[Chem. Pharm. Bull. 24(4) 672—682 (1976)]

UDC 547.963.32.04 : 547.857.04

Studies of Nucleosides and Nucleotides. LXVIII.[1] Purine Cyclonucleosides. (29). Synthesis and Properties of O-Cyclonucleosides derived from Hypoxanthine, Mercaptopurine, Methylmercaptopurine and Purine

Morio Ikehara and Masako Muraoka

Faculty of Pharmaceutical Sciences, Osaka University and Department of Chemistry, Japan Womens University[2]

(Received June 30, 1975)

The O-cyclonucleosides of hypoxanthine, 6-mercaptopurine, 6-methylmercaptopurine, and purine were synthesized. Cyclonucleosides of hypoxanthine, and 6-mercaptopurine were synthesized i) by the derivatization of corresponding adenine cyclonucleosides and ii) by the cyclization of appropriate 2′- or 3′-TPS-derivatives. Cyclonucleosides of 6-methylmercaptopurine and purine were synthesized from the corresponding 6-mercaptopurine cyclonucleosides by methylation and Raney Nickel desulfurization. Ultraviolet spectrum (UV), nuclear magnetic resonance (NMR), circular dichroism (CD) and Mass spectra of these cyclonucleosides were measured. It was found in the CD spectrum that the purine 8,2′-cyclonucleoside had a larger magnitude of Cotton effect than its 8,3′-counterpart, in contrast to the order found in other cyclonucleosides.

In the course of our studies on purine cyclonucleosides[3] we mainly investigated adenine derivatives. Although some reports on other purine cyclonucleosides of guanine,[1,4–6] N^6-methyladenosine[7] and hypoxanthine[8,9] have appeared, knowledge about these cyclonucleosides is rather limited. For hypoxanthine cyclonucleosides especially, only S-cyclonucleosides were reported.[10] In this paper we describe the synthesis and properties of various O-cyclonucleosides of hypoxanthine 6-mercaptopurine, 6-methylmercaptopurine and purine.

Since we have synthesized the 8,2′-,[11,12] 8,3′-[12] and 8,5′-O-cyclonucleosides[13] of adenine, the corresponding inosine derivatives (II, XII and XXII) could be obtained by deamination reactions using sodium nitrite in acetic acid. Yields were 76, 42 and 67%, respectively. The ultraviolet (UV) absorption maximum of 8,2′-O-cycloinosine (II) was at 251—252.5 nm, that of the 8,3′-compound (XII) at 254—257 nm, and that of the 8,5′-compound (XXII) at 251—255 nm. Shoulders at longer wave lengths were observed for 8,2′- and 8,3′-cyclonucleosides. These UV absorption properties are somewhat different from those of adenine cyclonucleosides[14] which had λ max shifted from low to high wavelength in the order 8,2′-, 8,3′- and 8,5′-compound.

1) Part LXVII: M. Ikehara, T. Maruyama, and N. Watanabe, *J. Carbohyd. Nucleoside. Nucleotide*, in press.
2) Location: *133-1, Yamadakami, Suita and Bunkyo-ku, Tokyo.*
3) M. Ikehara, *Accounts Chem. Res.*, **2**, 47 (1969).
4) M. Ikehara and K. Muneyama, *J. Org. Chem.*, **34**, 3039 (1967).
5) M. Ikehara and K. Muneyama, *J. Org. Chem.*, **34**, 3042 (1967).
6) K.K. Ogilvie, L. Slotin, J.B. Westmore, and D. Lin, *Cand. J. Chem.*, **50**, 1100 (1972).
7) M. Ikehara and K. Morisawa, *Chem. Pharm. Bull.* (Tokyo), **19**, 2593 (1971).
8) M. Kaneko, M. Kimura, and B. Shimizu, *Chem. Pharm. Bull.* (Tokyo), **20**, 635 (1972).
9) A. Yamazaki, M. Akiyama, I. Kumashiro, and M. Ikehara, *Chem. Pharm. Bull.* (Tokyo), **21**, 1143 (1973).
10) M. Ikehara and M. Muraoka, *Chem. Pharm. Bull.* (Tokyo), **20**, 550 (1972).
11) M. Ikehara, H. Tada, and M. Kaneko, *Tetrahedron*, **24**, 2389 (1968).
12) M. Ikehara and M. Kaneko, *Chem. Pharm. Bull.* (Tokyo), **18**, 2401 (1970).
13) M. Ikehara, M. Kaneko, and R. Okano, *Tetrahedron*, **26**, 5675 (1970).
14) M. Ikehara, M. Kaneko, Y. Nakahara, S. Yamada, and S. Uesugi, *Chem. Pharm. Bull.* (Tokyo), **19**, 1381 (1971).

The mass spectra (Table I) of these hypoxanthine cyclonucleosides (II, XII and XXII) showed strong peaks of the molecular ion, which revealed the fairly stable nature of these compounds as previously observed for other cyclonucleosides.[15] The nuclear magnetic resonance (NMR) spectrum of 8,3′-O-cycloinosine (II) in d_6-DMSO showed a coupling constant $J_{H1'-2'}$ equal to 5Hz. The other two compounds (XII and XXII) showed no coupling of 1′ and 2′ protons. This feature is consistent with previous observations[16] and suggests that the assigned structures of these cyclonucleosides are correct. Circular dichroism (CD) spectra of the hypoxanthine cyclonucleosides (II, XII and XXII) are shown in Fig. 1. As expected from previous observations on adenine cyclonucleosides,[3,14] all these compounds had positive Cotton bands around their UV absorption maxima and their order of magnitude was 8,5′->8,3′->8,2′-cyclonucleosides. These physical properties, together with the following "*de novo*" synthesis, suggested that the structures of compounds (II, XII and XXII) were correct. In order to confirm the structure of cyclonucleoside (II) and (XII), chemical synthesis by way of 2,4,6-tri-isopropylbenzensulfonyl-8-bromoadenosines were performed. These pathways were also used to obtain N^6-modified purine cyclonucleosides as described later.

8-Bromo-2′-TPS-adenosine[17] (III) was treated with sodium nitrite in DMF to give 8-bromo-2′-TPS-inosine (IV) in a yield of 86%. The structure of IV was confirmed by elemental analysis and UV absorption properties. The compound (IV) was heated with sodium acetate in an acetic acid-acetic anhydride mixture at 150° for 3 hrs. The 8-oxy-compound (Va) was obtained as a light yellow powder having UV absorption maxima at 257 nm in neutral and 270 nm in alkaline media. The compound (Va) was then deacylated by treatment with methanolic ammonia and cyclized by heating with sodium acetate in dimethyl formamide (DMF). Appropriate work-up and purification by paper chromatography gave 8,2′-O-cyclo-inosine (II) in a yield of 69%. This sample was completely identical with that obtained in the deamination of 8,2′-O-cycloadenosine by the criteria of mp, paper chromatography and UV absorption properties. An alternative cyclization using methanolic ammonia gave a 48% yield. In order to obtain the 6-mercapto derivative (VII) an attempt was made to thiolate compound (Va) with phosphorus pentasulfide.[18] However, cleavage of the 8,2′-O-cyclo bond occurred prior to thiolation. The compound (Va) was then thiolated by heating in pyridine with phosphorus pentasulfide. The 6-thio compound (VI) was obtained as a yellow glass with the characteristic UV absorption maxima at 312—337 nm. When compound

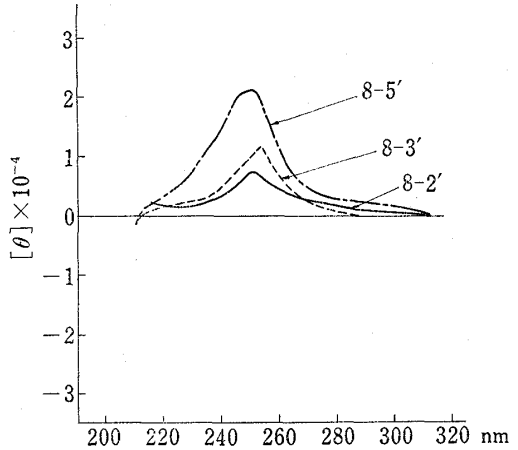

Fig. 1. CD Spectra of O-Cyclo-inosines

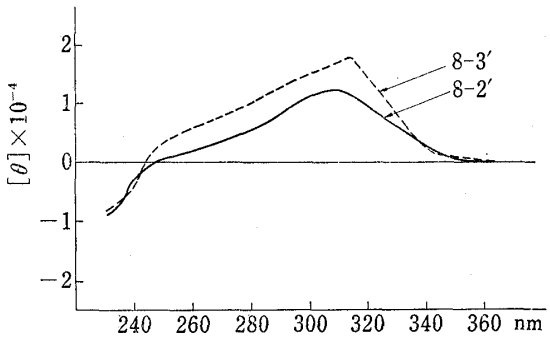

Fig. 2. CD Spectra of O-Cyclo-thioinosines

15) M. Ikeda, Y. Tamura, and M. Ikehara, *J. Heterocycl. Chem.*, **7**, 1377 (1970).
16) M. Ikehara and H. Tada, "The Purines-Theory and Experiment," ed. B. Pullman, The Israel Academy of Sciences and Humanities Jerusalem, 1972, p. 455.
17) M. Ikehara and M. Kaneko, *Tetrahedron*, **26**, 4251 (1970).
18) J.J. Fox, Wempen, A. Hampton, and I.L. Doerr, *J. Amer. Chem. Soc.*, **80**, 1669 (1958).

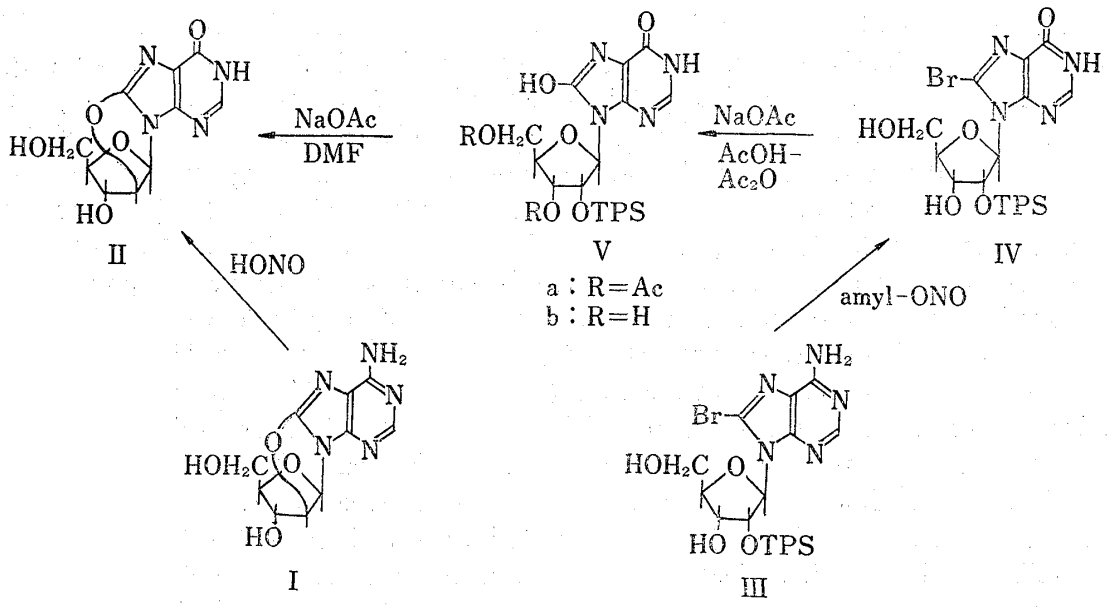

Chart 1

(VI) was cyclized by heating with sodium acetate in DMF or by methanolic ammonia, 8,2′-anhydro-6-mercapto-8-oxy-9-β-D-arabinofuranosylpurine (VII) was obtained in yields of 62 or 29%. The structure of VII was elucidated by its UV absorption properties and CD spectra (Fig. 2). The CD spectrum of VII had a [θ] peak at 310 nm which suggested the cyclonucleoside structure. Further confirmation of the structure was obtained by derivatizing VII to 6-methylmercaptopurine cyclonucleoside (VIII) with methyl iodide. The sample was identical with that obtained by the cyclization method described below. The crude compound (VI) was then methylated with methyl iodide in the presence of potassium

Table I. Principal Ions in Mass Spectra of Cyclonucleosides

Compound	M^+	M^+-31	M^+-48	M^+-59	M^+-77	M^+-89	M^+-101	a+1	a	a−1	b	c	d
8,2′-O-Inosine	266 (72.5)	235 (6.5)	218 (4.9)	207 (9.7)	189 (21)	177		153 (25.9)	152 (100)				
8,3′-O-Inosine	266 (31)			207 (27.5)	189 (6.9)	177 (12)	165 (58.6)	153 (37.9)	152 (100)				
8,5′-O-Inosine	266 (55.5)					177 (8.6)	165 (13.2)	153 (78)	152 (100)				
8,2′-O-Thioinosine	282 (10.7)		234 (50.0)			193 (10.7)	181 (10.7)	169 (14.0)	168 (92.8)	167 (14.0)	152 (75.0)	136 (53.5)	
8,3′-O-Thioinosine	282 (7.1)	251 (7.1)	234 (7.1)	223 (7.1)		193 (10.7)		169 (14.2)	168 (100)	167 (7.1)	152 (58.9)	136 (58.9)	
8,2′-O-CH_3S-Inosine	296 (61)	265 (6.5)	248 (4.8)	237 (6.5)	219 (4.8)	207 (12.8)	195 (6.5)	183 (3)	182 (95)	181 (100)			
8,3′-O-CH_3S-Inosine	296 (67.5)	265 (3.7)		237 (7.4)	219 (5.6)	207 (5.6)	195 (18.6)	183 (16.6)	182 (37)	181 (100)			
8,2′-O-Nebularine	250 (100)	219 (10.2)		191 (15.4)	173 (7.7)	161 (38.4)		137 (69.1)	136 (79.5)				120 (28.2)
8,3′-O-Nebularine	250 (100)	219 (14.9)		191 (29.6)	173 (14.9)	161 (26.0)		137 (55.5)	136 (44.5)				120 (33.3)

a : This ion corresponds to 6-substituted 8-oxypurine.
b : This ion corresponds to 8-oxyhypoxanthine.
c : This ion corresponds to 6-mercaptopurine.
d : This ion corresponds to purine.

Table II. NMR Signals of O-Cyclo Nucleosides

	2H	$6CH_3S$	1'H	2'H	3'H	4'H	5'H	2'OH	3'OH	5'OH
8,2'-O-Cyclo-inosine	7.91 s		6.79 d $J_{1'H-2'H}=5$	5.65 d	4.44 m $J_{3'H-4'H}=2.5$	4.07 6	3.18 m $J_{4'H-5'H}=6$		5.87 d $J_{3'H-3'OH}=5$	4.87 t
8,2'-O-Cyclo-methyl-thioinosine	8.56 s	2.61 s	6.50 d $J_{1'H-2'H}=6$	5.75 d	4.49 broad $J_{3'H-4'H}=2.5$	4.12 6	3.25 m $J_{4'H-5'H}=5$		5.92 d $J_{3'H-3'OH}=4$	4.83 t $J_{5'H-5'OH}=2.5$
8,3'-O-Cyclo-inosine	7.92 s		5.77 s	4.82 s	4.98 m $J_{3'H-4'H}=3$	4.49 6	3.54 m $J_{4'H-5'H}=6$	6.34 broad		
8,3'-O-Cyclo-methyl-thioinosine	8.58 s	2.62 s	5.94 s	4.89 d	5.13 d $J_{3'H-4'H}=3$	4.56 6	3.54 6 $J_{4'H-5'H}=6$	6.41 d $J_{2'H-2'OH}=4$		5.00 t $J_{5'H-5'OH}=6$
8,5'-O-Cyclo-inosine	8.02 s		5.93 s		3.90 m	4.70				

carbonate[19] and cyclized with sodium acetate in DMF. When compound IX was heated in DMF at 120° for 1.5 hr as in the case of adenosine[12] only the 8-oxy-arabino compound was obtained. The reaction conditions were then moderated to 40° for 30 min. In this case a compound (VIII) having a UV absorption maximum at 293—295 nm was obtained by thin-layer chromatography (TLC) purification. The elemental analysis showed that cyclization had occurred and *Rf* values in paper chromatography also suggested the cyclonucleoside structure. The mass spectrum (Table I) showed a molecular ion peak at *m/e* 296 together with peaks corresponding to 6-methylmercapto-8-oxypurine. The NMR spectrum (Table. II) showed signals of H-2 at 8.56, C_1'–H at 6.50 and C_2'–H at 5.75 δ with a coupling constant $J_{1'-2'}$ equal to 6 Hz. The CD spectrum (Fig. 3) showed a peak at 293 nm and a trough at 230 nm. These features are consistent with those expected for compound (VIII) having the 6-methylmercaptopurine-8,2'-O-cyclonucleoside structure.

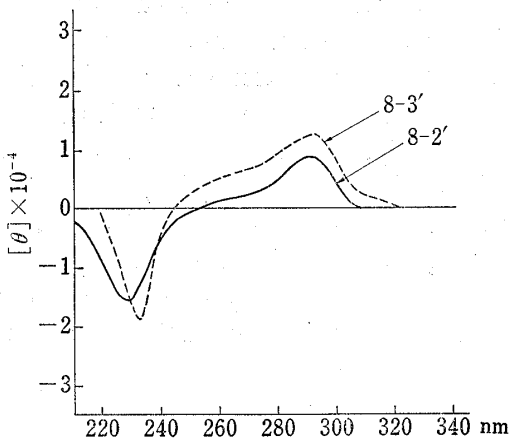

Fig. 3. CD Spectra of O-Cyclo-methyl-thioinosines

When compound (VIII) was desulfurized with Raney nickel, 8,2'-O-cyclo-8-oxynebularine (X) was obtained. UV absorption properties and mass spectra (M^+ 250) suggested that the structure was correct. The CD spectrum of compound X was compared with that of its 8,3'-counterpart (XX) described later. As shown in Fig. 4, the spectrum of X had a peak at 270 nm and a trough at 235 nm, which are analogous to other cyclonucleosides. However, the magnitude of the Cotton effect was smaller than that of its 8,3'-counterpart. This is the first case in which an 8,3'-cyclonucleoside has had a larger $[\theta]$ value than the 8,2'-cyclonucleoside and it suggests that the N^6-substituent in purine bases has a large effect on the magnitude of the Cotton effect which is also variable with ϕ_{CN}[20] the angle of the base and the furanose ring.

19) J.A. Montgomery, T.P. Johnston, A. Gallagher, C.R. Stringfellow, Jr. and F.M. Schabel, Jr., *J. Med. Pharm. Chem.*, 3, 265 (1961).
20) J. Donohue and K.N. Trueblood, *J. Mol. Biol.*, 2, 363 (1960).

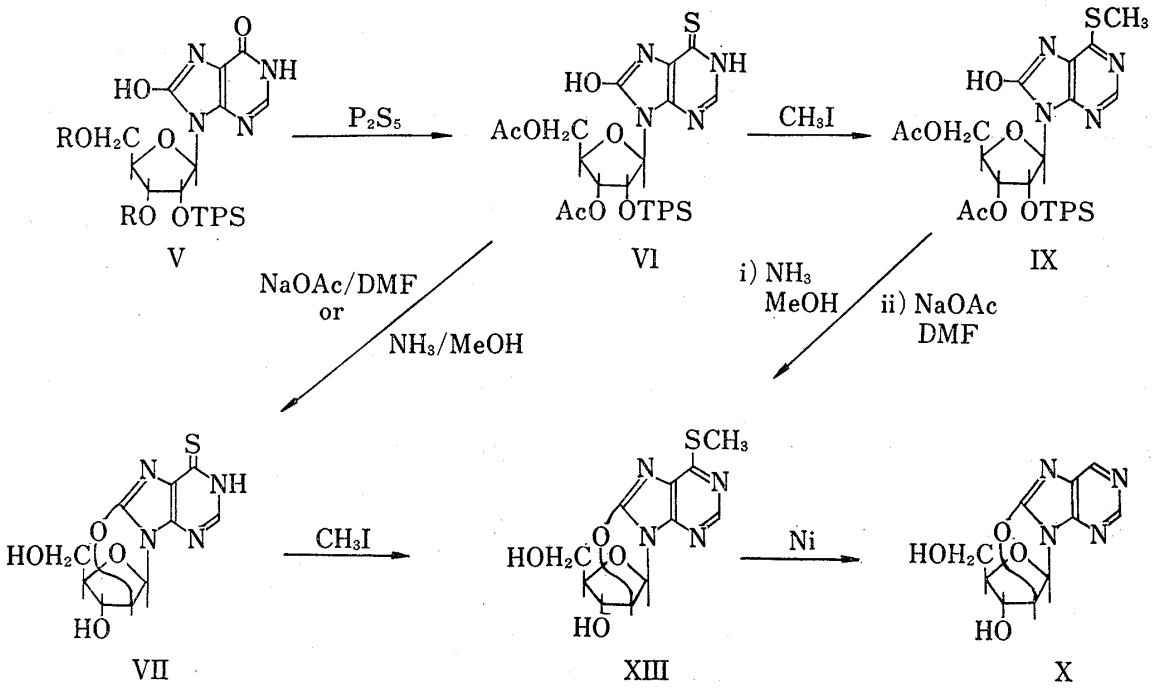

Chart 2

The series of reactions leading to 8,3′-O-cycloinosine (XII) from 3′-TPS-8-bromoadenosine (XIII) was conducted almost analogously to that leading to the 8,2′-compound. Compound (XIII) was deaminated with amyl nitrite to the inosine derivative (XIV), which was converted to the 8-oxy compound (XV) with sodium acetate. Yields in the two steps were 67 and 71%, respectively. Cyclization of XV was performed by heating with sodium acetate in DMF and 8,3′-O-cycloinosine (XII) was obtained along with 8-oxy-9-β-D-arabinofuranosylhypoxanthine, which may be formed by cleavage of XV by attack of the acetate ion. The sample of XII, thus obtained, was compared with that obtained above. They were identical in paper chromatography and UV absorption properties. In order to synthesize 6-mercaptopurine cyclonucleoside (XVII), the compound (XV) was thiolated at the N^6-position with

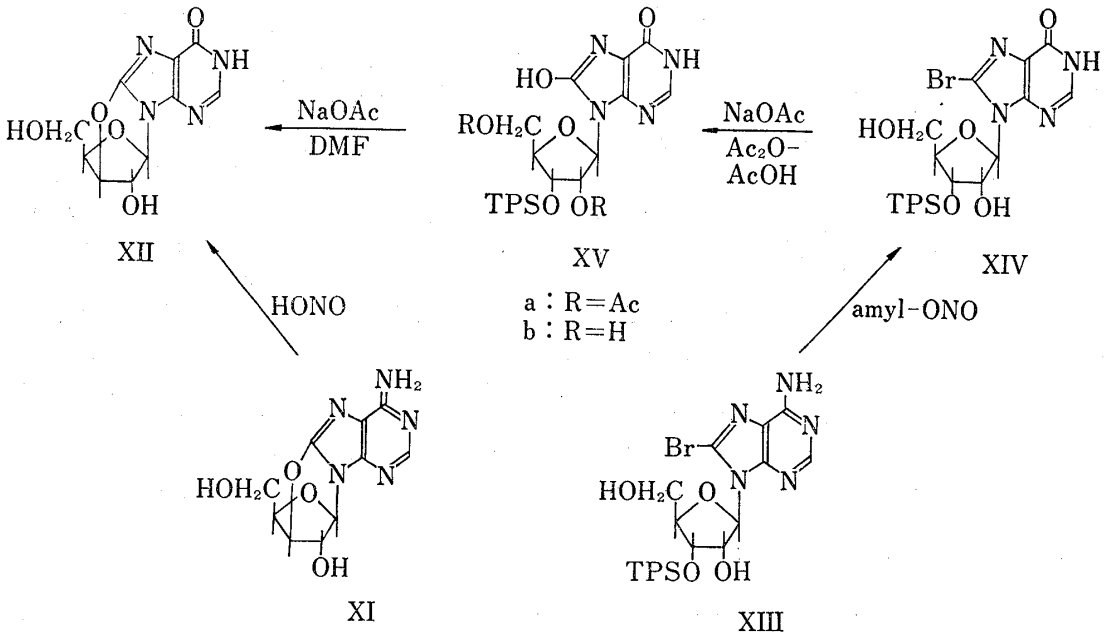

Chart 3

P_2S_5 to give XVI. Compound (XVI) was then cyclized by heating with sodium acetate in DMF to give the 8,3′-O-cyclonucleoside of 6-mercaptopurine (XVII) in a yield of 49%. UV absorption properties resembling those of the 8,2′-counterpart (VII) and a CD spectrum (Fig. 2) having $[\theta]$ peak at 135 nm suggested the structure of XVII to be correct. Methylation of the compound XVII with methyl iodide gave a 6-methylmercapto cyclonucleoside (XVIII), which was alternatively synthesized as described below. The compound XVI was methylated with methyl iodide and cyclized with sodium acetate as described in the case of the 8,2′-compound. The cyclonucleoside (XVIII) showed a UV absorption maximum at around 295 nm and a molecular ion peak at *m/e* 296 in its mass spectrum. The NMR signal of H-1′ appeared as a singlet peak and that of H-3′ at 5.13 δ suggesting that the 8,3′-cyclonucleoside structure was correct. The CD spectrum (Fig. 3) also showed a positive band at 294 nm and a trough at 233 nm having $[\theta]$ values larger than those of its 8,2′-counterpart. The 8,3′-O-cyclonucleoside structure of XVIII was thus confirmed. The Raney-nickel desulfurization of compound (XVIII) gave 8,3′-O-cyclo-8-oxynebularine (XX) and this structure was confirmed by UV absorption properties (ε max at 271—276 nm), mass peak at *m/e* 250 for the molecular ion, and mobility in paper chromatography in several solvent systems. As described before, the CD spectra of compound XX had a smaller Cotton band than that of 8,2′-O-cyclonebularine (X) (Fig. 4).

Finally the 8,5′-O-cyclonucleoside (XXII) of hypoxanthine was synthesized by two routes. 8,5′-O-Cyclonucleoside (XXI) was deaminated with sodium nitrite in acetic acid to give compound (XXII) in a yield of 67%. The structure of XXII was confirmed by elemental analysis and other physical properties. The UV absorption spectra showed λ max at 251—255 suggesting an inosine derivative and mass spectrum gave a molecular ion peak at *m/e* 266. A singlet signal of H-1′ in NMR was also consistent with 8,5′-O-cyclonucleoside structure. In the spectrum (Fig. 1) a peak at 250 nm, which had the largest $[\theta]$ of the three hypoxanthine cyclonucleosides, suggested the structure to be correct. An alternative synthetic pathway of XXII starting from 8-bromoinosine (XXIII) also confirmed this structure. In this case 8-bromoinosine[21] was protected on the 2′,3′-hydroxyls with the ethoxymethylidene group

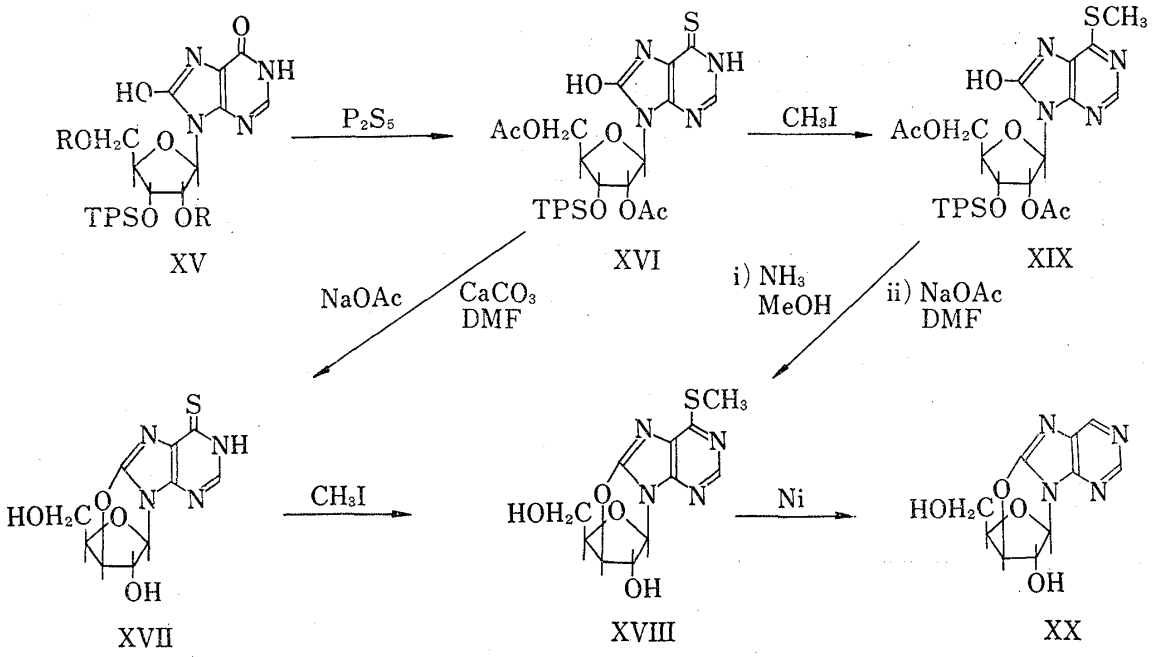

Chart 4

21) R.E. Holmes and R.K. Robins, *J. Amer. Chem. Soc.*, **86**, 1242 (1964).

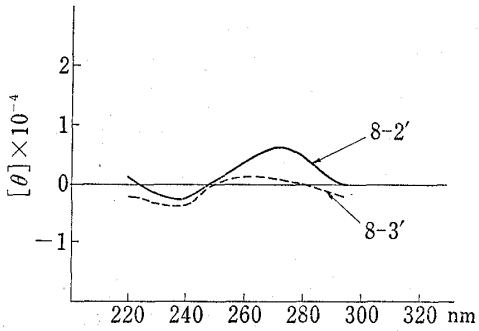

Fig. 4. CD Spectra of O-Cyclonebularines

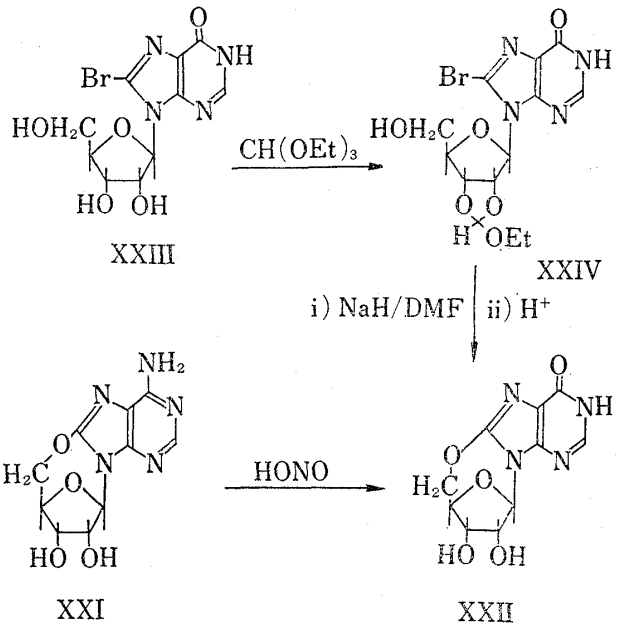

Chart 5

to give compound XXIV. When XXIV was treated at room temperature with excess sodium hydride in DMF then with acetic acid in ethanol, 8,5′-O-cyclo-8-oxyinosine (XXII) was obtained. This sample proved to be identical with that obtained by the deamination of 8,5′-O-cycloadenosine as described above. Several attempts to obtain 6-mercapto compounds failed presumably because of the labile nature of the 8,5′-O-cycloinosine. Thus, various O-cyclonucleosides of hypoxanthine, 6-mercaptopurine, 6-methylmercaptopurine and purine were synthesized and their physical properties were elucidated. Reactions of these cyclonucleosides will be reported in subsequent papers.

Experimental[22]

8-Bromo-2′-O-TPS-inosine (IV)——8-Bromo-2′-O-TPS-adenosine (III) (8.015 g, 13 mmoles) was dissolved in DMF (130 ml) and 2N HOAc (16.5 ml). The solution was stirred at room temperature and isoamyl nitrite (42 ml) was added. The stirring was continued for 6 hr and the disappearance of the starting material was confirmed by TLC ($CHCl_3$–EtOH, 19: 1, *Rf* 0.51). Ethanol (*ca.* 50 ml) was added to the reaction mixture which was evaporated *in vacuo* to *ca.* 130 ml. The solution was added dropwise to H_2O (1: 1) containing 13 g of $NaHCO_3$ with stirring. After 1 hr's stirring precipitates were collected by filtration, washed with H_2O, and dried. Yield was 6.87 g (85.6%). For elemental analysis a sample was recrystallized from benzene twice, mp 170—182°. *Anal.* Calcd. for $C_{25}H_{33}O_7N_4SBr$: 48.94; H, 5.42; N, 9.13. Found: C, 48.96; H, 5.35; N, 9.47. UV: λ_{max}^{MeOH} nm (ε) 238 (16000), 257 (sh, 11900), 280 (sh, 7200); $\lambda_{max}^{0.1NHCl}$ 239 (16200), 257 (sh, 12500), 280 (sh, 7400); $\lambda_{max}^{0.1NNaOH}$ 237 (14300), 262.5 (12100) 277 (sh, 11000). Paper chromatography: *Rf* (B) 0.92, *Rf* (C) 0.91.

8-Oxy-2′-O-TPS-3′,5′-di-O-acetylinosine (Va)——8-Br-TPS-inosine (3.05 g, 5 mmoles) was dissolved in acetic acid (50 ml) and acetic anhydride (50 ml). To the solution sodium acetate (6.0 g, 70 mmoles) was added and it was heated at 160° for 3 hrs with exclusion of moisture. After disappearance of the starting material was confirmed by TLC ($CHCl_3$–EtOH, 13: 3, *Rf* 0.28), the solution was evaporated *in vacuo*. Ethanol (50 ml) was added to the solution, which was kept at room temperature overnight. The solution was evaporated repeatedly with added ethanol. The residue was dissolved in $CHCl_3$, washed with water, dried

22) UV absorption spectra were taken with a Hitachi EPS-3T or 124 spectrophotometer, NMR spectra were taken with a Hitachi R-22 spectrometer operated at 90 mHz with tetramethylsilane as external standard. CD spectra were measured with a JASCO ORD/UV-5 spectropolarimeter in 10 mm path-length cells using aqueous solutions of 1.5 OD unit of nucleosides. Paper chromatography was performed on Toyo filter paper No. 51A unless otherwise noted in the following solvent systems: A, H_2O adjusted to pH 10 with NH_3; B, *n*-BuOH–H_2O (86: 14); C, iso-PrOH–conc·NH_3–H_2O (7: 1: 2). TLC was performed on Kieselgel unless otherwise noted.

over Na_2SO_4, and $CHCl_3$ was evaporated. 8-Oxy-TPS-diacetylinosine was obtained as a pale yellow powder in a yield of 2.84 g (89.5%). UV: $\lambda_{max}^{50\%EtOH}$ (nm) 238,258; $\lambda_{max}^{0.1NHCl}$ 238,257; $\lambda_{max}^{0.1NNaOH}$ 237,270: Paper chromatography: *Rf* (B) 0.89; *Rf* (C) 0.86.

8,2′-Anhydro-8-oxy-9-β-D-arabinofuranosylhypoxanthine (II)——i) 8,2′-O-Cycloadenosine (265 mg, 1 mmole) was dissolved in 2N HOAc (20 ml). To the solution sodium nitrite (345 mg, 5 mmoles) dissolved in H_2O (5 ml) was added. The reaction mixture was stirred at room temperature for 24 hr and completion of the reaction was examined with TLC on Avicel. The solvent was evaporated repeatedly with added EtOH until no odour of AcOH remained. The residue was recrystallized from water to give colourless plates, mp 265—273 (decomp), in a yield of 200 mg (76%). *Anal.* Calcd. for $C_{10}H_{10}O_5N_4$: C, 45.11; H, 3.79; N, 21.05; Found: C, 45.35; H, 3.83; N, 21.16. UV: $\lambda_{max}^{pH7.0}$ nm (ε) 251 (12900) 286 (sh, 3900); $\lambda_{max}^{0.1NHCl}$ 251 (13000), 286 (3900); $\lambda_{max}^{0.1NNaOH}$ 252.5 (13000). Mass spectrum peaks are as in Table I. NMR signals are summarized in Table II. CD: [θ] peak at 251 nm was 7500 and spectrum is shown in Fig. 1. Paper chromatography *Rf* (A) 0.81 *Rf* (B) 0.41 *Rf* (C) 0.52.

ii) 8-Oxy-2′-TPS-diacetylinosine (Va) (2.5 g) was dissolved in anhydrous MeOH (100 ml), which was saturated with NH_3 gas at 0°. The tightly stoppered flask was kept at room temperature for 3 days. MeOH was evaporated and 8-OH-TPS-inosine (Vb) was obtained as a yellow powder. This material (275 mg) was dissolved in DMF (20 ml) and anhydrous NaOAc (600 mg) was added. The mixture was heated at 100° for 30 min with stirring. After the reaction extent was examined by TLC, the solvent was evaporated *in vacuo*. The residue was applied to Whatman 3MM paper and developed in solvent C. The nucleoside migrating at *Rf* 0.40 was extracted with H_2O. Recrystallization of the material from water gave 8,2′-O-cycloinosine in a yield of 92 mg (69%). *Anal.* Calcd. for $C_{10}H_{10}O_5N_4$: C, 45.11; H, 3.79; N, 21.05. Found: C, 45.08; H, 3.72; N, 21.08. This sample was identical with that obtained above by criteria of mp, paper chromatography and UV absorption properties.

iii) 8-Oxy-TPS-diacetylinosine (Va) (317 mg) was dissolved in anhydrous MeOH (50 ml) and saturated with dry NH_3 at 0°. The solution was heated at 80° for 8.5 hr in a steel tube. MeOH was evaporated and the residue was purified as in ii). Yield was 64 mg (47.5%).

8,2′-Anhydro-6-mercapto-8-oxy-9-β-D-arabinofuranosylpurine (VII)——i) The crude 8-oxy-6-mercapto-2′-TPS-diacetylpurine riboside (VIa) (400 mg) was dissolved in anhydrous MeOH (50 ml), which was saturated with dry NH_3 at 0°. After 48 hr at room temperature, the reaction extent was examined by TLC ($CHCl_3$–EtOH, 18: 2, starting material *Rf* 0.46). MeOH was evaporated, and a yellowish residue was obtained. UV: $\lambda_{max}^{50\%EtOH}$ (nm) 237, 317; $\lambda_{max}^{0.1NHCl}$ 235, 340; $\lambda_{max}^{0.1NNaOH}$ 240 (sh), 322.5. The crude material (374 mg, 0.66 mmole), thus obtained, was dissolved in DMF (27 ml) and freshly prepared anhyd. NaOAc (758 mg) was added. The mixture was refluxed for 2 hr with exclusion of moisture. After cooling, salts were filtered off, DMF was evaporated *in vacuo*, and the residue was applied to Whatman 3MM paper. Development with solvent C gave a band migrating at *Rf* 0.44. Elution of this band with H_2O gave 115.3 mg (61.8%) of 6-mercaptopurine cyclonucleoside. UV: $\lambda_{max}^{pH7.0}$ (nm) 236,310; $\lambda_{max}^{0.1NHCl}$ 236 (sh), 296 (sh), 327.5; $\lambda_{max}^{0.1NNaOH}$ 236,310. CD: [θ] peak at 310 nm=12200. (see Fig. 2). Mass spectral data are summarized in Table I. PPC: *Rf* (A) 0.87, *Rf* (B) 0.22, *Rf* (C) 0.44.

ii) Well dried 8-oxy-6-mercapto-diacetyl-TPS compound (VIa) (206 mg) was dissolved in MeOH (30 ml) and saturated with NH_3 gas at 0°. The mixture was heated at 80° for 8 hr in a steel tube. MeOH was evaporated and the residue was purified as in the case of i). Yield was 25.8 mg (28.5%).

8,2′-Anhydro-6-methylmercapto-8-oxy-9-β-D-arabinofuranosylpurine (VIII)——i) 8-Oxy-TPS-diacetylinosine (Va) (0.825 g, 1.3 mmole) was dissolved in pyridine (30 ml). To the solution P_2S_5 (1.154 g, 5.2 mmoles) and H_2O (0.0936 ml, 5.2 mmoles) were added. The mixture was heated at 135° for 8 hr. The reaction extent was examined by TLC ($CHCl_3$–EtOH, 19: 1; Va *Rf* 0.38) and solvents evaporated *in vacuo*. To the residue H_2O (40 ml) was added and boiled for 5 min. After cooling the supernatant was decanted and the residual material was taken up in $CHCl_3$. The $CHCl_3$ solution was washed twice with H_2O and dried over Na_2SO_4 with addition of mercaptoethanol (1 drop). The solvent was evaporated and the residue was applied to a thin-layer plate, which was developed in $CHCl_3$–EtOH (19: 1). The band migrating at *Rf* 0.38 was eluted with MeOH (50 ml). The 6-Mercaptopurine derivative (VI) was obtained in a yield of 0.493 g (60.2%) as a yellow glass. UV: $\lambda_{max}^{50\%EtOH}$ nm 236,288 (sh), 312,340 (sh); $\lambda_{max}^{0.1NHCl}$ 236, 288 (sh), 337; $\lambda_{max}^{0.1NNaOH}$ 238,288 (sh), 319. Paper chromatography *Rf* (B) 0.95.

The crude 6-mercapto compound (VI) (0.455 g, 0.7 mmole) was dissolved in DMF (6 ml) and anhydrous K_2CO_3 (0.1 g, 0.73 mmole) was added. Methyl iodide (0.043 ml, 0.73 mmole) was added dropwise to the solution with stirring. After 1 hr stirring at room temperature, loss of the starting material was examined by TLC ($CHCl_3$–EtOH, 8: 2, *Rf* 0.83). The solvent was evaporated to afford compound (IX) as a glass. UV $\lambda_{max}^{50\%EtOH}$ nm 229, 299; $\lambda_{max}^{0.1NHCl}$ 229, 299; $\lambda_{max}^{0.1NNaOH}$ 290 (sh), 311.5.

This material was well dried over P_2O_5 *in vacuo* and dissolved in anhydrous methanolic ammonia (50 ml). After keeping the solution at room temperature for 3 days, the reaction extent was examined by TLC ($CHCl_3$–EtOH, 18: 2, IX, *Rf* 0.60). Evaporation of this mixture gave a residue having UV absorption properties resembling those of IX. The crude deacetylated material was dissolved in DMF (6 ml) and anhydrous NaOAc (0.861 g, 1.05 mmole) was added. The mixture was evaporated *in vacuo* and the residue was dissolved in a small amount of MeOH. Application of the residue to a preparative TLC plate and development with

$CHCl_3$–EtOH (18: 2) gave a band migrating at *Rf* 0.24. The band was extracted with MeOH (70 ml) and the 6-methylmercaptocyclonucleoside (VIII) was obtained in a yield of 0.116 g (30%). A sample was recrystallized twice from MeOH to give colourless needles, mp 183—186°. *Anal.* Calcd. for $C_{11}H_{12}O_4N_4S$: C, 44.60; H, 4.08; N, 18.91. Found: C, 45.06; H, 4.26; N, 18.10. UV: $\lambda_{max}^{pH7.0}$ nm (ε) 228 (12100), 293 (20500); λ_{max}^{pH1} 228 (12100); 293 (19200), 318 (sh, 1100); λ_{max}^{pH13} 227 (13600), 295 (17200), 315 (sh, 5600). Mass and NMR spectral data are summarized in Table I and II. CD: [θ] peak at 293 nm=8800, [θ] trough at 230 nm= −16,000 and spectrum is recorded in Fig. 3. Paper chromatography *Rf* (A) 0.74 *Rf* (B) 0.67 *Rf* (C) 0.61.

ii) The crude 8,2′-O-cyclonucleoside (VII) (96 mg, 0.34 mmole) was dissolved in DMF (3 ml). To the solution, anhyd. K_2CO_2 (50.3 mg) and CH_3I (0.0217 ml, 0.36 mmole) were added with stirring at 0°. The reaction mixture was stirred for 1 hr at 0° and applied to a preparative TLC. The band migrating at *Rf* 0.40 in the solvent $CHCl_3$–EtOH (17: 3) was eluted with MeOH. 8,2′-O-6-Methylmercapto-cyclonucleoside was obtained in a yield of 45.3 mg (45.2%). This material was identical with the sample obtained in i) by the criteria of *Rf*'s in PPC and UV absorption properties.

8,2′-Anhydro-8-oxynebularine (IX)——8,2′-O-Methylmercaptopurine cyclonucleoside (VIII) (80 mg) was dissolved in MeOH (20 ml) and freshly prepared Raney nickel (*ca.* 1 ml) was added. After heating of the mixture at reflux temperature for 1 hr, the supernatant was applied to Whatman 3MM paper and developed in solvent B. Neburaline cyclonucleoside (X) was obtained from the band migrating at *Rf* (B) 0.39. UV: $\lambda_{max}^{H_2O}$ nm 240, 270; $\lambda_{max}^{0.1NHCl}$ 274.5; $\lambda_{max}^{0.1NNaOH}$ 240 (sh) 271. Mass Spectrum: *m/e* 250 (M^+), 136 (oxypurine), 120(purine). (see Table I). CD: [θ] peak at 270 nm (6200), [θ]trough at 235 nm (−2500) calculated on the basis of ε=9330 for 8-ethoxy-9-methylpurine. (see Fig. 4). Paper chromatography *Rf* (A) 0.74 *Rf* (C) 0.64.

8-Bromo-3′-TPS-inosine (XIV)——8-Bromo-3′-TPS-adenosine (XIII) (3.06 g, 5 mmoles) was dissolved in DMF (50 ml). To the solution 2N acetic acid (8.3 ml) and isoamyl nitrite (20 ml, 15 mmoles) were added with stirring. After keeping the mixture for 2 days at room temperature, the reaction extent was examined by TLC ($CHCl_3$–EtOH, 19: 1, *Rf* 0.11). The mixture was evaporated to *ca.* 20 ml *in vacuo* and poured in water (500 ml) containing $NaHCO_3$ (5 g). Continuing the stirring for 30 min gave pale yellow precipitates, which were collected by filtration. After a water wash, the precipitates were recrystallized from 50% EtOH to give pale yellow plates, mp 170—171°. Yield was 2.272 g (68.6%). *Anal.* Calcd. for $C_{25}H_{33}O_7N_4SBr$: C, 48.94; H, 5.42; N, 9.13. Found: 48.72; H, 5.59; N, 9.11. UV: nm (ε) λ_{max}^{MeOH} 236.5 (16,800), 255 (sh, 13,100), 277 (8000), 280 (sh, 6600); $\lambda_{max}^{0.1NHCl}$ 236.5 (19700), 255 (sh, 15800), 275 (sh, 10000), 280 (sh, 7600); $\lambda_{max}^{0.1NNaOH}$ 233 (sh, 14500), 263 (12900), 276 (sh, 11500). Paper chromatography *Rf* (B) 0.92; *Rf* (C) 0.95.

8-Oxy-3′-TPS-inosine (XV)——Freshly prepared anhyd. NaOAc (447 mg) was dissolved in acetic acid (7.5 ml) with stirring and exclusion of moisture. 8-Bromo-3′-TPS-inosine (453 mg, 0.74 mmole) was added and the mixture was heated at 170° for 2 hr. After examination by TLC ($CHCl_3$–EtOH, 19: 1) the mixture was evaporated *in vacuo* to *ca.* 2 ml and poured in water (30 ml) with stirring. The stirring was continued for 30 min and precipitates were collected by filtration. After thorough washing with cold water the precipitates were dried to give a solid material (yield 365 mg). The solid was dissolved in $CHCl_3$ (50 ml), dried over Na_2SO_4, and evaporated to give a residue as a glass. The residue was dissolved in MeOH (100 ml), which was saturated with anhydrous NH_3 at 0°. After 42 hr at room temperature, the mixture was evaporated *in vacuo* and the residue was recrystallized from 50% EtOH to give colourless plates, mp 197—199° (decomp). Yield was 288.5 mg (70.9%). An analytical sample was recrystallized twice from 50% EtOH. *Anal.* Calcd. for $C_{25}H_{34}O_8N_4S$: C, 54.54; H, 6.23; N, 10.18; S, 5.81. Found: C, 54.21; H, 6.35; N, 10.05; S, 5.86. UV: λ_{max}^{MeOH} nm (ε) 235.5 (13600); 258.5 (11900), 278 (sh, 7700) 285 (sh, 7000); $\lambda_{max}^{0.1NHCl}$ 235.5 (13900), 275.5 (12200), 278.5 (7800), 285 (sh, 7600); $\lambda_{max}^{0.1NNaOH}$ 273.5 (14400). Paper chromatography *Rf* (B) 0.93 *Rf* (C) 0.95.

8,3′-Anhydro-8-oxy-9-β-D-xylofuranosylhypoxanthine (XII)——i) Well-dried 8-oxy-3′-TPS-inosine (550 mg, 1 mmole) was dissolved in DMF (38 ml) and freshly prepared anhydrous NaOAc (1.10 g) and anhyd. $CaCO_3$ (2.75 g) were added. The reaction mixture was refluxed with exclusion of moisture. Refluxing was stopped after 4 min. Examination of the reaction extent by TLC ($CHCl_3$–EtOH, 17: 3) showed the disappearance of the starting material (*Rf* 0.52) and the appearance of two spots having *Rf*s 0.06 and 0.24. Precipitating salts were removed by filtration, DMF was evaporated *in vacuo*, and the residue was applied to preparative TLC. Nucleoside extracted from the band migrating at *Rf* 0.06 was 8,3′-O-cycloinosine, which was identical with the sample obtained in i). Compound having *Rf* 0.24 showed UV: $\lambda_{max}^{50\%EtOH}$ nm 257.5, $\lambda_{max}^{0.1NHCl}$ 257.5, $\lambda_{max}^{0.1NNaOH}$ 273, which resembles 8-oxy inosine. Thus, this compound seems to be arabinosyl-8-oxyhypoxanthine.

ii) 8,3′-O-Cycloadenosine (527 mg, 2 mmoles) was dissolved in 2N AcOH (50 ml) and a saturated aqueous solution of $NaNO_2$ (552 mg, 8 mmole) was added with stirring. The reaction mixture was kept at room temperature overnight. After completion of the reaction was examined by TLC on Avicel (H_2O, pH 10), EtOH (20 ml) was added. The solvent was evaporated three times *in vacuo* and the final solution (20 ml) was adjusted to pH 6 with $NaHCO_3$. The solution was applied to a column of active charcoal (8 g), which was washed with H_2O (1.6 liter) and eluted with 50% EtOH containing 1% conc. NH_3 aq. Eluants were evaporated to dryness and the residue was recrystallized again from H_2O. *Anal.* Calcd. for $C_{10}H_{10}O_5N_4 \cdot 1/2H_2O$: C, 43.60; H, 4.03; N, 20.36. Found: C, 44.10; H, 3.97; N, 20.76. UV $\lambda_{max}^{pH7.0}$ nm (ε) 254 (14100), 285 (sh, 3700); λ_{max}^{pH2} 254 (14100), 285 (sh, 3700); λ_{max}^{pH12} 257 (15100). Mass spectrum peaks and NMR signals are listed in Table I and II. CD: [θ] peak at 254 nm 11500 (Fig 1). Paper chromatography: *Rf* (A) 0.81 *Rf* (B) 0.18, *Rf* (C) 0.49.

6-Mercapto-8-oxy-9-β-(2,5-di-O-acetyl-3-TPS-D-ribofuranosyl)purine (XVI)——Crude 8-oxy-3′-TPS-2′,-5′-diacetylinosine (1.9 g, 3 mmoles) and P_2S_5 (2.66 g, 12 mmoles) were dissolved in pyridine (70 ml). H_2O (0.216 ml, 12 mmoles) was added to the solution and heated at 135° for 10 hr with stirring. Loss of starting material (*Rf* 0.36) was examined by TLC ($CHCl_3$–EtOH, 10: 1) and pyridine was evaporated *in vacuo*. Co-evaporation with H_2O (20 ml) was repeated three times. To the residue was added H_2O (50 ml) and the solution was boiled for 5 min. After cooling, precipitates were extracted with $CHCl_3$, washed twice with H_2O, and dried over Na_2SO_4 with addition of one drop of β-mercaptoethanol. The solvent was evaporated and the residue was applied to preparative TLC ($CHCl_3$–EtOH, 19: 1). The band migrating at *Rf* 0.36 was extracted with MeOH. The solvent was evaporated to give a yellow glass (1.2 g, 61.5%). A part of the glass was recrystallized twice from 80% EtOH to pale yellow plates mp 218—220°, and subjected to elemental analysis. *Anal.* Calcd. for $C_{29}H_{38}O_9N_4S_2$: C, 53.53; H, 5.89; N, 8.61; S, 9.84. Found: C, 54.09; H, 5.87; N, 8.74; S, 10.24. UV λ_{max}^{MeOH} nm (ε) 235.5 (23200) 287 (sh, 8200) 315 (14800), 340 (12400); $\lambda_{max}^{0.1NHCl}$ 235.5 (22500), 287 (sh, 8200), 342.5 (17200); $\lambda_{max}^{0.1NNaOH}$ 243 (sh, 19700), 287.5 (sh, 7500), 322.5 (23200). Paper chromatography: *Rf* (B) 0.97, *Rf* (C) 0.96.

8,3′-Anhydro-6-mercapto-8-oxy-9-β-D-xylofuranosylpurine (XVII)——6-Mercapto-8-Oxy-3′-TPS-diacetylpurine riboside (XVI) (370 mg) was dissolved in anhydrous MeOH (30 ml) with ice cooling. The solution was saturated with dry NH_3 at 0° and kept at room temperature for 24 hr. MeOH was evaporated to give a glass. UV $\lambda_{max}^{50\%EtOH}$ (nm) 234 (sh), 298.5; $\lambda_{max}^{0.1NHCl}$ 234 (sh), 298.5; $\lambda_{nnax}^{0.1NNaOH}$ 234 (sh), 315.5. PPC: *Rf* (A) 0.78, *Rf* (B) 0.86, *Rf* (C) 0.91. The well-dried (over P_2O_5 *in vacuo*) material (306 mg, 0.54 mmole) was dissolved in DMF (20 ml) and freshly fused NaOAc (594 mg) and anhyd. $CaCO_3$ (1.782 g) were added. The mixture was refluxed for 3 min. After cooling salts were filtered off, DMF was evaporated *in vacuo*, and the residue was applied to Whatman 3MM paper. Development with the solvent C gave a band migrating at *Rf* 0.33. Elution of the nucleoside with H_2O and evaporation of H_2O gave 74 mg (48.5%) of 8,3′-O-cyclonucleoside. UV: $\lambda_{max}^{pH7.0}$ (nm) 235,315; $\lambda_{max}^{0.1NHCl}$ 232 (sh), 297 (sh), 327.5; $\lambda_{max}^{0.1NNaOH}$ 239,311. CD: [θ] peak at 315 nm=17800. PPC: *Rf* (A) 0.78, *Rf* (B) 0.26, *Rf* (C) 0.44. Mass spectral data are summarized in Table I.

6-Methylmercapto-8-oxy-β-(2,5-diacetyl-3-TPS-D-ribofuranosyl)purine (XIX)——i) 8-Oxy-TPS-diacetylthioinosine (XVI) (1.196 g, 1.84 mmole) and K_2CO_3 (0.276 g, 1.93 mmole) were suspended in DMF (20 ml) and methyl iodide (0.12 ml, 1.93 mmole) was added dropwise with stirring. The stirring was continued for 2 hrs and the reaction extent was examined by TLC ($CHCl_3$–EtOH, 19: 1, starting material *Rf* 0.80). The inorganic salt was filtered off and the filtrate was evaporated *in vacuo*. The oily residue was dried over P_2O_5 in a desiccator. UV: $\lambda_{max}^{50\%EtOH}$ (nm) 232,298.5; $\lambda_{max}^{0.1NHCl}$ 232, 298.5; $\lambda_{max}^{0.1NNaOH}$ 280 (sh), 311.5.

ii) 6-Mercaptopurine-8,3′-O-cyclonucleoside (XVII) (56.5 mg, 0.2 mmole) was dissolved in DMF (0.7 ml). K_2CO_3 (27.7 mg, 0.21 mmole) and CH_3I (0.013 ml, 0.21 mmole) were added with stirring. After stirring at room temperature for 1 hr, the salts were filtered off and washed with a small amount of DMF. The filtrate and washing were combined and evaporated *in vacuo*. The residue was applied to PPC in solvent A. The band migrating at *Rf* 0.69 was eluted with H_2O and 8,3′-methylmercaptocyclonucleoside was obtained in a yield of 7 mg (10.2%).

8,3′-Anhydro-6-methylmercapto-8-oxy-9-β-D-xylofuranosylpurine (XVIII)——The crude 8-oxy-TPS-diacetylmethylthio compound (XIX), obtained as above was dissolved in anhydrous methanolic ammonia (60 ml) and kept at room temperature for 42 hr. MeOH was evaporated and the residual oil was dried over P_2O_5. TLC ($CHCl_3$–EtOH, 18: 2) of this material showed *Rf* 0.70 and paper chromatography showed *Rf* (C) 8.90. To the residue were added anhydrous NaOAc (2.03 g), $CaCO_3$ (6.06 g), and DMF (70 ml). The mixture was heated at 175° for 1.5 min after commencement of boiling. The salt was filtered off and the filtrate was evaporated *in vacuo*. TLC ($CHCl_3$–EtOH, 18: 2) showed 2 spots having *Rf* 0.52 and 0.26. The compound having *Rf* 0.52 showed UV: $\lambda_{max}^{50\%EtOH}$ (nm) 298, $\lambda_{max}^{0.1NNaOH}$ 311 and was thought to be a 8-oxy-xylofuranosyl compound. The compound having *Rf* 0.26 was extracted with MeOH (100 ml) and the 8,3′-O-cyclo compound was obtained as a glass (0.34 g, 54%). Recrystallization twice from MeOH gave colorless needles, mp 235—238°. *Anal.* Calcd. for $C_{11}H_{12}O_4SN_4$: C, 44.60; H, 4.08; N, 18.91. Found: C, 44.67; H, 4.09; N, 18.58. UV: nm (ε) $\lambda_{max}^{pH7.0}$ 230 (12400), 294.5 (20300); λ_{max}^{pH2} 231 (12100), 295 (19200), 320 (sh, 1900); λ_{max}^{pH12} 231.5 (11300), 296 (20700). Mass spectrum showed a molecular ion peak at *m/e* 296 and others are listed in Table I. NMR signals are listed in Table II. CD: [θ] peak at 294 nm=12500 and [θ] trough at 233 nm (−19000). (see Fig. 3). Paper chromatography: *Rf* (A) 0.70, *Rf* (B) 0.71, *Rf* (C) 0.70.

8,3′-Anhydro-8-oxynebularine (XX)——8,3′-O-Cyclo-6-methylmercapto compound (XVIII) (74 mg) was dissolved in MeOH (20 ml) and freshly prepared Raney Ni (*ca.* 1 ml) was added. The mixture was heated at reflux temperature for 1 hr. The catalyst was filtered off and the filtrate was evaporated to dryness. The residue was applied to Whatman 3MM paper and developed in solvent B. The band migrating at *Rf* 0.39 was cut and eluted with H_2O. Evaporation of the solvent gave 8,3′-O-cyclonebularine. UV: $\lambda_{max}^{H_2O}$ (nm) 241,271; $\lambda_{max}^{0.1NHCl}$ 296; $\lambda_{max}^{0.1NNaOH}$ 241 (sh), 274. Mass spectrum gave a molecular ion peak at *m/e* 250. (see Table I). CD: [θ] peak at 265 nm=1000 (broad) and [θ] trough at 235 nm=−3700. (Fig. 4).

2′,3′-Ethoxymethylidene-8-bromoinosine (XXIV)——8-Bromoinosine[21] (1.74 g, 5 mmoles) was dissolved in anhydrous DMF (25 ml) with warming. After cooling to room temperature, ethylorthoformate (2.5 ml, 15 mmoles) and 5.65 M HCl–DMF (1.25 ml, 8 mmole) were added to the solution. The reaction mixture was kept at room temperature for 40 hr with exclusion of moisture. The reaction was stopped by adding triethyl-

amine (11 ml) and the precipitating salt was filtered off. The filtrate was evaporated *in vacuo* to give a glassy residue. The residue was extracted twice with EtOH and the extracts were evaporated. Recrystallization of this material twice from acetone gave colorless needles. UV: $\lambda_{max}^{50\%EtOH}$ (nm) 248.255 (sh); $\lambda_{max}^{0.1NHCl}$ 248, 255, $\lambda_{max}^{0.1NNaOH}$ 245. TLC: ($CHCl_3$–EtOH, 19: 2) *Rf* 0.44.

8,5′-Anhydro-8-oxy-inosine (XXII)——i) 8,5′-O-Cycloadenosine (327 mg, 1.25 mmole) was dissolved in 2N AcOH by heating at 30°. After cooling, a saturated aqueous solution of $NaNO_2$ (431 mg, 6.25 mmole) was added. After the reaction mixture was kept at room temperature for 5 days, precipitates were collected by filtration. The filtrate was evaporated to 1/3 volume and kept at 0—5° overnight. Crystals were collected by filtration and combined with the first crop. Recrystallization twice from H_2O gave 220 mg (67.3%) of 8,5′-O-cycloinosine as colorless needles, mp 250 (decomp). *Anal.* Calcd. for $C_{10}H_{10}O_5N_4H_2O$: C, 42.45; H, 4.26; N, 19.71. Found: C, 42.27; H, 3.95; N, 19.48. UV: $\lambda_{max}^{pH7.0}$ nm (ε) 252 (13500), λ_{max}^{pH1} 251 (13900) λ_{max}^{pH13} 255 (15300). Mass spectrum showed a signal of a molecular ion at *m/e* 266 (see Table I). NMR: (d_6-DMSO ppm) 12.37 (br, N_1–H), 8.02 (s, C_2–H), 5.93 (s, C_1, –H), 3.90—4.70 (C_2, to C_5, –H's). CD: [θ] peak at 250 nm= 22000. (see Fig. 1). Paper chromatography: *Rf* (A) 0.61, *Rf* (B) 0.11 *Rf* (C) 0.37.

ii) 2′,3′-Ethoxymethylidene-8-bromoinosine (1.209 g, 3 mmole) was dissolved in anhyd. DMF (18 ml) and NaH (0.69 g, containing 50% mineral oil and washed with anhyd. benzene (12 mmoles) was added with ice-cooling. The reaction mixture was stirred for 2 hr and kept at room temperature overnight. Excess NaH was decomposed by adding 99% EtOH, H_2O (60 ml) and the pH was adjusted with acetic acid to 7.0. The mixture was cooled to −5° for 1 hr and precipitates were collected by filtration. This material (322 mg, 1 mmole) was dissolved in 50% AcOH–EtOH (21 ml, 1: 1 vol/vol) and heated at 110° for 1 hr. Solvents were evaporated *in vacuo* and traces of acetic acid were removed by repeated evaporation with added EtOH. The residue was recrystallized from H_2O to give pale yellow needles (104 mg). Examination of this material by paper chromatography showed *Rf* (A) 0.61 and *Rf* (B) 0.11 corresponding to 8,5′-O-cycloinosine described in ii), but slightly contaminated with material having *Rf* (A) 0.71 and *Rf* (B) 0.20. The latter material seemed to be the 8-oxy compound by criteria of UV: (nm) $\lambda_{max}^{H_2O}$ 257, $\lambda_{max}^{H^+}$ 253.5, $\lambda_{max}^{OH^-}$ 258 nm.