(Supporting Information)

Specific and sensitive detection of *Plasmodium falciparum* lactate dehydrogenase by DNA-scaffolded silver nanoclusters combined with an aptamer

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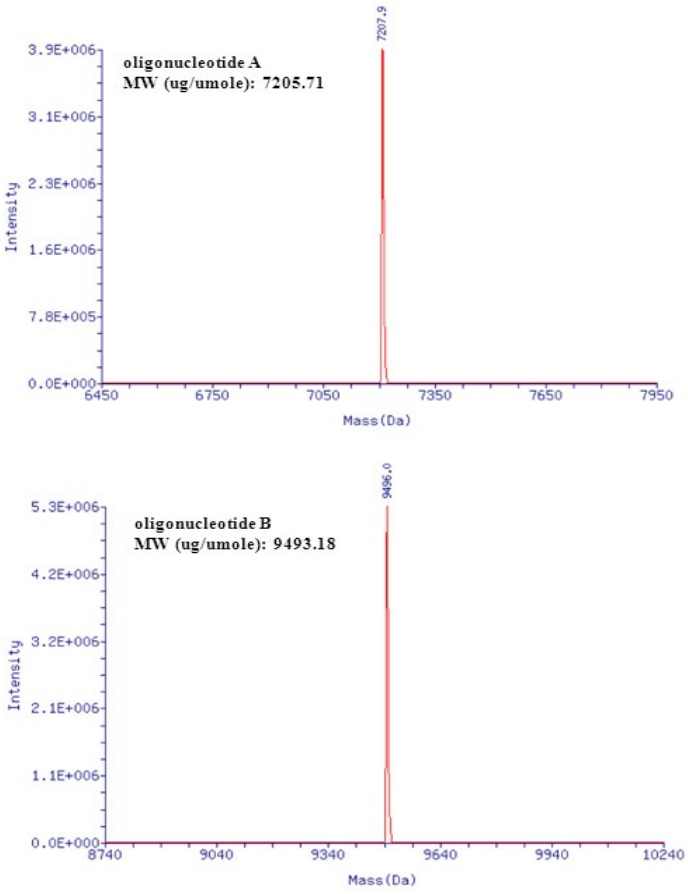

Fig. S1 HPLC-MS of the single strand oligonucleotide A and B, which are shown for instances the characterizations of single strand oligonucleotides.

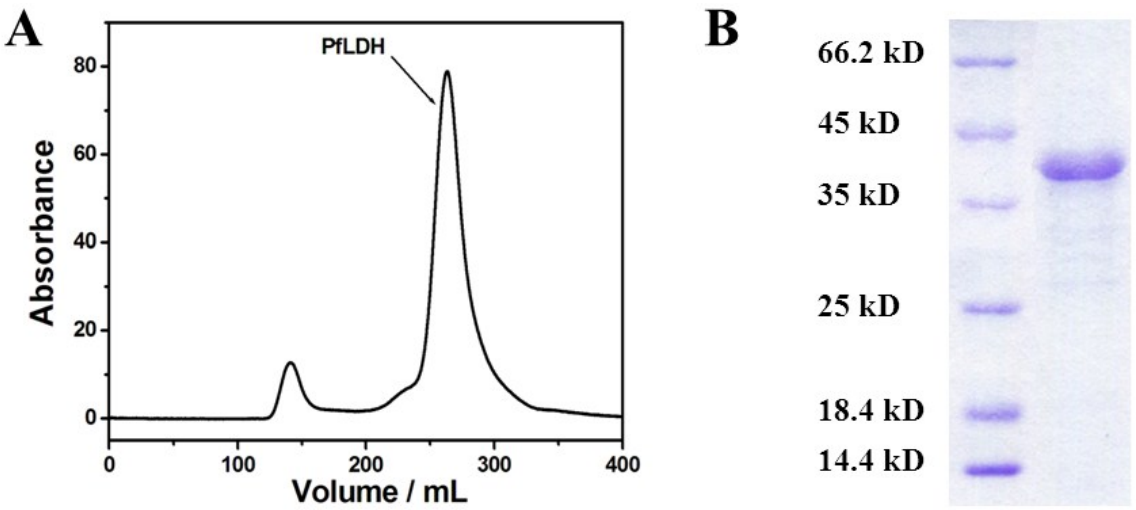

Fig. S2 A) The fast protein liquid chromatography (FPLC) plot and B) SDS-PAGE gel electrophoresis of the obtained PfLDH.

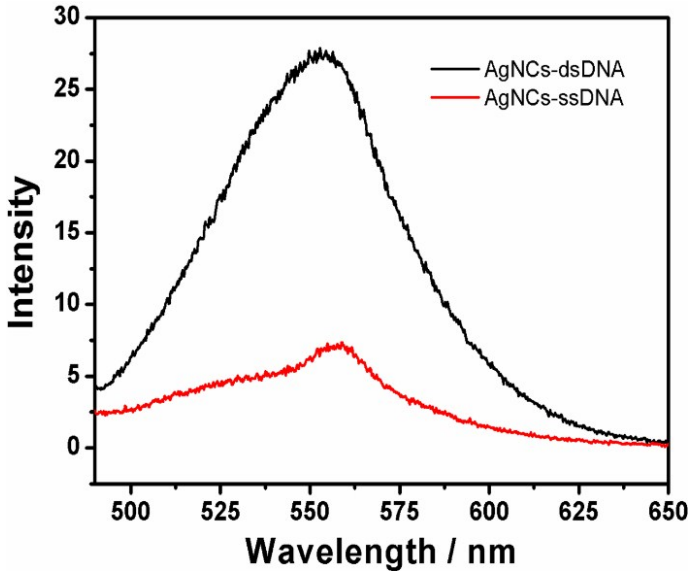

Fig. S3 Fluorescence spectrum of AgNCs-dsDNA (1.0 μM, black) and that of AgNCs-ssDNA (1.0 μM, red) in phosphate buffer solution (pH = 7.4, λ_{ex} = 470 nm), large difference of emission intensity was observed between them.

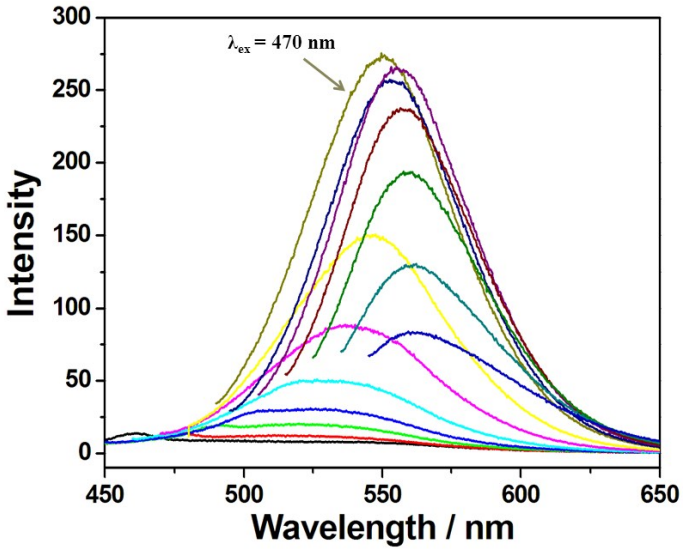

Fig. S4 Fluorescence spectra of AgNCs-dsDNA (10.0 μM) as a function of excitation wavelength (from left to right, which is set from 400 to 530 nm with an interval of 10 nm). The results showed both the emission wavelength and maximum intensity of AgNCs-dsDNA varied with the changes of excitation wavelength, illustrating the wide distribution of the particle sizes and being consistent with the previous related reports.[32-36]

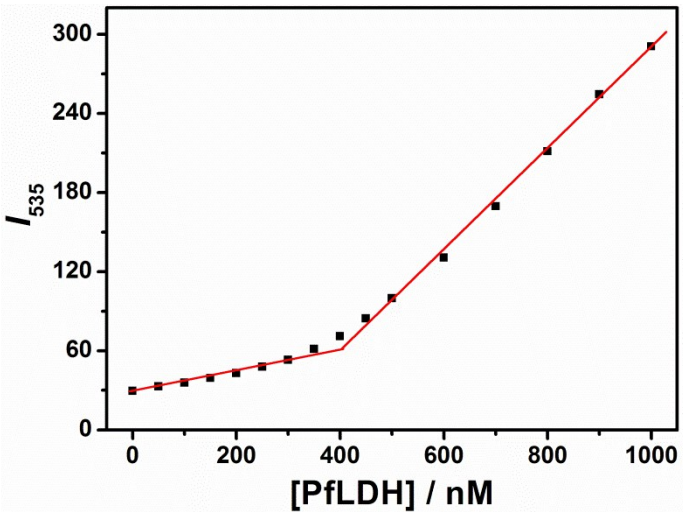

Fig. S5 The enlarged plot of Fig. 2B in the concentration range of 0 to 1000 nM, which showed two different slopes of enhancement, suggesting more than one binding site of AgNCs-dsDNA at PfLDH.

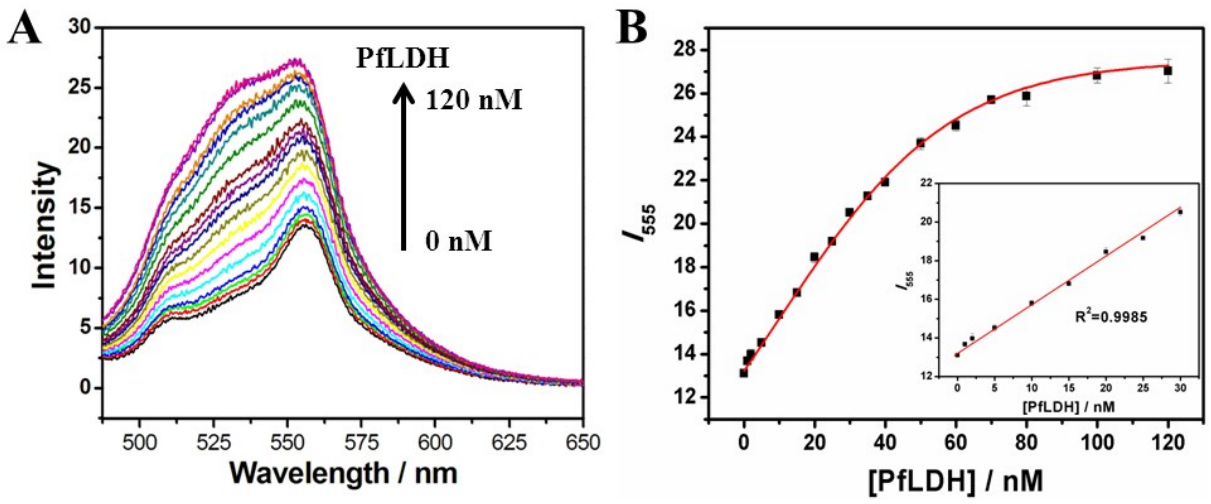

Fig. S6. A) Fluorescence spectra of AgNCs-dsDNA (0.10 μM) in phosphate buffer solution (pH = 7.4) upon adding various amounts of PfLDH (0−120 nM, λ_{ex} = 470 nm); B) The response plot of fluorescence intensity *vs* the concentration of PfLDH (the inset is the enlargement of Fig. S6B in the range of 0−30 nM, in which the limit of detection (LOD) was 0.20 nM, being calculated through 3-fold standard deviation of the blank intensity corresponding concentration of PfLDH at the linear response plot (I_{535} = *0.2396* × *[PfLDH]* + *13.19*), where *[PfLDH]* is the concentration of the corresponding PfLDH).

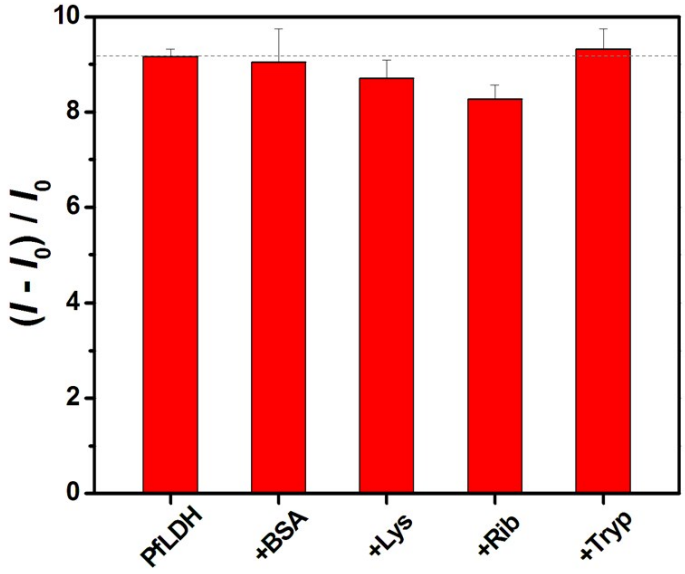

Fig. S7 The fluorescence enhanced ratios of AgNCs-dsDNA (1.0 μM) in phosphate buffer solution (pH = 7.4) in the presence of PfLDH (1.0 μM) together with indicated proteins (1.0 μM). The results indicated no much strong interference of either protein on the detection of PfLDH.

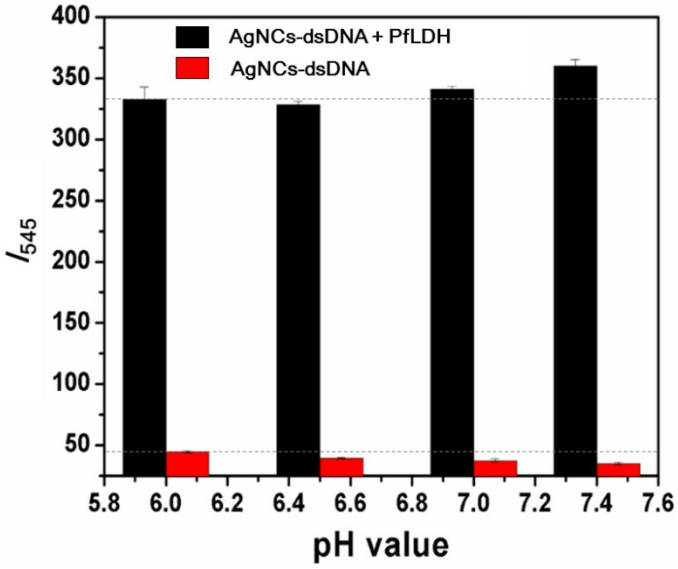

Fig. S8 The fluorescence intensities of AgNCs-dsDNA (1.0 μM) in the absence (red bar) and presence (black bar) of 1.0 μM PfLDH at different pH. The results showed the emission of the complex of AgNCs-dsDNA and PfLDH did not change much with pH. Finally, a near physiological pH at 7.4 was chosen to be used to detect PfLDH as an optimized value.

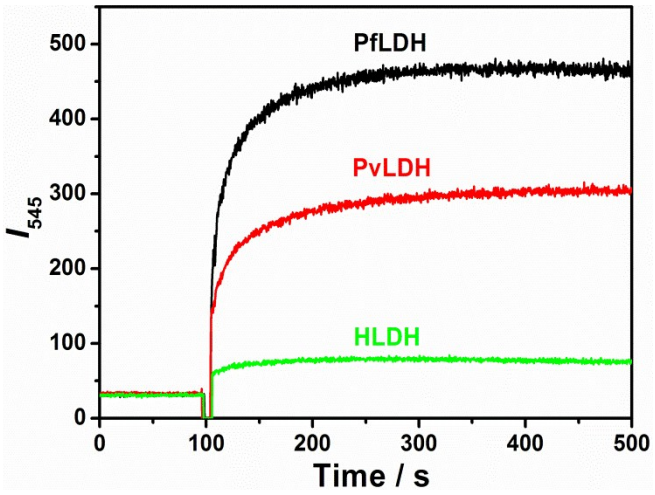

Fig. S9 Time-dependent fluorescence intensity of AgNCs-dsDNA (1.0 μM) in phosphate buffer solution (pH = 7.4), which are measured before (0−100 s) and after adding LDHs (1.0 μM; λ_{ex} = 470 nm, λ_{em} = 550 nm). The results exhibited the interaction of AgNCs-dsDNA and PfLDH was rapid and it reached a maximum within 2 min. Therefore, in following to get more accurate and stable fluorescence result the detection was conducted at 4 min after mixing protein and AgNCs-dsDNA.

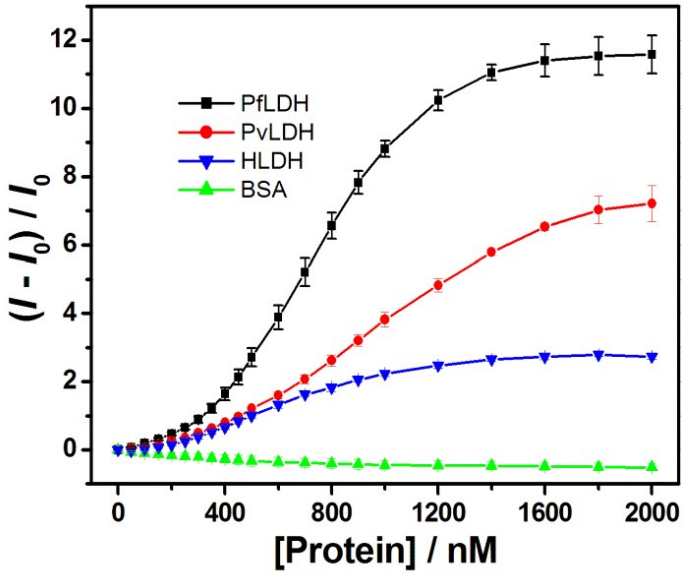

Fig. S10 The fluorescence enhanced ratios of AgNCs-dsDNA (1.0 μM) in the absence and presence of different amounts of proteins (0−2000 nM) in phosphate buffer solution (pH = 7.4). The emission of AgNCs-dsDNA was enhanced by PfLDH, PvLDH, HLDH at different extents.

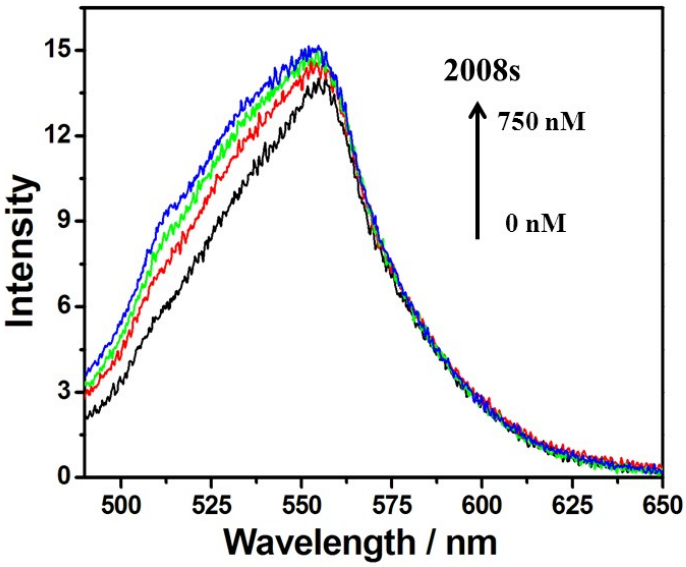

Fig. S11 Fluorescence spectra of AgNCs-dsDNA (0.5 μM) in the absence and presence of different amounts of 2008s (250, 500 and 750 nM, respectively) in phosphate buffer solution (pH = 7.4; λ_{ex} = 470 nm).

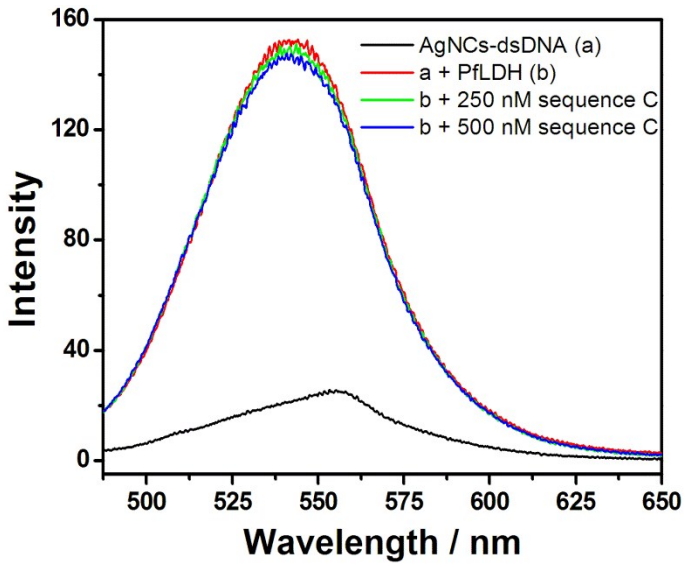

Fig. S12 Fluorescence spectra of AgNCs-dsDNA (0.5 μM) in the absence and presence of PfLDH (0.5 μM), and those after further adding different amounts of single strand oligonucleotide C (sequence C; 250, 500 nM) in phosphate buffer solution (pH = 7.4; λ_{ex} = 470 nm).

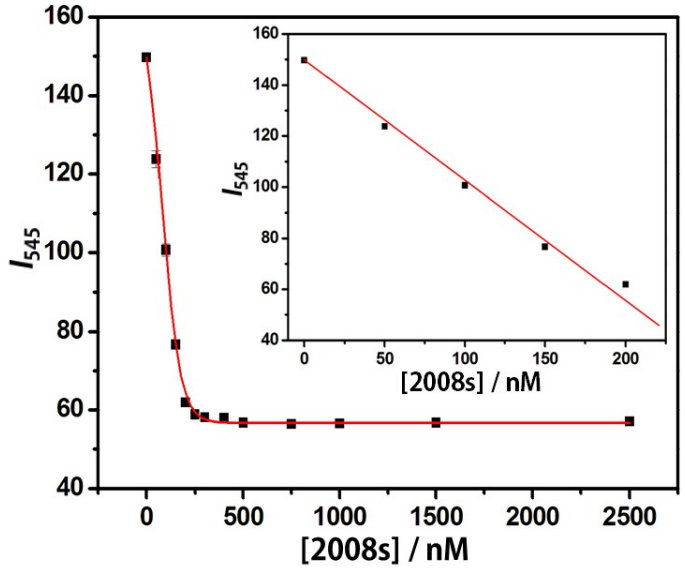

Fig. S13 The emission plot of the mixed solution of AgNCs-dsDNA (0.50 μM) and PfLDH (0.50 μM) in the absence and presence of different amount of aptamer (2008s; 0−2500 nM) in phosphate buffer solution (pH = 7.4). Inset showed the enlargement in the range of 0–200 nM, illustrating a good linear relationship between emission intensity and the concentration of aptamer.

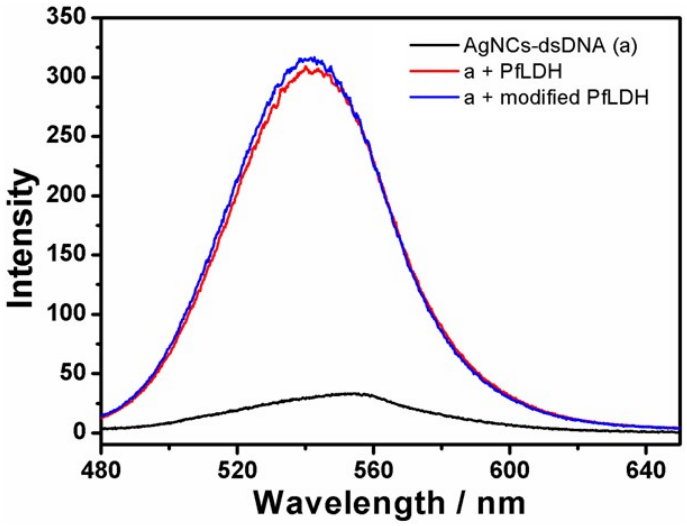

Fig. S14 Fluorescence spectra of AgNCs-dsDNA (1.0 μM) in the absence and presence of PfLDH or modified PfLDH (1.0 μM) in phosphate buffer solution (pH = 7.4; λ_{ex} = 470 nm). The PfLDH was modified by using 4 equiv. N-glycinylmaleimide and 120 min incubation after mixing.

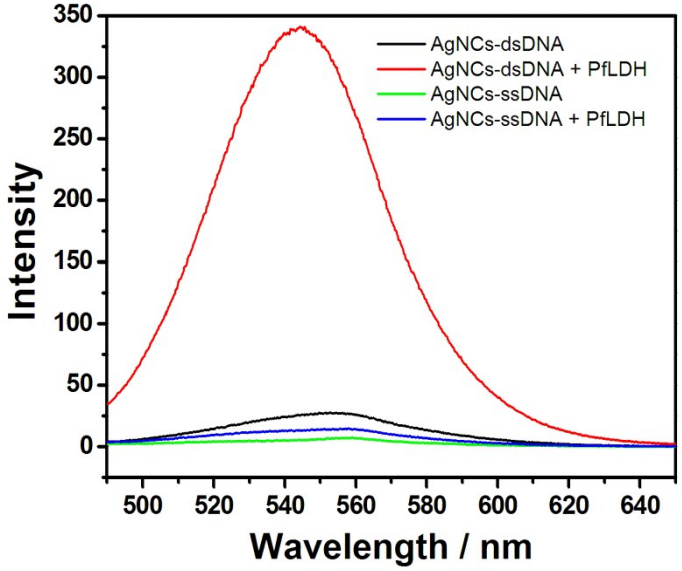

Fig. S15 Fluorescence spectra of AgNCs-dsDNA, AgNCs-ssDNA (1.0 μM) in the absence and presence of PfLDH (1.0 μM) in phosphate buffer solution (pH = 7.4; λ_{ex} = 470 nm), respectively. The results showed PfLDH induced a 9-fold enhancement of AgNCs-dsDNA emission but only 2-fold of that of AgNCs-ssDNA at identical condition.

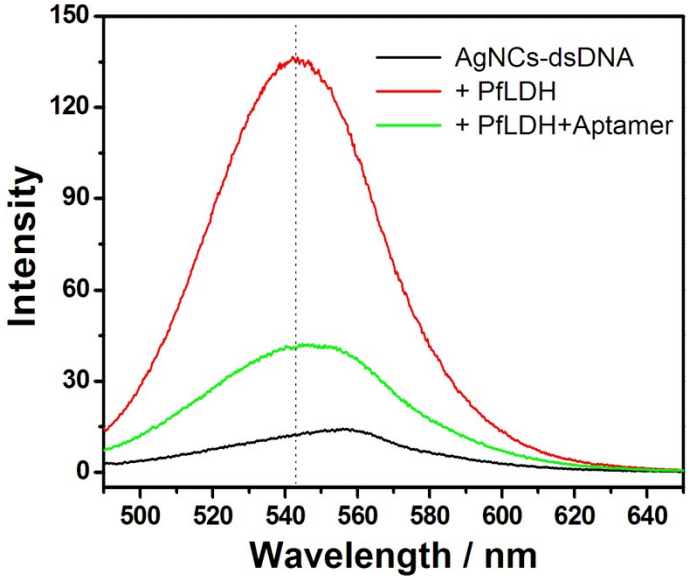

Fig. S16 Fluorescence spectra of AgNCs-dsDNA (0.50 μM) in phosphate buffer solution (pH = 7.4) in the absence and presence of 0.50 μM PfLDH, and further in competition with 0.25 μM aptamer (2008s). The results indicated that the presence of aptamer reduced the fluorescent emission, which might be caused by the replacement of AgNC-dsDNA by 2008s at the surface of PfLDH.

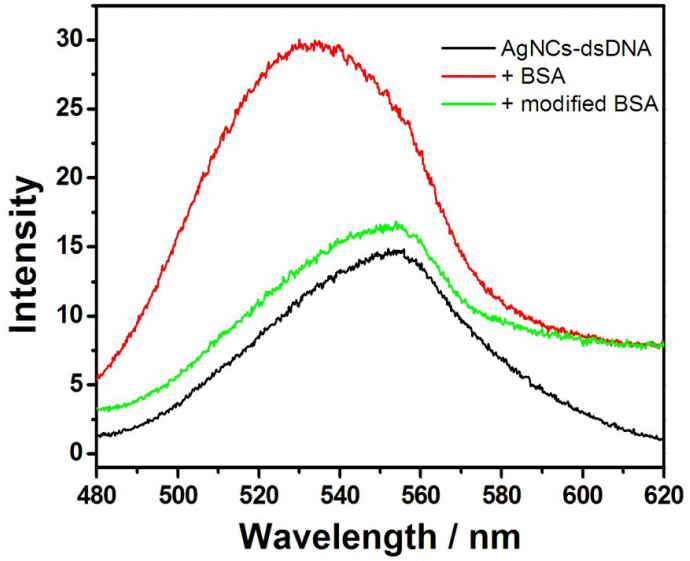

Fig. S17 The fluorescence spectra of AgNCs-dsDNA (0.50 μM) in the absence and presence of BSA (10 μM) or the modified BSA (10 μM) in phosphate buffer solution (pH = 7.4; λ_{ex} = 470 nm). The BSA was modified by using 1 equiv. N-glycinylmaleimide and 120 min incubation after adding.

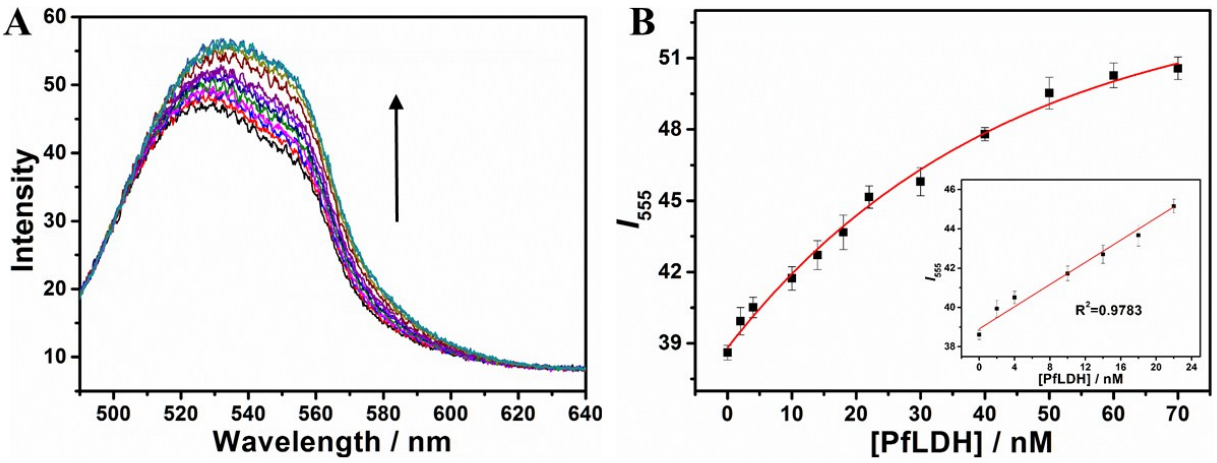

Fig. S18 A) The fluorescence spectra of AgNCs-dsDNA (0.10 μM) in absence and presence of different amounts of PfLDH (0−70 nM) in the diluted fetal calf serum (1% in phosphate buffer solution, pH = 7.4), which were measured 4 min after the addition of PfLDH (λ_{ex} = 470 nm); B) The plot of the corresponding fluorescence intensities in responding to the concentration of PfLDH, and inset shows the enlargement in the range of 0−25 nM, illustrating a good linear relationship between fluorescence intensity and concentration of PfLDH. The limit of detection (LOD) was shown to be 1.0 nM, being calculated through 3-fold standard deviation of the blank intensity corresponding concentration of PfLDH at the linear response plot (I_{555} = *0.2825 [PfLDH] + 39.91*, where *[PfLDH]* is the concentration of corresponding PfLDH.)

Table S1 DNA sequences used in the present study.

Names	Length / nt	Sequence (From 5′ to 3′)
A	24	TTTAAATAATATCCCTTAATCCCC
B	30	GGGGTGGGGTGGGGTGGGATATTATTTAAA
2008s	35	CTGGGCGGTAGAACCATAGTGACCCAGCCGTCTAC
B′	65	GGGGTGGGGTGGGGTGGGATATTATTTAAACTGGGCGGTAGAACCATAGTGACCCAGCCGTCTAC
C	29	CCAAGCTTTTAAGCAGCCAGAGAAACGTCA

Table S2 Comparison of different methods for the determination of PfLDH

Method	System	Range	LOD	Ref.
Colorimetry	AuNPs	10 pM-1 uM	10.3 pM	37
Colorimetry	AuNPs	1 pM-1 uM	2.94 pM	38
Colorimetry	AuNPs	0.1-1 nM	402 ± 40 pM	39
Immunomagnetic beads	antibody modified Magnetic particle	30-2000 pM	25.7±1.1 pM	40
Magnetic based ELISA	magnetic microparticles	1.0–85 pM	0.4 ± 0.2 pM	41
Electrochemistry	aptamer-graphene oxide	0.1-10 fM	0.5 fM	42
Fluorimetry	AgNCs-dsDNA, aptamer	1.0-1500 nM	0.2 nM	this work