Chiral Synthesis of 4-[1-(2-Deoxy-β-L-ribofuranosyl)] Derivatives of 2-Substituted 5-Fluoroaniline: "Cytosine Replacement" Analogues of Deoxy-β-L-cytidine

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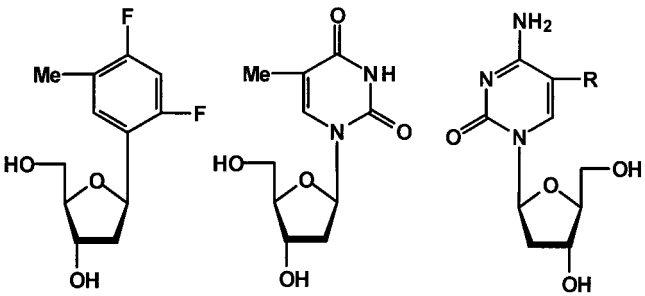
Introduction

L-Thymidine (L-TdR), a substrate for herpes simplex virus type 1 thymidine kinase (HSV-1 TK), reduces HSV-1 multiplication in HeLa cells. HSV-1 TK phosphorylates the L and D enantiomers of TdR to their corresponding monophosphates (MPs) with identical efficacy.[1] Similar results have been observed for the L-TdR analogues 5-iodo-2′-deoxyuridine (L-IUdR) and (*E*)-5-(2-bromovinyl)-2′-deoxyuridine (L-BVUdR), whose D enantiomers are potent, but cytotoxic, antiherpetic drugs. The approximately 1000-fold lower cytotoxicity of L-IUdR and L-BVUdR, relative to the D enantiomers, is due to the fact that L-IUdR lacks affinity for cellular TK and that L-IUdRMP and L-BVUdRMP, in contrast to their D enantiomers, do not inhibit thymidylate synthase (TS).[2] Thus, the viral TK enzyme, but not human cytosolic TK, lacks enantioselectivity for natural β-D- and unnatural β-L-nucleosides. Consequently, L-nucleosides have attracted the attention of medicinal chemists due to their unique potency, mechanism of action, and toxicity profile.[3] Some representative L-cytidine analogues such as 2′,3′-dideoxy-3′-thia-β-L-cytidine (3TC, Lamivudine),[4,5] 2′,3′-dideoxy-3′-thia-β-L-5-fluorocytidine (L-FTC),[6] and 2′,3′-dideoxy-β-L-5-fluorocytidine (L-FddC)[7,8] have shown promising antiviral activity. 3TC and L-FTC exhibit more potent antiviral activity against human immunodeficiency virus (HIV) and hepatitis B virus (HBV), and lower toxicity, in comparison to the D enantiomers.[5,9]

Nonpolar hydrophobic isosteres of β-D-pyrimidine nucleosides which retain close structural, steric, and isoelectronic relationships to the natural base, which are not likely to form hydrogen bonds, have been reported by Kool et al.[10] In this regard, the 2,4-difluoro-5-methylphenyl isostere (β-D-**1**) was designed as an unnatural mimic of thymidine (β-D-**2**). Furthermore, the 5′-tri-



phosphate of β-D-**1** (β-D-**1**-TP) was selectively inserted opposite adenine (A) into replicating DNA strands by the Klenow fragment (KF, exo⁻ mutant) of *Escherischia coli* DNA polymerase 1 with an efficacy (V_{max}/K_m) only 40-fold lower than that for β-D-**2**-TP.[11] These results indicated that the 2,4-difluoro-5-methylphenyl moiety of β-D-**1** is isoelectronic with the thymine base which it replaces and is utilized by KF polymerase.[12−14] It was envisaged that the structurally related 4-(2-substituted 5-fluoroaniline)-β-L-nucleoside mimics (**13a**,**b**), which are hybrids of the *C*-aryl (β-D-**1**) and deoxycytidine nucleosides (β-L-**3a**,**b**), may have interesting biological activity.[1,2] We now report the synthesis of the 4-[1-(2-deoxy-β-L-ribofuranosyl)] derivatives of 2-substituted-5-fluoroanilines (β-L-**13a**,**b**), which were designed as unnatural *C*-aryl 2′-deoxy-β-L-cytidine mimics.

Results and Discussion

The Heck-type coupling reaction constitutes a simple, yet direct, method to form a C−C bond between a suitably protected glycal and an activated iodo- or trifluoromethanesulfonate-substituted aryl (heteroaryl) reagent, to prepare the β-anomer of nucleosides in reasonable yields.[15] In this study, the Heck coupling reaction is a key method for the synthesis of unnatural deoxy-β-L-cytidine mimics, as illustrated in Scheme 1.

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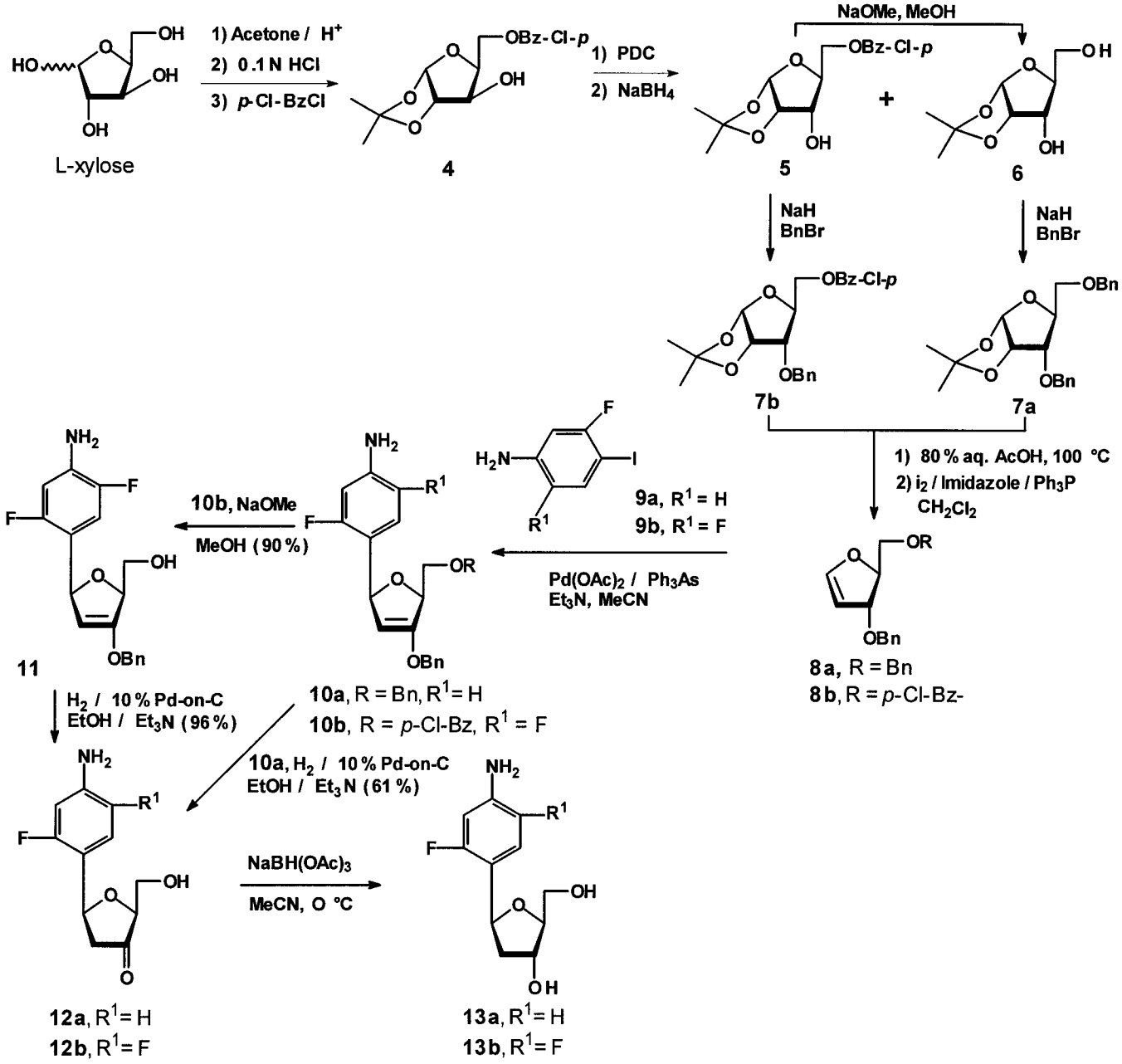
Scheme 1



The L-ribose derivatives **5** and **6** were prepared by adapting reported procedures.[16] Ketalization of L-xylose, in acetone in the presence of anhydrous $CuSO_4$ and a catalytic amount of concentrated H_2SO_4, followed by selective hydrolysis with 0.1 N HCl afforded 1,2-*O*-isopropylidene-α-xylofuranose.[17] Selective protection of the 5-OH using *p*-Cl-Bz-Cl in pyridine−CH_2Cl_2 at 0 °C gave **4**. The 3-OH substituent of the pentofuranose derivative **4** was oxidized by PDC in CH_2Cl_2, and the resulting ketone was reduced stereoselectively using $NaBH_4$ to afford a mixture of the desired ribose derivatives **5** (68%) and **6** (28%). Chu et al.[16] reported that the use of EtOAc−EtOH (1:2) as solvent prevented cleavage of a benzoyl group, although in our hands the diol **6** was produced using the same reaction conditions. The diol **6** was also readily prepared by treatment of **5** with NaOMe in MeOH. Reaction of **6** with benzyl bromide in THF in the presence of NaH gave the 3,5-di-*O*-benzyl L-ribose derivative **7a**. Careful treatment of the 5-*O*-(*p*-chlorobenzoyl) L-ribose derivative **5** with benzyl bromide and NaH in dry THF afforded the 5-*O*-(*p*-chlorobenzoyl)-3-benzyl L-ribose derivative **7b** in 87% yield.

Although numerous attempts have been made to prepare furanose glycals, two reactions have been the most useful in the carbohydrate field. For example, Garegg and Samuelsson have found that action of the iodine−triphenylphosphine−imidazole reagent on a *vic*-

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diol produced an unstable *vic*-diiodide that underwent an iodine-elimination reaction to afford the corresponding unsaturated sugar.[18] The second reaction is related to the Corey and Hopkins dideoxygenation procedure,[19] where the action of 1,3-dimethyl-2-phenyl-1,3,2-diazaphospholidine (DMPD) on a *vic*-diol thiocarbonate was shown to result in elimination to afford the olefin product, which has been used for the synthesis of enofuranosides[20] and furanoid glycals.[21] We adapted a method related to the Garegg and Samuelsson procedure[18] to prepare the ribofuranoid glycals **8a** and **8b**.

The 1,2-*O*-isopropylidenyl group in **7a** and **7b** was readily removed by treatment with 80% aqueous AcOH at 100 °C. The resulting *vic*-diols were treated directly with the iodine−triphenylphosphine−imidazole reagent in dry CH_2Cl_2 at 25 °C to afford the desired ribofuranoid glycals **8a** and **8b** in 45 and 62% yields, respectively. Garegg and Samuelsson performed similar reactions at reflux temperature in toluene for several hours to prepare unsaturated sugars.[18] In our study, the starting material (**7a**,**b**) immediately disappeared after addition of the reagent, to yield the corresponding glycal (**8**). This observation, which is identical with a report by Diaz et al.,[21] is attributed to the higher reactivity of the hydroxyl group at the anomeric position.

The Heck coupling reaction of 3-fluoro-4-iodoaniline (**9a**) or 2,5-difluoro-4-iodoaniline (**9b**) with glycal **8a** or **8b** in the presence of palladium(II) acetate, Ph_3As, and Et_3N in dry MeCN at 70 °C,[22] afforded **10a** (57%) or **10b** (71%). A variety of methods were investigated to convert the benzyl enol ether (**10**) to the ketone (**12**). Thus, hydrogenation of **10b** using 10% Pd on C and H_2 gas in THF, or EtOH, gave a complex mixture of products. Treatment of **10b** with TMSI gave the 5-*O*-(*p*-chlorobenzoyl)-protected ketone in low yield (25%), and subsequent reduction of this ketone with $NaB(OAc)_3H$ slowly produced a mixture of two C-3 OH isomers. Treatment of **10a**, or **10b**, with BCl_3 in CH_2Cl_2 at −78 °C also did not give the desired ketone, even though the starting compound was completely consumed. The instability of compounds **10a**,**b** is probably due to the high reactivity of the sugar ring oxygen atom, which is located at an allylic and benzylic position that makes it susceptible to attack by acid, resulting in sugar ring opening. It was therefore anticipated that the decomposition of **10a**, or **10b**, could be prevented if the reaction was performed under basic conditions. Accordingly, the Pd on C catalyzed hydrogenation of **10a** in anhydrous EtOH containing several drops of Et_3N at 60 °C afforded the hydroxy ketone **12a** in 61% yield. The 5-(*p*-chlorobenzoyl) group of **10b** was readily removed by treatment with NaOMe in MeOH, and the resulting benzyl enol ether (**11**) was converted to the hydroxy ketone **12b** in quantitative yield upon Pd on C catalyzed hydrogenation in anhydrous EtOH in the presence of several drops of Et_3N at 60 °C. Subsequent reduction of the hydroxy ketones **12a**,**b** with $NaB(AcO)_3H$ [22] in dry MeCN afforded the target deoxy-β-L-cytidine C-nucleoside mimics (**13a**,**b**). This stereospecific reduction apparently involves initial coordination of the borohydride reagent with the C-5′ hydroxyl and hydride delivery from the hindered β-face of the carbohydrate ring.[15f] The configuration and conformation of **13b** were analyzed by nuclear Overhauser enhancement (NOE) ^{1}H NMR difference spectroscopy (see Figure 1). Selective irradiation of the H-4′ signal resulted in an enhancement of the H-1′ signal (2.5%) and irradiation of the H-2″ signal gave a 6.3% enhancement of the H-1′ signal, which support the assignment of the β-configuration.[22] The C-6 hydrogen of the 4-amino-2,5-difluorophenyl moiety is oriented in the direction of the sugar ring, since NOE enhancements of the C-6 hydrogen were observed upon selective irradiation of H-1′ (1.6%), H-2′ (2.8%), and the CH_2OH (1.1%) signals.

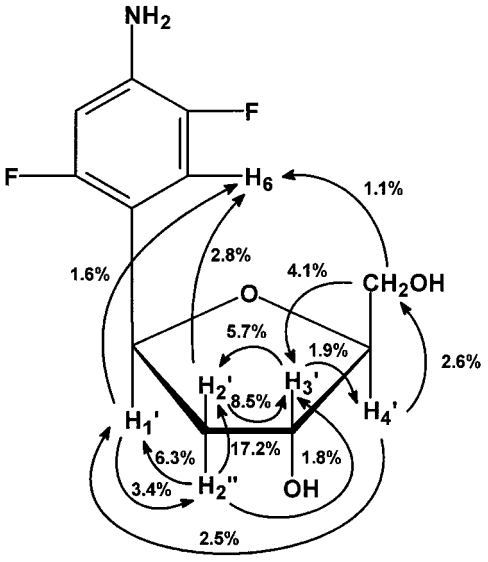

Figure 1. NOE determinations of the conformation and configuration of 2,5-difluoro-4-[1-(2-deoxy-β-L-ribofuranosyl)]-aniline (**13b**) in MeOH-d_4 at 22 °C.

Replacement of the natural cytosine base moiety in β-L-**3a**, or β-L-**3b**, by an unnatural aryl isostere such as a 2-substituted 5-fluoroaniline ring system could confer new properties that may be useful in the design of a novel class of third-generation *C*-aryl deoxy-β-L-cytidine mimics. These mimics offer a number of potential advantages, such as resistance to pyrimidine phosphorylases due to the absence of a glycosyl bond, decreased host cell toxicity due to their inability to inhibit TS, and resistance to deamination (inactivation) by deoxycytidine deaminase.

The ability of β-L-**13a** and β-L-**13b** to inhibit the proliferation of murine leukemia cells (L1210/0), murine mammary carcinoma cells (FM3*A*/0), and human T-lymphocyte cells (Molt4/C8, CEM/O) in cell cultures by 50% (IC_{50} ± SEM) were evaluated using a previously reported procedure.[23] In these assays **13a** and **13b** provided negligible (IC_{50}: 300−400 μM), or no (IC_{50} > 500 μM), inhibition of L1210/0 (IC_{50} = 402 ± 138 μM (**13a**) and 451 ± 10 μM (**13b**)), FM3A/0 (IC_{50} > 500 μM (**13a,b**)), Molt 4/C8 (IC_{50} = 297 ± 45 μM (**13a**) and > 500 μM (**13b**)), or CEM/0 (IC_{50} = 406 ± 133 μM (**13a**) and > 500 μM (**13b**)) cell proliferation. In addition **13a**,**b** did not protect (EC_{50} > 250 μM) human T-lymphocytes (CEM

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cells) against the cytopathogenicity induced by human immunodeficiency viruses (HIV-1, HIV-2).[24]

Experimental Section

General Methods. Melting points are uncorrected. ^{1}H NMR (300 MHz), ^{13}C NMR (75.5 MHz), and ^{19}F NMR (282.4 MHz) spectra were recorded with TMS (δ 0), $CDCl_3$ (δ 77) or DMSO-d_6 (δ 39.5), and external C_6F_6 (δ 0) as reference standards, respectively. NOE studies were performed under steady-state conditions using the Bruker NOE DIFF.AU software program (signal to noise ratio of 136 for a single pulse). MeOH-d_4 was dried using molecular sieves (type 3A, 1.6 mm pellets) and degassed by passage of dry argon at 22 °C just prior to use. Elemental analyses were performed by the MicroAnalysis Service Laboratory, Department of Chemistry, University of Alberta. Silica gel 60 (E. Merck Co.) was employed for all silica gel column flash chromatography separations. All reagents were purchased from the Aldrich Chemical Co.

5-*O*-(*p*-Chlorobenzoyl)-1,2-*O*-isopropylidene-α-L-xylofuranose (4). A mixture of L-xylose (24.5 g, 0.163 mol), dry $CuSO_4$ (50 g, 0.341 mol), and concentrated H_2SO_4 (2 mL) in acetone (200 mL) was stirred at 25 °C for 24 h. The mixture was filtered, and the solid was washed with acetone. The filtrate was neutralized with concentrated NH_4OH, and the resulting white solid was removed by suction filtration. Removal of the solvent in vacuo from the filtrate gave a syrup that was treated with aqueous 0.1 N HCl (150 mL) for 1 h at 25 °C, during which time the reaction mixture turned to a clear solution. The reaction was quenched with solid $NaHCO_3$ to a pH of 7.5. The solution was washed with ether once, and the aqueous fraction was evaporated in vacuo to yield a pale yellow syrup, which was dissolved in $CHCl_3$ (100 mL) prior to drying (Na_2SO_4). Filtration, and removal of the solvent in vacuo, afforded a pale yellow syrup (30.5 g, 98%). *p*-Chlorobenzoyl chloride (20.1 mL, 0.158 mol) was added dropwise over 30 min to an ice-cold solution of this syrup (30 g, 0.158 mol) and pyridine (27 mL) in dry CH_2Cl_2 (130 mL). The resulting mixture was stirred at 0 °C for 1 h and washed with aqueous 1 N HCl and saturated $NaHCO_3$, prior to drying the organic fraction (Na_2SO_4). After filtration, and removal of the solvent in vacuo, the residue obtained was recrystallized from ether to afford white crystals (37 g). The product remaining in the mother liquor was purified by flash chromatography (hexanes−EtOAc, 5:1 to 2:1 v/v) to give additional product (4.5 g), providing the title compound **4** (41.5 g, 80%) as white crystals: mp 102−104 °C; ^{1}H NMR ($CDCl_3$) δ 7.99 (d, J = 8.5 Hz, 2H), 7.43 (d, J = 8.5 Hz, 2H), 5.96 (d, J = 3.7 Hz, 1H), 4.77 (dd, J = 12.8, 8.8 Hz, 1H), 4.59 (d, J = 3.7 Hz, 1H), 4.5−4.4 (m, 2H), 4.19 (br s, 1H), 3.16 (br s, 1H, D_2O exchangeable), 1.51 (s, 3H), 1.33 (s, 3H); ^{13}C NMR ($CDCl_3$) δ 166.10, 139.84, 131.10, 128.70, 127.87, 111.83, 104.79, 85.17, 78.46, 74.64, 62.14, 26.76, 26.13. Anal. Calcd for $C_{15}H_{17}ClO_6$: C, 54.81; H, 5.21. Found: C, 54.72; H, 5.28.

5-*O*-(*p*-Chlorobenzoyl)-1,2-*O*-isopropylidene-α-L-ribofuranose (5) and 1,2-*O*-Isopropylidene-α-L-ribofuranose (6). PDC (30.7 g, 0.0816 mol) and Ac_2O (42.3 mL, 0.449 mol) were added to a solution of **4** (41 g, 0.125 mol) in dry CH_2Cl_2 (350 mL), and the resulting solution was stirred at reflux for 2 h. After the mixture was cooled to 25 °C, the solvent was removed in vacuo, and the residue obtained was dissolved in EtOAc (50 mL). The mixture was filtered through a silica gel pad, and the pad was washed with EtOAc−hexane (1:1 v/v). The solvent from the filtrate was removed in vacuo, and the residue was coevaporated with toluene (2 × 50 mL). The residue was recrystallized from hexanes−EtOAc to afford 5-*O*-(*p*-chlorobenzoyl)-1,2-*O*-isopropylidene-α-L-erythro-pentofuranos-3-ulose (35 g, 87%) as white crystals, mp 104−105 °C.

$NaBH_4$ (3.27 g, 86 mmol) was added in aliquots to a stirred solution of the above ketone (28 g, 85.76 mmol) in anhydrous EtOH (100 mL) and EtOAc (100 mL) at 0 °C; the mixture was stirred at 0 °C for 1 h and then neutralized with dilute AcOH. Removal of the solvent in vacuo gave a residue that was dissolved in EtOAc; this solution was passed through a silica gel pad, and the pad was washed with EtOAc. Removal of the solvent from the filtrate gave a residue that was recrystallized from hexanes−ether to give **5** (15.8 g, 56%) as white crystals. Removal of the solvent from the mother liquor gave a residue that was purified via flash chromatography (hexanes−EtOAc, 2:1 to 1:2 v/v) to provide additional **5** (3.4 g, 12%) and **6** (4.5 g, 28%), affording 19.2 g of **5** (68% total yield).

Compound 5: white crystals; mp 136−138 °C; ^{1}H NMR ($CDCl_3$) δ 7.98 (d, J = 8.5 Hz, 2H), 7.40 (d, J = 8.5 Hz, 2H), 5.84 (d, J = 4.0 Hz, 1H), 4.68 (dd, J = 12.2, 2.4 Hz, 1H), 4.60 (dd, J = 5.2, 4.0 Hz, 1H), 4.43 (dd, J = 12.2, 5.5 Hz, 1H), 4.07 (ddd, J = 8.8, 5.8, 2.4 Hz, 1H), 3.93 (m, 1H), 2.60 (br s, 1H, D_2O exchangeable), 1.58 (s, 3H), 1.37 (s, 3H); ^{13}C NMR ($CDCl_3$) δ 165.43, 139.55, 131.09, 128.66, 128.29, 112.80, 104.10, 78.50, 78.30, 72.28, 63.72, 26.54, 26.49. Anal. Calcd for $C_{15}H_{17}ClO_6$: C, 54.81; H, 5.21. Found: C, 54.68; H, 5.15.

Compound 6: white crystals; mp 87−89 °C (lit.[16] mp 86−87 °C); ^{1}H NMR ($CDCl_3$) δ 5.76 (d, J = 3.7 Hz, 1H), 4.54 (dd, J = 4.9, 3.7 Hz, 1H), 3.96 (dd, J = 8.8, 4.9 Hz, 1H), 3.89 (dd, J = 12.2, 2.4 Hz, 1H), 3.82 (ddd, J = 8.8, 3.3, 2.4 Hz, 1H), 3.69 (dd, J = 12.2, 3.3 Hz, 1H), 3.2 (br s, 2H, D_2O exchangeable), 1.53 (s, 3H), 1.33 (s, 3H); ^{13}C NMR ($CDCl_3$) δ 113.13, 104.37, 81.09, 79.25, 71.35, 61.23, 26.93 (2C).

1,2-*O*-Isopropylidene-α-L-ribofuranose (6). NaOMe (1.9 g, 35 mmol) was added to a suspension of **5** (8.5 g, 25.8 mmol) in MeOH (50 mL) at 25 °C, and the resulting mixture was stirred at 25 °C for 30 min. The reaction was quenched with solid NH_4-Cl, and the mixture was diluted with ether (150 mL), filtered, and washed with ether. Removal of the solvent from the filtrate gave a residue that was purified via flash chromatography (hexanes−EtOAc, 1:1 to 1:2 v/v) to give the diol **6** (4.81 g, 98%) as white crystals. The melting point and NMR spectral data for **6** were identical with those listed above.

3,5-Di-*O*-benzyl-1,2-*O*-isopropylidene-α-L-ribofuranose (7a). The diol **6** (4.8 g, 25 mmol) was added in aliquots to an ice-cold suspension of NaH (60% in mineral oil, 2.4 g, 60 mmol) in dry THF (80 mL) with stirring under argon, and the resulting mixture was stirred at 0 °C for 30 min, prior to addition of benzyl bromide (7.3 mL, 60 mmol). After the reaction mixture was stirred at 0 °C for 1 h, and then at 25 °C for 12 h, the reaction was quenched with solid NH_4Cl. The reaction mixture was diluted with ether (100 mL), and the mixture was washed with water and brine, prior to drying the organic fraction (Na_2SO_4). After filtration and removal of the solvent in vacuo, the residue obtained was purified via flash chromatography (hexanes−ether, 1:0 to 1:1 v/v) to afford **7a** (8.65 g, 92%) as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 7.4−7.2 (m, 10H), 5.79 (d, J = 4.0 Hz, 1H), 4.76 (d, J = 11.9 Hz, 1H), 4.60 (d, J = 7.0 Hz, 1H), 4.56 (d, J = 7.0 Hz, 1H), 4.58 (m, 1H), 4.52 (d, J = 11.9 Hz, 1H), 4.22 (ddd, J = 9.2, 4.0, 2.1 Hz, 1H), 3.89 (dd, J = 9.2, 4.3 Hz, 1H), 3.80 (dd, J = 11.3, 2.1 Hz, 1H), 3.60 (dd, J = 11.3, 4.0 Hz, 1H), 1.63 (s, 3H), 1.39 (s, 3H); ^{13}C NMR ($CDCl_3$) δ 138.08, 137.67, 128.25, 128.18, 127.82, 127.76, 127.54, 127.42, 112.73, 104.13, 77.43, 77.38, 73.44, 72.14, 68.28, 26.78, 26.59.

5-*O*-(*p*-Chlorobenzoyl)-3-*O*-benzyl-1,2-*O*-isopropylidene-α-L-ribofuranose (7b). Compound **5** (13.14 g, 40 mmol) and benzyl chloride (7.2 mL, 60 mmol) were added in aliquots to an ice-cold suspension of NaH (60% in mineral oil, 1.8 g, 45 mmol) in dry THF (50 mL) with stirring under argon. The reaction mixture was stirred at 0 °C for 30 min and then at 25 °C for 5 h. Solid NH_4Cl (5 g) was added to quench the reaction, and the mixture was stirred for another 10 min. The reaction mixture was diluted with ether (200 mL) and washed with water (150 mL). The aqueous fraction was extracted with ether (2 × 100 mL), and the combined organic extracts were washed with saturated aqueous NH_4Cl and brine prior to drying (Na_2SO_4). After filtration, and removal of the solvent in vacuo, the residue obtained was purified via flash chromatography (hexanes−EtOAc, 10:1 v/v) to give **7b** (14.49 g, 87%) as white crystals: mp 84−85 °C; ^{1}H NMR ($CDCl_3$) δ 7.89 (d, J = 8.2 Hz, 2H), 7.5−7.3 (m, 7H), 5.81 (d, J = 3.7 Hz, 1H), 4.85 (d, J = 11.9 Hz, 1H), 4.6−4.7 (m, 2H), 4.59 (d, J = 11.9 Hz, 1H), 4.45−4.35 (m, 2H), 3.79 (dd, J = 8.8, 4.3 Hz, 1H), 1.67 (s, 3H), 1.44 (s, 3H); ^{13}C NMR ($CDCl_3$) δ 165.16, 139.39, 137.23, 131.01, 128.54, 128.41, 128.28, 128.02, 127.93, 113.08, 104.20, 77.56, 76.96, 76.33, 72.11, 63.35,

(24) Balzarini, J.; Naesens, L.; Slachmuylders, J.; Niphuis, H.; Rosenberg, L.; Holý, A.; Schellekens, H.; De Clercq, E. *AIDS* **1991**, *5*, 21−28.

26.77, 26.54. Anal. Calcd for $C_{22}H_{23}ClO_6$: C, 63.08; H, 5.53. Found: C, 62.89; H, 5.37.

1,4-Anhydro-3,5-di-*O*-benzyl-2-deoxy-L-*erythro*-pent-1-enitol (8a). A solution of **7a** (4.63 g, 12.5 mmol) in 80% aqueous AcOH (80 mL) was stirred at 100 °C for 5 h. Removal of the solvent in vacuo gave a residue that was coevaporated with toluene once. The residue was dissolved in EtOAc (100 mL) and treated with solid $NaHCO_3$. Filtration and removal of the solvent in vacuo afforded the diol as a colorless syrup, which was used directly in the subsequent reaction.

Triphenylphosphine (7.22 g, 27.5 mmol) and imidazole (7.52 g, 110.6 mmol) were added in aliquots to a well-stirred solution of iodine (7.0 g, 27.5 mmol) in dry CH_2Cl_2 (100 mL) at 25 °C, during which time the reaction mixture turned to a pale yellow. The above diol in dry CH_2Cl_2 (20 mL) was added to the reaction mixture, which immediately changed color to brown. After it was stirred at 25 °C for 10 min, Et_3N (5 mL) was added to the mixture, and the reaction mixture was concentrated to half-volume before dilution with hexane (100 mL). The mixture was passed through a silica gel pad that was washed with a large volume of hexanes−ether (1:1 v/v) and then ether. The solvents from the combined organic eluants were removed, and the residue obtained was purified via flash chromatography (hexanes−ether, 6:1 v/v, the silica gel was treated with 1% Et_3N in hexane before packing the column to avoid on-column decomposition of the glycal **8a**) to afford the glycal **8a** (1.65 g, 45%) as a colorless syrup: ^{1}H NMR ($CDCl_3$) δ 7.4−7.2 (m, 10H), 6.62 (d, J = 2.7 Hz, 1 H), 5.20 (dd, J = 2.7, 2.1 Hz, 1H), 4.75−4.60 (m, 2H), 4.61 (d, J = 4.0 Hz, 2H), 4.56 (s, 2H), 3.59 (dd, J = 9.9, 6.1 Hz, 1H), 3.46 (d, J = 9.9, 5.3 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 150.20, 138.26, 137.83, 128.30, 128.27, 127.72, 127.61, 127.57, 127.48, 100.53, 84.81, 82.68, 73.38, 69.89, 69.55.

1,4-Anhydro-5-*O*-(*p*-chlorobenzoyl)-3-*O*-benzyl-2-deoxy-L-*erythro*-pent-1-enitol (8b). A solution of **7b** (5.25 g, 12.5 mmol) in 80% aqueous AcOH (80 mL) was stirred at 100 °C for 4 h. Removal of the solvent in vacuo gave a residue that was coevaporated with toluene once; the residue was dissolved in EtOAc (100 mL) and treated with solid $NaHCO_3$. Filtration, and removal of the solvent, afforded the diol as a colorless syrup, which was used directly in the subsequent reaction.

Triphenylphosphine (7.22 g, 27.5 mmol) and imidazole (7.52 g, 110.6 mmol) were added in aliquots to a well-stirred solution of iodine (7.0 g, 27.5 mmol) in dry CH_2Cl_2 (100 mL) at 25 °C; the reaction was completed and the product purified as described for the preparation of **8a** above to afford the glycal **8b** (2.55 g, 62%) as a colorless syrup: ^{1}H NMR ($CDCl_3$) δ 7.97 (d, J = 8.8 Hz, 2H), 7.43 (d, J = 8.8 Hz, 2H), 7.4−7.25 (m, 5H), 6.63 (dd, J = 2.7, 0.8 Hz, 1H), 5.27 (dd, J = 2.7, 2.4 Hz, 1H), 4.79 (td, J = 5.5, 3.0 Hz, 1H), 4.72 (ddd, J = 3.0, 2.4, 0.8 Hz, 1H), 4.56 (s, 2H), 4.35 (d, J = 5.5 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 165.32, 150.45, 139.68, 137.95, 131.09, 128.77, 128.45, 128.16, 127.74, 100.70, 83.57, 82.52, 69.79, 64.70. Anal. Calcd for $C_{19}H_{17}ClO_4$: C, 66.19; H, 4.97. Found: C, 66.24; H, 4.91.

4-(5-((Benzyloxy)methyl)-4-(benzyloxy)-2,5-dihydro-β-L-furan-2-yl)-3-fluoroaniline (10a). A freshly dried 100 mL round-bottom flask was charged with palladium acetate (0.1 g, 0.364 mmol) and triphenylarsine (0.4 g, 1.33 mmol) under argon. Dry MeCN (15 mL) was added, and the yellow suspension was stirred at 25 °C for 30 min. A solution of the glycal **8a** (0.592 g, 2 mmol), **9a** (0.592 g, 2.5 mmol), and Et_3N (0.6 mL) in MeCN (10 mL) was added with stirring, and the resulting solution was stirred at 70 °C for 24 h. The solvents were removed in vacuo, and the residue was purified via flash chromatography (hexanes−EtOAc, 3:1 v/v) to give **10a** (0.46 g, 57%) as a yellow syrup: IR (NaCl) 3471, 3368, 3229, 1664, 1630 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.45−7.2 (m, 11H), 6.35−6.3 (m, 2H), 6.10 (dd, J = 3.3, 1.2 Hz), 4.93 (br s, 3H), 4.83 (br s, 1H), 4.63 (d, J = 3.7 Hz, 2H), 3.85 (dd, J = 10.7, 2.4 Hz), 3.74 (dd, J = 10.7, 4.9 Hz, 1H), 3.7 (br s, 2H, D_2O exchangeable); ^{13}C NMR ($CDCl_3$) δ 160.95 (d, J = 243.9 Hz), 154.81, 147.67 (d, J = 11.0 Hz), 138.43, 136.15, 129.63 (d, J = 5.5 Hz), 128.41, 128.12, 127.98, 127.56, 127.34, 127.25, 119.16 (d, J = 13.2 Hz), 110.72, 101.30 (d, J = 25.3 Hz), 96.40, 81.49, 73.25, 72.27, 71.46.

4-[5-(((*p*-Chlorobenzoyl)oxy)methyl)-4-(benzyloxy)-2,5-dihydro-β-L-furan-2-yl]-2,5-difluoroaniline (10b). Reaction of **8b** with **9b**, using the method described for the preparation of **10a** above, and purification of the product (hexanes−EtOAc, 4:1 v/v) followed by recrystallization from hexanes−ether gave **10b** (0.81 g, 71%) as pale yellow needles: mp 137−138 °C; IR (NaCl) 3479, 3365, 1721, 1666, 1643 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.88 (d, J = 8.5 Hz, 2H), 7.36 (d, J = 8.5 Hz, 2H), 7.33 (br s, 5H), 7.02 (dd, J = 11.3, 6.4 Hz, 1H), 6.43 (dd, J = 10.7, 7.2 Hz, 1H), 6.03 (m, 1H), 5.03 (m, 1H), 4.93 (d, J = 5.5 Hz, 2H), 4.89 (m, 1H), 4.63 (dd, J = 11.9, 2.7 Hz, 1H), 4.55 (dd, J = 11.9, 4.0 Hz, 1H), 3.75 (br s, 2H, D_2O exchangeable); ^{13}C NMR ($CDCl_3$) δ 165.43, 156.25 (d, J = 240.6 Hz), 154.24, 147.2 (d, J = 235.3 Hz), 139.21, 135.72, 135.21 (dd, J = 15.4, 11.0 Hz), 131.00, 128.50, 128.41, 128.19, 127.41, 118.61 (dd, J = 16.0, 5.5 Hz), 113.96 (dd, J = 21.9, 6.0 Hz), 103.10 (dd, J = 27.5, 3.3 Hz), 96.29, 79.97, 78.31 (d, J = 2.2 Hz), 72.60, 65.06. Anal. Calcd for $C_{25}H_{20}ClF_2NO_4$: C, 63.63; H, 4.27; N, 2.97. Found: C, 63.41; H, 4.00; N, 2.86.

3-Fluoro-4-(β-L-glyceropentofuran-3-ulos-1-yl)aniline (12a). Pd on C (10% w/w, 0.1 g) was added to a solution of **10a** (0.38 g, 0.936 mmol) in dry EtOH (6 mL) and Et_3N (0.05 mL), and the resulting mixture was stirred at 60 °C under 1 atm of H_2 gas for 8 h. After filtration, and removal of the solvent in vacuo, the residue was purified via flash chromatography (hexanes−EtOAc, 1:1 v/v) to give the hydroxy ketone **12a** (0.13 g, 61%) as pale yellow crystals: mp 132−133 °C; IR (NaCl) 3446, 3364, 3220, 1752, 1635 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.20 (dd, J = 8.5, 8.2 Hz, 1H), 6.39 (dd, J = 8.5, 2.1 Hz, 1H), 6.30 (dd, J = 12.5, 2.1 Hz, 1H), 5.22 (dd, J = 11.0, 6.0 Hz, 1H), 3.90 (t, J = 3.0 Hz, 1H), 3.78 (d, J = 3.0 Hz, 2H), 3.62 (br s, 3H, D_2O exchangeable), 2.70 (dd, J = 18.1, 6.0 Hz, 1H), 2.48 (dd, J = 18.1, 11.0 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 214.2, 161.26 (d, J = 243.9 Hz), 148.61 (d, J = 9.9 Hz), 128.39 (d, J = 6.6 Hz), 115.4 (d, J = 14.5 Hz), 110.74 (d, J = 2.2 Hz), 101.68 (d, J = 25.3 Hz), 82.32, 72.1, 61.05, 43.87. Anal. Calcd for $C_{11}H_{12}FNO_3$: C, 58.66; H, 5.37; N, 6.22. Found: C, 58.29; H, 5.30; N, 6.15.

4-(5-(Hydroxymethyl)-4-(benzyloxy)-2,5-dihydro-β-L-furan-2-yl)-2,5-difluoroaniline (11). NaOMe (0.16 g, 3 mmol) was added to a suspension of **10b** (0.8 g, 1.75 mmol) in dry MeOH (30 mL) with stirring, and the reaction mixture was stirred at 25 °C for 1.5 h, during which time the reaction mixture changed to a clear solution. The reaction was quenched with solid NH_4Cl, and then ether (100 mL) was added. After filtration, and removal of the solvent in vacuo, the residue obtained was purified via flash chromatography (hexanes−EtOAc, 2:1 v/v) to give **11** (0.465 g, 90%) as a yellow syrup: IR (NaCl) 3465, 3362, 3224, 1663, 1645, 1518 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.45−7.3 (m, 5H), 7.05 (dd, J = 11.3, 6.4 Hz, 1H), 6.45 (dd, J = 11.0, 7.3 Hz, 1H), 6.03 (m, 1H), 4.95 (d, J = 3.1 Hz, 2H), 4.79 (m, 1H), 4.76 (m, 1H), 3.80 (d, J = 3.4, 2H); ^{13}C NMR ($CDCl_3$) δ 156.25 (d, J = 240.6 Hz), 155.39, 147.6 (d, J = 235.1 Hz), 135.85, 135.57 (dd, J = 15.4, 11.0 Hz), 128.53, 128.19, 127.42, 118.41 (dd, J = 16.0, 5.5 Hz), 114.22 (dd, J = 22.0, 5.5 Hz), 103.20 (dd, J = 28.6, 3.3 Hz), 95.53, 82.33, 77.88 (d, J = 3.3 Hz), 72.53, 62.95. Anal. Calcd for $C_{18}H_{17}F_2NO_3$: C, 64.85; H, 5.14; N, 4.20. Found: C, 64.55; H, 5.21; N, 3.98.

2,5-Difluoro-4-(β-L-glyceropentofuran-3-ulos-1-yl)-aniline (12b). Et_3N (3 drops) and 10% Pd on C (0.1 g) were added to a solution of **11** (0.42 g, 1.42 mmol) in dry EtOH (10 mL) with stirring, and the resulting mixture was stirred at 60 °C under 1 atm of H_2 gas for 2 h. After filtration, and removal of the solvent in vacuo, the residue was purified via flash chromatography (hexanes−EtOAc, 1.5:1 to 1:1 v/v) to give the hydroxy ketone **12b** (0.33 g, 96%) as a colorless syrup: IR (NaCl) 3465, 3368, 3231, 1758, 1649, 1522 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.14 (dd, J = 11.3, 6.4 Hz, 1H), 6.48 (dd, J = 11.3, 7.3 Hz, 1H), 5.29 (dd, J = 11.0, 5.8 Hz, 1H), 4.0 (t, J = 3.3 Hz, 1H), 3.95 (d, J = 3.3 Hz, 2H), 3.92 (br s, 2H, D_2O exchangeable), 2.84 (dd, J = 18.0, 5.8 Hz, 1H), 2.49 (dd, J = 18.0, 11.0 Hz, 1H), 2.47 (br s, 1H, D_2O exchangeable); ^{13}C NMR ($CDCl_3$) δ 213.41, 156.45 (dd, J = 240.6, 2.2 Hz), 147.47 (dd, J = 235.1, 2.2 Hz), 135.83 (dd, J = 15.4, 12.1 Hz), 115.41 (dd, J = 16.9, 5.5 Hz), 113.5 (dd, J = 22.0, 5.5 Hz), 103.3 (dd, J = 27.5, 4.4 Hz), 82.0, 71.76, 61.40, 44.10. Anal. Calcd for $C_{11}H_{11}F_2NO_3$: C, 54.32; H, 4.56; N, 5.76. Found: C, 54.61; H, 5.05; N, 5.51.

3-Fluoro-4-[1-(2-deoxy-β-L-ribofuranosyl)]aniline (13a). $NaB(OAc)_3H$ (0.26 g, 1.2 mmol) was added in one aliquot to an ice-cold solution of **12a** (0.09 g, 0.3 mmol) in dry MeCN (5 mL) with stirring under argon. After the mixture was stirred at 0 °C for 1.5 h, the reaction was quenched with MeOH (2 mL), and

the solvents were removed in vacuo. The residue was purified via flash chromatography (hexanes−EtOAc, 1:2 to 0:1 v/v) to give **13a** (0.075 g, 83%) as a pale yellow syrup, which was recrystallized from EtOH to give pale yellow crystals (0.020 g, 22%): mp 182−184 °C; $[\alpha]^{27}_D$ −27.6° (*c* 0.17, MeOH); UV (MeOH) λ_{max} 243.1 nm (ϵ 11 156), 288.5 (ϵ 1757); ^{1}H NMR (DMSO-d_6) δ 7.10 (dd, *J* = 8.8, 8.5 Hz, 1H), 6.34 (d, *J* = 8.5 Hz, 1H), 6.25 (d, *J* = 13.2 Hz, 1H), 5.31 (s, 2H, D_2O exchangeable), 5.06 (dd, *J* = 10.4, 5.5 Hz, 1H), 5.0 (d, *J* = 4.0 Hz, 1H, D_2O exchangeable), 4.69 (t, *J* = 5.5 Hz, 1H, D_2O exchangeable), 4.14 (m, 1H), 3.69 (m, 1H), 3.41 (m, 2H), 1.94 (dd, *J* = 12.6, 5.5 Hz, 1H), 1.78 (ddd, *J* = 12.6, 10.4, 5.5 Hz, 1H); ^{13}C NMR (DMSO-d_6) δ 160.52 (d, *J* = 241.7 Hz), 149.74 (d, *J* = 12.1 Hz), 128.12, 114.91 (d, *J* = 14.3 Hz), 109.73, 99.62 (d, *J* = 27.5 Hz), 87.05, 72.83, 72.30 (d, *J* = 8.8 Hz), 62.41 (d, *J* = 8.8 Hz), 41.76 (d, *J* = 3.3 Hz); ^{19}F NMR (DMSO-d_6) δ 46.03 (dd, *J* = 12.2, 9.2 Hz). Anal. Calcd for $C_{11}H_{14}FNO_3$: C, 58.14; H, 6.21; N, 6.16. Found: C, 58.08; H, 6.42; N, 6.09.

2,5-Difluoro-4-[1-(2-deoxy-β-L-ribofuranosyl)]aniline (13b). $NaB(OAc)_3H$ (1.1 g, 5.2 mmol) was added in one portion to an ice-cold solution of **12b** (0.31 g, 1.275 mmol) in dry MeCN (15 mL) with stirring under argon. After stirring at 0 °C for 2 h, the reaction was quenched with MeOH (5 mL), and the solvents were removed in vacuo. The residue was purified via flash chromatography (hexanes−EtOAc, 1:2 to 0:1 v/v) to give **13b** (0.26 g, 84%) as a pale yellow solid, which was recrystallized from ether to give pale yellow crystals (0.21 g, 68%): mp 163−165 °C; $[\alpha]^{27}_D$ −42.8° (*c* 0.26, MeOH); UV (MeOH) λ_{max} 237.3 nm (ϵ 12 656), 289.2 (ϵ 3663); ^{1}H NMR (MeOH-d_4) δ 7.09 (dd, *J* = 12.2, 6.7 Hz, 1H), 6.47 (dd, *J* = 11.9, 7.6 Hz, 1H), 5.25 (dd, *J* = 10.4, 5.2 Hz, 1H), 4.21 (m, 1H), 3.92 (m, 1H), 3.63 (m, 2H), 2.10 (dd, *J* = 12.5, 5.2 Hz, 1H), 1.91 (ddd, *J* = 12.5, 10.4, 5.5 Hz, 1H); ^{13}C NMR (DMSO-d_6) δ 155.53 (d, *J* = 238.4 Hz), 146.61 (d, *J* = 232.9 Hz), 136.58 (dd, *J* = 15.4, 12.1 Hz), 115.26 (dd, *J* = 16.5, 5.5 Hz), 112.95 (dd, *J* = 20.9, 6.6 Hz), 101.69 (dd, *J* = 27.5, 4.4 Hz), 87.17, 72.22, 72.14, 62.20, 42.02. Anal. Calcd for $C_{11}H_{13}F_2NO_3$: C, 53.88; H, 5.34; N, 5.71. Found: C, 53.64; H, 5.36; N, 5.60.

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