Acyclopyrimidine *C*-Nucleosides: Synthesis of 5-[(2,3-Dihydroxy-1-propoxy)methyl]pyrimidines [1]

Chung K. Chu* and Jungjin Suh

Department of Medicinal Chemistry and Pharmacognosy, College of Pharmacy, The University of Georgia, Athens, Georgia 30602

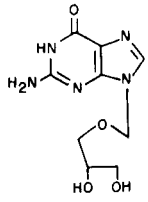
Received April 14, 1986

From the condensation of 5-hydroxymethyluracil and glycerine, 5-[(2,3-dihydroxy-1-propoxy)methyl]uracil (**3**) was synthesized, which was converted to the isocytosine derivative **9** by the ring-transformation reaction *via* dimethyluracil derivative **7**.

J. Heterocyclic Chem., **23**, 1621 (1986).

Acyclonucleosides are an important class of compounds as antiviral agents. Acyclovir (Zovirax) [2-4] has played a key role as a lead compound in this new class of nucleosides. Since 1982 ointment and intravenous dosage forms of acyclovir for herpes type-2 have been marketed and an oral form of the drug was also approved by the Food and Drug Administration in 1985. Due to the clinical efficacy of acyclovir, a number of purine and pyrimidine acyclic nucleosides were prepared. A comprehensive review on the chemistry and biology of acyclonucleosides was recently published in this Journal [5].

Among acyclovir analogues, 9-[(1,3-dihydroxy-2-propoxy)methyl]guanine (DHPG; 2'-NDG; Biolg-62; BW 757W) [6-9] was found to be more active than acyclovir *in vivo*. Unfortunately, this compound produces testicular atropy [10]. In connection with modification of the side chain of acyclovir, Lin and Lin [11], and Ashton *et al* [12] reported the synthesis of 9-[(2,3-dihydroxy-1-propoxy)-methyl]guanine (iso-NDG) and its antiviral activity.



iso-NDG

Recently, we reported the synthesis of acyclopyrimidine *C*-nucleosides with a side chain of acyclovir for antiviral screening [13]. Preliminary results indicated that the thymidine analog was found to give some activity (50% PFU reduction at 50 μM against HSV-1). The same compound was also reported by Melbnik *et al* [14]. As a part of our continuing efforts to synthesize acyclo-*C*-nucleosides for antiviral studies, we report the synthesis of 5-[(2,3-dihydroxy-1-propoxy)methyl]pyrimidines, which contain the iso-NDG type side chain.

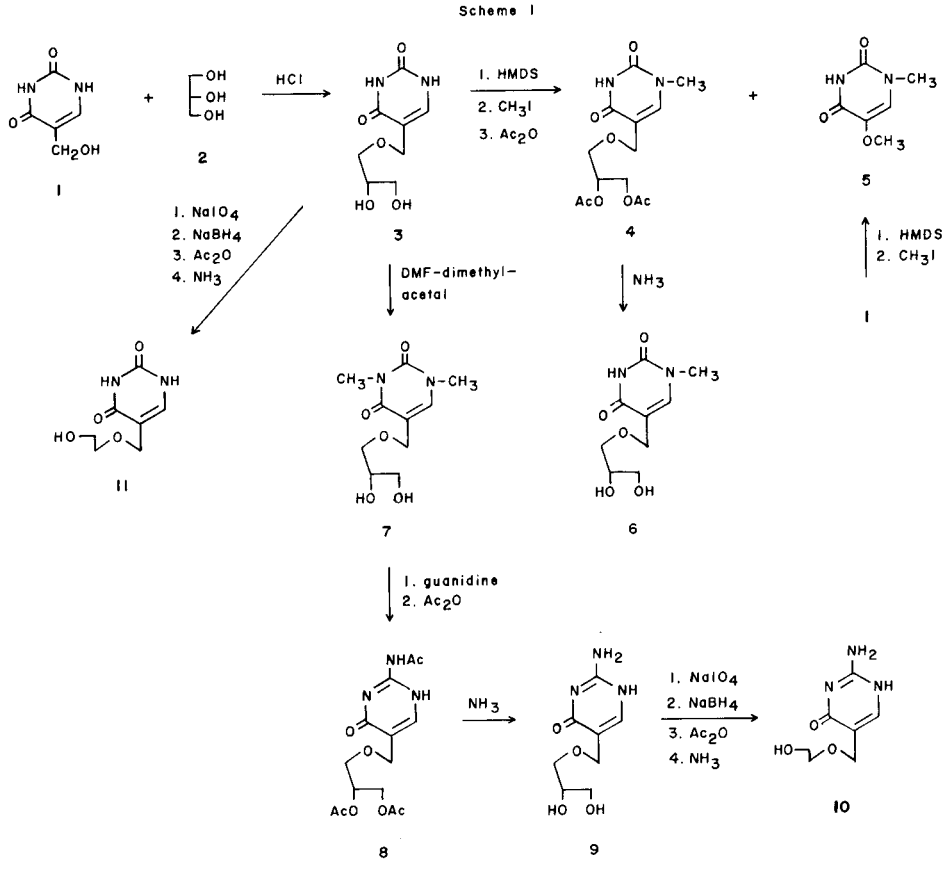
5-Hydroxylmethyluracil [13,15] and glycerine were condensed with concentrated hydrochloric acid without solvent to give a good yield (88%) of 5-[(2,3-dihydroxy-1-propoxy)methyl]uracil (**3**) (Scheme 1). The identification of **3** was made on the basis of pmr spectrum, in which a characteristic doublet and triplet for secondary and primary hydroxyl group, respectively, indicated the correct side chain as well as the typical uv pattern of the uracil moiety. In order to obtain the thymidine analog **6**, **3** was selectively methylated at the N_1 position by our previous method [13]. The reaction, however, gave a mixture of two products **5** and **6**. Since the desired product **6** is polar, acetylation was necessary to separate the mixture on a silica gel column. The desired product **6** was obtained from **4** by the treatment of the acetylated product with methanolic ammonia. Another compound from the methylation was found to be a methyl ether **5**, which was probably formed from **6** by methyl iodide and hydrogen iodide liberated during the reaction. Attempts to minimize the product **5** by varying conditions was not successful. This compound was also prepared from **1** for confirmation of the structure. The structural identification of **6** was based on the pmr and uv spectra described above for **3**.

In order to synthesize acyclonucleoside analogue **9** of pseudoisocytidine [16], uracil derivative **3** was converted to the dimethyluracil derivative **7** by dimethylformamide dimethylacetal. The dimethyluracil derivative was then treated with free guanidine to obtain an isocytidine analogue **9**. As described for **6**, **9** was also too polar to use a silica gel column. Thus, the reaction mixture with guanidine was acetylated and separated on a silica gel column to obtain **8**, which was then treated with methanolic ammonia to yield an acyclo-*C*-nucleoside analogue of pseudoisocytidine **9**.

It was of interest to convert **3** and **9** to **11** and **10**, respectively, confirming the structures of **3** and **9**, since we have previously reported the preparation of **10** and **11** by a different route [13]. The oxidation reaction with sodium periodate went smoothly, however, acetylation was also required to obtain the pure compounds **10** and **11**.

None of the compounds showed any significant activity against herpes type-1 and type-2 virus [17].

Scheme I



EXPERIMENTAL

Melting points were determined on a Thomas-Hoover capillary melting point apparatus and are uncorrected. The pmr spectra were recorded on a JEOL FX 90Q fourier transform spectrometer (90 MHz). Tetramethylsilane was the internal standard for organic solvents and sodium 3-(trimethylsilyl)-1-propane-1-sulfonate (DSS) was the internal standard for deuterium oxide: chemical shifts are reported in parts per million (δ), and signals are described as s (singlet), d (doublet), t (triplet), q (quartet), b (broad), m (multiplet). Ultraviolet spectra were recorded on a Bausch & Lomb Spectronic 2000 spectrometer. The tlc analysis was performed on Uniplates purchased from Analtech Co. or Pre-coated tlc sheets (Silica gel 60 F-254) by Em Laboratories, Inc. Elemental analysis were performed by Atlantic Microlab, Inc., Atlanta, GA.

5-[(2,3-Dihydroxy-1-Propoxy)methyl]uracil (**3**).

A mixture of 5-hydroxymethyluracil (10 g, 0.07 mole), glycerine (80 ml), and concentrated hydrochloric acid (6 ml) was heated at 60-70° for 2 hours. The mixture was then cooled to room temperature and the turbid mixture was triturated with absolute ethanol (100 ml). The resulting precipitate was filtered and the filter cake was collected and dried *in vacuo* to give white amorphous crystals (13.3 g, 88%). Recrystallization from ethanol gave an analytical sample, mp 181-183°; uv: λ max 261 nm (*p*H 1-7) and 285 (*p*H 13); ^{1}H nmr (dimethylsulfoxide-d_6): δ 3.20-3.70 (m, 5H, CH_2CHCH_2), 4.10 (s, 2H, benzylic), 4.46 (t, 1H, CH_2OH, J = 5.6 Hz, exchangeable), 7.41 (d, 1H, aromatic, JC_6NH = 5.5 Hz, collapsed to a singlet with deuterium oxide), 10.82 (d, 1H, N_1H, exchangeable), and 11.09 (b, 1H, N_3H, exchangeable).

Anal. Calcd. for $C_8H_{12}N_2O_5$: C, 44.44; H, 5.56; N, 12.96. Found: C, 44.55; H, 5.56; N, 12.96.

5-[(2,3-Diacetoxy-1-propoxy)methyl]-1-methyluracil (**4**).

A mixture of **3** (2.16 g, 0.01 mole), ammonium sulfate (0.1 g), and 1,1,1,3,3,3-hexamethyldisilazane (HMDS) (15 ml) was heated to reflux for 4 hours, during which the suspension became a homogenous solution. Excess HMDS was then removed *in vacuo* and the resulting syrup was dissolved in dry acetonitrile (30 ml). Methyl iodide (3 ml) was added to the solution and the mixture was stirred at room temperature for 24 hours. The mixture was then evacuated *in vacuo* and the residue was dissolved in methanol and neutralized with Amberlite IR-45 ($RNH_3^+OH^-$). The mixture was filtered and the filtrate was evaporated to a syrup, which was dissolved in dry pyridine (20 ml) and cooled in an ice-water mixture. Acetic anhydride (10 ml) was added to the solution and stirred at room temperature for 15 hours. The reaction mixture was then poured into an ice-water mixture. After decantation and washing with water, the resulting syrup was triturated with ethanol and then evaporated to dryness, which was separated on a silica gel column using chloroform-methanol (20:1) as the eluent. Due to the incomplete separation, further separation on preparative silica gel plates was required using chloroform-methanol (10/1) as the eluting solvent to give **4** (0.66 g, 21%) from the upper band, mp 123-125°; ^{1}H nmr (dimethylsulfoxide-d_6): δ 2.01 (s, 3H, CH_3CO_2), 2.02 (s, 3H, CH_3CO_2), 3.24 (s, 3H, N-CH_3), 3.55 (d, 2 H, -CH_2-O-, J = 5.2 Hz), 4.05-4.20 (m, 4H, AcO-CH_2 and benzylic), 5.05 (m, 1H, CH), 7.68 (s, 1H, aromatic), and 11.30 (b, 1H, NH, exchangeable).

Anal. Calcd. for $C_{13}H_{18}N_2O_7$: C, 49.68; H, 5.77; H, 8.91. Found: C, 49.77; H, 5.79; N, 8.88.

From the lower band 0.53 g of 1-methyl-5-(methoxymethyl)uracil (**5**) was obtained, mp 134-136°; uv λ max 268 nm (*p*H 1-7) and 266 (*p*H 13); ^{1}H nmr (chloroform-d): δ 3.38 (s, 3H, O-CH_3), 3.43 (s, 3H, N-CH_3), 4.21 (s,

2H, benzylic), 7.24 (s, 1H, aromatic), and 9.05 (b, 1H, NH, exchangeable).

Anal. Calcd. for $C_8H_{10}N_2O_4$: C, 49.41; H, 5.92; N, 16.46. Found: C, 49.20; H, 5.94; N, 16.41.

1-Methyl-5-(methoxymethyl)uracil (**5**) from **1**.

A mixture of 5-hydroxymethyluracil (1.42 g, 0.01 mole), ammonium sulfate (0.1 g) and HMDS (20 ml) was heated to reflux for 4 hours. The solution was then evaporated to a syrup and dissolved in dry acetonitrile (20 ml). Methyl iodide (2 ml) was added, and the mixture was stirred at room temperature for 24 hours. The solvent and excess methyl iodide were removed *in vacuo* and the residue was dissolved in methanol (100 ml), neutralized with Amberlite IR-45 ($RNH_3^+OH^-$), filtered, and the filtrate was evaporated. Acetic anhydride (5 ml) was added to the solid and the mixture was stirred at room temperature for 24 hours. The mixture was then poured into an ice-water mixture (30 ml). The resulting solid was separated and chromatographed on preparative silica gel plates using chloroform-methanol (20:1) to give a white crystalline product (0.9 g, 53%) after evaporation of the solvents, which was identical in all aspects with the sample isolated from the methylation of **3** to **6**.

5-[(2,3-Dihydroxy-1-propoxy)methyl]-1-methyluracil (**6**).

A mixture of diacetyl compound **4** (0.25 g) and methanolic ammonia (5 ml) was stirred at room temperature for 24 hours. After stirring, the solvent was evacuated to a syrup, which was redissolved in absolute ethanol and cooled in the refrigerator overnight. The resulting white precipitate was filtered and washed with cold ethanol, and then dried over phosphorus pentoxide to give 0.13 g (71%), mp 140-142°; uv: λ max 269 nm (*p*H 1-7) and 265 (*p*H 13); ^{1}H nmr (dimethylsulfoxide-d_6); δ 3.24 (s, 3H, N-CH_3), 3.32-3.41 (m, 5H, CH_2-CH-CH_2), 4.10 (s, 2H, benzylic), 4.60 (t, 1H, CH_2OH, J = 5.6 Hz, exchangeable), 4.65 (d, 1H, CH-OH, J = 4.4 Hz, exchangeable), 7.66 (s, 1H, aromatic), and 11.25 (b, 1H, NH, exchangeable).

Anal. Calcd. for $C_9H_{14}N_2O_5$: C, 46.95; H, 6.13; N, 12.17. Found: C, 47.06; H, 6.17; N, 12.12.

1,3-Dimethyl-5-[(2,3-dihydroxy-1-propoxy)methyl]uracil (**7**).

A mixture of **3** (2.16 g) and *N,N*-dimethylformamide dimethylacetal (20 ml) was refluxed for 3 hours. The mixture was evacuated *in vacuo* to give a pale yellow syrup, which was chromatographed on a vacuum flash silica gel column using a mixture of chloroform-methanol (10:1) as the eluting solvent to give a white solid (1.42 g, 58%) after evaporation of the solvents, mp 102-104°; uv: λ max 267 nm (*p*H 1-13); ^{1}H nmr (dimethylsulfoxide-d_6): δ 3.31 (s, 3H, N-CH_3), 3.32 (s, 3H, N-CH_3), 3.32 (s, 3H, N-CH_3), 3.30-3.65 (m, 5H, CH_2-CH-CH_2-), 4.11 (s, 2H, benzylic), 4.47 (t, 1H, CH_2-OH, exchangeable), 4.63 (d, 1H, CH-OH, exchangeable), 7.73 (s, 1H, aromatic).

Anal. Calcd. for $C_{10}H_{16}N_2O_5$: C, 49.18; H, 6.60; N, 11.47. Found: C, 49.22; H, 6.64; N, 11.45.

5-[(2,3-Diacetoxy-1-propoxy)methyl]-*N*^{2}-acetylisocytosine (**8**).

A mixture of **7** (4.88 g, 0.02 mole) and free guanidine (prepared from 19.1 g of guanidine hydrochloride and 300 ml of 1*N* sodium ethoxide solution) was heated at 90-100° for 1 hour under nitrogen. The reaction mixture was cooled and dissolved in water (40 ml), which was applied on an Amberlite IRC-50 column (2.5 x 20 cm) and eluted with water until no more uv absorbing material appeared. The eluent was evaporated to a solid, ethanol was added, triturated, and then the resulting precipitate was filtered to obtain a white solid (1.7 g). To a suspension of the above solid in pyridine (8 ml), acetic anhydride (3 ml) was added while cooling in an ice-water mixture. After stirring 24 hours, the mixture was poured into an ice-water mixture, extracted with chloroform (30 ml x 3), dried (magnesium sulfate), filtered, and the filtrate was evaporated to dryness to give 0.63 g . An analytical sample was obtained from preparative silica gel tlc plates using chloroform-methanol (10/1) as the eluent. ^{1}H nmr (dimethylsulfoxide-d_6): δ 2.00 (s, 3H, CH_3CO), 2.02 (s, 3H, CH_3CO), 2.15 (s, 3H, $NHCOCH_3$), 3.59 (d, 2H, CH_2O, J = 5.3 Hz), 4.10-4.30 (m, 4H, AcO-CH_2 and benzylic), 5.10 (m, 1H, CH), 7.76 (s, 1H, aromatic), and 11.75 (b, 1H, NH).

Anal. Calcd. for $C_{14}H_{19}N_3O_7$: C, 49.27; H, 5.61; N, 12.31. Found: C, 49.36; H, 5.66; H, 12.28.

5-[(2,3-Dihydroxy-1-propoxy)methyl]isocytocine (**9**).

A mixture of **8** (0.2 g) and methanolic ammonia (5 ml) was stirred at room temperature for 15 hours. The solvent was evacuated *in vacuo* and the residue was recrystallized from methanol to give white crystals (80 mg, 64%), mp 135-136°; uv: λ max 258 nm (*p*H 1), 287 (*p*H 7), 275 (*p*H 13); ^{1}H nmr (dimethylsulfoxide-d_6): δ 3.20-3.60 (m, 5H, CH_2CHCH_2), 4.11 (s, 2H, benzylic), 6.74 (b, 2H, NH_2, exchangeable) and 7.56 (s, 1H, aromatic).

Anal. Calcd. for $C_8H_{13}N_3O_4$: C, 44.65; H, 6.09; N, 19.53. Found: C, 44.74; H, 6.13; N, 19.46.

5-[(2-Hydroxyethoxy)methyl]isocytocine (**10**).

To a solution of **9** (0.86 g, 0.004 mole) in water (20 ml), sodium periodate (0.86 g, 0.004 mole) was added portionwise, during which the temperature was maintained at 20-25° in an ice-water bath, and the mixture was stirred at room temperature for 2 hours. The mixture was then poured into ethanol (20 ml). The resulting white precipitate was filtered off. The filtrate was evaporated to a syrup, which was dissolved in water (20 ml) and sodium borohydride solution (0.8 g in 20 ml of water) was added dropwise. After stirring for one hour at room temperature, the solution was neutralized with Dowex 50 (H^+), filtered and the filtrate was evaporated to a white solid. Pyridine (12 ml) and acetic anhydride (4 ml) were added to the solid and the mixture was stirred for 15 hours. The mixture was poured into an ice-water mixture (30 ml) and the resulting solid was obtained by filtration and chromatographed on a silica gel column using chloroform-methanol (20:1) as the eluent to give a white solid (0.27 g, 25%), mp 176-178°; ^{1}H nmr (dimethylsulfoxide-d_6): δ 2.01 (s, 3H, $NHCOCH_3$), 2.15 (s, 3H, CH_3CO_2), 3.62 (t, 2H, CH_2CH_2, J = 4.84 Hz), 4.13 (t, 2H, CH_2CH_2), 4.23 (s, 2H, benzylic), 7.75 (s, 1H, aromatic), and 11.80 (b, 1H, NH).

Anal. Calcd. for $C_{11}H_{15}N_3O_5$: C, 49.07, H, 5.62; N, 15.61. Found: C, 48.96; H, 5.62; N, 15.57.

The above acetylated compound (0.08 g) was treated with methanolic ammonia (2 ml) for 24 hours. The solvent was evacuated and the residue was recrystallized from ethanol to give **10** as white crystals (41 mg, 73%). This compound was identical in all respects with the previously reported compound [13].

5-[(2-Hydroxyethoxy)methyl]uracil (**11**).

To a stirred solution of **3** (2.16 g, 0.01 mole) in water (50 ml), sodium periodate (2.14 g, 0.01 mole) was added portionwise, during which the temperature was kept at 20-25° in an ice-water bath. After stirring for 2 hours at the temperature, the mixture was poured into ethanol (50 ml) and the resulting white precipitate was filtered off and the filtrate was evaporated to a solid. Water (50 ml) was added to the solid and sodium borohydride (2 g) in water (50 ml) was added dropwise. After 1 hour of stirring at room temperature, the solution was neutralized with Dowex 50 (H^+). The resin was removed by filtration and the filtrate was evaporated to a white solid containing boric acid. The latter was removed as methylborate by coevaporation with methanol several times. The residue was triturated with ethanol and filtered to give crude **11** (1.3 g, 60%). The crude compound (0.25 g) was suspended in a mixture of pyridine (4 ml) and acetic anhydride (1 ml) and the mixture was stirred in an ice-water bath for 4 hours and then at room temperature for 15 hours. The mixture was then poured into an ice-water mixture. The resulting solid was filtered and recrystallized from ethanol to give white crystals (0.15 g, 50%), mp 188-190°; ^{1}H nmr (dimethylsulfoxide-d_6); δ 2.01 (s, 3H, CH_3CO), 3.57 (t, 2H, CH_2CH_2, J = 4.85 Hz), 4.10 (t, 2H, CH_2CH_2), 4.12 (s, 2H, benzylic), 7.40 (s, 1H, aromatic), and 11.05 (b, 2H, NH).

Anal. Calcd. for $C_9H_{12}N_2O_5$: C, 47.37; H, 5.30; N, 12.28. Found: C, 47.16; H, 5.33; N, 12.20.

The above acetylated compound (80 mg) was treated with methanolic ammonia (3 ml) and stirred at room temperature for 24 hours. The sol-

vent was evaporated to give a white crystalline product which was recrystallized from absolute ethanol to give **11** (51 g, 78%). This compound was identical in all respects with the previously reported data [13].

REFERENCES AND NOTES

[1] This work was presented at the 10th International Congress of Heterocyclic Chemistry, Waterloo, Ontario, Canada, Aug., 1985, Paper No. P9-265.

[2] G. B. Elion, P. A. Furman, J. A. Fyfe, P. de Miranda, L. Beauchamp, and H. J. Schaeffer, *Proc. Nat. Acad. Sci. USA*, **74**, 5716 (1977).

[3] H. J. Schaeffer, L. Beauchamp, P. de Miranda, G. Elion, D. J. Bauer, and P. Collins, *Nature* (London), **272**, 583 (1978).

[4] The Proceedings of a Symposium on Acyclovir, *Am. J. Med.*, **73**, (1H) (1982).

[5] C. K. Chu and S. J. Cutler, *J. Heterocyclic Chem.*, **23**, 289 (1986).

[6] J. C. Martin, C. A. Dvorak, D. F. Smee, T. R. Matthews, and J. P. H. Verheyden, *J. Med. Chem.*, **26**, 759 (1983).

[7] K. K. Ogilvie, U. O. Cheriyan, B. K. Radatus, K. O. Smith, K. S. Galloway, and W. L. Kennell, *Can. J. Chem.*, **60**, 3005 (1982).

[8] A. K. Field, M. E. Davies, C. DeWitt, H. C. Perry, R. Liou, J. I. Germershausen, J. D. Karkas, W. T. Ashton, D. B. R. Johnson, and R. L. Tolman, *Proc. Nat. Acad. Sci.*, USA, **80**, 4139 (1983).

[9] H. J. Schaeffer, In "Nucleosides, Nucleotides and Their Biological Applications", J. L. Rideout, D. W. Henry, and L. M. Beaucham, eds, Academic Press, New York, 1983, pp 1-17.

[10] Private communication with Dr. R. L. Tolman of Merck Sharp & Dohme.

[11] T.-S. Lin and M.-C. Lin, *Tetrahedron Letters*, **25**, 905 (1984).

[12] W. T. Ashton, L. F. Canning, G. F. Reynolds, R. L. Tolman, J. D. Karkas, R. Liou, M.-E. M. Davies, C. M. DeWitt, H. C. Perry, and A. K. Field, *J. Med. Chem.*, **28**, 926 (1985).

[13] C. K. Chu, *J. Heterocyclic Chem.*, **21**, 9 (1984).

[14] S. Y. Melbnik, T. D. Miniker, I. V. Yartseva, T. P. Nedoresova, G. I. Potapova and M. N. Preobrazhenskaya, *Bioorg. Khim.*, **9**, 1395 (1983).

[15] R. E. Cline, R. M. Fink, and K. Fink, *J. Am. Chem. Soc.*, **81**, 2521 (1959).

[16] C. K. Chu, I. Wempen, K. A. Watanabe and J. J. Fox, *J. Org. Chem.*, **41**, 2793 (1976).

[17] We are grateful to Dr. Raymond F. Schinazi, Emory University School of Medicine and Veterans Administration for antiviral screening.