Journal of Medicinal Chemistry, 1971, Vol. 14, No. 12 **1155**

Nucleoside Peptides. 2. Synthesis of Certain 5-*N*-Aminoacyl and 5-*N*-Peptidyl Derivatives of 5-Aminouridine

GEORGE A. IVANOVICS, ROBERT J. ROUSSEAU,* AND ROLAND K. ROBINS

ICN Nucleic Acid Research Institute, Irvine, California 92664

Received April 26, 1971

Several 5-*N*-aminoacyl- and 5-*N*-peptidyl-5-aminouridines have been synthesized which represent a new class of nucleoside peptides. 5-Aminouridine (**1**) and 2′,3′,5′-tri-*O*-acetyl-5-aminouridine (**4c**) were coupled to CBZ-blocked amino acids and peptides by the acid chloride and DCC methods. Removal of the protecting groups gave the title compounds which were examined for their effects in several biological systems.

Roberts and Visser[1] first prepared 5-aminouridine (**1**) and discovered it possessed a wide range of biological activity including inhibition of the growth of fungi,[1] viruses,[2] and tumors.[3] This analog is incorporated into RNA of Ehrlich ascites cells and has been shown to be metabolized to 5-amino-UMP, 5-amino-UDP, 5-amino-UTP, and 5-amino-UDP-sugars.[4] The metabolite 5-amino-UMP was found to be a potent inhibitor of orotidylic acid decarboxylase and probably exerts biological action by interference with the *de novo* pathway of pyrimidine nucleotide biosynthesis.[4] It has also been observed that **1** inhibits incorporation of formate[5] into purines and pyrimidines and reduces the incorporation of phosphate into the phospholipids and the ribonucleic acid nucleotides[6] of rat livers.

Reasons for the study and synthesis of nucleoside peptides as possible medicinal agents have been outlined in a previous paper dealing with the syntheses of 5′-peptidyl derivatives submitted from these laboratories.[7] The 5 position of uridine was selected for peptide attachment since 5-alanyluracil has recently been isolated[8] from germinating pea seeds.

The active ester method of Bodanszky[9] has been utilized to couple an amino group of a nucleoside to amino acids in the presence of free OH groups.[7] When *N*-CBZ amino acid *p*-nitrophenyl esters[10] were treated with 5-aminouridine (**1**) in DMF there was no detectable product formation. On the other hand, activation of the 5-amino group of **1** using the phosphazo method[11,12] was unsuccessful in that it provided low yields and was complicated by numerous side products caused by presence of unprotected OH groups[13] of D-ribose. Similarly, the use of DCC[14] as a coupling agent resulted in a variety of side products.

A modification of the classic acid chloride method[15] provided the procedure of choice for the coupling of amino acids to **1**. When the *N*-CBZ derivative of L-Phe, was treated with the DMF–$SOCl_2$ adduct[16] at −20° and the resulting amino acid chloride was treated with **1**, 5-*N*-(*N*-CBZ-L-Phe)-5-aminouridine (**2a**) was isolated in 82% yield without major side products. Similarly **2b**, **2c**, and **2d** were produced in yields of 73, 67, and 87%, respectively (Scheme I). Attempts to remove the CBZ-blocking group with HBr–AcOH were accompanied by partial acetylation of the ribose moiety, therefore catalytic hydrogenolysis with Pd/C provided the desired aminoacyl-5-aminouridines (**3a**–**3d**) in excellent yield.

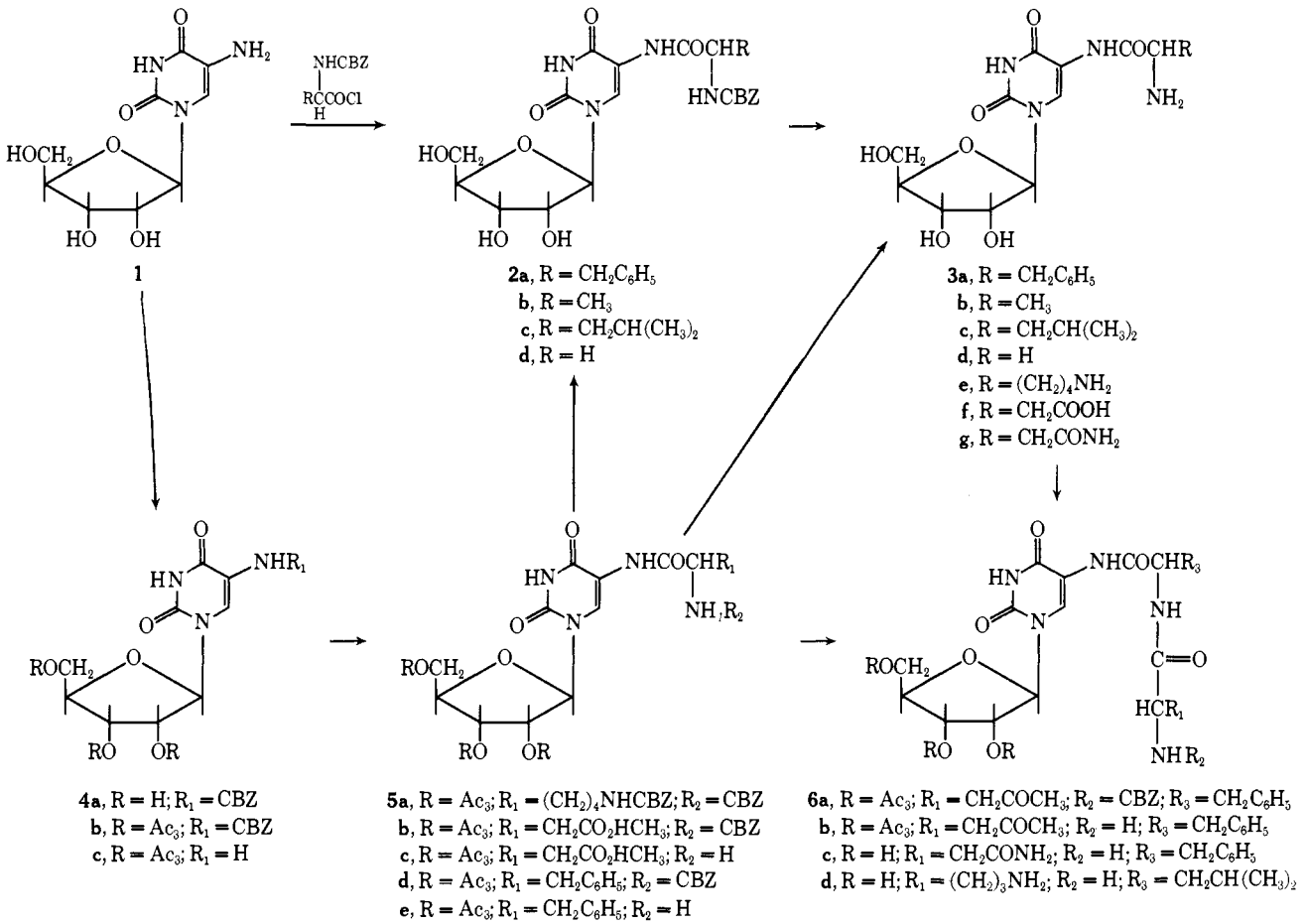
Treatment of N^{α},N^{ϵ}-di-CBZ-L-Lys with $SOCl_2$–DMF however, resulted in quantitative conversion to L-Lys-*N*-carboxyanhydride.[17] This limitation of the acid chloride method prompted a reinvestigation of the DCC coupling method *via* the appropriately blocked nucleoside 2′,3′,5′-tri-*O*-acetyl-5-aminouridine (**4c**). Synthesis of **4c** was accomplished by selective reaction of the 5-amino group of **1** with benzyloxycarbonyl chloride in basic media to afford 5-*N*-CBZ-5-aminouridine (**4a**). Subsequent acylation with a mixture of AcOH and pyridine provided 5-*N*-CBZ-2′,3′,5′-tri-*O*-acetyl-5-aminouridine (**4b**). Catalytic cleavage of the CBZ group gave the desired intermediate **4c** in excellent yield.

Facile coupling between N^{α},N^{ϵ}-di-CBZ-L-Lys and **4c** was achieved with DCC and afforded the blocked aminoacyl nucleoside **5a**. Catalytic removal of the CBZ group followed by treatment with methanolic NH_3 gave 5-N^{α}-(L-Lys)-5-aminouridine (**3e**) in 54% overall yield. Similarly *N*-CBZ-L-Asp-β-methyl ester was coupled to **4c** and yielded the blocked compound **5b**. Catalytic cleavage of the CBZ group gave the acetylated methyl ester **5c** which could be converted either to 5-*N*-(L-Asp)-5-aminouridine (**3f**) or 5-*N*-(L-Asn)-5-aminouridine (**3g**) by the action of aqueous base or methanolic NH_3, respectively.

The synthesis of 5-*N*-dipeptidyl derivatives of **1** was accomplished by coupling of the appropriately blocked amino acid with the intermediates 5-*N*-(L-Phe)-2′,3′,5′-tri-*O*-acetyl-5-aminouridine (**5e**) or 5-*N*-L-Leu-5-aminouridine (**3c**). Coupling of **5e** with *N*-CBZ-L-Asp-β-methyl ester was accomplished in good yield by the DCC method. The CBZ group of the product **6a** was removed with HBr–AcOH and the intermediate 5-*N*-[L-(β-*O*-methyl)Asp-L-Phe]-2′,3′,5′-tri-*O*-acetyl-5-

(1) M. Roberts and D. W. Visser, *J. Biol. Chem.*, **194**, 695 (1952).
(2) D. W. Visser, D. L. Lagerborg, and H. E. Pearson, *Proc. Soc. Exp. Biol. Med.*, **79**, 571 (1952).
(3) D. W. Visser, *Antimetab. Cancer Symp.*, **1953**, 59 (1955).
(4) D. A. Smith, P. Roy-Burman, and D. W. Visser, *Biochim. Biophys. Acta*, **119**, 221 (1966).
(5) W. C. Werkheiser and D. W. Visser, *Cancer Res.*, **15**, 644 (1955).
(6) W. C. Werkheiser, R. J. Winzler, and D. W. Visser, *ibid.*, **15**, 641 (1955).
(7) M. J. Robins, L. N. Simon, M. G. Stout, G. A. Ivanovics, M. P. Schweizer, R. J. Rousseau, and R. K. Robins, *J. Amer. Chem. Soc.*, **93**, 1474 (1971).
(8) E. G. Brown and A. V. Silver, *Biochim. Biophys. Acta*, **119**, 1 (1966).
(9) M. Bodanszky, M. Szelke, E. Tomorkeny, and E. Weiss, *Chem. Ind. (London)*, 1517 (1955).
(10) M. Bodanszky and V. du Vigneaud, *J. Amer. Chem. Soc.*, **81**, 5688 (1959).
(11) S. Goldschmidt and C. Jutz, *Chem. Ber.*, **86**, 1116 (1953).
(12) S. Goldschmidt and H. L. Kraus, *Angew. Chem.*, **67**, 471 (1955).
(13) J. Zahn and F. Schnabel, *Justus Liebigs Ann. Chem.*, **605**, 212 (1957).
(14) J. C. Sheehan and G. P. Hess, *J. Amer. Chem. Soc.*, **77**, 1067 (1955).
(15) M. Zaoral and Z. Arnold, *Tetrahedron Lett.*, 9 (1960).
(16) H. H. Bosshard, R. Mory, M. Schmid, and H. Zollinger, *Helv. Chim. Acta*, **42**, 1653 (1953).
(17) M. Bergman, L. Zervas, and W. F. Ross, *J. Biol. Chem.*, **111**, 245 (1935).

SCHEME I



aminouridine·HBr (**6b**) was converted to 5-*N*-(L-Asn-L-Phe)-5-aminouridine (**6c**) with methanolic NH_3.

N^{α}-CBZ-N^{ω}-nitro-L-Arg was coupled to free nucleoside amino acid **3c** by the action of DCC and *N*-hydroxysuccinimide in good yield. After removal of the CBZ- and NO_2-blocking groups by catalytic hydrogenolysis, the elemental analysis of the product isolated did not correspond to an arginine derivative but to 5-*N*-(L-ornithyl-L-Leu)-5-aminouridine (**6d**). This product was predicted since arginine is known to decompose to ornithine under basic conditions[18] similar to those used in the isolation of **6d**. Confirmation was obtained when **6d** was hydrolyzed by dil HCl to give L-Leu- and L-ornithine.

These compounds were tested for inhibition of herpes simplex, parainfluenza, rhino, and adeno virus (Table I). Different amino acid moieties attached to the 5-amino group of 5-aminouridine were found to influence the rate of multiplication of these viruses. L-Lys-5-aminouridine inhibited all of these virus strains at concns of 320 μg/ml, whereas L-Asp-5-aminouridine (**3f**) gave no inhibition of growth at concns as high as 1000 μg/ml. The L-Phe-(**3g**), L-Ala-(**3c**), Gly-(**3d**), and L-Asn-(**3g**) derivatives were intermediate in their antiviral properties. It is interesting to note that the dipeptide 5-*N*-(L-Asn-L-Phe)-5-aminouridine (**6c**) showed greater inhibition of rhino virus than 5-aminouridine (**1**) at concns of 100 μg/ml.

(18) D. E. Rivard and H. E. Carter, *J. Amer. Chem. Soc.*, **77**, 1260 (1955).

Experimental Section[19]

5-*N*-(*N*-CBZ-Aminoacyl)-5-aminouridines (Table II). Method A.—The appropriate *N*-CBZ-amino acid (1.0 mmole) was treated with 119 mg of $SOCl_2$ in 4 ml of DMF at −20°. The reaction mixt was protected from moisture during 1 hr at −5 to −8°, then added to a mixt of 284 mg (1.1 mmoles) of 5-aminouridine[20] (**1**) and 222 mg (2 mmoles) of Et_3N in 5 ml of DMF at −20°. The Et_3NHCl which pptd was removed by filtration. The filtrate was evapd to a syrup *in vacuo*. The syrup was triturated with a 1:1 mixt of EtOAc–Et_2O then with H_2O. The resulting solid was collected and recrystd from the appropriate solvent.

Method B.—Removal of CBZ group by catalytic hydrogenolysis. The calcd wt of CBZ-protected compd dissolved in the appropriate solvent (Table II) was hydrogenated with 50 mg of 10% Pd/C at atm pressure and room temp. After 2 hr the catalyst was removed by filtration and washed with H_2O (3 × 5 ml), and the combined filtrates were evapd to dryness *in vacuo* and recrystd.

5-*N*-CBZ-Aminouridine (4a).—A rapidly stirred soln of **1**[20] (2.58 g, 10 mmoles) and Na_2CO_3 (0.74 g, 7 mmoles) in 30 ml of H_2O was treated with a soln of benzyl chloroformate (1.87 g, 11 mmoles) in 10 ml of Et_2O at 0°. After the addn was complete the cooling bath was removed and the stirring was contd at room temp for 3 hr. The white product which was collected by filtration was triturated with Et_2O (50 ml) and H_2O (50 ml) and then recrystd from MeOH to yield 3.06 g of **4a** (77%): mp 192–193°; $[\alpha]^{25}D$ −33.3° (*c* 1, DMSO). *Anal.* ($C_{17}H_{19}N_3O_8$) C, H, N.

5-*N*-CBZ-(2′,3′,5′-Tri-*O*-Ac)-5-aminouridine (4b).—Compd **4a** (1.65 g, 3.09 mmoles) was treated with a mixt of Ac_2O (15 ml)

(19) Physical properties were detd with the following instruments: mp, Thomas Hoover app (uncorrected); uv spectra, Cary 15 uv spectrometer (pH 1 and pH 11); sp rot., Perkin-Elmer Model 141 polarimeter; pmr, Hitachi Perkin-Elmer R20A high-resolution nmr spectrometer (Me_4Si or DSS); and ir spectra, Perkin-Elmer Model 257 (KBr).

(20) M. Roberts and D. W. Visser, *J. Amer. Chem. Soc.*, **74**, 668 (1952).

TABLE I

EFFECT OF CERTAIN 5-*N*-AMINOACYL-5-AMINOURIDINE DERIVATIVES ON THE MULTIPLICATION OF VIRUSES IN KB CELLS

Compd	Adeno virus, % inhibn	Concn, μg/ml	Herpes virus, % inhibn	Concn, μg/ml	Parainfluenza virus, % inhibn	Concn, μg/ml	Rhino virus, % inhibn	Concn, μg/ml
3a			0	100	67	32	0	100[a]
			50	320[b]	67	100[a]	50	1000[b]
3b			0	100	0	100	0	100
			25	320[a]	0	320[a]	25	320[a]
3c			0	100	0	100	0	100
			25	320[b]	50	320[b]	0	320[b]
3d			0	32	0	32		
			25	100[a]	0	100		
3e	0	100	0	100	0	100	67	320
	75	320[b]	67	320[b]	50	320[b]	100	1000[b]
3f	0	320	0	320	0	320	0	320
	0	1000	0	1000[a]	0	1000	0	1000[a]
3g	0	320	0	320	0	320	0	320
	0	1000	0	1000	0	1000	75	1000
6c			0	100	0	32	43	100
			0	320[a]	0	100[a]	65	320[a]
6d			0	32	0	32	0	320
			0	100[a]	0	100[a]	67	1000[a]
1	0	32	0	32	0	32	0	32
	75	100[a]	0	100[b]	0	100[a]	25	100

[a] Microscopic examination revealed a slight difference in shape of KB cells but monolayer not effected. [b] Partial slight destruction of monolayer.

TABLE II

Compd	Recrystallization solvent	Method	Mp, °C	$[\alpha]^{25}D$	Formula	Anal.	Yield, %
2a	MeOH	A	182–184	−22.8 (*c* 1, DMSO)	$C_{26}H_{28}N_4O_9$	C, H, N	82
2b	95% EtOH	A	194–196	−56.8 (*c* 1, DMSO)	$C_{20}H_{24}N_4O_9 \cdot H_2O$	C, H, N	73
2c	EtOAc	A	71–73	−15.5 (*c* 1, DMSO)	$C_{23}H_{30}N_4O_9$	C, H, N	67
2d	MeOH	A	211–221	−34.4 (*c* 1, DMSO)	$C_{19}H_{21}N_4O_9$	C, H, N	87
3a	H_2O	B[a]	191–193	52.3 (*c* 1, 1 *N* HCl)	$C_{18}H_{22}N_4O_7$	C, H, N	86
3b	95% EtOH	B[b]	88–90		$C_{12}H_{18}N_4O_7$	C, H, N	72
3c	EtOH	B[c]	103.5–105.5	16.0 (*c* 1, 1 *N* HCl)	$C_{15}H_{24}N_4O_7$	C, H, N	82
3d	EtOH–H_2O	B[d]	71–73	−8.0 (*c* 1, 1 *N* HCl)	$C_{11}H_{16}N_4O_7$	C, H, N	93

[a] **2a** (1 mmole) in 8 ml of 85% EtOH. [b] **2b** (0.443 mmole) in 10 ml of 90% EtOH. [c] **2c** (0.39 mmole) on 6 ml of 95% EtOH. [d] **2d** (0.445 mmole) in 10 ml of 60% dioxane.

and pyridine (10 ml) at room temp. After 2 hr the soln was evapd to dryness *in vacuo*, and the residue was crystd from abs EtOH to yield 1.92 g (96%) of **4b**: mp 149–150°, $[\alpha]^{25}D$ −28.3° (*c* 1, DMSO). *Anal.* ($C_{23}H_{25}N_3O_{11}$) C, H, N.

2′,3′,5′-Tri-*O*-Ac-5-aminouridine (4c).—Compd **4b** (1.6 g, 3.08 mmoles) was dissolved in 50 ml of 95% EtOH and hydrogenated with 200 mg of 10% Pd/C at room temp and atm pressure. After 2.5 hr the catalyst was removed by filtration and washed with 95% EtOH (3 × 20 ml), and the combined filtrates were evapd to dryness. The product was recrystd from MeOH–H_2O to yield 1.030 g (87%): mp 66–68°; $[\alpha]^{25}D$ 30.8° (*c* 1, DMSO). *Anal.* ($C_{15}H_{19}N_3O_9$) C, H, N.

5-*N*-(N^{α},N^{ϵ}-Di-CBZ-L-Lys)-2′,3′,5′-tri-*O*-Ac-5-aminouridine (5a).—A cold soln of **4c** (1.34 g, 3.5 mmoles) and N_{ϵ},N_{ϵ}-dicarbobenzoxy-L-Lys (1.23 g, 3 mmoles) in 10 ml of EtOAc was treated with DCC (0.72 g, 3.5 mmoles). After 48 hr at 4° the pptd dicyclohexylurea was removed by filtration and the filtrate was washed with 5% citric acid, 5% $NaHCO_3$, and then H_2O. The solvent was removed under reduced pressure and the residual glass was purified by chromatog on a column (2.4 × 30 cm) packed with silica gel "Baker," in $CHCl_3$. The column was washed with $CHCl_3$ (300 ml) and the product was eluted with $CHCl_3$–EtOAc 1:1. The combined uv-absorbing fractions were evapd to dryness to yield an amorphous, white material: 1.78 g (76%); $[\alpha]^{25}D$ −41.5° (*c* 1, DMSO). *Anal.* ($C_{37}H_{43}N_5O_{14}$) C, H, N.

5-*N*-(L-Lys)-5-aminouridine (3e).—Compd **5a** (570 mg, 0.73 mmole) was dissolved in 20 ml of abs EtOH contg 1 ml of HOAc and hydrogenated with 10% of Pd/C (80 mg) at 45° and atm pressure. After 2 hr the catalyst was removed by filtration, washed with EtOH, and evapd to dryness *in vacuo*. The residual glass was treated with 50 ml of satd methanolic NH_3 at 0° and then the soln was kept at room temp for 16 hr. The solvent was removed under reduced pressure. The residue was dissolved in a small amt of H_2O and applied to a column of Dowex 50 [H^+]-(50–100 mesh, 1.5 × 25 cm). After washing the column with H_2O (300 ml) the product was eluted with 1 *N* NH_4OH. The uv-absorbing fraction was collected and evapd to a small vol. EtOH was added to the residue until incipient turbidity, then this mixt was allowed to cool to −4° for several hr and white crystals deposited to yield 282 mg (54%): mp 138–140°, $[\alpha]^{25}D$ −58.5° (*c* 1, H_2O). *Anal.* ($C_{15}H_{25}N_5O_7$) C, H, N.

5-*N*-(*N*-CBZ-β-*O*-Methyl-L-Asp)-2′,3′,5-tri-*O*-Ac-5-aminouridine (5b).—A soln of *N*-CBZ-L-Asp-β-methyl ester[21] (1.405 g, 5 mmoles), and **4c** (2.304 g, 6 mmoles) in EtOAc (25 ml) was treated with DCC (1.44 g, 1 mmole) at 0°. After 4 hr at room temp and 16 hr at 4° the *N*,*N*′-dicyclohexylurea was removed by filtration and the filtrate was washed with 5% citric acid, 5% $NaHCO_3$, and then H_2O. The solvent was removed under reduced pressure and the residue was dissolved in a small amt of $CHCl_3$ and applied to a column of silica gel "Baker," 2.5 × 30 cm, packed in $CHCl_3$. The column was washed first with $CHCl_3$ (1000 ml) and then the product was eluted with $CHCl_3$–EtOAc 2:1. The uv-absorbing fractions were collected and evapd to a small vol. Petr ether was carefully added to these fractions and a colorless cryst material deposited to yield 2.760 g (85%) of **5b**: mp 67–69°; $[\alpha]^{25}D$ −61.7° (*c* 1, DMSO). *Anal.* ($C_{28}H_{32}N_4O_{14}$) C, H, N.

5-*N*-(L-Asn)-5-aminouridine (3g).—A soln of **5b** (1.296 g, 2 mmoles) in 95% EtOH (50 ml) was hydrogenated with 100 mg of 10% Pd/C at 40° and atm pressure. After 2 hr the catalyst was removed by filtration and the filtrate was evapd to dryness

(21) H. Schwartz, F. M. Bumpus, and I. H. Page, *J. Amer. Chem. Soc.*, **79**, 5697 (1957).

in vacuo. Half of the residue was preserved for an alternative work-up (see **3f**). The other half was treated with satd methanolic NH_3 (40 ml) at 0°. After 16 hr at 4° the solvent was removed under reduced pressure and the residue was crystd from EtOH–H_2O to yield 258 mg (66%) of **3g**: mp 135–137°; $[\alpha]^{25}D$ 9.0° (*c* 1, 1 *N* HCl). *Anal.* ($C_{13}H_{19}N_4O_8 \cdot H_2O$) C, H, N.

5-*N*-(L-Asp)-5-aminouridine (3f).—The amt of crude 5-*N*-(β-*O*-methyl-L-Asp)-2′,3′,5′-tri-*O*-Ac-5-aminouridine which was prepared in the previous procedure (see **3g**) was treated with anhyd MeOH (30 ml) contg 80 mg of NaOMe. After the soln was refluxed for 15 min, then kept at room temp for 4 hr, the solvent was removed under reduced pressure. H_2O (8 ml) was added and the soln was kept at 4° for an additional 16 hr, percolated through a column of Amberlite IR 50 [H^+] 100–200 mesh, and then evapd to a small vol. The residue was kept at 4° for 16 hr, and colorless crystals deposited to yield 221 mg (58%) of product: mp 203–206°; $[\alpha]^{25}D$ −8.3° (*c* 1, 1 *N* HCl). *Anal.* ($C_{13}H_{18}N_4O_9 \cdot 1.5\ H_2O$) C, H, N.

5-*N*-(*N*-CBZ-L-Phe)-2′,3′,5′-tri-*O*-Ac-5-aminouridine (5d).—Compd **4c** (1.21 g; 31 mmoles) and *N*-CBZ-L-Phe (1.036 g; 34 mmoles) were dissolved in EtOAc (10 ml). The soln was treated with DCC (0.707 g; 34 mmoles) at 0°, then stirred at room temp for 18 hr. The dicyclohexylurea was removed by filtration, and the filtrate was washed, resp, with 5% citric acid, 5% HCl, H_2O, 5% $NaHCO_3$, and H_2O and then dried (Na_2SO_4). The solvent was evapd under reduced pressure and the residue was crystd from Et_2O–heptane to yield 1.26 g (61%): mp 71–73°; $[\alpha]^{25}D$ 34.7° (*c* 1, DMSO).

5-*N*-(L-Phe)-2′,3′,5′-tri-*O*-acetyl-5-aminouridine·HBr (5e).—Compd **5d** (665 mg; 1 mmole) was dissolved in EtOAc (1.5 ml) and the soln was treated with 35% HBr in HOAc (0.6 g; 2.6 mmoles) at room temp. After 1 hr Et_2O (3.5 ml) was added and colorless cryst product sepd to yield 573 mg (94%) of **5c**, mp 112–114°.

5-*N*-[*N*-CBZ-L-(β-*O*-methyl-L-Asp)-L-Phe]-2′,3′,5′-tri-*O*-Ac-5-aminouridine (6a).—*N*-CBZ-L-Asp-β-methyl ester (436 mg; 1.55 mmoles) was treated with excess Et_2NH (3.0 mmoles) in CH_2Cl_2 (10 ml). The solvents were removed under reduced pressure and the residue was dissolved in CH_2Cl_2 (12 ml). Compd **5e** (1.468 g; 2.4 mmoles) and DCC (618 mg; 3 mmoles) were added to the soln and it was kept at room temp for 18 hr. The dicyclohexylurea was removed by filtration, and a filtrate was washed with 5% HCl, 5% $NaHCO_3$, and H_2O and dried (Na_2SO_4). The soln was concd to a small vol and applied to a column of silica gel "Baker" (2.5 × 25 cm), packed in $CHCl_3$. The column was washed first with $CHCl_3$ (200 ml) then with $CHCl_3$–Me_2CO 10:1. The uv-absorbing fractions which were chromotog homogeneous were combined and evapd to dryness to yield 820 mg (66%) of **6a** as a colorless glass.

5-*N*-[L-β-*O*-Methyl)Asp-L-Phe]-2′,3′,5′-tri-*O*-Ac-5-aminouridine·HBr (6b).—Compd **6a** (800 mg; 1 mmole) was dissolved in EtOAc (6 ml) and was treated with 35% HBr in AcOH (0.8 ml). After 2 hr an addl 0.5 ml of AcOH–HBr was added and the soln was kept at room temp for 30 min. Then Et_2O (3 ml) was added slowly and colorless cryst material deposited to yield 610 mg (82%) of product: mp 118–120°; $[\alpha]^{25}D$ 14.5 (*c* 1, DMSO).

5-*N*-(L-Asn-L-Phe)-5-aminouridine (6c).—Compd **6b** (200 mg; 0.27 mmole) was treated with satd MeOH–NH_3 (10 ml) at room temp for 18 hr then the solvent was removed under reduced pressure. The residue was dissolved in a small amt of H_2O and applied to a column of Dowex 50 ([H^+] form, 100–200 mesh, 1 × 15 cm). The column was washed with H_2O, then the product was eluted by gradient elution (150 ml of 0.5 *N* NH_4OH, reservoir; 150 ml of H_2O) mixing chamber. The uv-absorbing fractions which were chromatog homogeneous were combined and lyophilized, then dried (P_2O_5 *in vacuo*) at 60° to yield 87 mg (62%) of **6c**: mp 122–123° (174–175° dec); $[\alpha]^{25}D$ 4.0° (*c* 1, 1 *N* HCl). *Anal.* ($C_{22}H_{28}N_6O_9 \cdot 0.5\ H_2O$) C, H, N.

5-*N*-(L-Ornithyl-L-Leu)-5-aminouridine (6d).—5-*N*-L-Leu-7,5-amiuouridine (**3c**) (254 mg; 0.685 mmole), N^{α}-CBZ-N^{ω}-nitro-L-Arg[22] (353 mg, 1.0 mmole), *N*-hydroxysuccinimide (115 mg, 1.0 mmole), and DCC (206 mg; 1.0 mmole) were dissolved in dry DMF (3 ml) and kept at room temp for 2 hr. After the reaction mixt was allowed to stand at 14° for 16 hr, the dicyclohexylurea was removed by filtration, and the filtrate was evapd to dryness under reduced pressure. The residue was dissolved in 5 ml of MeOH–NH_4OH (concd) (8:2), kept at room temp for 4 hr, applied to prep silica gel tl plates, and developed with solvent system MeOH–CH_2Cl_2–NH_4OH (concd), 2:2:1. The major uv-absorbing band was eluted with 80% MeOH (30 ml). The eluant was treated with 100 mg of Pd/C, then with H_2 at room temp and pressure. After 3 hr the catalyst was removed by filtration, the filtrate was evapd to dryness, and the residue was taken up with a small amt of H_2O and lyophilized to yield 102 mg (26%) of **6d**: mp 81–83°; $[\alpha]^{25}D$ 17.0° (*c* 1, 1 *N* HCl). *Anal.* ($C_{20}H_{34}N_6O_8 \cdot 4H_2O$) C, H, N.

Compd **6d** was hydrolyzed with 6 *N* HCl for 8 hr at 100° in a sealed tube. The hydrolysate was chromatogd against L-ornithine and L-Leu on silica gel [CH_2Cl_2–MeOH–NH_4OH (concd) (2:2:1)] and cellulose plates [*n*-BuOH–AcOH–H_2O (3:1:1)]. The ninhydrin-positive spots from the hydrolysate were found to be identical with L-ornithine and L-Leu.

Acknowledgments.—The authors wish to thank Dr. R. Sidwell and Mr. J. Huffman for performing the antiviral screening.

(22) J. P. Greenstein and M. Winitz, "The Chemistry of the Amino Acids," Wiley, New York, N. Y., 1961, p 1069.