Supporting Information for

Turn-on and label-free fluorescent detection of lead ion based on

target-induced G-quadruplex formation

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Experimental Details

Materials and Reagents. Oligonucleotides were obtained from Sangon Biotechnology Co. Ltd. (Shanghai, China). The stock solution of DNA samples were prepared in ultrapure water, and stored at -20℃. Concentrations of DNA were accurately quantified using UV absorbance at 260 nm. NMM was purchased from J&K Scientific Ltd. (Beijing, China). The stock solution of NMM was prepared in dimethyl sulfoxide (DMSO) and stored in darkness at -20 °C. The concentration of NMM was accurately quantified using UV-VIS spectrophotometer (λ = 379 nm, extinction coefficient = 1.45×10^5 M^{-1} cm^{-1}). Thioflavin T (ThT) was purchased from Sigma-Aldrich, Co. (St. Louis, MO, USA). Berberine chloride was purchased from Aladdin Industrial Inc. (Shanghai, China). Other chemicals were of analytical grade and purchased from Sinopharm Chemical Reagent Co. Ltd. (Shanghai, China). Milli-Q water was used to prepare solutions. Measurements were performed in 10 mM Tris-HAc buffer (pH 7.4), unless stated otherwise.

The DNA sequences are as follows:
AGRO100: 5'- GGTGGTGGTGGTTGTGGTGGTGGTGG-3';
G3T4: 5'-GGGTTTTGGGTTTTGGGTTTTGGG-3';
G3T4TT4: 5'-GGGTTTTGGGTGGGTTTTGGG-3'.

Instrumentation. F-4600 Fluorescence spectrometer (Hitachi, Tokyo, Japan) was employed to record fluorescence spectra. The excitation wavelength was fixed at 399, 425, and 365 nm for NMM, ThT and berberine, respectively. The CD spectra of DNAs in the Tris-HAc buffer were collected by a Chirascan-plus Circular Dichroism Spectrometers (Applied Photophysics Ltd, Surrey, UK). Three scans from 220 to 320 at 0.1 nm intervals were accumulated and averaged.

Fluorescence measurements. For metal ions sensing, DNA sequence and metal ions were added to Tris-HAc (10 mM, pH 7.4) working buffer, and incubated for 5 min at room temperature. Then NMM was added to the mixture before measurement of the fluorescence spectra. The assay procedures for ThT and berberine were the same as those for NMM, except that ThT and berberine with corresponding DNA sequences were used instead.

Circular Dichroism (CD) Measurements. DNA sequences and metal ions were added to Tris-HAc (10 mM, pH 7.4) working buffer, incubated for 5 min at room temperature, and CD spectra were

measured.

Analysis of real samples

Water sample was collected from Dushu Lake of Suzhou and first filtered through a 0.22 μm membranes before using. After adding different concentrations of Pb^{2+}, these samples were diluted with Tris-HAc buffer, mixed with AGRO100 (0.4 μM) and incubated for 5 min at room temperature. Then NMM was added to the mixture before measurement of the fluorescence spectra. Water samples were diluted 20-fold finally.

After adding 5 μM Pb^{2+} into fetal bovine serum, these samples were diluted with Tris-HAc buffer, mixed with AGRO100 (0.4 μM) and incubated for 5 min at room temperature. Then NMM was added to the mixture before measurement of the fluorescence spectra. Serum samples were diluted 50-fold finally.

Table S1. Comparison of G-quadruplex-based label-free Pb^{2+} biosensors

Methods	Linear range	Limit of detection	Remarks[a]	Testing time	Ref.
PS2.M/NMM	5 nM - 1 μM	1 nM	Signal off ($F_0/F \approx 11.5$)	> 30 min	1
PS2.M/ Iridium(III) complex	0 - 2.5 nM	600 pM	Signal on ($F/F_0 \approx 1.8$)	> 1 h	2
TBAA/cationic water-soluble conjugated polymer (PMNT)	0 - 120 nM	6 nM	Signal on ($F/F_0 \approx 3$)	several minutes	3
PS2.M capped CdS QDs	20 nM - 1 μM	10 nM	Signal on ($F/F_0 \approx 1.7$)	> 1 h	4
T30695/ZnPPIX	20 nM - 1 μM	20 nM	Signal on (using partly complementary strand to reduce background, $F/F_0 \approx 6$)	> 5 h	5
AGRO100 DNAzyme/AUR	0 - 1 μM	0.4 nM	Signal on ($F/F_0 \approx 6$)	> 3 h	6
PS2.M DNAzyme/ ABTS or luminol	10^{-7} M to 10^{-5} M or 10^{-9} M - $10^{-6.5}$ M	32 nM or 1 nM	Signal off (Pb^{2+}-induced decrease in peroxidase activity, $A_0/A \approx 1.3$, $F_0/F \approx 1.4$)	> 2 h	7
AGRO100/NMM	0 - 1 μM	3 nM	Signal on ($F/F_0 \approx 16$)	several minutes	This work

[a] F0 (or A0), F (or A) denote the fluorescence (or absorbance) of the sensor in the absence of Pb^{2+}, and presence of the linear upper bound Pb^{2+} concentration, respectively.
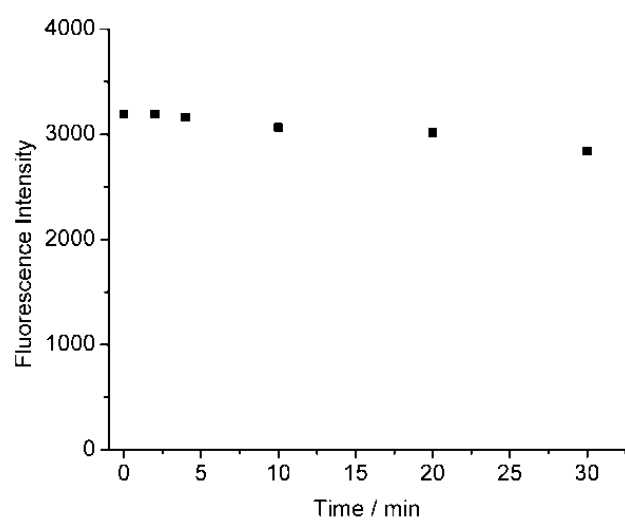
Figure S1. Fluorescence intensity at 610 nm of this AGRO100-NMM system against reaction time after all composition was mixed. Experimental conditions: 1 μM AGRO100, 1 μM NMM, and 1 μM Pb^{2+} in 10 mM Tris–HAc (pH 7.4) buffer solutions.
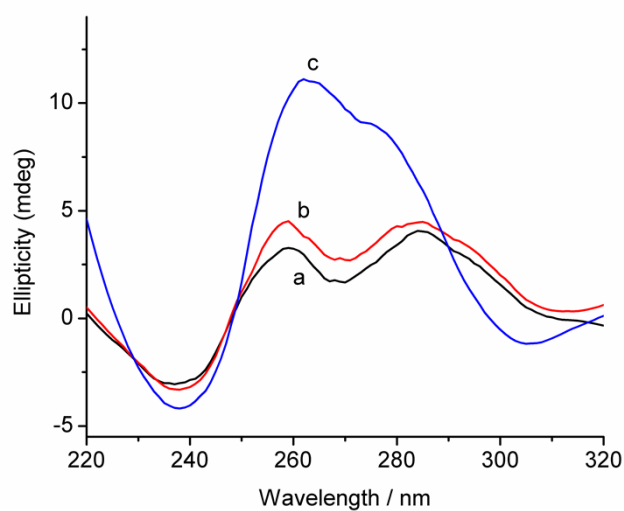

Figure S2. Circular dichroism (CD) spectra for characterizing the AGRO100 structural conversion in the absence (a), and presence of 100 μM K^+ (b) and 1 μM Pb^{2+} (c). Experimental conditions: 4 μM AGRO100 in 10 mM Tris–HAc (pH 7.4) buffer solutions.
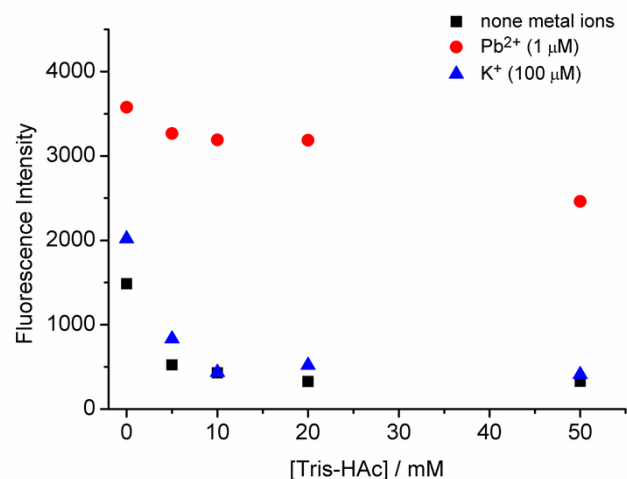
Figure S3. Effect of the concentration of Tris-HAc buffer on AGRO100-NMM probe in the absence, and presence of Pb^{2+} (1.0 μM) or K^+ ions (100 μM). The concentration of AGRO100 and NMM are 1.0, 1.0 μM, respectively.
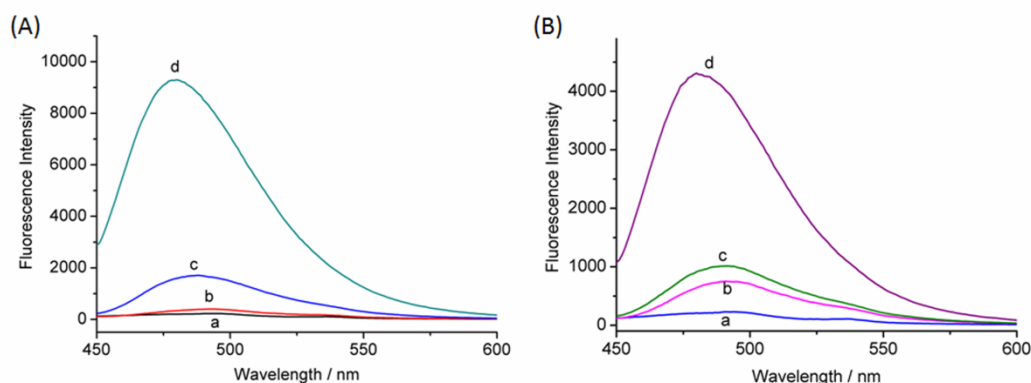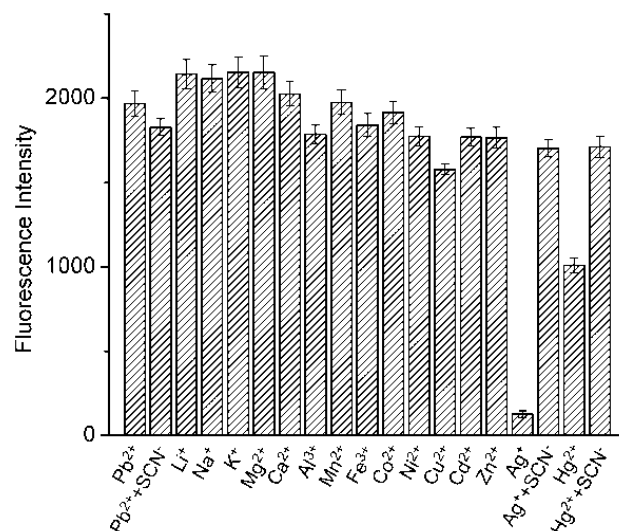

Figure S4. (A) Fluorescence spectra for turn-on detection of Pb^{2+} using ThT and G3T4: (a) ThT (0.4 μM); (b) ThT (0.4 μM) and G3T4 (0.4 μM); (c) ThT (0.4 μM), G3T4 (0.4 μM), and K^+ (100 μM); (d) ThT (0.4 μM), G3T4 (0.4 μM), and Pb^{2+} (1 μM). (B) Fluorescence spectra for turn-on detection of Pb^{2+} using ThT and G3T4TT4: (a) ThT (0.4 μM); (b) ThT (0.4 μM) and G3T4TT4 (0.4 μM); (c) ThT (0.4 μM), G3T4TT4 (0.4 μM), and K^+ (100 μM); (d) ThT (0.4 μM), G3T4TT4 (0.4 μM), and Pb^{2+} (1 μM).
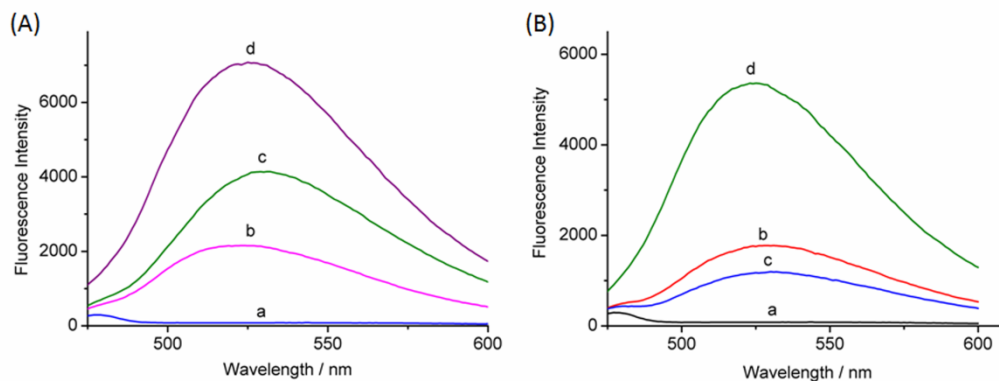

Figure S5. (A) Fluorescence spectra for turn-on detection of Pb^{2+} using Berberine and G3T4: (a)

Berberine (5 μM); (b) Berberine (5 μM) and G3T4 (1 μM); (c) Berberine (5 μM), G3T4 (1 μM), and K$^+$ (100 μM); (d) Berberine (5 μM), G3T4 (1 μM), and Pb^{2+} (5 μM). (B) Fluorescence spectra for turn-on detection of Pb^{2+} using Berberine and G3T4TT4: (a) Berberine (5 μM); (b) Berberine (5 μM) and G3T4TT4 (1 μM); (c) Berberine (5 μM), G3T4TT4 (1 μM), and K$^+$ (100 μM); (d) Berberine (5 μM), G3T4TT4 (1 μM), and Pb^{2+} (5 μM).

Figure S6. Competitive experiments of the AGRO100-NMM probe. The bars denote the fluorescence intensity when Pb^{2+} (1 μM) coexisted with other individual metal ions (100 μM Li$^+$, 100 μM Na$^+$, 100 μM K$^+$, 100 μM Mg^{2+}, 100 μM Ca^{2+}, 10 μM Al^{3+}, 10 μM Mn^{2+}, 10 μM Fe^{3+}, 3 μM Co^{2+}, 1 μM Ni^{2+}, 10 μM Cu^{2+}, 3 μM Cd^{2+}, 3 μM Zn^{2+}, 10 μM Ag$^+$, and 10 μM Hg^{2+}). 10 mM NaSCN was added to mask Ag$^+$ and Hg^{2+} ions. Error bars represent standard deviations from three repeated measurements.
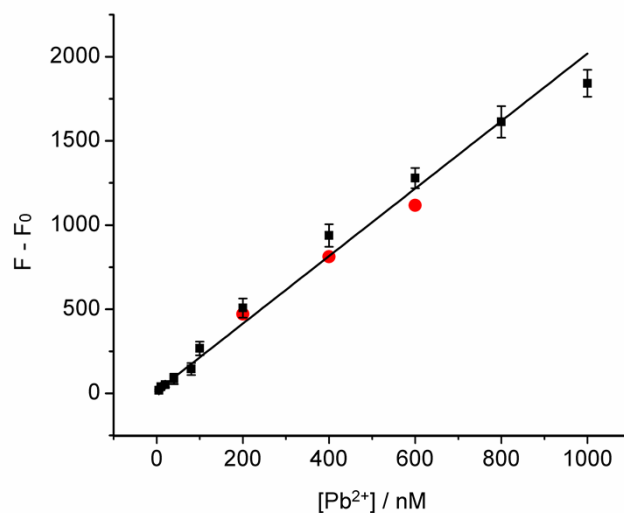
Figure S7. Detection of Pb^{2+} in lake water samples. ■: data obtained from sensitiviy experiments; ●: data of analyzing real water samples. 200, 400, and 600 nM Pb^{2+} were spiked into water samples, and the recovery values obtained were 115.4%, 100.2%, and 92.3%, respectively.

References

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