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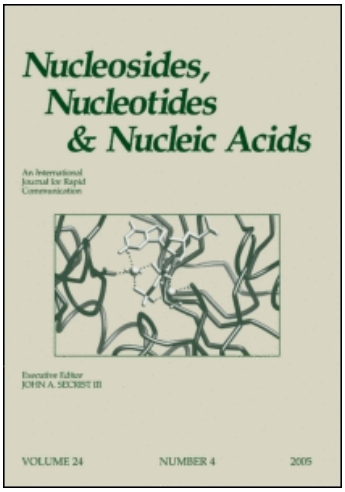

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Rakesh Kumar[a]; Edward E. Knaus[a]; Leonard I. Wiebe[a]
[a] Faculty of Pharmacy and Pharmaceutical Sciences, University of Alberta, Edmonton, Alberta, Canada

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NUCLEOSIDES & NUCLEOTIDES, 11(6), 1219-1228 (1992)

SYNTHESIS AND ANTI-HIV ACTIVITY OF 3'-*O*-FORMYL DERIVATIVES OF THYMIDINE AND 2'-DEOXYURIDINE

Rakesh Kumar, Edward E. Knaus and Leonard I. Wiebe*
Faculty of Pharmacy and Pharmaceutical Sciences,
University of Alberta, Edmonton, Alberta, Canada T6G 2N8

ABSTRACT: Reaction of the 5'-*O*-*t*-butyldimethylsilyl derivatives of thymidine and 2'-deoxyuridine with the Vilsmeier reagent ($POCl_3$/DMF), and removal of the *t*-butyldimethylsilyl protecting group, afforded 3'-*O*-formylthymidine (**5**) and 3'-*O*-formyl-2'-deoxyuridine (**6**), respectively. *In vitro* evaluation, determined as the ability of the test compound to inhibit HIV induced cytopathogenicity in CEM cells, indicated that **5** was moderately active, whereas **6** was inactive.

The clinical efficacy of 3'-azido-3'-deoxythymidine (AZT, **1a**), and related 2',3'-dideoxythymidines, for the treatment of human immunodeficiency virus (HIV) has stimulated the design and evaluation of structurally related thymidine analogues. Structural and electronic studies suggest that antiviral activity may be correlated with the presence of an electronegative atom at the C-3' position.[1] A quantitative structure-activity relationship study indicated that a small hydrophobic substituent at C-5 and a flat substituent at the C-3' position provided the best correlation with anti-HIV activity.[2] Replacement of the 3'-hydroxyl substituent present in thymidine by an azido substituent enhances binding to the nucleotide binding site of reverse transcriptase, the target viral RNA dependent DNA polymerase, and also precludes further elongation into the nascent strand. A comparison of the x-ray crystal structures of AZT[3,4] and thymidine 3'-phosphate[5] indicated that the N-3'-α and N-3'-γ azido nitrogen atoms occupy positions that are similar to those of the corresponding thymidine 3'-phosphate ester oxygen atoms when the thymine bases are superimposed.[3] Thus, it has been proposed that the charge distribution of the azido group may mimic the charge distribution on the phosphate group, and that these groups are accommodated at the nucleotide binding site in reverse transcriptase.

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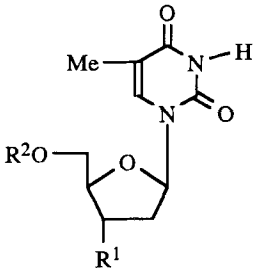
Electronically analogous compounds to AZT have been synthesized that contain a 3'-cyano, ethynyl, thiocyano, isothiocyano and isocyano substituent.[6] 3'-Formamido-3'-deoxythymidine (**1b**), which was inactive against HIV-1,[7] and a synthetic intermediate 5'-*O*-*t*-butyldimethylsilyl-3'-*O*-formylmethyl-3'-deoxythymidine (**1c**)[8] have also been reported. These latter results prompted us to investigate the 3'-*O*-formyl-3'-deoxythymidine (**5**), in which the 3'-*O*-formyl substituent was expected to be isosteric with the 3'-azido substituent present in AZT. This assumption was based on the expectation that the charge distribution in the 3'-*O*-formyl group of **5** could mimic the charge distribution in a 3'-azido group and a 3'-O-P-O bond.[3,4] Although 3'-*O*-formylthymidine and 3'-*O*-formyl-(*N*-acyl)-2'-deoxyribonucleosides have been used as building units in the synthesis of oligodeoxyribonucleotides,[9-11] their anti-HIV activity, to the best of our knowledge, has not been reported.



1a ; $R^1 = N_3$, $R^2 = H$

1b ; $R^1 = NHCHO$, $R^2 = H$

1c ; $R^1 = CH_2CHO$, $R^2 = Me_3CSi(Me)_2$-

5-Iodo-2'-deoxyuridine (**2a**) was used as a model compound in the initial studies to develop the synthetic methodology. Thus, reaction of **2a** with pivaloyl chloride in dry pyridine afforded 5-iodo-5'-*O*-pivaloyl-2'-deoxyuridine (**3a**, 49%). Reaction of **3a**, and 5'-*O*-pivaloylthymidine (**3b**),[12] with the Vilsmeier reagent $POCl_3$ in DMF at 0°C yielded 3'-*O*-formyl-5-iodo-5'-*O*-pivaloyl-2'-deoxyuridine (**4a**) and 3'-*O*-formyl-5'-*O*-pivaloylthymidine (**4b**) in 56 and 55% yields, respectively. It was necessary to protect the 5'-OH substituent of the 2'-deoxyuridines **2a-c**, since reaction with the Vilsmeier reagent was also expected to convert the 5'-OH substituent to a 5-chloro substituent.[13,14] Thus, a related reaction of **2a** with $POCl_3$ in the presence of Et_4N^+ Cl^- and *N,N*-diethylaniline in dry acetonitrile gave 3',5'-dichloro-5-iodo-2',3',5'-trideoxyuridine (**7**, 41% yield). Deprotection of **4a** and **4b** using either n-Bu_4N^+ F^- (TBAF) in tetrahydrofuran (Method A, Scheme 1), or a saturated solution of NH_3 in MeOH (Method B, Scheme 1), also cleaved the 3'-*O*-formyl moiety to yield 5-iodo-5'-*O*-pivaloyl-2'-deoxyuridine (**3a**) and 5'-*O*-pivaloylthymidine (**3b**), respectively. These results indicate that the 3'-*O*-formyl group is rapidly cleaved under basic reaction conditions. Therefore, the 5'-*O*-*t*-butyldimethylsilyl (TBDMS) derivatives of thymidine

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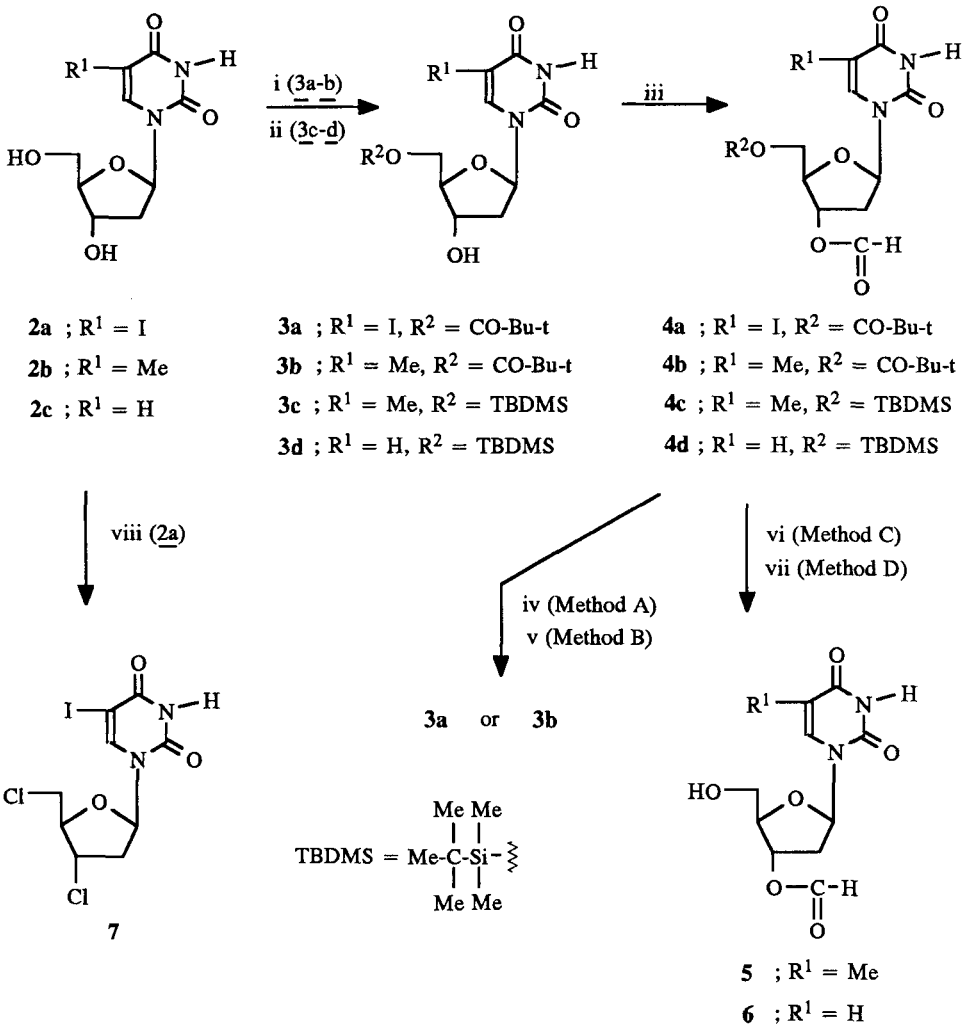

2a ; R^1 = I
2b ; R^1 = Me
2c ; R^1 = H

3a ; R^1 = I, R^2 = CO-Bu-t
3b ; R^1 = Me, R^2 = CO-Bu-t
3c ; R^1 = Me, R^2 = TBDMS
3d ; R^1 = H, R^2 = TBDMS

4a ; R^1 = I, R^2 = CO-Bu-t
4b ; R^1 = Me, R^2 = CO-Bu-t
4c ; R^1 = Me, R^2 = TBDMS
4d ; R^1 = H, R^2 = TBDMS

7

5 ; R^1 = Me
6 ; R^1 = H

Reagents: i, pivaloyl chloride, pyridine; ii, *t*-butyldimethylsilyl chloride, imidazole, DMF: iii, $POCl_3$, DMF, 0°C; iv, *n*-Bu_4N^+ F^-, THF, 25°C (for **4a** and **4b**), Method A; v, NH_3 in MeOH, 25°C (for **4a** and **4b**), Method B; vi, 50°C under vacuum (4 mm Hg) (for **4c** and **4d**), Method C; vii, 80% acetic acid, 60°C (for **4c** and **4d**), Method D; viii, Et_4N^+ Cl^-, *N,N*-diethylaniline, $POCl_3$, dry MeCN, reflux.

SCHEME 1

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(**3c**) and 2'-deoxyuridine (**3d**) were prepared since it was expected that the 3'-*O*-formyl group would be stable under the mild reaction conditions required to remove the 5'-*O*-TBDMS blocking group. It has been reported that the acid labile *O*-formyl group was not hydrolyzed employing mild acidic conditions required to cleave 5'-*O*-trityl derivatives.[15]

The reaction of 5'-*O*-TBDMS-thymidine (**3c**) with $POCl_3$ in DMF at 0°C afforded 3'-*O*-formyl-5'-*O*-TBDMS-thymidine (**4c**, 69%). In contrast, when the unpurified reaction product was heated at 50°C *in vacuo* (4 mm Hg), 3'-*O*-formylthymidine (**5**) was obtained in 53% yield (Method C, Scheme 1). Removal of the 5'-*O*-TBDMS group may be due to traces of phosphoric acid generated during the isolation procedure. Alternatively, treatment of **4c** with 80% acetic acid at 60°C yielded **5** in 51% yield. A similar reaction of 5'-*O*-TBDMS-2'-deoxyuridine (**3d**) with $POCl_3$ in DMF at 0°C, and heating the unpurified reaction product at 50°C under vacuum afforded 3'-*O*-formyl-5'-*O*-TBDMS-2'-deoxyuridine (**4d**, 27%) and 3'-*O*-formyl-2'-deoxyuridine (**6**, 27%). Deprotection of **4d** using 80% acetic acid gave **6** in 57% yield.

3'-*O*-Formylthymidine (**5**) and 3'-*O*-formyl-2'-deoxyuridine (**6**), were cytotoxic to uninfected host cells at concentrations greater than 2×10^{-5}M and 1×10^{-4}M, respectively. The *in vitro* anti-HIV assays indicated that **5** protected 30% of T_4 lymphocytes (CEM cell line), against HIV induced cytolysis at its non-cyctoxic host cell concentration of 2×10^{-5} M. In contrast, **6** provided less than 15% protection in the same assay at its non cytotoxic host cell concentration of 1×10^{-4}M.

These results indicate that **5** was a moderately active, whereas **6** was an inactive, anti-HIV agent. This antiviral screen is designed to detect agents, which interact with virions, cell or virus gene products to exhibit an antiviral effect, that protects cells from cytolysis at any stage of the virus reproductive cycle.[16] The anti-HIV activities obtained for **5** and **6** are readily explained by the known structure-activity correlation that a small hydrophobic substituent such as methyl at C-5 is required for activity.[2] The differences in anti-HIV activity exhibited by **5** and **6** may also be due, at least in part, to differences in the rate of phosphorylation by cellular kinases to their 5'-triphosphates, which is a prerequisite for inhibition of reverse transcriptase.

EXPERIMENTAL

Melting points were determined on a Buchi capillary apparatus and are uncorrected. Nuclear magnetic resonance spectra (^{1}H NMR, ^{13}C NMR) were recorded on a Bruker AM-300 spectrometer using tetramethylsilane as internal standard (^{1}H NMR). All exchangeable protons were confirmed by addition of D_2O. ^{13}C NMR spectra were determined using the J modulated spin echo technique where methyl and methine

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carbon resonances appear as positive peaks, and methylene and quaternary carbon resonances appear as negative peaks. Mass spectra (MS), using electron impact bombardment, were measured on a Hewlett-Packard 5995A spectrometer. Silica gel column chromatography was carried out using Merck 7734 silica gel (25μ particle size). Thin layer chromatography (TLC) was performed with Whatman MK6F silica gel microslides (25 μm thickness). 5'-*O*-Pivaloylthymidine (**3b**)[12], 5'-*O*-*t*-butyldimethylsilylthymidine (**3c**)[17] and 5'-*O*-*t*-butyldimethylsilyl-2'-deoxyuridine (**3d**)[17] were prepared according to the literature procedures.

5-Iodo-5'-*O*-pivaloyl-2'-deoxyuridine (3a).

Pivaloyl chloride (0.5 ml, 4.05 mmol) was added to a solution of 5-iodo-2'-deoxyuridine (**2a**, 1.0 g, 2.8 mmol) in dry pyridine (100 ml) at 0°C. The reaction mixture was stirred for 12 h at 25°C and poured onto ice-water (50 ml). Extraction with ethyl acetate (3 x 50 ml), washing the combined ethyl acetate extracts with water (2 x 50 ml), drying the organic fraction (Na_2SO_4) and removal of the solvent *in vacuo* afforded the impure product. Elution of the product from a silica gel column using MeOH:$CHCl_3$ (3:97, v/v) afforded **3a** (0.6 g, 49%); mp 70-75°C (sublimes). ^{1}H NMR ($CDCl_3$) δ: 1.30 (s, 9H, *t*-Bu), 2.12 (m, 1H, H-2'), 2.58 (m, 1H, H-2"), 4.23-4.50 (complex m, 5H, H-3', H-4', H-5', H-5", 3'-OH), 6.25 (t, $J_{1',2'}=J_{1',2"}=6$ Hz, 1H, H-1'), 7.90 (s, 1H, H-6), 8.96 (s, 1H, NH). MS m/z: 438 (M^+). *Anal.* Calcd for $C_{14}H_{19}IN_2O_6$: C, 38.36; H, 4.37; N, 6.39. Found: C, 38.33; H, 4.34; N, 6.20.

3'-*O*-Formyl-5-Iodo-5'-*O*-pivaloyl-2'-deoxyuridine (4a).

Phosphorous oxychloride (0.1 ml) was dissolved in dry DMF (10 ml) at 0°C, and 5-iodo-5'-*O*-pivaloyl-2'-deoxyuridine (**3a**, 0.1 g, 0.228 mmol) was added. The reaction mixture was stirred for 15 min at 0°C and then poured onto ice-water (50 ml). Extraction with ethyl acetate (2 x 50 ml), washing the extract with cold water (2 x 25 ml), drying the ethyl acetate fraction (Na_2SO_4) and removal of the solvent *in vacuo* afforded a residue which was recrystallized from ethyl acetate to yield **4a** (60 mg, 56%) as crystals; mp 155-158°C. ^{1}H NMR ($CDCl_3$) δ: 1.28 (s, 9H, *t*-Bu), 2.18 (m, 1H, H-2'), 2.65 (m, 1H, H-2"), 4.25-4.42 (m, 3H, H-4', H-5', H-5"), 5.38 (d, J=4.8 Hz, 1H, H-3'), 6.24 (d, $J_{1',2'}=8.4$ Hz of d, $J_{1',2"}=6.9$ Hz, 1H, H-1'), 7.88 (s, 1H, H-6), 8.08 (s, 1H, C*H*O), 9.18 (s, 1H, NH). ^{13}C NMR ($CDCl_3$) δ: 27.41 (*C*H_3), 38.26 (C-2'), 38.95 (Me_3-*C*-), 63.54 (C-5'), 68.93 (C-5), 73.66 (C-3'), 82.69 (C-1'), 85.62 (C-4'), 143.28 (C-6), 149.67 (C-2 *C*=O), 159.58 and 159.89 (C-4 *C*=O and t-Bu-*C*=O), 177.90 (-O-*C*HO). MS m/z: 466 (M^+). *Anal.* Calcd for $C_{15}H_{19}IN_2O_7$: C, 38.62; H, 4.07; N. 6.00. Found: C, 38.96; H, 4.17; N, 5.91.

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3'-*O*-Formyl-5'-*O*-pivaloylthymidine (4b).

Phosphorous oxychloride (0.25 ml) was dissolved in dry DMF (20 ml) at 0°C and 5'-O-pivaloylthymidine (**3b**, 0.25 g, 0.766 mmol) was added. The reaction was allowed to proceed for 20 min at 0°C, and the product was isolated and recrystallized as described for the preparation of **4a**, to yield **4b** as crystals (0.15 g, 55%); mp 158-160°C. ^{1}H NMR ($CDCl_3$) δ: 1.26 (s, 9H, *t*-Bu), 1.96 (s, 3H, C-5 Me), 2.20 (m, 1H, H-2'), 2.58 (m, 1H, H-2"), 4.28-4.48 (m, 3H, H-4', H-5', H-5"), 5.38 (m, 1H, H-3'), 6.32 (d, $J_{1',2'}$=8.4 Hz of d, $J_{1',2"}$=6.0 Hz, 1H, H-1'), 7.28 (s, 1H, H-6), 8.08 (s, 1H, O-C*H*O), 9.0 (s, 1H, NH). ^{13}C NMR ($CDCl_3$) δ: 12.45 (C-5 *C*H$_3$), 27.21 (*Me*$_3$C), 37.60 (C-2'), 38.86 (Me$_3$-*C*-CO), 63.54 (C-5'), 73.64 (C-3'), 82.09 (C-1'), 84.87 (C-4'), 111.56 (C-5), 134.30 (C-6), 150.18 (C-2 *C*=O), 159.95 (C-4 *C*=O), 163.45 (t-Bu-*C*O-), 177.86 (O-*C*HO). MS m/z: 354 (M^+). *Anal.* Calcd. for $C_{16}H_{22}N_2O_7$: C, 54.22; H, 6.26; N, 7.90. Found: C, 53.85; H, 6.32; N, 7.85.

3',5'-Dichloro-5-iodo-2',3',5'-trideoxyuridine (7).

A mixture of 5-iodo-2'-deoxyuridine (**2a**, 88 mg, 0.25 mmol), Et_4N^+ Cl^- (83 mg, 0.5 mmol), *N,N*-diethylaniline (0.32 ml) and phosphorous oxychloride (1.4 ml) in dry acetonitrile (5 ml) was refluxed for 10 min. The volatile components were removed *in vacuo*, and the oil-like residue remaining was dissolved in choroform prior to washing with cold water (2 x 10 ml). The chloroform extract was dried (Na_2SO_4), the solvent was removed *in vacuo* and the product obtained was purified by silica gel column chromatography using dichloromethane:methanol (199:1, v/v) as eluent. Removal of the solvent from the fractions containing the product yielded **7** (40 mg, 41%) which was recrystallized from ethyl acetate-hexanes; mp 160°C (decomp.). ^{1}H NMR ($CDCl_3$) δ: 2.43 (m, 1H, H-2'), 3.0 (m, 1H, H-2"), 3.80 (m, 2H, H-5', H-5"), 4.28 (m, 1H, H-4'), 4.60 (m, 1H, H-3'), 6.10 (d, $J_{1',2'}$=7.6 Hz of d, $J_{1',2"}$=2.9 Hz, 1H, H-1'), 8.02 (s, 1H, H-6), 8.93 (s, 1H, NH). ^{13}C NMR ($CDCl_3$) δ: 41.17 (C-5'), 42.88 (C-2'), 57.43 (C-3'), 67.84 (C-5), 83.29 (C-4'), 85.38 (C-1'), 144.30 (C-6), 149.78 (C-2 *C*=O), 159.74 (C-4 *C*=O). MS m/z: 391 (M^+, Cl^{35}), 395 (M^+, Cl^{37}). *Anal.* Calcd for $C_9H_9Cl_2IN_2O_3$: C, 27.64; H, 2.31; N, 7.16. Found: C, 27.42; H, 2.23; N, 7.08.

Reaction of 3'-*O*-formyl-5'-*O*-pivaloyl-2'-deoxyuridine (4a) and 3'-*O*-formyl-5'-*O*-pivaloylthymidine (4b) with tetra-*n*-butylammonium fluoride in tetrahydrofuran. Method A.

A solution of either **4a** or **4b** (0.1 mmol) was allowed to react with tetra-*n*-butylammonium fluoride (1 ml of a 1M solution) at 25°C for 90 min. The solvent was removed *in vacuo* and the residue obtained was purified by silica gel column

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chromatography using chloroform:methanol (95:5, v/v) as eluent to yield **3a** or **3b**, respectively.

Reaction of 3'-*O*-formyl-5-iodo-5'-*O*-pivaloyl-2'-deoxyuridine (4a) and 3'-*O*-formyl-5'-*O*-pivaloylthymidine(4b) with ammonia in methanol. Method B.

A solution of **4a** or **4b** (0.1 mmol) in a saturated solution of ammonia in methanol (5 ml) was stirred at 25°C for 20 min at which time TLC ($CHCl_3$:MeOH; 9:1, v/v) indicated the reaction was completed. The solvent was removed *in vacuo*, the residue obtained was suspended in MeOH to which a small amount of silica gel was added, and the solvent was removed. This material was placed on the top of a silica gel column which was eluted with chloroform:methanol (95:5, v/v) as eluent to yield **3a** or **3b**, respectively. The mp and ^1H NMR spectra for **3a** and **3b** were identical to those described previously.

5'-*O*-*t*-Butyldimethylsilyl-3'-*O*-formylthymidine (4c).

Phosphorous oxychloride (0.1 ml) was dissolved in dry DMF (20 ml) at a temperature below 5°C, and 5'-*O*-*t*-butyldimethylsilylthymidine (**3c**, 0.16 g, 0.45 mmol) was added. The reaction mixture was stirred at 0°C for 15 min and then cold water (50 ml) was added. This solution was extracted with ethyl acetate (3 x 50 ml), the ethyl acetate extract was washed with cold water (2 x 25 ml), the ethyl acetate extract was dried (Na_2SO_4), and the solvent was removed *in vacuo*. Recrystallization from diethyl ether-hexane afforded **4c** (120 mg, 69%) as a viscous oil. ^1H NMR ($CDCl_3$) δ: 0.16 (s, 6H, Si-Me_2), 0.95 (s, 9H, *t*-Bu), 1.98 (s, 3H, C-5 Me), 2.20 (m, 1H, H-2'), 2.50 (m, 1H, H-2"), 3.98 (m, 2H, H-5', H-5"), 4.18 (m, 1H, H-4'), 5.44 (m, 1H, H-3'), 6.42 (d $J_{1',2'}$=8.4 Hz of d, $J_{1',2"}$=4.8 Hz, 1H, H-1'), 7.58 (s, 1H, H-6), 8.11 (s, 1H, O-C*H*O), 8.16 (s, 1H, NH). *Anal.* Calcd for $C_{17}H_{28}N_2O_6Si$: C, 53.10; H, 7.33. Found: C, 53.27; H, 7.00.

3'-*O*-Formylthymidine (5). Method C.

5'-*O*-*t*-Butyldimethylsilylthymidine (**3c**, 0.3 g, 0.842 mmol) was allowed to react with phosphorous oxychloride (0.25 ml) in dry DMF (20 ml) using the procedure described for the synthesis of **4c**. The aqueous solution was extracted with ethyl acetate (3 x 50 ml), the ethyl acetate extract was dried (Na_2SO_4), the solvent was removed *in vacuo* and the oil-like residue obtained was heated at 50°C under reduced pressure (4 mm Hg) for 1 h. This product was then purified by silica gel column chromatography using chloroform:methanol (95:5, v/v) as eluent to yield **5** (120 mg, 53%) after recrystallization from EtOH; mp 153-155°C. ^1H NMR (Me_2SO-d_6) δ: 1.82 (s, 3H, Me), 2.32 (m, 2H, H-2', H-2"), 3.68 (m, 2H, H-5', H-5"), 4.05 (m, 1H, H-4'), 5.30 (m, 1H, 5'-OH), 5.38 (m, 1H, H-3'), 6.23 (d, $J_{1',2'}$=8.8 Hz of d, $J_{1',2"}$=5.4 Hz,

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1H, H-1'), 7.78 (s, 1H, H-6), 8.30 (s, 1H, OC*H*O), 11.42 (s, 1H, NH). ^{13}C NMR (CD_3OD) δ: 12.38 (*C*H_3), 38.40 (C-2'), 62.83 (C-5'), 75.71 (C-3'), 86.52 (C-4' or C-1'), 86.55 (C-4' or C-1'), 111.87 (C-5), 137.86 (C-6), 152.39 (C-2 *C*=O), 162.14 (C-4 *C*=O), 166.29 (O-*C*HO). *Anal.* Calcd. for $C_{11}H_{14}N_2O_6$: C, 48.88; H, 5.22; N, 10.36. Found: C, 48.57; H, 5.41; N, 9.98.

3'-*O*-Formylthymidine (5). Method D.

A solution of 5'-*O*-*t*-butyldimethylsilyl-3'-*O*-formylthymidine (**4c**, 0.1 g, 0.26 mmol) in 80% acetic acid (5 ml) was heated at 60°C for 1 h. Removal of the solvent *in vacuo* and purification of the product obtained by elution from a silica gel colunm using chloroform:methanol (95:5, v/v) as eluent gave **5** (36 mg, 51%) after recrystallization from EtOH. The melting point and ^{1}H NMR spectrum for **5** obtained in this reaction were identical to those described previously.

5'-*O*-*t*-Butyldimethylsilyl-3'-*O*-formyl-2'-deoxyuridine (4d) and 3'-*O*-formyl-2'-deoxyuridine (6).

Phosphorous oxychloride (0.25 ml) was dissolved in dry DMF (15 ml) at a temperature below 5°C prior to the addition of a solution of 5'-*O*-*t*-butyldimethylsilyl-2'-deoxyuridine (**3d**, 0.3 g, 0.877 mmol) in dry DMF (10 ml). The reaction mixture was stirred at 0°C for 25 min at which time cold water (25 ml) was added. Extraction with ethyl acetate (3 x 50 ml), drying (Na_2SO_4), and removal of the solvent *in vacuo* gave an oil-like residue which was heated at 50°C under reduced pressure (4 mm Hg) for 1 h. This material was purified by silica gel column chromatography using chloroform:methanol (98:2, v/v) as eluent to yield **4d** and **6**. Compound **4d:** (80 mg, 27%, syrup); ^{1}H NMR ($CDCl_3$) δ: 0.16 (s, 6H, Si-*Me*$_2$), 0.94 (s, 9H, *t*-Bu), 2.22 (m, 1H, H-2'), 2.55 (m, 1H, H-2"), 3.95 (m, 2H, H-5', H-5"), 4.20 (m, 1H, H-4'), 5.42 (m, 1H, H-3'), 5.75 (d, $J_{5,6}$=7.2 Hz, 1H, H-5), 6.42 (d, $J_{1',2'}$=8.7 Hz of d, $J_{1',2''}$=6.0 Hz, 1H, H-1'), 7.92 (d, $J_{5,6}$=7.2 Hz, 1H, H-6), 8.08 (s, 1H, O-C*H*O), 9.75 (s, 1H, NH). ^{13}C NMR ($CDCl_3$) δ: -5.61 and -5.66 (Si-*Me*$_2$), 18.22 (Me$_3$-*C*-), 25.78 (*Me*$_3$-C), 38.35 (C-2'), 63.39 (C-5'), 74.78 (C-3'), 85.07 (C-4' or C-1'), 85.28 (C-4' or C-1'), 102.66 (C-5), 139.61 (C-6), 150.38 (C-2 *C*=O), 160.15 (C-4 *C*=O), 163.28 (O-*C*HO). This product **4d** was used immediately in the subsequent reaction with 80% acetic acid to remove the 5'-*O*-TBDMS protecting group (Method D).

Compound **6:** (60 mg, 27% yield); mp 150-151°C. ^{1}H NMR (Me_2SO-d_6) δ: 2.30 (m, 2H, H-2', H-2"), 3.64 (m, 2H, H-5', H-5"), 4.02 (m, 1H, H-4'), 5.24 (m, 1H, 5'-OH), 5.32 (m, 1H, H-3'), 5.68 (d, $J_{5,6}$=8 Hz, 1H, H-5), 6.16 (t, $J_{1',2'}$=$J_{1',2''}$=7.8 Hz, 1H, H-1'), 7.88 (d, $J_{5,6}$=8 Hz, 1H, H-6), 8.22 (s, 1H, O-C*H*O), 11.36 (s, 1H, NH). ^{13}C NMR (CD_3OD) δ: 38.63 (C-2'), 62.81 (C-5'), 75.76

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(C-3'), 86.57 (C-1' or C-4'), 86.74 (C-1' or C-4'), 103.00 (C-5), 142.25 (C-6), 150.22 (C-2 *C*=O), 162.15 (C-4 *C*=O), 166.08 (O-*C*HO). *Anal.* Calcd. for $C_{10}H_{12}N_2O_6$: C, 46.87; H, 4.72; N, 10.93. Found: C, 46.71; H, 4.79; N, 10.52.

3'-*O*-Formyl-2'-deoxyuridine (6). Method D.

A solution of 5'-*O*-*t*-butyldimethylsilyl-3'-*O*-formyl-2'-deoxyuridine (**4d**, 60 mg, 0.175 mmol) in 80% acetic acid (3 ml) was heated at 60°C for 1 h. The solvent was removed *in vacuo* and the product was purified by silica gel column chromatography using methanol:chloroform (98:2, v/v) as eluent to yield **6** (26 mg, 57%). The melting point and ^{1}H NMR spectrum for **6** were identical to those described previously.

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