformation, respectively. If 8,2′-EA exists in their equilibrium, a 50% *exo* form and 50% C(1′)-*exo* conformation are estimated. However, the calculated J value (2.3 Hz) for the fragment H(2′)–C(2′)–C(3′)–H(3′) in the sugar moiety is significantly different from the observed one (5.5 Hz). This suggests that the contribution of the other sugar puckerings, for example C(3′)-*endo*, C(2′)-*exo* ($^3T_2$)[24] (Table IX) may be considerable, because the C(3′)-*endo*, C(2′)-*exo* conformation is assumed to exhibit a larger value (11.2 Hz) of $J_{2'3'}$. If a 20% C(3′)-*endo*, C(2′)-*exo* conformer exists, the observed value (5.5 Hz) is more consistent with the calculated value (4.0 Hz) than the value estimated from only the C(1′)-*exo* ⇋ O(4′)-*endo*, C(4′)-*exo* equilibrium (2.3 Hz) (Table IX). The conformation of the exocyclic bond in 8,2′-EA is estimated to be 55% *gauche*$^+$ and 45% others.

In total, 8,2′-MA and 8,2′-EA in solution take several sugar puckerings and exocyclic conformations, and 8,2′-EA adopts two kinds of conformations, even around the glycosidic bond.

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