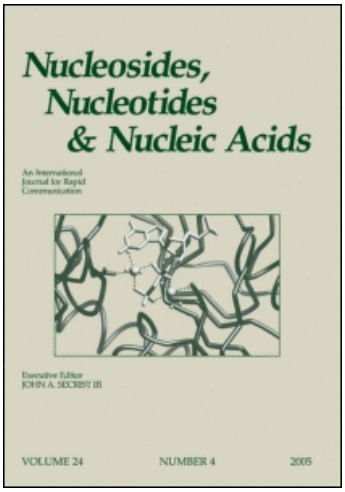

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Synthesis and anti-HCV Evaluation of 4'(α)-ethyl and 2'(β)-methyl-carbodine Analogues

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SYNTHESIS AND ANTI-HCV EVALUATION OF 4′(α)-ETHYL AND 2′(β)-METHYL-CARBODINE ANALOGUES

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□ *Novel 4′(α)-ethyl-2′(β)-methyl carbocyclic nucleoside analogues have been prepared and evaluated for inhibition of hepatitis C virus (HCV) RNA replication in cell culture. The construction of cyclopentene intermediate* ***12β*** *was successfully made via sequential Johnson-Claisen orthoester rearrangement and ring-closing metathesis (RCM) starting from Weinreb amide* ***5****. Selective dihydroxylation and desilylation gave the target carbodine analogues. The synthesized nucleoside analogues mentioned above* ***18*** *and* ***19*** *were assayed for their ability to inhibit HCV RNA replication in a subgenomic replicon Huh7 cell line (LucNeo#2). However, the synthesized nucleosides neither showed any significant antiviral activity nor toxicity up to 50 μM.*

Keywords Anti-HCV agent; carbodine; branched carbocyclic nucleoside

INTRODUCTION

Hepatitis C virus (HCV) infection is a leading cause of chronic hepatitis, liver cirrhosis and hepatoma carcinoma.[1] However, there is no effective chemotherapy for the treatment of HCV-infected people except immunotherapy using ribavirin in combination with interferon-α, which leads to a sustained virological response in only about half of the patients treated.[2]

Since nucleoside analogues have been used as a drug of choice in curing viral infection, many have been synthesized and evaluated for anti-HCV agent.[3] These nucleosides are incorporated into proviral RNA like a substrate after being converted to their corresponding triphosphates and act as chain terminators. Modification in the vicinity of the 2′-hydroxy of the ribose in natural ribonucleosides can produce effective RNA chain terminator.[4]

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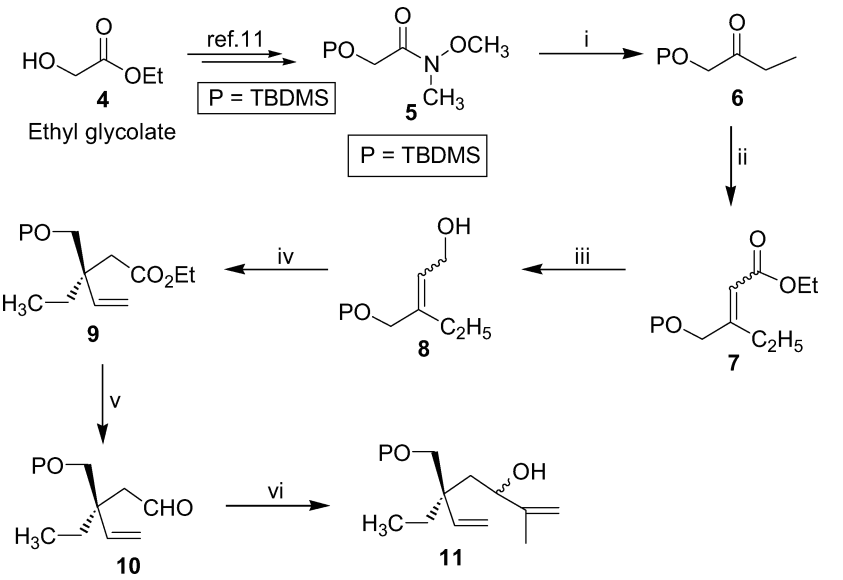
For example, 2′-methyl ribonucleosides yield compounds with excellent chain-terminating properties. Among them, 2′-*C*-methylcytidine[5] **1** and 2′-*C*-methyladenosine[6] **2** were discovered as potent anti-HCV agents and 2′-*C*-methylcytidine is in phase II clinical trials (Figure 1).

Recently, 4′-homologated stavudine[7] and thiostavudine[8] analogues are molecules of considerable interest. One of reasons for this prominence arises from the notable biological activities as antiviral and antitumor agents. Modeling studies demonstrated the presence of a narrow, relatively hydrophobic 4′-pocket that can accommodate these substitutions, contributing to the observed enhancement in potency.[9] Furthermore, 4′-azido-arabinocytidine[10] **3** showed excellent anti-HCV activity (IC_{50} 0.17 μM) in the genotype 1b subgenomic replicon system.

On the basis of these findings, that the branched nucleoside analogues exhibited excellent anti-HCV activities, we have determined to synthesize novel classes of nucleosides comprising 4′-ethyl and 2′-*C*-methyl carbodine analogues.

The target compounds were prepared as shown in Scheme 1 from the key starting material **5**, which was readily synthesized from our previous report.[11]

Transformation of the Weinreb amide[12] **5** to ketone derivative **6** turned out to be successful under the usual carbonyl addition condition using ethylmagnesium bromide. Olefination of **6** using triethyl phosphonoacetate under Horner-Wadsworth-Emmons (HWE) reaction condition[13] provided



SCHEME 1 Synthesis of route divinyl intermediate 11. Reagents: i) ethyMgBr, THF, 0°C; ii) triethylphophonoacetate, NaH, THF; iii) DI BAL-H, CH2Cl2; iv) triethylorthoacetate, propionic acid, overnight, 135–140°C; v) DI BAL-H, toluene, −78°C; vi) isopropenyl MgBr, THF, −78°C.

α,β-unsaturated ethyl ester **7** as cis/trans isomeric mixtures (Scheme 1). It is unnecessary to separate the isomers, because they will be merged into one isomer after [3,3]-sigmatropic rearrangement. Ester **7** was reduced to allylic alcohol **8** by using diisobutylaluminum hydride (DIBAL-H), which underwent regular Johnson-Claisen rearrangement[14] using triethyl orthoacetate with catalytic amount of propionic acid to give γ,δ–unsaturated ester **9**. Direct reduction of the ester **9** to the aldehyde **10** was possible in the mild conditions by slow addition of DIBAL-H in the toluene solvent system at −78°C. The aldehyde **10** was subjected to nucleophilic Grignard conditions by isopropenyl magenesiumbromide [$CH_2 = C(CH_3)MgBr$] to yield divinyl **11**.

Without separation of diastereomeric mixture, divinyl **11** was subjected to ring-closing metathesis (RCM) condition[15] using second generation Grubbs catalyst to provide cyclopentenol **12α** (38%) and **12β** (39%), which were readily separated by silica gel column chromatography. The relative stereochemical assignments of two isomers were readily made based on Nuclear Overhauser Enhancement (NOE) comparisons. Upon the irradiation of C_1-H, different NOE patterns were observed at the proximal hydrogens of compound **12β** [*methyloxy*-H (0.08%)], from those of compound **12α** [*methyloxy*-H (0.19%)] (Figure 2).

The allylic functionalization using palladium(0)-catalyzed reactions[16] have been the central role in synthetic organic chemistry, because they provide successful regio- and stereochemical products in allyic functional positions of cyclopentene intermediate. To obtain a desired carbocyclic nucleoside, cyclopentenol **12β** was transformed to **13** using ethyl

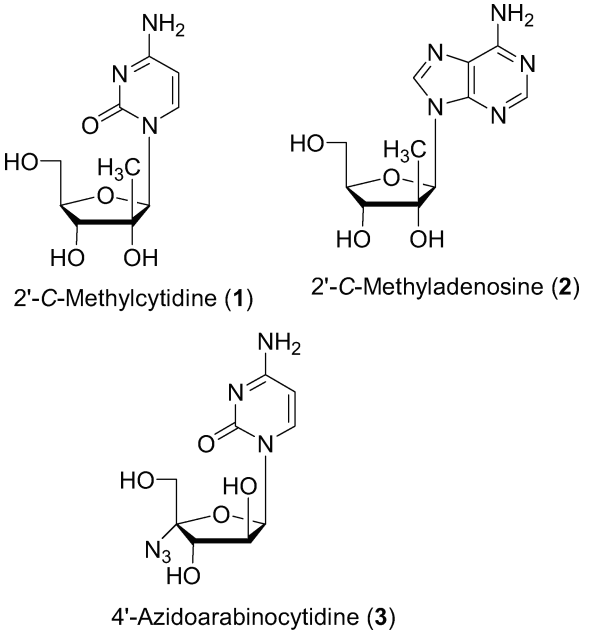


FIGURE 1 Structures of potent anti-HCV agents.

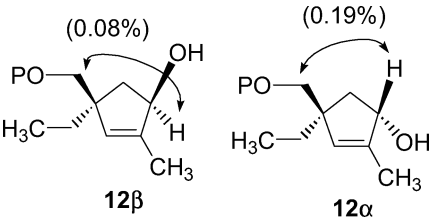


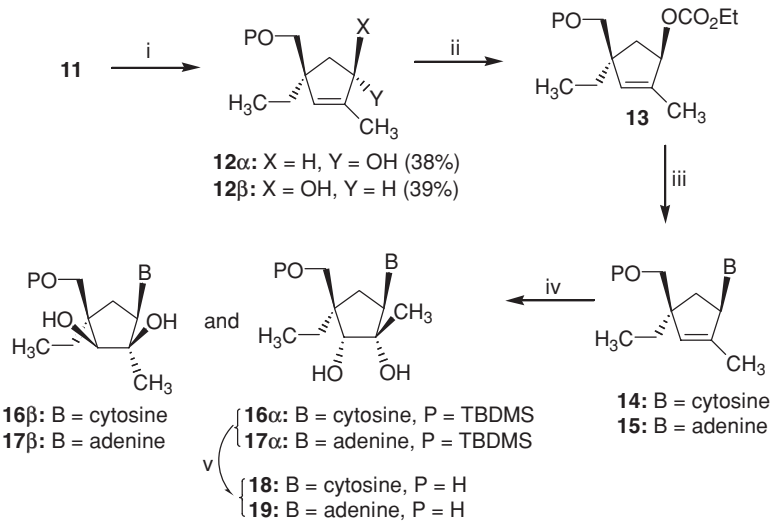
FIGURE 2 NOE differences of two isomers **12**β and **12**α.

chloroformate, which was readily coupled with adenine and cytosine generated by NaH/DMSO with use of catalyst [tris(dibenzylidene-acetone)-dipalladium(0)-chloroform] adduct to provide nucleoside analogue **14** and **15**. To get the carbodine analogues, the protected nucleosides **14** and **15** were oxidized by a catalytic amount of OsO_4 and NMO to give the dihydroxylated **16**α and **17**α as major reaction products compared to **16**β and **17**β. Their stereochemistries were also readily determined by NOE experiment. These stereochemical outcomes suggest that the bulky groups such as silylated hydroxymethyl group and nucleosidic bases (adenine and cytosine) reinforce the steric hindrance of the β-faces.[17] Furthermore, we can suppose that the cyclopentane ring causes the 4′-moiety with a protecting group on the 5′-hydroxy to be in equatorial positions, unlike the axial position that exist in normal nucleosides. This causes the 4′-substituent to be in axial down position, making the dihyroxylation from a-face more hindered. Removals of silyl protection group of **16**α and **17**α were performed by the treatment of tetrabutylammonium fluoride (TBAF) to give target nucleosides **18** and **19** (Scheme 2).

The synthesized nucleoside analogues mentioned above **18** and **19** were assayed for their ability to inhibit HCV RNA replication in a subgenomic replicon Huh7 cell line (LucNeo#2).[18] However, the synthesized nucleosides neither showed any significant antiviral activity nor toxicity up to 50 μM. Taking these data into account, it appears that the alkyl group equipped at the 4′-position makes the cyclopentane ring conformation be unfavorable for the phosphorylation at the 5'-hydroxyl group necessary for incorporation into the growing RNA chain by the polymerase.

EXPERIMENTAL

Melting points were determined on a Mel-temp II laboratory device and are uncorrected. NMR spectra were recorded on a JEOL 300 Fourier transform spectrometer (JEOL, Tokyo, Japan); chemical shifts are reported in parts per million (δ) and signals are reported as s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet), and dd (doublet of doublets). Ultraviolet (UV) spectra were obtained on a Beckman DU-7 spectrophotometer (Beckman, South Pasadena, CA, USA). The elemental analyses were performed using a Perkin-Elmer 2400 analyzer (Perkin-Elmer,



SCHEME 2 Synthesis of target carbocyclic nucleosides. Reagents: i) Grubbs catalyst (II), benzene; ii) $ClCO_2Et$, pyrimidine, DMAP; iii) cytosine, adenine, $Pd_2(dba)_3.CHCl_3$, $P(O\text{-}i\text{-}Pr)_3$, NaH, THF/DMSO; iv) OsO_4 NMO; v) TBAF, THF/CH_3CN, room temperature.

Norwalk, CT, USA). Thin layer chromatography (TLC) was performed on Uniplates (silica gel) purchased from Analtech Co. (7558, Newark, DE, USA). All reactions were carried out under an atmosphere of nitrogen unless specified. Dry dichloromethane, benzene and pyridine were obtained by distillation from CaH_2. Dry THF was obtained by distillation from Na and benzophenone immediately prior to use.

1-(t-Butyldimethylsilyloxy)-butan-2-one (6): To a solution of Weinreb amide **5** (3.0 g, 12.85 mmol) in dry THF (50 mL) was slowly added ethylmagnesium bromide (19.0 mL, 1.0 M solution in THF) at 0°C. After 6 hours, saturated NH_4Cl solution (19 mL) was added, and the reaction mixture was slowly warmed to room temperature. The mixture was extracted with EtOAc (2 × 100 mL). The combined organic layer was dried over $MgSO_4$, filtered, and evaporated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:15) to give **6** (2.11 g, 76%) as colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 4.08 (s, 2H), 2.43 (q, $J = 7.2$ Hz, 2H), 0.96 (t, J = 7. 2Hz, 3H), 0.81 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 211.79, 68.10, 25.71, 18.71, 14.14, −5.59.

(*E*) and (*Z*)-3-(t-Butyldimethylsilyloxymethyl)-3-ethyl-but-2-enoic acid ethyl ester (7): To a suspension of sodium hydride (0.48 g, 19.96 mmol) in distilled THF (90 mL) was added drop wise triethyl phosphonoacetate (4.47 g, 19.96 mmol) at 0°C and the mixture was stirred at room temperature for 1 hour. The ketone **6** (4.32 g, 19.96 mmol) was added to this mixture and the mixture was stirred for 2 hours. The solution was neutralized with AcOH (4.0 mL) and poured into H_2O (150 mL) and extracted with EtOAc

(150 × 2). The combined organic layer was washed with brine and dried over anhydrous $MgSO_4$, filtered and evaporated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:12) to give **7** (3.8 g, 70%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.85, 5.54 (s, s, 1H), 4.11 (m, 4H), 2.43 (m, 2H), 1.23 (m, 3H), 1.02 (m, 3H), 0.82 (m, 9H), 0.01 (s, 6H).

(*E*) and (*Z*)-3-(t-Butyldimethylsilyloxymethyl)-3-ethyl-but-2-en-1-ol (8): To a solution of **7** (4.6 g, 16.88 mmol) in CH_2Cl_2 (100 mL), DIBAL-H (35.45 mL, 1.0 M solution in hexane) was added slowly at −20°C, and stirred for 2 hours at the same temperature. To the mixture, methanol (35 mL) was added. The mixture was stirred at room temperature for 2 hours, and the resulting solid was filtered through a Celite pad. The filtrate was concentrated under vacuum and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:5) to give alcohol **8** (3.61 g, 93%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.57-5.43 (m, 1H), 4.10 (m, 2H), 2.08-1.95 (m, 2H), 1.01-0.93 (m, 3H), 0.82 (m, 9H), 0.02 (m, 6H).

(±)-3-(t-Butyldimethylsilyloxymethyl)-3-ethyl-pent-4-enoic acid ethyl ester (9): A solution of allylic alcohol **8** (5.5 g, 23.85 mmol) in triethyl orthoacetate (110 mL) and 0.1 mL of propionic acid was heated at 135–140°C overnight with stirring under condition for distillative removal of ethanol. The excess of triethyl orthoacetate was distilled off and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:20) to give **9** (5.59 g, 78%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.92 (d, J = 10.4 Hz, 1H), 5.88 (d, J = 11.2 Hz, 1H), 5.03 (dd, J = 7.8, 1.2 Hz, 1H), 4.02 (q, J = 7.2 Hz, 2H), 3.47 (d, J = 9.2 Hz, 1H), 3.42 (d, J = 9.2 Hz, 1H), 2.41 (dd, J = 14.8, 8.4 Hz, 2H), 1.29-1.22 (m, 5H), 0.99 (m, 3H), 0.81 (s, 9H), 0.46 (m, 1H), 0.28-0.21 (m, 4H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 171.92, 143.43, 114.21, 70.12, 60.21, 43.21, 27.12, 25.56, 18.43, 14.65, 9.43, −5.59.

(±)-3-(t-Butyldimethylsilyloxymethyl)-3-ethyl-pent-4-enal (10): To a solution of **9** (3.3 g, 10.98 mmol) in toluene (50 mL), DIBALH (16.1 mL, 1.5 M solution in toluene) was added slowly at −78°C, and stirred for 10 minutes at the same temperature. To the mixture, methanol (10 mL) was added. The mixture was stirred at room temperature for 1 hour, and the resulting solid was filtered through a Celite pad. The filtrate was concentrated under vacuum and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:15) to give **10** (1.69 g, 60%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 9.99 (d, J = 8.4 Hz, 1H), 5.98 (d, J = 12.2 Hz, 1H), 5.49 (d, J = 12.0 Hz, 1H), 5.20 (d, J = 14.2 Hz, 1H), 3.51 (d, J = 9.4 Hz, 1H), 3.40 (d, J = 9.4 Hz, 1H), 2.45 (dd, J = 14.4, 7.2 Hz, 2H), 1.52 (q, J = 7.0 Hz, 2H), 0.92 (t, J = 7.2 Hz, 3H), 0.82 (s, 9H), 0.02 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 202.82, 144.98, 113.54, 70.87, 51.54, 42.21, 27.67, 25.80, 18.52, 9.45, −5.62.

(*rel*)-(3*R* and 3*S*,5*S*)-5-(t-Butyldimethylsilanyloxymethyl)-5-ethyl-2-methyl-hepta-1,6-dien-3-ol (11): To a solution of **10** (2.7 g, 10.52 mmol) in dry THF (30 mL) was slowly added isopropenylMgBr (15.79 mL, 1.0 M solution in THF) at −78°C. After 6 hours, saturated NH_4Cl solution (15 mL) and water (60 mL) was sequentially added, and the reaction mixture was slowly warmed to room temperature. The mixture was extracted with EtOAc (70 mL) two times. The combined organic layer was dried over $MgSO_4$, filtered, and evaporated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:15) to give **11** (2.12 g, 71%) as colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.99-5.81 (m, 1H), 5.32-5.16 (m, 4H), 3.49-3.30 (m, 3H), 1.70-1.59 (m, 7H), 0.98 (t, $J = 7.0$ Hz, 3H), 0.81 (s, 9H), 0.01 (m, 6H).

(*rel*)-(1*R*,4*S*)-4-(t-Butyldimethylsilyloxymethyl)-4-ethyl-2-methyl-cyclopent-2-enol (12*β*); and (*rel*)-(1*S*,4*S*)-4-(t-butyldimethylsilyloxymethyl)-4-ethyl-2-methyl-cyclopent-2-enol (12*α*): To a solution of **11** (2.57 g, 8.61 mmol) in dry benzene (15 mL) was added 2nd generation Grubbs catalyst (127 mg 0.15 mmol). The reaction mixture was refluxed overnight, and cooled to room temperature. The mixture was concentrated in vacuum, and residue was purified by silica gel column chromatography (EtOAc/hexane, 1:10) to give cyclopentenol **12*β*** (908 mg, 39%) and **12*α*** (885 mg, 38%) as colorless oils, respectively. Cyclopentenol **12*β***: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.41 (s, 1H), 4.37 (d, $J = 4.2$ Hz, 1H), 3.48 (d, $J = 8.2$ Hz, 1H), 3.33 (d, $J = 8.2$ Hz, 1H), 1.94 (dd, $J = 13.6$, 6.8 Hz, 1H), 1.78 (dd, $J = 13.6$, 4.8 Hz, 1H), 1.69 (s, 3H), 1.32 (m, 2H), 0.96 (dd, $J = 5.8$, 2.1 Hz, 3H), 0.83 (s, 9H), 0.01 (s, 6H): ^{13}C NMR ($CDCl_3$) δ 140.76, 134.45, 75.10, 70.11, 51.34, 46.78, 28.21, 25.32,18.34, 14.02, 10.04, −5.50; Anal. Calcd. for $C_{15}H_{30}O_2Si$: C, 66.61; H, 11.18. Found: C, 66.76; H, 11.09.

Cyclopentenol **12*α***: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.50 (s, 1H), 4.50 (s, 1H), 3.40 (dd, $J = 14.2$, 5.2 Hz, 2H), 1.92 (dd, $J = 13.8$, 7.2 Hz, 1H), 1.80 (dd, $J = 13.8$, 6.2 Hz, 1H), 1.71 (s, 3H), 1.35-1.28 (m, 2H), 0.96 (t, $J = 6.2$ Hz, 3H), 0.83 (s, 9H), 0.01 (s, 6H): ^{13}C NMR ($CDCl_3$) δ 141.02, 134.32, 76.81, 70.78, 51.29, 47.43, 28.89, 25.54,18.77, 13.98, 10.15, −5.61; Anal. Calcd. for $C_{15}H_{30}O_2Si$: C, 66.61; H, 11.18. Found: C, 66.52; H, 10.99.

(*rel*)-(1*R*,4*S*)-1-Ethoxycarbonyloxy-4-(t-butyldimethylsilyloxymethyl)-4-ethyl-2-methyl-cyclopent-2-ene (13): To a solution of **12*β*** (2.34 g, 8.65 mmol) in anhydrous pyridine (15 mL) was added ethyl chloroformate (1.65 mL, 17.31 mmol) and DMAP (102 mg, 0.84 mmol). The reaction mixture was stirred overnight at room temperature. The reaction mixture was quenched with saturated $NaHCO_3$ solution (2 mL), stirred for 10 minutes and concentrated in reduced pressure. The residue was extracted with EtOAc/H_2O, dried over $MgSO_4$, filtered, and concentrated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:10)

to give **13** (2.25 g, 76%) as colorless syrup: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.53 (s, 1H), 4.55 (dd, J = 5.8 Hz, 1H), 4.15 (q, J = 7.2 Hz, 2H), 3.39 (dd, J = 13.8, 5.8 Hz, 2H), 1.91 (dd, J = 13.8, 7.6 Hz, 1H), 1.76 (dd, J = 13.8, 6.6 Hz, 1H), 1.70 (s, 3H), 1.31 (m, 2H), 1.24 (t. J = 7.2 Hz, 3H), 0.97 (t, J = 7.0 Hz, 3H), 0.82 (s, 9H), 0.01 (s, 6H): ^{13}C NMR ($CDCl_3$) δ 155.13, 145.01, 128.32, 83.54, 69.98, 62.99, 51.54, 43.65, 27.92, 25.71, 18.48, 17.31, 13.7, 9.62, -5.51; Anal. Calcd. for $C_{18}H_{34}O_4Si$: C, 63.11; H, 10.00. Found: C, 63.21; H, 9.93.

(*rel*)-(1′*R*,4′*S*)-1-[4-(t-Butyldimethylsilyloxymethyl)-4-ethyl-2-methyl-cyclopent-2-en-1-yl] cytosine (14): In order to generate nucleosidic base anion, cytosine (86.6 mg, 0.78 mmol) was added to a hexane washed NaH (18.7 mg, 0.78 mmol) in anhydrous DMSO (5.0 mL). The reaction mixture was stirred for 30 minutes at 50–55°C and cooled to room temperature. Simultaneously, P(O-*i*-Pr)$_3$ (65 mg, 0.312 mmol) was added to a solution of $Pd_2(dba)_3 \cdot CHCl_3$ (41.4 mg, 4.0 μmol) in anhydrous THF (4.0 mL), which was stirred for 30 minutes. To the cytosine solution of DMSO was sequentially added catalyst solution of THF and **13** (239.7 mg, 0.70 mmol) dissolved in anhydrous THF (4.0 mL). The reaction mixture was stirred overnight at refluxing temperature and quenched with water (2.0 mL). The reaction solvent was removed in vacuum. The residue was purified by silica gel column chromatography (MeOH/Hexane/EtOAc, 0.1:3:1) to give **14** (99 mg, 39%) as a white solid: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.20 (d, J = 6.8 Hz, 1H), 5.98 (s, 1H), 5.86 (d, J = 6.8 Hz, 1H), 4.59 (d, J = 4.8 Hz, 1H), 3.41 (dd, J = 13.8, 6.8 Hz, 2H), 2.42 (dd, J = 14.0, 8.8 Hz, 1H), 2.00 (dd, J = 14.0, 6.2 Hz, 1H), 1.72 (s, 3H), 1.34 (dd, J = 5.8, 2.4 Hz, 2H), 0.95 (m, 3H), 0.81 (s, 9H), 0.01 (s, 6H): ^{13}C NMR ($CDCl_3$) δ 165.76, 156.81, 145.76, 145.12, 133.45, 92.29, 70.07, 60.21, 45.65, 29.43, 25.54, 18.47, 13.98, 10.11, −5.49. Anal. Calcd. for $C_{19}H_{33}N_3O_2Si$: C, 62.77; H, 9.15; N, 11.56. Found: C, 62.67; H, 9.08; N, 11.48.

(*rel*)-(1′*R*,4′*S*)-9-[4-(t-Butyldimethylsilyloxymethyl)-4-ethyl-2-methyl-cyclopent-2-en-1-yl] adenine(15): Adenine nucleoside analogue **15** analogue was synthesized from **12** by the similar procedure as described for **13**: yield 36%; ^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.27 (s, 1H), 8.14 (s, 1H), 6.00 (s, 1H), 4.67 (s, 1H), 3.52 (d, J = 9.6 Hz, 1H), 3.40 (d, J = 9.6 Hz, 1H), 2.45 (dd, J = 13.8, 8.6 Hz, 1H), 2.02 (dd, J = 13.8, 6.6 Hz, 1H), 1.70 (s, 3H), 1.38 (m, 2H), 0.97 (dt, J = 5.8, 1.4 Hz, 3H), 0.83 (s, 9H), 0.02 (s, 6H): ^{13}C NMR ($CDCl_3$) δ 156.07, 152.72, 151.02, 145.43, 142.33, 132.62, 119.29, 69.54, 59.28, 46.56, 30.54, 25.65, 18.47, 14.02, 10.54, −5.55. Anal. Calcd. for $C_{20}H_{33}N_5OSi$: C, 61.98; H, 8.58; N, 18.07. Found: C, 62.09; H, 8.65; N, 17.96.

(*rel*)-(1′*R*,2′*S*,3′*S*,4′*R*)-1-[4-(t-Butyldimethylsilyloxymethyl)-4-ethyl-2-methyl-2,3-dihydroxy-cyclopentan-1-yl] cytosine (16α) and (*rel*)-(1′*R*,2′*R*,3′*R*,4′*R*)-1-[4-(t-Butyldimethyl silyloxymethyl)-4-ethyl-2-methyl-2,3-dihydroxy-cyclopentan-1-yl] cytosine (16β): To a stirred solution of **14** (305

mg, 0.84 mmol) in cosolvent (4.0 mL, acetone/water = 5:1) was added NMO (393 mg, 1.68 mmol), and OsO_4 (0.7 mL, 4% in water). The mixture was stirred overnight at 50 °C, and quenched with saturated Na_2SO_3 solution (4 mL). Resulting solid was removed by filtration through a pad of Celite, and filtrate was concentrated in reduced pressure. The residue was purified by silica gel column chromatography ($MeOH/CH_2Cl_2$, 1:7) to give **16α** (197 mg, 59%) and **16β** yield (62 mg,19%): Spectroscopical data for **16α:** ^1H NMR (DMSO-d_6, 300 MHz) δ 7.57 (d, J = 7.2 Hz, 1H), 7.02 (br d, 2H, D_2O exchangeable), 5.58 (d, J = 7.2 Hz, 1H), 5.12 (d, J = 4.4 Hz, 1H, D_2O exchangeable), 5.02 (s, 1H, D_2O exchangeable), 3.89 (d, J = 4.8 Hz, 1H), 3.51 (dd, J = 10.4 Hz, 1H), 3.37 (dd, J = 10.4 Hz, 1H), 3.28 (s, 1H), 1.67-1.58 (m, 2H), 1.37 (dd, J = 5.6, 2.4 Hz, 2H), 1.29 (s, 3H), 0.96 (m, 3H), 0.83 (s, 9H), 0.02 (s, 6H): ^{13}C NMR (DMSO-d_6) δ 165.34, 156.17, 145.28, 94.65, 82.34, 77.39, 68.43, 54.43, 39.47, 25.49, 21.49, 20.21, 18.71, 16.37, 13.67, 9.99, −5.58; Anal. Calcd. for $C_{19}H_{35}N_3O_4Si$: C, 57.40; H, 8.87; N, 10.57. Found: C, 57.51; H, 8.76; N, 10.58; Spectroscopical data for **16β:** ^1H NMR (DMSO-d_6, 300 MHz) δ 7.37 (d, J = 7.2 Hz, 1H), 6.98 (br d, 2H, D_2O exchangeable), 5.54 (d, J = 7.2 Hz, 1H), 5.15 (br s, 1H, D_2O exchangeable), 5.04 (s, 1H, D_2O exchangeable), 3.92 (dd, J = 4.8, 2.4 Hz, 1H), 3.47 (d, J = 10.2 Hz, 1H), 3.37 (d, J = 10.2 Hz, 1H), 3.23 (s, 1H), 1.69-1.59 (m, 2H), 1.35 (m, 2H), 1.27 (s, 3H), 0.95 (m, 3H), 0.82 (s, 9H), 0.01 (s, 6H): ^{13}C NMR (DMSO-d_6) δ 165.54, 156.32, 145.71, 93.32, 81.97, 77.39, 68.72, 53.42, 38.39, 25.51, 22.34, 20.95, 18.62, 17.07, 14.67, 9.98, −5.48; Anal. Calcd. for $C_{19}H_{35}N_3O_4Si$: C, 57.40; H, 8.87; N, 10.57. Found: C, 57.37; H, 8.92; N, 10.46.

(*rel*)-(1′*R*,2′*S*,3′*S*,4′*R*)-9-[4-(t-Butyldimethylsilyloxymethyl)-4-ethyl-2-methyl-2,3-dihydroxy-cyclopentan-1-yl] adenine (17α) and (*rel*)-(1′*R*,2′*S*,3′*S*, 4′*R*)-9-[4-(t-Butyldimethyl silyloxymethyl)-4-ethyl-2-methyl-2,3-dihydroxy-cyclopentan-1-yl] adenine (17β); Adenine nucleoside analogue **17α** and **17α** were synthesized from **15** by the similar procedure as described for **16α, 16β:** Spectroscopical data for **17α:** yield 56%; ^1H NMR (DMSO-d_6, 300 MHz) δ 8.18 (s, 1H), 8.11 (s, 1H), 7.18 (br s, 2H, D_2O exchangeable), 5.14 (d, J = 4.8 Hz, 1H, D_2O exchangeable), 5.06 (s, 1H, D_2O exchangeable), 3.94 (d, J = 4.8 Hz, 1H), 3.48 (dd, J = 10.4 Hz, 1H), 3.32 (dd, J = 10.4 Hz, 1H), 3.30 (s, 1H), 1.75-1.69 (m, 2H), 1.32 (s, 3H), 1.22 (dd, J = 5.6, 2.4 Hz, 2H), 0.96 (m, 3H), 0.81 (s, 9H), 0.02 (s, 6H): ^{13}C NMR (DMSO-d_6) δ 155.68, 152.21, 148.69, 142.28, 118.29, 82.12, 77.36, 67,23, 58.73, 38.76, 25.58, 21.76, 20.65, 18.44, 16.21, 10.01, −5.65; Anal calc for $C_{20}H_{35}N_5O_3Si$: C, 56.98; H, 8.37; N, 16.61. Found: C, 57.10; H, 8.42; N, 16.70; Spectroscopical data for **17β**: yield 16%; ^1H NMR (DMSO-d_6, 300 MHz) δ 8.15 (s, 1H), 8.12 (s, 1H), 7.1 (br s, 2H, D_2O exchangeable), 5.15 (br s, J = 4.8 Hz, 1H, D_2O exchangeable), 5.09 (br s, 1H, D_2O exchangeable), 3.91 (dd, J = 5.0, 2.4 Hz, 1H), 3.51 (d, J = 9.8 Hz, 1H), 3.32 (d, J = 9.8 Hz, 1H), 3.34 (s, 1H), 1.70-1.61 (m, 2H), 1.35 (s, 3H),

1.24 (dd, J = 5.4, 2.6 Hz, 2H), 0.94 (m, 3H), 0.82 (s, 9H), 0.01 (s, 6H): ^{13}C NMR (DMSO-d_6) δ 156.01, 152.32, 148.43, 141.97, 118.32, 81.87, 77.43, 68.21, 59.33, 39.65, 25.64, 22.39, 21.76, 18.62, 16.99, 9.94, −5.57; Anal calc for $C_{20}H_{35}N_5O_3Si$: C, 56.98; H, 8.37; N, 16.61. Found: C, 56.87; H, 8.29; N, 16.54.

(*rel*)-(1′*R*,2′*S*,3′*S*,4′*R*)-1-[4-(Hydroxymethyl)-4-ethyl-2-methyl-2,3-dihydroxy-cyclopentan-1-yl] cytosine (18). To a solution of **16α** (163 mg, 0.41 mmol) in cosolvent (6.0 mL, THF/CH_3CN = 1:1), was TBAF (0.615 mL, 1.0 M solution in THF) at 0°C. The mixture was stirred overnight at room temperature, and concentrated. The residue was purified by silica gel column chromatography (MeOH/CH_2Cl_2, 1:4) to give **18** (91 mg, 79%) as a white solid: m.p. 193–195°C; UV (H_2O) λ_{max} 271.5 nm; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 7.58 (d, J = 7.2 Hz, 1H), 7.03 (br d, 2H, D_2O exchangeable), 5.58 (d, J = 7.2 Hz, 1H), 5.09 (d, J = 4.8 Hz, 1H, D_2O exchangeable), 5.01 (s, 1H, D_2O exchangeable), 4.86 (t, J = 4.8 Hz, 1H, D_2O exchangeable), 3.86 (m, 1H), 3.51 (d, J = 10.2 Hz, 1H), 3.44 (d, J = 10.2 Hz, 1H), 3.28 (s, 1H), 1.67-1.58 (m, 2H), 1.32 (s, 3H), 1.22 (m, 2H), 0.95 (m, 3H); ^{13}C NMR (DMSO-d_6) δ 165.54, 155.29, 146.28, 95.78, 81.34, 78.27, 68.47, 56.26, 39.02, 22.34, 21.17, 17.32, 10.03; Anal. Calcd. for $C_{13}H_{21}N_3O_4$ (+1.0 MeOH): C, 53.32; H, 7.99; N, 13.32. Found: C, 53.45; H, 8.07; N, 13.42.

(*rel*)-(1′*R*,2′*S*,3′*S*,4′*R*)-9-[4-(Hydroxymethyl)-4-ethyl-2-methyl-cyclopent-2-en-1-yl] adenine (19): Adenine nucleoside analogue **19** was synthesized from **17α** by the similar condition as described for **19** as a white solid: m.p. 206–209°C; UV (H_2O) λ_{max} 260.5 nm; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.18 (s, 1H), 8.10 (s, H1), 7.18 (br d, 2H, D_2O exchangeable), 5.16 (d, J = 4.6 Hz, 1H, D_2O exchangeable), 5.09 (s, 1H, D_2O exchangeable), 4.82 (t, J = 4.8 Hz, 1H, D_2O exchangeable), 3.86 (dd, J = 5.2, 2.4 Hz, 1H), 3.50 (d, J = 10.2 Hz, 1H), 3.38 (d, J = 10.2 Hz, 1H), 3.31 (s, 1H), 1.82-1.73 (m, 2H), 1.33 (s, 3H), 1.29 (dd, J = 5.6, 2.4 Hz, 2H), 0.96 (m, 3H); ^{13}C NMR (DMSO-d_6) δ 155.68, 152.39, 147.39, 142.43, 119.33, 81.90, 77.42, 68.64, 57.66, 39.11, 22.03, 20.21, 16.92, 9.98; Anal calc for $C_{14}H_{21}N_5O_3$ (+ 1.0 H_2O): C, 51.68; H, 7.12; N, 21.52. Found: C, 51.56; H, 7.07; N, 21.47.

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