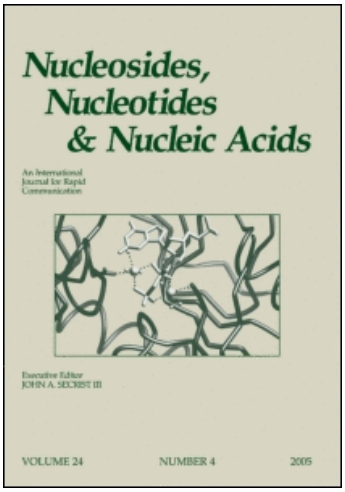


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Xinchao Chen[a]; Shintaro Matsumi[b]; Hiroaki Mitsuya[bc]; Jiri Zemlicka[ad]
[a] Developmental Therapeutics Program, Department of Chemistry, Barbara Ann Karmanos Cancer Institute, Wayne State University School of Medicine, Detroit, Michigan, USA [b] Experimental Retrovirology Section, HIV and AIDS Malignancy Branch, NCI, National Institutes of Health, Bethesda, Maryland, USA [c] Department of Internal Medicine II, Kumamoto University School of Medicine, Kumamoto, Japan [d] Developmental Therapeutics Program, Department of Chemistry, Barbara Ann Karmanos Cancer Institute, Wayne State University School of Medicine, Detroit, MI, USA

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# Synthesis of (*Z*)-(2,3-bis-Hydroxymethyl)-methylenecyclopropane Analogues of Purine Nucleosides

**Xinchao Chen,[1] Shintaro Matsumi,[2] Hiroaki Mitsuya,[2,3] and Jiri Zemlicka[1,*]**

[1]Developmental Therapeutics Program, Department of Chemistry, Barbara Ann Karmanos Cancer Institute, Wayne State University School of Medicine, Detroit, Michigan, USA
[2]Experimental Retrovirology Section, HIV and AIDS Malignancy Branch, NCI, National Institutes of Health, Bethesda, Maryland, USA
[3]Department of Internal Medicine II, Kumamoto University School of Medicine, Kumamoto, Japan



## ABSTRACT

Synthesis of (*Z*)-(2,3-bis-hydroxymethyl)methylenecyclopropane analogues of nucleosides adenosine **10a**, **10b**, **10c** and **17** is described. Epimerization of Feist's acid (**11**) using acetic anhydride gave cyclic anhydride **12** which was reduced in situ to give diol **13**. Acetylation (compound **14**) followed by addition of bromine led to dibromo derivative **15**. Alkylation-elimination of adenine with **15** afforded, after deacetylation, analogue **10a**. Similar treatment of 2-amino-6-chloropurine and 2,6-diaminopurine led to diacetates **16** and **18**. Deprotection then gave compounds **17** and **10c**. Hydrolysis of **17** furnished guanine analogue **10b**.

*Correspondence: Jiri Zemlicka, Developmental Therapeutics Program, Department of Chemistry, Barbara Ann Karmanos Cancer Institute, Wayne State University School of Medicine, Detroit, MI 48201-1379, USA; E-mail: zemlicka@kci.wayne.edu.



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Compounds **10a**, **10b** or **10c** were inactive against HCMV, HSV-1, HSV-2, EBV, VZV and HBV. Analogues **10a** and **10b** were also assayed for anti-HIV activity. Compound **10a** was effective in HIV-1/MT-2 culture with $EC_{50}/CC_{50}$ 33/> 100 μM but **10b** was inactive. Analogue **10a** was not a substrate for adenosine deaminase.

*Key Words:* Nucleoside analogues; Methylenecyclopropanes; Anti-HIV agents; Alkylation-elimination.

## INTRODUCTION

Methylenecyclopropane analogues of purine nucleosides are new antiviral agents with a broad-spectrum of activity.[1–3] Compounds **1** with *Z*-configuration are the most effective antivirals whereas the *E*-isomers **2** are in most cases devoid of a significant potency. Structure-activity relationships studies in this series of analogues have shown that an intact cyclopropane ring attached to a double bond carrying purine base in *Z* configuration is crucial for high efficacy of these analogues. Thus, the *Z*- and *E*-isomeric analogues **3** and **4** were inactive[4] although spiropentanes **5**, **6** and **7** lacking a double bond were inhibitors of replication of human cytomegalovirus (HCMV) or Epstein-Barr virus (EBV) in vitro.[5] Expansion of the cyclopropane ring of synadenol (**1a**) had a detrimental effect on antiviral potency as shown in Ref. [6] for methylenecyclobutane **8**. Although analogue **9** which combines structural features of the *Z*- and *E*-isomers **1** and **2** was also synthesized[7] no biological data were reported. This compound has the 3′,4′ hydroxymethyl groups in a *trans* (*E*) disposition. We have now prepared the *cis* (*Z*) isomer **10a** as well as corresponding guanine and 2,6-diaminopurine compounds **10b** and **10c** and investigated their biological activity.

## RESULTS AND DISCUSSION

Commercially available Feist's acid (**11**) was a convenient starting material for synthesis of all intended analogues. Epimerization of **11** using acetic anhydride under catalysis with potassium acetate to give the cyclic anhydride **12** was described.[8] Although this method was used in subsequent work[9,10] distillation of **12** was beset with a safety hazard.[10] Acetic anhydride was also used for conversion of *cis*/*trans* cyclopropane 1,2-dicarboxylic acids to the respective cyclic anhydride.[11] We have modified this procedure for epimerization of **11** using acetic anhydride and a catalytic amount of HCl (Sch. 1). After evaporation of volatile components in vacuo, the crude anhydride **12** was reduced in situ with DIBALH to give *cis*-diol **13** in 63% yield. The $^1$H NMR spectrum was similar to that described[9] and it was distinctly different from the respective *trans*-diol prepared by reduction of Feist's acid (**11**) with $LiAlH_4$. Routine acetylation of **13** afforded diacetate **14** (89%). Addition of bromine via pyridinium perbromide in $CH_2Cl_2$ afforded smoothly the *E*,*Z*-dibromo derivative **15** (86%) which was considered the reagent of choice for alkylation-elimination procedure as based on our previous experience.[1,12]





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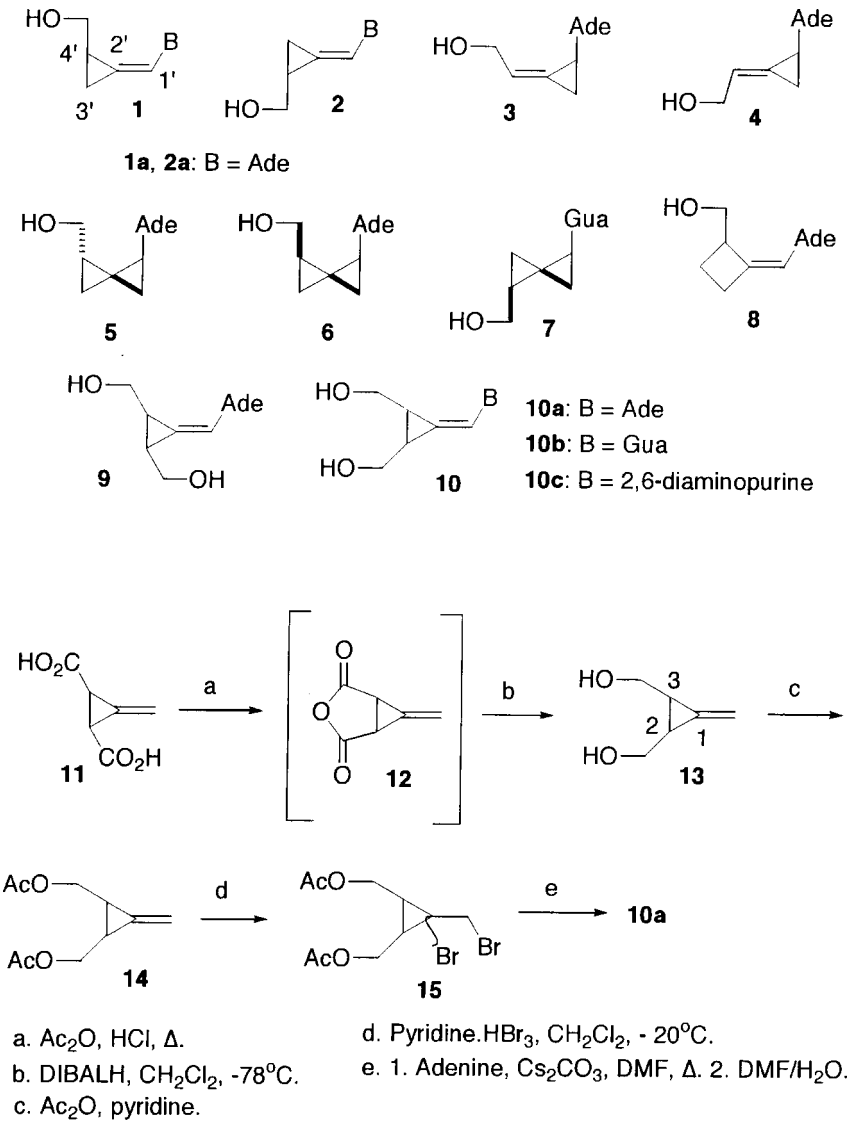


*Scheme 1.*

Reaction of **15** with adenine using $Cs_2CO_3$ in DMF (80°C, 30 h) gave, after deacetylation, analogue **10a** (50%). In case of 2-amino-6-chloropurine, the intermediary diacetate **16** was isolated in 65% yield (Sch. 2). Deprotection with $K_2CO_3$ in aqueous methanol furnished compound **17** (87%) Routine hydrolysis with formic acid afforded guanine analogue **10b** in 70% yield. Alkylation-elimination procedure with the dibromo derivative **15** and 2,6-diaminopurine led to diacetate **18** (59%). Deacetylation with $K_2CO_3$ in aqueous methanol then provided analogue **10c** in 90% yield.





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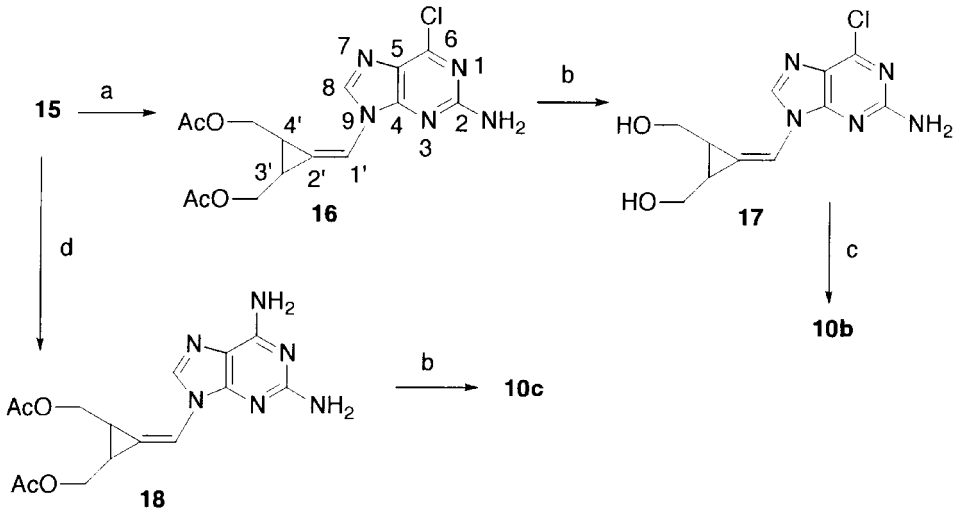


a. 2-Amino-6-chloropurine, $Cs_2CO_3$, DMF, Δ.
b. $K_2CO_3$, MeOH/$H_2O$.
c. 80 % $HCO_2H$, Δ.
d. 2,6-Diaminopurine, $Cs_2CO_3$, DMF, Δ.

*Scheme 2.*

Although *Z*,*E*-isomerism is absent in compounds **10a**, **10b** and **10c**, NMR spectra indicate a significant lack of symmetry. In several aspects, $^1$H NMR spectra resemble hybrids between the *Z*- and *E*-isomers **1** and **2**.[1,2] For example, the 3′-$CH_2$ (*trans* to the base) of adenine analogue **10a** occurs as a single multiplet at δ 3.60 as found in the *E*-isomer **2a**. By contrast, the *cis*-4′-$CH_2$ forms two distinctly different multiplets at δ 3.47 and 3.72, respectively, resembling the *Z*-isomers **2a**.[1] The OH signals (δ 4.76 and 5.03), $H_{3'}$ and $H_{4'}$ (δ 2.07 and 2.27) are also different. The chemical shift of $H_g$ (δ 8.72) is close to that of synadenol (**1a**, δ 8.74) indicating a significant deshielding by the 4′-$CH_2$OH. A similar differentiation is also seen in the $^{13}$C NMR spectra where non-equivalent 3′- and 4′-$CH_2$ as well as $C_{3'}$ and $C_{4'}$ are discernable. The $^1$H and $^{13}$C NMR spectra of **10a** in $CD_3OD$ are clearly different from those of the *trans* isomer **9**[7] corroborating further the assigned structure.

None of analogues **10a**, **10b** or **10c** exhibited a significant antiviral activity[13] against HBV, HCMV, HSV-1, HSV-2, VZV ($EC_{50} > 100\,\mu M$) and EBV ($EC_{50} > 50\,\mu M$). Adenosine analogue **10a** was effective against HIV-1 and MT-2 culture with $EC_{50}/CC_{50}$ $33/>100\,\mu M$. It was more active than the *E*-isomer **2a** but about 40 times less potent[14] than synadenol (**1a**). Guanine analogue **10b** was inactive ($EC_{50} > 100\,\mu M$). Compound **10a** was not a substrate for adenosine deaminase under the conditions employed for similar derivatives[1,4] using adenosine as a positive control. Thus, simultaneous presence of both 3′,4′ (*cis*) $CH_2$OH groups interferes with antiviral activity characteristic[1] for the 4′-substituted analogues (*Z*-isomers **1**) as well as with enzymic deamination[1,4] which prefers 3′-substituted compound (*E*-isomer **2a**).





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## EXPERIMENTAL SECTION

### General Methods

See[1]. UV spectra were taken in ethanol. NMR spectra were measured in $CD_3SOCD_3$ unless stated otherwise. Mass spectra (MS) were determined in an electron impact (EI) or electrospray ionization (ESI) mode.

**(*Z*)-2,3-bis(hydroxymethyl)methylenecyclopropane (13).** A mixture of Feist's acid (**11**, 2.50 g, 17.60 mmol), acetic anhydride (25 mL) and conc. HCl (5 drops) was stirred at 80–90°C for 2 days under $N_2$. The solution was evaporated to give crude anhydride **12** which was dissolved in $CH_2Cl_2$ (30 mL). DIBALH (1 M solution in hexane, 93 mL, 93 mmol) was then added dropwise at −78°C with stirring which was continued at 0°C for 3 h. The resultant solution was then cooled again to −78°C. The reaction was quenched by a careful addition of 2 M HCl (30 mL) and it was allowed to warm to room temperature. Ether (100 mL) was added, the organic phase was washed with 2 M HCl (2 × 30 mL), water, saturated $NaHCO_3$ and brine and it was dried over $Na_2SO_4$. Solvent was evaporated and the residue was chromatographed on a silica gel column using 30% EtOAc in hexane to give diol **13** (1.25 g, 63%). $^1$H NMR (400 MHz, $CDCl_3$) δ 2.07 (m, 2H, $H_2 + H_3$), 2.79 (bs, 2H, OH), 3.36 (m, 2H) and 4.05 (m, 2H, $CH_2O$), 5.37 (t, 2H, J = 2.4 Hz, $CH_2$=); $^{13}$C NMR (100 MHz) 22.3 ($C_2$, $C_3$), 61.2 ($CH_2O$), 105.1 ($CH_2$=), 135.0 ($C_1$); EI-HRMS calcd for $C_6H_{11}O_2$ (M + H) 115.0759, found 115.0761.

**(*Z*)-2,3-bis(acetoxymethyl)methylenecyclopropane (14).** Acetic anhydride (1.25 g, 12.0 mmol) was added dropwise to a solution of diol **13** (570 mg, 5.0 mmol) and pyridine (1.20 g, 15.0 mmol) in $CH_2Cl_2$ (20 mL) at room temperature with stirring. The reaction mixture was stirred for 16 h, ether (80 mL) was then added and the organic phase was washed with water (2 × 30 mL), saturated $CuSO_4$ (2 × 30 mL), brine (30 mL) and it was dried over $Na_2SO_4$. After evaporation of solvent, the residue was chromatographed using 10% EtOAc in hexane to give compound **13** (879 mg, 89%). $^1$H NMR (500 MHz, $CDCl_3$) δ 1.99–2.06 and 2.03 (overlapped m and s, 8H, $H_2 + H_3$ and $CH_3$), 3.95 (dd, 2H, J = 12.5 and 9.0 Hz) and 4.23 (dd, 2H, J = 12.0 and 6.5 Hz, $CH_2O$), 5.47 (t, 2H, J = 2 − 2.5 Hz, $CH_2$=); $^{13}$C NMR (125 MHz) 19.0 and 21.1 ($C_2 + C_3$ and $CH_3$), 62.6 ($CH_2O$), 106.1 ($CH_2$=), 133.5 ($C_1$), 171.0 (CO); EI-HRMS calcd for $C_{10}H_{15}O_4$ (M + H) 199.0970; found 199.0963.

**(*E*,*Z*)-1-Bromo-1-bromomethyl-2,3-bis(acetoxymethyl)cyclopropane (15).** Pyridinium tribromide (795 mg, 2.53 mmol) was added to a solution of compound **14** (400 mg, 2.02 mmol) in $CH_2Cl_2$ (10 mL) at −20°C with stirring. The stirring was continued at 0°C for 2.5 h, the reaction mixture was diluted with ether (50 mL), it was filtered and the solids were washed with ether (2 × 10 mL). The organic phase was washed with saturated $Na_2S_2O_3$ (3 × 20 mL), saturated $NaHCO_3$ (2 × 20 mL), brine (20 mL) and it was dried over $Na_2SO_4$. The solvents were evaporated and the residue was chromatographed using 20% EtOAc in hexane to give *E*,*Z* dibromide **15** (615 mg, 86%). $^1$H NMR (400 MHz, $CDCl_3$) δ 2.06 (s overlapped with m) and





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2.19 (m, 8H, $H_2 + H_3$ and $CH_3$), 3.73 and 3.82 (2s, 2H, $CH_2Br$), 4.09–4.11, 4.23 and 4.27 (3 m, 4H, $CH_2O$); $^{13}C$ NMR (100 MHz) 21.06, 21.09, 27.5, 32.2, 36.8, 59.4, 62.9, 170.8; ESI-MS (MeOH, NaCl) 379, 381, 383 (53.6, 100.0, 53.3, M + H). Anal. Calcd for $C_{10}H_{14}Br_2O_4$: C, 33.55; H, 3.94; Br, 44.64. Found: C, 33.60; H, 4.06; Br, 44.79.

**(*Z*)-9-[2,3-bis(hydroxymethyl)cyclopropylidenemethyl]adenine (10a).** A mixture of dibromide **15** (395 mg, 1.10 mmol), adenine (195 mg, 1.44 mmol) and $Cs_2CO_3$ (2.36 g, 6.60 mmol) in DMF (20 mL) was stirred at 80°C for 30 h under $N_2$. After cooling, the reaction mixture was filtered and the solids were washed with DMF ($2 \times 30$ mL,[15]). The filtrate was evaporated and the residue was chromatographed using 5–10% MeOH in $CH_2Cl_2$ to give compound **10a** (135 mg, 50%). Mp 249–250°C; UV max 288 (shoulder, ε 5,300), 276 nm (ε 8,300), 260 (shoulder, ε 11,000), 229 nm (ε 25,100); $^1H$ NMR (400 MHz) δ 2.07 (poorly resolved dd, 1H) and 2.27 (poorly resolved ddd, 1H, $H_{3'} + H_{4'}$), 3.47 (m, 1H) and 3.72 (m, 1H, 4′-$CH_2O$), 3.60 (m, 2H, 3′-$CH_2O$), 4.76 (t, 1H, J = 6.0 Hz, 3′-$CH_2$**OH**), 5.03 (split t, 1H, J = 5.6 Hz and 4.0 Hz, 4′-$CH_2$**OH**), 7.33 (bs, 2H, $NH_2$), 7.46 (s, 1H, $H_{1'}$), 8.16 (s, 1H, $H_2$), 8.72 (s, 1H, $H_8$); $^{13}C$ NMR (100 MHz) 21.7, 24.0 ($C_{3'}$, $C_{4'}$), 59.6, 59.8 ($CH_2O$), 111.0 ($C_{1'}$), 118.4, 119.1 ($C_{2'}$, $C_5$), 138.3 ($C_8$), 148.8 ($C_4$), 153.7 ($C_2$), 156.7 ($C_6$); $^1H$ NMR (500 MHz, $CD_3OD$) δ 2.25 (ddt, 1H, J = 8.5, 7.5 and 2.0 Hz) and 2.41 (ddt, 1H, J = 8.0, 8.5 and 2.0 Hz, $H_{3'} + H_{4'}$), 3.63 (dd, 1H, J = 11.7, 7.5 and 8.5 Hz) and 3.78 (dd, 1H, J = 11.7, 7.5 and 8.5 Hz, 4′-$CH_2O$), 3.85 (ddd, 2H, J = 12.0, 7.0 and 6.0 Hz, 3′-$CH_2O$), 7.52 (t, 1H, J = 2.0 Hz, $H_{1'}$), 8.22 (s, 1H, $H_2$), 8.74 (s, 1H, $H_8$); $^{13}C$ NMR (125 MHz) 21.2, 23.6 ($C_{3'}$, $C_{4'}$), 59.5, 59.8 ($CH_2O$), 110.8 ($C_{1'}$), 118.7, 119.0 ($C_{2'}$, $C_5$), 138.8 ($C_8$), 148.3 ($C_4$), 153.0 ($C_2$), 156.2 ($C_6$); ESI-MS (MeOH, NaCl) 517 (2M + Na, 22.2), 270 (M + Na, 76.1), 248 (M + H, 100.0). Anal. Calcd for $C_{11}H_{13}N_5O_2$: C, 53.43; H, 5.30; N, 28.32. Found: C, 53.59; H, 5.65; N, 28.51.

**(*Z*)-2-Amino-6-chloro-9-[2,3-bis(acetoxymethyl)cyclopropylidenemethyl]purine (16).** A mixture of dibromide **15** (899 mg, 2.51 mmol), 2 amino-6-chloropurine (510 mg, 3.02 mmol) and $Cs_2CO_3$ (4.90 mg, 15.1 mmol) in DMF (60 mL) was stirred at 90°C for 9 h under $N_2$. After cooling, the solids were filtered off, they were washed with DMF ($4 \times 30$ mL) and the filtrate was evaporated. The crude product was chromatographed using 5% MeOH in $CH_2Cl_2$ to give compound **16** (594 mg, 64.5%). Mp 187–188°C; UV max 310 (ε 8,600), 235 (ε 32,600), 204 nm (ε 18,100); $^1H$ NMR (500 MHz) δ 1.93 (s, 3H) and 2.04 (s, 3H, $CH_3$), 2.11 (m, 1H) and 2.34 (m, 1H, $H_{3'} + H_{4'}$), 4.01 (dd, 1H) and 4.05–4.17 (m, 3H, $CH_2O$), 7.03 (bs, 2H, $NH_2$), 7.38 (s, 1H, $H_{1'}$), 8.39 (s, 1H, $H_8$); $^{13}C$ NMR (125 MHz) 20.1, 21.2, 21.4, 22.7 ($C_{3'}$, $C_{4'}$, $CH_3$), 65.1, 65.4 ($CH_2O$), 113.1 ($C_{1'}$), 117.1 ($C_{2'}$), 123.8 ($C_5$), 140.6 ($C_8$), 150.4 ($C_4$), 153.3 ($C_2$), 160.8 ($C_6$), 170.8, 171.0 (CO); ESI-MS (MeOH, NaCl) 757, 755, 753 (2M + Na, 6.6, 42.9, 58.9), 390, 388 (M + Na, 18.2, 45.5), 368, 366 (M + H, 32.1, 100.0). Anal. Calcd for $C_{15}H_{16}ClN_5O_4$: C, 49.26; H, 4.41; N, 19.15; Cl, 9.69. Found: C, 49.38; H, 4.36; N, 19.32; Cl, 9.53.

**(*Z*)-2-Amino-6-chloro-9-[2,3-bis(hydroxymethyl)cyclopropylidenemethyl]purine (17).** Diacetate **16** (520 mg, 1.42 mmol) and $K_2CO_3$ (395 mg, 2.86 mmol) were stirred in 88% MeOH (35 mL) at room temperature for 2.5 h. Acetic acid (0.4 mL) was added

and the solution was stirred for another 0.5 h. The solvent was evaporated and the residue was chromatographed using 10–15% MeOH in $CH_2Cl_2$ to give compound **17** (345 mg, 87%). Mp 195–198°C; UV max 309 (ε 8,800), 235 (ε 32,900), 204 nm (ε 20,500); $^1$H NMR (500 MHz) δ 1.78 (ddd, 1H) and 1.94 (ddd, 1H, $H_{3'} + H_{4'}$), 3.24–3.44 (2m's and dd overlapped with $H_2O$) and 3.83 (dd, 1H, $CH_2O$), 4.79 (bs, 1H) and 5.10 (bs, 1H, OH), 7.01 (bs, 2H, $NH_2$), 7.30 (s, 1H, $H_{1'}$), 8.77 (s, 1H, $H_8$); $^{13}$C NMR (125 MHz) 22.7, 25.2 ($C_{3'}$, $C_{4'}$), 62.9, 63.1 ($CH_2O$), 110.8 ($C_{1'}$), 119.7 ($C_{2'}$), 123.8 ($C_5$), 140.5 ($C_8$), 150.2 ($C_4$), 153.0 ($C_2$), 160.7 ($C_6$); ESI-MS (MeOH, NaCl) 589, 587, 585 (2M + Na, 3.6, 31.7, 44.9), 284, 282 (M + H, 31.4, 100.0). Anal. Calcd for $C_{11}H_{12}ClN_5O_2$: C, 46.90; H, 4.29; N, 24.86; Cl, 12.59. Found: C, 46.79; H, 4.28; N, 24.77; Cl, 12.66.

**(*Z*)-9-[2,3-bis(hydroxymethyl)cyclopropylidenemethyl]guanine (10b).** A solution of 2-amino-6-chloropurine derivative **17** (285 mg, 1.01 mmol) in 80% formic acid (10 mL) was heated at 90°C for 5 h. The solvent was evaporated, the residue was dissolved in water (30 mL) and the solution was lyophilized. The crude product was dried in vacuo (oil pump) for 16 h at room temperature. It was stirred in 6% methanolic $NH_3$ (7 mL) at room temperature for 16 h. The product was filtered off, it was washed with MeOH (10 mL) and then crystallized from 90% MeOH to give compound **10b** (189 mg, 70%). Mp 245°C (decomp.); UV max 271 (ε 14,000), 230 nm (ε 37,100); $^1$H NMR (400 MHz) δ 1.72 (m, 1H) and 1.87 (m, 1H, $H_{3'} + H_{4'}$), 3.25–3.46 (3m's overlapped with $H_2O$, 3H) and 3.79 (m, 1H, $CH_2O$), 4.77 (t, 1H, J = 6.0 Hz) and 5.06 (t, 1H, J = 5.2 Hz, OH), 6.50 (bs, 2H, $NH_2$), 7.20 (s, 1H, $H_{1'}$), 8.36 (s, 1H, $H_8$), 10.64 (s, 1H, NH); $^{13}$C NMR (100 MHz) 22.5, 25.1 ($C_{3'}$, $C_{4'}$), 62.9, 63.2 ($CH_2O$), 111.2 ($C_{1'}$), 116.9, 118.4 ($C_{2'}$, $C_5$), 134.8 ($C_8$), 150.2 ($C_4$), 154.6 ($C_2$), 157.3 ($C_6$); ESI-MS (MeOH, NaCl) 549 (2M + Na, 16.7), 527 (2M + H, 42.3), 286 (M + Na, 22.0), 264 (M + H, 100.0). Anal. Calcd for $C_{11}H_{13}N_5O_3 \cdot 0.5\ H_2O$: C, 48.53; H, 5.18; N, 25.72. Found: C, 48.46; H, 5.23; N, 25.33.

**(*Z*)-2,6-Diamino-9-[2,3-bis(acetoxymethyl)cyclopropylidenemethyl]purine (18).** A mixture of dibromide **15** (100 mg, 0.279 mmol), 2,6-diaminopurine (55 mg, 0.363 mmol) and $Cs_2CO_3$ (545 mg, 1.67 mmol) in DMF (15 mL) was stirred at 80–90°C for 15 h under $N_2$. After cooling, the solids were filtered off and they were washed with DMF (2 × 20 mL). Filtrate was evaporated and the residue was chromatographed using 5% MeOH in $CH_2Cl_2$ to give compound **18** (57 mg, 59%). Mp 189–190°C; UV max 279 (ε 9,800), 219 nm (ε 30,200). $^1$H NMR (400 MHz) δ 1.98 (s, 3H) and 2.04 (s, 3H, $CH_3$), 2.27 (ddd, 1H), 2.50 (ddd overlapped with $CHD_2SOCD_3$, $H_{3'} + H_{4'}$), 4.06 (ddd, 2H), 4.26 (dd, 1H, J = 12.0, 6.4 Hz) and 4.34 (dd, 1H, J = 11.6, 6.4 Hz, $CH_2O$), 5.90 (bs, 2H) and 6.81 (bs, 2H, $NH_2$), 7.32 (t, 1H, $H_{1'}$), 8.01 (s, 1H, $H_8$); $^{13}$C NMR (100 MHz) 18.3, 20.7, 21.3, 21.4 ($C_{3'}$, $C_{4'}$ and $CH_3$), 62.4, 62.5 ($CH_2O$) 113.3, 113.4, 113.5 ($C_{5'}$, $C_{1'}$, $C_{2'}$), 134.7 ($C_8$), 151.3 ($C_4$), 156.8 ($C_2$), 161.3 ($C_6$), 170.8, 171.0 (CO); ESI-MS (MeOH, NaCl) 715 (2M + Na, 20.1), 369 (M + Na, 100.0), 347 (M + H, 92.4). Anal. Calcd for $C_{15}H_{18}N_6O_4$: C, 52.02; H, 5.24; N, 24.26. Found: C, 52.24; H, 5.40; N, 24.38.

**(*Z*)-2,6-Diamino-9-[2,3-bis(hydroxymethyl)cyclopropylidenemethyl]purine (10c).** Diacetate **18** (150 mg, 0.434 mmol) and $K_2CO_3$ (130 mg, 0.954 mmol) were stirred in





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83% methanol (6 mL) at room temperature for 3 h. The solvent was removed and the residue was chromatographed using 25% MeOH in $CH_2Cl_2$ to give compound **10c** (101 mg, 90%). Mp 265–267°C; UV max 279 (ε 9,700), 219 nm (ε 30,600); $^1$H NMR (400 MHz) δ 2.02 (ddd, 1H) and 2.22 (ddd, 1H, $H_{3'} + H_{4'}$), 3.45 (m, 1H), 3.55 and 3.62 (2 overlapped m's, 3H, $CH_2O$), 4.73 (t, 1H, J = 5.6 Hz) and 4.96 (t, 1H, J = 5.2 Hz, OH), 5.85 (bs, 2H) and 6.75 (bs, 2H, $NH_2$), 7.24 (s, 1H, $H_{1'}$), 8.27 (s, 1H, $H_8$); $^{13}$C NMR (100 MHz) 21.6, 23.9 ($C_{3'}$, $C_{4'}$), 59.6, 59.9 ($CH_2O$), 111.2, 113.4, 116.4 ($C_{1'}$, $C_{2'}$, $C_5$), 134.7 ($C_8$), 151.1 ($C_4$), 156.8 ($C_2$), 161.3 ($C_6$); ESI-MS (MeOH, NaCl) 547 (2M + Na, 33.5), 285 (M + Na, 72.5), 263 (M + Na, 92.2), 102 (100.0). Anal. Calcd for $C_{11}H_{14}N_6O_2$: C, 50.38; H, 5.38; N, 32.04. Found: C, 50.48; H, 5.46; N, 31.85.

## HIV-1 Assays

The activity of **10a** and **10b** against HIV-1 was determined as previously described with minor modifications.[16,17] Briefly, MT-2 cells (2,000 cells/well) in 96-well flat-bottomed microtiter culture plates were exposed or unexposed to 100 $TCID_{50}$ (50% tissue culture infectious dose) of HIV-$1_{LAI}$ in the presence of various concentrations of each compound and incubated at 37°C for 7 days. Drug concentrations that suppressed HIV-1 replication by 50% ($EC_{50}$) were determined using the 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide (MTT) assay. The cytotoxic concentration of each compound that reduced cellular growth or viability of uninfected MT-2 cells by 50% ($CC_{50}$) was also assessed in this assay. All assays were performed in triplicate.



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