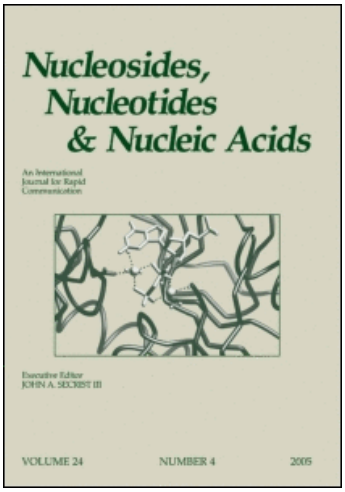

Nucleosides, Nucleotides and Nucleic Acids
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597286

Inhibitors of Adenosine Deaminase: Synthesis of Coformycin and 3'-Deoxycoformycin

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To cite this Article Hawkins, L. D. , Hanvey, J. C. , Boyd Jr., F. L. , Baker, David C. and Showalter, H. Hollis(1983) 'Inhibitors of Adenosine Deaminase: Synthesis of Coformycin and 3'-Deoxycoformycin', Nucleosides, Nucleotides and Nucleic Acids, 2: 5, 479 — 494
To link to this Article: DOI: 10.1080/07328318308079413
URL: http://dx.doi.org/10.1080/07328318308079413

NUCLEOSIDES AND NUCLEOTIDES, 2(5), 479-494 (1983)

INHIBITORS OF ADENOSINE DEAMINASE. SYNTHESIS OF COFORMYCIN AND 3'-DEOXYCOFORMYCIN.

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Abstract. Glycosylation of the heterocycle, 6,7-dihydroimidazo[4,5-d][1,3]diazepin-8(3H)-one, with suitably protected sugars under the influence of Lewis acid catalysts gave the β-D-ribo- and 3'-deoxy-β-D-erythropentofuranosyl nucleosides. Deprotection and reduction of the keto nucleosides with sodium borohydride gave the (8R)- and (8S)-3-β-D-glycofuranosyl-3,6,7,8-tetrahydroimidazo[4,5-d]-[1,3]diazepin-8-ols, the (8R)-isomers of which are potent inhibitors of adenosine deaminase.

INTRODUCTION

Work from these laboratories has resulted in a definitive synthesis[1-3] for pentostatin [2'-deoxycoformycin, (8R)-3-(2-deoxy-β-D-erythropentofuranosyl)-3,6,7,8-tetrahydroimidazo[4,5-d][1,3]diazepin-8-ol (**1**)] which is the most potent inhibitor[4] known for adenosine deaminase

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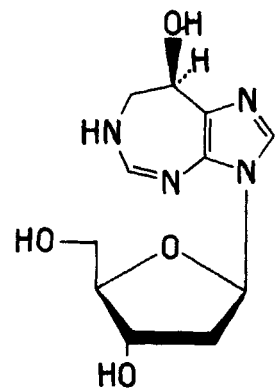
(EC 3.5.4.4, "ADase"), a ubiquitous enzyme that is responsible for the deamination and deactivation of adenine nucleosides that are either in use (e.g., 9-β-D-arabinofuranosyladenine) or potentially useful as drugs against certain viral diseases and cancer.[5] Co-administration of pentostatin (1), for example, has been shown to potentiate the effects of several adenine nucleosides,[5] and pentostatin is currently under Phase I clinical trials against acute myelogenous leukemia.[6]

The goals of the present research included an investigation of certain congeners of pentostatin (1) with the aim of (1) synthesizing a compound that would be stabilized toward hydrolysis at the glycosylic linkage relative to 1, which is structurally a 2'-deoxynucleoside with inherent stability problems;[7] (2) limiting synthetic products to the β-D-anomers that are the known[1,3] active species; (3) retaining potent ADase-inhibitor activity while possibly increasing selectivity for the ADase enzyme over related enzyme systems.



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RESULTS AND DISCUSSION

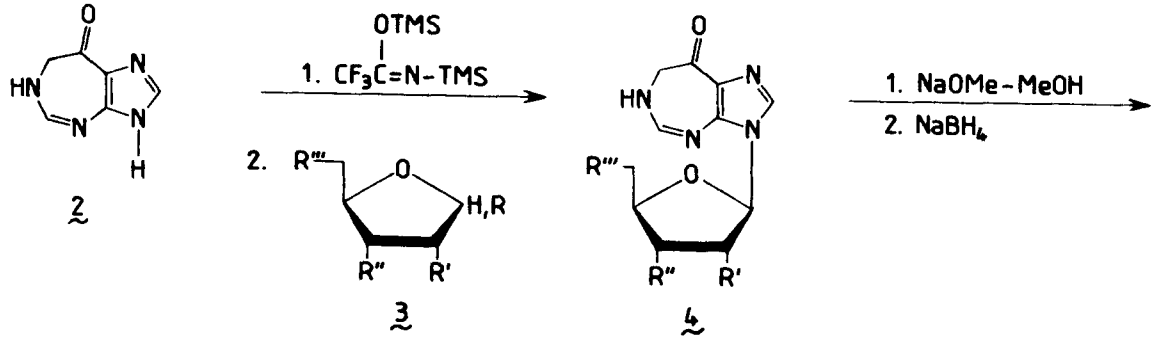
Rationale. In selecting possible target molecules, adenosine analogs were identified which were all good

substrates for ADA, preferably with K_m values either as small as or smaller than that for adenosine. Based on calculations made from the relationship between the K_m's for adenosine and 2'-deoxyadenosine and the ratio between the K_i's for coformycin (**5a**) and pentostatin (**1**), which are respectively the β-D-ribo-[8] and 2'-deoxy-β-D-erythropentofuranosyl (i.e., "2'-deoxyribo-") compounds, the effects of the particular sugar on the binding constants appear to correlate between the two series, i.e., between pentostatin-type inhibitors (K_i's) and adenine-type substrates (K_m's).[9] Thus one would expect[9] a 3'-deoxy-β-D-erythropentofuranosyl analog of pentostatin to have a K_i intermediate between those for pentostatin and coformycin (**5a**). Such a compound could also fulfill requirement no. 2, as the sugar reagent used in the chemical synthesis, 1,2-di-O-acetyl-5-O-benzoyl-3-deoxy-D-erythropentofuranose (**3b**), should offer anchimeric assistance during the nucleoside condensation, giving rise to the β-D-anomer as the chief product.

Chemistry. Prior to our attempting a synthesis of the 3'-deoxy (cordycepin) analog, we wished to test the techniques on a synthesis of the β-D-ribofuranosyl analog, coformycin. Previous work had shown that classical tin(IV) chloride-catalyzed procedures[10] could be modified[2,3] and adapted to the synthesis of pentostatin by using the per(trimethyl)silylated heterocycle **2**, prepared from 4-methyl-5-nitroimidazole.[3] Furthermore, the process has been extended[2] to the less reactive sugar, 2,3,5-tri-O-benzoyl-D-ribofuranosyl chloride (**3a**),[11] with exclusively the β-D-anomeric product, 2,3,5-tri-O-benzoyl-β-D-ribofuranosyl-6,7-dihydroimidazo[4,5-d][1,3]diazepin-8(3H)-one (**4a**), being isolated in 51% yield.[2] In order to achieve this success, certain precautions were found necessary: (1)

rigorously anhydrous conditions; (2) a heterocycle absolutely free from solvent and TMS-reagents; (3) acetonitrile as solvent, with the reaction being conducted at -10 to -20 °C. The product **4a**, purified by column chromatography, failed to crystallize, but gave an acceptable ^{1}H NMR spectrum and elemental analysis. The β-D-anomer was indicated by the H-1' signal, a doublet at δ 6.32 with $J_{1',2'}$ = 4.5 Hz. There was no evidence for the α-anomer by either HPLC or ^{1}H NMR analysis of the product.

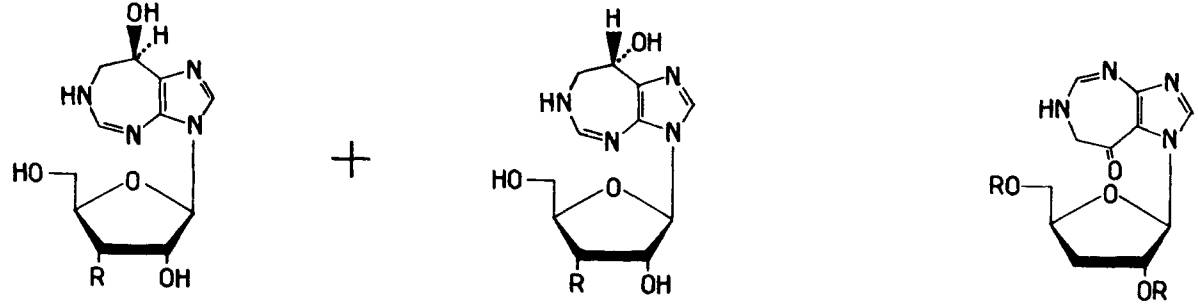
Removal of the benzoate protecting groups from **4a** with sodium methoxide - methanol, followed by reduction of the crude, free-hydroxy ketone[12] afforded a mixture of the (8R)-3-β-D-ribofuranosyl-3,6,7,8-tetrahydroimidazo[4,5-d][1,3]-diazepin-8-ol (**5a**) and the (8S)-isomer **6a** in good yield. These diastereomers were separated by preparative reverse-phase chromatography in aqueous acetonitrile to give compounds **5a** and **6a**, each in >99% isomeric purity as determined by HPLC. The faster-eluting component **5a** was found to be identical with authentic coformycin [the natural (8R)-isomer] by reverse-phase HPLC and by both UV and ^{1}H NMR spectroscopy. The slower-eluting component **6a**, shown to be decidedly different from **5a** by HPLC, exhibited a UV spectrum identical with **5a** at pH 1, 7 and 10, and differed only in minor detail from **5a** in its 200 MHz, ^{1}H NMR spectrum (See Experimental Section.); **6a** gave a m/z = 284 (theory: 284) in its field-desorption mass spectrum. Examination of the ORD data for both **5a** and **6a** revealed widely divergent values for the isomers (See Experimental Section.), leading one to conclude that the product **6a** is indeed the 8-epimer of **5a**, based on previous observations for **1** and its (8S)-isomer,[3] where the chirality of the 8-position, as expected, was found to be the dominant factor



2 ≈ 3 ≈ 4 ≈

a series: R = Cl-, R′ = R″ = R‴ = C_6H_5CO-

b series: R = R′ = AcO-, R″ = H-, R‴ = C_6H_5CO-



5 ≈ + 6 ≈ 7 ≈

a series: R = HO-

b series: R = H-

R = C_6H_5C(=O)-

in determining optical rotation values [(8R)-isomer **5a**, positive at 589 nm; (8S)-isomer **6a**, strongly negative at 589 nm]. Such constitutes a definitive synthesis of coformycin (**5a**)[8] and its (8S)-isomer (**6a**).

Under a set of conditions similar to those described in the previous paragraph, 1,2-di-O-acetyl-5-O-benzoyl-3-deoxy-D-erythropentofuranose[13] (**3b**) was condensed with the per(TMS)-derivative of **2**. Conditions that were found best for this condensation included the use of trimethylsilyl trifluoromethanesulfonate[14] as the Lewis acid catalyst. The reaction was conducted under strictly anhydrous conditions at -20 to -25 °C in acetonitrile - 1,2-dichloroethane. By this method was isolated, upon workup and crystallization, a 22% yield of 3-(2-O-acetyl-5-O-benzoyl-3-deoxy-β-D-erythropentofuranosyl)-6,7-dihydro[4,5-d][1,3]diazepin-8(3H)-one (**4b**), which was fully characterized by UV, ^{1}H NMR and elemental analysis. Along with **4b**, a very minor by-product was isolated (4% yield) having similar spectral (UV, ^{1}H NMR) properties to those of **4b**. The ^{1}H NMR for the by-product **7**, after column chromatography, indicated a β-D-nucleoside (H-1', singlet for **4b**; narrow doublet for **7**), and its field-desorption mass spectrum gave a m/z = 412. Inasmuch as the product was observed to partly rearrange to **4b** upon simple column chromatography, the isomeric N-1 species **7** shown in the scheme is postulated, although other N-derivatives, perhaps a kinetic product involved in the formation of **4b**, cannot be rigorously excluded. What is noteworthy from the observations on the above condensation reaction is that only the β-D-sugar derivative **3b**, from a mixture of anomeric 1-acetates having a ca. 60:40 β:α-ratio as determined by ^{1}H NMR, was observed to react to form the

nucleoside **4b**. The α-D-anomer of **3b** was recovered quantitatively from the reaction mixture and could not be induced to react under Lewis acid-catalyzed conditions to form a nucleoside. Such a finding is in line with the theorized role of Lewis acids in the condensation mechanism as proposed by Vorbrüggen,[15] and perhaps explains the lowered yields obtained in our hands with other sugar derivatives in comparison with those reactions that involve **3a**, which is indicated by ^{1}H NMR to be the pure β-D-anomer.

As for **4a**, the protected nucleoside **4b** was subjected to sodium methoxide - methanol transesterification, followed by reduction of the crude ketonucleoside with sodium borohydride to give the diastereomeric nucleosides (8R)-3-(3-deoxy-β-D-erythropentofuranosyl)-3,6,7,8-tetrahydroimidazo[4,5-d][1,3]diazepin-8-ol **5b** and its (8S)-epimer **6b**, which were separated by reverse-phase chromatography. The fact that both the ^{1}H NMR spectra and the UV spectra for **5b** and **6b** were nearly identical, together with the wide divergence observed in the ORD rotations, indicate the 8-epimers, a finding in agreement with our earlier observations on both **5a** and **6a** and on pentostatin **1**.[3] Field-desorption mass spectrometry gave m/z = 268 (theory = 268) for both compounds.

Inhibition of Adenosine Deaminase. The 3'-deoxycoformycin **5b** and the (8S)-epimer were each assayed against calf mucosal adenosine deaminase (Sigma, Type I) using the method of Cha[16] developed for tight-binding inhibitors. Compound **5b** exhibited a $K_i = 9 \times 10^{-12}$ M, a value that supports the predicted value. The (8S)-epimer **6b** was less tight-binding, with a K_i ca. 10^{-5} M, which is of the same order found with the (8S)-isomer of pentostatin.[3] The synthetic coformycin (**5a**) was found to be equipotent with the natural product.[17]

EXPERIMENTAL

General Methods. Melting points were taken on a Thomas-Hoover capillary melting point apparatus and are uncorrected. Solvent evaporations were conducted on a rotary evaporator at ca. 40 °C. Ultraviolet (UV) spectral data were determined either on a Cary model 118C or on a Varian model 635 double-beam instrument; (solvent, pH 1) = ca. 3 mL of solvent + 1 drop of 5N hydrochloric acid; (solvent, pH 11) = ca. 3 mL of solvent + 1 drop 6N potassium hydroxide. Infrared (IR) data were determined either on a Digilab model 11 FT-IR [indicated IR (FT)] or on a Perkin-Elmer model 710B. ^{1}H NMR were determined at 200 MHz as ca. 1% solutions on a Nicolet NT-200 instrument. Chemical shifts are reported downfield (δ units) from internal tetramethylsilane. Field-desorption mass spectra (MS) were determined using a VG model ZAB-29 mass spectrometer. Thin-layer chromatography (TLC) was carried out on E. Merck silica gel plates (0.2 mm), with both 254-nm UV and phosphomolybdic acid spray visualization. Column chromatography was conducted in open columns slurry packed with E. Merck Silica Gel-60 (70 - 230 mesh, ASTM). Preparative, adsorption-mode LC was conducted using a Waters Associates Prep 500A unit with silica gel cartridges. Preparative reverse-phase LC was carried out on a custom-built system at medium pressures (50 - 100 p.s.i.) using a column of octadecyl (C-18)-derived silica gel (20% carbon by analysis).

Nucleoside Condensation. A. 3-(2,3,5-Tri-*O*-benzoyl-β-*D*-ribofuranosyl)-6,7-dihydroimidazo[4,5-*d*][1,3]-diazepin-8-(3*H*)-one (4a). To a flame-dried, 50-mL, 3-neck round-bottom flask equipped with a serum cap, drying tube, and

stirring bar was added 635 mg (2.4 mmol) of 6,7-dihydroimidazo[4,5-d][1,3]diazepin-8(3H)-one hydrochloride·methylsulfoxide (2),[3] 1.34 mL (5 mmol) of bis(trimethylsilyl)trifluoroacetamide (Petrarch), and 2.5 mL of pyridine (distilled from calcium hydride). The suspension was stirred overnight during which time solution resulted. Volatiles were evaporated at 60 °C in vacuo, the syrupy residue was evaporated with acetonitrile (2 x 50 mL, Burdick and Jackson), and the resultant solid was dried at 60 °C in vacuo for 3 days. The solid residue was suspended in 15 mL of acetonitrile, and the mixture was cooled to -20 °C, to which was added 0.56 mL (4.8 mmol) of anhydrous tin(IV) chloride (Baker), and the mixture was stirred for 5 min during which time complete dissolution occurred. A 25 °C solution of 960 mg (2 mmol) 2,3,5-tri-O-benzoyl-D-ribofuranosyl chloride[11] in 10 mL of acetonitrile was added at once. The black solution was maintained at -10 °C to -20 °C for 4.5 h, then poured into a solution of saturated sodium bicarbonate. Ethyl acetate was added, and the mixture was stirred for 30 min, filtered over Celite, and the layers were separated. The aqueous phase was extracted with ethyl acetate, and the combined organic phases were dried ($MgSO_4$) and concentrated to a glass. The residue, showing essentially only one anomer by HPLC (94:6 dichloromethane - methanol, 10 μm, 0.46 x 25 cm, 2.0 mL/min flowrate), was dissolved in a minimum of ethyl acetate and charged onto a bed of 25 g of silica gel. The column was eluted with 150 mL of 25:75 toluene - ethyl acetate, 100 mL of 12:88 toluene - ethyl acetate, then with ethyl acetate until TLC showed complete elution of product. Solvent from the combined product fractions was evaporated, and the residue was pumped under vacuum to leave 610 mg (51%) of a

white glass: mp 122-128 °C, which could not be crystallized; R_f = 0.44 (90:10 ethyl acetate - methanol); $[\alpha]_D^{23}$ = -69.2° (c 1, methanol); UV (methanol) 349 (ε 3,900), 230 (61,000); (pH 1) 348 nm (2,800), 268 (8,900), 229 (47,000); (pH 11) 349 (4,350), 228 (56,500); IR (FT, KBr) 3430 (NH), 1734 (C=O) cm^{-1}; ^1H NMR (200 MHz, chloroform-d) δ 3.96 [d, 2H, $J_{H,NH}$ = 4.0 Hz, (s with D_2O), H-7,7_a], 4.61 - 4.88 (m, 3H, H-4', H-5',$5'_a$), 6.09 (t, 1H, $J_{2',3'}$ = 5.4 Hz, H-3'), 6.22 (t, 1H, H-2'), 6.32 (d, 1H, $J_{1',2'}$ = 4.5 Hz, H-1'), 7.33 - 7.66, 7.93 - 8.09 (m, 17H aryl/heteroaryl).

Anal. Calcd. for $C_{32}H_{26}N_4O_8$: C, 64.65; H, 4.41; N, 9.42. Found: C, 64.59; H, 4.49; N, 9.32.

B. 3-(2-O-Acetyl-5-O-benzoyl-3-deoxy-β-D-erythropentofuranosyl)-6,7-dihydroimidazo[4,5-d][1,3]diazepin-8(3H)-one (4b) and the 1-isomer, 7. By a process, essentially that described under **A**, 0.69 g (2.6 mmol) of **2** was trimethylsilylated with 2.91 g (11.3 mmol) of bis(trimethylsilyl)trifluoroacetamide, rendered free of reagents, dissolved in 4 mL of acetonitrile and mixed with 1.49 g (4.6 mmol) of 1,2-di-O-acetyl-5-O-benzoyl-3-deoxy-D-erythropentofuranose (**3b**)[13] (ca. 60:40 β:α-anomers) dissolved in 5 mL of 1,2-dichloroethane. To this mixture, maintained under a nitrogen atmosphere, was added dropwise at -20 °C 0.88 g (4.0 mmol) of trimethylsilyltrifluoromethanesulfonate (Fluka). After 5 h the reaction was quenched, and the products were chromatographed with a linear gradient of 0 → 10% methanol in chloroform to give 50 mg (4%) of **7** (amorphous solid) and 0.24 g (22%) of **4b** (crystallized from ethyl acetate).

Physical data for **4b**: mp 200 - 201 °C (dec); $[\alpha]_D^{21}$ = -51° (c 0.3, chloroform); R_f= 0.27 (9:1 chloroform -

methanol); UV (methanol, pH 1) 223 nm (ε39,300), 350 (4,240), (pH 7) 230 (42,390), 282 (2,650), 300 (2,910), 350 (3,900), (pH 10) 227 (48,570), 280 (sh), 298 (2,560), 350 (4,220); IR (KBr) 3220 (br-s, NH), 1720 (br-s, CO_2R), 1665 cm^{-1} (s, RCOR'); ^1H NMR (9:1 chloroform-d:D_2O) δ 2.12 - 2.17 [1H, m (within singlet), H-3'], 2.15 (3H, s, $COCH_3$), 2.37 - 2.54 [1H, m (7 lines), H-3'$_a$], 3.96 (2H, s, H-7), 4.5 - 4.75 [2H, m (6 lines), H-5',5'$_a$), 5.58 (1H, d, J=5.4 Hz, H-4'), 6.07 (1H, s, H-1') 7.39 - 7.63 (5H, m, COPh), 8.01 and 8.05 (2H, s, s, H-2 and H-5).

Anal. Calcd for $C_{20}H_{20}N_4O_6 \cdot 0.4\ H_2O$: C, 57.89; H, 4.73; N, 13.27. Found: C, 57.65; H, 5.03; N, 12.99.

Physical data for **7**: R_f= 0.29 (9:1 chloroform - methanol); UV (methanol, pH 1) 212 nm, 227, 275 (sh), (pH 7) 211, 225, 275 (sh), (pH 10) 214, 229 (sh) and 275 (sh); IR (chloroform) 1720 cm^{-1} (s, CO_2R); NMR (chloroform-d) δ 2.05 - 2.38 (5H, m, H-3',3'$_a$ and $COCH_3$), 3.93 (2H, d, J<1 Hz, H-7), 4.37 - 4.65 (2H, m, H-5',5'$_a$), 5.53 - 5.59 (1H, m, H-2'), 5.93 (1H, d, $J_{1',2'}$ = ~3 Hz, H-1'), 6.75 - 6.85 (1H, m, NH), 7.35 - 7.65 (5H, m, COPh), 8.01 (1H, s, H-5) 8.05 (1H, s, H-2). (Note: Due to the partial isomerization of **7** to **4b** upon silica gel chromatography, ^1H NMR peaks for **4b** were also observed in the spectrum.) MS (field desorption) m/z = 412 (theory 412).

(8R)-3-β-D-ribofuranosyl-3,6,7,8-tetrahydroimidazo-[4,5-d][1,3]diazepin-8)-ol (5a) and (8S)-3-β-D-ribofurano-syl-3,6,7,8-tetrahydroimidazo[4,5-d][1,3]diazepin-8-ol (6a). To a stirred solution of 0.48 g (0.8 mmol) of 2,3,5-tri-O-benzoyl-β-D-ribofuranosyl-6,7-dihydroimidazo[4,5-d]-[1,3]diazepin-8(3H)-one (**4a**) in 20 mL of dry methanol under dry conditions was added 0.65 g (2.8 mmol) of sodium at 5 °C. The mixture was allowed to warm to room temperature

and stir for Ø.5 h, at the end of which time the solution was neutralized by the addition of dry ice. [TLC (1:9 methanol - ethyl acetate) indicated complete deprotection of **4a** (R_f= Ø.67) to yield the deprotected ketone[12] (R_f= Ø.Ø3).] The solvent was evaporated to give a yellow residue which solidified upon trituration with ether. The solid was filtered, washed with ether and used directly in the following step.

To the above solid dissolved in 4Ø mL of water and 1Ø mL of methanol was added 16.4 mg (Ø.4 mmol) of sodium borohydride. The mixture was stirred for 1 h at room temperature, at the end of which time the excess reducing reagent was decomposed by the addition of dry ice, and the methanol was removed by evaporation, leaving an aqueous solution that was decolorized, filtered and lyophilized to a fluffy solid. Purification over a C-18 reverse-phase column by preparative HPLC (2 x 33 cm column), eluting with 2-L of a linear gradient of Ø% to 2.5% acetonitrile in water, gave after lyophilization 35.7 mg (16%) of >99% of pure **6a** and 41.2 mg (18%) of >99% pure **5a** as determined by C-18 reverse-phase HPLC.

Physical data for **5a**: $[\alpha]_D^{21}$ = +37.Ø° (c Ø.5, water), $[\alpha]_{578}$ = +38.6°, $[\alpha]_{546}$ = +51.2°, $[\alpha]_{436}$ = +184°; k'= 8 (HPLC: RP-18, 2.5:97.5 acetonitrile - Ø.Ø5 M aqueous sodium phosphate, 2 mL·min^{-1}); UV (methanol, pH 1) 262 nm, 211, (pH 7) 285, 211, (pH 1Ø) 285, 211; IR (KBr) 329Ø (br-s, OH and NH_2) and 162Ø cm^{-1} (s, aromatic); ^{1}H NMR (D_2O) δ3.33 - 3.56 [2H, m (6 lines), H-7,7$_a$], 3.77 - 3.86 (2H, m, H-5',5'$_a$ 4.2 - 4.3 (1H, m, H-4'), 4.3 - 4.35 (1H, m, H-3'), 4.62 - 4.82 (1H, m, H-2'), 5.14 - 5.16 (1H, m, H-8), 5.84 (1H, d, $J_{1',2'}$= 6.5 Hz, H-1'), 7.18 (1H, s, H-5), 7.68 (1H, s, H-2); MS (field desorption) m/z = 284 (theory 284).

Compound **5a** was essentially identical with authentic coformycin[17] by the above physical measurements.

Physical data for **6a**: $[\alpha]_D^{21}$ = -124° (c 1, water), $[\alpha]_{578}$ = -130°, $[\alpha]_{546}$ = -154°, $[\alpha]_{436}$ = -330°; k'= 12.4 (HPLC as for **5a**), UV (water, pH 1) 262 nm, 202 (9,460), (pH 7) 284, 205, (pH 10) 284, 205; IR (KBr) 3300 (br-s), OH) and 1620 (s, aromatic); ^{1}H NMR (D_2O) δ 3.34 - 3.56 [2H, m (6 lines), H-7,7_a], 3.77 - 3.81 (2H, m, H-5',$5'_a$), 4.20 - 4.24 (1H, m, H-4'), 4.31 - 4.36 (1H, m, H-3'), 4.67 - 4.82 (1H, m, H-2'), 5.14 - 5.16 (1H, m, H-8), 5.83 (1H, d, $J_{1',2'}$= 6.7 Hz, H-1'), 7.18 (1H, s, H-5), 7.67 (1H, s, H-2); MS (field desorption) m/z = 284 (theory 284).

(8R)-3-(3-Deoxy-β-D-erythropentofuranosyl)-3,6,7,8-tetrahydroimidazo[4,5-d][1,3]diazepin-8-ol (5b) and (8S)-3-(3-deoxy-β-D-erythropentofuranosyl)-3,6,7,8-tetrahydroimidazo[4,5-d][1,3]diazepin-8-ol (6b). As for **4a**, 0.24 g (0.6 mmol) of **4b** was deprotected with sodium methoxide, and the resulting crude ketone was reduced with 50 mg (1.32 mmol) of sodium borohydride. The crude mixture of diastereomers was separated by reverse-phase chromatography using 95:5 water - methanol to give 50 mg (32%) of **5b** and 60 mg (39%) of **6b**.

Physical data for **5b**: $[\alpha]_D^{21}$ = +17.0° (c 1, water), $[\alpha]_{578}$ = +18.6°, $[\alpha]_{546}$ = +24.3°, $[\alpha]_{436}$ = +76.3°; k'= 12.1 (RP-18 HPLC, 10:90 acetonitrile - 0.05 M aqueous sodium phosphate, 2 mL·min^{-1}); UV (water, pH 1) 215 nm, 262, (pH 7) 215, 285, (pH 10) 215 and 285; IR (KBr) 3600 - 3000 cm^{-1} (br, OH and NH); ^{1}H NMR (D_2O) δ 2.13 - 2.23 (2H, m, H-3,$3'_a$), 3.15 - 3.85 (4H, m, H-5',5_a and H-7,7_a), 4.50 - 4.60 (1H, m, H-4'), 5.12 - 5.21 (1H, m, H-8), 5.85 (1H, d, $J_{1',2'}$= 3.4 Hz, H-1'), 7.19 (1H, s, H-5), 7.66 (1H, s, H-2); MS (field desorption) m/z = 268 (theory 268).

Physical data for **6b**: $[\alpha]_D^{21} = -68.2^\circ$ (c 1, water), $[\alpha]_{578} = -71.4^\circ$, $[\alpha]_{546} = -83.0^\circ$, $[\alpha]_{436} = -182^\circ$; k'= 15.6 (HPLC as for **5b**); UV (methanol, pH 1) 215, 262, (pH 7) 215, 285, (pH 10) 215 and 285; IR (KBr) 3300 (br-s, OH and NH) and 1630 cm^{-1} (s, aromatic); ^1H NMR (D_2O) δ 2.13 - 2.23 (2H, m, H-3',3'$_a$), 3.36 (1H, dd, $J_{7,8} < 1$, $J_{7,7a} = 13.6$ Hz, H-7), 3.51 (1H, dd, $J_{7a,8} = 4.2$ Hz, H-7$_a$), 3.66 (1H, dd, $J_{4',5'} = 4.6$ Hz, $J_{5',5'a} = 12.5$ Hz, H-5'), 3.85 (1H, dd, $J_{4',5'a} = 2.8$ Hz, H-5'$_a$), 4.5 - 4.6 (1H, m, H-4'), 4.65 - 4.7 (1H, m, H-2'), 5.14 (1H, d, H-8), 5.85 (1H, d, $J_{1',2'} = 3.42$ Hz, H-1'), 7.19 (1H, s, H-5), 7.65 (1H, s, H-2); MS (field desorption) m/z = 268 (theory 268).

ACKNOWLEDGMENTS.

C. N. Richmond is thanked for the 200-MHz ^1H NMR spectra. A special acknowledgment is made to R. Hale and W. Tindall of the Tennessee Eastman Corporation for the field-desorption mass spectra. This work was supported, in part, by the National Institutes of Health Grant No. CA-26465 (to D.C.B.) and Contract No. NO1-CM-23708 (to W.-L./P.-D.).

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9. The correlations are as follows:

Substrate	$K_m \times 10^6$	Ratios
Adenosine (Ade)	25[a] (33)[b]	
2'-Deoxyadenosine (2'-dAde)	7[a] (22)[b]	Ade/2'-dAde = 3.57[a] (Ade/2'-dAde = 1.5)[b]
3'-Deoxyadenosine (3'-dAde)	- (25)[b]	(Ade/3'-dAde = 1.3)[b]

Inhibitor	$K_i \times 10^{12}$	Ratio
Coformycin (**5a**)	10[a]	
Pentostatin (**1**)	2.5[a]	**5a**/**1** = 4.0

[a] Values (using human erythrocytic adenosine deaminase) are taken from Agarwal, R. P.; Cha, S; Crabtree, G. W.; Parks, R. E., Jr. In "Chemistry and Biology of Nucleosides and Nucleotides", Harmon, R. E.; Robins, R. K.; Townsend, L. B., Eds.; Academic Press: New York, 1978; pp. 158 - 197.

[b] Values (using calf mucosal adenosine deaminase) are taken from Frederiksen, S. Archiv. Biochem. Biophys. **1966**, 113, 383 - 388. These values are comparable to those found elsewhere.

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12. The free hydroxy ketone, isolated by neutralizing the sodium methoxide with carbon dioxide, evaporating the methanol to dryness, extracting the methyl p-toluoylate with ether, then lyophilizing to dryness an aqueous solution of the nucleoside, gave an ^{1}H NMR spectrum as follows: (90 MHz, D_2O) δ 3.70 - 3.85 (m, 2H, H-5',5'$_a$), 4.18 [m (4 lines), 1H, H-4'], 4.31 (t, 1H, H-3'), 4.60 (t, 1H, H-2'), 5.91 (d, 1H, H-1'), 7.47 (s, 1H, H-2) and 7.85 (s, 1H, H-5).

13. Sugar **3b** was prepared via 1,2:5,6-di-O-isopropylidene-α-D-ribohexofuranos-3-ulose 4-tolylsulfonylhydrazone (Nair, V.; Sinhababu, A. K. J. Org. Chem. **1978**, 49, 5013 - 5017) by the process described by Murray, D. H.; Prokop, J. in "Synthetic Procedures in Nucleic Acid Chemistry," Zorbach, W. W.; Tipson, R. S., Eds.; Wiley: New York, 1978; Part 1, pp. 193 - 197.

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17. Authentic coformycin was supplied by H. Dion and associates, W.-L./P.-D. $[\alpha]_D^{23} = +34.6^{o}$ (c 1, water).

Received October 6, 1983