Electronic Supplementary Material (ESI) for Chemical Communications

Cationic polymer and aptamers mediated aggregation of gold nanoparticles for the colorimetric detection of arsenic(III) in aqueous solution

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Electronic Supplementary Information

(Including Experimental details, Optimization of sensing conditions, Supplementary figures and table)

Experimental details:

(1) Reagents and chemicals

The sequence of Ars-3 aptamer is reference to previous literatures,[1] and was synthesized by Sangon Biotechnology Co., Ltd. (Shanghai, China). Its secondary structure and sequence are showed in Fig. S1(a,b). Before use, the Ars-3 aptamer was dissolved in 50 mM N-(2-hydroxyethyl) piperazine-N-2-ethanesulfonic acid (HEPES) buffer solution of pH 7.2. Poly(diallyldimethylammonium chloride) (PDDA) was obtained from Sigma-Aldrich (Milwaukee, WI, USA). $HAuCl_4$ and 3-(N-morpholino) propanesulfonic acid (MOPS) were obtained from Sangon Biotechnology Inc. (Shanghai, China). 96-well microplate was purchased from Thermo Fisher Scientific Inc. (Nunclon, Denmark). Unless otherwise mention, all other reagents were analytical grade and used without further purification or treatment. Ultrapure water (Milli-Q plus, Millipore Inc., Bedford, MA) was used throughout.

(2) Instrumentation

Colorimetric assays were recorded on Microplate Spectrophotometer M200 Pro (Tecan Group Ltd, Switzerland). A model of J-815 CD spectrometer (Jasco, Japan) was employed to characterize the structure change of Ars-3 aptamer. Analytical transmission electron microscope JEM-2010HT (Hitachi, Japan) was used to observe the images of AuNPs.

(3) Preparation of AuNPs

AuNPs were synthesized by sodium citrate reduction of $HAuCl_4$ following a literature procedure.[2] All glassware used in this procedure was cleaned in a bath of freshly prepared 3:1(v/v) HNO_3-HCl, then rinsed thoroughly in

ultrapure water and dried in air. AuNPs were prepared by adding 3.5 mL of 1% (w/v) trisodium citrate to a boiling solution of $HAuCl_4$ (100 mL, 0.01% (w/w)) and stirred for 30 min, within the time, the color of the solution changed from light grey, blue, purple, to wine red. The mixture continued to stir for 10 min after removal from the heater. Finally, the cooled solution was filtrated by 0.2 μm ultrafiltration membrane to remove aggregated particles, and then stored in dark glass bottles at 4 °C for further use.

(4) Procedure of As(III) determination

An appropriate volume of 500 nM Ars-3 aptamer solution and 5 μL $NaAsO_2$ solution [As(III)] with varying concentration were mixed thoroughly in a 2 mL plastic tube, and then diluted to 390 μL with MOPS buffer (pH 7.0) and incubated at 25 °C for 30 min. The blank sample was added 5 μL ultrapure water instead As(III) solution. Subsequently, 10 μL PDDA was added into the above mixed solutions and incubated at 25 °C for another 30 min. Finally, 100 μL AuNPs stock solution was added to give a final volume of 500 μL. After incubation at 25 °C for 5 min, 200 μL of above samples were moved into a 96-well microplate for colorimetric assay. The absorption spectra (the wavelength range from 400 nm to 800 nm) and absorbance values at 520 nm (A520) and 650 nm (A650) were measured by Microplate Spectrophotometer M200 Pro. The value of A650/A520 was calculated to indicate the aggregation of AuNPs in solutions. To test the selectivity of the biosensor, other metal salts including Na_2HAsO_4 [As(V)], $Pb(NO_3)_2$, $Hg(NO_3)_2$, $Cd(NO_3)_2$, $AgNO_3$, $CaCl_2$, $ZnCl_2$, $FeCl_3$, $MgSO_4$, $MnSO_4$, $NiSO_4$, $CuSO_4$ and $FeSO_4$ were used.

Optimization of sensing conditions:

The concentration of PDDA and Ars-3 aptamer are critical to the performance of biosensor. To optimize the sensing conditions, the varying concentrations of PDDA were added into the constant AuNPs solutions. The absorption spectra and absorbance values of AuNPs solutions are showed in Fig. S5. The results confirm that 1.52 nM of PDDA is sufficient to aggregate all of AuNPs. The effect of Ars-3 aptamer is presented in Fig. S6, and the result exhibits that 5 nM of aptamer is suitable for the biosensor. The pH value of sensing solutions can change the charge of aptamers and AuNPs, which may affect the colorimetric assays. We regulated the desired pH of sensing solutions by the dilute HCl and KOH solutions. The results show that the suitable pH for biosensor is about 7.0 (Fig. S7). The higher and lower pH value of sensing solutions could cause the aggregation of AuNPs. Considering the adverse effects of sodium on the AuNPs, we used the sodium-free buffer (MOPS, pH 7.2) as the major sensing solution throughout the experiments. Unless otherwise mention, the following experiments were done at the optimized conditions.

Calculation of the detection limit:

According to the previous report,[3, 4] 3σ/slope was used to determine the detection limit of the biosensor. Some details on how to calculate the detection limit are showed below. The standard deviation (σ) of the instrument for colorimetric assays (Microplate Spectrophotometer) was obtained from statistical analysis of the assay results of blank sample.

	1	2	3	4	5	6	7	8	Standard Deviation
A650/A520	0.33012	0.32974	0.32904	0.32955	0.32866	0.32849	0.32811	0.32849	7.100×10^{-4}

The linear fitting equation at low As(III) concentrations was $y=0.3351+4.019 \times 10^{-4} x$ (R=0.996), so the slope is 4.019×10^{-4}, thus 3σ/slope is calculated as 5.3.

Supplementary figures and table:

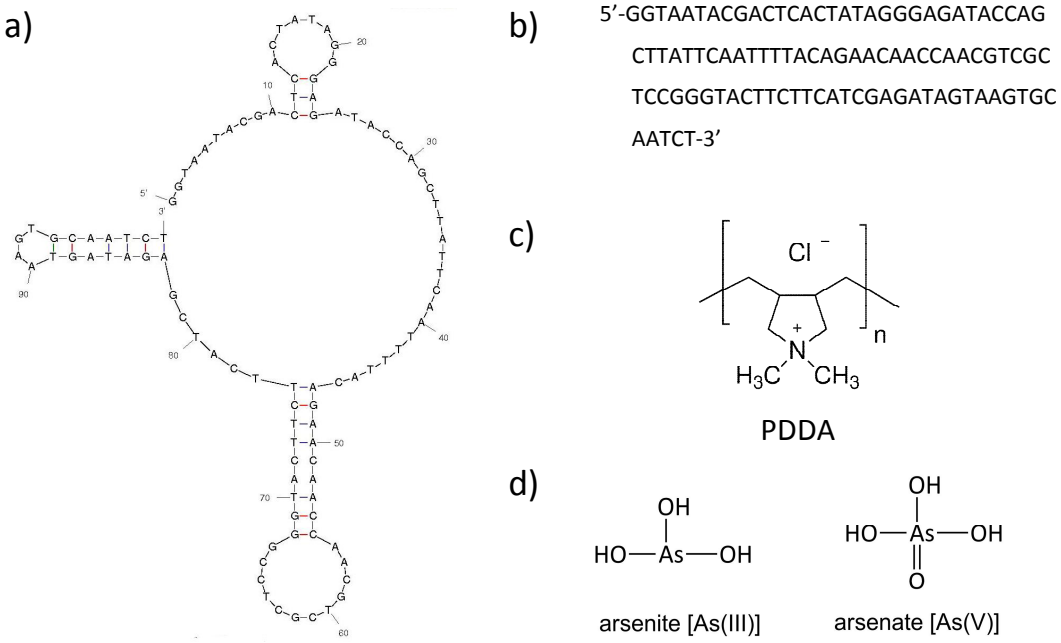

Fig. S1. All components of the biosensor used in this work. (a) Secondary structure of Ars-3 aptamer predicted using Zuker's algorithm Mfold.[1] (b) Sequence of Ars-3 aptamer. (c) Chemical structure of poly(diallyldimethylammonium chloride). (d) Chemical structure of arsenite [As(III)] and arsenate [As(V)] in aqueous solution.

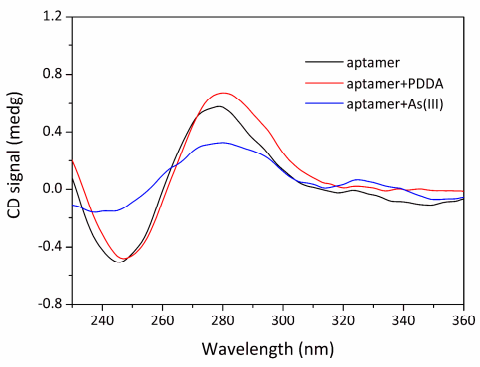

Fig. S2 CD spectra of Ars-3 aptamer solutions treated with PDDA and As(III). Experimental conditions: 500 nM Ars-3 aptamer, 76 nM PDDA and 2000 ppb As(III).

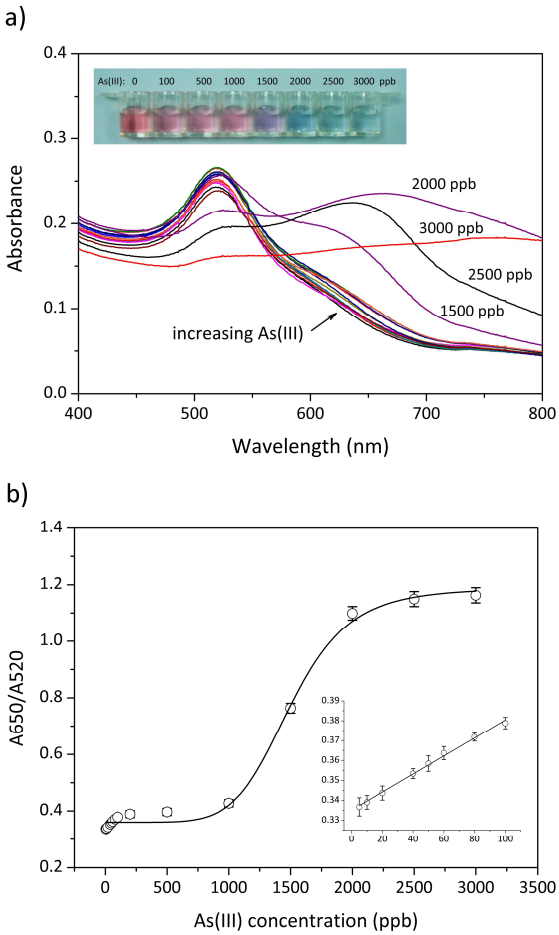

Fig. S3 Sensitivity of the biosensor for As(III) detection. (a) The absorption spectra of the sensing solutions treated with 0, 5, 10, 20, 40, 50, 60, 80, 100, 200, 500, 1000, 1500, 2000, 2500 and 3000 ppb As(III). Inset: Visual colour changes of the sensing solutions. (b) Calibration curve of the biosensor. The curve was fitted to Logistic plot with a correlation coefficient of 0.974. Inset: The values of A650/A520 at low As(III) concentrations, which was fitted to a Linear plot with a correlation coefficient of 0.996.

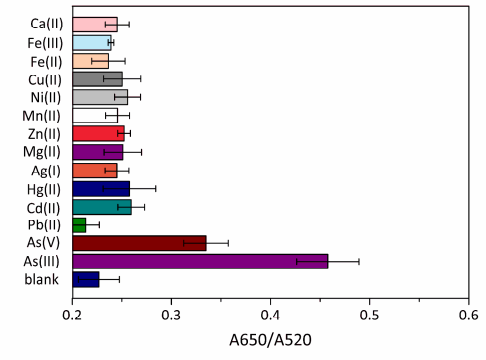

Fig. S4 Selectivity of the biosensor for As(III) detection. The concentrations of metal ions were all 100 ppb.

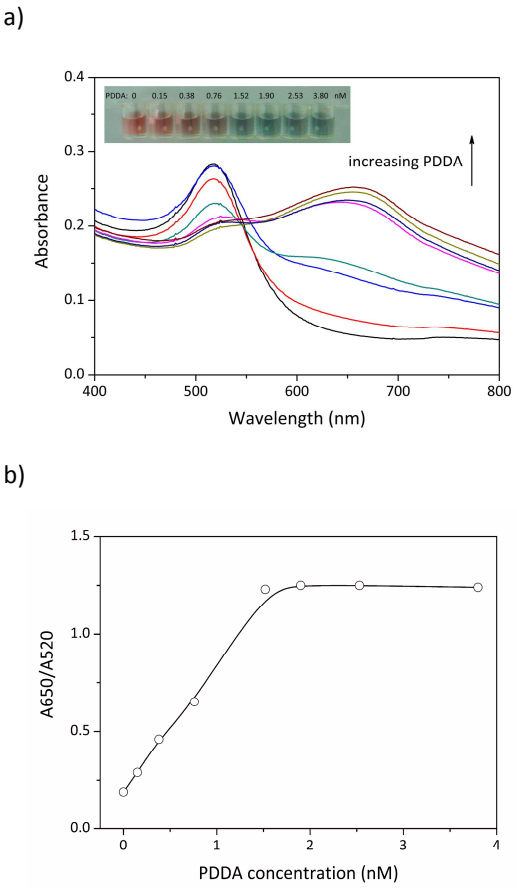

Fig. S5 Effect of PDDA concentrations on the aggregation of AuNPs. (a) The absorption spectra of the AuNPs solutions treated with increasing concentration of PDDA. Inset: Visual colour changes of the AuNPs solutions. (b) The variation of A650/A520 in AuNPs solutions treated with the increasing concentration of PDDA.

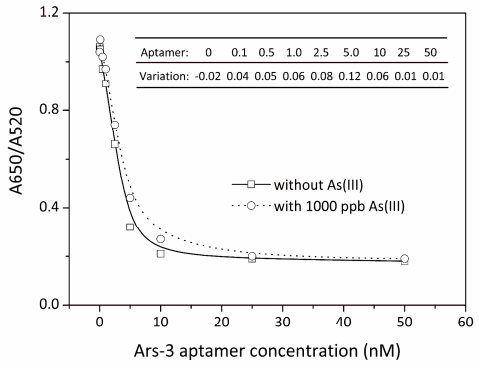

Fig. S6 Effect of Ars-3 aptamer concentrations on the aggregation of AuNPs. The concentration of PDDA was 1.52 nM.

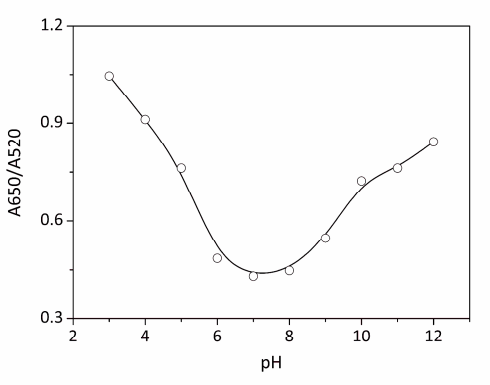

Fig. S7 Effect of pH values on the aggregation of AuNPs. The experimental conditions: 1.52 nM PDDA, 5 nM Ars-3 aptamer and 1000 ppb As(III).

Table S1 Determination of As(III) in water samples

Sample	Mean found ± SD (ppb)	Mean recovery (%)	RSD (%)
$As^{(III)}(10)^{\dagger}$, $Pb^{(II)}(20)$, $Cd^{(II)}(20)$, $Fe^{(II)}(10)$, $Mg^{(II)}(10)$, NO_3^- (80), SO_4^{2-} (20)	11.4 ± 1.6	114	14.0
$As^{(III)}(20)$, $Hg^{(II)}(30)$, $Cd^{(II)}(30)$, $Ag^{(I)}(10)$, $Fe^{(III)}(20)$, $Ca^{(II)}(10)$, NO_3^- (130), Cl^- (60)	21.5 ± 2.3	108	10.7
$As^{(III)}(60)$, $Pb^{(II)}(80)$, $Ni^{(II)}(20)$, $Cu^{(II)}(60$, $Zn^{(II)}(40)$, NO_3^- (160), SO_4^{2-} (80) , Cl^- (80)	56.6± 3.4	94.3	6.0
$As^{(III)}(80)^a$, $Pb^{(II)}(10)$, $Cd^{(II)}(20)$, $Fe^{(II)}(40)$, $Mn^{(II)}(20)$, NO_3^- (60), SO_4^{2-} (60)	82.4± 5.8	103	7.0

† Final concentration (ppb) of ions was added.

Reference

1. M. Kim, H. J. Um, S. Bang, S. H. Lee, S. J. Oh, J. H. Han, K. W. Kim, J. Min and Y. H. Kim, *Environ Sci Technol.* 2009, **43**, 9335-9340.
2. Y. Wang, F. Yang and X. R. Yang, *Biosens Bioelectron.* 2010, **25**, 1994-1998.
3. P. Zhou, Y. G. Wu, S. S. Zhan, L. R. Xu, W. W. Shi, T. Xi and X. J. Zhan, *Chem Commun.* 2011, **47**, 6027-6029.
4. J. W. Liu, A. K. Brown, X. L. Meng, D. M. Cropek, J. D. Istok, D. B. Watson and Y. Lu, *P Natl Acad Sci USA.* 2007, **104**, 2056-2061.