

Electrochemical Aptasensor Based on Prussian Blue-Chitosan-Glutaraldehyde for the Sensitive Determination of Tetracycline

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Abstract: In this paper, a novel and sensitive electrochemical aptasensor for detecting tetracycline (TET) with prussian blue (PB) as the label-free signal was fabricated. A PB-chitosan-glutaraldehyde (PB-CS-GA) system acting as the signal indicator was developed to improve the sensitivity of the electrochemical aptasensor. Firstly, the PB-CS-GA was fixed onto the glass carbon electrode surface. Then, colloidal gold nanoparticles (AuNPs) were dropped onto the electrode to immobilize the anti-TET aptamer for preparation of the aptasensor. The stepwise assembly process of the aptasensor was characterized by cyclic voltammetry (C-V) and scanning electron microscope (SEM). The target TET captured onto the electrode induced the current response of the electrode due to the non-conducting biomolecules. Under the optimum operating conditions, the response of differential pulse voltammetry (DPV) was used for detecting the concentration of TET. The proposed aptasensor showed a high sensitivity and a wide linear range of $10^{-9} \sim 10^{-5}$ M and $10^{-5} \sim 10^{-2}$ M with the correlation coefficients of 0.994 and 0.992, respectively. The detection limit was 3.2×10^{-10} M (RSD 4.12%). Due to its rapidity, sensitivity and low cost, the proposed aptasensor could be used as a pre-scanning method in TET determination for the analysis of livestock products.

Keywords: Aptasensor; Tetracycline; Colloidal gold nanoparticle; Chitosan

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Introduction

Tetracyclines (TCs) have been extensively used as a bacteriostatic and antibiotic drug in livestock production to control bacterial infections and to increase the growth rate of animals owing to its broad-spectrum antibacterial activity [1]. Tetracycline (TET), oxytetracycline hydrochloride (OTC), chlortetracycline (CTC) and doxycycline hydrochloride (DXC) are members of the polyketide family, which includes a number of important pharmaceutical compounds. They are widely used as bactericidal agents in both human and veterinary medicine during recent decades [2]. However, the heavy use of TET and other TCs in aquaculture and

farming industry may lead to the antibiotics residues in dairy food products [3-6]. It has been concerned that the use of antibiotics in food-producing animals, particularly long-term use for growth promotion, contributes to the emergence of antibiotic-resistant bacteria in animals. These resistant bacteria may spread from animals to human beings via the food chain. They may also transfer their antibiotic-resistant genes into human pathogenic bacteria, leading to failure risk of antibiotic treatment for possibly life-threatening human conditions [7]. To safeguard human health, many organizations in the world have officially established a maximum antibiotics residues limit of 100 $\mu\text{g}/\text{kg}$ in the food-producing species [8-10]. So it is of grave importance

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to develop a quick and convenient method to test and identify antibiotics residues in livestock products.

Over the past decade, many studies had been established for the determination of TET residues. Kurittu *et al.* [11] used microbiological inhibition test technique to measure TET residues, but the test lacked specificity and sensitivity. Therefore, a lot of traditional analytical methods have been developed for the replacement of microbiological assays, such as high performance liquid chromatography (HPLC) [12-14], capillary electrophoresis (CE) [15,16], fluorescence (FL) [17,18] and mass spectrometric detection (MS) [19,20]. These traditional analytical methods are highly sensitive and specific, but they are not suitable for widespread and rapid detection. Because they need expensive equipments and complicated procedures for sample pretreatment. In view of these disadvantages, a simple, sensitive and selective method for the determination of TET residue is required.

In recent years, new rapid electrochemical methods for TET detection appeared frequently, such as immunosensors [21], enzyme sensors [22] and aptasensor [23]. Aptamers are nucleic acid ligands selected from random sequence pools *in vitro*, which can be naturally folded into different three-dimensional structures and have the capability of binding specifically to bio-surfaces [24-27]. Aptamers against a specific target can be obtained by an iterative approach called Systematic Evolution of Ligands by Exponential Enrichment (SELEX) [25]. Compared to antibodies, aptamers have more advantages, such as low cost, stability, simplicity, easy modification, target versatility and can be synthetically manufactured. To the best of our knowledge, electrochemical aptasensors for TET detection have been applied to detect TET in the analysis of the food safety [23,28]. Kim *et al.* [28] developed an electrochemical aptasensor based on the screen printed gold electrode using square-wave voltammetry (SWV) technique for the detection of TET, but the proposed aptasensor suffered the drawbacks of low sensitivity, and the stability and reproducibility of the method were not discussed.

Nanometer materials, as the new modified sensing interface, can facilitate the immobilization of biomolecules and improve electrochemical properties of the transducer, such as owning low-background current, high signal to noise ratio and fast electron transfer [29]. Advanced inorganic nanoparticles are currently the most popular material with a wide range of application including optics, electricity and sensors [30]. Among various nanomaterials, colloidal gold nanoparticles (AuNPs) have gained considerable attention because of their large specific surface area, good biocompatibility and high surface free energy [31,32]. Chitosan (CS) possesses excellent membrane-forming ability, high permeability towards water, good adhesion, outstanding biocompatibility, nontoxicity, high me-

chanical strength and susceptibility to chemical modification due to the protonation and insolubility in solution with pH above pKa (6.3) for the deprotonation [33]. It possesses abundant primary amino groups, so it has a high affinity for Au.

In this work, we proposed a novel label-free electrochemical aptasensor based on prussian blue (PB) as the signal for the determination of TET. Herein, PB-chitosan-glutaraldehyde (PB-CS-GA) was synthesized for the first layer modificatory material of the electrode, which could adsorb AuNPs and act as "molecular wire" to facilitate the electron transfer. AuNPs serving as the second layer modificatory material were used for two purposes: (a) to further accelerate the electron transfer (b) to bond and immobilize the anti-TET aptamer onto the electrode via Au-N bond. TET was captured by the anti-TET aptamer immobilized AuNPs/PB-CS-GA modified electrode, which was folded into specific three-dimensional structures. Each step modification of the electrode was investigated by cyclic voltammetry (C-V), and the concentration of TET was analyzed by differential pulse voltammetry (DPV). Due to the inhibition ability of the charge transfer of non-conducting biomolecules, the decrease level of peak currents before and after the TET captured by anti-TET aptamer indicated the concentration of TET. The constructed aptasensor preparation process and determination ability toward its target were studied in detail and it was applied to determine TET in milk samples with satisfactory results.

Experimental

Materials

The anti-TET oligonucleotides purchased from Shanghai Sangon Biological Engineering Technology & Services Co., Ltd. (Shanghai, China) with the following sequences were designed by Niazi [34] and Zhou [23]: 5'-NH₂-(CH₂)-CGT ACG GAA TTC GCT AGC CCC CCG GCA GGC CAC GGC TTG GGT TGG TCC CAC TGC GCG TGG ATC CGA GCT CCA CGT G -3' and probe aptamer 5'-GCA TGC CTT AAG CGA TCG GGG GGC CGT CCG GTG CCG AAC CCA ACC AGG GTG ACG CGC ACC TAG GCT CGA GGT GCA C-6 FAM (FITC)-3' Chloroauric acid (HAuCl₄) and ethanol were obtained from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). Chitosan (CS) was purchased from Sangon Biotech Co., Ltd. (Shanghai, China). Phosphate buffer solution (PBS, pH = 7.4) was prepared from 1.45 g Na₂HPO₄·12H₂O, 0.1 g KH₂PO₄, 0.1 g KCl and 4 g NaCl in 500 mL ultrapure water with its pH adjusted by HCl and NaOH solutions. The other chemicals were analytical reagent grade and used without further purification. Ultrapure water (18.2 MΩ·cm) was produced

by Millipore (PALL, Germany).

Apparatus and measurements

The C-V and differential pulse voltammetry (DPV) were carried out on a CHI660D electrochemical workstation (Chenhua Co., Shanghai, China). A three-electrode system was selected to measure electrochemical response. Modified glass carbon electrode (GCE) (3 mm in diameter) was used as working electrode. Platinum wire acted as auxiliary electrode, and saturated calomel electrode was used as reference electrode. Scanning electron microscope (SEM) was carried out on JSM-6360LV SEM (Japan). Fourier transformation infrared (FT-IR) spectroscopy was recorded with a Nicolet 380 FTIR spectrometer (Thermo Fisher Scientific Co., USA).

Preparation of colloidal gold nanoparticles

The colloidal AuNPs were prepared according to the reference with slight modifications [35]. First of all, all glassware used in these preparations needed to be thoroughly cleaned in aqua regia (HCl:HNO₃ = 3:1). Then the glassware was rinsed with ultrapure water, and oven-dried prior to be used. Then 2.5 mL of 1% sodium citrate solution was quickly added to 100 mL 0.01% HAuCl₄ solution, and the mixture solution was brought to boiling with vigorous stirring. Then the color changed from pale-yellow to claret-red within 2 min. Keeping the solution boiling for 10 min, the mixture was continued to be stirred for another 10 min until the solution returning to room temperature. Finally, the colloidal AuNPs were stored in a refrigerator with a dark-colored glass bottle.

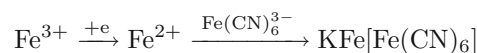
Preparation of CS-GA composites

1% (1 g/100 mL) chitosan solution was prepared by dissolving 1.0 g chitosan flakes into 100 mL 1.0% acetic acid and stirring for 3 h. Then 5% GA solution was added into the 1% CS solution by 10 min sonication to get CS-GA solution. The solution was stored in refrigerator.

Fabrication of the aptasensor

The GCE was sonicated in "piranha solution" (H₂SO₄:30% H₂O₂ = 7:3, V/V), and rinsed with ultrapure water before dried naturally. Next, the GCE was polished to a mirror-like appearance with 0.05 μm alumina slurry, and then sonicated in nitric acid (V/V, 1:1), ethanol and ultrapure water for 5 min, respectively. Firstly, the bare GCE was immersed in the CS-GA solution containing 2.5 mM FeCl₃, 2.5 mM K₃[Fe(CN)₆], 0.1 M KCl, 0.01 M HCl with a constant potential of 0.4 V for electrodeposition for 300 s. PB was deposited onto the surface of the electrode when FeCl₃ (Fe³⁺) and K₃[Fe(CN)₆] (Fe(CN)₆³⁻) were coexis-

tent in the electrodeposition solution, and the reactions were as follows:



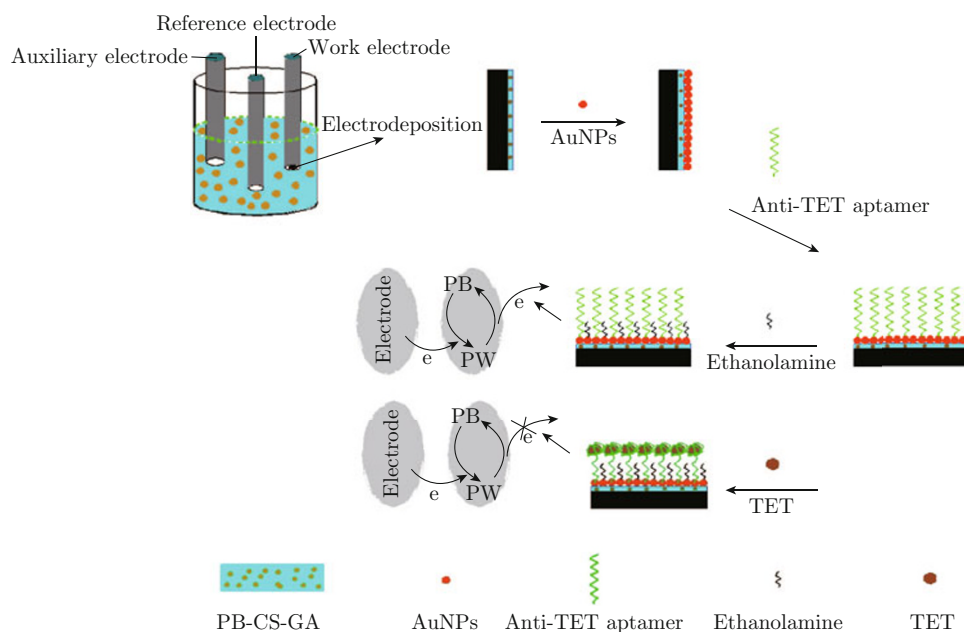
After that, the electrode was cyclic scanned in the potential range from -0.2 to +0.6 V at the scan rate of 50 mV/s until the C-V curve stable, and the PB-CS-GA/GCE was obtained. Then 5 μL AuNPs were added onto the surface of PB-CS-GA/GCE and dried in the air at room temperature (noted as AuNPs/PB-CS-GA/GCE). Then the AuNPs/PB-CS-GA/GCE removed physically adsorbed colloidal AuNPs with ultrapure water and dried by nitrogen. 5 μL anti-TET aptamer of 5 mM was dropped onto the AuNPs/PB-CS-GA/GCE. Then a rubber cap was coated on the electrode to prevent evaporation of the aptamer solution. 4 h later, unbound anti-TET aptamer was removed by ultrapure water and then anti-TET/AuNPs/PB-CS-GA/GCE was passivated by 1 M ethanolamine (EA) for 1 h to obtain EA/anti-TET/AuNPs/PB-CS-GA/GCE (ethanolamine was used to remove non-specific adsorption on the anti-TET/AuNPs/PB-CS-GA/GCE surface). Finally, the EA/anti-TET/AuNPs/PB-CS-GA/GCE was rinsed with ultrapure water and stored at 4°C in refrigerator. The obtained electrode was used as the aptasensor in this work, and the stepwise assembly of the proposed aptasensor was shown in Scheme 1.

Hybridization of anti-TET aptamer with probe aptamer

5 μL anti-TET aptamer with 5 mM concentration was dropped onto the AuNPs/PB-CS-GA modified glass slide. 4 h latter, 2 μL probe aptamer (2 mM) was added and the hybridization reaction was carried out at room temperature in the dark environment for 2 h. The obtained sample was rinsed with 0.1 M Tris-HCl buffer to remove unbound probe aptamer, and then dried in the air atmosphere.

Electrochemical measurements

The electrochemical characteristics of the modified electrode were characterized by C-V and DPV in PBS (pH = 7.4). Electrochemical measurements were established in a conventional electrochemical cell. The formation of anti-TET/TET complexes was performed by dropped 10 μL of different concentration of the target TET on the EA/anti-TET/AuNPs/PB-CS-GA/GCE at 37°C for 1 h. After being carefully washed with ultrapure water to remove unbound TET, the electrode was characterized by DPV for the detection of TET. The DPV measurements were performed in the potential range from -0.1 V to +0.4 V with pulse amplitude of 50 mV.

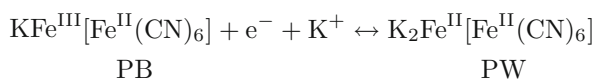


Scheme 1 The stepwise fabrication process of the aptasensor.

Results and discussion

Sensing mechanisms of the aptasensor

In this study, the aptasensor was constructed by covalently attaching an amino-modified anti-TET aptamer on the AuNPs/PB-CS-GA coated GCE surface. It contained 76-base sequence and specifically bound to TET with high affinity. The AuNPs were employed as the carriers of the anti-TET aptamer to amplify the change of peak currents. The PB was used as a mediator to generate the electron flow between bulk solution and work electrode, which was shown in Scheme 1. The anti-TET aptamer modified electrodes presented a stable and free configuration on the GCE surface to give a significantly strong faradaic current. The formation of anti-TET/TET complex hindered the electron-transfer for the PB and Prussian white (PW) transition, thus producing a detectable decreasing signal. The electrochemical data analysis was carried out by the change of the DPV peak currents before and after the TET treatment in PBS (pH = 7.4). The reaction between PB and PW was as follows:



Morphology characterization of the aptasensor

The surface morphology of the PB-CS-GA/GCE and AuNPs/PB-CS-GA/GCE were characterized by SEM. The SEM image of the PB-CS-GA modified GCE was shown in Fig. 1(a). It exhibits the morphology with a rough, porous, and homogeneous layer with packed

uniform particles. When the AuNPs were covered over the PB-CS-GA modified GCE, the surface exhibited a film with many globular features (Fig. 1(b)). Furthermore, to monitor anti-TET aptamer immobilizing process, fluorescent images before and after the probe aptamer assembly were indicated. In Fig. 1(c) and Fig. 1(d), the results showed that the FITC labeled anti-TET aptamer immobilized on AuNPs/PB-CS-GA modified glass released visible green fluorescence under the fluorescent microscopy, while the anti-TET aptamer covered AuNPs/PB-CS-GA modified glass exhibited no fluorescence.

Characterization of the aptasensor

The FT-IR was applied to character the formation and properties of the films. FT-IR spectra of CS, PB and PB-CS-GA were measured and shown in Fig. 2. It could be seen in FT-IR spectra of CS (Fig. 2(a)), the band around the wave number of 3400 cm^{-1} showed an obvious absorption peak which could be attributed to the stretching vibration of —OH or —NH groups. The absorption peaks around the wave number of 1560 cm^{-1} and 1400 cm^{-1} corresponded to the bending vibration of N—H and C—H, respectively. The FT-IR spectra in Fig. 2(b) showed stretching vibration of —CN group which was contained in molecule of PB from 2080 to 2040 cm^{-1} . Figure 2(c) contained all the characteristic peaks appeared in CS and PB, and another typical absorbance band attributed to stretching vibration of —C=O group (around 1640 cm^{-1}) existing in GA, which indicated that the PB-CS-GA was successfully deposited onto the surface of the electrode.

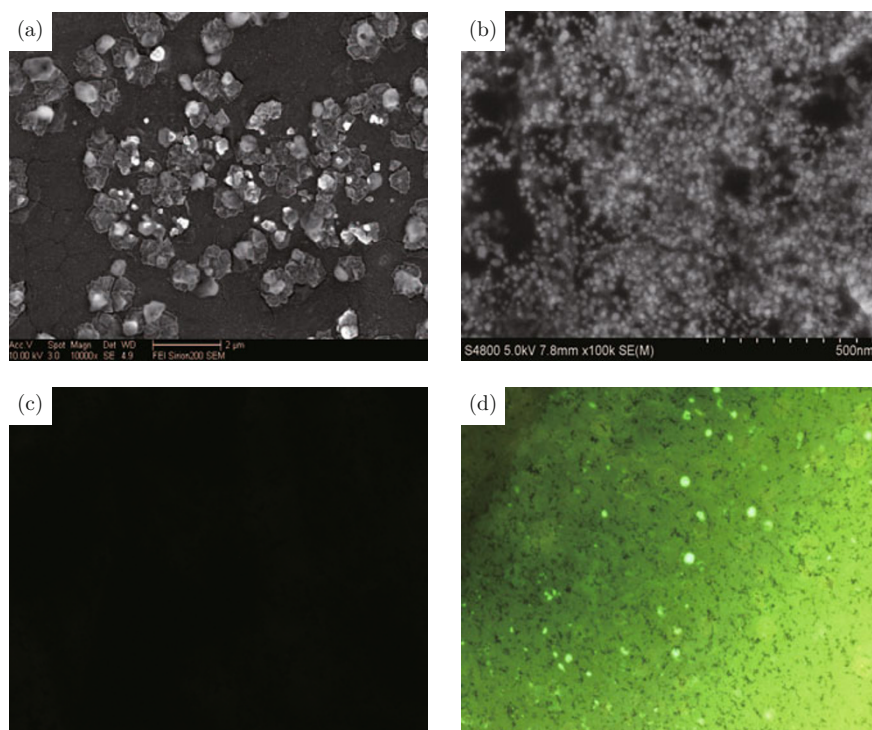


Fig. 1 SEM images of (a) PB-CS-GA/GCE; (b) AuNPs/PB-CS-GA/GCE; Fluorescence microscope images of the anti-TET modified glass (c) and the probe aptamer hybridized with the anti-TET (d).

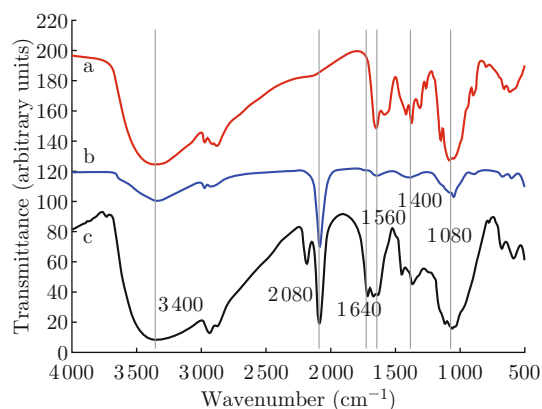


Fig. 2 The FT-IR spectra of (a) CS; (b) PB and (c) PB-CS-GA.

C-V is an effective and convenient technique for probing the feature of the modified electrode surface. Herein C-V was employed to investigate the electrochemical properties of each immobilization step. Figure 3 showed the C-Vs of different electrodes in the PBS buffer (pH = 7.4). No redox peak can be observed for the bare GCE (Fig. 3(a)). Figure 3(b) showed reversible oxidation-reduction peaks of PB in PBS. Compared to the bare GCE, the electrochemical response of the electrode indicated the conversion of PW and PB on the electrode. The current clearly increased when AuNPs were deposited on the electrode, indicating that the use of AuNPs significantly enhanced the conductivity of the electrode as a conductive wire or an electron-conducting

tunnel (Fig. 3(c)). While the anti-TET aptamer was immobilized successfully on the modified electrode, the peak currents decreased (Fig. 3(d)). This was because the immobilized anti-TET aptamer hindered electron transmission. After EA was used to block nonspecific sites and the TET was captured by the anti-TET aptamer, the diffusion of the conductive ion participated in the reaction between PB and PW was hindered, so the peak current further decreased in turn (Fig. 3(e) and 3(f)).

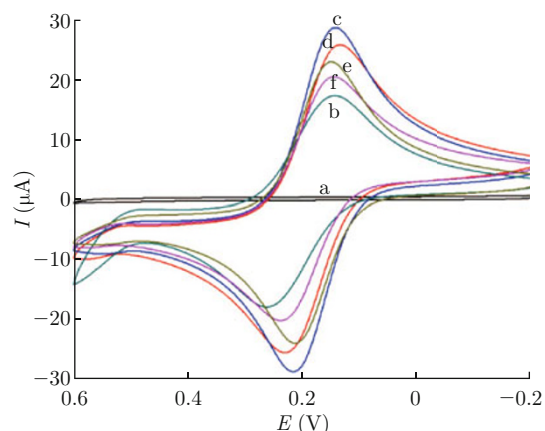


Fig. 3 C-Vs of the bare GCE (a); PB-CS-GA/GCE (b); AuNPs/PB-CS-GA/GCE (c); anti-TET/AuNPs/PB-CS-GA/GCE (d); EA/anti-TET/AuNPs/PB-CS-GA/GCE (e) and TET/EA/anti-TET/AuNPs/PB-CS-GA/GCE (f) in the pH 7.4 PBS in the potential range of potential range from -0.2 to $+0.6$ V at scan rate of 50 mV/s.

Optimization of experimental conditions for aptasensor

Optimal concentration of the anti-TET for the aptasensor

The concentration of the anti-TET was optimized to obtain a high sensitivity of the fabricated electrochemical aptasensor. The effect of the concentration of anti-TET on ΔI (the decrease of DPV peak currents before and after the TET treatment.) was investigated from 2 μM to 6 μM . The results were shown in Fig. 4(a). It could be seen that the ΔI of aptasensor increased rapidly with the increase of anti-TET concentration in the range of 2~5 μM . After the concentration was higher than 5 μM , the ΔI almost kept constant, which indicated 5 μM anti-TET was sufficient to cover the AuNPs/PB-CS-GA modified GCE surface. Therefore, 5 μM anti-TET was selected as the optimum loading amount in this study.

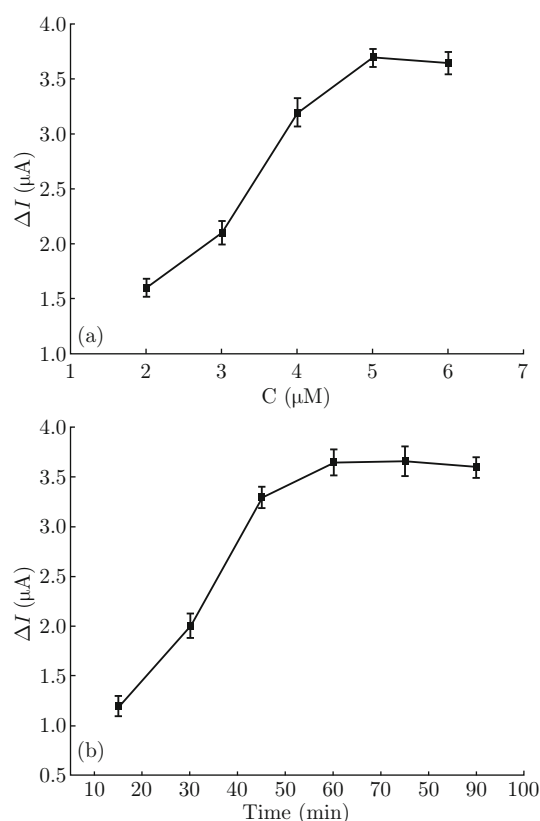


Fig. 4 Optimization of the experimental parameters: effects of (a) the concentration of the anti-TET and (b) the TET incubation time.

Optimal incubation time for the aptasensor

The incubation time of the anti-TET with the TET was an important parameter for the aptasensor. It could be seen that a visible difference in the increase of ΔI with different incubation times. As shown in

Fig. 4(b), the ΔI increased gradually with the augment of incubation time. However, when the incubation time reached 60 min, the signal response sustained a stable value. The result indicated that the interaction of aptamer with TET reached to equilibrium. Thus, the incubation time was controlled to be 60 min.

Performance of the aptasensor

Detection of TET

DPV technique could provide a better peak resolution and current sensitivity, which was very suitable for detecting the concentration of TET. The performed aptasensors were evaluated by the relationship between the ΔI and TET concentrations. Under the optimal experimental conditions, DPVs for TET detection were obtained. As shown in Fig. 5(a), the decrease of peak current was proportional to the logarithm of the TET

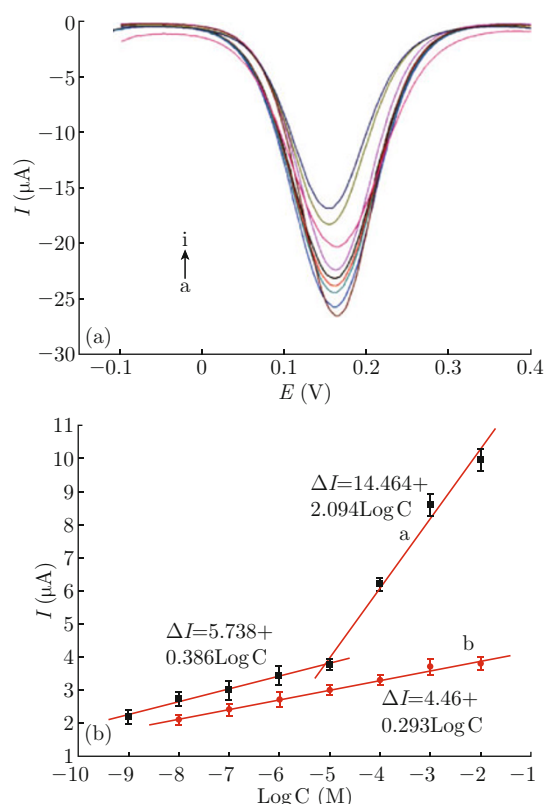


Fig. 5 (a) DPVs of the aptasensor incubated with different concentrations of TET (a-i: the concentrations of TET are 0 M, 1×10^{-9} M, 1×10^{-8} M, 1×10^{-7} M, 1×10^{-6} M, 1×10^{-5} M, 1×10^{-4} M, 1×10^{-3} M, 1×10^{-2} M) were performed in the pH 7.4 PBS in the potential range from -0.1 V to +0.4 V with the parameters: pulse amplitude 50 mV, pulse width 0.2 s, pulse period 0.5 s; (b) The linear relationship between the peak current changes (ΔI) in the DPV and the logarithmic values of the TET concentrations. Error bars showed the standard deviations of measurements taken from three independent experiments.

Table 1 Comparison with other methods for the determination of TET

Method of detection	Limit of detection (M)	Linear range (M)	References
Fluorescence optical fiber sensor	1.06×10^{-7}	6.98×10^{-7} - 8.73×10^{-5}	[37]
Molecularly Imprinted Technique	5.58×10^{-8}	2.25×10^{-7} - 2.25×10^{-5}	[38]
high-throughput suspension array technology	3.4×10^{-9}	1.5×10^{-8} - 6.7×10^{-6}	[39]
high pressure liquid chromatographic	2.1×10^{-8}	–	[40]
aptasensor	3.2×10^{-10}	1×10^{-9} - 1×10^{-2}	this work

concentration. In Fig. 5(b), curve a indicates the current variables of DPV showing a good linear relationship with the logarithmic values of the TET concentrations in two ranges of 1×10^{-9} M \sim 1×10^{-5} M and 1×10^{-5} M \sim 1×10^{-2} M. The linear equation was ΔI (μ A) = $5.738 + 0.386 \text{ Log}C$ (M) and ΔI (μ A) = $14.464 + 2.094 \text{ Log}C$ (M), with a correlation coefficient of 0.994 and 0.992, respectively. The limit of detection (LOD) of TET was calculated as three times higher than the standard deviation for the average measurements of blank samples (LOD = 3 RSD/slope) [36]. Therefore, the LOD was determined to be as low as 3.2×10^{-10} M with relative standard deviation (RSD) of 4.12%, lower than that of TET detection in other ways showed in Table 1. As comparison, the current responses of the aptasensor (without PB-CS-GA composite, EA/anti-TET/AuNPs/GCE, curve b in Fig. 5(b)) were recorded in 0.1 mol/L PBS (pH = 7.4) containing 5.0 mM $\text{K}_3[\text{Fe}(\text{CN})_6]$, and the linear range was 1×10^{-8} M \sim 1×10^{-2} M with the detection limit of 7.4×10^{-9} M. It was observed that the EA/anti-TET/AuNPs/PB-CS-GA modified electrode led a wider dynamic measurement range and a higher sensitivity compared with EA/anti-TET/AuNPs modified electrode. Based on the above work, we could conclude that PB mixed into CS-GA as the signal indicator could improve the sensitivity of the electrochemical aptasensor.

The specificity of the aptasensor

The specificity performance of the aptasensor for TET was further studied in this work. The selectivity test was carried out by measuring and comparing the response of TET (5 μ M) to some possible interferences including a structurally similar tetracycline derivative OTC (5 μ M) and two structurally distinct molecules of Kanamycin Monosulfate (5 μ M) and Gentamycin Sulfate (5 μ M). It could be observed in Fig. 6, the TET sample existed a significant peak current change, while the same concentrations of the other three chemicals had slight emissions. These tests indicated that the developed strategy could be used to identify TET with high specificity.

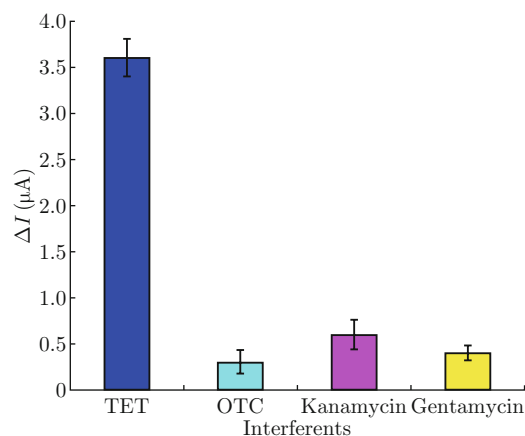


Fig. 6 The specificity of the aptasensor in the presence of 5 μ M TET, 5 μ M OTC, 5 μ M Kanamycin Monosulfate, 5 μ M Gentamycin Sulfate.

Stability and reproducibility of the aptasensor

The stability of the aptasensor was studied by comparing the ΔI after a storage period of 7, 14 and 21 days. The electrodes were stored in refrigerator at 4°C and ΔI was periodically measured. The response current retained above 96%, 93% and 89% of its initial response, respectively, which indicated the storage stability of the aptasensor was quite good.

The reproducibility of the aptasensor was evaluated by detecting the ΔI response with five electrodes prepared in the same way. The five aptasensors were incubated with 10^{-5} M TET, and an acceptable reproducibility with relative standard deviation (RSD) of 5.8% was obtained. The experimental result indicated the proposed aptasensor had a good reproducibility.

Determination of TET in milk samples

In order to evaluate the feasibility of the aptasensor system for possible applications, the standard addition method was used. The milk samples were prepared as follows: the milk was firstly diluted at the ratio of 1:10 and ultracentrifugated at 30 000 rpm for 90 min. In this way, the intermediate layer milk serum was obtained without fat and casein. Then we added TET to the collected TET-free milk serum to the final concen-

tration of 5×10^{-8} M, 5×10^{-6} M and 5×10^{-4} M. As shown in Table 2, the recovery tests were performed using 3 replicates with the aptasensor that prepared in the same way. The recovery of 92~106% indicated that the aptasensor was available for the analysis of TET in milk samples.

Table 2 Determination of TET in milk samples

Sample	Added (M)	Total found (M)	RSD (% (n=3))	Recovery (%)
1	5×10^{-8}	4.8×10^{-8}	5.3	96
2	5×10^{-6}	5.3×10^{-6}	6.2	106
3	5×10^{-4}	4.6×10^{-4}	5.4	92

Conclusions

In this work, a novel label-free aptasensor based on AuNPs/PB-CS-GA for sensitive determination of tetracycline was developed. The PB-CS-GA has performed higher sensitivity of the electrochemical aptasensor due to the PB as the signal indicator. In addition, the PB-CS-GA composites were stable and could be easily prepared. Compared to the electrochemical aptasensor for TET reported by Kim *et al.* [28]. The AuNPs/PB-CS-GA/GCE system showed good conductivity and biocompatibility and exhibited higher sensitivity and stability. Its electrochemical signal had a good linear relationship with the TET concentration in the range of 1×10^{-9} M~ 1×10^{-5} M and 1×10^{-5} M~ 1×10^{-2} M. This strategy contributed to improve the sensitivity and stability of aptasensor, thus, it provided a novel promising platform of aptasensor for TET detection.

Acknowledgements

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