Supporting information for

Rational design of signal-on biosensors by using Photoinduced Electron Transfer between Ag Nanoclusters and split G-quadruplex halves/Hemin Complex

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Experimental section

Materials. All oligonucleotides were purchased from Genscript Biotechnology Co., Ltd (Nanjing, China) and their sequences were listed in Table S1. The stock solutions of DNA were prepared in 20 mM phosphate buffer. Silver nitrate (AgNO$_3$), sodium borohydride (NaBH$_4$), adenosine and hemin were purchased from Sigma-Aldrich (St. Louis, MO, USA). The stock solution of hemin (5 µM) was prepared in dimethyl sulfoxide (DMSO), stored in darkness at -20 °C. Other chemicals were of reagent grade and were used without further purification. Solutions were prepared with ultrapure water, which was purified with Milli-Q purification system (Branstead) to a specific resistivity of > 18.2 MΩ cm).

Fluorescent Adenosine Assays: In adenosine assay, DNA1 and DNA3 (1.0 µM each) in 20 mM phosphate buffer (5.0 mM Mg(Ac)$_2$, pH 7.4) was heated at 88 °C for 10 min, and gradually cooled to room temperature. A 6 µM solution of AgNO$_3$ was added to the DNA solutions, After 1.0 h at room temperature, 6 µM NaBH$_4$ was added into the mixture. After 14.0 h reaction in the dark, appropriate concentrations of adenosine, 1.0 µM hemin and 50 mM KNO$_3$ were added, and the mixture was incubated for another 2.0 h, and then the fluorescence intensity was measured. In the experiment of selectivity, Concentration of adenosine is 80 µM and for the other small molecules are 10 mM (the guanosine concentration is 0.5 mM).
Fluorescent RNA Assays: In a typical RNA assay, DNA2 and DNA3 (1.0 µM each) in 20 mM phosphate buffer (5.0 mM Mg(Ac)$_2$, pH 7.4) was heated at 88 °C for 10 min, and gradually cooled to room temperature. A 6.0 µM solution of AgNO$_3$ was added to the DNA solutions. After 1.0 h at room temperature, 6.0 µM NaBH$_4$ was added into the mixture. After 14.0 h reaction in the dark, appropriate concentrations of target RNA, 1.0 µM hemin and 50 mM KNO$_3$ were added, and the mixture was incubated for another 2.0 h, and then the fluorescence intensity was measured. In the experiment of selectivity, the concentrations of target RNA and others are 1.4 µM.

Instrumentation: The fluorescence spectra were recorded by a SpectraMax M5 Multi-Mode Microplate Readers (Molecular Devices, USA), using a Costar 96 well microtiter plate (No. 3599, Corning, New York, USA). The DNA-AgNCs displayed fluorescence emission at 655 nm upon excitation at 585 nm.
DNA1: 5’-GGG TTG GGT CTT TCA GTC CGT CAA CCT GGG GGA GTA TIG CGG AGG AAG GTG GGT AGG G-3’
DNA2: 5’-GGG TTG GGT CTT TCA GTC CGT CAA ACT ATA CAA CCT ACT ACC TCA GGG TAG GG-3’
DNA3: 5’-TGA CGG ACT GAC CTC CTT CCT CCC TAC GTG CTA AGA-3’
DNA3-zero: 5’-TGA CGG ACT GAA AGA CTT CCT CCC TAC GTG CTA AGT GCT-3’
DNA3-two: 5’-TGA CGG ACT GAA ACC TCC TTC CTC CCT ACG TGC TGA-3’
DNA3-six: 5’-TGA CGG ACT CCT CTT TCC TCC CTA CGT GCT GAA AGA-3’
DNA3-eight: 5’-TGA CGG ACC TCC TTC CTC CCT ACG TGC TCT GAA AGA-3’
Let.-7a: 5’-UGA GGU AGU AGG UUG UAU AGU U-3’
Let.-7b: 5’-UGA GGU AGU AGG UUG UGU GGU U-3’
Let.-7c: 5’-UGA GGU AGU AGG UUG UAU GGU U-3’

Fig. S1 Oligonucleotides sequence used in this strategy. The colors of the sequences are the same as given in Scheme 1.
Fig. S2 Quenching efficiency of the position of the DNA-templated loop from zero to eight bases away from the 3’-end of DNA3
Fig. S3 Fluorescence emission spectra of Ag nanoclusters generated under different pH conditions.
Fig. S4 (A) Fluorescence peak intensity quantification for DNA1/DNA3/AgNO₃ for different time after adding NaBH₄. (B) Fluorescence peak intensity quantification for DNA-AgNCs for different time after adding K⁺ and hemin.