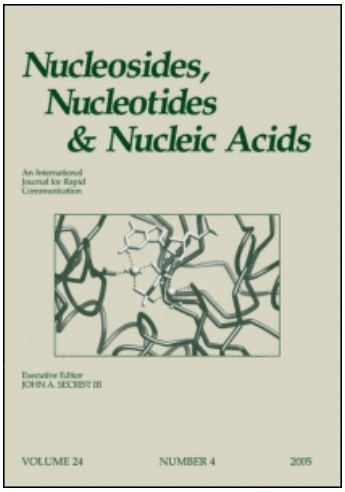

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DESIGN AND SYNTHESIS OF DUALLY BRANCHED 5′-NORCARBOCYCLIC ADENOSINE PHOSPHONODIESTER ANALOGUE AS A NEW ANTI-HIV PRODRUG

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□ *A novel 3′,4′-dimethyl-5′-norcarbocyclic adenosine phosphonic acid was prepared using acyclic stereoselective route from 4-hydroxybutan-2-one (**4**). To improve the cellular permeability and enhance the anti-HIV activity of this phosphonic acid, a (bis)SATE phosphonodiester nucleoside prodrug (**20**) was prepared and its chemical stability was evaluated. The newly synthesized bis(SATE) analogue (**20**) and its parent nucleoside phosphonic acid (**18**) were assayed for anti-HIV activity using an in vitro assay system in a CEM cell line.*

Keywords Anti-HIV agent; bis(SATE) derivative; nucleoside adenine phosphonic acid

INTRODUCTION

Emerging drug-resistant viral strains and drug toxicity are major problems in antiviral chemotherapy,[1] which has lead to research for structurally modified nucleosides. Although the pharmacophore of nucleoside antiviral activity is not completely defined, 5′-nornucleoside phosphonic acid analogues such as d4AP (**1**),[2] 2′-Fd4AP (**2**),[3] and 4′-ethynyl-cpAP (**3**)[4] as potential anti-HIV agents have encouraged the search for novel nucleosides in this class of compounds (Figure 1).

A nucleotide is essentially a nucleoside monophosphate analogue. However, the phosphonate has certain advantages over its phosphate counterpart as it is metabolically stable due to the fact that its phosphorus-carbon bond is not susceptible to hydrolytic cleavage.[5] Moreover, a nucleotide can skip the requisite first phosphorylation, which is a crucial step for the activation of nucleosides. Although triphosphates of the nucleoside analogues exhibit excellent antiviral potency, only a few nucleoside derivatives exhibit biological

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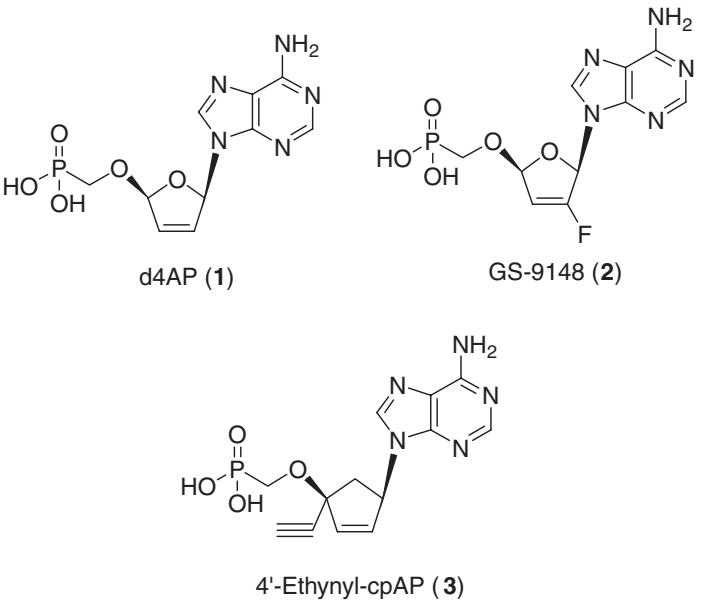
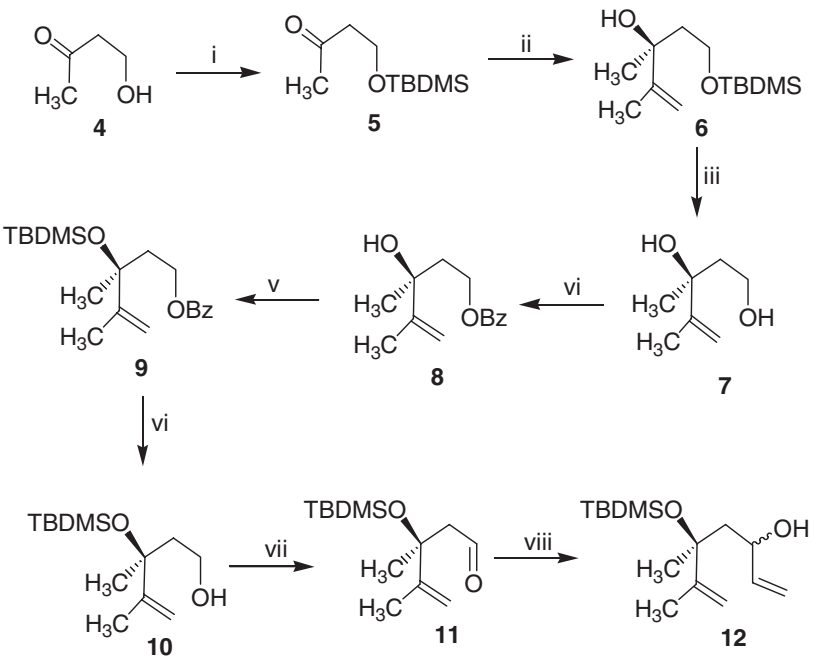


FIGURE 1 Structures of 5′-nornucleoside analogues as potent anti-HIV agents.

activity in cell culture assays. This might be due to poor cellular penetration coupled with insufficient metabolism of these nucleoside derivatives to 5′-triphosphates. The poor oral bioavailability of these nucleoside analogues is due to the negative charges of the phosphonate present in nucleoside phosphonic acid at physiological pH. Therefore, temporary masking of these charges with neutral groups to form more lipophilic derivatives capable of crossing the gastrointestinal wall and reverting back to the parent nucleoside phosphonic acid was attempted.[6]

Stimulated by the finding that 5′-nornucleosides and their phosphonic acid derivatives exhibit excellent antiviral activities, we sought to synthesize a novel class of nucleosides comprising 3′,4′-dimethyl carbocyclic 5′-norcarbocyclic phosphonic acid and its bis(SATE) phosphonodiester derivative. Here we report on the synthesis, antiviral activity, and chemical stability of the bis(SATE) prodrug.

The target compounds were prepared from commercially available starting material (**4**) as shown in Scheme 1. The alcohol functional group of (**4**) was temporarily protected with *t*-butyldimethylchlorosilane (TBDMSCl) to give the ketone derivative (**5**), which was subject to carbonyl addition using isopropenylmagnesium bromide [$CH_2{=}C(CH_3)MgBr$] to yield the tertiary alcohol derivative (**6**). In order to differentiate between the two hydroxyl groups, the protective silicon group on the primary hydroxyl was replaced with a benzoyl group by sequential desilylation and selective benzoylation to provide (**8**). The tertiary hydroxyl group of (**8**) was successfully silylated



SCHEME 1 Synthesis of divinyl intermediate **12**. Reagents: i) TBDMSCl, imidazole, CH_2CI_2; ii) isopropenylMgBr, THF; iii) TBAF, THF; iv) BzCI, DMAP, pyridine; v) TBDMSOTf, DMAP, TEA, CH_2Cl_2; vi) NH_3/MeOH; vii) $(COCI)_2$, DMSO, TEA, CH_2CI_2; viii) vinylMgBr, THF.

using trimethylsilyl trifluoromethanesulfonate (TMSOTf) to give the fully protected compound (**9**). The benzoyl protection group of the primary hydroxyl was removed under methanolic ammonia conditions to provide the alcohol derivative (**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 by vinylmagnesium bromide to yield divinyl (**12**).

Without separation of the diastereomeric mixture, divinyl (**12**) was subjected to ring-closing metathesis (RCM) conditions using second-generation Grubbs catalyst[78] to provide cyclopentenol (**13a**) (36%) and (**13b**) (35%), which were readily separated by silica gel column chromatography. The relative stereochemical assignments of the two isomers were made readily based on NOE comparisons. Upon irradiation of C_1-H, weak NOE patterns were observed at the proximal hydrogens of compound (**13b**) [C_4-CH_3 (0.09%)] versus those of compound (**13a**) [C_4-CH_3 (0.21%)] (Figure 2).

To synthesize the desired carbocyclic nucleoside analogues, the protected cyclopentenol (**13b**) was treated with 6-chloropurine in the presence of diisopropyl azodicarboxylate (DIAD) and PPh_3 to give (**14**) with the correct configuration through a chirality inversion.[9] The oxygen at C_4 of (**14**) was phosphonated after the silyl protection was removed. Treatment of the

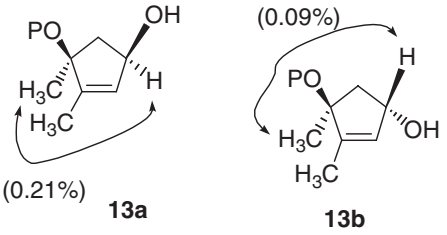
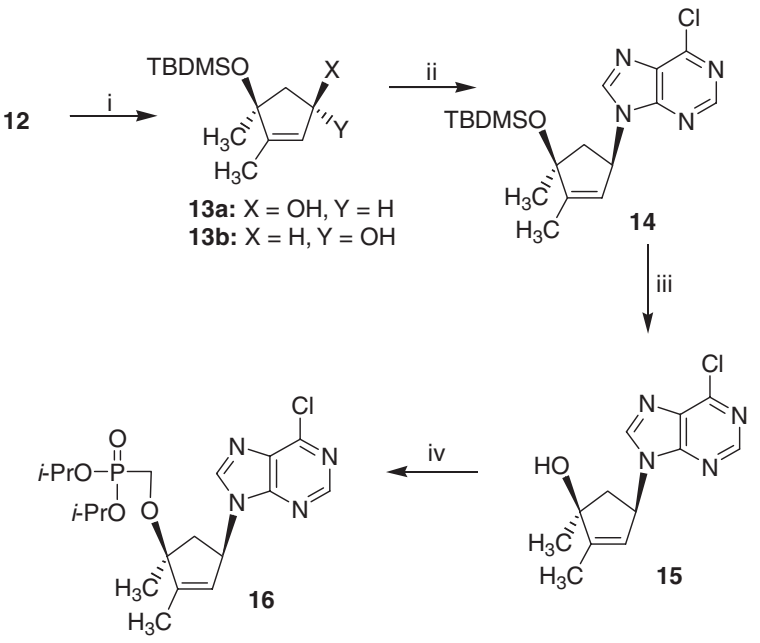


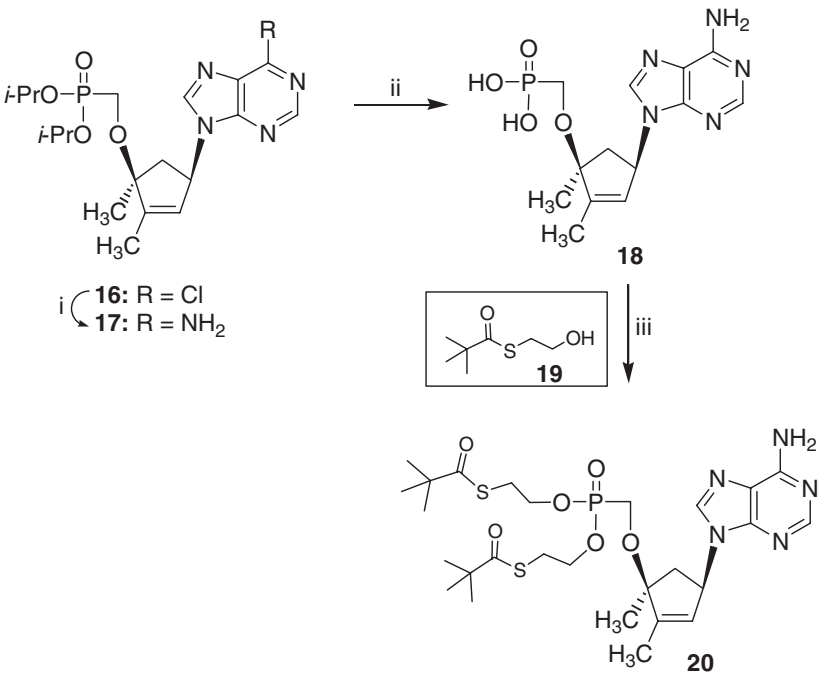
FIGURE 2 NOE difference between the hydrogens of **13a** and **13b**.

nucleoside (**15**) with diisopropylphosphonomethyl bromide[10] yielded the desired compound (**16**) (Scheme 2). The chlorine group of (**16**) was then transformed to amine with methanolic ammonia in a steel bomb at 80°C to give the adenine phosphonate derivative (**17**). The nucleoside phosphonate mimics the overall shape and geometry of a nucleoside monophosphate. Hydrolysis of (**17**) by treatment with bromotrimethylsilane afforded the adenine phosphonic acid (**18**).[11] To synthesize the thioester-protected analogue, compound (**18**) was reacted with thioester (**19**)[12] in the presence of 1-(2-mesitylenesulfonyl)-3-nitro-1*H*-1,2,4-triazole (MSNT)[13] to provide the bis(SATE) derivative as a target compound (**20**) (Scheme 3).

The newly synthesized bis(SATE) analogue (**20**) and its parent nucleoside phosphonic acid (**18**) were assayed for anti-HIV activity using an in vitro



SCHEME 2 Synthesis of carbocyclic adenine phosphonate **16**. Reagents; i) Grubbs (II), benzene; ii) 6-chloropurine, DIAD, THF; iii) TBAF, THF; iv) $(i\text{-PrO})_2POCH_2Br$, LiO-*t*-Bu, THF.



SCHEME 3 Synthesis of target bis(SATE) prodrug of adenine analogue **20**. Reagents: i) NH_3/MeOH, steel bomb, 50°C; ii) TMSBr, CH_3CN; iii) thioester, **19**, 1-(2-mesitylenesulfonyl)-3-nitro-1*H*-1,2,4-triazole, pyridine.

assay system in a CEM cell line that is suitable for monitoring the anti-HIV activities of compounds (Table 1).[14]

As shown in Table 1, nucleoside prodrug (**20**) exhibited an increased toxicity-dependent anti-HIV activity over its parent nucleoside phosphonic acid (**18**).

In conclusion, the anti-HIV activity of the bis(t-Bu-SATE) prodrug was 4-fold higher than that of the parent nucleoside phosphonic acid. The synthesis of other nucleoside analogues (U,T,C), together with chemical and enzymatic stability data will be reported elsewhere.

TABLE 1 Anti-HIV activity of synthesized compounds

Compound No.	Anti-HIV EC_{50} (μM)	Cytotoxicity CC_{50} (μM)
18	82.3	>100
20	21.6	81.5
ABC	0.17	>100

ABC: abacavir
EC_{50}: concentration (μM) required to inhibit the replication of HIV-1 by 50%.
CC_{50}: concentration (μM) required to reduce the viability of unaffected cells by 50%.

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). Mass spectrometry (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). 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.

4-(*t*-Butyldimethylsilanyloxy) butan-2-one (5)

To a solution of 4-hydroxy-butan-2-one **4** (10.0 g, 113.5 mmol) and imidazole (11.59 g, 170.24 mmol) in CH_2Cl_2 (250 mL), TBDMSCl (18.82 g, 124.85 mmol) was added slowly at 0°C and stirred overnight at room temerpature. The reaction solvent was evaporated under reduced pressure. The residue was poured into water (200 mL) and extracted with ethyl acetate (200 mL) two times. The combined organic layer was dried over anhydrous $MgSO_4$, filtered, and concentrated under reduced pressure. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:25) to give compound **5** (21.36 g, 93%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 3.91 (t, $J = 7.0$ Hz, 2H), 2.72 (t, $J = 7.0$ Hz, 2H), 2.17 (s, 3H), 0.81 (s, 9H), 0.01 (s, 6H);^{13}C NMR ($CDCl_3$) δ 208.6, 59.3, 46.2, 25.5, 18.7, −5.5.

(±)-5-(t-Butyldimethylsilanyloxy)-2,3-dimethyl-pent-1-en-3-ol (6)

To a solution of **5** (1.11 g, 5.5 mmol) in dry THF (15 mL) was slowly added isopropenylMgBr (11.10 mL, 0.5 M solution in THF) at −20°C and stirred for 4 hours at the same temperature. Saturated NH_4Cl solution (10 mL) was added to the mixture, which was slowly warmed to room temperature. The mixture was further diluted with water (50 mL) and extracted with EtOAc (50 mL) two times. The combined organic layer was washed with brine, dried over $MgSO_4$, filtered, and evaporated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 20) to give **6** (968 mg, 72%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.38 (d, $J = 2.0$ Hz, 1H), 5.12 (t, $J = 1.9$ Hz, 1H), 3.78 (dd, $J = 10.4$, 1.2 Hz, 2H), 1.74 (s, 3H), 1.62–1.55

(m, 2H), 1.35 (s, 3H), 0.82 (s, 9H), 0.01 (s, 6H);^{13}C NMR ($CDCl_3$) δ 152.2, 109.4, 76.6, 57.4, 48.6, 25.4, 24.3, 18.5, 13.8, −5.2.

(±)-3,4-Dimethyl-pent-4-ene-1,3-diol (7)

To a solution of **6** (1.2 g, 4.9 mmol) in THF (10 mL) at 0°C, TBAF (7.3 mL, 1.0 M solution in THF) was added. The mixture was stirred overnight at room temperature and concentrated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 3:1) to give **7** (523 mg, 82%): ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.35 (d, J = 2.1 Hz, 1H), 5.15 (d, J = 2.0 Hz, 1H), 3.67 (dd, J = 10.2, 1.4 Hz, 2H), 1.73 (s, 3H), 1.61–1.54 (m, 2H), 1.36 (s, 3H);^{13}C NMR ($CDCl_3$) δ 152.7, 105.2, 77.5, 56.7, 45.2, 24.9, 14.1.

(±)-Benzoic Acid 3-hydroxy-3,4-dimethyl-pent-4-enyl Ester (8)

To a solution of **7** (1.4 g, 10.76 mmol) in anhydrous pyridine (15 mL) was added benzoyl chloride (1.66 g, 11.83 mmol) and dimethylamino pyridine (DMAP) (262 mg, 2.15 mmol) at 0∞C. The reaction mixture was stirred for 2 hours at 0∞C and further stirred overnight at room tmperature. The reaction mixture was quenched with saturated $NaHCO_3$ solution (6 mL), stirred for 20 minutes and concentrated under reduced pressure. The residue was diluted with water (60 mL) and extracted with EtOAc (60 mL) twice. The combined organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:10) to give **8** (1.76 g, 70%) as a colorless syrup: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.04 (m, 2H), 7.58 (m, 1H), 7.47 (m, 2H), 5.34 (t, J = 2.0 Hz, 1H), 5.18 (d, J = 2.0 Hz, 1H), 4.21 (dd, J = 9.4, 1.8 Hz, 2H), 1.89–1.79 (m, 2H), 1.73 (s, 3H), 1.37 (s, 3H);^{13}C NMR ($CDCl_3$) δ 167.3, 151.4, 132.7, 130.4, 128.4, 107.2, 78.3, 59.2, 43.7, 25.2, 14.2; Anal. Calc. for $C_{14}H_{18}O_3$: C, 71.77; H, 7.74; Found: C, 71.75; H, 7.71.

(±)-Benzoic Acid 3-(tert-butyl-dimethyl-silanyloxy)-3,4-dimethyl-pent-4-enyl Ester (9)

To a solution of **8** (984 mg, 4.2 mmol) in anhydrous CH_2Cl_2 (15 mL) was added TEA (1.01 g, 9.99 mmol) and DMAP (131 mg, 1.07 mmol) at 0°C. *t*-Butyldimethylsilyl trifluomethane-sulfonate (TBDMSOTf) (1.45 g, 5.49 mmol) was added to this mixture; the reaction mixture was stirred overnight at room temperature and quenched with cold H_2O (5 mL). The mixture was concentrated under reduced pressure, diluted with water (60 mL), and extracted with EtOAc (60 mL) two times. Combined organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:20) to give **9** (1.21 g, 83%) as a colorless syrup: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.01 (m, 2H), 7.46 (m, 1H), 7.39 (m, 2H), 5.38 (d, J = 2.1 Hz, 1H), 5.12 (t, J = 2.0

Hz, 1H), 4.32–4.24 (m, 2H), 1.93–1.86 (m, 2H), 1.70 (s, 3H), 1.39 (s, 3H), 0.82 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 166.5, 152.0, 133.5, 130.8, 128.9, 128.1, 105.8, 78.7, 58.5, 42.5, 25.8, 24.6, 18.4, 14.0, −5.3; Anal. Calc. for $C_{20}H_{32}O_3Si$: C, 68.92; H, 9.25; Found: C, 68.89; H, 9.21.

(±)-3-(tert-Butyl-dimethyl-silanyloxy)-3,4-dimethyl-pent-4-en-1-ol (10)

A solution of **9** (2.2 g, 6.31 mmol) was dissolved in saturated methanolic ammonia (20 mL) at 0°C and stirred overnight at room temperature. The mixture was concentrated under reduced pressure and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:4) to give **10** (1.4 g, 91%) as a colorless syrup: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.37 (t, J = 2.0 Hz, 1H), 5.14 (d, J = 2.0 Hz, 1H), 3.61–3.53 (m, 2H), 1.72 (s, 3H), 1.63–1.55 (m, 2H), 1.49 (s, 3H), 0.81 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 151.7, 109.3, 71.9, 55.0, 46.6, 26.2, 25.4, 18.5, 13.8, −5.4; Anal. Calc. for $C_{13}H_{28}O_2Si$: C, 63.87; H, 11.55; Found: C, 63.90; H, 11.52.

(±)-3-(tert-Butyl-dimethyl-silanyloxy)-3,4-dimethyl-pent-4-enal (11)

To a stirred solution of oxalyl chloride (0.0864 mL, 0.979 mmol) in CH_2Cl_2 (8 mL) was added a solution of DMSO (0.0864 mL, 1.296 mmol) in CH_2Cl_2 (1.2 mL) dropwise at −78°C. The resulting solution was stirred at −78°C for 10 minutes, and a solution of alcohol **10** (133 mg, 0.545 mmol) in CH_2Cl_2 (5 mL) was added dropwise. The mixture was stirred at −78°C for 20 minutes and TEA (0.384 mL, 2.71 mmol) was added. The resulting mixture was warmed to 0°C and stirred for 30 minutes. H_2O (7 mL) was added, and the solution was stirred at room temperature for 20 minutes. The mixture was diluted with water (60 mL) and then extracted with EtOAc (60 mL) two times. The combined organic layer was washed with brine, dried over $MgSO_4$ and filtered. The filtrate was concentrated under reduced pressure and the residue was purified by silica gel column chromatography (EtOAc/hexane, 1:25) to give aldehyde compound **11** (123 mg, 93%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 9.91 (s, 1H), 5.36 (d, J = 2.1 Hz, 1H), 5.15 (d, J = 2.0 Hz, 1H), 2.51 (dd, J = 10.2, 6.2 Hz, 2H), 1.74 (s, 3H), 1.51 (s, 3H), 0.81 (s, 9H), 0.02 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 200.9, 151.8, 110.3, 70.3, 56.2, 26.0, 25.7, 18.6, 13.9, −5.6.

(*rel*)-(3*R* and *3S*,5*S*)-5-(tert-Butyl-dimethyl-silanyloxy)-5,6-dimethyl-hepta-1,6-dien-3-ol (12)

To a solution of **11** (4.2 g, 17.32 mmol) in dry THF (20 mL) was slowly added vinylMgBr (20.78 mL, 1.0 M solution in THF) at −20°C. After 5 hours, saturated NH_4Cl solution (20 mL) was added to the mixture, and

the reaction mixture was slowly warmed to room temperature. The mixture was further diluted with water (100 mL) and extracted with EtOAc (100 mL) two times. The combined organic layer was washed with brine, dried over $MgSO_4$, filtered, and evaporated. The residue was purified by silica gel column chromatography (EtOAc/hexane, 1:15) to give **12** (3.84 g, 82%) as a colorless oil: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.75 (dd, J = 16.2, 10.2 Hz, 1H), 5.21 (m, 2H), 5.04 (d, J = 10.3 Hz, 1H), 3.85–3.78 (m, 1H), 1.65–1.56 (m, 2H), 1.73 (s,s, 3H), 1.50 (s, 3H), 0.83 (s, 9H), 0.01 (m, 6H); ^{13}C NMR ($CDCl_3$) δ 151.3, 142.2, 115.2, 105.2, 71.7, 66.5, 50.6, 26.6, 25.4, 18.4, 13.9, −5.3.

(*rel*)-(1*S* and 4*S*)-4-(tert-Butyl-dimethyl-silanyloxy)-3,4-dimethyl-cyclopent-2-enol (13a) and (*rel*)-(1*R* and 4*S*)-4-(tert-Butyl-dimethyl-silanyloxy)-3,4-dimethyl-cyclopent-2-enol (13b)

To a solution of **12** (1.39 g, 5.16 mmol) in dry benzene (8 mL) was added second-generation Grubbs catalyst (76.2 mg 0.09 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, 1:10) to give cyclopentenol **13a** (450 mg, 36%) and **13b** (437 mg, 35%) as colorless oils, respectively. Cyclopentenol **13a**: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.41 (s, 1H), 4.09 (dd, J = 6.4, 2.0 Hz, 1H), 2.16 (dd, J = 12.2, 8.6 Hz, 1H), 1.98 (dd, J = 12.2, 6.8 Hz, 1H), 1.70 (s, 3H), 1.53 (s, 3H), 0.81 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 143.2, 122.9, 77.5, 68.4, 44.8, 26.8, 25.5, 18.5, 14.3, -5.6; Anal. Calc. for $C_{13}H_{26}O_2Si$: C, 64.41; H, 10.81; Found: C, 64.43; H, 10.84; Cyclopentenol **13b**: ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.39 (s, 1H), 4.12 (t, J = 6.8 Hz, 1H), 2.15 (dd, J = 10.8, 8.8 Hz, 1H), 2.01 (dd, J = 10.7, 7.2 Hz, 1H), 1.75 (s, 3H), 1.52 (s, 3H), 0.82 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 142.9, 123.1, 78.2, 69.0, 45.6, 25.9, 25.2, 18.2, 14.8, −5.7; Anal. Calc. for $C_{13}H_{26}O_2Si$: C, 64.41; H, 10.81; Found: C, 64.38; H, 10.80.

(*rel*)-(1′*R*,4′*S*)-9-[4-(*t*-Butyldimethylsilyloxy)-3,4-dimethyl-cyclopent-2-en-1-yl] 6-chloropurine (14)

To a solution containing compound **13b** (131 mg, 0.542 mmol), triphenylphosphine (570 mg, 2.17 mmol) and 6-chloropurine (167 mg, 1.085 mmol) in anhydrous THF (6 mL), diisopropyl azodicarboxylate (DIAD) (219 mg, 1.085 mmol) was added dropwise at −20°C for 30 minutes under nitrogen. The reaction mixture was stirred for 3 hours at 0°C and further stirred overnight at room temperature under nitrogen. The solvent was concentrated under reduced pressure and the residue was purified by silica gel column chromatography (EtOAc/hexane, 2.5:1) to give compound **14** (86 mg, 42%) as a white solid: m.p. 167–169°C;^{1}H NMR ($CDCl_3$, 300 MHz) δ

8.72 (s, 1H), 8.50 (s, 1H), 5.41 (s, 1H), 4.56 (dd, $J = 7.8$, 2.2 Hz, 1H), 2.52 (dd, $J = 10.6$, 8.6 Hz, 1H), 2.13 (dd, $J = 10.6$, 6.8 Hz, 1H), 1.76 (s, 3H), 1.54 (s, 3H), 0.81 (s, 9H), 0.01 (s, 6H); ^{13}C NMR ($CDCl_3$) δ 153.2, 151.7, 149.6, 144.3, 137.7, 133.2, 123.2, 78.3, 53.6, 41.7, 26.2, 25.1, 18.5, 17.5, 13.5, −5.4; Anal. Calc. for $C_{18}H_{27}ClN_4OSi$: C, 57.05; H, 7.18; N, 14.78; Found: C, 57.09; H, 7.22; N, 14.81.

(*rel*)-(1′*R*,4′*S*)-9-[4-Hydroxy-3,4-dimethylcyclopent-2-en-1-yl] 6-chloropurine (15)

To a solution of **14** (180 mg, 0.475 mmol) in THF (6.0 mL), TBAF (0.569 mL, 1.0 M solution in THF) was added at 0°C. The mixture was stirred overnight at room temperature and concentrated. The residue was purified by silica gel column chromatography (MeOH/Hexane/EtOAc, 0.1:4:1) to give **15** (85 mg, 87%) as a white solid: m.p. 164–166°C;^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.76 (s, 1H), 8.55 (s, 1H), 5.40 (s, 1H), 5.21 (s, 1H, D_2O exchangeable), 4.52 (t, $J = 7.2$ Hz, 1H), 2.60 (dd, $J = 10.8$, 8.7 Hz, 1H), 2.12 (dd, $J = 10.9$, 6.8 Hz, 1H), 1.75 (s, 3H), 1.51 (s, 3H); ^{13}C NMR (DMSO-d_6) δ 153.4, 151.7, 159.2, 147.7, 137.5, 134.6, 124.4, 78.3, 56.9, 43.5, 26.4, 17.5, 13.5; Anal. Calc. for $C_{12}H_{13}ClN_4O \cdot 0.5$ MeOH: C, 53.48; H, 5.38; N, 19.96; Found: C, 53.51; H, 5.36; N, 19.93.

(*rel*)-(1′*R*,4′*S*)- Diisopropyl [9-(4-Hydroxy-3,4-dimethylcyclopent-2-en-1-yl)] 6-chloropurine] Methylphosphonate (16)

Both LiO*t*-Bu (7.6 mL of 1.0 M solution in THF, 3.8 mmol) and a solution of diisopropyl bromomethylphosphonate (0.832 g, 3.21 mmol) in 6 mL of THF were slowly added to a solution of the carbocyclic purine analogue **15** (627 mg, 2.37 mmol) in 6 mL of THF at 0°C and stirred for 5 hours at room temperature under anhydrous conditions. The mixture was quenched by adding water (6 mL) and further diluted with additional H_2O (70 mL). The aqueous layer was extracted with EtOAc (3 × 70 mL). The combined organic layer was washed with brine, dried over $MgSO_4$, and concentrated in vacuo. The residue was purified by silica gel column chromatography (MeOH/Hexane/EtOAc, 0.05:3:1) to give **16** (619 mg, 59%) as a foamy syrup: m.p. 121–123°C;^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.73 (s, 1H), 8.52 (s, 1H), 5.45 (s, 1H), 4.72 (m, 2H), 4.52 (dd, $J = 7.4$, 1.8 Hz, 1H), 3.79 (dd, $J =$ 8.8, 2.8 Hz, 2H), 2.51 (dd, $J = 10.8$, 8.7 Hz, 1H), 2.08 (dd, $J = 10.8$, 6.8 Hz, 1H), 1.76 (s, 3H), 1.52 (s, 3H), 1.33 (m, 12H); ^{13}C NMR ($CDCl_3$) δ 152.9, 152.4, 150.3, 147.8, 137.7, 134.7, 124.6, 84.5, 72.2, 63.6, 53.8, 38.4, 24.2, 22.5, 14.4; Anal. Calc. for $C_{19}H_{28}ClN_4O_4P \cdot 1.0$ MeOH: C, 50.64; H, 6.80; N, 11.81; Found: C, 50.67; H, 6.82; N, 11.79.

(*rel*)-(1′*R*,4′*S*)-Diisopropyl [9-(4-Hydroxy-3,4-dimethylcyclopent-2-en-1-yl)] adenine] Methylphosphonate (17)

A solution of **16** (187 mg, 0.422 mmol) in saturated methanolic ammonia (8 mL) was stirred overnight on a steel bomb at 50°C, and the volatile components were evaporated. The residue was purified by silica gel column chromatography ($MeOH/CH_2Cl_2$, 1:7) to give **17** (123 mg, 69%) as a solid: m.p. 145–147°C; ^1H NMR (DMSO-d_6, 300 MHz) δ 8.28 (s, 1H), 8.14 (s, 1H), 5.45 (s, 1H), 4.70 (m, 2H), 4.49 (t, $J = 7.8$ Hz, 1H), 3.73 (dd, $J = 8.6$, 6.4 Hz, 2H), 2.57 (dd, $J = 10.6$, 8.6 Hz, 1H), 2.15 (dd, $J = 10.6$, 7.0 Hz, 1H), 1.78 (s, 3H), 1.53 (s, 3H), 1.33 (m, 12H); ^{13}C NMR (DMSO-d_6) δ 154.8, 152.7, 151.5, 147.4, 137.5, 130.6, 118.8, 83.5, 72.6, 66.7, 54.2, 38.6, 26.4, 22.6, 14.3; Anal. Calc. for $C_{19}H_{30}N_5O_4P \cdot 1.0$ MeOH: C, 52.74; H, 7.52; N, 15.37; Found: C, 52.73; H, 7.49; N, 15.39.

(*rel*)-(1′*R*,4′*S*)-[9-(4-Hydroxy-3,4-dimethylcyclopentyl)] adenine] Methylphosphonic Acid (18)

To a solution of the phosphonate **17** (103 mg, 0.244 mmol) in CH_3CN (10 mL) was added trimethylsilyl bromide (407 mg, 2.66 mmol). The mixture was heated under reflux for 24 hours and then concentrated under reduced pressure. The residue was partitioned between CH_2Cl_2 (50 mL) and distilled H_2O (50 mL). The aqueous layer was washed out with CH_2Cl_2 and then freeze-dried to give target compound **18** (69 mg, 84%) as a yellowish foamy solid. m.p. 109–111°C; ^1H NMR (DMSO-d_6, 300 MHz) δ 8.29 (s, 1H), 8.13 (s, 1H), 7.15 (br s, 2H), 5.33 (s, 1H), 4.50 (d, $J = 7.8$ Hz, 1H), 3.79 (dd, $J = 8.8$, 6.8 Hz, 2H), 2.54 (dd, $J = 10.8$, 8.8 Hz, 1H), 2.12 (dd, $J = 10.8$, 6.8 Hz, 1H), 1.73 (s, 3H), 1.46 (s, 3H); ^{13}C NMR (DMSO-d_6) δ 154.4, 152.3, 151.1, 146.7, 137.4, 124.7, 118.7, 84.7, 66.8, 54.2, 38.5, 23.2, 14.3; Anal. Calc. for $C_{13}H_{18}N_5O_4P \cdot 1.0\ H_2O$: C, 43.69; H, 5.64; N, 19.60; Found: C, 43.73; H, 5.61; N, 19.55.

(*rel*)-(1′*R*,4′*S*)-Bis(SATE) phosphoester of [9-(4-methyloxyphosphonate-3,4-dimethylcyclopentyl)] Adenine (20)

A solution of adenine phosphonic acid derivative **18** (72 mg, 0.212 mmol) and tri-*n*-butylamine (117 mg, 0.636 mmol) in methanol (4.5 mL) was mixed for 30 minutes and concentrated under reduced pressure. The residue was thoroughly dried with anhydrous ethanol and toluene. The resulting foamy solid was dissolved in anhydrous pyridine (15 mL) to which thioester **19** (649 mg, 4.0 mmol) and 1-(2-mesitylenesulfonyl)-3-nitro-1*H*-1,2,4-triazole (251 mg, 0.848 mmol) were added. The mixture was stirred overnight at room temperature and quenched with tetrabutylammonium bicarbonate buffer (12.0 mL, 1 M solution, pH 8.0). The mixture was concentrated under reduced pressure and the residue was diluted with water (70 mL) and

extracted with $CHCl_3$ (80 mL) two times. The combined organic layer was washed with brine, dried over Na_2SO_4, filtered, and evaporated. The residue was purified by silica gel column chromatography (MeOH/Hexane/EtOAc, 0.05:4:1) to give **20** (46 mg, 35%) as a white solid: m.p. 121–123°C; UV (MeOH) λ_{max} 262.5 nm;^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.31 (s, 1H), 8.16 (s, 1H), 5.39 (s, 1H), 4.49 (d, $J = 6.8$ Hz, 1H), 4.02 (m, 4H), 3.56 (d, $J = 9.6$ Hz, 2H), 3.16 (t, $J = 6.4$ Hz, 4H), 2.21–2.13 (m, 2H), 1.71 (s, 3H), 1.50 (s, 3H), 1.22–1.16 (s, 18H); ^{13}C NMR ($CDCl_3$) δ 204.8, 154.4, 152.7, 148.6, 145.5, 125.4, 118.6, 83.6, 62.6, 60.3, 53.4, 46.6, 38.4, 30.1, 26.8, 23.4, 13.9; Anal. Calc. for $C_{27}H_{42}N_5O_6PS_2 \cdot 1.0$ MeOH: C, 49.15; H, 7.03; N, 10.61. Found: C, 49.20; H, 6.97; N, 10.58.

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