4',1'a-METHANOCARBOCYCLIC ADENOSINE ANALOGUES AS POTENTIAL INHIBITORS OF *S*-ADENOSYLHOMOCYSTEINE HYDROLASE

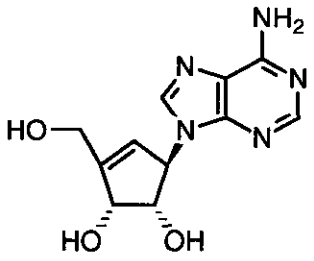
Lak S. Jeong,[1] Victor E. Marquez,[1(*)] Chong-Shen Yuan,[2] and Ronald T. Borchardt[2]

[1]Laboratory of Medicinal Chemistry, Developmental Therapeutics Program, Division of Cancer Treatment, National Cancer Institute, NIH, Bethesda, Maryland 20892, USA and [2]Department of Biochemistry, The University of Kansas, Lawrence, Kansas 66045, USA

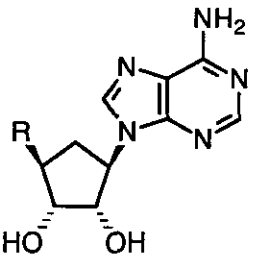
Abstract –Cyclopropane-fused carbocyclic adenosine (**4**) and its 5'-carboxaldehyde analogue (**5**) were synthesized as potential inhibitors of *S*-adenosylhomocysteine hydrolase. The key bicyclo[3.1.0]hexane alcohol (**8**) was obtained from the optically pure cyclopentenone synthon (**6**), and attachment of the purine base was performed in a single step from the methylsulfonate ester of **8** (compound (**9**)). Both target compounds behaved as weak inhibitors of the hydrolase.

S-Adenosylhomocysteine (AdoHcy) hydrolase catalyzes the interconversion of AdoHcy into adenosine and homocysteine and plays an important role in regulating *S*-adenosyl-L-methionine (AdoMet)-dependent methyl transferase activity.[1] It has been established that the accumulation of AdoHcy levels that result from AdoHcy hydrolase inhibition correlates well with the antiviral activity of these inhibitors, and hence, the development of more selective and powerful inhibitors of this enzyme continues to be an attractive goal for chemotherapeutic purposes.[2,3]

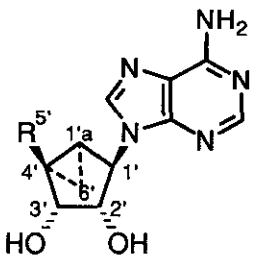
Neplanocin A (**1**) and aristeromycin (**2**) are the paradigm inhibitors of AdoHcy hydrolase.[4-6] Therefore, we decided to investigate the effects of fusing a cyclopropane ring at the site of the double bond in neplanocin A, which essentially transforms the molecule into an aristeromycin analogue (**4**) with a rigid carbasugar moiety possessing a North-type form or ring pucker,[7,8] as defined in the pseudorotational cycle.[9] Since both adenosine-5'-carboxaldehyde[10] and aristeromycin-5'-carboxaldehyde (**3**)[11] are similarly



Neplanocin A (**1**)



R = CH_2OH (Aristeromycin, **2**)
R = CHO (**3**)



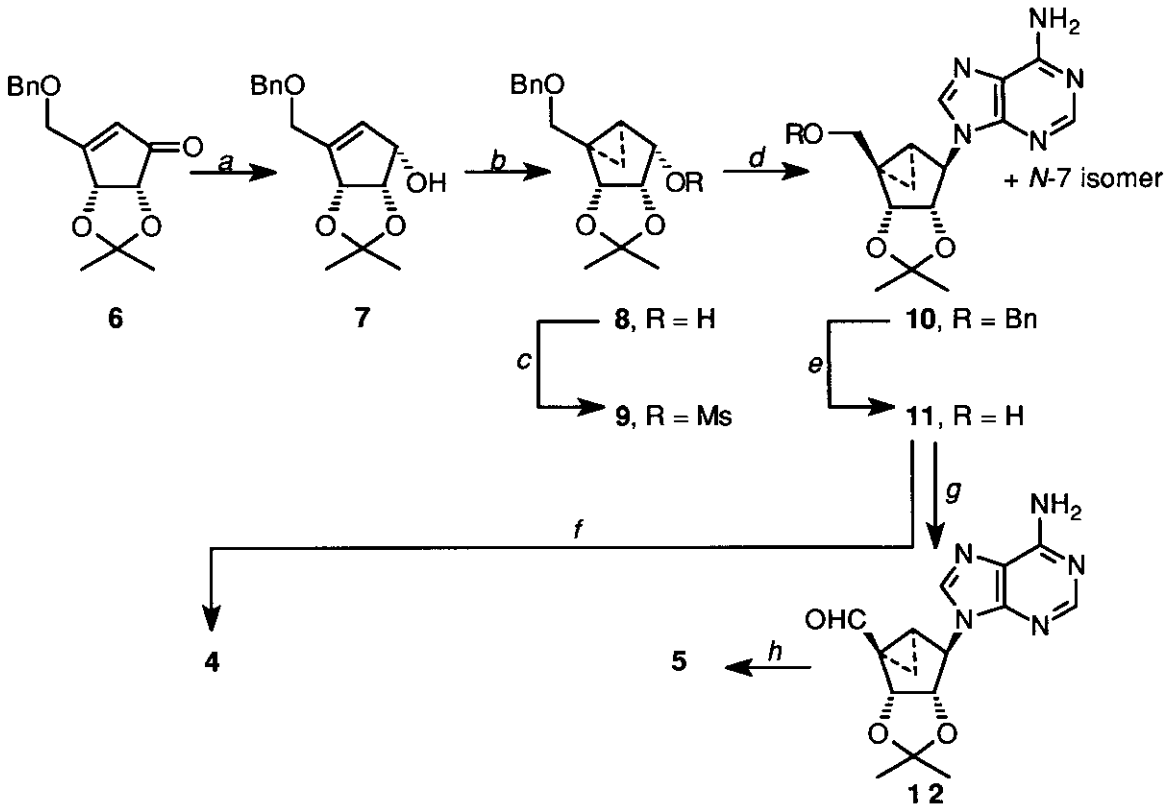
R = CH_2OH (**4**)
R = CHO (**5**)

potent inhibitor of *S*-AdoHcy hydrolase, the corresponding 5'-carboxyaldehyde (**5**) of the cyclopropane-fused aristeromycin analogue was also selected as a target.

Synthesis of compound (**4**) proceeded as shown in Scheme 1. Our readily available optically active cyclopentenone (**6**) was reduced to a single diastereomeric allylic alcohol (**7**) as previously reported.[12] Stereoselective Simmonds-Smith cyclopropanation of **7** using Et_2Zn and CH_2I_2 gave an excellent yield of the bicyclo[3.1.0]hexane alcohol (**8**),[8] which was readily converted to the mesylate (**9**) quantitatively. Condensation of **9** with adenine in the presence of K_2CO_3 and 18-crown-6 ether gave the desired *N*-9 product (**10**) as the major component (68%) together with a small amount of the *N*-7 analogue (8%). Deprotection of the 5'-*O*-benzyl ether using palladium black and formic acid, followed by removal of the isopropylidene group with 80% acetic acid, gave target compound (**4**).[13] The primary alcohol function in **11** was oxidized to the aldehyde (**12**) using tetrapropylammonium perruthenate (TPAP), and deprotection of the isopropylidene function afforded the second target compound (**5**).[14]

The compounds were evaluated as inhibitors of *S*-AdoHcy hydrolase using pure recombinant human placental enzyme (Table 1). Compounds (**4**) and (**5**) were preincubated with the enzyme at various concentrations ranging from 0.1 to 100 μM for 10 min at 37 °C. The residual enzyme activity was then measured by incubation of the enzyme for 5 min in the presence of 100 μM of [2,8-^{3}H]AdoHcy and 4 units of adenosine deaminase. A known potent inhibitor of the enzyme, 9-(*trans*-2',*trans*-3'-dihydroxycyclopent-4'-enyl)adenine (**13**),[15] was included as a control. Both **4** and **5** behaved as extremely weak

Scheme 1.



Reagents and Conditions. [*a*] Ref. 12, [*b*] Et_2Zn, CH_2I_2, CH_2Cl_2, 0 °C to room temperature, 15 h (90%), [*c*] MsCl, pyridine, 0 °C, 4 h (100%), [*d*] adenine, K_2CO_3, 18-crown-6, DMF, 100 °C, 18 h (68%), [*e*] Pd black, HCO_2H, MeOH, room temperature, 24 h (75%), [*f*] 80% AcOH, 80 °C, 5 h (80%), [*g*] TPAP, CH_3CN, CH_2Cl_2, room temperature, 30 h (60%), [*h*] 88% HCO_2H, room temperature, 5 h followed by neutralization with aq. 29% NH_4OH, room temperature, 10 min (90%).

inhibitors of the enzyme (30-40% inhibition at 100 μM). This poor inhibition of AdoHcy hydrolase shown by compound (**4**) is similar to that demonstrated earlier for the structurally related neplanocin C, the 6'-oxa-bicyclo[3.1.0]hexane analogue of **4**,[6] which suggests that the conformation of the ring,[7] and not the presence or absence of the oxygen, is responsible for the loss of activity. In fact, the *S*-

AdoHcy hydrolase inhibitory activity of neplanocin C assayed in monolayers of *L929* cells at 1 μM concentration was very similar (*ca.* 18% inhibition) to that achieved by compound (**4**) with the cloned enzyme at the same concentration.[6] It was surprising, however, that the 5'-carboxaldehyde analogue (**5**) was even weaker than **4**, in view of the potent activity of aristeromycin-5'-carboxaldehyde.[11] It can be tentatively concluded that either the bulk of the cyclopropane ring prevents the molecule from interacting efficiently with the enzyme, or that the rigid North-conformation induced by the bicyclo[3.1.0]hexane skeleton[7,8] is inadequate for good substrate recognition. In order to test the latter hypothesis, the synthesis of the corresponding 1',1'a-methano carbocyclic adenosine analogues which would have a South-conformation is being actively pursued in this laboratory.

Table 1. Concentration-dependent inhibition of AdoHcy hydrolase by **4**, **5**, and reference compound (**13**).

		% Activity Remaining				
Compounds	(μM)	0.1	1	10	50	100
4		94.3	80.5	71.9	63.2	60.3
5		92.5	81.5	76.0	73.7	70.6
13		34.2	3.1	1.7	—	1.5

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13. Compound (**4**); white solid, mp 130 °C (decomp.); $[\alpha]_D^{25} = -28.0°$ (c 0.10, MeOH); uv (H_2O, pH 7) λ_{max} 260.8 nm (ε 16,550), uv (H_2O, pH 2) λ_{max} 259.0 nm (ε 16,250), uv (H_2O, pH 11) λ_{max} 261.8 nm (ε 24,990); ^{1}H nmr (DMSO-d_6) δ 8.41 (s, 1 H, H-8), 8.12 (s, 1 H, H-2), 7.23 (br s, 2 H, NH_2), 5.19 (br s, 1 H, OH), 5.09 (br s, 1 H, OH), 4.72 (s, 1 H, H-1'), 4.55 (d, *J* = 6.2 Hz, 1 H, H-3'), 4.50 (br s, 1 H, OH), 4.08 (d, *J* = 11.2 Hz, 1 H, H-5'), 3.65 (d, *J* = 5.9 Hz, 1 H, H-2'), 3.13 (d, *J* = 11.2 Hz, 1 H, H-5"), 1.45 (dd, *J* = 8.5, 3.3 Hz, 1 H, H-6'), 1.35 (t, *J* = 4.4 Hz, 1 H, H-1'a), 0.61 (dd, *J* = 8.5, 4.7 Hz, 1 H, H-6"); ^{13}C nmr (DMSO-d_6) δ 156.0, 152.4, 148.9,

138.8, 118.8, 76.1, 70.2, 62.3, 60.8, 36.5, 23.1, 11.5; FAB MS *m/z* 278 (MH+). Anal. Calcd $C_{12}H_{15}N_5O_3 \cdot 0.5H_2O$: C, 50.35; H, 5.63; N, 24.46. Found: C, 50.24; H, 5.61; N, 24.36.

14. Compound (**5**); hygroscopic white solid, mp 134 °C (decomp.); $[\alpha]_D^{25} = -20.0$ ° (c 0.10, MeOH); uv (H_2O, pH 7) λ_{max} 261.0 nm (ε 36,500), uv (H_2O, pH 2) λ_{max} 258.8 nm (ε 24,480), uv (H_2O, pH 11) λ_{max} 262.6 nm (ε 39,520); ^{1}H nmr (D_2O) δ 8.91 (s, 1 H, CHO), 8.02 (s, 1 H, H-8), 7.88 (s, 1 H, H-2), 4.95 (d, *J* = 6.4 Hz, 1 H, H-3'), 4.65 (d, *J* = 2.6 Hz, 1 H, H-1'), 4.11 (dd, *J* = 6.3, 3.0 Hz, 1 H, H-2'), 2.42 (dd, *J* = 9.0, 5.4 Hz, 1 H, H-6'), 1.99 (t, *J* = 5.6 Hz, 1 H, H-1'a), 1.77 (dd, *J* = 9.1, 6.1 Hz, 1 H, H-6"); ^{13}C nmr (D_2O+MeOH-d_4) δ 202.2, 155.5, 152.5, 148.7, 139.9, 118.6, 76.8, 67.5, 61.1, 45.6, 30.0, 17.2; FAB MS *m/z* 276 (MH+). Anal. Calcd $C_{12}H_{13}N_5O_3 \cdot 1.9H_2O$: C, 46.57; H, 5.47; N, 22.63. Found: C, 46.78; H, 5.15; N, 22.36.

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Received, 27th July, 1995