DOI 10.5012/bkcs.2010.31.11.3348

Synthesis and Anti-HIV Activity of Novel 4'-Ethyl-5'-norcarbocyclic Adenosine Phosphonic Acid Analogues

Jin Cheol Yoo, Hua Li, Wonjae Lee, and Joon Hee Hong*

BK-21 Project Team, College of Pharmacy, Chosun University, Kwangju 501-759, Korea
**E-mail: hongjh@chosun.ac.kr*
Received July 20, 2010, Accepted September 15, 2010

Novel 4'-ethyl-5'-norcarbocyclic adenosine phosphonic acid analogues were synthesized from propionaldehyde **5** through a *de novo* acyclic synthetic route using reiterative Grignard additions and ring-closing metathesis (RCM) as key reactions. The synthesized nucleoside phosphonic acids analogues **17**, **18**, **19**, and **21** were subjected to antiviral screening against human immunodeficiency virus.

Key Words: Anti-HIV agents, 4'-Ethyl branched nucleoside, Phosphonic acid nucleosides

Introduction

4'-Branched nucleosides were first investigated by Maag *et al.*[1] in 1992, and 4'-azido-thymidine (**1**) exerts potent activity against HIV-1. Extensive structure-activity studies found that other 4'-position lipophilic substituents, such as 4'-ethylthymidine (**2**),[2] also exhibited high antiviral activity against HIV. Molecular modeling studies demonstrated the presence of a narrow, relatively hydrophobic 4'-pocket that can accommodate these substitutions, contributing to enhanced potency.[3]

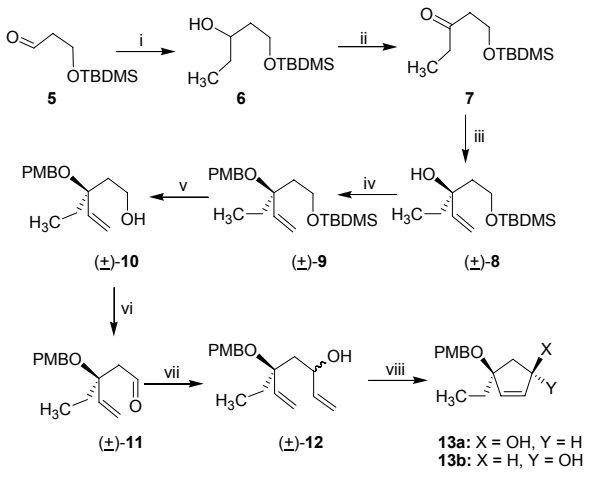
5'-Nornucleoside phosphonic acid analogues such as d4AP (**3**)[4] may be potential anti-HIV agents and have encouraged the search for novel nucleosides in this class of compounds.[5] The spatial location of the oxygen atom, namely the β-position from the phosphorus atom in the nucleoside analogue, plays a critical role for antiviral activity by increasing binding capacity of the phosphonate analogues to target enzymes.[6] The phosphonate has certain advantages over its phosphate counterpart as it is metabolically stable because its phosphorus-carbon bond is not susceptible to hydrolytic cleavage.[7] Moreover, a phosphonate nucleoside analogue can skip the requisite first phosphorylation, a rate-limiting crucial step for the activation of nucleosides that ultimately leads to triphosphates.[8]

Carbocyclic nucleosides are a group of compounds structurally similar to natural nucleosides in which the furanose oxygen is replaced by a methylene group. Replacement of the furanose ring oxygen by carbon is of particular interest because the resulting carbocyclic nucleosides possess greater metabolic stability to phosphorylase,[9] which cleaves the glycosidic bond of nucleosides. The recent discovery of 4'-ethynyl-cpAP (**4**)[3] as an anti-HIV agent gave strong impetus to the search for novel nucleosides in this class of compounds.

Stimulated by these findings that 4'-branched nucleoside analogues and 5'-nornucleoside phosphate had excellent biological activities, we sought to synthesize a novel class of nucleosides comprising 4'-ethyl branched carbocyclic-5'-norcarbocyclic phosphonic acid analogues to search for more efficient therapeutic agents against HIV and to provide analogues used in probing the conformational preferences of enzymes associated with the metabolism of nucleosides and nucleotides.

Results and Discussions

As depicted in Scheme 1, the target compounds were prepared from protected propionaldehyde **5**.[10] The aldehyde functional group of **5** was subjected to a carbonyl addition reaction by ethylmagnesium bromide to furnish the secondary alcohol **6**, which was subjected to oxidation using pyridium chlorochromate (PCC) to provide ketone derivative **7**. The corresponding ketone functional group of **7** was again subjected to an addition reaction by vinylmagnesium bromide to give the tertiary hydroxyl analogue (±)-**8**, which was successfully protected using *p*-methoxybenzyl chloride (PMBCl) to give the fully protected compound (±)-**9**. Removal of the silyl protecting group of (±)-**9** using *t*-butylammonium fluoride (TBAF) provided the primary alcohol (±)-**10**, which was oxidized to the aldehyde (±)-**11** using Swern oxidation conditions (DMSO, oxalyl chloride, TEA). The aldehyde (±)-**11** was subjected to nucleophilic Grignard conditions with vinylmagnesium bromide to give divinyl (±)-**12**, which was subjected to ring-closing metathesis (RCM) conditions using a 2nd generation Grubbs catalyst[11] to provide ethyl-



Reagents: i) ethylMgBr, THF; ii) PCC, CH_2Cl_2; iii) vinylMgBr, THF; iv) PMBCl, NaH, DMF; v) TBAF, THF; vi) $(COCl)_2$, DMSO, TEA; vii) vinylMgBr, THF; viii) Grubbs (II), CH_2Cl_2.

Scheme 1. Synthesis of key cyclopentenol intermediate **13b**

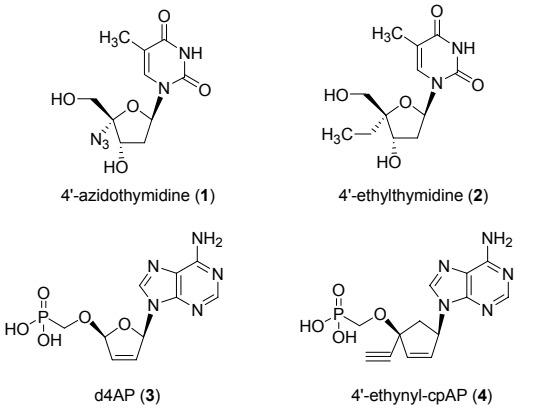


Figure 1. Some structures of nucleoside analogues as potent anti-HIV agents.

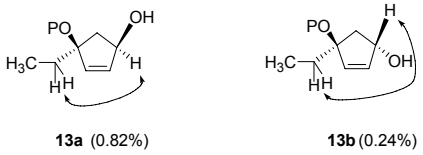


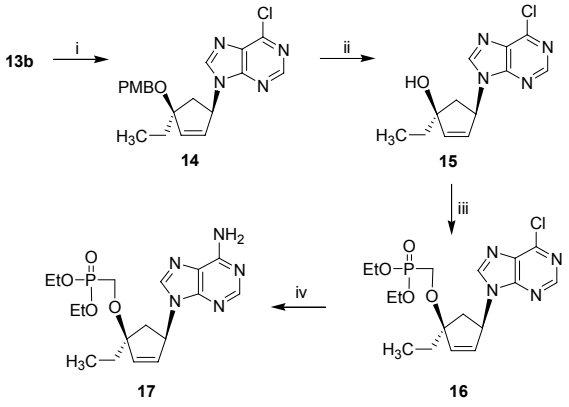
Figure 2. NOE differences between the proximal hydrogens of **13a** and **13b**.

ated cyclopentenol **13a** (39%) and **13b** (40%), which were readily separated by silica gel column chromatography. The NOE experiments with cyclpopentenols **13a** and **13b** confirmed these assignments. As expected, NOE enhancements were found between the *cis*-oriented hydrogens. Upon irradiation of C_1-H, weak NOE patterns were observed at the proximal hydrogens of compound **13b** [C_4-CH_2 (0.24%)] *versus* those of compound **13a** [C_4-CH_2 (0.82%)] (Figure 2).

To synthesize the desired 5'-norcarbocyclic adenine nucleoside analogue, the protected cyclopentenol **13b** was treated with 6-chloropurine under Mitsunobu conditions (DIAD and PPh_3). Slow addition of diisopropyl azodicarboxylate (DIAD) to a mixture of cyclopentenol **13b**, triphenylphosphine, and the 6-chloropurine in anhydrous tetrahydrofuran gave a yellow solution that was stirred for 3 h at −20 °C to give a protected 6-chloropurine analogue **14** (Scheme 2).[12] The PMB protection group was removed with 2,3-dichloro-5,6-dicyano-*p*-benzoquinone (DDQ)[13] to the 5'-nornucleoside analogue **15**, which was treated with diethylphosphonomethyl triflate[14] using lithium *t*-butoxide to yield the nucleoside phosphonate **16**. The chlorine group of **16** was then converted to amine with methanolic ammonia at 70 °C to give a corresponding adenine phosphonate derivative **17**.

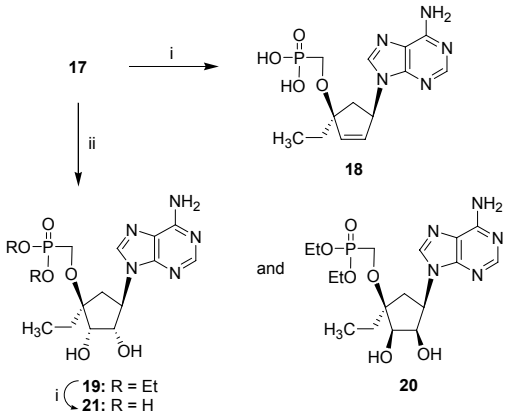
The nucleoside phosphonate mimics the overall shape and geometry of a nucleoside monophosphate. Hydrolysis of **17** by treatment with bromotrimethylsilane in CH_3CN in the presence of 2,6-lutidine gave an adenine phosphonic acid derivative **18**.[15]

Bishydroxylation[16] of the double bond in **17** was accomplished with a catalytic amount of osmium tetraoxide (OsO_4) and 4-methyl-morpholine *N*-oxide (NMO) as the oxidant to give the dihydroxylated isomer **19** (34%) and **20** (29%), with a relatively low stereoselectivity (Scheme 3).[17] Their stereochemistries were also readily determined by NOE experiments. These stereochemical outcomes suggest that the steric environments of α and



Reagents: i) 6-chloropurine, DIAD, THF; ii) DDQ, CH_2Cl_2/H_2O, rt;; iii) $(EtO)_2POCH_2OTf$, LiO-*t*-Bu, THF; iv) NH_3/MeOH, 70 °C.

Scheme 2. Synthesis of 5'-norcarbocyclic adenosine phosphonate



Reagents: i) TMSBr, 2,6-lutidine, CH_3CN; ii) OsO_4, NMO, acetone/*t*-BuOH/H_2O.

Scheme 3. Synthesis of desired 5'-norcarbocyclic adenosine phosphonic acid analogues

Table 1. Anti-HIV activity of synthesized compounds

Compound No.	anti-HIV-1 EC_{50} (μM)[b]	Cytotoxicity CC_{50} (μM)[c]
17	55	95
18	21	58
19	> 100	> 100
21	> 100	> 100
PMPA[a]	3.6	> 100

[a]**PMPA**: 9-(2-phosphonylmethoxypropyl)adenine. [b]EC_{50} (μM): Concentration (μM) required to inhibit the replication of HIV-1 by 50%. [c]CC_{50} (μM): Concentration (μM) required to reduce the viability of unaffected cells by 50%.

β-faces on the cyclopentene ring might be equivalent. Hydrolysis of the diethyl phosphonate functional groups of **19** by a similar procedure described for **18** gave an adenine phosphonic acid derivative **21**.

The synthesized nucleoside phosphonate and phosphonic acid analogues **17**, **18**, **19**, and **21** were evaluated for antiviral activity against HIV-1.[18]

As shown in Table 1, nucleoside phosphonic acid **18** exhibited greater toxicity-dependent anti-HIV activity than its parent

nucleoside phosphonate **17**. However, nucleotide analogs **19** and **21** did not show anti-HIV activity nor cytotoxicity up to 100 μM.

In summary, on the basis of potent anti-HIV activity of 4'-alkyl branched nucleoside and 5'-norcarbocyclic nucleoside analogues, we have designed and successfully synthesized novel 4'-ethyl-5'-norcarbocyclic nucleotide analogues starting from propionaldehyde. In this series, adenosine phosphonic acid derivative **18** showed moderate toxicity-dependent anti-HIV-1 activity.

Experimental Section

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). UV spectra were obtained on a Beckman DU-7 spectrophotometer (Beckman, South Pasadena, CA, USA). MS spectra were collected in electrospray ionization (ESI) mode. The elemental analyses were performed using a Perkin-Elmer 2400 analyzer (Perkin-Elmer, Norwalk, CT, USA). TLC was performed on Uniplates (silica gel) purchased from Analtech Co. (7558, Newark, DE, USA). All reactions were performed 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*-Butyldimethylsilanyloxy)-pentan-3-ol (6). To a solution of **5** (2.5 g, 13.27 mmol) in dry THF (25 mL) was slowly added ethylmagnesium bromide (15.9 mL, 1.0 M solution in THF) at −20 °C and stirred 5 h at 0 °C. Saturated NH_4Cl solution (16 mL) was added to the mixture, which was slowly warmed to rt. The mixture was further diluted with water (80 mL) and extracted with EtOAc (2 × 80 mL). The combined organic layer was washed with brine, dried over anhydrous $MgSO_4$, filtered, and evaporated under reduced pressure. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:6) to give **6** (2.58 g, 89%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 3.71 (t, J = 6.8 Hz, 2H), 3.32 (m, 2H), 1.45 (m, 2H), 0.97 (t, J = 6.9 Hz, 3H), 0.81 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 70.7, 58.8, 42.5, 32.1, 25.5, 18.5, 9.8, −5.4.

1-(*t*-Butyldimethylsilanyloxy)-pentan-3-one (7). To a solution of compound **6** (1.39 g, 6.4 mmol) in CH_2Cl_2 (50 mL), 4 Å molecular sieves (3.75 g) and PCC (3.45 g, 16.05 mmol) were added slowly at 0 °C, and stirred overnight at room temperature. To the mixture, excess diethyl ether (400 mL) was then added. The mixture was stirred vigorously for 3 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, 1:15) to give compound **7** (1.08 g, 78%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 4.09 (t, J = 7.0 Hz, 2H), 2.68 (t, J = 7.0 Hz, 2H), 2.52 (q, J = 7.2 Hz, 2H), 1.09 (t, J = 7.2 Hz, 3H), 0.82 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 206.7, 60.4, 34.3, 25.6, 18.3, 10.5, −5.5.

(±)-5-(*t*-Butyldimethylsilanyloxy)-3-ethyl-pent-1-en-3-ol (8). To a solution of **7** (2.5 g, 11.55 mmol) in dry THF (25 mL) was slowly added vinylmagnesium bromide (12.7 mL, 1.0 M solution in THF) at −20 °C and stirred 4 h at 0 °C. Saturated NH_4Cl solution (17 mL) was added to the mixture, which was slowly warmed to rt. The mixture was further diluted with water (80 mL) and extracted with EtOAc (2 × 80 mL). The combined organic layer was washed with brine, dried over anhydrous $MgSO_4$, filtered, and evaporated under reduced pressure. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:12) to give **8** (2.25 g, 80%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.89 (m, 1H), 5.21-5.12 (m, 2H), 3.82 (t, J = 6.2 Hz, 2H), 1.70 (t, J = 6.3 Hz, 2H), 1.45 (q, J = 6.9 Hz, 2H), 0.97 (t, J = 6.8 Hz, 3H), 0.81 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 144.1, 112.7, 71.5, 58.9, 46.2, 34.6, 25.5, 18.5, 11.8, −5.6; Anal. Calc. for $C_{13}H_{28}O_2Si$: C, 63.87; H, 11.55; Found: C, 63.92; H, 11.52.

(±)-*t*-Butyl-[3-ethyl-3-(4-methoxybenzyloxy)-pent-4-enyloxy]-dimethylsilane (9). NaH (60% in mineral oil, 207 mg, 83.21 mmol) was added portion-wise to a solution of alcohol **8** (3.5 g, 14.31 mmol) and *p*-methoxybenzyl chloride (2.46 g, 15.74 mmol) in DMF (20 mL) at 0 °C. The reaction mixture was stirred at room temperature overnight. The solvent was concentrated under reduced pressure and the residue was quenched with H_2O followed by extraction with diethyl ether. The organic layer was washed with brine, dried over anhydrous $MgSO_4$, and concentrated under reduced pressure. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:15) to give **9** (4.33 g, 83%) as a colorless oil. ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.27 (m, 2H), 6.88 (m, 2H), 5.95-5.86 (m, 1H), 5.15-5.03 (m, 2H), 4.49 (s, 2H), 3.81 (s, 3H), 3.73 (t, J = 6.8 Hz, 2H), 1.65 (t, J = 6.7 Hz, 2H), 1.46 (m, 2H), 0.95 (t, J = 6.9 Hz, 3H), 0.82 (s, 9H), 0.02 (s, 6H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 159.3, 143.7, 136.2, 131.5, 128.7, 117.6, 112.9, 72.4, 71.2, 69.2, 58.3, 55.6, 43.1, 32.6, 25.3, 18.4, 10.6, -5.6; Anal. Calc. for $C_{21}H_{36}O_3Si$: C, 69.18; H, 9.95; Found: C, 69.23; H, 9.89.

(±)-3-Ethyl-3-(4-methoxybenzyloxy)-pent-4-en-1-ol (10). To a solution of **9** (300 mg, 0.823 mmol) in THF (8.0 mL), TBAF (1.0 mL, 1.0 M solution in THF) was added at 0 °C. The mixture was stirred for 6 h at room temperature and concentrated in vacuo. The residue was purified by silica gel column chromatography (Hexane/EtOAc, 5:1) to give **10** (183 mg, 89%): ^{1}H NMR ($CDCl_3$, 300MHz) δ 7.29 (m, 2H), 6.89 (m, 2H), 5.94-5.85 (m, 1H), 5.20-5.09 (m, 2H), 4.50 (s, 2H), 3.83 (s, 3H), 3.75 (t, J = 6.7 Hz, 2H), 1.66 (t, J = 6.8 Hz, 2H), 1.45 (q, J = 6.8 Hz, 2H), 0.97 (t, J = 6.8 Hz, 3H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 159.7, 143.8, 138.5, 132.1, 129.0, 119.2, 115.1, 71.9, 71.0, 68.3, 58.8, 54.8, 44.7, 33.1; Anal. Calc. for $C_{15}H_{22}O_3$: C, 71.97; H, 8.86; Found: C, 71.94; H, 8.91.

3-Ethyl-3-(4-methoxybenzyloxy)-pent-4-enal (11). To a stirred solution of oxalyl chloride (212 mg, 1.67 mmol) in CH_2Cl_2 (10 mL) was added a solution of DMSO (195 mg, 2.5 mmol) in CH_2Cl_2 (5.0 mL) dropwise at −78 °C. The resulting solution was stirred at −78 °C for 10 min, and a solution of alcohol **10** (210 mg, 0.839 mmol) in CH_2Cl_2 (10 mL) was added dropwise. The mixture was stirred at −78 °C for 30 min and TEA (507 mg, 5.01 mmol) was added. The resulting mixture was warmed to 0 °C and stirred for 30 min. H_2O (15 mL) was added,

and the solution was stirred at room temperature for 30 min. The mixture was diluted with water (120 mL) and then extracted with EtOAc (2 × 120 mL). The combined organic layer was washed with brine, dried over anhydrous $MgSO_4$, and filtered. The filtrate was concentrated under reduced pressure and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:15) to give aldehyde compound **11** (189 mg, 91%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 9.82 (s, 1H), 7.30 (m, 2H), 6.91 (m, 2H), 5.93-5.85 (m, 1H), 5.20-5.11 (m, 2H), 4.49 (s, 2H), 3.78 (s, 3H), 2.55 (dd, *J* = 10.0. 8.2 Hz, 2H), 1.47 (q, *J* = 6.9 Hz, 2H), 0.98 (t, *J* = 6.8 Hz, 3H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 205, 160.0, 144.1, 138.3, 133.6, 128.9, 121.6, 114.1, 71.9, 69.1, 55.1, 32.6, 9.8.

(*rel*)-(3*R* and 3*S*,5*S*)-5-Ethyl-5-(4-methoxybenzyloxy)-hepta-1,6-dien-3-ol (12). Divinyl analogue **12** was prepared from aldehyde **11** using the similar procedure as described for **8** as a diastereomeric mixture: yield 87%; ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.29-7.27 (m, 2H), 6.89 (m, 2H), 5.96-5.83 (m, 2H), 5.16-4.99 (m, 4H), 4.59 (s, 2H), 3.89 (m, 1H), 3.78 (s, 3H), 1.65-1.58 (m, 2H), 1.48 (m, 2H), 0.97 (t, *J* = 6.9 Hz, 3H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 159.6, 143.7, 141.2, 137.5, 134.0, 127.5, 121.9, 113.1, 112.4, 71.5, 69.6, 68.2, 57.5, 47.0, 33.3, 9.6.

(*rel*)-(1*S*,4*S*)-4-Ethyl-4-(4-methoxybenzyloxy)-cyclopent-2-enol (13a) and (*rel*)-(1*R*,4*S*)-4-ethyl-4-(4-methoxybenzyloxy)-cyclopent-2-enol (13b). To a solution of **12** (205 mg, 0.74 mmol) in dry methylene chloride (6 mL) was added 2nd generation Grubbs catalyst (38.0 mg 0.0452 mmol). The reaction mixture was refluxed overnight and cooled to room temperature. The mixture was concentrated in vacuo, and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:12) to give cyclopentenol **13a** (71.6 mg, 39%) and **13b** (73.5 mg, 40%).

Data for 13a: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.31 (m, 2H), 6.88 (m, 2H), 5.65 (d, *J* = 5.4 Hz, 1H), 5.36 (m, 1H), 4.68 (s, 2H), 4.08 (m, 1H), 3.79 (s, 3H), 2.16 (dd, *J* = 13.4. 8.8 Hz, 1H), 2.02 (dd, *J* = 13.4, 6.8 Hz, 1H), 1.52 (m, 2H), 0.97 (t, *J* = 7.0 Hz, 3H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 159.5, 139.3, 136.6, 134.4, 130.5, 129.6, 118.4, 78.9, 71.8, 68.6, 57.0, 42.8, 32.6, 9.4; Anal. Calc. for $C_{15}H_{20}O_3$: C, 72.55; H, 8.12; Found: C, 72.58; H, 8.15.

Data for 13b: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.29 (m, 2H), 6.91 (m, 2H), 5.58 (d, *J* = 5.4 Hz, 1H), 5.36 (dd, *J* = 5.3, 4.2 Hz, 1H), 4.69 (s, 2H), 4.03 (dd, *J* = 6.0, 4.8 Hz, 1H), 3.81 (s, 3H), 2.19 (dd, *J* = 13.6. 8.2 Hz, 1H), 2.06 (dd, *J* = 13.5, 7.6 Hz, 1H), 1.49 (m, 2H), 0.98 (t, *J* = 6.9 Hz, 3H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 159.7, 138.8, 135.7, 130.4, 130.5, 129.6, 117.4, 79.1, 72.3, 69.3, 56.7, 43.2, 33.4, 9.6; Anal. Calc. for $C_{15}H_{20}O_3$: C, 72.55; H, 8.12; Found: C, 72.49; H, 8.08.

(*rel*)-(1'*R*,4'*S*)-9-[4-Ethyl-(4-methoxybenzyloxy)-cyclopent-2-enyl]-6-chloropurine (14). To a solution containing compound **13b** (112 mg, 0.45 mmol), triphenylphosphine (415 mg, 1.584 mmol), and 6-chloropurine (139 mg, 0.90 mmol) in anhydrous THF (7.0 mL), diisopropyl azodicarboxylate (DIAD) (182 mg, 0.90 mmol) was added dropwise at −20 °C for 30 min under nitrogen. The reaction mixture was stirred for 3 h at −20 °C under nitrogen and further stirred overnight at rt. The solvent was concentrated under reduced pressure and the residue was purified by silica gel column chromatography (EtOAc/hexane, 2:1) to give compound **14** (74 mg, 43%): mp 156 - 158 °C; UV (MeOH) λ_{max} 264.5 nm; ^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.72 (s, 1H), 8.44 (s, 1H), 7.30 (m, 2H), 6.90-6.87 (m, 2H), 5.64 (d, *J* = 5.3 Hz, 1H), 5.35 (dd, *J* = 5.2, 4.2 Hz, 1H), 4.64 (s, 2H), 4.43 (m, 1H), 3.80 (s, 3H), 2.21 (dd, *J* = 13.5. 8.4 Hz, 1H), 2.04 (dd, *J* = 13.6, 7.2 Hz, 1H), 1.53 (m, 2H), 0.96 (t, *J* = 6.9 Hz, 3H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 159.6, 152.6, 151.3, 147.9, 141.5, 137.5, 135.3, 133.4, 117.7, 81.3, 70.9, 57.1, 54.6, 38.7, 32.6, 9.7; Anal. Calc. for $C_{20}H_{21}ClN_4O_2$ (+ 0.5 EtOAc): C, 61.60; H, 5.87; N, 13.06; Found: C, 61.56; H, 5.90; N, 13.11.

(*rel*)-(1'*R*,4'*S*)-9-(4-Ethyl-4-hydroxycyclopent-2-enyl)-6-chloropurine (15). To a solution of compound **14** (435 mg, 1.13 mmol) in CH_2Cl_2/H_2O mixture (10 mL, 10:1 v/v) was added DDQ (280 mg, 1.24 mmol) and the mixture was stirred for 4 h at room temperature. Saturated $NaHCO_3$ (1.5 mL) was added to quench the reaction and further diluted with water (80 mL). The mixture was extracted with CH_2Cl_2 (3 × 80 mL) and the combined organic layer was dried over anhydrous $MgSO_4$, and filtered. The filtrate was concentrated under reduced pressure and the residue was purified by silica gel column chromatography (EtOAc/hexane/MeOH, 4:1:0.04) to give compound **15** (200 mg, 67%): mp 169 - 171 °C; UV (MeOH) λ_{max} 264.0 nm; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.75 (s, 1H), 8.46 (s, 1H), 5.59 (d, *J* = 5.2 Hz, 1H), 5.36 (m, 1H), 5.09 (s, 1H), 4.50 (m, 1H), 2.25 (dd, *J* = 13.6. 8.5 Hz, 1H), 2.06 (dd, *J* = 13.5, 7.4 Hz, 1H), 1.51 (m, 2H), 0.98 (t, *J* = 7.0 Hz, 3H); ^{13}C NMR (DMSO-d_6, 75 MHz) δ 152.3, 151.6, 150.5, 148.2, 138.5, 136.6, 132.4, 76.8, 52.9, 40.3, 34.3, 9.3; Anal. Calc. for $C_{12}H_{13}ClN_4O$: C, 54.45; H, 4.95; N, 21.17; Found: C, 54.51; H, 4.91; N, 21.21.

(*rel*)-(1'*R*,4'*S*)-Diethyl [9-(4-Hydroxy-4-ethylcyclopent-2-en-1-yl)-6-chloropurine] methylphosphonate (16). Both LiO*t*-Bu (2.084 mL of 0.5 M solution in THF, 1.042 mmol) and a solution of diethyl phosphonomethyltriflate (313 mg, 1.042 mmol) in 10.0 mL of THF were slowly added to a solution of the 6-chloropurine analogue **15** (138 mg, 0.521 mmol) in 5.0 mL of THF at 0 °C and stirred overnight at rt under anhydrous conditions. The mixture was quenched by adding saturated NH_4Cl solution (3 mL) and further diluted with additional H_2O (80 mL). The aqueous layer was extracted with EtOAc (3 × 80 mL). The combined organic layer was dried over anhydrous $MgSO_4$ and concentrated in vacuo. The residue was purified by silica gel column chromatography (MeOH/Hexane/EtOAc, 0.05:4:1) to give **16** (153 mg, 71%): mp 142 - 144 °C; UV (MeOH) λ_{max} 265.0 nm; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.74 (s, 1H), 8.48 (s, 1H), 5.62 (d, *J* = 5.4 Hz, 1H), 5.35 (m, 1H), 4.51 (m, 1H), 4.18 (m, 4H), 4.09 (d, *J* = 8.0 Hz, 2H), 2.28 (dd, *J* = 13.6. 8.7 Hz, 1H), 2.04 (dd, *J* = 13.6, 7.5 Hz, 1H), 1.52 (m, 2H), 1.36 (m 6H), 0.98 (m, 3H); ^{13}C NMR (DMSO-d_6, 75 MHz) δ 152.3, 151.9, 149.3, 146.6, 145.7, 136.5, 132.6, 81.8, 66.5, 65.2, 63.6, 54.1, 38.2, 32.5, 16.8, 9.6; Anal. Calc. for $C_{17}H_{24}ClN_4O_4P$ (+ 1.0 MeOH): C, 48.38; H, 6.31; N, 12.54; Found: C, 48.44; H, 6.27; N, 12.49.

(*rel*)-(1'*R*,4'*S*)-Diethyl [9-(4-Hydroxy-1,4-dimethylcyclopent-2-en-1-yl)-adenine] methylphosphonate (17). A solution of **16** (132 mg, 0.318 mmol) in saturated methanolic ammonia (8 mL) was stirred at 70 °C in a steel bomb, and the volatiles were evaporated. The residue was purified by silica gel column chromatography ($MeOH/CH_2Cl_2$, 1:10) to give **17** (74 mg, 59%): mp 171 - 173 °C; UV (MeOH) λ_{max} 260.5 nm; ^{1}H NMR

(DMSO-d_6, 300 MHz) δ 8.32 (s, 1H), 8.12 (s, 1H), 7.17 (br s, 2H), 5.67 (d, J = 5.2 Hz, 1H), 5.31 (dd, J = 5.3, 4.2 Hz, 1H), 4.52 (m, 1H), 4.17 (m, 4H), 4.05 (d, J = 8.2 Hz, 2H), 2.26 (dd, J = 13.5. 8.8 Hz, 1H), 2.05 (m, 1H), 1.51 (m, 2H), 1.34 (m 6H), 0.98 (t, J = 6.9 Hz, 3H); ^{13}C NMR (DMSO-d_6, 75 MHz) δ 155.0, 153.3, 144.2, 137.7, 134.2, 131.8, 119.2, 82.4, 67.1, 66.0, 64.2, 54.5, 39.1, 33.7, 17.2, 9.9; Anal. Calc. for $C_{17}H_{26}N_5O_4P$ (+ 0.5 MeOH): C, 51.09; H, 6.86; N, 17.02; Found: C, 51.15; H, 6.92; N, 16.98.

(*rel*)-(1'*R*,4'*S*)-[9-(4-Ethylcyclopenten-1-yl)-adenine]-4-methylphosphonic acid (18). To a solution of the phosphonate **17** (125 mg, 0.316 mmol) in anhydrous CH_3CN (10 mL) and 2,6-lutidine (0.8 mL) was added trimethylsilyl bromide (483 mg, 3.16 mmol). The mixture was heated overnight at 60 °C and then concentrated under reduced pressure. The residue was partitioned between CH_2Cl_2 (70 mL) and distilled clean water (60 mL). The aqueous layer was washed out with CH_2Cl_2 two times and then freeze-dried to give phosphonic acid **18** (73 mg, 68%) in a yellowish form: UV (H_2O) λ_{max} 260.5 nm; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.29 (s, 1H), 8.11 (s, 1H), 7.16 (br s, 2H), 5.64 (d, J = 5.3 Hz, 1H), 5.32 (dd, J = 5.4, 4.2 Hz, 1H), 4.50 (m, 1H), 4.11 (d, J = 8.0 Hz, 2H), 2.29 (m, 1H), 2.02 (dd, J = 13.6, 7.2 Hz, 1H), 1.52 (m, 2H), 0.96 (m, 3H); ^{13}C NMR (DMSO-d_6, 75 MHz) δ 154.7, 152.9, 142.7, 136.8, 135.5, 132.7, 120.1, 81.9, 65.2, 56.3, 38.7, 34.1, 9.6; Anal. Calc. for $C_{13}H_{18}N_5O_4P$ (+ 2.0 H_2O): C, 41.60; H, 5.91; N, 18.66; Found: C, 41.56; H, 5.92; N, 18.59.

(*rel*)-(1'*R*,2'*S*,3'*S*,4'*S*)-Diethyl [9-(2,3-dihydroxy-4-ethyl-cyclopent-1-yl)-adenine]-4-methylphosphonate (19) and (*rel*)-(1'*R*,2'*R*,3'*R*,4'*S*)-diethyl [9-(2,3-dihydroxy-4-ethyl-cyclopent-1-yl)-adenine]-4-methylphosphonate (20). Compound **17** (158 mg, 0.4 mmol) was dissolved in a cosolvent system (10 mL) (acetone:*t*-BuOH:H_2O = 6:1:1) along with 4-methylmorpholine *N*-oxide (82 mg, 0.8 mmol). Subsequently, OsO_4 (0.19 mL, 0.03 mmol, 4% wt % in H_2O) was added. The mixture was stirred overnight at rt and quenched with saturated Na_2SO_3 solution (3 mL). The resulting solid was removed by filtration through a pad of Celite, and filtrate was concentrated under reduced pressure. The residue was purified by silica gel column chromatography (MeOH/CH_2Cl_2, 1:6) to give **19** (58 mg, 34%) and **20** (49 mg, 29%): Spectroscopical data for **19:** ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.30 (s, 1H), 8.13 (s, 1H), 7.15 (br s, 2H), 4.25 (m, 4H), 4.14 (d, J = 8.2 Hz, 2H), 3.75-3.68 (m, 2H), 3.32 (m, 1H), 2.13 (dd, J = 13.6, 8.7 Hz, 1H), 1.94 (dd, J = 13.6, 7.2 Hz, 1H), 1.48 (m, 2H), 1.31 (m 6H), 0.98 (t, J = 6.9 Hz, 3H); ^{13}C NMR (DMSO-d_6, 75 MHz) δ 154.6, 153.2, 147.7, 138.4, 119.3, 78.4, 77.6, 69.1, 66.2, 65.1, 63.7, 48.6, 28.8, 26.4, 17.1, 9.9; Anal. Calc. for $C_{17}H_{28}N_5O_6P$ (+ 1.0 MeOH): C, 46.85; H, 6.99; N, 15.17; Found: C, 46.91; H, 7.05; N, 15.13.

Spectroscopical data for 20: ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.34 (s, 1H), 8.15 (s, 1H), 7.17 (br s, 2H), 4.27(m, 4H), 4.12 (d, J = 8.1 Hz, 2H), 3.76 (dd, J = 6.4, 5.2 Hz, 1H), 3.66 (d, J = 6.0 Hz, 1H), 3.25 (m, 1H), 2.15 (dd, J = 13.7, 8.8 Hz, 1H), 1.96 (dd, J = 13.6, 7.4 Hz, 1H), 1.50 (m, 2H), 1.32 (m 6H), 0.97 (m, 3H); ^{13}C NMR (DMSO-d_6, 75 MHz) δ 154.5, 151.7, 146.8, 134.6, 120.7, 79.3, 76.2, 68.5, 65.8, 64.2, 63.0, 47.3, 30.6, 27.5, 16.8, 9.5; Anal. Calc. for $C_{17}H_{28}N_5O_6P$ (+ 1.0 MeOH): C, 46.85; H, 6.99; N, 15.17; Found: C, 46.79; H, 6.95; N, 15.20.

(*rel*)-(1'*R*,2'*S*,3'*S*,4'*S*)-[9-(2,3-Dihydroxy-4-ethylcyclopent-1-yl)] adenine]-4-methylphosphonic acid (21). The final adenosine phosphonic acid **21** was synthesized from **19** using a similar procedure described for **18** as a formy solid: yield 60%; UV (H_2O) λ_{max} 261.0 nm; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.29 (s, 1H), 8.14 (s, 1H), 3.98 (d, J = 8.0 Hz, 2H), 3.78 (m, 1H), 3.68 (d, J = 6.1 Hz, 1H), 3.22 (m, 1H), 2.13 (dd, J = 13.8, 8.7 Hz, 1H), 1.95 (dd, J = 13.8, 7.6 Hz, 1H), 1.51 (m, 2H), 0.95 (m, 3H); ^{13}C NMR (DMSO-d_6, 75 MHz) δ 154.9, 152.5, 147.4, 132.8, 119.6, 78.6, 77.3, 67.8, 64.9, 63.1, 48.1, 31.4, 28.8, 10.0; Anal. Calc. for $C_{13}H_{20}N_5O_6P$ (+ 3.0 H_2O): C, 36.54; H, 6.13; N, 16.39; Found: C, 36.49; H, 6.08; N, 16.44.

Acknowledgments. This work was supported by Research Grant of Chosun University (2008).

References

1. Maag, H.; Rydzewski, R. M.; McRoberts, M. J.; Crawford-Ruth, D.; Verheyden, J. P.; Prisbe, E. J. *J. Med. Chem.* **1992**, *35*, 1440.
2. Sugimoto, I.; Shuto, S.; Mori, S.; Shigeta, S.; Matsuda, A. *Bioorg. Med. Chem. Lett*. **1999**, *9*, 385.
3. Boojamra, C. G.; Parrish, J. P.; Sperandio, D.; Gao, Y.; Petrakovsky, O. V.; Lee, S. K.; Markevich, D. Y.; Vela, J. E.; Laflamme, G.; Chen, J. M.; Ray, A. S.; Barron, A. C.; Sparacino, M. L.; Desai, M. C.; Kim, C. U.; Cihlar, T.; Mackman, R. L. *Bioorg. Med. Chem*. **2009**, *17*, 1739.
4. (a) Mackman, R. L.; Boojamra, C. G.; Prasad, V.; Zhang, L.; Lin, K. Y.; Petrakovsky, O.; Babusis, D.; Chen, J.; Douglas, J.; Grant, D.; Hui, H. C.; Kim, C. U.; Markevitch, D. Y.; Vela, J.; Ray, A.; Cihlar, T. *Bioorg. Med. Chem. Lett*. **2007**, *17*, 6785. (b) Ray, A. S.; Vela, J. E.; Boojamra, C. G.; Zhang, L.; Hui, H.; Callebaut, C.; Stray, K.; Lin, K. Y.; Gao, Y.; Mackman, R. L.; Cihlar, T. *Antimicrob. Agents Chemother*. **2008**, *52*, 648.
5. (a) Ray, A. S.; Vela, J. E.; Boojamra, C. G.; Zhang, L.; Hui, H.; Callebaut, C.; Stray, K.; Lin, K. Y.; Gao, Y.; Mackman, R. L.; Cihlar, T. *Antimicrob. Agents Chemother*. **2008**, *52*, 648. (b) Boojamra, C. G.; Mackman, R. L.; Markevitch, D. Y.; Prasad, V.; Ray, A. S.; Douglas, J.; Grant, D.; Kim, C. U.; Cihlar, T. *Bioorg. Med. Chem. Lett*. **2008**, *18*, 1120. (c) Mackman, R. L.; Lin, K. Y.; Boojamra, C. G.; Hui, H.; Douglas, J.; Grant, D.; Petrakovsky, O.; Prasad, V.; Ray, A. S.; Cihlar, T. *Bioorg. Med. Chem. Lett*. **2008**, *18*, 1116.
6. Kim, C. U.; Luh, B. Y.; Misco, P. F.; Bronson, J. J.; Hitchcock, M. J.; Ghazzouli, I.; Martin, J. C. *J. Med. Chem*. **1990**, *33*, 1207.
7. Wu, T.; Froeyen, M.; Kempeneers, V.; Pannecouque, C.; Wang, J.; Busson, R.; De Clercq, E.; Herdewijn, P. *J. Am. Chem. Soc*. **2005**, *127*, 5056.
8. Kim, C. U.; Luh, B. Y.; Martin, J. C. *J. Org. Chem*. **1991**, *56*, 2642.
9. Stoeckler, J. D.; Cambor, C.; Parks, R. E., Jr. *Biochemistry* **1980**, *19*, 102.
10. Heapy, A. M.; Bramble, M. A. *Tetrahedron* **2010**, *66*, 5424.
11. (a) Jeong, L. S.; Lee, J. A. *Antiviral Chem. Chemother.* **2004**, *15*, 235. (b) Amblard, F.; Nolan, S. P.; Agrofoglio, L. A. *Tetrahedron* **2005**, *61*, 7067.
12. (a) Liu, L. J.; Kim, S. W.; Lee, W.; Hong, J. H. *Bull. Korean Chem. Soc*. **2009**, *30*, 2989; (b) Li, H.; Kim, S. W.; Hong, J. H. *Bull. Korean Chem. Soc*. **2010**, *31*, 2180.
13. Oh, H. S.; Kang, H. Y. *Tetrahedron* **2010**, *66*, 4307.
14. (a) Phillion, D. P.; Andrew, S. S. *Tetrahedron Lett*. **1986**, *27*, 1477. (b) Xu, Y.; Flavin, M. T.; Xu, Z.-Q. *J. Org. Chem*. **1996**, *61*, 7697.
15. Hocková, D.; Holý, A.; Masojídková, M.; Keough, D. T.; De Jersey, J.; Guddat, L. W. *Bioorg. Med. Chem*. **2009**, *17*, 6218.
16. Koh, Y. H.; Shim, J. H.; Wu, J. Z.; Zhong, W.; Hong, Z.; Girardet, J. L. *J. Med. Chem*. **2005**, *48*, 2867.
17. Trost, B. M.; Kuo, G. H.; Benneche, T. *J. Am. Chem. Soc*. **1988**, *110*, 621.
18. Lian, L. J.; Yoo, J. C.; Hong, J. H. *Nucleos. Nucleot. Nucl*. **2009**, *28*, 150.