Supporting Information

Synthesis of gem-difluorinated nucleoside analogues of the liposidomycins and evaluation as MraY inhibitors

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Synthesis and characterization data of compounds 5, 10, 11, 3, 14, 15, 7, 16-19 and 4.

COSY NMR and NOESY NMR spectra of compounds 4 and 11
Tetrahydrofuran (THF) was distilled from sodium metal. Dichloromethane (CH$_2$Cl$_2$), acetonitrile (CH$_3$CN), nitromethane (CH$_3$NO$_2$), toluene and pyridine were distilled from CaH$_2$. $^1$H NMR spectra were recorded on a Bruker AM300 spectrometer. $^{19}$F NMR spectra were recorded on a Bruker AM300 spectrometer (FCCl$_3$ as outside standard and low field is positive). All the melting points and optical rotations are uncorrected. The following abbreviations were used to explain the multiplicities: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet.

2,3-Di-O-acetyl-5-azido-5-deoxy-D-ribofuranose (5). To a cooled solution (0°C) of 1,2,3-Tri-O-acetyl-5-azido-5-deoxy-D-ribofuranose 9 (960 mg, 3.19 mmol) in dry CH$_2$Cl$_2$ (190 mL) was slowly added HBr-HOAc (30%, 1.69 mL). After the mixture was stirred at the same temperature for 2 h, saturated aq. NaHCO$_3$ was added to quench the reaction. The organic layer was dried over anhydrous Na$_2$SO$_4$, and concentrated in vacuo. The residue was purified by silica gel column chromatography (petroleum ether: ethyl acetate = 4: 3) to afford the compound 5 (422 mg, 51% yield): clear oil, $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 5.60-5.09 (m, 3H, H-1, H-2 and H-3), 4.37-4.22 (m, 1H, H-4), 3.65-3.59 (m, 1H, H-5a), 3.50-3.43 (m, 1H, H-5b), 3.17 (s, 1H, OH), 2.16-2.07 (m, 6H, CH$_3$); $^{13}$C NMR (75.5 MHz, CDCl$_3$) $\delta$, major 169.8 (COCH$_3$), 169.7 (COCH$_3$), 100.2 (C-1), 79.8 (C-4), 75.5 (C-2), 71.5 (C-3), 52.9 (C-5), 20.5 (COCH$_3$), 20.4 (COCH$_3$), minor 170.0 (COCH$_3$), 169.6 (COCH$_3$), 95.6 (C-1), 80.6 (C-4), [71.1, 71.0 (C-2, C-3)], 52.1 (C-5), 20.7 (COCH$_3$), 20.6 (COCH$_3$); MS (ESI) m/z 277.1 (M+NH$_4^+$); IR (thin film) 3457 (OH), 2940 (CH), 2104 (N 3), 1741 (C=O), 1438, 1375, 1239, 965 cm$^{-1}$; HRMS (ESI) calcd for C$_9$H$_{13}$N$_3$O$_6$Na 282.0697, found 282.0704.

5-O-[5''-Azido-5'''-deoxy-2'',3''-O-diacetyl-beta-D-ribofuranosyl]-3-deoxy-3,3-O-difluoro-1-(uracil-1'y)-beta-D-arabinofuranose (10). A solution of compound 5 (406 mg, 1.57 mmol), CCl$_3$CN (0.79 mL, 7.85 mmol) and DBU (1 drop) in CH$_2$Cl$_2$ (16 mL)
was stirred at room temperature for 1.5 h. Then solvent was removed, and the residue was purified by flash chromatography on a silica gel column (petroleum ether: ethyl acetate = 3: 1, with 1% Et₃N) to give the corresponding trichloroacetimidate derivative (480 mg, 76% yield), which was used in the next step without further characterization. To a suspension of the trichloroacetimidate derivative (480 mg, 1.19 mmol), compound 6 (256 mg, 0.97 mmol) and 4 Å MS (970 mg) in dry CH₃CN (10 mL) at -20 °C, was slowly added TMSOTf (0.07 mL, 0.30 mmol) in dry CH₃CN (2 mL). After being stirred at the same temperature for 1 h, the reaction was quenched with Et₃N (2 mL), filtered, and concentrated. The residue was purified by silica gel column chromatography (dichloromethane: methanol = 60 : 1) to give the compound 10 (333 mg, 68% yield) as a white solid: mp 59-61 °C; [α]²¹_D = +1.4 (c 0.95, CH₃OH); ¹H NMR (300 MHz, CD₃OD) δ 7.86 (d, J₅₆,₆ 8.1 Hz, 1H, H-6), 6.03 (dd, J₁°₁,2°₂ 7.2 Hz, J₁°₂,₂°₇ 1.8 Hz, 1H, H-1°), 5.94 (d, J₅₆,₆ 7.8 Hz, 1H, H-5), 5.54-5.31 (m, 2H, H-2°° and H-3°°), 5.18 (s, 1H, H-1°°), 4.50-4.32 (m, 3H, H-5°₂, H-2° and H-4°°), 4.20 (dd, J₅°₂,₅°₂ 11.7 Hz, J₄°₅°₂ 13.2 Hz, 1H, H-5°b), 3.80 (dd, J₅°₁,₅°₂ 11.1 Hz, J₄°₂,₂°₂ 2.7 Hz, 1H, H-4°), 3.66 (dd, J₅°₁,₅°₂ 13.2 Hz, J₄°₂,₂°₂ 3.3 Hz, 1H, H-5°a), 3.48 (dd, J₅°₁,₅°₂ 13.2 Hz, J₄°₂,₂°₂ 5.1 Hz, 1H, H-5°b), 2.15 (s, 3H, CH₃), 2.09 (s, 3H, CH₃); ¹⁹F NMR (282 MHz, CD₃OD) δ –117.6 (ddd, J₂₃₉.₇ Hz, 14.1 Hz, 12.1 Hz, 1F), –123.0 (ddd, J₂₃₉.₉ Hz, 10.2 Hz, 5.9 Hz, 1F); ¹³C NMR (75.5 MHz, CD₃OD) δ 171.6 (COCH₃), 171.3 (COCH₃), 165.8 (C-4), 152.3 (C-2), 141.4 (C-6), 124.0 (t, J₁₃₅,₂₅ 257.2 Hz, C-3°), 107.0 (C-1°°), 104.0 (C-5°), 87.1 (d, J₁₃₅,₂₅ 17.7 Hz, C-1°), 80.9 (C-4°°), 80.2 (dd, J₅₃₅,₂₅ 33.5 Hz, 27.6 Hz, C-4°), 75.7 (C-2°°), 75.0 (dd, J₅₃₅,₂₅ 25.0 Hz, 17.1 Hz, C-2°), 72.4 (C-3°°), 66.4 (dd, J₅₃₅,₂₅ 8.4 Hz, 7.4 Hz, C-5°), 53.6 (C-5°°), 20.4 (COCH₃), 20.3 (COCH₃); MS (ESI) m/z 506.1 (M+H⁺), 523.2 (M+NH₄⁺), 528.2 (M+Na⁺); IR (thin film) 3406 (OH), 2938 (CH), 2107 (N=O), 1755 (C=O), 1684 (C=O), 1464, 1247, 1054 cm⁻¹; HRMS (ESI) calcd for C₁₈H₂₁F₂N₅O₁₀Na 528.1149, found 528.1149.

5-O-[5’’-Azido-5’’-deoxy-β-D-ribofuranosyl]-3-deoxy-3,3-O-difluoro-1-(uracil-1’-yl)-β-D-arabinofuranose (11). Compound 10 (110 mg, 0.20 mmol) was dissolved
in saturated methanolic ammonia (40 mL) and methanol (20 mL). The resulting reaction mixture was stirred for 12 h. After removal of the volatile materials, the residue was purified by silica gel chromatography (dichloromethane: methanol = 10:1) to give compound 11 (80 mg, 95% yield) as a white solid: mp 65-68 °C; [α]21D = −4.4 (c 0.66, CH3OH); 1H NMR (300 MHz, CD3OD) δ 7.80 (d, J5,6 8.1 Hz, 1H, H-6), 5.95 (d, J1,F 6.9 Hz, 1H, H-1’), 5.79 (d, J5,6 8.1 Hz, 1H, H-5), 4.93 (s, 1H, H-1”), 4.37-4.27 (m, 2H, H-2’ and H-5’a), 4.10-4.08 (m, 3H, H-5’b, H-3’” and H-4’”), 3.95 (d, J2” 3.6 Hz, 1H, H-2”), 3.68 (dd, J4’,5’a 11.1 Hz, J4’,5’b 3.6 Hz, 1H, H-4’), 3.55 (dd, J5”a, 5”b 13.5 Hz, J4”,5”a 2.7 Hz, 1H, H-5”a), 3.34 (dd, J5”a, 5”b 13.5 Hz, J4”,5”b 5.1 Hz, 1H, H-5”b); 19F NMR (282 MHz, CD3OD) δ −118.4 (dt, J241.7 Hz, 11.6 Hz, 1F), −123.2 (dt, J239.7 Hz, 9.6 Hz, 1F); 13C NMR (75.5 MHz, CD3OD) δ 165.7 (C-4), 152.2 (C-2), 141.6 (C-6), 124.0 (dd, JC1,F 257.3 Hz, 254.4 Hz, C-3’), 109.5 (C-1”), 103.6 (C-5), 87.4 (d, JC1,F 10.0 Hz, C-1’), 82.9 (C-4”), 80.4 (dd, JC4,F 29.6 Hz, 24.5 Hz, C-4’), 76.1 (C-2”), 75.2 (dd, JC2,F 31.1 Hz, 18.3 Hz, C-2’), 72.7 (C-3”), 66.2 (t, JC5,F 3.6 Hz, C-5’), 54.1 (C-5”); MS (ESI) m/z 422.0 (M+H+), 439.0 (M+NH4+), 443.9 (M+Na+); IR (thin film) 3343 (OH), 2936 (CH), 2106 (N3), 1700 (C=O), 1466, 1284, 1137, 1045 cm⁻¹; HRMS (ESI) calcd for C14H17F2N5O8Na 444.0937, found 444.0929.

5-O-5’”-Deoxy-β-D-ribofuranosyl]-3-deoxy-3,3-O-difluoro-1-(uracil-1’-yl)-β-D-arabinofuranose (3). A suspension of Pd/C (52 mg) and 11 (52 mg, 0.12 mmol) in MeOH (8 mL) was stirred under a hydrogen atmosphere for 30 min. Filtration and removal of the solvent gave the crude product, which was purified by column chromatography on silica gel (dichloromethane: methanol = 1:1) to give compound 3 (40 mg, 82% yield) as a white solid: mp 124-127 °C; [α]23D = −16.9 (c 0.15, CH3OH); 1H NMR (300 MHz, CD3OD) δ 7.77 (d, J5,6 7.8 Hz, 1H, H-6), 5.98 (dd, J1,F 6.3 Hz, J1’,F 1.2 Hz, 1H, H-1’), 5.83 (d, J5,6 7.8 Hz, 1H, H-5), 4.98 (s, 1H, H-1”), 4.43-4.36 (m, 2H, H-2’ and H-5’a), 4.12-3.99 (m, 4H, H-5’b, H-3” and H-4’”), 3.76 (dd, J4’,5’a 11.4 Hz, J4’,5’b 4.5 Hz, 1H, H-4’), 2.98 (dd, J5”a, 5”b 13.2 Hz, J4”,5”a 3.0 Hz, 1H, H-5”a), 2.78 (dd, J5”a, 5”b 13.2 Hz, J4”,5”b 7.2 Hz, 1H, H-5”b); 19F NMR
(282 MHz, CD$_3$OD) $\delta$ –118.8 (ddd, $J_{C3,F}$ 258.2 Hz, 255.9 Hz, C-3'), 109.3 (C-1''), 103.7 (C-5), 87.8 (d, $J_{C1,F}$ 9.8 Hz, C-1'), 84.6 (C-4''), 80.2 (dd, $J_{C4,F}$ 28.9 Hz, 24.1 Hz, C-4''), 76.3 (C-2''), 75.2 (dd, $J_{C2,F}$ 25.7 Hz, 15.9 Hz, C-2''), 73.8 (C-3''), 65.8 (t, $J_{C5,F}$ 4.8 Hz, C-5'), 45.9 (C-5''); MS (ESI) m/z 396.0 (M+H$^+$); IR (thin film) 3308 (OH, NH$_2$), 2926 (CH), 1699 (C=O), 1488, 1455, 1290, 1116, 1033 cm$^{-1}$; HRMS (ESI) calcd for C$_{14}$H$_{20}$F$_2$N$_3$O$_8$ 396.1213, found 396.1221.

(2$R$,3$R$)-3-O-Benzyl-4,4-difluoro-5-hexene-1,2,3-triol (14). To a solution of compound 13 (3.600 g, 12.06 mmol) in MeOH (36 mL) was added p-toluenesulfonic acid hydrate (2.200 g, 11.56 mmol). The reaction mixture was stirred overnight at room temperature, then was quenched with solid K$_2$CO$_3$ (1.700 g, 12.32 mmol). The reaction mixture was diluted with 150 mL of EtOAc and washed three times with 150 mL of brine, dried over anhydrous Na$_2$SO$_4$. After filtration and removal of the solvent in vacuo, the residue was purified by flash chromatography on silica gel (petroleum ether: ethyl acetate = 1:1) to afford compound 14 (2.832 g, 91% yield) as a clear oil: $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 7.40-7.30 (m, 5H, H$_{aryl}$-Bn), 6.15-5.97 (m, 1H, H-5), 5.76 (d, $J_{5,6a}$ 17.4 Hz, 1H, H-6a), 5.55 (d, $J_{5,6b}$ 11.1 Hz, 1H, H-6b), 4.92-4.82 (m, 1H, H$_{alkyl}$-Bn), 4.68-4.60 (m, 1H, H$_{alkyl}$-Bn), 3.86-3.56 (m, 4H, H-1a, H-1b, H-2, H-3), 2.37 (s, 2H, OH); $^{19}$F NMR (282 MHz, CDCl$_3$) $\delta$ –101.2 (dt, $J$ 254.1 Hz, 14.1 Hz, 0.04F), –102.3 (dt, $J$ 254.9 Hz, 11.0 Hz, 0.96F), –105.7 (ddd, $J$ 259.4 Hz, 11.0 Hz, 5.9 Hz, 0.04F), –107.1 (dt, $J$ 254.9 Hz, 10.7 Hz, 0.96F); MS (ESI) m/z 259.1 (M+H$^+$), 276.1 (M+NH$_4^+$), 281.1 (M+Na$^+$); IR (thin film) 3394 (OH), 3025 (C=H), 1716, 1498, 1421, 1212, 1102, 989 cm$^{-1}$; Anal. Calcd for C$_{13}$H$_{16}$F$_2$O$_3$: C, 60.46; H, 6.24; Found: C, 60.29; H, 6.20.

(2$R$,3$R$)-1-O-Benzoyl-3-O-benzyl-4,4-difluoro-5-hexene-1,2,3-triol (15). To a solution of compound 14 (2.648 g, 10.26 mmol) in anhydrous CH$_2$Cl$_2$ (50 mL) and
pyridine (25 mL) was slowly added a solution of BzCl (1.24 mL, 10.26 mmol) in CH$_2$Cl$_2$ (9 mL) at -78 °C. After the mixture was stirred at the same temperature for 2 h, MeOH (10 mL) was added and the mixture was stirred for 30 min. Then water was added to quench the reaction. The layers were separated and the aqueous layer was extracted with ether. The combined organic layers were washed with 1 N HCl, saturated aqueous NaHCO$_3$ and brine. After the resultant solution was dried over anhydrous Na$_2$SO$_4$, and filtered, the solvent was removed in vacuo. The residue was purified by silica gel column chromatography (petroleum ether: ethyl acetate = 4: 1) to give compound 15 (3.195 g, 86% yield) as a clear oil: $^1$H NMR (300 MHz, CDCl$_3$) δ 8.04-7.95 (m, 2H, H$_{aryl}$-Bz), 7.58 (t, $J$ 7.5 Hz, 1H, H$_{aryl}$-Bz), 7.44 (t, $J$ 7.5 Hz, 2H, H$_{aryl}$-Bz), 7.35-7.29 (m, 5H, H$_{aryl}$-Bn), 6.20-6.03 (m, 1H, H-5), 5.79 (d, $J_{5,6a}$ 17.4 Hz, 1H, H-6a), 5.56 (d, $J_{5,6b}$ 11.1 Hz, 1H, H-6b), 4.95-4.85 (m, 1H, H$_{alkyl}$-Bn), 4.69 (m, 1H, H$_{alkyl}$-Bn), 4.60 (dd, $J_{1a,1b}$ 11.7 Hz, $J_{1a,2}$ 2.4 Hz, 1H, H-1a), 4.48 (dd, $J_{1a,1b}$ 11.7 Hz, $J_{1b,2}$ 6.3 Hz, 1H, H-1b), 4.20-4.10 (m, 1H, H-2), 3.92-3.84 (m, 1H, H-3), 2.44 (s, 1H, OH); $^{19}$F NMR (282 MHz, CDCl$_3$) δ –101.3 (dt, $J$ 255.8 Hz, 11.3 Hz, 0.06F), –101.7 (dt, $J$ 256.9 Hz, 9.0 Hz, 0.94F), –104.2 (dt, $J$ 257.2 Hz, 8.2 Hz, 0.06F), –106.6 (dt, $J$ 258.3 Hz, 9.0 Hz, 0.94F); MS (ESI) m/z 363.1 (M+H$^+$), 380.2 (M+NH$_4^+$), 385.0 (M+Na$^+$); IR (thin film) 3474 (OH), 3035 (=CH), 1721 (C=O), 1498, 1421, 1276, 1114, 990 cm$^{-1}$; Anal. Calcd for C$_{20}$H$_{20}$F$_2$O$_4$: C, 66.29; H, 5.56; Found: C, 66.44; H, 5.64.

5-O-Benzoyl-3-O-benzyl-2-deoxy-2,2-difluoro-\textit{\textit{D}}-arabinofuranose (7). Ozone was bubbled through a solution of compound 15 (2.864 g, 7.91 mmol) in dichloromethane (60 mL) at -78 °C. The reaction was monitored by TLC, then the solvent was removed in vacuo. The residue was purified by silica gel column chromatography (chloroform: methanol = 60: 1) to give compound 7 (2.477 g, 86% yield) as a clear oil: $^1$H NMR (300 MHz, CDCl$_3$) δ 7.98-7.94 (m, 2H, H$_{aryl}$-Bz), 7.61-7.55 (m, 1H, H$_{aryl}$-Bz), 7.46-7.40 (m, 2H, H$_{aryl}$-Bz), 7.33-7.27 (m, 5H, H$_{aryl}$-Bn), 5.32 (d, $J_{1,F}$ 6.6 Hz, 0.55H, H-1), 5.24 (d, $J_{1,F}$ 6.9 Hz, 0.45H, H-1), 4.89 (d, $J$ 11.7 Hz, 1H, H$_{alkyl}$-Bn), 4.62 (d, $J$ 11.4 Hz, 1H, H$_{alkyl}$-Bn), 4.60-4.24 (m, 4H, H-3, H-4, H-5a
and H-5b), 4.03-3.96 (m, 1H, OH); \(^{19}\)F NMR (282 MHz, CDCl\(_3\)) \(\delta \) –109.5 (ddd, \(J\) 247.9 Hz, 14.4 Hz, 9.3 Hz, 0.56F), –121.9 (dd, \(J\) 238.6 Hz, 5.6 Hz, 0.44F), –127.1 (ddd, \(J\) 240.3 Hz, 14.9 Hz, 6.8 Hz, 0.44F), –128.6 (d, \(J\) 247.0 Hz, 0.56F); MS (ESI) \(m/z\) 365.1 (M+H\(^+\)), 382.2 (M+NH\(_4^+\)), 387.0 (M+Na\(^+\)), 403.0 (M+K\(^+\)); IR (thin film) 3437 (OH), 2951 (CH), 1724 (C=O), 1497, 1454, 1275, 1097, 1028 cm\(^{-1}\); Anal. Calcd for C\(_{19}\)H\(_{18}\)F\(_2\)O\(_5\): C, 62.63; H, 4.98; Found: C, 62.24; H, 5.02.

**Ethanimidic acid,**

\(N\)-(5"]-benzoyl-3"]-O-benzyl-2"]-deoxy-2"]',2"]'-difluoro-D-ribofuranosyl)-2,3-O-isopropylidene1-(uracil-1"]'-yl)-\(\beta\)-D-arabinofuranosyl ester (16).** A solution of compound 7 (730 mg, 2.00 mmol), CCl\(_3\)CN (1.00 mL, 10.00 mmol) and DBU (1 drop) in CH\(_2\)Cl\(_2\) (20 mL) was stirred at room temperature for 1.5 h. Then solvent was removed, and the residue was purified by flash chromatography on a silica gel column (petroleum ether: ethyl acetate = 5: 1, with 1% Et\(_3\)N) to give the corresponding trichloroacetimidate derivative (880 mg, 87% yield), which was used in the next step without further characterization. To a suspension of the trichloroacetimidate derivative (880 mg, 1.73 mmol), compound 8 (580 mg, 2.08 mmol) and 4 Å MS (1380 mg) in dry CH\(_3\)CN (20 mL) at -20 \(^\circ\)C, was slowly added TMSOTf (0.43 mL, 1.90 mmol). After being stirred at the same temperature for 6 h, the reaction was quenched with Et\(_3\)N (3 mL), filtered, and concentrated. The residue was purified by silica gel column chromatography (petroleum ether: ethyl acetate = 1: 2) to give the compound 16 (720 mg, 62% yield) as a white solid: \(^1\)H NMR (300 MHz, CDCl\(_3\)) \(\delta \) 8.79 (s, 0.45H, NH), 8.49 (s, 0.55H, NH), 8.02-7.97 (m, 2H, H\(_{\text{aryl}}\)-Bz), 7.60-7.55 (m, 1H, H\(_{\text{aryl}}\)-Bz), 7.45-7.41 (m, 2H, H\(_{\text{aryl}}\)-Bz), 7.33-7.30 (m, 6H, H\(_{\text{aryl}}\)-Bn and H-6), 5.75-5.63 (m, 2H, H-1"]’ and H-5), 5.29 (d, \(J_{1",\text{p}}\) 10.2 Hz, 0.55H, H-1"]’’), 5.16 (d, \(J_{1",\text{p}}\) 10.8 Hz, 0.45H, H-1"]’’), 4.94-3.97 (m, 11H, H-2’, H-3’, H-4’, H-5’a, H-5’b, H-3”, H\(_{\text{alkyl}}\)-Bn, H-4”, H-5”a and H-5”b), 1.97 (s, 3H, CH\(_3\)), 1.57 (s, 3H, CH\(_3\)), 1.35 (s, 3H, CH\(_3\)); \(^{19}\)F NMR (282 MHz, CDCl\(_3\)) \(\delta \) –101.1 (ddd, \(J\) 237.7 Hz, 17.2 Hz, 10.4 Hz, 0.55F), –119.3 (dd, \(J\) 231.8 Hz, 9.3 Hz, 0.45F), –121.9 (dd, \(J\) 236.6 Hz, 4.5 Hz, 0.55F),
Conversion of Compound 7 (728 mg, 2.00 mmol) to Compound 17 was accomplished using the similar condition as described for compound 16 with CH₃NO₂ as the solvent instead of CH₃CN. Compound 17 (403 mg, 32% in two steps) was obtained as a white solid: 

\[ \text{1H NMR (300 MHz, CDCl₃) } \delta 8.47 (s, 0.28H, NH), 8.37 (s, 0.72H, NH), 8.00-7.93 (m, 2H, H ary-Bz), 7.64-7.47 (m, 1H, H ary-Bz), 7.45-7.37 (m, 2H, H ary-Bz), 7.34-7.28 (m, 6H, H ary-Bn and H-6), 6.07 (d, J_I'-3 3.0 Hz, 0.72H, H-1'), 5.75-5.56 (m, 1H, H-1' and H-5), 5.57 (d, J_3,5 8.1 Hz, 0.28H, H-5), 5.08 (d, J_1,5 1.5 Hz, 0.72H, H-1''), 4.98 (d, J_1,5 3.0 Hz, 0.28H, H-1''), 4.89-3.96 (m, 10H, H-2', H-3', H-4', H-5'a, H-5'b, H-3'', H alkyl-Bn, H-4'' and H-5''a), 3.83-3.70 (m, 1H, H-5''b), 1.58 (s, 2.16H, CH₃), 1.50 (s, 0.84H, CH₃), 1.31 (s, 0.85H, CH₃), 1.30 (s, 2.15H, CH₃); \n
\[ \text{19F NMR (282 MHz, CDCl₃) } \delta -105.7 (ddd, J 249.3 Hz, 17.2 Hz, 8.5 Hz, 0.72F), -121.2 (dd, J 239.4 Hz, 7.9 Hz, 0.28F), -125.3 (ddd, J 239.7 Hz, 16.1 Hz, 8.5 Hz, 0.28F), -127.4 (d, J 251.5 Hz, 0.72F); \n
\[ \text{MS (ESI) } m/z 631.3 (M+H⁺), 653.2 (M+Na⁺); \n
\[ \text{IR (thin film) 2937 (CH), 1720 (C=O), 1695 (C=O), 1456, 1384, 1275, 1112, 713 cm⁻¹; } \n
\[ \text{Anal. Calcd for C₃₁H₃₀F₂N₂O₁₀: } C, 59.04; H, 5.11; N, 4.44; \text{ Found: C, 58.90; H, 5.20; N, 4.37.} \n
5-O-[5''-Benzoyl-3''-O-benzyl-2''-deoxy-2'',2''-difluoro-β-D-ribofuranosyl]-2,3-O-isopropylidene-1-(uracil-1'-yl)-β-D-arabinofuranose (17). Compound 17 (277 mg, 0.44 mmol) was dissolved in saturated methanolic ammonia (33 mL) and methanol (11 mL). The resulting reaction mixture was stirred for 36 h. After removal of the volatile materials, the residue was purified by silica gel chromatography (petroleum ether: ethyl acetate = 1: 1) to give compound 18 (204 mg, 88 % yield) as a white solid: 

\[ \text{1H NMR (300 MHz, CDCl₃) } \delta 8.47 (s, 0.28H, NH), 8.37 (s, 0.72H, NH), 8.00-7.93 (m, 2H, H ary-Bz), 7.64-7.47 (m, 1H, H ary-Bz), 7.45-7.37 (m, 2H, H ary-Bz), 7.34-7.28 (m, 6H, H ary-Bn and H-6), 6.07 (d, J_I'-3 3.0 Hz, 0.72H, H-1'), 5.75-5.56 (m, 1H, H-1' and H-5), 5.57 (d, J_3,5 8.1 Hz, 0.28H, H-5), 5.08 (d, J_1,5 1.5 Hz, 0.72H, H-1''), 4.98 (d, J_1,5 3.0 Hz, 0.28H, H-1''), 4.89-3.96 (m, 10H, H-2', H-3', H-4', H-5'a, H-5'b, H-3'', H alkyl-Bn, H-4'' and H-5''a), 3.83-3.70 (m, 1H, H-5''b), 1.58 (s, 2.16H, CH₃), 1.50 (s, 0.84H, CH₃), 1.31 (s, 0.85H, CH₃), 1.30 (s, 2.15H, CH₃); \n
\[ \text{19F NMR (282 MHz, CDCl₃) } \delta -105.7 (ddd, J 249.3 Hz, 17.2 Hz, 8.5 Hz, 0.72F), -121.2 (dd, J 239.4 Hz, 7.9 Hz, 0.28F), -125.3 (ddd, J 239.7 Hz, 16.1 Hz, 8.5 Hz, 0.28F), -127.4 (d, J 251.5 Hz, 0.72F); \n
\[ \text{MS (ESI) } m/z 631.3 (M+H⁺), 653.2 (M+Na⁺); \n
\[ \text{IR (thin film) 2937 (CH), 1720 (C=O), 1695 (C=O), 1456, 1384, 1275, 1112, 713 cm⁻¹; } \n
\[ \text{Anal. Calcd for C₃₁H₃₀F₂N₂O₁₀: } C, 59.04; H, 5.11; N, 4.44; \text{ Found: C, 58.90; H, 5.20; N, 4.37.} \n
5-O-[3''-O-Benzyl-2''-deoxy-2'',2''-difluoro-β-D-ribofuranosyl]-2,3-O-isopropylidene-1-(uracil-1'-yl)-β-D-arabinofuranose (18).
NMR (300 MHz, CDCl₃) δ 8.99 (s, 0.28H, NH), 8.66 (s, 0.72H, NH), 7.62 (d, J₅,₆ 7.8 Hz, 0.72H, H-6), 7.36-7.28 (m, 5.28H, Haryl-Bn and H-6), 6.06 (d, J₁,₂ 2.7 Hz, 0.72H, H-1’), 5.73-5.66 (m, 1.28H, H-1’ and H-5), 5.08-3.76 (m, 11H, H-2’, H-3’, H-4’, H-5’a, H-5’b, H-1”, H-3”, H₃kyt-Bn, H-4” and H-5”a), 3.67 (dd, J₅₄,₅₅ 12.3 Hz, J₄’,₅’b 4.5 Hz, 0.72H, H-5”b), 3.55 (dd, J₅₄,₅₅ 12.9 Hz, J₄’,₅’b 3.0 Hz, 0.28H, H-5”b), 2.08 (s, 1H, OH), 1.57 (s, 3H, CH₃), 1.36 (s, 0.85H, CH₃), 1.30 (s, 2.15H, CH₃); ¹⁹F NMR (282 MHz, CDCl₃) δ –105.9 (ddd, J 250.1 Hz, 17.8 Hz, 8.2 Hz, 0.72F), –120.6 (ddd, J 237.2 Hz, 16.6 Hz, 9.9 Hz, 0.28F), –127.0 (d, J 249.6 Hz, 0.72F); MS (ESI) m/z 527.2 (M+H +), 549.2 (M+Na +); IR (thin film) 3231 (OH), 2937 (CH), 1693 (C=O), 1458, 1384, 1274, 1084, 701 cm⁻¹; Anal. Calcd for C₂₄H₂₈F₂N₂O₉: C, 54.75; H, 5.36; N, 5.32; Found: C, 54.86; H, 5.04; N, 5.36.

5-O-[5’'-Azido-3’'-O-benzyl-2’',5''-dideoxy-2’',2’'-difluoro-β-D-ribofuranosyl]-2,3-O-isopropylidene-1-(uracil-1’'-yl)-β-D-arabinofuranose (19). Compound 18 (126 mg, 0.24 mmol) was dissolved in dry CH₂Cl₂ (1 mL). Then, fresh distilled pyridine (0.042 mL, 0.48 mmol) was added. After the resulting mixture was cooled to –55 °C, a solution of trifluoromethanesulfonic anhydride (0.065 mL, 0.40 mmol) in dry CH₂Cl₂ (0.4 mL) was added dropwise to the mixture with stirring. Then, the reaction mixture was stirred for about 2 h at about –30 °C. Water and NaHCO₃ solution were successively added after the mixture was warmed to the room temperature. The mixture was extracted with CH₂Cl₂, dried over anhydrous Na₂SO₄ and purified by silica gel column chromatography (petroleum ether: ethyl acetate = 2: 1) to afford a clear oil (135 mg). A solution of the product in acetone (30 mL) was cooled to 0 °C by ice bathe. Then, sodium azide (78 mg, 1.2 mmol) was added carefully with stirring. The reaction mixture was stirred at room temperature for 1h. Water was added to quench the reaction. The combined organic layer was washed with brine and dried over anhydrous Na₂SO₄, and concentrated in vacuo. The residue was quickly purified by silica gel column chromatography (petroleum ether: ethyl acetate = 2: 1) to afford the compound 19 (77 mg, 58% yield in two steps) as a white solid: ¹H NMR (300
MHz, CDCl$_3$) $\delta$ 8.58 (s, 0.28H, NH), 8.46 (s, 0.72H, NH), 7.59 (d, $J_{5,6}$ 8.1 Hz, 0.72H, H-6), 7.42-7.30 (m, 5.28H, H$_{aryl}$-Bn and H-6), 6.06 (d, $J_{1',2'}$ 3.0 Hz, 0.72H, H-1'), 5.82 (d, $J_{1',2'}$ 2.1 Hz, 0.28H, H-1'), 5.72 (d, $J_{5,6}$ 7.8 Hz, 0.72H, H-5), 5.64 (d, $J_{5,6}$ 9.6 Hz, 0.28H, H-5), 5.07 (d, $J_{1''', F}$ 7.8 Hz, 0.72H, H-1'''), 4.97 (d, $J_{1''', F}$ 8.1 Hz, 0.28H, H-1'''), 4.88-3.74 (m, 9H, H-2', H-3', H-4', H-5'a, H-5'b, H-3'', H$_{alkyl}$-Bn and H-4''), 3.50-3.32 (m, 1H, H-5''a), 3.31-3.25 (m, 1H, H-5''b), 1.58 (s, 3H, CH$_3$), 1.36 (s, 0.85H, CH$_3$), 1.31 (s, 2.15H, CH$_3$); 19F NMR (282 MHz, CDCl$_3$) $\delta$ –105.7 (ddd, $J$ 251.8 Hz, 15.2 Hz, 7.9 Hz, 0.72F), –120.5 (dd, $J$ 238.8 Hz, 6.5 Hz, 0.28F), –124.6 (ddd, $J$ 239.4 Hz, 16.1 Hz, 7.3 Hz, 0.28F), –126.9 (d, $J$ 250.4 Hz, 0.72F); MS (ESI) m/z 552.2 (M+H$^+$), 574.3 (M+Na$^+$); IR (thin film) 2938 (CH), 2107 (N 3), 1695 (C=O), 1458, 1384, 1277, 1087, 700 cm$^{-1}$; Anal. Calcd for C$_{24}$H$_{27}$F$_2$N$_5$O$_8$: C, 52.27; H, 4.93; N, 12.70; Found: C, 52.27; H, 4.69; N, 12.76.

5-O-[5"-Amino-2"',5"'-dideoxy-2"',2"'-difluoro-β-D-ribofuranosyl]-1-(uracil-1'yyl) -β-D-arabinofuranose (4). Compound 19 (64 mg, 0.12 mmol) was dissolved in dry CH$_2$Cl$_2$ (6 mL). After the resulting mixture was cooled to –60 ºC, BCl$_3$ (1.20 mL, 1.0 M in heptane, 1.20 mmol) was added dropwise to the mixture with stirring. The mixture was then warmed to the room temperature and stirred overnight. The reaction was quenched with MeOH (3 mL), and concentrated. The residue was purified by silica gel column chromatography (dichloromethane: methanol = 5: 1) to give the compound 4 (21 mg, 45% yield) and the α isomer (8 mg, 18% yield) as white solids: compound 4, mp 248-250 ºC; [α]$^{$27}$D = +22.0 ($c$ 0.12, CH$_3$OH); $^1$H NMR (300 MHz, CD$_3$OD) $\delta$ 8.01 (d, $J_{5,6}$ 8.1 Hz, 1H, H-6), 5.98 (d, $J_{1',2'}$ 4.2 Hz, 1H, H-1'), 5.68 (d, $J_{5,6}$ 8.1 Hz, 1H, H-5), 5.15 (d, $J_{1''', F}$ 7.5 Hz, 1H, H-1'''), 4.23-4.18 (m, 3H, H-2', H-3' and H-4'), 4.13-4.08 (m, 1H, H-4'''), 4.00 (d, $J_{5,6}$ 9.8 Hz, 1H, H-5'a), 3.93 (dd, $J_{3'',5''}$ 18.3 Hz, $J_{3'',4''}$ 5.7 Hz, 1H, H-3''), 3.89 (dd, $J_{5'',6''}$ 21.0 Hz, $J_{4'',5''}$ 2.1 Hz, 1H, H-5'b), 3.07 (dd, $J_{5'',6''}$ 13.8 Hz, $J_{4'',5''}$ 3.9 Hz, 1H, H-5''a), 2.93 (dd, $J_{5'',6''}$ 13.5 Hz, $J_{4'',5''}$ 7.5 Hz, 1H, H-5''b); $^{19}$F NMR (282 MHz, CD$_3$OD) $\delta$ -109.7 (ddd, $J$ 247.0 Hz, 17.8 Hz, 8.5 Hz, 1F), -128.4 (d, $J$ 247.0 Hz, 1F); $^{13}$C NMR (75.5 MHz, CD$_3$OD) $\delta$ 167.1 (C-4),
153.1 (C-2), 143.0 (C-6), 124.0 (dd, $J_{C2''} = 202.2$ Hz, $184.8$ Hz, C-2''), 103.1 (C-5), 102.3 (dd, $J_{C1''} = 32.5$ Hz, $16.3$ Hz, C-1''), 90.5 (C-1'), [84.9, 84.8 (C-4', C-4'')], 76.0 (C-2'), 73.8 (dd, $J_{C3''} = 24.0$ Hz, $14.7$ Hz, C-3''), 72.0 (C-3'), 67.7 (C-5'), 44.1 (C-5''); MS (ESI) $m/z$ 396.2 (M+H$^+$), 418.0 (M+Na$^+$); IR (thin film) 3312 (OH, NH$_2$), 2926 (CH), 1684 (C=O), 1508, 1465, 1266, 1221, 1033 cm$^{-1}$; HRMS (ESI) calcd for C$_{14}$H$_{20}$F$_2$N$_3$O$_8$ 396.1213, found 396.1217.
COSY NMR spectrum of compound 4
NOESY NMR spectrum of compound 4
COSY NMR spectrum of compound 11
NOESY NMR spectrum of compound 11