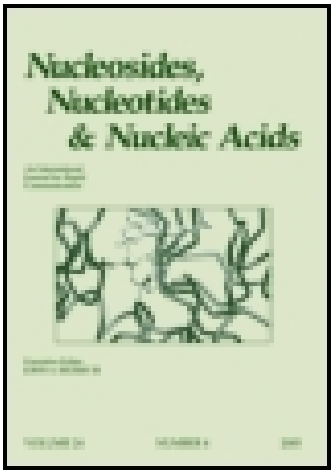


Nucleosides, Nucleotides and Nucleic Acids

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/lncn20

Pyrimidine-Purine and Pyrimidine Heterodinucleosides Synthesis Containing a Triazole Linkage

R. Lucas [a] , P. H. Elchinger [a] , P. A. Faugeras [a] & R. Zerrouki [a]

[a] Université de Limoges, Laboratoire de Chimie des Substances Naturelles, Faculté des Sciences et Techniques , 123, Limoges, France

Published online: 19 Apr 2010.

To cite this article: R. Lucas , P. H. Elchinger , P. A. Faugeras & R. Zerrouki (2010) Pyrimidine-Purine and Pyrimidine Heterodinucleosides Synthesis Containing a Triazole Linkage, Nucleosides, Nucleotides and Nucleic Acids, 29:3, 168-177, DOI: 10.1080/15257771003708579

To link to this article: http://dx.doi.org/10.1080/15257771003708579

Nucleosides, Nucleotides and Nucleic Acids, 29:168–177, 2010

ISSN: 1525-7770 print / 1532-2335 online
DOI: 10.1080/15257771003708579

Taylor & Francis
Taylor & Francis Group

PYRIMIDINE-PURINE AND PYRIMIDINE HETERODINUCLEOSIDES SYNTHESIS CONTAINING A TRIAZOLE LINKAGE

R. Lucas, P. H. Elchinger, P. A. Faugeras, and R. Zerrouki
Université de Limoges, Laboratoire de Chimie des Substances Naturelles, Faculté des Sciences et Techniques, 123Limoges, France

□ *This article describes a synthetic route to generate two purine-pyrimidine and pyrimidine heterodinucleosides. Both microwave activated regioselective alkylation using hydride and copper-catalyzed-azide-alkyne-cycloaddition (CuAAC) were used in order to perform the synthesis.*

Keywords Oligonucleotides analogues; click chemistry; microwave-activated synthesis

INTRODUCTION

Synthetic oligonucleotide analogues are of considerable importance because of their ability to inhibit gene expression through the antisense effect.[1] One of the most important modifications is the complete substitution of the phosphate internucleoside bridge,[2] in order to achieve stronger affinity for the nucleic acid target, enhanced resistance, and improved membrane permeability and cellular uptake. The Huigsen 1,3-dipolar cycloaddition between azides and alkynes, performed by recently discovered copper (I) catalysis[3] appears as a promising way to generate oligomers and particularly oligonucleotide analogues, which can resist chemical or enzymatic depolymerization. Moreover the exclusive regioselective formation of 1,4-disubstituted-1,2,3-triazole conserves the directional character of DNA strands. In addition the introduction of a **dT-A** or pyrimidine-purine step into a straight and rigid A-tract of oligonucleoside can cause a positive roll deformation that kinks the DNA helix at this step.[4] In this way, our research effort in this area has focused on the synthesis of an anti HIV-1 sequence (Figure 1)[5] and the introduction of a **dT-A** step in this sequence. Previous studies, introducing a triazole linkage in oligonucleosides backbone, have been realized using thymidine as nucleoside (homothymidines) in both

Received 4 February 2010; accepted 10 February 2010.
Address correspondence to R. Zerrouki, Université de Limoges, Laboratoire de Chimie des Substances Naturelles, Faculté des Sciences et Techniques, 123, Av. Albert Thomas, 87060 Limoges, France. E-mail: Rachida.zerrouki@unilim.fr

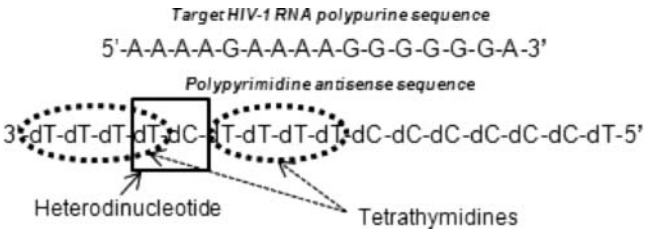


FIGURE 1 Anti HIV-1 sequence.

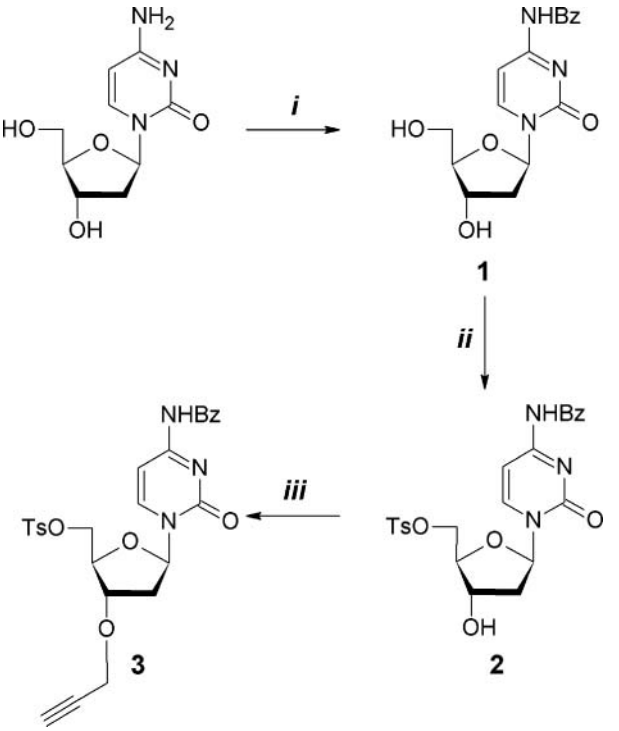
solution-phase and solid phase synthesis.[6] However, no other pyrimidine or purine nucleosides have been inserted in aoligonucléoside sequences using a "click backbone."

In this article, we are interested in the feasibility of coupling two different nucleosides by click chemistry and synthesis of two hetero-dinucleoside analogues (**dC-dT** and **dT-A**) in which glycosidic moieties are linked by a triazole group between positions 3′ and 5′.

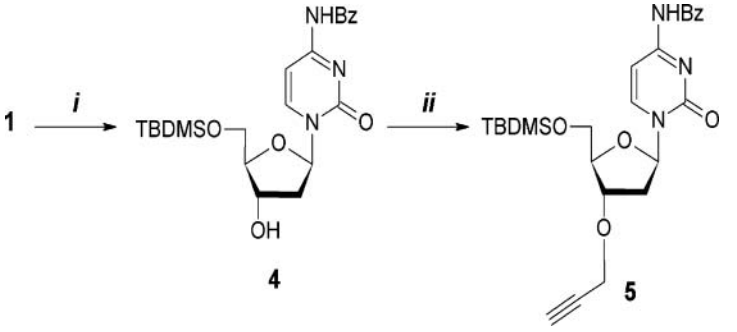
RESULTS AND DISCUSSION

Toward a Cytidine-Thymidine heterodimer (dC-dT)

The synthesis of the first precursor **3** began benzoylating commercially available 2′-deoxycytidine, using an established transient protection method,[7] in order to protect the exo-cyclic amine on the nucleobase to give compound **1** (87%). Then tosylation[8] of resultant N^4-benzoyl-2′-deoxycytidine **1** using tosyl chloride (1.6 equiv.) and standard conditions in pyridine, at 0°C produced compound **2** in low yield (49%; Scheme 1). In addition to 5′-*O*-tosyldeoxycytidine, 3′,5′-di-*O*-tosyldeoxycytidine also is observed. This result strongly suggest that the slight difference in reactivity between 5′ and 3′ positions is the cause of relative low yield of tosylation of primary hydroxyl group. Alkyne **3** was obtained by regioselective 3′-*O*-alkylation using a two-step method.[9] During the first step compound **2** was activated with 2.5 equivalents of NaH in THF, then, for the second step, 2.5 equivalents of propargyl bromide was added and the mixture was activated again. No evolution of hydrogen gas was observed and a large amount of reactant (5 equiv.) was necessary in order to obtain the compound **3** in 51% yield. This strategy is not efficient to prepare compound **3** because the over yield is very low (22% from deoxycytidine). Finally, for obtention of the alkyne compound, we decided to use another strategy (Scheme 2). Precursor **5** was synthesized from N^4-benzoyl-2′-deoxycytidine **1**, first by selective protection of 5′-hydroxyl group with *tert*-butyldimethylsilyl chloride (1.6 equiv.) in dry dimethylformamide (DMF) in the presence of imidazole (3.0 equiv.) giving compound **4** in 84% yield.[10] The 3′-hydroxyl group was

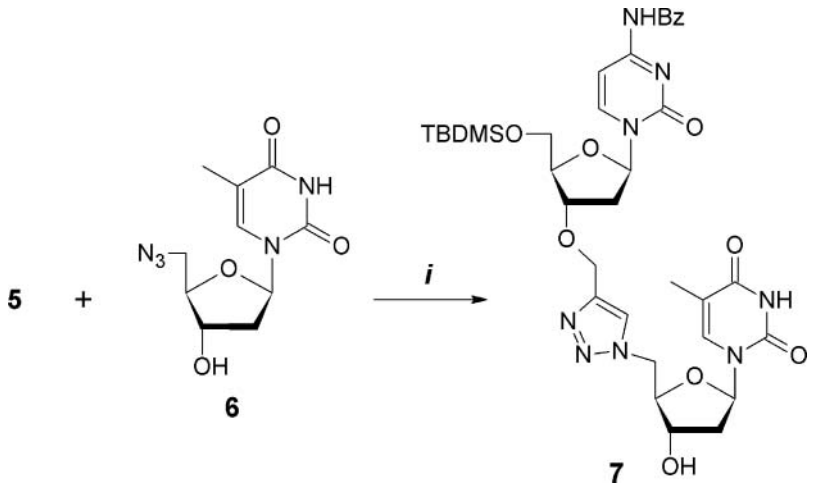


SCHEME 1 Synthesis of propargylated 2′-deoxycytidin precursor (**3**). Reagents and conditions: (i) 1-Me_3SiCl (5 equiv.), 2-BzCl (5 equiv.), Py (10 mL); (ii) TsCl (1.6 equiv.), Py, 0°C; (iii) 1-NaH (2.5 equiv.), THF, MW (2 minutes, 200 W, 40°C) 2-propargylbromide (2.5 equiv.); (6 minutes, 200 W, 40°C).



SCHEME 2 Synthesis of the second propargylated 2′-deoxycytidine precursor (**5**). Reagents and conditions: (i) Imidazole (3 equiv.), TBDMSCl (1.6 equiv.), DMF; (ii) 1-NaH (2.5 equiv.), THF, MW (2 minutes, 200 W, 40°C) 2-propargylbromide (2.5 equiv.), MW (6 minutes, 200 W, 40°C).

then alkylated using Chattopadhyaya's method[9] with NaH (2.5 equiv.) and propargylbromide (2.5 equiv.) in THF under MW activation to give alkyne compound **5** in 91% yield (67% from deoxycytidine). The azido-precursor was 5′-azido-5′-deoxythymidine.[6] Tosylation of thymidine followed by an azidation gave the expected precursor **6** in a two steps good yield of 67% from thymidine.

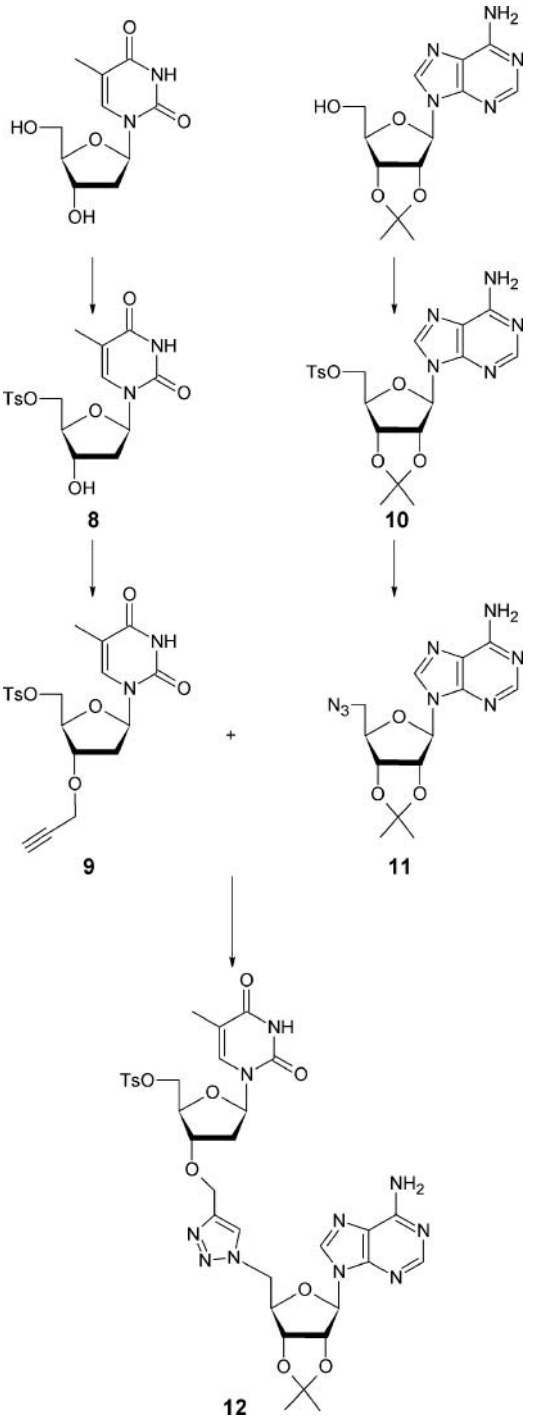


SCHEME 3 Synthesis of the **dC-dT** heterodinucleotide analogue (**7**). Reagents and conditions: (i) $CuSO_4$ (0.1 equiv.), Na ascorbate (0.6 equiv.), H_2O/EtOH (1/1), MW (1 minutes, 200 W, 80°C).

Derivatives **5** and **6** were then coupled using copper(I)-catalyzed cycloaddition in order to obtain silylated heterodimer **7** (Scheme 3). Optimization of this reaction has been described in a previous article.[11] Alkyne and azide precursors were suspended in a 1:1 mixture of water and ethanol, together with Cu(I) catalyst and sodium ascorbate. After 3 minutes of MW irradiation the heterodimer **7** was isolated in 70% yield.

Synthesis of an Thymidine-Adenosine heterodimer (dT-A)

The synthesis of hetero-dinucleoside analogue **dT-A** is presented in Scheme 4. The first precursor synthesized was 3′-O-propargyl-5′-O-tosylthymidine. Selective tosylation of the primary hydroxyl group of thymidine gave the already known product **8.** Alkyne **9** was obtained by regioselective 3′-O-alkylation using Chattopadhyaya's two-step method under microwave irradiation. During the first step compound **8** was activated (40°C, 200 W, 3 minutes) with 2.5 equivalents of NaH in THF then in the second step 2.5 equivalents of propargyl bromide was added and the mixture was subjected to microwave irradiation for 3 minutes. The reaction produced compound **9** in 96% yield.[6] The synthesis of second precursor **11** began with tosylating commercially available 2′,3′-*O*-isopropylidene adenosine. Then using previous method used for hydroxyl group protection, the protected compound **10** was synthesized in moderate yield (50%). In addition to 5′-O-tosyl-2′,3′-O-isopropylidene adenosine, cycloadenosine **10′** was also is observed (Figure 2). Compound **10′** results from the intramolecular nucleophilic displacement by N^3 (Figure 2). It was reported that, upon rotation of the adenine ring of 5′-protected adenosine, the N^3 of the base can



SCHEME 4 Synthesis of the **dT-A** heterodinucleotide analogue (**12**).

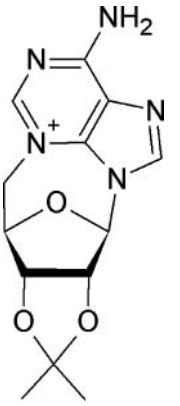


FIGURE 2 Cycloadenosine **10′**.

readily displace the 5′-activation to give the ionic cycloadenosine, causing a steep decrease of the overall yield of **10**.[12] In fact, intramolecular displacement of sulfonate is so rapid that it interferes with product isolation and purification.

Derivatives **9** and **11** were then clicked using copper(I)-catalyzed cycloaddition in order to obtain silylated heterodimer **12**. Alkyne and azide precursors were suspended in a 1:1 mixture of water and ethanol, together with Cu(I) catalyst and sodium ascorbate. After 3 minutes of irradiation, heterodimer **12** was obtained after purification in 87% yield. The tosyl group in 5′ of the **dT-A** could be used for further azidation in order to generate an oligonucléotides analogue.

CONCLUSION

In conclusion we have investigated the first total synthesis of two triazole heterodimers (**dC-dT** and **dT-A**) using a "click chemistry" reaction and MW irradiation to reduce reaction time. This work is presently continuing toward solid-phase synthesis of a pyrimidine antisense sequence with promising results.

EXPERIMENTAL

All the solvents and chemicals were commercially available and, unless otherwise stated, were used as received. Reactions were monitored by thin layer chromatography (TLC) on precoated 0.2 mm silica gel 60 F_{254} (Merck) plates and visualized in several ways: with an ultraviolet light source at 254 nm, by spraying with sulphuric acid (6N) and heating to 200°C. Microwave irradiations were performed by the means of an Ethos 1600 MicroSynth reactor from Milestone. Temperature was measured with a fiber optic thermometer (ATC-FO)/Ethos. ^{1}H NMR spectra were recorded at 400.13 MHz with a

Brüker DPX spectrometer (Germany). Chemical shifts (δ) are expressed in ppm with Me_4Si as an internal standard ($\delta = 0$). Data are reported as follows: chemical shift, multiplicity (s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet and br, broad), coupling constants (Hz) and assignment.

Tosylation of N^4-Benzoyl-2′-deoxycytidine

N^4-benzoyl-2′-deoxycytidine **1** (223 mg, 0.68 mmol) was solubilized in anhydrous pyridine with 1.6 equivalents of tosyl chloride (206 mg, 1.08 mmol). This solution was placed under argon at 0°C during 3 hours. Then solvent was removed under reduced pressure and the crude residue purified using flash chromatography (CH_2Cl_2/petroleum ether, from 8/2 to pure petroleum ether). Pure product **2** was recovered as a white solid in 49% yield (160 mg).

Silylation of N^4-Benzoyl-2′-deoxycytidine

N^4-benzoyl-2′-deoxycytidine **1** (150 mg, 0.45 mmol) was solubilized in anhydrous DMF with 3.0 equivalents of imidazole (92.5 mg, 1.36 mmol) and 1.6 equivalents of *tert*-butyldimethylsilyl chloride (109 mg, 0.73 mmol). This solution was placed under argon at 0°C during 3 hours. Then solvent was removed under reduced pressure and the crude residue purified using flash chromatography. Pure product **4** was recovered as a white solid in 84% yield (184 mg).

General Procedure for Propargylation

To compound **4** (50 mg, 0.10 mmol) in dry THF (15 mL), were added 2.5 equivalents of NaH (60%, 10 mg, 0.26 mmol) and the mixture was activated by microwave irradiation (2 minutes, 200 W, 40°C). Two and half equivalents of propargyl bromide (30 μL, 0.26 mmol) were then added and the mixture was activated by microwave irradiation (6 minutes, 200W, 40°C). After work-up (NH_4Cl/H_2O), the product was purified by chromatography (thick layer plates, AcOEt/MeOH (9/1)). Compound **5** was recovered in 91% yield (39 mg).

General Procedure for Click Reaction

Compounds **9** (217 mg, 0.50 mmol), **11** (167 mg, 0.50 mmol), sodium ascorbate (300 μL, 1 M, 0.6 equiv.), and copper sulfate pentahydrate (7.5 mg, 0.1 equiv.) were suspended in a 1:1 mixture of water and ethanol (4 mL each) in a 25 mL bicol (open reaction vessel). The mixture was then irradiated for 1 minutes at 80°C, using an irradiation power of 200 W. After work-up (THF), the crude residue was concentrated in vacuo and product **12** was recovered as a white solid in 85% yield (326.2 mg).

CHARACTERIZATIONS

Spectroscopic data of selected new compounds:

Compound ***4****:* R_f = 0.52 (AcOEt/EtOH: 9/1); ^{1}H NMR (DMSO d_6): *cytidine*: 11.22 (s, 1H, NH), 8.31 (br s, 1H, H_5), 7.36 (br d, J = 6.8 Hz, H_5); *benzoyl:* 8.00 (dd, 2H, J = 1.2 Hz, J = 8.5 Hz, H_{2-6}), 7.62 (br t, 1H, J = 7.5 Hz, H_4), 7.51 (br t, 2H, J = 7.5 Hz, H_{3-5}); *ose*: 6.13 (t, 1H, J = 6.1 Hz, $H_{1'}$), 5.33 (d, 1H, J = 4.4 Hz, 1H, $O_{3'}$-H), 4.22 (m, 1H, $H_{3'}$), 3.93 (br dd, 1H, J = 3.1 Hz, J = 11.5 Hz, $H_{5'a}$), 3.87 (dd, 1H, J = 3.3 Hz, J = 11.5 Hz, $H_{5'b}$), 2.36 (ddd, 1H, J = 4.8 Hz, J = 6.1 Hz, J = 13.3 Hz, $H_{2'a}$), 2.07 (dt, 1H, J = 6.1 Hz, J = 13.3 Hz, $H_{2'b}$); *TBDMS*: 0.88 (s, 9H, C-CH_3), 0.09 (s, 6H, Si-CH_3); ^{13}C NMR (DMSO d_6): *thymine*: 162.9 (C4), 154.3 (C2), 144.4 (C6), 95.8 (C5); *benzoyl:* 167.3 (C=O), 133.1 (C1), 132.7 (C4), 128.4 (C2, C3, C5, C6); *ose*: 87.3 (C4′), 86.2 (C1′), 69.6 (C3′), 62.6 (C5′), 41.2 (C2′); *TBDMS*: 25.7 (C—$\underline{C}H_3$), 17.9 ($\underline{C}$—CH_{3a}), −5.5 (Si—CH_{3a}), −5.6 (Si—CH_{3b}).

Compound ***5****:* R_f = 0.45 (AcOEt/MeOH: 9/1); ^{1}H NMR (DMSO d_6): *cytidine*: 11.26 (s, 1H, NH), 8.28 (br s, 1H, H_5), 7.36 (br d, J = 7.4 Hz, H_5); *benzoyl:* 8.01 (dd, 2H, J = 7.2 Hz, J = 1.2 Hz, H_{2-6}), 7.62 (br t, 1H, J = 7.4 Hz, H_4), 7.51 (br t, 2H, J = 7.6 Hz, H_{3-5}); *ose*: 6.09 (br t, 1H, J = 6.4 Hz, $H_{1'}$), 4.27 (m, 1H, $H_{3'}$), 4.14 (br dd, 1H, J = 3.1 Hz, J = 6.2 Hz, $H_{4'}$), 3.87 (br dd, 1H, J = 3.7 Hz, J = 11.4 Hz, $H_{5'a}$), 3.78 (br dt, 1H, J = J = 3.3 Hz, 11.4 Hz, $H_{5'b}$), 2.54 (ddd, 1H, J = 3.1 Hz, J = 6.1 Hz, J = 13.3 Hz, $H_{2'a}$), 2.14 (dt, 1H, J = 6.3 Hz, J = 13.3 Hz, $H_{2'b}$); *TBDMS*: 0.89 (s, 9H, C—CH_3), 0.10 (s, 6H, Si—CH_{3a}), 0.09 (s, 6H, Si—CH_{3a}); *propargyl*: 4.23 (br d, 1H, J = 2.3 Hz, J = 18,0 Hz, CH_2), 3.47 (t, 1H, J = 2.3 Hz, C—H); ^{13}C NMR (DMSO d_6): *thymine*: 163.0 (C4), 154.2 (C2), 144.3 (C6), 95.9 (C5); *benzoyl:* 167.3 (C=O), 133.1 (C1), 132.7 (C4), 128.4 (C2, C3, C5, C6); *ose*: 86.5 (C1′), 84.5 (C4′), 77.7 (C3′), 62.8 (C5′), 40.4 (C2′); *TBDMS*: 25.7 (C—$\underline{C}H_3$), 17,9 ($\underline{C}$—CH_3), −5.6 (CH_{3a}), −5.6 (CH_{3b}); *propargyl*: 80.0 (C), 77.4 (CH), 55.9 (CH_2); MS (IS) m/z = 484.4 (MH^+), 506.4 (MNa^+), 989,7 (M_2Na^+).

Compound ***7****:* R_f = 0.20 ($CHCl_3$/EtOH: 9/1); ^{1}H NMR (DMSO d_6): 11.29 (s, 2H, NH), 2.52–2.06 (m, 4H, $H_{2'\ \text{cytidine and thymine}}$); *cytidine*: 8.28 (d, 1H, J = 7.4 Hz, H_5), 7.34 (d, 1H, J = 7.4, H_6); *ose*: 6.10 (t, 1H, J = 6.4, $H_{1'}$), 4.29 (m, 1H, $H_{3'}$), 4.14 (m, 1H, $H_{4'}$), 3.85 (dd, 1H, J = 3.5, J = 11.5, $H_{5'}$), 3.75 (dd, 1H, J = 3.2, J = 11.5, $H_{5'}$); *TBDMS*: 0.86 (s, 9H, C—$(CH_3)_3$), 0.07 (s, 3H, Si—CH_{3a}), 0.06 (s, 3H, Si—CH_{3b}); *benzoyl*: 8.01 (dd, 2H, J = 1.2 Hz, J = 7.4 Hz), 7.62 (br. t, 1H, J = 7.4 Hz), 7.51 (br. t, 2H, J = 7.7 Hz); *linker*: 8.09 (s, 1H, $H_{triazole}$), 4.57 (d, 2H, J = 12.1, $CH_{2triazole}$); *thymine*: 7.35 (s, 1H, H_6), 1.80 (s, 3H, CH_3); *ose*: 6.15 (t, 1H, J = 6.9, $H_{1'}$), 5.50 (br. s, 1H, OH), 4.71 (dd, 1H, J = 4.3 Hz, J = 14.4 Hz, $H_{5'}$), 4.70 (dd, 1H, J = 3.4 Hz, J = 14.4 Hz, $H_{5'}$), 4.20 (m, 1H, $H_{3'}$), 4.08 (m, 1H, $H_{4'}$); ^{13}C NMR (DMSO d_6), 41.4 ($C2'_{\text{cytidine or thymidine}}$), 37.5 ($C2'_{\text{cytidine or thymidine}}$); *cytidine*: 163.0 (C4), 154.2 (C2), 144.3 (C6), 95.9 (C5); *ose*: 86.5 (C1′), 84.9 (C4′), 78.1 (C3′); *benzoyl*: 167.2 (C=O), 133.1 (C1), 132.7 (C4), 128.4 (C2, C3, C5, C6); *TBDMS*: 25.7 (C—$\underline{C}H_3$), 17.9 ($\underline{C}$—CH_3);

linker: 143.7 ($C_{triazole}$), 124.7 ($\underline{C}H_{triazole}$), 61.8 ($CH_{2triazole}$), −5.6 ($CH_{3a}$), −5.7 ($CH_{3b}$); *thymine*: 163.6 (C4), 150.4 (C2), 136.0 (C6), 109.8 (C5), 12.1 (CH_3); *ose*: 84.0 (C1′), 83.9 (C4′), 79.1 (C3′).

*Compound **10**:* R_f = 0.21 ($CHCl_3$/EtOH: 9/1); ^{1}H NMR (DMSO d_6): *adenine*: 8.21 (s, 1H, H_8), 8.03 (s, 1H, H_2), 7.36 (br s, NH_2); *ose*: 6.18 (d, 1H, J = 1.8 Hz, $H_{1'}$), 5.33 (dd, 1H, J = 1.8 Hz, J = 6.2 Hz, $H_{2'}$), 5.00 (m, 1H, $H_{3'}$), 4.33 (m, 1H, $H_{4'}$), 4.33 (dd, 1H, J = J = 3.9 Hz, 12.2 Hz, $H_{5'a}$), 4.22 (dd, 1H, J = 2.8 Hz, J = 7.0 Hz, $H_{5'b}$); *isopropylidene:* 1.51 (s, 3H, CH_{3a}), 1.29 (s, 1H, CH_{3b}); *tosyl:* 7.56 (d, 2H, J = 8.2 Hz, $H_{ortho/S}$), 7.25 (d, 2H, J = 8.2 Hz, $H_{meta/S}$), 2.36 (s, 3H, CH_3).

*Compound **11**:* R_f = 0.50 (CH_2Cl_2/EtOH: 9/1); ^{1}H NMR (DMSO d_6): *adenine*: 8.35 (s, 1H, H_8), 8.18 (s, 1H, H_2), 7.35 (s, NH_2); *ose*: 6.22 (d, 1H, J = 2.6 Hz, $H_{1'}$), 5.52 (dd, 1H, J = 2.6 Hz, J = 6.2 Hz, $H_{2'}$), 5.01 (dd, 1H, J = 3.0 Hz, J = 6.2 Hz, $H_{3'}$), 4.31 (m, 1H, $H_{4'}$), 3.63 (dd, 1H, J = 7.2 Hz, 13.0 Hz, $H_{5'a}$), 3.55 (dd, 1H, J = 4.9 Hz, J = 13.0 Hz, $H_{5'b}$); *isopropylidene:* 1.55 (s, 3H, CH_{3a}), 1.13 (s, 1H, CH_{3b}); ^{13}C NMR (DMSO d_6): *adenine*: 156.2 (C6), 152.8 (C2), 148.8 (C4), 140.0 (C8), 119.2 (C5); *ose*: 84.3 (C1′), 80.6 (C2′), 77.8 (C3′), 69.9 (C4′), 35.3 (C5′); *isopropylidene:* 113.5 (C), 26.9 (CH_{3a}), 25.1 (CH_{3b}).

*Compound **12**:* R_f = 0.27 ($CHCl_3$/EtOH: 9/1); ^{1}H NMR (DMSO d_6): *adenine*: 8.28 (s, 1H, H_8), 8.19 (s, 1H, H_2), 7.36 (s, NH_2); *ose*: 6.23 (d, 1H, J = 2.0 Hz, $H_{1'}$), 5.45 (dd, 1H, J = 2.0 Hz, J = 6.2 Hz, $H_{2'}$), 5.15 (dd, 1H, J = 3.4 Hz, J = 6.2 Hz, $H_{3'}$), 4.77 (dd, 1H, J = 5.4 Hz, J = 14.1 Hz, $H_{5'b}$), 4.67 (dd, 1H, J = 7.7 Hz, 14.1 Hz, $H_{5'a}$), 4.55 (m, 1H, $H_{4'}$); *isopropylidene:* 1.53 (s, 3H, CH_{3b}), 1.31 (s, 1H, CH_{3a}); *linker*: 7.95 (s, 1H, $H_{triazole}$), 4.47 (d, 2H, J = 12.4, $CH_{2triazole}$); *thymine*: 11.31 (s, 1H, NH), 7.39 (s, 1H, H_6), 1.77 (s, 3H, CH_3); *ose*: 6.08 (d, 1H, J = 6.8 Hz, J = 7.4 Hz, $H_{1'}$), 4.26 (dd, 1H, J = 3.5 Hz, J = 10.9 Hz, $H_{5'b}$), 4.20 (dd, 1H, J = 5.5 Hz, J = 10.9 Hz, $H_{5'a}$), 4.13 (ddd, 1H, J = 3.0 Hz, J = 5.5 Hz, J = 6.0 Hz, $H_{3'}$), 4.06 (ddd, 1H, J = 3.2 Hz, J = 5.4 Hz, J = 6.2 Hz, $H_{4'}$), 2.19 (m, 1H, $H_{2'}$); *tosyl:* 7.78 (d, 2H, J = 8.2 Hz, $H_{ortho/S}$), 7.45 (d, 2H, J = 8.2 Hz, $H_{meta/S}$), 2.39 (s, 3H, CH_3); ^{13}C NMR (DMSO d_6): *adenine*: 156.2 (C6), 152.8 (C2), 148.6 (C4), 140.1 (C8), 119.2 (C5); *ose*: 89.0 (C1′), 84.5 (C4′), 83.3 (C2′), 81.5 (C3′), 51.0 (C5′); *isopropylidene:* 113.7 (C), 26.9 (CH_{3a}), 25.2 (CH_{3b}); *linker:* 143.5 ($C_{triazole}$), 124.5 ($\underline{C}H_{triazole}$), 62.0 ($CH_{2triazole}$); *thymine:* 163.6 (C4), 150.3 (C2), 135.9 (C6), 109.9 (C5); *ose:* 84.3 (C1′), 80.8 (C4′), 78.2 (C3′), 70.1 (C5′), 35.6 (C2′), 12.1 (CH_3); *tosyl:* 145.1 ($C_{p/S}$), 132.1 ($C_{ipso/S}$), 130.2 ($C_{m/S}$), 127.6 ($C_{o/S}$), 21.1 (CH_3).

REFERENCES

1. a) Abes, S.; Moulton, H.M.; Clair, P.; Prevot, P.; Youngblood, D.S.; Wu, R.P.; Iversen, P.L.; Lebleu, B. Vectorization of morpholino oligomers by the (R-Ahx-) 4 peptide allows efficient splicing correction in the absence of endosomolytic agents. *J. Control. Rel.* **2006**, 116, 304–313; b) Helene, C.; Saison-Behmoaras, The antisense strategy: New therapeutic approaches [La strategie antisens: nouvelles approaches therapeutiques]. *E. Med./Sci.* **1994**, 10, 253–273.

2. a) Pérez-Rentero, S.; Alguacil, J.; Robles, J. Novel oligonucleotide analogues containing a morpholinoamidine unit. *Tetrahedron* **2009**, 65, 1171–1179; b) Michel, T.; Martinand-Mari, C.; Debart, F.; Lebleu, B.; Robbins, I.; Vasseur, J.-J. Cationic phosphoramidate α-oligonucleotides efficiently target single-stranded DNA and RNA and inhibit hepatits C virus IRES-mediated translation. *Nucleic Acids Res.* **2003**, 31, 5282–5290; c) Rozners, E.; Katkevica, D.; Bizdena, E.; Strömberg, R. Synthesis and properties of RNA analogues having amides as interuridine linkages at selected positions. *J. Am. Chem. Soc.* **2003**, 125, 12125–12136.
3. a) Tornoe, C.W.; Meldal, M. Peptidotriazoles: Copper(I)-catalysed 1,3-dipolar cycloadditions on solid-240 phase, in *Peptides 2001, Proceedings of the American Peptide Symposium*, ed. M. Lebl and R.A. Houghten, American Peptide Society and Kluwer Academic Publishers, San Diego, 2001, pp 263–264; b) Rostovtsev, V.V.; Green, L.G.; Fokin, V.V.; Sharpless, K.B. A stepwise huisgen cycloaddition process: Copper(I)-catalyzed regioselective "ligation" of azides and terminal alkynes. *Angew. Chem. Int. Ed.* **2002**, 41, 2596–2599; c) Tornoe, C.W.; Christensen, C.; Meldal, M. Peptidotriazoles on solid phase: [1,2,3]-Triazoles by regiospecific copper(I)-catalyzed 1,3-dipolar cycloadditions of terminal alkynes to azides. *J. Org. Chem.* **2002**, 67, 3057–3064; d) Helms, B.; Mynar, J.L.; Hawker, C.J.; Frechet, J.M. J.; Sharpless, K.B. Dendronized linear polymers via "click chemistry." *J. Am. Chem. Soc.* **2004**, 126, 15020–15021; e) Wu, P.; Feldman, A.K.; Nugent, A.K.; Hawker, C.J.; Scheel, A.; Voit, B.; Pyun, J.; Frechet, J.M.J.; Sharpless, K.B.; Fokin, V.V. Efficiency and fidelity in a click-chemistry route to triazole dendrimers by the copper(I)-catalyzed ligation of azides and alkynes. *Angew. Chem. Int. Ed.* **2004**, 43, 2938–2954.
4. Mack, D.R.; Chiu, T.K.; Dickerson, R.E. Intrinsic bending and deformability at the T-A step of CCTTTAAAGG: A comparative analysis of T-A and A-T steps within A-tracts. *J. Mol. Biol.* **2001**, 312, 1037–1049.
5. Hiratou, T.; Tsukahara, S.; Miyano-Kurosaki, N.; Takai, K.; Yamamoto, N.; Takaku, H. Inhibition of HIV-1 replication by a two-strand system (FTFOs) targeted to the polypurine tract. *FEBS Lett.* **1999**, 456, 186–190.
6. a) Nuzzi, A.; Massi, A.; Dondoni, A. Model studies toward the synthesis of thymidine oligonucleotides with triazole internucleosidic linkages via iterative Cu(I)-promoted azide-alkyne ligation chemistry. *QSAR Comb. Sci.* **2007**, 26, 1191–1199; b) Lucas, R.; Zerrouki, R.; Granet, R.; Krausz, P.; Champavier, Y. A rapid efficient microwave-assisted synthesis of a 3',5'-pentathymidine by copper(I)-catalyzed [3+2] cycloaddition. *Tetrahedron* **2008**, 64, 5467–5471; c) Isobe, H.; Fujino, T.; Yamazaki, N.; Guillot-Nieckowski, M.; Nakamura, E. Triazole-linked analogue of deoxyribonucleic acid (TLDNA): Design, synthesis, and double-strand formation with natural DNA. *Org. Lett.* **2008**, 10, 3729–3732; d) Fujino, T.; Yamazaki, N.; Hiroyuki, I. Convergent synthesis of oligomers of triazole-linked DNA analogue (TLDNA) in solution phase. *Tetrahedron Lett.* **2009**, 50, 4101–4103.
7. Ti, G.S.; Gaffney, B.L.; Jones, R.A. Transient protection: Efficient one-flask syntheses of protected deoxynucleosides. *J. Am. Chem. Soc.* **1982**, 104, 1316–1319.
8. a) Wang, H.; Cheng, C.; Zheng, H.; Liu, X.; Fang, C.; Xu, S.; Zhao, Y. Study on disulfur-backboned nucleic acid: Part 2. Efficient synthesis of 3',5'-dithiothymidine. *Chem. Lett.* **2005**, 34, 432–433; b) Kochetkova, S.V.; Timofeev, E.N.; Korobeinikova, E.A.; Kolganova, N.A.; Florentiev, V.L. Oligodeoxynucleotides with extended zwitter-ionic internucleotide linkage. *Tetrahedron* **2001**, 57, 10287–10292.
9. a) Wu, J.C.; Xi, Z.; Gioeli, C.; Chattopadhyaya, J. Intramolecular cyclization-trapping of carbon radicals by olefins as means to functionalize 2'- and 3'-carbons in β-D-nucleosides. *Tetrahedron* **1991**, 47, 2237–2254; b) Teste, K.; Colombeau, L.; Hadj-Bouazza, A.; Lucas, R.; Zerrouki, R.; Krausz, P.; Champavier, Y. Solvent-controlled regioselective protection of 5'-O-protected thymidine. *Carbohydr. Res.* **2008**, 343, 1490–1495; c) Lucas, R.; Teste, K.; Zerrouki, R.; Champavier, Y.; Guilloton, M. Chelation-controlled regioselective alkylation of pyrimidine 2'-deoxynucleosides. *Carbohydr. Res.* **2010**, 345, 199–207.
10. Alexander, R.L.; Morris-Natschke, S.L.; Ishaq, K.S.; Fleming, R.A.; Kucera, G.L. Synthesis and cytotoxic activity of two novel 1-dodecylthio-2-decyloxypropyl-3-phosphatidic acid conjugates with gemcitabine and cytosine arabinoside. *J. Med. Chem.* **2003**, 46, 4205–4208.
11. Lucas, R.; Neto, V.; Hadj Bouazza, A.; Zerrouki, R.; Granet, R.; Krausz, P.; Champavier, Y. Microwave-assisted synthesis of a triazole-linked 3'–5' dithymidine using click chemistry. *Tetrahedron Lett.* **2008**, 49, 1004–1007.
12. Liu, F.; Austin, D.J. Synthesis of 5'-functionalized adenosine: Suppression of cyclonucleoside formation. *Tetrahedron Lett.* **2001**, 42, 3153–3154.