Pergamon

Bioorganic & Medicinal Chemistry, Vol. 3, No. 9, pp. 1223–1229, 1995

0968-0896/95 $9.50 + .00

0968-0896(95)00089-5

Stereoselective Synthesis, Chemistry and Antiviral Evaluation of 1-(2,3-Dideoxy-3-*C*-hydroxymethyl-β-D-*threo*-pentofuranosyl)thymine Derivatives

Jesper Wengel,*† Raymond F. Schinazi‡ and Marvin H. Caruthers

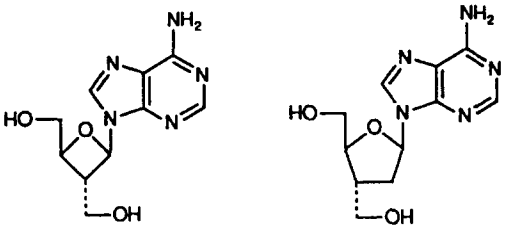
Department of Chemistry and Biochemistry, University of Colorado, Boulder, CO 80309-0215, U.S.A.

Abstract—A series of novel 3'-*C*-branched 2',3'-dideoxynucleosides have been synthesized and evaluated as anti-HIV agents. Hydroboration of 2',3'-dideoxy-3'-*C*-methylene nucleoside proceeded regio- and stereoselectively to give 1-(2,3-dideoxy-3-*C*-hydroxymethyl-5-*O*-trityl-β-D-*threo*-pentofuranosyl)thymine (**5**) after oxidation. 3'-*C*-Chloromethyl and 3'-*C*-iodomethyl 5'-*O*-protected 2',3'-dideoxynucleosides **9** and **12** were obtained from **5** by reaction with carbon tetrachloride/triphenylphosphine and methyltriphenoxyphosphonium iodide, respectively. Arbuzov reaction of **12** with triethyl phosphite afforded 3'-*C*-(diethylphosphono)methyl 5'-*O*-protected 2',3'-dideoxynucleoside **14**. Compounds **5**, **9**, **12** and **14** were detritylated to give 1-(3-*C*-chloromethyl-2,3-dideoxy-β-D-*threo*-pentofuranosyl)thymine (**10**), 1-(2,3-dideoxy-3-*C*-hydroxymethyl-β-D-*threo*-pentofuranosyl)-thymine (**11**), 1-(2,3-dideoxy-3-*C*-iodomethyl-β-D-*threo*-pentofuranosyl)thymine (**13**) and 1-(2,3-dideoxy-3-*C*-(*O*,*O*'-diethylphosphono)methyl-β-D-*threo*-pentofuranosyl)thymine (**15**), respectively. These nucleoside analogues were evaluated for antiviral activity against human immunodeficiency virus type 1 (HIV-1) and herpes simplex virus type 1 (HSV-1) *in vitro*. Compound **5** demonstrated selective antiviral activity against HIV-1 but not HSV-1.

Introduction

The causative agent for acquired immunodeficiency syndrome (AIDS) is the retrovirus human immunodeficiency virus type 1 (HIV-1). Several 2',3'-dideoxynucleosides have shown promising antiviral activity against HIV-1 *in vitro* and *in vivo* as their 5'-triphosphates are potent inhibitors of the retrovirus reverse transcriptase.[1–3] These derivatives, 3'-azido-3'-deoxythymidine (AZT), 2',3'-dideoxyinosine (DDI), 2',3'-dideoxycytidine (DDC) and 2',3'-didehydro-2',3'-dideoxythymidine are the only drugs approved for clinical use despite their considerable cytotoxic effects. Therefore, there is an urgent need for additional new compounds with improved potency and selectivity in their antiviral action.

The naturally occurring nucleoside oxetanocin A (**1**) is known to exhibit promising anti-HIV-1 activity.[4,5] Recently, it was shown that 2',3'-dideoxy-3'-*C*-hydroxymethyladenosine (**2**)[6] and 2',3'-dideoxy-3'-*C*-hydroxymethylcytidine[7] display an anti HIV-1 activity profile similar to **1** (Scheme 1). This has caused an increased interest in 3'-*C*-branched 2',3'-dideoxynucleosides, and it prompted us to investigate the synthesis, chemistry and antiviral activity of novel β-D-*threo* configurated 3'-*C*-branched 2',3'-dideoxythymidine nucleoside analogs.



1 **2**

Scheme 1.

Generally, synthesis of 3'-*C*-branched 2',3'-dideoxynucleosides has involved 3'-keto carbohydrates as key intermediates in multi-step syntheses often suffering from lack of stereoselectivity.[6,8,9] Other examples have been reported where D- and L-2',3'-dideoxy-3'-*C*-hydroxylmethyl nucleosides are synthesized in nine steps from chiral epoxy alcohols[7,10] and the corresponding 3'-*C*-fluoromethyl and 3'-*C*-azidomethyl derivatives from D-2',3'-dideoxy-3'-*C*-hydroxymethyl nucleosides.[11] Synthesis of 3'-deoxy-3'-*C*-formyluridine and its subsequent 3'-epimerization and reduction to both epimers of the corresponding 2',3'-dideoxy-3'-*C*-hydroxymethyl uracil nucleosides has been achieved[12] via ring contraction of a 3'-aminohexopyranosyl nucleoside. Recently, Bamford *et al.*[13] have reported stereo- and regiospecific synthesis of 2',3'-dideoxy-3'-*C*-hydroxymethyl uracil nucleosides by ring opening of a 2',3'-epoxide intermediate with 4,5-dihydro-2-lithio-5-methyl-1,3,5-dithiazine followed by reaction with HgO/$HgCl_2$, reduction and deoxygenation. After completion of the work described here, synthesis of the two epimers of 2',3'-dideoxy-3'-*C*-hydroxymethyl-5'-*O*-monomethoxy-

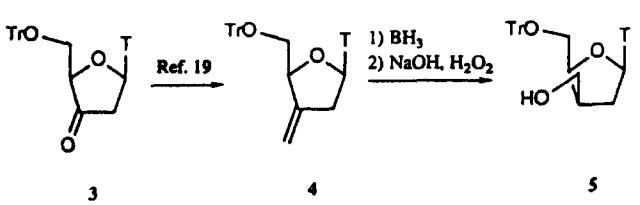
†Present address: Department of Chemistry, Odense University, Campusvej 55, DK-5230 Odense M, Denmark.
‡Address: Laboratory of Biochemical Pharmacology, Emory University School of Medicine and Veterans Affairs Medical Center, Decatur, GA 30033, U.S.A.

tritylthymidine from the corresponding 2',3'-dideoxy-3'-nitro nucleoside has been reported and the 3'-*threo* isomer **11** was isolated after chromatographic separation and deprotection.[14] Summarizing, it is clear that the efforts to synthesize 2',3'-dideoxy-3'-*C*-hydroxymethyl nucleosides and their derivatives have been limited, and that hitherto known approaches suffer from elaborate and multiple step procedures. In this paper we report a short and stereoselective synthesis of 1-(2,3-dideoxy-3-*C*-hydroxymethyl-β-D-*threo*-pentofuranosyl)-thymine (**11**) and derivatives thereof. Besides, results from evaluation of antiviral activities against HIV-1 and HSV-1 are reported.

Results and Discussion

Chemistry

For the two step synthesis of 1-(2,3-dideoxy-3'-*C*-hydroxymethyl-5-*O*-trityl-β-D-*threo*-pentofuranosyl) thymine (**5**) from 3'-keto derivative **3** (Scheme 2), we used a strategy earlier developed for synthesis of the corresponding 5'-*O*-dimethoxytrityl protected nucleoside used in syntheses of 3'-*C*-hydroxymethyl linked and functionalized oligodeoxynucleotide analogs.[15,16] Thus, by the procedure of Froehlich *et al.*[17] we obtained 3'-keto-5'-*O*-tritylthymidine (**3**) in 70% yield (reported[17] 80%). As Samano and Robins[18] have reported successful oxidations of both purine and pyrimidine 2'-deoxynucleosides, the methods reported here for the synthesis of **5** and **9–15** can probably be used for synthesis of analogous purine and pyrimidine nucleosides. Wittig methylenation with methyl triphenylphosphonium iodide/*n*-butyllithium failed to give 3'-*C*-methylene nucleoside **4**. The basic conditions induced quantitative β-elimination of the thymine base. However, using $Zn/CH_2Br_2/TiCl_4$[19,20] as reported for the methylenation of **3**, 2',3'-dideoxy-3'-*C*-methylene nucleoside **4** was obtained in 78% yield (reported[19] 95% yield). Hydroboration of **4** with borane:1,4-oxathiane[21] followed by oxidation with alkaline hydrogen peroxide afforded 1-(2,3-dideoxy-3-*C*-hydroxymethyl-5-*O*-trityl-β-D-*threo*-pentofuranosyl)-thymine (**5**) in 75% yield. The β-D-*threo* configuration of the product is consistent with the *syn*-addition mechanism of hydroboration with attack from the less sterically hindered α-face of the pentose ring. As no other product could be detected by ^1H NMR analysis of the crude product, the hydroboration was completely regio- and stereoselective.



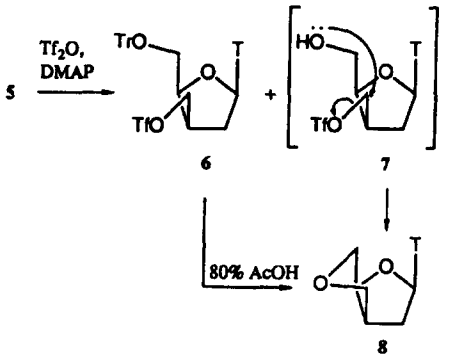
3 4 5

3-5: T = thymine-1-yl

Scheme 2.

As a potential substrate for nucleophilic substitution reactions, the triflate **6** was prepared from **5** in 63% yield by esterification with trifluoromethanesulfonic anhydride (Scheme 3). Analytical TLC showed an additional product (less polar than **5**) which was identified as the bicyclic compound **8** (26% yield). The formation of **8** was induced by the presence of small amounts of trifluoromethanesulfonic acid formed during esterification. A four-fold excess of DMAP was not able to neutralize this acid quickly enough to prevent partial detritylation of triflate **6**. It was impossible to isolate the putative intermediate **7**, presumably because the 5'-OH sterically is in a position favorable for intramolecular nucleophilic attack on the activated 3'-*C*-hydroxymethyl functionality. To support this explanation, the triflate **6** was exposed to the standard detritylating conditions (80% acetic acid, 100 °C, 10 min) which afforded the bicyclic compound **8** in near quantitative yield.

Synthesis of 3'-*C*-iodomethyl nucleoside **12** and the corresponding 3'-azido nucleoside was attempted by nucleophilic substitution on the triflate **6** (tetrabutylammonium iodide, methylene chloride, 30 min to 10 h; sodium iodide, acetone, 30 min to 15 h; sodium iodide, acetonylacetone, 20–90 °C, 30 min to 15 h; sodium azide, DMF, 20–90 °C, 30 min to 8 h) but in no case was substitution observed. Probably, the bulky 5'-*O*-trityl group and the nucleobase sterically block the nucleophiles from approaching the activated 3'-*C*-hydroxymethyl.



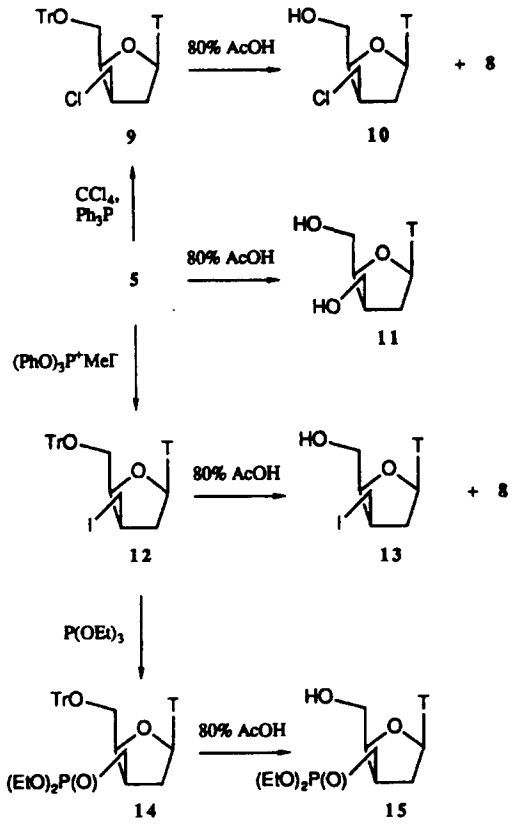
6 7 8

5-8: T = thymine-1-yl

Scheme 3.

3'-*C*-Chloromethyl-2',3'-dideoxy-5'-*O*-trityl nucleoside **9** was prepared in 84% yield by reaction of **5** with carbon tetrachloride and triphenylphosphine.[22] 2',3'-Dideoxy-3'-*C*-iodomethyl-5'-*O*-trityl nucleoside **12** was prepared from **5** in 81% yield by reaction with methyltriphenoxyphosphonium iodide.[23] 3'-*C*-Iodomethyl derivative **12** was subsequently submitted to Arbuzov reaction[24] with triethyl phosphite to give 2',3'-dideoxy-3'-*C*-(*O*,*O*'-diethylphosphono)methyl-5'-*O*-trityl nucleoside **14** in 63% yield (Scheme 4).

The nucleosides **5**, **9**, **12** and **14** were deprotected with 80% acetic acid at 100 °C. This afforded 3'-*C*-chloromethyl-2',3'-dideoxynucleoside **10** in 79% yield, 2',3'-



Scheme 4.

dideoxy-3'-*C*-hydroxymethyl nucleoside **11** in 91% yield, 2',3'-dideoxy-3'-*C*-iodomethyl nucleoside **13** in 60% yield and 2',3'-dideoxy-3'-*C*-(*O*,*O*'-diethylphosphono)methyl nucleoside **15** in 85 % yield. Additionally, the bicyclic compound **8** was isolated in 12 and 26% yield during deprotection of 3'-*C*-chloromethyl-5'-*O*-trityl nucleoside **9** and 3'-*C*-iodomethyl-5'-*O*-trityl nucleoside **12**, respectively. Prolonged exposure of **12** to deprotection conditions increased the yield of **8** and decreased the yield of 3'-*C*-iodomethyl 5'-*O*-deprotected nucleoside **13**. The yields obtained for the bicyclic compound **8** during deprotection of the triflate **6** (~100%), the 3'-*C*-iodomethyl nucleoside **12** (26%) and the 3'-*C*-chloromethyl nucleoside **9** (12%), reflect the relative leaving group ability of the 3'-*C*-methyl substituents (triflate > iodide > chloride). This further supports the mechanism suggested in Scheme 2 for formation of **8**.

The structural assignment of the nucleosides was based on ^1H, ^{13}C, ^{31}P, 2D-COSY ^1H NMR and mass spectra. NOE ^1H NMR on **5** was used to prove the configuration of the nucleosides: the NOE between H-1', H-2'α, H-3' and H-4' were all relatively strong, indicating the positioning of these protons to the α-side of the pentose ring. The NOE between H-6 in the pyrimidine ring and H-2'β was much stronger than the NOE between H-6 and any of the other protons in the pentose ring. Finally, the NOE between H-3' and H-2'α was much stronger than the NOE between H-3' and H-2'β. This unambiguously proves the β-D-*threo* configuration of the nucleosides **5**–**15**.

Antiviral evaluation

Selected nucleosides were tested as potential antiviral agents against HIV-1 in human PBM cells using the methodology described in Schinazi *et al.*[25] As can be seen from the data presented in Table 1, only compound **5** has modest activity against HIV-1 (EC_{50} = 3.3 μM) with no cytotoxicity when tested up to 100 μM. In contrast, AZT used as a positive control had an EC_{50} of 0.003 μM. Compounds **11** and **15** had weak activity against HIV-1. Compounds **5**, **8**, **10**, **11**, **13** and **15** were also tested for their ability to inhibit HSV-1 in Vero cells using a plaque reduction assay.[26] None of these compounds had significant activity against HSV-1. In contrast, acyclovir used as a positive control had an EC_{50} of 0.12 μM in the same assay. Compounds were also evaluated for cytotoxicity in human PBM cells and Vero cells, as described previously.[25] None of the compounds tested demonstrated cytotoxicity in PBM cells when evaluated up to 100 μM. However, compound **5** was found to be cytotoxic to rapidly dividing Vero cells.

Conclusion

The procedures described here constitute an efficient stereoselective synthetic route to novel 3'-*C*-branched 2',3'-dideoxy-β-D-*threo* nucleosides. Thus, derivatives

Table 1. Antiviral effect of selected compounds against HIV-1 and HSV-1[a]

Compound	Anti-HIV-1 in PBM cells EC_{50} (μM)	Cytoxicity in PBM cells IC_{50} (μM)	Anti-HSV-1 in Vero cells EC_{50} (μM)	Cytotoxicity in Vero cells IC_{50} (μM)
5	3.3	>100	>10[b]	4.6
8	>100	>100	>100	>100
10	>100	>100	>100	>100
11	87.7	>100	>100	>100
13	>100	>100	>100	>100
15	32.0	>100	>100	>100
AZT	0.003	>100	>100	29.0

[a]Assay completed according to Refs 25 and 26.
[b]This compound was cytotoxic at 100 μM when the cells were confluent.

5–15 were synthesized from the parent nucleoside thymidine in only four to six steps. Equivalent procedures should be applicable for synthesis of analogous derivatives of other pyrimidine and purine nucleosides. The anti-HIV activity of β-D-*threo* configurated 2',3'-dideoxy-3'-*C*-hydroxymethyl nucleoside **5**, together with the recently reported anti-HIV activity of an analogously configurated 3'-*N*-hydroxyamino substituted thymine nucleoside,[27] indicates that efforts towards synthesis and biological evaluation of other structurally related nucleosides should be undertaken.

Experimental

General

NMR spectra were recorded at 299.9 MHz for ^{1}H NMR, 75.4 MHz for ^{13}C NMR, 282.2 MHz for ^{19}F NMR and 121.4 MHz for ^{31}P NMR. Magic bullet was used as matrix for the FAB-MS experiments. Melting points are uncorrected. Analytical TLC was performed on precoated TLC sheets (Merck silica gel 60 F_{254} 0.2 mm) with CH_2Cl_2:MeOH (19:1) (A) and CH_2Cl_2:MeOH (9:1) (B) as solvent systems. Merck silica gel 230–400 mesh was used for column chromatography.

*1-(2,3-Dideoxy-3-*C*-hydroxymethyl-5-*O*-trityl-β-D-*threo*-pentofuranosyl)thymine (5).* 3'-*C*-Methylene nucleoside **4** (750 mg, 1.56 mmol) in anhydrous THF (3 mL), was added dropwise under Ar to a well-stirred solution of BH_3:oxathiane (0.19 mL of a 7.8 M solution in oxathiane, 1.50 mmol) in anhydrous THF (4 mL) at room temperature. As analytical TLC (solvent system A) showed no more starting nucleoside **4** after 30 min, a 2 M solution of NaOH (0.78 mL, 1.56 mmol) was added. The reaction mixture was then immersed in a cooling bath (0 °C) and 30% aqueous H_2O_2 (0.16 mL, 1.56 mmol) was added dropwise. After stirring at room temperature for 45 min, the reaction mixture was poured into ice/H_2O (20 mL) and extracted with Et_2O (2 × 20 mL). The organic phase was successively washed with H_2O (3 × 20 mL) and a saturated aqueous solution of $NaHCO_3$ (15 mL), and dried over anhydrous Na_2SO_4. After filtration and evaporation of the solvents, the crude product was purified by flash chromatography on a silica gel column (150 g, 8 × 6 cm) packed in CH_2Cl_2. The column was eluted with CH_2Cl_2 (300 mL) and CH_2Cl_2:MeOH (500 mL, 99.5:0.5; 500 mL, 99:1; 500 mL, 98:2) to give 580 mg (75%) of pure[28] **5** as a white foam after evaporation of the solvents: R_f 0.27 (solvent system A), 0.68 (solvent system B); ^{1}H NMR ($CDCl_3$): δ 1.52 (*s*, 3H, Me), 1.80–1.86 (*m*, 1H, H-2'β), 2.31–2.40 (*m*, 1H, H-2'α), 2.59–2.63 (*m*, 1H, H-3'), 3.21 (*dd*, 1H, *J* = 10.7 Hz and 2.9 Hz, H-5'a), 3.47–3.56 (*m*, 3H, CH_2-a", CH_2-b", H-5'b), 4.21–4.26 (*m*, 1H, H-4'), 6.09 (*dd*, 1H, *J* = 8.3 Hz and 5.9 Hz, H-1'), 7.18–7.39 (*m*, 15H, trityl), 7.53 (*s*, 1H, H-6), 9.18 (*s*, 1H, NH); ^{13}C NMR ($CDCl_3$): δ 12.00 (Me), 33.88 (C-2'), 42.25 (C-3'), 61.40, 62.88 (CH_2", C-5'), 78.53 (C-4'), 84.10 (CPh_3), 88.10 (C-1'), 111.11 (C-5), 127.55, 128.13, 128.55, 142.87 (trityl), 135.55 (C-6), 150.59 (C-2), 163.84 (C-4).

*1-(2,3-Dideoxy-3-*C*-((trifluoromethyl)sulfonyloxy)methyl-5-*O*-trityl-β-D-*threo*-pentofuranosyl)thymine (6) and 1-((5-*O*,3-*C*-hydroxymethyl-*O*)-anhydro)-2,3-dideoxy-3-*C*-hydroxymethyl-β-D-*threo*-pentofuranosyl)thymine (8).* Compound **5** (80 mg, 160 μmol) in anhydrous CH_2Cl_2 (2 mL) was added dropwise under Ar during 10 min to a stirred solution of trifluoromethanesulfonic anhydride (68 mg, 240 μmol) and DMAP (73 mg, 600 μmol) in dry CH_2Cl_2 (2 mL) at room temperature. After 30 min, analytical TLC (solvent system A) showed no more starting nucleoside **5** but two less polar products. The reaction mixture was poured into a cold, saturated aqueous solution of $NaHCO_3$ (10 mL), and CH_2Cl_2 (10 mL) was added. The organic phase was separated and the aqueous phase extracted with CH_2Cl_2 (15 mL). The combined organic phase was evaporated and the residue was purified by flash chromatography on a silica gel column (20 g, 2 × 5 cm) packed in CH_2Cl_2 and eluted with CH_2Cl_2 (200 mL) and CH_2Cl_2:MeOH (400 mL, 99.5:0.5; 500 mL, 99:1) to give 74 mg (63 %) of pure **6** as a white foam after evaporation of the solvents; R_f 0.42 (solvent system A); ^{1}H NMR ($CDCl_3$): δ 1.38 (*s*, 3H, Me), 1.80–1.88 (*m*, 1H, H-2'β), 2.38–2.47 (*m*, 1H, H-2'α), 2.56–2.64 (*m*, 1H, H-3'), 3.18 (*dd*, 1H, *J* = 10.5 and 3.4 Hz, H-5'a), 3.41–3.53 (*m*, 3H, CH_2-a", CH_2-b", H-5'b), 4.21–4.26 (*m*, 1H, H-4'), 6.11 (*dd*, 1H, *J* = 8.1 Hz and 5.9 Hz, H-1'), 7.17–7.38 (*m*, 15H, trityl), 7.63 (*s*, 1H, H-6), 8.88 (*s*, 1H, NH); ^{13}C NMR ($CDCl_3$): δ 11.71 (Me), 35.14 (C-2'), 42.25 (C-3'), 61.39, 63.29 (CH_2", C-5'), 79.03 (C-4'), 84.58 (CPh_3), 87.48 (C-1'), 110.88 (C-5), 127.38, 127.94, 128.80, 143.24 (trityl), 135.85 (C-6), 150.53 (C-2), 163.90 (C-4); ^{19}F NMR ($CDCl_3$): δ 115.52 (CF_3). Additionally, 10 mg (26 %) of pure **8** was obtained as a white foam after evaporation of the solvents from the fractions containing the more polar product: R_f 0.33 (solvent system A); ^{1}H NMR ($CDCl_3$): δ 1.58–1.67 (*m*, 1H, H-2'β), 1.87 (*s*, 3H, Me), 2.58–2.68 (*m*, 1H, H-2'α), 2.93–2.98 (*m*, 1H, H-3'), 3.44 (*dd*, 1H, *J* = 10.7 Hz and 3.7 Hz, H-5'a), 3.57 (*dd*, 1H, *J* = 9.5 Hz and 6.2 Hz, CH_2-a"), 3.81 (*d*, 1H, *J* = 9.5 Hz, CH_2-b"), 4.12 (*d*, 1H, *J* = 10.7 Hz, H-5'b), 4.63 (*dd*, 1H, *J* = 7.3 Hz and 3.7 Hz, H-4'), 6.05 (*dd*, 1H, *J* = 7.8 Hz and 5.9 Hz, H-1'), 7.30 (*s*, 1H, H-6), 9.47 (*s*, 1H, NH); ^{13}C NMR ($CDCl_3$): δ 12.50 (Me), 38.32 (C-2'), 42.47 (C-3'), 74.15, 74.50 (CH_2", C-5'), 83.45 (C-4'), 85.58 (C-1'), 111.38 (C-5), 134.83 (C-6), 150.59 (C-2), 163.95 (C-4); HRMS (EI) m/z 238.0951 (M, calcd 238.0954). Moreover, compound **8** was synthesized in small scale by the following procedure: triflate **6** (5 mg, 8 μmol) was dissolved in 80% HOAc (0.5 mL) and stirred 10 min at 100 °C. Analytical TLC showed one spot with R_f 0.33 (solvent system A) and complete disappearance of **6**. The mixture was evaporated and purified by flash chromatography on silica gel column (8 g, 2 × 2 cm) packed in CH_2Cl_2 and eluted with CH_2Cl_2:MeOH (200 mL, 99.5:0.5; 100 mL, 98:2). After evaporation of the solvents, the product was obtained in near quantitative yield with ^{1}H NMR data identical to those given above for compound **8**.

1-(3-C-Chloromethyl-2,3-dideoxy-5-O-trityl-β-D-threo-pentofuranosyl)thymine (9). CCl_4 (60 mg, 390 μmol) was added to a solution of **5** (72 mg, 144 μmol) and triphenylphosphine (50 mg, 191 μmol) in anhydrous DMF (3 mL). After 5 h at room temperature, analytical TLC (solvent system A) showed no more starting nucleoside **5**. The solvent was evaporated *in vacuo* and the crude product was purified on a silica gel column (20 g, 2 × 6 cm) packed in CH_2Cl_2. The column was first washed with CH_2Cl_2 (200 mL) and then the product was eluted with CH_2Cl_2:MeOH (700 mL, 99.5:0.5) to give 63 mg (84%) of pure **9** as a white foam after evaporation of solvents: R_f 0.63 (solvent system A); ^{1}H NMR ($CDCl_3$): δ 1.36 (*s*, 3H, Me), 2.01–2.09 (*m*, 1H, H-2'β), 2.52–2.60 (*m*, 1H, H-2'α), 2.88–2.96 (*m*, 1H, H-3'), 3.26 (*dd*, 1H, *J* = 10.9 and 2.6 Hz, H-5'a), 3.45–3.58 (*m*, 2H, CH_2-a", CH_2-b"), 3.68 (*dd*, 1H, *J* = 10.9 and 3.5 Hz, H-5'b), 4.27–4.32 (*m*, 1H, H-4'), 6.25 (*dd*, 1H, *J* = 8.9 and 5.2 Hz, H-1'), 7.28–7.43 (*m*, 15H, trityl), 7.79 (*s*, 1H, H-6), 9.30 (*s*, 1H, NH); ^{13}C NMR ($CDCl_3$): δ 11.53 (Me), 36.30 (C-2'), 42.96, 43.09 (CH_2", C-3'), 62.90 (C-5'), 78.38 (C-4'), 83.90 (CPh_3), 87.94 (C-1'), 111.43 (C-5), 127.57, 128.10, 128.71, 142.81 (trityl), 135.53 (C-6), 150.62 (C-2), 163.68 (C-4).

1-(3-C-Chloromethyl-2,3-dideoxy-β-D-threo-pentofuranosyl)thymine (10). Compound **9** (55 mg, 106 μmol) was dissolved in 80% HOAc (1.5 mL) and stirred for 15 min at 100 °C after which analytical TLC (solvent system A) showed no more starting nucleoside **9**. The mixture was evaporated and purified by flash chromatography on a silica gel column (20 g, 2 × 6 cm) packed in CH_2Cl_2 and eluted with CH_2Cl_2 (200 mL) and CH_2Cl_2:MeOH (400 mL, 99.5:0.5; 500 mL, 99:1) to give 23 mg (79%) of pure **10** as a white foam after evaporation of the solvents: R_f 0.26 (solvent system A); ^{1}H NMR ($CDCl_3$): δ 1.91 (*s*, 1H, Me), 2.06–2.17 (*m*, 1H, H-2'β), 2.39–2.47 (*m*, 1H, H-2'α), 2.90–3.01 (*m*, 1H, H-3'), 3.79–4.03 (*m*, 4H, CH_2-a", CH_2-b", H-5'a, H-5'b), 4.23–4.26 (*m*, 1H, H-4'), 6.06 (*dd*, 1H, *J* = 9.0 and 5.6 Hz, H-1'), 7.68 (*s*, 1H, H-6), 9.34 (*s*, 1H, NH); ^{13}C NMR ($CDCl_3$): δ 12.50 (Me), 35.58 (C-2'), 43.11, 43.14 (CH_2", C-3'), 62.38 (C-5'), 79.64 (C-4'), 85.88 (C-1'), 111.03 (C-5), 136.87 (C-6), 150.68 (C-2), 164.04 (C-4); MS (FAB, positive mode) m/z (rel. intensity) 275 (MH^+, 17), 127 (100). Moreover, 3 mg (12%) of pure **8** was isolated after evaporation of the solvents from the fractions containing the less polar product.

1-(2,3-Dideoxy-3-C-hydroxymethyl-β-D-threo-pentofuranosyl)thymine (11). Compound **5** (60 mg, 120 μmol) was dissolved in 80% acetic acid (1.5 mL) and stirred for 5 min at 100 °C after which analytical TLC (solvent system B) showed no more starting nucleoside **5**. The mixture was evaporated and purified by flash chromatography on a silica gel column (20 g, 2 × 6 cm) packed in CH_2Cl_2 and eluted with CH_2Cl_2 (200 mL) and CH_2Cl_2:MeOH (200 mL, 99.5:0.5; 300 mL, 97:3) to give 28 mg (91%) of pure **11** as a white foam after evaporation of the solvent: R_f 0.35 (solvent system B); ^{1}H NMR (D_2O): δ 1.60 (*s*, 1H, Me), 1.71–1.77 (*m*, 1H, H-2'β), 2.24–2.33 (*m*, 1H, H-2'α), 2.55–2.64 (*m*, 1H, H-3'), 3.45–3.68 (*m*, 4H, CH_2-a", CH_2-b", H-5'a, H-5'b), 4.06–4.12 (*m*, 1H, H-4'), 5.89 (*dd*, 1H, *J* = 7.9 and 6.2 Hz, H-1'), 7.54 (*s*, 1H, H-6); ^{13}C NMR (CD_3OD): δ 12.22 (Me), 35.25 (C-2'), 43.55 (C-3'), 61.34, 62.59 (CH_2", C-5'), 81.81 (C-4'), 85.96 (C-1'), 111.12 (C-5), 138.17 (C-6), 152.34 (C-2), 166.34 (C-4); MS (FAB, positive mode) m/z (rel intensity) 257 (MH^+, 20), 155 (100). This compound has been synthesized earlier by another method with NMR data reported in other solvents.[14]

1-(2,3-Dideoxy-3-C-iodomethyl-5-O-trityl-β-D-threo-pentofuranosyl)thymine (12). Methyltriphenoxyphosphonium iodide (110 mg, 243 μmol) and **5** (60 mg, 120 μmol) were dissolved in anhydrous DMF (3 mL). After 15 min at room temperature, analytical TLC showed no more starting nucleoside **5**. MeOH (0.1 mL) was added, and after an additional 15 min the mixture was evaporated *in vacuo*. The residue was dissolved in $CHCl_3$ (20 mL), extracted with aqueous 10% $Na_2S_2O_3$ (15 mL) and H_2O (15 mL). The organic phase was dried over anhydrous Na_2SO_4, filtered, and concentrated to a gum. The crude product was dissolved in CH_2Cl_2 and purified on a silica gel column (20 g, 2 × 6 cm) packed in CH_2Cl_2 (300 mL). Elution of the product with CH_2Cl_2:MeOH (700 mL, 99.5:0.5) gave 59 mg (81%) of pure **12** as a white foam after evaporation of the solvents: R_f 0.67 (solvent system A); ^{1}H NMR ($CDCl_3$): δ 1.23 (*s*, 3H, Me), 1.95–2.02 (*m*, 1H, H-2'β), 2.50–2.58 (*m*, 1H, H-2'α), 2.88–3.13 (*m*, 3H, H-3', CH_2-a", CH_2-b"), 3.21 (*dd*, 1H, *J* = 11.0 and 2.4 Hz, H-5'a), 3.63 (*dd*, 1H, *J* = 11.0 and 3.2 Hz, H-5'b), 4.16–4.20 (*m*, 1H, H-4'), 6.16 (*dd*, 1H, *J* = 9.3 and 5.1 Hz, H-1'), 7.21–7.35 (*m*, 15H, trityl), 7.77 (*s*, 1H, H-6), 8.99 (*s*, 1H, NH); ^{13}C NMR ($CDCl_3$): δ 1.79 (CH_2"), 11.43 (Me), 39.20 (C-2'), 43.68 (C-3'), 63.00 (C-5'), 78.89 (C-4'), 83.42 (CPh_3), 88.01 (C-1'), 111.44 (C-5), 127.63, 128.10, 128.77, 142.68 (trityl), 135.67 (C-6), 150.56 (C-2), 163.79 (C-4).

1-(2,3-Dideoxy-3-C-iodomethyl-β-D-threo-pentofuranosyl)thymine (13). Compound **12** (50 mg, 82 μmol) was dissolved in 80% HOAc (1.51 mL) and stirred for 20 min at 100 °C after which analytical TLC (solvent system A) showed no more starting nucleoside **12**. The mixture was evaporated and purified by flash chromatography on a silica gel column (20 g, 2 × 6 cm) packed in CH_2Cl_2 (200 mL). Elution with CH_2Cl_2:MeOH (500 mL, 99.5:0.5; 500 mL, 99:1) afforded 18 mg (60%) of pure **13** as a glass after evaporation of the solvents: R_f 0.27 (solvent system A); ^{1}H NMR ($CDCl_3$): δ 1.84 (*s*, 3H, Me), 2.04–2.08 (*m*, 1H, H-2'β), 2.37–2.45 (*m*, 1H, H-2'α), 2.90–2.97 (*m*, 1H, H-3'), 3.35–3.87 (*m*, 4H, CH_2-a", CH_2-b", H-5'a, H-5'b), 4.10–4.15 (*m*, 1H, H-4'), 5.93 (*dd*, 1H, *J* = 9.4 and 5.2 Hz, H-1'), 7.56 (*s*, 1H, H-6), 8.48 (*s*, 1H, NH); ^{13}C NMR (CD_3OD): δ 2.64 (CH_2"), 12.29 (Me), 39.93 (C-2'), 45.47 (C-3'), 62.37 (C-5'), 81.88 (C-4'), 85.96 (C-1'), 111.26 (C-5), 138.22 (C-6), 152.47 (C-2), 166.38 (C-4); MS (FAB, positive mode) m/z (rel. intensity) 367 (MH^+, 6), 127 (100). Moreover, 5 mg (26%) of pure **8** was isolated after evaporation of the solvents from the fractions containing the less polar product.

1-(2,3-Dideoxy-3-C-(O,O'-diethylphosphono)methyl-5-O-trityl-β-D-threo-pentofuranosyl)thymine (14). Compound **12** (100 mg, 164 μmol) was refluxed in triethyl phosphite (2 mL). After 6 h, analytical TLC showed only a trace of starting nucleoside **12**. The mixture was evaporated *in vacuo* and the residue purified on a silica gel column (20 g, 2 × 6 cm) packed in CH_2Cl_2. The column was eluted with CH_2Cl_2 (200 mL) and CH_2Cl_2:MeOH (400 mL, 99.5:0.5; 400 mL, 99:1; 400 mL, 98:2) to give 64 mg (63%) of pure **14** as a white foam after evaporation of the solvents: R_f 0.23 (solvent system A), 0.74 (solvent system B); ^{1}H NMR ($CDCl_3$): δ 1.14–1.34 (*m*, 9H, 2 × Et, Me), 1.98–2.06 (*m*, 1H, H-2'β), 2.48–2.54 (*m*, 1H, H-2'α), 2.71–2.80 (*m*, 1H, H-3'), 3.09 (*dd*, 1H, *J* = 11.0 and 1.9 Hz, H-5'a), 3.63 (*dd*, 1H, *J* = 11.0 and 3.2 Hz, H-5'b), 3.84–4.20 (*m*, 7H, 2 × Et, CH_2-a", CH_2-b", H-4'), 6.18 (*dd*, 1H, *J* = 9.2 and 5.0 Hz, H-1'), 7.21–7.34 (*m*, 15H, trityl), 7.74 (*s*, 1H, H-6), 8.86 (*s*, 1H, NH); ^{13}C NMR ($CDCl_3$): δ 11.28 (Me), 16.05–16.41 (*m*, 2 × Et), 24.51 (*d*, *J* = 143 Hz, CH_2"), 34.43 (*d*, *J* = 4.4 Hz, C-3'), 37.49 (*d*, *J* = 6.0 Hz, C-2'), 61.45–61.69 (*m*, 2 × Et), 63.26 (C-5'), 79.39 (*d*, *J* = 14.9 Hz, C-4'), 83.90 (CPh_3), 87.90 (C-1'), 111.27 (C-5), 127.59, 127.99, 128.77, 142.69 (trityl), 135.89 (C-6), 150.55 (C-2), 163.72 (C-4); ^{31}P NMR ($CDCl_3$): δ 29.94 (CH_2"-P).

1-(2,3-Dideoxy-3-C-(O,O'-diethylphosphono)methyl-β-D-threo-pentofuranosyl)thymine (15). Compound **14** (50 mg, 81 μmol) was dissolved in 80% HOAc (1.5 mL) and stirred 20 min at 100 °C after which analytical TLC (solvent system B) showed no more starting nucleoside **14**. The mixture was evaporated and purified by flash chromatography on a silica gel column (20 g, 2 × 6 cm) packed in CH_2Cl_2. The column was eluted with CH_2Cl_2 (200 mL) and CH_2Cl_2:MeOH (400 mL, 99.5:0.5; 400 mL, 98.5:1.5; 300 mL, 97:3) to give 26 mg (85%) of pure **15** as a white foam after evaporation of the solvents: R_f 0.46 (solvent system B); ^{1}H NMR ($CDCl_3$): δ 1.30–1.36 (*m*, 6H, 2 × Et), 1.91 (*s*, 3H, Me), 2.08–2.18 (*m*, 1H, H-2'β), 2.42–2.50 (*m*, 1H, H-2'α), 2.83–2.87 (*m*, 1H, H-3'), 3.80–3.96 (*m*, 2H, H-5'a, H-5'b), 4.07–4.21 (*m*, 7H, 2 × Et, CH_2-a", CH_2-b", H-4'), 6.01 (*dd*, 1H, *J* = 9.2 and 5.3 Hz, H-1'), 7.70 (*s*, 1H, H-6), 8.95 (*s*, 1H, NH); ^{13}C NMR ($CDCl_3$): δ 12.51 (Me), 16.37–16.66 (*m*, 2 × Et), 25.17 (*d*, *J* = 145 Hz, CH_2"), 34.61 (*d*, *J* = 4.9 Hz, C-3'), 37.47 (*d*, *J* = 6.8 Hz, C-2'), 61.82–62.00 (*m*, 2 × Et), 62.69 (C-5'), 80.70 (*d*, *J* = 12.7 Hz, C-4'), 86.09 (C-1'), 110.86 (C-5), 137.03 (C-6), 150.61 (C-2), 163.84 (C-4); ^{31}P NMR ($CDCl_3$): δ 30.60 (CH_2"-P); MS (FAB, positive mode) m/z (rel. intensity) 377 (MH^+, 8), 251 (100).

Acknowledgements

This research was supported by National Institutes of Health GM25680 and the Department of Veteran Affairs. We thank the Danish Technical Research Council for supporting J. W. We also thank M. Ashley for his help with the NOE ^{1}H NMR and R. Mathis, D. Cannon and A. McMillan for excellent technical assistance for the virological portion of the work.

References and Note

1. Nasr, M.; Litterst, C.; McGowan, J. *Antiviral Res.* **1990**, *14*, 125.

2. DeClerq, E. *AIDS Res. Human Retroviruses* **1992**, *8*, 119.

3. Schinazi, R. F.; Mead, J. R.; Feorino, P. M. *AIDS Res. Human Retroviruses* **1992**, *8*, 963.

4. Hoshino, H.; Shimizu, N.; Shimada, N.; Takita, T.; Takeuchi, T. *J. Antibiot.* **1987**, *40*, 1077.

5. Seki, J.-I.; Shimada, N.; Takahashi, K.; Takita, T.; Takeuchi, T.; Hoshino, H. *Antimicrob. Agents Chemother.* **1989**, *33*, 773.

6. Tseng, C. K.-H.; Marquez, V. E.; Milne, G. W. A.; Wysocki, Jr R. J.; Mitsuya, H.; Shirasaki, T.; Driscoll, J. S. *J. Med. Chem.* **1991**, *34*, 343.

7. Svansson, L.; Kvarnström, I.; Classon, B.; Samuelsson, B. *J. Org. Chem.* **1991**, *56*, 2993.

8. Acton, E. M.; Goerner, R. N.; Uh, H. S.; Ryan, K. J.; Henry, D. W. *J. Med. Chem.* **1979**, *22*, 518.

9. Yoshimura, J. *Adv. Carbohydr. Chem. Biochem.* **1984**, 42, 69.

10. Kvarnström, I.; Svansson, L.; Svensson, C.; Svensson, S. C. T.; Classon, B.; Samuelsson, B. *Nucleosides Nucleotides* **1992**, *11*, 1367.

11. Svansson, L.; Kvarnström, I.; Classon, B.; Samuelsson, B. *Nucleosides Nucleotides* **1992**, *11*, 1353.

12. Shuto, S.; Iwano, T.; Inoue, H.; Ueda, T. *Nucleosides Nucleotides* **1982**, *1*, 263.

13. Bamford, M. J.; Coe, P. L.; Walker, R. T. *J. Med. Chem.* **1990**, *33*, 2494.

14. Garg, N.; Plavec, J.; Chattopadhyaya, J. *Tetrahedron* **1993**, *49*, 5189.

15. Svendsen, M. L.; Wengel, J.; Dahl, O.; Kirpekar, F.; Roepstorff, P. *Tetrahedron* **1993**, *49*, 11341.

16. Jørgensen, P. N.; Stein, P. C.; Wengel, J. *J. Am. Chem. Soc.* **1994**, *116*, 2231.

17. Froehlich, M. L.; Swartling, D. J.; Lind, R. E.; Mott, A. W.; Bergstrom, D. E. *Nucleosides Nucleotides* **1989**, *8*, 1529.

18. Samano, V.; Robins, M. J. *J. Org. Chem.* **1990**, *55*, 5186.

19. Sharma, M.; Bobek, M. *Tetrahedron Lett.* **1990**, *31*, 5839.

20. Lombardo, L. *Tetrahedron Lett.* **1982**, *23*, 4293.

21. Brown, H. C.; Mandal, A. K. *Synthesis* **1980**, 153.

22. Verheyden, J. P. H.; Moffatt, J. G. *J. Org. Chem.* **1972**, *37*, 2289.

23. Verheyden, J. P. H.; Moffatt, J. G. *J. Org. Chem.* **1970**, *35*, 2319.

24. Arbuzov, B. A. *Pure Appl. Chem.* **1964**, *9*, 307.

25. Schinazi, R. F.; Sommadossi, J. P.; Saalman, V.; Canon, D. L.; Xie, M.-W.; Hart, G. C.; Smith, G. A.; Hahn, E. F. *Antimicrob. Agents Chemother.* **1990**, 34, 1061.

26. Schinazi, R. F.; Fox, J. J.; Watanabe, K. A.; Nahmias, A. J. *Antimicrob. Agents Chemother.* **1986**, 29, 77.

27. Tronchet, J. M. J.; Zsély, M.; Capek, K.; Komaromi, I.; Geoffroy, M.; De Clercq, E.; Balzarini, J. *Nucleosides Nucleotides* **1994**, *13*, 1871.

28. To verify the purity of compounds **5**, **6** and **8–15**, copies of the ^{13}C NMR spectra were enclosed with this manuscript at submission.

(*Received in U.S.A.* 13 *January* 1995; *accepted* 9 *March* 1995)