Supplementary Information

Colorimetric Hg^{2+} Detection with a Label-Free and Fully DNA-Structured Sensor Assembly Incorporating G-Quadruplex Halves

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Fig. S1 Logarithmic plot of the calibration curve as shown in Fig.1 B indicates a linear domain for Hg^{2+} concentrations less than 200 nM, based on which a detection limit of 4.5 nM was obtained by linear extrapolation according to a commonly used 3σ/slope rule.

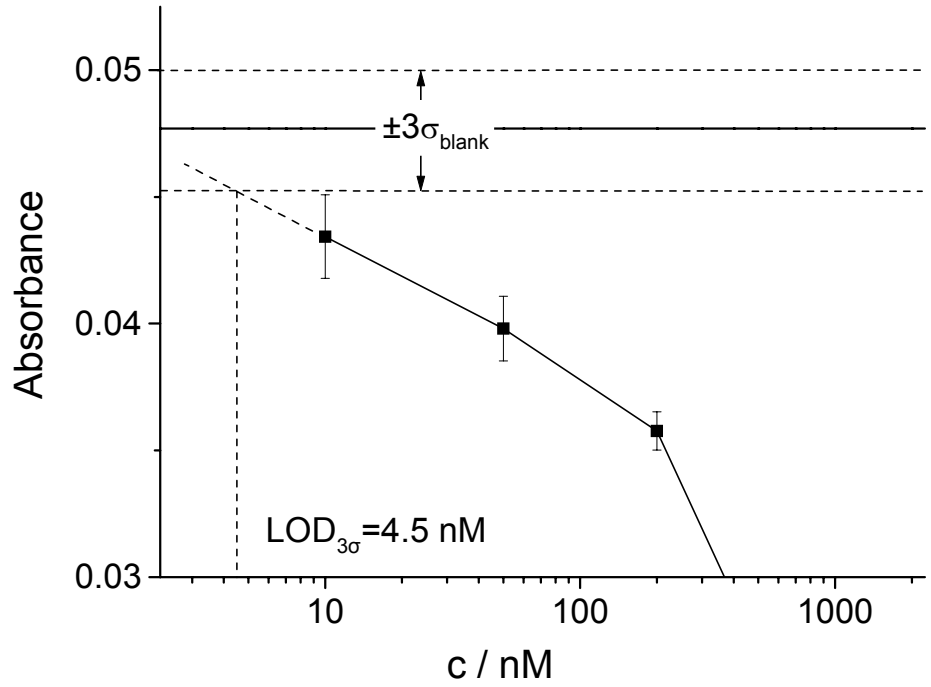
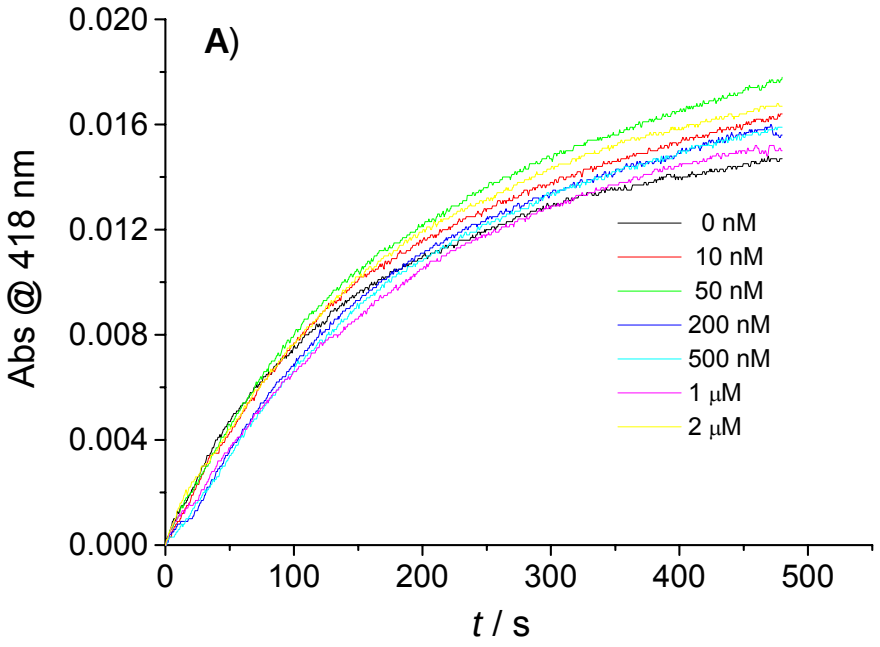


Fig. S2 A control experiment to test any possible suppression of the peroxidase activity of hemin by Hg^{2+}. The concentration of hemin was 0.25 μM (this concentration of hemin was 2.5 times of that used in a real assay in order to get stronger absorbance signals for measurements). A) Time-dependent absorbance curves at 418 nm with reaction durations of 8 min. The concentrations of Hg^{2+} were: a) 0 nM, b) 10 nM, c) 50 nM, d) 200 nM, e) 500 nM, f) 1 μM, and g) 2 μM. B) Scatter plot of the absorbances at 418 nm after 8 min reactions (refer to the absorbance-time curves on panel A) in the presence of Hg^{2+} from 0 to 2000 nM. A red horizontal line in B) corresponds to the average level of all the absorbances after 8 min reactions at different Hg^{2+} concentrations.



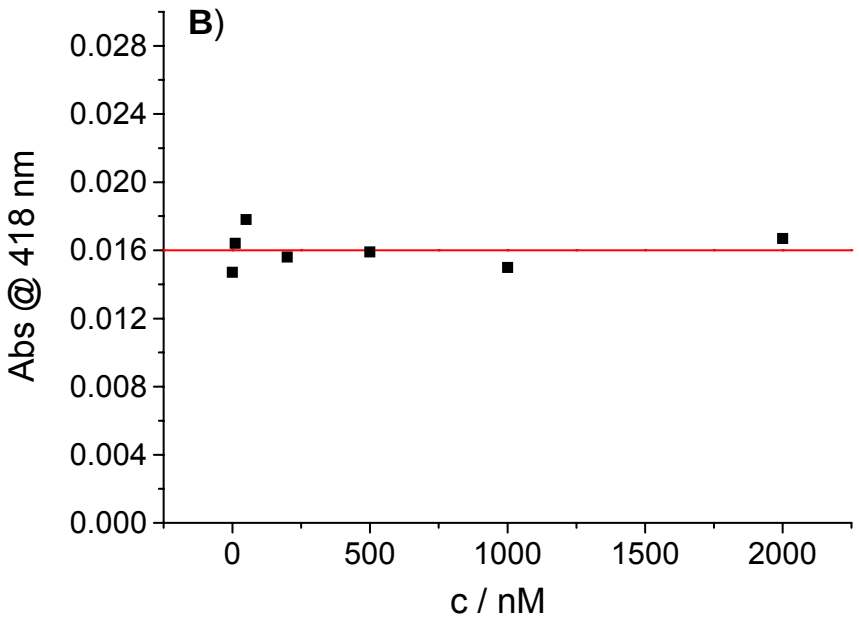


Fig. S3 A control experiment to test possible suppression of the peroxidase activity of a G-quadruplex-hemin complex by Hg^{2+}. All concentrations were identical to Fig. 1. The results showed negligible changes of absorbance after the introduction of Hg^{2+} at various concentrations up to 2 μM. A horizontal line in red as shown in panel B) corresponds to the average value of all absorbances on the scattered data points. Sequence of the G-quadruplex used here was: 5' GGGTAGGGCGGGTTGGG 3'

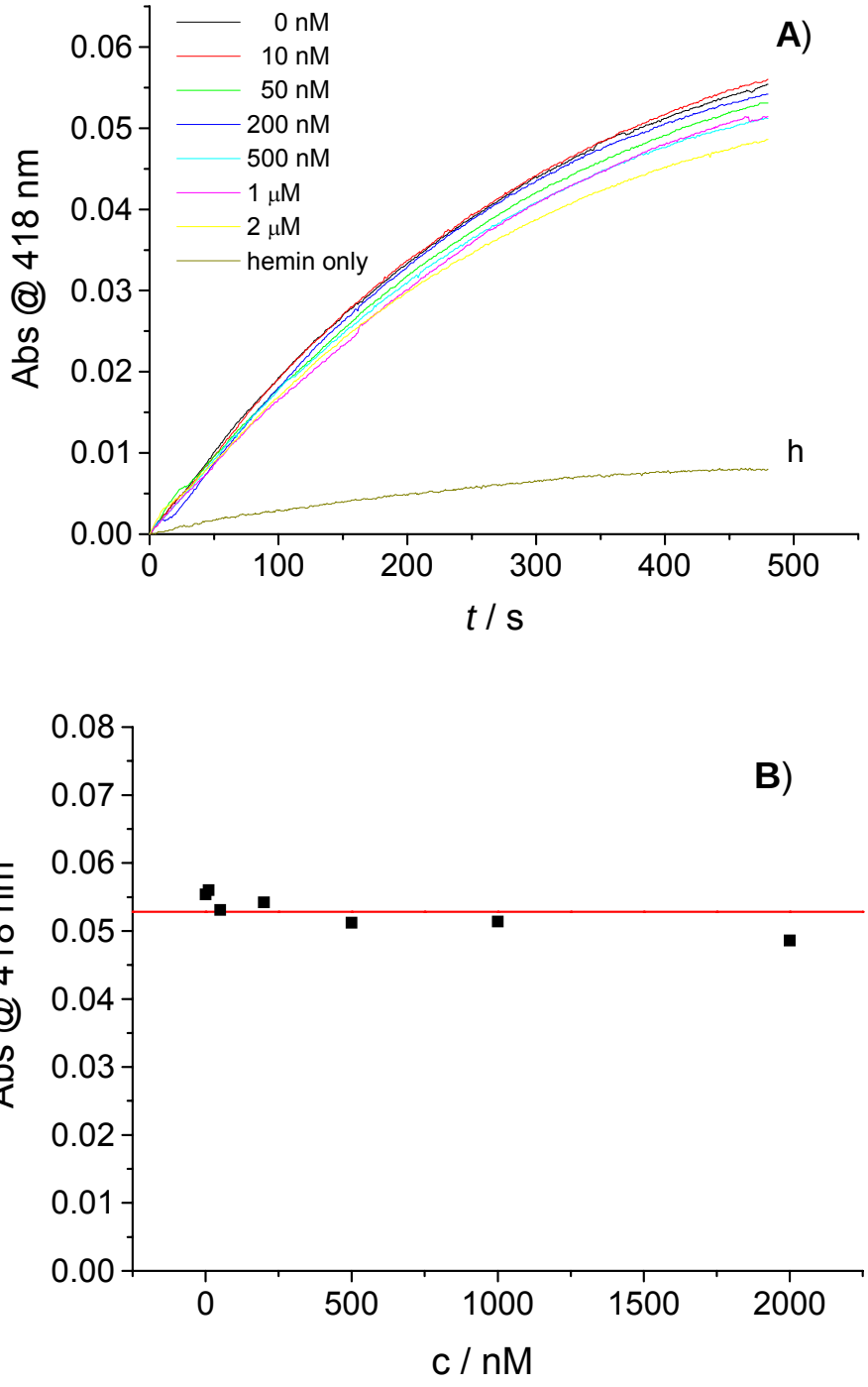


Fig. S4 A control experiment to test possible decomposition of H_2O_2 in the presence of Fe^{3+}. In this experiment, 10 μM Fe^{3+} was pre-mixed with H_2O_2. Followed by an incubation at room temperature for different times (as shown on the figure), the mixture was added to a DNAzyme-hemin assay solution to trigger the oxidation of ABTS. *The results showed negligible changes of absorbance for different interaction times between Fe^{3+} and H_2O_2. The G-quadruplex enzyme used here was: 5'GGGTAGGGCGGGTTGGG 3'.*

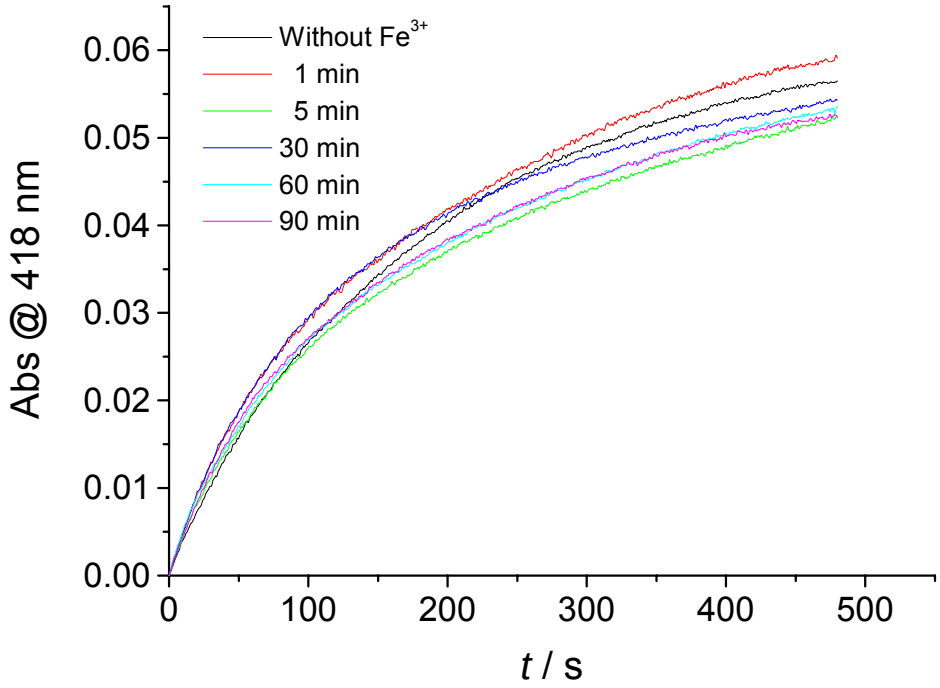


Fig. S5 A control experiment to test possible interactions between Fe^{3+} and the DNAzyme that led to a suppressed catalytic activity of the enzyme. In this experiment, 10 μM Fe^{3+} was incubated with the DNA enzyme (same as Figures S3 and S4) for different times (as shown on the figure) before hemin and other chemicals were added to initiate the oxidation process. The results showed that a longer interaction with Fe^{3+} resulted in a stronger suppression of the enzyme activity. For convenience, hemin was allowed to interact with the enzyme for 2 min before the absorbances were recorded, and this (compared to 1 hour incubations in real tests) accounted for the as-recorded greater absorbances. *These data implied that Fe^{3+} did have an interaction with the DNA structured enzyme and resulted in suppressed kinetics of the following oxidation process.*

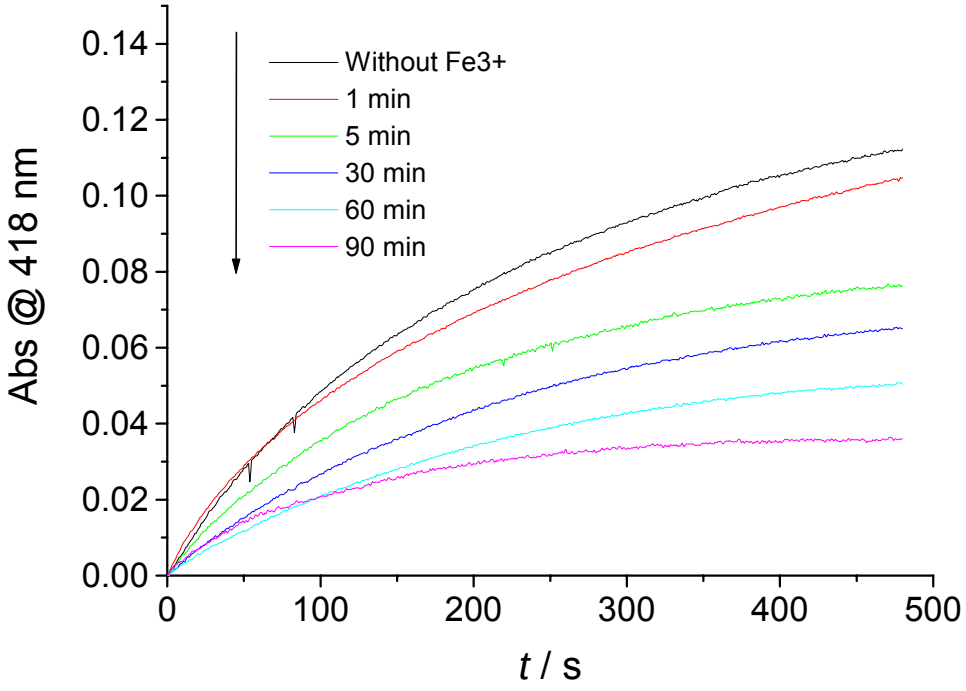
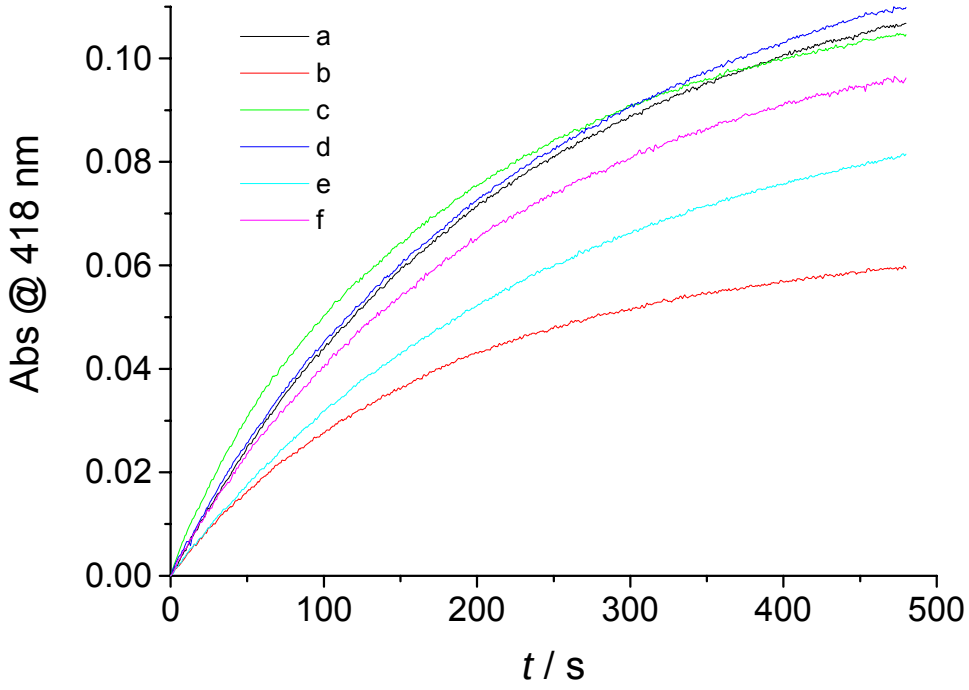


Fig. S6 A control experiment to show that the influence of Fe^{3+} on the catalytic activity of the enzyme could be eliminated by addition of citrate to complex Fe^{3+}. The data also showed that if Fe^{3+} was allowed to incubate with DNA first, it would require longer time (could be facilitated by an incubation at elevated temperature) to completely restore the enzyme activity. For convenience, hemin was allowed to interact with the enzyme for a 2 min duration before the absorbances were recorded, and this (compared to 1 hour incubation in real tests) accounted for the as-recorded greater absorbances. *The results implied that the adverse effect of Fe^{3+} on the enzyme activity was not due to a permanent degradation of DNA, but, more probably, was a result from a strong interaction between Fe^{3+} and DNA (for example, complexation), which inhibited the correct formation of the quadruplex-hemin structure.*



(a) Absorbance-time curve in the absence of Fe^{3+};

(b) 10 μM Fe^{3+} was allowed to incubate with the DNA for 30 min;

(c) 1 mM citrate was added to the system before Fe^{3+} was introduced and a waiting time of 30 min was allowed before hemin and other chemicals were added;

(d) 10 μM Fe^{3+} and 1 mM citrate was pre-mixed and then added to the assay solution followed by a 30 min incubation at room temperature before hemin and other chemicals were added;

(e) 1 mM citrate was added after Fe^{3+} was allowed to interact with the DNAzyme for 30 min, hemin and other chemical were introduced 30 min later to allow a complexation of Fe^{3+} by citrate;

(f) Same as (e) except that the solution was incubated at 50 °C for 20 min after citrate was added.

Table S1. Summary of some literature reported Hg^{2+} sensors.

Ref.	Method	Detection technique	LOD
1	FRET-MSO	Fluorometric	40 nM
2	CP-MSO	Colorimetric	2.5 μM
		Fluorometric	42 nM
3	MSO-Functionalized AuNPs	Colorimetric	100 nM
4	DNAzymes	Fluorometric	2.4 nM
5	AuNPs and MSO	Fluorometric	10 nM
5	DNA-based machine amplification	Colorimetric	1 nM
6	MSO-functionalized AuNPs	Colorimetric	1~3 μM
7	AuNPs and MSO	Fluorometric	40 nM
8	DNA-functionalized AuNPs	Fluorescence polarization	1 nM
9	MSO and DNA intercalating dye	Fluorometric	1.33 nM
10	AuNPs and silver-amplification	Scanometric	10 nM
11	MSO and FRET	Fluorometric	3.2 nM
12	AuNPs-OliGreen	Fluorometric	25 nM
13	DNAzyme	Colorimetric	50 nM
This work	DNAzyme and MSO	Colorimetric	4.5 nM

[a] CP: conjugated polymers; [b] MSO: Mercury-specific oligonucleotide; [c] AuNPs: gold nanoparticles.

References to Table S1

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