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Synthesis and properties of terthiophene-modified oligodeoxynucleotides

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Abstract—Synthesis and properties of oligodeoxynucleotides (ODNs) containing terthiophene (Thp) were described. One-electron oxidation of Thp-modified ODN resulted in the formation of Thp radical cation ($Thp^{\bullet+}$), which remained stable in the experimental time window up to 200 μs, showing that charge may be carried along DNA by Thp as $Thp^{\bullet+}$.

DNA is a versatile molecule that can be used to construct nanometer-sized higher-ordered assemblies and architectures based on the stored information.[1–4] In view of the potential use of DNA as a molecular wire for electronic applications, mechanistic studies of charge transfer in DNA have attracted considerable attention.[5–13] Gel electrophoretic analysis[14,15] and our recent time-resolved transient absorption measurement[16] clearly demonstrated that the hole generated on DNA upon one-electron oxidation can migrate along DNA over 100 Å. However, since DNA is inherently unstable upon oxidation, DNA itself cannot be a good conductive material.[17–20] Another promising way is to use DNA as a scaffold for arranging organic molecules whose radical ion is stable. Previously, we have shown that a charge can be carried by pyrene and phenothiazine as their radical cations along DNA.[21] However, only a few compounds, e.g., pyrene,[22,23] phenothiazine,[24–26] methylindole,[27,28] and ferrocene,[29,30] were tested as carriers of positive charge along DNA. Here, we report the synthesis of terthiophene-modified oligodeoxynucleotides (ODNs), showing that a hole can be carried as a radical cation of Thp ($Thp^{\bullet+}$) along DNA.

Polythiophenes and substituted polythiophenes have received much attention as among the most studied conducting polymers worldwide.[31–33] We selected Thp to assemble along DNA because $Thp^{\bullet+}$ is shown to be stable in water.[34–36] The oxidation potential of Thp ($E^0 = 1.23$ V vs NHE in CH_3CN)[37] is lower than that of guanine which has the lowest oxidation potential among the four DNA bases ($E^0 = 1.47$ V vs NHE in CH_3CN).[38] Therefore, similar to the previously studied pyrene[22,23] and phenothiazine,[24–26] Thp was expected to serve as a hole trap in DNA and may be useful to carry a hole along DNA. The minor groove of DNA has been demonstrated to serve as a good candidate for arranging heterocyclic compounds.[39–41] Thus, we synthesized a nucleoside derivative that possesses a Thp group on guanine N2 (Th**G**) to arrange Thp along the minor groove of DNA through the reaction of 3′,5′-protected 2-fluorodeoxyinosine derivative (**4**) with 5-(3-aminopropyl)-2,2′,5′,2″-terthiophene (**3**) as shown in Scheme 1.[42,43] Th**G** was converted to the phosphoramidite derivative (**9**) and Th**G**-modified ODNs were synthesized by DNA synthesizer according to the standard procedure. Incorporation of Th**G** into ODNs was confirmed by digestion with snake venom phosphodiesterase/nuclease P1/alkaline phosphatase to 2′-deoxyribonucleosides and MALDI-TOF mass spectra.[44]

First, to check the duplex stability of the Th**G**-modified ODNs, melting temperatures were measured (Table 1). Th**G**-modified ODN (**11**) showed slightly higher thermostability compared to the corresponding unmodified ODN (**10**). Incorporation of two Th**G** (**12**) showed a similar T_m value with that of **10**. Thus, Th**G** was incorporated into DNA without large disturbance of the duplex stability.

Keywords: DNA; Modified oligonucleotide; Charge transfer; Thiophene.
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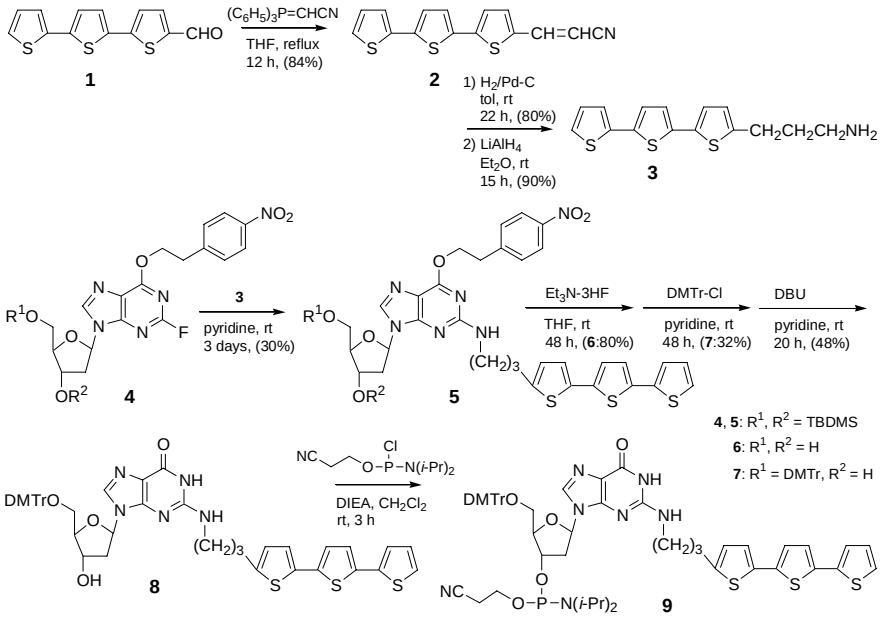


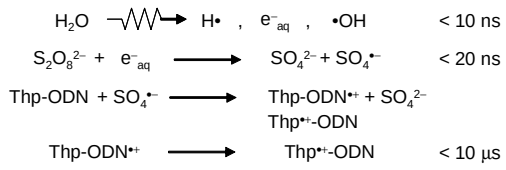
Scheme 1. Synthetic route of the Th**G**-phosphoramidite **9**.

Table 1. Melting temperatures of Th**G**-modified ODNs

ODNs	Sequence	T_m (°C)[a]
10	5′GCAGAACACG 3′CGTCTTGTGC	34
11	5′GCATh**G**AACACG 3′CGTCTTGTGC	36
12	5′GCATh**G**AACACG 3′CGTCTTTh**G**TGC	34

[a] UV melting measurements were carried out in a pH 7.0 Na phosphate buffer 20 mM at a total strand concentration of 8 μM.

Next, to obtain the information on the charge-carrying properties of Th**G**, pulse radiolysis of Th**G**-modified ODN was performed. A hole was produced in Th**G**-modified ODN from the reaction with $SO_4^{\bullet-}$ that was generated during the pulse radiolysis (28 MeV, 8 ns) of Ar-saturated aqueous solution containing 10 mM $K_2S_2O_8$, 100 mM *t*-BuOH, 20 mM Na phosphate buffer (pH 7.0), and 0.1 mM ODN (strand conc) (Scheme 2). There are two possible processes for the formation of Thp$^{\bullet+}$: the direct diffusional collision between $SO_4^{\bullet-}$ and Th**G**, and the hole transfer from the radical cation of nucleobases generated in DNA to Th**G** as shown in Scheme 2. [22,23] Interestingly, a transient absorption with a peak at 560 nm assigned to Thp$^{\bullet+}$ was observed after the electron pulse during the pulse radiolysis and was stable within our experimental timescale up to 200 μs (Fig. 1).



Scheme 2. Mechanistic scheme for generation of oxidizing reagent $SO_4^{\bullet-}$ during pulse radiolysis, hole generation, and transfer in Thp-modified ODN.

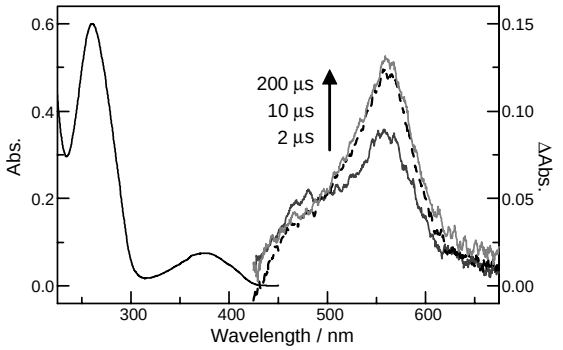


Figure 1. Ground-state absorption (left axis) and transient absorption spectra (right axis) obtained at 2, 10, and 200 μs during the pulse radiolysis of ODN **11**. Ground-state absorption spectrum was measured in a 20 mM Na phosphate buffer (pH 7.0) at a total strand concentration of 8 μM. Pulse radiolysis was carried out in Ar-saturated aqueous solution in the presence of 10 mM $K_2S_2O_8$, 100 mM *t*-BuOH, and 20 mM Na phosphate buffer (pH 7.0), at a total strand concentration of 200 μM.

In conclusion, a phosphoramidite derivative of Th**G** was synthesized to assemble Thp along the DNA minor groove as a hole carrier. It has been demonstrated that Th**G** can be incorporated into DNA without large alternation of the duplex stability. Pulse radiolysis of Th**G**-modified ODN clearly demonstrated that Thp$^{\bullet+}$ can be stably generated in DNA, showing that Thp serves as a hole trap. Hence, by arranging several Th**G** along DNA, it may be useful to carry a hole along DNA.

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Supplementary data

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.bmcl.2005.07.004.

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