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The triphosphate of β-D-4′-*C*-ethynyl-2′,3′-dideoxycytidine is the preferred enantiomer substrate for HIV reverse transcriptase

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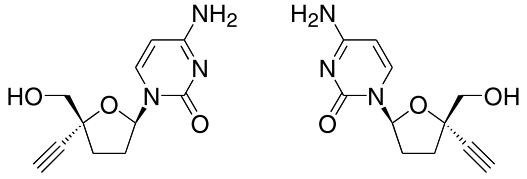
Abstract—The enantioselective synthesis of the β-D (**1**) enantiomer of 4′-*C*-ethynyl-2′,3′-dideoxycytidine confirms an earlier stereochemical assignment that was strictly based on the ability of HIV reverse transcriptase and its M184V mutant to discriminate between the D- and L-configuration of nucleoside 5′-triphosphates.

1. Introduction

The β-D (**1**) and β-L (**2**) enantiomers of 4′-*C*-ethynyl-2′,3′-dideoxycytidine were previously isolated by a lipase-catalyzed trans-esterification resolution performed on the N^4-benzoyl protected nucleoside racemate.[1] Each pure enantiomer was obtained with 98% and 91% ee, respectively.



1 **2**

The assignment of the absolute stereochemistry of **1** and **2** was based on the ability of HIV reverse transcriptase (RT) to discriminate between the corresponding 5′-triphosphates, which was further confirmed by assaying against an RT mutant (M184V) with a marked preference for incorporating nucleosides in the D-configuration.[1] Although the conclusions from these studies seemed well-substantiated, we wished to provide absolute structural proof for the β-D (**1**) enantiomer by chemical synthesis. Taking advantage of a recently developed asymmetric synthesis of γ-lactone **3a**,[2] which was utilized for the synthesis of chiral (*R*)-diacylglycerol lactones, we developed an asymmetric approach for the synthesis of β-D (**1**) to confirm the earlier assignment based strictly on the biological data.

2. Results and discussion

The trityl group on the available chiral γ-lactone (**3a**) was not ideal to keep for an extended synthesis because of its ease of cleavage under acidic conditions, including during silica gel chromatography. Therefore, the more acid robust *t*-butyldiphenylsilyl ether was put in place leading to compound **3c** after two steps in 86% overall yield (Scheme 1). Reduction of the lactone with diisobutylaluminum hydride (DIBAL-H) proceeded in 91% yield and acetylation of the resulting lactol (**4a**) with acetic anhydride in pyridine led to the corresponding acetate **4b** in 96% yield. During catalytic transfer hydrogenation to remove the benzyl group in **4b**, the OAc group was exchanged to form the methyl glycoside **4c**, and although such transformation was not intended, the outcome was inconsequential for the ensuing elaboration of the nucleoside (Scheme 2).

The free OH group in **4c** was protected as an acetate (**4d**, Scheme 2) with the hope that, at least to some degree, distant neighboring group participation by the acetate would form an intermediate cation that would favor attack from the β-face (Fig. 1). Thus, the methyl glycoside

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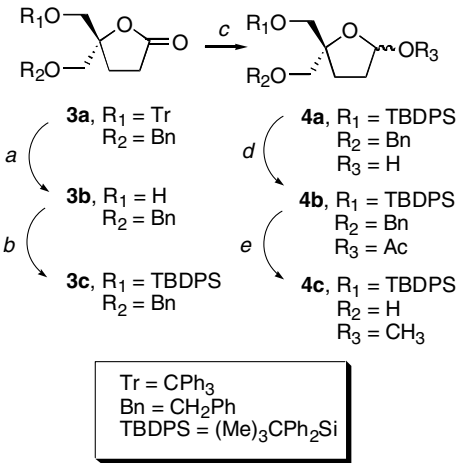


Scheme 1. Reagents and conditions: (a) TFA, CH_2Cl_2; (b) TBDPS-Cl, imidazole, CH_2Cl_2; (c) DIBAL-H, toluene, −78 °C; (d) Ac_2O, pyridine; (e) HCO_2H/MeOH, Pd-black.

4d reacted under standard Vorbrüggen conditions with persilylated N^4-benzoylcytosine and trimethylsilyl triflate (TMSOTf) as a Lewis acid catalyst to give the expected nucleoside products with a β-**5**/α-**6** ratio of 1.14 (Scheme 2). Although we do not have any direct evidence to support the distant participation of the acetate, use of this strategy improved the β/α anomer ratio significantly relative to the 0.51 ratio obtained in the absence of any participating group (Scheme 2).[1] This type of distant neighboring anchimeric assistance has been observed in the synthesis of nucleosides.[3,4]

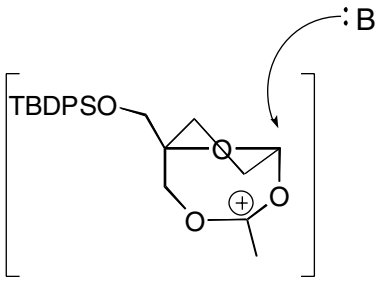
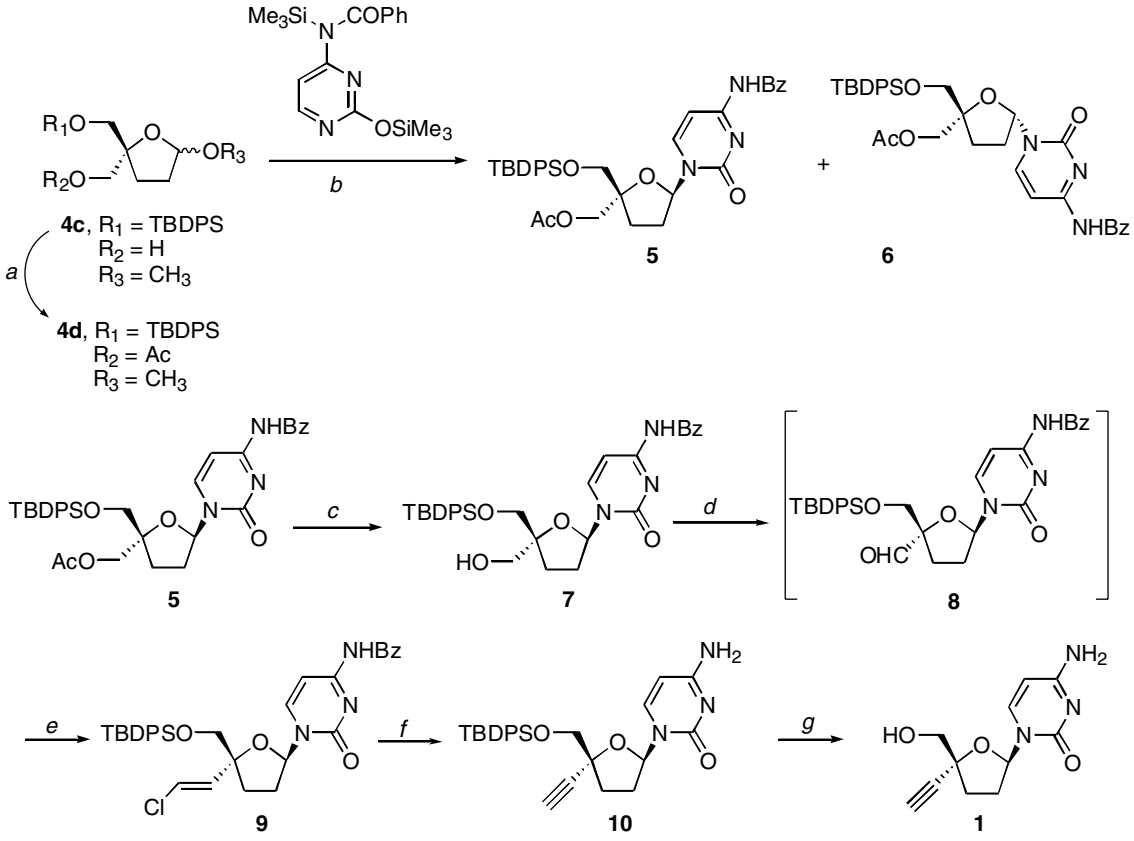


Figure 1. Proposed sugar carbocation intermediate formed through anchimeric assistance of the distant acetate.

Separation of both β- and α-anomers was accomplished by flash column chromatography and a complete identification of the stereochemistry was accomplished by examining the coupling pattern of the anomeric protons. Typically, for the β-anomer the anomeric proton appears as a pseudotriplet.[5] This assignment for the β-anomer had been confirmed previously by X-ray crystallography during the synthesis of (±)-**1**.[1] The remaining sequence of steps involved: (1) hydrolysis of the acetate to give compound **7** and (2) oxidation of the free alcohol to form a formyl



Scheme 2. Reagents and conditions: (a) Ac_2O, pyridine; (b) TMSOTf, CH_2ClCH_2Cl; (c) guanidine, EtOH/CH_2Cl_2 (9:1); (d) DCC, DMSO, Cl_2CHCO_2H; (e) $ClCH_2P(C_6H_5)_3Cl$, *n*-BuLi, THF, −78 °C; (f) *n*-BuLi, THF, −78 °C; (g) $NH_4F{\cdot}0.5H_2O$, MeOH, 80 °C.

group. Without isolation, Wittig olefination was performed on intermediate **8** with (chloromethyl)triphenylphosphonium chloride to produce the chlorovinyl intermediate **9**. Because of some persistent contamination with DCC from the oxidation step, even after flash column chromatography, the following conversion was performed without further purification. The chlorovinyl group was converted to the ethynyl group after treatment with *n*-butyllithium in tetrahydrofuran to give the penultimate intermediate **10**, obtained in 56% overall yield from **7**. Final fluoride deprotection of the silyl ether afforded the desired, enantiomer (+)-β-**1**, which was identical in all respects to the sample that was resolved chemico-enzymatically.[1]

3. Conclusions

In conclusion, we have described an enantioselective method for the synthesis of the β-D (**1**) enantiomer of 4′-*C*-ethynyl-2′,3′-dideoxycytidine starting from a chiral γ-lactone (**3a**). The synthesis of **1** verifies our earlier assignment, which was based on the ability of HIV RT and its M184V mutant to discriminate between the D- and L-configuration of nucleoside triphosphates.[6,7] Our results provide further support for the use of RT as an effective enzyme to help assign the absolute configuration of nucleoside 5′-triphosphates.

4. Experimental

4.1. General

All chemical reagents were commercially available. Flash column chromatography was performed on silica gel 60, 230–240 mesh (E. Merck), and analytical TLC was performed on Analtech Uniplates silica-gel GF. Routine ^{1}H NMR spectra were recorded at 400 MHz on a Varian INOVA instrument. Chemical shifts are reported in δ (ppm) relative to internal tetramethylsilane. Optical rotations were obtained using JASCO model P-1010 polarimeter. Positive-ion fast-bombardment mass spectra (FABMS) were recorded on a VG 7070E mass spectrometer at an accelerating voltage of 6 kV and a resolution of 2000. Glycerol was used as the sample matrix and ionization was effected by a beam of xenon atoms. Elemental analyses were performed by Atlantic Microlab Inc., Norcross, GA.

4.1.1. (*R*)-5-(hydroxymethyl)-5-[(phenylmethoxy)methyl]-3,4,5-trihydrofuran-2-one (3b). A solution of **3a** (5.26 g, 10.90 mmol) in anhydrous CH_2Cl_2 (50 mL) was treated with an equal volume of 50% trifluoroacetic acid (TFA) in CH_2Cl_2 and stirred at room temperature for 1 h. After the addition of CH_2Cl_2 (100 mL), the solution was washed with water (2× 100 mL), dried ($MgSO_4$), and concentrated under vacuum. The crude product was purified by flash column chromatography using a step gradient of hexanes/EtOAc (from 4:1 to 1:2) to give 2.31 g (88.9%) of **3b** as a foam; $[\alpha]_D^{23}$ +10.08 (*c* 0.74, $CHCl_3$); ^{1}H NMR ($CDCl_3$)δ 7.20–7.35 (m, 5H, Ph), 4.50 (s, 2H, C*H*$_2$Ph), 3.70 (d, 1H, *J* = 12.1 Hz, C*H*HOBn), 3.58 (d, 1H, *J* = 12.1 Hz, CH*H*OBn), 3.58 (d, 1H, *J* = 10.5 Hz, C*H*HOH), 3.50 (d, 1H, *J* = 10.5 Hz, CH*H*OH), 2.48–2.67 (m, 2H, H-3$_{a,b}$), 2.05 (m, 2H, H-4$_{a,b}$); FAB MS (*m*/*z*, relative intensity) 237.2 (MH^+, 22). Anal. Calcd for $C_{13}H_{16}O_4$: C, 66.09; H, 6.83. Found: C, 66.23; H, 6.85.

4.1.2. (*R*)-5-[(2,2-dimethyl-1,1-diphenyl-1-silapropoxy)methyl]-5-[(phenylmethoxy)-methyl]-3,4,5-trihydrofuran-2-one (3c). A solution of **3b** (2.30 g, 9.73 mmol) in anhydrous CH_2Cl_2 was treated with imidazole (4.22 g, 61.98 mmol) and *tert*-butyldiphenylsilyl chloride (TBDPS-Cl, 5.30 mL, 20.63 mmol). After stirring at room temperature for 1.5 h, the solvent was removed under vacuum and the residue was taken up in EtOAc (200 mL). The organic solution was washed with water (2× 100 mL), dried ($MgSO_4$), and concentrated under vacuum. The crude product was purified by flash column chromatography using a step gradient of hexanes:EtOAc (from 10:1 to 4:1) to give 4.51 g (97.6%) of **3c** as an oil; $[\alpha]_D^{23}$ +9.28 (*c* 0.79, $CHCl_3$); ^{1}H NMR ($CDCl_3$)δ 7.19–7.60 (m, 15H, Ph), 4.46 (AB q, 2H, *J* = 11.9 Hz, C*H*$_2$Ph), 3.69 (d, 1 H, *J* = 10.9 Hz, C*H*HOBn), 3.61 (d, 1H, *J* = 10.9 Hz, CH*H*OBn), 3.50 (AB q, 2 H, *J* = 10.3 Hz, CH_2OSi), 2.55 (m, 2H, H-3$_{a,b}$), 2.11 (irregular t, 2H, H-4$_{a,b}$), 1.00 (s, 9H, $SiC(CH_3)_3$); FAB MS (*m*/*z*, relative intensity) 475.4 (MH^+, 20). Anal. Calcd for $C_{29}H_{34}O_4Si$: C, 73.38; H, 7.22. Found: C, 73.38; H, 7.19.

4.1.3. 5-[(2,2-Dimethyl-1,1-diphenyl-1-silapropoxy)methyl]-5-[(phenylmethoxy)methyl]-oxolan-2-ol (4a). A solution of **3c** (3.02 g, 6.36 mmol) in dry toluene (50 mL) was cooled to −78 °C and treated slowly with a solution of diisobutylaluminum hydride (DIBAL, 13.4 mL, 1 M in hexane). After 1 h stirring the reaction was quenched with methanol (15 mL) and the resulting mixture was warmed to room temperature. The solvent was removed under vacuum and the residue was dissolved in EtOAc (200 mL) and extracted with water (50 mL). The organic layer was separated, dried ($MgSO_4$), and concentrated. The crude product was purified by flash column chromatography using a step gradient of hexanes:EtOAc (from 10:1 to 4:1) to give 2.765 g (91.2%) of **4a** as an oily mixture of anomers; ^{1}H NMR ($CDCl_3$)δ 7.20–7.65 (m, 15H, Ph), 5.35 and 5.43 (m, 1H, C*H*OH), 4.50 (m, 2H, C*H*$_2$Ph), 3.32–3.57 (m, 4H, C*H*$_2$OBn and C*H*$_2$OSi), 1.50 (br s, 1 H, OH), 1.63–2.13 (m, 4H, H-3$_{a,b}$ and H-4$_{a,b}$), 1.00–1.06 (s, 9H, $SiC(CH_3)_3$); FAB MS (*m*/*z*, relative intensity) 469.2 (MH^+–H_2O, 10). Anal. Calcd for $C_{29}H_{36}O_4Si$: C, 73.07; H, 7.61. Found: C, 73.31; H, 7.67.

4.1.4. 5-[(2,2-Dimethyl-1,1-diphenyl-1-silapropoxy)methyl]-5-[(phenylmethoxy)methyl]-oxolan-2-yl acetate (4b). A solution of **4a** (2.76 g, 5.79 mmol) in anhydrous pyridine (50 mL) was treated with acetic anhydride (4 mL, 42.31 mmol) and stirred at room temperature for 24 h. The solvent was removed under vacuum and the crude product was purified by flash column chromatography using EtOAc:hexanes:Et_3N (40:10:1) to give 2.94 g (98%) of **4b** as an oily mixture of anomers; ^{1}H NMR ($CDCl_3$)δ 7.15–7.65 (m, 15H, Ph), 6.21 and 6.23 (doublet, 1 H, *J* = 4.68 and 4.69 Hz, C*H*OAc),

4.40–4.60 (m, 2 H, CH_2Ph), 3.400–3.70 (m, 4H, CH_2OBn and CH_2OSi), 1.80–2.20 (m, 4H, H-3$_{a,b}$ and H-4$_{a,b}$), 2.00 and 1.92 (s, 3H, OCOCH_3), 1.00–1.06 (s, 9H, SiC(CH_3)$_3$); FAB MS (*m*/*z*, relative intensity) 459.2 (MH^+–CH_3CO_2H, 12) and FAB MS (*m*/*z*, relative intensity) 557.2 ($M+K^+$, 30). Anal. Calcd for $C_{31}H_{38}O_5Si$: C, 71.78; H, 7.38. Found: C, 71.86; H, 7.37.

4.1.5. {2-[(2,2-Dimethyl-1,1-diphenyl-1-silapropoxy)methyl]-5-methoxyoxolan-2-yl}methan-1-ol (4c). A solution of **4b** (2.94 g, 5.66 mmol) in 4.4% HCO_2H in methanol (100 mL) maintained under a blanket of argon was treated with palladium black (0.75 g) and stirred. After 1 h, an equal amount of catalyst was added and stirred for 1.5 h. The mixture was filtered through a pad of Celite® and the pad was washed with MeOH (50 mL). The filtrate was evaporated to dryness under vacuum and the residue was purified by flash column chromatography using a step gradient of hexanes/EtOAc (8:1 to 2:1) to give 0.809 g (92.9%) of **4c** as a thick oil containing a mixture of anomers; ^{1}H NMR ($CDCl_3$)δ 7.25–7.65 (m, 10H, Ph), 4.84 and 4.97 (m, 1H, J = 4.68 and 4.69 Hz, CHOCH$_3$), 3.40–3.74 (m, 4H, CH_2OH and CH_2OSi), 3.14 and 3.30 (s, 3H, OCH_3), 1.75–2.00 (m, 4H, H-3$_{a,b}$ and H-4$_{a,b}$), 1.00–1.06 (s, 9H, SiC(CH_3)$_3$); FAB MS (*m*/*z*, relative intensity) 369.1 (MH^+–CH_3OH, 40) and FAB MS (*m*/*z*, relative intensity) 439.1 ($M+K^+$, 55). Anal. Calcd for $C_{23}H_{32}O_4Si{\cdot}0.25H_2O$: C, 68.19; H, 8.09. Found: C, 68.08; H, 8.10.

4.1.6. {2-[(2,2-Dimethyl-1,1-diphenyl-1-silapropoxy) methyl]-5-methoxyoxolan-2-yl}methyl acetate (4d). A solution of **4c** (0.785 g, 1.95 mmol) in anhydrous pyridine (25 mL) was treated with acetic anhydride (0.58 mL, 6.13 mmol) and stirred at room temperature for 24 h. The solvent was removed under vacuum and the crude product was purified by flash column chromatography using a step gradient of hexanes/EtOAc (15:1 to 8:1) to give 0.842 g (97.1%) of **4d** as a thick oil containing a 1:1 mixture of anomers; ^{1}H NMR ($CDCl_3$)δ 7.30–7.65 (m, 10H, Ph), 4.98 (br d, 0.5H, J = 3.5 Hz, CHOCH$_3$), 4.92 (m, 0.5H, CHOCH$_3$), 4.26 (d, 0.5H, J = 11.1 Hz, CHHOAc), 4.15 (q, 1H, J = 11.1 Hz, CH_2OAc), 4.85 (d, 0.5 H, J = 11.1 Hz, CHHOAc), 3.73 (d, 0.5H, J = 9.9 Hz, CHHOSi), 3.61 (q, 1H, J = 9.9 Hz, CH$_2$OSi), 3.42 (d, 0.5H, J = 9.9 Hz, CHHOSi), 3.25 and 3.18 (s, 3H, OCH_3), 2.00 and 2.01 (s, 3H OCOCH_3), 1.76–2.05 (m, 4H, H-2$_{a,b}$ and H-3$_{a,b}$), 1.00–1.06 (s, 9H, SiC(CH_3)$_3$); FAB MS (*m*/*z*, relative intensity) 411.2 (MH^+–CH_3OH, 40) and FAB MS (*m*/*z*, relative intensity) 481.3 ($M+K^+$, 100). Anal. Calcd for $C_{25}H_{34}O_5Si$: C, 67.84; H, 7.74. Found: C, 67.87; H, 7.84.

4.1.7. (2*R*,5*R*)-{2-[(2,2-dimethyl-1,1-diphenyl-1- silapropoxy)methyl]-5-[2-oxo-4-(phenylcarbonylamino)hydropyrimidinyl]oxolan-2-yl}methyl acetate (5). A suspension of N^4-benzoylcytosine (0.70 g, 3.25 mmol) in anhydrous CH_3CN (10 mL) was treated with bis(trimethylsilyl)trifluoroacetamide (BSTFA, 10 mL). After 30 min the reaction became homogeneous, the solvent was removed under vacuum, and the residual oil dissolved in anhydrous 1,2-dichloroethane (30 mL). This solution was cooled to 0 °C and treated with a solution of **4d** (0.703 g, 1.58 mmol) in anhydrous 1,2-dichloroethane (30 mL). Trimethylsilyl triflate (0.55 mL, 3.01 mmol) was added, the ice bath was removed, and the solution was stirred for 2.5 h. The reaction was quenched by the addition of an aqueous saturated solution of $NaHCO_3$ (10 mL) and after adding CH_2Cl_2 (50 mL) the product was extracted into the organic phase. The organic solution was dried ($MgSO_4$) and evaporated to dryness under vacuum. The crude product obtained was purified by flash column chromatography using a step gradient of hexanes/EtOAc (2:1 to 1:4) to give the less polar α-isomer **6** (0.464 g, 46.4%) followed by the desired, more polar β-isomer **5** (0.525 g, 53%).

Compound **5**: $[\alpha]_D^{23}$ +35.93 (*c* 0.54, $CHCl_3$); ^{1}H NMR ($CDCl_3$)δ 8.12 (d, 1H, J = 7.8 Hz, H-6), 7.30–7.90 (m, 16H, H-5, Ph), 7.25 (br s, 1H, NH), 6.10 (irregular t, 1H, $J \approx$ 5.3 Hz, H-1′), 4.04 (AB s, 2H, CH_2OAc), 3.73 (AB q, 2H, J = 10.2 H, CH_2OSi), 2.68 (m, 1H, H – 3′$_a$), 1.80–2.10 (m, 3H, H – 3′$_b$, H – 2′$_{a,b}$), 1.98 (s, 3H, OCOCH_3), 1.00 (s, 9H, SiC(CH_3)$_3$); FAB MS (*m*/*z*, relative intensity) 626.4 (MH^+, 12). Anal. Calcd for $C_{35}H_{39}N_3O_6Si{\cdot}0.25H_2O$: C, 66.69; H, 6.32; N, 6.67. Found: C, 66.71; H, 6.41; N, 6.52.

Compound **6**: ^{1}H NMR ($CDCl_3$)δ 8.70 (br s, 1H, NH), 8.20 (d, 1H, J = 7.4 Hz, H-6), 7.30–7.90 (m, 16H, H-5, Ph), 5.92 (irregular dd, 1H, $J \approx$ 6.2, 4.0 Hz, H-1′), 4.44 (d, 1H, J = 12.1 Hz, CHHOAc), 4.22 (d, 1H, J = 12.1 Hz, CHHOAc), 3.54 (AB q, 2H, J = 10.3 Hz, CH_2OSi), 2.55 (m, 1H, H – 3′$_a$), 2.00 (m, 2H, H – 2′$_{a,b}$), 1.80 (m, 1H, H – 3′$_b$), 2.04 (s, 3H, OCOCH$_3$), 1.00 (s, 9H, SiC(CH_3)$_3$); FAB MS (*m*/*z*, relative intensity) 626.4 (MH^+, 9).

4.1.8. (2*R*,5*R*) – *N*-(1-{5-[(2,2-dimethyl-1,1-diphenyl-1-silapropoxy)methyl]-5-(hydroxymethyl)oxolan-2-yl}-2-oxohydropyrimidin-4-yl)benzamide (7). A solution of nucleoside **5** (0.707 g, 1.11 mmol) in anhydrous CH_2Cl_2 (3 mL) at 0 °C was treated with a solution of guanidine in ethanol (27 mL) and allowed to reach room temperature after 30 min [The solution of guanidine was prepared from guanidine hydrochloride (0.70 g, 7.32 mmol) dissolved in ethanol (7 mL) and treated with sodium ethoxide (3.5 mL of 21 wt% in ethanol, 10.7 mmol) at room temperature for 20 min. The precipitate was removed and the filtrate containing free guanidine was used immediately]. After the 30 min treatment with guanidine, the solvent was removed under vacuum and the crude residue was purified by flash column chromatography using a step gradient of hexanes/EtOAc (1:1 to 100% EtOAc) followed by CH_2Cl_2:MeOH (20:1) to give 0.577 g (87.5%) of **7** as a white foam; $[\alpha]_D^{23}$ +45.00 (*c* 0.86, $CHCl_3$); ^{1}H NMR ($CDCl_3$)δ 8.13 (d, 1H, J = 7.4 Hz, H-6), 7.30–7.90 (m, 16H, H-5, Ph), 7.25 (br s, 1H, NH), 6.13 (irregular t, 1H, $J \approx$ 5.5 Hz, H-1′), 3.76 (AB q, 2H, J = 10.9 Hz, CH_2OSi), 3.53 (AB s, 2H, CH_2OH), 2.66 (m, 1H, H – 4′$_a$), 1.86–2.40 (m, 3H, H – 4′$_b$, H – 3′$_{a,b}$), 1.00 (s, 9H, SiC(CH_3)$_3$); FAB MS (*m*/*z*, relative intensity) 584.1 (MH^+, 55). Anal. Calcd for $C_{33}H_{37}N_3O_5Si{\cdot}H_2O$:

C, 65.86; H, 6.53; N, 6.98. Found: C, 65.98; H, 6.39; N, 6.75.

4.1.9. (2*R*,5*R*)-4-amino-1-{5-[(2,2,-dimethyl-1,1- diphenyl-1-silapropoxy)methyl]-5-ethynyloxolan-2-yl}-1-hydropyrimidin-2- one (10). A stirred solution of **7** (1.39 g, 2.38 mmol) and dicyclohexylcarbodiimide (DCC, 1.48 g, 7.17 mmol) in anhydrous DMSO (50 mL) was cooled to 0 °C and treated with dichloroacetic acid (0.10 mL, 1.21 mmol). The ice bath was removed and stirring continued for 2 h. The suspension was filtered and the solid washed with EtOAc (50 mL). The filtrate was extracted with water (2× 50 mL) and the organic phase was dried ($MgSO_4$) and concentrated under vacuum. The crude product was purified by flash column chromatography using a step gradient of hexanes/EtOAc (1:1 to 100% EtOAc) to give 1.30 g (94.2%) of **8** as a foam. Because this product showed some contamination with DCC it was used directly in the next step.

A suspension of (chloromethyl)trimethylphosphonium chloride (2.10 g, 6.04 mmol) in dry THF (20 mL) was cooled to −78 °C and treated with *n*-BuLi (1.6 M in hexanes, 3.8 mL). After stirring for 1 h, a solution of aldehyde **8** in THF (50 mL) was added. The dry ice/acetone cooling bath was replaced with a wet-ice bath (0 °C) and stirred for 3 h. The reaction was quenched with a saturated aqueous solution of NH_4Cl (5 mL) and the resulting mixture was extracted with EtOAc (2× 50 mL). The combined organic extract was washed with water (50 mL), dried ($MgSO_4$), and concentrated under vacuum. The crude product was purified by flash column chromatography using a step gradient of EtOAc/hexanes (1:1 to 4:1) to give 1.85 g of **9** still contaminated with DCC. This material was also used directly in the following step.

A solution of **9** in dry THF (20 mL) at −78 °C was kept under a blanket of argon and treated with *n*-BuLi (1.6 M in hexanes, 30 mL). The solution was stirred under argon for 2 h and the reaction was quenched with a saturated aqueous solution of NH_4Cl (20 mL). The solution was then allowed to reach room temperature and extracted with EtOAc (2× 100 mL). The combined organic extract was washed with water (75 mL), dried ($MgSO_4$), and concentrated under vacuum. The crude product was purified by flash column chromatography using a step gradient of EtOAc/hexanes (2:1 to 100% EtOAc) and CH_2Cl_2/MeOH (20:1 to 10:1) to give 0.595 g of **10** in 52.6% after three steps; $[\alpha]_D^{23}$ +31.58 (*c* 0.52, $CHCl_3$); ^1H NMR ($CDCl_3$)δ 7.95 (d, 1H, J = 7.4 Hz, H-6), 7.30–7.62 (m, 10H, Ph), 6.20 (irregular dd, 1H, $J \approx 6.9$, 2.9 Hz, H-1′), 5.20 (d, 1H, J = 7.4 Hz, H-5), 4.30 (d, 1H, J = 11.3 Hz, C*H*HOSi), 3.75 (d, 1H, J = 11.3 Hz, CH*H*OSi), 2.72 (m, 1H, H − $4'_a$), 2.30–2.40 (m, 1H, H − $4'_b$), 2.38 (s, 1H, CC*H*), 1.90–2.60 (m, 2H, H − $3'_{a,b}$), 1.00 (s, 9H, $SiC(CH_3)_3$); FAB MS (*m*/*z*, relative intensity) 474.1 (MH^+, 14). HRMS (FAB) calcd for $C_{27}H_{31}N_3O_3Si$ (MH^+, *m*/*z*) 474.2213. Found 474.2232.

4.1.10. (2*R*,5*R*)-(4-amino-1-[5-ethynyl-5-(hydroxymethyl)oxolan-2-yl]-1-hydropyrimidin-2-one [(+)-1-(2′,3′-dideoxy-4-*C*-ethynyl-ribo-pentofuranosyl)cytosine](+)-1. Ammonium fluoride (2.39 g, 51.9 mmol) was added to a stirred solution of **10** (0.824 g, 1.73 mmol) in methanol (60 mL) and heated to reflux for 1.5 h. The solution was allowed to reach room temperature, concentrated under vacuum to a reduced volume, and directly loaded onto a silica-gel column to be flashed chromatographed using a step gradient of CH_2Cl_2/MeOH (20:1 to 2:1). The collected fractions containing the product were combined and rechromatographed using a reverse-phase C-18 column eluted with water, followed by a step gradient of H_2O/MeOH (10:1 to 5:1). The desired fractions were collected, combined, and lyophilized to give 0.339 g (82.9%) of **1** as a white foam, identical in all respects to the previously made chiral material obtained by the lipase-catalyzed resolution; $[\alpha]_D^{23}$ +45.84 (*c* 0.59, H_2O).

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