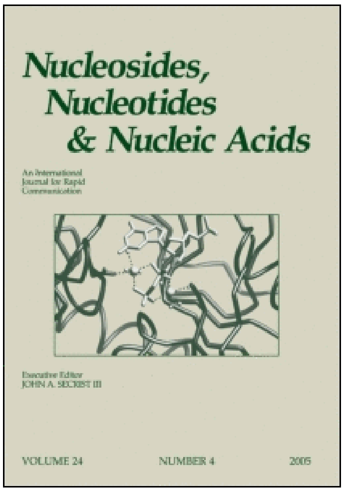

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5'-*C*-Chain-Extended Adenosine Derivatives Related to Sinefungin. Synthesis and Biological Activity

John A. Secrist III[a]; Ratnakar R. Talekar[a]
[a] Kettering-Meyer Laboratories, Southern Research Institute, Birmingham, AL

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5'-*C*-CHAIN-EXTENDED ADENOSINE DERIVATIVES RELATED TO SINEFUNGIN. SYNTHESIS AND BIOLOGICAL ACTIVITY.

John A. Secrist III* and Ratnakar R. Talekar

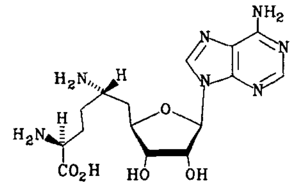
Kettering-Meyer Laboratories, Southern Research Institute,
P. O. Box 55305, Birmingham, AL 35255-5305

Summary. The synthesis of four 5'-chain-extended adenosine derivatives is described. Three of these nucleosides were prepared by manipulation of lactone **6**, which is derived from N^6-benzoyl-2',3'-*O*-isopropylideneadenosine 5'-aldehyde by chain extension through Wittig chemistry. Functional group manipulation allowed introduction of the C-6' amide and amine groups, and the chain terminal amino or hydroxyl group. The fourth compound was prepared in five steps from the natural product sinefungin. Antiviral data and certain enzyme inhibition data are presented on the target compounds.

INTRODUCTION

The nucleoside antibiotic sinefungin (1, SF), isolated from *Streptomyces griseolus*,[1] has attracted significant attention because of its interesting structure as well as its potent biological activity as an inhibitor of methyltransferase enzymes.[2] SF is an amino acid-containing nucleoside that has an ornithine residue attached through the terminal carbon to C-5' of adenosine, thereby producing a decose as the carbohydrate moiety of the nucleoside. SF bears a strong structural resemblance to S-adenosylmethionine (AdoMet), with the methylated sulfur in AdoMet being replaced by a $-CH(NH_2)-$ group. Of particular interest is that the spatial orientation of this amino group in SF is identical to the methyl in AdoMet.[3] SF has been found to have a variety of activities both in vitro and in vivo including antifungal,[4] antiviral,[2,5-8] and antiparasitic activities.[2,9-18] These biological activities may be attributed to the aforementioned inhibition of a variety of methyltransferases utilizing AdoMet, many examples of which exist,[19-31] or perhaps to the inhibition by SF of an enzyme on the polyamine pathway.[32] A series of synthetic efforts directed towards either SF or analogues have been reported,[33-38] and a variety of interesting chemical approaches have been developed.

Our initial efforts in this area focused on the utilization of the Wittig reaction to carry out chain extension at C-5' of a suitably protected adenosine 5'-aldehyde.[37] The Wittig reagents that we prepared were designed to allow for manipulation at C-6′ (where the amino group of SF resides) as well as at the terminus of the carbohydrate chain. Herein we describe the extension of this effort to the synthesis of several chain-extended adenosines (**2**-**5**) that were designed as potential inhibitors of polyamine biosynthesis, and some biological data on these compounds.[39] The effects of these compounds on the enzymes of the polyamine pathway, especially spermine synthase, have already been described.[40]



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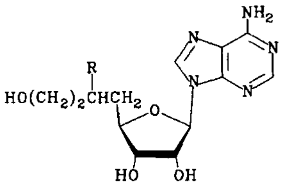
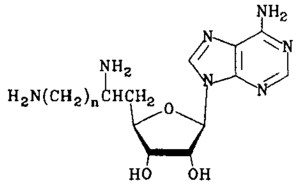


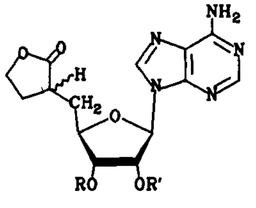
2) R = $CONH_2$ (6R,S)
3) R = NH_2 (6R,S)



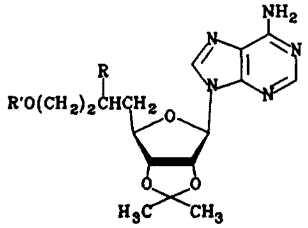
4) n = 2 (6R,S)
5) n = 3 (6S)

CHEMISTRY

The starting material for compounds **2**-**4** is the lactone **6**, which is prepared from adenosine as described[37] previously through a series of steps utilizing a Wittig reaction as the key transformation. Lactone ring opening with ammonia followed by protection of the generated side-chain hydroxyl group of **8** as a tert-butyldiphenylsilyl ether provided **9**. Rearrangement of the amide with [bis(trifluoroacetoxy)iodo]benzene[41] to afford **10** followed by deblocking with aqueous hydrochloric acid provided 6′-amino compound **3**. Removal of the isopropylidene group from **6** followed by lactone opening of **7** with ammonia readily provided the corresponding 6′-carboxamide **2**.



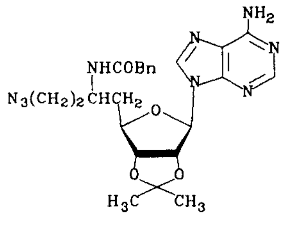
6) R,R′ = $C(CH_3)_2$
(ca. 2:1 mixture of R and S at C-6′)
7) R,R′ = H



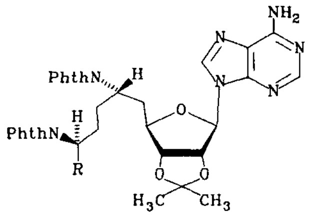
8) R = $CONH_2$, R′ = H
9) R = $CONH_2$, R′ = TBDPS
10) R = NH_2, R′ = TBDPS

The two diamino compounds **4** and **5** were prepared by very different routes. The diamine **4**, with a propyl spacer between the amino groups, was generated from urethane azide **11**, which was prepared as previously described,[37] the key step being a Curtius rearrangement to incorporate the 6′ carbon-nitrogen bond. Hydrogenolysis of **11** in aqueous formic acid provided **4**, isolated as the formate salt.

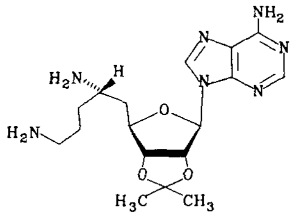
The diamine **5**, with a butyl spacer between the amino groups, was prepared from sinefungin (**1**)[42] in five steps. Sinefungin was first blocked as the 2′,3′-*O*-isopropylidene derivative, and then treated with *N*-carboethoxyphthalimide in order to generate **12**, with the two basic side-chain amines blocked. Irradiation of **12** in a Pyrex tube with a medium-pressure Hanovia lamp[43] resulted in two products, one of which was the desired decarboxylated bis-phthalimide **13**. The other product, which had lost a phthalimide group, was not fully characterized. Treatment of **13** with hydrazine produced **14**, which upon treatment with aqueous hydrochloric acid produced the desired diamine **5** as the hydrochloride salt.



11



12) R = CO_2H
13) R = H



14

Compounds **2-4** were isolated and characterized as mixtures at C-6′, the result of starting lactone **6** being an inseparable mixture at this center. We were unable to obtain any physical separation of the isomers for any of the three compounds by either tlc or hplc, though some ^1H NMR signals showed the presence of the two isomers. Compound **5**, derived from sinefungin, was, of course, a single compound. Purification of the three 6′-amino compounds **3-5** proved to be difficult, and NMR data were also difficult to obtain. For some reason these compounds hold onto metal ions tightly, and apparently a trace amount of a paramagnetic metal ion, which we were unable to remove in spite of extensive efforts, broadened the ^1H NMR spectra, and also prevented us from obtaining ^{13}C NMR spectra. It was, however, possible to obtain reasonable analytical data on the compounds.

BIOLOGICAL EVALUATION

As mentioned above, compounds **2-5** were examined for their effects on polyamine synthesis.[40] The two diamino compounds **4** and **5** were found to be moderate inhibitors of *E. coli* AdoMet decarboxylase, but none of the four inhibited the enzyme from rat prostate. None of the four was a potent inhibitor of rat prostate spermidine synthase. The three 6′-amino compounds **3-5** were all inhibitors of rat brain spermine synthase, with **4** being the most potent inhibitor. In SV-3T3 cells, **4** was found to reduce the spermine content, although there was a compensatory increase in spermidine levels.

Antiviral activity was assessed in several systems. In a standard Herpes simplex type 1 assay in Vero cell monolayers assessing the inhibition of cytopathic effects, compounds **2-5** were inactive.

The effects of **2-4** on vaccinia virus plaque production in mouse fibroblast cells (clone L929) were also examined.[6] Neither **2** nor **3** showed any effects up to 800 μM, either antiviral or cytotoxic, while **4** showed weak activity, with an MIC_{50} of *ca.* 700 μM, and some cytotoxicity at the highest concentration examined. In agreement with these observations, no cytotoxicity was seen toward L1210 cells in a suspension culture assay for any of the compounds up to concentrations in excess of 100 μM. Additionally, none of the four compounds was inhibitory toward vaccinia virion mRNA (guanine-7) methyltransferase, as determined in the laboratory of Dr. R. T. Borchardt.

On the basis of the potent inhibitory activity of sinefungin toward the enzyme 1-aminocyclopropanecarboxylic acid synthase,[44-46] which utilizes AdoMet as substrate, compounds **2-4** were examined as inhibitors of this enzyme by Dr. P. K. Chiang. None of these compounds was a potent inhibitor, with **4** showing an I_{50} of 660 μM and **3** an I_{50} of 800 μM.[48] The amide **2** was not an inhibitor at the highest levels examined. These results lend further support to the observation that the carboxylic acid group in sinefungin is an important contributor to its inhibitory properties toward this enzyme.

EXPERIMENTAL SECTION

Infrared spectra were recorded on a Nicolet 10MX infrared spectrometer. ^1H NMR and ^{13}C NMR spectra were recorded on a Nicolet NMC 300NB spectrometer operating at 300.635 MHz (^1H) or 75.16 MHz (^{13}C). Chemical shifts are expressed in parts per million downfield from tetramethylsilane. Microanalyses were performed by the Molecular Spectroscopy Section of Southern Research Institute. HPLC data were recorded on a Hewlett-Packard HP 3380A chromatograph at 254 nm using a Partisil 10 SCX cation-exchange column, 20 min linear gradient, 0.1-0.5 *M* $NH_4H_2PO_4$, pH 4.4, 1.5 mL/min. Mass spectra were recorded on a Varian MAT 311A mass spectrometer in the fast atom bombardment (FAB) mode.

The vaccinia virus plaque assay procedure was performed as described in reference 6, except that the incubation temperature was 37 °C and the vaccinia virus strain used was the Lederle CA strain.

9-[6-*C*-(*R,S*)-Carboxamido-5,6,7-trideoxy-β-D-*ribo*-octofuranosyl]adenine (2). A solution of lactone **6**[37] (375 mg, 1 mmol) in 44% aqueous formic acid (10 mL) was allowed to stand at room temperature for 24 h, at which time TLC analysis indicated the complete loss of starting material. The solution was lyophilized and the product isolated by preparative TLC (9:1 $CHCl_3$-CH_3OH) to afford 267 mg of **7** (80%), FABMS *m/z* 336 $(M + H)^+$.

The lactone **7** was dissolved in 30 mL of ethanol that had been saturated with ammonia gas at 0 °C, and allowed to stand at 5-10 °C for five days, by which time TLC analysis indicated the presence of one new compound. The solvent was removed under reduced pressure, and the residue was dried (0.1 mm Hg, 24 h) to afford **2** as a colorless foam, 267 mg (95%); IR (KBr) 3344, 3340, 3335, 3271, 3193, 3186, 1653, 1601, 1576, 1047 cm^{-1}; ^1H NMR (Me_2SO-d_6) δ 1.4-2.0 (m, 4, 5′-CH_2, 7'-CH_2), 2.40 (m, 1, H-6'), 3.35 (m, 2, 8'-CH_2), 3.81, 3.90 (2 m, 1, H-4'), 4.00 (m, 1, H-3'), 4.38 (br s, 1, OH), 4.62, 4.73 (2 apparent t, 1, H-2', J = 2 Hz), 5.27, 5.48 (2 br s, 2, OH), 5.87 (2 d, 1, H-1', J = 2 Hz), 6.80 (br s, 1, NH), 7.26 (br s, 1, NH), 7.28 (br s, 2, 6-NH_2), 8.15, 8.16 (2 s, 1, H-2 or H-8), 8.34 (s, 1, H-2 or H-8); ^{13}C NMR (Me_2SO-d_6) δ 34.90, 36.24 (C-7'), 58.84 (C-8'), 72.84, 73.15, 73.40, 73.67 (C-2',3'), 81.77 and 82.39 (C-1' or C-4'), 87.45 (C-1' or C-4'), 119.11 and 119.20 (C-5), 139.60 and 139.90 (C-8), 149.34 and 149.41 (C-4), 152.15 (C-2), 155.97 (C-6), 176.36 and 176.68 ($CONH_2$), C-5' and C-6' hidden by solvent; exact mass (FAB) calcd 353.157 (M + H)$^+$; found, 353.158. The NMR spectra showed the presence of the appropriate amounts of ethanol and water. *Anal.* Calcd for $C_{14}H_{30}N_6O_5 \cdot 0.5EtOH \cdot 0.8H_2O$: C, 46.22; H, 6.36; N, 21.56. Found: C, 46.23; H, 6.17; N, 21.33.

9-[6(*R*,*S*)-Amino-5,6,7-trideoxy-β-D-*ribo*-octofuranosyl]adenine (3). A solution of lactone 6[37] (1.875 g, 5 mmol) in ethanolic ammonia (100 mL, saturated at 1 °C) was allowed to stand at 5 °C for five days, at which time TLC analysis (9:1 CH_2Cl_2-CH_3OH) indicated the presence of only one material, different from **6**. Solvent was removed under reduced pressure to leave the intermediate hydroxy amide as a foam, 1.89 g (96%), that was used directly in the next step: FABMS 393 (M + H)$^+$, 258 (sugar)$^+$; IR (KBr) 3391, 3344, 3335, 3193, 1665, 1645, 1600, 1375, 1210, 1080 cm^{-1}; ^1H NMR (Me_2SO-d_6) δ 1.31, 1.54 (2 s, 2, $C(CH_3)_2$), 1.7-2.0 (m, 4, 5'-CH_2, 7'-CH_2), 3.40 (m, 1, H-6'), 4.02 (m, 2, 8'-CH_2), 4.3 (m, 1, H-4'), 4.88 (m, 1, H-3'), 5.44 (m, 1, H-2'), 6.11 (m, 1, H-1'), 7.10, 7.28 (2 s, 2, $CONH_2$), 7.36 (br s, 2, Ad-NH_2), 8.17, 8.32, 8.34 (3 s, 2, H-2, H-8).

The hydroxy amide (630 mg, 1.6 mmol) was dissolved in DMF (5 mL) and stirred with imidazole (109 mg, 1.6 mmol) and *tert.*-butyldiphenylchlorosilane (439 mg, 1.6 mmol). After 24 h at room temperature, TLC analysis (95:5 CH_2Cl_2-CH_3OH) indicated one new, faster-moving material. Solvent was removed under reduced pressure and the residue was chromatographed over silica gel, eluting with 95:5 CH_2Cl_2-CH_3OH to afford 964 mg (95%) of **7**, which was used directly: FABMS *z/e* 631 (M + H)$^+$, 496 (sugar)$^+$; IR (KBr) 3375, 3175, 2930, 2855, 1675, 1642, 1598, 1575, 1427, 1374, 1210, 1112, 1085, 703 cm^{-1}.

To a solution of the amide **7** (1.22 g, 1.94 mmol) in 50% aqueous DMF (8 mL) was added [bis(trifluoroacetoxy)iodo]benzene (886 mg, 2.06 mmol), and the reaction mixture was stirred for 2 h. Pyridine (2 equiv) was added, and the mixture stirred for an additional 2 h. Solvent was removed under reduced pressure, and the residue was chromatographed over silica gel to afford 1.1 g (94%) of the amino compound **8**: FABMS *z/e* 603 (M + 1)$^+$, 468 (sugar)$^+$; IR (KBr) 3340 (br), 3185, 2930, 2860, 1682, 1643, 1206, 1181, 1156, 1135, 1112, 1106, 1089, 702 cm^{-1}; ^1H NMR (Me_2SO-d_6, major isomer) δ 0.86 (s, 9, $C(CH_3)_3$), 1.32, 1.55 (2 s, 6, $C(CH_3)_2$), 1.6-2.0 (m, 4, 5'-CH_2, 7'-CH_2), 3.15 (m, 1, H-6'), 3.7 (m, 8'-CH_2, NH), 4.34 (m, 1, H-4'), 4.90 (m, 1, H-3'), 5.43 (m, 1, H-2'), 6.13 (d, 1, H-1', J = 1 Hz), 7.32 (br s, 2, Ad-NH_2), 7.2-7.7 (m, aromatic), 8.15, 8.33 (2 s, 2, H-2, H-8).

The silyl amine **8** (353 mg, 0.59 mmol) was dissolved in a minimum amount of THF and then 0.1 *N* HCl was added (25 mL). The solution was allowed to stand at room temperature for 10-14 days. The acid was neutralized with Amberlite IR-45 (2 g), and the resin filtered off and washed with water. The filtrate was lyophilized to afford 178 mg of **3** as a hygroscopic foam homogeneous by TLC. HPLC purity, 97.4%, retention time 8.9 min; IR (KBr) 3319, 3183, 1644, 1600, 1575, 1480, 1440, 1330, 1250, 1050, 725 cm^{-1}; ^{1}H NMR (Me_2SO-d_6) δ 1.74, 2.04 (2 br s, 4, 5'-CH_2, 7'-CH_2), 3.36, 3.54, 4.11, 4.55, 4.65, 5.0, 5.5 (br, -CH_2O-, -CHO-, -CHN-, OH, NH), 5.89 (d, 1, H-1'), 7.30 (br s, 2, Ad-NH_2), 8.18, 8.37 (2 br s, 2, H-2, H-8); ^{13}C spectrum was again not obtainable; exact mass (FAB) calcd $(M + H)^+$ 325.162; found, 325.165. We were unable to obtain satisfactory microanalytical data on this compound.

9-[6(*R,S*),8-Diamino-5,6,7,8-tetradeoxy-β-D-*ribo*-octofuranosyl]adenine (4). Purified urethane azide **11**[37] (343 mg, 0.655 mmol) was dissolved in 50% aqueous formic acid (40 mL) and shaken in a Parr apparatus at 50 psi of hydrogen for 5 days over 10% Pd/C (245 mg). The catalyst was filtered off and washed with water (10 mL), and the filtrate lyophilized. The residue was taken up in water (10 mL) and lyophilized twice more, and the resulting pale, yellow foam was dried (0.1 mm Hg) for 24 h. The product, a hygroscopic foam, was a hydrated formate salt weighing 277 mg (75% based upon analytical data; IR (KBr) 3505, 3411, 3398, 3393, 3388, 3380, 1630, 1598, 1342, 1332 cm^{-1}; ^{1}H NMR (Me_2SO-d_6) δ 1.9 (br s, 4, 5'-CH_2, 7'-CH_2), 2.92, 3.20, 4.10, 4.60 (br, 6, H-2',3',4',6', 8'-CH_2), 5.88 (s, 1, H-1'), 8.17, 8.34 (2 s, 2, H-2, H-8), 7.32, 8.40 (2 br s, 4, 2 NH_2), 6.74 (br, HCO_2H, NH); exact mass (FAB) calcd 324.178 $(M + H)^+$; found 324.176. We were unable to record a ^{13}C NMR spectrum. *Anal.* Calcd for $C_{13}H_{21}N_7O_3 \cdot 4HCO_2H \cdot 3H_2O$: C, 36.36; H, 6.24; N, 17.46. Found: C, 36.17; H, 6.00; N, 17.29.

9-[6(*S*),9-Diamino-5,6,7,8,9-pentadeoxy-β-D-*ribo*-nonfuranosyl]adenine (5). A solution of sinefungin (**1**, 1.143 g, 3 mmol)[42] and perchloric acid (70-72%, 1 mL) in acetone was stirred for 2 h in the presence of 4A molecular sieves (1 g). The solution was filtered, neutralized with triethylamine, and evaporated to dryness. The residue was dissolved in DMF (20 mL) and stirred at room temperature with *N*-carboethoxyphthalimide (1.315 g, 6 mmol) for 24 h. After acidification with glacial acetic acid the reaction mixture was evaporated to dryness. Isolation of the bis phthalimido compound **12** was accomplished by column chromatography, eluting with 9:1 CH_2Cl_2-CH_3OH, to afford 1.064 g (52%) of reasonably pure material: FABMS 682 $(M + H)^+$.

A solution of **12** (569 mg, 0.83 mmol) in acetone (150 mL) was irradiated for 4 h in a Pyrex tube using a medium-pressure Hanovia lamp. TLC analysis showed the absence of starting material and the presence of two products. Separation was accomplished by thick-layer chromatography, eluting with CH_2Cl_2-CH_3OH (7:3) to yield 230 mg (43%) of the decarboxylated material **13**. The other product, which had lost a phthalimide group, was not fully characterized. Spectral data for **13**: IR (KBr) 3350, 3175, 2980, 2935, 1772, 1710, 1635, 1596, 1397, 1372, 1330, 1209, 1076, 721 cm^{-1}; FABMS *z/e* 638 $(M + H)^+$, 503 $(\text{sugar} + 1)^+$; ^{1}H NMR (Me_2SO-d_6) δ 1.07 and 1.32 (2 s, 6,

$C(CH_3)_2$), 1.42, 1.61, 1.91 (3 m, 4, 7'-CH_2, 8'-CH_2), 2.10, 2.46 (2 m, 2, 5'-CH_2), 3.49 (m, 2, 9'-CH_2), 3.93 (m, 1, H-4'), 4.20 m, 1, H-6'), 4.75 (dd, 1, H-3', $J_{3',4'}$ = 4 Hz), 5.37 (dd, 1, H-2', $J_{2',3'}$ = 6 Hz), 6.03 (d, 1, H-1', $J_{1',2'}$ = 3 Hz), 7.32 (br s, 2, NH_2), 7.81 (m, 4, phthalimide), 8.15, 8.31 (2 s, 2, H-2, H-8); ^{13}C NMR (Me_2SO-d_6) δ 24.63, 24.80, 26.66, 29.16 ($C(CH_3)_2$, C-7', C-8'), 35.17, 36.75 (C-5', C-9'), 47.81 (C-6'), 82.64, 82.90, 83.53 (C-2',3',4'), 88.07 (C-1'), 113.36 ($C(CH_3)_2$), 118.98 (C-5), 122.78, 131.16, 131.42, 134.11, 134.20 (phthalimide aromatic carbons), 139.83 (C-8), 148.67 (C-4), 152.58 (C-2), 155.97 (C-6), 167.68, 168.00 (phthalimide carbonyls). Bis phthalimide compound **13** (310 mg, 0.486 mmol) was dissolved in methanol (15 mL) and treated with hydrazine (94 mg, 2.93 mmol), and the solution stirred at room temperature for 48 h, at which time TLC analysis ($CHCl_3$-EtOH-conc. NH_4OH, 3:2:1) showed one band, ninhydrin positive. The mixture was evaporated to dryness, and the residue dissolved in methanol and absorbed onto Amberlite CG-50 (100-200 mesh, 2 mL) in a column over 1 h. The column was washed with 10% aqueous methanol until no UV-absorbing material was detected. The diamino compound **14** was then eluted with 4 M NH_4OH, monitoring fractions by TLC. Appropriate fractions were lyophilized to a foam (175 mg, 94%) that showed one spot by TLC analysis; FABMS 378 $(M + H)^+$.

Removal of the isopropylidene group was accomplished by dissolving **14** (140 mg, 0.37 mmol) in 0.1 *N* HCl (30 mL) and allowing the solution to stand for seven days at room temperature. The acid was neutralized with Amberlite IR-45 ion-exchange resin (2.0 g). The resin was filtered off, washing with water, and the filtrate was lyophilized to afford 112 mg of the very hygroscopic hydrochloride salt **5**, which showed a single peak by HPLC analysis (retention time 15.4 min); IR (KBr) 3310 (br), 3175, 2900 (br), 1642, 1599, 1475, 1330, 1250, 1125, 1040 cm^{-1}; ^1H NMR (Me_2SO-d_6) δ 1.71, 2.07 (2 m, 6, 5'-CH_2, 7'-CH_2, 8'-CH_2), 2.77 (m, 2, 9'-CH_2), 3.22 (m, 1, H-6'), 4.01 (m, 1, H-3'), 4.21 (m, 1, H-4'), 4.66 (m, 1, H-2'), 5.98 (d, 1, H-1', $J_{1',2'}$ = 5 Hz), 8.24 (br, OH, NH), 8.67, 8.85 (2 s, 2, H-2, H-8); exact mass (FAB) calcd 338.194 $(M + H)^+$; found 338.196. The ^1H NMR spectrum was broadened, apparently by metal ions, and we were unable to get a ^{13}C NMR or microanalytical data.

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REFERENCES

1. a) Boeck, L. D.; Clem, G. M.; Wilson, M. M.; Westhead, J. E. *Antimicrob. Agents Chemother.*, **1973**, *3*, 49-56. b) Hamill, R. L.; Hoehn, M. *11th Annual ICAAC Meeting*, **1971**, Abstr. 21.
2. Suhadolnik, R. J. "*Nucleosides as Biological Probes*"; Wiley: New York, **1979**.
3. Nagarajan, R., private communication.
4. Gordee, R. S.; Butler, T. F. *J. Antibiotics*, **1973**, *26*, 466-470.
5. Nagarajan, R.; Chao, B.; Dorman, D. E.; Nash, S. M.; Occolowitz, J. L.; Schabel, A. *17th Annual ICAAC Meeting*, New York, **1977**, Abstr. 50.
6. Pugh, C. S. G.; Borchardt, R. T.; Stone, H. O. *J. Biol. Chem.*, **1978**, *253*, 4075-4077.
7. Long, W. K.; Fronko, G. E.; Lindemeyer, R. G.; Wu, B.; Henderson, E. E. *J. Virol.*, **1987**, *61*, 221-224.
8. Nagarajan, R. *U. S. Patent*, 4,158,056, 1979.
9. Neal, R. A.; Iwobi, M. V.; Robert-Gero, M. *C. R. Acad. Sci., Ser. 3*, **1989**, *308*, 485-488.
10. Ferrante, A.; Ljungstroem, I.; Lederer, E. *C. R. Acad. Sci., Ser. 3*, **1988**, *306*, 109-113.
11. Lawrence, F.; Robert-Gero, M. *Bull. Soc. Fr. Parasitol.*, **1986**, *4*, 193-196.
12. Nolan, L. L. *Antimicrob. Agents Chemother.*, **1987**, *31*, 1542-1548.
13. Paolantonacci, P.; Lawrence, F.; Nolan, L. L.; Robert-Gero, M. *Biochem. Pharmacol.*, **1987**, *36*, 2813-2820.
14. Bacchi, C. J.; Berens, R. L.; Nathan, H. C.; Klein, R. S.; Elegbe, I. A.; Rao, K. V. B.; McCann, P. P.; Marr, J. J. *Antimicrob. Agents Chemother.*, **1987**, *31*, 1406-1413.
15. Zweygarth, E.; Schillinger, D.; Kaufmann, W.; Roettcher, D. *Trop. Med. Parasitol.*, **1986**, *37*, 255-257.
16. Paolantonacci, P.; Lawrence, F.; Robert-Gero, M. *Antimicrob. Agents Chemother.*, **1985**, *28*, 528-531.
17. Neal, R. A.; Croft, S. L.; Nelson, D. J. *Trans. R. Soc. Trop. Med. Hyg.*, **1985**, *79*, 122-128.
18. Dube, D. K.; Mpimbaza, G.; Allison, A.C.; Lederer, E.; Rovis, L. *Am. J. Trop. Med. Hyg.*, **1983**, *32*, 31-33.
19. Fuller, R. W.; Nagarajan, R. *Biochem. Pharmacol.*, **1978**, *27*, 1981-1983.
20. Borchardt, R. T.; Eiden, L. E.; Wu, B.; Rutledge, C. O. *Biochem. Biophys. Res. Commun.*, **1979**, *89*, 919-924.
21. Borchardt, R. T.; Kuonen, D.; Huber, J. A.; Moorman, A. *Mol. Pharmacol.*, **1982**, *21*, 181-186.
22. Kreuzman, A. J.; Turner, J. R., Yeh, W.-K. *J. Biol. Chem.*, **1988**, *263*, 15626-15633.
23. Bauer, N. J.; Kreuzman, A. J.; Dotzlaf, J. E.; Yeh, W.-K. *J. Biol. Chem.*, **1988**, *263*, 15619-15625.
24. Ghosh, S. K.; Paik, W. K.; Kim, S. *J. Biol. Chem.*, **1988**, *263*, 19024-19033.
25. Billich, A.; Zocher, R. *Biochemistry*, **1987**, *26*, 8417-8423.

26. Amur, S. G.; Shanker, G.; Cochran, J. M.; Ved, H. S.; Pieringer, R. A. *J. Neurosci. Res.*, **1986**, *16*, 367-376.

27. Ferenz, H.-J.; Peter, M. G.; Berg, D. *Agric. Biol. Chem.*, **1986**, *50*, 1003-1008.

28. O'Leary, M.; Hansen, J. A.; Mirkin, B. L.; O'Dea, R. F. *Biochem. Pharmacol.*, **1983**, *32*, 2339-2343.

29. Gupta, A.; Jensen, D.; Kim, S.; Paik, W. K. *J. Biol. Chem.*, **1982**, *257*, 9677-9683.

30. Trivedi, L.; Gupta, A.; Ki Paik, W.; Kim, S. *Eur. J. Biochem.*, **1982**, *128*, 349-354.

31. McCammon, M. T.; Parks, L. W. *J. Bacteriol.*, **1981**, *145*, 106-112.

32. Bachrach, U.; Schnur, L. F.; El-On, J.; Greenblatt, C. L.; Pearlman, E.; Robert-Gero, M.; Lederer, E. *FEBS Lett.*, **1980**, *121*, 287-291.

33. Buchanan, J. G.; Flinn, A.; Mundill, P. H. C.; Wightman, R. H. *Nucleosides & Nucleotides*, **1986**, *5*, 313-323.

34. Mizuno, Y.; Tsuchida, K.; Tampo, H. *Chem. Pharm. Bull.*, **1984**, *32*, 2915-2924.

35. Geze, M.; Blanchard, P.; Fourrey, J. L.; Robert-Gero, M. *J. Am. Chem. Soc.*, **1983**, *105*, 7638-7640.

36. Moorman, A. R.; Martin, T.; Borchardt, R. T. *Carbohydr. Res.*, **1983**, *113*, 233-239.

37. Lyga, J. W.; Secrist III, J. A. *J. Org. Chem.*, **1983**, *48*, 1982-1988.

38. Mock, G. A.; Moffatt, J. G. *Nucleic Acids Res.*, **1982**, *10*, 6223-6234.

39. A brief preliminary account of this work has been presented: Montgomery, J. A.; Secrist III, John A. *Biol. Methylation Drug., (Proc. Symp.)*, Borchardt; R. T.; Creveling, C. R.; Ueland, P. M., Eds., Humana, Clifton, NJ, **1986**, p. 409-416.

40. Pegg, A. E.; Coward, J. K.; Talekar, R. R.; Secrist III, J. A. *Biochemistry*, **1986**, *25*, 4091-4097.

41. Loudon, G. M.; Radhakrishna, A. S.; Almond, M. R.; Blodgett, J. K.; Boutin, R. H. *J. Org. Chem.*, **1984**, *49*, 4272-4276.

42. Sinefungin was generously supplied by Dr. R. Nagarajan of Eli Lilly and Company.

43. Sato, Y.; Nakai, H.; Mizoguchi, T.; Kawanishi, M.; Hatanaka, Y.; Kanaoka, Y. *Chem. Pharm. Bull.*, **1982**, *30*, 1263-1270.

44. Miura, G. A.; Chiang, P. K. *Anal. Biochem.*, **1985**, *147*, 217-221.

45. Icekson, I.; Apelbaum, A. *Biochem. Biophys. Res. Commun.*, **1983**, *113*, 586-591.

46. Smith, Jr., D. D.; Norton, S. J. *Biochem. Biophys. Res. Commun.*, **1980**, *94*, 1458-1462.

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