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ORIGINAL PAPER

Acetylene black paste electrode modified with a molecularly imprinted chitosan film for the detection of bisphenol A

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Abstract We report on a voltammetric sensor for bisphenol A (BPA) that is based on an acetylene-black paste electrode modified with a chitosan film molecularly imprinted for BPA. The sensor responds linearly to BPA in the 80 nM to 10 μM concentration range, and the detection limit is 60 nM (at an S/N of 3). The use of a molecular imprint provides an efficient way for eliminating interferences from potentially interfering substances. The high sensitivity, selectivity and stability of the sensor demonstrate its practical application for the determination of BPA in plastic samples.

Keywords Molecular imprinting · Bisphenol A · Chitosan · Acetylene black · Voltammetric sensor

Introduction

Nowadays, it is well known that some pesticides and industrial products called endocrine-disruptors chemicals (EDCs) exhibit hormones-like behavior [1]. The effects of EDCs on human reproduction are probably due to their ability to mimic the function of natural estrogens as well as to disrupt the synthesis and metabolism of such hormones by binding the hormonal receptors. Bisphenol A [BPA, 2,2-bis(4-hydroxyphenyl)propane] is an important organic chemical as an intermediate in the manufacture of polycarbonate plastic, epoxy resin, flame retardants and other special products. Recently, BPA has received a great deal of attention from regulatory agencies and scientists, because it has estrogenic activity and serves as an environmental endocrine disruptor. Furthermore many kinds of abnormalities, including the cancers and other diverse pleiotropic actions in the brain and cardiovascular system had been found with long contact to BPA [2]. For these instances, a rapid, simple, sensitive, selective and economic method for the determination of BPA is now highly in demand.

Until now, BPA has been determined by several techniques such as fluorimetry [3], liquid chromatography (HPLC) [4], gas chromatography (GC) [5], enzyme-linked immunosorbent assay (ELISA) [6], and flow injection chemiluminescence [7]. Although these methods can offer good selectivity and detection limit, they are very expensive and time consuming in practice. Accordingly, the design and development of portable devices such as sensors rather than laboratory based instruments for monitoring BPA at trace level is a prime challenge. In this regard, electrochemically analytical technique is an alternative to determine low concentration of BPA in environment. Some chemically modified electrodes have been reported [8–13]. However, these electrochemical sensors suffer from a main drawback, which is moderate selectivity. Therefore, it is necessary to seek a facile, cheap, stable and highly selective method to determine BPA.

Compared with conventional methods, molecularly imprinted sensors (MIS) have recently attracted considerable interest because of their high selectivity for target molecules, relative simplicity, low cost and inherent miniaturization. As a recognition element for sensors, molecularly imprinted polymers (MIPs) are made by synthesizing highly crosslinked polymers in the presence of "imprint" molecules (the template). The template molecule produces cavities specific to its detection in a bulk solution. Removal of the template leaves cavities in the structure with the size, shape and interaction patterns complementary to the template molecule. Then the polymer can rebind template molecule with very high affinity and specificity [14, 15]. Up to now, many MIS based on various functional materials have been

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reported [16–18]. Chitosan (CHIT) is a linear copolymer of glucosamine and *N*-acetylglucosamine units. It displays an excellent film-forming ability, good adhesion, biocompatibility, and high mechanical strength. However, to the best of our knowledge, the construction of an electrochemical sensor for the detection of BPA using molecularly imprinted chitosan film as a recognition element has not been reported in any literature.

Although the selectivity was improved using molecular imprinted technique, low sensitivity still exists in the application. In order to enhance the sensitivity of the sensor, acetylene black (AB), a special kind of carbon black, another carbon nano material, was introduced during the preparation of imprinted sensor owing to its unique properties involving huge surface area, strong adsorptive ability and excellent electric conductivity. In previous reports, AB was successfully applied in the electrode fabrications to increase the determining sensitivity of different species [19–23]. In this paper, direct electrochemical oxidation of BPA was investigated on an acetylene black paste electrode (ABPE) modified with molecularly imprinted chitosan film for the first time. The experimental results demonstrated that the combination of AB particles with molecularly imprinted technique offered an attractive route to enhance the sensitivity and selectivity of the sensor. By clarifying the recognition mechanism and optimizing some of the operating conditions, this study provided a promising approach in sensor preparation and a practical method in BPA analysis.

Experimental

Chemicals and solutions

Acetylene black (AB, purity >99.99 %) was purchased from STREM Chemicals, USA (http://www.strem.com). Chitosan (95 % deacetylation) was purchased from Shanghai Biochemical Co. Ltd., China (http://www.shbiochemical.com). Bisphenol A (BPA) was obtained from Sinopharm Chemical Reagent Co. Ltd., China (http://shreagent.en.alibaba.com). The standard stock solution (1×10^{-2} M) of BPA was prepared by dissolving BPA into ethanol and kept in darkness at 4 °C. Working solutions were freshly prepared before use by diluting the stock solution. 0.1 M phosphate buffer with pH 4.0 was used as the supporting electrolyte. All reagents were of analytical grade and used directly. All the solutions were prepared with redistilled water from quartz and all experiments were carried out at room temperature.

Apparatus

Fourier transform infrared (FTIR) spectroscopic measurements were performed on IRPrestige-21 fourier transform infrared spectrometer (Shimadzu corp., Japan. http://www.shimadzu.com). The morphologies of different electrodes were studied on a JEOL JSM-6610LV scanning electron microscope (Jeol/Ntc.,Japan. http://www.jeol.cn) at an accelerating voltage of 30 kV. Cyclic voltammetry (CV) was performed on a CHI 660D electrochemical workstation (Chenhua Instrument Co. Ltd., Shanghai, China. http://www.chinstr.com) controlled by a microcomputer with CHI660 software. A model JP-303 polarographic analyzer (Chengdu Instrument Factory, Chengdu, China. http://www.scchengyi.com) was used to give second-order derivative linear sweep voltammograms for electroanalytical measurements. A three-electrode system was used, where the molecular imprinted electrode was used as the working electrode, a platinum wire as the counter electrode and a saturated calomel electrode (SCE) as the reference electrode. All potentials reported were versus the SCE. pH measurements were performed with a pH-3c Model pH meter (Shanghai Leichi Instrument Factory, Shanghai, China. http://www.lei-ci.com) using a combined glass electrode. High-performance liquid chromatography (HPLC) was performed on Waters model 510 system (Waters Ltd., USA. http://www.waters.com) comprising a Kromasil 100-5C18 (250 mm×4.6 mm) column equipped with a Waters 2487 dual λ absorbance detector using a mobile phase consisting of 20 mM PBS (pH 7)-acetonitrle (65:35, v/v) at a flow rate of 0.8 mL min^{-1}.

Fabrication of the BPA imprinted sensor

The acetylene black paste electrode (ABPE) was prepared by thoroughly mixing 1.20 g acetylene black powder with 0.30 g solid paraffin in a mortar, and then the mixture was heated for about 5 min until the solid paraffin melted. The resultant paste was tightly pressed into the end cavity (3 mm in diameter) of the electrode body immediately, and the surface was polished with a piece of smooth paper until it had a shiny appearance. Then the electrode was subjected to cyclic potential sweeps between 0.2 and 1.2 V in 1.0 M H_2SO_4 until a stable cyclic voltammogram was obtained. For the preparation of the molecularly imprinted electrode, a 0.5 wt% chitosan solution was prepared by dissolving 500 mg chitosan solid in 100 mL 1.0 % (v/v) acetic acid aqueous solution, 0.01 M BPA was prepared by dissolving BPA into anhydrous ethanol. Then 1.2 mL 0.01 M BPA (template) was added to 5 mL 0.5 wt% chitosan (functional polymer). The mixture was stirred gently for 2 h, and the thus obtained solution (3 μL) was drop-coated onto the surface of the pretreated ABPE, and the solvent was left to evaporate under ambient conditions. Subsequently, the modified electrode was immersed into 0.5 M sulfuric acid (crosslinking agent) at room temperature for 6 h, thoroughly washed with distilled water, air-dried overnight and used the next day. The electrode was tagged as BPA-MIP/ABPE. Removal of the template molecules was achieved by cyclic potential sweeping the modified electrode in 0.1 M HCl solution. The cyclic voltammetry was carried out by potential scanning repeatedly between 0.2 and 1.2 V till there was no signal of BPA. The obtained imprinted sensor was denoted as MIP/ABPE. As a control, the reference non-imprinted electrode (NIP/ABPE) was prepared in the same way mentioned above but without the addition of the template molecules. That is, 1.2 mL anhydrous ethanol was added to 5 mL 0.5 wt% chitosan, and then an aliquot (3 μL) of the solution was casted onto the surface of ABPE. The NIP/ABPE had the same treatment as the MIP/ABPE in order to ensure that the effects observed are only attributed to the imprinting features. The modified electrodes were stored in the air at room temperature before use.

Real sample preparation

Four kinds of plastic products such as polycarbonate (PC) infant nursing bottle, PC water bottle, poly (vinyl chloride) (PVC) bottle and PVC food package bag were purchased from a local supermarket. The sample solutions were prepared according to the literature [12] with a little modification. Briefly, the plastic samples were cut into pieces by scissors, washed twice with redistilled water and then dried. Afterwards, the plastic products were treated as following process to obtain sample solutions. 3.0 g of plastic sample and 30 mL of doubly distilled water were added into a flask fitted with a condenser. The flask was then placed in a water bath at 70 ◦C for 72 h. After cooling to room temperature, the condenser was washed with doubly distilled water and the sample solution was filtrated, and the filtrate was collected. Finally, doubly distilled water was added to make up 100 mL.

Electrochemical measurements

A 0.1 M phosphate buffer with pH 4.0 was used as the electrolyte for BPA analysis. Cyclic voltammetry or second-order derivative linear sweep voltammetry were carried out in the potential range from 0.2 V to 1.2 V with scan rate of 0.1 V s^{-1}, and the oxidation peak current at about 0.78 V was measured. Prior to BPA stripping, the incubation time of the imprinted sensor in BPA test solution was 120 s. After every measurement, the template-entrapped electrode was subjected to cyclic potential sweeps repeatedly between 0.2 and 1.2 V in 0.1 M HCl to remove BPA molecules for reuse. Complete regeneration was verified by the disappearance of the oxidation peak of BPA. The same procedure was carried out in the sample analysis and all electrochemical experiments were performed at room temperature.

Results and discussion

Template removal of MIP/ABPE using CV is presented in Electronic Supplementary Material (ESM).

Characterization of the MIP/ABPE

Figure 1 presents the FTIR spectra of pure chitosan film, sulfuric acid-crosslinked chitosan film, BPA and chitosan-BPA film. Pure chitosan film exhibits absorption bands at 3,447 cm^{-1} (O-H stretching vibration), 3,363 cm^{-1} (N-H_2 stretching vibration), 2,884 cm^{-1}(C-H stretching vibration), 1,657 cm^{-1} (Amide I, C=O stretching vibration), 1,591 cm^{-1} (NH_2 deformation vibration), 1,379 cm^{-1}(CH_3 deformation vibration), 1,152 cm^{-1} (C-O-C deformation vibration) and 662 cm^{-1} (sensitive band of chitosan crystallization [24]), respectively (Fig. 1a). After crosslinked with sulfuric acid, as shown in Fig. 1b, the chitosan O-H and N-H absorption bands gradually became less distinct, while the broad band near 3,250 cm^{-1} became evident. This absorption is assigned to the stretching vibration of N^+-H. The position of the absorption due to the C-H stretching vibration shifted to higher wave numbers (from 2,884 cm^{-1} to 2,954 cm^{-1}). This shift is consistent with decreasing crystallinity of chitosan [25]. Further IR spectral changes could be seen that the band characteristic of NH_2 bending vibrations (1,591 cm^{-1}) gradually weakened, and new absorption

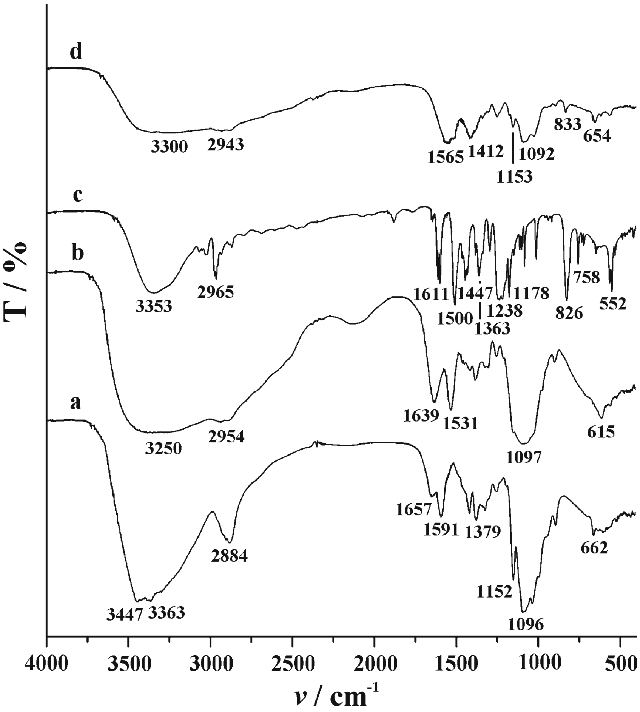


Fig. 1 FTIR spectra of pure chitosan film (**a**), sulfuric acid-crosslinked chitosan film (**b**), BPA (**c**) and chitosan-BPA film (**d**) in the range of 400–4,000 cm^{-1}

bands characteristic of NH_3^+ bending vibrations appeared at 1,639 cm^{-1} and 1,531 cm^{-1}. These results suggest that the NH_2 groups in the chitosan chains were protonated by the H^+ supplied by sulfuric acid. It can also be seen that the small band at 1,152 cm^{-1} became obscured by the broader band at 1,097 cm^{-1}, and the band at 662 cm^{-1} disappeared while another band at 615 cm^{-1} became dominant. These spectral changes can be attributed to the presence of SO_4^{2-} ions [24, 25]. Based on these above results, crosslinking of the membrane occurs when a $SO_4{}^{2-}$ anion interacts with two NH_3^+ groups by ionic bonding. As shown in Fig. 1c, the spectrum of BPA exhibits the characteristic bands of –OH stretching vibration at about 3,353 cm^{-1}, C–H stretching vibration at about 2,965 cm^{-1}, benzene ring skeleton C=C stretching vibration at 1,450~1,600 cm^{-1}, C–O stretching vibration at 1,020~1,275 cm^{-1}, C-C stretching vibration at 750~1,200 cm^{-1}. In the Fig. 1d, the broaden absorption peak at about 3,300 cm^{-1} is corresponding to the hydrogen bond strength. In comparison with that of Fig. 1a, the absorption band corresponding to NH_2 deformation vibration moves from 1,591 to 1,565 cm^{-1}. These changes are due to the formation of hydrogen bonds between $–NH_2$ and –OH in the functional polymer CHIT and –OH in the template molecule BPA.

Scanning electron microscopy (SEM) was employed to investigate the surface morphologies of different electrodes. The SEM images were shown in Fig. 2. On the surface of ABPE, the layer of irregular and large flakes of an AB particle was isolated (Fig. 2a), while the non-imprinted film on NIP/ABPE was relatively flat and compact (Fig. 2b). It is obvious that CHIT, a linear hydrophilic polysaccharide, has excellent membrane forming ability. In contrast with NIP/ABPE, the top views of MIP/ABPE changed significantly. As shown in Fig. 2c, after removal of the template BPA molecules, a three-dimensional network porous structure was presented on the surface of MIP/ABPE, which indicated that the specific cavities were left in the chitosan network.

Cyclic voltammetry (CV) of ferricyanide is an effective and convenient tool to monitor the surface status and the barrier of the modified electrode during each assembly step. Figure 3 shows the CV responses of 1 mM $K_3[Fe(CN)_6]$ solution in 0.1 M KCl at the ABPE, BPA-MIP/ABPE, MIP/ABPE respectively. As is shown, the electrochemical response for $Fe(CN)_6^{3-/4-}$ is a quasi-reversible process with the peak-to-peak separation of 87 mV at 100 mV s^{-1} at ABPE (curve a). While at the BPA-MIP/ABPE, a significant increase in redox peak current is observed, and the peak-to-peak separation is 80 mV (curve b), which could be attributed to the affinity of positively charged chitosan to negatively charged $Fe(CN)_6^{3-/4-}$ [26]. Once the template BPA was extracted from the imprinted matrix, the redox current of $Fe(CN)_6^{3-/4-}$ further increased, as shown in curve c. This current was 2.25 times larger than that measured at the BPA-MIP/ABPE. This could be explained that after removal of the template, the $K_3[Fe(CN)_6]$ molecules could pass through the vacant recognition sites or binding cavity and reached the surface of the electrode more easily.

Electrochemical behavior of BPA at the MIP/ABPE

The electrochemical response of the MIP/ABPE to BPA was compared with the bare ABPE and NIP/ABPE. Figure 4A shows the cyclic voltammograms of the bare ABPE, MIP/ABPE and NIP/ABPE to 5.0 μM BPA in 0.1 M phosphate buffer with pH 4.0 after 120 s incubation time. In the case of the bare ABPE, a small peak (0.779 V, 1.084 μA) can be observed (Fig. 4A, curve a). After the imprinted chitosan film was modified onto the ABPE surface, a very strong and well-defined oxidation peak at 0.780 V was clearly observed (Fig. 4A, curve b). The oxidation peak current of the imprinted electrode is about 5.3-fold compared to that of the bare ABPE. Furthermore, the electrochemical response of the NIP/ABPE to BPA was also examined. Under the same conditions, it could be seen that the oxidation peak current of BPA at the NIP/ABPE (Fig. 4A, curve c) was almost the same as that obtained at bare ABPE, indicating that the non-imprinted chitosan film has no obviously accumulation efficiency towards BPA. Besides, it can be seen that the background current of MIP/ABPE (curve b) was higher than that of bare ABPE (curve a) and NIP/ABPE (curve c), which may be related to

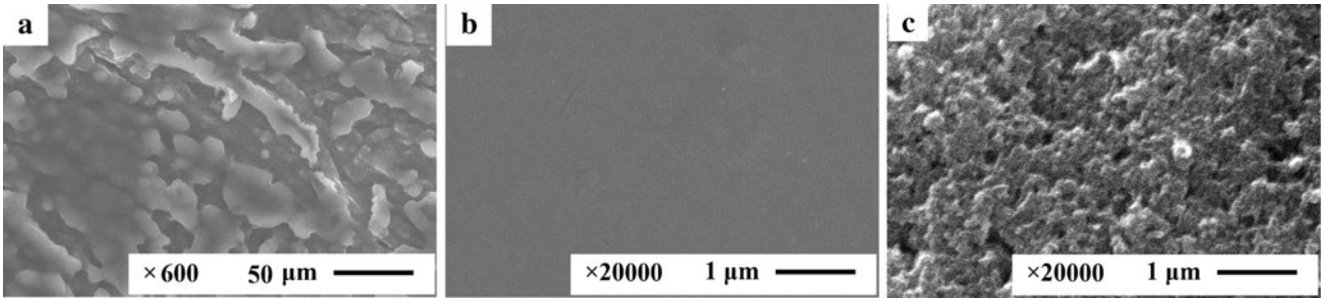


Fig. 2 SEM images of ABPE (**a**), NIP/ABPE (**b**) and MIP/ABPE (**c**). Accelerating voltage: 30 kV

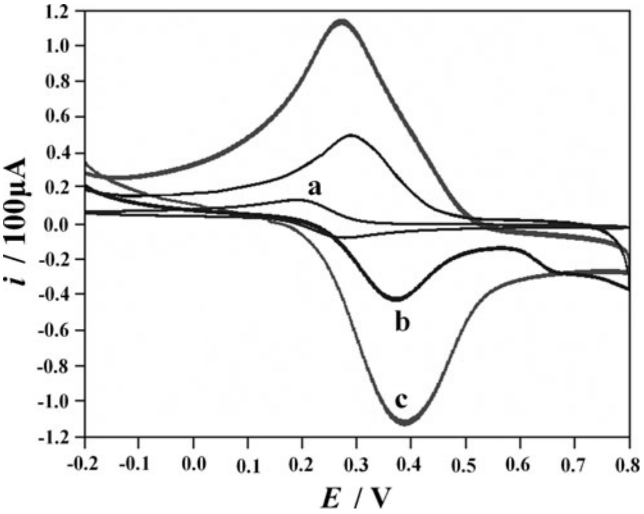


Fig. 3 Cyclic voltammograms for different electrodes in the presence of 1 mM $K_3[Fe(CN)_6]$ and 0.1 M KCl: bare ABPE (**a**), BPA-MIP/ABPE (**b**) and MIP/ABPE (**c**). Scan rate: 0.1 V s^{-1}

the larger surface area of MIP/ABPE. Second-order derivative linear sweep voltammetry is a widely used analytical technique for the enhancement of sensitivity and specificity in quantitative analysis [27–29]. The rebinding ability of the imprinted film was also tested by second-order derivative linear sweep voltammetry experiments (Fig. 4B). Under the same conditions, similar phenomena can be observed. After adding 5 μM BPA into the phosphate buffer, the imprinted electrode (Fig. 4B, curve b) exhibited a much higher current response compared to the bare electrode (Fig. 4B, curve a) and the non-imprinted electrode (Fig. 4B, curve c), indicating that the template BPA imprinted procedure was successful. A much higher sensitivity to BPA by the imprinted electrode should be mainly attributed to three advantages: (i) the AB powder could enhance the electrical conductivity of the sensor and have strong adsorptive ability towards the BPA molecules, (ii) the AB powder could increase surface area of electrode and thus improve the amount of effective imprinted sites on the sensor surface which would greatly improve the enrichment of electrode to BPA species in solution, (iii) the imprinted cavities in the chitosan matrix and the functional groups on the cavities produced by the template BPA molecules on the imprinted electrode shows much higher binding properties to BPA than non-imprinted ones.

Optimization of fabrication conditions of MIP/ABPE

Several key parameters including the concentration of chitosan, the ratio of template to functional polymer, the volume of CHIT-BPA suspension loaded onto the ABPE surface, and the effect of cross-linking agents, were optimized for fabrication of MIP/ABPE (see detailed results in ESM). The results showed that the best electrochemical response of BPA was obtained for 0.5 wt% chitosan. Different imprinted electrodes with different template to functional polymer ratio (the mass ratio between BPA and chitosan vary from 1:5 to 1:20) were prepared, and the results showed that the current response to BPA reached the maximum with the mass ratio of 1:9. The relationship between the volume of CHIT-BPA suspension loaded onto the ABPE surface and the oxidation peak current of BPA has been examined. As shown in Fig. S2 (see ESM), an optimal volume of 3 μL was chosen for analytical purposes. In this study, glutaraldehyde, epichlorohydrin and sulfuric acid were used as cross-linking agent, respectively. The results showed that the peak current of BPA is largest in the case of sulfuric acid. The structure and properties of sulfuric acid-

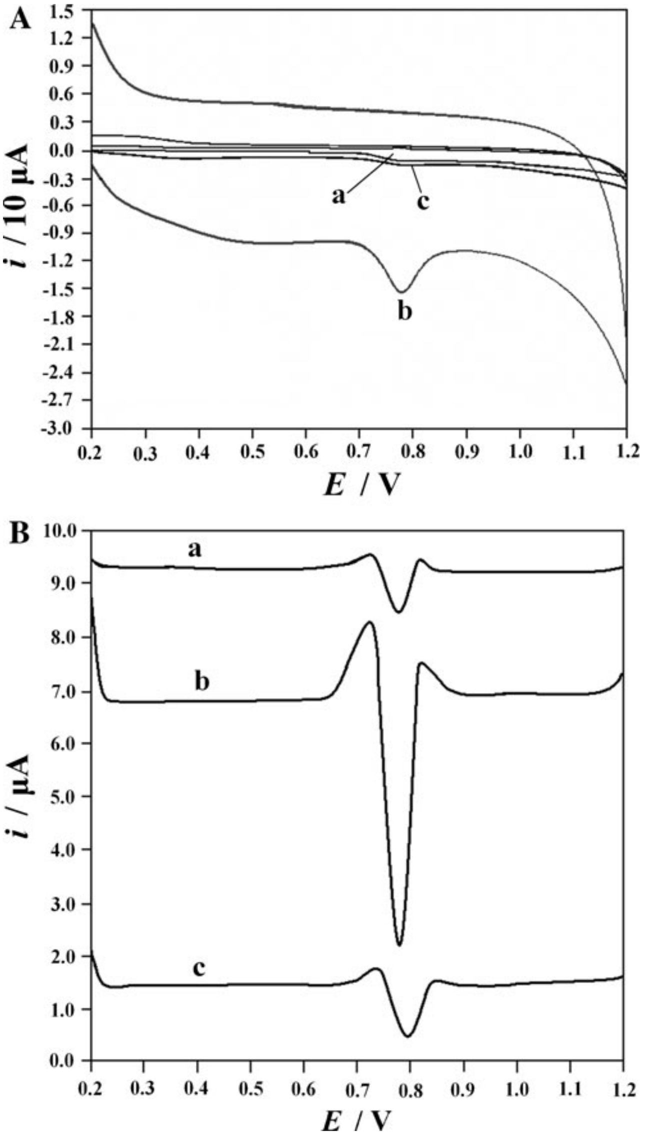
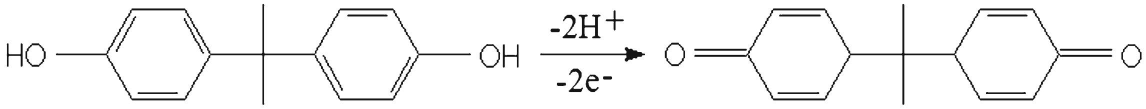


Fig. 4 Cyclic voltammograms (**A**) and second-order derivative linear sweep voltammograms (**B**) recorded at the bare ABPE (**a**), MIP/ABPE (**b**), and NIP/ABPE (**c**) in the presence of 5.0 μM BPA in 0.1 M phosphate buffer (pH 4.0). Incubation time: 120 s, scan rate: 0.1 V s^{-1}



Scheme 1 The reaction mechanism of bisphenol A on the MIP/ABPE

crosslinked chitosan films was studied by infrared spectroscopy, elemental analysis, and X-ray diffraction in previous reports [24, 25], and the results indicated that the strong interactions between chitosan NH_3^+ groups and SO_4^{2-} anions improved the stability of the film structure and the ionic permeability, which may relate to the excellent performance of imprinted chitosan film [30].

Electrode process

The oxidation peak currents of 5 μM BPA at different scan rates from 30 mV s^{-1} to 300 mV s^{-1} were measured by cyclic voltammetry. It was found that the oxidation peak current was proportional to the scan rate. The regression equation was $i_p = 0.0294v + 0.6995$ (i_p in μA and v in mV s^{-1}), with a reasonable correlation coefficient of 0.9929, suggesting that the oxidation process of BPA at the MIP/ABPE was controlled by adsorption. Similarly, a linear relationship between E_p and Napierian logarithm of v ($\ln v$) is also observed in the range of 30–300 mV s^{-1}. The equation can be expressed as E_p (V)=0.023 $\ln v + 0.6741$ (R=0.9931). As for an electron transfer controlled and totally irreversible electrode process, according to Laviron's theory [31], the slope of the line is equal to $RT/\alpha nF$, therefore, αn is calculated to be 1.12. Generally, α is assumed to be 0.5 in totally irreversible electrode process [32], so the electron transfer number (n) is around 2.

The effect of the pH value of the buffer solution on the electrochemical behavior of BPA on the MIP/ABPE was also studied in 0.1 M phosphate buffer. As shown in Fig. S3 (see ESM), with pH increasing, the peak potential shifted negatively, indicating that the oxidation of BPA on MIP/ABPE is a deprotonation process. The relationship between the anodic peak potential and the solution pH value could be fitted to the regression equation of E_p (V)=−0.0598 pH+1.0222, the correlation coefficient was 0.9975. The slope is found to be −0.0598 V/pH, which is very close to the theoretical value of −0.059 V/pH, indicating equal number of electrons and protons involved in the oxidation. Since $n \approx 2$ determined above, the electrooxidation of BPA at the MIP/ABPE is a two-electron and two-proton process, which can be illustrated as Scheme 1.

Optimization of analytical parameters

In order to find the optimum conditions with the highest sensitivity and larger linear dynamic range for the determination of BPA, the influence of various parameters, including supporting electrolyte, solution pH and incubation time, were studied (see ESM). It was found that BPA had the best electrochemical responses in 0.1 M phosphate buffer. As shown in Fig. S3 (see ESM), the effect of pH on the peak current of BPA was investigated, and the buffer solution of pH 4.0 was selected for further experiments. The experimental results also showed that the incubation time has effect on the peak current (Fig. S4, ESM). In all further studies, an incubation time of 120 s was used for further experiments as a compromise between sensitivity and analysis speed.

Calibration graph

Under the optimal experimental conditions described above, the analytical performance of the imprinted sensor to BPA was examined. Fig. S5 (see ESM) displayed the second-order derivative linear sweep voltammotric responses of the MIP/ABPE in 0.1 M phosphate buffer with pH 4.0 containing different concentrations of BPA after the electrode had been incubated for 120 s. It can be seen that the peak current increased with the concentration of BPA increasing. The inset of Fig. S5 shows the calibration curves of the imprinted electrode and the non-imprinted electrode. For the imprinted electrode, the peak currents were proportional to the concentrations of BPA in the range of 80

Table 1 Comparison of the efficiency of some molecularly imprinted electrochemical sensors for the determination of bisphenol A

Technique	Molecularly imprinted electrochemical sensor	Linear range/M	Detection limit/M	References
Amperometry	Molecularly imprinted poly(2-aminothiophenol)-gold nanoparticles modified glassy carbon electrode	$8.0\times10^{-6}\sim6.0\times10^{-2}$	1.38×10^{-7}	[33]
Cyclic voltammetry	Surface molecularly imprinted ordered mesoporous silica polymer modified carbon paste electrode	$1.000\times10^{-7}\sim5.000\times10^{-4}$	3.222×10^{-8}	[34]
Amperometry	Molecularly imprinted sol–gel/gold nanoparticles-multi-walled carbon nanotubes modified gold electrode	$1.13\times10^{-7}\sim8.21\times10^{-3}$	3.6×10^{-9}	[35]
Derivative voltammetry	Molecularly imprinted chitosan film modified acetylene black paste electrode	$8.0\times10^{-8}\sim1.0\times10^{-5}$	6.0×10^{-8}	This work

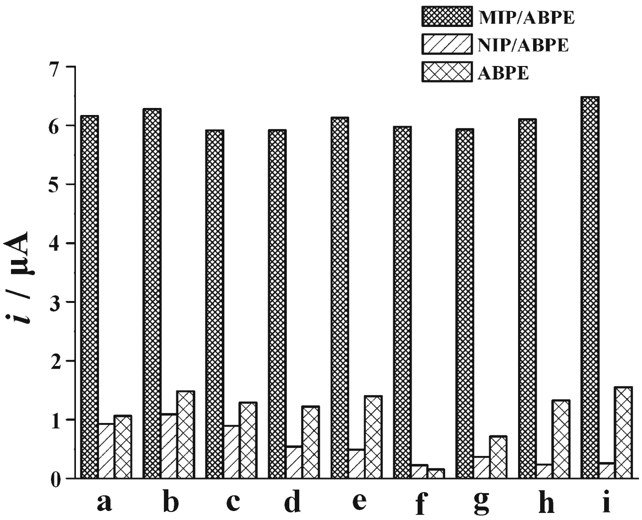


Fig. 5 Peak currents of (**a**) 5.0 μM BPA in 0.1 M phosphate buffer (pH 4.0), (**b**) a+5.0 μM 4-tert-butylphenol, (**c**) a+50 μM phenol, (**d**) a+0.5 mM p-nitrophenol, (**e**) a+0.5 mM o-nitrophenol, (**f**) a+50 μM o-aminophenol, (**g**) a+0.5 mM p-aminophenol, (**h**) a+0.5 mM hydroquinone, (**i**) a+0.5 mM catechol

nM~10 μM. The liner regression equation was i_p (μA)= 1.1126 c (μM)+0.5721, the correlation coefficient was 0.9984. The limit of detection (S/N=3) was calculated to be 60 nM. For the non-imprinted electrode, the linear detection range was 0.2 μM ~ 6.0 μM. The liner regression equation was i_p (μA)=0.1956 c (μM)+0.098 with correlation coefficient of 0.9947, and the peak current appeared a leveled-off trend after the concentration was higher than 6.0 μM. Compared with the MIP/ABPE, the NIP/ABPE gave a lower current, lower sensitivity and narrower linear detection range, which can be explained by the lack of specific binding sites.

Only in a few cases other molecularly imprinted electrochemical sensors for the determination of BPA were reported [33–35]. In order to make a realistic comparison with previous procedures, a detailed comparison of the performance of different imprinted electrochemical sensors for the determination of BPA was summarized in Table 1. As can be seen from Table 1, the presented method can provide comparable linear range and detection limit with other previous procedures. Besides, this new method shows a significant improvement in simplifying electrode preparation, saving time and lowing cost, which demonstrated that the developed electrode has satisfactory analytical performance and it can be a feasible sensor for the determination of BPA.

Selectivity

Selectivity is one of potential merits for an imprinted sensor. The selectivity of the imprinted sensor to BPA was evaluated by testing the second-order derivative linear sweep voltammetric response of BPA or BPA in the presence of some analogues and potential interfering substances, which include 4-tert-butylphenol, phenol, p-nitrophenol, o-nitrophenol, o-aminophenol, p-aminophenol, hydroquinone and catechol, respectively. As shown in Fig. 5, the presence of these species hardly caused any significant change of peak current of BPA at the MIP/ABPE, in which peak current only slightly varied from 96.1 % to 105.3 % of that of BPA alone. It was obvious that structural analogs could not rebind on the electrode, and had small response on MIP sensor. Thus they would not interfere with the determination of BPA. In contrast, the voltammetric response of BPA at the bare ABPE or NIP/ABPE changed greatly in the presence of interferences. This could be explained by the oxidation and adsorption competition of the interferences and BPA at the electrode surface.

Reproducibility and stability

The most attractive feature with the use of MIP/ABPE for determination of BPA is the easy renewal of surface for the next use. The reproducibility of the measurements was evaluated by measuring the second-order derivative linear sweep voltammetric responses of 5.0 μM BPA at the MIP/ABPE. The relative standard deviation (RSD) for seven determinations is about 2.2 %. The MIP/ABPE was stored under desiccated conditions at room temperature when not in use. The sensor retained 94 % of the initial current after 10 days.

Table 2 Determination of BPA in plastic samples (n=4)

Sample[a]	Determined by this method				Determined by HPLC[b]/μM
	Measured[b]/μM	Added/μM	Found[b]/μM	Recovery/%	
Infant nursing bottle (PC)	0.374(±0.012)	0.50	0.896(±0.033)	104.4	0.438(±0.030)
Water bottle (PC)	0.253(±0.008)	0.50	0.746(±0.025)	98.6	0.317(±0.026)
Water bottle (PVC)	0.518(±0.020)	0.50	1.025(±0.037)	101.4	0.583(±0.045)
Food package bag (PVC)	0.736(±0.030)	0.50	1.247(±0.042)	102.2	0.758(±0.043)

[a] All samples were collected from local supermarkets

[b] Average ± confidence interval, the confidence level is 95 %

Analytical application of the electrochemical sensor

In an attempt to explore the imprinted sensor for practical applications, the sensor was applied to determine BPA in different kinds of plastic samples. The sample solutions were prepared as described above. An aliquot of the sample solutions was taken and BPA was determined following above developed procedure using the standard addition method. The results were shown in Table 2 and the recoveries are in the range of 98.6–104.4 %. High-performance liquid chromatography (HPLC) was also used to detect the content of BPA to testify the accuracy of this method. The results obtained by the MIP/ABPE were in good agreement with the results obtained by HPLC. Therefore, the method has good accuracy and great potential in the practical sample analysis without the pretreatment of samples.

Conclusion

A electrochemical sensor for the determination of BPA has been developed via drop-coating method to form a thin molecularly imprinted chitosan film on the surface of an acetylene black paste electrode. This sensor shows a high degree of sensitivity, selectivity and broad linear measuring range. The construction of this sensor is relatively simple and it provides a rapid and economical electrochemical method for the determination of BPA. In addition, it showed good repeatability and stability. Furthermore, the molecularly imprinted sensor had been successfully used to analyze trace BPA in the plastic samples.

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