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Synthesis and Antiviral Evaluation of Novel 3'- and 4'-Doubly Branched Carbocyclic Nucleosides as Potential Antiviral Agents

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A series of 3'- and 4'-branched carbocyclic nucleosides **25, 26, 27, 28, 29** and **30** were synthesized starting from simple acyclic ketone derivatives. The construction of the required quaternary carbon was made using a [3,3]-sigmatropic rearrangement. In addition, the installation of a methyl group in the 3'-position was accomplished using a Horner-Wadsworth-Emmons (HWE) reaction with triethyl 2-phosphonopropionate. Bis-vinyl was successfully cyclized using a Grubbs' catalyst (II). Natural bases (adenine, cytosine, uracil) were efficiently coupled with the use of a Pd(0) catalyst.

Key words: Doubly branched carbocyclic nucleosides, [3,3]-Sigmatropic rearrangement, Antiviral agents

INTRODUCTION

The discovery of novel nucleosides as antiviral and anticancer agents has been the goal of nucleoside chemists for a several decades. In particular, since the emergence of the HIV pandemic, extensive efforts have been concentrated on various modifications in the sugar moiety of nucleosides, resulting in FDA approved anti-HIV agents such as AZT (Furman *et al.*, 1986), ddC (Yarchoan *et al.*, 1988), ddI (Yarchoan *et al.*, 1989), d4T (Lin *et al.*, 1987), 3TC (Schinazi *et al.*, 1992), and abacavir (Daluge *et al.*, 1997). In addition, several nucleosides have been synthesized as anti-HBV agents including 3TC (Dienstag *et al.*, 1995), DAPD (Schinazi *et al.*, 1994), L-F-ddC (Lin *et al.*, 1994), L-FMAU (Chu *et al.*, 1995), and entecavir (Levine *et al.*, 2002), which are being developed at various stages (Fig. 1). However, the toxicities (Martin *et al.*, 1994; Parker *et al.*, 1994) associated with these nucleosides as well as the emergence of resistant viral strains (Shirasaka *et al.*, 1995, Chatis *et al.*, 1992) has prompted nucleoside chemists to search for additional novel and structurally diverse compounds with a minimal overlapping resistance and toxicity profiles. Among the several approaches to modify the structure of the nucleosides, carbocyclic nucleosides (Borthwick *et al.*, 1992; Agrofoglio *et al.*, 1994; Crimmins *et al.*, 1998) have been attracted with great interest because the replacement of the furanose ring offers greater metabolic stability to the endogenous phosphorylases (Herdewijn *et al.*, 1985), which cleave the glycosidic linkage. Another interesting feature of carbocyclic nucleosides is that a number of carbocyclic adenosine analogues are assumed to exert their antiviral action *via* the inhibition of *S*-adenosylhomocystenine hydrolase (Ueland *et al.*, 1982; Palmer *et al.*, 1979). Moreover, this mechanism might be

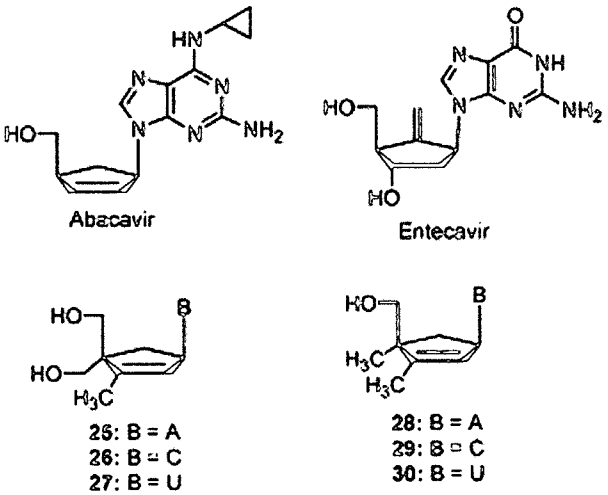

Fig. 1. Rationale of target nucleosides

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exploited in a combination therapy in association with the nucleosides with a different mechanism of action. In view of these interesting mechanisms and antiviral activities of carbocyclic nucleosides, this study synthesized and assayed novel 3'-methyl and 4'-alkyl doubly branched carbocyclic nucleosides.

MATERIALS AND METHODS

All the chemicals were of reagent grade and were used as purchased. All the moisture-sensitive reactions were performed in an inert atmosphere with either N_2 or Ar using distilled dry solvents. The NMR spectra were recorded on a bruker 300 Fourier transform spectrometer.

4,4'-Bis-(*t*-butyldimethylsilyloxy)-2-methylbut-2-en-1-ol (5)

To a solution of compound **3** (10 g, 24.83 mmol) in CH_2Cl_2 (300 mL), DIBALH (52.1 mL, 1.0 M solution in hexane) was added slowly at -50°C, and stirred for 2 h at the same temperature. To the resulting mixture, methanol (50 mL) was added. The mixture was stirred at room temperature for 2 h, 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, 1:7) to give the alcohol **5** (8.68 g, 97%) as a colorless oil: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 4.25 (s, 4H), 4.17 (s, 2H), 1.71 (s, 3H), 0.93 (s, 18H), 0.05 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 141.21, 125.39, 64.85, 59.40, 58.70, 25.83, 20.34, 18.24, -5.35;

(*E*)- and (*Z*)-4-(*t*-Butyldimethylsilyloxy)-2,3-dimethyl-but-2-en-1-ol (6)

Compound **6** was prepared from compound **4** using the method described for the allylic alcohol **5**: Yield: 90%; ^{1}H-NMR ($CDCl_3$, 300 MHz) as mixture δ 4.21 (s, 2H), 4.19 (s, 2H), 4.03 (s, 2H), 1.77, 1.74 (s, s, 3H), 1.64, 1.62 (s, s, 3H), 0.91 (s, 9H), 0.07 (s, 6H).

3,3'-Bis-(*t*-butyldimethylsilyloxy)-4-methylpent-4-enoic acid ethyl ester (7)

A solution of allylic alcohol **5** (15 g, 41.58 mmol) in triethyl orthoacetate (300 mL) and 1.5 mL of propionic acid was heated at 140°C overnight with constant stirring to allow for the removal of ethanol. An excess of triethyl orthoacetate was removed by distillation, and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:50) to give compound **7** (15.22 g, 85%) as a colorless oil: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 4.87 (s, 1H), 4.62 (s, 1H), 4.05 (q, *J* = 7.5 Hz, 2H), 3.65 (dd, *J* = 15.6, 9.0 Hz, 4H), 2.41 (s, 2H), 1.61 (s, 3H), 1.12 (t, *J* = 7.5 Hz, 3H), 0.94 (s, 18H), 0.02 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 171.92, 139.76, 114.48, 64.67, 59.88, 45.98, 36.84, 25.85, 20.34, 18.25, 14.25, -5.26.

(±)-3-(t-Butyldimethylsilyloxymethyl)-3,4-dimethylpent-4-enoic acid ethyl ester (8)

Compound **8** was prepared from compound **6** as described for compound **7**: yield 87%; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 4.85 (s, 1H), 4.65 (s, 1H), 4.05 (q, *J* = 7.2 Hz, 2H), 3.56 (dd, *J* = 9.3 Hz, 2H), 3.41 (d, *J* = 9.3 Hz, 2H), 2.42 (d, *J* = 3.2 Hz, 2H), 1.62 (s, 3H), 1.23 (t, *J* = 7.3 Hz, 3H), 1.12 (s, 3H), 0.91 (s, 9H), 0.05 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 171.96, 143.13, 113.02, 69.93, 59.84, 41.33, 25.81, 22.60, 20.70, 20, 45, 18.20, 14.26, -5.58.

3,3'-Bis-(*t*-butyldimethylsilyloxymethyl)-4-methylpent-4-enol (9)

To a solution of compound **7** (8.5 g, 19.73 mmol) in CH_2Cl_2 (300 mL), DIBALH (41.43 mL, 1.0 M solution in Hexane) was added slowly at 0°C, and stirred for 30 min at the same temperature. To the mixture, methanol (40 mL) was added. The mixture was stirred at room temperature for 2 h, 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, 1:25) to give compound **9** (7.09 g, 96%) as a colorless oil: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 4.82 (s, 1H), 4.62 (s, 1H), 3.57 (dd, *J* = 12.9, 9.6 Hz, 6H), 1.69 (s, 3H), 1.64 (dd, *J* = 6.0, 3.6 Hz, 2H), 0.87 (s, 18H), 0.04 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 146.45, 112.24, 63.41, 59.16, 47.99, 33.63, 25.81, 20.30, 18.16, -5.57.

(±)-3-(*t*-Butyldimethylsilyloxymethyl)-3,4-dimethylpent-4-enol (10)

Compound **10** was prepared from compound **8** using the method described for compound **9**; Yield 93%; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 4.79 (s,1H), 4.70 (s, 1H), 3.56 (t, *J* = 5.4 Hz, 2H), 3.52 (d, *J* = 9.6 Hz, 1H), 3.39 (d, *J* = 9.6 Hz, 1H), 1.73 (d, *J* = 4.2 Hz, 1H), 1.69 (s, 3H), 1.67 (d, *J* = 4.2 Hz, 1H), 1.00 (s, 3H), 0.85 (s, 9H), 0.02 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 149.49, 111.10, 69.77, 59.60, 43.10, 38.95, 25.93, 21.55, 20.00, 18.22, -5.29.

3,3'-Bis-(*t*-butyldimethylsilyloxymethyl)-4-methylpent-4-enal (11)

To a solution of compound **9** (5.0 g, 12.86 mmol) in CH_2Cl_2 (100 mL), 4Å molecular sieves (7.5 g) and PCC (6.93 g, 32.15 mmol) were added slowly at 0°C, and stirred for 3 h at room temperature. To the mixture, excess diethyl ether (500 mL) was added. The mixture was vigorously stirred for 2 h at the same temperature, and the resulting solid was filtered through a short silica gel column. The filtrate was concentrated under vacuum and

the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:50) to give compound **11** (4.47 g, 90%) as a colorless oil: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 9.65 (m, 1H), 4.96 (s, 1H), 4.75 (s, 1H), 3.64 (dd, *J* = 13.5, 9.3 Hz, 4H), 2.43 (s, 2H), 1.76 (s, 3H), 0.84 (s, 18H), 0.04 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 202.89, 145.02, 113.28, 65.01, 48.62, 45.44, 25.79, 20.42, 18.17, -5.67.

(±)-3-(*t*-Butyldimethylsilyloxymethyl)-3,4-dimethyl-pent-4-enal (12)

Compound **12** was prepared from compound **10** using the method described for compound **11**: Yield 92%; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 9.68 (m, 1H), 4.89 (s, 1H), 4.69 (s, 1H), 3.56 (d, *J* = 9.0 Hz, 1H), 3.42 (d, *J* = 9.0 Hz, 1H), .2.37 (d, *J* = 3.3 Hz, 2H), 1.75 (s, 3H), 1.13 (s, 3H), 0.83 (s, 9H), 0.01 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 203.35, 147.54, 112.23, 69.73, 49.53, 43.55, 25.81, 22.21, 20.03, 18.22, -5.61.

(±)-5,5'-Bis-(*t*-butyldimethylsilyloxymethyl)-6-methyl-hepta-1,6-dien-3-ol (13)

To a cooled (-78°C) solution of compound **11** (7.0 g, 18.1 mmol) in dry THF (120 mL) vinylmagnesium bromide (21.7 mL, 1.0 M solution in THF) was added slowly. After 2 h, a saturated NH_4Cl solution (22 mL) was added, and the reaction mixture was warmed slowly to room temperature. The mixture was extracted with EtOAc (2 × 200 mL). The combined organic layer was dried over $MgSO_4$, filtered, and evaporated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:20) to give compound **13** (6.0 g, 80%) as a colorless oil: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 5.82-5.71 (m, 1H), 5.17 (s, 1H), 5.11 (s, 1H), 4.96 (d, *J* = 9.9 Hz, 1H), 4.89 (s, 1H), 4.64 (s, 1H), 4.13 (t, *J* = 6.9 Hz, 1H), 3.74 (d, *J* = 9.3 Hz, 1H), 3.64 (d, *J* = 9.3 Hz, 1H), 3.54 (d, *J* = 9.3 Hz, 1H), 3.48 (d, *J* = 9.3 Hz, 1H), 3.40 (s, 1H), 1.68 (s, 3H), 1.63-1.43 (m, 2H), 0.82, 0.80 (s, s, 18H), 0.03, 0.01 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 146.59, 141.78, 113.25, 112.78, 69.26, 64.38, 63.71, 48.32, 38.86, 25.80, 20.39, 18.15, -5.54, -5.66.

(*rel*)-(3*R* and 3*S*,5*S*)-5-(*t*-Butyldimethylsilyloxymethyl)-5,6-dimethyl-hepta-1,6-dien-3-ol (14)

Compound **14** was prepared from compound **12** using the method described for compound **13**: yield 82%; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 5.19-5.11 (m, 1H), 5.19-5.11 (m, 1H), 4.98-4.93 (m, 1H), 4.85-4.84 (m, 2H), 4.12 (m, 1H), 3.62-3.37 (m, 2H), 1.71-1.52 (m, 5H), 1.05, 1.01 (s, s, 3H), 0.84, 0.83 (s, s, 9H), 0.04, 0.01 (s, s, 6H).

(±)-4,4'-Bis-(*t*-butyldimethylsilyloxymethyl)-3-methyl-cyclopent-2-enol (15)

To a solution of compound **13** (2.5 g, 6.02 mmol) in dry CH_2Cl_2 (7 mL) Grubbs' catalyst (II) (255 mg 0.3 mmol) in dry CH_2Cl_2 (3 mL) was added slowly over a 10-minute period under a N_2 atmosphere. The reaction mixture was refluxed overnight, and cooled to room temperature. The mixture was then concentrated under vacuum, and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:10) to give the cyclopentenol, **15** (2.06 g, 89%) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 5.58 (s, 1H), 4.40 (t, *J* = 9.0 Hz, 1H), 3.65 (d, *J* = 9.6 Hz, 1H), 3.50-3.39 (m, 3H), 2.75 (d, *J* = 10.8 Hz, 1H), 2.04 (dd, *J* = 14.1, 6.9 Hz, 1H), 1.63 (s, 3H), 0.88 (s, 18H), 0.05 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 145.14, 131.13, 73.61, 66.77, 64.72, 56.60, 42.21, 26.98, 18.48, 13.28, -5.62.

(*rel*)-(1*R*,4*S*)-4-(*t*-Butyldimethylsilyloxymethyl)-3,4-dimethyl-cyclopent-2-enol (16β); and (*rel*)-(1*S*,4*S*)-4-(*t*-Butyldimethylsilyloxymethyl)-3,4-dimethyl-cyclopent-2-enol (16α)

Compound **16β** and **16α** were prepared from compound **14** using the method described for compound **15**: yield for **16β**, 48%, yield for **16α**, 47%; Compound **16β**: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 5.51 (s, 1H), 4.31 (t, *J* = 9.3 Hz, 1H), 3.39 (d, *J* = 9.3, Hz, 1H), 3.23 (d, *J* = 9.3 Hz, 1H), 1.90 (dd, *J* = 14.1, 6.6 Hz, 1H), 1.66 (dd, *J* = 14.1, 6.9 Hz, 1H), 1.48 (s, 3H), 0.81 (s, 3H), 0.78 (s, 9H), 0.04 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 146.66, 129.87, 73.39, 67.51, 50.63, 46.83, 25.99, 21.94, 18.51, 11.99, -5.27: Compound **16α**: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 5.53 (s, 1H), 4.30 (d, *J* = 6.3 Hz, 1H), 3.38 (dd, *J* = 12.6, 9.3, Hz, 2H), 1.88 (dd, *J* = 13.4, 6.4 Hz, 1H), 1.65 (dd, *J* = 13.4, 6.9 Hz, 1H), 1.53 (s, 3H), 0.85 (s, 3H), 0.80 (s, 9H), 0.03 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 145.98, 129.89, 73.40, 67.67, 51.63, 46.83, 26.05, 22.04, 18.61, 12.45, -5.57.

(±)-1-Ethoxycarbonyloxy-4,4'-bis-(*t*-butyldimethylsilyloxymethyl)-3-methyl-cyclopent-2-ene (17)

To a solution of compound **15** (5.58 g, 14.43 mmol) in anhydrous pyridine (50 mL) ethyl chloroformate (2.76 mL, 28.87 mmol) and DMAP (0.17 g, 1.4 mmol) were added. The reaction mixture was stirred overnight at room temperature. The reaction mixture was quenched using a saturated $NaHCO_3$ solution (2 mL) and concentrated under vacuum. The residue was extracted with EtOAc, dried over $MgSO_4$, filtered, and concentrated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:50) to give compound **17** (5.42 g, 82%) as a colorless syrup: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 5.50 (s, 1H), 5.43 (d, *J* = 6.9 Hz, 1H), 4.15 (q, *J* = 7.5 Hz, 2H), 3.65-3.49 (m, 4H), 2.19 (dd, *J* = 14.4, 7.8 Hz, 1H), 1.80-1.68 (m, 3H), 1.26 (t, *J* = 7.5 Hz, 3H), 0.85 (s, 18H), 0.03 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 155.14, 151.30, 125.29, 81.80, 66.24, 65.10, 63.49, 56.70, 37.52, 25.83, 18.21, 14.38, -5.56.

(*rel*)-(1*R*,4*S*)-1-Ethoxycarbonyloxy-4-(*t*-butyldimethylsilyloxymethyl)-3,4-dimethylcyclopent-2-ene (18)
Compound **18** was prepared from compound **16** using the method described for compound **17**: Yield 80%: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 5.44 (s, 1H), 5.41 (s, 1H), 4.41 (q, *J* = 7.8 Hz, 2H), 3.41 (s, 2H), 1.96 (d, *J* = 5.4 Hz, 2H), 1.67 (s, 3H), 1.26 (t, *J* = 7.8 Hz, 3H), 1.00 (s, 3H), 0.86 (s, 9H), 0.02 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 155.15, 153.96, 123.42, 81.57, 69.63, 63.48, 50.91, 41.74, 25.94, 22.00, 18.24, 14.32, 13.39, -5.51.

(±)-9-[4-Bis-(*t*-butyldimethylsilyloxymethyl)-3-methylcyclopent-2-en-1-yl] adenine (19)
To pure NaH (23.4 mg, 0.98 mmol) in anhydrous DMSO (3.4 mL), adenine (134 mg, 0.98 mmol) was added. The reaction mixture was stirred for 30 min at 50-55 °C and cooled to room temperature. Simultaneously, P(O-*i*-Pr)$_3$ (0.096 mL, 0.22 mmol) was added to a solution of $Pd_2(dba)_3.CHCl_3$ (4.6 mg, 2.5μmol) in anhydrous THF (3.0 mL), which was stirred for 40 min. To the adenine solution in DMSO, the catalyst solution of THF and **17** (403 mg, 0.88 mmol) dissolved in anhydrous THF (3 mL) were added slowly. The reaction mixture was stirred overnight at a refluxing temperature and quenched with water (2 mL). The reaction solvent was removed under reduced pressure. The residue was purified by silica gel column chromatography (MeOH/CH_2Cl_2, 1:10) to give compound **19** (221 mg, 50%) as a white solid: mp 181-183 °C; UV (MeOH) λ_{max} 261.5 nm; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 8.34 (s, 1H), 7.92 (s, 1H), 5.63 (dd, *J* = 8.1, 2.7 Hz, 1H), 5.54 (d, *J* = 14.1 Hz, 1H), 3.68 (d, *J* = 10.2 Hz, 1H), 3.53 (s, 3H), 3.50 (d, *J* = 10.2 Hz, 1H), 2.57 (dd, *J* = 13.8, 8.1 Hz, 1H), 1.90 (dd, *J* = 13.8, 5.7 Hz, 1H), 1.80 (s, 3H), 0.87, 0.85 (s, s, 18H), 0.03, 0.01 (s, s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 155.26, 152.73, 149.97, 139.20, 125.00, 119.78, 66.02, 65.12, 57.87, 57.24, 39.33, 25.90, 18.38, 13.92, -5.55.

(±)-1-[4-Bis-(*t*-butyldimethylsilyloxymethyl)-3-methylcyclopent-2-en-1-yl] cytosine (20)
Compound **20** was prepared from compound **17** using the method described for compound **19**; Yield 40%; mp 170-173 °C; UV (MeOH) λ_{max} 271.5 nm; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 7.51 (d, *J* = 7.2 Hz, 1H), 5.95 (d, *J* = 7.2 Hz, 1H), 5.56 (s, 1H), 4.42 (br s, 1H), 3.65 (d, *J* = 9.4 Hz, 1H), 3.50-3.39 (m, 3H), 2.76 (d, *J* = 10.2 Hz, 1H), 2.04 (dd, *J* = 14.2, 7.0 Hz, 1H), 1.62 (s, 3H), 0.87 (s, 18H), 0.04 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 165.14, 155.90, 142.91, 141.20, 131.12, 93.21, 73.61, 66.77, 64.72, 56.60, 42.21, 25.78, 18.38, 13.20, -5.52.

(±)-1-[4-Bis-(*t*-butyldimethylsilyloxymethyl)-3-methylcyclopent-2-en-1-yl]uracil (21)
Compound **21** was synthesized from compound **17** using the described for compound **19**: yield 36%; mp 168-170°C; UV (MeOH) λ_{max} 266.5 nm; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 7.69 (d, *J* = 8.0 Hz, 1H), 5.60 (d, *J* = 8.0 Hz, 1H), 5.57 (d, *J* = 8.1 Hz, 1H), 5.48 (d, *J* = 12.1 Hz, 1H), 3.68 (d, *J* = 10.0 Hz, 1H), 3.50 (m, 3H), 2.51 (dd, *J* = 12.8, 8.2 Hz, 1H), 1.91 (dd, *J* = 12.8, 5.6 Hz, 1H), 1.82 (s, 3H), 0.85 (s, 18H), 0.03, 0.01 (s, 12H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 163.09, 151.27, 145.14, 141.89, 140.21, 100, 01, 66.02, 65.12, 57.87, 57.24, 39.33, 25.90, 18.38, 13.92, -5.55.

(*rel*)-(1′*R*,4′*S*)-9-[4-(*t*-Butyldimethylsilyloxymethyl)-3,4-dimethylcyclopent-2-en-1-yl] adenine (22)
Compound **22** was prepared from compound **18** using the method described for compound **19**: yield 54%; mp 188-190 °C; UV (MeOH) λ_{max} 262 nm; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 8.45 (s, 1H), 7.29 (s, 1H), 5.51 (s, 1H), 5.45 (d, *J* = 6.6 Hz, 1H), 3.40 (d, *J* = 9.0, Hz, 1H), 3.31 (d, *J* = 9.0 Hz, 1H), 1.91 (dd, *J* = 13.8, 6.4 Hz, 1H), 1.66 (dd, *J* = 13.8, 6.4 Hz, 1H), 1.49 (s, 3H), 0.88 (s, 3H), 0.84 (s, 9H), 0.02 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 155.36, 152.41, 150.51, 146.66, 139.30, 129.87, 119.07, 73.39, 67.51, 50.63, 46.83, 26.10, 22.34, 18.61, 12.12, -5.37.

(*rel*)-(1′*R*,4′*S*)-1-[4-(*t*-Butyldimethylsilyloxymethyl)-3,4-dimethylcyclopent-2-en-1-yl] cytosine (23)
Compound **23** was prepared from compound **18** using the method described for compound **19**: yield 41%; mp 169-172 °C; UV (MeOH) λ_{max} 271 nm; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 7.48 (d, *J* = 7.0 Hz, 1H), 5.94 (d, *J* = 7.1 Hz, 1H), 5.53 (s, 1H), 5.40 (s, 1H), 3.40 (dd, *J* = 13.6, 9.3, Hz, 2H), 1.88 (dd, *J* = 12.6, 7.0 Hz, 1H), 1.64 (dd, *J* = 12.0, 7.0 Hz, 1H), 1.50 (s, 3H), 0.85 (s, 3H), 0.84 (s, 9H), 0.03 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 165.77, 155.89, 146.66, 142.09, 130.85, 93.87, 77.26, 67.05, 51.24, 47.38, 25.98, 22.23, 19.01, 11.90, -5.45.

(*rel*)-(1′*R*,4′*S*)-1-[4-(*t*-Butyldimethylsilyloxymethyl)-3,4-dimethylcyclopent-2-en-1-yl]uracil (24)
Compound **24** was prepared from compound **18** using the method described for compound **19**: yield 39%; mp 171-173°C; UV (MeOH) λ_{max} 268.5 nm; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 7.42 (d, *J* = 7.8 Hz, 1H), 5.53 (s, 1H), 5.44 (d, *J* = 7.8 Hz, 1H), 5.23 (d, *J* = 4.3 Hz, 1H), 3.49 (d, *J* = 8.6, Hz, 1H), 3.36 (d, *J* = 8.6 Hz, 1H), 1.78 (dd, *J* = 13.4, 7.2 Hz, 1H), 1.67 (dd, *J* = 13.4, 7.2 Hz, 1H), 1.67 (s, 3H), 0.85 (s, 3H), 0.79 (s, 9H), 0.05 (s, 6H); ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 164.34, 151.27, 144.45, 142.99, 123.45, 101.02, 75.78, 67.51, 55.24, 45.73, 26.21, 21.95, 18.57, 12.23, -5.65.

(±)-9-[4,4′-Bis-(hydroxymethyl)-3-methylcyclopent-2-en-1-yl]adenine (25)
To a solution of compound **19** (200 mg, 0.397 mmol) in THF (3 mL), TBAF (1.19 mL, 1.0 M solution in THF) at

0°C was added. The mixture was stirred at room temperature for 5 h, and concentrated. The residue was purified by silica gel column chromatography (MeOH/CH_2Cl_2, 1:4) to give compound **25** (93 mg, 86%) as a white solid: mp 200-203°C; UV (H_2O) λ_{max} 261.5 nm; ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 8.11 (s, 1H), 8.05 (s, 1H), 7.18 (br s, 2H), 5.52 (s, 1H), 5.49 (m, 1H), 4.71 (dt, *J* = 14.7, 5.1 Hz, 2H), 3.48 (dd, *J* = 11.1, 5.7 Hz, 1H), 3.37 (dd, *J* = 11.1, 5.0 Hz, 1H), 2.53 (dd, *J* = 13.8, 9.0 Hz, 1H), 1.90 (dd, *J* = 13.5, 5.1 Hz, 1H), 1.72 (s, 3H); ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 155.94, 152.17, 149.19, 148.43, 138.83, 125.20, 118.93, 64.03, 63.38, 57.90, 57.01, 38.16, 13.18.

(±)-1-[4,4'-Bis-(hydroxymethyl)-3-methylcyclopent-2-en-1-yl]cytosine (26)

Compound **26** was prepared from compound **20** using the method described for compound **25**; Yield: 81%; mp 168-171°C; UV (H_2O) λ_{max} 271 nm; ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 7.50 (d, *J* = 7.2 Hz, 1H), 5.91 (d, *J* = 7.2 Hz, 1H), 5.56 (s, 1H), 5.48 (m, 1H), 4.62 (dd, *J* = 12.4, 5.6 Hz, 2H), 3.65 (d, *J* = 9.4 Hz, 1H), 3.58 (m, 2H), 2.76 (dd, *J* = 13.8, 4.8 Hz, 1H), 2.00 (dd, *J* = 13.8, 6.8 Hz, 1H), 1.72 (s, 3H); ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 165.14, 155.90, 142.91, 141.20, 131.12, 93.21, 73.61, 66.77, 64.72, 56.60, 42.21, 13.20.

(±)-1-[4,4'-Bis-(hydroxymethyl)-3-methylcyclopent-2-en-1-yl]uracil (27)

Compound **27** was prepared from compound **21** using the compound described for compound **25**; Yield: 75%; mp 166-169 °C; UV (H_2O) λ_{max} 267.5 nm; ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 7.38 (d, *J* = 8.0 Hz, 1H), 5.45 (d, *J* = 7.8 Hz, 1H), 5.50 (dd, *J* = 10.2, 5.6 Hz, 1H), 5.42 (d, *J* = 8.4 Hz, 1H), 3.72 (dd, *J* = 12.8, 6.6 Hz, 2H), 3.48 (m, 2H), 2.54 (dd, *J* = 12.6, 6.2 Hz, 1H), 1.90 (dd, *J* = 12.6, 5.2 Hz, 1H), 1.80 (s, 3H); ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 164.89, 152.37, 146.10, 142.89, 144.21, 101, 21, 66.12, 65.12, 57.87, 57.24, 38.73, 13.92.

(*rel*)-(1'*R*,4'*S*)-9-[4-(Hydroxymethyl)-3,4-dimethylcyclopent-2-en-1-yl]adenine (28)

Compound **28** was prepared from compound **22** using the compound described for compound **25**; Yield: 86%; mp 189-191°C; UV (H_2O) λ_{max} 261.5 nm; ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 8.33 (s, 1H), 8.01 (s, 1H), 5.85 (s, 1H), 5.55 (d, *J* = 6.6 Hz, 1H), 3.40 (d, *J* = 9.0, Hz, 1H), 3.31 (d, *J* = 9.0 Hz, 1H), 2.49 (dd, *J* = 14.0, 6.4 Hz, 1H), 1.70 (dd, *J* = 14.0, 7.0 Hz, 1H), 1.55 (s, 3H), 0.92 (s, 3H); ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 155.94, 152.40, 149.51, 140.66, 139.30, 129.87, 119.02, 74.21, 67.54, 50.73, 46.80, 22.34, 12.12.

(*rel*)-(1'*R*,4'*S*)-1-[4-(Hydroxymethyl)-3,4-dimethylcyclopent-2-en-1-yl]cytosine (29)

Compound **29** was prepared from compound **23** using the method described for compound **25**; Yield: 70%; mp 165-168 °C; UV (H_2O) λ_{max} 271.5 nm; ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 7.46 (d, *J* = 7.8 Hz, 1H), 5.89 (m, 1H), 5.74 (d, *J* = 7.8 Hz, 1H), 5.53 (s, 1H), 3.43 (dd, *J* = 13.2, 9.0, Hz, 2H), 2.21 (dd, *J* = 13.2, 9.0 Hz, 1H), 1.74 (dd, *J* = 13.2, 6.3 Hz, 1H), 1.66 (s, 3H), 0.87 (s, 3H); ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 165.98, 156.02, 143.66, 142.11, 129.80, 93.07, 78.26, 21.05, 50.9, 47.28, 22.56, 12.30.

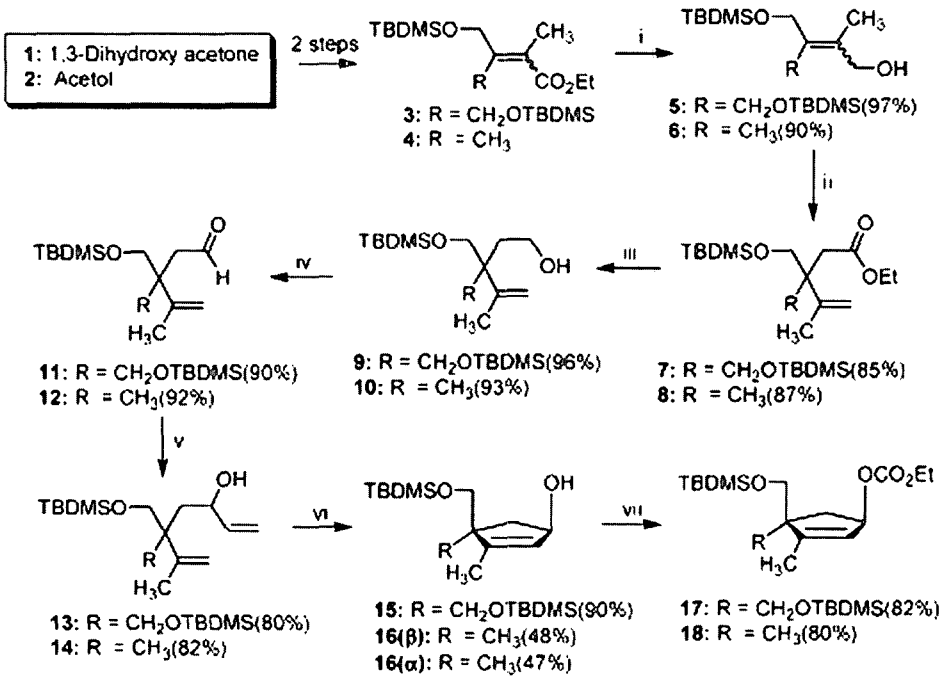
(*rel*)-(1'*R*,4'*S*)-1-[4-(Hydroxymethyl)-3,4-dimethylcyclopent-2-en-1-yl]uracil (30)

Compound **30** was prepared from compound **24** using the method described for compound **25**; Yield: 79%; mp 168-171°C; UV (H_2O) λ_{max} 268 nm; ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 7.67 (d, *J* = 7.8 Hz, 1H), 5.63 (m, 1H), 5. 54 (d, *J* = 7.8 Hz, 1H), 5.33 (d, *J* = 4.3 Hz, 1H), 3.49 (dd, *J* = 8.6, Hz, 2H), 2.23 (dd, *J* = 13.0, 9.0 Hz, 1H), 1.77 (dd, *J* = 13.0, 6.8 Hz, 1H), 1.67 (s, 3H), 0.89 (s, 3H); ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 163.24, 151.56, 145.75, 143.12, 123.45, 102.12, 74.23, 67.41, 55.36, 45.42, 21.95, 12.23.

RESULTS AND DISCUSSION

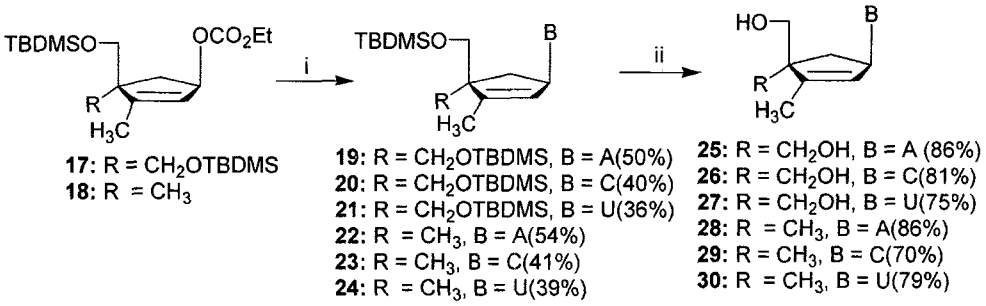
Although the synthetic methods for 4'α-*C* alkyl branched furanose (Kitano *et al.*, 1997; Waga *et al.*, 1993) and carbocyclic nucleosides (Kato *et al.*, 1999) have been reported, no synthetic example of a branched nucleoside with alkyl substituents at both 3'- and 4'-positions has been reported. The lack of suitable examples may be due to the synthetic difficulties in obtaining the necessary quaternary carbon. As shown in Scheme 1, it was envisioned that the ring-closing metathesis of the proper bis-olefin **13** and **14**, which could be readily synthesized via a sequential [3,3]-sigmatropic rearrangement and carbonyl addition beginning with simple acyclic precursors, would produce doubly branched cyclopentenes, **15** and **16**, as the key intermediates.

The protection of the hydroxyl on the commercially available starting materials, **1** and **2**, with TBDMSCl (*t*-butyldimethylchlorosilane) followed by the Horner-Wadsworth-Emmons (HWE) reaction (Jeong *et al.*, 1998) provided the α,β-unsaturated ethyl ester, **3** and **4**, as *cis*/*trans* isomeric mixtures. It was unnecessary to separate these isomers, because they were merged into one racemic mixture in the subsequent reaction. Esters **3** and **4**, were reduced to their respective allylic alcohols, **5** and **6**, using diisobutylaluminum hydride (DIBALH) in a high yield, which were subjected to a normal Johnson's orthoester Claisen rearrangement (Hong *et al.*, 2000, Hong *et al.*, 1999) using triethyl orthoacetate to give the γ,δ–unsaturated esters, **7** and **8**, in an 85-87% yield. The addition of DIBALH to a solution of the esters, **7** and **8**, in CH_2Cl_2 at 0°C furnished the alcohols, **9** and **10**, which were sequentially subjected to PCC oxidation and carbonyl



Reagents i) Dibal-H, CH_2Cl_2, -50°C; ii) Triethylorthoacetate, propionic acid, 140°C; iii) Dibal-H, CH_2Cl_2, 0°C; iv) PCC, 4A MS, CH_2Cl_2, rt; v) CH_2=CHMgBr, THF, Dibal-H, -78°C; vi) Grubbs' catalyst (II) CH_2Cl_2, reflux; vii) $ClCO_2Et$, DMAP, pyridine, rt

Scheme 1. Synthesis of allylic coupling intermediates



Reagents: i) Bases (adenine, cytosine, uracil), $Pd_2(dba)_3.CHCl_3$, P(O-*i*-Pr)$_3$, NaH, THF/DMSO, reflux, overnight; ii) TBAF, THF,rt.

Scheme 2. Synthesis of final nucleosides

addition by CH_2=CHMgBr to yield the bis-olefins, **13** and **14**, as stereoisomeric mixtures.

Bis-olefin **13** was subjected to the standard ring-closing metathesis conditions using a Grubb's catalyst (II) [(Im) $Cl_2PCy_3RuCHPh$] provide the cyclopentenols, **15**. In the case of compound **14**, the stereoisomers, **16(α)** and **16 (β)**, were prepared in equal amounts. The stereochemistry of compound **16(α)** and **16(β)** was unambiguously assigned based on the NOE correlations between the proximal hydrogen and methyl group (H-1, vs. CH_3-4).

In order to couple the cyclopentenols with the bases (adenine, cytosine and uracil) using a simple nucleophilic substitution type reaction, compounds **15** and **16(α)** were subjected to a mesylation reaction (MsCl, TEA, CH_2Cl_2). Unexpectedly, the reactions had a very low yield (20-30%) and were irreproducible. Therefore, attention was turned to palladium(0) catalyzed reactions (Hong *et al.*, 2002; Hong *et al.*, 2003) for the coupling of bases.

The cyclopentenols, **15** and **16(β)**, were activated to compounds **17** and **18** using ethyl chloroformate in a high yield (80-82%), which were coupled with an adenine anion generated by NaH/DMSO using the well-known coupling catalyst [tris(dibenzylidene-acetone)-dipalladium(0)-chloroform] adduct to give compounds

Table I. The antiviral activities of the synthesized compounds

	HIV-1 EC_{50} (μg/mL)	HSV-1 EC_{50} (μg/mL)	HSV-2 EC_{50} (μg/mL)	EMCV EC_{50} (μg/mL)	cytotoxicity IC_{50} (μg/mL)
25	38.58	>100	>100	46.6	>100
26	>100	>100	>100	>100	>100
27	>100	>100	>100	>100	>100
28	>100	>100	>100	>100	>100
29	>100	>100	>100	55.4	>100
30	>100	>100	>100	69.87	>100
AZT	0.002	ND	ND	ND	5.41
ACV	ND	1.95	1.95	ND	>10
Ribavirin	ND	ND	ND	20.56	300.00

ND: Not Determined

19~24. The required stereochemistry of the nucleosides **19~24** were successfully controlled from the β-configuration of compounds **17** and **18** *via* a Pd (0) catalyzed π–allyl complex mechanism. The desilylations of compounds **19~24** were performed by a treatment with tetrabutylammonium fluoride (TBAF) to give the final nucleosides **25~30** in a 70-86% yield.

The antiviral assays against the HIV-1, HSV-1, HSV-2 and EMCV were performed and the results are shown in Table I. As shown in Table I, Adenine **25** showed moderate activity against HIV-1 and EMCV. In addition, cytosine **29** and the uracil analogue **30** showed weak antiviral activity against the EMCV.

In summary, a concise synthetic method for synthesizing 3'-methyl and 4'α-alkyl doubly branched carbocyclic nucleosides from simple α-hydroxy ketone derivatives was developed. This procedure highlights the simplicity and efficiency in constructing the vicinal alkyl branches at the cyclopentene ring systems of nucleosides.

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REFERENCES

Agrofoglio, L., Suhas, E., Farese, A., Condom, R., Challand, S., Earl, R. A., and Guedj, R., Synthesis of carbocyclic nucleosides. *Tetrahedron*, 50, 10611-10670 (1994).

Borthwick, A. D. and Biggadike, K., Synthesis of chiral carbocyclic nucleosides. *Tetrahedron*, 48, 571-623 (1992).

Chu, C. K., Ma, T. W., Shanmuganathan, K., Wang, C.-G., Xiang, Y.-J., Pai, S. B., Yao, G.-Q., Sommadossi, J.-P., and Cheng, Y.-C., Use of 2'-fluoro-5-methyl-β-L-arabinofuranosyluracil as a novel antiviral agent for hepatitis-B virus and epstein-barr-virus. *Antimicrob. Agents Chemother.*, 39, 979-981 (1995).

Chatis, P. A. and Crumpacker, C. S., Resistance of herpesvirus to antiviral drugs. *Antimicrob. Agents Chemother.*, 36, 1589-1595 (1992).

Crimmins, M. T., King, B. W., Zuercher, W. J., and Choy, A. L., An efficient, general asymmetric synthesis of carbocyclic nucleosides: application of an asymmetric aldol/ring-closing metathesis strategy. *J. Org. Chem.*, 65, 8499-8500 (2000).

Daluge, S. M., Good, S. S., Faletto, M. B., Miller, W. H., StClair, M. H., Boone, L. R., Tisdale, M., Parry, N. R., Reardon, J. E., Dornsife, R. E., Averett, D. R., and Krenitsky, T. A., 1592U89, a novel carbocyclic nucleoside analog with potent, selective anti-human immunodeficiency virus activity. *Antimicrob. Agents Chemother.*, 41, 1082-1093 (1997).

Dienstag, J. L., Perrillo, R. P., Schiff, E. R. Bartholomew, M., Vicary, C., and Rubin, M., A preliminary trial of lamivudine for chronic hepatitis-B infection. *New Engl. J. Med.*, 333, 1657-1661 (1995).

Furman, P. A. Fyfe, J. A., St. Clair, M. H., Weinhold, K., Rideout, J. L., Freeman, G. A., Nusinoff-Lehrman, S., Bolognesi, D. P., Broder, S., Mitsuya, H., and Barry, D. W., Phosphorylation of 3'-azido-3'-deoxythymidine and selective interaction of the 5'-triphosphate with human immunodeficiency virus reverse transcriptase. *Proc. .Natl. Acad. Sci. USA*, 83, 8333-8337 (1986).

Jeong, L. S., Lee, Y. A., Moon, H. Y., and Chun, M. W., Synthesis and antiviral activity of apio dideoxy nucleosides with azido or amino substituent. *Nucleosides & Nucleotides*, 17, 1479-1487 (1998).

Herdewijn, P., De Clercq, E., Balzarini, J., and Vanderhaeghe, H. Synthesis and antiviral activity of the carbocyclic analogues of (*E*)-5-(2-halovinyl)-2'-deoxyuridines and (*E*)-5-(2-halovinyl)-2'-deoxycytidines. *J. Med. Chem.*, 28, 550-555 (1985).

Hong, J .H., Gao, M. Y., and Chu, C. K., Synthesis of novel 3'-*C*-methyl-4'-thioapionucleosdies *via* highly enantioselective elaboration of quaternary carbon [3,3]-sigmatropic rearrangement. *Tetrahedron Lett.*, 40, 231-234 (1999).

Hong, J. H., Gao, M. Y., Choi, Y., Cheng, Y.-C., Schinazi, and Chu, C. K., Synthesis of novel 3'-*C*-methylapionucleosides: an asymmetric construction of a quaternary carbon by Claisen rearrangement. *Carbohydrate Res.*, 328, 37-48 (2000).

Hong, J. H., Shim, M. J., Ro, B. O., and Ko, O. H., An efficient synthesis of novel carbocyclic nucleosides with use of ring-closing metathesis from D-lactose. *J. Org. Chem.*, 67, 6387-6840 (2002).

Hong, J. H., Oh, C. H., and Cho, J. H., Stereocontrolled synthesis of novel 6'(α)-hydroxy carbovir analogues. *Tetrahedron*, 59, 6103-6108 (2003).

Kato, K., Suzuki, H., Tanaka, H., Miyasaka, T., Baba, M., Yamaguchi, K., and Akita, H., Stereoselective synthesis of 4-α-carbovir derivative based on an asymmetric synthesis of chemoenzymatic procedure. *Chem. Pharm. Bull.*, 47, 1256-1264 (1999).

Kitano, K. and Miura, S., Synthesis of 4'-*C*-fluoromethyl-nucleosides as potential antineoplastic agents. *Tetrahedron*, 53, 13315-13322 (1997).

Levine, S., Hernandez, D., Yamanaka, G., Zhang, S., Rose, R., Weinheimer, S., and Colonno, R. J., Efficacies of entecavir against lamivudine-resistance hepatitis B virus replication and recombinant polymerase *in vitro*. *Antimicrob. Agents Chemother.*, 46, 2525-2532 (2002).

Lin, T.-S., Schinazi, R. F., and Prusoff, W. H., Potent and selective *in vitro* activity of 3'-deoxythymidine-2'-ene-(3'-deoxy-2',3'-dideoxydehydrothymidine) against human immunodeficiency virus. *Biochem. Pharmacol.*, 36, 2713-2718 (1987).

Lin, T. S., Luo, M. Z., Liu, M. C., Pai, S. B., Dutschman, G. E., and Cheng, Y.-C., Synthesis and biological evaluation of 2',3'-dideoxy-L-pyrimidine nucleosides as potential antiviral agents against human-immunodeficiency-virus (HIV) and hepatitis-B-virus (HBV). *J. Med. Chem.*, 37, 798-803 (1994).

Martin, J. L., Brown, C. E., Mattews-Davis, N., and Reardon, J. E., Effects of antiviral nucleoside analogs on human DNA polymerase and mitochondrial DNA synthesis. *Antimicrob. Agents Chemother.*, 38, 2743-2749 (1994).

Palmer, J. L. and Abeles, R. H., The mechanism of action of *S*-adenosylhomocysteinase. *J. Biol. Chem.*, 254, 1217-1226 (1979).

Parker, W. B. and Cheng, Y. C., Mitochondrial toxicity of antiviral nucleoside analogues. *J. NIH Res.*, 6, 57-61 (1994).

Shirasaka, T., Kavlick, M. F., Ueno, T., Gao, W. Y., Kojima, E., Alcaide, M. L., Chokekijchai, S., Roy, B. M., Arnol, E., Yarchoan, R., and Mitsuya, H., Emergence of human-immunodeficiency-virus type-1 variants with resistance to multiple dideoxynucleosides in patients receiving therapy with dideoxynucleosides. *Proc. Natl. Acad. Sci. USA*, 92, 2398-2402 (1995).

Schinazi, R. F., Chu, C. K., Peck, A., McMillan, A., Mathis, R. Cannon, D., Jeong, L. S., Beach, J. W., Choi, W. B., Yeola, S., and Liotta, D. C., Activities of the four optical isomers of 2',3'-dideoxy-3'-thiacytidine (BCH-189) against human immunodeficiency virus type I in human lymphocytes. *Antimicrob. Agents Chemother.*, 36, 672-676 (1992).

Schinazi, R. F., McClure, H. M., Boudinot, F. D., Xiang, Y.-J., and Chu, C. K., Development of (-)-β-D-2,6-diaminopurines dioxolane as a potential antiviral agent. *Antiviral Res.*, 23, suppl. 81 (1994).

Ueland, P. M., Pharmacological and biochemical aspects of *S*-adenosylhomocysteine and *S*-adenosylhomocysteine hydrolase. *Pharmacol. Rev.*, 34, 223-253 (1982).

Waga, T., Nishizaki, T., Miyakawa, J., Ohrui, H., and Meguro, H., Synthesis of 4'-*C*-methyl nucleosides. *Biosci. Biotechnol. Biochem.*, 57, 1433-1438 (1993).

Yarchoan, R., Thomas, R. V., Allain, J.-P., McAtee, N., Dubinsky, R., Mitsuya, H., Lawley, T. J., Safai, B., Myers, C. E., Perno, C. F., Klecker, R. W., Wills, R. J., Fischl, M. A., McNeely, M. C., Pluda, J. M., Leuther, M., Collins, J. M., and Broder, S., The phase I studies of 2',3'-dideoxycytidine in severe human immunodeficiency virus infection as single agent and alternating with zidovudine (AZT). *Lancet*, 1, 76-81 (1988).

Yarchoan, R., Mitsuya, H., Thomas, R. V., Pluda, J. M., Hartman, N. R., Perno, C. F., Marczyk, K. S., Allain, J.-P., Johns, D. G., and Broder, S., *In vivo* activity against HIV and favorable toxicity profile of 2',3'-dideoxyinosine. *Science*, 245, 412-415 (1989).