Fabrication of a Versatile Atpasensing Chip for Aflatoxin B1 in Photothermal and Electrochemical Dual Modes

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Abstract
The dual-mode sensing strategy not only has the inherent characteristics of each response mode but also can mutually verify the detection results obtained by different modes, which will effectively improve the accuracy and reliability of detection. The electrochemical-photothermal dual mode not only has two mode signals, but also has the advantages of fast response, high sensitivity, good selectivity, low cost, and simple operation. Efficient integration and accurate detection of pathogenic bacteria to achieve early warning of food safety is one of the most valuable antifouling methods. Here, we report an integrated design strategy to establish a photothermal and electrochemical dual modes for sensitive detection of aflatoxin B1 (AFB1). The aptasensor was fabricated by loading the Au@Fe3O4 onto the indium tin oxides (ITO) conductive glass modified AuNPs nano-layer (ITO/AuNPs) surface through DNA hybridization between aptamer and cDNA. After adding AFB1, aptamer tends to form aptamer-AFB1 complex, resulting in part of Au@Fe3O4 falling off the ITO/AuNPs surface and entering the reaction solution. 3,3′,5,5′-tetramethylbenzidine (TMB) and H2O2 system produces color change under the catalysis of Au@Fe3O4 and realizes the conversion of light and heat under the excitation of near-infrared light. Photothermal signal analysis is realized with the help of thermometer. In addition, combined with electrochemical impedance spectroscopy analysis with ITO/AuNPs electrode, a dual channel method is provided for AFB1 detection. Under the optimal conditions, the developed aptasensor realized sensitive and specific detection of AFB1 from 10 pg mL−1 to 300 ng mL−1 with low detection limit of 5 pg mL−1 in photothermal mode. Importantly, this work provided a promising prospect for the application of photothermal effect coupled with electrochemical aptasensor in food safety.

Keywords Food safety · Atpasensor · Aflatoxin B1 · Photothermal analysis · Dual modes

Introduction
Aflatoxin B1 (AFB1) is a highly toxic substance (Wang et al. 2018), which can cause liver cancer and even death to human beings and animals (Bashiry et al. 2021; Ahmad Al-Jaal et al. 2019; Buszewska-Forajta 2020). Considering its carcinogenic effects on human beings, many countries have established the maximum level of AFB1 between 0.05 and 20 ng mL−1 in all food and agriculture products (Babu and Muriana 2011). The European Union stipulates that the content of aflatoxin B1 in human consumer goods shall not exceed 2 μg kg−1 (Goud et al. 2016). Traditional methods of AFB1 detection include high-performance liquid chromatography and enzyme-linked immunoassay methods (Xing et al. 2020; Xia et al. 2018; Xiong et al. 2018). The methods mentioned above can accurately and sensitively detect aflatoxin B1, but the defects of time-consuming and high detection cost limit their development (Xue et al. 2019; Xiong et al. 2020). Therefore, we urgently need a fast, low-cost detection method (Wang et al. 2019).

Since the advent of aptamers in the 1990s, researchers have worked hard to the research of aptamers with many advantages (Liu et al. 2018; Chen et al. 2017a). The special spatial configuration of aptamers is easy to form different three-dimensional structures, such as spiral, hairpin,
stem ring, convex ring, and other structures, which can easily capture the target based on the interaction of various functional groups (Yang et al. 2018; Barthelmébs et al. 2011; Zhang et al. 2012). Compared to antibodies, the aptamer obtained by screening is easier to prepare and store (Hansen et al. 2006; Wang et al. 2016a). Nowadays, various aptasensor methods came into being and were successfully applied to the efficient detection of aflatoxin B1, such as colorimetric, electrochemistry, and fluorescence aptasensors (Hao et al. 2018; Seok et al. 2015; Luo et al. 2019; Zheng et al. 2016). Among them, the colorimetric system (3,3′,5,5′-tetramethylbenzidine (TMB) and H2O2) mediated by Fe3O4 is considered to be one of the most promising technologies for the determination of AFB1 because it can usually change the color in the process of colorimetric analysis (Woo et al. 2013; Fu et al. 2018). It has been reported that temperature changes in the TMB-H2O2 colorimetric system are more sensitive than color changes, so it is expected to improve the sensitivity of its colorimetric determination through photothermal analysis. At the same time, studies have proved that the TMB-H2O2 colorimetric system has a high photothermal conversion efficiency when the near infrared (NIR) is activated (Luo et al. 2020). Portable devices (NIR thermometers) can quantitatively monitor the temperature changes in the TMB-H2O2 colorimetric system under laser irradiation. Therefore, photothermal analysis has great potential in food analysis sensing applications. In the past few years, photothermal-sensing technology has developed rapidly and made great strides, but the dream of researchers pursuing the high-sensitivity sensing technology is as good as ever. The previous research results of our research group have proved that the enzyme-like catalytic activity of Au@Fe3O4 is 1.5 times that of Fe3O4, which greatly increases the sensitivity of the sensor (Wang et al. 2016b). However, the above single detection mode will inevitably be affected by the possible simultaneous interferences, different operators, instruments, and non-standard detection processes. The dual-mode sensing strategy not only has the inherent characteristics of each response mode, but also can mutually verify the detection results obtained by different modes, which will effectively improve the accuracy and reliability of detection (Fu et al. 2021).

The electrochemical-photothermal dual mode not only has two mode signals, but also has the advantages of fast response, high sensitivity, low cost and simple operation. Here, a versatile aptasensing chip was researched for the photothermal and electrochemical dual modes sensitive detection of AFB1 in this work. Compared with single-mode electrochemical signal, photothermal signal can effectively improve the accuracy and reliability of detection and achieve mutual verification and joint use. ITO conductive glass modified AuNPs nano-layer (ITO/AuNPs) cannot only effectively improve the electronic conductivity, but also effectively link the thiol terminal modified aptamer. First, the aptamer of the terminal modified thiol group connected to the ITO/AuNPs surface (Apt-ITO/AuNPs). Second, the complementary DNA (cDNA) of the terminal amino modified connected to the Au@Fe3O4 (cDNA-Au@Fe3O4). At last, the aptasensor fabricated by loading the cDNA-Au@Fe3O4 onto the Apt-ITO/AuNPs surface obtained by hybridization of aptamer and cDNA. After adding AFB1, aptamer tends to form aptamer-AFB1 complex, resulting in part of Au@Fe3O4 falling off the ITO/AuNPs surface and entering the reaction solution. TMB-H2O2 system with magnetically collected Au@Fe3O4 produces color change under the catalysis of Au@Fe3O4 and photothermal signal analysis under the excitation of near infrared light realized with the help of a thermometer. In addition, combined with electrochemical impedance spectroscopy (EIS) analysis with ITO/AuNPs electrode, a dual mode provided for AFB1 detection.

Experimental Section

Reagents and Apparatus

Diethylene glycol (DEG), glutaraldehyde, ethylene glycol (EG), HAuCl4·4H2O, FeCl3·6H2O, (3-aminopropyl) triethoxysilane (APTS), and polyethylene glycol (PEG) were obtained from Sinopharm Chemical Reagent Co., Ltd. (Beijing, China). OTA, Tris (2-chloroethyl) phosphate (TCEP), aflatoxins B1 (AFB1), 3,3′,5,5′-tetramethylbenzidine (TMB), fumonisin B1 (FB1), and 6-mercaptop-1-hexanol (MCH) were obtained from Sigma-Aldrich. 5′-GAT CGG GTG TGG GTG CCG TAA AGG GAG CAT CCG ACA-SH-3′ (aptamer) (Le et al. 2012; Chen, et al. 2017b) and 5′-CCT TTA CGC CAC CCA CAC CGG ATC-NH2-3′ (cDNA) were purchased from Sangon Biotech Co., Ltd. (Shanghai, China).

The morphologies of samples were observed with the help of transmission electron microscopy (TEM) (HitachS-2400 N, Japan). A three-electrode system is composed of ITO conductive glass modified gold working electrode, Ag/AgCl as reference electrode, and platinum wire as counter electrode. X-ray photoelectron spectroscopy (XPS) was obtained by the ESCALAB 250 multi technology surface analysis system of the Waltham thermal power company in Massachusetts. UV–vis spectra were obtained on a UV-2450 spectrophotometer (Shimadzu, Tokyo, Japan). A diode laser with output power from 0 to 2.5 W and wavelength of 808 nm was purchased from Yuanming Laser Technology Co., Ltd (Ningbo, China). An ordinary digital thermometer was purchased from Boyang Instrument Co., Ltd (Zhengzhou, China).
Synthesis of Au@Fe₃O₄ Nano-Heterojunction

The Au@Fe₃O₄ nano-heterojunction was prepared by one-step hydrothermal method, which was previously reported by our group (Wang et al. 2016b). Typically, 2 mmol of FeCl₃·6H₂O was ultrasonically dispersed in 20 mL of mixture containing EG and DEG (volume ratio of 1:1). Subsequently, sodium acetate and PEG (1.5 g and 1 g) were poured into the above mixture with stirring for 30 min. Finally, HAuCl₄ (2%, 0.5 mL) was injected into the above solution, stirred for 30 min, and transferred to a hydrothermal reactor for reaction at 200 °C for 6 h. The obtained Au@Fe₃O₄ nano-heterojunction (MB) was washed in water and alcohol with the help of magnets for many times, which was dispersed in 40 mL of ethanol in refrigerator for future use.

Preparation of Complementary DNA Conjugated Au@Fe₃O₄ Nano-Heterojunction

The complementary DNA (cDNA) conjugated MB (cDNA-MB) mainly referred to our previous preparation method (Wang et al. 2016b). In short, 0.5 mL of APTS was added with 20 mL of MB prepared above, and after continuous stirring for 1 h, the amino-terminated MB was successfully prepared after repeated water and alcohol washing. Next, 5 mL of glutaraldehyde was added to 20 mL of the above product, shaken at 37 °C for 2 h and purified several times. Finally, 100 μL cDNA was added to the above solution with shaking for 2 h. After washing three times with Tris–HCl buffer, cDNA-MB was successfully prepared and re-dispersed in 20 mL of Tris–HCl buffer (10 mM) for future use.

Preparation of ITO Conductive Glass Substrate with Au Plating Layer

The pretreatment of ITO electrode was performed by the following procedure. Firstly, the ITO electrodes (1.5 × 2 cm²) were sonicated orderly in dilute ammonia water, ethanol, and water for 10 min, respectively. Then, the ITO electrodes were dried under infrared light, and the perforated brown tape with a square hole (the sides of the square are 1 cm) was covered on the surface of ITO electrodes. The electrodeposition of Au NPs on ITO was carried out in 2 mM HAuCl₄ using the cyclic voltammetry (CV) for 30 cycles (potential range: −0.2 to 1.2 V, scan rate of 100 mV s⁻¹). After washing with water and drying under nitrogen stream, the Au NPs modified ITO (ITO/AuNPs) was then obtained.

Preparation of Aptamer Conjugated of ITO Conductive Glass Substrate with AuNPs

Two hundred fifty-microliter aptamer (Apt,100 μM) solution was fully mixed with Tris–HCl buffer (10 mM) containing TCEP (100 mM) and purified after shaking at 37 °C for 1 h to remove excess TCEP. Thirdly, ITO conductive glass substrate with AuNPs plating layer (ITO/AuNPs) was purified by ultrasound for 30 s, incubated with the activated aptamer for 30 min. Finally, after purification, salt aging, and MCH, the prepared Apt-ITO/AuNPs were immersed into Tris–HCl buffer (10 mM) and stored in refrigerator for future use (Wang et al. 2015b).

Preparation of the AptaSensor

After shaking the mixture of cDNA-MB and Apt-ITO/AuNPs for 1 h at 37 °C, the multifunctional aptaSensor MB-cDNA/Apt-ITO/AuNPs was successfully prepared after purification, which was immersed in Tris–HCl buffer (10 mM) and stored a in refrigerator for standby.

Assay Procedure

The aptaSensor of MB-cDNA/Apt-ITO/AuNPs was initially rinsed with acetate buffer solution (ABS) and dried with nitrogen. The aptaSensor was immersed in 3 mL different concentrations of AFB1 standard solution for 30 min at 4 °C. Then, the aptaSensor was used as a working electrode for the EIS measurement in 2 mL electrolyte solution including the redox probe [Fe(CN)₆]³⁻/⁴⁻. With the help of a magnet, the cDNA-MB collected from the reaction solution is transferred to a centrifuge tube containing TMB-H₂O₂ solution. Finally, the temperature of the blue solution after reaction was measured by a thermometer after illumination at 808 nm for 20 s.

Results and Discussion

Characterization of the Au@Fe₃O₄ Nano-Heterojunction

This kind of Au(core)@Fe₃O₄(shell) nano-heterojunction not only has strong magnetism, but also has stronger catalytic performance than pure Fe₃O₄ nanoparticles, which can be proved from our previous reports (Wang et al. 2016b). It can be seen from Fig. 1A, B that the dark AuNPs ball occupies the center of the whole ball, with a diameter of 30 nm, surrounded by slightly lighter Fe₃O₄, with a diameter of 130 nm. The XRD (Fig. 1C) further proved the successful preparation of the Au@Fe₃O₄ nano-heterojunction. The Au@Fe₃O₄ has four more diffraction peaks, which are 38.1°, 44.3°, 64.5°, and 77.6° ascribed to the (111), (200), (220), and (311) planes (curve a) for the AuNPs of face-centered-cubic (JCPDS No. 04–0784) (curve c) compared with the XRD diffraction peaks of pure Fe₃O₄ (curve b). The Au@Fe₃O₄ nanoparticles we prepared have the same typical ferromagnetism as pure Fe₃O₄, with small
magnetic permanence and coercively. As can be seen from Fig. 1D, the saturation magnetism of ferromagnetic material is 50.2 emu g\(^{-1}\) (curve b), less than that of pure Fe\(_3\)O\(_4\) (35 emu g\(^{-1}\)) (curve a), which is mainly due to the integration of AuNPs. It can also be found that the Au@Fe\(_3\)O\(_4\) could quickly concentrate around the magnet and could be quickly and uniformly dispersed by hand shaking in 3 s (inset of Fig. 1D), showing that the Au@Fe\(_3\)O\(_4\) has very similar properties to superparamagnetic particles (Xie et al. 2010).

Characterization of the Complementary DNA Conjugated Au@Fe\(_3\)O\(_4\) Nano-Heterojunction

cDNA can be successfully connected to the surface of Au@Fe\(_3\)O\(_4\) mainly through condensation reaction. According to our previous research results (Wang et al. 2016b), the Au@Fe\(_3\)O\(_4\) surface has a large number of amino groups after APTS modification, and the cDNA end is modified with amino groups. With the help of glutaraldehyde, the cDNA successfully connected to the Au@Fe\(_3\)O\(_4\) surface. We can support this conclusion by UV absorption spectrum. Figure 2A shows that curve a, b are the UV absorption spectra of cDNA and Au@Fe\(_3\)O\(_4\) respectively. Observing curve c, there are corresponding characteristic peaks of cDNA and Au@Fe\(_3\)O\(_4\) at 265 and 585 nm, indicating that cDNA is successfully connected to the surface of Au@Fe\(_3\)O\(_4\).

Fabrication the Aptamer Conjugated ITO Conductive Glass Substrate with AuNPs

The CVs in a scanning potential range between 0 and 2 V in 0.5 M H\(_2\)SO\(_4\) were used to demonstrate the successful deposition of AuNPs on ITO conductive glass layer (Fig. 2B). Two clear peaks were observed from AuNPs-ITO, the one occurring at 1.2 V relative to oxidation of gold and the other appearing at 0.6 V corresponding to the reduction of gold (Ren et al. 2017; Wang et al. 2018; An et al. 2019). For comparison, the CV of bare ITO under the same condition is also displayed as curve a of Fig. 2B, in which we could not observe the characteristic peaks mentioned above. These results further provided good reasons to judge the formation of deposited AuNPs on ITO. Aided by Au–S linkage, the aptamer connected to the AuNPs modified electrode surface, which is seen from Fig. 2C. The XPS spectrum of the electrode modified with aptamer has an obvious P 2p characteristic peak (curve b), which is mainly due to the existence of phosphate skeleton in aptamer, and the center of the peak is at 133.3 eV. In contrast, the XPS spectrum of the electrode without modified aptamer had no characteristic peak of P 2p (curve a).

Optimization of Experimental Conditions

The above data show that we successfully modified the aptamer to the electrode surface. However, the maximum
load of aptamer per unit area and the time required are two important parameters. Figure 2D shows the electrode impedance value of modified aptamer increases with the increase of aptamer concentration (0.1, 0.5, 1, 5 μM). When the concentration of aptamer reaches 1 μM, the impedance value reaches the highest platform. Therefore, we selected the aptamer as 1 μM for the best loading concentration of aptamer.

Figure 3A shows the relationship between loading time and impedance value when the fixed concentration of aptamer is 1 μM. It can be seen that the impedance value increases with the extension of loading time (3, 6, 12, 24 h), but the impedance value increases slowly after 12 h. Therefore, we chose 12 h as the time for the aptamer loading on ITO/AuNPs in this study.

Under the optimization condition, the multifunctional aptasensor connects the cDNA-MB to the Apt-ITO/AuNPs based on the base matching principle between the aptamer and its complementary chain. The step-by-step preparation process of the electrode is completed by monitoring the impedance value of [Fe(CN)₆]³⁻/⁴⁻ (Fig. 3B). The inset of Fig. 3B shows the classic Randles equivalent circuit. It can be seen from the Nyquist plots that any fabrication steps of the electrode will be reflected by the change of the semicircle diameter of \( R_{et} \). Because the electron transfer process of the redox probe on the modified electrode surface is blocked, the interface \( R_{et} \) increases sharply from 350 (curve a) to 950 Ω (curve b) after the aptamer linked on the ITO/AuNPs. After MCH treatment, the \( R_{et} \) value was changed to be 1750 Ω (curve c) (Wang et al. 2015a). Finally, when cDNA-MB was modified to the Apt-ITO/AuNPs, the \( R_{et} \) increased to 3600 Ω (curve d). In order to clarify the maximum concentration of loaded cDNA-MB per unit area of ITO/AuNPs, we use the EIS value of [Fe(CN)₆]³⁻/⁴⁻ to monitor the change trend of loaded concentration of cDNA-MB (Fig. 3C). With the continuous increase of cDNA-MB concentration (from 0.1 to 5 μL mL⁻¹), the impedance diameter also increases. Finally, when the MB concentration is 5 μL mL⁻¹, the impedance diameter no longer increases. Therefore, we choose the concentration of MB as 5 μL mL⁻¹ as the most optimal load. As can be seen from Fig. 3D, the optimum temperature and time for the reaction of the prepared electrode with 50 pg mL⁻¹ AFB₁ are 37 °C and 60 min, respectively.

**Fabrication of the APTasensor and Its Working Principle**

ITO conductive glass is an important substrate for the successful construction of aptasensor. Its conductive layer modified AuNPs nano-layer can not only effectively improve the electronic conductivity, but also effectively link the thiol terminal modified aptamer. See Scheme 1 for the detailed
preparation: Firstly, thiol terminal modified aptamers were modified on ITO/AuNPs surface by Au–S bond. Then, MCH was introduced to block redundant reaction sites of ITO/AuNPs. Our previous studies showed that the surface of MB was rich in hydroxyl groups (Wang et al. 2016b), and the terminal of cDNA was modified with amino groups. With the help of glutaraldehyde, cDNA successfully connected to the surface of MB. Finally, ITO/AuNPs-Apt/cDNA-MB aptasensor was successfully obtained at 37 °C with the help of the specific reaction between aptamer and cDNA. When the aptasensor prepared here was immersed in the solution to be tested containing the target AFB1, AFB1 and its aptamer specifically preferentially bind to promote the separation of cDNA-MB from ITO/AuNPs into the solution to be tested. With the increased concentration of AFB1, the amount of cDNA-MB in the solution to be tested increases. At this time, use a magnet to collect cDNA-MB in the solution to be measured, and then add it to the ABS solution containing TMB and H2O2. Previous studies have shown that MB has peroxidase like enzyme, so cDNA-MB can catalyze H2O2 and oxidize TMB to make it blue. The greater the concentration of cDNA-MB, the darker the blue. The temperature of blue solution does not change under the irradiation of 808 nm laser. At this time, the temperature change was
recorded by a thermometer. The aptasensor was then used to make a three-electrode system into the electrolyte solution including the redox probe for EIS measurements.

**Optimization of Parameters**

To clarify the catalytic performance of MB for TMB and H$_2$O$_2$, we use Vis–NIR spectroscopy to help us analyze the results. As displayed in Fig. 4A, in the absence of cDNA-MB, the visible near infrared absorption of TMBox can be observed at 650 and 890 nm, indicating that MB nanoenzyme has oxidase like activity. With the increase of MB concentration from 5 to 50 μM, the above characteristic absorbance increases to a maximum and stops. It is well known that a class of nano materials with enzyme catalytic activity cannot oxidize TMB to TMBox under neutral conditions, high temperature, and short time. Figure 4B,C shows that the optimum temperature, reaction time and pH value of the MB catalytic system are 37 °C, 15 min, and 4, respectively. Furthermore, the blue solution after the reaction was placed under 808 nm laser beam to obtain the optimal irradiation time. Figure 4D shows that the temperature elevation range increases with the increase of irradiation time (range of 0~5 min) but turned down at 10 min. The temperature does not rise but decreases, which can be attributed to the photo-bleaching of TMBox during long-term irradiation (Zheng et al. 2013; Cheng et al. 2013). Thus, 5 min was used as the irradiation time in this study to avoid the photo bleaching for the aptasensor.

**Fig. 4** Vis–NIR spectra of 250 μM TMB reaction solutions upon different Au@Fe$_3$O$_4$ concentrations (A). Influence of the reaction time and temperature for the Vis–NIR of the TMB upon the 50 μM Au@Fe$_3$O$_4$ (B). Influence of pH for the Vis–NIR of the TMB upon the 50 μM Au@Fe$_3$O$_4$ (C). The effect of irradiation time on aptasensor performance (D). Inset: The responses of the aptasensor with different targets.
Linear Range of the Aptasensor

Under the above conditions, the sensitivity of the aptasensor for AFB1 detection was investigated. As shown in Fig. 5A, with the increase of AFB1 concentration, the impedance radius of aptasensor decreases. The linear relationship between $R_{et}$ value and AFB1 concentration and the relationship between $\Delta R_{et}$ ($\Delta R_{et} = R_{et} - R_{et0}$) value and logarithm of AFB1 concentration are shown in Fig. 5B. The linear equation is as follows: $\Delta R_{et} = -2734 - 1360 \log (c/\text{ng mL}^{-1})$ ($R^2 = 0.996$), and the concentration range is 50 pg mL$^{-1}$–500 ng mL$^{-1}$. The limit of detection (LOD) is calculated as 25 pg mL$^{-1}$, which is much lower than the determined maximum tolerance level of AFB1 in food and feed based on S/N = 3. The aptasensor has a good linear relationship between temperature increases ($\Delta T = T - T_0$) with the logarithm of target AFB1 concentration range from 10 pg mL$^{-1}$ to 300 ng mL$^{-1}$ ($R^2 = 0.994$) (Fig. 5C). The LOD was calculated to be 5 pg mL$^{-1}$, which was lower than the reported colorimetric sensor (Yang et al. 2011; Pradhan et al. 2007) (Fig. 5D). Table 1 lists the detection results of aflatoxin B1 by relevant aptasensors. It can be seen that the aptasensor we developed has a wider detection range than other sensors (Wang et al. 2018; Chen et al. 2017b; Xie et al. 2014).

Selectivity and Reproducibility

In order to verify that the aptasensor has specificity only for AFB1, the aptasensors reacted with the targets (AFB1, OTA, OTB, FB1, and AFM1). The cDNA-MB collected by the magnet is added to the reaction solutions with TMB and H$_2$O$_2$; only the reaction system added with AFB1 has obvious blue change, indicating that the aptasensor has specific selection for AFB1 (inset of Fig. 4D). We performed five

![Fig. 5](image-url) (A) EIS profiles of the aptasensor corresponding to various AFB1 concentrations (from a to h: 0, 0.1, 1, 20, 100, 200, 300, and 500 ng mL$^{-1}$). (B) Relationship between $\Delta R_{et}$ and the different concentrations of AFB1. Inset: the calibration curve for the EIS assay. (C) Temperature of the proposed aptasensor systems upon various AFB1 concentrations (from a to i: 0, 0.01, 0.1, 1, 10, 50, 100, 200, and 300 ng mL$^{-1}$). (D) Relationship between $T - T_0$ and the different concentrations of AFB1. Inset: Calibration curve for AFB1 determination

| Methods          | Liner range (ng mL$^{-1}$) | LOD (ng mL$^{-1}$) | References         |
|------------------|----------------------------|--------------------|--------------------|
| Colorimetric     | 8–250                      | 8                  | Yang, et al. 2011  |
|                  | 0.0004–20                  | 0.00012            | Xie, et al. 2014   |
| EIS              | 0.03–3.3                   | 0.13               | Pradhan, et al. 2007|
|                  | 0.02–50                    | 0.015              | Wang, et al. 2018  |
|                  | 0.03–33                    | 0.02               | Chen, et al. 2017b |
| Photothermal     | 0.001–300                  | 0.0005             | This work          |

EIS electrochemical impedance spectroscopy
AFB1 repeated measurements with a relative standard deviation (RSD) of less than 5.1%.

### Analysis of Real Serum Samples

We applied the aptasensor to the photothermal detection of AFB1 in peanut samples, and the detection results are shown in Table 2. We selected three different contaminated peanut samples with AFB1 concentrations of 0.1, 1, and 10 ng mL\(^{-1}\), respectively. The recovery of AFB1 is 96%–97%, and the RSD value of the recovery is less than 6.3%, which show that the photothermal aptasensor can be used for the determination of AFB1 in peanut samples.

### Conclusions

In this work, a versatile aptasensing chip was proposed for the detection of AFB1 in photothermal and electrochemical dual modes. Using the advantages of multi-functional signal probe and near-infrared laser irradiation, the aptasensor realizes the non-contact and accurate control of the signal. In addition, combined with electrochemical impedance detection technology, this method realizes the dual channel synchronous detection of AFB1, which increases the accuracy and selectivity of detection. Photothermal induced technology is a promising method to realize high-sensitivity signal amplification. Therefore, the combination of photothermal technology and other detection methods to meet the needs of food safety is the development trend in the future.

### Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

### Code Availability

Not applicable.

### Declarations

**Competing interests** The authors declare no competing interests.

**Conflict of Interest** Chengquan Wang declares that he has no conflict of interest. Xin Zhao declares that he has no conflict of interest. Cheng-dong Gu declares that he has no conflict of interest. Foyan Xu declares that he has no conflict of interest. Wuhao Zhang declares that he has no conflict of interest. Xingyi Huang declares that he has no conflict of interest. Jing Qian declares that he has no conflict of interest.

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**Author Contribution** CW: Supervision, conceptualization, methodology, investigation, data curation, writing—review and editing. XZ: Investigation, data curation and writing—original draft. CG: Preparation electrode chip. FX: Sample pretreatment. WZ: Writing—review and editing. XH: Supervision, conceptualization, investigation, data curation. JQ: Supervision, conceptualization, methodology, investigation, data curation, writing—review and editing.

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**Consent to Participate** Informed consent not applicable.

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