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Studies of Nucleosides and Nucleotides. XLIII.[1] Purine Cyclonucleosides. (10). Optical Rotatory Dispersion and Circular Dichroism of Adenine 8-Cyclonucleosides

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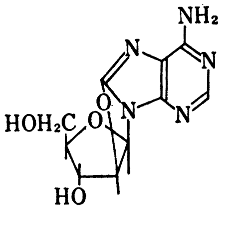
Optical rotatory dispersion and circular dichroism of 8-cyclonucleosides derived from adenosine were measured.

In the β-cyclonucleosides magnitude of rotatory strengths increased in the order 8,2'-<8,3'-<8,5'-cyclonucleoside both in the case of O- and S-cyclonucleosides. All of these cyclonucleosides had positive Cotton curve at absorption maxima.

In α-cyclonucleoside the sign of the Cotton effect inverted to negative and the magnitude is almost same with β-cyclonucleoside.

The synthesis and properties of various cyclonucleosides were reported.[3,4] Cyclonucleosides are unique in the respect of the structure, possessing anhydro linkages, which fix the furanose and the base moiety at certain angle. This situation may serve as a good model for the extreme conformation of ordinary nucleosides, in which the base is rather freely rotatable despite barriers caused by the interference of adjacent atoms.[5] Especially in the purine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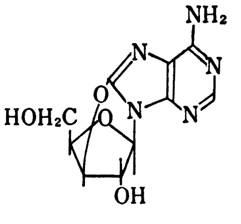
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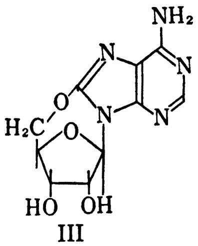
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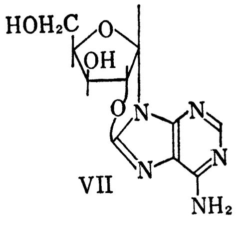
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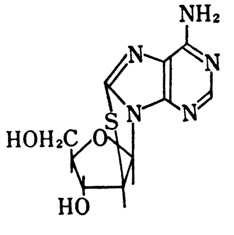
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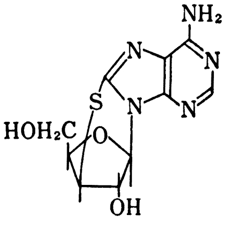
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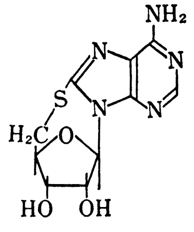
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[1] Part XLII: M. Ikehara and M. Kaneko, *Chem. Pharm. Bull.* (Tokyo), **18**, 2401 (1970).

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nucleosides, *syn* and *anti* conformation may be taken according to environmental conditions.[6-9] We have measured, therefore, optical rotatory dispersion and circular dichroism of various adenine 8-cyclonucleosides. In this paper we summarize the results and discuss about the relationship between rotational angle of the base and the magnitude of Cotton effect.

O-Cyclonucleosides

As shown in Fig. 1 and Table I, purine cyclonucleosides having O-anhydro linkages, 8,2′-anhydro-8-oxy-9-β-D-arabinofuranosyladenine[10] (8,2′-O-cycloadenosine) (I), 8,3′-anhydro-8-oxy-9-β-D-xylofuranosyladenine[4,11] (8,3′-O-cycloadenosine) (II) and 8,5′-anhydro-8-oxy-adenosine[12] (8,5′-O-cycloadenosine) (III), had ultraviolet (UV) absorption maxima around 260 nm in pH region 1—13. These absorption maxima could be assigned to B_{1u} and B_{2u} band as denoted by Clark and Tinoco.[13] Although position of the maximum is the almost same with that of 8-methoxyadenosine[14] (Table I), a slight bathochromic shift (0.5—3 nm) of the absorption maxima was observed in cyclonucleosides. There appeared shoulders at about 280 nm in acidic condition and it is the largest in 8,2′-O-cyclonucleoside (I). This shoulder may be ascribed to splitting of B band, which has been clearly shown in magnetic circular dichroism spectra of compound I.[15]

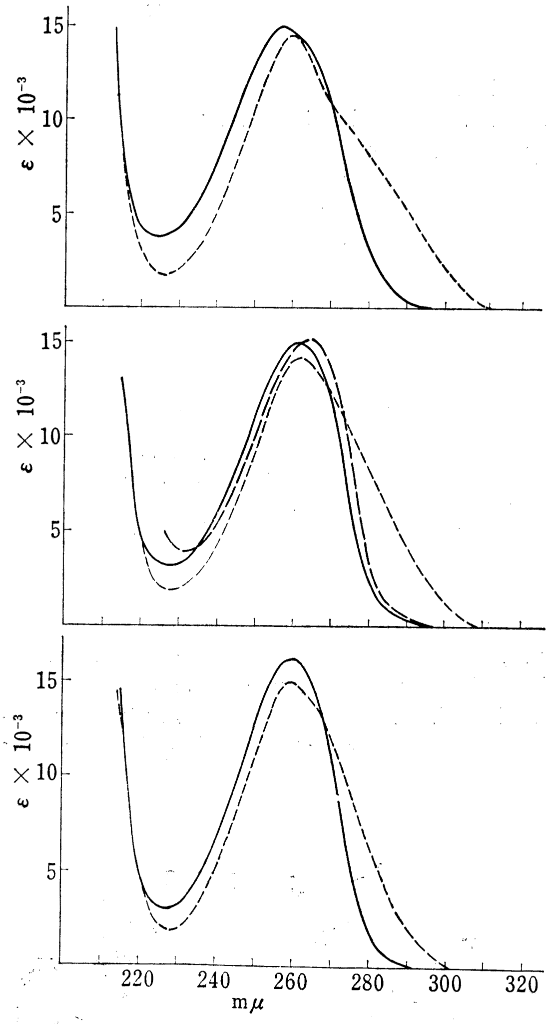

Fig. 1. Ultraviolet Absorption Spectra of O-Cycloadenosine

a. 8,2′-, b. 8,3′-, c. 8,5′-cyclonucleoside
------ taken at pH 2.0, ---- at pH 7.0, —— at pH 12.0

Optical rotatory dispersion (ORD) spectra of O-cyclonucleosides I—III were shown in Table II and Fig. 2. The curve of β-cyclonucleosides have a peak at 274—275 nm and a trough at 220—230 nm. The positive Cotton effect around 260 nm agreed with position

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TABLE I. Ultraviolet Absorption Properties of Adenine Cyclonucleosides and Related Compounds

Compound	λ_{max} (nm) (ε)		
	pH2	pH7	pH12
8,2′-O-Cycloadenosine (I)	260 (14600) 280 (sh) (8500)	257 (15000)	257 (14800)
8,3′-O-Cycloadenosine (II)	262 (14300) 280 (sh)	262 (14900)	265 (15200)
8,3′-O-Cyclo-2′-deoxyadenosine	262 (14200)	263 (14200)	263 (14600)
8,5′-O-Cycloadenosine (III)	260 (15000)	260.5 (16200)	261 (16400)
8,2′-O-α-Cycloadenosine	260 (16570)	257 (16800)	260 (16800)
8,2′-S-Cycloadenosine (IV)	277 (20100)	275.5 (20300)	276 (20300)
8,3′-S-Cycloadenosine (V)	283 (22000) 292 (sh) (21000)	282.5 (22000) 290 (sh) (15000) 276 (sh) (20000)	284.5 (22000) 278 (sh) (19200) 293 (sh) (16000)
8,5′-S-Cycloadenosine (VI)	284 (19500) 276 (sh) (17100) 294 (sh) (13900)	285.5 (17900) 278 (sh) (16400) 294 (sh) (12100)	285.5 (17800) 278 (sh) (16300) 295 (sh) (12100)
8-Methoxyadenosine	261	259	259
8-Methylmercaptoadenosine	281	280	279

of the main absorption band in B region. The sign and magnitude of the Cotton effect of cyclonucleosides are in contrast to natural nucleosides of purine, which have a small negative Cotton effect in neutral solution.[16] Amplitude of the Cotton effect is in the order of magnitude, 8,2′- $<$8,3′- $<$8,5′-cyclonucleoside and was 15800—19800, 28400—29000 and 47000, respectively. This tendency does not change in 8,3′-O-cyclonucleoside of 2′-deoxyadenosine, which had amplitude of 24600.[17] When we examine molecular models of 8,2′-(I), 8,3′- (II) and 8,5′-cyclonucleoside (III), the torsion angle[18] of the base is −108°, −76° and −38° (±20°),[19] respectively (see Fig. 3). One can therefore postulate that rotation of the base around glycosidic linkage from 8,2′ to 8.5′ direction causes increase in the magnitude of positive sign. The same proposition was made by Rogers and Ulbricht.[20] This change in the magnitude of the Cotton effect may be interpreted by a theory presented by Miles, *et al.*[21] According to them, change in magnitude of

TABLE II. Optical Rotatory Dispersion of Purine Cyclonucleosides

Compound	Peak (nm)	$[\phi]_{peak}$	Trough (nm)	$[\phi]_{trough}$	Amplitude ($\times 10^{-2}$)
8,2′-O-Cycloadenosine (I)	275	4300	220	−11500	158
8,3′-O-Cycloadenosine (II)	275	2900	220	−26200	291
8,3′-O-Cyclo-2′-deoxyadenonosine	275	3800	220	−24600	284
8,5′-O-Cycloadenosine (III)	274	19000	223	−28000	470
8,2′-S-Cycloadenosine (IV)	295	0	266 (sh)	−14900	149
			235	−24900	249
8,3′-S-Cycloadenosine (V)	295	11900	270 (sh)	−11900	238
			245	−18700	306
8,5′-S-Cycloadenosine (VI)	303	22400	260 (sh)	−56000	784
	225	93000	246	−82000	1044

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[17] M. Ikehara, M. Kaneko and Y. Nakahara, *Tetrahedron Letters*, **1968**, 4707.
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[21] D.W. Miles, S.J. Hahn, R.K. Robins, M.J. Robins and H. Eyring, *J. Phys. Ckem.*, **72**, 1483 (1968).

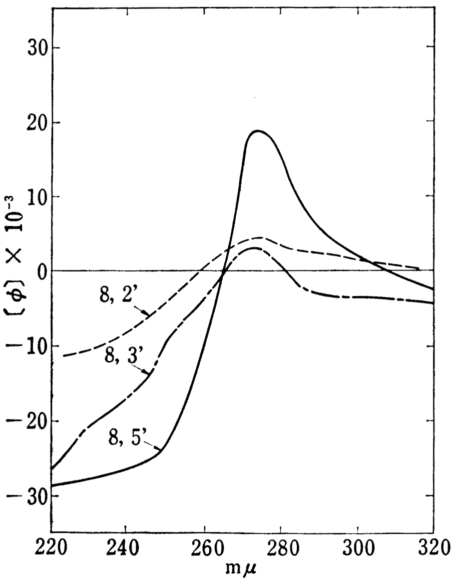

Fig. 2. Optical Rotatory Dispersion Spectratra of O-Cyclonucleoside

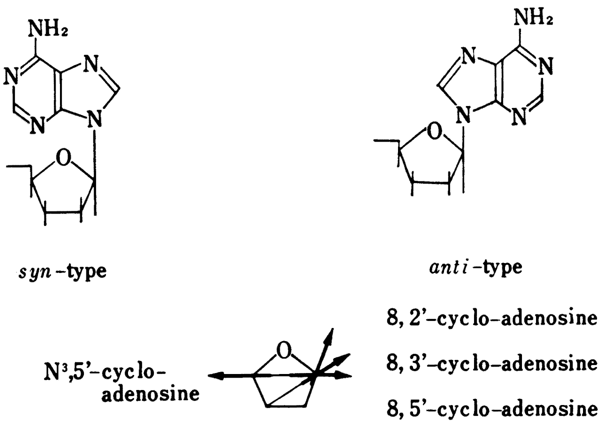

Fig. 3. Schematic Representation of Adenine Cyclonucleosides

the Cotton effect could be ascribed to variable interaction between transition moments of the sugar and the base moiety according to the relative angle of the components. Difference in conformation of the furanose moiety in ribo (2'-*exo*) and deoxyribo (2'-*endo*) nucleoside might be neglected by formation of anhydro bond, which attracted carbohydrate carbon atom to the endo conformation.[22] This point was proved by comparison of nuclear magnetic resonance (NMR) signals of H_1, of these cyclonucleosides (Table IV). Coupling constants, $J_{1'-2'}$ were zero in 8,3'-cyclonucleosides showing that H_1, and H_2' formed dihedral angle[23] of about

Table III. Circular Dichroism of Purine Cyclonucleosides

Compound	Peak(nm)	$[\theta]_{peak}$	Trough(nm)	$[\theta]_{trough}$
8,2'-O-Cycloadenosine (I)	246	8000	210	
	263 (sh)[a]	4000		
	222 (sh)	0		
8,3'-O-Cycloadenosine (II)	250	14000	210	
	260 (sh)	11000		
	233 (sh)	6000		
8,5'-O-Cycloadenosine (III)	255	32000	206	−38000
			229 (sh)	0
8,2'-S-Cycloadenosine (IV)	270	10000	225	−20000
	240 (sh)	2000		
8,3'-S-Cycloadenosine (V)	274	18000	229	−25000
	289 (sh)	8000	241 (sh)	−9000
	250 (sh)	3000		
8,5'-S-Cycloadenosine (VI)	280	34000	236	−85000
	213	34000	205	30000
	293 (sh)	20000		
	273 (sh)	31000		

a) shoulder

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22) M. Ikehara and H. Tada, *Chem. Pharm. Bull.* (Tokyo), **15**, 94 (1967).
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Table IV. NMR Spectra of Adenine Cyclonucleosides

Compound	2-H (ppm)	6-NH$_2$	1′-H	$J_{1'-2'}$(cps)
8,2′-O-Cycloadenosine (I)	8.04 (s)[a]	6.81 (s)	6.49 (d)[b]	5.4
8,3′-O-Cycloadenosine (II)	7.99 (s)	6.78 (s)	5.74 (s)	0
8,5′-O-Cycloadenosine (III)	8.10 (s)	7.04 (s)	6.05 (d)	0
8,2′-O-Cyclo-α-adenosine	8.04 (s)	6.80 (s)	6.52 (d)	5.0
8,2′-S-Cycloadenosine (IV)	8.09 (s)	7.08 (s)	6.53 (d)	6.6
8,3′-S-Cycloadenosine (V)	8.08 (s)	7.10 (s)	5.84 (s)	0
8,5′-S-Cycloadenosine (VI)	8.12 (s)	7.32 (s)	6.28 (s)	0

a) singlet *b*) doublet

90°, whereas 8,2′-cyclonucleosides showed $J_{1'-2'}$ 5—6 cps. This fact supports the 3′-endo conformation for 8,3′-cyclonucleosides.

Relationship of the torsion angle and the magnitude of the Cotton effect is much clearly shown in circular dichroism (CD) curves (Table III and Fig. 4). CD bands at 245—255 nm around major absorption region showed regular increase in positive sign from 8,2′- to 8,5′-cyclonucleoside, 8,3′-cyclonucleoside being in the middle. There appears a significant shift of the extremes of the Cotton band from 245 nm of 8,2′-(I), 250 nm of 8,3′-(II) to 255 nm of 8,5′-cyclonucleoside (III). This shift may be explained by assuming a weak splitting of the Cotton band in B region presumably due to a steric factor caused by the formation of five, six and seven-membered ring adjacent to the adenine moiety.

Thus far the amplitude of the Cotton effect around major absorption bands of O-cyclonucleosides was proved to increase in the order of 8,2′- < 8,3′- < 8,5′-cyclonucleoside and the sign of the Cotton effect is positive when nucleosidic configuration was β.

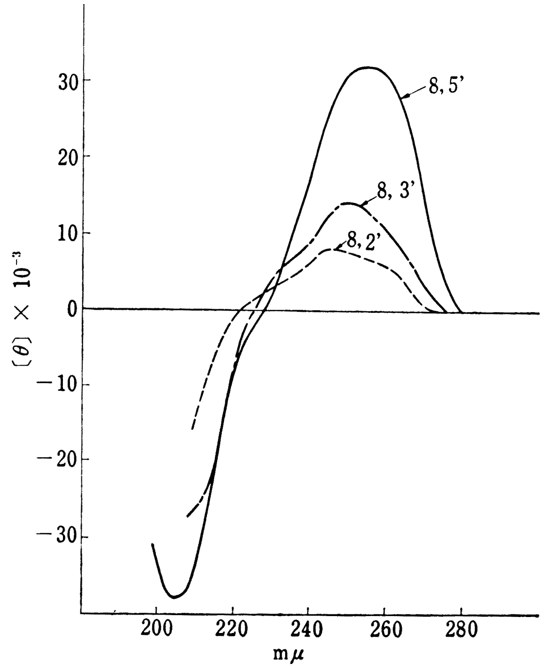

Fig. 4. Circular Dichroism Spectra of O-Cycloadenosine

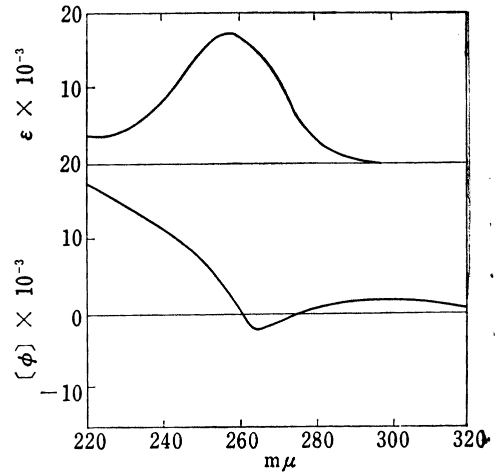

Fig. 5. Optical Rotatory Dispersion Spectra of 8,2′-O-Cyclo-α-adenosine

There exists only one example of α-8,2′-O-cyclonucleoside[17] (VII). Although this compound showed almost same UV absorption properties with those of β-8,2′-O-cyclonucleosides, the sign of the Cotton effect inverted to negative (Fig. 5). This is in agree with the fact that α-adenosine had positive Cotton effect in contrast to that of β-nucleosides.[24] Thus it was concluded that either in α- or β-nucleosides, the sign of the Cotton effect inverted from the original nucleoside by forming cyclonucleosides.

S-Cyclonucleosides

As shown in Fig. 6, UV absorption maxima of S-cyclonucleosides of adenine, 8,2′-anhydro-8-mercapto-9-β-D-arabinofuranosyladenine[22] (8,2′-S-cycloadenosine) (IV), 8,3′-anhydro-8-mercapto-9-β-D-xylofuranosyladenine[22] (8,3′-S-cycloadenosine) (V) and 8,5′-anhydro-8-mercaptoadenosine[25] (8,5′-S-cycloadenosine) (VI), appearred at the position *ca.* 20 nm shifted towards long wave length region from those of O-cyclonucleosides. The maximum of 8,2′-S-cyclonucleoside (IV) appeared at 275—277 nm and that of 8,3′-(V) and 8,5′-S-cyclonucleoside (VI) at 282.5—286 nm, which were in the similar position with B_{1u} and B_{2u} band of 8-methylmercaptoadenosine (279—281nm).[26] However, in 8,3′- and 8,5′-cyclonucleoside shoulders at 276—278 nm and at 290—296 nm appeared. The short wave length shoulder may be ascribed to *n*-*π*-transition and the shoulder at 290—296 nm may be ascribed to charge transfer band as described by Cheong, *et al.*[27] A slight bathochromic shift of UV maxima and that of shoulders may be due to the steric factor, which was caused by the formation of 5—7 membered ring fursed to C_8-N_9 of the adenine ring as in O-cyclonucleosides.

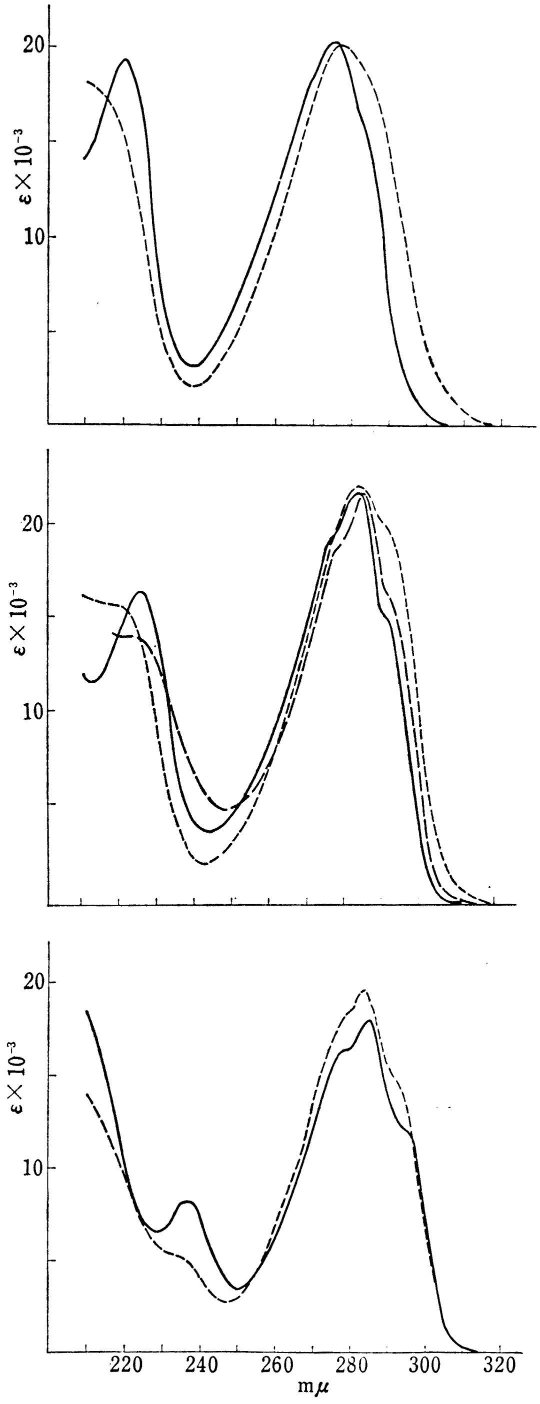

Fig. 6. Ultraviolet Absorption Spectra of S-Cycloadenosine

a. 8,2′-, b. 8,3′-, c. 8,5′-cycloadenosine
------ taken at pH 2.0, —— at pH 7.0, ---- at pH 12.0

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[24] T. Nishimura, B. Shimizu and I. Iwai, *Biochim. Biophys. Acta*, **157**, 221 (1968).

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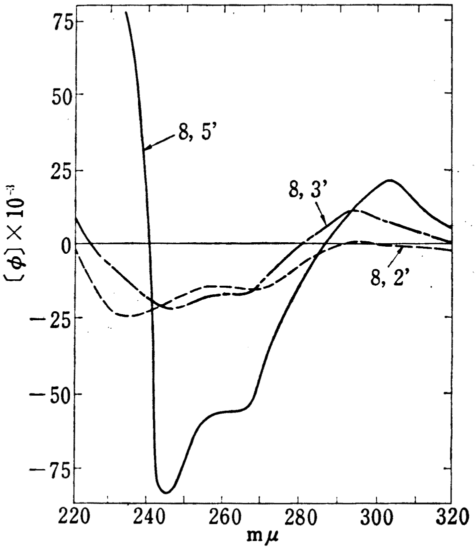

Fig. 7. Optical Rotatory Dispersion Spectra of S-Cycloadenosine

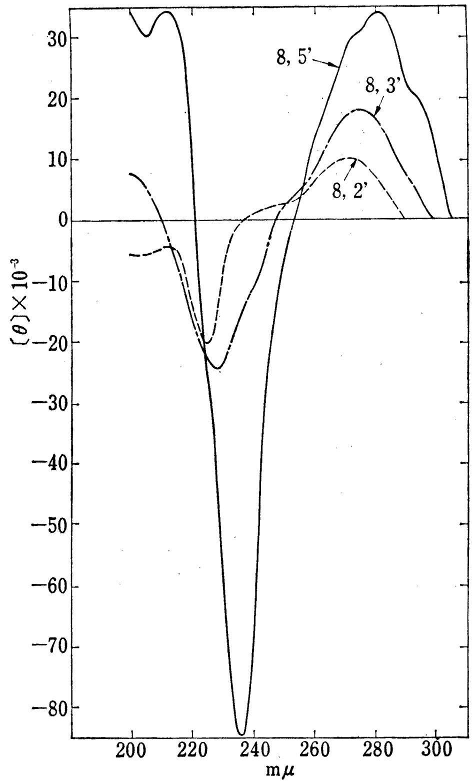

Fig. 8. Circular Dichroism Spectra of S-Cycloadenosine

ORD of S-cyclonucleosides (Fig. 7) showed a large peak at 295—303 nm, a trough at 235—246 nm and a shoulder at 260—270 nm. These extremes construct positive Cotton effect around 280 nm, which is consistent with the B region absorption band. This situation was much clearly shown in CD curves of S-cyclonucleosides (Fig. 8). Peaks at 272—282 nm showed a positive Cotton effect in B region and trough at 222—237 nm showed a negative one. In the case of 8,5′-cyclonucleoside (VI), one more peak at 216 nm was observed. These properties of Cotton effect both in ORD and CD showed a regular increase in the order 8,2′-<8,3′-< 8,5′-cyclonucleoside. This tendency is the same with that found in O-cyclonucleoside series. S-Cyclonucleosides showed a negative Cotton band at around 220—240 nm. Especially in 8,5′-cyclonucleoside, negative value of this band is very large. As the position of this band shifted bathochromically about 20 nm from that of O-cyclonucleosides, we tentatively assigned this band to E_{1u}. When we illustrate the position of the base on furanose ring schematically as shown in Fig. 3, 8,2′-, 8,3′- and 8,5′-cyclonucleoside may be represented by arrows, which were drawn as the head points pyrimidine portion and the tail directs imidazole portion of the adenine ring. Studies of X-ray crystallography[28] and examinations of molecular models showed that the base moiety in these cyclonucleosides was fixed at torsion angle of −108°, −76° and −68° for 8,2′-, 8,3′- and 8,5′-cyclonucleoside. For the latter compound (VI), the angle was thought to be around −38° (*exo*-type).[25] However X-ray crystallography showed an *endo*-type ($\phi_{CN}=-68°$) conformation.

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[28] K. Tomita, T. Fujiwara, T. Nishida and M. Ikehara, *Acta Crystallog.*, **A25**, 2178 (1969); *idem*, *Biochem. Biophys. Res. Commun.*, **41**, 1043 (1970).

Several investigators have shown[29-31] that N^3, 5'-cyclonucleoside derived from adenosine[32] had a negative Cotton effect and this compound must be represented by an arrow pointing to the left in Fig. 3. If we define the conformation of the cyclonucleoside in which the arrow is pointing to right as "*anti*" and that to left as "*syn*", 8-cyclonucleosides are in *anti* conformation and N^3,5'-cyclonucleoside is *syn* conformation. Therefore one can postulate that in purine β-cyclonucleoside, those having *anti* conformation show positive and those having *syn* conformation show negative Cotton effects. Furthermore, the magnitude of Cotton effect varies as the plane of the base rotates around nucleosidic linkage and the order of magnitude is 8,2'-$<$8.3'-$<$8,5'-cyclonucleoside both in O- and S-cyclonucleosides. Although example is limited to one case, α-8,2'-O-cyclonucleoside showed the magnitude of the Cotton effect in the same range with β-anomer and its sign was negative.

This kind of change in amplitude of the Cotton effect was also reported previously by Rogers and Ulbricht in pyrimidine cyclonucleosides.[33] In this case also torsion angle and the magnitude of Cotton effects were correlated. Thus it was hoped that these changes of the Cotton effect could be applied to the usual nucleosides in which base moieties were relatively freely rotatable around glycosidic linkage. Since 8-cyclonucleosides have 8-substitution, which causes a complex effect to ORD and CD curves and make it difficult to compare results found in cyclonucleosides directly to those of ordinary nucleosides. However, careful comparison of ORD and CD curves with those of 8-substituted nucleosides might useful for the determination of the position of the base moiety. The work along this line is in progress in our laboratory.

Experimental

Nucleosides——Nucleosides were synthesized according to the literature and checked for its purity by paper chromatography, UV absorption properties and elemental analysis.

UV Absorption Spectra——UV absorption spectra were taken with Hitachi EPS-3T spectrophotometer.

NMR Measurements——NMR spectra were taken with Hitachi H-6013 high resolution spectrometer operated at 60 mc with tetramethylsilane as internal standard.

ORD and CD Measurements——ORD and CD were taken with a JASCO ORD/UV-5 spectropolarimeter installed with CD attachment. Materials were dissolved in a concentration to obtain $OD_{max}=1.0$—2.0 in water. Light path was 10 mm. Before and after each run, zero lines were drawn for the proof of accuracy. Each curves were taken at least twice until a constant curve was obtained. Calibration of magnitude was made with d-10-camphorsulfonic acid as a standard. Temperature within cell holder was $15\pm2°$.

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31) Negative sign was found also in N^3,5'-cyclonucleoside derived from guanosine (unpublished results by K. Muneyama).

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