Synthesis of 4-Substituted-1,2,3-Triazole Carbanucleoside Analogues of Ribavirin via Click Chemistry.

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General Experimental Details

All chemicals used were of reagent grade and were obtained from Aldrich Chemical Co and used without further purification. All air-sensitive reactions were carried out under argon. Flash chromatography was performed on silica gel (Merck 60, 230-240 mesh) and analytical TLC on pre-coated silica gel plates (Merck 60 F_{254}, 0.25 mm). Melting points were measured in a Reichert Kofler Thermopan and are uncorrected. Infrared spectra were recorded in a Perkin-Elmer 1640 FTIR spectrophotometer. ^{1}H and ^{13}C NMR spectra were recorded in a Bruker AMX 300

spectrometer at 300 and 75.47 MHz, respectively, using TMS as internal standard (chemical shifts in δ values, *J* in Hz). Mass spectra were recorded on a Hewlett-Packard HP5988A or a Micromass Autospec spectrometers. Microanalyses were performed in a FISONS EA 1108 Elemental Analyser at the University of Santiago Microanalysis Service; all results shown are within ± 0.4% of the theorethaical values. X-ray diffraction data were collected in an BRUKER Smart-CCD-1000 automatic diffractometer.

***tert*-Butyldiphenylsilyl (cyclopent-3-enyl)methyl ether (14).** A mixture of **12** (4.22 g, 43.06 mmol), imidazole (6.44 g, 94.59 mmol) and TBDPSCl (7.21 g, 47.76 mmol) in CH_2Cl_2 (75 mL) was stirred under Ar atmosphere at room temperature for 1.5 hours. The crude reaction mixture was then transferred to an extraction funnel, diluted with CH_2Cl_2 (250 mL) and washed with sat. $NaHCO_3$ solution (3 × 125 mL). The aqueous layer was then extracted with more CH_2Cl_2 (3 × 125 mL) and the combined organic layer washed with sat. NH_4Cl solution (3 × 125 mL). After that, the halogenated extract was dried (Na_2SO_4) and evaporated under reduced pressure affording **14** (8.72 g, 98%) as a yellowish oil. An analytical sample was obtained by column chromatography on silica gel using hexane-EtOAc 100:1 as eluent.

14 : Colourless oil; ν_{max}/cm^{-1} 3070, 3051, 2930, 2874, 2856, 2351, 1590, 1472, 1427, 1389, 1112, 1030, 1002, 739, 608, 590; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.07 (9H, s, $C(CH_3)_3$), 2.12-2.18 (2H, m), 2.40-2.49 (2H, m), 2.53-2.56 (1H, m), 3.58 (2H, d, *J* 6.8, OCH_2), 5.65 (2H, virtual s), 7.36-7.45 (6H, m), 7.67-7.72 (4H, m) ppm; ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 19.7 (C), 27.3 ($C(CH_3)_3$), 35.9 (CH_2), 39.8 (CH), 68.1 (CH_2), 128.0 (CH), 129.9 (CH), 130.0 (CH), 134.6 (C), 135.2 (CH), 136.0 (CH) ppm; HRMS *m/z* calcd for $C_{22}H_{28}IN_3OSi$, 336.1909; found, 336.1936.

(±)-(*c*-3-Azido-*t*-4-iodo-*r*-1-cyclopentyl)methyl *tert*-butyldiphenylsilyl ether (15). To a suspension of NaN_3 (0.97 g, 1.49 mmol) in dry CH_3CN (1 mL) under Ar flux at -10ºC, was added a solution of ICl (0.11 g, 0.67 mmol) in dry CH_3CN (1 mL). That was stirred for 10 min, and then, a solution of 14 (0.2 g, 0.59 mmol) in dry CH_3CN (2 mL) was added dropwise. The mixture was allowed to reach room temperature and stirred for 5 hours. The reaction mixture was then poured into H_2O (20 mL) and extracted with

diethyl ether (3 x 40 mL). The organic extract was successively washed with 60% $Na_2S_2O_3$ (30 mL), H_2O (2 × 20 mL) and brine (20 mL), dried (Na_2SO_4) and evaporated under reduced pressure. The crude product (0.21 g), was purified by column chromatography (silica gel, hexane as eluent). The nonvoid fractions afforded 15 as a yellowish oil (0.16 g, 55%). An analytical sample was obtained after a second purification of a small portion of the above-mentioned material by column cromatography on silicagel using hexane as eluent.

15: colourless sticky oil; ν_{max}/cm^{-1} 3069, 2929, 2857, 2100, 1466, 1428, 1256, 1109, 1005, 798, 701; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.06 (9H, s, $C(CH_3)_3$), 1.44-1.53 (1H, m, 5-H*H*), 2.15-2.19 (2H, m, 2-H*H* + 5-*H*H), 2.20-2.25 (1H, m, 2-*H*H), 2.46-2.49 (1H, m, 1-H), 3.58 (2H, d, *J* 5.9, OCH_2), 4.06-4.13 (2H, m, 3-H + 4-H), 7.36-7.46 (6H, m), 7.62-7.66 (4H, m) ppm; ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 19.27 (C), 26.8 ($C(CH_3)_3$), 27.7 (CH), 32.5 (CH_2), 39.0 (CH), 39.5 (CH_2), 66.3 (CH_2), 71.4 (CH), 127.7 (CH), 129.7 (CH), 133.5 (C), 135.6 (CH) ppm; *m/z* (EI 70eV) 448 (M-*tert*-Bu, 7%), 405 (25), 309 (16), 293 (17), 224 (17), 207 (20), 199 (42), 183 (25), 181 (27), 105 (24), 80 (30), 79 (100), 77 (18), 57 (16); HRMS *m/z* calcd for $C_{22}H_{28}IN_3OSi$, 505.1046; found, 505.1068.

(±)-*cis*-(4-Azidocyclopent-2-enyl)methyl *tert*-butyldiphenylsilyl ether (17). A mixture of **15** (100 mg, 0.198 mmol) and DABCO (44 mg, 0.40 mmol) in benzene (2 mL) was stirred under reflux for 42 hours. After that, more DABCO (22 mg, 0.20 mmol) was added and the reflux kept for 13 hours until the reaction was considered completed by TLC analysis. The solvent was then evaporated under reduced pressure, and the resulting brownish residue (120 mg) was purified by chromatography (silica gel, hexane-EtOAc 30:1 as eluent) and concentration of the nonvoid fractions to dryness afforded **17** (32 mg, 43%).

17: yellowish oil, ν_{max}/cm^{-1} 2959, 2270, 2094, 1728, 1428, 1259, 1174, 1107, 808, 702; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.07 (9H, s, $C(CH_3)_3$), 1.58 (1H, dt, *J* 14.1 and 5.3, 5-H*H*), 2.41 (1H, dt, *J* 14.1 and 8.2, 5-*H*H), 2.89-2.94 (1H, m, 4-H), 3.56-3.67 (2H, m, OCH_2), 4.37-4.41 (1H, m, 1-H), 5.79 (1H, dt, *J* 5.6 and 2.0, 2-H), 6.04 (1H, dt, *J* 5.6 and 1.8, 3-H), 7.36-7.46 (6H, m), 7.63-7.73 (4H, m) ppm; ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 19.6 (C), 26.8 ($C(CH_3)_3$), 32.9 (CH_2), 47.6 (CH), 66.7 (CH), 67.3 (CH_2),

127.7 (CH), 129.7 (CH), 129.7 (CH), 133.9 (C), 135.6 (CH), 137.7 (CH) ppm; *m/z* (EI 70eV) 292 (M-*tert*-Bu, 5%), 278 (5), 277 (21), 224 (56), 199 (57), 191 (20), 135 (30), 105 (38), 91 (24), 80 (18), 79 (100), 78 (18), 77 (49); HRMS *m/z* calcd for $C_{22}H_{27}N_3OSi$, 377.1923; found, 377.1948.

Methyl (±)-1-[(*c*-4-hydroxymethyl)-*t*-2-iodo-*r*-1-cyclopentyl]-1*H*-1,2,3-triazole-5-carboxylate (18) and methyl (±)-1-[(*c*-4-hydroxymethyl)-*t*-2-iodo-*r*-1-cyclopentyl]-1*H*-1,2,3-triazole-4-carboxylate (19). A mixture of **13a** (150 mg, 0.56 mmol) and methyl propiolate (0.26 mL, 2.97 mmol) was stirred at 50ºC for 4 hours. The non-reacted methyl propiolate was removed under reduced pressure and the resulting oily residue was purified by chromatography (silica gel hexane-EtOAc, 2:1 and 1:1 mixtures as successive eluents). Upon concentration to dryness, the combined early nonvoid fractions eluted with the first solvent mixture afforded **18** (27 mg, 14%), and with the second eluent produced **19** (156 mg, 79%).

18: colourless oil, ν_{max}/cm^{-1} 3379, 3062, 2948, 1731, 1530, 1440, 1317, 1259, 771; ^1H NMR (300 MHz, $CDCl_3$) δ 2.02-2.11 (1H, m, 5-H*H*), 2.32-2.41 (1H, m, 5-*H*H), 2.43-2.68 (4H one of them D_2O exch., m, OH), 3.73-3.75 (2H, m, HOC*H*$_2$), 3.95 (3H, s, OCH_3), 4.67 (1H, virtual q, *J* 7.4, 2-H), 5.85 (1H, virtual q, *J* 7.6, 1-H), 8.14 (1H, s, 4-$H_{triazole}$) ppm; ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 25.7 (CH), 33.9 (CH_2), 39.4 (CH), 39.6 (CH_2), 52.7 (CH_3), 65.4 (CH_2), 69.6 (CH), 128.1 (CH), 138.0 (C), 158.9 (C) ppm; *m/z* (FABMS) 352.0 (M+1, 19 %); HRMS *m/z* calcd for $C_{10}H_{14}IN_3O_3$, 351.008; found, 351.0033.

19: colourless oil, ν_{max}/cm^{-1} 3379, 3176, 3136, 2948, 1729, 1546, 1439, 1377, 1223, 1123, 1043, 731; ^1H NMR (300 MHz, $CDCl_3$) δ 2.06-2.11 (1H, D_2O exch., m, OH), 2.20 (1H, dt, *J* 14.0 and 8.1, 5-H*H*), 2.35 (1H, dt, *J* 14.0 and 8.7, 5-*H*H), 2.42-2.52 (2H, m), 2.58-2.62 (1H, m), 3.74-3.76 (2H, m, HOC*H*$_2$), 3.95 (3H, s, OCH_3), 4.47 (1H, virtual q, *J* 7.9, 2-H), 5.05 (1H, virtual q, *J* 8.2, 1-H), 8.30 (1H, s, 5-$H_{triazole}$) ppm; ^{13}C NMR (75.5 MHz, $CDCl_3$) δ 25.6 (CH), 33.6 (CH_2), 38.9 (CH), 39.9 (CH_2), 52.3 (CH_3), 65.0 (CH_2), 71.7 (CH), 127.3 (CH), 139.7 (C), 161.0 (C) ppm; *m/z* (FAB) 352.0 (M+1, 20%); HRMS *m/z* calcd for $C_{10}H_{14}IN_3O_3$, 351.008; found, 351.0103.

(±)-1-[(*c*-4-Hydroxymethyl)-*t*-2-iodo-*r*-1-cyclopentyl]-1*H*-1,2,3-triazole-4-carboxamide (20). A solution of **19** (60 mg, 0.19 mmol) in MeOH (4 mL) and 30% NH_4OH aqueous solution (4 mL) was stirred, and after 3 hours, 30% NH_4OH aqueous solution (4 mL) were added and the stirring kept for 1.5 hours more. The MeOH was then evaporated and the resulting aqueous phase extracted with EtOAc (3 × 10 mL). After that, the organic layer was dried (Na_2SO_4) and evaporated under reduced pressure. The crude product obtained was chromatographed on silica gel using CH_2Cl_2-MeOH 20:1 as eluent, yielding **20** (45 mg, 71%).

20: white solid; mp 162-164ºC (recrystallized from EtOAc/MeOH); ν_{max}/cm^{-1} 3410, 3253, 3183, 2923, 1632, 1414, 1301, 1043; ^{1}H NMR (300 MHz, DMSO*d*$_6$) δ 1.69-1.79 (1H, m), 2.18-2.34 (4H, m), 3.35-3.49 (2H, m, HOC*H*$_2$, this signal is simplified to a duplet (*J* 6.9) when treated with D_2O), 4.55 (1H, virtual q, *J* 9.4, 2-H), 4.74 (1H, t, *J* 5.3, D_2O exch., OH), 5.15 (1H, virtual q, *J* 9.7, 1-H), 7.47 (1H, s, D_2O exch., NH*H*), 7.85 (1H, s, D_2O exch., N*H*H), 8.66 (s, 1H, 5-$H_{triazole}$) ppm; ^{13}C NMR (75.47 MHz, DMSO*d*$_6$) δ 25.6 (CH), 35.2 (CH_2), 40.2 (CH), 40.9 (CH_2), 65.8 (CH_2), 72.6 (CH), 127.0 (CH), 143.6 (C), 164.6 (C) ppm; *m/z* (FAB) 307.1 ((M + 2)-CH_2OH, 35%); $C_9H_{13}IN_4O_2$ (336.1296): calcd. C, 32.16; H, 3.90; N, 16.67; found C, 31.48; H, 4.05; N, 16.88%.

(±)-*tert*-Butyldiphenylsilyl [*t*-3-iodo-*c*-4-(4-phenyl-1*H*-1,2,3-triazol-1-yl)-*r*-1-cyclopentyl]methyl ether (22a) and (±)-*tert*-butyldiphenylsilyl *t*-3-iodo-*c*-4-(5-iodo-4-phenyl-1*H*--1,2,3-triazol-1-yl)-*r*-1-cyclopentyl)methyl ether (22b). *Method A*: The concentration to dryness, the combined early nonvoid fractions eluted with hexane afforded unreacted **15** (4%), and the fractions eluted with hexane/EtOAc 50:1 afforded **22b** (13%), and the fractions eluted with hexane/EtOAc 20:1 afforded **22a** (71%).

Method B: The nonvoid fractions eluting with hexane/EtOAc 10:1 afforded **22a** (76%), that presented identical spectroscopic features as using method A.

22a: whitish oil, ν_{max}/cm^{-1} 2932, 1465, 1407, 1385, 1110, 909, 755, 699; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.08 (9H, s), 2.17-2.28 (1H, m), 2.31-2.50 (3H, m), 2.54-2.63 (1H, m), 3.72 (2H, d, *J* 5.9, HOC*H*$_2$), 4.48 (1H, virtual q, *J* 7.9, 3-H), 5.03 (1H, virtual q, *J* 8.2, 4-H), 7.34-7.46 (9H, m), 7.65-7.68 (4H, m), 7.79 (1H, s, 5-$H_{triazole}$), 7.80-7.84 (2H, m) ppm; ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 10.3 (C), 25.9 (CH), 26.9 ($3CH_3$), 34.0 (CH_2),

39.2 (CH), 40.0 (CH_2), 66.2 (CH_2), 71.4 (CH), 119.0 (CH), 125.8 (CH), 127.8 (CH), 128.2 (CH), 128.8 (CH), 129.8 (CH), 129.8 (CH), 130.5 (C), 133.4 (C), 135.6 (CH), 147.5 (C) ppm; *m/z* (ESI-TOF) 608.16 (M + 1, 20%); HRMS *m/z* calcd for $C_{30}H_{34}IN_3OSi$, 607.1516; found, 607.1550.

22b: yellow oil, ν_{max}/cm^{-1} 2926, 1466, 1407, 11466, 320, 1109, 800, 756, 700; ^1H NMR (300 MHz,$CDCl_3$) δ 1.07 (9H, s), 2.13 (1H, dt, *J* 13.5 and 8.7, 5-*H*H), 2.29-2.49 (3H, m), 2.64-2.69 (1H, m), 3.73 (2H, d, *J* 6.2, HOC*H*$_2$), 4.46 (1H, virtual q, *J* 7.5, 3-H), 5.14 (1H, virtual q, *J* 8.1, 4-H), 7.37-7.50 (9H, m), 7.65-7.68 (4H, m), 7.93-7.96 (2H, m) ppm; ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 19.3 (C), 25.5 (CH), 26.9 ($3CH_3$), 34.3 (CH_2), 39.7 (CH), 39.8 (CH_2), 66.2 (CH_2), 71.1 (CH), 127.5 (C), 127.6 (CH), 127.7 (CH), 127.9 (CH), 128.5 (CH), 129.6 (CH), 129.8 (CH), 130.2 (C), 133.5 (C), 135.6 (CH), 149.4 (C) ppm; *m/z* (FAB) 733.84 (M, 69%). HRMS *m/z* calcd for $C_{30}H_{33}I_2N_3OSi$, 733.0482; found, 733.0506.

(±)-{*t*-3-Iodo-*c*-4-[4-(2-methoxyphenyl)-1*H*-1,2,3-triazol-1-yl]-*r*-1-cyclopentyl}methanol (25a). *Method A*: The nonvoid fractions eluting with hexane-EtOAc 7:1 afforded **25a** accompanied by a small quantity of a non-identified compound. A second column chromatography on silica gel of the above-mentioned mixture using CH_2Cl_2-MeOH 80:1 allowed us to isolate **25a** (63%).

25a: yellow oil, ν_{max}/cm^{-1}3367, 2929, 1549, 1459, 1249, 1044, 801, 755; ^1H NMR (300 MHz, $CDCl_3$) δ 1.64 (1H, br s, OH), 2.21-2.28 (1H, m), 2.33-2.39 (1H, m), 2.41-2.49 (1H, m), 2.55 (1H, dt, *J* 13.9 and 8.7), 2.64-2.74 (1H, m), 3.78 (2H, dd, *J* 5.6 and 4.2, HOC*H*$_2$), 3.95 (3H, s, C*H*$_3$), 4.57 (1H, virtual q, *J* 7.0, 3-H), 5.09 (1H, virtual q, *J* 7.5, 4-H), 6.98 (1H, d, *J* 7.7), 7.08 (1H, t, *J* 7.5), 7.30-7.33 (1H, m), 8.15 (1H, s, 5-$H_{triazole}$), 8.33 (1H, dd, *J* 7.7 and 1.4) ppm; ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 27.0 (CH), 33.6 (CH_2), 39.4 (CH), 40.1 (CH_2), 55.4 (CH_3), 65.3 (CH_2), 71.2 (CH), 110.8 (CH), 119.2 (C), 121.1 (CH), 123.0 (CH), 127.6 (CH), 129.0 (CH), 142.9 (C), 155.7 (C) ppm; *m/z* (ESI-TOF) 400.04 (M + 1, 100%); HRMS *m/z* calcd for $C_{15}H_{18}IN_3O_2$, 399.0444; found, 399.0465.

(±)-{3-[4-(4-Methoxyphenyl)-1*H*-1,2,3-triazol-1-yl]cyclopent-3-enyl}methanol (34b). To a solution of **24a** (50 mg, 0.12 mmol) in dry benzene (1.5 mL) DBU (71 mg, 70 µl, 0.45 mmol) was added. The reaction mixture was stirred under reflux for 30

hours. After that, the solvent was evaporated under reduced pressure, and the subsequent residue was chromatographied on silica gel using hexane-EtOAc 2:1 1:1 as eluents. The non-void fractions afforded **34b** (11 mg, 32%).

34b. beige solid; mp 123-124ºC; ν_{max}/cm^{-1} 3257, 2923, 1658, 1615, 1561, 1497, 1459,1245, 1034, 805; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.58-1.71 (1H, m, D_2O exch., OH), 2.33-2.41 (1H, m), 2.68-2.87 (3H, m), 3.10-3.17 (1H, m), 3.70 (2H, d, *J* 6.4, HOC*H*$_2$), 3.85 (3H, s, CH_3), 6.03-6.07 (1H, m, 4-H), 6.97 (2H, d, *J* 8.79), 7.8 (2H, d, *J* 8.79), 7.85 (1H, s, 5-$H_{triazole}$) ppm; ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 29.7 (CH_2), 34.5 (CH_2), 38.2 (CH), 55.3 (CH_3), 65.2 (CH_2), 66.4 (CH_2), 114.3 (CH), 115.6 (CH), 116.4 (CH), 122.9 (C), 127.1 (CH), 136.5 (CH), 147.5 (C), 159.7 (C) ppm; *m/z* (ESI-TOF) 272.13 (M + 1, 100%); $C_{15}H_{17}N_3O_2$ (271.3144): calcd. C, 66.40; H, 6.32; N, 15.49; found C, 66.69; H, 6.62; N, 15.71%.

Antiviral Activity Assays

Antiviral activity against parainfluenza-3 virus, reovirus-1, Sindbis virus, Coxsackie B4 virus, Punta Toro virus, HSV-1, HSV-2, vaccinia virus, vesicular stomatitis virus, and respyratory syncytial virus was determined essentially as described previously.[1, 2] Antiviral activity was expressed as EC_{50} or concentration required to reduce virus-induced cytopathogenicity as a function of the concentration of the compounds. Cytostatic measurements based on the inhibition of HEL cell growth were performed as follows: HEL cells were seeded at a rate of 5×10^3 cells/well into 96-well microtiter plates and allowed to proliferate for 24 h. Then medium containing different concentrations of the test compound was added. After 3 days of incubation at 37ºC, the cell number was determined with a Coulter counter. The cytostatic concentration was calculated as the CC_{50}, or the compound concentration required to reduce cell growth by 50% relative to the number of cells in the untreated controls. CC_{50} values were estimated from graphic plots of the number of cells (percentage of control) as a function of the concentration of the test compounds. Cytotoxicity was expressed as minimum cytotoxic concentration (MCC) or the

[1] E. De Clercq, A. Holý, I. Rosenberg, T. Sakuma and P. C. Maudgal *Nature* 1986, **323**, 464.
[2] J. Balzarini, L.Naesens, J. Slachmuylders, H. Niphuis, I. Rosenberg, A. Holý, H. Schellekens and E. De Clercq, *AIDS* 1991, **5**, 21.

compound concentration that causes a microscopically detectable alteration of cell morphology of the confluent cell cultures that were exposed the compound.

Table 4 . Antiviral Activity* and Cytotoxicity** of Compounds **21a-28a**, **30**a, **31a**, **23b**, **27b**, **28b**, **30b**, **36**, **38** and **39**.

Virus (strain)	CELL	21a	22a	23a	23b	24a	25a	26a	27a	27b	28a	28b	30a	30b	31a	36	38	39	BVDU[a]	Ribavirin	GCV[b]	ACV[c]
HSV-1 (KOS)	HEL	>20	>4	>20	12	>20	>20	>100	>100	>20	>100	>20	>100	>4	>100	>100	>20	>100	0.08	>250	0.032	0.4
HSV-2 (G)	HEL	>20	>4	>20	12	>20	>20	>100	>100	>20	60	>20	>100	>4	>100	>100	>20	>100	30	150	0.032	0.4
Vaccinia	HEL	12	>4	20	12	>20	>20	>100	>100	>20	60	>20	100	>4	>100	>100	>20	>100	2	150	>100	>250
Vesicular stomatitis	HEL	>20	>4	>20	>20	>20	>20	>100	>100	>20	>100	>20	>100	>4	>100	>100	>20	>100	>250	150	>100	>250
HSV-1 (TK^- KOS ACV)	HEL	>20	>4	>20	20	>20	>20	>100	>100	>20	100	>20	>100	>4	>100	>100	>20	>100	50	>250	30	150
Cytotoxicity	HEL	100	20	100	≥20	100	100	>100	>100	100	>100	≥20	>100	20	>100	>100	100	>100	>250	>250	>100	>250
																					(S)-DHPA[d]	
Vesicular stomatitis	Hela	60	>4	>20	>4	>100	>20	>100	>100	100	>20	>20	≥100	>4	>100	>100	>20	>100	>250	30	50	
Coxsackie B4	Hela	60	>4	>20	>4	>100	>20	>100	>100	60	>20	>20	60	>4	>100	>100	>20	>100	>250	150	>250	
Respiratory syncytial	Hela	>100	>4	>20	>4	>100	>20	>100	>100	>100	>20	>20	>100	4	>100	>100	>20	>100	>250	2	30	
Cytotoxicity	Hela	>100	20	100	20	>100	100	>100	>100	>100	100	100	≥100	20	>100	>100	100	>100	>250	>250	>250	
Parainfluenza-3	Vero	>20	>4	>20	>100	60	20	>100	>100	>20	>100	>20	>100	>4	>100	>100	>100	>100	>400	70	40	
Reovirus-1	Vero	>20	>4	>20	>100	>100	>100	>100	>100	>20	>100	>20	>100	>4	>100	>100	>100	>100	>400	70	100	
Sindbis	Vero	>20	>4	>20	20	>100	60	>100	>100	>20	>100	>20	>100	>4	100	>100	>100	>100	>400	200	>400	
Coxsackie B4	Vero	20	4	>20	20	60	>100	60	>100	>20	>100	>20	>100	>4	>100	>100	>100	>100	>400	>400	>400	
Punta Toro	Vero	20	>4	20	>100	60	20	>100	>100	>20	>100	>20	>100	4	>100	>100	>100	>100	>400	70	>400	
Cytotoxicity	Vero	100	20	100	>100	>100	>100	>100	>100	100	>100	100	>100	20	>100	>100	>100	>100	>400	>400	>400	

*MIC_{50} or Minimun inhibitory concentration (μg/mL) required to reduce virus-induced cytopatogenicity by 50%.
**MCC or Minimum cytotoxic concentration (μg/mL) required to cause a microscopically detectable alteration of normal cell morphology.
Cell lines used: human embryonic skin-muscle (E_6SM) fibroblasts, human epithelial (Hela) cells and African green monkey (Vero) kidney cells.
[a]Brivudin, [b]gancyclovir, [c]acyclovir, [d](*S*)-9-(2,3-dihydroxypropyl)adenine.

Table 5. Antiviral Activity of Compounds **21a-25a** against CMV and VZV in Human Embryonic Lung (HEL) Cells

comp	Davis strain	TK^+ VZV OKA strain	TK^- VZV 07/1 strain	Cell morphology (MCC)[b]	Cell growth (CC_{50})[c]
21a	>20	>20	>20	100	43.8
22a	100	>100	>100	>100	>100
22b	>100	>100	>100	>100	>100
23a	>4	>4	>20	≥20	32.2
24a	>20	46	>20	≥100	47.9
25a	>20	4.5	20	100	43.3
35	54.7	56	69	>100	34.4

Table 6. Antiviral Activity of Compounds **23b**, **26a-28a**, **26b-28b**, **30a**, **31a**, **36**, **38**, and **39** against Feline Corona Virus and Feline Herpes Virus in Crandell-Rees Feline Kidney (CRFK) Cells.

	CC_{50}[a] (μg/mL)	EC_{50}[b] (μg/mL)	
Compounds	Cell growth	Feline Corona Virus	Feline Herpes Virus
23b	33.7	>20	>20
26a	81.9	>20	>20
27a	>100	>100	>100
27b	>100	>100	>100
28a	>100	88.8	>100
28b	14.6	>4	>4
30a	>100	>100	>100
30b	18.0	>4	>4
31a	>100	>100	>100
36	>100	>100	>100
38	68.2	>20	>20
39	>100	>100	>100

[a]50% Cytotoxic concentration, as determined by measuring the cell viability with the colorimetric formazan-based MTS assay.
[b]50% Effective concentration or concentration producing 50% inhibition of virus-induced cytopathic effect, as determined by measuring the cell viability with the colorimetric formazan-based MTS assay.