Acta Chemica Scandinavica, 1993: 47: 889–895
Printed in Belgium – all rights reserved

ACTA
CHEMICA SCANDINAVICA
ISSN 0904–213X

Synthesis of New Acyclic Nucleosides

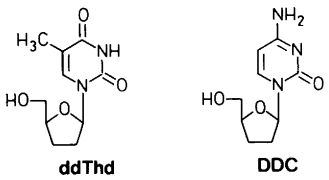
Charlotte Schou,[a] Erik B. Pedersen[a,*] and Claus Nielsen[b]

[a]Department of Chemistry, Odense University, DK-5230 Odense M, Denmark and [b]Retrovirus Laboratory, Department of Virology, Statens Seruminstitut, Artillerivej 5, DK-2300 Copenhagen, Denmark

Schou, C., Pedersen, E. B. and Nielsen, C., 1993. Synthesis of New Acyclic Nucleosides. – Acta Chem. Scand. 47: 889–895.

The preparation of 1-(4,5-dihydroxypentyl)uracils and the corresponding cytosine derivatives as acyclic analogs of 3′-deoxythymidine and 2′,3′-dideoxycytidine (DDC), respectively, is described. The synthesis consists of alkylation of sodium salts of pyrimidine bases with 4-pentenyl *p*-toluenesulfonate **2** followed by oxidation of the double bond with peroxyformic acid to afford the 1-(4,5-dihydroxypentyl)uracils. 1-(4,5-Dihydroxypentyl)uracil was further reacted to give the corresponding cytosine derivatives via the 4-(1,2,4-triazol-1-yl) derivative. 1-(5-hydroxy-4-methoxypentyl)uracil (**9a**) was obtained together with the 4-hydroxy-5-methoxypentyl derivative **9b** from 1-(4-pentenyl)uracil (**3b**) by epoxidation and subsequent acid-catalyzed cleavage of the epoxide by methanol. 6-(Thymin-1-yl) and 6-(cytosin-1-yl) derivatives of (*Z*)-2,3,6-trideoxy-D-*erythro*-2-hexenitol **17a,c** were synthesized by alkylation of the sodium salts of thymine and N^4-isobutyrylcytosine with 3,4-di-*O*-acetyl-6-*O*-*p*-toluenesulfonyl-D-glucal (**14**) followed by hydrolysis, reduction with sodium borohydride and deprotection.

The advent of acquired immunodeficiency syndrome (AIDS) and the identification of the retrovirus human immunodeficiency virus (HIV), as the causative agent of AIDS,[1] has increased the interest in compounds that can block the replication of retroviruses. The present paper deals with the synthesis of 4,5-dihydroxypentyl derivatives of pyrimidine nucleobases which are acyclic analogs of 2′,3′-dideoxycytidine (DDC)[2] and 3′-dideoxythymidine (ddThd),[2] known to be active against HIV.

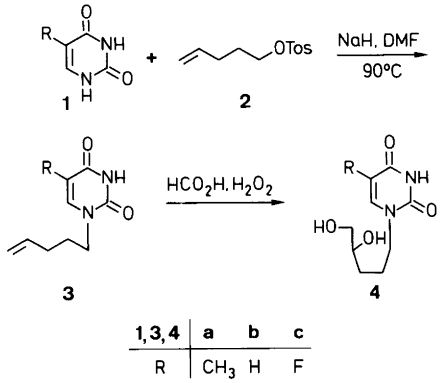


Scheme 1.

1-(5-Hydroxy-4-methoxypentyl)uracil was also a target molecule because the electronic properties of the 5′-hydroxy group resembles those of 2′,3′-dideoxy nucleosides. In recent years a number of unsaturated acyclic nucleosides have shown activity against a wide range of viruses[3] and some have shown activity against HIV.[4] In the present paper syntheses of some chiral unsaturated acyclic nucleoside analogs are also reported using tri-*O*-acetylglucal as the starting material.

Results and discussion

The key precursor **2** in the synthesis of the 4,5-dihydroxypentyl- and 5-hydroxy-4-methoxypentyl derivatives was prepared in three steps[5,6] from tetrahydrofurfuryl alcohol.

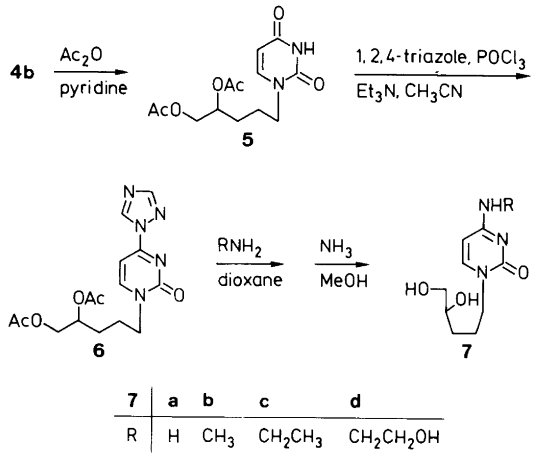


Scheme 2.

Compound **2** was reacted with the sodium salts of uracils **1** in *N*,*N*-dimethylformamide overnight at 90°C to give the 1,3-alkylated and the 1-alkylated pyrimidine bases according to the procedure of Sasaki *et al.*[7] The 1-alkylated product was isolated by chromatography to give **3a–c** in 39–53% yield. **3a–c** were oxidized with peroxyformic acid, which was prepared *in situ* from formic acid and hydrogen peroxide.[8] The resulting formyloxy hydroxy compound was hydrolyzed in refluxing sodium hydroxide for 1 h. After silica chromatography **4b** could be isolated as pure crystals, whereas **4a,c** were

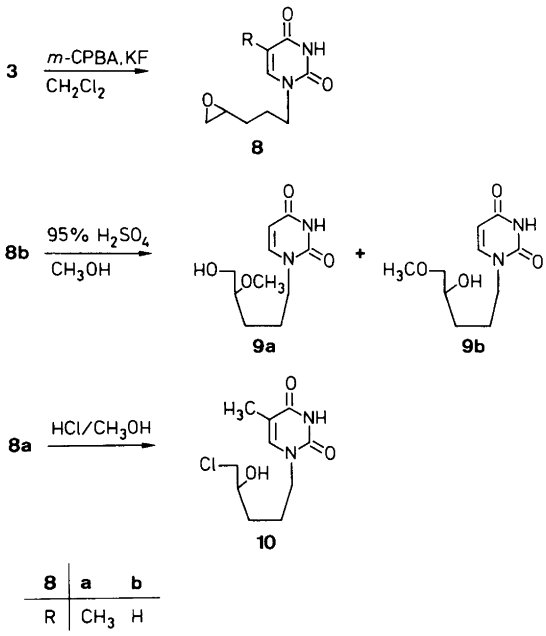
* To whom correspondence should be adressed.

isolated as slightly impure oils which could only be crystallized after HPLC purification.



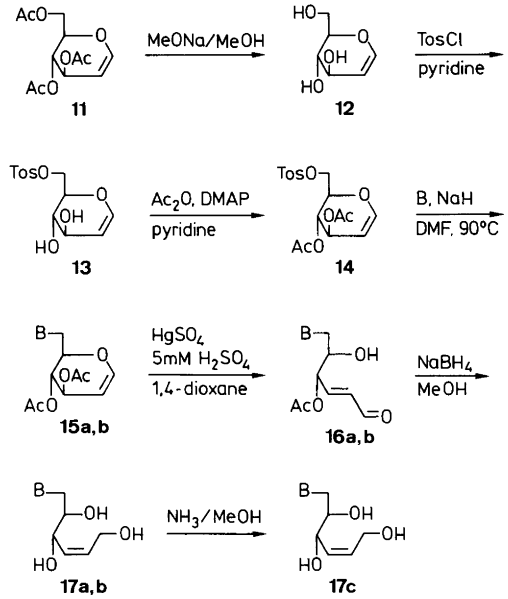
Scheme 3.

For the preparation of the cytosine derivatives **7a–d**, **4b** was acetylated with acetic anhydride in pyridine to give **5** as an oil which was chromatographed to remove the residual acetic acid that otherwise would interfere with the following reaction. Compound **5** was reacted with phosphoryl chloride and 1,2,4-triazole in triethylamine and acetonitrile to give **6**,[9] with 1,2,4-triazol-1-yl as a good leaving group for nucleophilic substitution reactions. These were performed with ammonia, methylamine, ethylamine, and ethanolamine in dioxane[10] and the subsequent deprotection of the hydroxy groups were performed in methanolic ammonia to give **7a–d** in 15–46% yield. Nucleophilic substitutions by 1,1-dimethylhydrazine and ethane-1,2-diamine were also tried, but they resulted in complex reaction mixtures.



Scheme 4.

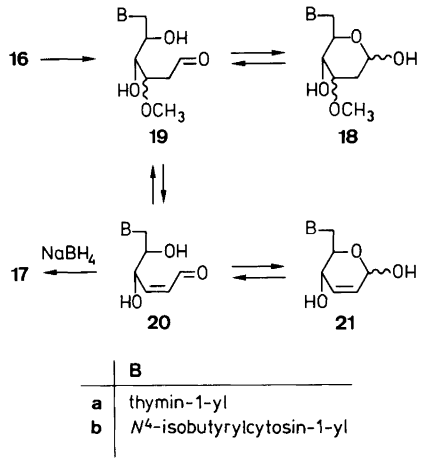
For the preparation of **9a,b** and **10**, **3a,b** were epoxidized with a complex of *m*-chloroperbenzoic acid (*m*-CPBA) with potassium fluoride. Potassium fluoride is believed to activate the *m*-CPBA by means of hydrogen bonding.[11] When the reaction was finished, the excess of *m*-CPBA was precipitated by further addition of activated potassium fluoride and removed by filtration. In this way, further effort to isolate the labile epoxide was avoided because evaporation of the solvent afforded the epoxide **8** contaminated by starting material only. Acid-catalyzed cleavage of the epoxide **8b** with 95% sulfuric acid and methanol was expected to give the abnormal isomer **9a** by a borderline S_N1 reaction at the more sterically hindered carbon (C-4'),[12] but the ratio between **9a** and **9b** was found to be 1 : 3 based on a ^{13}C NMR spectrum of the mixture. Compound **9b** was isolated by chromatography and identified by means of a coupled ^{13}C NMR spectrum which showed a coupling of 3 Hz between the methoxy carbon and 5'-H. Compound **9b** was also oxidized with pyridinium chlorochromate in dichloromethane to give the corresponding ketone[13] as a proof of the correct identification. For the epoxide cleavage a saturated solution of hydrogen chloride in methanol was also attempted, but **10** was isolated as the sole product. MS showed an M^+ peak at *m/z* 246 which proved that the chloride ion had acted as the nucleophile and the coupled ^{13}C NMR spectrum showed that C-5' had shifted upfield compared with the 4,5-dihydroxypentyl derivative indicating that the attack had occurred at C-5'.



Scheme 5.

The synthese of the chiral trihydroxyhexenyl derivatives **17a–c** started with the deacetylation of 3,4,6-tri-*O*-acetyl-

D-glucal **11** with sodium methoxide in methanol[14] and subsequent selective tosylation of the primary hydroxy group to give **13**. This was acetylated with acetic anhydride and pyridine in dichloromethane with 4-dimethylaminopyridine (DMAP) as a catalyst to give **14** which was subsequently used for alkylation of the sodium salts of thymine and N^4-isobutyrylcytosine[15] in *N*,*N*-dimethylformamide. Silica gel TLC (10% MeOH in CH_2Cl_2) showed that many products had been formed. For the pyrimidine derivatives the products with R_f ca. 0.5 were isolated to give the 1-alkylated derivatives **15a,b** in 28–36% yield. **15a,b** were hydrolyzed in dilute sulfuric acid in 1,4-dioxane and with mercuric sulfate as catalyst[16] to give the α,β-unsaturated aldehyde **16a,b**. **16a** was reduced by sodium borohydride in methanol, to give as the main product **18a** (26%) (a Michael addition of methoxide ion to the α,β-unsaturated aldehyde followed by a ring closure to the pyranose form). Compound **17a** was isolated as a by-product (7%). Compound **16b** was reduced by being added in portions together with sodium borohydride to methanol to give the corresponding alcohol **17b** in 21% yield.



Scheme 6.

Although 3′-H of compounds **17** appeared as multiplets in the ^{1}H NMR spectra, a small ΣJ value, as deduced from its line broadening, may be indicative of a *Z* configuration of the double bond. This can be explained as shown in Scheme 6 by an equilibrium between the *trans*-α,β-unsaturated aldehyde **16** and the *cis*-α,β-unsaturated aldehyde **20**, probably via addition and elimination of methoxide via the intermediate **19**, which also explains the formation of **18**. Compound **21** could act as a pool of **20** until reduced to **17**. The 4′-*O*-acetyl group was removed during the reduction because of the basic conditions, but for removal of the N^4-isobutyryl from **17b**, to give **17c**, methanolic ammonia was required.

The acyclic analogs **4a–d** and **7a–d** of ddThd and DDC, respectively were selected together with the compounds **17a,c** and a mixture of the compounds **9a** and **9b** for *in vitro* studies of biological effects. The compounds did not show any significant activity at 100 μM against Herpes Simplex Virus type 1 (HSV-1), strain McIntyre, when tested in a continuous cell line from rabbit cornea (SIRC) which was maintained in Eagle's MEM containing 1% fetal calf serum (FCS) and test compounds. The compounds were also devoid of activity at 100 μM against HIV-1 (strain HTLV-IIIB) when MT-4 cells were incubated with virus, washed and added in a proportion of 1 : 10 to uninfected MT-4 cells which had been preincubated in test compound containing culture medium (RPM 1640 containing 10% FCS) for 2 h. The MT-4 cells were maintained in the culture medium likewise containing the test compound. Expression of HIV in culture medium was quantitated by HIV antigen detection ELISA.

Experimental

NMR spectra: Bruker AC 250 FT spectrometer. FAB MS, EI MS and peak matching: Varian Mat 311A spectrometer (70 eV). Column chromatography: silica gel (0.040–0.063 mm, Merck). Reversed-phase HPLC: RP-18, 15 μm, 300A. TLC: 60 F-254 precoated plates (Merck).

1-(4-Pentenyl)thymine (**3a**). A mixture of thymine (23.9 g, 0.19 mol) and 50% oil-immersed sodium hydride (6.38 g, 0.13 mol) in DMF (1300 ml) was stirred at 70–80°C for 1 h and cooled to room temperature. 4-Pentenyl *p*-toluenesulfonate **2** (28.6 g, 0.12 mol) was added and the mixture was stirred at 90°C overnight. TLC (5% MeOH in $CHCl_3$) showed two main products and some unchanged thymine. The mixture was neutralized with acetic acid, evaporated under reduced pressure and partitioned between $CHCl_3$ (550 ml) and water (200 ml). The sparingly soluble thymine was recovered by filtration and washed with $CHCl_3$. The separated organic layer was dried with $MgSO_4$ and evaporated. The oil was subjected to silica gel column chromatography with MeOH in $CHCl_3$ (gradient 5–9%) to give **3a** which crystallized after standing for a few days. The crystals were washed with ether–petroleum ether to give 10.7 g (53%). M.p. 105–106°C. MS: *m/z* (%) 194 (37, M^+). ^{1}H NMR ($CDCl_3$): δ 1.92 (s, 3 H, CH_3), 1.80 (quintet, $J_{1',2'} = J_{2',3'} = 7$ Hz, 2 H, 2′-H), 2.11 (q, $J_{2',4'} = 7$ Hz, 2 H, 3′-H), 3.73 (t, 2 H, 2′-H), 5.01 (dd, $J_{cis} = 10$ Hz, $J_{gem} = 1.1$ Hz, 1 H, 5′-Ha), 5.05 (dd, $J_{trans} = 17$ Hz, 1 H, 5′-Hb), 5.79 (ddt, 1 H, 4′-H), 7.06 (s, 1 H, 6-H), 10.45 (br, 1 H, NH). ^{13}C NMR ($CDCl_3$): δ 11.9 (CH_3), 27.6 (C-2′), 30.0 (C-3′), 47.6 (C-1′), 110.1 (C-5), 115.4 (C-5′), 136.6 (C-4′), 140.6 (C-6), 151.0 (C-2), 164.6 (C-4).

1-(4-Pentenyl)uracil (**3b**). This compound was prepared in 49% yield from 4-pentenyl tosylate and uracil as described for **3a**. The isolated oil was crystallized from acetone. M.p. 79–80°C. MS: *m/z* (%) 180 (30, M^+). ^{1}H NMR (DMSO-d_6): δ 1.69 (quintet, $J_{1',2'} = J_{2',3'} = 7$ Hz, 2 H, 2′-H), 2.04 (q, $J_{3,4'} = 7$ Hz, 2 H, 3′-H), 3.68 (t, 2 H, 1′-H), 4.99 (dd, $J_{cis} = 10$ Hz, $J_{gem} = 1.4$ Hz, 1 H, 5′-Ha),

5.06 (dd, J_{trans} = 17 Hz, 1 H, 5'-Hb), 5.58 (d, J = 8 Hz, 1 H, 5-H), 5.82 (ddt, 1 H, 4'-H), 7.66 (d, 1 H, 6-H), 11.26 (br, 1 H, NH). ^{13}C NMR (DMSO-d_6): δ 27.5 (C-2'), 30.0 (C-3'), 47.1 (C-1'), 100.8 (C-5), 115.2 (C-5'), 137.5 (C-4'), 145.7 (C-6), 150.9 (C-2), 163.8 (C-4).

1-(4-Pentenyl)-5-fluorouracil (**3c**). This compound was prepared as an oil in 39% yield from 4-pentenyl tosylate and 5-fluorouracil as described for **3a**. ^{1}H NMR ($CDCl_3$): δ 1.81 (quintet, $J_{1',2'} = J_{2',3'}$ = 7 Hz, 2 H, 2'-H), 2.12 (q, $J_{3',4'}$ = 7 Hz, 2 H, 3'-H), 3.75 (t, 2 H, 1'-H), 5.02 (dd, J_{cis} = 10 Hz, J_{gem} = 1.2 Hz, 1 H, 5'-Ha), 5.05 (dd, J_{trans} = 17 Hz, 1 H, 5'-Hb), 5.80 (ddt, 1 H, 4'-H), 8.09 (d, $J_{6,F}$ = 7 Hz, 1 H, 6-H). ^{13}C NMR ($CDCl_3$): δ 27.6 (C-2'), 30.0 (C-3'), 47.2 (C-1'), 130.2 (d, $^2J_{6,F}$ = 33 Hz, C-6), 139.0 (d, $^1J_{5,F}$ = 229 Hz, C-5), 149.6 (C-2), 157.5 (d, $^2J_{4,F}$ = 26 Hz, C-4).

1-(4,5-Dihydroxypentyl)thymine (**4a**). A solution of 1-(4-pentenyl)thymine (5.41 g, 28 mmol) in formic acid (70 ml) was stirred at 70–80°C for 10 min and cooled to room temperature. Then 30% hydrogen peroxide (1.85 ml, 28 mmol) was added dropwise and the reaction mixture was stirred overnight at room temperature. The formic acid was removed under reduced pressure and 2 M NaOH (100 ml) was added. The mixture was refluxed for 1 h and neutralized with hydrochloric acid. The water was evaporated off under reduced pressure, absolute ethanol was added to the residue and the sodium chloride was removed by filtration and the filtrate was washed with absolute ethanol. The ethanol was removed under reduced pressure and the residue was chromatographed on silica gel with 20% MeOH in $CHCl_3$. The main fraction gave 4.9 g (77%) of **4a** as a viscous oil. This oil (200 mg) was further purified on HPLC (5% EtOH in water) to a colourless oil which crystallized after a few days from ether to give 150 mg of **4a**. M.p. 106–109°C. MS: *m/z* (%) 228 (15, M^+). ^{1}H NMR (CD_3OD): δ 1.3–1.9 (m, 4 H, 2'-H and 3'-H), 1.91 (s, 3 H,. CH_3), 3.49 (d, $J_{4',5'}$ = 5 Hz, 2 H, 5'-H), 3.64 (tt, $J_{3',4'}$ = 7 Hz, 1 H, 4'-H), 3.79 (t, $J_{1',2'}$ = 7 Hz, 2 H, 1'-H), 7.48 (s, 1 H, 6-H). ^{13}C NMR (CD_3OD): δ 12.2 (CH_3), 26.3 (C-2'), 31.1 (C-3'), 49.3 (C-1'), 67.2 (C-5'), 72.7 (C-4'), 111.1 (C-5), 143.2 (C-6), 153.0 (C-2), 166.9 (C-4). Anal. ($C_{11}H_{16}N_2O_4$) C, H, N.

1-(4,5-Dihydroxypentyl)uracil (**4b**). This compound was prepared in 43% yield from **3b** as described for **4a**. After separation on a silica gel column the colorless oil crystallized from acetone. M.p. 139°C. MS: *m/z* (%) 214 (3, M^+). ^{1}H NMR (DMSO-d_6): δ 1.1–1.8 (m, 4 H, 2'-H and 3'-H), 3.27 (d, $J_{4',5'}$ = 7 Hz, 2 H, 5'-H), 3.43 (m, 1 H, 4'-H), 3.68 (t, $J_{1'2'}$ = 7 Hz, 2 H, 1'-H), 3.86 (br, 2 H, 2 × OH), 5.57 (d, J = 8 Hz, 1 H, 5-H), 7.68 (d, 1 H, 6-H), 8.47 (s, 1 H, NH). ^{13}C NMR (DMSO-d_6): δ 24.9 (C-2'), 30.1 (C-3'), 47.7 (C-1'), 65.8 (C-5'), 70.7 (C-4'), 100.7 (C-5), 145.8 (C-6), 151 (C-2), 163.9 (C-4). Anal. ($C_9H_{14}N_2O_4$) C, H, N.

1-(4,5-Dihydroxypentyl)-5-fluorouracil (**4c**). This compound was prepared in 40% yield from **3c** as described for **4a**. 600 mg of **4c** were further purified on HPLC (5% EtOH in water). The colorless oil afforded 400 mg of white crystals after a few days in ether. M.p. 110–113°C. MS: *m/z* (%) 232 (5, M^+). ^{1}H NMR (CD_3OD): δ 1.3–2.0 (m, 4 H, 2'-H and 3'-H), 3.50 (d, $J_{4',5'}$ = 5 Hz, 2 H, 5'-H), 3.64 (tt, $J_{3',4'}$ = 7 Hz, 1 H, 4'-H), 3.79 (t, $J_{1',2'}$ = 7 Hz, 2 H, 1'-H), 7.89 (d, $J_{6,F}$ = 6 Hz, 1 H, 6-H). ^{13}C NMR (CD_3OD): δ 26.1 (C-2'), 30.9 (C-3'), 49.8 (C-1'), 67.2 (C-5'), 72.7 (C-4'), 131.2 (d, $^2J_{6,F}$ = 33 Hz, C-6), 141.6 (d, $^1J_{5,F}$ = 232 Hz, C-5), 151.6 (C-2), 160.0 (d, $^2J_{4,F}$ = 25 Hz). Anal. ($C_9H_{13}FN_2O_4$) C, H, N.

1-(4,5-Diacetoxypentyl)uracil (**5**). Compound **4b** (2.2 g, 0.01 mol) in acetic anhydride (40 ml) was cooled to 0°C and pyridine (120 ml) was added. The ice bath was removed and the progress of the reaction was followed by TLC (5% MeOH in $CHCl_3$). After 4 h the solvents were evaporated off under reduced pressure and the residue was chromatographed on silica gel (5% MeOH in $CHCl_3$) to give 2.89 g (97%) of **5** as a colorless oil. ^{1}H NMR (DMSO-d_6): δ 1.5–1.8 (m, 4 H, 2'-H and 3'-H), 2.06 (s, 3 H, $COCH_3$), 2.08 (s, 3 H, $COCH_3$), 3.75 (t, $J_{1',2'}$ = 7 Hz, 2 H, 1'-H), 4.22 (m, 2 H, 5'-H), 5.07 (m, 1 H, 4'-H), 5.66 (d, J = 8 Hz, 1 H, 5-H), 7.70 (d, 1 H, 6-H), 11.38 (br, 1 H, NH). ^{13}C NMR (DMSO-d_6): δ 20.4, 20.7 (2 × $COCH_3$), 24.2 (C-2'), 26.8 (C-3'), 47.0 (C-1'), 64.2 (C-5'), 70.6 (C-4'), 100.9 (C-5), 145.5 (C-6), 150.9 (C-2), 163.7 (C-4), 170.0, 170.1 (2 × $COCH_3$).

1-(4,5-Diacetoxypentyl)-4-(1,2,4-triazol-1-yl)pyrimidin-2(1H)-one (**6**). Triethylamine (13 ml, 93 mmol) was added dropwise to a stirred, cooled (ice–water bath) mixture of 1,2,4-triazole (2.2 g, 94 mmol), phosphoryl chloride (2.0 ml, 21 mmol) and acetonitrile (55 ml). To these was added a solution of **5** (2.89 g, 9.7 mmol in acetonitrile (35 ml) and the reaction mixture was stirred at room temperature overnight. Triethylamine (10 ml) and water (2 ml) were added and, after 10 min, the solvents were evaporated off under reduced pressure. The residue was partitioned between chloroform (100 ml) and saturated aqueous sodium hydrogen carbonate (80 ml). The organic layer was separated off and the aqueous layer was further extracted with chloroform (2 × 50 ml). The combined organic layers were dried ($MgSO_4$), evaporated under reduced pressure and crystallization from acetone afforded 2.49 g (74%) of **6**. M.p. 125–126°C. ^{1}H NMR (DMSO-d_6): δ 1.5–1.8 (m, 4 H, 2'-H and 3'-H), 2.01 (s, 3 H, $COCH_3$), 2.03 (s, 3 H, $COCH_3$), 3.94 (t, $J_{1',2'}$ = 7 Hz, 2 H, 1'-H), 4.11 (m, 2 H, 5'-H), 5.00 (m, 1 H, 4'-H), 6.95 (d, J = 7 Hz, 1 H, 5-H), 8.40 (s, 1 H, 5''-H), 8.49 (d, 1 H, 6-H), 9.41 (s, 1 H, 3''-H). ^{13}C NMR (DMSO-d_6): δ 20.4, 20.6 (2 × $COCH_3$), 23.9 (C-2'), 26.8 (C-3'), 49.9 (C-1'), 64.2 (C-5'), 70.5 (C-4'), 93.4 (C-5), 143.3 (C-5''), 153.2, 153.9 (C-3'' and C-6), 154.2 (C-2), 158.4 (C-4), 169.9, 170.0 (2 × $COCH_3$).

1-(4,5-Dihydroxypentyl)cytosine (**7a**). A solution of **6** (430 mg, 1.23 mmol) and 25% aqueous ammonia (4 ml) in dioxane (10 ml) was stirred for 2 days at room temperature and evaporated under reduced pressure. The oil obtained was chromatographed (gradient 0–10% MeOH in $CHCl_3$) to give an oil which crystallized from ether–petroleum ether to give 120 mg (46%) of **7a**. M.p. 140–143°C. MS: *m/z* (%) 213 (2, M^+). ^{1}H NMR (DMSO-d_6): δ 1.1–1.8 (m, 4 H, 2′-H and 3′-H), 3.1–3.6 (m, 4 H, 5′-H, 4′-H and OH), 3.62 (t, $J_{1',2'} = 7$ Hz, 2 H, 2′-H), 4.54 (br, 1 H, OH), 5.65 (d, $J = 7$ Hz, 1 H, 5-H), 7.03 (br s, 2 H, NH_2), 7.57 (d, 1 H, 6-H). ^{13}C NMR (DMSO-d_6): δ 25.2 (C-2′), 30.2 (C-3′), 48.8 (C-1′), 58.8 (C-5′), 70.8 (C-4′), 93.0 (C-5), 146.0 (C-6), 155.8 (C-2), 165.8 (C-4). Anal. ($C_9H_{15}N_3O_3 \cdot \frac{1}{2} H_2O$) C, H, N.

1-(4,5-Dihydroxypentyl)-N^4-methylcytosine (**7b**). This compound was prepared in 15% yield from **6** (349 mg, 1 mmol) and 40% aqueous methylamine (6 ml) in dioxane (18 ml) as described for **7a**. Reaction time was 12 h. After chromatography the oil was further purified on HPLC (2% EtOH in water) to give 50 mg of **7b** as an oil. MS: *m/z* (%) 227 (5, M^+). ^{1}H NMR (CH_3OD): δ 1.3–2.0 (m, 4 H, 2′-H and 3′-H), 2.91 (s, 3 H, CH_3), 3.48 (d, $J_{4',5'} = 5$ Hz, 2 H, 5′-H), 3.64 (tt, $J_{3',4'} = 7$ Hz, 1 H, 4′-H), 3.81 (t, $J_{1',2'} = 7$ Hz, 2 H, 1′-H), 5.83 (d, $J = 7$ Hz, 1 H, 5-H), 7.50 (d, 1 H, 6-H). ^{13}C NMR (CD_3OD): δ 26.5 (C-2′), 27.7 (CH_3), 31.2 (C-3′), 50.8 (C-1′), 67.2 (C-5′), 72.8 (C-4′), 96.7 (C-5), 145.7 (C-6), 159.5 (C-2), 166.3 (C-4). HRMS $C_{10}H_{17}N_3O_3$: Calcd. 227.1269. Found 227.1275.

1-(4,5-Dihydroxypentyl)-N^4-ethylcytosine (**7c**). This compound was prepared in 51% yield from **6** (349 mg, 1 mmol) and 50% aqueous ethylamine (6 ml) as described for **7a**. Reaction time was 16 h and **7c** was isolated as an oil. MS: *m/z* (%) 241 (12, M^+). ^{1}H NMR (CD_3OD): δ 1.22 (t, $J_{1'',2''} = 7$ Hz, 3 H, 2″-H), 1.3–1.9 (m, 4 H, 2′-H and 3′-H), 3.3–3.6 (m, 4 H, 2″-H and 5′-H), 3.66 (m, 1 H, 4′-H), 3.82 (t, $J_{1',2'} = 7$ Hz, 2 H, 1′-H), 5.91 (d, $J = 7$ Hz, 1 H, 5-H), 7.55 (d, 1 H, 6-H). ^{13}C NMR (CD_3OD): δ 14.4 (C-2″), 26.4 (C-2′), 31.0 (C-3′), 36.4 (C-1″), 50.7 (C-1′), 67.0 (C-5′), 72.6 (C-4′), 97.0 (C-5), 145.7 (C-6), 159.4 (C-2), 165.3 (C-4). HRMS $C_{11}H_{19}N_3O_3$: Calcd. 241.1426. Found 241.1414.

1-(4,5-Dihydroxypentyl)-N^4-(2-hydroxyethyl)cytosine (**7d**). This compound was prepared in 29% yield from **6** (330 mg, 0.95 mmol) and ethanolamine (6 ml) as described for **7a**. After chromatography the oil was further purified on HPLC (2% EtOH in water) to give **7d** as an oil. MS: *m/z* (%) 257 (5, M^+). ^{1}H NMR (CD_3OD): δ 1.3–2.0 (m, 4 H, 2′-H and 3′-H), 3.4–3.7 (m, 7 H, 4′-H, 5′-H, 1″-H and 2″-H), 3.82 (t, $J_{1',2'} = 7$ Hz, 2 H, 1′-H), 5.89 (d, $J = 7$ Hz, 1 H, 5-H), 7.52 (d, 1 H, 6-H). ^{13}C NMR (CD_3OD): δ 26.5 (C-2′), 31.2 (C-3′), 44.1 (C-1″), 50.8 (C-1′), 61.5 (C-2″), 67.2 (C-5′), 72.8 (C-4′), 96.8 (C-5), 146.0 (C-6), 159.4 (C-2), 166.0 (C-4). HRMS $C_{11}H_{19}N_3O_4$: Calcd. 257.1375. Found 257.1362.

1-(4,5-Epoxypentyl)thymine (**8a**). A mixture of *m*-chloroperbenzoic acid (mCPBA) (9.87 g, 57.2 mmol), KF (3.32 g, 57.2 mmol) and **3a** (1.0 g, 5.56 mmol) in CH_2Cl_2 (100 ml) was stirred at room temperature overnight. The reaction mixture was filtered and freshly activated (1 h, 100°C, 0.1 Torr) KF (3.32 g, 57.2 mmol) was added to ensure complete elimination of the acids. Filtration and evaporation under reduced pressure afforded crude epoxide which was used without any further purification for preparation of **10**. ^{13}C NMR ($CDCl_3$): δ 11.9 (CH_3), 25.3 (C-2′), 28.7 (C-3′), 46.4 (C-5′), 47.6 (C-1′), 51.3 (C-4′), 110.1 (C-5), 140.3 (C-6), 150.8 (C-2), 164.3 (C-4).

1-(4,5-Epoxypentyl)uracil (**8b**). This compound was prepared from **3b** (1.0 g, 5.56 mmol) as described for **8a**. The crude epoxide was used without further purification for the preparation of **9a,b**. ^{13}C NMR ($CDCl_3$): δ 25.5 (C-2′), 28.8 (C-3′), 46.5 (C-5′), 48.2 (C-1′), 51.5 (C-4′), 102.0 (C-5), 144.6 (C-6), 150.9 (C-2), 164.0 (C-4).

1-(5-Hydroxy-4-methoxypentyl)uracil (**9a**) *and 1-(4-hydroxy-5-methoxypentyl)uracil* (**9b**). To the crude **8b** (5.56 mmol) was added 95% sulfuric acid (0.1 ml) in methanol (50 ml) and the reaction mixture was stirred overnight at room temperature. The methanol was evaporated off under reduced pressure and the obtained oil was chromatographed (5% EtOH in $CHCl_3$) to give 720 mg (57%) of a mixture of the two isomers **9a** and **9b** (1 : 3). After repeated chromatography 300 mg of **9b** were isolated as an oil.

9a: ^{13}C NMR ($CDCl_3$): δ 24.8 (C-2′), 27.2 (C-3′), 48.5 (C-1′), 57.0 (OCH_3), 62.6 (C-5′), 80.8 (C-4′), 101.9 (C-5), 144.5 (C-6), 151.0 (C-2), 164.2 (C-4).

9b: MS: *m/z* (%) 228 (3, M^+). ^{1}H NMR ($CDCl_3$): δ 1.5–2.0 (m, 4 H, 2′-H and 3′-H), 3.44 (d, $J_{4',5'} = 6$ Hz, 2 H, 5′-H), 3.47 (s, 3 H, OCH_3), 3.7–3.9 (m, 3 H, 1′-H and 4′-H), 5.79 (d, $J = 8$ Hz, 1 H, 5-H), 7.42 (d, 1 H, 6-H), 10.49 (br, 1 H, NH). ^{13}C NMR ($CDCl_3$): δ 25.0 (C-2′), 29.3 (C-3′),. 48.4 (C-1′), 58.7 (OCH_3), 69.4 (C-4′), 76.7 (C-5′), 101.8 (C-5), 144.7 (C-6), 151.0 (C-2), 164.2 (C-4). HRMS $C_{10}H_{16}N_2O_4$: Calcd. 228.1111. Found 228.1112.

1-(5-Chloro-4-hydroxypentyl)thymine (**10**). 50 ml of dry methanol were saturated with hydrogen chloride and added to the crude **8a** (14.3 mmol) and the reaction mixture was stirred overnight at room temperature. The oil obtained was chromatographed (5% MeOH in CH_2Cl_2) to give 300 mg (9%) of **10** as white crystals. M.p. 143–145°C. MS: *m/z* (%) 246 (19, M^+), 248 (6, $M^+ + 2$). ^{1}H NMR (DMSO-d_6): δ 1.2–1.7 (m, 4 H, 2′-H and 3′-H), 1.77 (s, 3 H, CH_3), 3.4–3.7 (m, 5 H, 1′-H, 4′-H and 5′-H), 5.12 (d, $J = 5$ Hz, 1 H, OH). 7.54 (s, 1 H, 6-H), 11.2 (s, 1 H, NH). ^{13}C NMR (DMSO-d_6): δ 11.9 (q, CH_3), 24.7 (t, C-2′), 30.6 (t, C-3′), 47.0 (t, C-1′), 49.6

(t, C-5'), 69.5 (d, C-4'), 108 (s, C-5), 141.4 (d, C-6), 150.9 (s, C-2), 164.3 (s, C-4). HRMS $C_{10}H_{15}ClN_2O_3$: Calc. 246.0771. Found 246.0760.

6-O-p-Toluenesulfonyl-D-glucal (**13**). A stirred solution of D-glucal **12** (26.9 g, 0.184 mol) in dry pyridine (150 ml) was cooled with an ice–water bath and *p*-toluenesulphonyl chloride (35.1 g, 0.184 mol) was added portionwise. The reaction mixture was left overnight at 5°C and the pyridine was evaporated off under reduced pressure. The oil was applied to a silica gel column and eluted with CH_2Cl_2 to give 20.8 g (38%) of **13** as a colorless oil. ^{1}H NMR ($CDCl_3$): δ 2.40 (s, 3 H, CH_3), 3.69 (m, 1 H, 4'-H), 3.91 (m, 1 H, 3'-H), 4.1–4.4 (m, 3 H, 5'-H and 6'-H), 4.6–4.8 (m, 3 H, 3'-H and 2 × OH), 6.15 (d, 6 Hz, 1 H, 1'-H), 7.34 (d, J = 8 Hz, 2 H, aryl), 7.78 (d, 2 H, aryl). ^{13}C NMR ($CDCl_3$): δ 21.2 (CH_3), 68.4, 68.7, 69.0 (C-4', C-5' and C-6'), 75.3 (C-3'), 103.1 (C-2'), 127.6 (C-2), 129.5 (C-3), 132.2 (C-1), 143.1 (C-1'), 144.6 (C-4).

3,4-Di-O-acetyl-6-O-p-toluenesulfonyl-D-glucal (**14**). A stirred solution of **13** (15.5 g, 52 mmol) in CH_2Cl_2 (150 ml) was cooled to 0°C and acetic anhydride (20 ml, 210 mmol), pyridine (17 ml, 210 mmol) and 4-dimethylaminopyridine (0.5 g, 4 mmol) was added and the reaction was stirred at room temperature overnight. The mixture was evaporated under reduced pressure and afforded **14** as white crystals which were washed with acetone (15.0 g, 75%). M.p. 100–101°C. ^{1}H NMR ($CDCl_3$): δ 2.03 (s, 6 H, 2 × $COCH_3$), 2.45 (s, 3 H, CH_3), 4.2–4.3 (m, 3 H, 5'-H and 6'-H), 4.82 (dd, $J_{1',2'}$ = 6.1, $J_{2',3'}$ = 3.4, 1 H, 2'-H), 5.13 (m, 1 H, 4'-H), 5.27 (m, 1 H, 3'-H), 6.35 (d, 1 H, 1'-H), 7.36 (d, J = 8 Hz, 2 H, aryl), 7.70 (d, 2 H, aryl). ^{13}C NMR ($CDCl_3$): δ 20.5, 20.7 (2 × CO-CH_3), 21.4 (CH_3), 66.3, 66.5, 66.8 (C-4', C-5' and C-6'), 73.0 (C-3'), 98.7 (C-2'), 127.8 (C-2), 129.7 (C-3), 132.4 (C-1), 144.9 (C-4), 145.1 (C-1'), 169.2, 170.0 (2 × $COCH_3$).

3,4-Di-O-acetyl-6-deoxy-6-thymin-1-yl-D-glucal (**15a**). This compound was prepared from thymine (8.40 g, 66 mmol), 50% oil-immersed sodium hydride (2.24 g, 47 mmol) and **14** (15.36 g, 40 mmol) as described for **3a**. The reaction time was 36 h. After silica gel chromatography (gradient 0–10% MeOH in CH_2Cl_2) **15a** (TLC 10% MeOH in CH_2Cl_2, R_f = 0.50) was crystallized from ether–petroleum ether to give 3.82 g (28%). M.p. 78–81°C. ^{1}H NMR (DMSO-d_6): δ 1.76 (s, 3 H, CH_3), 2.03 (s, 3 H, $COCH_3$), 2.06 (s, 3 H, $COCH_3$), 3.98 (m, 2 H, 6'-H), 4.40 (m, 1 H, 5'-H), 4.88 (dd, $J_{1',2'}$ = 6.1 Hz, $J_{2',3'}$ = 3.6 Hz, 1 H, 2'-H), 4.99 (m, 1 H, 4'-H), 5.19 (m, 1 H, 3'-H), 6.61 (d, 1 H, 1'-H), 7.44 (s, 1 H, 6-H), 11.32 (br, 1 H, NH). ^{13}C NMR ($CDCl_3$): δ 11.9 (CH_3), 20.6, 20.7 (2 × $COCH_3$), 46.4 (C-6'), 65.7 (C-5'), 67.7 (C-4'), 73.1 (C-3'), 98.6 (C-2'), 108.4 (C-5), 141.6 (C-6), 145.4 (C-1'), 150.9 (C-2), 164.1 (C-4), 169.3, 169.7 (2 × $COCH_3$).

3,4-Di-O-acetyl-6-deoxy-6-(N^4-isobutyrylcytosin-1-yl)-D-glucal (**15b**). This compound as prepared in 36% yield from N^4-isobutyrylcytosine (4.53 g, 25 mmol), 50% oil-immersed sodium hydride (1.2 g, 25 mmol) and **14** (9.50 g, 25 mmol) as described for **15a**. M.p. 148–151°C. ^{1}H NMR (DMSO-d_6): δ 1.08 (d, J = 7 Hz, 6 H, CH_3), 2.04 (s, 3 H, $COCH_3$), 2.07 (s, 3 H, $COCH_3$), 2.72 (septet, 1 H, CH), 4.15 (m, 2 H, 6'-H), 4.47 (m, 1 H, 5'-H), 4.91 (dd, $J_{1',2'}$ = 6.2 Hz, $J_{2',3'}$ = 3.6 Hz, 1 H, 2'-H), 5.03 (m, 1 H, 4'-H), 5.20 (m, 1 H, 3'-H), 6.60 (d, 1 H, 1'-H), 7.21 (d, J = 7 Hz, 1 H, 5-H), 7.99 (d, 1 H, 6-H), 10.84 (br, 1 H, NH). ^{13}C NMR (DMSO-d_6): δ 18.9 (CH_3), 20.5, 20.7 (2 × $COCH_3$), 34.8 (CH), 48.9 (C-6'), 65.6 (C-5'), 67.8 (C-4'), 72.3 (C-3'), 95.1 (C-5), 98.7 (C-2'), 145.3 (C-1'), 150.9 (C-6), 155.0 (C-2), 162.6 (C-4), 169.3, 169.7 (2 × $COCH_3$), 177.7 (NH–CO).

General procedure for the preparation of **16a,b**. To a stirred solution of the glucal **15a,b** (2.5 mmol) in dioxane (4 ml) was added mercuric sulfate (100 mg) and sulfuric acid (5 mM, 40 ml). The reaction mixture was stirred for 4 h at room temperature and barium carbonate was added for neutralization. The suspension was filtered and the filtrate was evaporated under reduced pressure. The residue was dissolved in CH_2Cl_2 (100 ml) and dried over Na_2SO_4. Evaporation of the solvent afforded, in 95% yield, the desired product which was crystallized form ether–petroleum ether.

(2E,4S,5R)-4-Acetoxy-5-hydroxy-6-thymin-1-yl-2-hexenal (**16a**). M.p. 116–120°C. ^{1}H NMR ($CDCl_3$): δ 1.79 (s, 3 H, CH_3), 2.19 (s, 3 H, $COCH_3$), 3.49 (m, 1 H, 5'-H), 4.18 (m, 2 H, 6'-H), 4.44 (m, 1 H, OH), 5.69 (m, 1 H, 4'-H), 6.26 (ddd, $J_{2',3'}$ = 16 Hz, $J_{1',2'}$ = 8 Hz, $J_{2',4'}$ = 1.4 Hz, 1 H, 2'-H), 6.98 (dd, $J_{3',4'}$ = 5 Hz, 1 H, 3'-H), 7.14 (s, 1 H, 6-H), 9.59 (d, 1 H, 1'-H), 10.74 (s, 1 H, NH). ^{13}C NMR ($CDCl_3$): δ 11.9 (CH_3), 20.7 ($COCH_3$), 51.7 (C-6'), 68.8 (C-5'), 73.7 (C-4'), 109.1 (C-5), 132.7 (C-2'), 143.3 (C-6), 150.2 (C-3'), 151.4 (C-2), 165 (C-4), 169.8 ($COCH_3$), 193.1 (C-1').

(2E,4S,5R)-4-Acetoxy-5-hydroxy-6-(N^4-isobutyryl-cytosin-1-yl)-2-hexenal (**16b**). ^{1}H NMR ($CDCl_3$): δ 1.19 (d, J = 7 Hz, 6 H, CH_3), 2.19 (s, 3 H, $COCH_3$), 2.69 (septet, 1 H, CH), 3.69 (m, 1 H, 5'-H), 4.2–4.3 (m, 3 H, 6'-H and OH), 5.81 (m, 1 H, 4'-H), 6.25 (dd, $J_{2',3'}$ = 16 Hz, $J_{1',2'}$ = 8 Hz, 1 H, 2'-H), 6.97 (dd, $J_{3',4'}$ = 4 Hz, 1 H, 3'-H), 7.36 (d, J = 7 Hz, 1 H, 5-H), 7.73 (d, 1 H, 6-H), 9.56 (d, 1 H, 1'-H). ^{13}C NMR ($CDCl_3$): δ 18.9 (CH_3), 20.8 ($COCH_3$), 36.3 (CH), 52.9 (C-6'), 69.1 (C-5'), 73.6 (C-4'), 96.6 (C-5), 132.6 (C-2'), 150.6 (C-3' and C-6), 156.7 (C-2), 165.7 (C-4), 170.1 ($COCH_3$), 178.1 (NH–CO), 193.1 (C-1').

(Z)-6-(Thymin-1-yl)-2,3,6-trideoxy-D-erythro-2-hexenitol (**17a**). To a stirred solution of **16a** (800 mg, 2.7 mmol) in methanol (50 ml) was added sodium borohydride (720 mg, 19 mmol) portionwise over 0.5 h and the reac-

tion mixture was stirred overnight. After neutralization with conc. HCl the solvent was removed under reduced pressure and the residue almost dissolved in hot absolute ethanol. After cooling the NaCl was filtered off and the filtrate was evaporated. The oil obtained was applied to a silica gel column and eluted with 5% MeOH in CH_2Cl_2 to give **18a** (200 mg, 26%, $R_f = 0.40$, $\alpha : \beta = 2 : 1$) and **17a** (50 mg, 7%, $R_f = 0.16$) as oils.

17a: FAB MS: *m/z* (%) 257 (72, $M + H^+$). ^{1}H NMR (DMSO-d_6): δ 1.75 (s, 3 H, CH_3), 3.2–3.5 (m, 6 H, 5′-H, 6′-H and 3 × OH), 3.56 (m, 1 H, 4′-H), 3.92 (dd, J_{gem} = 14 Hz, $J_{1'a,2'}$ = 5 Hz, 1′-Ha), 3.99 (dd, $J_{1'b,2'}$ = 2.5 Hz, 1 H, 1′-Hb), 5.02 (m, 1 H, 2′-H), 5.73 (m, 1 H, 3′-H), 7.39 (s, 1 H, 6-H), 11.19 (s, 1 H, NH). ^{13}C NMR (DMSO-d_6): δ 11.9 (CH_3), 50.5 (C-6′), 61.1 (C-1′), 71.4 (C-5′), 72.9 (C-4′), 107.2 (C-5), 129.7 (C-2′), 131.1 (C-3′), 143.1 (C-6), 151.0 (C-2), 164.4 (C-4).

18a: MS: *m/z* (%) 286 (10%, M^+).

18aα: ^{13}C NMR (DMSO-d_6): δ 12.0 (CH_3), 35.4 (C-2′), 49.3 (C-6′), 56.6 (OCH_3), 69.3 (C-5′), 72.7 (C-4′), 77.4 (C-3′), 90.5 (C-1′), 107.6 (C-5), 142.8 (C-6), 150.9 (C-2), 164.5 (C-4).

18aβ: ^{13}C NMR (DMSO-d_6): δ 12.0 (CH_3), 37.6 (C-2′), 49.3 (C-6′), 56.5 (OCH_3), 72.1 (C-4′), 73.2 (C-5′), 79.8 (C-3′), 93.5 (C-1′), 107.6 (C-5), 142.8 (C-6), 151.0 (C-2), 164.4 (C-4).

(Z)-*6-(*N^4*-Isobutyrylcytosin-1-yl)-2,3,6-trideoxy-*D*-erythro-2-hexenitol* (**17b**). Compound **16b** (850 mg, 2.4 mmol) and sodium borohydride (640 mg, 17.0 mg) were mixed and added portionwise over 0.5 h to stirred methanol (50 ml). The reaction mixture was stirred for 3 h at room temperature and neutralized with conc. HCl. The solvent was evaporated off under reduced pressured and the residue almost dissolved in hot absolute ethanol. After cooling the NaCl was filtered off and the filtrate was evaporated. After chromatography (5% MeOH in CH_2Cl_2) 150 mg (21%) of **17b** was obtained as an oil. ^{1}H NMR (DMSO-d_6): δ 1.08 (d, J = 7 Hz, 6 H, CH_3), 2.72 (septet, 1 H, CH), 3.20 (br, 3 H, 3 × OH), 3.4–3.7 (m, 3 H, 5′-H and 6′-H), 3.99 (m, 1 H, 4′-H), 4.0–4.4 (m, 2 H, 1′-Ha and 1′-Hb), 5.10 (m, 1 H, 2′-H), 5.76 (m, 1 H, 3′-H), 7.19 (d, J = 7 Hz, 1 H, 5-H), 7.93 (d, 1 H, 6-H). ^{13}C NMR (DMSO-d_6): δ 19.1 (CH_3), 35.1 (CH), 52.9 (C-6′), 61.4 (C-1′), 71.2 (C-5′), 73.2 (C-4′), 94.9 (C-5), 129 (C-2′), 131.4 (C-3′), 151.8 (C-6), 156.1 (C-2), 162.5 (C-4), 177.9 (NHCO).

(Z)-*6-(Cytosin-1-yl)-2,3,6-trideoxy-*D*-erythro-2-hexenitol* (**17c**). This compound was prepared by dissolving **17b** (150 mg) in methanolic ammonia (50 ml). After 5 days at room temperature the solvent was evaporated off under reduced pressure and the oil was chromatographed (gradient 5–15% MeOH in CH_2Cl_2) to give **17c** as an oil (50 mg, 43%). FAB MS: *m/z* (%) 242 (77, $M + H^+$), ^{1}H NMR (DMSO-d_6): δ 3.4–3.7 (m, 6 H, 5′-H, 6′-H and 3 × OH), 3.82 (m, 1 H, 4′-H), 3.9–4.1 (m, 2 H, 5′-Ha and 1′-Hb), 5.10 (m, 1 H, 2′-H), 5.6–5.8 (m, 2 H, 3′-H and 5-H), 7.49 (d, J = 7 Hz, 1 H, 6-H). ^{13}C NMR (DMSO-d_6): δ 51.6 (C-6′), 61.1 (C-1′), 72.0 (C-5′), 72.5 (C-4′), 92.8 (C-5), 129.8 (C-2′), 130.9 (C-3′), 147.5 (C-6), 156.7 (C-2), 166.0 (C-4).

References

1. Gallo, R. C., Salahuddin, S. Z., Popovic, M., Shearer, G. M., Kaplan, M., Haynes, B. F., Palker, T. J., Redfield, R., Olesky, J., Safari, B., White, G., Foster, P. and Markham, P. D. *Science 224* (1984) 500.
2. De Clercq, E. *Adv. Drug Res. 17* (1988) 1.
3. Haines, D. R., Tseng, C. K. H. and Marquez, V. E. *J. Med. Chem. 30* (1987) 943.
4. Hayashi, S., Phadtarc, S., Zemlicka, J., Matsukura, M., Mitsuya, H. and Broder, S. *Proc. Natl. Sci. USA 85* (1988) 6127.
5. Brooks, L. A. and Snyder, H. R. *Org. Synth. Coll. Vol. 3*, 698.
6. Cerny, M. *Collect. Czech. Chem. Commun. 42* (1977) 3069.
7. Sasaki, T., Minamoto, K., Suzuki, T. and Yamashita, S. *Tetrahedron 36* (1980) 865.
8. Roveri, P., Caurini, V. and Gatti, R. *Il Farmaco-Ed. Sci. 39* (1984) 346.
9. Divakar, K. J. and Reese, C. B. *J. Chem. Soc., Perkin Trans. 1* (1982) 1171.
10. Sung, W. L. *J. Org. Chem. 47* (1982) 3623.
11. Camps, F., Coll, J., Messeguer, A. and Pericàs, M. A. *Tetrahedron Lett. 22* (1981) 3895.
12. Swern, D., Billen, G. N. and Knight, H. B. *J. Am. Chem. Soc. 71* (1949) 1152.
13. Corey, E. J. and Suggs, J. W. *Tetrahedron Lett.* (1975) 2647.
14. Whistler, R. L. and Wolfram, M. L. *Methods in Carbohydrate Chemistry*, Vol. II, Academic Press, New York and London 1963.
15. Sugiura, Y., Furuya, S. and Furukawa, Y. *Chem. Pharm. Bull. 36* (1988) 3253.
16. Gonzalez, F., Lesage, S. and Perlin, A. S. *Carbohydr. Res. 42* (1975) 267.

Received November 20, 1992.