somatostatin on inhibition of insulin release in vivo. It is, however, less potent than somatostatin on inhibition of glucagon release. Des-Asn[5]-[D-Trp[8]]somatostatin is thus the first analogue of somatostatin with high potency and dissociated biological activity.

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Nucleic Acids. 16. Orally Active Derivatives of *ara*-Cytidine[1,2]

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Water-soluble derivatives of *ara*-cytidine (cytarabine, Cytosar) were prepared and tested for antitumor, immunosuppressive, and antiarthritic activities in animals after oral administration. The compounds tested included the 5′-palmitate, 5′-benzoate, and 5′-adamantoate esters of *ara*-cytidine, made water soluble by use of their hydrochloride salts or peptidyl derivatives, and two basic 5′ esters (5′-nicotinoate and 5′-quinuclidinate) as their hydrochloride salts. Five of the compounds had antitumor activity superior to that found with *ara*-cytidine itself after oral administration in the L1210 leukemic mouse assay. One of these, 5′-adamantoyl-*ara*-cytidine hydrochloride, had antitumor activity after oral administration approaching that achieved with parenterally administered *ara*-cytidine.

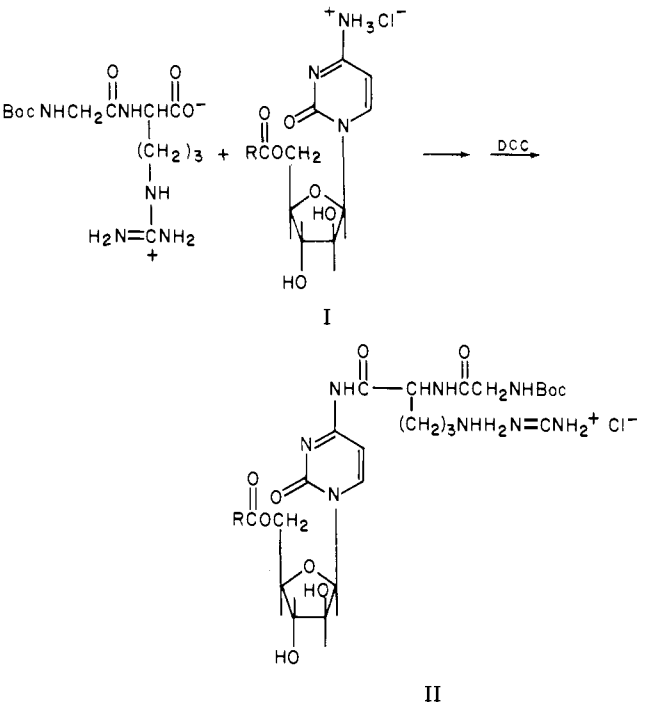
In previous papers[1] we described the systematic modification of relatively insoluble 5′-acylates (esters) of the antitumor, antiviral, immunosuppressive nucleoside, *ara*-cytidine (cytarabine, Cytosar), in order to adjust predictably the nucleoside's pharmacologic properties. By these structural modifications we were able to affect the solubility, the enzymatic hydrolysis of the ester to the active nucleoside, and the catabolic conversion to *ara*-uridine and thus increase the depot activity. This paper describes results of our modifications of the most biologically interesting of these 5′-esters in an attempt to prepare a derivative that will produce as much activity in man when administered orally as *ara*-cytidine administered parenterally.

Orally administered *ara*-cytidine has not been shown to be active in man, although in mice it is about one-fifth as active when administered orally as when given parenterally.[3] Pharmacokinetic analyses suggest low rate and extent of absorption and rapid elimination (deamination and excretion) so that plasma levels are never sufficient to produce therapeutic activity.[4]

Desirable Properties. Ideally, an orally active derivative of *ara*-cytidine should have adequate dissolution and absorption rates in the gastrointestinal tract and resistance to deamination by pyrimidine aminohydrolase found in the gut and other tissues. If the drug, once absorbed, is rapidly localized in lipid depots, it should be protected from deamination. Diffusion from the depot sites should maintain therapeutic blood levels. Finally, the substituent groups used to confer these properties upon the compound should be subject to rapid removal in vivo[1] to release *ara*-cytidine for subsequent conversion to *ara*-CTP, the active form of the drug.[5]

The presence of an appropriate ester group at the 5′ position achieves localization of the drug at the site of parenteral administration and/or in lipid depots. It also prevents deamination. However, such esters are only sparingly soluble in water and, apparently because of slow dissolution, produce only minimal activity when administered orally in aqueous suspensions. It occurred to us that they might be made adequately soluble by forming acid addition salts of the esters or by attaching a suitable peptidyl substituent at N^4. The covalently bonded substituents could then be removed enzymatically in vivo. Our earlier results with N^4-amide derivatives of *ara*-cytidine suggested that such derivatives were inactive,[6,7] but these results were from in vitro experiments, which have not quantitatively predicted in vivo activity[1,8]—presumably because the systems lack the requisite amidase activity. We reasoned that if a substituent placed on the N^4-amino group were an amino acid or peptide, hydrolytic enzymes ubiquitous in vivo would convert it to the free amine at N^4. This should be especially likely if the amino acid linked to N^4 were lysine or arginine, because trypsin or trypsin-like enzymes capable of splitting lysyl and arginylamido bonds are found in the gut, blood, and tissues.

Scheme I



For our studies we chose the *tert*-butoxycarbonyl-glycyl-L-arginyl derivatives (II in Scheme I) in which the carboxyl group of arginine is attached to the N^4-amino group of cytosine. We thought that combining a glycine residue with a protonated arginine residue would confer water solubility on the derivative, while the arginylamido

Table I. Oral Antitumor Activity (L1210 Mouse Leukemia) of *ara*-Cytidine Derivatives

Compd no.	Derivative[b]	Mol wt	Route of admin	Vehicle[c]	% increase in life-span (% ILS)[a] 100 μmol/kg/day	200 μmol/kg/day	400 μmol/kg/day
1	*ara*-Cytidine (parent)	243	Oral	A	15	38	62
2	Palmitate	518	Oral	C	46	69	92
3	Benzoate	384	Oral	B	15	31	62
4	Adamantoate	442	Oral	D	46	77	130
5	Peptidyl	618	Oral	A	0	14	14
6	Peptidyl palmitate	831	Oral	A	30[d]	44[d]	110[d]
7	Peptidyl benzoate	697	Oral	A	14	21	43
8	Peptidyl adamantoate	755	Oral	A	21	43	93
9	Nicotinoate	381	Oral	C	31	46	69
10	Quinuclidinate	389	Oral	B	8	0	15
1 + THU	*ara*-Cytidine + THU[e]	243	Oral	A	62	92	138
1	*ara*-Cytidine	243	Intraperitoneal	A	69	100	131

[a] Calculated from median survival times of treated and control groups. [b] Compound 1 was given as the free base, compounds 2-9 as hydrochloride salts, and compound 10 as the dihydrochloride salt. [c] Vehicle A = 0.9% (w/v) aqueous NaCl (saline). Vehicle B = saline. Vehicle C = 0.01 N HCl. Vehicle D = 0.1 N HCl. Vehicles B, C, and D: before the material was added to the vehicle it was wetted with a few drops of ethanol, <5% v/v. [d] Average of two experiments. [e] THU was administered orally at a dosage of 10 mg/kg/day on days 1-5.

bond would be split in vivo.

Methods. Synthesis of Peptidyl Derivatives. The N^4-*tert*-butoxycarbonylglycyl-L-arginyl derivatives of 5′-palmitoyl-, 5′-benzoyl-, and 5′-adamantoyl-*ara*-cytidine esters were prepared. Synthesis was accomplished by coupling the *tert*-butoxycarbonylglycyl-L-arginine with the hydrochloride of *ara*-cytidine using dicyclohexylcarbodiimide (DCC) as the coupling agent. Use of the hydrochloride salts was necessary to provide for protonation of the strongly basic guanido group of arginine. Details of the synthesis are in the Experimental Section.

Biological Test Systems. Ten compounds listed in Table I were tested for antitumor activity when given orally to L1210 leukemic mice.[1] Activity of the orally administered compounds was compared to that of *ara*-cytidine given intraperitoneally. Mice were inoculated intraperitoneally on day 0 with 1×10^6 L1210 cells per mouse. Treatment was started 24 h later and continued daily for 5 days in the test groups of eight animals each. Survival time was determined to the nearest day. Percent increase in life-span (% ILS) was calculated from median survival times of the control group and treated groups. In this test a compound is considered to have significant antitumor activity if it increases life-span 25% or more.

The compounds evaluated for antitumor activity after oral administration were *ara*-cytidine (compound 1); the palmitate **2**, benzoate **3**, and adamantoate **4** esters; the peptidyl derivative **5** of *ara*-cytidine; the peptidyl derivatives **6**–**8** of the palmitate, benzoate, and adamantoate esters; and two water-soluble esters, 5′-nicotinoate **9** and 5′-quinuclidinate **10**.[1] *ara*-Cytidine was used as the free base; the quinuclidinate was a dihydrochloride salt; and the other compounds were monohydrochloride salts. *ara*-Cytidine was also given in combination with the deaminase inhibitor, tetrahydrouridine (THU).[9] All agents were in solution; vehicles are indicated in Table I. Dosages were given on a μmol/kg basis because the molecular weights of the individual compounds cover a very broad range (243–831 daltons; see Table I). The intraperitoneal injection volume was 0.2 ml. In an experiment not indicated in Table I, the palmitate ester **2** was given orally as a suspension of the free base in aqueous methylcellulose, 200 μmol/kg/day.

The compounds were also tested for immunosuppressive activity in a standard hemagglutinin assay in the mouse.[6] The dosage was 400 μmol/kg/day given on days 1–5. Other assays were the experimental allergic encephalomyelitis (EAE) system in the rat;[10] adjuvant-induced polyarthritis in the rat;[11] and acute inflammation (hindpaw edema) in the rat.[11] In these three assays dosage was 200 μmol/kg/day, and the vehicle was 0.01 N HCl. Animals were dosed on days 1–13 in the EAE assay and twice daily on days 0–14 in the other two assays.

Results

Properties of the Peptidyl Derivatives. Yields of the N^4-*tert*-butoxycarbonylglycyl-L-arginyl derivatives of 5′-palmitoyl-, 5′-benzoyl-, and 5′-adamantoyl-*ara*-cytidine, after purification by chromatography, varied from 25 to 35%. The hydrochloride salts of all three derivatives were water soluble to at least the extent of 25 mg/ml in saline solution. Removal of the *tert*-butoxy protecting groups did not afford easily characterizable compounds of high water solubility. Consequently, the additional protecting group was retained.

Biological Activity. Table I shows the antitumor activity achieved by each compound tested in the L1210 mouse leukemia assay. The palmitate ester given as a suspension of free base, 200 μmol/kg/day, produced a 39% increase in life-span. Figure 1 shows dose–response curves for seven compounds. Figure 2 indicates the relative antitumor activity of ten compounds and also compares results in the immunosuppression (mouse hemagglutinin) assay with those in the antitumor assay for one dosage level (400 μmol/kg/day given on days 1–5). Table II summarizes results in the EAE, polyarthritis, and acute inflammation assays.

Discussion and Conclusions

Of all the 5′-esters of *ara*-cytidine tested, the 5′-adamantoate ester (compound **4**) showed the greatest antitumor activity after oral administration, approaching the activity shown by parenterally administered *ara*-cytidine (1) or by the orally administered combination of *ara*-cytidine and the deaminase inhibitor THU (Table I, Figure 1). A disadvantage of the adamantoate is that it must be administered in 0.1 N HCl to keep it in solution. Thus it would be impractical for therapeutic use.

The adamantoate ester **4** and three other derivatives, peptidyl palmitate **6**, peptidyl adamantoate **8**, and pal-

Table II. Results in Immunoinflammatory Assays with Orally Administered *ara*-Cytidine Derivatives in the Rat

Derivative	Compd no.	Percent inhibition: EAE Day 14	EAE Day 17	Adjuvant-induced polyarthritis Day 15	Adjuvant-induced polyarthritis Day 21	Acute inflam
ara-Cytidine (parent)	1			20	7	−4
Peptidyl *ara*-cytidine	**5**	0	0	10	0	22
Benzoate	**3**	0	0	47	20	15
Peptidyl benzoate	7	13.5	5.3	52	35	9
Adamantoate	**4**	5.4	7.9	31	28	4
Peptidyl adamantoate	**8**	5.4	10.5	50	18	1
Palmitate	**2**	8.1	13.2	11	6	13
Peptidyl palmitate	**6**	35.1	10.5			6
Quinuclidinate	**10**			5	0	−2
Nicotinoate	9			0	0	4

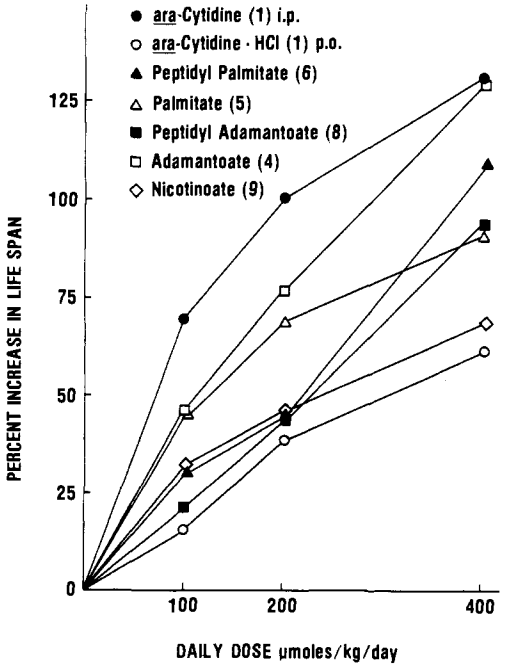


Figure 1. Dose-response curves of antitumor activity (L1210 mouse leukemia) for orally administered compounds (1, 4-6, 9) and parenterally administered *ara*-cytidine.

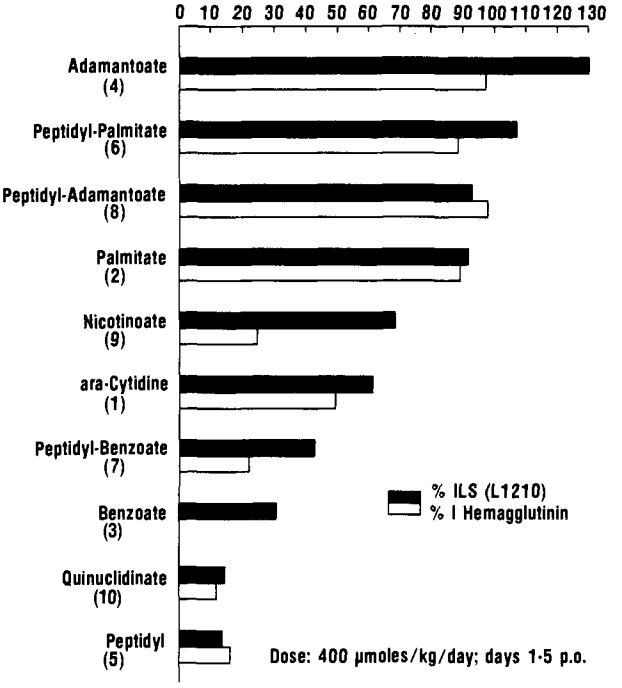


Figure 2. Comparison of antitumor activity (% ILS in the L1210 leukemic mouse) with immunosuppression (percent inhibition of hemagglutinin formation in the mouse).

mitate 2, were more active than *ara*-cytidine hydrochloride itself given orally in the antitumor and immunosuppression assays and ranked roughly in that order (Figures 1 and 2). At one dosage level (200 μmol/kg/day), the palmitate ester 2 given as a suspension of the free base showed more antitumor activity (39% ILS) than the peptidyl palmitate (30% ILS) but less than a solution of the hydrochloride salt of the ester (46% ILS).

The nicotinoate 9 appeared to be slightly more active than oral *ara*-cytidine, while the benzoate, either as the hydrochloride salt 3 or the peptidyl derivative 7, was less effective (Table I). The quinuclidinate 10 and the peptidyl derivative 5 of *ara*-cytidine itself were essentially inactive when given orally.

As was found with parenteral administration of the 5′-esters of *ara*-cytidine,[8] there was good correlation between the results of the antitumor assay and those of the hemagglutinin assay when the compounds were administered orally (Figure 2). The notable exceptions were that the nicotinoate 9 and peptidyl benzoate 7 derivatives failed to show significant immunosuppressive activity but exhibited considerable antitumor activity at the 400 μmol/kg/day dosage level.

At the dosages used, none of the derivatives given orally had much effect on experimental allergic encephalomyelitis in the rat (Table II). The peptidyl palmitate 6 did show minimal activity in this system. None of the compounds showed significant inhibitory effect on adjuvant-induced polyarthritis or acute inflammation in the rat.

The lack of activity in the rat EAE and arthritis systems suggests the potential of species specificity. This has already been alluded to in regard to the lack of activity of orally administered *ara*-cytidine in man. Species differences in absorption, distribution, metabolism, and excretion of the various derivatives greatly complicate the design and testing processes. For example, we have no good animal model for catabolism of *ara*-cytidine derivatives. Deamination occurs primarily in the liver in man but in the kidney in the mouse. The rat apparently has little deaminase activity, while the monkey—a logical candidate for testing—has even higher total levels of enzyme activity than man.[12]

The antitumor activity of one of the orally administered derivatives of *ara*-cytidine, the 5′-adamantoate hydrochloride, approaches that of parenterally administered *ara*-cytidine. However, we do not consider this or any of

the 5′ derivatives yet tested to be optimal agents. The vehicle (0.1 N HCl) required to dissolve the adamantoate hydrochloride is certainly impractical. Furthermore, while the antitumor activity of some of the esters given orally did not compare too badly with that of the parent compound given parenterally, it was nowhere near that observed when the 5′ derivatives themselves were given parenterally. The benzoate, adamantoate, palmitate, and other esters injected parenterally as suspensions of the free base all yielded much greater therapeutic effects than *ara*-cytidine itself given parenterally. The therapeutic value of these derivatives in humans remains to be demonstrated.

Experimental Section

The 5′-esters of *ara*-cytidine used in this work were prepared as previously described.[1] *tert*-Butoxyglycyl-L-arginine was obtained from Fox Chemical Co., Los Angeles, Calif. Commercial grade solvents were used without further purification.

Hydrochloride Salts of 5′-Esters of *ara*-Cytidine. The ester[1b] (0.1 mol) was dissolved in about 150 ml of MeOH with the aid of 10 ml of concentrated HCl. Ether was added to opalescence and crystallization was induced. Additional Et_2O was added to complete crystallization. The product was collected by filtration, washed with Et_2O, and dried in vacuo.

5′-Palmitoyl-*ara*-cytidine Hydrochloride (2). The yield was 90%, mp 171–172°. Anal. ($C_{25}H_{43}N_3O_6$·HCl) H, N, Cl; C: calcd, 57.95; found, 58.34.

5′-Benzoyl-*ara*-cytidine Hydrochloride (3). The yield was 97%, mp 200–201°. Anal. ($C_{16}H_{17}N_3O_6$·HCl) C, H, N; Cl: calcd, 9.24; found, 9.68.

5′-(1-Adamantoyl)-*ara*-cytidine Hydrochloride (4). The yield was 89%, mp 247° dec. Anal. ($C_{20}H_{27}N_3O_6$·HCl) C, H, N, Cl.

N^4-(*tert*-Butoxycarbonylglycyl-L-arginyl)-*ara*-cytidine Acetate Salt (5). *ara*-Cytidine hydrochloride (112 g, 0.4 mol) and 132 g (0.4 mol) of *tert*-butoxycarbonylglycyl-L-arginine were dissolved in 1 l. of dimethylformamide (DMF), the mixture was cooled to 5°, and 165 g (0.8 mol) of dicyclohexylcarbodiimide (DCC) was added. The mixture was stirred at 5° for 72 h. The dicyclohexylurea was removed by filtration and the solvent was evaporated in vacuo. Partial purification of the product was accomplished by chromatography over silica gel (Merck-Darmstadt, 0.05–0.2 mm) using methyl ethyl ketone–Me_2CO–H_2O (72:20:8) as solvent. The column (4 in. i.d.) was loaded with 11 kg of silica gel which had been equilibrated with solvent. After 40 l. of effluent, 2-l. fractions were collected. The product, along with some *ara*-cytidine, emerged in fractions 13–31. The material (52.7 g) was recovered by evaporation of the solvent in vacuo. Final purification was accomplished by continuous flow electrophoresis, using the Hannig Model FF electrophoretic separator. The experimental procedure used was similar to that described by Ko et al.[13] The sweeping buffer employed (pH 7) was made up by diluting 270 ml of 2,4-lutidine and 33 ml of glacial AcOH to 30 l. with H_2O. In a typical run, 8 g of the partially purified product above was dissolved in 200 ml of H_2O and the pH was adjusted to 7.0. The electrophoresis was conducted at 1700 V and 100 mA, with a dosing rate of 6–8 ml/h. The separation was monitored by thin-layer chromatography (silica gel, MEK–Me_2CO–H_2O, 65:20:15). The purified product was recovered by lyophilization of appropriate fractions: recovery, 19.3 g (8% of theory). The NMR spectrum was completely consistent with the assigned structure. Anal. ($C_{22}H_{36}N_8O_9$·CH_3COOH) H, N; C: calcd, 46.75; found, 45.27.

5′-Palmitoyl-N^4-(*tert*-butoxycarbonylglycyl-L-arginyl)-*ara*-cytidine Hydrochloride (6). 5′-Palmitoyl-*ara*-cytidine hydrochloride (15.5 g, 0.03 mol) was dissolved in 100 ml of dimethylacetamide (DMA), *tert*-butoxycarbonylglycyl-L-arginine (9.9 g, 0.03 mol) was added, the solution was cooled in an ice bath, and 7.4 g (20% excess) of DCC was added. The mixture was stirred in the cold for 90 h. The dicyclohexylurea was removed by filtration and washed with DMA (added to filtrate). An equal volume of H_2O was added, followed by 1 l. of EtOAc. A crystalline solid which separated was removed by filtration and washed with a mixture of DMA, EtOAc and H_2O, then EtOAc, and finally with Et_2O: wt, 4.3 g; identified by TLC (silica gel G, MEK–Me_2CO–H_2O, 65:20:15) as 5′-palmitoyl-*ara*-cytidine. The EtOAc–H_2O phases above were separated and the organic phase was extracted with 200 ml of H_2O. This caused separation of additional 5′-palmitoyl ester: wt, 1.9 g. The combined aqueous extracts were concentrated in vacuo to leave an oil: wt, 20 g.

TLC (as above) indicated that the product, which absorbs uv light and gives a positive Sakaguchi test for arginine, was contaminated with unreacted 5′-palmitoyl-*ara*-cytidine and *tert*-butoxycarbonylglycylarginine. The peptidyl derivative was purified by chromatography over silica gel (Merck-Darmstadt, 0.05–0.2 mm). The column (45 mm × 115 cm) contained 800 g of silica gel, and the solvent system used was MEK–Me_2CO–H_2O (72:20:8). The material (about 18 g of the crude product above) was dissolved in 50 ml of the solvent and absorbed to the column. After the column had been eluted with 500 ml of solvent, 25-ml fractions were collected. The pure product (as determined by TLC) emerged in fractions 71–160 and was recovered by evaporation in vacuo of the solvent to a low volume, followed by lyophilization. The pure product, 5′-palmitoyl-N^4-(*tert*-butoxycarbonylglycyl-L-arginyl)-*ara*-cytidine hydrochloride, amounted to 9.0 g (36%): $[\alpha]^{25}D$ +62° (*c* 1, H_2O); ir spectrum ν_{max}^{mull} 3340, 3160, 1720, 1650, 1560, 1310, 1250, 1165, 1105, 1070, 1050, 940, 865, 805. The NMR spectrum was consistent with the proposed structure. Anal. ($C_{38}H_{66}N_8O_{10}$·HCl) C, H, N, Cl.

5′-Benzoyl-N^4-(*tert*-butoxycarbonylglycyl-L-arginyl)-*ara*-cytidine Hydrochloride (7). This compound was prepared and purified essentially as described for the palmitoyl ester above. The yield of purified product was 25%: $[\alpha]^{25}D$ +36° (*c* 0.9, H_2O); uv spectrum $\nu_{max}^{H_2O}$ 216 nm (ε 17 200), 236 (17 000), 282 (5100), and 300 (5450); ir spectrum ν_{max}^{mull} 3310, 3260, 3160, 1720, 1655, 1560, 1490, 1310, 1275, 1250, 1165, 1100, 1070, 1050, 1025, 800, 715. The NMR spectrum was reasonable for the proposed structure. Anal. ($C_{29}H_{40}N_8O_{10}$·HCl) C, H, N; Cl: calcd, 5.09; found, 5.69.

5′-(1-Adamantoyl)-N^4-(*tert*-butoxycarbonylglycyl-L-arginyl)cytarabine Hydrochloride (U-33 091A, 8). This derivative was prepared and purified essentially as described above for the palmitoyl ester. The yield of purified product was 24%: $[\alpha]^{25}D$ +53° (c 0.95, H_2O); uv spectrum $\nu_{max}^{H_2O}$ sh 213 nm (ε 17 200), 247 (12 550), and 301 (7150); ir spectrum ν_{max}^{mull} 3330, 3320 sh, 1720, 1660, 1565, 1495, 1310, 1275, 1235, 1170, 1105, 1070, 805. The NMR spectrum was reasonable for the proposed structure. Anal. ($C_{33}H_{50}N_8O_{10}$·HCl) C, H, N; Cl: calcd, 6.81; found, 7.37.

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Synthesis of Tetrazole Ribonucleosides and Their Evaluation as Antiviral Agents. 2.[1] 5-Amino-1-(β-D-ribofuranosyl)-1*H*-tetrazole and 5-Amino-2-(β-D-ribofuranosyl)-2*H*-tetrazole

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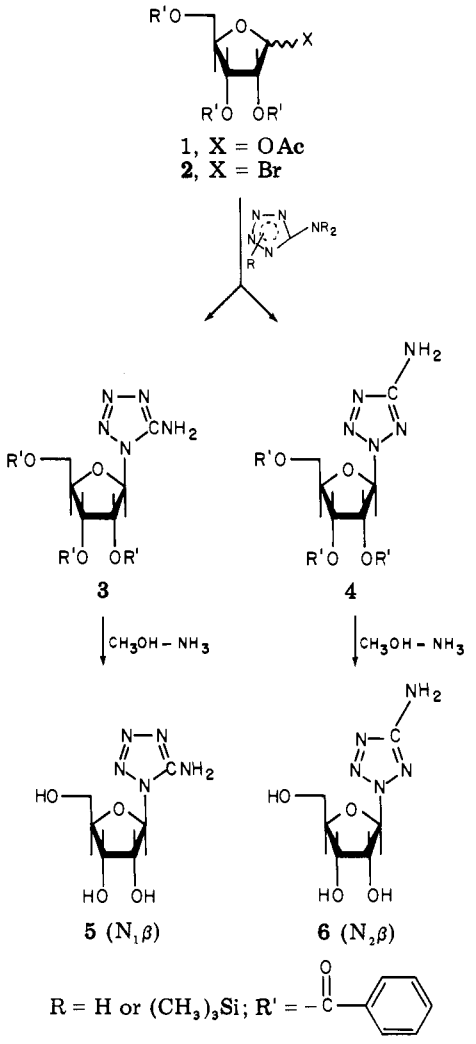
Synthesis of 5-amino-1-(β-D-ribofuranosyl)-1*H*-tetrazole and 5-amino-2-(β-D-ribofuranosyl)-2*H*-tetrazole is described. X-Ray crystallography was first used to establish the stereochemical configuration of the two isomers. By conducting ^{13}C NMR analysis on these isomers with known structures, i.e., $N_1\beta$ and $N_2\beta$, a correlation is developed for determining the *N*-ribosyl attachment site of tetrazole ribonucleosides. Results are also presented on antiviral testing of these synthetic 5-aminotetrazole ribonucleosides against influenza A2/Asian/J305 virus infection in mice.

The recent discoveries[2,3] of potent antiviral activities in several classes of nucleosides have given impetus to acceleration of research in this area. Our own interest in the synthesis of tetrazole ribonucleosides was stimulated by the discovery of antiviral activity of Virazole[2a] (ribavirin). In a previous report[4] we have described the synthesis, structure elucidation, and antiviral testing of the parent tetrazole ribonucleoside and two of its 5-substituted derivatives. This paper presents the synthesis, structural assignment, and antiviral evaluation of 5-amino-1-(β-D-ribofuranosyl)-1*H*-tetrazole (**5**) and 5-amino-2-(β-D-ribofuranosyl)-2*H*-tetrazole (**6**). Although ^{13}C spectroscopy was utilized in the assignment of the site of N-glycosylation of tetrazoles in our previous work,[4] the conclusions were based on analogy to the parent 1-(β-D-ribofuranosyl)tetrazole and N_1-methyl- and N_2-methyltetrazole derivatives. This work provides us with an isomeric pair of 5-aminotetrazole ribonucleosides which possess similar (β) anomeric configuration but differ in the site of *N*-ribosyl substitution. The availability of these compounds has enabled us to develop a correlation between ^{13}C spectroscopy and the *N*-ribosyl attachment site.

Results and Discussion

Ribosylation of 5-Aminotetrazole. The synthesis of 5-amino-1-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-1*H*-tetrazole (**3**) and 5-amino-2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-2*H*-tetrazole (**4**) was achieved by using two well-documented procedures (Scheme I): (a) by a reaction between fully silylated 5-aminotetrazole and 2,3,5-tri-*O*-benzoyl-D-ribofuranosyl bromide (**2**) in acetonitrile at room temperature; (b) by an acid-catalyzed fusion[5] of 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-ribofuranose (**1**) and 5-aminotetrazole at 165°. The resulting nucleoside mixtures differed dramatically in both yields and relative ratios of the $N_1\beta$ (**3**) and $N_2\beta$ (**4**) isomers in the two methods. From the silyl method the yield of the total ribosylated 5-aminotetrazole was 72% with a ratio of 55:45 between $N_1\beta$ and $N_2\beta$ isomers. The acid-catalyzed fusion method, on the other hand, resulted in quite poor yield of the coupled product which contained $N_2\beta$ as the predominant component, $N_1\beta$ isomer being present only in trace amounts (TLC). The tri-*O*-benzoylated nucleosides **3** and **4** were then deblocked by the standard methanolic-ammonia

Scheme I



treatment to obtain 5-amino-1-(β-D-ribofuranosyl)-1*H*-tetrazole (**5**) and 5-amino-2-(β-D-ribofuranosyl)-2*H*-tetrazole (**6**), respectively.

Structural Determination of 5-Aminotetrazole Ribonucleosides. Although a number of exceptions to