Electronic Supplementary Material (ESI) for Analyst

Visual Detection of Potassium by A Cyanine Dye Supramolecular Aggregate Responsive to G-quadruplex Motif Transition

Hongxia Sun, Junfeng Xiang, Wei Gai, Qian Shang, Qian Li, Aijiao Guan, Qianfan Yang, Yan Liu, Yalin Tang* and Guangzhi Xu

Experimental

Sample preparation

MTC was prepared according to the method suggested by Hamer and Ficken,[1,2] and the purity has been proved by nuclear magnetic resonance (NMR) and mass spectroscopy (MS). The oligonucleotide, $d(AG_3(T_2AG_3)_3)$ (H22) was purchased from Invitrogen (Beijing, China), purified by PAGE. Methanol, KCl, Tris, HCl, and EDTA were all in analytical grade. Ultrapure water was prepared by Milli-Q gradient ultrapure water system. All the samples were measured in 10 mM Tris-HCl buffer solution (pH 7.2). All experiments were made in buffer solutions containing 10 mM Tris-HCl and 1 mM EDTA. The G-quadruplexes were prepared by simply dissolving H22 with the buffer solution with Na^+.

Spectroscopy measurement

CD spectra were collected from 200 to 350 nm on a Jasco-815 automatic recording spectropolarimeter with a 1-cm pathlength quartz cell at 25℃. Spectra were collected with scan speed of 1000 nm/min and a response time of 0.25 s. Each spectrum was the average of three scans. A buffer blank correction was made for all spectra. The cuvette-holding chamber was flushed with a constant stream of dry N_2 gas to avoid water condensation on the cuvette exterior.

Ultraviolet (UV) spectra were measured on an Agilent 8453 UV-visible spectrophotometer at the wavelength range 200～1000 nm using a 1 cm path cell at room temperature. Ultrapure water was used as reference.

Fluorescence spectra were recorded on a Hitachi F4500 spectrofluorometer (Japan) in a 1-cmx0.2-mm path-length quartz cell at room temperature. Xenon arc lamp was used as the excitation light source. The excitation and emission slits were both 10 nm. Excitation was set at 550 nm, and emission was collected from 560 to 720 nm. The scan speed was 240 nm/min.

Application.

Urine was used to confirm the feasibility of this aptasensor for analysis of real-world sample. Urine samples were harvested from five persons in a hospital, which have been measured by an automatic chemistry analyzer (Beckman LX-20). Add 500 μL buffer solution with 40μM H22, 150 mM Na^+, and 50 mM K^+, and 80 μL methanol solution with 200 μM MTC into the five urine samples (1 mL), then observe the colour change under an environment without sunlight.

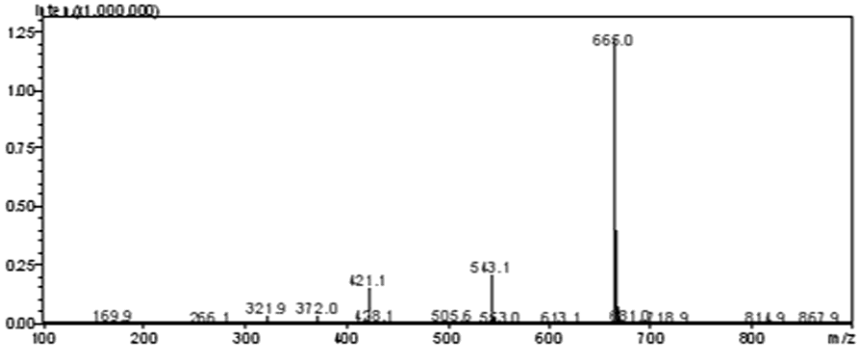
1 F. M. Hamer, *The chemistry of Hererocyclic Compounds*, Interscience, New York, 1964, pp. 148.

2 G. E. Ficken, *The Chemistry of Synthetic Dyes*, Academic Press, New York, 1971, pp. 228.

Identification of Cyanine Dye MTC

The structure and purification of cyanine dye MTC were identified by MS-ESI, elemental analysis, NMR and absorption spectroscopy.

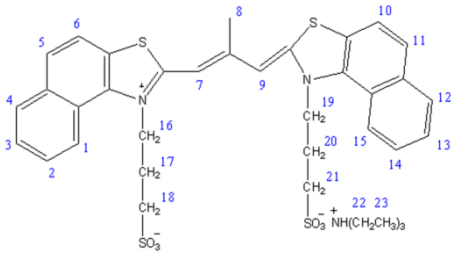
MS-ESI



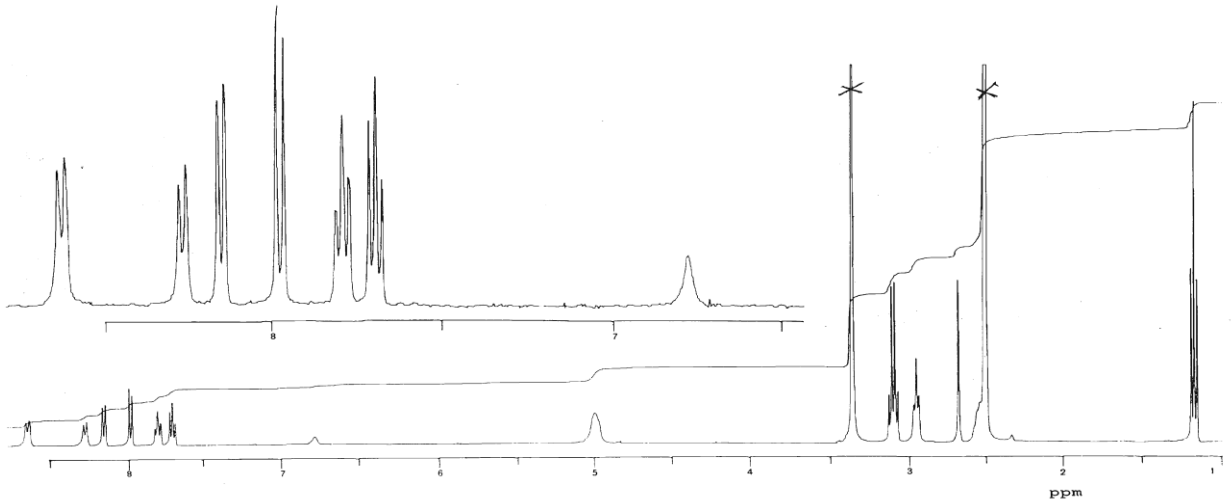
The MS-ESI spectrum of cyanine dye MTC

From the mass spectrum, the measured molecular weight of MTC is 666, which is consistent with the calculated value, 665.

NMR spectrum



The numbering scheme of molecular structure of MTC



The ^{1}H-NMR spectrum of cyanine dye MTC in DMSO-d_6

The full assignments of the proton peaks of MTC

Proton number	Proton kind	^{1}H peak	Proton number	Proton kind	^{1}H peak
1	Aromatic	8.16-8.14, d	13	Aromatic	7.70, t
2	Aromatic	7.80, t	14	Aromatic	7.80, t
3	Aromatic	7.70, t	15	Aromatic	8.16-8.14, d
4	Aromatic	7.99-7.97, d	16	Secondary	2.96, t
5	Aromatic	8.28-8.26, d	17	Secondary	--*
6	Aromatic	8.64-8.62, d	18	Secondary	4.99, s
7	Tertiary	6.777, s	19	Secondary	2.96, t
8	Primary	2.685, s	20	Secondary	--*
9	Tertiary	6.777, s	21	Secondary	4.99, s
10	Aromatic	8.64-8.62, d	22	Secondary	3.13-3.07, q
11	Aromatic	8.28-8.26, d	23	Primary	1.17, t
12	Aromatic	7.99-7.97, d			

* The peaks of these secondary protons are covered by the peak of the solvent DMSO-d_6

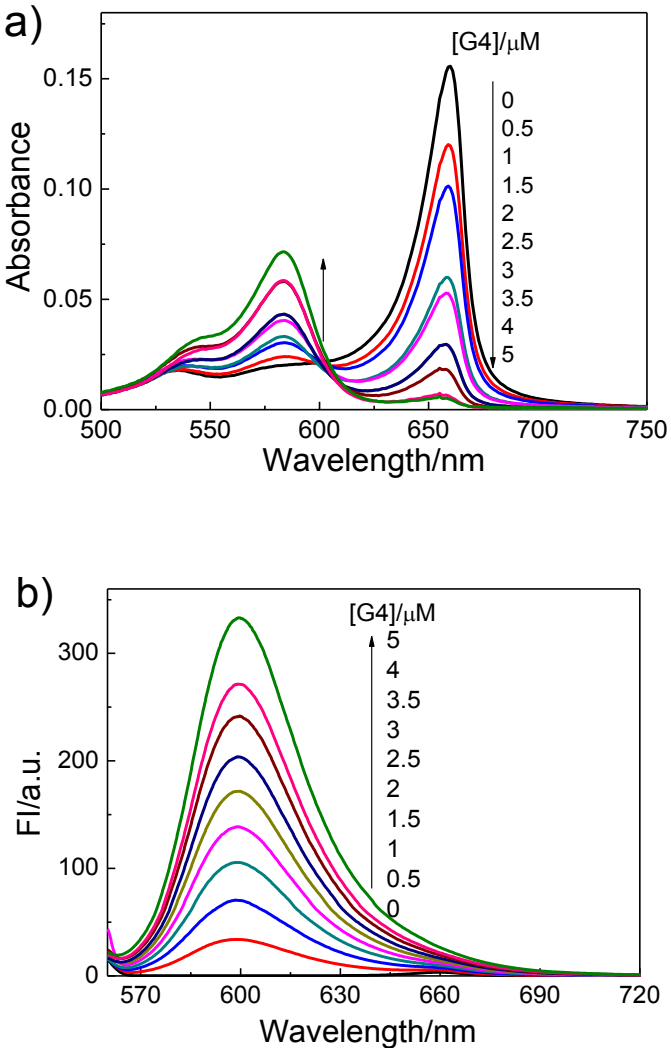


Fig. S1 The a) absorption spectra and b) fluorescence spectra of 3.3μM MTC in Tris-HCl buffer solution containing 20mM KCl with increasing amount of H22 G-quadruplexes (G4).

The absorbance at 580 nm belongs to MTC monomers and that at 660 nm belongs to MTC J-aggregates.[3] With an increase amount of mixed-type G-quadruplexes, the absorbance at 660nm decreased while that at 580 nm increased, corresponding to the switch from MTC J-aggregate to monomer. The result indicates a strong binding effect between MTC monomers and the mixed-type G-quadruplexes.

Similarly, a sharp increase of fluorescence intensity at 600 nm, which belongs to MTC monomers, with adding the telomeric mixed-type G-quadruplexes also indicates the strong interaction between MTC and mixed-type G-quadruplexes.

3 H. Sun, J. Xiang, Q. Yang, Q. Shang, Q. Zhou, Y. Zhang and G. Xu, Y. Tang, *Appl. Phys. Lett.*, 2011, **98**, 031103.

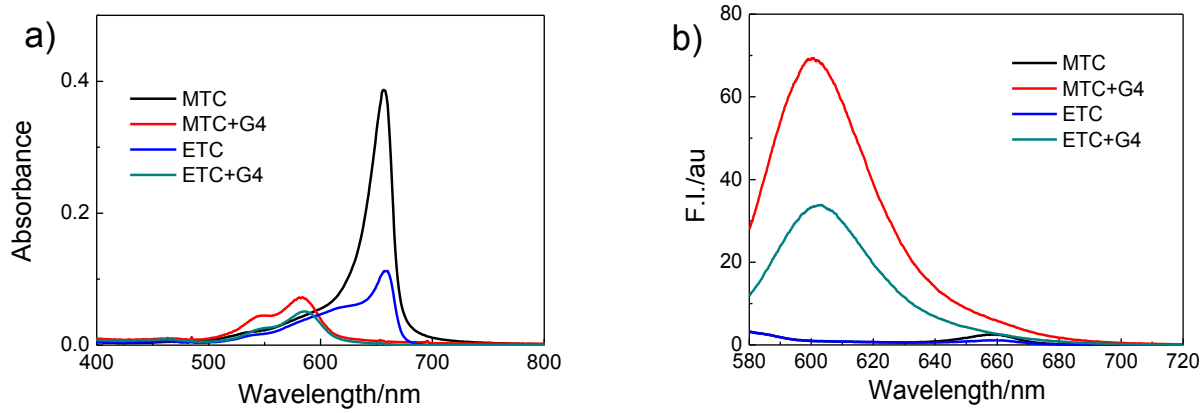


Fig. S2 The a) absorption and b) fluorescence spectra of 3 μM ETC and MTC without and with 2.4 μM G-quadruplexes (G4) present in Tris-HCl buffer solution with 50 mM K^+.

Both of MTC and ETC form J-aggregate in the buffer solution containing metal ions, which exhibit a major absorption band at about 660 nm. The fluorescence of J-aggregates is very weak. When interacting with G-quadruplexes, the MTC and ETC J-aggregates are converted to monomers with an absorption band at 585 nm. Meanwhile, a new fluorescence band at 598 nm belonging to MTC or ETC monomers also appears. Compared with ETC, MTC exhibits more obvious change in absorbance and fluorescence under the same conditions, meaning the MTC aggregate has more excellent spectral properties for recognizing G-quadruplexes.

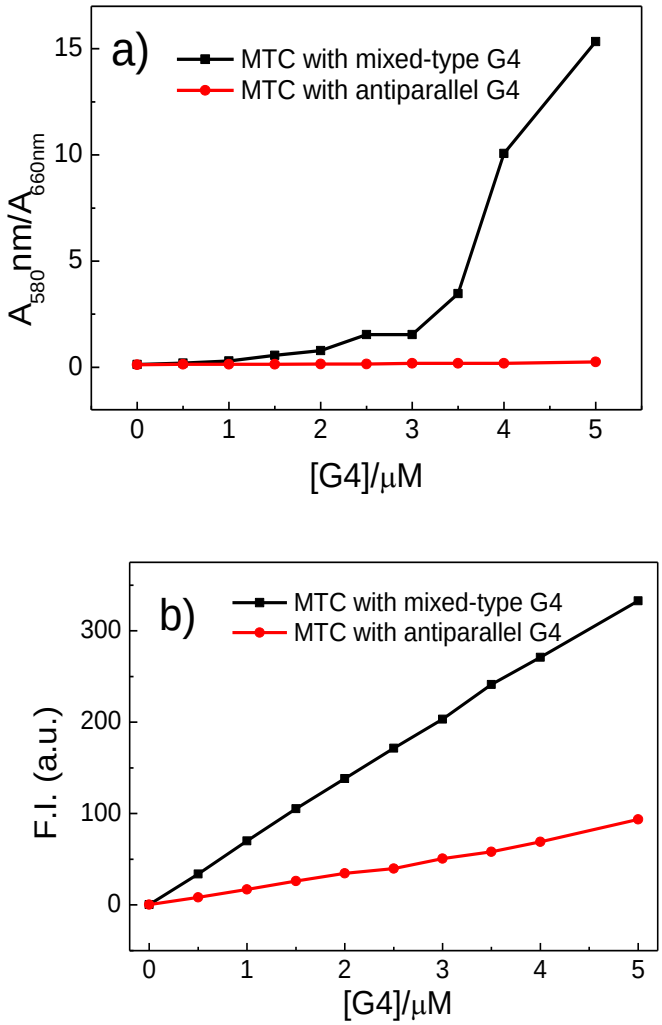


Fig. S3 a) The curves of the ratio A_{580nm}/A_{660nm} versus the concentration of telomeric G-quadruplexes (G4) with mixed-type (black line) and antiparallel (red line) motif, and b) The curves for the fluorescent intensity of 3.3 μM MTC at 600 nm versus the concentration of telomeric G4 with mixed-type (black line) and antiparallel (red line) motif.

The increased absorbance ratio at 580 nm and 660 nm as well as the enhancement of fluorescence intensity at 600 nm reflects the switch from MTC J-aggregates to monomers.

Compared with the mixed-type G-quadruplexes, the antiparallel G-quadruplexes aroused the switch of MTC more weakly, meaning the weaker interaction between the antiparallel G-quadruplexes and MTC.

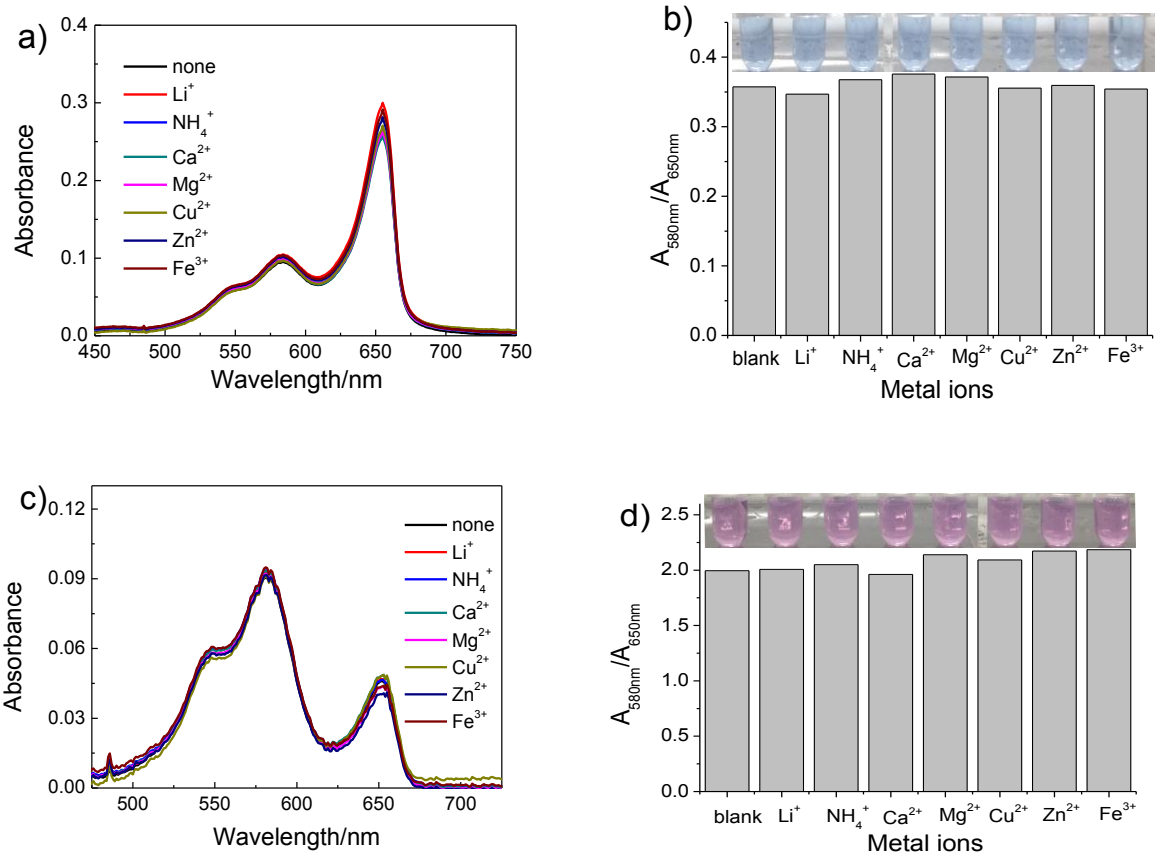


Fig. S4 The absorption spectra of 4 μM MTC with 8μM H22 G-quadruplexes without and with 100μM various metal ions in Tris-HCl buffer solution containing 140 mM NaCl together with a) 20 mM or c) 100 mM KCl. b) and d) The columns of A_{580nm}/A_{650nm} versus various metal ions. The inserted photograph is the image of the probe solution without and with various metal ions.

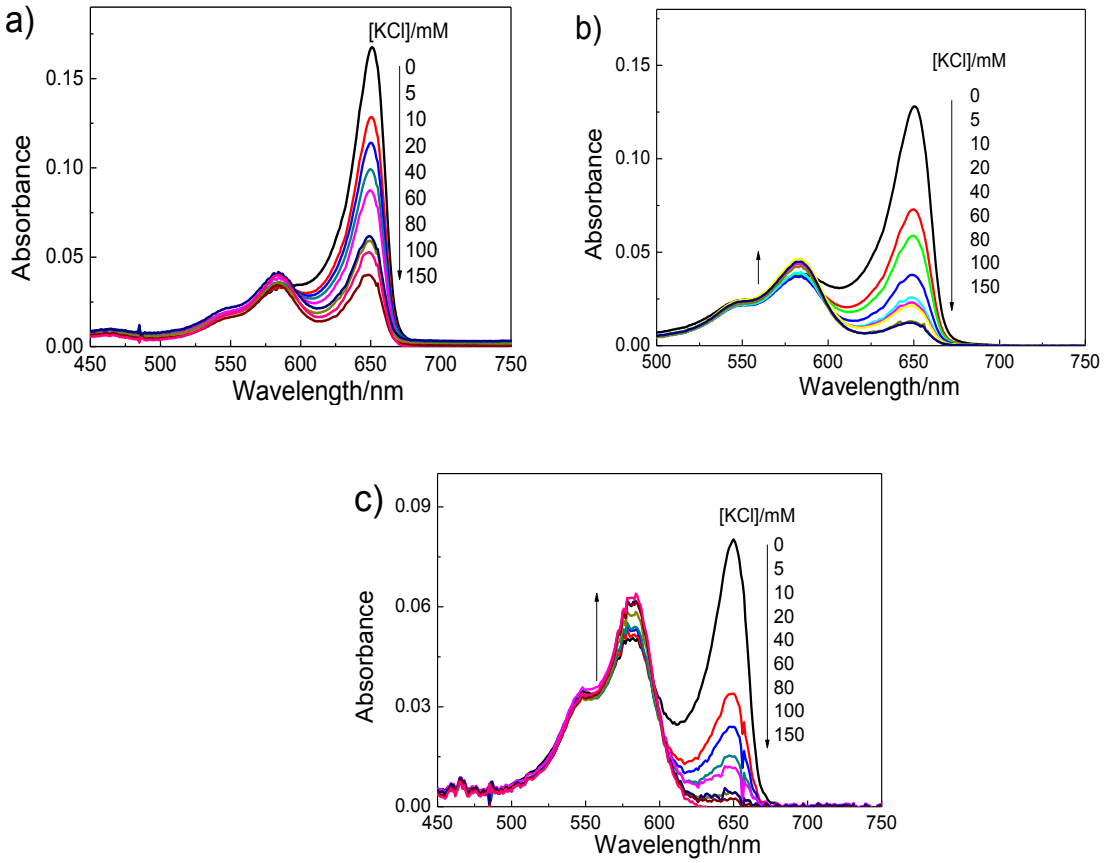


Fig. S5 The absorption spectra of 3 μM MTC with increasing amounts of KCl in the presence of a) 6 μM, b) 9 μM, and c) 12 μM H22 G-quadruplexes in Tris-HCl buffer solution containing 140 mM NaCl.

Because the colour of the probe solution depends on MTC absorption, one can imagine a colour discrepancy according to the absorption spectra. Comparing above data, we find that less K^+ can induce MTC switch when more G-quadruplexes are present. As shown in Fig. S5c, 5 mM KCl has aroused an obvious switch of MTC, implicating solution colour has changed. Additionally, a much lower K^+ level may also cause a colour change if more G-quadruplexes are present.