Available online at www.sciencedirect.com

Bioorganic & Medicinal Chemistry

Bioorganic & Medicinal Chemistry 14 (2006) 5866–5875

# Synthesis and biological evaluation of carboacyclic nucleosides with (*Z*) and (*E*)-9-[4,4-bis(hydroxymethyl)]-2-butenyl side chain

Ye Tang, Ramaiah Muthyala*,† and Robert Vince*

*Center for Drug Design, Academic Health Center and Department of Medicinal Chemistry, College of Pharmacy,*
*University of Minnesota, Minneapolis, MN 55455, USA*

Received 31 March 2006; revised 12 May 2006; accepted 15 May 2006
Available online 8 June 2006

**Abstract**—A series of carboacyclic nucleosides with (*Z*) and (*E*)-9-[4,4-bis(hydroxy-methyl)]-2-butenyl side chains were synthesized as ring open analogs of cyclohexene nucleosides **1** and bioisosteres of ganciclovir **3**. The (*E*)-isomers were obtained to compare the side chain geometry effect on antiviral activity.

## 1. Introduction

To overcome the drug-resistant virus strains and toxicity problems in antiviral chemotherapy, structurally modified nucleosides are continually being synthesized. Among them, carbocyclic and acyclic nucleosides are two successful classes because of their unique chemical and metabolic features. Carbocyclic nucleosides replace the oxygen atom of the ribofuranose with a methylene group, resulting in metabolic stability to endogenous phosphorylase.[1] A well-known example of this class is carbovir, a selective and potent inhibitor of HIV-1 replication, which led to the FDA approval of abacavir for the treatment of AIDS.[2,3]

Recently, cyclohexene nucleoside analogs of **1** (Fig. 1) have been reported.[4,5] The guanosine analogs were found to possess good activity against a broad spectrum of herpes viruses ($IC_{50} = 0.002–0.3$ μg/mL).[6] The stereoelectronic properties of these cyclohexene nucleosides were close to their natural counterparts because the double bond could assume the function of the oxygen atom of a furanose. The cyclohexene ring was considered a bioisostere of the furanose moiety.[7]

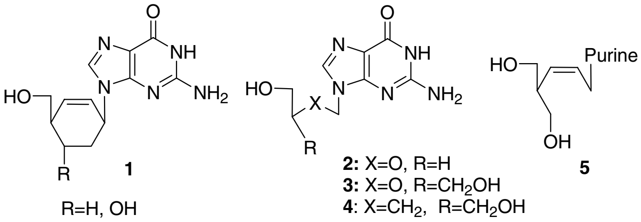


**Figure 1.** Carbocyclic and acyclic nucleoside analogs.

Interest in acyclic nucleoside research started from the discovery of acyclovir **2** (ACV, 'Zoviraz®'), which demonstrated to be a potent anti-viral agent against herpes diseases.[8] Acyclovir has been the gold standard for the treatment of HSV-1 and HSV-2 infections. Besides ACV, ganciclovir (GCV) **3** and penciclovir (PCV) **4** have also been used clinically as antiherpetic drugs.[9]

These successful modifications from a sugar moiety to an acyclic chain demonstrate that the total structure of the carbohydrate portion may not be required for biological activity.[10] Studies on acyclic nucleoside analogs as substrates and/or inhibitors for various enzymes demonstrate the importance of the acyclic chain and its ability to mimic the interaction of the glycosyl portion of the natural nucleoside with the enzyme (kinases).[11] Therefore, if the acyclic chain could adopt an optimal conformation for phosphorylation, biological activity would be expected to occur.

Based on the promising antiviral results of cyclohexene nucleosides, we designed a new series of unsaturated

---

*Keywords*: Carboacyclic; Nucleosides; Antiviral; Bioisosteres.

* Corresponding authors. Tel.: +1 612 624 9911; fax: +1 612 624 0139 (R.V.); tel.: +1 612 624 7120 (R.M.); e-mail addresses: muthy003@umn.edu; vince001@umn.edu

† Present address: Center for Orphan Drug Development, College of Pharmacy, Department of Medicinal Chemistry, 2001, 6th St., SE, University of Minnesota, Minneapolis, MN 55455, USA.

0968-0896/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmc.2006.05.029

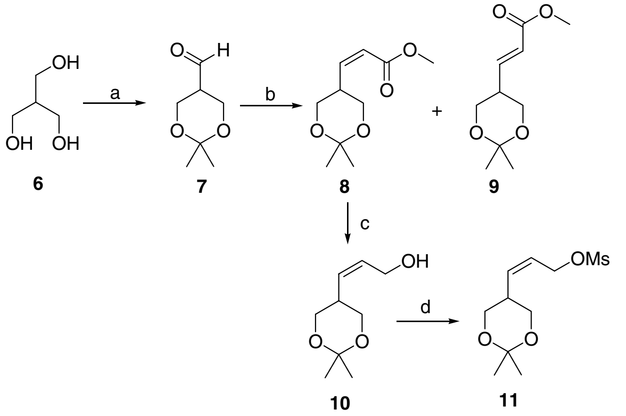
carboacyclic nucleosides **5** (Fig. 1), which can be considered as the corresponding ring open analogs. We hypothesize that the ring open modification would allow more flexibility on the side chain than from a cyclohexene ring. Also the existence of the double bond would still maintain a slight rigidity, as compared to **3** and **4**. The designed carboacyclic nucleoside may represent a compromise between a flexible side chain and the rigid cyclohexene moiety. We surmised that such a modification would also allow the nucleoside to stay in the favorable conformation that could interact with activating enzymes. These proposed carboacyclic nucleosides have two terminal primary hydroxyl groups, which could provide a substrate for phosphorylation by viral or cellular kinases. In addition, the carbon–carbon double bond within the acyclic chain can be regarded as an isostere of the oxygen in the side chain of GCV **3**. Compared to **3**, introducing the carboacyclic side chain would allow these nucleoside analogs to be more resistant to chemical and enzymatic degradation.

## 2. Results and discussion

### 2.1. Synthesis

The commercially available 2-(hydroxymethyl)propane-1,3-diol **6** was chosen as the appropriate starting material. Protection of **6** with 2,2-dimethoxypropane following Swern oxidation gave the aldehyde **7**.[12] A Horner–Wadsworth–Emmons (HWE) olefination[13] of **7** with the Still's reagent, bis(2,2,2-trifluoroethyl)-methoxycarbonyl methyl phosphonate, in the presence of LiCl and 1,8-diazabicyclo[5,4,0]-undec-7-ene(DBU), afforded two unsaturated ester products **8** and **9**, with the ratio of 4 to 1. Reduction of **8** with diisobutylaluminum hydride (DIBAL-H) led to the formation of an allylic alcohol **10**, which underwent mesylation to afford compound **11** (Scheme 1).

The mesylate **11** was coupled with adenine in the presence of NaH and DMF to generate the adenine derivative **12** (Scheme 2). A Mitsunobu reaction[14] between the alcohol **10** and adenine also gave compound **12** in a similar yield. Acid hydrolysis of **12** with 1 N HCl solution removed the isopropylidene group and gave the adenine analog **13**. Condensation between the mesylate **11** and 6-chloropurine provided two products as regioisomers, the $N_9$-substituted **14** and the $N_7$-substituted **15**, in a 4.5/1 ratio. The two regioisomers **14** and **15** were characterized by 2D-NMR experiments (HMBC). The correlation between H-1′ and C-4 and C-8 of the purine ring in compound **14** assigned the N-9 regiochemistry. For compound **15**, correlation was observed between H-1′, and C-5 and C-8 in the purine ring (Fig. 2).[15]



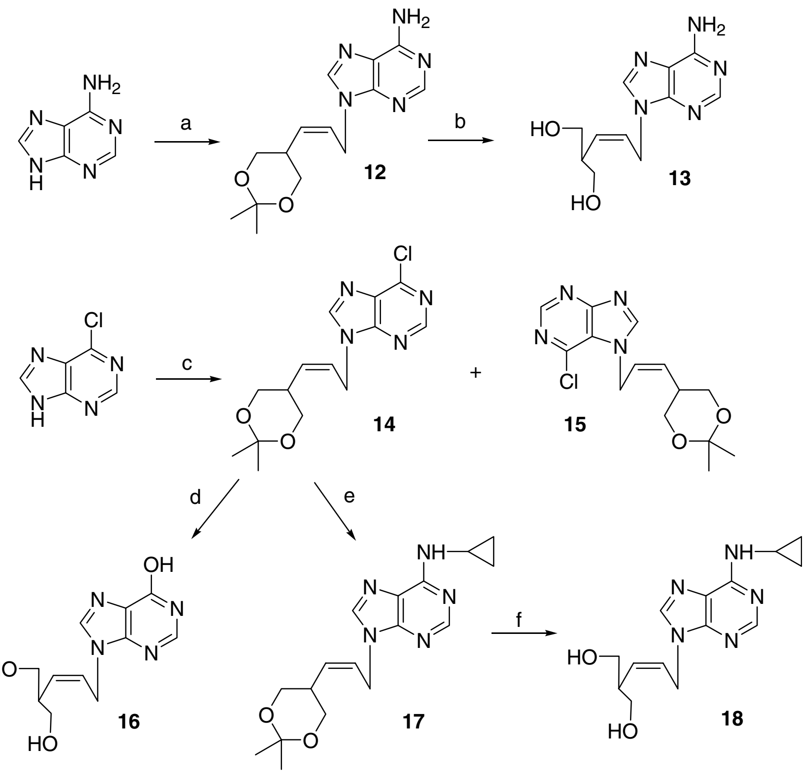
**Scheme 1.** Reagents and conditions: (a) i—2,2-dimethoxypropane, *p*-TsOH, THF, rt, 98%; ii—$(COCl)_2$, DMSO, $Et_3N$, $CH_2Cl_2$, −65 °C to rt, 63%; (b) $(CF_3CH_2O)_2P(O)CH_2COOCH_3$, LiCl, DBU, THF, −5 °C to 0 °C, 67%, **8/9** 4:1; (c) DIBAL-H, $CH_2Cl_2$, −78 °C, 84%; (d) MsCl, $Et_3N$, $CH_2Cl_2$, 0 °C, 87%.

An inosine derivative **16** was obtained by refluxing compound **14** in 1 N HCl. Displacement of the 6-chloro group of **14** with cyclopropyl-amine in ethanol and subsequent acid hydrolysis gave the cyclopropylamino derivative **18**. The syntheses of the 2,6-disubstituted purine analogs were accomplished via a similar synthetic method (Scheme 3). The 2-amino-6-chloropurine was condensed with mesylate **11** or the alcohol **10**, to afford the purine intermediate **19**. Treatment of **19** with 1 N HCl in tetrahydrofuran at room temperature for 30 min generated compound **20**, with no effect on the 6-chloro group. When compound **19** was refluxed with 1 N HCl,[16] the guanine analog **21** was obtained. The diaminopurine analog **22** was obtained by displacing the chloro group of **19** with liquid ammonia, followed by 1 N HCl hydrolysis. When compound **19** was treated with acidic resin in the presence of methanol, in addition to the expected deprotection, the chlorine was displaced with a methoxy group and compound **23** was obtained.

Because two unsaturated esters, **8** and **9**, were separated from the HWE olefination reaction, the same synthetic method was used to synthesize the purine derivatives incorporating the E-olefin moiety of the carboacyclic side chain. Reduction of **9** with DIBAL-H led to the formation of an allylic alcohol **24**, which underwent mesylation to afford **25** (Scheme 4). Condensation between the mesylate **25** and 6-chloropurine provided the $N_9$-substituted purine derivative **26** and the $N_7$-substituted purine derivative **27**, in a 2/1 ratio. Displacement of the chlorine of **26** with liquid ammonia, followed by 1 N HCl hydrolysis, afforded the adenine analog **29**. Coupling between 2-amino-6-chloropurine and mesylate **25** generated the purine intermediate **30**. Acid hydrolysis of compound **30** in THF at room temperature for 30 min yielded compound **31**. Refluxing **30** in 1 N HCl solution gave the guanine analog **32**. The diaminopurine analog **33** was obtained by displacing the halide of the compound **31** with liquid ammonia.

### 2.2. Biological results and conclusion

The synthesized unsaturated carboacyclic nucleoside analogs were evaluated against Herpes simplex virus types 1 and 2 (HSV-1, HSV-2) in VERO cell lines. Preliminary data indicated that the cyclopropylamino adenine analog **18** inhibited both herpes viruses with an $IC_{50}$ of 34.6 μM. The (*Z*)-guanine analog **21** had an $IC_{50}$ of 37.7 μM against HSV-2 and less potency



**Scheme 2.** Reagents and conditions: (a) **11**, NaH, DMF, 90 °C, 46.5% or **10**, DIAD, $PPh_3$, dioxane, 0 °C to rt, 44%; (b) 1 N HCl, rt, 85%; (c) **11**, NaH, DMF, rt, 40%, **14**/**15** 4.5:1; (d) 1 N HCl, reflux, 51.4%; (e) cyclopropylamine, ethanol, rt, 75%; (f) 1 N HCl, rt, 95%.

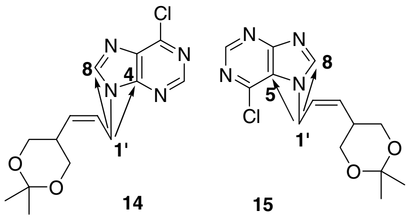


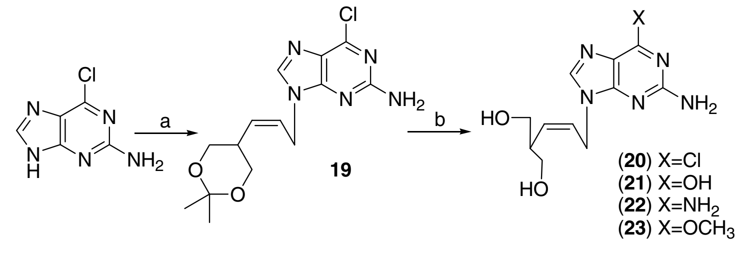
**Figure 2.** HMBC correlation comparison between **14** and **15**.

($IC_{50} > 37.7\ \mu M$) against HSV-1. The (*E*)-guanine analog **32** showed less potency ($IC_{50} > 37.7\ \mu M$) against both herpes viruses, as compared to acyclovir, a potent anti-herpes compound ($IC_{50} = 0.44\ \mu M$ for both HSV-1 and HSV-2). All compounds were evaluated for antiviral activity against HIV-1 and no activity was observed. Their antitumor activities were tested against a P388 mouse leukemia cell line and were devoid of cytotoxicity. The (*Z*) and (*E*) adenine and 2,6-diamino purine analogs (compounds **13**, **22**, **29**, and **33**) were tested for their adenosine deaminase activities. Compound **13** was found to be a slow substrate for adenosine deaminase. The other purine analogs were resistant to adenosine deaminase. The two adenine analogs, **13** and **29**, were found to be weak inhibitors for adenosine deaminase.

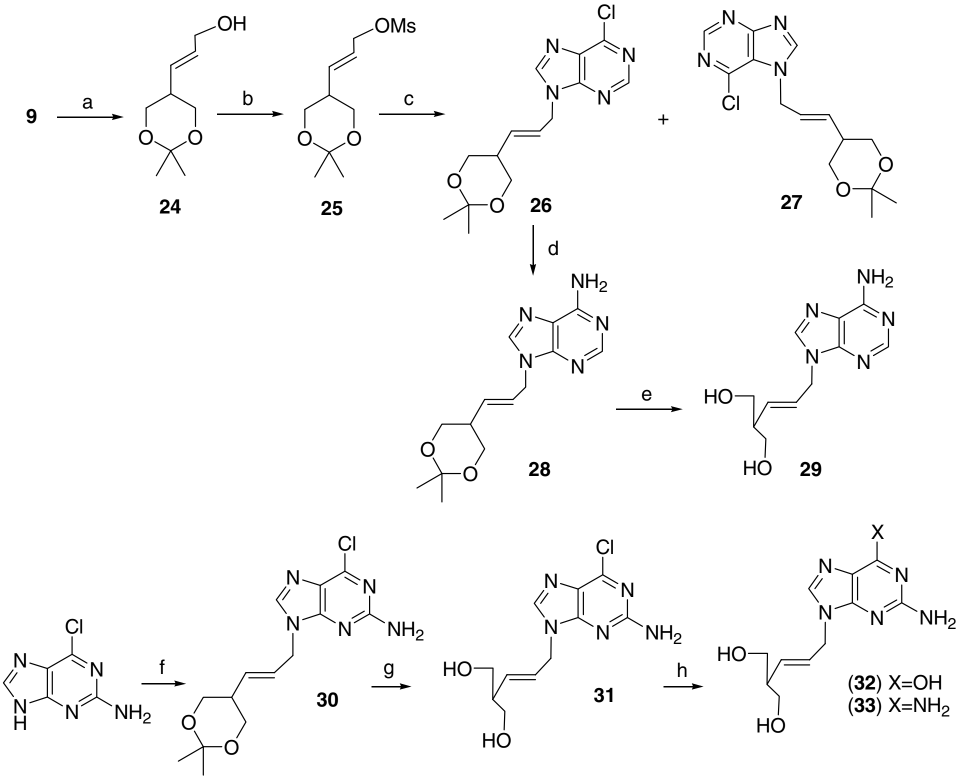
## 3. Experimental

### 3.1. Chemical experimental

All reactions involving moisture-sensitive reagents were conducted in oven-dried glassware under argon atmosphere. Solvents were dried when necessary. All chemicals and solvents were of reagent grade unless otherwise specified. The chemicals and solvents used were obtained from Aldrich Chemical Company, Milwaukee, WI. The $^1H$, $^{13}C$ NMR spectra were



**Scheme 3.** Reagents and conditions: (a) **11**, NaH, DMF, 90 °C, 35% or **10**, DIAD, $PPh_3$, dioxane, 0 °C to rt, 31%; (b) for **20**: 1 N HCl, rt, THF, 83%; for **21**: 1 N HCl, reflux, 63%; for **22**: liquid ammonia, 70 °C, 42% then 1 N HCl, rt, 60%; for **23**: Amberlyst 15 acidic resin, methanol, rt, 78%.



**Scheme 4.** Reagents and conditions: (a) DIBALH, $CH_2Cl_2$, −78 °C, 98%; (b) MsCl, $Et_3N$, $CH_2Cl_2$, 0 °C, 85%; (c) 6-chloropurine, NaH, DMF, rt, 44%, **26**/**27** 2:1; (d) liquid ammonia, 75 °C, 80%; (e) 1 N HCl, rt, 74.6%; (f) **25**, NaH, DMF, 90 °C, 37%; (g) 1 N HCl, THF, rt, 99%; (h) for **32**: 1 N HCl, reflux, 85%; for **33** liquid ammonia, 75 °C, 89%.

obtained on Varian Unity VAC-300 spectrometers and referenced to the solvent. Chemical shifts are expressed in parts per million (ppm, $\delta$) and coupling constants ($J$) are in Hertz (Hz). The peak multiplicities are abbreviated as follows: broad, br; singlet, s; doublet, d; triplet, t; quartet, q; pentet, p; septet, s; and multiplet, m. Positive-ion fast atom bombardment mass spectra (FAB-MS) were obtained on a VG-7070E-HF spectrometer. Positive-ion electron spray ionization mass spectra (ESI-MS) were obtained on a Bruker Biotof II spectrometer. Column chromatography was performed on Merck Science silica gel 60, 230–400 mesh. Thin-layer chromatography (TLC) was performed on Merck Science silica gel 60 F254 glass plates (0.25 mm thickness). Plates were visualized by UV light or by exposure to iodine vapor unless otherwise specified. The melting points were determined in an open capillary tube heated in a Mel-Temp II melting point apparatus and are uncorrected. The elemental analyses were performed by M-H-W Laboratories in Phoenix, AZ.

### 3.1.1. (*Z*)-3-(2,2-Dimethyl-[1,3]dioxan-5-yl)-acrylic acid methyl ester (8) and (*E*)-3-(2,2-dimethyl-[1,3]dioxan-5-yl)-acrylic acid methyl ester (9).

Anhydrous LiCl (513 mg, 12.1 mmol) was suspended under argon in THF (30 mL) and bis(2,2,2-trifluiroethyl)methoxycarbonylmethyl phosphonate (4.05 g, 12.1 mmol) was added at room temperature. The mixture was stirred for 15 min at room temperature and then cooled to 0 °C, then DBU (1.88 g, 12.1 mmol) was added. This mixture was stirred at −10 °C for 30 min, then aldehyde **7** (640 mg, 4.44 mmol) diluted in THF (5 mL) was added dropwise via syringe. Stirring continued for 1 h at −5 °C, saturated $NH_4Cl$ solution was added to quench the reaction. The mixture was extracted with ether (50 mL 2×) and the organic layer was combined and dried over $Na_2SO_4$. Filtration and removal of solvent gave a yellow liquid which was purified through a silica gel column eluted with ether/*n*-hexanes (1/6:1/4) to afford the compound **8** (471 mg, 53%) as a colorless liquid, $R_f$ 0.49 ($Et_2O$/*n*-hexanes 1:1). $^1H$ NMR ($CDCl_3$, 300 MHz): $\delta$ 6.38–6.31 (dd, 1H, $J = 11.42$), 5.85–5.81 (dd, $J = 11.43$), 4.01–3.96 (dd, 2H), 3.66–3.61 (m, 2H), 3.64 (s, 3H), 3.54–3.52 (m,1H), 1.38 (s, 3H), 1.36 (s, 3H); $^{13}C$ NMR ($CDCl_3$, 75 MHz): $\delta$ 166.35, 147.46, 121.27, 98.14, 63.80, 51.57, 34.82, 24.76, 23.72; HR-ESI: $(M+Na)^+$, calcd, 223.0940. Found: 223.0929; compound **9** (130 mg, 14%) was obtained as a colorless solid. mp: 32–33 °C, $R_f$ value 0.40 ($Et_2O$/*n*-hexanes 1:1). $^1H$ NMR ($CDCl_3$, 300 MHz): $\delta$ 6.80–6.72 (dd, 1H, $J = 15.82$), 5.88–5.83 (dd, 1H, $J = 15.82$), 3.88–3.82 (dd, 2H), 3.74–3.71 (d, 2H), 3.66 (s, 3H), 2.65–2.57 (m, 1H), 1.38 (s, 3H), 1.35 (s, 3H); $^{13}C$ NMR ($CDCl_3$, 300 MHz): $\delta$ 166.60, 144.95, 123.23, 98.17, 63.42, 51.99, 38.14, 27.07, 21.36; HR-ESI: $(M+Na)^+$, calcd, 223.0940. Found: 223.0951. Anal. Calcd for $C_{10}H_{16}O_4$: C, 59.98; H, 8.05. Found: C, 59.70; H, 7.89.

### 3.1.2. (*Z*)-3-(2,2-Dimethyl-[1,3]dioxan-5-yl)-prop-2-en-1-ol (10).

To a solution of compound **8** (600 mg, 3.0 mmol) in $CH_2Cl_2$ (15 mL) cooled at −78 °C was added dropwise a solution of DIBAL-H (1.0 M in cyclohexane, 9 mL, 9.0 mmol). After addition, the mixture was stirred at −78 °C for 2 h. The reaction was quenched with $CH_3OH$ (1 mL), then diluted with $CH_2Cl_2$ (100 mL) and washed with saturated $NH_4Cl$ (50 mL). The organic phase was separated and aqueous phase was extracted with $CH_2Cl_2$ (100 mL 2×). The combined organic phase was washed with brine (200 mL) and dried over $Na_2SO_4$. Filtration and removal of solvent gave a yellow liquid which was purified through a silica gel column eluting with ethyl ester/*n*-hexanes (v/v, 1/1) to afford the alcohol **10** (437 mg, 84%) as a colorless liquid. $R_f$ value 0.23 (ethyl acetate/*n*-hexanes 1:1). $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 5.72–5.64 (m, 1H, $J$ = 11.13, 6.73), 5.25–5.18 (dd, 1H, $J$ = 11.13, 4.39), 4.17-4.15 (dd, 2H, $J$ = 6.73), 3.73–3.57 (m, 4H), 2.86–2.76 (m, 1H, $J$ = 4.68), 1.69 (br, 1H, OH), 1.39 (s, 3H), 1.34 (s, 3H). $^{13}$C NMR ($CDCl_3$, 300 MHz): $\delta$ 132.72, 128.03, 97.82, 64.31, 58.98, 34.70, 28.33, 20.27; HR-ESI: $(M+Na)^+$, calcd, 195.0991. Found: 195.1000.

**3.1.3. (*Z*)-Methanesulfonic acid 3-(2,2-dimethyl-[1,3]dioxan-5-yl)-allyl ester (11).** Alcohol **10** (258 mg, 1.5 mmol) was dissolved in $CH_2Cl_2$ (15 mL) and cooled at 0 °C. $Et_3N$ (228 mg, 2.25 mmol) was added during stirring. Keep stirring at 0 °C for 10 min, then MsCl (195 mg, 1.7 mmol) was added slowly. This mixture was stirred at 0 °C for 1.5 h. Water (5 mL) was added and stirred for 30 min the organic layer was separated, washed with saturated $NaHCO_3$ (30 mL), brine (50 mL), and dried over $Na_2SO_4$. Removal of solvent afforded a faint yellow syrup as compound 102 (325 mg, 86.7%), which was used immediately to the next step without further purification. $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 5.73–5.56 (m, 2H), 4.73 (d, 2H), 3.80–3.74 (dd, 2H), 3.66–3.59 (dd, 2H), 2.95 (s, 3H), 2.84–2.75 (m, 1H), 1.38 (s, 3H), 1.35 (s, 3H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 132.37, 123.73, 96.55, 63.74, 62.47, 37.11, 33.27, 25.77, 19.81. LR-ESI: $(M+Na)^+$, 273.1.

**3.1.4. (*Z*)-9-[3-(2,2-Dimethyl-[1,3]dioxan-5-yl)-allyl]-9*H*-purin-6-ylamine (12).** *Preparation method 1.* A mixture of adenine (123 mg, 0.9 mmol) and NaH (36 mg, 60% in mineral oil, 0.9 mmol) in DMF (10 mL) was stirred at 90 °C for 1 h. Heating was stopped and at 60 °C, mesylate **11** (150 mg, 0.6 mmol) taken in DMF (4 mL) was added via syringe. After addition, the reaction mixture was stirred at 90 °C for 16 h. The reaction was stopped and cooled to room temperature. The solvent was evaporated to obtain a yellow solid residue. To this residue was added chloroform (50 mL) and washed with saturated $NaHCO_3$ (25 mL), brine (30 mL). The organic layer was separated and dried over $Na_2SO_4$. Removal of solvent gave a yellow residue, which was purified through a silica gel column eluting with $CHCl_3/CH_3OH$ (v/v, 30/1) to afford compound **12** (57.5 mg, 46.5%) as a white solid.

*Preparation method 2.* Diisopropal azodicarboxylate (414 µl, 2.0 mmol) taken in dry dioxane (6 mL) was added slowly into a stirred solution of alcohol **10** (172 mg, 1.0 mmol), adenine (270 mg, 2.0 mmol), and triphenylphosphine (525 mg, 2.0 mmol) in dry dioxane (15 mL) at 0 °C. After addition, the suspension solution was allowed to warm to room temperature and stirred for 24 h. The reaction mixture was filtered to remove the insoluble solid. The filtrate was concentrated to give a brown syrup, which was purified through a silica gel column eluting with $CHCl_3/CH_3OH$ (v/v, 50/1, then 30/1) to afford **12** (126 mg, 43.6%) as a white solid. $R_f$ value 0.68 ($CHCl_3/CH_3OH$ 4:1); mp 181–183 °C; $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 8.29 (s, 1H, *H*-2), 7.71 (s, 1H, *H*-8), 5.95 (br, 2H, $NH_2$, $D_2O$ exchangeable), 5.71–5.68 (m, 1H, $J$ = 10.84, 7.03), 5.54–5.47(m, 1H, $J$ = 10.25), 4.81–4.78 (d, 2H, $J$ = 7.74), 3.86–3.81 (dd, 2H, $J$ = 4.4), 3.70–3.63 (dd, 2H,$J$ = 9.13), 3.04–2.97 (m, 1H, $J$ = 9.08, 4.4), 1.40 (s, 3H), 1.37 (s, 3H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 155.71, 153.25, 150.07, 139.85, 131.47, 126.57, 119.82, 98.02, 63.96, 41.07, 34.70, 27.65, 20.90; HR-ESI: $(M+Na)^+$, calcd 312.1430. Found: 312.1425. Anal. Calcd. for $C_{14}H_{19}O_2N_5$: C, 58.12; H, 6.62; N, 24.21. Found: C, 57.98; H, 6.48; N, 24.49.

**3.1.5. (*Z*)-2-[3-(6-Amino-purin-9-yl)-propenyl]-propane-1,3-diol (13).** To compound **12** (75 mg, 0.26 mmol) in a 25 mL flask charged with stirrer at room temperature was added 1 N HCl (3 mL) and this mixture was stirred at room temperature for 3 h. The reaction mixture was neutralized to pH 7 with saturated $NaHCO_3$ solution. The solvent was removed and a yellow solid (100 mg) was obtained, which was separated through a silica gel column eluting with chloroform/methanol (v/v, 10/1) to yield compound **13** (55 mg, 85%) as a white solid. $R_f$ value 0.19 (chloroform/methanol 4:1); mp: 203–205 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 8.13 (s, 2H, *H*-2, *H*-8), 7.21 (s, 2H, br, $NH_2$, $D_2O$ exchangeable), 5.71–5.63 (m, 1H), 5.55–5.48 (m, 1H), 4.82–4.80 (d, 2H), 4.62–4.59 (t, 2H, 2 OH, $D_2O$ exchangeable), 3.53–3.46 (m, 2H), 3.41–3.37 (m, 2H), 2.85(m, 1H); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 156.54, 152.98, 149.87, 141.06, 134.87, 126.20, 119.33, 62.55, 43.89, 41.15; HR-ESI: $(M+Na)^+$, calcd 272.1117. Found: 272.1114. Anal. Calcd. For $C_{11}H_{15}O_2N_5$: C, 53.00; H, 6.07; N, 28.09. Found: C, 52.91; H, 5.99; N, 27.93.

**3.1.6. (*Z*)-6-Chloro-9-[3-(2,2-methyl-[1,3]dioxan-5-yl)-allyl]-9*H*-purine (14) and (*Z*)-6-chloro-7-[3-(2,2-methyl-[1,3]dioxan-5-yl)-allyl]-7*H*-purine (15).** To a mixture of 6-chloropurine (0.69 g, 4.4 mmol) and sodium hydride (0.21 g, 5.28 mmol, 60% in mineral oil) in dry DMF (20 mL) under argon protection at room temperature was added the mesylate **102** (1.1 g, 4.4 mmol) diluted in DMF (10 mL). The reaction mixture was stirred at room temperature under argon for 16 h. Water (10 mL) was added to the reaction mixture and stirred for 20 min. The mixture was extracted with chloroform (100 mL, twice) and the organic layer was combined and dried over $Na_2SO_4$. Removal of the solvent gave a brown syrup residue (1.5 g), which was separated through a silica gel column eluting with ethyl acetate/methanol (v/v, 150/1 to 100/1 to 40/1) to yield compound **14** (450 mg, 33%) as a white solid, $R_f$ value 0.44 (ethyl acetate/methanol 50:1); mp: 98–100 °C; $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 8.75 (s, 1H, *H*-2), 8.12 (s, 1H, *H*-8), 5.80–5.61 (m, 2H, $J$ = 10.84, 7.03), 4.99–4.96 (dd, 2H, $J$ = 7.03), 3.96–3.90 (dd, 2H, $J$ = 4.68), 3.80–

3.73 (dd, 2H), 3.19–3.06 (m, 1H,$J = 4.69$), 1.48 (s, 3H), 1.45 (s, 3H); $^{13}$C NMR (CDCl$_3$, 75 MHz): $\delta$ 152.19, 151.74, 151.25, 144.59, 132.51, 131.79, 125.46, 98.06, 63.87, 41.60, 34.79, 27.61, 20.90; HR-ESI: (M+Na)$^+$, calcd 331.0932. Found: 331.0946. Anal. Calcd for $C_{14}H_{17}ClO_2N_4$: C, 54.46; H, 5.55; N, 18.15. Found: C, 54.60; H, 5.68; N, 18.15. Compound **15** (100 mg, 7.4%) was obtained as a white solid, $R_f$ value 0.21 (ethyl acetate/methanol 50:1); mp: 86–87 °C; $^1$H NMR (CDCl$_3$, 300 MHz): $\delta$ 8.80 (s, 1H, *H*-2), 8.24 (s, 1H, *H*-8), 5.77–5.55 (m, 2H, $J = 10.8$, 6.6), 5.18–5.15 (dd, 2H, $J = 6.3$), 3.83–3.77 (dd, 2H, $J = 4.8$), 3.74–3.67 (t, 2H), 3.01–2.92 (m, 1H,$J = 4.5$), 1.42 (s, 3H), 1.37 (s, 3H); $^{13}$C NMR (CDCl$_3$, 75 MHz): $\delta$ 162.13, 152.61, 148.54, 143.13, 131.99, 126.55, 122.58, 98.08, 63.77, 44.82, 35.12, 27.75, 20.68; HR-ESI: (M+Na)$^+$, calcd 331.0932. Found: 331.0972.

**3.1.7. (*Z*)-2-[3-(6-Hydroxy-purin-9-yl)-propenyl]-propane-1,3-diol (16).** To compound **14** (120 mg, 0.39 mmol) in a 50 mL flask charged with stirrer at room temperature was added 1 N HCl (3 mL) and this mixture was heated at 90 °C for 3 h. The reaction mixture was neutralized to pH 7 with saturated NaHCO$_3$ solution. The solvent was removed and a yellow solid (110 mg) was obtained. This solid was crystallized from hot water and ethanol to yield compound **16** (50.0 mg, 51.4%) as a white solid. mp: 164–166 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 12.30 (br s, 1H, 6-OH, D$_2$O exchangeable), 8.07 (s, 1H,), 8.01 (s, 1H,), 5.68–5.47 (m, 2H), 4.80 (d, 2H), 4.60 (t, 2H, 2 OH, D$_2$O exchangeable), 3.48–3.31 (m, 4H), 2.80 (m, 1H); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 157.2, 148.7, 146.1, 140.5, 135.1, 125.8, 124.4, 62.3, 43.8, 41.5; HR-ESI: (M+Na)$^+$, calcd 273.0963. Found: 273.1103. Anal. Calcd For $C_{11}H_{14}O_3N_4$ + 1HCl: C, 45.93; H, 5.25; N, 19.48. Found: C, 45.53; H, 5.00; N, 19.48.

**3.1.8. (*Z*)-Cyclopropyl-{9-[3,2,2-dimethyl-[1,3]dioxan-5-yl]-allyl}-9*H*-purine-6-yl}-amine (17).** A solution of compound **14** (100 mg, 0.32 mmol) and cyclopropylamine (0.71 ml, 9.6 mmol) in ethanol (15 mL) was stirred at room temperature for 2 days. The solvent then was evaporated to give a yellow solid residue (100 mg). This residue was crystallized from ether (5 mL) to yield compound **106** as a white solid. (80 mg, 75%). mp: 102–104 °C; $^1$H NMR (CDCl$_3$, 300 MHz): $\delta$ 8.41(s, 1H), 7.67 (s, 1H), 6.31 (br s, 1H, D$_2$O exchangeable), 5.74–5.66 (m, 1H), 5.54–5.47 (m, 1H), 4.80 (d, 2H), 3.86–3.81 (m, 2H), 3.70–3.63 (m, 2H), 3.00 (m, 1H), 1.41 (s, 3H), 1.37 (s, 3H), 1.19 (s, 1H), 0.86 (d, 2H), 0.58 (s, 2H); $^{13}$C NMR (CDCl$_3$, 75 MHz): $\delta$ 155.9, 153.4, 149.2, 139.2, 131.3, 126.6, 120.0, 97.9, 63.9, 40.8, 34.6, 27.5, 24.0, 20.8, 7.7; HR-ESI: (M+Na)$^+$, calcd 352.1749, Found: 352.1944. Anal. Calcd For $C_{17}H_{23}O_2N_5$: C, 61.99; H, 7.04; N, 21.26. Found: C, 61.91; H, 6.97; N, 21.09.

**3.1.9. (*Z*)-2-[3-(6-Cyclopropylamino-purin-9-yl)-propenyl]-propane-1,3-diol (18).** To compound **17** (70 mg, 0.21 mmol) in a 25 mL flask charged with stirrer at room temperature was added 1 N HCl (5 mL) and this mixture was stirred at room temperature for 5 h. The reaction mixture was neutralized to pH 7 with 1 N NaOH solution. The solvent was removed and a brown solid residue (85 mg) was obtained, which was separated through a silica gel column eluting with chloroform/methanol (v/v, 20/1) to afford a syrup-like residue (65 mg). This residue was crystallized from ethanol–ether to yield compound **52** (58 mg, 95%) as a white solid, mp: 149–150 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 8.21 (br s, 1 H), 8.11 (s, 1H),7.84 (br s, 1H, D$_2$O exchangeable), 5.70–5.61(m, 1H, $J = 6.74$, 10.84), 5.54–5.47(m, 1H, $J = 9.95$, 10.84), 4.81–4.79(d, 2H, $J = 6.73$), 4.58(t, 2H, 2 OH, D$_2$O exchangeable, $J = 5.56$), 3.52–3.35 (m, 4H, $J = 5.56$, 10.54), 2.87–2.80(m, 1H, $J = 5.86$, 9.98), 2.49 (t, 1H), 0.7–0.58 (m, 4H); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 156.1, 152.8, 140.8, 134.8, 126.1, 119.6, 62.5, 43.8, 41.1, 24.7, 7.2; HR-ESI: (M+Na)$^+$, calcd: 312.1436. Found: 312.1448. Anal. Calcd For $C_{14}H_{19}O_2N_5$: C, 58.12; H, 6.62; N, 24.21. Found: C, 58.34; H, 6.85; N, 24.29.

**3.1.10. (*Z*)-6-Chloro-9-[3-(2,2-dimethyl-[1,3]dioxan-5-yl)-allyl]-9*H*-purin-2-ylamine (19).** *Preparation method 1.* A mixture of 2-amino-6-chloro purine (393 mg, 2.25 mmol), potassium carbonate (320 mg, 2.31 mmol)m and mesylate **11** (450 mg, 1.80 mmol) in dry DMF (30 mL) was stirred at 90 °C for 18 h under argon protection. The reaction was stopped and the solvent was evaporated to obtain a yellow solid residue. To this residue was added CHCl$_3$ (50 mL) and washed with NaHCO$_3$ (30 mL), brine (50 mL), the organic layer was separated and dried over Na$_2$SO$_4$. Removal of the solvent gave a yellow residue, which was purified through a silica gel column eluting with CHCl$_3$/CH$_3$OH (v/v, 50/1 to 30/1) to yield compound **19** (300 mg, 35.4%) as a yellow solid. mp: 193–194 °C.

*Preparation method 2.* Diisopropal azodicarboxylate (830 μl, 4.0 mmol) taken in dry dioxane (10 mL) was added slowly into a stirred solution of alcohol **10** (344 mg, 2.0 mmol), 2-amino-6-chloro purine (700 mg, 4.0 mmol), and triphenylphosphine (1060 mg, 4.0 mmol) in dry dioxane (20 mL) at 0 °C. After addition, the suspension solution was allowed to warm to room temperature and stirred for 24 h. The reaction mixture was filtered to remove the insoluble solid. The filtrate was concentrated to give a brown syrup, which was purified through a silica gel column eluting with CHCl$_3$/CH$_3$OH (v/v, 200/1, 150/1, 120/1) to yield **19** (200 mg, 31%) as a white solid. mp: 192–194 °C.

$^1$H NMR (CDCl$_3$, 300 MHz): $\delta$ 7.74 (s, 1H, H8), 5.75–5.66 (m, 1H), 5.57–5.50 (m, 1H), 5.16 (br s, 2H, NH$_2$, D$_2$O exchangeable), 4.76–4.73 (d, 2H), 3.90–3.84 (m, 2H), 3.78–3.71 (m, 2H), 3.26–3.14(m, 1H), 1.48 (s, 3H), 1.44 (s, 3H); $^{13}$C NMR (CDCl$_3$, 75 MHz): $\delta$ 159.25, 153.70, 151.48, 141.72, 131.74, 126.17, 125.43, 98.04, 64.00, 40.92, 34.91, 28.12, 20.48. Anal. Calcd for $C_{14}H_{18}O_2N_5Cl$: C, 51.93; H, 5.61; N, 21.63. Found: C, 51.85; H, 5.49; N, 21.75.

**3.1.11. (*Z*)-2-[3-(2-Amino-6-chloro-purin-9-yl)-propenyl]-propane-1,3-diol (20).** To a solution of compound **19** (200 mg, 0.62 mmol) in THF (10 mL) was added 2 N

HCl (0.5 mL) at room temperature. A white precipitate formed. Stirring continued for 30 min. Then the reaction mixture was diluted with THF and filtered to yield compound **20** (185 mg, 83%, HCl salt form) as a white solid. mp: 170 °C (decomposed). $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 8.33 (s, 1H), 6.38 (br s, 5H, 2 OH, $NH_2$, HCl, $D_2O$ exchangeable), 5.69–5.61(m, 1H, $J = 6.73$, 10.84), 5.55–5.49 (t, 1H, $J = 9.96$, 10.54), 4.73–4.71 (d, 2H, $J = 6.9$), 3.49–3.44 (dd, 2H, $J = 5.86$, 10.55), 3.37–3.31 (dd, 2H, $J = 6.44$), 2.85–2.76 (m, 1H, $J = 5.86$, 6.15, 9.96); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 160.4, 154.0, 149.4, 143.3, 135.6, 125.2, 122.4, 62.4, 43.9, 41.5. Anal. Calcd for $C_{11}H_{14}ClO_2N_5$ + 0.5 $H_2O$ + 0.5 HCl: C, 42.49; H, 5.02; N, 22.52. Found: C, 42.44; H, 4.95; N, 22.60.

**3.1.12. (*Z*)-2-[3-(2-Amino-6-hydroxy-purin-9-yl)-propenyl]-propane-1,3-diol (21).** To compound **19** (90.0 mg, 0.278 mmol) in a 50 mL flask charged with stirrer at room temperature was added 1 N HCl (9 mL) and this mixture was heated under reflux for 6 h. The reaction mixture was concentrated to give a brown syrup, which was dissolved in water (3 mL) and neutralized to pH 7 with 6 N NaOH solution. A white precipitate formed and this suspension was refrigerated for 1 h. Filtration gave a white solid, which was crystallized from hot water to yield compound **21** (46.5 mg, 63.1%) as a white solid. mp: 278–281 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 10.54 (s, 1H, 6-OH, $D_2O$ exchangeable), 7.64 (s, 1H, H8), 6.38 (br s, 2H, $NH_2$, $D_2O$ exchangeable), 5.57–5.43 (m, 2H), 4.58–4.53 (m, 4H, 2 OH, $D_2O$ exchangeable + N–$CH_2$), 3.45–3.33 (m, 4H), 2.75 (m, 1H); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 157.40, 154.6, 151.53, 137.60, 134.65, 126.23, 117.11, 62.50, 43.86, 40.88; Anal. Calcd for $C_{11}H_{15}O_3N_5$ + 0.6$H_2O$: C, 47.86; H, 5.91; N, 25.36. Found: C, 48.07; H, 5.93; N, 24.55.

**3.1.13. (*Z*)-2-[3-(2,6-Diamino-purin-9-yl)-propenyl]-propane-1,3-diol (22).** Liquid ammonia (~20 mL) was passed into a solution of **19** (100 mg, 0.31 mmol) in methanol (5 mL) at −78 °C in a bomb. This bomb was sealed and heated at 70 °C for 48 h. Ammonia and methanol were evaporated. The yellow solid (90 mg) left was purified through a silica gel column eluting with $CHCl_3/CH_3OH$ (v/v, 10/1) to afford a yellow solid (70 mg), which was crystallized from $CH_3CN$ to yield compound (*Z*)-9-[3-(2,2-dimethyl-[1,3]dioxan-5-yl)-allyl]-9*H*-purine-2,6-diamine (40 mg, 42%) as a light-yellow solid, mp: 172–174 °C; $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 7.48 (s, 1H), 5.76–5.67 (m, 1H, $J = 7.03$, 10.84), 5.52–5.45 (t, 1H, $J = 9.95$, 10.84), 5.40 (br s, 2H, $D_2O$ exchangeable), 4.73 (br s, 2H, $D_2O$ exchangeable), 4.69–4.66 (dd, 2H, $J = 7.03$), 3.90–3.84 (dd, 2H, $J = 4.98$), 3.76–3.69 (t, 2H, $J = 9.37$), 3.20–3.07 (m, 1H, $J = 9.96$, 9.67, 4.98), 1.47 (s, 3H), 1.43 (s, 3H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 160.0, 155.9, 152.1, 137.4, 130.8, 127.0, 114.6, 97.9, 64.0, 40.4, 34.7, 27.9, 20.5; HR-ESI: $(M+Na)^+$, calcd: 327.1545. Found: 327.1523. Anal. Calcd for $C_{14}H_{20}O_2N_6$: C, 55.25; H, 6.62; N, 27.61. Found: C, 54.88; H, 6.89; N, 27.49.

To the above compound (*Z*)-9-[3-(2,2-dimethyl-[1,3]dioxan-5-yl)-allyl]-9*H*-purine-2,6-diamine (480 mg, 1.58 mmol) in a 100 mL flask charged with stirrer at room temperature were added 1 N HCl (10 mL) and ethanol (2 mL). This mixture was stirred at room temperature for 6 h. The reaction mixture was neutralized with 5 N NaOH solution until pH reached 8–9. White precipitate appeared. This precipitate was filtered off and dried under vacuum to give a white solid (294 mg). Crystallization from ethanol yielded compound **22** (252 mg, 60%) as a light yellow solid. mp 192–194 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 7.67 (s, 1H), 6.66 (br s, 2H, $D_2O$ exchangeable), 5.73 (br s, 2H, $D_2O$ exchangeable), 5.64–5.56 (m, 1H,$J = 6.9$, 10.8), 5.50–5.43 (t, 1H, $J = 10.8$), 4.65 ( br s, 2H, 2 OH, $D_2O$ exchangeable), 4.62–4.59 (d, 2H, $J = 6.9$), 3.50–3.33 (m, 4H, $J = 6.3$), 2.84–2.76 (m, 1H, $J = 6.9$, 9.9); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 160.7, 156.6, 152.0, 137.6, 134.3, 126.5, 113.8, 62.5, 43.9, 40.8; HR-ESI: $(M+Na)^+$, calcd: 287.1232. Found: 287.1229. Anal. Calcd for $C_{11}H_{16}O_2N_6$: C, 49.99; H, 6.10; N, 31.80. Found: C, 49.17; H, 6.41; N, 31.42.

**3.1.14. (*Z*)-2-[3-(2-Amino-6-methoxy-purin-9-yl)-propenyl]-propane-1,3-diol (23).** To compound **19** in a 100 mL flask charged with stirrer was added dry methanol (30 mL) under argon. Amberlyst 15 acidic resin was added. Stirring continued at room temperature for 8 h. The reaction mixture was filtered and the filtrate was concentrated to give a yellow solid (230 mg), which was purified through a silica gel column eluting with ethyl acetate/$CH_3OH$ (v/v, 20/1 to 10/1) to yield compound **23** (143 mg, 78.9%) as a yellow solid. mp 144–146 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 7.83 (s, 1H), 6.38 (br s, 2H, $NH_2$, $D_2O$ exchangeable), 5.66–5.58 (m, 1H,$J = 7.03$, 10.54), 5.51–5.44 (m, 1H, $J = 9.96$, 10.54), 4.67 (d, 2H, $J = 7.03$), 4.57 (br s, 2H, 2 OH, $D_2O$ exchangeable), 3.93(s, 3H, –$OCH_3$), 3.50–3.45 (m, 2H, $J = 5.75$, 9.54), 3.38–3.33 (m,2H, $J = 9.46$), 2.86–2.74 (m, 1H, $J = 5.86$, 9.67); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 161.1, 160.3, 154.4, 139.9, 134.7, 126.1, 114.3, 62.5, 53.9, 43.8, 40.8. Anal. Calcd for $C_{12}H_{17}O_3N_5$: C, 51.60; H, 6.14; N, 25.08. Found: C, 51.49; H, 6.36; N, 25.04.

### 3.2. (*E*)-3-(2,2-Dimethyl-[1,3]dioxan-5-yl)-prop-2-en-1-ol (24)

To a solution of ester **9** (525 mg, 2.625 mmol) in $CH_2Cl_2$ (15 mL) cooled at −78 °C was added dropwise a solution of DIBAL-H (8.0 mL, 8.0 mmol, 1.0 M in $CH_2Cl_2$). After addition, the mixture was stirred at −78 °C for 2 h. The reaction was quenched with $CH_3OH$ (1 mL), then diluted with $CH_2Cl_2$ (100 mL) and washed with rochelle salt solution (50% saturated solution, 50 mL). The organic phase was separated and aqueous phase was extracted with $CH_2Cl_2$ (100 mL 2×). The combined organic phase was washed with brine (200 mL) and dried over $Na_2SO_4$. Filtration and removal of solvent gave a yellow liquid which was purified through a silica gel column eluting with ethyl ester/hexanes (v/v, 1/4) to yield compound **24** (443 mg, 98%) as colorless liquid.

$R_f$ value 0.21 in ethyl acetate/*n*-hexanes 1:1; $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 5.81–5.70 (m, 1H, J = 15.6, 5.4), 5.58–5.49 (m, 1H, $J = 15.3$, 4.8), 4.11–4.09 (dd, 2H, $J = 5.1$), 3.87–3.81 (m, 2H), 3.74–3.67 (m, 2H), 2.63–2.51 (m, 1H, $J = 4.77$), 2.06 (br, 1H, OH), 1.44 (s, 3H), 1.41 (s, 3H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 132.13, 128.18, 97.81, 64.40, 63.47, 37.99, 28.00, 20.53.

**3.2.1. (*E*)-Methanesulfonic acid 3-(2,2-dimethyl-[1,3]dioxan-5-yl)-allyl ester (25).** Alcohol **24** (247 mg, 1.44 mmol) was dissolved in $CH_2Cl_2$ (15 mL) and cooled at 0 °C. $Et_3N$ (218 mg, 2.15 mmol) was added during stirring. Keep stirring at 0 °C for 10 min, then MsCl (201 mg, 2.15 mmol) was added slowly. This mixture was stirred at 0 °C for 2 h. Water (5 mL) was added and stirred for 30 min the organic layer was separated, washed with saturated $NaHCO_3$ (30 mL), brine (50 mL), and dried over $Na_2SO_4$. Removal of solvent afforded a faint yellow syrup as compound **25** (348 mg, 97%), which was used immediately to the next step without further purification. $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 5.75–5.56 (m, 2H), 4.55 (d, 2H), 3.80–3.72 (dd, 2H), 3.67–3.56 (dd, 2H), 2.90 (s, 3H), 2.48–2.38 (m, 1H), 1.30 (s, 3H), 1.27 (s, 3H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 135.0, 124.9, 97.8, 70.4, 63.7, 38.2, 37.8, 26.8, 21.5.

**3.2.2. (*E*)-6-Chloro-9-[3-(2,2-methyl-[1,3]dioxan-5-yl)-allyl]-9*H*-purine (26) and (*E*)-6-chloro-7-[3-(2,2-methyl-[1,3]dioxan-5-yl)-allyl]-7*H*-purine (27).** To a mixture of 6-chloropurine (323 mg, 2.1 mmol) and NaH (84 mg, 2.1 mmol, 60% in mineral oil) in dry DMF (20 mL) under argon protection at room temperature was added the mesylate **25** (348 mg, 1.39 mmol) diluted in DMF (5 mL). The reaction mixture was stirred at room temperature under argon for 40 h. The reaction was stopped and solvent was evaporated at reduced pressure to give a yellow solid. To this solid residue was added chloroform and the insoluble solid was filtered off. The filtrate was concentrated to afford a yellow solid residue (350 mg), which was separated through a silica gel column eluting with ethyl acetate/methanol (v/v, 80/1) to yield compound **26** (160 mg, 37%) as a yellow solid, $R_f$ value 0.41 (ethyl acetate/methanol 50:1); mp: 83–84 °C; $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 8.69 (s, 1H, H-2), 8.05 (s, 1H, H-8), 5.81–5.61 (m, 2H), 4.82–4.80 (d, 2H), 3.85–3.80 (dd, 2H), 3.66–3.59 (dd, 2H), 2.52–2.44 (m, 1H), 1.35 (s, 3H), 1.34 (s, 3H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 152.2, 151.7, 151.2, 144.9, 133.8, 131.8, 125.4, 98.0, 63.9, 46.1, 37.9, 26.5, 21.9. Anal. Calcd for $C_{14}H_{17}ClO_2N_4$: C, 54.46; H, 5.55; N, 18.15. Found: C, 54.25; H, 5.31; N, 18.00.

Compound **27** (80 mg, 18%) was obtained as a white solid, $R_f$ value 0.15 (ethyl acetate/methanol 50:1); mp: 103–106 °C; $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 8.89 (s, 1H, H-2), 8.25 (s, 1H, H-8), 5.91–5.82 (m, 1H, J = 15.6, 6), 5.68–5.60 (m, 1H, $J = 15.6$), 5.10–5.08 (d, 2H, J = 6), 3.92–3.86 (m, 2H, $J = 4.8$), 3.71–3.64 (m, 2H), 2.59–2.48 (m, 1H, $J = 4.5$), 1.37 (s, 6H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 162.10, 152.71, 148.82, 143.18, 133.59, 126.37, 122.55, 98.10, 63.88, 49.16, 37.99, 26.31, 22.12; HR-ESI: $(M+Na)^+$, calcd 331.0932. Found: 331.0941.

**3.2.3. (*E*)-9-[3-(2,2-Dimethyl-[1,3]dioxan-5-yl)-allyl]-9*H*-purin-6-ylamine (28).** Liquid ammonia (20 mL) was passed into **26** (120 mg, 0.39 mmol) at −78 °C in a bomb. This bomb was sealed and heated at 75 °C for 20 h. Excessive ammonia and methanol were evaporated. To the yellow solid residue left was added methanol and the insoluble solid was filtered off. The filtrate was concentrated to give a white solid (112 mg), which was purified through a silica gel column eluting with $CHCl_3/CH_3OH$ (v/v, 10/1) to yield compound **28** as a white solid (90 mg, 80%); mp 205–206 °C; $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 8.37 (s, 1H, H2), 7.81 (s, 1H, H8), 6.02 (br, 2H, $NH_2$, $D_2O$ exchangeable), 5.86–5.77 (m, 1H, $J = 15.9$, 5.4), 5.72–5.65 (m, 1H, $J = 15.3$), 4.80–4.78 (d, 2H,$J = 5.7$), 3.90–3.84 (dd, 2H), 3.73–3.66 (dd, 2H), 2.60–2.51 (m, 1H), 1.43 (s, 3H), 1.41 (s, 3H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 160.08, 153.04, 152.88, 140.31, 132.21, 126.38, 119.66, 97.83, 63.83, 45.27, 37.69, 26.79, 21.08; HR-ESI: $(M+Na)^+$, calcd 312.1430. Found: 312.1433. Anal. Calcd for $C_{14}H_{19}O_2N_5$: C, 58.12; H, 6.62; N, 24.21. Found: C, 58.16; H, 6.68; N, 23.96.

**3.2.4. (*E*)-2-[3-(6-Amino-purin-9-yl)-propenyl]-propane-1,3-diol (29).** To compound **28** (70 mg, 0.24 mmol) in a 25 mL flask charged with stirrer at room temperature was added 1 N HCl (3 mL) and this mixture was stirred at room temperature for 3 h. The reaction mixture was neutralized to pH 7 with saturated $NaHCO_3$ solution. The solvent was removed and a yellow solid (100 mg) was obtained. To this solid was added methanol (20 mL) and the insoluble solid was filtered off. The filtrate was concentrated to give a yellow solid (82 mg), which was separated through a silica gel column eluting with chloroform/methanol (10:1) to yield compound **29** (45 mg, 74.6%) as a yellow solid, mp: 168–170 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 8.12 (s, 1H), 8.06 (s, 1H), 7.20 (s, br, 2H, $D_2O$ exchangeable), 5.77–5.56 (m, 4H, $J = 5.86$, 15.52), 4.70–4.68 (d, 2H, $J = 5.57$), 4.45–4.41 (t, 2H, $J = 5.28$, 2 OH, $D_2O$ exchangeable), 3.45–3.31 (m, 4H, $J = 5.27$), 2.25–2.19 (m, 1H); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 156.5, 153.0, 149.8, 141.0, 135.0, 126.2, 119.3, 62.2, 47.7, 45.4; HR-ESI: $(M+Na)^+$, calcd, 272.1117. Found: 272.1107. Anal. Calcd for $C_{11}H_{15}O_2N_5$: C, 53.00; H, 6.07; N, 28.10. Found: C, 53.22; H, 6.16; N, 28.30.

**3.2.5. (*E*)-6-Chloro-9-[3-(2,2-dimethyl-[1,3]dioxan-5-yl)-allyl]-9*H*-purin-2-ylamine (30).** A mixture of 2-amino-6-chloro purine (393 mg, 2.25 mmol) and potassium carbonate (320 mg, 2.31 mmol) in DMF (20 mL) was stirred at 60 °C for 20 min, then mesylate **25** (450 mg, 1.80 mmol) in dry DMF (10 mL) was added. The reaction mixture was stirred under argon protection at 90 °C for 20 h. The reaction was stopped and water (15 mL) was added. The mixture was extracted with chloroform (100 mL, three times). The organic layer was combined and dried over $Na_2SO_4$. Removal of the solvent afforded a yellow solid residue (400 mg), which was purified through a silica gel column eluting with $CHCl_3/CH_3OH$ (v/v, 10/1) to yield compound **30** (216 mg, 37%) as a yellow solid. mp: 145–147 °C. $^1$H NMR ($CDCl_3$, 300 MHz): $\delta$ 7.73 (s, 1H), 5.79–5.70 (m, 1H, $J = 5.86$, 15.52), 5.62–5.55 (m, 1H, $J = 7.91$, 15.53), 5.29 (br s, 2H, $NH_2$, $D_2O$ exchangeable), 4.65 (d, 2H, $J = 5.86$), 3.88–3.82 (dd, 2H), 3.69–3.63 (dd, 2H), 2.57–2.45 (m, 1H, $J = 4.69, 7.91$), 1.40 (s, 3H), 1.38 (s, 3H); $^{13}$C NMR ($CDCl_3$, 75 MHz): $\delta$ 159.2, 153. 7, 151.4, 142.0, 132.6, 126.0, 125.3, 98.0, 64.0, 45.4, 37.9, 26.7, 21.6. Anal. Calcd for $C_{14}H_{18}O_2N_5Cl$: C, 51.93; H, 5.61; N, 21.63. Found: C, 52.15; H, 5.79; N, 22.08; HR-ESI: $(M+Na)^+$, calcd 346.1046. Found: 346.1033.

**3.2.6. (*E*)-2-[3-(2-Amino-6-chloro-purin-9-yl)-propenyl]-propane-1,3-diol (31).** To a solution of compound **30** (140 mg, 0.43 mmol) in THF (10 mL) was added 2 N HCl (0.4 mL) at room temperature. A white precipitate formed. Stirring continued for 30 min. Then the reaction mixture was diluted with THF and filtered to yield compound **31** (137 mg, 99%, HCl salt form) as a white solid. Mp: 152–155 °C (decomposed). $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 8.34 (s, 1H), 6.70 (br s, 4H, 2 OH, $NH_2$, $D_2O$ exchangeable), 5.74–5.55 (m, 2H, $J = 5.57, 15.53$), 4.63–4.61 (d, 2H, $J = 5.57$), 3.43–3.31 (m, 4H, $J = 6.15$), 2.26–2.15 (m, 1H, $J = 6.15$); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 160.5, 154.0, 149.4, 143.3, 135.5, 125.3, 122.2, 62.0, 47.7, 45.7; HR-ESI: $(M+Na)^+$, calcd: 306.0728. Found: 306.0725.

**3.2.7. (*E*)-2-[3-(2-Amino-6-hydroxy-purin-9-yl)-propenyl]-propane-1,3-diol (32).** To compound **31** (100 mg, 0.31 mmol) in a 50 mL flask charged with stirrer at room temperature was added 1 N HCl (8 mL) and this mixture was heated under reflux for 3 h. The reaction was cooled and mixture was neutralized to pH 7 with 4 N NaOH solution. A white precipitate formed and this suspension was refrigerated for 1 h. The solid was filtered out and dried under vacuum to give a white solid, which was crystallized from hot water to afford compound **32** (70 mg, 85%) as a white solid. Mp: 266–269 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 10.54 (s, 1H), 7.61 (s, 1H), 6.43 (br s, 2H, $NH_2$, $D_2O$ exchangeable), 5.70–5.49 (m, 2H), 4.49–4.47 (d, 2H), 4.44–4.40 (t, 2H, OH, $D_2O$ exchangeable), 3.45–3.30 (m, 4H), 2.21 (m, 1H); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 157.4, 154.1, 151.5, 137.6, 134.6, 126.3, 117.1, 62.2, 47.76, 45.17. Anal. Calcd for $C_{11}H_{15}O_3N_5$: C, 49.81; H, 5.70; N, 26.40. Found: C, 49.96; H, 5.79; N, 26.53.

**3.2.8. (*E*)-2-[3-(2,6-Diamino-purin-9-yl)-propenyl]-propane-1,3-diol (33).** Liquid ammonia (15 mL) was passed into **31** (60 mg, 0.21 mmol) at −78 °C in a bomb. This bomb was sealed and heated at 75 °C for 24 h. Excessive ammonia was evaporated. The yellow solid residue left in the bomb was absorbed onto silica gel and purified through a silica gel column eluting with chloroform/methanol (v/v, 5/1) to yield compound **33** (50 mg, 89%) as a yellow solid, mp 209–212 °C; $^1$H NMR (DMSO-$d_6$, 300 MHz): $\delta$ 7.66 (s, 1H), 7.37 (br s, 2H, $D_2O$ exchangeable), 6.79 (br s, 2H, $D_2O$ exchangeable), 5.89 (br s, 2H, $D_2O$ exchangeable), 5.71–5.50 (m, 2H, $J = 5.5, 15.8$), 4.50 (d, 2H), 3.40 (d, 4H, $J = 6.16$), 2.23–2.18 (m, 1H, $J = 5.8, 6.15$); $^{13}$C NMR (DMSO-$d_6$, 75 MHz): $\delta$ 160.3, 156.2, 152.0, 137.9, 134.4, 126.5, 113.6, 62.2, 47.7, 44.9; HR-ESI: $(M+Na)^+$, calcd: 287.1226. Found: 287.1225. Anal. Calcd for $C_{11}H_{16}O_2N_6$: C, 49.99; H, 6.10; N, 31.80. Found: C, 49.29; H, 6.37; N, 31.19.

## 3.3. Biological experimental

**3.3.1. Antiviral Test.** The in vitro antiviral assays were carried out by virus-induced cytopathogenic effect s(CPE) inhibition studies. Confluent monolayers of African green monkey kidney cells (VERO cell lines) grown in minimal essential media supplemented with 5% fetal calf serum in COSTAR 96-well microtiter plates were infected with an inoculum of HSV-1, strain F. Final concentrations of 100 μg/mL followed by 10-fold serial dilutions of the test compounds were added to the wells (100–0.1 μg/mL). Virus controls contained no test compounds. Other wells remained free of virus to determine the drug toxicity effects against the Vero cells. Acyclovir, a potent anti-HSV agent, was used as the positive control. The 96-well plates were incubated for 3 days at 37 °C in a humidified atmosphere containing 5% $CO_2$ until the maximum CPE were observed in the virus control cultures. The cell monolayers were examined microscopically for virus-induced CPE and for drug cytotoxicity. The $IC_{50}$ value of the inhibitor is defined as the concentration of the drug that decreases the total number of plaques 50% to that present in the control wells with no test compound.

**3.3.2. Adenosine deaminase assay.** Calf spleen adenosine deaminase (100 milliunits, Sigma A-5168) was obtained from Sigma Chemical Company, St. Louis, Missouri. The standard conditions for substrate activity are as follows, adenosine deaminase, phosphate buffer (0.05 M, pH 7.3) is combined with 100 μ sample for a total volume of 1 mL (one unit of adenosine deaminase will deaminate 1.0 μmol adenosine to inosine per minute at pH 7.5 and 25 °C). The reaction was incubated overnight at 25 °C and monitored at the absorbance of the peak height, a decrease in absorbance at the wavelength peak indicates that a reaction did occur and determines whether the sample was a substrate or not for this enzyme. The wavelength at which to look for substrate activity was determined by reading 100 μM sample in 50 mM phosphate buffer, pH 7.3, at 25 °C for 2 s (the conditions for all enzymatic reactions are at 25 °C using a Beckman Coulter DU-7400 Spectrophotometer). The inhibitory effects of the test compounds against adenosine deaminase were evaluated by comparing the rate of deamination of adenosine with or without the presence of test compound. If deamination rate was delayed by the coexistence of the test compound, the compound was considered an inhibitor of adenosine deaminase.

## Acknowledgments

We thank Jay Brownell and Christine Dreis in our laboratory at the University of Minnesota for performing the in vitro antiviral (HSV-1 and HSV-2) assays and the adenosine deaminase assay. The anti-HIV-1 assay was conducted by Dr. Roger Ptak, Department of Infection Disease Research, Southern Research Institute, and evaluated by Dr. William M. Shannon.

## References and notes

1. Herdewijn, P.; De Clercq, E.; Balzarini, J.; Vanderhaeghe, H. *J. Med. Chem.* **1985**, *28*, 550–555.
2. Vince, R.; Hua, M.; Brownell, J.; Daluge, S.; Lee, F.; Shannon, W. M.; Lavelle, G. C.; Qualls, J.; Weislow, O. S.; Kiser, R.; Canonico, P. G.; Schultz, R. H.; Narayanan, V. L.; Mayo, J. G.; Shoemaker, R. H.; Boyd, M. R. *Biochem. Biophys. Res. Commun.* **1988**, *156*, 1046–1053.
3. Vince, R.; Hua, M. *J. Med. Chem.* **1990**, *33*, 17–21.
4. Konkel, M. J.; Vince, R. *Nucleosides Nucleotides* **1995**, *14*, 2061–2077.
5. Wang, J.; Herdewijn, P. *J. Org. Chem.* **1999**, *64*, 7820–7827.
6. Wang, J.; Froeyen, M.; Hendrix, C.; Andrei, G.; Snoeck, R.; De Clercq, E.; Herdewijn, P. *J. Med. Chem.* **2000**, *43*, 736–745.
7. Herdewijn, P.; De Clercq, E. *Bioorg. Med. Chem. Lett.* **2001**, *11*, 1591–1597.
8. Elion, G. B.; Furman, P. A.; Fyfe, J. A.; de Miranda, P.; Beauchamp, L.; Schaeffer, H. J. *Proc. Natl. Acad. Sci. U.S.A.* **1977**, *74*, 5716–5720.
9. De Clercq, E.; Andrei, G.; Snoeck, R.; De Bolle, L.; Naesens, L.; Degreve, B.; Balzarini, J.; Zhang, Y.; Schols, D.; Leyssen, P.; Ying, C.; Neyts, J. *Nucleosides, Nucleotides Nucleic Acids* **2001**, *20*, 271–285.
10. Hua, M.; Korkowski, P. M.; Vince, R. . *J. Med. Chem.* **1987**, *30*, 198–200.
11. Ashton, W. T.; Meurer, L. C.; Cantone, C. L.; Field, A. K.; Hannah, J.; Karkas, J. D.; Liou, R.; Patel, G. F.; Perry, H. C.; Wagner, A. F. . *J. Med. Chem.* **1988**, *31*, 2304–2315.
12. Maezaki, N.; Murakami, M.; Soejima, M.; Tanaka, T.; Imanishi, T.; Iwata, C. *Chem. Pharm. Bull.* **1996**, *44*, 1146–1151.
13. Ruebsam, F.; Evers, A. M.; Michel, C.; Giannis, A. *Tetrahedron* **1997**, *53*, 1707–1714.
14. Mitsunobu, O. *Synthesis* **1981**, 1–28.
15. Guillarme, S.; Legoupy, S.; Aubertin, A.-M.; Olicard, C.; Bourgougnon, N.; Huet, F. *Tetrahedron* **2003**, *59*, 2177–2184.
16. Lee, H.; Vince, R. *J. Pharm. Sci.* **1980**, *69*, 1019–1021.