Supporting Information (SI)

A sensitive strategy for label-free and time-resolved fluorescence assay of thrombin using Tb-complex and unmodified gold nanoparticles

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1. The schematic diagram of absorbance spectra of GNPs and fluorescence emission spectra excited at 311 nm of Tb-complexes (Figure S-1).

2. The investigation results of some potential influencing factors toward the time-resolved fluorescence intensity of Tb-complexes in the present assay, including GNPs, ssDNA (aptamer or random nucleic acid sequence), thrombin, and binding buffer (Figure S-2 and Figure S-3).

3. The study results of the impact of ssDNA, thrombin, and binding buffer toward GNPs (Figure S-4).

4. The derived calibrations of the thrombin concentration with absorbance (Figure S-5).

Figures

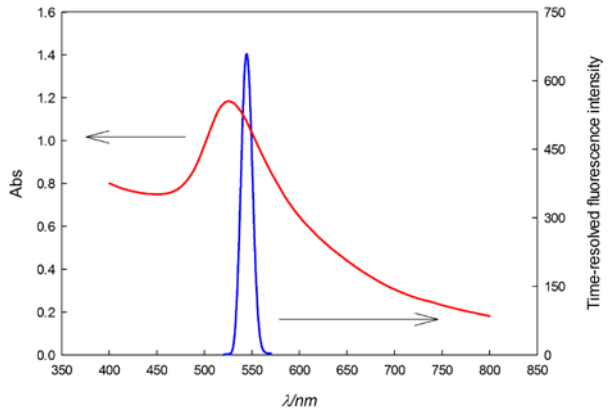


Figure S-1. The schematic diagram of the absorbance spectra of GNPs (red) and time-resolved fluorescence emission spectra excited at 311 nm of Tb-complexes (blue).

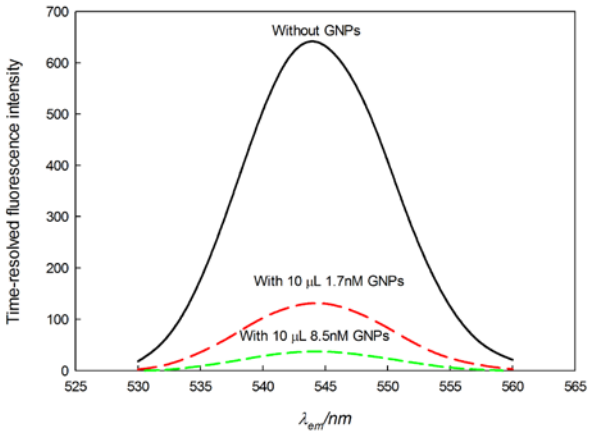


Figure S-2. Time-resolved fluorescence emission spectra of Tb-complexes with different concentrations of GNPs. It demonstrates that the GNPs can quench the emission spectra of Tb-complexes.

Supplementary Material for Analyst

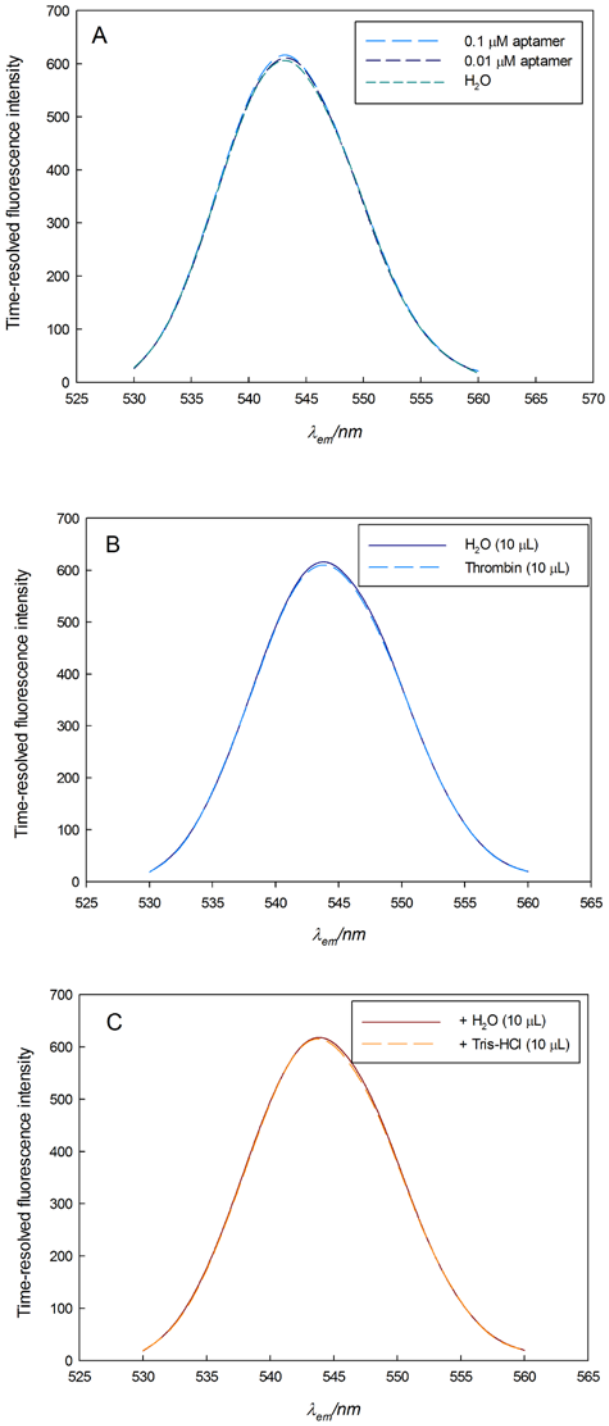
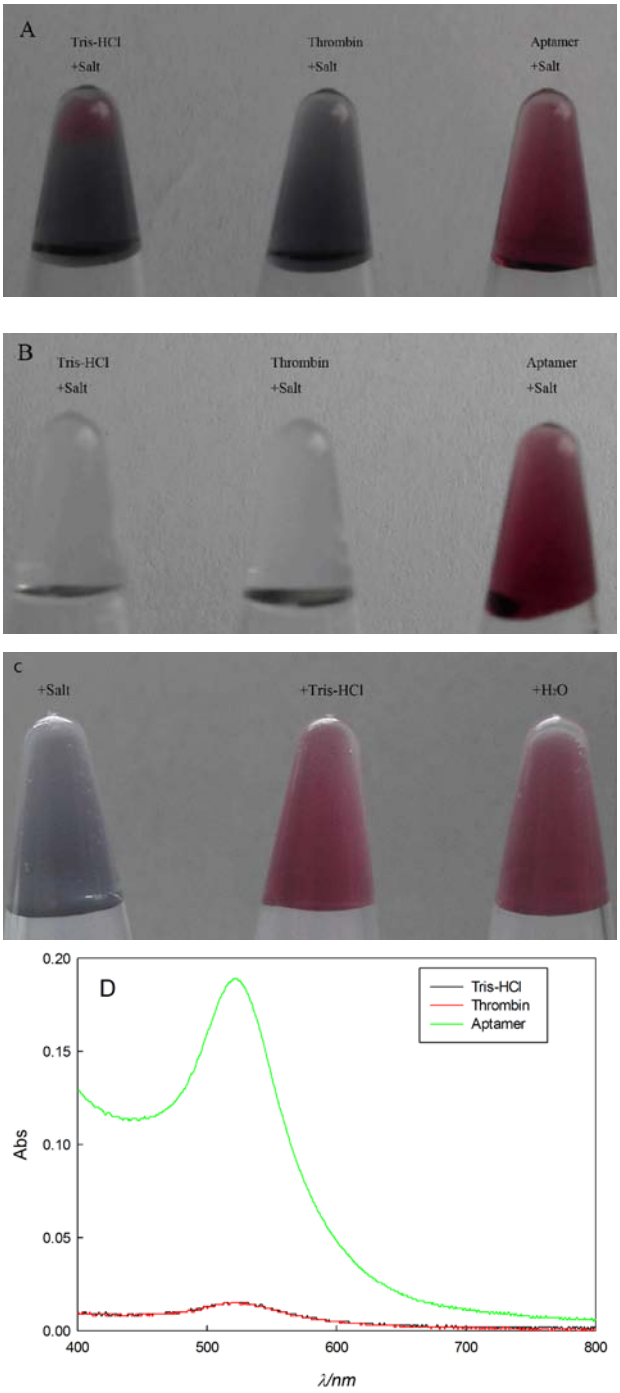


Figure S-3. Time-resolved fluorescence emission spectra for aptamer/Tb-complexes (A), thrombin/Tb-complexes (B), and Tris-HCl/Tb-complexes (C). $C_{aptamer}$=0, 0.01, and 0.1 μM, $C_{thrombin}$=20 nM, the Tris-HCl is the binding buffer (20 mM Tris-HCl, pH=7.4, containing 100 mM NaCl, 1 mM $CaCl_2$, 1 mM $MgCl_2$, and 5 mM KCl). These results revealed that the ssDNA, thrombin and binding buffer have no effect toward the fluorescence intensity of Tb-complexes.

Supplementary Material for Analyst



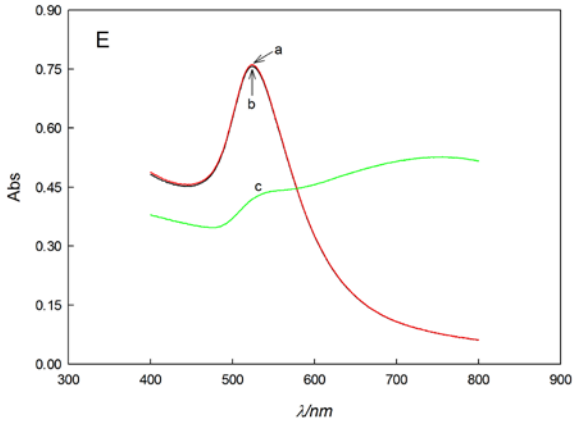


Figure S-4. The color changes of GNPs in Tris-HCl, Thrombin, Aptamer solutions with high salt before centrifugation (A) and after centrifugation (B), and compared the colors of GNPs in Tris-HCl solution to salt solution and H_2O (C). The absorbance spectra of Tris-HCl/GNPs/Salt, Thrombin/GNPs/Salt, and Aptamer/GNPs/Salt mixture solutions after centrifugation (D), and Tris-HCl/GNPs, H_2O/GNPs, and Salt/GNPs without centrifugation (E), a: Tris-HCl/GNPs, b: H_2O/GNPs, c: Salt/GNPs. $C_{aptamer}$= 0.25 μM, $C_{thrombin}$= 20 nM, the Tris-HCl is the binding buffer. These results shown that the ssDNA can prevent the aggregation of GNPs from high concentration salt, while the thrombin cannot. And, the binding buffer cannot lead to the aggregation of GNPs

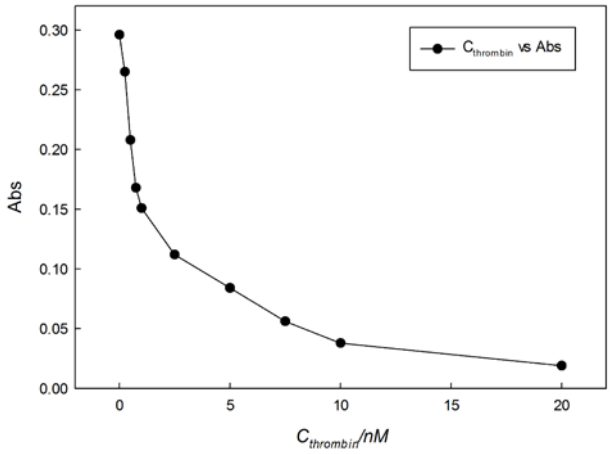


Figure S-5. The calibration curve of different concentrations of thrombin toward absorbance spectra.