Notes

3-Deaza- and 7-Deaza-5′-noraristeromycin and Their Antiviral Properties

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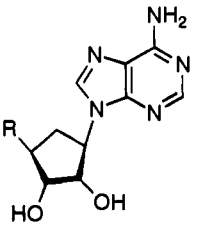
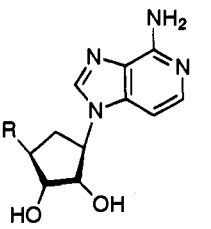
An enantiospecific synthesis of 3-deaza-5′-noraristeromycin as its dihydrochloride ((−)-**6**) has been accomplished in six steps beginning with the reaction of (+)-(1*R*,4*S*)-4-hydroxy-2-cyclopenten-1-yl acetate with 4-chloro-1*H*-imidazo[4,5-*c*]pyridine. The preparation of 7-deaza-5′-noraristeromycin ((−)-**7**) was described previously. Compounds (−)-**6** and (−)-**7** were evaluated for antiviral activity against a large number of viruses. Compound (−)-**6** produced an antiviral activity pattern similar to 5′-noraristeromycin but was less potent. Compound (−)-**6** inhibited CEM cell proliferation at a 50% inhibitory concentration of 27 μg/mL but proved not inhibitory to HEL cell proliferation and not toxic to E_6SM, HeLa, Vero, and MDCK cells at concentrations up to 200 μg/mL. While (−)-**6** showed inhibition of *S*-adenosyl-L-homocysteine (AdoHcy) hydrolase, it was less inhibitory than 5′-noraristeromcyin. Compound (−)-**7** displayed no antiviral properties or inhibitory effects toward AdoHcy hydrolase.

It is clear that new antiviral agents derived from aristeromycin (**1**) must be incapable of undergoing phosphorylation at the C-5′ center if undesirable side effects are to be avoided.[1] In that regard, 5′-noraristeromycin (**2**)[2] and its deoxy derivative **3**[3] have been found to possess selective antiviral properties, which is likely due to their inhibition of *S*-adenosyl-L-homocysteine (AdoHcy) hydrolase.[2,3] Prompted by similar properties for the 3-deaza analogue of **3** (that is, **4**)[3] and for 3-deazaneplanocin A (**5**),[4] 3-deaza-5′-noraristeromycin (**6**) became a logical extension of our studies[2] with **2**.

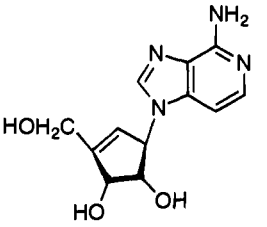
Also, the variation of **1** that led to **2** suggested that a similar alteration of 7-deazaaristeromycin (or carbocyclic tubercidin) could provide a compound (that is, **7**) with antiviral activity that was not dependent on C-5′ phosphorylation and thus circumvent the cytotoxicity associated with 7-deazapurine carbocyclic nucleosides.[5] Thus, 7-deaza-5′-noraristeromycin (**7**) has also been considered.



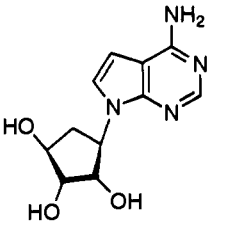
(-)-**1**, R=CH_2OH
(-)-**2**, R=OH
(-)-**3**, R=H



4, R=H
6, R=OH



(-)-**5**



(-)-**7**

Chemistry

The preparation of **6** began with the coupling of (+)-(1*R*,4*S*)-4-hydroxy-2-cyclopenten-1-yl acetate (**8**)[6] with 4-chloro-1*H*-imidazo[4,5-*c*]pyridine[7] in the presence of tetrakis(triphenylphosphine)palladium to yield enantiospecifically[6] **9** (Scheme 1). Acetylation of **9** to **10** was followed by standard vicinal glycolization conditions of osmium tetraoxide/*N*-methylmorpholine *N*-oxide to provide the "ribo" product **11**.[8] Due to the reduced reactivity of the 4-chloro atom of **11** toward nucleophilic displacement (compared to 6-chloropurine),[9] conversion of **11** into **6** followed a standard procedure[10] for 3-deazapurines by reacting **11** with hydrazine followed by treatment of the resultant hydrazino derivative with Raney nickel. Due to problems in purifying **6** as the free base, it was characterized as its dihydrochloride salt.

The synthesis of (−)-**7** has been previously described.[6]

Antiviral Results

Compounds (−)-**6** and (−)-**7** were evaluated against a wide variety of both DNA viruses and RNA viruses (Table 1). Compound (−)-**6** displayed the same antiviral activity spectrum as (−)-5′-noraristeromycin ((−)-**2**) but was clearly less active as an antiviral agent. In that regard, (−)-**6** retained activity against vaccinia virus (VV), vesicular stomatitis virus (VSV), parainfluenza-

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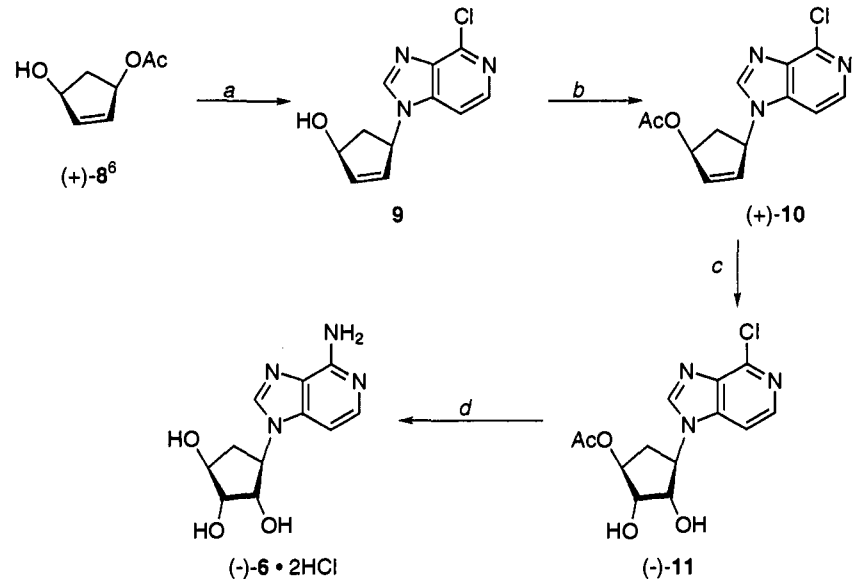
Scheme 1



Reaction conditions: *a*, (i) 4-chloro-1*H*-imidazo[4,5-*c*]pyridine[7] and NaH in THF then add (+)-**8**/$(Ph_3P)_4Pd/PPh_3$ in THF at room temperature and raise temperature to 55 °C; *b*, Ac_2O and pyridine in CH_2Cl_2 containing 4-(N,N-dimethylamino)pyridine; *c*, OsO_4/60% aq. N-methylmorpholine N-oxide in THF-H_2O; *d*, (i) N_2H_4 hydrate/100-120 °C; (ii) Raney Ni in H_2O/90 °C; (iii) HCl in EtOH

3, and reo-1 although it was about 10 times less potent against these viruses than (−)-**2**. On the other hand, in contrast to (−)-**2**, (−)-**6** lost activity against cytomegalovirus and the arenoviruses Junin and Tacaribe. Compound (−)-**7** virtually lost activity against all viruses when viewed in comparison to (−)-**2**. None of the compounds showed anti-HIV activity under the experimental conditions where 3′-azido-2′,3′-dideoxythymidine (AZT) proved active at 0.004 μM (data not shown).

Since the antiviral activity of 5′-noraristeromycin ((−)-**2**) was believed[2a] to be due to its inhibition of *S*-adenosyl-L-homocysteine (AdoHcy) hydrolase, both (−)-**6** and (−)-**7** were evaluated for their ability to inhibit this enzyme. The IC_{50} (μM) values were found to be as follows: (−)-**6**, 0.14 ± 0.042; (−)-**7**, ≥89; (−)-**2**, 0.042 ± 0.011; and neplanocin A, 0.0042 ± 0.0024. The order of decreasing inhibitory effects of these compounds on AdoHcy hydrolase activity (that is, (−)-**2** > (−)-**6** > (−)-**7**) follows the order of decreasing antiviral activity for this series. This observation further supports inhibition of AdoHcy hydrolase as the mechanism for the antiviral activity of the 5′-noraristeromycin class of adenosine analogues. Since recent modeling studies by Borchardt[11] indicate that all nitrogens of the purine ring are necessary for binding to AdoHcy hydrolase, it is not surprising that (−)-**6** is less potent than 5′-noraristeromycin ((−)-**2**) in its inhibition of this enzyme. AdoHcy hydrolase has been previously shown[12] to be a suitable target enzyme for inhibition of those viruses that are highly dependent on cellular methylation reactions, which suggests that further study should be carried out to ascertain the effects of the 5′-noraristeromycin series on viral required methylations.

Experimental Section

Materials and Methods. Unless otherwise noted, the reactions were carried out using freshly distilled solvents under anhydrous conditions in an argon atmosphere. The glassware was dried overnight in an oven at 100 °C. All reactions were monitored by thin-layer chromatography (TLC) using 0.25-mm E. Merck silica gel 60-F_{254} precoated silica gel glass plates with visualization by irradiation with a Mineralight UVGL-25 lamp, exposure to iodine vapor, or spraying with 3% phenol in 5% ethanolic H_2SO_4 and subsequent heating at 200 °C. The column chromatography purifications were performed on Aldrich flash chromatography silica gel 60 (particle size 0.035−0.07 mm; 220−440 mesh ASTM) by eluting with the indicated solvent system. The ^1H NMR and ^{13}C NMR spectra were recorded on either a JEOL FX90Q or a Bruker AMX-360 spectrometer in $CDCl_3$ or DMSO-d_6 referenced to internal tetramethylsilane (TMS) at 0.0 ppm. The spin multiplicities are indicated by the symbols s (singlet), d (doublet), t (triplet), m (multiplet), and br (broad). The UV spectrum for (−)-**6** was obtained using an IBM 9420 spectrophotometer. Melting point data was obtained using a Mel-Temp capillary melting point apparatus and are uncorrected. Optical rotations were recorded on Perkin-Elmer 241MC polarimeter. The microanalyses were performed by M-H-W Laboratories, Phoenix, AZ, on samples that were homogeneous by TLC analysis.

(1*S*,4*R*)-4-(4-Chloro-1*H*-imidazo[4,5-*c*]pyridin-1-yl)cyclopent-2-en-1-yl Acetate ((+)-10). To a solution of 4-chloro-1*H*-imidazo[4,5-*c*]pyridine[7] (5.39 g, 35.23 mmol) in dry THF (50 mL) was added NaH (60% dispersion in mineral oil, 1.41 g, 35.21 mmol). The reaction mixture was mechanically stirred at the room temperature for 30 min, followed by the addition of tetrakis(triphenylphosphine)palladium (2.0 g, 28.17 mmol), triphenylphosphine (1 g, 3.81 mmol), and a solution of (+)-**8**[6] (5 g, 35.21 mmol) in dry THF (100 mL). This mixture was stirred at 55 °C for 24 h. The volatiles were removed by rotary evaporation *in vacuo* at 18 mmHg and 50 °C. The residue was slurried in CH_2Cl_2 (250 mL) and filtered to remove insoluble solids, and the filtrate was washed with brine (2 × 200 mL), dried ($MgSO_4$), and evaporated to dryness. The residue (**9**) was dissolved in CH_2Cl_2 (250 mL) containing pyridine (15 mL) and 4-(*N*,*N*-dimethylamino)pyridine (200 mg). To this solution was added Ac_2O (50 mL) slowly, and this mixture was stirred overnight at the room temperature. After the mixture was washed with saturated aqueous $NaHCO_3$ (3 × 200 mL), the organic phase was dried ($MgSO_4$) and filtered, and the filtrate was evaporated to dryness. The residual oil was purified by

Table 1. Activity of Compounds (−)-6 and (−)-7 against Different Viruses in Different Cell Systems

virus	cell	MIC$_{50}$[a] (μg/mL) (−)-**6**	(−)-**7**	(−)-**2**	neoplanocin A
HSV-1 (KOS)	E_6SM	>400	300	>400	70
HSV-2 (G)	E_6SM	>400	>400	>400	>100
TK$^-$ HSV-1 (B2006)	E_6SM	300	300		20
TK$^-$ HSV-1 (VMW 1837)	E_6SM	300	300		70
VV	E_6SM	0.4	150	0.04	0.2
VSV	E_6SM	0.7	70	0.1	2
VSV	HeLa	2	300	0.2	2
Coxsackie B4	HeLa	>400	>400	>400	>40
Polio-1	HeLa	>400	>400	>400	>40
RSV (Long)	HeLa	≥150	100	>200	>1.2
Parainfluenza virus type 3 (ATCC VR-93)	Vero	1	100	0.07	0.2
Reo-1 (ATCC VR-230)	Vero	7	>200	0.7	0.7
Sindbis (ATCC VR-67)	Vero	≥200	>200	>400	2
Semliki forest (ATCC VR-67)	Vero	>200	>200	>400	7
Arenavirus (Junin)	Vero	>40	≥20	6	1
Arenavirus (Tacaribe)	Vero	>40	≥20	8	1
HIV-1 (III_B)	CEM	>20	>500	>0.16	
HIV-2 (ROD)	CEM	>20	>500	>0.16	
Influenza A (Ishikawa/222/82/H_3N_2)	MDCK	150	>200	>100	0.3−1.2
Influenza B (Singapore/222/79)	MDCK	>200	>200	>100	>4
CMV (AD-169)	HEL	>40	>40	0.01−0.05	0.6
CMV (Davis)	HEL	>40	>40	0.03−0.07	0.4
VZV (Oka)	HEL	>40	>40	>50	4
VZV (YS)	HEL	>40	≥40	>50	5
TK$^-$ VZV (07-1)	HEL	≥30	>40		4
TK$^-$ VZV (YS-R)	HEL	>40	≥40		3
morphology	E_6SM	>400	>400	>400	100
morphology	HeLa	>400	>400	>400	1−40
morphology	Vero	>200	≥400	>400	40
morphology	MDCK	>200	>200	>400	4
viability	CEM	27	>500	0.4	
proliferation	HEL	>200	>200	50	20

[a] Concentration required to reduce virus-induced cytopathicity (all viruses), cell viability (CEM), or cell proliferation (HEL) by 50%, or concentration required to cause a microscopically detectable alteration of normal cell morphology (E_6SM, HeLa, Vero, MDCK).

silica gel column chromatography (eluent AcOEt) to give **10** as a colorless foam (8.26 g, 85%): $[\alpha]^{25}_D$ +78.0° (*c* 0.5, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 2.15 (dt, *J* = 15.2 and 8.50 Hz, 1 H, H_a-5), 2.31 (s, 3 H, Me), 3.05 (dt, *J* = 15.2 and 8.50 Hz, 1 H, H_b-5), 5.03 (m, 1 H, H-1), 5.57 (d, *J* = 5.87 Hz, 1 H, H-4), 5.99 (dd, *J* = 5.57 and 2.32 Hz, 1 H, H-2), 6.63 (d, *J* = 5.57 Hz, I H, H-3), 7.33 (d, 1 H, heterocyclic H-7), 7.78 (d, 1 H, heterocyclic H-6), 8.64 (s, 1 H, heterocyclic H-2); ^{13}C NMR ($CDCl_3$) δ 25.38, 40.15, 58.70, 73.68, 98.67, 117.20, 127.54, 129.77, 137.67, 148.35, 149.80, 152.30, 171.43. Anal. ($C_{13}H_{12}ClN_3O_2$) C, H, N.

(1*S*,2*R*,3*S*,4*R*)-4-(4-Chloro-1*H*-imidazo[4,5-*c*]pyridin-1-yl)-2,3-dihydroxycyclopent-1-yl Acetate ((−)-11). To a solution of compound **10** (6.5 g, 23.47 mmol) in THF−H_2O (40:1, 100 mL) was added 60% aqueous *N*-methylmorpholine *N*-oxide (9 mL, 52.09 mmol) and osmium tetroxide (60 mg). The reaction mixture was stirred at the room temperature for 20 h. Sodium bisulfite (3 g) and H_2O (5 mL) were added into the reaction flask, and the resulting mixture was stirred at room temperature for 30 min. After removal of the solvent by rotary evaporation, the residue was purified by column chromatography using silica gel (AcOEt−MeOH, 9:1). The fractions containing product were evaporated to dryness to give **11** (6.38 g, 88%) as an analytically pure white solid: mp 60−63 °C; $[\alpha]^{25}_D$ −38.2° (*c* 0.5, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 1.80 (m, 1 H, H_a-5), 2.35 (s, 3 H, CH_3), 3.05 (m, 1 H, H_b-5), 3.78 (m, 3 H, H-1, H-2, and H-3), 4.57 (m, 2 H, H-4 and OH), 4.98 (m, 1 H, OH), 7.33 (d, 1 H, heterocyclic H-7), 7.78 (d, 1 H, heterocyclic H-6), 8.64 (s, 1 H, heterocyclic H-2); ^{13}C NMR ($CDCl_3$) δ 25.40, 39.54, 58.40, 73.79, 76.38, 77.15, 100.67, 102.87, 122.94, 149.87, 149.99, 151.65, 171.43. Anal. ($C_{13}H_{14}ClN_3O_4$) C, H, N.

(1*S*,2*R*,3*S*,4*R*)-4-(4-Amino-1*H*-imidazo[4,5-*c*]pyridin-1-yl)cyclopentane-1,2,3-triol Dihydrochloride ((−)-6). A solution of **11** (5 g, 16.08 mmol) in hydrazine monohydrate (100 mL) was heated at 100−120 °C for 6 h. After cooling to room temperature, the excess solvent was removed by rotary evaporation. The residue was dissolved in distilled H_2O (100 mL), and freshly prepared W_2 Raney Ni (prepared from 30 g of alloy) was added to it. The new suspension was heated at 90 °C for 2 h. The hot reaction mixture was filtered through a pad of Celite and the catalyst washed with hot H_2O (3 × 20 mL). The combined filtrates were evaporated to dryness. The oily residue was dissolved in hot EtOH, treated with decolorizing carbon, and filtered and the filtrate evaporated to dryness. This residue was dissolved in HCl saturated absolute EtOH (30 mL). After the mixture was refrigerated for a few days, the white solid that resulted was collected by filtration, washed with dry acetone, and dried over P_2O_5 *in vacuo* to give 1.83 g (46%) of **6** as its dihydrochloride: mp 250 °C (decomposed); $[\alpha]^{25}_D$ −40.6° (*c* 1, H_2O); UV λ_{max} at pH 1, 260 (log ε 10.0), 268 (9.8); at pH 7, 263 (10.3), 268 sh; at pH 13, 268 (9.8). The UV spectrum, which agrees well with 3-deazaaristeromycin,[10] provides proof that substitution in the first step occurred at the N-9 position and not at N-7: ^{1}H NMR (DMSO-d_6) δ 1.78 (m, 1 H, H_a-5), 3.15 (m, 1 H, H_b-5), 3.60 (m, 3 H, H-1, H-2, and H-3), 4.55 (m, 2 H, H-4 and OH), 5.00 (m, 2 H, 2 × OH), 7.33 (d, 1 H, heterocyclic H-7), 7.78 (d, 1 H, heterocyclic H-6), 8.48 (s, 2 H, NH_2), 8.64 (s, 1 H, heterocyclic H-2); ^{13}C NMR ($CDCl_3$) δ 40.04, 58.38, 74.09, 76.40, 77.00, 100.87, 102.65, 123.00, 150.02, 150.21, 156.64. Anal. ($C_{11}H_{14}N_4O_3$·2HCl) C, H, N.

Antiviral Activity Assays. The antiviral assays, other than the anti-HIV-1 assays, were based on an inhibition of virus-induced cytopathicity in either E_6SM, HeLa, Vero,

MDCK, or HEL cell cultures, following previously established procedures.[13-15] Briefly, confluent cell cultures in microtiter trays were inoculated with 100 $CCID_{50}$ of virus, 1 $CCID_{50}$ being the virus dose required to infect 50% of the cell cultures. After a 1 h virus adsorption period, residual virus was removed, and the cell cultures were incubated in the presence of varying concentrations (400, 200,100, ... μg/mL) of the test compounds. Viral cytopathicity was recorded as soon as it reached completion in the control virus-infected cell cultures.

Cytostatic Activity Assays. The cytostatic assays were performed as previously described.[16] Briefly, 100 μL aliquots of the cell suspensions (5×10^5 murine leukemia L1210 or 7.5×10^5 human T-lymphocyte Molt-4 or CEM cells/mL) were added to the wells of a microtiter plate containing 100 μL of varying concentrations of the test compounds. After a 2-day (L1210) or 3-day (Molt-4 and CEM) incubation period at 37 °C in a humidified CO_2-controlled incubator, the number of viable cells was determined using a Coulter Counter. Cytostatic activity is expressed as the compound concentration that reduced the number of viable cells by 50% (CC_{50}). The cytotoxicity measurements were based on microscopically visible alteration of normal cell morphology (E_6SM, HeLa, Vero, MDCK) or inhibition of normal cell growth (HEL), as previously described.[16]

Inhibition of HIV-Induced Giant Cell Formation. CEM cell cultures were suspended at 250,000-300,000 cells/mL of culture medium and infected with HIV-1 (III_B) or HIV-2 (ROD) at 100 $CCID_{50}$/mL. Then, 100 μL of the infected cell suspension were transferred to 200 μL microtiter plate wells containing 100 μL of serial dilutions of the test compound solutions. After 4 days of incubation at 37 °C, cell cultures were examined for syncytium formation as previously described.[17]

Inhibition of AdoHcy Hydrolase Activity. Measurements of L929 cell AdoHcy hydrolase activity inhibition were carried out as described by Cools and co-workers.[18]

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References

(1) Wolfe, M. S.; Borchardt, R. T. *S*-Adenosyl-L-homocysteine Hydrolase as a Target for Antiviral Chemotherapy. *J. Med. Chem.* **1991**, *34*, 1521-1530.

(2) (a) Patil, S. D.; Schneller, S. W.; Hosoya, M.; Snoeck, R.; Andrei, G.; Balzarini, J.; De Clercq, E. Synthesis and Antiviral Properties of (±)-5′-Noraristeromycin and Related Purine Carbocyclic Nucleosides. A New Lead for Anti-Human Cytomegalovirus Agent Design. *J. Med. Chem.* **1992**, *35*, 3372-3377. (b) Siddiqi, S. M.; Chen, X.; Schneller, S. W.; Ikeda, S.; Snoeck, R.; Andrei, G.; Balzarini, J.; De Clercq, E. Antiviral Enantiomeric Preference for 5′-Noraristeromycin. *J. Med. Chem.* **1994**, *37*, 551-554.

(3) Hasobe, M.; Liang, R.; Ault-Riche, D. B.; Borcherding, D. R.; Wolfe, M. S.; Borchardt, R. T. (1′*R*,2′*S*,3′*R*)-9-(2′,3′-Dihydroxycyclopentan-1′-yl)-adenine and -3-Deazaadenine: Analogues of Aristeromycin which Exhibit Potent Antiviral Activity with Reduced Cytotoxicity. *Antiviral Chem. Chemother.* **1993**, *4*, 245-248.

(4) Tseng, C. K. H.; Marquez, V. E.; Fuller, R. W.; Goldstein, B. M.; Haines, D. R.; McPherson, H.; Parsons, J. L.; Shannon, W. M.; Arnett, G.; Hollingshead, M.; Driscoll, J. S. Synthesis of 3-Deazaneplanocin A, a Powerful Inhibitor of *S*-Adenosylhomocysteine Hydrolase with Potent and Selective in Vitro and in Vivo Antiviral Activities. *J. Med. Chem.* **1989**, *32*, 1442-1446.

(5) Secrist, J. A., III; Clayton, S. J.; Montgomery, J. A. (±)-3-(4-Amino-1*H*-pyrrolo[2,3-*d*]pyrimidin-1-yl)-5-(hydroxymethyl)-(1α,2α,3β,5β)-1,2-cyclopentanediol, the Carbocyclic Analogue of Tubercidin. *J. Med. Chem.* **1984**, *27*, 534-536.

(6) Siddiqi, S. M.; Chen, X.; Schneller, S. W. Enantiospecific Synthesis of 5′-Noraristeromycin and Its 7-Deaza Derivative and a Formal Synthesis of (−)-5′-Homoaristeromycin. *Nucleosides Nucleotides* **1993**, *12*, 267-278.

(7) Mizuno, Y.; Itoh, T.; Saito, K. Studies on Condensed Systems of Aromatic Nitrogenous Series. XXIV. Synthesis of 4-Substituted 1*H*-Imidazo[4,5-*c*]pyridines. *Chem. Pharm. Bull.* **1964**, *12*, 866-872.

(8) (a) Trost, B. M.; Kuo, G.-H.; Benneche, T. A. Transition Metal-Controlled Synthesis of (±)-Aristeromycin and (±)-2′,3′-Aristeromycin. An Unusual Directive Effect in Hydroxylations. *J. Am. Chem. Soc.* **1988**, *110*, 621-622. (b) Deardorff, D. R.; Shulman, M. J.; Sheppeck, J. E., II. Palladium-Assisted Route to Carbocyclic Nucleosides: A Formal Synthesis of (±)-Aristeromycin. *Tetrahedron Lett.* **1989**, *30*, 6625-6628.

(9) Rousseau, R. J.; Townsend, L. B.; Robins, R. K. The Synthesis of 4-Amino-β-D-ribofuranosylimidazo[4,5-*c*]pyridine (3-Deazaadenosine) and Related Nucleosides. *Biochemistry* **1966**, *5*, 756-760.

(10) Montgomery, J. A.; Clayton, S. J.; Thomas, H. J.; Shannon, W. M.; Arnett, G.; Bodner, A. J.; Kion, I.-K.; Cantoni, G. L.; Chiang, P. K. Carbocyclic Analogue of 3-Deazaadenosine: A Novel Antiviral Agent Using *S*-Adenosylhomocysteine Hydrolase as a Pharmacological Target. *J. Med. Chem.* **1982**, *25*, 626-629.

(11) Yeh, J. C.; Borchardt, R. T.; Vedani, A. A Molecular Model for the Active Site of *S*-Adenosyl-L-homocysteine Hydrolase. *J. Comput.-Aided Mol. Design* **1991**, *5*, 213-234.

(12) Cools, M.; De Clercq, E. Correlation between the Antiviral Activity of Acyclic and Carbocyclic Adenosine Analogues in Murine L929 Cells and Their Inhibitory Effect on L929 Cell *S*-Adenosylhomocysteine Hydrolase. *Biochem. Pharmacol.* **1989**, *38*, 1061-1067.

(13) Schols, D.; De Clercq, E.; Balzarini, J.; Baba, M.; Witvrouw, M.; Hosoya, M.; Andrei, G.; Snoeck, R.; Neyts, J.; Pauwels, R.; Nagy, M.; Györgyi-Edelényi, J.; Machovich, R.; Horváth, I.; Löw, M.; Görög, S. Sulphated Polymers are Potent and Selective Inhibitors of Various Enveloped Viruses, including Herpes Simplex Virus, Cytomegalovirus, Vesicular Stomatitis Virus, Respiratory Syncytial Virus, and Toga-, Arena-, and Retroviruses. *Antiviral Chem. Chemother.* **1990**, *1*, 233-240.

(14) De Clercq, E.; Descamps, J.; Verhelst, G.; Walker, R. T.; Jones, A. S.; Torrence, P. F.; Shugar, D. Comparative Efficacy of Antiherpes Drugs against Different Strains of Herpes Simplex Virus. *J. Infect. Dis.* **1980**, *141*, 563-574.

(15) De Clercq, E.; Holy, A.; Rosenberg, I.; Sakuma, T.; Balzarini, J.; Maudgal, P. C. A Novel Selective Broad-spectrum anti-DNA Virus Agent. *Nature* **1986**, *323*, 464-467.

(16) De Clercq, E.; Balzarini, J.; Torrence, P. F.; Mertes, M. P.; Schmidt, C. L.; Shugar, D.; Barr, P. J.; Jones, A. S.; Verhelst, G.; Walker, R. T. Thymidylate Synthetase as a Target Enzyme for the Inhibitory Activity of 5-Substituted 2′-Deoxyuridines on Mouse Leukemia L-1210 Cell Growth. *Mol. Pharmacol.* **1981**, *19*, 321-330.

(17) Balzarini, J.; Naesens, L.; Schlachmuylders, J.; Niphuis, H.; Rosenberg, I.; Holy, A.; Schellekens, H.; De Clercq, E. 9-(2-Phosphonylmethoxyethyl)adenine (PMEA) Effectively Inhibits Retrovirus Replication *In Vitro* and Simian Immunodeficiency Virus Infection in Rhesus Monkeys. *AIDS* **1991**, *5*, 21-28.

(18) Cools, M.; Balzarini, J.; De Clercq, E. Mechanism of Antiviral and Cytotoxic Action of (±)-6′-β-fluoroaristeromycin, a Potent Inhibitor of S-Adenosyl-homocysteine Hydrolase. *Mol. Pharmacol.* **1991**, *39*, 718-724.

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