Electronic Supplementary Material (ESI) for ChemComm.

Supporting Information

Highly sensitive and selective mercury sensor based on mismatched base pairing with ^{dioxT}T

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Materials

Oligonucleotides containing ^{diox}**T** was prepared by followed previous method.[1] Other oligomers were synthesized and HPLC purified by SIGMA-Genosys. All other chemicals and solvent including metal salts, $HgCl_2$, $AgNO_3$, $MgCl_2$, $NiCl_2$, $ZnCl_2$, $CaCl_2$, $CoCl_2$, $Cu(NO_3)_2$, $CdCl_2$ and $Pd(NO_3)_2$, were purchased from Sigma-Aldrich Chemicals Co., Wako Pure Chemical Ind. Ltd., TCI, or Kanto Chemical Co. Inc.

UV-melting measurement

Melting temperatures were determined by measuring changes in absorbance at 260 nm as a function of temperature using a JASCO V-650 UV/VIS spectrophotometer. JASCO PAC-743R equipped with a high performance temperature controller and micro auto eight-cell holder. Absorbance was recorded in the forward and reverse direction at temperatures from 5 to 95 °C at a rate of 1 °C/min. Before starting melting temperature experiment, the samples were denatured at 95 °C for 5 min and annealed slowly to RT then stored at 5 °C.

Steady state fluorescence measurement

Fluorescence measurements were conducted using fluorescence cells with a 0.5-cm path length on a JASCO FP-6300 Spectrofluorometer equipped with a JASCO EHC-573 temperature controller. The emission spectra were recorded from 350 nm to 600 nm with an excitation wavelength at 328 nm.

Time resolved fluorescence measurement

Fluorescence decay curves were collected on a HORIBA Fluorocube 3000U-SHK using an LED laser source for excitation. All samples were excited at 317 nm and the fluorescent decay was observed at 380 nm. TAC range was 100 ns and repetition rate was 1 MHz.

CD Spectroscopy

CD spectra of 2 μM oligonucleotide solutions collected in 0.5-nm steps from 320 to 220 nm were measured using JASCO J-805LST Spectrometer in a 1 cm quartz cuvette at 20 ^{o}C. Each spectrum shown is the average of two individual scans.

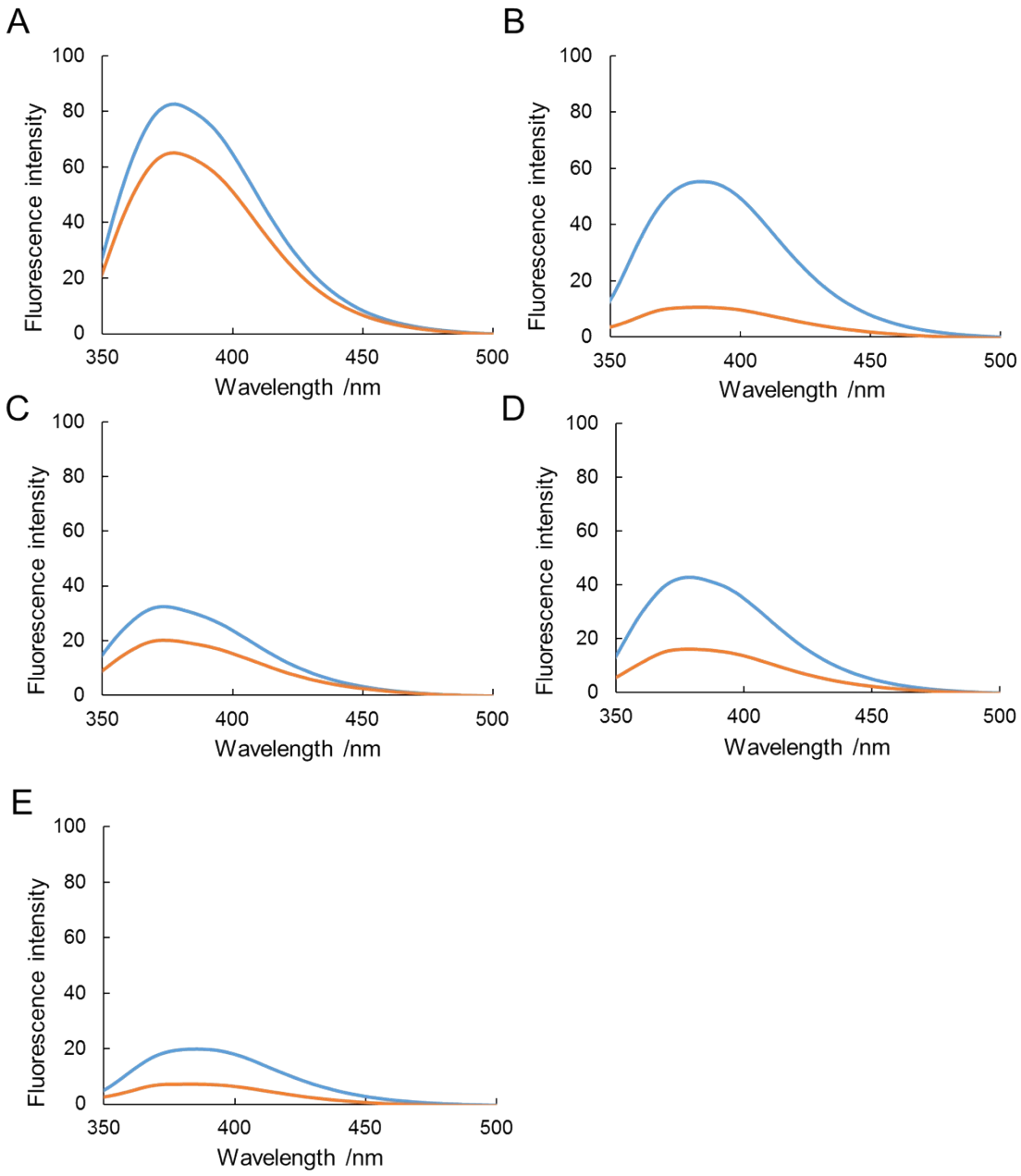

Figure S1. Fluorescent spectra of duplexes with (A) ^{diox}**T**-A, (B) ^{diox}**T**-T, (C) ^{diox}**T**-G, (D) ^{diox}**T**-C and (E) ^{diox}**T**-^{diox}**T** mismatched base pair in absence (blue) and presence (orange) of 1 eq Hg^{2+} ion. The metal ion was added, and the sample was incubated for 3 h.

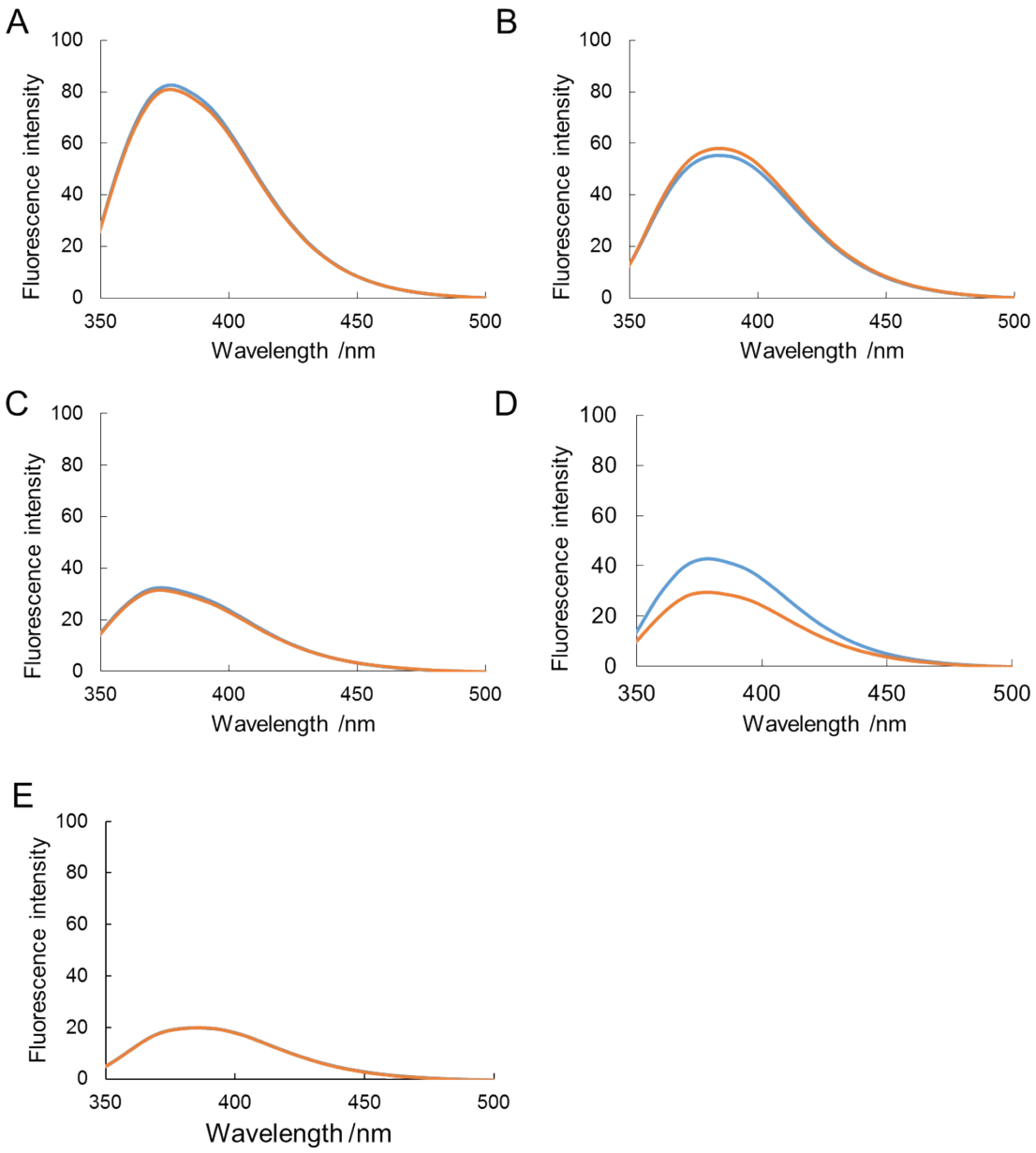

Figure S2. Fluorescent spectra of duplexes with (A) ^{diox}**T**-A, (B) ^{diox}**T**-T, (C) ^{diox}**T**-G, (D) ^{diox}**T**-C and (E) ^{diox}**T**-^{diox}**T** mismatched base pair in absence (blue) and presence (orange) of 1 eq Ag^{+} ion. The metal ion was added, and the sample was incubated for 3 h.

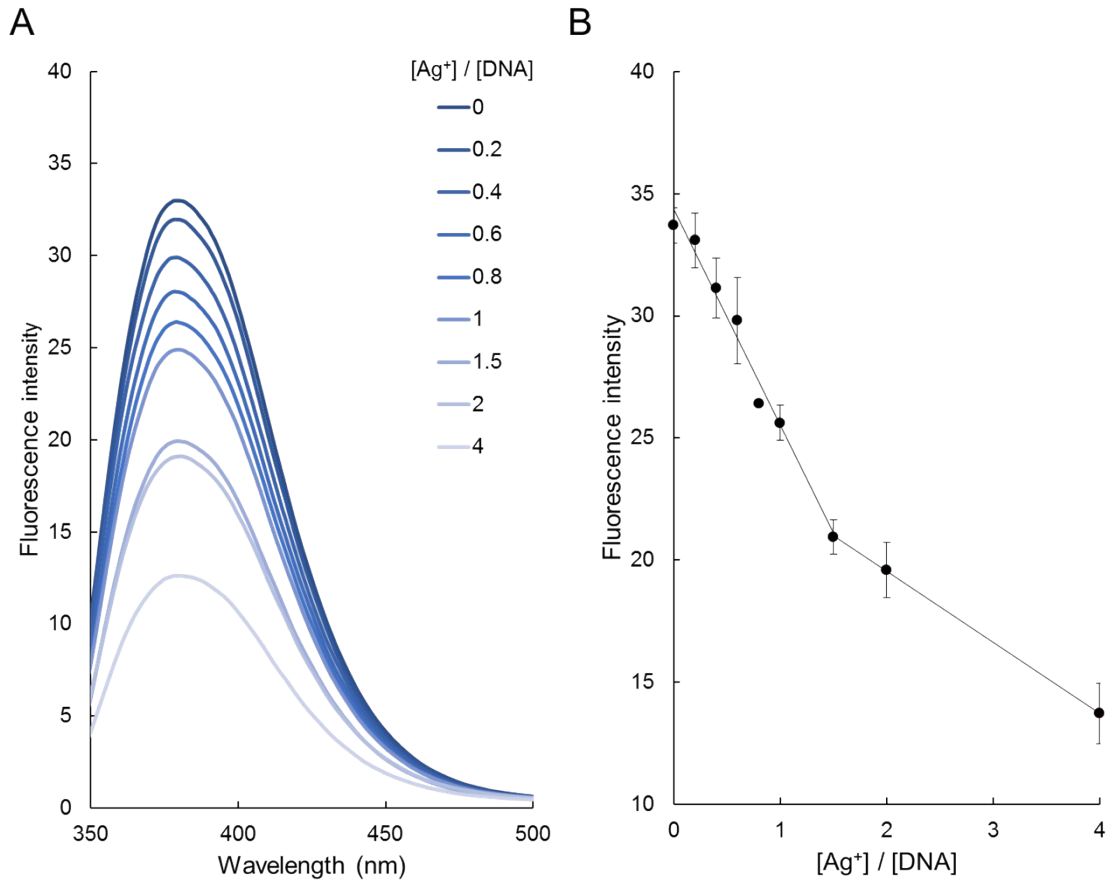

Figure S3. (A) Fluorescence spectra of DNA-containing mismatched base pair ^{diox}**T**-C upon titration of Ag^+ ion (B) Plot of fluorescence intensity based on the ratio of [Ag^+] to [DNA]. All samples contained 2 μM DNA and 100 mM $NaNO_3$ in 10 mM MOPS buffer (pH 7.0). After the metal ion were added, the samples were incubated for 3 h.

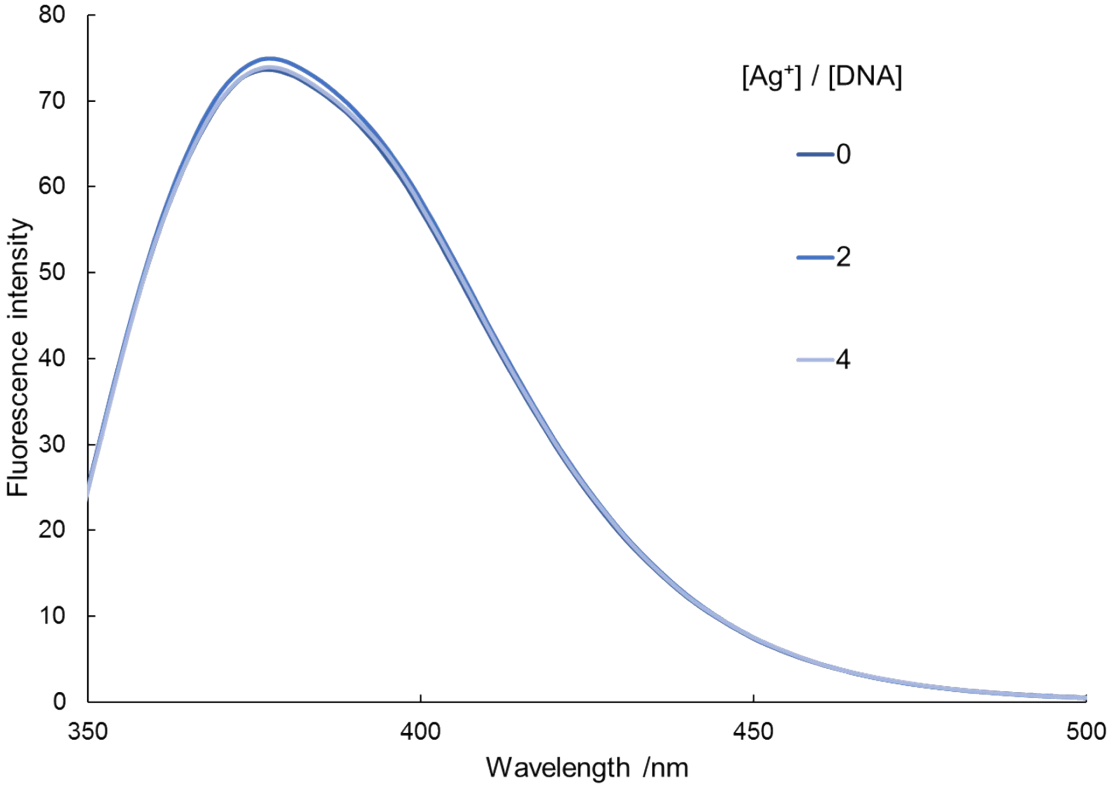

Figure S4. Fluorescence spectra of DNA-containing mismatched base pair ^{dioxT}-A upon addition of 0, 2, 4 eq Ag^+ ion. All samples contained 2 μM DNA and 100 mM $NaNO_3$ in 10 mM MOPS buffer (pH 7.0). After the metal ion were added, the samples were incubated for 3 h.

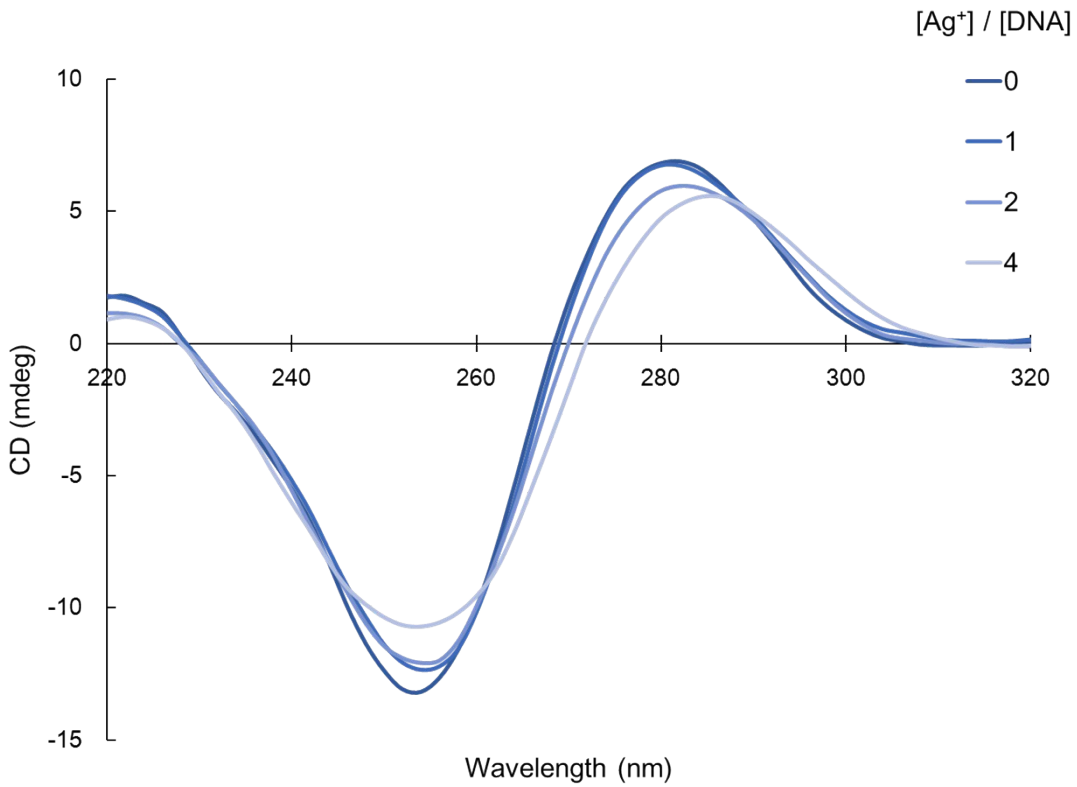

Figure S5. CD spectra of DNA-containing mismatched base pair ^{diox}**T**-C upon addition of 0, 1, 2, 4 eq Ag^+ ion. All samples contained 2 μM DNA and 100 mM $NaNO_3$ in 10 mM MOPS buffer (pH 7.0). After the metal ion were added, the samples were incubated for 3 h.

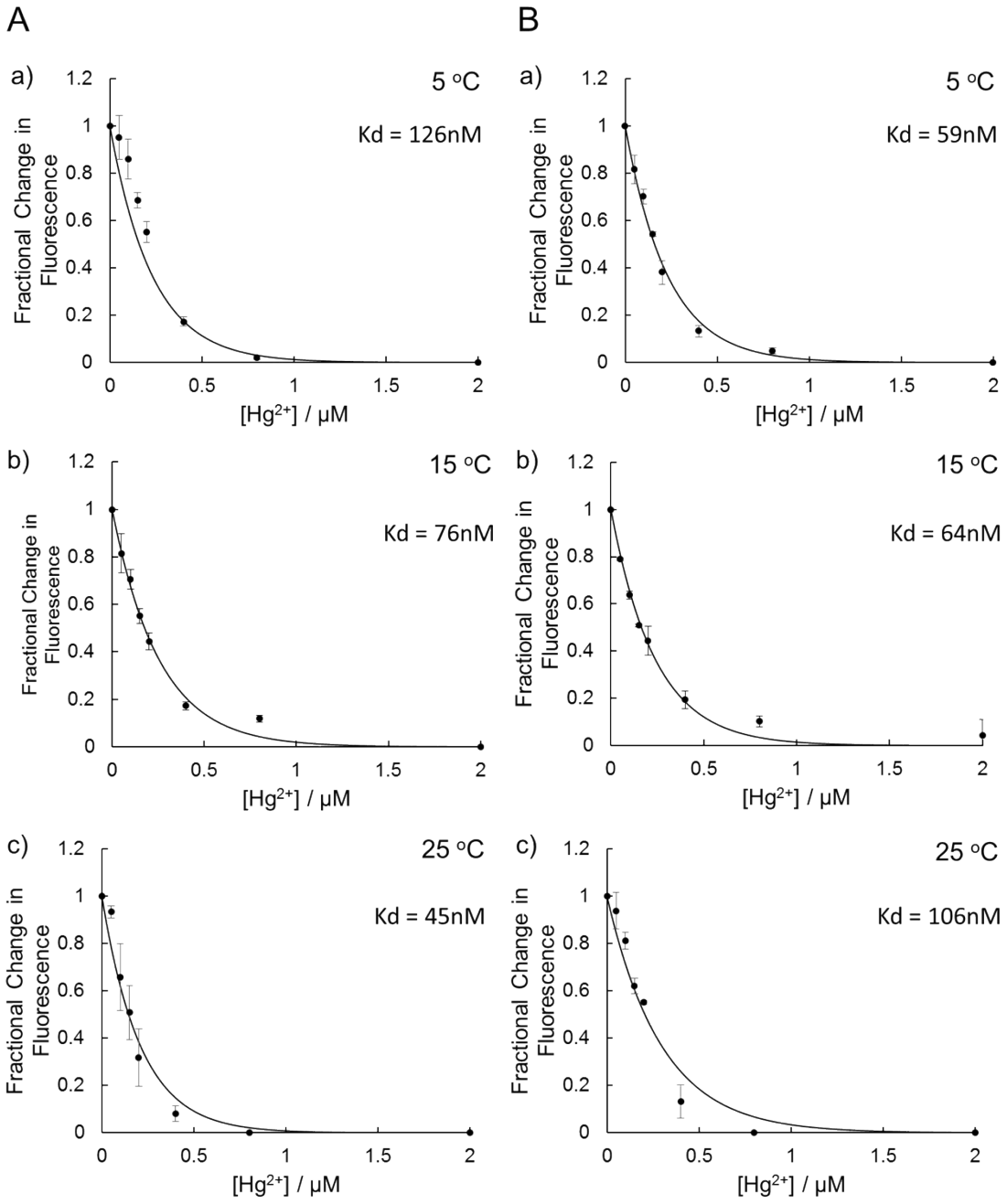

Figure S6. Normalized fluorescence decreases of duplexes upon addition of Hg^{2+} ion at 5 °C (a), 15 °C (b) and 25 °C (c); (A) ^{dioxT}**T**-T mismatched base pair, (B) ^{dioxT}**T**-C mismatched base pair. The fitting curve was obtained by origin 2018. All samples contained 200 nM DNA and 100 mM $NaNO_3$ in 10 mM MOPS buffer (pH 7.0). After the metal ions were added, the samples were incubated for 30 min. All experiments were performed in duplicates, average values were plotted.

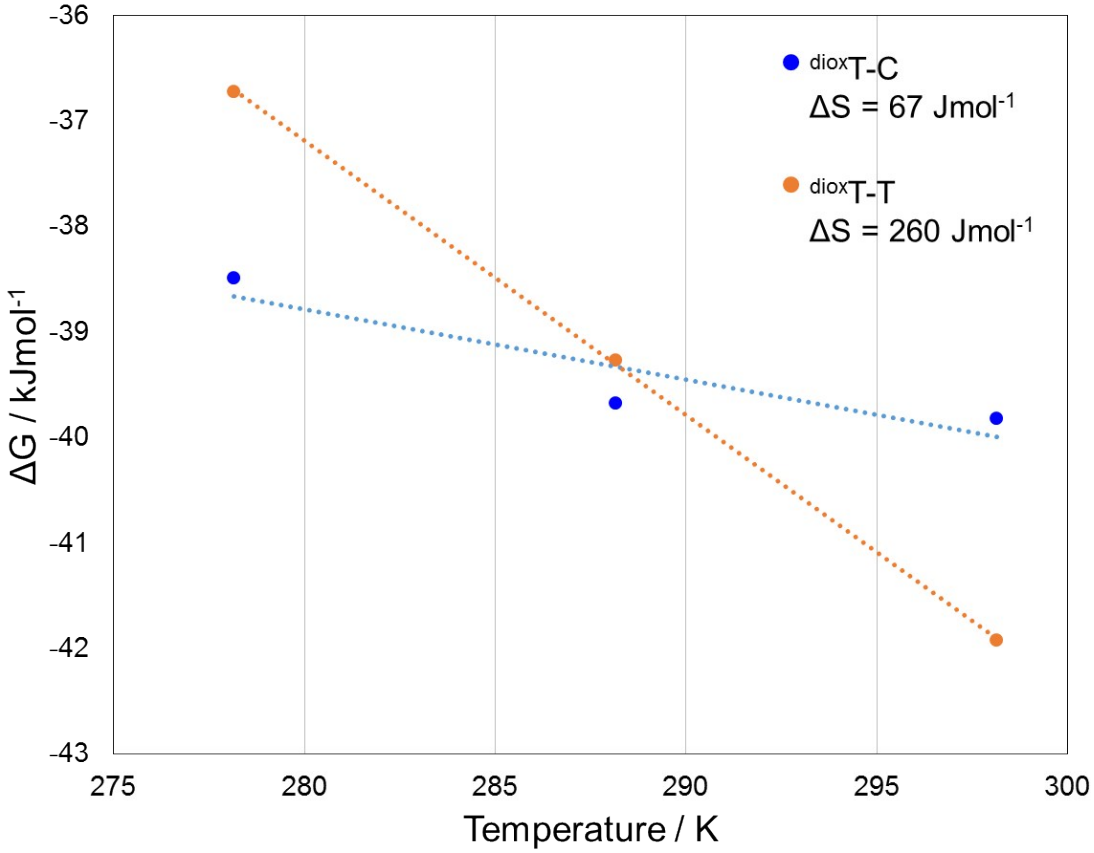

Figure S7. Plot of ΔG for **^{diox}T**-T (orange) and **^{diox}T**-C (blue) mismatched base pair, in which ΔG is calculated by ΔG = -RTln($1/K_d$), versus temperature. The slope of linear regression indicates the ΔS of **^{diox}T**-T and **^{diox}T**-C mismatched base pair. All experiments were performed in duplicates, average values were plotted.

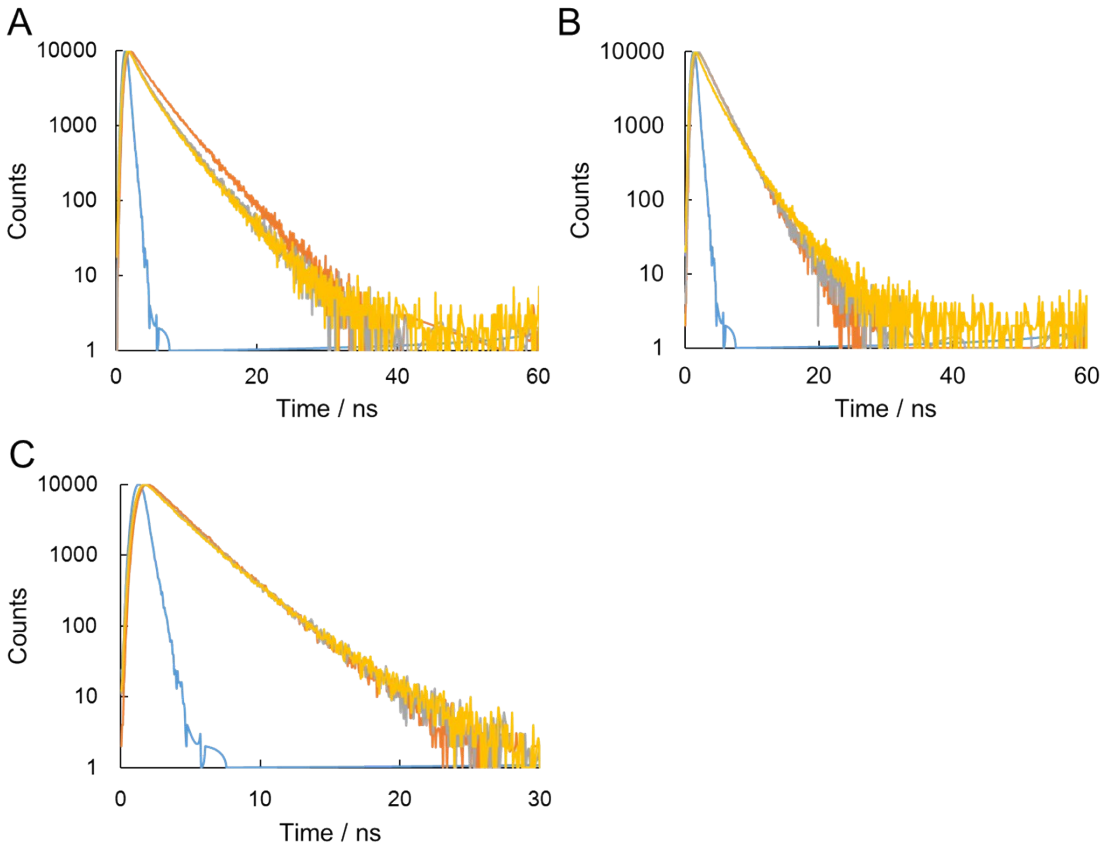

Figure S8. Fluorescence decay curve of duplexes with (A) ^{diox}**T**-T mismatched base pair and (B) ^{diox}**T**-C mismatched base pair in absence (orange) and presence of 1 eq (gray) and 2 eq (yellow) of Hg^{2+} ion. (C) ^{diox}**T**-C mismatched base pair in absence (orange) and presence of 1 eq (gray) and 2 eq (yellow) of Ag^{+} ion.

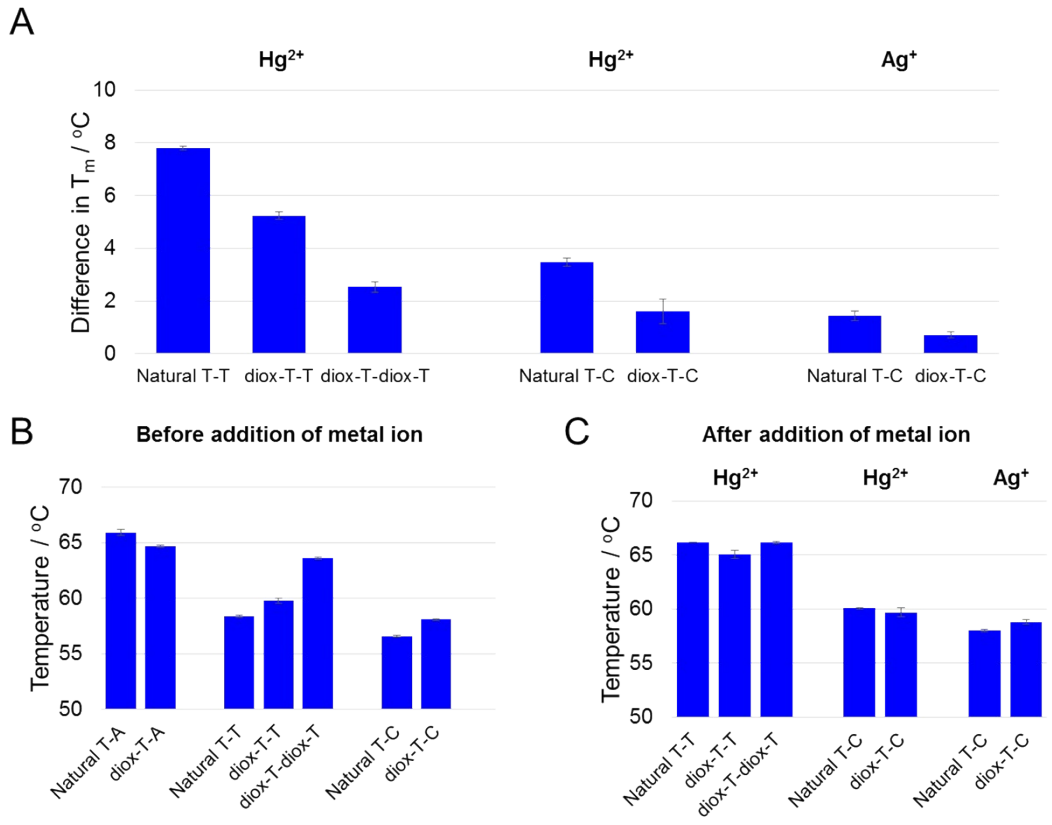

Figure S9. (A) Differences in Tm with standard error between mismatched (T-T, T-C, ^{diox}**T**-T, ^{diox}**T**-^{diox}**T**, ^{diox}**T**-C) in absence and presence of 1 eq Hg^{2+} ion or 1 eq Ag^{+} ion. Melting temperature of duplexes with each mismatched base pair in (B) absence and (C) presence of 1eq Hg^{2+} ion or 1 eq Ag^{+} ion. The values were obtained from duplicated experiment. The metal ions were added, and the sample was incubated for 3 h.

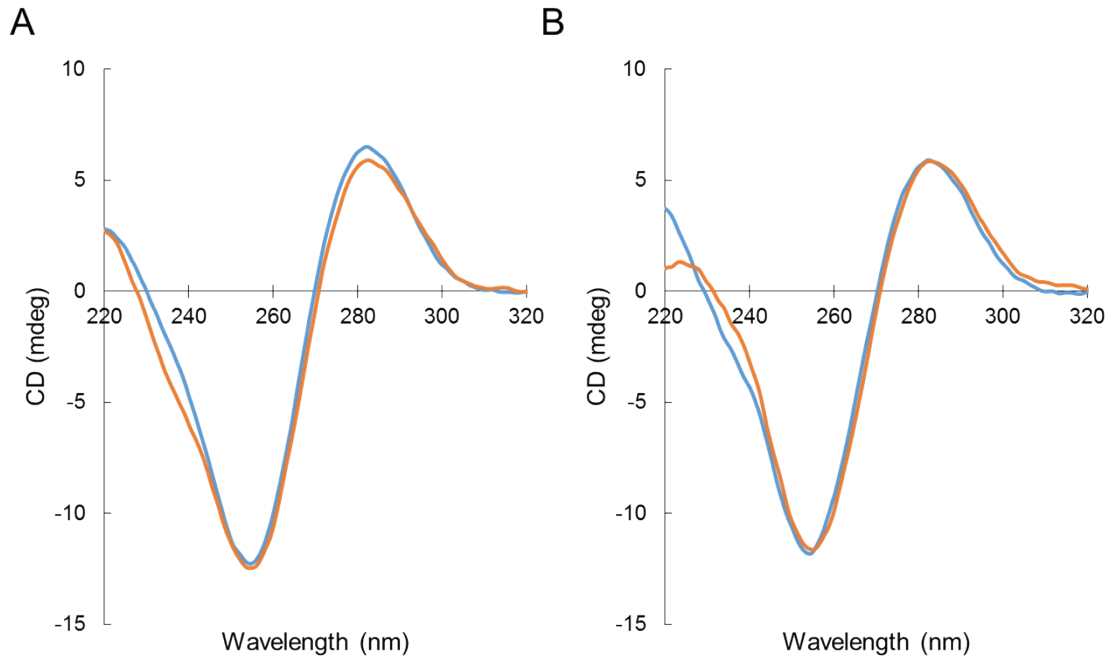

Figure S10. CD spectra of duplexes with (A) ^{diox}**T**-T or (B) ^{diox}**T**-C mismatched base pair in the absence (blue) and presence (orange) of 1eq Hg^{2+} ion. The metal ion was added, and the sample was incubated for 3 h.

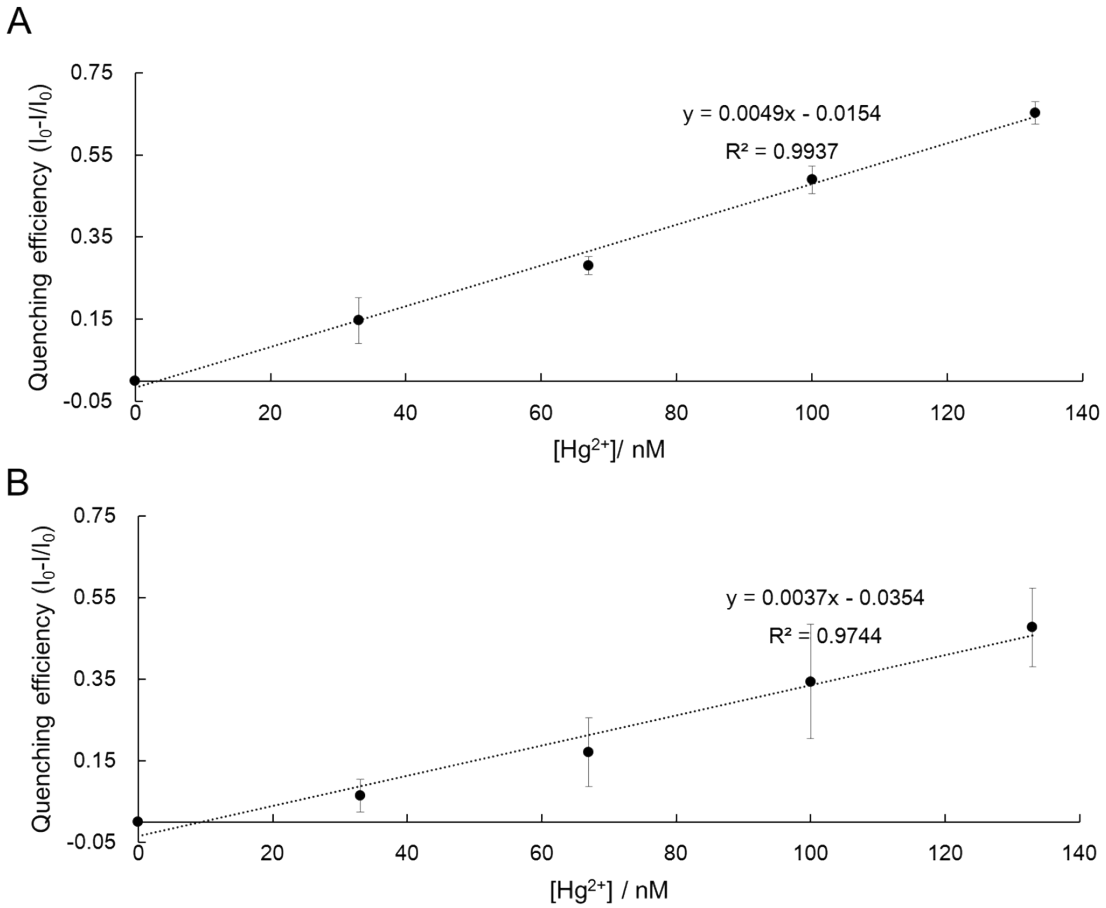

Figure S11. Calibration curve for Hg^{2+} determination by duplex containing (A) **^{diox}T**-T (A) and (B) ^{diox}T-C mismatched base pair. All samples contained 133 nM DNA and 100 mM $NaNO_3$ in 10 mM MOPS buffer (pH 7.0). After the metal ions were added, the samples were incubated for 30 min.

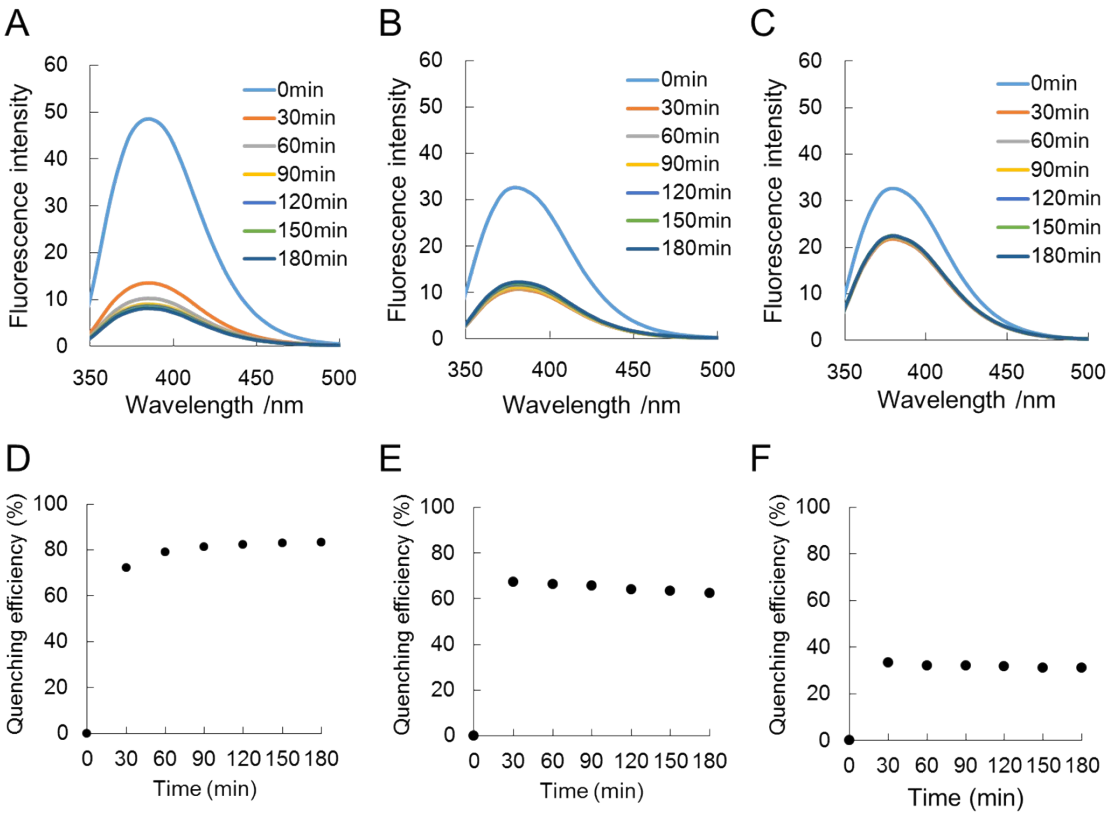

Figure S12. Time-dependent fluorescence spectra with duplexes containing ^{diox}**T**-T or ^{diox}**T**-C mismatched base pair in presence of 1 eq Ag^{+} ion or Hg^{2+} ion. (A) ^{diox}**T**-T mismatched base pair in presence of 1eq Hg^{2+} ion, (B) ^{diox}**T**-C mismatched base pair in presence of 1eq Hg^{2+} ion, (C) ^{diox}**T**-C mismatched base pair in presence of Ag^{+} ion. Time-dependent quenching efficiency of (D) ^{diox}**T**-T mismatched base pair in presence of Hg^{2+} ion, (E) ^{diox}**T**-C mismatched base pair in presence of Hg^{2+} ion, (F) ^{diox}**T**-C mismatched base pair in presence of Ag^{+} ion.

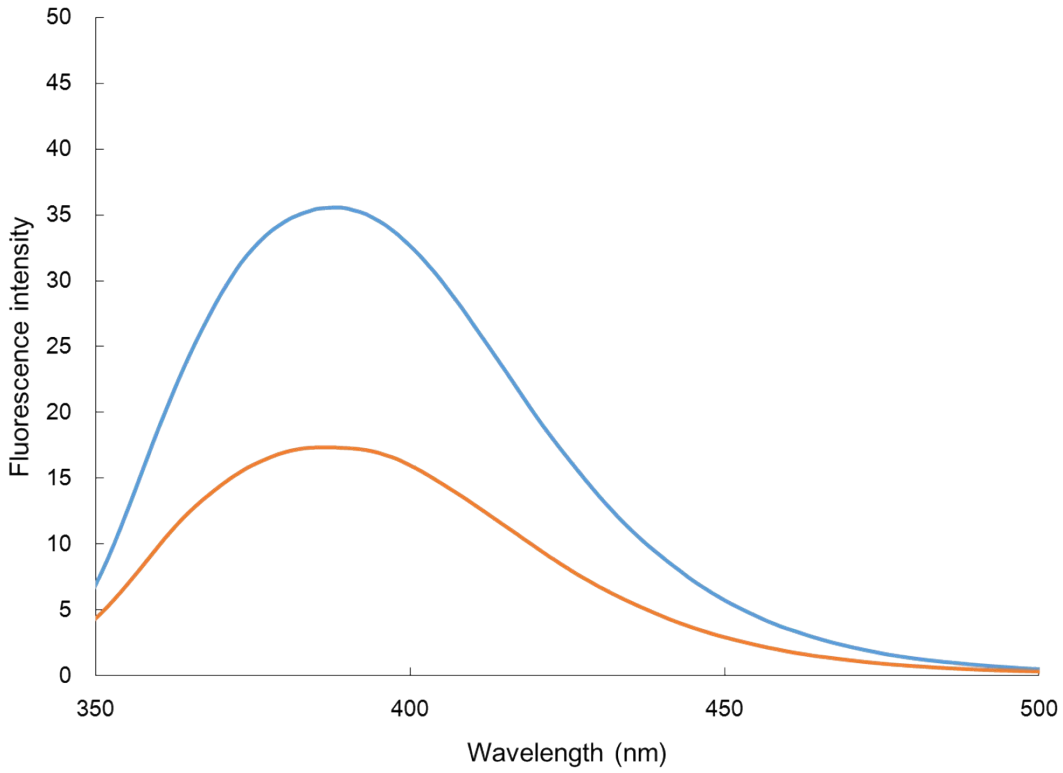

Figure S13. Fluorescence spectra of single stranded DNA containing ^{diox}**T** in absence (blue) and presence (orange) of Hg^{2+} ion.

Table S1. Time-resolved fluorescence parameters of double stranded DNA containing ^{diox}**T**-T or ^{diox}**T**-C mismatched base pair in absence and presence of mercury ion.

	τ1 (ns)	A1	τ2 (ns)	A2	τ3 (ns)	A3	χ^2	$\langle\tau\rangle^a$ (ns)
^{diox}T-T	1.50	0.23	4.06	0.77			0.97	3.47
^{diox}T-T + 1eq Hg^{2+}	4.85	0.1	2.03	0.41	4.16	0.49	1.10	2.92
^{diox}T-T + 2eq Hg	4.37	0.12	2.06	0.44	4.16	0.44	0.89	2.79
^{diox}T-C	1.36	0.23	2.62	0.77			0.90	2.33
^{diox}T-C + 1eq Hg^{2+}	1.61	0.38	2.80	0.62			1.18	2.35
^{diox}T-C + 2eq Hg^{2+}	1.53	0.14	1.60	0.38	3.39	0.48	1.05	2.25

$^a\langle\tau\rangle = \alpha_1\tau_1 + \alpha_2\tau_2 + \alpha_3\tau_3$

Reference

1 S. Hirashima, J. H. Han, H. Tsuno, Y. Tanigaki, S. Park, H. Sugiyama, *Chem. – Eur. J.,* Accepted. DOI: 10.1002/chem.201900843