Supporting Information

Experiment section

Chemicals

The DNA oligonucleotides were obtained from Shanghai Sangon Biological Engineering Technology & Services Co., Ltd. of China. The sequences of oligonucleotides are as follows:

MB: FAM-5' CAGTCT **GGGTCGTGATGTG** AGACTG 3'- DABCYL

Mismatched ssDNA-1: 5'- CACATCTCGACCC-3'

Mismatched ssDNA-2: 5'- CACATCTCGTCCC-3'

Mismatched ssDNA-3: 5'- CACTTCTCGTCCC-3'

Mismatched ssDNA-4: 5'- CTCTTCTCGTCCC-3'

The stem-loop oligonucleotide is modified with a carboxyfluorescein (FAM) dye at the 5' end and a quencher 4-(4-dimethylaminophenylazo) benzoic acid (DABCYL) at the 3' end, respectively. It can form the stem-loop structure at an appropriate ionic strength. The part (in bold) is the loop of MB. Mismatched ssDNA-1, -2, -3 and -4 have different numbers T-T mismatched base pairs (numbers are 1, 2, 3 and 4, respectively, in underline), which can't match with the loop of MB.

All solutions were prepared by using metal free reagents and water was purified with Milli Q purification system (Millipore). DNA buffer solutions (the concentration of oligonudeotides is 10 μM) were prepared as the stock solutions by dissolving oligonudeotides into 40 mM Tris-acetate buffer solutions containing 0.1M NaCl and 0.05 M $MgCl_2$ (pH 7.4) (except for the ionic strength optimized experiment).

All chemicals used were analytical grade or better. The $Hg(NO_3)_2$ stock solution (1.0×10^{-3} M) was prepared with 2 or 3 drops of concentrated nitric acid. Other metal ion solutions were prepared from nitrate salts.

Apparatus

The fluorescence measurements were performed at room temperature on a Varian Cary Eclipse Fluorescence Spectrophotometer except specific indication. The emission spectra were collected from 500 to 600 nm with the excitation wavelength of 480 nm. Both the excitation and emission slit widths were set to 10.0 nm.

Assay of Hg^{2+}

Above stock solutions of MB and mismatched ssDNA were diluted to 100 nM by Tris-acetate (pH 7.4), respectively. The equal volume (150 μl for each) solutions of MB and mismatched ssDNA were mixed each other. Before fluorescence measurements, aliquots of Hg^{2+} aqueous were added into above mixture at 37□ for 50 min (except for the temperature and time-course study). The final volume is 300 μl. Then the fluorescence was measured for excitation at 480 nm and the emission spectra were collected from 500 to 600 nm. Maximum fluorescence intensity value was used for quantification. (Note: Due to the high toxicity of Hg^{2+}, after assays the solutions should be discarded following the waste disposal procedure.)

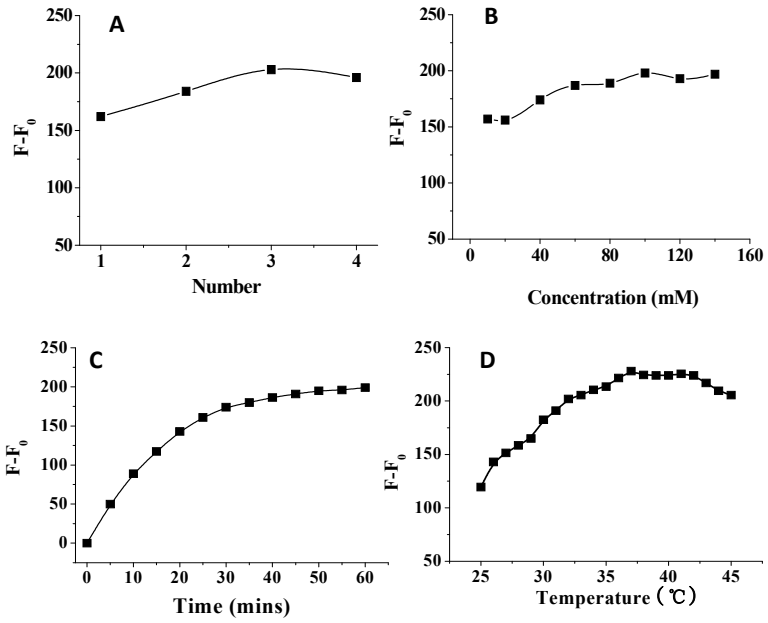

Figure S1 Optimization of the measuring system. Influence of: (A) the number of unmatched T residues; (B) the concentration of NaCl; (C) incubation time; (D) incubation temperature. F and F_0 are the fluorescence intensity with and without 2×10^{-7} M Hg^{2+} ions, respectively. Other conditions were the same as those described in Figure 2.

Table 1 Determination of Hg^{2+} ions in water samples using the proposed method

	Added(nM)	Founded(nM) (mean[a] ± SD[b])	Recovery (%)
Sample 1	10	10.9±0.5	109
Sample 2	25	24.1±0.8	96.4
Sample 3	50	51.2±8	102.2
Sample 4	75	78.4±5	104.5
Sample 5	100	93.4±12	93.4
Sample 6	150	148±10	98.6

[a] Mean of three determinations.

[b] SD, standard deviation.