reaction mixture was cooled and most of the ethanol was removed in vacuo. The residue was diluted with 100 mL of water and the pH adjusted to 6 by careful addition of 2 N hydrochloric acid. Extraction with chloroform (2 × 300 mL) followed by drying the extracts over anhydrous sodium sulfate and evaporation of the solvent in vacuo afforded a nearly colorless crystalline solid. Recrystallization from ethanol gave 6.16 g (78%) of fine colorless crystals, mp 185–188 °C. Properties of **6**, and of 11, 12, and 13 prepared in similar manner, are included in Table I.

Tetrabenazine Assay. The assay was conducted and the data were analyzed as described in ref 1.

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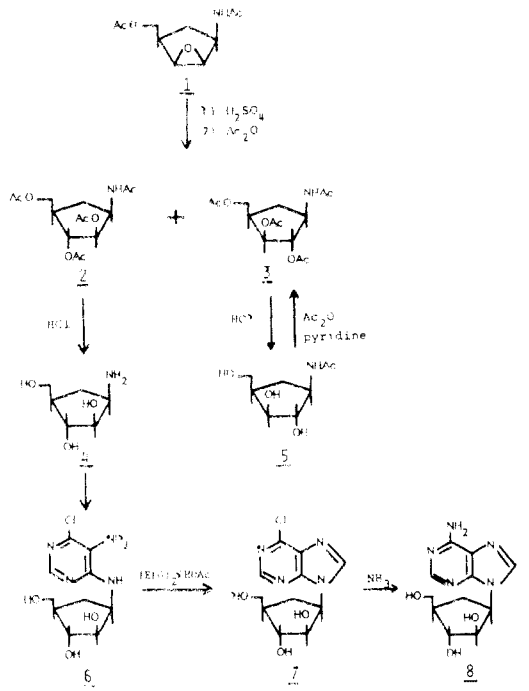
Communications to the Editor

Carbocyclic Arabinosyladenine, an Adenosine Deaminase Resistant Antiviral Agent

Sir:

The antiviral nucleoside 9-β-D-arabinofuranosyladenine (*ara*-A) was first synthesized in a program designed to produce anticancer agents.[1] Recent interest in the promising antiviral activity of *ara*-A has been extensively reviewed.[2-4] Broad spectrum activity of *ara*-A against DNA viruses[5] and significant therapeutic activity of *ara*-A against experimental herpes simplex keratitis and herpes simplex and vaccinial encephalitis have been reported.[2] A major liability in the use of *ara*-A lies in the fact that the nucleoside is rapidly deaminated by a commonly occurring enzyme, adenosine deaminase.[5,6] Although the deamination product, 9-β-D-arabinofuranosylhypoxanthine (*ara*-H), is also active against DNA viruses, it is considerably less active than *ara*-A.[7] A major effort to circumvent the deamination problem employs the use of *ara*-A in combination with adenosine deaminase inhibitors[8-11] such as deoxycoformycin or *erythro*-9-(2-hydroxy-3-nonyl)adenine.[12] A more desirable approach to the development of a more active antiviral or antitumor agent would involve the use of a deamination resistant *ara*-A derivative.

We report here the synthesis of carbocyclic arabinosyladenine (*C-ara*-A), an adenosine deaminase resistant *ara*-A analogue with in vitro antiviral and antitumor activity. Hydrolysis of the easily synthesized epoxide 1[13] (2% H_2SO_4, 100 °C, 1 h) and subsequent acetylation gave a mixture of **2** and **3**. The major isomer, (±)-4α-acetamido-2β,3α-diacetoxy-1α-cyclopentanemethyl acetate (**2**), was separated from the mixture with one crystallization as colorless prisms (53% from EtOAc, mp 137–137.5 °C). Anal. ($C_{14}H_{21}NO_7$) C, H, N. When **2** was subjected to mild acidic hydrolysis (2 N HCl, 70 °C, 1 h), amine **4** was formed, since acyl migration to the adjacent *cis*-hydroxyl facilitates hydrolysis of the acetamide.[13] Subjection of a mixture of **2** and **3** to the same hydrolysis conditions gave a mixture of amine **4** and acetamide **5**. This mixture was separated by passage through an IRA-120(H^+) resin. Reacetylation of **5** gave pure **3** (14% from 1) as a colorless syrup. Anal. ($C_{14}H_{21}NO_7$) C, H, N. Amine **4**, a hygroscopic gum, was immediately condensed with 5-amino-4,6-dichloropyrimidine, giving 5-amino-4-*N*-[2α,3β-dihydroxy-4α-(hydroxymethyl)cyclopent-1α-yl]amino-6-chloropyrimidine (**6**) as a white powder (72% from ethanol, mp 184–186 °C). Anal. ($C_{10}H_{15}N_4O_3Cl$) C, H, N, Cl. Ring closure of **6** with diethoxymethyl acetate gave the 6-chloropurine **7** as white granules (72% from ethanol, mp 212–214 °C dec). Anal. ($C_{11}H_{13}N_4O_3Cl$) C, H, N, Cl. Treatment of **7** with liquid ammonia gave the desired (±)-9-[2α,3β-dihydroxy-4α-(hydroxymethyl)cyclopent-1α-yl]adenine (*C-ara*-A) (**8**) as a white powder [76% from water; mp 252.5–254.5 °C dec; UV max in nm (ε × 10^{-3}) (0.1 N HCl) 258.5 (14.8), 210 sh (21.3); (H_2O) 260 (15.4); (0.1 N NaOH) 260 (15.5)]. Anal. ($C_{11}H_{15}N_5O_3 \cdot H_2O$) C, H, N. A more detailed presentation of the chemistry of these compounds and related derivatives will be published.[14]



The cytotoxicity of *C-ara*-A was evaluated by growing P-388 mouse lymphoid leukemia cells in the presence of either **8** or *ara*-A using a method previously described.[15] Both *ara*-A and *C-ara*-A exhibited LD_{50} concentrations of

Table I. In Vitro Antiviral Activity of Carbocyclic *ara*-A

Challenge virus	Virus rating (VR)[a]	MED_{50},[b] μg/mL
Herpes simplex virus, type 1	2.2	9.0
	3.5	2.8
Vaccinia virus	1.5	9.0
	1.7	9.0

[a] Virus rating (VR): a weighted measurement of antiviral activity, based on the in vitro inhibition of virus-induced cytopathogenic effects (cpe) and the cytotoxicity exhibited by the drug, determined by a modification of the method of Ehrlich et al.[16] (see text). A VR ≥ 1.0 indicates definite (+) antiviral activity; a VR of 0.5–0.9 indicates marginal to moderate (±) antiviral activity; and a VR <0.5 indicates no (−) apparent antiviral activity. [b] Minimum effective dose, 50% (MED_{50}): the minimum drug dose required for 50% inhibition of virus-induced cpe.

1×10^{-5} M. In contrast to *ara*-A, the carbocyclic analogue **8** is completely resistant to deamination by adenosine deaminase. Thus, under conditions in which *ara*-A is completely deaminated (1 μmol/min per unit of enzyme) by calf intestinal adenosine deaminase (type III, Sigma), no detectable deamination of **8** was observed. In addition, *C-ara*-A did not inhibit the enzymatic deamination of either *ara*-A or adenosine.

C-ara-A was examined by Shannon and Arnett (Southern Research Institute, Birmingham, Ala.) for in vitro antiviral activity against two representative DNA-containing animal viruses by the quantitative determination of its ability to inhibit virus-induced cytopathogenic effects (cpe) in infected cultures. The viruses employed in these assays were herpes simplex virus (HSV) type 1 (strain HF) and vaccinia virus (VV) (Strain Lederle Chorioallantoic). Both viruses were propagated and assayed for infectivity in continuous-passage human epidermoid carcinoma of the larynx (H.Ep.-2) cells. A virus rating (VR) was calculated for the activity of *C-ara*-A against each virus by the use of a modification of the method of Ehrlich et al.[16] previously described by Sidwell et al.,[17] except that triplicate cultures rather than duplicate cultures were employed for each assay. The results are shown in Table I. As can be seen, the carbocyclic analogue of *ara*-A demonstrated highly significant antiviral activity against HSV and VV with VR's ranging from 1.5 to 3.5. The approximate MED_{50} for *C-ara*-A appears to be about 9 μg/mL, a concentration which is noncytotoxic to the host cells.

The easy accessibility of this novel arabinosyl nucleoside analogue, in addition to such desirable features as hydrolytic stability, adenosine deaminase resistance, and significant antiviral activity, makes *C-ara*-A an excellent candidate for detailed evaluation as a chemotherapeutic agent. Further work is continuing in this laboratory and elsewhere to study the metabolism, antiviral spectrum, and therapeutic effects of *C-ara*-A in animals.

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