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Synthesis of 8,2'-Methanoguanosine and 9-(α -D-Arabinofuranosyl)-8,2'-methanoguanine (Nucleosides and Nucleotides. LXVII.¹⁾)

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Guanosine fixed in the high-anti conformation by means of an 8,2'-methylene bridge was prepared. *N*²-Acetyl-*O*⁶-ethylguanosine (**2**) was converted to the 3',5'-*O*-(tetraisopropylidisiloxane-1,3-diyl) derivative (**3**). Oxidation of **3** to the 2'-keto derivative (**4**) and successive coupling with methylenetriphenylphosphorane gave the 2'-methylidene derivative (**5**) and its α -anomer. The 2'-methylidene function of **5** was hydroxylated, and the 2'-hydroxymethyl group was modified to give the phenylthiomethyl derivative (**6**). Photocyclization of **6** followed by deprotection of the sugar and base protecting groups furnished 8,2'-methanoguanosine (**12**). The α anomer of **12** was likewise prepared. The circular dichroism spectra of **12**, its α -anomer, and related compounds were measured.

Keywords—8,2'-methanoguanosine; 9-(α -D-arabinofuranosyl)-8,2'-methanoguanine; C-cycloguanosine; Wittig reaction; photocyclization; nucleoside conformation; CD; NMR

In our earlier studies on the synthesis of carbon-bridged cycloguanosines, we have synthesized 5'-deoxy-8,5'-cycloguanosine²⁾ and its 2', 3'-cyclic phosphate.³⁾ This nucleotide is useful for investigations on the mode of binding of guanylates to ribonuclease (RNase) T₁ and on the mechanism of cleavage by RNase T₁.^{3,4)} For further study of the stereochemistry of the enzyme-substrate binding of RNase T₁, it is expected that other C-cycloguanosines having different glycosyl torsion angles, such as 8,2'- or 8,3'-cycloguanosine derivatives, would also be useful. This paper deals with the synthesis of 8,2'-methanoguanosine in which the base portion is fixed in a high-anti conformation. Very recently, 2'-deoxy-8,2'-methanoguanosine was prepared by an intramolecular ionic cyclization reaction.⁵⁾

We have recently reported the synthesis of carbon-bridged 8,2'-cycloadenosines by a photocyclization of 2'-phenylthiomethyladenosines.⁶⁾ This method was expected to be quite effective for the present purpose. As is well known, the solubility of guanine derivatives is very low and gel formation is quite general during attempts at crystallization, but protection of the base portion by *O*⁶-alkylation is an effective means of increasing the solubility in organic solvents and reducing the reactivity of the N-1 position of the guanine moiety.⁷⁾ Therefore, in the present study we used the *O*⁶-ethylated guanosine as a starting material. Treatment of *N*²,2',3',5'-*O*-tetraacetylguanosine (**1**) with Mitsunobu reagents (triphenylphosphine and diethyl azodicarboxylate)⁸⁾ and ethanol afforded, after de-*O*-acetylation, *N*²-acetyl-*O*⁶-ethylguanosine⁵⁾ (**2**). The 3'- and 5'-hydroxyl groups of **2** were then protected with 1,1,3,3-tetraisopropyl-1,3-dichlorodisiloxane⁹⁾ (TIPDSCl₂) to give the 3',5'-*O*-silyl derivative (**3**). Oxidation of **3** with dimethylsulfoxide and oxalyl chloride¹⁰⁾ afforded the *O*⁶-ethyl-2'-ketoguanosine (**4**), which was used for the next step without purification. Treatment of crude **4** with methylenetriphenylphosphorane at 0 °C gave the expected 2'-methylidene derivative (**5**).

Although **5** was homogeneous as checked by chromatography on silica gel, the nuclear magnetic resonance (NMR) spectrum of **5** showed that it was an approximately equimolar

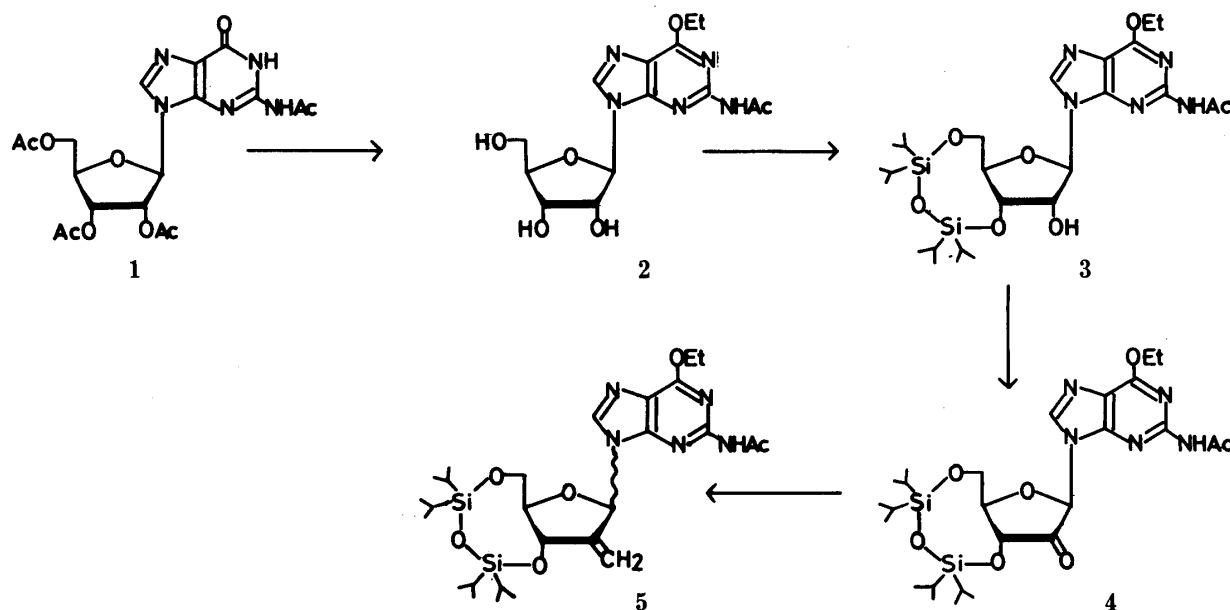


Chart 1

mixture of anomers. Therefore, anomerization of the 2'-ketoguanosine (4) under the coupling conditions must have occurred, although such anomerization had not been observed in the Wittig reaction of the 2'-ketoadenosine derivative.⁶⁾ Compound 5 was hydroxylated by treatment with osmium tetroxide and *N*-methylmorpholine *N*-oxide, then the 2'-hydroxymethyl compounds were converted to the β -(6) and α -anomer (7) of 2'-phenylthiomethylguanosine by mesylation and successive substitution with thiophenoxide. Compounds 6 and 7 were separated by silica gel column chromatography in yields of 23 and 21%, respectively. The configurations of the 2'-position of 6 and 7 were assigned as *R* and *S*, respectively, assuming that the hydroxylation occurred from the less hindered side¹¹⁾ (*trans* to the base orientation) in each case.

Photoirradiation of 6 in acetonitrile in the presence of trimethyl phosphite^{1,6)} gave the 8,2'-cyclo products (8 and its *N*-deacetylated derivative, 9) in 67% yield. The mixture was deacetylated, desilylated, and then re-acetylated to give the di-*O*-acetate (10). Treatment of 10 with iodotrimethylsilane resulted in de-*O*-ethylation to give 11, which was deacetylated by treatment with triethylamine in methanol to furnish 8,2'-methanoguanosine (12) in a crystalline form. The NMR spectrum and other instrumental analyses confirmed the structure. In paper electrophoresis in a borate buffer, 12 migrated similarly to guanosine, which supports the presence of a 2',3'-*cis* diol system in 12.

The α -anomer 7 was likewise photoirradiated to give a mixture of 8,2'-methano-cyclo derivatives (13, R=H or Ac), which was deacetylated, desilylated, and re-acetylated to furnish the *O*⁶-ethyl cycloguanosine (14). Deprotection of 14 by treatment with iodotrimethylsilane followed by methanolic triethylamine gave crystalline 9- α -D-arabinofuranosyl-8,2'-methanoguanine (15). The structure of 15 was confirmed by comparison of its NMR spectrum with those of the β -anomer 12 and 8,2'-methanoadenosine.⁶⁾ The low field shift of one of the bridge-methylene signals and reversal of the chemical shifts of H-3', and H-4' are noteworthy in the α -anomer (15) as compared to the β -anomers of cycloadenosine and -guanosine (12). The slower migration of 15 on paper electrophoresis in borate buffer is also consistent with the *trans* configuration of the 2',3'-diol system.

The circular dichroism (CD) spectra of 10 and 14 are also informative regarding the anomer relations. Compounds 10 and 14 showed inverted CD spectra with respect to each

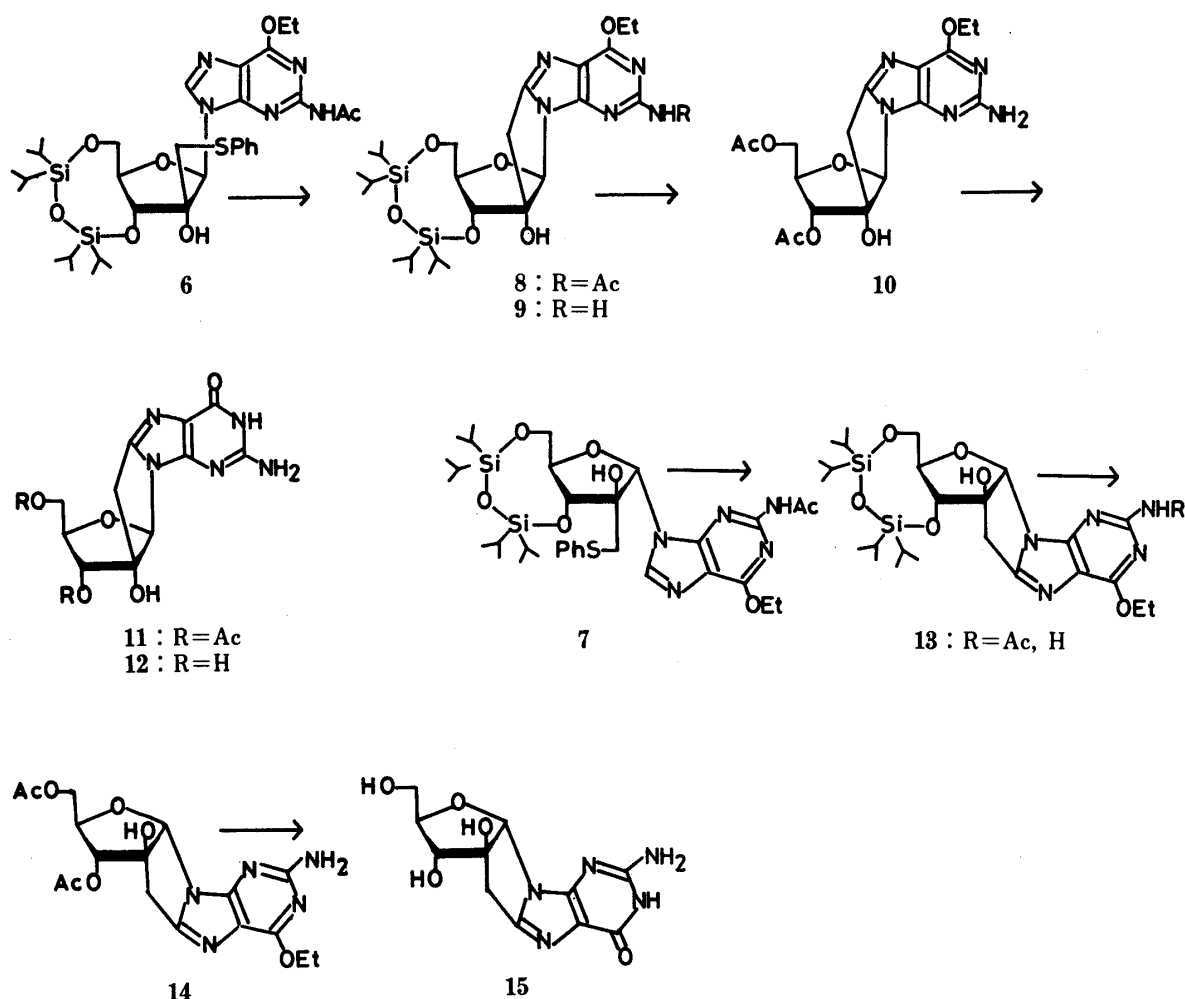


Chart 2

other in methanol (the two spectra are virtual mirror images), whereas the ultraviolet (UV) spectra are closely similar (Fig. 1). In contrast, the CD spectra of **12** and **15** are apparently more complex (Fig. 2). In the case of the β -anomer **12**, the CD band at the 280 nm region (the B_{2u} transition) is positive and that at the 250 nm region (B_{1u} transition) is negative. The CD bands of the α -anomer **15** exhibited positive bands both in the B_{1u} and B_{2u} transitions. On protonation at pH 1.0, the overall pattern of the CD spectrum of **12** remained essentially unchanged, though the molar ellipticity at the B_{1u} region was increased. Compound **15** also showed an increase at the B_{1u} region. In **15**, however, inversion of the sign at the longer wavelength (B_{2u}) region upon protonation was noted.

We have already reported the CD spectra of 5'-deoxy-8,5'-cycloguanosine 2',3'-cyclic phosphate and 3'-phosphate,^{3,4)} and pointed out the inversion of the CD band (negative to positive) on N-7 protonation, as in the cases of naturally occurring guanosine and its phosphates. Therefore, it may be concluded that the inversion of the sign of the CD spectrum of guanosine by N-7 protonation is not a general feature of guanosines, but the phenomenon is rather a function of the glycosyl torsion angle as well as the nature of the transition. Therefore, it follows that guanosine having the glycosyl torsion angle fixed at the anti conformation (8,5'-cycloguanosines) shows positive CD bands on N-7 protonation and guanosine with high-anti conformation (**12**) shows a negative band on N-7 protonation. However, since guanosine derivatives generally show the partially separated UV spectra of B_{1u} and B_{2u} transitions, and these two transitions should both contribute to the CD

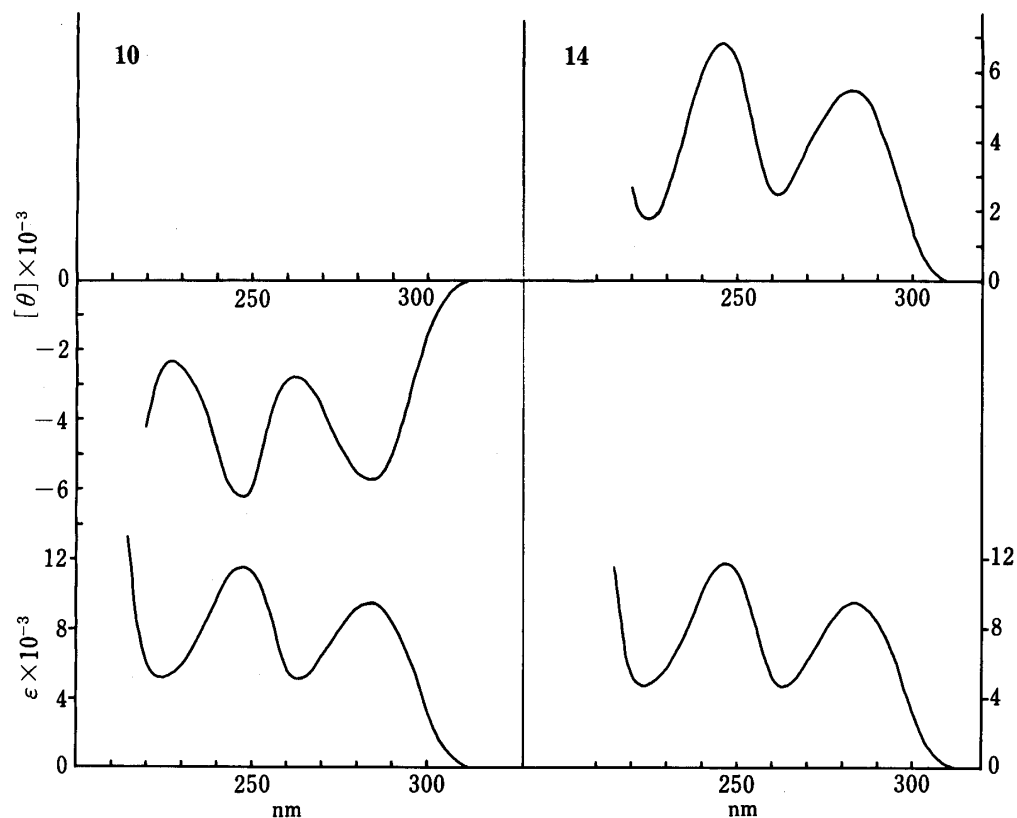


Fig. 1. CD Spectra of 3',5'-Di-*O*-acetyl-*O*⁶-ethyl-8,2'-methanoguanosine (10) and 2-Amino-6-ethoxy-9-(3,5-di-*O*-acetyl- α -D-arabinofuranosyl)purine (14) in Methanol

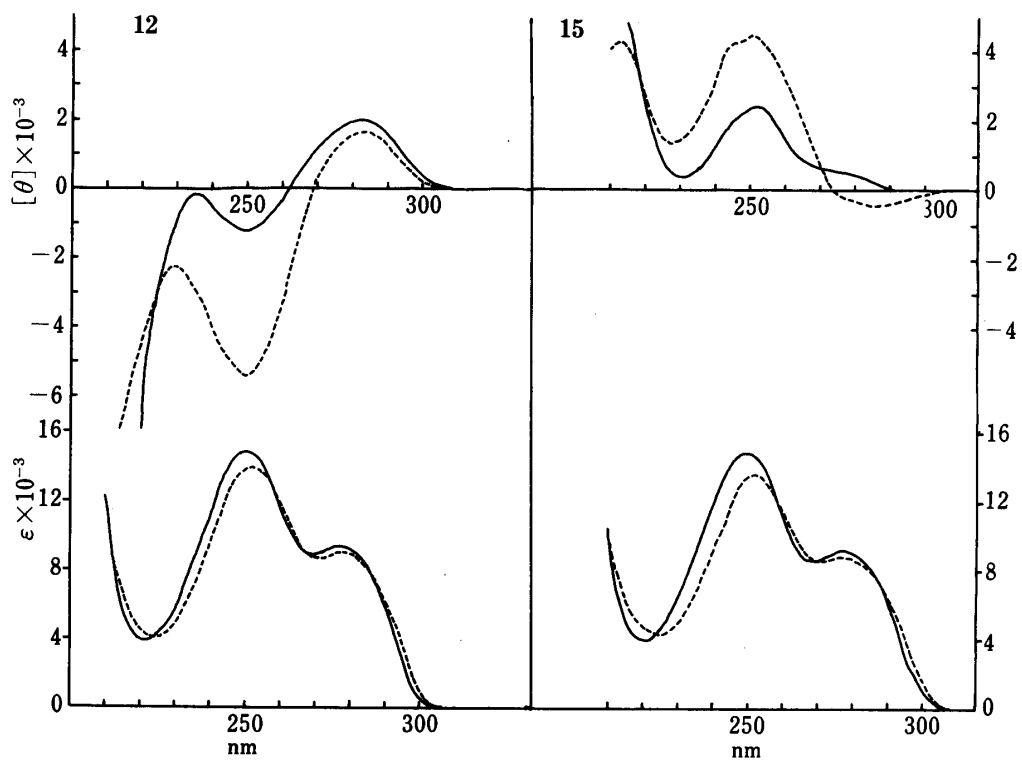


Fig. 2. CD Spectra of 8,2'-Methanoguanosine (12) and 9-(α -D-Arabinofuranosyl)-8,2'-methanoguanine (15) in Water

— in H₂O; ---- in 0.1 N HCl.

spectra, further experiments on the synthesis of various C-cycloguanosines will be necessary to fully understand the CD spectra in terms of glycosyl torsion angles and transition moments. In fact, 2'-deoxy-8,2'-methanoguanosine was reported to exhibit somewhat different CD and UV spectra from those of **12** or other guanosine derivatives.⁵⁾

Experimental

All melting points were determined on a Yanagimoto MP-3 micromelting point apparatus and are uncorrected. The UV spectra were measured with a Shimadzu UV-260 spectrophotometer, and NMR spectra were taken on a JEOL JNM-FX 100FT or 200FT spectrometer with tetramethylsilane as an internal standard. Chemical shifts are reported as δ (ppm) and the signal splitting is described as s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet) or b (broad). Mass spectra (MS) were taken on a JEOL D-300 spectrometer, and CD spectra were recorded on a JEOL J-500A spectropolarimeter at room temperature. The photoreaction was carried out in an apparatus fitted with an Eikosha PIL-60 60W low-pressure Hg vapor lamp (quartz filter) in an argon atmosphere. The starting nucleoside, guanosine, was from Yamasa Shoyu Co. Silica gel used for column chromatography or preparative thin layer chromatography (PTLC) was Wako gel C-200.

The abbreviation TIPDS is used for the tetraisopropylidisiloxane-1,3-diyl protecting group in the present experiment.

***N*²-Acetyl-*O*⁶-ethylguanosine (2)**—This compound was prepared by a method essentially similar to that reported.⁵⁾ Triphenylphosphine (9.7 g, 1.2 eq), diethyl azodicarboxylate (5.8 ml, 1.2 eq) and EtOH (2.0 ml, 1.1 eq) were added to a solution of *N*²-2',3',5'-*O*-tetraacetylguanosine (**1**, 13.9 g, 30.8 mmol) in dioxane (300 ml) and the whole was stirred for 15 min at room temperature. The solvent was removed *in vacuo* and the residue was dissolved in a mixture of MeOH (200 ml) and conc. NH₄OH (200 ml). The mixture was stirred for 3 h at room temperature. The precipitated **2** was collected (3.66 g) by filtration and the filtrate was concentrated. The residue was triturated in CHCl₃ (200 ml) to give additional **2** (3.86 g). Further **2** (1.40 g) was obtained by silica gel column (200 g) chromatography of the mother liquor (eluted with 12% MeOH-CHCl₃). Total yield was 8.92 g (82%). An aliquot was recrystallized from H₂O to obtain a pure sample for analyses. mp 195–196 °C. UV $\lambda_{\text{max}}^{\text{H}_2\text{O}}$ nm (ϵ): 260 (16300), 218 (20600). MS *m/z*: 353 (*M*⁺). NMR (DMSO-*d*₆ + D₂O) δ : 8.43 (1H, s, H-8), 5.89 (1H, d, H-1', *J* = 5.9 Hz), 4.67–4.46 (3H, m, H-2', CH₃CH₂O), 4.18 (1H, dd, H-3', *J*_{2',3'} = 5.1 Hz, *J*_{3',4'} = 3.4 Hz), 3.93 (1H, m, H-4') 3.59 (2H, m, H-5'ab), 2.22 (3H, s, Ac), 1.40 (3H, t, CH₃CH₂O, *J* = 7.1 Hz). Anal. Calcd for C₁₄H₁₉N₅O₆: C, 47.59; H, 5.42; N, 19.82. Found: C, 47.55; H, 5.35; N, 19.53.

3',5'-*O*-TIPDS-*N*²-acetyl-*O*⁶-ethylguanosine (3)—A solution of **2** (9.49 g, 26.86 mmol) and TIPDSCl₂ (9.3 ml, 1.1 eq) in pyridine (200 ml) was stirred for 3 h at room temperature. A small volume of H₂O was added, the solvent was removed *in vacuo* and the residue was partitioned between EtOAc (200 ml) and H₂O (100 ml). The organic layer was separated and passed through a Whatman 1PS filter paper, then the filtrate was concentrated *in vacuo*. The residue was taken up in toluene and a trace of H₂O was removed by co-distillation. The residue was dissolved in CHCl₃, and applied to a column of silica gel (300 g). The eluate with 1–2% MeOH-CHCl₃ was concentrated to leave **3** (14.02 g, 88%) as a foam. UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm 268; $\lambda_{\text{max}}^{\text{H}_2\text{O}}$: 272, 255. MS *m/z*: 595 (*M*⁺), 552 (*M* – isoPr). NMR (CDCl₃) δ : 8.00 (1H, s, H-8), 7.86 (1H, br s, HN-2), 5.98 (1H, d, H-1', *J* = 1.7 Hz), 4.69–4.40 (4H, m, H-2', 3', CH₃CH₂O), 4.15–4.10 (3H, m, H-4', 5'ab), 3.16 (1H, d, HO-2', *J* = 2.0 Hz), 2.59 (3H, s, Ac), 1.10–0.98 (28H, m, isoPr).

3',5'-*O*-TIPDS-*N*²-acetyl-*O*⁶-ethyl-2'-ketoguanosine (4)—DMSO (4.67 ml, 2.8 eq) in CH₂Cl₂ (20 ml) was added dropwise to a solution of (COCl)₂ (2.67 ml, 1.3 eq) in CH₂Cl₂ (100 ml) at –60––70 °C. After 30 min, **3** (14.0 g, 23.5 mmol) in CH₂Cl₂ (70 ml) was added over a period of 30 min. Et₃N (16.4 ml, 5 eq) was added to the solution after 30 min of stirring, and the whole was then brought to room temperature. The mixture was partitioned between CHCl₃ (100 ml) and H₂O (200 ml), the aqueous layer being acidified to pH 4 by addition of 1 *N* HCl, and the organic layer was separated. The aqueous layer was extracted with CHCl₃ (50 ml), and the combined organic layer was concentrated. The residue was dissolved in CHCl₃ and applied to a column of silica gel (300 g). The eluate with 10–20% EtOAc-CHCl₃ was concentrated to leave **4** (9.7 g, 69%) as a foam, which was dried over P₂O₅ overnight and used for the next step without further purification.

3',5'-*O*-TIPDS-*N*²-acetyl-*O*⁶-ethyl-2'-deoxy-2'-methylideneguanosine (5)—Compound **4** (9.7 g, 16.3 mmol) in THF (35 ml) was added dropwise to a solution of methylenetriphenylphosphorane⁶⁾ (2 eq) in THF and DMSO at 0 °C, and the solution was stirred for 1 h at that temperature. Stirring was continued for a further 2.5 h at room temperature. After neutralization of the mixture by addition of AcOH, the solvent was removed *in vacuo* and the residue was partitioned between EtOAc and H₂O (200 ml each). The organic layer was passed through a Whatman 1PS filter paper and the solvent was evaporated off. The residue was applied to a column of silica gel (200 g). The eluate with 1% EtOH in CHCl₃ was concentrated and the residue was subjected to silica gel column chromatography several times. From the concentrate of the eluate, a small amount of **5** and a 1:1 mixture of **5** and its α -anomer (as judged from the NMR signal of H-1') were obtained in a combined yield of 1.47 g (15%). MS *m/z*: 591 (*M*⁺), 576 (*M* – Me), 548 (*M* – isoPr). NMR (CDCl₃) δ : 7.90 (1H, s, H-8), 7.85 (1H, br s, HN), 6.59 (1H, dd, H-1'), 5.48 (2H, m,

=CH₂ at 2'), 5.08 (1H, m, H-3'), 4.59 (2H, q, CH₃CH₂O), 4.15–4.08 (2H, m, H-5'ab), 3.80 (1H, m, H-4', $J_{3,4'} = 8.8$ Hz), 2.61 (3H, s, Ac), 1.49 (3H, t, CH₃CH₂O), 1.11–1.07 (28H, m, isoPr). The signal of H-1' of the α -anomer appeared at 6.70 ppm as a broad triplet. Other signals appeared at the same positions as or overlapped with those of the β -anomer, and were not characterized, since the pure α -anomer could not be isolated.

3',5'-O-TIPDS-N²-acetyl-O⁶-ethyl-2'-(R)-phenylthiomethylguanosine (6)—A mixture of **5** and its α -anomer (1.14 g, 1.92 mmol) was dissolved in a mixture of THF (9 ml), *tert*-BuOH (9 ml), and H₂O (3 ml). *N*-Methylmorpholine-*N*-oxide (0.26 g, 1 eq) and OsO₄ (0.98 ml of 0.5% *tert*-BuOH solution, 0.01 eq) were added to the solution and the whole was stirred at 4 °C for 3 d. Further *N*-oxide (80 mg) and OsO₄ solution (0.5 ml) were added and the reaction mixture was stirred for an additional 3 d. The mixture was partitioned between EtOAc and 1 *N* NaHSO₃ (30 ml each), and the organic layer was separated, and passed through a Whatman 1 PS filter paper. The filtrate was concentrated. The residue was applied to a column of silica gel (50 g). The eluate with 1–2% MeOH in CHCl₃ was concentrated to leave the 2'-(*R* and *S*)-hydroxymethyl derivatives (0.925 g, 77%). The compound was dissolved in pyridine (10 ml) and MsCl (0.17 ml, 1.5 eq) was added to the solution. After 2 h, a small volume of H₂O was added, the solvent was removed *in vacuo*, and the residue was partitioned between EtOAc and sat. NaHCO₃ (20 ml each). The organic layer was separated and the solvent was removed. The residue was dried by codistillation with toluene, and the residue was dissolved in DMF (10 ml). PhSH (0.152 ml) and KO*tert*-Bu (158 mg) were added and the solution was stirred for 1 h at room temperature. After neutralization of the solution by addition of 1 *N* HCl, the solvent was removed and the residue was partitioned between EtOAc and H₂O saturated with NaCl. The organic layer was separated, the solvent was removed, and the residue was subjected to silica gel column chromatography several times. Compound **6** (321 mg, 23%) was obtained as a foam from the slower-eluting fractions with 2–4% EtOAc in CHCl₃. UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm 257. MS m/z : 717 (M⁺). NMR (CDCl₃) δ : 8.10 (1H, s, H-8), 7.81 (1H, br s, HN), 7.17–6.96 (5H, m, SPh), 6.14 (1H, s, H-1'), 4.76 (1H, d, H-3'), 4.53 (2H, q, CH₃CH₂O), 4.31–4.00 (3H, m, H-4', 5'ab), 3.40 (1H, s, HO-2'), 3.24 (1H, d, H-6'a, $J_{a,b} = 13.2$ Hz), 2.95 (1H, d, H-6'b), 2.58 (3H, s, Ac), 1.14–1.05 (28H, m, isoPr).

2-Acetamido-6-ethoxy-9-[3,5-O-TIPDS-2(S)-phenylthiomethyl- α -D-arabionofuranosyl]purine (7)—Compound **7** (288 mg, 21%) was obtained by evaporation of the solvent from the above faster-eluting fractions. UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm 258. MS m/z : 717 (M⁺). NMR (CDCl₃) δ : 8.21 (1H, s, H-8), 8.02 (1H, br s, HN), 7.15–7.03 (5H, m, SPh), 6.17 (1H, s, H-1'), 4.67–4.28 (5H, m, H-3', 4', HO-2', CH₃CH₂O), 4.07 (2H, m, H-5'ab), 3.35 (1H, d, H-6'a, $J_{a,b} = 13.7$ Hz), 3.20 (1H, d, H-6'b), 2.50 (3H, s, Ac), 1.49 (3H, t, CH₃CH₂O), 1.13–1.06 (28H, m, isoPr).

3',5'-O-TIPDS-N²-acetyl-O⁶-ethyl-8,2'-methanoguanosine (8) and 3',5'-O-TIPDS-O⁶-ethyl-8,2'-methanoguanosine (9)—Compound **6** (380 mg, 0.53 mmol) and trimethyl phosphite (1 ml) were dissolved in acetonitrile (350 ml) and the solution was irradiated for 1.5 h at room temperature. The solvent was removed *in vacuo* and the residue was applied to a column of silica gel (30 g). The eluate with 1–2% EtOH in CHCl₃ was concentrated to give **8** and **9** as a 45:55 mixture (208 mg, 67%). A portion was separated by PTLC (CHCl₃–acetone, 1:1) to obtain **8** and **9**.

Physical constants of **8**. UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm: 268; $\lambda_{\text{max}}^{\text{pH}2}$: 270.5, 258.1. MS m/z : 607 (M⁺), 564 (M–isoPr). NMR (CDCl₃) δ : 7.93 (1H, br s, HN), 5.94 (1H, s, H-1'), 4.58 (2H, q, CH₃CH₂O), 4.18–3.68 (4H, m, H-3', 4', 5'ab), 3.32 (1H, d, H-6'a, $J = 16.2$ Hz), 3.15 (1H, d, H-6'b), 2.58 (3H, s, Ac), 1.49 (3H, t, CH₃CH₂O), 1.08–0.93 (28H, m, isoPr).

Physical constants of **9**. UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm: 283, 248; $\lambda_{\text{max}}^{\text{pH}2}$: 290, 245. MS m/z : 565 (M⁺), 522 (M–isoPr). NMR (CDCl₃) δ : 5.89 (1H, s, H-1'), 4.96 (2H, br s, NH₂), 4.52 (2H, q, CH₃CH₂O), 4.25–3.68 (4H, m, H-3', 4', 5'ab), 3.72 (1H, s, HO-2'), 3.30 (1H, d, H-6'a, $J = 17.3$ Hz), 3.09 (1H, d, H-6'b), 1.44 (3H, t, CH₃CH₂O), 1.08–0.94 (28H, m, isoPr).

3',5'-Di-O-acetyl-O⁶-ethyl-8,2'-methanoguanosine (10)—A mixture of **8** and **9** (200 mg, 0.34 mmol) in MeOH saturated with NH₃ (25 ml) was heated at 100 °C for 3 d in a sealed tube. The solvent was removed and the residue was treated with Bu₄NF (0.3 ml of 1 *M* THF) in THF (5 ml) at room temperature for 1.5 h. The solvent was removed and the residue was dissolved in acetonitrile (5 ml) containing Ac₂O (71 μ l), Et₃N (105 μ l), and *N*⁴-dimethylaminopyridine (3 mg). The solution was stirred for 15 min, then the solvent was removed and the residue was applied to a column of silica gel (20 g). The eluate with 2–4% MeOH in CHCl₃ was concentrated to leave **10** (69 mg, 69%). UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm (ϵ): 284 (9500), 246 (11500); λ_{min} : 264 (5040). CD (MeOH) [θ] (nm): –5760 (284), –2780 (262), –6250 (248), –2330 (227). MS m/z : 407 (M⁺), 392 (M–Me). NMR (CDCl₃) δ : 5.89 (1H, s, H-1'), 5.08 (2H, br s, NH₂), 5.02 (1H, d, H-3', $J_{3,4'} = 6.4$ Hz), 4.60–4.32 (3H, m, H-4', CH₃CH₂O), 4.26 (1H, dd, H-5'a, $J_{4',5'a} = 4$ Hz, $J_{5'a,b} = 12$ Hz), 4.05 (1H, dd, H-5'b, $J_{4',5'b} = 4.2$ Hz), 3.62 (1H, d, H-6'a, $J_{6'a,b} = 18.1$ Hz), 3.19 (1H, d, H-6'b), 2.20, 1.91 (3H each, s, Ac), 1.44 (3H, t, CH₃CH₂O).

3',5'-Di-O-acetyl-8,2'-methanoguanosine (11)—Me₃SiCl (113 μ l) was added to a solution of NaI (143 mg) in acetonitrile in the dark. After 5 min, a solution of **10** (71 mg) in acetonitrile (2 ml) was added and the whole was stirred at 50 °C for 6 h. H₂O (0.65 ml) and then Et₃N (133 μ l) were added under cooling in an ice bath. After being stirred for 5 min, the solution was concentrated and the residue was subjected to PTLC (CHCl₃–MeOH, 5:1). The appropriate band was extracted with the same solvent to give **11** (41 mg, 62%). Crystallization from H₂O–EtOH gave a pure sample, mp 237 °C (dec). UV $\lambda_{\text{max}}^{\text{MeOH-H}_2\text{O}}$ nm: 277, 250; $\lambda_{\text{max}}^{\text{pH}11}$: 254. MS m/z : 379 (M⁺). NMR (CDCl₃) δ : 10.66 (1H, br s, HN-1), 6.52 (2H, br s, H₂N), 6.40 (1H, s, H-1'), 5.63 (1H, s, HO-2'), 4.94 (1H, s, H-3', $J_{3,4'} = 6.8$ Hz),

4.48—4.11 (2H, m, H-4', 5'a), 3.98 (1H, dd, H-5'b), 3.40 (1H, d, H-6'a, $J_{6'a,b} = 17$ Hz), 2.85 (1H, d, H-6'b), 2.12, 1.89 (3H each, s, Ac).

8,2'-Methanoguanosine (12)—Compound 11 (18 mg) was added to 20% Et₃N-MeOH (3 ml) and the suspension was stirred overnight at 50 °C. The solvent was removed *in vacuo* and the residue was crystallized from H₂O-MeOH to give 12 (6.5 mg, 47%), mp > 300 °C. UV $\lambda_{\max}^{\text{H}_2\text{O}}$ nm (ϵ): 277 (9300), 250 (14800); $\lambda_{\max}^{\text{H}^+}$: 278 (8900), 252 (13800); $\lambda_{\max}^{\text{H}^{11}}$: 270 (11100), 255 (11600), 214 (18100). pK_a value determined from the UV spectra: 2.05. CD (H₂O) [θ] (nm): +2020 (282), 0, (262), -1200 (250). NMR (D₂O) δ : 5.72 (1H, s, H-1'), 4.04 (1H, ddd, H-4'), 3.82 (1H, d, H-3', $J_{3',4'} = 8.3$ Hz), 3.74 (1H, dd, H-5'a, $J_{4',5'a} = 2.7$ Hz, $J_{5'a,b} = 12.7$ Hz), 3.32 (1H, d, H-5'b, $J_{4',5'b} = 5.4$ Hz), 3.19 (1H, d, H-6'a), 2.86 (1H, d, H-6'b, $J_{6'a,b} = 17.1$ Hz). Relative migration with respect to guanosine on paper electrophoresis (0.2 M boric acid-sodium borate, pH 7.5, 700 V, 70 min): 1.14. Anal. Calcd for C₁₁H₁₃N₅O₅ · 1/4 H₂O: C, 44.08; H, 4.54; N, 23.36. Found: C, 44.08; H, 4.32; N, 23.08.

2-Acetamido-(and 2-amino)-6-ethoxy-8,2'-methano-9-[3,5-O-TIPDS- α -D-arabinofuranosyl]purine (13)—Compound 7 (340 mg, 0.474 mmol) and trimethyl phosphite (1 ml) in acetonitrile (300 ml) were irradiated for 1.5 h at room temperature. The solvent was removed *in vacuo* and the residue was applied to a column of silica gel (30 g). The eluate with 3—4% EtOH in CHCl₃ was concentrated to leave 13 (R=Ac and R=H, 46: 54, 202 mg, 73%). A part of 13 was separated by PTLC as in the case of 8 and 9. Physical constants of 13 (R=Ac). UV $\lambda_{\max}^{\text{MeOH}}$ nm: 273; $\lambda_{\max}^{\text{H}^+}$: 270, 254. MS m/z : 607 (M⁺), 564 (M - isoPr). NMR (CDCl₃) δ : 8.02 (1H, s, HN), 5.92 (1H, s, H-1'), 5.22 (1H, br s, HO-2'), 4.53 (3H, m, H-3', CH₃CH₂O), 3.98—3.79 (3H, m, H-5'ab, 6'a), 3.46 (1H, m, H-4', $J_{3',4'} = 10$ Hz), 2.87 (1H, d, H-6'b, $J_{6'a,b} = 19$ Hz), 2.51 (3H, s, Ac), 1.49 (3H, t, CH₃CH₂O), 1.16—1.04 (28H, m, isoPr). Physical constants of 13 (R=H). UV $\lambda_{\max}^{\text{MeOH}}$ nm: 284, 247; $\lambda_{\max}^{\text{H}^+}$: 292, 245. MS m/z : 565 (M⁺), 522 (M - isoPr). NMR (CDCl₃) δ : 5.79 (1H, s, H-1'), 5.52 (1H, br s, HO-2'), 5.02 (2H, br s, H₂N), 4.64—4.36 (3H, m, H-3', CH₃CH₂O), 3.95 (2H, m, H-5'ab), 3.80 (1H, d, H-6'a, $J_{6'a,b} = 19.0$ Hz), 3.50 (1H, m, H-4', $J_{3',4'} = 9.3$ Hz), 2.78 (1H, d, H-6'b), 1.45 (3H, t, CH₃CH₂O), 1.15—1.03 (28H, m, isoPr).

2-Amino-6-ethoxy-8,2'-methano-9-[3,5-di-O-acetyl- α -D-arabinofuranosyl]purine (14)—Compound 13 (173 mg) was converted to 14 (56 mg, 46%) by the method described for the preparation of 10. UV $\lambda_{\max}^{\text{MeOH}}$ nm (ϵ): 284 (9500), 247 (11700); $\lambda_{\min}$: 263 (4640), 224 (4640). CD (MeOH) [θ] (nm): +5530 (284), +2460 (262), +6840 (246), +1760 (225). MS m/z : 407 (M⁺), 392 (M - Me). NMR (CDCl₃) δ : 6.09 (1H, s, H-1'), 5.02 (2H, br s, H₂N), 4.95 (1H, d, H-3'), 4.52 (2H, q, CH₃CH₂O), 4.36—3.95 (3H, m, H-4', 5'ab), 3.40 (1H, d, H-6'a, $J_{6'a,b} = 18.3$ Hz), 2.92 (1H, d, H-6'b), 2.23, 2.11 (3H each, s, Ac), 1.45 (3H, t, CH₃CH₂O).

9-(α -D-Arabinorufanosyl)-8,2'-methanoguanine (15)—Compound 14 (50 mg) was treated with chlorotrimethylsilane (78 μ l) and NaI (101 mg) in acetonitrile (4 ml) for 4 h, then H₂O (0.5 ml) and Et₃N (94 μ l) were added to the mixture. The solvent was removed and the residue was subjected to PTLC (*n*-BuOH-H₂O, 86:14). The appropriate band was extracted with 30% MeOH-CHCl₃. The solvent was removed *in vacuo* to leave 18 mg of the de-ethyl derivative. This compound was taken up in 20% Et₃N-MeOH (5 ml) and the suspension was refluxed for 3 h. The solvent was removed and the residue was crystallized from aqueous EtOH to give 15 (5.3 mg, 15%), mp 300 °C. UV $\lambda_{\max}^{\text{H}_2\text{O}}$ nm: 278, 249. CD (H₂O) [θ], assuming ϵ_{249} as 14800 (nm): +2440 (252). FD-MS m/z : 295 (M⁺). NMR (D₂O) δ : 5.78 (1H, s, H-1'), 4.05 (1H, d, H-3', $J_{3',4'} = 9.3$ Hz), 3.70—3.49 (3H, m, H-4', 5'ab), 3.52 (1H, d, H-6'a), 2.71 (1H, d, H-6'b, $J_{6'a,b} = 18.6$ Hz). Relative migration with respect to guanosine on paper electrophoresis (0.2 M boric acid-sodium borate, pH 7.5, 700 V, 70 min): 0.34.

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