Triplex- and Duplex-Forming Abilities of Oligonucleotides Containing 2′-Deoxy-5-trifluoromethyluridine and 2′-Deoxy-5-trifluoromethycytidine

Yuta Ito, Misaki Matsuo, Takashi Osawa, and Yoshiyuki Hari*

Faculty of Pharmaceutical Sciences, Tokushima Bunri University; Nishihama, Yamashiro-cho, Tokushima 770–8514, Japan.
Received July 3, 2017; accepted July 24, 2017

A facile synthesis of 2′-deoxy-5-trifluoromethyluridine and 2′-deoxy-5-trifluoromethycytidine phosphoramidites from commercially available 2′-deoxyuridine and 2′-deoxycytidine was achieved, respectively. The obtained phosphoramidites were incorporated into oligonucleotides, and their binding affinity to double-stranded DNA (dsDNA) and single-stranded RNA (ssRNA) was evaluated by UV-melting experiments. The triplex-forming abilities of oligonucleotides containing 5-trifluoromethylpyrimidine nucleobases with dsDNA were decreased. Especially, the stability of the triplex containing a trifluoromethylcytosine (CF₃C)₃-GC base triplet was low, likely due to the low pKₐ of protonated CF₃C by the electron-withdrawing trifluoromethyl group. A slight decrease in stability of the duplex formed with ssRNA by oligonucleotides including 5-trifluoromethylpyrimidine nucleobases was only observed, suggesting that they might be applicable to various ssRNA-targeted technologies using features of fluorine atoms.

Key words 5-trifluoromethylpyrimidine nucleobase; oligonucleotide; duplex-forming ability; triplex-forming ability.

Chemically modified oligonucleotides are expected to be the next-generation medicine owing to their specific recognition of nucleic acid sequences through formation of base pairs. In particular, artificial oligonucleotides with high binding affinity towards double-stranded DNA (dsDNA) and single-stranded RNA (ssRNA) can be applied to antigen and antisense technologies. Therefore, chemical modifications of nucleobase, sugar and phosphate moieties have been investigated. Among these modifications, introduction of a substituent at the 5-position of pyrimidine bases is particularly useful because the substituent does not interfere with Watson–Crick or Hoogsteen base pairings, particularly useful because the substituent does not interfere with Watson–Crick or Hoogsteen base pairings, especially with ssRNA and dsDNA, respectively. Herein, we describe the syntheses of oligonucleotides containing CF₃U and CF₃C, and their binding affinities with dsDNA and ssRNA.

Results and Discussion

Syntheses of CF₃U- and CF₃C-Modified Oligonucleotides

CF₃U and CF₃C phosphoramidites were synthesized as depicted in Charts 1 and 2, respectively. For the introduction of a CF₃ group at the 5-position of a pyrimidine base, we used direct radical trifluoromethylation developed by Baran’s group. Treatment of 2′-deoxyuridine (1) with CF₃SO₂Na and t-BuOOH in H₂O afforded 2′-deoxy-5-CF₃-uridine (2) in 59% yield. Protection of the primary alcohol of 2 by 4,4′-dimethoxytrityl chloride (DMTrCl) followed by phosphorylation of 3 gave the CF₃U phosphoramidite 4.

The protected CF₃C phosphoramidite 11 was synthesized from commercially available 2′-deoxycytidine (5). Radical tri- fluoromethylation of 5 afforded 2′-deoxy-5-CF₃-cytidine (6) in 55% yield. Subsequent silylation of 3′- and 5′-hydroxyl groups gave compound 7, which was treated under N-acetylation conditions to furnish mono-acetylated 8 and di-acetylated 9 in 46 and 51% yields, respectively. Treatment of a mixture of 8 and 9 with tetrabutylammonium fluoride (TBAF) followed by dimethoxytritylation led to compound 10 in 53% yield over 2 steps. Finally, the synthesis of phosphoramidite 11 was carried out by phosphorylation of 10. The phosphoramidites 4 and 11
were incorporated into oligonucleotides using an automated DNA synthesizer with common phosphoramidite chemistry, and their oligonucleotides were purified by reversed-phase HPLC (RP-HPLC) and characterized by electrospray ionization-time-of-flight (ESI-TOF) mass spectrometry.

**UV-Melting Experiments** Initially, the triplex-forming ability of oligonucleotides containing CF3U and CF3C with dsDNA targets (YZ=AT, GC) was evaluated by UV-melting experiments and compared to that of 5-methyl and 5-unsubstituted congeners (Table 1). Unfortunately, the melting temperature ($T_m$) of the triplex-forming oligonucleotide (TFO) 12c ($X$=CF3U) for the AT base pair decreased by 4–5°C compared to that of TFOs 12a ($X$=U) and 12b ($X$=T). For the GC base pair, the $T_m$ of the CF3C-modified TFO 12f was 18°C, which was a drastic decrease of 11–13°C compared to TFOs 12d ($X$=C) and 12e ($X$=mC). To investigate the cause of this decrease, we measured the $pK_a$ of 6 (Fig. 2). The absorbance at 277 nm ($A_{277}$), which is the maximum absorption wavelength of 6 at pH 1.92, was plotted at each pH unit. The curve-fitting analysis based on the Henderson–Hasselbalch equation demonstrated that the $pK_a$ of protonated 6 was approximately 2.6 (see Experimental for details). Therefore, the protonated form of 6 was found to be much more acidic than those of 2′-deoxycytidine ($pK_a=4.4$) and 2′-deoxy-5-methylcytidine ($pK_a=4.5$).20 This result implies that protonation of the N-3 position of CF3C would not occur at neutral pH; therefore, recognition of the CF3C base for a GC base pair would be limited to one hydrogen bond. On the other hand, the $pK_a$ (7.4) of CF3U is also lower than those of uracil (9.5) and thymine (9.8).12 A partial deprotonation of CF3U under the measurement condi-
tions might significantly affect a decrease in the $T_m$ of triplex including a CF$_3$U-AT base triplet. Since it is known that T or C can interact with a CG base pair through a single hydrogen bond, the triplex-forming ability towards dsDNA including a CG base pair was investigated. Interestingly, whereas the TFO 12c containing CF$_3$U led to a decrease (3–4°C) in the $T_m$ compared with those containing U (12a) and T (12b), the $T_m$ of the TFO 12f containing CF$_3$C was almost same as those containing C (12d) and mC (12e). The $T_m$ of the triplex including a CF$_3$C-GC base triplet was comparable to that including a CF$_3$C-CG base triplet, which also strongly suggests that CF$_3$C forms only a single hydrogen bond with a GC base pair at neutral pH. In addition, the obtained results suggest that CF$_3$C can be a newly promising scaffold for CG base pair recognition in triplex DNA formation though C or mC analgous have been explored towards recognition of a CG base pair.4,22,23

**Table 1.** $T_m$ (°C) of Triplexes between Oligonucleotides 12a–f and Hairpin dsDNA

| TFO (X) | YZ | TFO (X) | YZ |
|---------|----|---------|----|
| AT      | CG | GC      | CG |
| 12a (U) | 34 | 20      | 18 |
| 12b (T) | 35 | 19      | 18 |
| 12c (CF$_3$U) | 30 | 16      | 17 |
| 12d (C) | 29 | 18      |    |
| 12e (mC) | 31 | 18      |    |
| 12f (CF$_3$C) | 18 | 17      |    |

*a) Conditions: 10 mM sodium cacodylate buffer (pH 7.4), 100 mM KCl, 50 mM MgCl$_2$, and 1.89 µM of each oligonucleotide. Câ€”2'-Deoxy-5-methylcytidine.*
creased binding affinity with a TCF3C behavior with ssDNA. Replacement of three C bases (12i) in the oligonucleotides containing 5-methyl analogs, and T oligonucleotides with ssRNA was examined (Table 2).

Table 2. $T_m$ (°C) of Duplexes between Oligonucleotides 12a-f and ssRNA

| ssDNA (X) | 5′-TTTTTCTCAGAGAGAGAA-3′ | ssDNA (X) | 5′-TTTTTCTCAGAGAGAGAA-3′ |
|-----------|-----------------------------|-----------|-----------------------------|
| 12a (U)   | 50                          | 12d (C)   | 56                          |
| 12b (T)   | 51                          | 12e (°C)  | 57                          |
| 12c (CF3U)| 48                          | 12f (CF3C)| 55                          |

a) Conditions: 10mm sodium cacodylate buffer (pH 7.4), 100mm NaCl and 3µM of each oligonucleotide. C–2′-Deoxy-5-methylcytidine.

Next, the duplex-forming ability of CF3U and CF3C modified oligonucleotides with ssRNA was examined (Table 2). $T_m$s of the oligonucleotides containing 5-methyl analogs, 12b (X=T) and 12c (X=CF3C), increased by 1°C compared to those of the oligonucleotides containing 5-unsubstituted analogs, 12a (X=U) and 12d (X=C). In contrast, the $T_m$s of CF3U- and CF3C-modified oligonucleotides, 12e and f, were slightly lower than those of 12a and d.

Table 3. $T_m$ (°C) of Duplexes between Oligonucleotides 12e and g-j, and ssRNA

| Sequence of ssDNA | $T_m$ | $\Delta T_m$ per mod. |
|-------------------|-------|-----------------------|
| 5′-TTTTTUCUCCTCTCT-3′ (12g) | 56 | 0.3 |
| 5′-TTTTTUCUCCTCTCT-3′ (12e) | 57 | 1.7 |
| 5′-TTTTTUCUCCTCTCT-3′ (12h) | 55 | 0.7 |
| 5′-TTTTTUCUCCTCTCT-3′ (12j) | 57 | 0.7 |

a) Conditions: 10mm sodium cacodylate buffer (pH 7.4), 100mm NaCl and 3µM of each oligonucleotide. C–2′-Deoxy-5-methylcytidine.

Next, the duplex-forming ability of CF3U and CF3C modified oligonucleotides with ssRNA was examined (Table 2). $T_m$s of the oligonucleotides containing 5-methyl analogs, 12b (X=T) and 12c (X=CF3C), increased by 1°C compared to those of the oligonucleotides containing 5-unsubstituted analogs, 12a (X=U) and 12d (X=C). In contrast, the $T_m$s of CF3U- and CF3C-modified oligonucleotides, 12e and f, were slightly lower than those of 12a and d.

The duplex-forming ability of oligonucleotides containing three CF3U and CF3C bases with ssRNA was also investigated (Table 3, Fig. S1). The $T_m$ of 12g bearing three U bases was 56°C, which was comparable to that of the thymine congener 12e. On the other hand, the triplic CF3C-modified oligonucleotide 12h, the $T_m$ of which was 51°C, destabilized the duplex by 1.7°C per modification. Since it was previously reported by Sigurdsson’s group that replacement of thymine by CF3U led to a decrease of 3.1°C in the $T_m$ of the duplex with ssDNA,[2] these results demonstrate that the duplex-forming ability of CF3U-modified oligonucleotide with ssRNA is similar to its behavior with ssDNA. Replacement of three C bases (12i) with CF3C ones (12e) resulted in an increase in the $T_m$ by 2°C. The triplic CF3C-modified oligonucleotide 12j exhibited slightly decreased binding affinity with a $T_m$ of 53°C. The $\Delta T_m$ per modification of CF3C-modified oligonucleotide was −0.7°C, which was slightly improved compared to that of CF3U (−1.7°C). These results suggest that an oligonucleotide containing CF3C as well as that containing CF3U might become an important material for nucleic acid technologies, especially targeting ssRNA, using features of fluorine atoms.

Conclusion

CF3U and CF3C phosphoramidites were easily synthesized and incorporated into 15-mer oligonucleotides. The UV-melting experiments revealed that introduction of a CF3 group at the C-5 position of pyrimidine bases resulted in a decrease in the duplex-forming affinity with dsDNA in comparison with that observed in methyl and unsubstituted analogs. Although the duplex-forming ability of oligonucleotides including CF3U and CF3C with ssRNA also slightly decreased, the stability of the duplex was considered to be sufficiently practicable. Therefore, we expect that these oligonucleotides would be applicable to ssRNA-targeted technologies using features of fluorine atoms such as 19F-NMR spectroscopy and 19F-labeled positron emission tomography (PET) imaging. Further studies on the application of CF3U- and CF3C-modified oligonucleotides are currently underway in our laboratory.

Experimental

General

All moisture-sensitive reactions were conducted in well-dried glassware under Ar atmosphere. Anhydrous CH2Cl2, tetrahydrofuran (THF), pyridine and N,N-dimethylformamide (DMF) were used as purchased. 1H-NMR, 13C-NMR, 19F-NMR and 31P-NMR spectra were recorded on a Bruker AVANCE III HD 500 equipped with a BBO cryoprobe or Agilent 400-MR. Chemical shift values were reported in ppm downfield from internal tetramethylsilane ($\delta=0.00$ ppm) for 1H-NMR, residual CDCl3 ($\delta=7.26$ ppm) for 13C-NMR, internal hexafluorobenzene ($\delta=0.00$ ppm) for 19F-NMR, and external 5% H3PO4 ($\delta=0.00$ ppm) for 31P-NMR. IR spectra were recorded on a JASCO FT/IR-4200 spectrometer. High-resolution mass spectrometry (HR-MS) was performed on a Waters SYNAPT G2-Si (Quadrupole/TOF). For column chromatography, silica gel PSQ 60B (Fuji Silysam, Japan) was used. The progress of the reaction was monitored by analytical TLC on pre-coated aluminum sheets (Silica gel 60F254 by Merck, Germany). For HPLC, a JASCO EXTREMA (PU-4180, CO-4060 and UV-4075) with a fraction collector CHF122SC (ADVANTEC), was used. Syntheses of oligonucleotides were performed on an automated DNA synthesizer (Gene Design nS-III). UV-melting experiments were carried out using JASCO V-730 UV/Vis spectrophotometer equipped with a $T_m$ analysis accessory. Solution pH values were measured with a pH meter LAQUAact D-41 (HORIBA, Japan).

Synthesis of Phosphoramidites 4 and 11

2′-Deoxy-5′-trifluoromethyluridine (2) To a solution of 2′-deoxyuridine (1, 381 mg, 1.67mmol) and CF3SO2Na (782mg, 5.01mmol) in H2O (5mL), t-BuOOH (70% solution in H2O, 0.81mL, 8.35mmol) was slowly added at 0°C. The reaction mixture was then warmed up to room temperature. After being stirred for 3h, the reaction mixture was concentrated in vacuo. The crude residue was purified by column chromatography (CHCl3–MeOH=10:1 to 5:1) to give compound 2 as a white solid (264mg, 59%). The NMR spectral data were identical to those reported in the literature.[39]

2′-Deoxy-5′-O-(4,4′-dimethoxytrityl)-5′-trifluoromethyluridine (3) To a solution of compound 2 (70mg, 0.24mmol) in pyridine, DMTrCl (96mg, 0.28mmol) was added at room temperature under Ar atmosphere. After being stirred for 41.5h, the reaction was quenched with MeOH and then concentrated in vacuo. The crude residue was purified by column chromatography (CHCl3–MeOH=100:1) to give compound 3 as a white solid (101mg, 72%). The NMR spectral data were identical to those reported in the literature.[39]

3′-O-[2-Cyanoethoxy(dimethylamino)phosphino]-2′-deoxy-5′-O-(4,4′-dimethoxytrityl)-5′-trifluoromethyluridine
(4) To a solution of compound 3 (232 mg, 0.39 mmol) and t-Pr,NEt (0.41 mL, 2.34 mmol) in CHCl₃ (5.0 mL), t-Pr,NP(Ch)OCH₂CH₂CN (0.13 mL, 0.59 mmol) was added at 0°C under Ar atmosphere. The reaction mixture was stirred at room temperature for 3 h. After addition of sat. NaHCO₃ aq., the reaction mixture was extracted with AcOEt. The combined organic layer was washed with water and brine, dried over Na₂SO₄, and concentrated in vacuo. The crude residue was purified by column chromatography (hexane–AcOEt=5:1) to give compounds 6 (446 mg, 46%) and 9 (536 mg, 51%).

Compound 8

Colorless oil. IR (ATR) cm⁻¹: 3425, 2953, 2931, 2858, 1722, 1691, 1657, 1579, 1473, 1363, 1254. ¹H-NMR (500 MHz, CDCl₃) δ: 0.07–0.09 (12H, m), 0.87–0.90 (18H, m), 1.96–2.05 (1H, m), 2.25–2.74 (4H, m), 3.75–3.97 (1H, m), 3.87–3.93 (1H, m), 4.10 (1H, brs), 4.37 (1H, brs), 6.18 (1H, brs), 7.58 (0.7H, brs), 8.28 (0.3H, brs), 8.47 (0.7H, brs), 13.1 (0.3H, brs).

¹³C-NMR (125 MHz, CDCl₃) δ: −5.83, −5.78, −5.0, −4.8, 17.9, 18.2, 25.6, 25.7, 26.6, 28.8, 42.3, 42.7, 62.8, 72.5, 86.9, 88.3, 92.0, 97.2 (q, J = 34.00 Hz), 104.4 (q, J = 31.50 Hz), 121.6 (q, J = 270.0 Hz), 142.2 (q, J = 270.0 Hz), 140.1, 143.8, 146.7, 153.0, 153.7, 156.7, 171.8, 188.1. ¹F-NMR (376 MHz, CDCl₃) δ: 99.4, 102.0. HR-MS (ESI-TOF): Calcd for C₂₅H₂₁F₃N₂O₂Si₃ [M+H]⁺ 566.2693. Found 566.2696.

Compound 9

White foam. IR (ATR) cm⁻¹: 2954, 2930, 2859, 1732, 1699, 1643, 1515, 1471, 1464, 1448, 1406, 1369, 1349, 1315, 1274, 1255, 1236. ¹H-NMR (500 MHz, CDCl₃) δ: 0.06 (6H, s), 0.10 (3H, s), 0.11 (3H, s), 0.87 (9H, s), 0.91 (9H, s), 2.11 (1H, dd, J = 13.7, 5.0, 6.0 Hz), 2.33 and 2.36 (6H, brs), 2.76 (1H, dd, J = 13.7, 5.0, 2.5 Hz), 3.79 (1H, dd, J = 11.5, 2.5 Hz), 3.95 (1H, dd, J = 11.5, 2.5 Hz), 4.16–4.18 (1H, m), 4.40–4.42 (1H, m), 6.19 (1H, dd, J = 6.5, 6.5 Hz), 8.74 (1H, s). ¹³C-NMR (125 MHz, CDCl₃) δ: −5.7, −5.6, −4.9, −4.6, 18.0, 18.4, 25.7, 25.8, 26.0, 42.9, 62.9, 72.7, 89.5, 89.9, 106.2 (q, J = 35.05 Hz), 121.9 (q, J = 270.0 Hz), 146.2 (q, J = 6.5 Hz), 153.7, 163.1, 170.4, 171.1. ¹F-NMR (376 MHz, CDCl₃) δ: 100.6. HR-MS (ESI-TOF): Calcd for C₂₅H₂₁F₃N₂O₂Si₃ [M+Na]⁺ 630.2618. Found 630.2617.

N-Acetyl-2′-deoxy-5′-O-(4,4′-dimethoxytrityl)-5-trifluoromethylcytidine (10) To a solution of a mixture of 8 and 9 (982 mg, 1.67 mmol, ca. 8:9 = 1:2) in THF (20 mL), TBAF (1 M in THF, 5.0 mL, 5.0 mmol) was added at room temperature. After being stirred for 0.5 h, the reaction mixture was concentrated in vacuo. The crude residue was purified by column chromatography (CHCl₃–MeOH=10:1) to give compound 10 as a pale yellow foam (566 mg, 53% in 2 steps). IR (ATR) cm⁻¹: 3418, 2936, 1717, 1656, 1607, 1577, 1542, 1508, 1486, 1446, 1370, 1250. ¹H-NMR (500 MHz, CDCl₃) δ: 2.18–2.32 (2H, m), 2.43 (0.8H, brs), 2.55–2.62 (0.2H, m), 2.70 (2.2H, m), 2.80–2.86 (0.8H, m), 3.34–3.44 (2H, m), 3.79 (6H, s), 4.14 and 4.22 (1H, brs), 4.43 (1H, brs), 6.17 (1H, dd, J = 6.0, 6.0 Hz), 6.82 (1H, d, J = 8.0 Hz), 7.20–7.40 (9H,
J 3.0 Hz), 3.52–3.88 (10H, m), 4.26 and 4.31 (1H, br s), 4.48–4.56 (1H, br s), 8.14–8.50 (1H, m), 13.1 (0.3H, br s).

In the triplex-forming experiment, oligonucleotides and hairpin dsDNA were dissolved in 10 mm sodium cacodylate buffer (pH 7.4) containing 100 mm KCl and 50 mm MgCl₂, to give a final concentration of 1.89 μM, respectively. The samples were annealed in boiling water followed by slow cooling to 5°C. The melting profiles at 260 nm were recorded from 5 to 90°C (for triplex) and from 10 to 80°C (for duplex) at a scan rate of 0.5°C/min. The two-point average method was employed to obtain the Tm, and the final values were determined by averaging three independent measurements, which were accurate within a 1°C range.

**pKₐ Calculation**
The absorbance of the solution of 6 in citrate buffer (prepared at each pH) was measured in the wavelength range of 220 to 340 nm using JASCO V-730. The pKₐ of 6 was calculated by fitting the plot to a rearrangement of the Henderson–Hasselbalch Eq. 1.

\[
A_{277} = \frac{\epsilon_{\alpha} \cdot C_0}{1 + 10^{pK_{a}-pH}} + \frac{\epsilon_{\beta} \cdot 10^{pK_{a}-pH}}{1 + 10^{pK_{a}-pH}},
\]

where \(A_{277}\) is the absorbance at 277 nm, \(C_0\) is the concentration of 6 (6.77×10⁻³ M), and \(\epsilon_{\alpha}\) and \(\epsilon_{\beta}\) are the molar absorptivity of protonated and neutral forms of 6 at 277 nm, respectively. SigmaPlot (Systat Software, Inc.) was used for the fitting. The data obtained were 2.56±0.11 for \(pK_{a}\), 6.807±0.161 for \(\epsilon_{\alpha}\), and 12718±514 for \(\epsilon_{\beta}\) (R²=0.98).

**Acknowledgment** This work was partially supported by the Japan Society for the Promotion of Science (JSPS) KAKENHI Grants (JP17K15431 and JP17K05943).

**Conflict of Interest** The authors declare no conflict of interest.

**Supplementary Materials**
The online version of this article contains supplementary materials.

**References**

1) Moribiro K., Kasahara Y., Obika S., *Mol. Biosyst.*, 13, 235–245 (2017).
2) Wan W. B., Seth P. P., *J. Med. Chem.*, 59, 9645–9667 (2016).
3) Sharma V. K., Sharma R. K., Singh S. K., *MedChemComm*, 5, 1454–1471 (2014).
4) Hari Y., Obika S., Imanishi T., *Eur. J. Org. Chem.*, 2012, 2875–2887 (2012).
5) Luyten I., Herdewijn P., *Eur. J. Org. Chem.*, 33, 515–576 (1998).
6) Xodo L. E., Manzini G., Quadrifiglio F., van der Marel G. A., van Boom J. H., *Nucleic Acids Res.*, 19, 5625–5631 (1991).
7) Butkus V., Klimašauskas S., Petrusrašienė L., Maneliene Z., Janušaitis A., Minchenkova L. E., Schyolkin A. K., *Nucleic Acids Res.*, 15, 8467–8478 (1987).
8) Songhi Y. S., Hoke G. D., Freier S. M., Zounes M. C., Gonzalez C., Cummins L., Sassor H., Cook P. D., *Nucleic Acids Res.*, 21, 3197–3203 (1993).
9) Povsic T. J., Dervan P. B., *J. Am. Chem. Soc.*, 111, 3059–3061 (1989).
10) Amosova O. A., Fresco J. R., *Nucleic Acids Res.*, 27, 4632–4635 (1999).
11) Theruvathu J. A., Kim C. H., Rogstad D. K., Neidigh J. W., Sowers L. C., *Biochemistry*, 48, 7539–7546 (2009).
12) Heidelberger C., *Cancer Res.*, 30, 1549–1569 (1970).
13) Nakamura S., Yang H., Hirata C., Kersaudy F., Fujimoto K., *Org. Biomol. Chem.*, 15, 5109–5111 (2017).
14) Nakamura S., Fujimoto K., *Chem. Commun.*, 51, 11765–11768 (2015).
15) Gmeiner W. H., Pon R. T., Lown J. J., *J. Org. Chem.*, 56, 3602–3608 (1991).
16) Sugimoto N., Matsumura A., Sasaki M., Satake H., Takeda S., Wataya Y., *Chem. Express*, **7**, 405–408 (1992).

17) Markley J. C., Chirakul P., Sologub D., Sigurdsson S. T., *Bioorg. Med. Chem. Lett.*, **11**, 2453–2455 (2001).

18) Purser S., Moore P. R., Swallow S., Gouverneur V., *Chem. Soc. Rev.*, **37**, 320–330 (2008).

19) Ji Y., Brueckl T., Baxter R. D., Fujiwara Y., Seiple I. B., Su S., Blackmond D. G., Baran P. S., *Proc. Natl. Acad. Sci. U.S.A.*, **108**, 14411–14415 (2011).

20) Karino N., Ueno Y., Matsuda A., *Nucleic Acids Res.*, **29**, 2456–2463 (2001).

21) Rusling D. A., Brown T., Fox K. R., “Sequence-specific DNA Binding Agents,” Chap. 1, ed. by Neidle S., Waring M. J., RSC Publishing, Cambridge, 2006, pp. 1–28.

22) Hari Y., Akabane M., Obika S., *Chem. Commun.*, **49**, 7421–7423 (2013).

23) Akabane-Nakata M., Obika S., Hari Y., *Org. Biomol. Chem.*, **12**, 9011–9015 (2014).

24) Hansen A. S., Thalhammer A., El-Sagheer A. H., Brown T., Schofield C. J., *Bioorg. Med. Chem. Lett.*, **21**, 1181–1184 (2011).

25) Musumeci D., Irace C., Santamaria R., Montesarchio D., *MedChemCommun.*, **4**, 1405–1410 (2013).