

Supporting Information

for

Effect of uridine protecting groups on the diastereoselectivity of uridine-derived aldehyde 5'-alkynylation

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**Description of materials and methods,
and preparation and characterization of new compounds**

***N*³-Allyl-2',3'-*O*-isopropylideneuridine (1b).** Compound **1b** was synthesized according to the general procedure for *N*-allylation of protected uridine derivatives from acetone **1a**¹ (1.0 g, 3.52 mmol, 1 equiv). Flash chromatography of the residue (cyclohexane/EtOAc 1:1) afforded **1b** as a gum (981 mg, 86% yield): *R*_f 0.34 (cyclohexane/EtOAc 1/1); [α]_D - 28 (*c* 0.5, CH₂Cl₂); IR (film) 3607*br*, 2297*m*, 1700*s*, 1676*s*, 1453*m*, 1040*m*; ¹H NMR δ 7.21 (d, 1H, *J*_{H6-H5} = 8.5 Hz, H₆), 5.87 (ddt, 1H, *J*_{H8-H9a} = 17.0 Hz, *J*_{H8-H9b} = 10.0 Hz, *J*_{H8-H7a} = *J*_{H8-H7b} = 6.0 Hz, H₈), 5.77 (d, 1H, *J*_{H5-H6} = 8.5 Hz, H₅), 5.54 (d, 1H, *J*_{H1'-H2'} = 3.0 Hz, H_{1'}), 5.29-5.24 (m, 1H, H_{9a}), 5.22-5.18 (m, 1H, H_{9b}), 5.07 (dd, 1H, *J*_{H2'-H3'} = 6.5 Hz, *J*_{H2'-H1'} = 3.0 Hz, H_{2'}), 4.99 (dd, 1H, *J*_{H3'-H2'} = 6.5 Hz, *J*_{H3'-H4'} = 3.5 Hz, H_{3'}), 4.54-4.52 (m, 2H, H₇), 4.31-4.29 (m, 1H, H_{4'}), 3.93 (dt, 1H, *J*_{H5'a-H5'b} = 12.0 Hz, *J*_{H5'a-H4'} = *J*_{H5'a-OH} = 3.0 Hz, H_{5'a}), 3.81 (ddd, 1H, *J*_{H5'b-H5'a} = 12.0 Hz, *J*_{H5'b-H4'} = 3.0 Hz, *J*_{H5'b-OH} = 7.5 Hz, H_{5'b}), 2.70 (dd, 1H, *J*_{OH-H5'a} = 3.0 Hz, *J*_{OH-H5'b} = 7.5 Hz, OH), 1.58 (s, 3H, H_{7'}), 1.37 (s, 3H, H_{7'}); ¹³C NMR δ 162.2 (C₄), 150.9 (C₂), 140.9 (C₆), 131.3 (C₈), 118.5 (C₉), 114.5 (C_{6'}), 102.4 (C₅), 97.4 (C_{1'}), 87.1 (C_{4'}), 83.8 (C_{2'}), 80.5 (C_{3'}), 62.9 (C_{5'}), 43.2 (C₇), 27.5, 25.4 (C_{7'}); HRMS ESI⁺ calcd for C₁₅H₂₁N₂O₆⁺ (M + H)⁺ 325.1394, found 325.1388.

2',3'-*O*-Isopentylideneuridine (2a). To a suspension of uridine (1.0 g, 4.09 mmol, 1 equiv) in pentanone (21.5 mL, 203.1 mmol, 49.6 equiv), was added sulfuric acid (300 μL, 4.09 mmol, 1 equiv) and anhydrous MgSO₄ (985 mg, 8.19 mmol, 2 equiv). The suspension was stirred at rt for 48 h, neutralized by a dropwise addition of triethylamine and concentrated in vacuo. Flash chromatography of the resulting orange oil (cyclohexane/EtOAc 1:9 to 0:10) afforded the isopentylidene **2a** as a white foam (1.0 g, 78% yield): *R*_f 0.35 (Cyclohexane/EtOAc 1/9); mp 112-115 °C; [α]_D - 35 (*c* 1.0, CH₂Cl₂); IR (film) 2999*br*, 1691*s*, 1463*m*, 1384*w*, 1265*m*, 1087*m*; ¹H NMR δ 9.30 (br s, 1H, NH), 7.36 (d, 1H, *J*_{H6-H5} = 8.5 Hz, H₆), 5.73 (dd, 1H, *J*_{H5-H6} = 8.5 Hz, *J*_{H5-NH} = 1.5 Hz, H₅), 5.60 (d, 1H, *J*_{H1'-H2'} = 3.0 Hz, H_{1'}), 5.06 (dd, 1H, *J*_{H2'-H3'} = 7.0 Hz, *J*_{H2'-H1'} = 3.0 Hz, H_{2'}), 4.97 (dd, 1H, *J*_{H3'-H2'} = 7.0 Hz, *J*_{H3'-H4'} = 4.0 Hz, H_{3'}), 4.32-4.30 (m, 1H, H_{4'}), 3.92 (dt, 1H, *J*_{H5'a-H5'b} = 12.0 Hz, *J*_{H5'a-H4'} = *J*_{H5'a-OH} = 4.0 Hz, H_{5'a}), 3.85-3.80 (m, 1H, H_{5'b}), 2.99 (dd, 1H, *J*_{OH-H5'a} = 4.0 Hz, *J*_{OH-H5'b} = 7.0 Hz, OH), 1.80 (q, 2H, *J*_{H7'-H8'} = 7.5 Hz, H_{7'}), 1.63 (q, 2H, *J*_{H7'-H8'} = 7.5 Hz, H_{7'}), 0.99 (t, 3H, *J*_{H8'-H7'} = 7.5 Hz, H_{8'}), 0.89 (t, 3H, *J*_{H8'-H7'} = 7.5 Hz, H_{8'}); ¹³C NMR δ 163.2 (C₄), 150.4 (C₂), 143.1 (C₆), 119.1 (C_{6'}), 102.8 (C₅), 96.3 (C_{1'}), 87.3 (C_{4'}), 84.1 (C_{2'}), 80.6 (C_{3'}), 62.9 (C_{5'}), 29.5, 29.4 (C₇), 8.5, 7.9 (C₈); HRMS ESI⁻ calcd for C₁₄H₁₉N₂O₆⁻ (M - H)⁻ 311.1249, found 311.1252.

2',3'-*O*-Cyclohexylideneuridine (3a). To a solution of uridine (1.0 g, 4.09 mmol, 1 equiv) in cyclohexanone (6 mL, 57.9 mmol, 14.1 equiv) was added sulfuric acid (cat). The suspension was stirred at 50 °C for 12 h, cooled to rt, neutralized by a dropwise addition of triethylamine and concentrated *in vacuo*. Flash chromatography of the resulting orange oil (Cyclohexane/EtOAc 6/4 to

¹ Eppacher, S.; Solladie, N.; Bernet, B.; Vasella, A. *Helv. Chim. Acta* **2000**, *83*, 1311–1330.

3/7) afforded the cyclohexylidene **3a** as a white foam (1.18 g, 89% yield): R_f 0.22 (Cyclohexane/EtOAc 3/7); mp 136-140 °C; $[\alpha]_D - 36$ (c 1.0, CH₂Cl₂); IR (film) 2917 br , 1691 s , 1463 m , 1388 w ; ¹H NMR δ 9.22 (br s, 1H, NH), 7.39 (d, 1H, $J_{H_6-H_5} = 8.0$ Hz, H₆), 5.74 (d, 1H, $J_{H_5-H_6} = 8.0$ Hz, H₅), 5.60 (d, 1H, $J_{H_{1'}-H_{2'}} = 3.0$ Hz, H_{1'}), 5.04 (dd, 1H, $J_{H_{2'}-H_{3'}} = 6.5$ Hz, $J_{H_{2'}-H_{1'}} = 3.0$ Hz, H_{2'}), 4.94 (dd, 1H, $J_{H_{3'}-H_{2'}} = 6.5$ Hz, $J_{H_{3'}-H_{4'}} = 3.5$ Hz, H_{3'}), 4.30-4.28 (m, 1H, H_{4'}), 3.92 (dt, 1H, $J_{H_{5'a}-H_{5'b}} = 12.0$ Hz, $J_{H_{5'a}-H_{4'}} = J_{H_{5'a}-OH} = 3.0$ Hz, H_{5'a}), 3.84-3.79 (m, 1H, H_{5'b}), 2.99 (dd, 1H, $J_{OH-H_{5'a}} = 3.0$ Hz, $J_{OH-H_{5'b}} = 7.5$ Hz, OH), 1.79-1.76 (m, 2H, H_{8'a}), 1.69-1.64 (m, 2H, H_{8'b}), 1.59-1.54 (m, 4H, H_{7'}), 1.43-1.39 (m, 2H, H_{9'}); ¹³C NMR δ 163.3 (C₄), 150.5 (C₂), 143.2 (C₆), 115.4 (C_{6'}), 102.8 (C₅), 96.2 (C_{1'}), 87.2 (C_{4'}), 83.3 (C_{2'}), 80.1 (C_{3'}), 62.8 (C_{5'}), 37.3, 34.9 (C_{7'}), 25.0, 24.1 (C_{8'}), 23.7 (C_{9'}); HRMS ESI⁺ calcd for C₁₅H₂₁N₂O₆⁺ (M + H)⁺ 325.1394, found 325.1391.

***N*³-Allyl-2',3'-*O*-(*tert*-butyldimethylsilyl)uridine (4b)**

Compound **4b** was synthesized according to the general procedure for *N*-allylation of protected uridine derivatives from compound **4a**² (400 mg, 0.85 mmol, 1 equiv). Flash chromatography of the residue (cyclohexane/EtOAc 8:2) afforded **4b** as a white solid (404 mg, 93% yield): R_f 0.25 (cyclohexane/EtOAc 8:2); mp 201-207 °C; $[\alpha]_D - 21$ (c 1.0, CH₂Cl₂); IR (film) 3710 br , 3005 br , 2369 w , 1708 m , 1664 s , 1461 m , 940 m ; ¹H NMR δ 7.51 (d, 1H, $J_{H_6-H_5} = 8.0$ Hz, H₆), 5.87 (ddt, 1H, $J_{H_8-H_{9a}} = 17.0$ Hz, $J_{H_8-H_{9b}} = 10.0$ Hz, $J_{H_8-H_{7a}} = J_{H_8-H_{7b}} = 6.0$ Hz, H₈), 5.76 (d, 1H, $J_{H_5-H_6} = 8.0$ Hz, H₅), 5.48 (d, 1H, $J_{H_{1'}-H_{2'}} = 6.0$ Hz, H_{1'}), 5.25 (d, 1H, $J_{H_{9a}-H_8} = 17.0$ Hz, $J_{H_{9a}-H_{7a}} = J_{H_{9a}-H_{7b}} = 2.0$ Hz, H_{9a}), 5.17 (dt, 1H, $J_{H_{9b}-H_8} = 10.0$ Hz, $J_{H_{9a}-H_{7a}} = J_{H_{9a}-H_{7b}} = 2.0$ Hz, H_{9b}), 4.61 (t, 1H, $J_{H_{2'}-H_{3'}} = 6.0$ Hz, $J_{H_{2'}-H_{1'}} = 6.0$ Hz, H_{2'}), 4.55-4.42 (m, 2H, H₇), 4.18 (dd, 1H, $J_{H_{3'}-H_{2'}} = 6.0$ Hz, $J_{H_{3'}-H_{4'}} = 3.0$ Hz, H_{3'}), 4.09-4.06 (m, 1H, H_{4'}), 3.89 (dt, 1H, $J_{H_{5'a}-H_{5'b}} = 12.0$ Hz, $J_{H_{5'a}-H_{4'}} = J_{H_{5'a}-OH} = 2.5$ Hz, H_{5'a}), 3.72 (ddd, 1H, $J_{H_{5'b}-H_{5'a}} = 12.0$ Hz, $J_{H_{5'b}-H_{4'}} = 2.5$ Hz, $J_{H_{5'b}-OH} = 8.5$ Hz, H_{5'b}), 3.21 (dd, 1H, $J_{OH-H_{5'a}} = 2.5$ Hz, $J_{OH-H_{5'b}} = 8.5$ Hz, OH), 0.92 (s, 9H, -C(CH₃)₃), 0.86 (s, 9H, -C(CH₃)₃), 0.09 (s, 3H, CH₃), 0.09 (s, 3H, CH₃), 0.05 (s, 3H, CH₃), -0.02 (s, 3H, CH₃); ¹³C NMR δ 162.4 (C₄), 150.9 (C₂), 141.3 (C₆), 131.4 (C₈), 118.6 (C₉), 101.9 (C₅), 94.9 (C_{1'}), 86.8 (C_{4'}), 73.3 (C_{2'}), 72.4 (C_{3'}), 62.1 (C_{5'}), 43.2 (C₇) 25.9, 25.9 (-C(CH₃)₃), 18.2, 18.1 (-C(CH₃)₃), -4.3, -4.4, -4.6, -4.7 (-C(CH₃)₂); HRMS ESI⁺ calcd for C₂₄H₄₅N₂O₆Si₂⁺ (M + H)⁺ 513.2811, found 513.2813.

***N*³-Allyl-2',3'-*Di-O*-(*triisopropylsilyl*)uridine (5b).** Compound **5b** was synthesized according to the general procedure for *N*-allylation of protected uridine derivatives from the compound **5a**³ (500 mg, 0.89 mmol, 1 equiv). This compound was obtained as a white solid (525 mg, 98% yield): R_f 0.34 (cyclohexane/EtOAc 7:3); mp 188–190 °C; $[\alpha]_D - 58$ (c 0.5, CHCl₃); IR (film) 3464, 2945, 2865,

² Maturano, M.; Catana, D.-A.; Lavedan, P.; Tarrat, N.; Saffon, N.; Payrastre, C.; Escudier, J.-M. *Eur. J. Org. Chem.* **2012**, 721–730.

³ Hwu, J. R. Jain, M. L.; Tsai, F.-Y.; Tsay, S.-C. Balakumar, A.; Hakimelahi, G. H. *J. Org. Chem.* **2000**, *65*, 17, 5077-5088.

1697, 1658, 1658, 1455, 1391, 1172; 1095, 991, 824; ¹H NMR δ 7.51 (d, $J_{H_6-H_5} = 8.1$ Hz, 1H, H₆), 5.86 (ddt, $J_{H_8-H_9a} = 16.2$ Hz, $J_{H_8-H_9b} = 10.2$ Hz, $J_{H_8-H_7a} = 6.0$ Hz, 1H, H₈), 5.79 (d, $J_{H_5-H_6} =$ Hz, 1H, H₅), 5.61 (d, $J_{H_{1'}-H_{2'}} = 7.0$ Hz, 1H, H_{1'}), 5.33 - 5.25 (m, 1H, H_{9a}), 5.23 - 5.16 (m, 1H, H_{9b}), 4.93 (dd, $J_{H_{1'}-H_{2'}} = 7.0$ Hz, $J_{H_{2'}-H_{3'}} = 4.2$ Hz, 1H, H_{2'}), 4.58 - 4.49 (m, 2H, H₇), 4.42 (dd, $J_{H_{3'}-H_{2'}} = 4.2$ Hz, $J_{H_{3'}-H_{4'}} = 1.4$ Hz, 1H, H_{3'}), 4.17 - 4.09 (m, 1H, H_{4'}), 3.90 (dt, $J_{H_{5'a}-H_{5'b}} = 12.3$ Hz, $J_{H_{5b'}-H_{4'}} = 2.9$ Hz, 1H, H_{5'b}), 3.75 (ddd, $J_{H_{5'a}-H_{5'b}} = 12.3$ Hz, $J_{H_{5'a}-OH} = 7.8$ Hz, $J_{H_{5'a}-H_{4'}} = 2.4$ Hz, 1H, H_{5'a}), 3.50 (dd, $J_{H_{5'a}-OH} = 7.7$ Hz, $J_{H_{5b'}-H_{4'}} = 2.9$ Hz, 1H, OH), 1.13 - 1.00 (m, 42H, (-CH(CH₃)₂)); ¹³C NMR δ 162.29 (C₄), 150.89 (C₂), 141.75 (C₆), 131.21 (C₈), 118.46 (C₉), 102.04 (C₅), 94.41 (C_{1'}), 86.44 (C_{4'}), 74.15 (C_{3'}), 73.61 (C_{2'}), 62.65 (C_{5'}), 43.07 (C₇), 18.04, 18.1, 18.0, 17.9, 12.9, 12.6 ((-C(CH₃)₃)); HRMS ESI⁺ calcd for C₃₀H₅₇N₂O₆Si₂⁺ (M + H)⁺ 597.3750, found 597.3757.

5'(R)-C-(Triethylsilylethynyl)-2',3'-O-isopropylideneuridine (11bb). Compound **6b** was synthesized from compound **1b** (236 mg, 0.73 mmol, 1 equiv) according to the general procedure for 5 the oxidation of uridine derivatives. Complete conversion was observed in 1 h and aldehyde **11b** was directly engaged in alkynylation reaction. Data for **11b**: R_f 0.37 (cyclohexane/EtOAc 1:1); ¹H NMR δ 9.37 (s, 1H, H_{5'}), 7.20 (d, 1H, $J_{H_6-H_5} = 8.0$ Hz, H₆), 5.83 (ddt, 1H, $J_{H_8-H_9a} = 17.0$ Hz, $J_{H_8-H_9b} = 10.0$ Hz, $J_{H_8-H_7a} = J_{H_8-H_7b} = 6.0$ Hz, H₈), 5.81 (d, 1H, $J_{H_5-H_6} = 8.0$ Hz, H₅), 5.48 (s, 1H, H_{1'}), 5.25 (dd, 1H, $J_{H_{3'}-H_{2'}} = 6.0$ Hz, $J_{H_{3'}-H_{4'}} = 1.5$ Hz, H_{3'}), 5.24-5.20 (m, 1H, H_{9a}), 5.19-5.16 (m, 1H, H_{9b}), 5.10 (d, 1H, $J_{H_{2'}-H_{3'}} = 6.0$ Hz, H_{2'}), 4.55 (d, 1H, $J_{H_{4'}-H_{3'}} = 1.5$ Hz, H_{4'}), 4.51 (ddt, 1H, $J_{H_{7a}-H_{7b}} = 14.5$ Hz, $J_{H_{7a}-H_8} = 6.0$ Hz, $J_{H_{7a}-H_{9a}} = J_{H_{7a}-H_{9b}} = 1.5$ Hz, H_{7a}), 4.42 (ddt, 1H, $J_{H_{7b}-H_{7a}} = 14.5$ Hz, $J_{H_{7b}-H_8} = 6.0$ Hz, $J_{H_{7b}-H_{9a}} = J_{H_{7b}-H_{9b}} = 1.5$ Hz, H_{7b}), 1.55 (s, 3H, H_{7'}), 1.38 (s, 3H, H_{7'}); ¹³C NMR δ 199.3 (C_{5'}), 162.0 (C₄), 151.1 (C₂), 141.9 (C₆), 131.1 (C₈), 118.5 (C₉), 113.7 (C_{6'}), 102.7 (C₅), 101.2 (C_{1'}), 94.3 (C₂), 85.1 (C_{4'}), 84.2 (C_{3'}), 43.0 (C₇), 26.6, 24.9 (C_{7'}). Compound **11bb** was synthesized according to the general procedure for the 5'-alkynylation of uridine-derived aldehydes: a dropwise addition of the crude aldehyde **6b** (235 mg, 0.73 mmol, 1 equiv) solubilized in dry THF (7.2 mL), on a freshly prepared triethylsilylacetylide magnesium bromide solution (2.18 mmol, 3 equiv) in THF (7.2 mL). After work up, the crude mixture **11bb** was isolated as a 65/35 mixture of diastereomers. Flash chromatography of the residue (cyclohexane/EtOAc 1:1) afforded alcohol **11bb** as a colourless gum (176 mg, 52% combined yield). The major diastereoisomer has been isolated with 24% yield as a colourless gum: R_f 0.34 (cyclohexane/EtOAc 1:1); [α]_D - 30 (*c* 1.0, CH₂Cl₂); IR (film) 3710*br*, 2356*w*, 1704*m*, 1662*s*, 1451*m*, 939*m*; ¹H NMR δ 7.54 (d, 1H, $J_{H_6-H_5} = 8.0$ Hz, H₆), 5.85 (ddt, 1H, $J_{H_8-H_9a} = 17.0$ Hz, $J_{H_8-H_9b} = 10.0$ Hz, $J_{H_8-H_7a} = J_{H_8-H_7b} = 6.0$ Hz, H₈), 5.77 (d, 1H, $J_{H_{1'}-H_{2'}} = 4.0$ Hz, H_{1'}), 5.74 (d, 1H, $J_{H_5-H_6} = 8.0$ Hz, H₅), 5.27-5.22 (m, 1H, H_{9a}), 5.19-5.15 (m, 1H, H_{9b}), 5.01 (dd, 1H, $J_{H_{3'}-H_{2'}} = 6.5$ Hz, $J_{H_{3'}-H_{4'}} = 2.5$ Hz, H_{3'}), 4.86 (dd, 1H, $J_{H_{2'}-H_{3'}} = 6.5$ Hz, $J_{H_{2'}-H_{1'}} = 4.0$ Hz, H_{2'}), 4.65-4.62 (m, 1H, H_{5'}), 4.56-4.46 (m, 2H, H₇), 4.36 (t, 1H, $J_{H_{4'}-H_{3'}} = J_{H_{4'}-H_{5'}} = 2.5$ Hz, H_{4'}), 3.65 (brd, 1H, $J_{OH-H_5'} = 3.0$ Hz, OH), 1.58 (s, 3H, H_{11'}), 1.35 (s, 3H, H_{11'}), 0.98 (t, 9H, $J_{H_9'-H_8'} = 7.5$ Hz, H_{9'}), 0.62 (q,

6H, $J_{H9'-H8'} = 7.5$ Hz, H_{8'}); ¹³C NMR δ 162.5 (C₄), 150.9 (C₂), 139.9 (C₆), 131.3 (C₈), 118.5 (C₉), 114.3 (C_{10'}), 103.2 (C_{6'}), 101.9 (C₅), 95.7 (C_{1'}), 90.3 (C_{7'}), 87.8 (C_{4'}), 83.6 (C_{2'}), 80.5 (C_{3'}), 63.2 (C_{5'}), 43.3 (C₇), 27.3, 25.2 (C_{11'}), 7.5 (C_{9'}), 4.3 (C_{8'}); HRMS ESI⁺ calcd for C₂₃H₃₅N₂O₆Si⁺ (M + H)⁺ 463.2259, found 463.2266.

5'(R)-C-(Triethylsilylethynyl)-2',3'-O-isopentylideneuridine (12ab). Compound **7a** was synthesized from compound **2a** (180 mg, 0.57 mmol, 1 equiv) according to the general procedure for the oxidation of uridine derivatives. Complete conversion was observed in 45 min and aldehyde **7a** was directly engaged in alkynylation reaction. Data for **7a**: R_f 0.30 (cyclohexane/EtOAc 1:1); ¹H NMR δ 9.44 (s, 1H, H_{5'}), 8.59 (br s, 1H, NH), 7.27 (d, 1H, $J_{H6-H5} = 8.0$ Hz, H₆), 5.76 (d, 1H, $J_{H5-H6} = 8.0$ Hz, H₅), 5.50 (s, 1H, H_{1'}), 5.24 (dd, 1H, $J_{H3'-H2'} = 7.0$ Hz, $J_{H3'-H4'} = 2.0$ Hz, H_{3'}), 5.11 (d, 1H, $J_{H2'-H3'} = 7.0$ Hz, H_{2'}), 4.58 (d, 1H, $J_{H4'-H3'} = 2.0$ Hz, H_{4'}), 1.76 (q, 2H, $J_{H7'-H8'} = 7.5$ Hz, H_{7'}), 1.63 (q, 2H, $J_{H7'-H8'} = 7.5$ Hz, H_{7'}), 0.96 (t, 3H, $J_{H8'-H7'} = 7.5$ Hz, H_{8'}), 0.90 (t, 3H, $J_{H8'-H7'} = 7.5$ Hz, H_{8'}); ¹³C NMR δ 199.4 (C₅), 162.6 (C₄), 150.3 (C₂), 143.9 (C₆), 118.3 (C_{6'}), 102.8 (C₅), 100.4 (C_{1'}), 94.3 (C_{4'}), 85.2 (C_{2'}), 84.1 (C_{3'}), 29.3, 28.8 (C_{7'}), 8.3, 8.4 (C_{8'}). Compound **12ab** was synthesized according to the general procedure for the 5'-alkynylation of uridine-derived aldehydes: a dropwise addition of a solution of the crude aldehyde **7a** (179 mg, 0.57 mmol, 1 equiv) in dry THF (5.6 mL), on a freshly prepared triethylsilylacetylide magnesium bromide solution (1.49 mmol, 2.6 equiv) in THF (5.6 mL). After work up, the crude mixture **12ab** was isolated as a 65/35 mixture of diastereomers. Flash chromatography of the residue (cyclohexane/EtOAc 7:3 to 6:4) afforded alcohol **12ab** as a white solid (139 mg, 54% combined yield). The major diastereoisomer (5'R)-**12ab** has been isolated with 15% yield as a white foam. Data for (5'R)-**12ab** R_f 0.37 (Cyclohexane/EtOAc 1/1); mp 149-152 °C; [α]_D -26 (c 0.5, CH₂Cl₂); IR (film) 3320br, 2356w, 1702m, 1664s, 1450m, 940m; ¹H NMR δ 9.50 (br s, 1H, NH), 7.52 (d, 1H, $J_{H6-H5} = 8.0$ Hz, H₆), 5.81 (d, 1H, $J_{H1'-H2'} = 4.0$ Hz, H_{1'}), 5.73 (dd, 1H, $J_{H5-H6} = 8.0$ Hz, $J_{H5-NH} = 1.5$ Hz, H₅), 5.03 (dd, 1H, $J_{H3'-H2'} = 6.5$ Hz, $J_{H3'-H4'} = 2.5$ Hz, H_{3'}), 4.89 (dd, 1H, $J_{H2'-H3'} = 6.5$ Hz, $J_{H2'-H1'} = 4.0$ Hz, H_{2'}), 4.65 (t, 1H, $J_{H5'-H4'} = J_{H5'-OH} = 2.5$ Hz, H_{5'}), 4.35 (t, 1H, $J_{H4'-H5'} = J_{H4'-H3'} = 2.5$ Hz, H_{4'}), 3.36 (brs, 1H, OH), 1.80 (q, 2H, $J_{H11'-H12'} = 7.0$ Hz, H_{11'}), 1.63 (q, 2H, $J_{H11'-H12'} = 7.0$ Hz, H_{7'}), 1.01 (t, 3H, $J_{H12'-H11'} = 7.0$ Hz, H_{12'}), 0.99 (t, 9H, $J_{H8'-H9'} = 8.0$ Hz, H_{8'}), 0.88 (t, 3H, $J_{H12'-H11'} = 7.0$ Hz, H_{12'}), 0.62 (q, 6H, $J_{H9'-H8'} = 8.0$ Hz, H_{9'}); ¹³C NMR δ 163.2 (C₄), 150.4 (C₂), 141.2 (C₆), 119.0 (C_{10'}), 103.2 (C_{6'}), 102.8 (C₅), 94.5 (C_{1'}), 90.3 (C_{7'}), 87.8 (C_{4'}), 83.4 (C_{2'}), 80.4 (C_{3'}), 63.2 (C_{5'}), 29.4, 29.3 (C_{11'}), 8.4, 7.9 (C_{12'}), 7.5 (C_{9'}), 4.3 (C_{8'}); HRMS ESI⁺ calcd for C₂₂H₃₅N₂O₆Si⁺ (M + H)⁺ 451.2259, found 451.2270.

5'(R)-C-(Triethylsilylethynyl)-2',3'-O-cyclohexylideneuridine (13ab). Compound **8a** was synthesized from compound **3a** (250 mg, 0.77 mmol, 1 equiv) according to the general procedure for the oxidation of uridine derivatives. Complete conversion was observed in 1 h and aldehyde **8a** was directly engaged in alkynylation reaction. Data for **8a**: R_f 0.32 (cyclohexane/EtOAc 6/4); ¹H NMR δ

9.45 (s, 1H, H₅), 8.53 (br s, 1H, NH), 7.26 (d, 1H, $J_{H_6-H_5} = 8.0$ Hz, H₆), 5.76 (d, 1H, $J_{H_5-H_6} = 8.0$ Hz, H₅), 5.50 (s, 1H, H_{1'}), 5.22 (dd, 1H, $J_{H_3'-H_2'} = 6.5$ Hz, $J_{H_3'-H_4'} = 2.0$ Hz, H_{3'}), 5.11 (d, 1H, $J_{H_2'-H_3'} = 6.5$ Hz, H_{2'}), 4.56 (d, 1H, $J_{H_4'-H_3'} = 2.0$ Hz, H_{4'}), 1.77-1.74 (m, 2H, H_{8'a}), 1.70-1.64 (m, 2H, H_{8'b}), 1.63-1.55 (m, 4H, H_{7'}), 1.44-1.41 (m, 2H, H_{9'}); ¹³C NMR δ 199.3 (C_{5'}), 162.7 (C₄), 150.4 (C₂), 144.1 (C₆), 114.7 (C_{6'}), 102.9 (C₅), 100.5 (C_{1'}), 94.4 (C_{4'}), 84.6 (C_{2'}), 83.4 (C_{3'}), 36.4, 34.4 (C_{7'}), 25.1, 24.1 (C_{8'}), 23.8 (C_{9'}). Compound **13ab** was synthesized according to the general procedure for the 5'-alkynylation of uridine-derived aldehydes: a dropwise addition of a solution of the crude aldehyde **8a** (249 mg, 0.77 mmol, 1 equiv) in dry THF (7.5 mL), on a freshly prepared triethylsilylacetylide magnesium bromide solution (1.89 mmol, 2.45 equiv) in THF (7.5 mL). After work up, the crude mixture **13ab** was isolated as a 70/30 mixture of diastereomers. Flash chromatography of the residue (Cyclohexane/EtOAc 7/3, to 1/1) afforded alcohol (5'*R*)-**13ab** as a white solid (225 mg, 65% combined yield). The major diastereoisomer has been isolated with 18% yield as a white foam. Data for (5'*R*)-**13ab** R_f 0.34 (cyclohexane/EtOAc 1:1); mp 150-152 °C; $[\alpha]_D - 22$ (*c* 0.5, CH₂Cl₂); IR (film) 2936*br*, 1692*s*, 1462*w*, 1275*s*, 1098*m*, 940*m*; ¹H NMR δ 8.59 (br s, 1H, NH), 7.50 (d, 1H, $J_{H_6-H_5} = 8.5$ Hz, H₆), 5.76 (d, 1H, $J_{H_1'-H_2'} = 3.0$ Hz, H_{1'}), 5.73 (d, 1H, $J_{H_5-H_6} = 8.5$ Hz, H₅), 5.02 (dd, 1H, $J_{H_3'-H_2'} = 6.5$ Hz, $J_{H_3'-H_4'} = 3.0$ Hz, H_{3'}), 4.91 (dd, 1H, $J_{H_2'-H_3'} = 6.5$ Hz, $J_{H_2'-H_1'} = 3.0$ Hz, H_{2'}), 4.66 (t, 1H, $J_{H_5'-H_4'} = J_{H_5'-OH} = 3.0$ Hz, H_{5'}), 4.36 (t, 1H, $J_{H_4'-H_5'} = J_{H_4'-H_3'} = 3.0$ Hz, H_{4'}), 3.21 (d, 1H, $J_{OH-H_5'} = 3.0$ Hz, OH), 1.80-1.77 (m, 2H, H_{12'a}), 1.70-1.65 (m, 2H, H_{12'b}), 1.65-1.52 (m, 4H, H_{11'}), 1.46-1.34 (m, 2H, H_{13'}), 1.10 (t, 9H, $J_{H_9'-H_8'} = 7.5$ Hz, H_{9'}), 0.62 (q, 6H, $J_{H_9'-H_8'} = 7.5$ Hz, H_{8'}); ¹³C NMR δ 162.8 (C₄), 150.3 (C₂), 142.2 (C₆), 115.4 (C_{10'}), 103.1 (C_{6'}), 102.9 (C₅), 95.0 (C_{1'}), 90.4 (C_{7'}), 87.5 (C_{4'}), 82.4 (C_{2'}), 79.8 (C_{3'}), 63.4 (C_{5'}), 37.3, 34.7 (C_{11'}), 25.1, 24.1 (C_{12'}), 23.6 (C_{13'}), 7.5 (C_{9'}), 4.3 (C_{8'}); HRMS ESI⁻ calcd for C₂₃H₃₃N₂O₆Si⁻ (M - H)⁻ 461.2113, found 461.2125.

5'(S)-(Triethylsilylethynyl)-2',3'-di-O-(tert-butyltrimethylsilyl)uridine (14ab). Compound **9a** was synthesized from alcohol **4a** (370 mg, 0.78 mmol, 1 equiv) according to the general procedure for the oxidation of uridine derivatives. Complete conversion was observed in 45 min and aldehyde **9a** was directly engaged in alkylation reaction. Spectral data were in agreement with literature.² Compound **14ab** was synthesized according to the general procedure for the 5'-alkynylation of uridine-derived aldehydes: a dropwise addition of a solution of the crude aldehyde **9a** (370, mg, 0.78 mmol, 1 equiv) in dry THF (7.7 mL), on a freshly prepared triethylsilylacetylide magnesium bromide solution (1.92 mmol, 2.5 equiv) in THF (7.7 mL). After work up, the crude mixture **14ab** was isolated as a 85/15 mixture of diastereomers. Flash chromatography of the residue (cyclohexane/EtOAc 8:2 to 7:3) afforded **14ab** as a white solid (308 mg, 64% combined yield). The major diastereoisomer (5'*S*)-**14ab** has been isolated with 48% yield. Data for (5'*S*)-**14ab**: R_f 0.47 (cyclohexane/EtOAc = 1:1); mp 182 °C; $[\alpha]_D - 42$ (*c* 0.5, CH₂Cl₂); IR (film) 3201*br*, 2870*w*, 1670*s*, 1460*m*, 1362*m*, 1081*s*, 723*s*; ¹H NMR δ 9.83 (s, 1H, NH), 7.60 (d, 1H, $J_{H_6-H_5} = 8.0$ Hz, H₆), 5.73 (dd,

1H, $J_{H5-H6} = 8.0$ Hz, $J_{H5-NH} = 1.5$ Hz, H₅), 5.61 (d, 1H, $J_{H1'-H2'} = 6.0$ Hz, H_{1'}), 4.62 (t, 1H, $J_{H3'-H2'} = 5.0$ Hz, $J_{H3'-H4'} = 5.0$ Hz, H_{3'}), 4.54 (dd, 1H, $J_{H2'-H3'} = 5.0$ Hz, $J_{H2'-H1'} = 6.0$ Hz, H_{2'}), 4.17 (dd, 1H, $J_{H5'-H4'} = 5.0$ Hz, $J_{H5'-OH'} = 3.5$ Hz, H_{5'}), 4.02 (t, 1H, $J_{H4'-H5'} = 5.0$ Hz, $J_{H4'-H3'} = 5.0$ Hz, H_{4'}), 3.50-3.45 (m, 1H, OH), 0.99 (t, 9H, $J_{H9'-H8'} = 8.0$ Hz, H_{9'}), 0.89 (s, 9H, -C(CH₃)₃), 0.86 (s, 9H, -C(CH₃)₃), 0.62 (q, 6H, $J_{H8'-H9'} = 8.0$ Hz, H_{8'}), 0.07 (s, 6H, CH₃), 0.05 (s, 3H, CH₃), 0.03 (s, 3H, CH₃); ¹³C NMR δ 162.9 (C₄), 150.4 (C₂), 143.0 (C₆), 104.8 (C_{6'}), 102.4 (C₅), 93.2 (C_{1'}), 89.3 (C₇), 87.6 (C_{4'}), 73.4 (C_{2'}), 72.5 (C_{3'}), 62.5 (C_{5'}), 25.9, 25.9 (-C(CH₃)₃), 18.2, 18.0 (-C(CH₃)₃), 7.5 (C₈), 4.4 (C₉), -4.4, -4.4, -4.3, -4.7 (-C(CH₃)₂); HRMS HRMS ESI⁺ calcd for C₂₉H₅₅N₂O₆Si₃⁺ (M + H)⁺ 611.3368, found 611.3371. **Characteristic signals for (5'R)-14ab:** ¹H NMR δ 9.68 (s, 1H, NH), 7.63 (d, 1H, $J_{H6-H5} = 8.0$ Hz, H₆), 4.30-4.26 (m, 1H, H_{4'}), 4.10-4.09 (m, 1H, H_{2'}).

N³-Allyl-2',3'-di-O-(tert-butyldimethylsilyl)-5'-(triethylsilylethynyl)-uridine (14bb). Compound **14bb** was synthesized according to the general procedure for 5'-uridine aldehydes alkynylation by a dropwise addition of a solution of the crude aldehyde **9b** (synthesized according to the general procedure for 5'-uridine aldehydes preparation from corresponding alcohol **4b**, (400 mg, 0.78 mmol, 1 equiv)) in dry THF (7.8 mL), on a freshly prepared triethylsilylacetylide magnesium bromide solution (1.95 mmol, 2.5 equiv) in THF (7.8 mL). After work up, the crude mixture **14bb** was isolated as a 91:9 mixture of diastereomers. Flash chromatography of the residue (cyclohexane/EtOAc 9:1) afforded **14bb** as a white solid 300 mg, 59% combined yield), the major diastereoisomer **14bb** has been isolated with 39% yield as a viscous oil: R_f 0.38 (cyclohexane/EtOAc 8:2); [α]_D - 13 (c 0.5, CHCl₃); IR (film) 2953, 2929, 2858, 2357, 2342, 1716, 1663, 1457, 1388, 1253, 1102, 932; ¹H NMR δ 7.47 (d, $J_{H6-H5} = 8.1$ Hz, 1H, H₆), 5.84 (ddt, $J_{H8-H9a} = 16.2$ Hz, $J_{H8-H9b} = 10.2$ Hz, $J_{H8-H7a} = J_{H8-H7b} = 6.0$ Hz, 1H, H₈), 5.76 (d, $J_{H5-H6} = 8.1$ Hz, 1H, H₅), 5.61 (d, $J_{H1'-H2'} = 6.9$ Hz, 1H, H_{1'}), 5.31 - 5.23 (m, 1H, H_{9a}), 5.20 - 5.13 (m, 1H, H_{9b}), 4.62 - 4.54 (m, 2H, H_{2'} + H_{5'}), 4.54 - 4.49 (m, 2H, H₇), 4.15 (dd, 1H, $J_{H3'-H2'} = 4.6$ Hz, $J_{H3'-H4'} = 1.6$ Hz, H_{3'}), 4.04 (dd, $J_{H4'-H5'} = 4.2$ Hz, $J_{H4'-H3'} = 1.6$ Hz, 1H, H_{4'}), 3.47 (br s, 1H, OH), 0.97 (t, $J_{H9'-H8'} = 7.9$ Hz, 1H, H_{9'}), 0.91 (s, 9H, -C(CH₃)₃), 0.83 (s, 9H, -C(CH₃)₃), 0.60 (q, $J_{H8'-H9'} = 7.9$ Hz, 6H, H_{8'}), 0.10 (s, 3H, CH₃), 0.09 (s, 3H, CH₃), 0.01 (s, 3H, CH₃), -0.08 (s, 3H, CH₃); δ 162.3 (C₄), 150.8 (C₂), 141.0 (C₆), 131.3 (C₈), 118.5 (C₉), 104.8 (C_{6'}), 102.0 (C₅), 93.5 (C_{1'}), 89.0 (C₇), 88.00 (C_{4'}), 73.1 (C_{2'}), 72.7 (C_{3'}), 62.8 (C_{5'}), 43.17 (C₇), 25.9, 25.8 (-C(CH₃)₃), 18.1, 18.3 (-C(CH₃)₃), , 7.50 (C₉), 4.33 (C₈), -4.3, -4.4, -4.4, -4.8 (-C(CH₃)₂); HRMS ESI⁺ calcd for C₃₂H₅₉N₂O₆Si₃⁺ (M + H)⁺ 651.3675, found 651.3680.

5'(S)-(Trimethylsilylethynyl)-2',3'-di-O-(triisopropylsilyl)-uridine (15aa). Compound **15aa** was synthesized according to the general procedure for the 5'-alkynylation of uridine-derived aldehydes: a dropwise addition of a solution of the crude aldehyde **10a** (synthesized from alcohol **5a**³ (450 mg, 0.80 mmol, 1 equiv) according to the general procedure for the oxidation of uridine derivatives) in dry THF (8.0 mL), on a freshly prepared trimethylsilylacetylide magnesium bromide solution (4.04

mmol, 5 equiv) in THF (8.0 mL). After work up, the crude mixture **15aa** was isolated as a 90:10 mixture of diastereomers. Flash chromatography of the residue (cyclohexane/EtOAc 7:3) afforded **15aa** as a white solid (348 mg, 66% combined yield). The major diastereoisomer (5'*S*)-**15aa** has been isolated with 36% yield as a white solid. R_f 0.39 (cyclohexane/EtOAc 7:3); mp 195 - 199 °C; $[\alpha]_D$ -40 (*c* 0.2, CHCl₃); IR (film) 2944, 2867, 1712, 1681, 1464, 1386, 1262, 1250, 883; ¹H NMR δ 9.28 (br s, 1H, NH), 7.30 (d, $J_{H_6-H_5}$ = 8.0 Hz, 1H, H₆), 5.56 (d, $J_{H_5-H_6}$ = 8.0 Hz, 1H, H₅), 5.42 (d, $J_{H_{1'}-H_{2'}}$ = 7.0 Hz, 1H, H_{1'}), 4.73 (dd, $J_{H_{2'}-H_{1'}}$ = 7.0 Hz, $J_{H_{2'}-H_{3'}}$ = 4.0 Hz, 1H, H_{2'}), 4.51 (t, $J_{H_{5'}-H_{4'}}$ = 5.4 Hz, $J_{H_{5'}-OH'}$ = 5.4 Hz, 1H, H_{5'}), 4.26 (dd, $J_{H_{3'}-H_{2'}}$ = 4.0 Hz, $J_{H_{3'}-H_{4'}}$ = 1.0 Hz, 1H, H_{3'}), 3.92 (dd, $J_{H_{4'}-H_{5'}}$ = 5.4 Hz, $J_{H_{4'}-H_{3'}}$ = 1.0 Hz, 1H, H_{4'}), 3.46 (d, $J_{H_{5'}-OH}$ = 5.4 Hz, 1H, OH), 0.84 - 0.96 (m, 42H, -CH(CH₃)₂), -0.00 (s, 9H, Si(CH₃)₃); ¹³C NMR δ 163.5 (C₄), 150.8 (C₂), 144.1 (C₆), 103.5 (C_{6'}), 102.7 (C₅), 94.1 (C_{1'}), 91.9 (C₇), 87.6 (C_{4'}), 74.2 (C_{3'}), 73.4 (C_{2'}), 62.8 (C_{5'}), 18.46 (-CH(CH₃)₂), 18.42 (-CH(CH₃)₂), 18.22 (-CH(CH₃)₂), 18.08 (-CH(CH₃)₂), 13.15 (-CH(CH₃)₂), 12.84 (-CH(CH₃)₂), 0.00 (-Si(CH₃)₃); HRMS ESI⁺ calcd for C₃₂H₆₁N₂O₆Si₃⁺ (M + H)⁺ 653.3837, found 653.3832.

5'(S)-(Triethylsilylethynyl)-2',3'-di-O-(triisopropylsilyl)-uridine (15ab). Compound **15ab** was synthesized according to the general procedure for the 5'-alkynylation of uridine-derived aldehydes: a dropwise addition of a solution of the crude aldehyde **10a** (synthesized from corresponding alcohol **5a**³ (500 mg, 0.90 mmol, 1 equiv) according to the general the oxidation of uridine derivatives) in dry THF (9.0 mL), on a freshly prepared triethylsilylacetylide magnesium bromide solution (4.50 mmol, 5 equiv) in THF (9.0 mL). After work up, the crude mixture **15ab** was isolated as a 95:5 mixture of diastereoisomers. Flash chromatography of the residue (cyclohexane/EtOAc 7:3) afforded **15ab** as a white solid 406 mg, 65% combined yield). The major diastereoisomer (5'*S*)-**15ab** has been isolated with 32% yield as a white solid. R_f 0.40 (cyclohexane/EtOAc 7:3); mp 216 - 220 °C; $[\alpha]_D$ -19 (*c* 0.2, CHCl₃); IR (film) 3675, 2971, 2900, 1704, 1689, 1463, 1393, 1074, 883; ¹H NMR δ 9.34 (br s, 1H, NH), 7.53 (d, $J_{H_6-H_5}$ = 8.0 Hz, 1H, H₆), 5.75 (dd, $J_{H_5-H_6}$ = 8.0 Hz, J_{H_5-NH} = 2.2 Hz, 1H, H₅), 5.62 (d, $J_{H_{1'}-H_{2'}}$ = 6.8 Hz, 1H, H_{1'}), 4.90 (dd, $J_{H_{2'}-H_{1'}}$ = 6.8 Hz, $J_{H_{2'}-H_{3'}}$ = 4.0 Hz, 1H, H_{2'}), 4.66 (t, $J_{H_{5'}-H_{4'}}$ = 5.5 Hz, $J_{H_{5'}-OH'}$ = 5.5 Hz, 1H, H_{5'}), 4.42 (dd, $J_{H_{3'}-H_{2'}}$ = 4.0 Hz, $J_{H_{3'}-H_{4'}}$ = 1.5 Hz, 1H, H_{3'}), 4.11 (dd, $J_{H_{4'}-H_{5'}}$ = 4.8 Hz, $J_{H_{4'}-H_{3'}}$ = 1.5 Hz, 1H, H_{4'}), 3.54 (d, $J_{H_{5'}-OH}$ = 5.5 Hz, 1H, OH), 1.15 - 1.01 (m, 42H, -CH(CH₃)₂), 0.98 (t, $J_{H_{9'}-H_{8'}}$ = 7.9 Hz, 9H, H_{9'}), 0.61 (q, $J_{H_{8'}-H_{9'}}$ = 7.9 Hz, 6H, H_{8'}); ¹³C NMR δ 163.3 (C₄), 150.4 (C₂), 143.7 (C₆), 104.4 (C_{6'}), 102.3 (C₅), 93.6 (C_{1'}), 89.0 (C₇), 87.4 (C_{4'}), 73.9 (C_{3'}), 73.4 (C_{2'}), 62.6 (C_{5'}), 18.2 (-CH(CH₃)₂), 18.1 (-CH(CH₃)₂), 17.9 (-CH(CH₃)₂), 17.8 (-CH(CH₃)₂), 12.8 (-CH(CH₃)₂), 12.6 (-CH(CH₃)₂), 7.3 (C₉), 4.1 (C₈); HRMS ESI⁺ calcd for C₃₅H₆₇N₂O₆Si₃⁺ (M + H)⁺ 695.4307, found 695.4311.

5'(S)-(Trisopropylsilylethynyl)-2',3'-Di-O-(triisopropylsilyl)uridine (15ac). Compound **15ac** was synthesized according to the general procedure for the 5'-alkynylation of uridine-derived aldehydes: a dropwise addition of a solution of the crude aldehyde **10a** (synthesized according to the general

procedure for 5'-uridine aldehydes preparation from corresponding alcohol **5a**,³ (500 mg, 0.90 mmol, 1 equiv)) in dry THF (9 mL), on a previously prepared triethylsilylacetylide magnesium bromide solution (4.50 mmol, 5 equiv) in THF (9 mL). After work up, the crude mixture **15ac** was isolated as a 99/1 mixture of diastereomers. Flash chromatography of the residue (cyclohexane/EtOAc 8:2) afforded **15ac** as a white solid 404 mg, 61% combined yield), the major diastereoisomer (5'*S*)-**15ac** has been isolated with 61% yield as a white solid: R_f 0.39 (cyclohexane/EtOAc 7:3); mp > 260 °C; $[\alpha]_D - 2$ (*c* 0.5, CHCl₃); IR (film) 3852, 3661, 2971, 2900, 1684, 1405, 1394, 1382, 1241, 1229, 882, 647; ¹H NMR δ 8.26 (br s, 1H, NH), 7.54 (d, $J_{H_6-H_5} = 8.0$ Hz, 1H, H₆), 5.72 (d, $J_{H_5-H_6} = 8.0$ Hz, $J_{H_5-NH} = 2.4$ Hz, 1H, H₅), 5.59 (d, $J_{H_{1'}-H_{2'}} = 6.8$ Hz, 1H, H_{1'}), 4.89 (dd, $J_{H_{2'}-H_{1'}} = 6.8$ Hz, $J_{H_{2'}-H_{3'}} = 4.0$ Hz, 1H, H_{2'}), 4.57 (dd, $J_{H_{5'}-OH'} = 5.8$ Hz, $J_{H_{5'}-H_{4'}} = 3.6$ Hz, 1H, H_{5'}), 4.37 (dd, $J_{H_{3'}-H_{2'}} = 4.0$ Hz, $J_{H_{3'}-H_{4'}} = 1.5$ Hz, 1H, H_{3'}), 4.11 (dd, $J_{H_{4'}-H_{5'}} = 3.6$ Hz, $J_{H_{4'}-H_{3'}} = 1.5$ Hz, 1H, H_{4'}), 3.35 (d, $J_{H_{5'}-OH} = 5.8$ Hz, 1H, OH), 1.11 - 1.01 (m, 63H, (-CH(CH₃)₂)); ¹³C NMR δ 162.7 (C₄), 150.3 (C₂), 143.8 (C₆), 105.2 (C_{6'}), 102.4 (C₅), 93.7 (C_{1'}), 88.0 (C₇), 87.7 (C_{4'}), 74.2 (C_{3'}), 73.6 (C_{2'}), 63.0 (C_{5'}), 18.6 (-CH(CH₃)₂), 18.3 (-CH(CH₃)₂), 18.2 (-CH(CH₃)₂), 18.1 (-CH(CH₃)₂), 18.0 (-CH(CH₃)₂), 13.0 (-CH(CH₃)₂), 12.8 (-CH(CH₃)₂), 11.2 (-CH(CH₃)₂); HRMS ESI⁺ calcd for C₃₈H₇₃N₂O₆Si₃⁺ (M + H)⁺ 737.4771, found 737.4775.

N³-Allyl-2',3'-di-O-(triisopropylsilyl)-5'-(trisopropylsilylethynyl)-uridine (15bc). Compound **15bc** was synthesized according to the general procedure for the 5'-alkynylation of uridine-derived aldehydes: a dropwise addition of a solution of the crude aldehyde **10b** (synthesized according to the general procedure for 5'-uridine aldehydes preparation from corresponding alcohol **5b**, (500 mg, 0.83 mmol, 1 equiv)) in dry THF (8.3 mL), on a freshly prepared triethylsilylacetylide magnesium bromide solution (4.18 mmol, 5 equiv) in THF (9.0 mL). After work up, the crude mixture **15bc** was isolated as a 90:10 mixture of diastereomers. Flash chromatography of the residue (cyclohexane/EtOAc 9:1) afforded **15bc** as a white solid 280 mg, 43% combined yield). The major diastereoisomer **15bc** has been isolated with 27% yield as a yellow solid: R_f 0.54 (cyclohexane/EtOAc 8:2); mp 217-219 °C; $[\alpha]_D - 5$ (*c* 0.5, CHCl₃); IR (film) 3403, 2943, 2865, 1712, 1664, 1459, 1389, 1338, 1140, 938; ¹H NMR δ 7.42 (d, $J_{H_6-H_5} = 8.1$ Hz, 1H, H₆), 5.91 - 5.80 (m, 1H, 1H, H₈), 5.77 (d, $J_{H_5-H_6} = 8.1$ Hz, 1H, H₅), 5.56 (d, $J_{H_{1'}-H_{2'}} = 7.3$ Hz, 1H, H_{1'}), 5.31 - 5.23 (m, 1H, H_{9a}), 5.19 - 5.13 (m, 1H, H_{9b}), 4.97 (dd, $J_{H_{1'}-H_{2'}} = 7.3$ Hz, $J_{H_{2'}-H_{3'}} = 4.0$ Hz, 1H, H_{2'}), 4.58 - 4.48 (m, 3H, H₇ + H_{5'}), 4.37 (dd, $J_{H_{3'}-H_{2'}} = 4.0$ Hz, $J_{H_{3'}-H_{4'}} = 1.5$ Hz, 1H, H_{3'}), 4.09 (dd, $J_{H_{4'}-H_{5'}} = 3.4$ Hz, $J_{H_{4'}-H_{3'}} = 1.5$ Hz, 1H, H_{4'}), 3.75 (d, $J_{H_{5'}-OH} = 6.6$ Hz, 1H, OH), 0.63 - 1.13 (m, 63H, (-CH(CH₃)₂)); ¹³C NMR δ 162.37 (C₄), 150.97 (C₂), 142.23 (C₆), 131.33 (C₈), 118.56 (C₉), 105.43 (C_{6'}), 102.19 (C₅), 94.87 (C_{1'}), 88.05 (C_{4'}), 87.67 (C₇), 74.56 (C_{3'}), 73.11 (C_{2'}), 63.24 (C_{5'}), 43.15 (C₇), 18.63, 18.33, 18.27, 18.11, 18.0, 13.0, 12.7, 11.2 ((-C(CH₃)₃)); HRMS ESI⁺ calcd for C₄₁H₇₇N₂O₆Si₃⁺ (M + H)⁺ 777.5089, found 777.5104.

5'(R)-(Triethylsilylethynyl)uridine (16).

At 0 °C, to a suspension of alcohol (5'R)-**11ab1** (113 mg, 0.27 mmol, 1 equiv.) in water (1 mL), TFA was added dropwise (4 mL). The mixture was stirred at 0 °C for 10 min and then at r.t. for 1 h 30. The mixture was then concentrated to dryness and the residue was purified by flash chromatography (EtOAc) to furnish the triol (5'R)-**16** as a white foam (103 mg, 99% yield): R_f 0.12 (EtOAc); mp 167-169 °C; $[\alpha]_D - 34$ (*c* 0.8, MeOH); IR (film) 3199 br , 2830 w , 1665 s , 1461 m , 1352 m , 1080 s , 756 s ; $^1\text{H NMR}$ (CD_3OD) δ 7.98 (d, 1H, $J_{\text{H}_6\text{-H}_5} = 8.0$ Hz, H₆), 6.02 (d, 1H, $J_{\text{H}_1\text{'-H}_2\text{'}} = 7.0$ Hz, H₁'), 5.68 (d, 1H, $J_{\text{H}_5\text{-H}_6} = 8.0$ Hz, H₅), 4.57-4.55 (m, 1H, H₅'), 4.29 (d, 1H, $J_{\text{H}_3\text{'-H}_2\text{'}} = 6.0$ Hz, H₃'), 4.23 (dd, 1H, $J_{\text{H}_2\text{'-H}_3\text{'}} = 6.0$ Hz, $J_{\text{H}_2\text{'-H}_1\text{'}} = 7.0$ Hz, H₂'), 4.11-4.09 (m, 1H, H₄'), 1.01 (t, 9H, $J_{\text{H}_9\text{'-H}_8\text{'}} = 8.0$ Hz, H₉'), 0.60 (q, 6H, $J_{\text{H}_8\text{'-H}_9\text{'}} = 8.0$ Hz, H₈'); $^{13}\text{C NMR}$ δ 165.9 (C₄), 152.6 (C₂), 142.5 (C₆), 103.1 (C₆'), 102.8 (C₅), 89.3 (C₁'), 89.5 (C₇'), 88.9 (C₄'), 75.6 (C₂'), 71.9 (C₃'), 63.9 (C₅'), 7.8 (C₉'), 5.1 (C₈'); HRMS ESI⁺ calcd for C₁₇H₂₇N₂O₆Si⁺ (M + H)⁺ 383.1633, found 383.1632.

5'(S)-(Triethylsilylethynyl)uridine (16).

At 0 °C, to a suspension of alcohol (5'S)-**11ab1**¹ (68 mg, 0.11 mmol, 1 equiv.) in water (650 μL), TFA was added dropwise (1.65 mL). The mixture was stirred at 0 °C for 10 min and then at r.t. for 18 h. The mixture was then concentrated to dryness and the residue was purified by flash chromatography (EtOAc) to furnish the triol (5'S)-**16** as a white foam (38 mg, 91% yield): R_f 0.12 (EtOAc); mp 132-135 °C; $[\alpha]_D + 12$ (*c* 0.9, MeOH); IR (film) 3201 br , 2842 w , 1667 s , 1452 m , 1356 m , 1080 s , 820 s ; $^1\text{H NMR}$ (CD_3OD) δ 7.98 (d, 1H, $J_{\text{H}_6\text{-H}_5} = 8.0$ Hz, H₆), 5.98 (d, 1H, $J_{\text{H}_1\text{'-H}_2\text{'}} = 6.0$ Hz, H₁'), 5.68 (d, 1H, $J_{\text{H}_5\text{-H}_6} = 8.0$ Hz, H₅), 4.55 (d, 1H, $J_{\text{H}_5\text{'-H}_4\text{'}} = 3.0$ Hz H₅'), 4.25 (t, 1H, $J_{\text{H}_2\text{'-H}_3\text{'}} = 6.0$ Hz, $J_{\text{H}_2\text{'-H}_1\text{'}} = 6.0$ Hz, H₂'), 4.20 (dd, 1H, $J_{\text{H}_3\text{'-H}_2\text{'}} = 6.0$ Hz, $J_{\text{H}_3\text{'-H}_4\text{'}} = 3.0$ Hz, H₃'), 4.05 (t, 1H, $J_{\text{H}_4\text{'-H}_3\text{'}} = 3.0$ Hz, $J_{\text{H}_4\text{'-H}_5\text{'}} = 3.0$ Hz, H₄'), 1.01 (t, 9H, $J_{\text{H}_9\text{'-H}_8\text{'}} = 8.0$ Hz, H₉'), 0.63 (q, 6H, $J_{\text{H}_8\text{'-H}_9\text{'}} = 8.0$ Hz, H₈'); $^{13}\text{C NMR}$ δ 166.2 (C₄), 152.7 (C₂), 142.6 (C₆), 107.5 (C₆'), 103.0 (C₅), 89.8 (C₁'), 88.6 (C₇'), 88.4 (C₄'), 75.7 (C₂'), 71.9 (C₃'), 63.4 (C₅'), 7.8 (C₉'), 5.3 (C₈'); HRMS ESI⁺ Calcd for C₁₇H₂₇N₂O₆Si⁺ (M + H)⁺ 383.1633, found 383.1634.

5'(R)-(ethynyl)-uridine (17).

Compound **17** has been synthesized from alkyne (5'R)-**11ab1**¹ (40 mg, 0.13 mmol, 1 equiv) according to the procedure used for the preparation of (5'S)-**17**. Crystallization of the residue (DCM/MeOH 9/1) furnished (5'R)-**17** as white crystals (32 mg, 92% yield); R_f 0.23 (DCM/MeOH 80/20); mp 224 °C; $[\alpha]_D - 27$ (*c* 0.9, H₂O); IR (film) 3270 s , 2852 m , 1682 s , 1456 m , 1383 m ; $^1\text{H NMR}$ (CD_3OD) δ 8.00 (d, 1H, $J_{\text{H}_6\text{-H}_5} = 8.5$ Hz, H₆), 6.04 (d, 1H, $J_{\text{H}_1\text{'-H}_2\text{'}} = 7.0$ Hz, H₁'), 5.71 (d, 1H, $J_{\text{H}_5\text{-H}_6} = 8.5$ Hz, H₅), 4.53 (t, 1H, $J_{\text{H}_5\text{'-H}_4\text{'}} = J_{\text{H}_5\text{'-H}_7\text{'}} = 2.5$ Hz, H₅'), 4.30 (dd, 1H, $J_{\text{H}_3\text{'-H}_2\text{'}} = 5.5$ Hz, $J_{\text{H}_3\text{'-H}_4\text{'}} = 2.5$ Hz, H₃'), 4.22 (dd, 1H, $J_{\text{H}_2\text{'-H}_1\text{'}} = 7.0$ Hz, $J_{\text{H}_2\text{'-H}_3\text{'}} = 5.5$ Hz, H₂'), 4.07 (t, 1H, $J_{\text{H}_4\text{'-H}_3\text{'}} = J_{\text{H}_4\text{'-H}_5\text{'}} = 2.5$ Hz, H₄'), 2.98 (d, 1H, $J_{\text{H}_7\text{'-H}_5\text{'}} = 2.5$ Hz, H₇'); $^{13}\text{C NMR}$ (CD_3OD) δ 166.2 (C₄), 152.8 (C₂), 142.7 (C₆), 103.2 (C₅), 89.5

(C_{1'}), 89.2 (C_{4'}), 82.8 (C_{6'}), 76.2 (C_{2'}), 75.6 (C_{7'}), 71.9 (C_{2'}), 63.3 (C_{5'}); HRMS ESI⁻ calcd for C₁₁H₁₁N₂O₆⁺ (M + H)⁻ 267.0626, found 267.0623.

5'(S)-(ethynyl)-uridine (17).

On a solution of alcohol (5'S)-**11ab**¹ (56 mg, 0.132 mmol, 1 equiv.) in MeOH (1,5 mL) was added dropwise at 0 °C a solution 1.5 M of sodium hydroxide. After 5 h, a saturated aqueous solution of NH₄Cl was added dropwise and MeOH was removed in vacuo. The aqueous phase was then extracted with EtOAc and the combined organic layers were dried over Na₂SO₄, filtered and concentrated in vacuo. The crude terminal alkyne (40 mg, 0.130 mmol) was crystallised and immediately deprotected. To a suspension of terminal alkyne in pure H₂O (0.7 mL), TFA (2.8 mL) was added dropwise at 0 °C. The pale yellow resulting solution was stirred at r.t. (TLC monitoring) and then concentrated to dryness. Flash chromatography of the residue (DCM/MeOH 95/5 to 90/10) afforded (5'S)-**17** as a white solid (18.6 mg, 72% yield); R_f 0.23 (DCM/MeOH 80/20); mp 220-222 °C; [α]_D + 40 (c 0.1, MeOH); IR (film) 3270_{br}, 2852_m, 1682_s, 1456_m, 1383_m; ¹H NMR (CD₃OD) δ 8.00 (d, 1H, J_{H6-H5} = 8.0 Hz, H₆), 5.98 (d, 1H, J_{H1'-H2'} = 6.0 Hz, H_{1'}), 5.72 (d, 1H, J_{H5-H6} = 8.0 Hz, H₅), 4.54 (dd, 1H, J_{H5'-H4'} = 3.5 Hz, J_{H5'-H7'} = 2.5 Hz, H_{5'}), 4.23 (t, 1H, J_{H2'-H1'} = J_{H2'-H3'} = 6.0 Hz, H_{2'}), 4.20 (dd, 1H, J_{H3'-H2'} = 6.0 Hz, J_{H3'-H4'} = 3.5 Hz, H_{3'}), 4.03 (t, 1H, J_{H4'-H3'} = J_{H4'-H5'} = 3.5 Hz, H_{4'}), 2.91 (d, 1H, J_{H7'-H5'} = 2.5 Hz, H_{7'}); ¹³C NMR (CD₃OD) δ 166.2 (C₄), 152.7 (C₂), 142.7 (C₆), 103.1 (C₅), 90.0 (C_{1'}), 88.4 (C_{4'}), 83.7 (C_{6'}), 75.4 (C_{3'}), 75.2 (C_{7'}), 71.9 (C_{2'}), 62.7 (C_{5'}); HRMS ESI⁻ calcd for C₁₁H₁₁N₂O₆⁺ (M + H)⁻ 267.0626, found 267.0623.