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Synthesis and structures of deoxyribonucleoside analogues for triazole-linked DNA (^{TL}DNA)

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ABSTRACT

Deoxyribonucleoside analogues bearing acetylene group at the pseudo-5′-position and azido group at the pseudo-3′-position have been synthesized by transglycosylation reaction of deoxythymidine analogue with adenine, cytosine, and guanine nucleobases as nucleophiles. The structures of analogues were studied in crystalline state by X-ray crystallography as well as in solution phase by NMR spectroscopy and showed the puckering conformations similar to the natural congeners.

Artificial oligonucleotides with non-natural backbones are an important class of compounds that cannot be biologically degraded but can be used as biological tools upon binding to the natural target strand of DNA. The oligonucleotide needs a sequence of four different nucleobases to target the natural strand where four deoxyribonucleosides, that is, deoxythymidine (dT), deoxyadenosine (dA), deoxycytidine (dC), and deoxyguanosine (dG), are coding genes in the complementary sequence.[1] Recently, we have developed a triazole-linked analogue of DNA (^{TL}DNA, Fig. 1) as a new artificial oligonucleotide and demonstrated that the oligonucleotide of 10-mer forms a stable double helix with a natural DNA strand.[2] However, the oligomer was synthesized only with dT analogue and therefore targeted a poly-dA strand. In this Letter, we report on the synthesis of analogues of the other deoxyribonucleosides, dA, dC, and dG via transglycosylation reaction. Structural analysis shows that the analogues have conformational flexibility similar to the natural congeners.

As we have previously validated a method for the large-scale synthesis of dT analogue **1**,[2] transglycosylation reaction[3] seems a straightforward route to the nucleobase diversity. Thus, the transglycosylation of **1** with protected adenine **2A** proceeded with catalytic amount of Me_3SiOTf in refluxing 1,2-dichloroethane to afford the desired analogues **3A** in moderate yield.[4] The stereoselectivity of the glycosylation was similar to that of natural congeners,[3] and we obtained α-anomer (**3A**-α) in 29% yield and β-anomer (**3A**-β) in 26% yield after the separation by middle pressure liquid chromatography (MPLC; Scheme 1). Transglycosylation of **1** with the other nucleobases (**2C** and **2G**) also proceeded under similar conditions, and the desired analogues **3C** and **3G** were obtained in moderate yield, respectively.[5] Although the anomers of dC analogue **3C** could not be chromatographically separated, benzoylation of amine residue of the base moiety led to the successful MPLC separation (Eq. 1). Protecting groups of all the nucleosides were removed under basic conditions to give the corresponding nucleosides **5** in moderate to good yield (Scheme 2 and Supplementary data). The structures of each anomer were first established spectroscopically and later confirmed by X-ray analysis of the single crystals (vide infra).

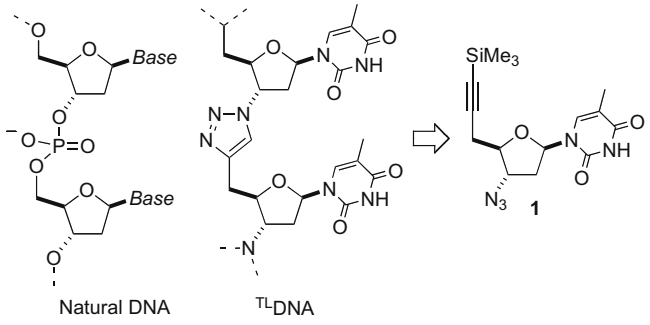

Figure 1. Structures of natural DNA, ^{TL}DNA, and dT analogue **1**.

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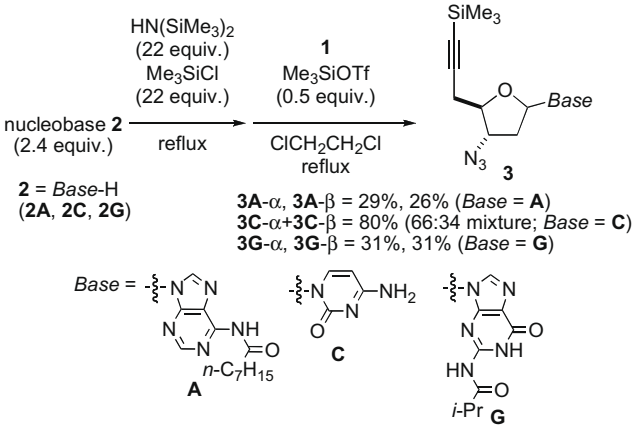
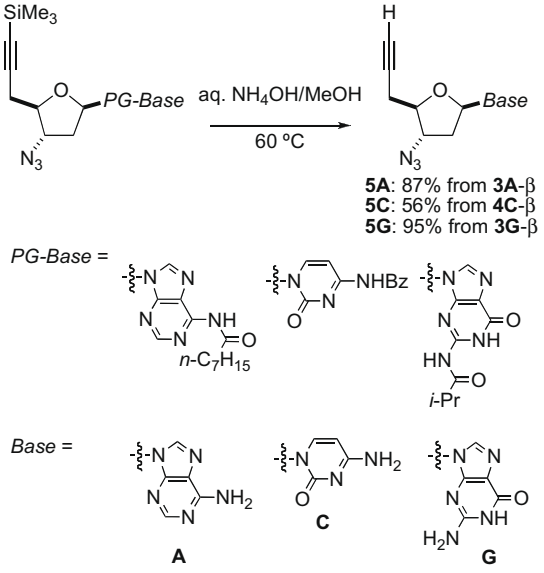
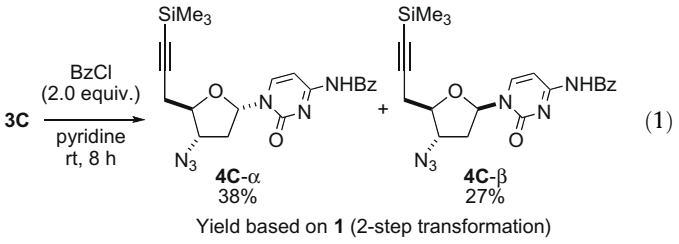


Scheme 1. Synthesis of deoxyribonucleoside analogues.



Scheme 2. Deprotection of deoxyribonucleoside analogues.



Yield based on **1** (2-step transformation)

With single crystals of dT **1**, dA **5A**, dC **5C**, and dG **3G**-β, the structural analysis by X-ray crystallography unequivocally established the structures and revealed the preferred conformations in solid state. Representative molecular structures and the puckering parameters are shown in Figure 2 (see Supplementary data for the α-anomers). In all the analogues, the glycosidic linkage to the nucleobase adopts *anti* conformations favorable for the base pairing. The azido group preferred the geometry *trans* to the C2′–C3′ bond,[6] which may help to free the group from steric hindrance for the subsequent click coupling.[2,7] Pyrimidine analogues (dT **1** and dC **5C**) adopt two puckering conformations both with geometries in the South of pseudorotational cycle (S-type, 2′-*endo*).[3,8] On the other hand, the puckering of purine analogues (dA **5A** and dG **3G**-β) was in the North (N-type, 3′-*endo*).[9]

In solution, puckering conformations of β-anomers equilibrate between S-type and N-type, as such observed with natural congeners. Proton NMR spectra of β-analogues were recorded in DMSO at room temperature (25 °C),[10] and the puckering population was estimated by Altona–Sundaralingam approximation using the vicinal coupling constants (^3J) of H1′, H2′ and H3′, H4′ (Table 1).[1,11] Thus, in all the analogues, the population of S-type conformer was slightly higher than that of N-type and estimated as 56% for dT (**1**), 56% for dA (**3A**-β), 53% for dC (**4C**-β), and 53% for dG (**3G**-β), respectively.

In summary, we have developed a method for the synthesis of deoxyribonucleoside analogues bearing adenine, cytosine and guanine nucleobases. All the analogues showed the puckering flexibility similar to the natural congeners, which may help the duplex

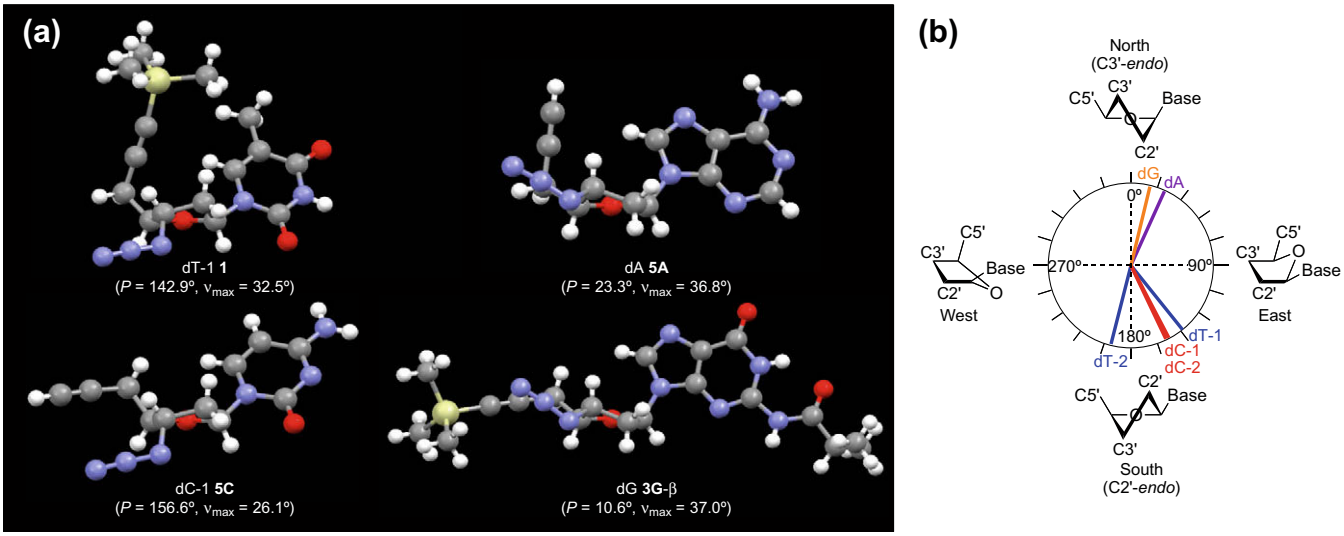


Figure 2. (a) Representative molecular structures and the structural parameters of analogues obtained by X-ray diffraction analysis of the single crystals. The structures are viewed along the plane of C1′–O4′–C4′. One of the two conformers found for dT and dC is shown, respectively. The phase angle (P) and the puckering amplitude (ν_{max}) are shown. Solvent molecules were omitted for clarity. Carbon: gray, hydrogen: white, nitrogen: blue, oxygen: red, and silicon: yellow. (b) Locations of puckering conformations of analogues in the pseudorotational cycle where twist and envelope forms appear alternatively every 18°.

Table 1
Vicinal coupling constants and puckering population in DMSO

Analogue	$^3J_{H1',H2'}$ (Hz)	$^3J_{H3',H4'}$ (Hz)	%South
1 (dT)	6.8	5.2	57
3A-β (dA)	7.2	5.6	56
4C-β (dC)	6.2	5.6	53
3G-β (dG)	6.4	5.6	53

formation of the oligonucleotides. The puckering preference of the new analogues may also be informative for the 3′-azidonucleosides of biological importance.[12] With the library of four nucleoside analogues, we can now design and synthesize functional ^{TL}DNA that targets a specific natural complementary strand. In addition, the transglycosylation reaction should be applicable to artificial nucleobases, and we will explore the scope in future.

Acknowledgments

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Supplementary data

Crystallographic data (excluding structure factors) for the structures in this paper have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication (CCDC 761746–761752). These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif. Supplementary data (experimental details, spectral data and X-ray crystallographic data) associated with this article can be found, in the online version, at doi:10.1016/j.tetlet.2010.02.046.

References and notes

1. Saenger, W. *Principle of Nucleic Acid Structure*; Springer: New York, 1984.
2. Isobe, H.; Fujino, T.; Yamazaki, N.; Guillot-Nieckowski, M.; Nakamura, E. *Org. Lett.* **2008**, *10*, 3729–3732; Fujino, T.; Yamazaki, N.; Isobe, H. *Tetrahedron Lett.* **2009**, *50*, 4101–4103.
3. Imazawa, M.; Eckstein, F. *J. Org. Chem.* **1978**, *43*, 3044–3048; Vorbrüggen, H.; Bennua, B. *Chem. Ber.* **1981**, *114*, 1279–1286.
4. Although the transglycosylation also proceeded with $SnCl_4$ as a Lewis acid, the yield was much lower than that with Me_3SiOTf. Vorbrüggen, H.; Krolikiewicz, K.; Bennua, B. *Chem. Ber.* **1981**, *114*, 1234–1255.
5. The reaction with guanine nucleophile gave two other transglycosylated products (ca. 4% yield, respectively) which are tentatively assigned as 7-glycosylated byproducts (Ref. 3). We did not observe similar isomeric byproducts with adenine nucleophile.
6. Van Roey, P.; Salerno, J. M.; Duax, W. L.; Chu, C. K.; Ahn, M. K.; Schinazi, R. F. *J. Am. Chem. Soc.* **1988**, *110*, 2277–2282.
7. Bock, V. D.; Hiemstra, H.; van Maarseveen, J. H. *Eur. J. Org. Chem.* **2006**, 51–68.
8. The puckering of furanose rings can be described by the pseudorotational cycle which correlates all possible twist and envelope conformations to the pseudorotional phase angle *P*. The puckering amplitude ν_{max} shows the degree of distortion of the ring from the planar form.
9. Observed phase angles of the analogues were in the range similar to the natural congeners (Ref. 1). See also: Altona, C.; Sundaralingam, M. *J. Am. Chem. Soc.* **1972**, *94*, 8205–8212; Altona, C.; Sundaralingam, M. *J. Am. Chem. Soc.* **1973**, *94*, 8205–8212.
10. Coupling analysis in aqueous solution was not successful due to low solubility of the analogues.
11. The population of S-type conformer was estimated by the following equation: $\%South = 100 \times (^3J_{H1',H2'}/(^3J_{H1',H2'} + {}^3J_{H3',H4'}))$.
12. Mathé, C.; Périgaud, C. *Eur. J. Org. Chem.* **2008**, 1489–1505; Sørensen, M. H.; Nielsen, C.; Nielsen, P. *J. Org. Chem.* **2001**, *66*, 4878–4886.