Supplementary Data

A comprehensive evaluation of a typical plant telomeric G-quadruplex (G4) DNA reveals the dynamics of G4 formation, rearrangement, and unfolding

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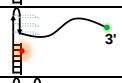
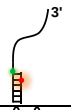
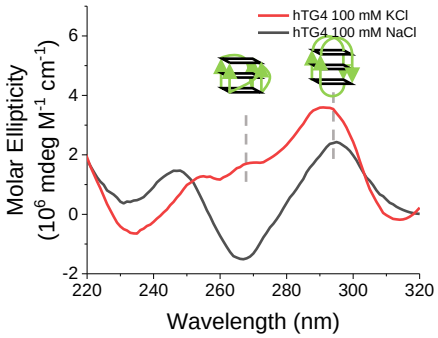
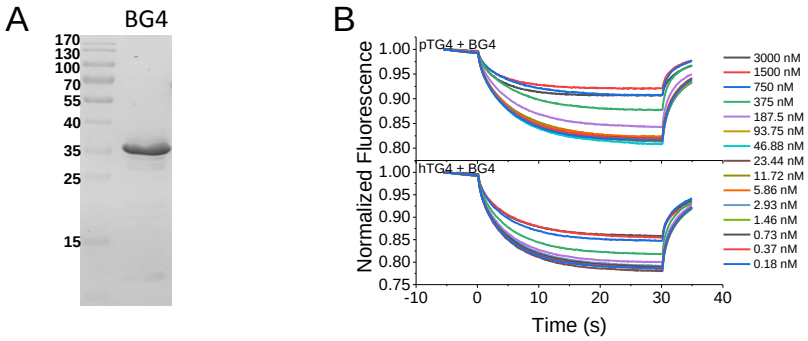
Supplementary Table S1. Structures and sequences of the substrates used in this study.

Name	Structure	Sequences (5'-3') of substrates for smFRET
DG4		CCTTACGTGCACAGGAGTT**GGGTTTAGGGTTTAGGGTTTAGGG**TT CTCCTGTGCACGTAAGG-biotin
DG3S		CCTTACGTGCACAGGAGTT**GGGTTTAGGGTTTAGGG**TTTTTTTTT CTCCTGTGCACGTAAGG-biotin
DG2S		CCTTACGTGCACAGGAGTT**GGGTTTAGGG**TTTTTTTTTTTTTTTT CTCCTGTGCACGTAAGG-biotin
DG4S		CCTTACGTGCACAGGAGTT**GGGTTTAGGGTTTAGGGTTTAGGG**TTTTTTTTTTT CTCCTGTGCACGTAAGG-biotin
DS		CCTTACGTGCACAGGAGTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTT CTCCTGTGCACGTAAGG-biotin
DG4S'		CCTTACGTGCACAGGAGTT**GGGTTTAGGGTTTAGGGTTTAGGG**TTTTTTTTTTTT CTCCTGTGCACGTAAGG-biotin
		Sequences (5'-3') of substrates for CD
pTG4		**GGGTTTAGGGTTTAGGGTTTAGGG**
		Sequences (5'-3') of substrates for MST
FAM-hTG4		FAM-T**GGGTTAGGGTTAGGGTTAGGG**
FAM-pTG4		FAM-T**GGGTTTAGGGTTTAGGGTTTAGGG**

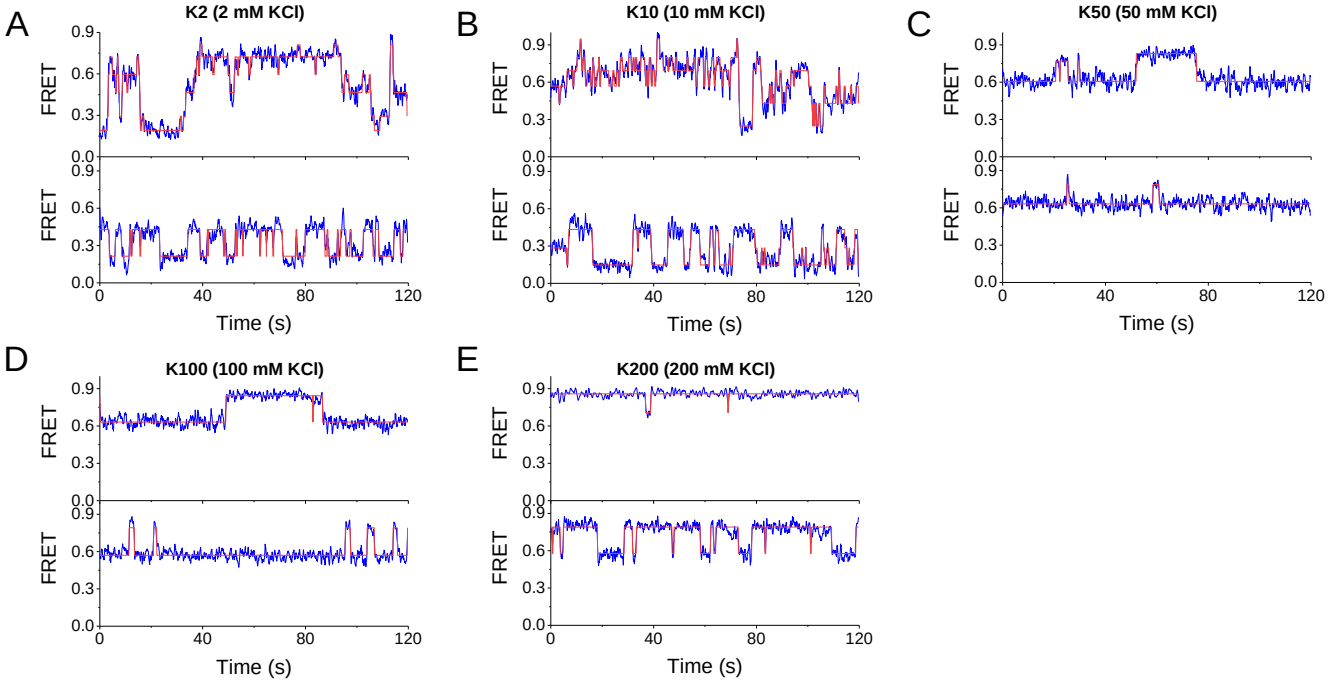
Color: Red, Cy5; Green, Cy3; blue, FAM. **Bold**: **G4/G3/G2 sequences**. Underline: dsDNA forming sequences.



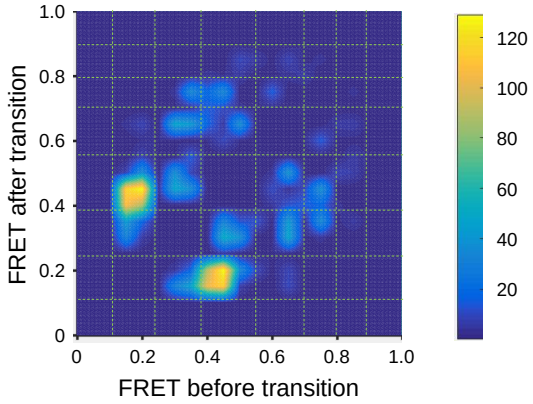
Supplementary Figure S1. CD spectra of hTG4 in 100 mM NaCl (black line) and 100 mM KCl (red line).



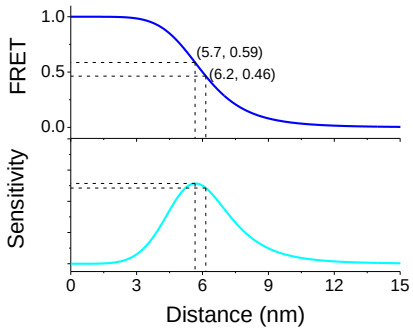
Supplementary Figure S2. (**A**) Analysis of the purified BG4 by SDS-PAGE. (**B**) MST time traces for the interactions of pTG4 and hTG4 with BG4 at 22 ℃.



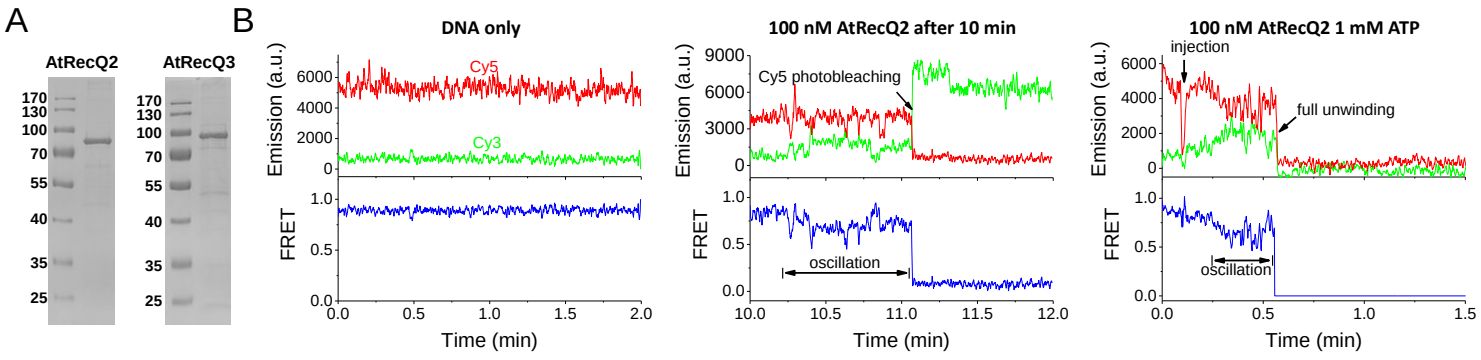
Supplementary Figure S3. Representative FRET traces of DG4 in 2 mM (A), 10 mM (B), 50 mM (C), 100 mM (D) and 200 mM (E) K^+. Dynamic states were determined by hidden Markov modeling (red line).



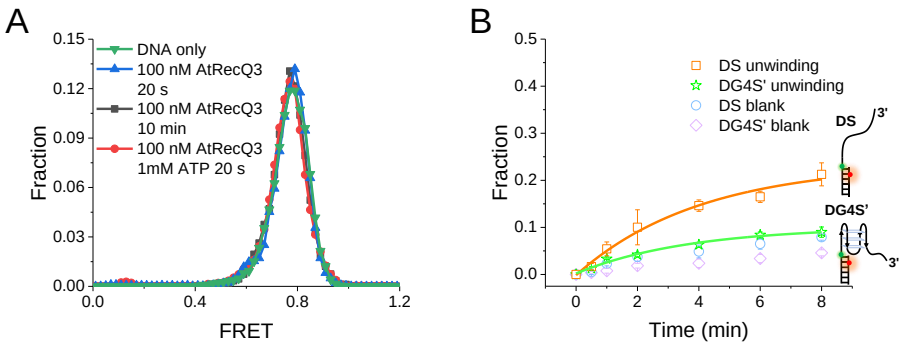
Supplementary Figure S4. The transition density plots (TDP) for 2 mM KCl from 47 single-molecule traces.



Supplementary Figure S5. The change of Cy3-Cy5 FRET is the most prominent around 0.5.



Supplementary Figure S6. (**A**) Analysis of the purified AtRecQ2 and AtRecQ3 by SDS-PAGE. (**B**) Representative single-molecule traces under different experimental treatments shown on the top of the panels in 100 mM KCl and 10 mM $MgCl_2$. FRET oscillations should be pTG4 unfolding. In the presence of 100 nM AtRecQ2 and 1 mM ATP (right panel), the oscillations were able to be terminated by the unwinding of duplex DNA (full unwinding) sometime, judging from the simultaneous disappearance of both dyes, because of the extremely low probability for Cy3-Cy5 simultaneous photobleaching.



Supplementary Figure S7. AtRecQ3 can not unfold pTG4. (**A**) FRET histograms for DG4S alone, in the presence of 100 nM AtRecQ3, and of 100 nM AtRecQ3 with 1 mM ATP at different times. (**B**) Unwinding fractions of DS and DG4S' at different times after addition of 100 nM AtRecQ3 and 1 mM ATP in the presence of 100 mM KCl and 10 mM $MgCl_2$.