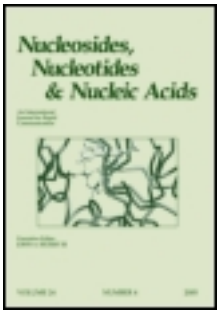

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Synthesis and Biological Evaluation of 4′-C-Methyl Nucleosides

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SYNTHESIS AND BIOLOGICAL EVALUATION OF 4'-*C*-METHYL NUCLEOSIDES†

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Abstract: A series of 2'-deoxy, 2',3'-unsaturated and 2',3'-dideoxynucleoside analogues, which have an additional methyl group at the 4'-position, have been synthesized. When evaluated for their inhibitory activity against HIV in MT-4 cell, 2'-deoxy-4'-C-methyl nucleosides exhibited potent activity.

Several types of viral infection cause severe diseases, including cancer. The HIV, the causative agent of AIDS, has recently spread all over the world to become a serious life-threatening disease. Therefore, the development of drugs to prevent such diseases is an important research subject. A number of sugar-modified nucleosides, especially since the discovery of AZT, 2',3'-dideoxy nucleosides and 2',3'-didehydro-dideoxy analogs as chemotherapeutic agents for AIDS, have been synthesized[1] to test against such diseases. After conversion to the triphosphate in cells, these analogs may either inhibit HIV reverse transcriptase, or be incorporated by it into the growing chain of DNA, resulting in chain termination or disruption.

Modification of the sugar fragment of 2'-deoxy nucleosides has resulted in some very potent inhibitors. Recently, Magg et al.[2] reported on the synthesis and anti-HIV activity of various 4'-azido- and 4'-methoxynucleosides with electronegative substituents at 4'-position. In the previous paper[3], we described the preparation of the 4'-modified nucleoside that have an additional C-C linked methyl group. The 4'-*C*-methyl nucleoside analogs are particularly interesting because the reactivities of 3' and 5'-hydroxyl groups differ from those of natural nucleosides due to the neopentyl character and further the steric effect of the 4'-*C*-methyl group can be also expected.

† This paper is dedicated to Dr. Yoshihisa Mizuno on the occasion of his 75th birthday .

In this paper, we report the syntheses and the HIV-inhibitory activity of the 4'-*C*-methyl-2'-deoxy nucleosides to evaluate the effect of 4'-*C*-methyl group and also report the preparation of the unsaturated (D4) and dideoxy (DD) derivatives of them which are expected to act as the chain terminators of viral DNA synthesis. Our target nucleosides are shown in Fig. 1.

RESULTS AND DISCUSSION

CHEMISTRY

Since we have reported[3] in detail the preparation of 4-*C*-methyl-D-ribose derivatives **1**, we would like to discuss here the preparation of nucleosides from **1**.

The application of Saneyoshi's protocol[4] to the 4-*C*-methyl ribose derivatives **1** gave an unseparable mixture (50:50) of 9- (**2a**) and 7-adenyl derivative (**2b**) in a 26% yield (Fig. 2). The change of the catalyst from stannic chloride to trimethylsilyltrifluoromethanesulfonate (TMSOTf) improved the yield up to 72% but the ratio of **2a** and **2b** could not be improved (58:42). Since the transition state of this glycosylation reaction can be speculated as shown in Fig. 3, we expected that the ratio of 9 and 7-adenylation could be change by the bulkiness of the 6-substituent of adenine. Thus, the glycosylation reaction was also carried out with N^6-benzoyladenine. Condensation of **1** with N^6-benzoyladenine in dry acetonitrile in the presence of TMSOTf at 0 °C gave a mixture of 9-adenyl derivative **3a** (74%) and 7-adenyl derivative **3b** (8.9%). These compounds were separated by silica gel column chromatography.

Treatment of **3a** with saturated methanolic ammonia at room temperature for 18 h and subsequent deoxygenation by Robins's protocol[5] gave 2'-deoxy derivative (**6**) in a 40% yield. Birch reduction of **6** gave 2'-deoxy-4'-*C*-methyladenosine (**7**) in an 87% yield (Scheme 1). In the deoxygenation procedure, use of excess phenoxythiocarbonyl chloride gave 4'-*C*-methyl purine derivative **8** in a 12% yield.

Thymidine derivative was also prepared from **9** by similar procedure to that described for 2'-deoxyadenosine derivative **7** (Scheme 2). The catalytic hydrogenation of **12** gave **13** in a 23% yield. However, the hydrogenolysis of benzyl groups did not proceed completely, due to the presence of a small amount of residual tin compound. Therefore, we chose a different route in which the *O*-debenzylation was performed before the deoxygenation of 2'-hydroxyl group. Mansuri[6] reported the preparation of 3',5'-di-*O*-acetyl-2'-bromothymidine in a 97% yield by treatment of ribofuranosylthymine with acetyl bromide. Thus, treatment of thymidine derivative **14**[3] with acetyl bromide in dry acetonitrile at reflux gave **15** and subsequent reduction of **15** with tributyltin hydride in toluene-benzene in the presence of 2,2'-azobisisobutyronitrile (AIBN) gave **16** in a 72% yield. The major by-product of this sequence was 2,2'-anhydro derivative **17**, which were formed during the reaction with acetyl bromide. Treatment of **16** with saturated methanolic ammonia at room temperature for 18 h gave 4'-*C*-methylthymidine (**13**) in a 62% yield (Scheme 3).

The approach to 4'-*C*-methyl-2'-deoxycytidine utilized a similar procedure to that described for 4'-*C*-methylthymidine (**13**). Compound **20**, which was prepared from 4'-*C*-methyluridine

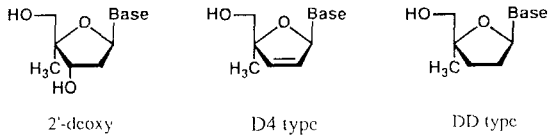


FIG. 1

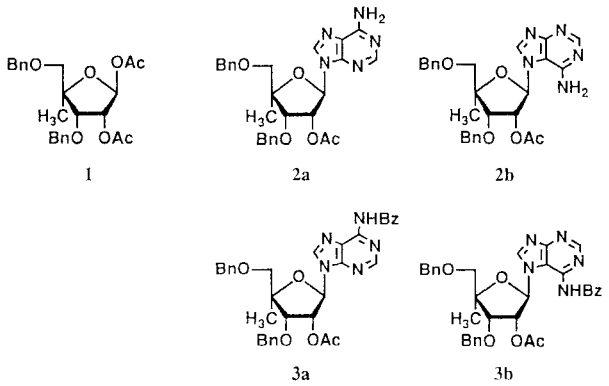


FIG. 2

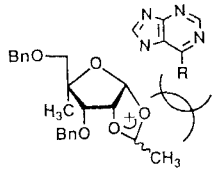
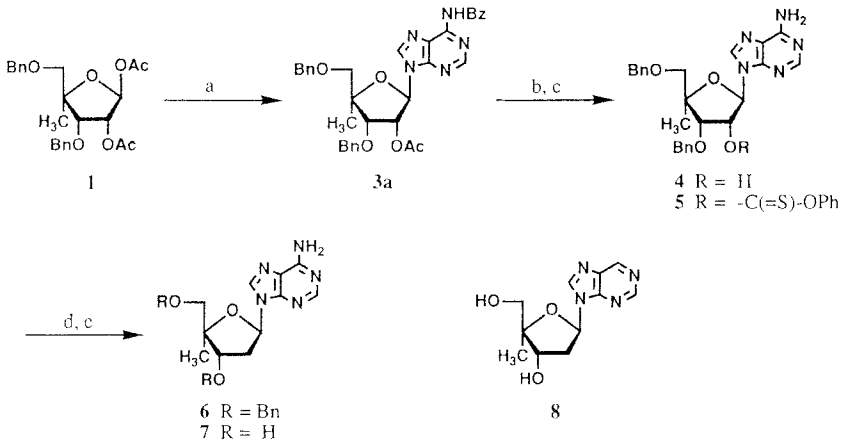


FIG. 3

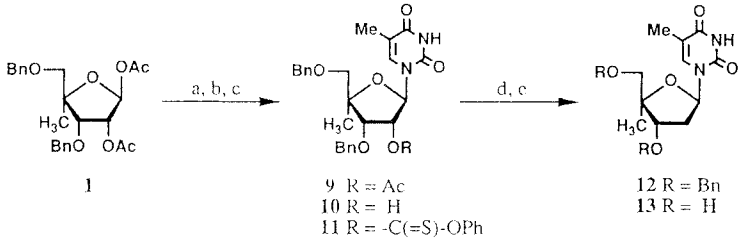
(**18**)[3] via its bromo acetyl derivative **19**, was converted to 2'-deoxy-4'-*C*-methylcytidine (**23**) by Reese's method [7] through 1,2,4-triazolyl derivative **22** in a 68% yield (*Scheme* 3).

Next we turned our attention to the preparation of D4- and DD-derivatives. One of the method to prepare D4-derivatives is to convert the vicinal 2',3'-diol to vicinal bromoacetate and reduce it with Zn/Cu reagent. However, treatment of 4'-*C*-methyladenosine (**24**)[3] with 2-acetoxyisobutyl bromide (Mattocks's bromide) gave a very complex mixture including compound **25**. Therefore, in the case of **24**, we selected the Corey's direct conversion of vicinal diols to double bond. Reaction of **24** with *tert*-butyldimethylsilyl chloride (TBDMSCl) in N,N-dimethylformamide (DMF) in the presence of imidazole at 60°C gave 5'-*O*-silyl protected derivative **26** in a 65%



a) N^6-Bz-Ad, TMSOTf / CH_3CN, b) NH_3-MeOH, c) PhOC(=S)-Cl, DMAP / CH_3CN, d) n-Bu_3SnH, AIBN / $PhCH_3$, e) Na / liq. NH_3

SCHEME 1

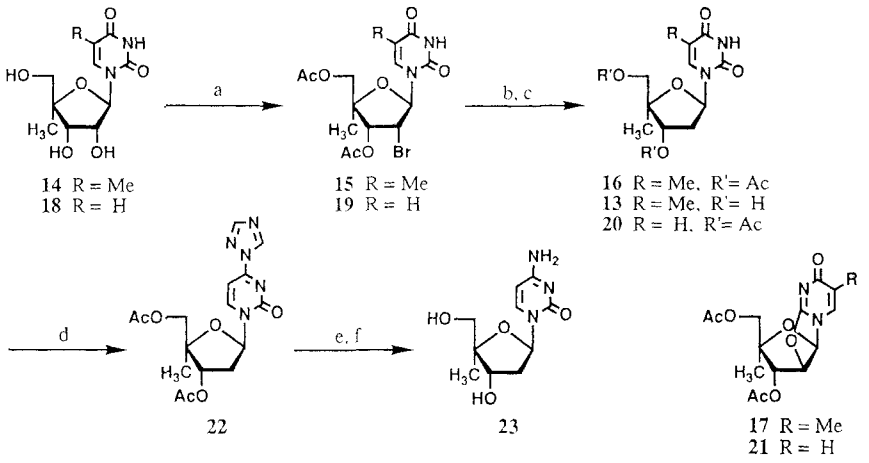


a) TMS-Th, TMSOTf / DCE, b) NH_3 / MeOH, c) PhOC(=S)-Cl, DMAP / CH_2Cl_2, d) n-Bu_3SnH, AIBN / $PhCH_3$, e) H_2, Pd-black / MeOH

SCHEME 2

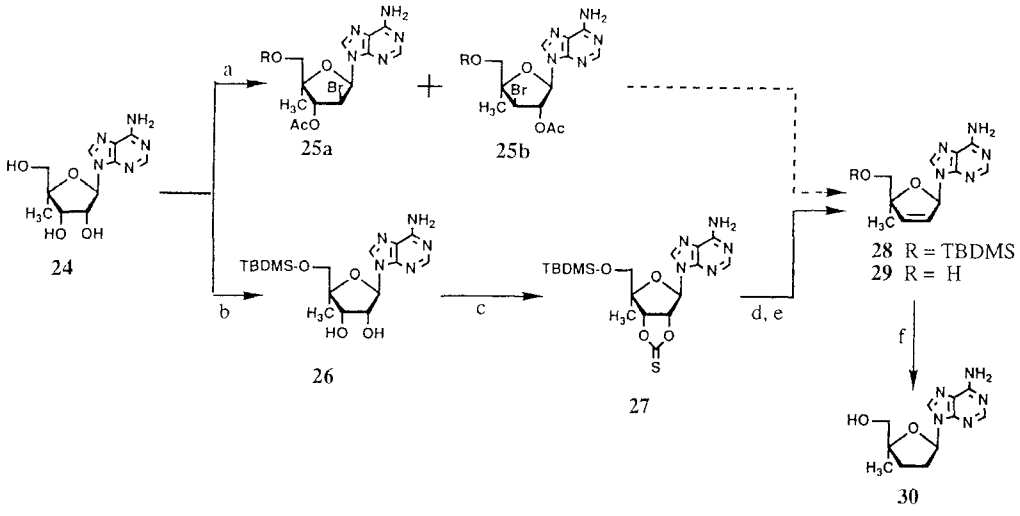
yield. Thionocarbonate **27**, which was prepared by the reaction of **26** with thiocarbonyldiimidazole in a 88% yield, was deoxygenated to **28** by treatment with 1,3-dimethyl-2-phenyl-1,3,2-diazaphospholidine in a 63% yield. Deprotection of **28** with 1M tetrabutylammonium fluoride-tetrahydrofurane (THF) gave the desired D4A analogue (**29**) in a 74% yield. DDA analogue **30** was prepared by hydrogenation of **29** over Pd-black in methanol in a 68% yield (Scheme 4).

The syntheses of D4T and DDT analogues followed the Mansuri's protocol[6]. Reaction of **15** with Zn/Cu couple in methanol gave olefinic product **31** in a 28% yield, which was *O*-



a) AcBr / CH_3CN, b) n-Bu_3SnH, AIBN / $PhCH_3$ - PhH, c) NH_3 / MeOH, d) ClPh-$POCl_2$, Tz / pyr, e) NH_4OH / Dioxane, f) NH_3 / MeOH

SCHEME 3



a) Mattocks-Br / CH_3CN, b) TBDMSCl, Im / DMF, c) $Im_2C{=}S$ / DMF, d) DMPDAP / THF, e) Bu_4NF / THF, f) H_2, Pd-black / MeOH

SCHEME 4

deacetylated to give D4T analogue **32** in a 98% yield. Hydrogenation of **31** over 10% Pd-carbon followed by deacetylation gave DDT analogue **34** in a 81% yield (Scheme 5).

The reaction of cytidine derivative **35** with Mattock's reagent did not give the desired bromoacetate similar with adenosine derivative **24**. The preparation of 2',3'-unsaturated nucleoside via cyclic thionocarbonate was also applied to cytidine derivative to give **39** in a 24% yield from **35**, which on desilylation with tetrabutylammonium fluoride, followed by *N*-deacetylation gave D4C analogue **41** in a 69% yield (Scheme 6). However, attempted reduction of **40** to the corresponding saturated cytidine analogue (**44**) was not successful. Therefore, DDC analogue **44** was synthesized from **43**, which was prepared from 4'-*C*-methyluridine (**18**)[3] by Mansuri's protocol[6] via D4U analogue **42** in a 26% overall yield, by the Reese's method through 1,2,4-triazolyl derivative in a 21% yield (Scheme 5).

The 2,2'-anhydro derivative **21**, which was obtained as by-product in the preparation of **20**, was converted to ara-C analogue **46** in a 49% yield through **45** (Scheme 7).

BIOLOGICAL ACTIVITY

The 4'-*C*-methyl nucleosides were evaluated for their in vitro inhibitory effect on the cytopathicity of HIV (HTLV-III$_B$ strain) in MT-4 cells with AZT, DDT and DDA as references.

All of the 2'-deoxy derivatives were potent inhibitors of HIV. IC_{50}'s ranged from 0.072 μM for cytidine derivative (**23**) to 50 μM for purine derivative (**8**). Thymidine derivative (**13**) was found to inhibit HIV replication at a concentration that was 14-fold below the cytotoxicity threshold. It was found that, although the cytidine derivative (**23**) was 100-fold more potent than compound **13**, it was also the most cytotoxic compound among the 4'-*C*-methyl nucleosides tested. Compound **23** was also tested for its ability to inhibit the growth of P388 mouse leukemia cells in vitro and was proved to have markedly effective activity (IC_{50} = 1.7 μM).

Within the class of dehydro-dideoxy derivatives, D4T analogue (**32**) was shown a substantial level of anti-HIV activity, and D4A and D4C analogues (**29**, **41**) were inactive.

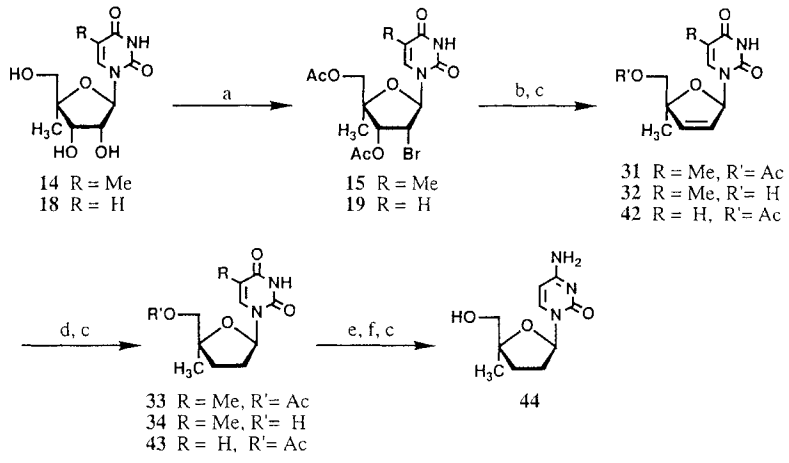
The arabinoside **46** was higher cytotoxity than 2'-deoxy derivative **23**.

It has been a common knowledge that D4 and DD types of sugar modified nucleosides exhibit a significant anti-HIV activity regardless the difference of nucleobases, and the nucleoside derivatives must be devoid of the 3'-hydroxyl group to be potential anti-HIV.

Some results presented here are in contrast with the general knowledge of the structure-activity relationships. The contrasts are as follows. 1) In the case of adenosine analogues, D4 type was found to be inactive and DD type exhibited good anti-HIV activity. On the other hand, in the thymidine series, DD type was inactive and D4 type showed a significant activity. 2) The 2'-deoxy type was found to be more potent than the corresponding D4 and DD types.

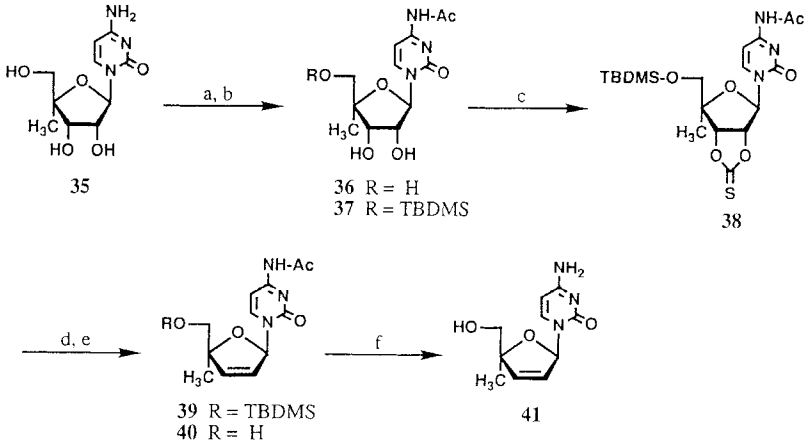
Although the reasons for these results are not clear at the present time, these structure-activity relation is very interesting and useful to design new potential anti viral nucleosides.

Lastly, we also showed that the deletion of the 3'-hydroxyl group is not indispensable for anti-HIV activity as in the case of Magg et al.[2]



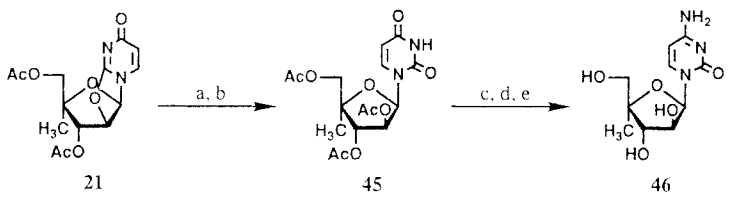
a) AcBr / CH_3CN, b) Zn-Cu / MeOH, c) NH_3 / MeOH, d) H_2, 10%Pd-C / MeOH, e) $ClPhP(=O)Cl_2$, Tz / pyr, f) NH_4OH / Dioxane

SCHEME 5



a) Ac_2O / MeOH, b) TBDMSCl, Im / DMF, c) $Im_2C=S$ / DMF, d) DMPDAP / THF, e) Bu_4NF / THF, f) NH_3 / MeOH

SCHEME 6



21 45 46

a) 1N NaOH / 50% EtOH, b) Ac_2O / pyr, d) $ClPhPOCl_2$, Tz / pyr, d) NH_4OH / Dioxane, e) NH_3 / MeOH

SCHEME 7

TABLE In vitro antiviral activity of 4'-*C*-methyl nucleosides against HIV-1 in MT-4 cells.

Comp.	IC_{50} (μM)	CC_{50} (μM)	Index
13	7.2	104	14
23	0.072	0.13	1.8
8	50	50	1.0
29	> 500	> 500	
32	21	330	16
41	350	350	1.0
30	30	400	13
34	> 500	> 500	
46	toxic	1.2	
AZT	0.0099	20	2020
DDA	47	> 500	> 11
DDT	4.1	> 500	> 120

EXPERIMENTAL

Melting point (mp) values were taken with a Shibata melting apparatus and are uncorrected. ^{1}H-NMR spectra were recorded with a JEOL JNMEX-270 spectrometer at 28 °C, in $CDCl_3$using Me_4Si or D_2O using DSS as an internal standard. UV spectra were recorded with a HITACHI U-2000 spectrophotometer. Mass spectra were recorded with a HITACHI M-80B spectrometer at 70eV. IR spectra were recorded on with a SHIMAZU FTIR-8100M spectrometer. Specific rotation values were mesured with a JASCO DIP-360 at 589nm. Merck silica gel Art. 9385 was used for column chromatography and Merck silica gel Art. 5554 for analytical thin layer chromatography.

N^6-Benzoyl-2'-*O*-acetyl-3',5'-di-*O*-benzyl-4'-*C*-methyladenosine (3a). N^6-Benzoyladenine (2.2 g, 9.34 mmol) was suspended in a solution of **1**[3] (4.0 g, 9.34 mmol) in dry acetonitrile (200 ml) at 0 °C. A solution of TMSOTf (3.6 ml, 18.7 mmol) in dry acetonitrile (80 ml) was added and the mixture was stirred at the same temperature. After 1 h, sodium bicarbonate (2.0 g) was added to a reaction mixture and filtered. The filtrate was evaporated and the residue was partitioned with $CHCl_3$ and H_2O. The $CHCl_3$ layer was dried ($MgSO_4$) and evaporated. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt-hexane (50:1:30:10) as an eluent, to give **3a** (5.4 g, 95%) as a pale yellow syrup, $[\alpha]_D^{25}$ +8.9° (*c* 1.0, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 9.21 (1 H, br s, NH), 8.76 (1 H, s, 2), 8.36 (1 H, s, 8), 8.02 (2 H, d, *J* = 7.1 Hz, Bz), 7.52 (3 H, m, Bz), 7.32 (10 H, m, Ph), 6.36 (1H, d, *J* = 4.2 Hz, 1'), 5.89 (1 H, dd, *J* = 4.2, 5.4 Hz, 2'), 4.62 (1 H, d, *J* = 11 Hz, Bn), 4.55 (1 H, d, *J* = 5.4 Hz, 3'), 4.51 (1 H, d, *J* = 11 Hz, Bn), 4.49 (1 H, d, *J* = 12 Hz, Bn), 4.44 (1 H, d, *J* = 12 Hz, Bn), 3.56 (1 H, d, *J* = 10 Hz, 5'), 3.37 (1 H, d, *J* = 10 Hz, 5'), 2.10 (3 H, s, Ac), 1.35 (3 H, s, Me); IR ν_{max} (neat) cm^{-1}: 3200, 3000, 1740, 1700, 1600 1570, 1500, 1440, 1220, 1090, 1020, 900, 800, 740, 700; EI-MS *m/z*: 607 (M^+); HR-MS calcd for $C_{34}H_{33}N_5O_6$ (M^+) 607.2429, found 607.2432.

3',5'-Di-*O*-benzyl-2'-deoxy-4'-*C*-methyladenosine (6). A solution of **3a** (5.4 g, 8.89 mmol) in saturated methanolic ammonia (350 ml) was stirred at room temperature for 18 h and then evaporated to dryness. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt-hexane (20:2:15:5) as an eluent, to give **4** (3.5 g, 86%) as crystal. To a stirred suspension of **4** (4.4 g, 9.53 mmol) and 4-(dimethylamino)pyridine (3.5 g, 28.6 mmol) in dry acetonitrile (250 ml), phenylchlorothionocarbonate (2.0 ml, 14.3 mmol) was added dropwise under nitrogen at room temperature. After 22 h, the solvent was evaporated to dryness in vacuo to give a residue which was partitioned between $CHCl_3$ and H_2O. The $CHCl_3$ layer was dried ($MgSO_4$) and evaporated. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt-hexane (50:1:30:10) as an eluent, to give **5** (5.0 g, 90 %) as a pale yellow syrup. A stirred solution of tributyltin hydride (6.6 g, 22.6 mmol) and AIBN (1.4 g, 8.37 mmol) in dry toluene (200 ml) was heated at 60 °C. A solution of **5** (5.0 g, 8.37 mmol) in dry benzene (150 ml) was added dropwise to an above hot solution. The resulting solution was heated under reflux for 4 h, and then the solvent was evaporated. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt-hexane (50:1:30:10) as an eluent, to give **6** (2.0 g, 52%) as a pale yellow syrup, $[\alpha]_D^{25}$ +9.0° (*c* 0.6, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.33 (1 H, s, 2), 8.17 (1 H, s, 8), 7.33 (10 H, m, Ph), 6.42 (1 H, dd, *J* = 5.9, 6.4 Hz, 1'), 5.56 (2 H, br s, NH_2), 4.61 (1 H, d, *J* = 12 Hz, Bn), 4.56 (1 H, d, *J* = 12 Hz, Bn), 4.51 (2 H, d, *J* = 12 Hz, Bn), 4.41 (1 H, t, *J* = 5.9 Hz, 3'), 3.59 (1 H, d, *J* = 10 Hz, 5'), 3.46 (1 H, d, *J* = 10 Hz, 5'), 2.70 (2 H, m, 2'), 1.33 (3 H, s, Me); IR ν_{max} (neat) cm^{-1}: 3300, 3150, 2900, 1640, 1600, 1470, 1420, 1330, 1300, 1210, 1100, 740, 700; EI-MS *m/z*: 445 (M^+); HR-MS calcd for $C_{25}H_{27}N_5O_3$ (M^+) 445.2144, found 445.2173.

2'-Deoxy-4'-*C*-methyladenosine (7). Sodium (260 mg, 11.2 mmol) was added to a stirred suspension of **6** (1.0 g, 2.24 mmol) in liquid ammonia (100 ml) at -78 °C, and the mixture was stirred for 10 min at the same temperature. Ammonium chloride was added to the mixture, and the mixture was evaporated. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH (5:1) as an eluent, to give **7** (520 mg, 87%) as a crystal, mp 128 °C, $[\alpha]_D^{25}$ -5.2° (*c* 1.0, MeOH), ^{1}H-NMR (D_2O) δ: 8.29 (1 H, s, 2), 8.17 (1 H, s, 8), 6.41 (1 H, t, *J* = 6.6 Hz, 1'), 4.62 (1 H, dd, *J* = 5.3, 5.9 Hz, 3'), 3.66 (2 H, s, 5'), 2.93 (1 H, ddd, *J* = 5.9, 6.4, 14 Hz, 2'), 2.64 (1 H, ddd, *J* = 5.3, 6.6, 14 Hz, 2'), 1.29 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3400, 3250, 1650, 1620, 1590, 1500, 1440, 1350, 1320, 1260, 1220, 1060, 980, 810; EI-MS *m/z*: 265 (M^+); HR-MS calcd for $C_{11}H_{15}N_5O_3$ (M^+) 265.1175, found 265.1170; UV λ_{max} (H_2O) nm (ε): 260 (11900), 209 (13900).

9-(2-Deoxy-4-*C*-methyl-β-D-ribofuranosyl)purine (8). Compound **8** was prepared from crude **6** in a 12% yield by the similar procedure to that described for **7**. **8**: mp 170-171 °C, $[\alpha]_D^{25}$ +3.1° (*c* 0.6, MeOH), ^{1}H-NMR (D_2O) δ: 9.13 (1 H, s, 2), 8.95 (1 H, s, 8), 8.71 (1 H, s, 6), 6.59 (1 H, t, *J* = 6.3 Hz, 1'), 4.70 (1 H, dd, *J* = 5.6, 6.3 Hz, 3'), 3.67 (2 H, s, 5'), 3.05 (1 H, dt, *J* = 6.3, 14 Hz, 2'), 2.71 (1 H, ddd, *J* = 5.6, 6.3, 14 Hz, 2'), 1.31 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3320, 3080, 1600, 1500, 1410, 1300, 1220, 1150, 1090, 1070, 1040, 950, 760, 740; EI-MS *m/z*: 250 (M^+); HR-MS calcd for $C_{11}H_{14}N_4O_3$ (M^+) 250.1065, found 250.1071; UV λ_{max} (H_2O) nm (ε): 263 (8380), 197 (26200).

1-(2-*O*-Acetyl-3,5-di-*O*-benzyl-4-*C*-methyl-β-D-ribofuranosyl)thymine (9). A mixture of thymine (1.2 g, 9.34 mmol), hexamethyldisilazane (2.0 ml) and trimethylchlorosilane (0.5 ml) was refluxed overnight and then concentrated. To a solution of the silylated thymine and **1**[3] (2.0 g, 4.67 mmol) in 1,2-dichloroethane (20 ml) was added TMSOTf (1.4 ml, 7.00 mmol) in 1,2-dichloroethane (5 ml). The mixture was stirred for 1 h at room temperature and diluted with $CHCl_3$. The mixture was washed with water, dried over $MgSO_4$ and evaporated. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt-hexane (50:1:20:10) as an eluent. Evaporation of the appropriate fractions and crystallization of the residue from AcOEt-hexane yielded 2.2 g (95%) of **9** as crystal, mp 156 °C, $[\alpha]_D^{25}$ +51.6° (*c* 1.0, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.49 (1 H, br s, NH), 7.52 (1 H, s, 6), 7.33 (10 H, m, Ph), 6.20 (1 H, d, J = 4.6 Hz, 1'), 5.38 (1 H, dd, J = 4.6, 5.9 Hz, 2'), 4.63 (1 H, d, J = 12 Hz, Bn), 4.48 (3 H, m, Bn), 4.31 (1 H, d, J = 5.9 Hz, 3'), 3.61 (1 H, d, J = 10 Hz, 5'), 3.38 (1 H, d, J = 10 Hz, 5'), 2.11 (3 H, s, Ac), 1.51 (3 H, s, Me), 1.28 (3 H, s, 4'Me); IR ν_{max} (KBr) cm^{-1}: 3150, 3050, 2850, 1750, 1710, 1680, 1460, 1370, 1280, 1230, 1100, 1060, 1010, 750, 700; EI-MS *m/z*: 494 (M^+); Calcd for $C_{27}H_{30}N_2O_7$: C, 65.58; H, 6.12; N, 5.67, *Anal.* Found: C, 65.39; H, 5.96; N, 5.73.

3',5'-Di-O-benzyl-4'-*C*-methylthymidine (12). Employing conditions analogous to those used for the preparation of **6**, **9** (810 mg, 1.80 mmol) gave **12** (250 mg, 32%) as a pale yellow syrup via **10** and **11**. **12**: $[\alpha]_D^{25}$ +50.6° (*c* 1.0, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.27 (1 H, br s, NH), 7.66 (1 H, d, *J* = 1.2 Hz, 6), 7.32 (10 H, m, Ph), 6.27 (1 H, t, *J* = 6.1 Hz, 1'), 4.62 (1 H, d, *J* = 12 Hz, Bn), 4.56 (1 H, d, *J* = 12 Hz, Bn), 4.52 (1 H, d, *J* = 12 Hz, Bn), 4.46 (1 H, d, *J* = 12 Hz, Bn), 4.29 (1 H, dd, *J* = 5.6, 6.6 Hz, 3'), 3.65 (1 H, d, *J* = 10 Hz, 5'), 3.49 (1 H, d, *J* = 10 Hz, 5'), 2.51 (1 H, ddd, *J* = 5.6, 6.6, 14 Hz, 2'), 2.26 (1 H, ddd, *J* = 5.9, 6.6, 14 Hz, 2'), 1.55 (3 H, d, *J* = 1.2 Hz, Me), 1.26 (3 H, s, 4'Me); IR ν_{max} (neat) cm^{-1}: 3150, 3050, 2900, 1690, 1460, 1360, 1290, 1200, 1100, 860, 750, 700; EI-MS *m/z*: 436 (M^+); HR-MS calcd for $C_{25}H_{28}N_2O_5$ (M^+) 436.1997, found 436.1981.

4'-*C*-Methylthymidine (13) : From 3',5'-di-*O*-benzyl-4'-*C*-methylthymidine (12). A solution of **12** (220 mg, 0.50 mmol) in MeOH (40 ml) was shaken under hydrogen with Pd-black (100 mg) for 3 h. The catalyst was removed by filtration, and the filtrate was evaporated. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH (7:1) as an eluent, to give **13** (30 mg, 23%) as a crystal and starting material **12** (65 mg). **From 3',5'-Di-*O*-acetyl-4'-*C*-methylthymidine (16).** A solution of **16** (640 mg, 1.88 mmol) in saturated methanolic ammonia (40 ml) was stirred at room temperature for 18 h and then evaporated to dryness. The residue was recrystalized from MeOH-Et_2O to give **13** (300 mg, 62%) as a crystal, mp 167 °C, $[\alpha]_D^{25}$ +33.6° (*c* 1.1, MeOH), ^{1}H-NMR (D_2O) δ: 7.70 (1 H, d, *J* < 1 Hz, 6), 6.24 (1 H, t, *J* = 6.3 Hz, 1'), 4.48 (1 H, t, *J* = 6.3 Hz, 3'), 3.65 (2 H, s, 5'), 2.48 (2 H, t, *J* = 6.3 Hz, 2'); IR ν_{max} (KBr) cm^{-1}: 3500, 3200, 2950, 1700, 1470, 1400, 1290, 1200, 1130, 1070, 1010, 960, 850, 820, 780; EI-MS *m/z*: 256 (M^+); HR-MS calcd for $C_{11}H_{16}N_2O_5$ (M^+) 256.1057, found 256.1061; UV λ_{max} (MeOH) nm (ε): 267 (9030).

3',5'-Di-*O*-acetyl-4'-*C*-methylthymidine (16) and 2,2'-Anhydro-3',5'-di-*O*-acetyl-4'-*C*-methylthymidine (17). Acetyl bromide (1.7 ml, 21.3 mmol) was added dropwise over 0.5 h to a suspension of 1-(4-methyl- β-D-ribofuranosyl)thymine (**14**)[3] (1.0 g, 3.67 mmol) in acetonitrile (45 ml) heated at reflux. On completion of addition, the solution was allowed to cool and then concentrated. The residue was dissolved in methylene chloride (100 ml) and washed with water. The organic phase was dried (Na_2SO_4) and concentrated to leave crude **15**. The solution of crude **15** in dry benzene (10 ml) was added dropwise to a stirred hot mixture of tributyltin hydride (2.1 g, 7.35 mmol) and AIBN (600 mg) in dry toluene (20 ml) at 60 °C. The resulting mixture was refluxed for 4 h and then evaporated under reduced pressure. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt-hexane (40:1:30:10) as an eluent, to give two fractions. The first-eluted fraction gave **16** (900 mg, 72%) as a colorless syrup, $[\alpha]_D^{25}$ +15.7° (*c* 0.7, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 9.35 (1 H, br s, NH), 7.34 (1 H, d, *J* = 1.3 Hz, 6), 6.29 (1 H, dd, *J* = 6.3, 7.3 Hz, 1'), 5.33 (1 H, dd, *J* = 3.6, 6.9 Hz, 3'), 4.21 (1 H, d, *J* = 12 Hz, 5'), 4.16 (1 H, *J* = 12 Hz, 5'), 2.46 (1 H, ddd, *J* = 3.6, 6.3,

14 Hz, 2'), 2.34 (1 H, ddd, J = 6.9, 7.3, 14 Hz, 2'), 2.14 (6 H, s, Ac), 1.94 (3 H, d, J = 1.3 Hz, Me), 1.28 (3 H, s, 4'Me); IR ν_{max} (neat) cm^{-1}: 3190, 3050, 1750, 1690, 1470, 1380, 1240, 1120, 1050, 960; EI-MS *m/z*: 340 (M^+); HR-MS calcd for $C_{15}H_{20}N_2O_7$ (M^+) 340.1268, found 340.1240. The second fraction gave **17** (220 mg, 18%) as a crystal, mp 199-200°C, $[\alpha]_D^{25}$ -61.0° (*c* 1.0, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 7.19 (1 H, d, J = 1.3 Hz, 6), 6.17 (1 H, d, J = 6.3 Hz, 1'), 5.57 (1 H, br s, 3'), 5.34 (1 H, dd, J = 6.3, <1 Hz, 2'), 4.15 (1 H, d, J = 12 Hz, 5'), 3.86 (1 H, d, J = 12 Hz, 5'), 2.19 (3 H, s, Ac), 2.00 (3 H, s, Ac), 1.98 (3 H, d, J = 1.3 Hz, Me), 1.26 (3 H, s, 4'Me); IR ν_{max} (KBr) cm^{-1}: 3070, 1750, 1670, 1620, 1560, 1490, 1380, 1450, 1220, 1070, 1040, 1030, 1000, 790; EI-MS m/z: 338 (M^+); HR-MS calcd for $C_{15}H_{18}N_2O_7$ (M^+) 338.1111, found 338.1111.

3',5'-Di-*O*-acetyl-2'-deoxy-4'-*C*-methyluridine (20) and 2,2'-Anhydro-3',5'-di-*O*-acetyl-2'-deoxy-4'-*C*-methyluridine (21). Employing conditions analogous to those used for the preparation of **16**, **18**[3] (1.08 g, 4.18 mmol) gave **20** (910 mg, 67%) as crystal and **21** (300 mg, 22%) as crystal via **19**. **20**: mp 141 °C, $[\alpha]_D^{25}$ +39.0° (*c* 1.0, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.29 (1 H, br s, NH), 7.55 (1 H, d, J = 7.9 Hz, 6), 6.25 (1 H, t, J = 6.6 Hz, 1'), 5.76 (1 H, dd, J = 2.0, 7.9 Hz, 5), 5.32 (1 H, dd, J = 3.6, 6.6 Hz, 3'), 4.19 (1 H, d, J = 12 Hz, 5'), 4.14 (1 H, d, J = 12 Hz, 5'), 2.52 (1 H, ddd, J = 3.6, 6.6, 14 Hz, 2'), 2.35 (1 H, dt, J = 6.6, 14 Hz, 2'), 2.13 (3 H, s, Ac), 2.11 (3 H, s, Ac), 1.29 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3040, 1740, 1700, 1670, 1460, 1390, 1230, 1200, 1130, 1110, 1060, 1030, 860; EI-MS *m/z*: 326 (M^+); HR-MS calcd for $C_{14}H_{18}N_2O_7$ (M^+) 326.1112, found 326.1083. Compound **21**: mp 181 °C, $[\alpha]_D^{25}$ -52.3° (*c* 0.8, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 7.32 (1 H, d, J = 7.6 Hz, 6), 6.19 (1 H, d, J = 6.3 Hz, 1'), 6.07 (1 H, d, J = 7.6 Hz, 5), 5.58 (1 H, s, 3'), 5.37 (1 H, d, J = 6.3 Hz, 2'), 4.16 (1 H, d, J = 12 Hz, 5'), 3.89 (1 H, d, J = 12 Hz, 5'), 2.19 (3 H, s, Ac), 2.02 (3 H, s, Ac), 1.27 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3000, 1760, 1660, 1620, 1540, 1480, 1370, 1220, 1090, 1040, 850; EI-MS *m/z*: 324 (M^+); HR-MS calcd for $C_{14}H_{16}N_2O_7$ (M^+) 324.0956, found 324.0958.

2'-Deoxy-4'-*C*-methylcytidine (23). A solution of **20** (1.0 g, 3.06 mmol) and 4-chlorophenylphosphorodichloridate (1.1 ml, 6.44 mmol) in dry pyridine (20 ml) was stirred at 0 °C for 2 min. 1,2,4-Triazole (1.6 g, 23.0 mmol) was then added to the solution, and the mixture was stirred for 48 h at room temperature. The mixture was concentrated under reduced pressure to give a syrupy residue, which was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt (40:1:30) as an eluent, to give **22** (830 mg, 72%) as crystal. Compound **22** (500 mg, 1,32 mmol) was dissolved in NH_4OH-dioxane (1:3, v/v, 20 ml), and then the mixture was stirred for 3 h at room temperature and concentrated under reduced pressure to give a syrup. This syrup was dissolved in saturated methanolic ammonia (20 ml), and stirred for 16 h at room temperature and concentrated to give another syrup. The syrup was purified by silica gel column

chromatography, using $CHCl_3$-MeOH (5:1) as an eluent. Evaporation of the appropriate fractions and crystallization of the residue from EtOH-Et_2O yielded 300 mg (94%) of **23** as crystal, mp 188 °C, $[\alpha]_D^{25}$ +81.6° (*c* 1.0, MeOH), ^{1}H-NMR (D_2O) δ: 7.88 (1 H, d, *J* = 7.6 Hz, 6), 6.21 (1 H, t, *J* = 6.6 Hz, 1'), 6.05 (1 H, d, *J* = 7.6 Hz, 5), 4.42 (1 H, t, *J* = 5.9 Hz, 3'), 3.64 (2 H, s, 5'), 2.52 (1 H, ddd, *J* = 5.9, 6.6, 14 Hz, 2'), 2.43 (1 H, ddd, *J* = 5.9, 6.6, 14 Hz, 2'), 1.25 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3430, 3340, 3220, 1650, 1620, 1540, 1480, 1410, 1370, 1290, 1060, 970; CI-MS *m/z*: 242 (M^++H); Calcd for $C_{10}H_{15}N_3O_4$: C, 49.79; H, 6.27; N, 17.42, *Anal.* Found: C, 49.69; H, 6.20; N, 17.13; UV λ_{max} (H_2O) nm (ε): 272 (10700), 198 (23000).

5'-*O*-(*tert*-Butyldimethylsilyl)-4'-*C*-methyladenosine (26). To a stirred suspension of 4'-*C*-methyladenosine (**24**)[3] (2.0 g, 7.11 mmol) and imidazole (1.2 g, 17.8 mmol) in DMF (70 ml) was added tert-butyldimethylsilyl chloride (1.2 g, 7.82 mmol). The reaction mixture was heated at 60 °C with the exclusion of moisture for 3 h. The solvent was removed under vacuum, and the residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH (20:1) as an eluent, to give **26** (1.8 g, 65%) as a crystal, mp 183 °C, $[\alpha]_D^{25}$ -24.4° (*c* 0.6, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.29 (1 H, s, 2), 8.06 (1 H, s, 8), 5.93 (1 H, d, *J* = 6.3 Hz, 1'), 5.73 (2 H, br s, NH_2), 4.73 (1 H, dd, *J* = 5.3, 6.3 Hz, 2'), 4.28 (1 H, d, *J* = 5.3 Hz, 3'), 3.62 (1 H, d, *J* = 11 Hz, 5'), 3.57 (1 H, d, *J* = 11 Hz, 5'), 1.40 (3 H, s, 4'Me), 0.74 (9 H, s, Bu^t), -0.01 (3 H, s, Me), -0.08 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3340, 3220, 2930, 1660, 1650, 1600, 1490, 1420, 1330, 1300, 1260, 1210, 1100, 840, 780; EI-MS *m/z*: 395 (M^+); HR-MS calcd for $C_{17}H_{29}N_5O_4Si$ (M^+) 395.1934, found 395.1960.

5'-*O*-(*tert*-Butyldimethylsilyl)-2',3'-didehydro-2',3'-dideoxy-4'-*C*-methyladenosine (28). To a solution of **26** (1.8 g, 4.65 mmol) in DMF (25 ml) was added 1,1'-thionocarbonyldiimidazole (1.0 g, 5.58 mmol), and the mixture was heated at 80 °C for 1 h. The solvent was removed under vacuum, and the residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH (20:1) as an eluent. Evaporation of the appropriate fractions and crystallization of the residue from $CHCl_3$-hexane yielded 1.8 g (88%) of **27** as crystal. To a ice-cooled solution of **27** (1.5 g, 3.43 mmol) in dry THF (15 ml) was added dropwise 1,3-dimethyl-2-phenyl-1,3,2-diazaphospholidine (2.0 g, 10.3 mmol). The reaction mixture was stirred under nitrogen at 0 °C for 10 min and then at room temperature for 3 h. The solvent was removed under vacuum, and the residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH (30:1) as an eluent. Evaporation of the appropriate fractions and crystallization of the residue from benzene-hexane yielded 780 mg (63%) of **28** as a crystalline product, mp 142 °C, $[\alpha]_D^{25}$ +1.3° (*c* 0.6, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.38 (1 H, s, 2), 8.09 (1 H, s, 8), 7.11 (1 H, dd, *J* = 1.3, 1.7 Hz, 1'), 6.35 (1 H, dd, *J* = 1.7, 5.9 Hz, 2'), 5.98 (1 H, dd, *J* = 1.3, 5.9 Hz, 3'), 5.68 (2 H, br s, NH_2), 3.65 (2 H, s, 5'), 1.37 (3 H, s, 4'Me), 0.89 (9 H, s, Bu^t), 0.05 (3 H, s, Me), 0.03 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3180,

2930, 1650, 1600, 1470, 1420, 1330, 1250, 1200, 1100, 1040, 980, 840, 780; EI-MS *m/z*: 361 (M^+); HR-MS calcd for $C_{17}H_{27}N_5O_2Si$ (M^+) 361.1932, found 361.1936.

2',3'-Didehydro-2',3'-dideoxy-4'-*C*-methyladenosine (29). To a solution of **28** (730 mg, 2.20 mmol) in dry THF (10ml), cooled in an ice bath, was added a 1M solution of *tetra*-n-butylammonium fluoride in THF (4.0 ml, 4.04 mmol). The mixture was stirred for 40 min at room temperature and concentrated. The resulting residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH (10:1) as an eluent. Evaporation of the appropriate fractions and crystallization of the residue from MeOH yielded 370 mg (74%) of **29** as a crystalline product, mp 164 °C, $[\alpha]_D^{25}$ +94.2° (*c* 0.6, MeOH), ^{1}H-NMR (DMSO-d_6) δ: 8.18 (1 H, s, 2), 8.14 (1 H, s, 8), 7.23 (2 H, br s, NH_2), 6.95 (1 H, dd, *J* = 1.3, 2.0 Hz, 1'), 6.40 (1 H, dd, *J* = 2.0, 5.9 Hz, 2'), 6.06 (1 H, dd, *J* = 1.3, 5.9 Hz, 3'), 5.10 (1 H, t, *J* = 5.6 Hz, OH), 3.49 (1 H, dd, *J* = 5.6, 12 Hz, 5'), 3.43 (1 H, dd, *J* = 5.6, 12 Hz, 5'), 1.26 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3330, 3180, 1660, 1600, 1480, 1420, 1370, 1330, 1290, 1240, 1200, 1080, 970, 840; EI-MS *m/z*: 247 (M^+); HR-MS calcd for $C_{11}H_{13}N_5O_2$ (M^+) 247.1067, found 247.1048, UV λ_{max} (MeOH) nm (ε): 259 (16100).

2',3'-Dideoxy-4'-methyladenosine (30). A solution of **29** (250 mg, 1.01 mmol) in MeOH (60 ml) was hydrogenated at 1.0 kg/cm^2 in the presence of Pd-black (100 mg) for 3 h. The reaction mixture was filtered and the filtrate was evaporated. Crystallization of the crude product from MeOH-Et_2O gave **30** (170 mg, 68%) as crystal, mp 180-181 °C, $[\alpha]_D^{25}$ -8.8° (*c* 0.5, MeOH), ^{1}H-NMR (D_2O) δ: 8.32 (1 H, s, 2), 8.20 (1 H, s, 8), 6.34 (1 H, dd, *J* = 4.6, 6.3 Hz, 1'), 3.63 (1 H, d, *J* = 12 Hz, 5'), 3.57 (1 H, d, *J* = 12 Hz, 5'), 2.53-2.78 (2 H, m, 2'), 2.33 (1 H, dt, *J* = 8.3, 13 Hz, 3'), 2.02 (1 H, ddd, *J* = 5.6, 8.3, 13 Hz, 3'), 1.32 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3320, 3180, 1650, 1600, 1570, 1470, 1420, 1370, 1340, 1300, 1250, 1210, 1080, 1000, 940; EI-MS *m/z*: 249 (M^+); HR-MS calcd for $C_{11}H_{15}N_5O_2$ (M^+) 249.1225, found 249.1233, UV λ_{max} (H_2O) nm (ε): 260 (18000), 207 (25500).

5'-*O*-Acetyl-2',3'-didehydro-3'-deoxy-4'-*C*-methylthymidine (31). Acetyl bromide (5.0 ml, 60.1 mmol) was added dropwise over 0.5 h to a suspension of 1-(4-methyl-β-D-ribofuranosyl)thymine (**14**)[3] (2.82 g, 10.4 mmol) in acetonitrile (120 ml) heated at reflux. On completion of addition, the solution was allowed to cool and then concentrated. The residue was dissolved in methylene chloride and washed with water. The organic phase was dried (Na_2SO_4) and concentrated to leave crude **15**. The Zn/Cu couple (ca. 14 g) was suspended in methanol (100 ml), and crude **15** (4.34 g, 10.4 mmol) was added, and the mixture was stirred for 0.5 h. The mixture was filtered and concentrated. The residue was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt-hexane (40:1:30:10) as an eluent. Evaporation of the appropriate fractions and crystallization of the residue from AcOEt-hexane yielded 810 mg (28%) of **31** as crystal, mp 158 °C, $[\alpha]_D^{25}$ -10.0° (*c* 1.0, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 7.33 (1 H, d, *J* = 1.2 Hz, 6), 6.99 (1 H, dd, *J* = 1.2, 2.0 Hz, 1'), 6.23 (1 H, dd, *J* = 2.0, 5.9 Hz, 3'),

5.82 (1 H, dd, J = 1.2, 5.9 Hz, 2'), 4.36 (1 H, d, J = 12 Hz, 5'), 4.06 (1 H, d, J = 12 Hz, 5'), 2.09 (3 H, s, Ac), 1.92 (3 H, d, J = 1.2 Hz, Me), 1.38 (3 H, s, 4'Me); IR ν_{max} (KBr) cm^{-1}: 3150, 3000, 2800, 1740, 1680, 1460, 1420, 1360, 1250, 1220, 1110, 1080, 1020, 890, 830, 770; EI-MS *m/z*: 280 (M^+); Calcd for $C_{13}H_{16}N_2O_5$: C, 55.71; H, 5.75; N, 10.00, *Anal.* Found: C, 55.51; H, 5.65; N, 10.04.

2',3'-Didehydro-3'-deoxy-4'-*C*-methylthymidine (32). Compound **32** (330 mg) was prepared from **31** in a 98% yield by the similar procedure to that described for **13**. **32**: mp 140 °C, $[\alpha]_D^{25}$ +83.5° (*c* 1.0, MeOH), ^{1}H-NMR (D_2O) δ: 7.67 (1 H, s, 6), 6.95 (1 H, br s, 1'), 6.43 (1 H, dd, J = 1.7, 6.1 Hz, 3'), 5.91 (1 H, br d, 2'), 3.71 (1 H, d, J = 13 Hz, 5'), 3.57 (1 H, d, J = 13 Hz, 5'), 1.85 (3 H, s, Me), 1.31 (3 H, s, 4'Me); IR ν_{max} (KBr) cm^{-1}: 3400, 3150, 3000, 1680, 1640, 1460, 1390, 1250, 1070, 980, 900, 820, 770; EI-MS *m/z*: 238 (M^+); Calcd for $C_{11}H_{14}N_2O_4 \cdot 1/5H_2O$: C, 54.63; H, 6.00; N, 11.58, *Anal.* Found: C, 54.37; H, 5.73; N, 11.48; UV λ_{max} (MeOH) nm (ε): 266 (7460), 210 (7590).

5'-*O*-Acetyl-3'-deoxy-4'-*C*-methylthymidine (33). A solution of **31** (500 mg, 1.78 mmol) in methanol (20 ml) was stirred under hydrogen with 10% Pd-carbon (250 mg) for 1 h. The catalyst was removed by filtration and the filtrate was evaporated to give crude crystal, which were recrystalized from AcOEt-hexane to give **33** (460 mg, 91%) as crystal, mp 130 °C, $[\alpha]_D^{25}$ +10.9° (*c* 1.0, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.34 (1 H, br s, NH), 7.42 (1 H, d, J = 1.3 Hz, 6), 6.13 (1 H, t, J = 5.9 Hz, 1'), 4.26 (1 H, d, J = 12 Hz, 5'), 4.11 (1 H, d, J = 12 Hz, 5'), 2.47-2.57 (1 H, m, 2'), 2.14 (3 H, s, Ac), 1.80-2.10 (3 H, m, 2', 3'), 1.94 (3 H, d, J = 1.3 Hz, Me), 1.32 (3 H, s, 4'Me); IR ν_{max} (KBr) cm^{-1}: 3190, 3060, 1740, 1690, 1480, 1380, 1270, 1230, 1120, 1070, 1040, 960, 900, 850; EI-MS *m/z*: 282 (M^+); HR-MS calcd for $C_{13}H_{18}N_2O_5$ (M^+) 282.1214, found 282.1204.

3'-Deoxy-4'-*C*-methylthymidine (34). Compound **34** (280 mg) was prepared from **33** in a 89% yield by the similar procedure to that described for **13**. **34**: mp 125-126 °C, $[\alpha]_D^{25}$ +29.6° (*c* 1.0, MeOH), ^{1}H-NMR (D_2O) δ: 7.76 (1 H, d, J = 1.0 Hz, 6), 6.18 (1 H, dd, J = 4.3 Hz, 6.9, 1'), 3.67 (1 H, d, J = 12 Hz, 5'), 3.63 (1 H, d, J = 12 Hz, 5'), 2.49-2.64 (1 H, m, 2'), 2.10-2.23 (2 H, m, 2', 3'), 1.90 (3 H, d, J = 1.0 Hz, Me), 1.84-1.94 (1 H, m, 3'), 1.26 (3 H, s, 4'Me); IR ν_{max} (KBr) cm^{-1}: 3370, 2920, 1710, 1650, 1460, 1410, 1280, 1260, 1140, 1110, 1080, 1060, 1000, 930, 860, 840; EI-MS *m/z*: 240 (M^+); HR-MS calcd for $C_{11}H_{16}N_2O_4$ (M^+) 240.1109, found 240.1109, UV λ_{max} (H_2O) nm (ε): 268 (10800), 207 (10800).

N^4-Acetyl-4'-*C*-methylcytidine (36). A mixture of 4'-*C*-methylcytidine (**35**)[3] (1.9 g, 7.39 mmol) and acetic anhydride (2 ml) in methanol (90 ml) was heated under reflux with stirring. At hourly intervals, acetic anhydride (2 ml x 2) was added to a mixture. After 4 h, the reaction mixture cooled at room temperature and evaporated. The residue was purified by silica gel column

chromatography, using $CHCl_3$-MeOH (5:1) as an eluent, to give **36** (1.5 g, 66%) as crystal, mp 191 °C, $[\alpha]_D^{25}$ +18.4° (*c* 0.9, MeOH), ^{1}H-NMR (D_2O) δ: 8.32 (1 H, d, *J* = 7.6 Hz, 6), 7.35 (1 H, d, *J* = 7.6 Hz, 5), 6.01 (1 H, d, *J* = 4.9 Hz, 1'), 4.47 (1 H, dd, *J* = 4.9, 5.9 Hz, 2'), 4.24 (1 H, d, *J* = 5.9 Hz, 3'), 3.68 (2 H, s, 5'), 2.25 (3 H, s, Ac), 1.33 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3530, 3270, 1720, 1650, 1490, 1430, 1390, 1320, 1310, 1190, 1100, 1060, 810, 790; EI-MS *m/z*: 299 (M^+); HR-MS calcd for $C_{12}H_{17}N_3O_6$ (M^+) 299.1116, found 299.1131.

N^4-Acetyl-2',3'-didehydro-2',3'-dideoxy-4'-*C*-methylcytidine (40). Employing conditions analogous to those used for the preparation of **29**, compound **36** (1.6 g, 5.18 mmol) gave **40** (410 mg, 33%) as an amorphous, $[\alpha]_D^{25}$ +65.8° (*c* 0.3, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.09 (1 H, d, *J* = 7.6 Hz, 6), 7.40 (1 H, d, *J* = 7.6 Hz, 5), 6.93 (1 H, dd, *J* = 1.3, 1.7 Hz, 1'), 6.13 (1 H, dd, *J* = 1.7, 5.9 Hz, 2'), 6.04 (1 H, dd, *J* = 1.3, 5.9 Hz, 3'), 4.43 (1 H, d, *J* = 12 Hz, 5'), 4.04 (1 H, d, *J* = 12 Hz, 5'), 2.27 (3 H, s, Ac), 1.41 (3 H, s, Me); IR ν_{max} (neat) cm^{-1}: 3090, 2980, 1750, 1660, 1560, 1500, 1400, 1370, 1310, 1240, 1090, 1050, 990, 850, 790; EI-MS *m/z*: 265 (M^+); HR-MS calcd for $C_{12}H_{15}N_3O_4$ (M^+) 265.1062, found 265.1061.

2',3'-Didehydro-2',3'-dideoxy-4'-*C*-methylcytidine (41). Compound **41** (100 mg) was prepared from **40** (150 mg, 0.57 mmol) in a 79% yield by the similar procedure to that described for **13**. **41**: mp 161 °C, $[\alpha]_D^{25}$ +90.7° (*c* 0.5, MeOH), ^{1}H-NMR (D_2O) δ: 7.78 (1 H, d, *J* = 7.3 Hz, 6), 6.95 (1 H, br d, *J* <1 Hz, 1'), 6.40 (1 H, dd, *J* = 5.9, <1 Hz, 2'), 5.98 (1 H, d, *J* = 7.3 Hz, 5), 5.92 (1 H, dd, *J* =5.9, <1 Hz, 3'), 3.68 (1 H, d, *J* = 13 Hz, 5'), 3.57 (1 H, d, *J* = 13 Hz, 5'), 1.32 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3410, 1650, 1590, 1520, 1470, 1400, 1360, 1290, 1230, 1090, 1060, 1030, 850; CI-MS *m/z*: 224 (M^++H); HR-MS calcd for $C_{10}H_{13}N_3O_3$ (M^+) 223.0956, found 223.0958, UV λ_{max} (MeOH) nm (ε): 271 (7930).

5'-*O*-Acetyl-2',3'-dideoxy-4'-*C*-methyluridine (43). Employing conditions analogous to those used for the preparation of **33**, compound **18** (1.7 g, 6.39 mmol) gave **43** (440 mg, 26%) as a pale yellow syrup, $[\alpha]_D^{25}$ +21.7° (*c* 1.0, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 9.50 (1 H, br s, NH), 7.68 (1 H, d, *J* = 8.2 Hz, 6), 6.11 (1 H, dd, *J* = 4.6, 6.3 Hz, 1'), 5.73 (1 H, d, *J* = 8.2 Hz, 5), 4.23 (1 H, d, *J* = 12 Hz, 5'), 4.11 (1 H, d, *J* = 12 Hz, 5'), 2.51-2.66 (1 H, m, 2'), 2.12 (3 H, s, Ac), 1.97-2.11 (2 H, m, 2', 3'), 1.89-1.91 (1 H, m, 3'), 1.32 (3 H, s, Me); IR ν_{max} (neat) cm^{-1}: 3200, 3050, 1730, 1700, 1680, 1650, 1470, 1280, 1240, 1100, 1050, 950, 810; EI-MS *m/z*: 268 (M^+); HR-MS calcd for $C_{12}H_{16}N_2O_5$ (M^+) 268.1058, found 268.1091.

2',3'-Dideoxy-4'-*C*-methylcytidine (44). Employing conditions analogous to those used for the preparation of **23**, compound **43** (400 mg, 1.49 mmol) gave **44** (70 mg, 21%) as an amorphous, $[\alpha]_D^{25}$ +67.6° (*c* 1.0, MeOH), ^{1}H-NMR (D_2O) δ: 7.93 (1 H, d, *J* = 7.6 Hz, 6), 6.14 (1 H, dd, *J* = 3.6, 6.6 Hz, 1'), 6.03 (1 H, d, *J* = 7.6 Hz, 5), 3.68 (1 H, d, *J* = 12 Hz, 5'), 3.48

(1 H, d, *J* = 12 Hz, 5'), 2.55-2.64 (1 H, m, 2'), 2.01-2.14 (2 H, m, 2', 3'), 1.82-1.92 (1 H, m, 3'), 1.27 (3 H, s, Me); IR ν_{max} (neat) cm^{-1}: 3350, 3200, 1650, 1490, 1360, 1290, 1190, 1070, 1010, 950, 790; SI-MS *m/z*: 226 $(M+H)^+$; HR-MS calcd for $C_{10}H_{15}N_3O_3$ (M^+) 225.1112, found 225.1152, UV λ_{max} (MeOH) nm (ε): 273 (7400).

1-(2',3',5'-Tri-*O*-acetyl-4'-*C*-methyl-β-D-arabinofuranosyl)uracil (45). A mixture of **21** (1.2 g, 3.70 mmol) and 1N NaOH (18 ml) in 50% ethanol (150 ml) was stirred at room temperature for 2 h. The solution was neutralized with AcOH/EtOH (1:1, v/v) to ~pH 7. The solvent was removed under vacuum and the residue was dissolved in pyridine (20 ml). Acetic anhydride (2.5 ml, 26.3 mmol) was added to a pyridine solution and the resulting mixture was stirred at room temperature for 18 h. The reaction mixture was concentrated to give another syrup. The syrup was purified by silica gel column chromatography, using $CHCl_3$-MeOH-AcOEt-hexane (20:2:15:5) as an eluent, to give **45** (760 mg, 53%) as crystal, mp 154 °C, $[\alpha]_D^{25}$ +82.1° (*c* 0.7, $CHCl_3$), ^{1}H-NMR ($CDCl_3$) δ: 8.28 (1 H, br s, NH), 7.52 (1 H, d, *J* = 8.3 Hz, 6), 6.32 (1 H, d, *J* = 4.6 Hz, 1'), 5.73 (1 H, dd, *J* = 8.3 Hz, <1, 5), 5.47 (1 H, dd, *J* = 3.0, 4.6 Hz, 2'), 5.31 (1 H, d, *J* = 3.0 Hz, 3'), 4.27 (1 H, d, *J* = 12 Hz, 5'), 4.23 (1 H, d, *J* = 12 Hz, 5'), 2.15 (3 H, s, Ac), 2.14 (3 H, s, Ac), 2.04 (3 H, s, Ac), 1.31 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3030, 1750, 1710, 1680, 1380, 1280, 1240, 1120, 1050, 910, 810; CI-MS *m/z*: 385 (M^++H); HR-MS calcd for $C_{16}H_{20}N_2O_9$ (M^+) 384.1167, found 384.1149.

1-(4'-*C*-Methyl-β-D-arabinofuranosyl)cytosine (46). Employing conditions analogous to those used for the preparation of **23**, compound **45** (730 mg, 1.90 mmol) gave **46** (450 mg, 92%) as crystal, mp 205-206 °C (dec.), $[\alpha]_D^{25}$ +91.0° (*c* 1.1, MeOH), ^{1}H-NMR (D_2O) δ: 7.88 (1 H, d, *J* = 7.6 Hz, 6), 6.24 (1 H, d, *J* = 5.9 Hz, 1'), 6.04 (1 H, d, *J* = 7.6 Hz, 5), 4.54 (1 H, t, *J* = 5.9 Hz, 2'), 4.14 (1 H, d, *J* = 5.9 Hz, 3'), 3.70 (2 H, s, 5'), 1.24 (3 H, s, Me); IR ν_{max} (KBr) cm^{-1}: 3180, 1650, 1510, 1290, 1250, 1190, 1140, 1100, 1060, 820, 790; CI-MS *m/z*: 258 (M^++H); Calcd for $C_{10}H_{15}N_3O_5$: C, 46.69; H, 5.88; N, 16.33, *Anal.* Found: C, 46.45; H, 5.93; N, 15.92; UV λ_{max} (H_2O) nm (ε): 272 (10200), 197 (22400).

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REFERENCES

1) a) Mitsuya, H; Yarchoan, R; Broder, S. *Science* **249**, 1533 (1990); b) Mitsuya, H; Broder, S. *Proc. Natl. Acad. Sci. U.S.A.* **83**, 1911 (1986); c) De Clercq, E; Van Aerschot, A; Herdewijn, P; Baba, M; Pauwels, R; Balazarini,J. *Nucleosides Nucleotides* **8**, 659 (1989).

2) Magg, H; Rydzewski, R. M; McRoberts, M. J; Crawford-Ruth, D; Verheyden, J. P. H; Prisbe, E. J. *J. Med. Chem.* **35**, 1440 (1992).

3) Waga, T; Nishizaki, T; Miyakawa, I; Ohrui, H; Meguro, H. *Biosci. Biotech. Biochem.* **57**, 1433 (1993).

4) Saneyoshi, M; Satoh, E. *Chem. Pharm. Bull.* **27**, 2518 (1979).

5) Robins, M. J ; Wilson, J. S; Hansske, F. *J. Am. Chem. Soc.* **105**, 4059 (1983).

6) Mansuri, H. M; Starrett, J. E; Wos, J. A; Tortolani, D. R; Brodfuehrer, P. R; Howell, H. G; Martin, J. C. *J. Org. Chem.* **54**, 4780 (1989).

7) Divaker, K. J; Reese, C. B. *J. Chem. Soc. Perkin Trans. 1* **1982**, 1171.