# Charge transfer in DNA assemblies: effects of sticky ends†

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**Transient absorption measurements of charge transfer (CT) demonstrated that the CT in the DNA assembly constructed by simply mixing DNAs with sticky ends occurs over 200 Å selectively to the complementary sticky end sequences.**

DNA nanostructures have been constructed by the self-assembly of DNA tiles. The DNA tiles are connected by the sticky end, single-stranded overhangs with a particular DNA sequence[1,2] where the self-assembly of the complementary sticky end DNA, sometimes followed by a ligation reaction, forms double-helical segments that facilitate the connection between the DNA tiles.[3] Although a variety of DNA nanostructures have been demonstrated, their ability to mobilise charge remains unclear.[4] Therefore, examination of the charge transfer (CT) especially between tiles would be important for the further development of DNA nanowires and DNA nanotubes.

The CT in DNA has been extensively studied. It is of interest that the CT in DNA occurs over distances longer than 200 Å.[5–9] Previous reports suggest that this long-range CT in DNA has a connection with biological signaling.[10] Moreover, it is reported that the long-range CT can be used for DNA mismatch and SNP detection.[11,12] Recently, we have reported that the CT in DNA self-assemblies occurs over 140 Å, but the detailed mechanism of CT in the DNA sticky end is still unclear.[13] A further understanding of CT in the DNA sticky end is important for the development of DNA nanowires, understanding biological signaling, and biological applications. In this communication, we report the effects of the sticky end lengths and sequences on the CT in DNA self-assemblies using time-resolved transient absorption measurements.

Naphthalimide (photosensitizer, **NI**) and phenothiazine (charge acceptor, **PTZ**) modified DNAs with different lengths or sequences of sticky end were synthesized in order to examine CT in DNA assemblies in detail. Upon 355 nm laser excitation of **NI**, the charge-separated state (**NI** radical anion and guanine (G) radical cation) is produced *via* CT between the adenines. After charge hopping between guanines (Gs), the charge on the G can be accessible to the **PTZ** [$E_{ox}$ = 0.76 V *versus* NHE] which has an oxidation potential lower than G, producing **PTZ** radical cation (**PTZ**$^{\bullet+}$) (Fig. 1). Thus, CT in DNA assemblies can be observed by the formation of **PTZ**$^{\bullet+}$.[13–17]

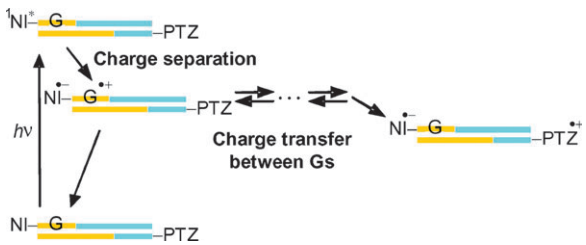


**Fig. 1** Kinetic scheme for CT in DNA assemblies.

Firstly, we examined the effects of the sticky end on CT in DNA assemblies. The CT in a DNA assembly was monitored by the formation of **PTZ**$^{\bullet+}$ at 520 nm for a direct measurement of the CT kinetics in the DNA assembly. The formations of the DNA assemblies were analyzed by gel mobility shift assays (Fig. S1†).[18,19] The annealed DNA assemblies and a 20 bp DNA ladder were loaded into a 15% non-denaturing polyacrylamide gel and resolved by electrophoresis. DNA assemblies with 10 nt sticky ends (**ST10**, 41-mer) and 40 bp markers have almost the same mobility on the gel, indicating that they formed DNA assemblies. We compared the CT rate of a DNA assembly of the 10 nt sticky end (without a 5′-terminal phosphate group, **ST10**) with that of full-length DNA (**F**), which was synthesized by an enzymatic ligation reaction (details shown in the ESI†). There was little difference in the CT rate between these DNAs (Fig. 2b), in agreement with Barton's previous report[20] using an electrochemical measurement, in which the CT efficiency was only slightly affected by the presence of the sticky end. Next, the DNA assembly with a 5′-terminus phosphorylated DNA sticky end (**P**) was examined. Again, the CT kinetics were almost the same as the fully-matched sequence and 10 nt sticky end sequence (without phosphate group) (Fig. 2b). These results indicate that the CT in DNA is unaffected by the phosphate group, at least on the microsecond time scale. In order to examine the effects of the sticky end lengths on the CT, DNA assemblies with 8, 6, 4 and 2 nt sticky ends (**ST8**, **ST6**, **ST4** and **ST2**) were also investigated. Again, the formations of DNA assemblies were analyzed (Fig. S1†). We observed the existence of the DNA assemblies with **ST8**, but did not observe those of **ST6**, **ST4** and **ST2**. Interestingly, CT in DNA assemblies with **ST8**, **ST6** and **ST4**, except for **ST2**, was observed and the CT kinetics among these assemblies were almost the same (Fig. S2†). These results might be derived from the differential conditions between gel electrophoresis and solution structures used in laser experiments. In other

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† Electronic supplementary information (ESI) available: Experimental details. Fig. S1 and S2: characterization and schematic illustration of DNA assemblies. See DOI: 10.1039/b801876f

 

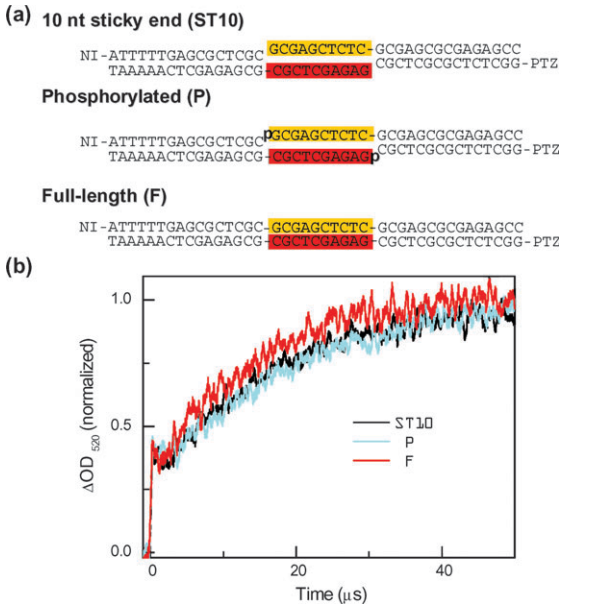


**Fig. 2** (a) Schematic illustration of DNA assemblies for the examination of the sticky end effect on CT in assembly. (b) Time profiles of the transient absorption of $\mathbf{PTZ}^{\bullet+}$ monitored at 520 nm during the 355 nm laser flash photolysis of **ST10** (black), **P** (phosphorylated, cyan) and **F** (full-length, red), respectively. The represented profiles were obtained from the accumulation of 32 laser shots.

words, we have achieved the direct observation of the weak interactions between the sticky ends, at least 4 nt, with the time-resolved laser flash experiments. Anyway, these results suggest that the CT in DNA assemblies actually occurs without an interruption process by sticky ends as small as 4 nt. DNA tiles used to construct DNA nanostructures are often connected by 5–6 nt sticky end DNAs.[3] Thus, the sticky end has little effect on CT rate and efficiency, strongly suggesting that CT between DNA tiles, which are connected by the sticky ends, is possible.

Next, we investigated the importance of the sticky end sequence complementarity. Four DNAs (two pairs of complementary sequences (**N1** + **P1** (**ST10**), **N2** + **P2**) and two pairs of non-complementary sequences of 10 nt sticky end (**N1** + **P2**, **N2** + **P1**)) were examined by the gel shift assay and transient absorption measurement to investigate the selectivity of the sticky end sequences (Fig. 3a and Fig. S1†). The gel shift assays show that DNA assemblies are formed only when the sticky ends have complementary sequences (**N1** + **P1** (**ST10**), **N2** + **P2**). In the transient absorption measurements, the formation of $\mathbf{PTZ}^{\bullet+}$ was observed when the sticky end DNAs were complementary to each other (**N1** + **P1**, **N2** + **P2**). On the other hand, the signals of $\mathbf{PTZ}^{\bullet+}$ were not observed when the non-complementary sticky end DNAs were mixed (**N1** + **P2**, **N2** + **P1**) (Fig. 3b). These results clearly show that sticky end complementary is critical for CT in DNA. Finally, an attempt was made to achieve the long-range CT over 200 Å by combining **N1** and **P2** with a connection unit (**C**). The formation of the $\mathbf{PTZ}^{\bullet+}$ signal was observed when the **C**, which has complementary sticky ends at the DNA terminals for **N1** and **P2**, was added (**N1** + **C** + **P2**) (Fig. 3c). This result corresponds to the results of the gel electrophoresis experiment that show a

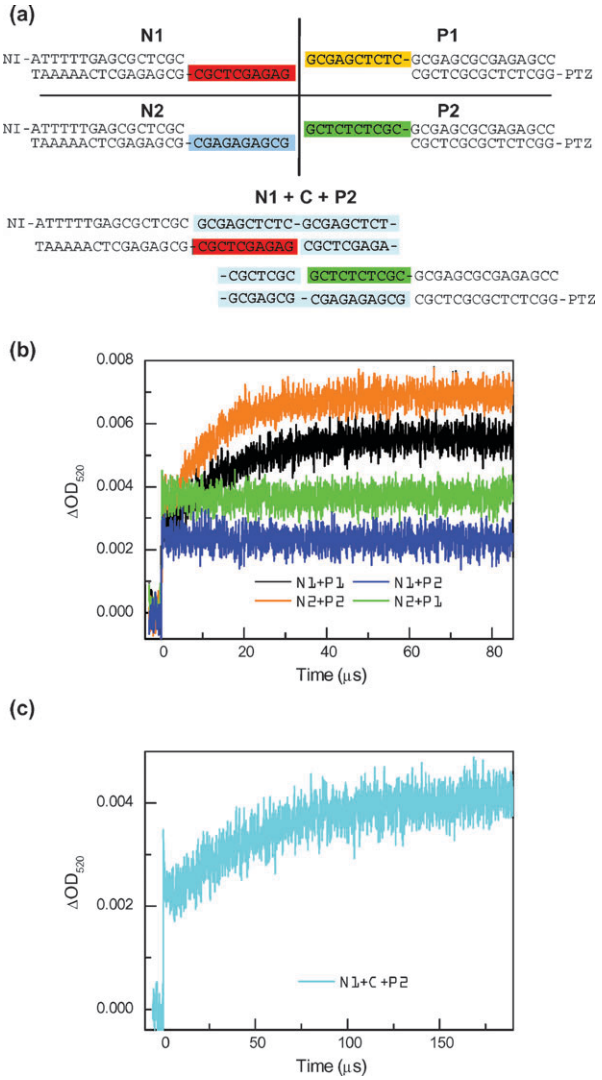


**Fig. 3** (a) Schematic illustration of DNA assemblies for the examination of the importance of sticky end sequence complementarity. **N1** + **P1** (**ST10**), **N2** + **P2**, and **N1** + **C** + **P2** have complementary sequences of nucleotides, respectively. **N1** + **P2** and **N2** + **P1** are pairs of non-complementary sticky end DNAs. (b) Time profiles of the transient absorption of $\mathbf{PTZ}^{\bullet+}$ monitored at 520 nm during the 355 nm laser flash photolysis of **N1** + **P1** (**ST10**, black), **N2** + **P2** (orange), **N1** + **P2** (blue) and **N2** + **P1** (green), respectively. (c) Time profiles of the transient absorption of $\mathbf{PTZ}^{\bullet+}$ monitored at 520 nm during the 355 nm laser flash photolysis of **N1** + **C** + **P2** (cyan). The represented profiles were obtained from the accumulation of 32 laser shots.

DNA assembly is formed. Thus, selective CT in only the complementary sequences was observed. Therefore, the DNA assembly system constructed by simply mixing complementary sequences certainly makes CT over 200 Å in DNA assemblies possible.

In this study, we have demonstrated that CT in DNA assemblies occurs without CT inhibition by the sticky end and is highly sensitive to the complementary sequence of the sticky end DNA. Continued studies, which focus on the CT in DNA tiles constructed from crossover DNA, are now underway.

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