Electronic Supplementary Material (ESI) for Analyst.

Novel fluorescent cationic benzothiazole dye response to G-quadruplex aptamer as a novel K^+ sensor

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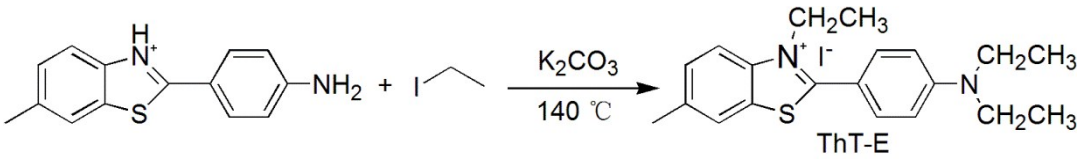
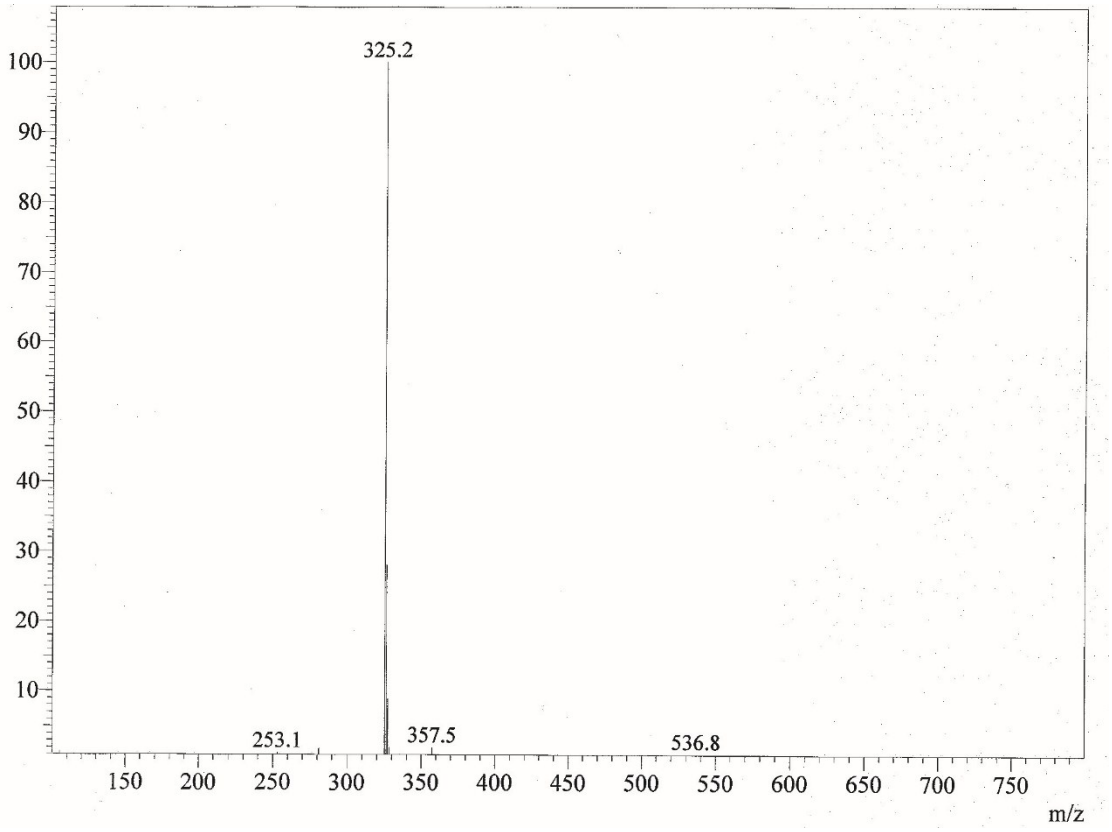
Scheme S1. Synthesis of ThT-E



Identification of ThT-E

a) Mass Spectrum

The calculated exact mass of $C_{20}H_{25}N_2S^+$ is 325.17.



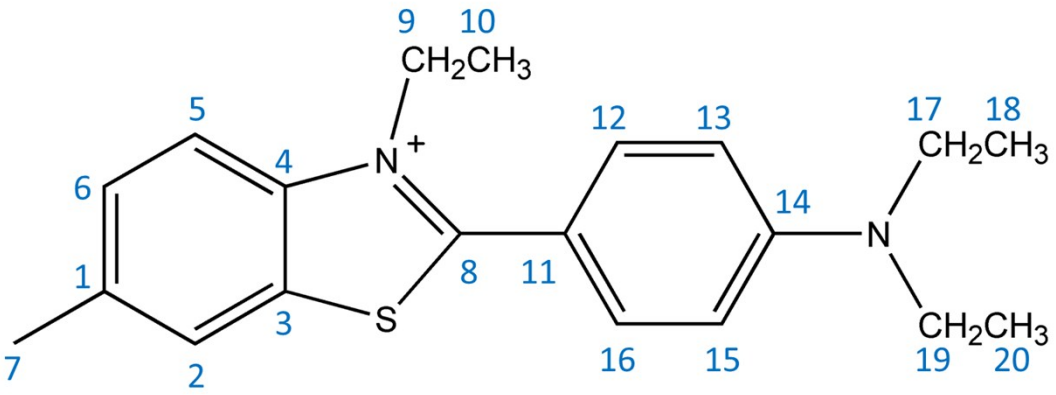
b) Elemental Analysis

Molecular formula of ThT-E: $C_{20}H_{25}IN_2S$

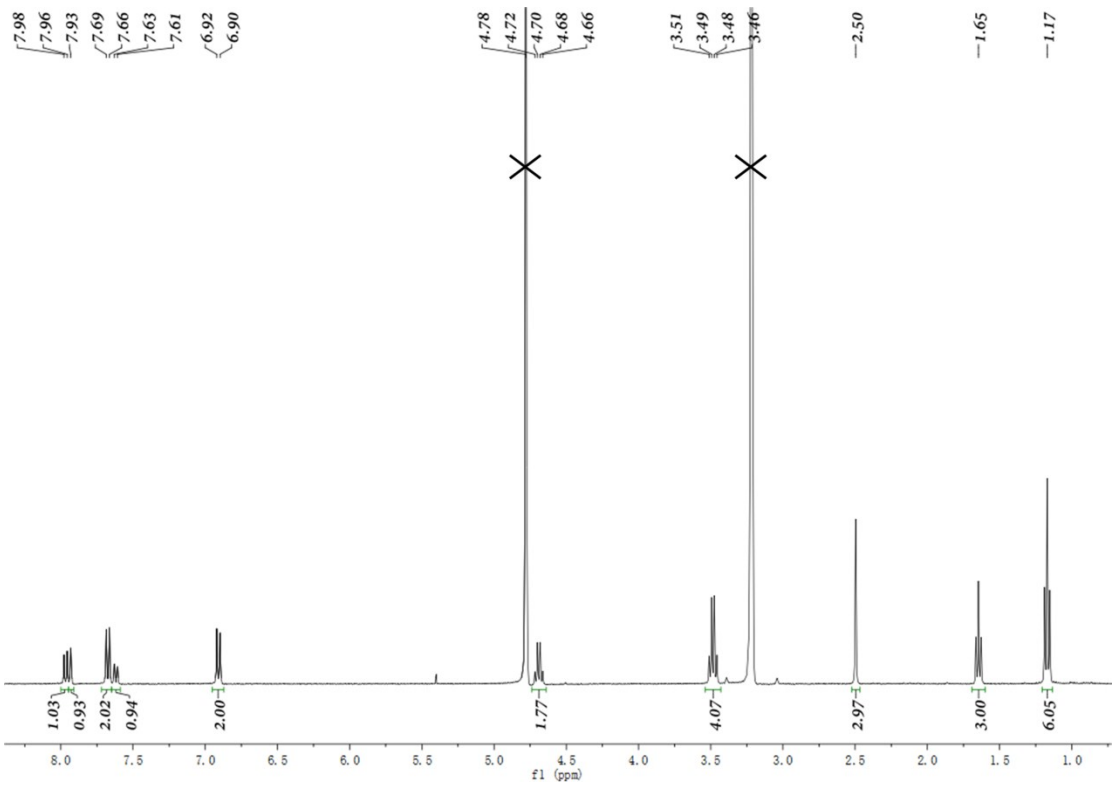
Element	N	C	H
Calculated Value（%）	6.19	53.10	5.57
Experimental Value (%)	5.91	52.26	5.57

c) NMR Spectrum

The numbering scheme of ThT-E



The 1H-NMR spectrum of ThT-E in CD_3OD

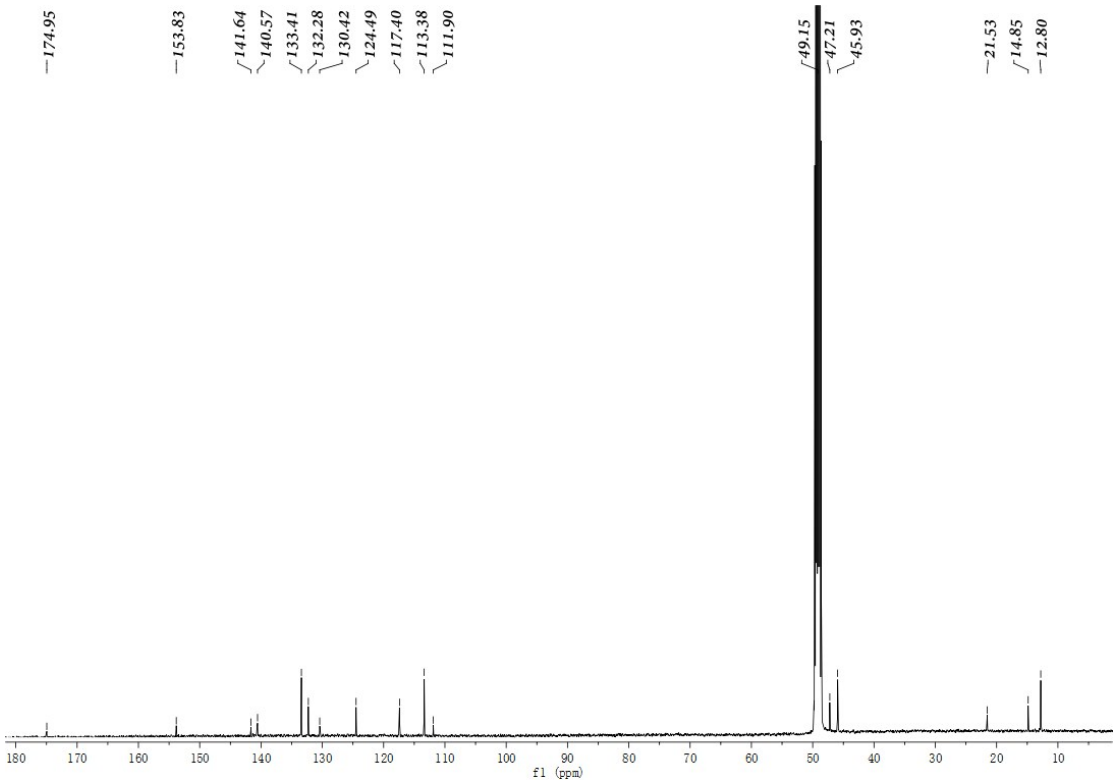


* The peaks of these protons are covered by the peak of the solvent CD_3OD

The full assignments of the proton peaks of ThT-E

Proton number	1H peak	Proton number	1H peak
1	--*	11	--*
2	7.93, s	12	7.69-7.66，d
3	--*	13	6.92-6.90, d
4	--*	14	--*
5	7.98-7.96, d	15	6.92-6.90, d
6	7.63-7.61, d	16	7.69-7.66，d
7	2.50, s	17	3.51-3.46, q
8	--*	18	1.17, t
9	4.72-4.66, q	19	3.51-3.46, q
10	1.65, t	20	1.17, t

The 13C-NMR spectrum of ThT-E in CD_3OD



* The peaks of these 13C are covered by the peak of the solvent CD_3OD

The full assignments of the 13C peaks of ThT-E

Proton number	13C peak	Proton number	13C peak
1	140.57	11	111.90
2	124.49	12	113.38
3	130.42	13	133.41
4	141.64	14	153.83
5	117.40	15	133.41
6	132.28	16	113.38
7	21.53	17	45.93
8	174.95	18	12.80
9	47.21	19	45.93
10	14.85	20	12.80

Experimental

Synthesis and identification of the ThT-E. To a sealed tube that contained a stir bar were added 2-(4-aminophenyl)-6-methylbenzothiazole (1.2 g, 5 mM), iodoethane (2.34 g, 15 mM) and potassium carbonate (1.38 g, 10 mM). The resulting mixture was stirred and heated at 140 ℃ for 14 h. Upon completion of the reaction, the mixture was cooled to room temperature and concentrated with the aid of a rotary evaporator. And the residue was purified by column chromatography on silica gel (the percent of methanol in dichloromethane from 2% to 20%). The pure product was isolated as a yellow solid (1.38 g, 61 percent). The purity was identified by mass spectrum (MS), nuclear magnetic resonance (NMR) and elemental analysis.

Chemicals and Materials. 2-(4-Aminophenyl)-6-methylbenzothiazole and iodoethane were obtained from J&K Company, Ltd. Dichloromethane and methanol were obtained from Beijing Chemical Plant (Beijing, China). Oligonucleotide oligo-3 was purchased from Zixi Bio Tech Co., Ltd (Beijing, China) and purified by high–performance liquid chromatography (HPLC). Oligo-3 was dissolved in ultrapure water, and then heated to 90 for 5 min following with gradually cooling to room temperature. Before the first using, the concentration of oligo-3 was determined by measuring the absorbance at 260 nm. The metal salts (NaCl, LiCl, $CaCl_2$, $ZnSO_4$, $MgSO_4$, NH_4Cl, $FeCl_3$, $CuSO_4$, $(CH_3COO)_2Pb$, $CoSO_4$) were purchased from Beijing Chemical Company and used without further purification. Ultrapure water was prepared with deionized water purified by Milli-Q Gradient ultrapure water system (Millipore) and used throughout all experiments.

Spectroscopy measurements. The fluorescence spectra were carried out on a Hitachi F-4600 spectrophotometer. Excitation and emission slits were set as 5 and 10 nm, respectively. Voltage was 700 V and the scan speed was set as 1200 nm/min. The concentration of ThT-E was fixed as 3 μM. CD spectra were taken on a Jasc0-810 automatic recording spectrometer from 200-500 nm with a 10-mm path-length quartz cuvette. The scan speed was set as 500 nm min^{-1} and each spectrum was the average of three scans. The lake water and urine were collected from Tsinghua University and two heathy volunteers, respectively. The concentrations of K^+ in urine were measured by an Inductively Coupled Plasma-Atomic Emission Spectrometer Prodigy 7. Before the test, the urine samples were centrifuged and diluted 500 and 40 folds for ICP-MS and fluorescence methods, respectively. All samples were maintained for 2 h before the spectra measurements.

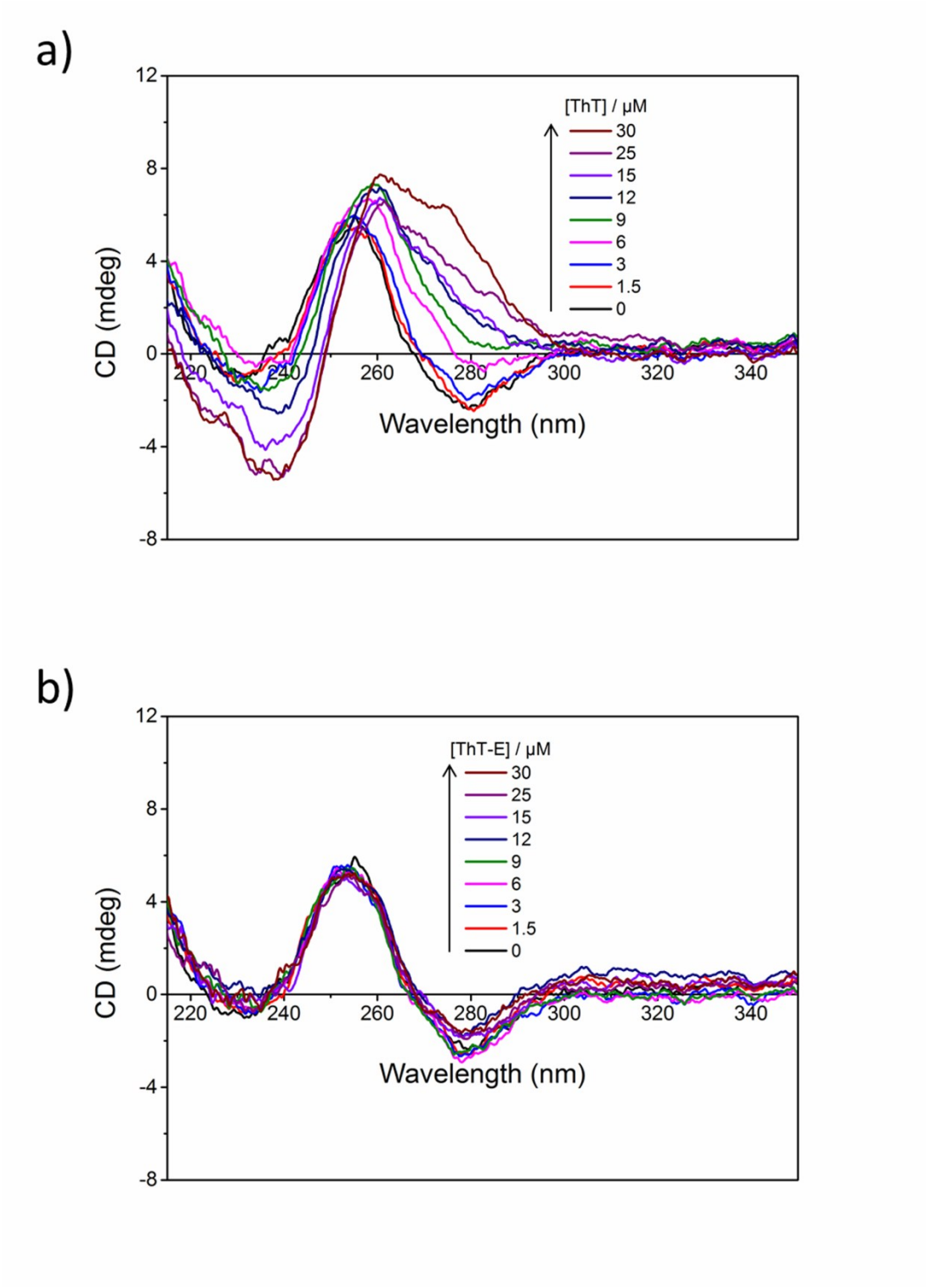


Figure S1. Circular dichroism spectra recorded for oligo-3 DNA (3 μM） solution, in a 20 mM Tris-HCl (pH 7.4). (A) ThT; (B) ThT-E (μM): (1) 0, (2) 1.5, (3) 3, (4) 6, (5) 6, (6) 9, (7) 12, (8) 15, (9) 25, (10) 30.

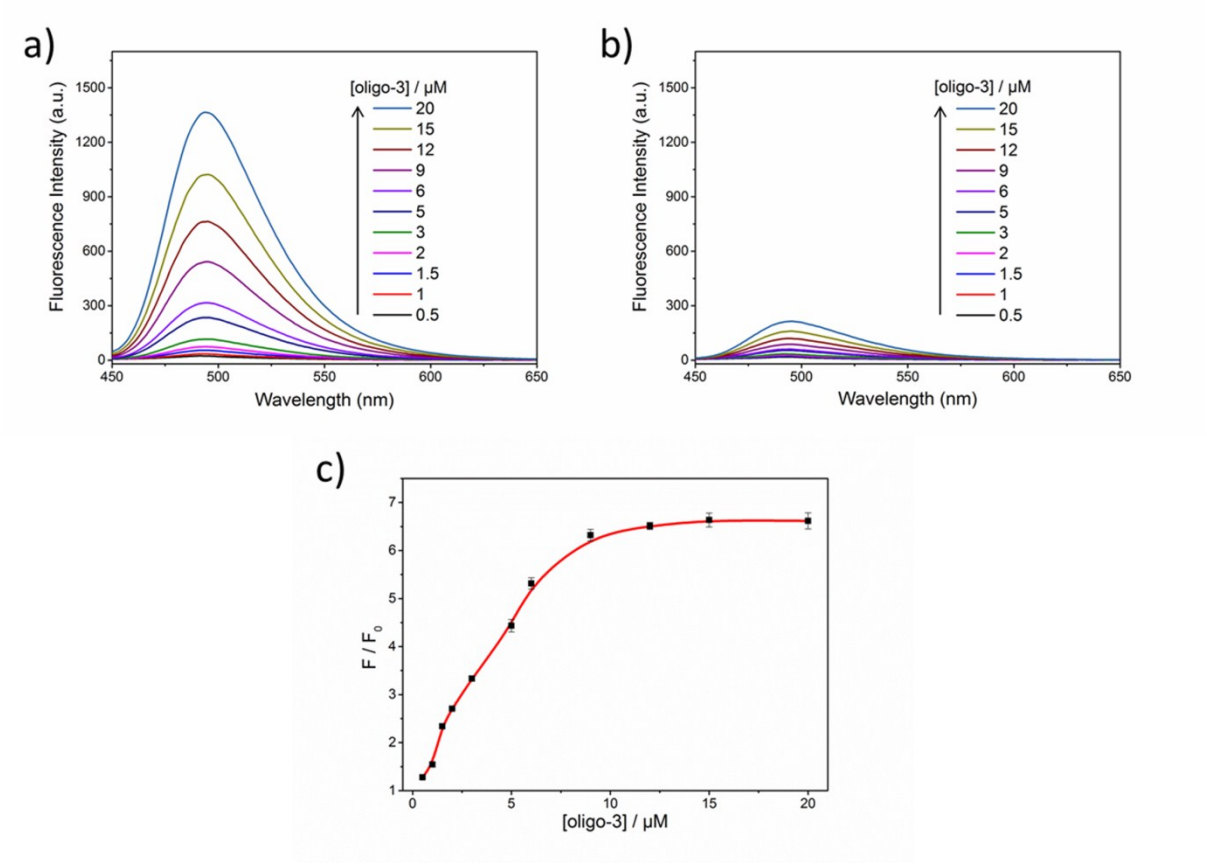


Figure S2. The fluorescence spectra of 3 μM ThT-E with increasing the amounts of the oligo-3 with (a) and without (b) 5 mM K^+ in 20 mM Tris-HCl buffer solution (pH=7.4). (c) Optimization of the aptamer concentration for the K^+ assay. F and F_0 stand for the fluorescence intensity of ThT-E at 492 nm in the presence or absence 5 mM K^+ solution, respectively.

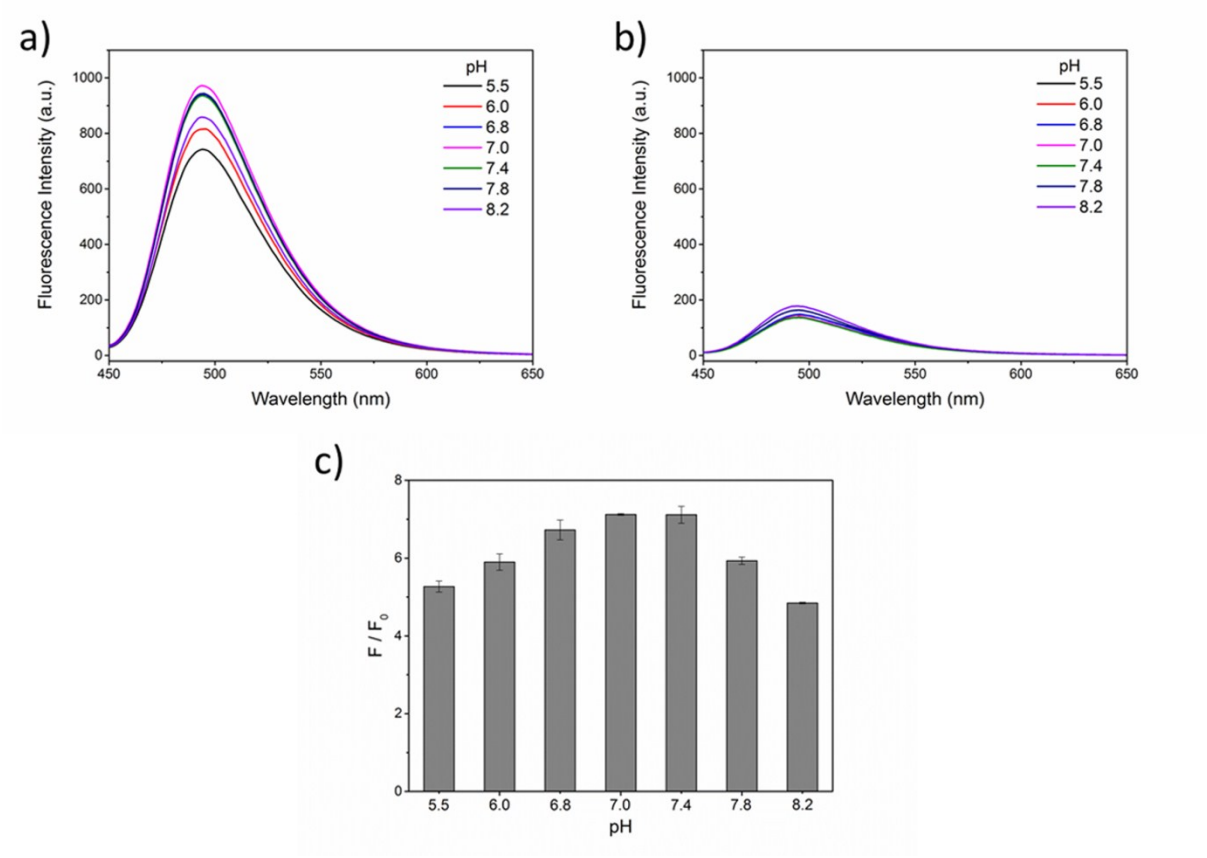


Figure S3. The fluorescence spectra of 3 μM ThT-E with 15 μM oligo-3 solution under different pH condition in the presence (a) and absence (b) 5 mM K^+ in 20 mM Tris-HCl buffer solution. (c) Optimization of the pH condition for the K^+ assay. F and F_0 stand for the fluorescence intensity of ThT-E at 492 nm in the presence or absence 5 mM K^+ solution, respectively.

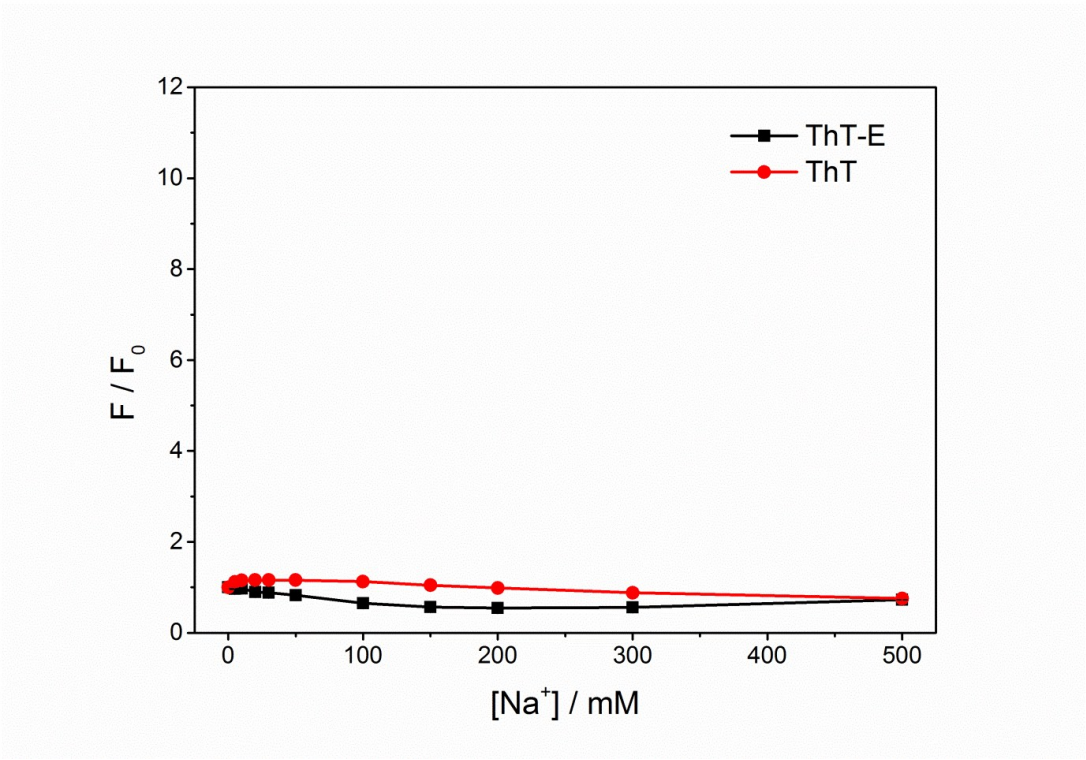


Fig.S4 Plot of the signal change (F/F_0) with increasing the concentration ranged from 0 to 500 mM of Na^+ in the presence of 15 μM oligo-3, 3 μM ThT-E and ThT, respectively.