Published in final edited form as:
J Org Chem. 2013 January 18; 78(2): 723–727. doi:10.1021/jo302038d.

Synthesis of Cyclopentanyl Carbocyclic 5-Fluorocytosine ((−)-5-Fluoro-Carbodine) Using a Facially Selective Hydrogenation Approach

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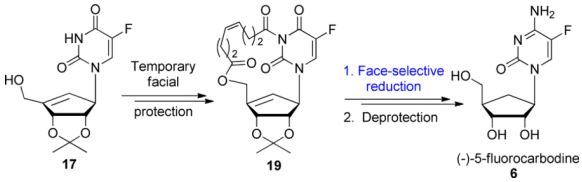
Abstract



An efficient synthetic route to biologically relevant (−)-5-fluorocarbodine **6** was developed. Direct coupling of N^6 protected 5-fluorouracil **15** with cyclopentenyl intermediate **13**, followed by formation of a macrocycle between the base and the carbocyclic sugar moiety, *via* ring closing metathesis, allowed for a facial selective hydrogenation of the sugar double bond to give, exclusively, the desired 4'-β stereoisomer.

Carbocyclic nucleosides, in which the furanose ring oxygen atom has been replaced by a carbon atom, are, in general, more chemically and enzymatically stable and some of them have demonstrated interesting biological properties.[1] Thus, naturally occurring (−)-aristeromycin (**1**)[2] and (−)-neplanocin A (**2**)[3] manifest cytotoxic and antiviral properties while the drugs abacavir (**3**)[4] and entecavir (**4**)[5] are Food and Drug Administration (FDA) approved for the treatment of human immunodeficiency virus (HIV) and hepatitis B virus (HBV), respectively (Figure 1.).

The carbocyclic cytosine analogs (−)-carbodine (**5**) and (−)-5-fluorocarbodine (**6**) was reported to possess significant antitumor and antiviral activities. For instance, compound (**6**) showed selective submicromolar IC_{50}'s against a human T cell lymphoblast cell line and human B cell lymphoma cell line but also potent inhibition of HCV RNA replication (ΔCt of 6 at 10 μM in a Huh-7 cell based subgenomic replicon assay).[6] In addition, compounds (**5**) and (**6**) demonstrated selective activity against Venezuela Equine Encephalitis Virus (VEEV)[7] and various strains of H5N1 influenza.[8]

The most recently reported syntheses of (−)-carbodine (**5**) and (−)-5-fluorocarbodine (**6**) used a linear approach (Scheme 1) from known cyclopentenyl intermediate including

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Supporting Information
Copies of NMR spectra. This material is available free of charge *via* the Internet at http://pubs.acs.org.

stepwise construction of the heterocyclic base. The key intermediate **7** was prepared from D-ribose in 13 steps by a sequence that used protecting/deprotection strategies and each step required purification by column chromatography on silica gel. After building the pyrimidine base, intermediate **8** was either converted, after a series of protection/deprotection, to its cytosine counterpart then fluorinated (ref 8), or directly fluorinated and then converted to its cytosine analog (ref 6). Overall, both approaches are lengthy (18 and 19 steps) and resulted in the formation of (**6**) in an overall yield of 2[6] and 10%.[8]

Preparation of (−)-carbodine (**5**) has also been achieved by either enzymatic hydrolysis[9] or selective hydrogenation of cyclopentenyl cytosine.[10] However, both of these methods resulted in poor overall yields and low selectivities. In the later approach, the authors demonstrated that the use of diimide as a source of hydrogen was not completely diastereoselective and led to a mixture of **5**, along with isocarbodine (**10**) (Scheme 2). Furthermore, separation of the two isomers appeared problematic and purification of enantiomerically pure **5** could only be achieved using semi-preparative HPLC. With this last result in mind, we hypothesized that the introduction of a temporary sugar-base bridging group on compound **A** (Scheme 2; X = OH) would act as an umbrella, covering the β face of the cyclopentene and allow for selective α face hydrogenation. Furthermore, we envisioned a sequence to form a cyclic eight member linker using a ring-closing metathesis (RCM)[11] reaction that would connect the 5'-O of the sugar and 3-N of the uridine base as depicted in Scheme 2 compound **B**. Facial selectivity of the subsequent hydrogenation could also be rationalized by the calculated[12] increased ring strain of the macrocycle for beta face hydrogenation that would ultimately lead to isocarbodine (**10**) formation.

In order to prepare 5-fluorocarbodine (**6**), the key cyclopentenol derivative **13** was synthesized in 9 steps from D-ribose, using a slightly modified known methodology (Scheme 3).[13] Thus, chiral intermediate **11** was selectively protected by treatment with TBDPSCl in presence of DMAP to give compound **12**. Diene **12** was then cyclized *via* a ring-closing metathesis reaction in the presence of catalytic amount of 2nd generation Grubbs' catalyst **D**, affording desired cyclopentenol **13**. Meanwhile, *N*3-benzoyl-5-fluorouracil **15** was prepared in 2 steps from 5-fluorouracil **14** (Scheme 4) by benzoylation followed by selective deprotection using K_2CO_3 in dioxane.[14]

Coupling of chiral intermediate **13** with 3-*N*-Bz-5-F-uracil **15** under Mitsunobu conditions provided the desired *N*-alkylated derivative as a mixture with its *O*-alkylated isomer and DIAD byproducts (Scheme 4). The partially column purified mixture was directly deprotected by treatmentwith 7 *N* NH_3 in MeOH, followed by NH_4F in MeOH, to afford pure carbocyclic 5-fluorouridine derivative **17** along with pure *O*-alkylated compound **16** (ratio 4:1) in 93% yield over 3 steps. At this stage, introduction of pentenoyl moieties on both 5'-OH and 3-NH positions of compound **17** had to be optimized. While use of pyridine either alone or with DMAP or Et_3N gave the desired compound in low yields (the monoalkylated compound being predominant), the use of Et_3N in dichloromethane lead to the nearly quantitative formation of dialkylated compound **18**. With intermediate **18** in hand, a ring closing metathesis reaction to form the desired macrocycle **19** was investigated. Evaluation of solvents, catalyst loading and reaction time was undertaken. The best results were obtained when compound **18** was treated with 2.0 mol% of 2nd generation Grubbs' catalyst **D** (Figure 2) in toluene at 80 °C for 1 h.

As predicted, hydrogenation of unsaturated derivative **19** in the presence of Pd/C in EtOAc lead to complete stereoselective formation of cyclopentane derivative **15** in 85% yield. The absolute stereochemistry of the final product was determined by ^{1}H-NMR and 1D-NOE experiments on compound **21**, which was obtained after removal of the linker using NH_3 in MeOH. The NOE experiment showed an enhancement of the 4' proton when the 1' proton

was irradiated indicating that they reside on the same face of the sugar moiety. Finally, carbocyclic 5-F-uridine **21** was converted to its cytosine counterpart following a previously reported method.[15] The primary alcohol in compound **21** was first protected using TBSCl. Compound **22** was then being reacted with 2,4,6-triisopropylbenzenesulfonyl chloride followed by NH_4OH to give intermediate **23**. Final deprotection was achieved using HCl in MeOH providing the target 5-fluoro-carbodine **6** in 98% isolated yield.

CONCLUSION

In summary, an efficient synthetic route to 5-fluoro-carbodine **6** was developed utilizing a key α-face-selective hydrogenation driven by the introduction of a temporary macrocyclic tether. This approach represents a noteworthy improvement to the existing syntheses of biologically relevant compound **6** and allowed us to prepare **6** in 23% overall yield (compared with 2 to 10% yields from previous approaches) with no major purification problems nor use of special synthetic techniques. We are currently evaluating our umbrella approach's compatibility with other reactions to modify the olefin functionality and further explore the scope of this innovative methodology.

EXPERIMENTAL SECTION

General Considerations

Nuclear magnetic resonance (NMR) spectra (^1H, ^{13}C and ^{19}F) were recorded on a 400 MHz FT-NMR spectrometer at ambient temperature, with tetramethylsilane (TMS) as an internal standard. Chemical shifts (δ) are reported in parts per million (ppm), and signals are quoted as s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet), br (broad), dd (doublet of doublets), dt (doublet of triplets) or ddd (doublet of doublets of doublets). High-resolution mass spectra (HRMS) were recorded on a linear ion trap LTQ-FT mass spectrometer with electrospray ionization (ESI). Thin-layer chromatography (TLC) was performed on 0.25 mm silica gel. Purifications were carried out by silica gel column chromatography (60 Å, 63–200 μm, or 40–75 μm).

(R)-1-((4R,5S)-5-(3-(tert-Butyldiphenylsilyloxy)prop-1-en-2-yl)-2,2-dimethyl-1,3-dioxolan-4-yl)prop-2-en-1-ol (12)—To a solution of compound **11** (4.0 g, 20.5 mmol) in 100 mL of anhydrous CH_2Cl_2 was added DMAP (0.2 g, 1.4 mmol), TBDPSCl (6.3 g, 24.6 mmol) and Et_3N (3.1 g, 30.8 mmol) at 0 °C under N_2 atmosphere. The reaction mixture was stirred for 12 h at room temperature and quenched with 5.0 mL of MeOH. The solution was adsorbed on silica gel and purified by silica gel column chromatography (hexane:EtOAc = 20:1 to 2:1 v/v) to give compound **12** as a white foam (8.7 g, 19.1 mmol) in 93% yield. HRMS-ESI$^+$ calcd. for $C_{27}H_{37}O_4Si$ ($M+H^+$) 453.2462, found 453.2456; ^1H NMR (400 MHz, $CDCl_3$) δ 7.71-7.67 (m, 4H), 7.46-7.37 (m, 6H), 6.03-5.95 (m, 1H), 5.47 (dd, *J* = 13.2, 1.2 Hz, 2H), 5.32 (dt, *J* = 17.2, 1.2 Hz, 1H), 5.21 (dt, *J* = 10.4, 1.6 Hz, 1H), 4.65 (d, *J* = 5.6 Hz, 1H), 4.24 (s, 2H), 4.04-3.99 (m, 1H), 3.94 (dd, *J* = 8.8, 6.0 Hz, 1H), 2.63 (d, *J* = 3.2 Hz, 1H), 1.44 (s, 3H), 1.32 (s, 3H), 1.09 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ; 143.2, 138.0, 135.9, 135.8, 132.9, 132.8, 130.2, 130.1, 128.1, 128.0, 116.2, 113.7, 108.2, 80.9, 77.6, 70.3, 66.2, 27.5, 27.0, 25.4, 19.4.

(3aS,4S,6aR)-6-((tert-Butyldiphenylsilyloxy)methyl)-2,2-dimethyl-4,6a-dihydro-3aH-cyclopenta[d][1,3]dioxol-4-ol (13)—To a solution of compound **12** (8.2 g, 18.1 mmol) in 500 mL of anhydrous CH_2Cl_2 was added a solution of 2nd Grubb's catalyst (0.1 mol%, 0.02 g, 0.02 mmol) in 100 mL of anhydrous CH_2Cl_2 at room temperature under argon atmosphere. After stirring for 24 h, the reaction mixture was treated with 5.0 mL of DMSO, additionally stirred for 1 h at room temperature and concentrated under reduced

pressure. The residue was purified by silica gel column chromatography (hexane:EtOAc = 10:1 to 1:1 v/v) to give compound **13** as a white foam (7.40 g, 17.43 mmol) in 96% yield. HRMS-ESI$^+$ calcd. for $C_{25}H_{33}O_4Si$ (M+H$^+$) 425.2149, found 425.2143; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.69-7.65 (m, 4H), 7.44-7.34 (m, 6H), 5.84 (d, *J* = 1.6 Hz, 1H), 4.86 (d, *J* = 5.6 Hz, 1H), 4.74 (t, *J* = 5.2 Hz, 1H), 4.56 (m, 1H), 4.38 (dd, *J* = 15.2, 20.0 Hz, 2H), 2.70 (d, *J* = 10.0 Hz, 1H), 1.35 (s, 3H), 1.33 (s, 3H), 1.07 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 145.5, 135.7, 133.5, 129.9, 129.5, 127.9, 112.7, 83.0, 78.2, 73.5, 61.0, 27.8, 27.0, 26.9, 19.5.

N_3-Benzoyl-5-fluorouracil (15)—To a solution of 5-fluorouracil **14** (3.6 g, 27.3mmol) in 50 mL of anhydrous CH_3CN was added BzCl (9.3 g, 68.2 mmol), pyridine (10.8 g, 136.5mmol) at 0 °C under N_2 atmosphere. The solution was stirred for 24 h at room temperature and concentrated under reduced pressure. The residue was dissolved in 200 mL of CH_2Cl_2 and then washed with cold water (50 mL × 3) and concentrated under reduced pressure. The residue was re-dissolved in 50 mL of 1,4-dioxane and treated with aqueous K_2CO_3 (20 mL, 0.5 M in water solution) at 0 °C. After stirring for 30 min at room temperature, the pH of the solution was lowered to ca. 5.0 by careful addition of glacial acetic acid. The solution was concentrated *in vacuo* and the residue was treated with a saturated solution of $NaHCO_3$ (100 mL). After stirring for 1 h, the white solid was filtered and washed with cold water (20 mL × 5) then dried under vacuum for 48 h. The crude product was purified by silica gel column chromatography (hexane:EtOAc = 1:1 to EtOAc) to give compound **15** as a white amorphous solid (5.6 g, 23.7 mmol) in 87% yield. MS-ESI$^+$ *m/z* 235 (M+H$^+$); ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.54 (br, 1H), 8.06 (d, *J* = 6.4 Hz, 1H), 8.01 (d, *J* = 8.4 Hz, 2H), 7.77 (t, *J* = 7.6 Hz, 1H), 7.58 (dd, *J* = 8.4, 7.6 Hz, 2H).

5-Fluoro-2-(((3aS,4R,6aR)-6-(hydroxymethyl)-2,2-dimethyl-4,6a-dihydro-3aH-cyclopenta[d][1,3]dioxol-4-yl)oxy)pyrimidin-4(3H)-one (16) and 5-Fluoro-1-((3aS,4R,6aR)-6-(hydroxymethyl)-2,2-dimethyl-4,6a-dihydro-3aH-cyclopenta[d][1,3]dioxol-4-yl)pyrimidine-2,4(1H,3H)-dione (17)—To a solution of Ph_3P (11.1 g, 42.4 mmol) in 150 mL of CH_3CN was added DIAD (8.6 g, 42.4 mmol) at 0 °C under N_2 atmosphere. After 30 min, to the reaction mixture was added a solution of compound **13** (7.2 g, 17.0 mmol) in 150 mL of CH_3CN and *N*-Bz-5-fluorouracil **15** (6.0 g, 25.4 mmol) at −78 °C. The reaction mixture was stirred for 24 h at room temperature and then concentrated under reduced pressure. The residue was purified by silica gel column chromatography (hexane:EtOAc = 4:1 to 1:1 v/v) to give a non separable mixture of *N*-/*O*-alkylated products along with DIAD byproducts. This mixture was dissolved in 100 mL of MeOH and treated with 100 mL of 7 *N* NH_3 in MeOH at 0 °C. The solution was stirred for 12 h at room temperature and concentrated under reduced pressure. The residue was then directly treated with a solution of ammonium fluoride (1.6 g, 42.4 mmol) in 200 mL of MeOH at room temperature and the solution was stirred at 50 °C for 6 h. After removal of the volatiles under vacuum, the residue was purified by silica gel column chromatography (hexane:EtOAc = 1:5 to EtOAc v/v) to give **16** as an off white gum (0.94 g, 3.2 mmol) and **17** as an off white gum (3.8 g, 12.6 mmol) in 93% yield over 3 steps. Compound **16**: MS-ESI$^+$ *m/z* 299 (M+H$^+$); HRMS-ESI$^+$calcd. for $C_{13}H_{16}FN_2O_5$ (M+H$^+$) 299.1039, found 299.1035; ^{1}H NMR (400 MHz, CD_3OD) δ 7.77 (d, *J* = 3.2 Hz, 1H), 5.86 (d, *J* = 1.6 Hz, 1H), 5.72 (s, 1H), 5.19 (d, *J* = 5.6 Hz, 1H), 4.73 (d, *J* = 5.6 Hz, 1H), 4.24 (dd, *J* = 16.0, 10.8 Hz, 2H), 1.38 (s, 3H), 1.34 (s, 3H); ^{13}C NMR (100 MHz, CD_3OD) δ 154.7, 149.3, 146.8, 136.0, 123.7, 113.7, 87.3, 85.0, 84.4, 59.00, 27.8, 26.2; Compound **17**: MS-ESI$^+$ *m/z* 299 (M+H$^+$); HRMS-ESI$^+$calcd for $C_{13}H_{16}FN_2O_5$ (M+H$^+$) 299.1038, found 299.1036; ^{1}H NMR (400 MHz, CD_3OD) δ 7.54 (d, *J* = 6.4 Hz, 1H), 5.60 (m, 1H), 5.36 (s, 1H), 5.24 (d, *J* = 6.0 Hz, 1H), 4.62 (d, *J* = 5.6 Hz, 1H), 4.29 (s, 2H), 1.40 (s, 3H), 1.33 (s, 3H); ^{13}C NMR (100 MHz, CD_3OD) δ 155.0, 151.5, 143.1, 140.8, 127.8, 122.6, 113.5, 85.6, 84.8, 68.9, 60.0, 27.7, 26.1; ^{19}F NMR (376 MHz, CD_3OD) δ −169.69 (d, *J* = 6.0 Hz).

((3aR,6R,6aS)-6-(3-But-3-enoyl-5-fluoro-2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-2,2-dimethyl-6,6a-dihydro-3aH-cyclopenta[d][1,3]dioxol-4-yl)methyl but-3-enoate (18)—To a solution of compound **17** (1.4 g, 4.7mmol) in 40 mL of anhydrous CH_2Cl_2 was added Et_3N (2.4 g, 23.5 mmol) and 4-pentenoyl chloride (1.7 g, 14.1 mmol) at 0 °C under argon atmosphere. After stirring for 4 h at room temperature, the solution was concentrated under reduced pressure. The residue was purified by silica gel column chromatography (hexane:EtOAc = 4:1 to 1:1 v/v) to give a compound **18** as a white foam (2.1 g, 4.5 mmol) in 97% yield. MS-ESI$^+$ *m/z* 463 (M+H$^+$); HRMS-ESI$^+$calcd for $C_{23}H_{27}FN_2O_7Na$ (M+Na$^+$) 485.1705, found 485.1696; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.09 (d, *J* = 5.6 Hz, 1H), 5.90-5.78 (m, 2H), 5.57 (s, 1H), 5.39 (s, 1H), 5.21 (d, *J* = 5.6 Hz, 1H), 5.15-5.01 (m, 4H), 4.82 (q, *J* = 15.2 Hz, 2H), 4.59 (d, *J* = 5.6 Hz, 1H), 2.97 (t, *J* = 7.2 Hz, 2H), 2.51 (m, 4H), 2.43 (m, 2H), 1.43 (s, 3H), 1.35 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.9, 172.8, 158.7, 149.3, 148.0, 136.6, 135.6, 124.9, 122.9, 116.5, 116.1, 113.4, 94.6, 84.1, 83.5, 68.1, 60.9, 39.8, 33.4, 28.9, 27.5, 27.4, 25.9; ^{19}F NMR (376 MHz, $CDCl_3$) δ −165.50 (d, *J* = 4.5 Hz).

Compound 19—To a solution of compound **18** (1.0 g, 2.16 mmol) in 500 mL of anhydrous toluene was added 2nd Generation Grubbs' catalyst **D** (2.0 mol%, 0.037 g, 0.043 mmol) at room temperature under an argon atmosphere. After stirring for 1 h at 80 °C, the reaction mixture was treated with 2.0 mL of DMSO at room temperature and additionally stirred for 1 h. The solution was concentrated under reduced pressure and then purified by silica gel column chromatography (hexane:EtOAc = 2:1 v/v) to give compound **19** as a white foam (0.85 g, 2.0 mmol) in 90% yield. MS-ESI$^+$ *m/z* 435 (M+H$^+$); HRMS-ESI$^+$calcd for $C_{21}H_{24}FN_2O_7$ (M+H$^+$) 435.1562, found 435.1560; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.29 (d, *J* = 5.2 Hz, 1H), 5.62 (s, 1H), 5.56-5.40 (m, 3H), 5.25 (d, *J* = 11.6 Hz, 1H), 4.90 (d, *J* = 5.6 Hz, 1H), 4.42 (s, 1H), 4.25 (d, *J* = 11.2 Hz, 1H), 3.07 (ddd, *J* = 18.0, 8.0, 2.4 Hz, 1H), 2.86 (ddd, *J* = 18.0, 10.4, 2.4 Hz, 1H), 2.60 (m, 1H), 2.50-2.30 (m, 3H), 2.28-2.15 (m, 2H), 1.40 (s, 3H), 1.36 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 174.5, 172.7, 155.9, 146.9, 141.1, 138.7, 129.8, 129.1, 127.7, 125.5, 112.3, 84.5, 82.7, 74.9, 58.6, 39.4, 32.8, 27.4, 25.8; ^{19}F NMR (376 MHz, $CDCl_3$) δ −165.63 (d, *J* = 6.0 Hz), −166.20 (d, *J* = 6.0 Hz).

Compound 20—To a solution of compound **19** (2.5 g, 5.8 mmol) in 100 mL of EtOAc was added Pd/C catalyst (10 wt %, 0.25 g). The solution was stirred for 12 h under H_2 atmosphere (1 atm) and then treated with Celite. After stirring for 30 min, the suspension was carefully filtered, the filtrate concentrated under vacuum, and then purified by silica gel column chromatography (hexane:EtOAc = 1:1 v/v) to give compound **20** as a white foam (2.1 g, 4.9 mmol) in 85% yield. MS-ESI$^+$ *m/z* 439 (M+H$^+$); HRMS-ESI$^-$calcd for $C_{21}H_{27}FN_2O_7Cl$ (M+Cl$^-$) 473.1496, found 473.1499; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.28 (d, *J* = 5.6 Hz, 1H), 5.00 (dd, *J* = 6.4, 2.4 Hz, 1H), 4.88 (t, *J* = 5.6 Hz, 1H), 4.70 (d, *J* = 12.0, 1.2 Hz, 1H), 3.99 (dd, *J* = 12.0, 1.2 Hz, 1H), 3.91 (m, 1H) 2.95 (ddd, *J* = 16.0, 8.4, 3.2 Hz, 1H), 2.73 (ddd, *J* = 16.0, 9.6, 3.2 Hz, 1H), 2.55 (ddd, *J* = 16.0, 8.4, 5.2 Hz, 1H), 2.33-2.17 (m, 4H), 1.90 (m, 1H), 1.79 (s, 1H), 1.66-1.40 (m, 6H), 1.39-1.25 (m, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 174.4, 174.1, 147.6, 141.1, 138.7, 130.1, 129.8, 113.0, 94.6, 83.4, 80.3, 69.7, 61.0, 45.7, 40.1, 33.6, 31.2, 28.1, 27.1, 25.6, 24.4; ^{19}F NMR (376 MHz, $CDCl_3$) δ −166.59 (d, *J* = 4.5 Hz),

5-Fluoro-1-((3aS,4R,6R,6aR)-6-(hydroxymethyl)-2,2-dimethyltetrahydro-3aH-cyclopenta[d][1,3]dioxol-4-yl)pyrimidine-2,4(1H,3H)-dione (21)—A solution of compound **20** (2.0 g, 4.58 mmol) in 50 mL of MeOH was treated with 20 mL of 7 *N* NH_3 in MeOH at 0 °C in a sealed tube. After stirring for 12 h at 80 °C, the solution was concentrated under reduced pressure and the residue was purified on a silica gel pad (CH_2Cl_2:MeOH = 30:1 to 10:1 v/v) to give compound **21** as a beige gum (1.3 g, 4.3 mmol)

in 94% yield. MS-ESI$^+$ *m/z* 301 (M+H$^+$); HRMS-ESI$^+$calcd for $C_{13}H_{18}FN_2O_5$ (M+H$^+$) 301.1194, found 301.1193; ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.01 (br, 1H), 7.48 (d, *J* = 5.6 Hz, 1H), 4.77 (dd, *J* = 7.2, 5.6 Hz, 1H), 4.61 (dd, *J* = 7.2, 4.8 Hz, 1H), 4.57 (m, 1H), 3.83 (dd, *J* = 10.4, 4.8 Hz, 1H), 3.74 (dd, *J* = 10.8, 4.8 Hz, 1H), 2.80 (br, 1H) 2.37-2.27 (m, 2H), 2.12-2.03 (m, 1H), 1.54 (s, 3H), 1.31 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 157.5, 157.2, 149.8, 141.9, 139.6, 127.9, 127.6, 113.2, 83.0, 81.4, 64.2, 63.3, 45.6, 32.0, 27.7, 25.4; ^{19}F NMR (376 MHz, $CDCl_3$) δ −169.67 (d, *J* = 6.0 Hz).

1-((3aS,4R,6R,6aR)-6-((tert-Butyldiphenylsilyloxy)methyl)-2,2-dimethyltetrahydro-3aH-cyclopenta[d][1,3]dioxol-4-yl)-5-fluoropyrimidine-2,4(1H,3H)-dione (22)—To a solution of compound **21** (1.3 g, 4.5 mmol) in 30 mL of anhydrous CH_2Cl_2 was added imidazole (0.74 g, 10.9mmol) and TBDPSCl (1.7 g, 5.4 mmol) at 0 °C under N_2 atmosphere. The reaction mixture was stirred for 12 h at room temperature and treated with 5.0 mL of MeOH. After stirring for 1 h at room temperature, the solution was concentrated under reduced pressure and purified by silica gel column chromatography (hexane:EtOAc = 2:1 to 1:4 v/v) to give compound **22** as an off white foam (2.3 g, 4.3mmol) in 98% yield. MS-ESI$^+$ *m/z* 539 (M+H$^+$);HRMS-ESI$^+$calcd for $C_{29}H_{36}FN_2O_5Si$ (M+H$^+$) 539.2372, found 539.2368; ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.78 (br, 1H), 7.66-7.62 (m, 4H), 7.40-7.36 (m, 6H), 7.23 (d, *J* = 5.6 Hz, 1H), 4.71 (m, 1H), 4.63 (dd, *J* = 6.8, 6.0 Hz, 1H), 4.51 (dd, *J* = 7.2, 6.8 Hz, 1H), 3.79 (d, *J* = 5.2 Hz, 2H), 2.33 (m, 1H), 2.25 (m, 1H), 2.00 (q, *J* = 12.0 Hz, 1H), 1.53 (s, 3H), 1.29 (s, 3H), 1.09 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 157.2, 157.0, 149.7, 142.1, 139.7, 135.83, 135.78, 133.5, 133.4, 130.1, 128.0, 126.6, 126.3, 113.9, 82.8, 80.7, 64.3, 63.8, 45.2, 32.4, 27.8, 27.2, 25.5, 19.6; ^{19}F NMR (376 MHz, $CDCl_3$) δ −165.85 (d, *J* = 4.5 Hz).

4-Amino-1-((3aS,4R,6R,6aR)-6-((tert-butyldiphenylsilyloxy)methyl)-2,2-dimethyltetrahydro-3aH-cyclopenta[d][1,3]dioxol-4-yl)-5-fluoropyrimidin-2(1H)-one (23)—To a solution of compound **22** (2.2 g, 4.1 mmol) and DMAP (2.0 g, 16.4 mmol) in 50 mL of anhydrous CH_3CN was added 2,4,6-trisiopropyl benzenesulfonyl chloride (3.1 g, 10.3mmol) and Et_3N (1.7 g, 16.4 mmol) at room temperature under N_2 atmosphere. The reaction mixture was stirred for 48 h at room temperature and treated with 25 mL of 30% NH_4OH solution. After stirring for 4 h at room temperature, the solution was poured into 150 mL of a cold water-$CHCl_3$ solution (1:2 v/v) and then extracted with $CHCl_3$ (30 mL × 3). The combined organic layers were washed with a saturated NH_4Cl aqueous solution (100 mL), dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was purified by silica gel column chromatography (CH_2Cl_2 to CH_2Cl_2:MeOH = 10:1 v/v) to give compound **23** as a gum (2.0 g, 3.7mmol) in 89% yield. MS-ESI$^+$ *m/z* 538 (M+H$^+$); HRMS-ESI$^+$calcd for $C_{29}H_{37}FN_3O_4Si$ (M+H$^+$) 538.2532, found 538.2525; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.83, (br, 1H), 7.66-7.63 (m, 4H), 7.45-7.36 (m, 6H), 7.24 (d, *J* = 5.6 Hz, 1H), 5.60 (br, 1H), 4.80 (dd, *J* = 7.2, 5.2 Hz, 1H), 4.52-4.44 (m, 2H), 3.82 (dd, *J* = 10.0, 4.4 Hz, 1H), 3.74 (dd, *J* = 10.0, 6.0 Hz, 1H), 2.33-2.25 (m, 2H), 2.16 (m, 1H), 1.50 (s, 3H), 1.26 (s, 3H), 1.07 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 158.1, 154.6, 137.8, 135.8, 135.7, 135.4, 133.7, 133.6, 129.9, 128.4, 128.1, 127.9, 113.3, 83.0, 81.1, 66.1, 64.8, 46.2, 32.8, 27.8, 27.1, 25.5, 19.5; ^{19}F NMR (376 MHz, $CDCl_3$) δ −169.48 (d, *J* = 7.5 Hz).

4-Amino-1-((1R,2S,3R,4R)-2,3-dihydroxy-4-(hydroxymethyl)cyclopentyl)-5-fluoropyrimidin-2(1H)-one (6)—A solution of compound **23** (2.5 g, 4.7mmol) in 100 mL of MeOH was treated with 50 mL of HCl in MeOH at 0 °C. After stirring for 3 h at room temperature, the acidic solution was concentrated under reduced pressure. The residue was dissolved in water (50 mL) and washed with EtOAc (20 mL × 5). The aqueous layer was concentrated under reduced pressure. The residue was dissolved in EtOH (10 mL) and the solution was concentrated under reduced pressure. After this process was repeated 5 times,

the final residue was dried under vacuum for 72 h at room temperature to give pure compound **6** as a white amorphous solid (1.4 g, 4.6 mmol) as a HCl salt in 98% yield. MS-ESI$^+$ *m/z* 260 (M+H$^+$); HRMS-ESI$^+$calcd for $C_{10}H_{15}FN_3O_4$ (M+H$^+$) 260.1041, found 260.1039; ^{1}H NMR (400 MHz, CD_3OD) δ 8.35 (d, *J* = 7.2 Hz, 1H), 4.70 (q, *J* = 9.2 Hz, 1H), 4.12 (dd, *J* = 5.6, 9.2 Hz, 1H), 3.85 (dd, *J* = 5.6, 3.2 Hz, 1H), 3.53 (d, *J* = 5.2 Hz, 2H), 2.19 (m, 1H), 2.05 (m, 1H), 1.46 (m, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ 154.65, 154.42, 148.52, 139.97, 137.78, 133.74, 133.46, 75.44, 73.49, 64.83, 64.53, 46.50, 28.67; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.75 (br, 1H), 9.14 (br, 1H), 8.53 (d, *J* = 7.2 Hz, 1H), 5.32 (br, 3H), 4.65 (q, *J* = 9.2 Hz, 1H), 4.01 (dd, *J* = 9.2, 5.6 Hz, 1H), 3.72 (dd, *J* = 4.8, 2.4 Hz, 1H), 3.42 (dd, *J* = 10.4, 6.4 Hz, 1H), 3.35 (dd, *J* = 10.4, 5.6 Hz, 1H), 2.04 (m, 1H), 1.93 (m, 1H), 1.28 (m, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 153.1, 152.9, 147.8, 136.2, 133.9, 131.8, 131.5, 73.4, 71.2, 62.9, 61.8, 44.81, 27.8; ^{19}F NMR (376 MHz, CD_3OD) δ −170.99 (d, *J* = 7.52 Hz).

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

This work was supported in part by NIH grant 5P30-AI-50409 (CFAR) and by the Department of Veterans Affairs. Dr. Schinazi is the founder and a major shareholder of RFS Pharma, LLC. Emory received no funding from RFS Pharma, LLC to perform this work and *vice versa*.

REFERENCES

1. a) Marquez VE, Lim M-I. Med. Res. Rev. 1986; 6:1–40. [PubMed: 3512934] b) Borthwick AD, Biggadike K. Tetrahedron. 1992; 48:571–623.c) Agrofoglio L, Suhas E, Farese A, Condom R, Challand R, Earl RA, Guedj R. Tetrahedron. 1994; 50:10611–10670.d) Jenkins GN, Turner NJ. Chem. Soc. Rev. 1995; 24:169–176.e) Trost BM, Van Vranken DL. Chem. Rev. 1996; 96:395–422. [PubMed: 11848758] f) Ferrero M, Gotor V. Chem. Rev. 2000; 100:4319–4347. [PubMed: 11749350] g) Schneller SW. Curr. Top. Med. Chem. 2002; 2:1087–1092. [PubMed: 12171514] h) Pathak T. Chem. Rev. 2002; 102:1623–1667. [PubMed: 11996546] i) Rodriguez JB, Comin MJ. Mini Rev. Med. Chem. 2003; 3:95–114. [PubMed: 12570843] j) Agrofoglio LA, Gillaizeau I, Saito Y. Chem. Rev. 2003; 103:1875–1916. [PubMed: 12744695] k) Jeong LS, Lee JA. Antiviral Chem. Chemother. 2004; 15:235–250.l) Kurteva VB, Afonso CAM. Chem. Rev. 2009; 109:6809–6857. [PubMed: 19772301]
2. a) Kusaka T, Yamamoto H, Shibata M, Murori M, Hicki T, Mizuno K. J. Antibiot. 1968; 21:255–263. [PubMed: 5671989] b) Shealy YF, Clayton JD. J. Pharm. Sci. 1973; 62:858–859. [PubMed: 4705710]
3. a) Yaginuma S, Tsujino M, Sudate Y, Hayashi M, Otani M. J. Antibiot. 1981; 34:359–366. [PubMed: 7275815] b) De Clercq E. Antimicrob. Agents Chemother. 1985; 28:84–89. [PubMed: 2994559]
4. a) Kimberlain DW, Coen DM, Biron KK, Cohen JI, Lamb RA, Mckinlay M, Emini EA, Whitley RJ. Antiviral Res. 1995; 26:369–401. [PubMed: 7574541] b) Foster RH, Faulds D. Drugs. 1998; 55:729–736. [PubMed: 9585869]
5. a) Bisacchi GS, Chao ST, Bachard C, Daris JP, Innaimo S, Jacobs GA, Kocy O, Lapointe P, Martel A, Merchant Z, Slusarchyk WA, Sundeen JE, Young MG, Colonno R, Zahler R. Bioorg. Med. Chem. Lett. 1997; 7:127–132.b) Yamanaka G, Wilson T, Innaimo S, Bisacchi GS, Egli P, Rinehart JK, Zahler R, Colonno RJ. Antimicrob. Agents Chemother. 1999; 43:190–193. [PubMed: 9869593]
6. Stuyver, L.; Watanabe, KA. Patent. WO 02/32920 A2. 2002.
7. Julander JG, Bowen RA, Rao JR, Day C, Shafer K, Smee DF, Morrey JD, Chu CK. Antiviral Res. 2008; 80:309–315. [PubMed: 18675850]
8. Chu, CK.; Rao, J. Patent. WO 2008/124157 A1. 2008.
9. Ohno M. Nucleosides Nucleotides. 1985; 4:21.

10. Russ PL, Hegedus L, Kelley JA, Barchi JJ Jr, Marquez VE. Nucleosides Nucleotides. 1992; 11:351.

11. For a review on metathesis strategy in nucleoside chemistry see: Amblard F, Nolan SP, Agrofoglio LA. Tetrahedron. 2005; 61:7067.

12. The two possible reduction products were energy minimized with the MM2 force field in Chem 3D Ultra version 10. 0.

13. Cho JH, Bernard DL, Sidwell RW, Kern ER, Chu CK. J. Med. Chem. 2006; 49:1140–1148. [PubMed: 16451078]

14. a) Cruickshank K, Jiricny J, Reese CB. Tetrahedron Lett. 1984; 25:681–684.b) Frieden M, Giraud M, Reese CB, Song Q. J. Chem. Soc. Perkin.Trans. 1. 1998:2827–2832.c) Lucey NM, McCormick JE, McElhinney RS. J. Chem. Soc. Perkin. Trans. 1. 1990:795–802.d) Lee Y-S, Kim BH. Bioorg. Med. Chem. Lett. 2002; 12:1395–1397. [PubMed: 11992785] e) Shatila RS, Bouhadir KH. Tetrahedron Lett. 2006; 47:1767–1770.f) Ludek OR, Meier C. Eur. J. Org. Chem. 2006:941–946.

15. a) Shuto S, Obara T, Saito Y, Andrei G, Snoeck R, De Clercq E, Matsuda A. J. Med. Chem. 1996; 39:2392–2399. [PubMed: 8691433] b) Jin YH, Liu P, Wang J, Baker R, Huggins J, Chu CK. J. Org. Chem. 2006; 68:9012–9018. [PubMed: 14604375]

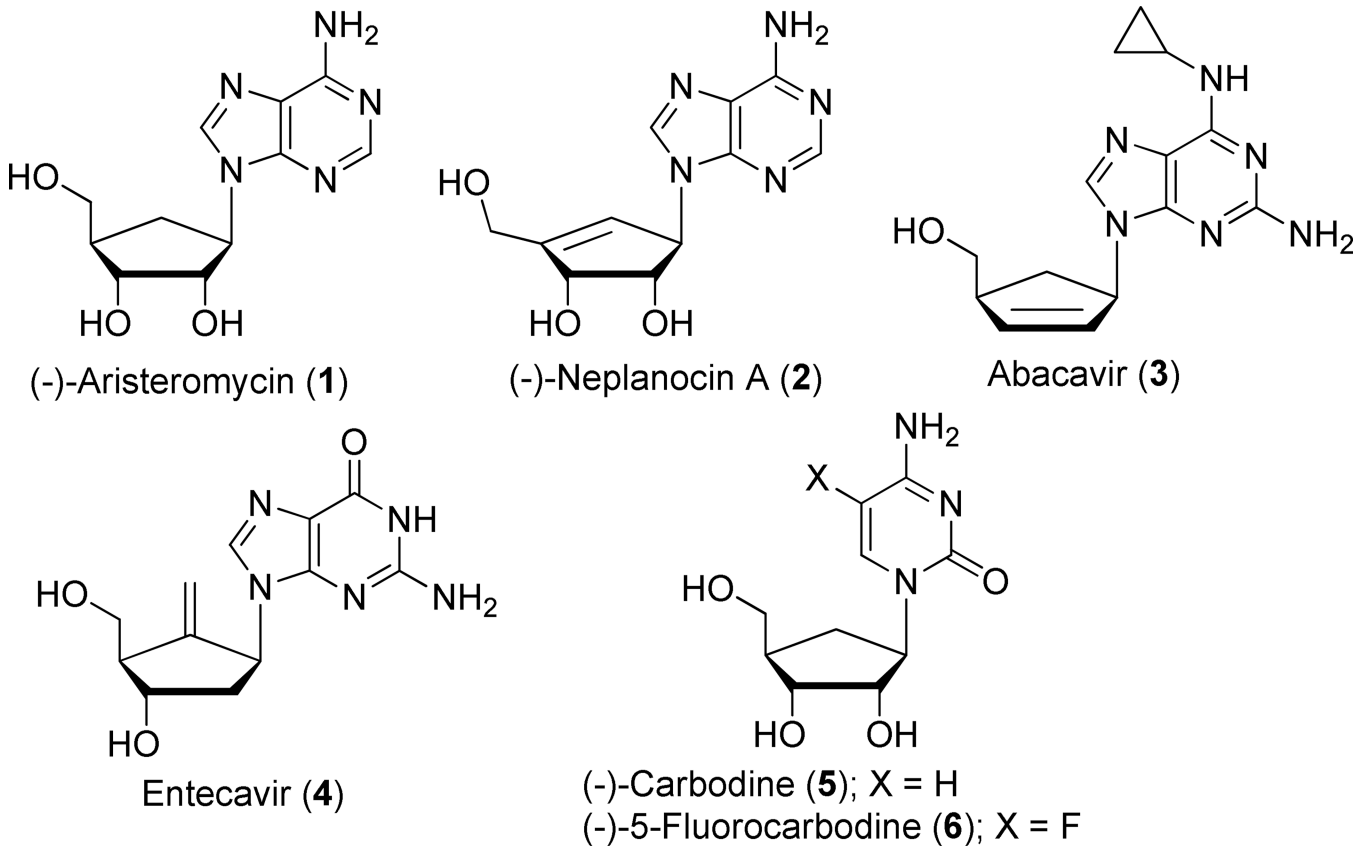
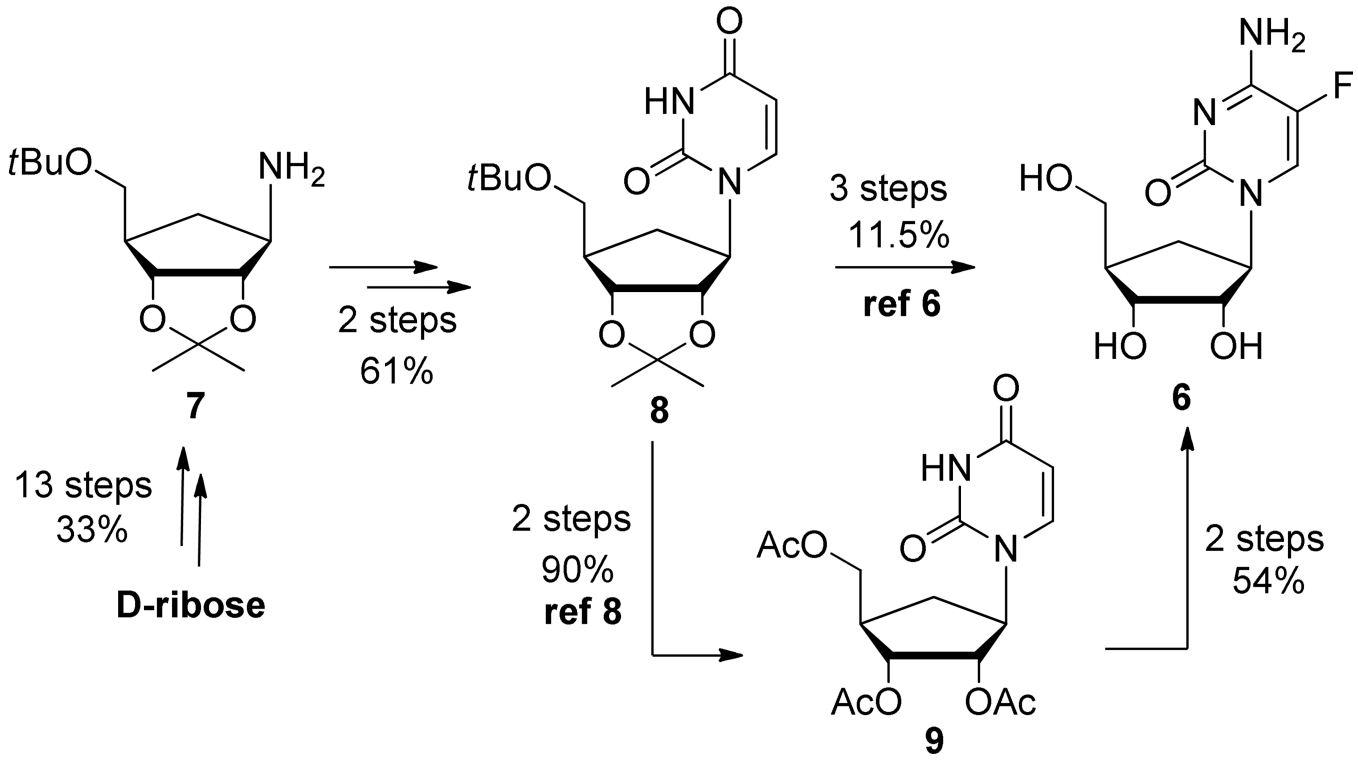
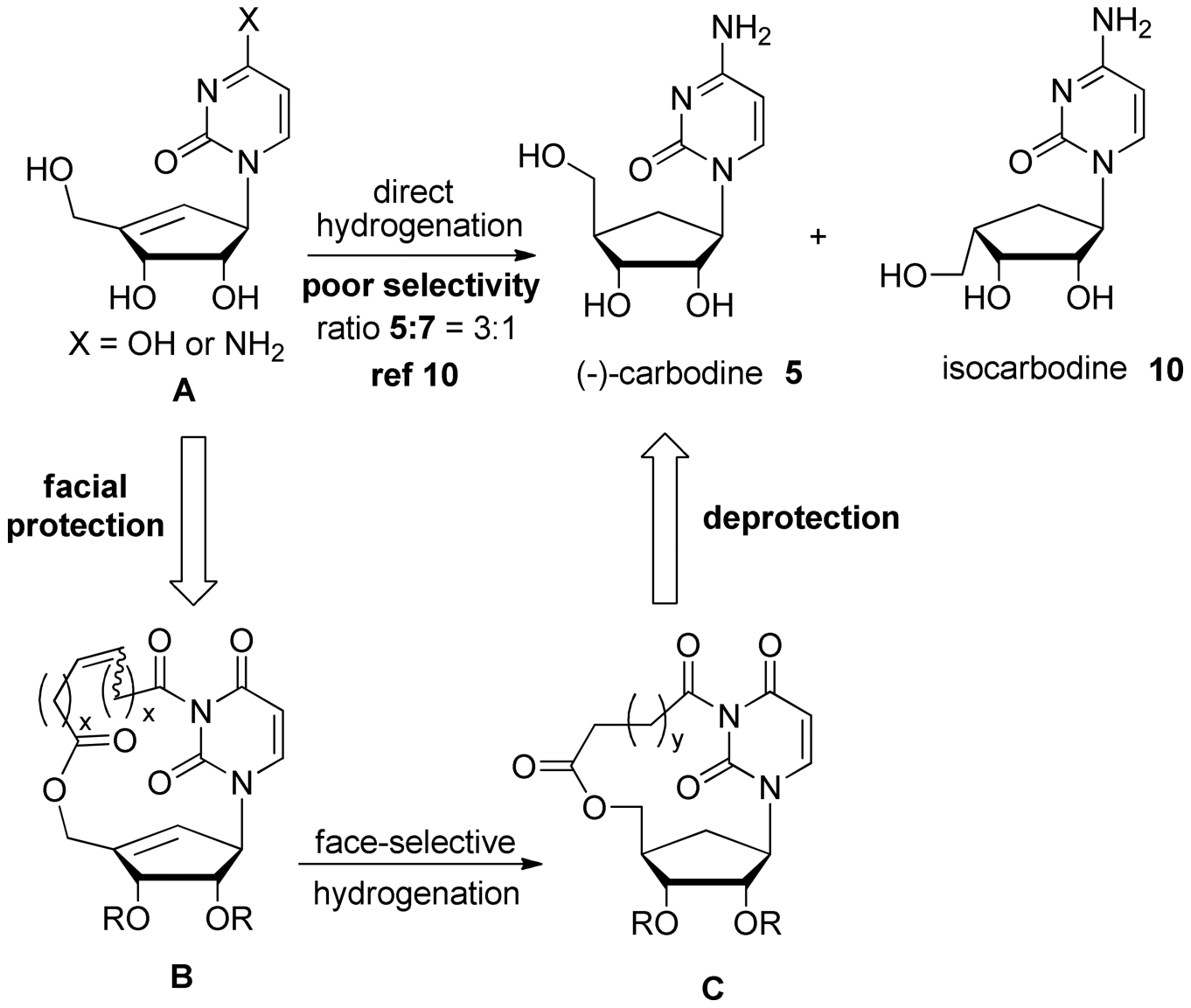


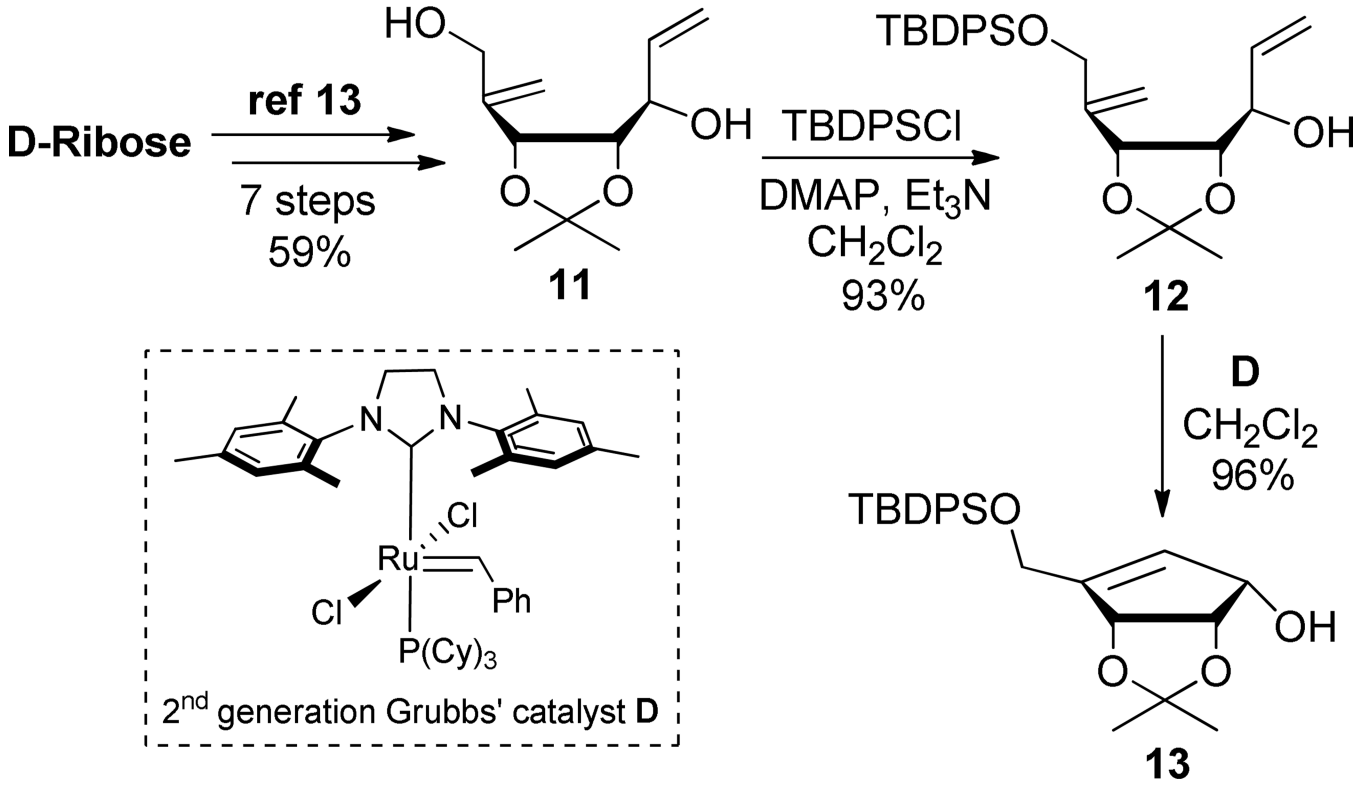
Figure 1.
Structures of Biologically Relevant Carbocyclic Nucleosides



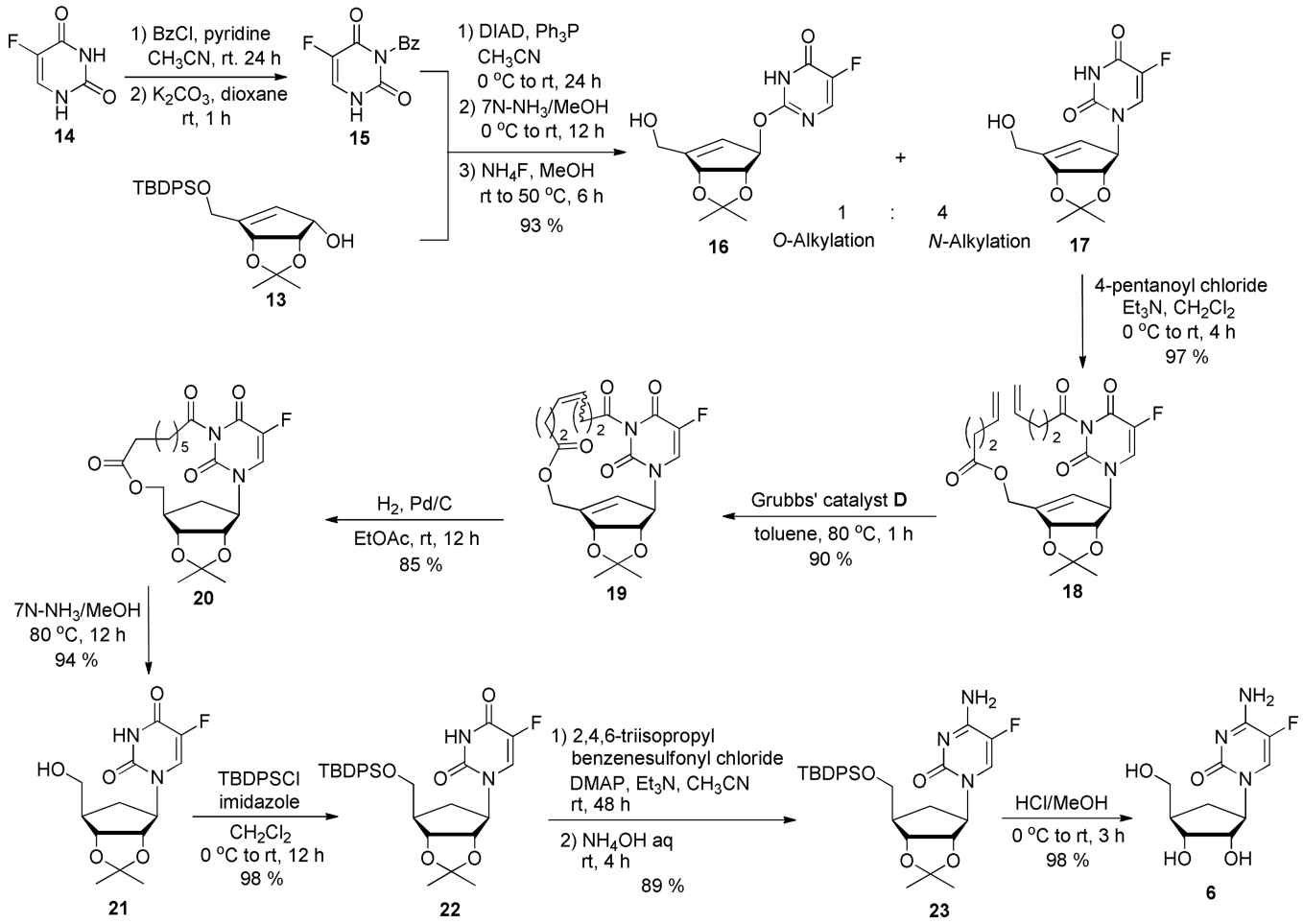
Scheme 1.
Linear Approaches for the Synthesis of 5-Fluorocarbodine (**6**)



Scheme 2.
Facially Selective Hydrogenation



Scheme 3.
Preparation of Protected Sugar **13**



Scheme 4.
Synthesis of 5-Fluorocarbodine **6**