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Determination of Bisphenol a with Poly (p-Aminobenzoic Acid) Modified Gold Electrode by Using Differential Pulse Voltammetry

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Abstract

Bisphenol A (BPA) is important matter in chemical industry used in the produce of some food contact materials such as plastics, adhesives and coatings. But BPA has also some toxic effects for human endocrine system and environment. Therefore, novel sensitive and selective methods for determination the level of BPA in environmental, medical samples and foods new measurement methods need to be developed. A novel, sensitive, simple and cheap electrochemical sensor based on poly (p-aminobenzoic acid) p(p-ABA) modified gold electrode (AuE) was developed for the measurement of BPA in water samples. Firstly, AuEs were modified with polymerization of the p-amino benzoic acid as a monomer by using bulk electrolysis in 0.01 M HCl at -250 mV. Then the electrochemical oxidation of BPA carried out in 0.1 M phosphate buffer solution (PBS) pH=6 on p(p-ABA) modified AuE. The effects of various electrochemical parameters, such as scan rate, electroyte type and electrolyte pH were examined. Differential pulse voltammetry (DPV) was used as an ideal analytical method for the qualitative and quantitative detect of BPA in water samples. The developed electrochemical sensor has linear signal to BPA in the range of 2–100 μM. Prepared electrode was showed the limit of detection (LOD) of 0.26 μM and R^2 of 0.999. Additionally, the developed sensor s used to determination the BPA in bottled drinking water sold in Turkish markets by using DPV, and obtains satisfactory results. The recovery values of the bottle water samples in the freezer, at room temperature and in the car were calculated as 102.22%, 104.68% and 106.06% respectively.

1. INTRODUCTION

Bisphenol A (4,4'-isopropylidendiphenol or 2,2'-bis (4-hydroxyphenyl) propane) is an important substance used in industry for plastic food packaging, certain coatings and many adhesives [1,2]. Bisphenol A (BPA) as monomer, coatings, antioxidants and lac is widely used for fabrication of phenolic resins, polyacrylates, epoxy resins, polycarbonates, polyethylene teraphatale (PET) and polyvinyl chloride (PVC) [2,3]. Thus, human exposure to bisphenols occurs primarily through the ingestion of canned drink and food, as well as skin absorption and dust inhalation [4]. For this reason personal exposure to BPA is quite high in daily life. Although the daily intake for BPA was determined by the US Environmental Protection Agency (US EPA) and the European Food Safety Authority to be 50 μg/kg body weight in one day [5], Health Canada accepted this amount as 25 μg/kg [1].

BPA is an estrogenic environmental toxin and passes through food plastic baby bottles and food plastic beverage bottles containing BPA [1,3,6,7]. Water temperature and hardness are the important factors affecting the breakaway of BPA from plastic bottles [7]. BPA can mimic the natural hormone, the main cause of which is occupying hormone receptors and preventing hormone system, which an important threat to human health. Because of the harm of BPA to the environment and people health, many researches [8-10] have begun to pay more attention to the adverse effects of using BPA-containing products. In order to

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reduce environmental and biological damages, it is requisite to improve and develop existing detection and measurement techniques of BPA [6].

Conventional measurement methods of BPA in some food and water include gas chromatography (GC) [1,10-13], LC-MS [2,14] fluorimetry [15,16], liquid chromatography (HPLC) [17,18] and flow injection located chemiluminescence [19]. Although the aforementioned methods provide a very accurate precision and accurate definition, some factors, such as costly and long response time restrict their application. On the contrary, electrochemical methods are would rather due to their simple preparation procedure, rapid response, cheap, good selectivity and high sensitivity. Despite the fact that BPA is an electroactive space, it is very difficult to determine directly with bare electrode by conventional electrochemical method due to the weak response of BPA. Several modified voltammetric electrodes have been developed to investigate the electrochemical behavior of BPA. In particular, selective permeable based membranes, molecular imprinting structures or conductive polymeric membranes were used to provide BPA selectivity in these studies [3, 6, 20-26]. There are many studies in the literature to detect a more sensitive BPA concentration. In these studies, carbon nanotubes and some nanoparticles added to the conductive membranes are generally used [27-28]. The electrocatalytic effect of the materials added to the conductive polymeric structures has been utilized [28-31]. However, clustering and homogeneous distribution in these particles is a major problem [32-33]. Another important problems are the increased electrode preparation stages and nanoparticles are very expensive. Despite some satisfactory results, faster, simpler and more sensitive electrochemical methods for BPA measurement need to be investigated and developed.

In present study, a novel, sensitive, simple and cheap electrochemical sensor was improved on p(p-ABA) modified gold electrode (AuE) for the measurement and detect of BPA in water samples. The AuEs were modified by polymerization of the p-amino benzoic acid monomer using a bulk electrolysis method at 0.01 M HCl at -250 mV. Subsequently p (p-ABA) modified AuE was used for electrochemical oxidation of BPA in 0.1 M PBS. The effects of electrolyte type, electrolyte pH and scan rate on the oxidation signal of BPA were investigated. The qualitative and quantitative determination of BPA in water samples was occured by using ideal analytical method DPV. The electrooxidation properties of BPA at the prepared p(p-ABA)/AuE were fully investigated by voltammetric methods. The new electrode was given a very good performance for determining BPA in terms of high sensitivity and good selectivity. Furthermore, BPA content was determined using p (p-ABA) modified gold electrode in water samples in plastic bottles.

2. EXPERIMENTAL

2.1. Reagents and Apparatus

The chemicals used in the study were taken with high purity and used without any purification. . p-ABA, used for the modification was obtained, from Fluka. BPA was purchased from Aldrich. BPA solutions were prepared in ethanol:ultra pure water (1:4) just before running each experiment. The PBS solution was prepared by mixing 125 mM NaCl, 2.7 mM KCl, and 0.1 M Na_2HPO_4 solutions. Aqueous solutions were prepared using ultra-pure water. This ultra pure water was obtained by Millipore brand Elix 20. .

Electropolymerization was conducted by bulk electrolysis mode with a BAS 100 BW model computerized electrochemical analyzer (Bioanalytical Systems, Inc., West Lafeyette, IN, United States) in a three-electrode cell with a gold (MF-2014) working electrode, Ag/AgCl (3 M KCl) reference electrode (CHI111) and a Pt wire auxiliary electrode. The pH of the solution was measured with a HI 2211 model pH/ORP Meter. pH meter was calibrated by using standard buffers (pH=4.00, pH=7.00 and pH=10.00) obtained from Merck.

The Au working electrodes used in the study were cleaned with a diamond polishing kit and an alumina polishing kit. After each cleaning step, the electrodes were washed several times with ultrapure water and sonicated for one to two minutes at an ultrasonic bath. (Branson model 3510).

All glassware and pyrex electrolysis cells were kept in 6 M HNO_3 overnight to remove impurities. All electrochemical measurements were carried out at room conditions.

Conducting polymer modification on the gold electrode was achieved in a three-electrode single-compartment cell.

Modified and prepared electrodes were washed with ultra pure water and then dried at room temperature before using.

3. RESULT AND DISCUSSIONS

3.1. Modification of Electrodes

In the study, Au working electrodes used were mechanically cleaned, polished with diamond and alumina polishing kits and modified with selective polymeric film coating on their surfaces by electropolymerization technique..

Firstly, cyclic voltammogram was obtained of p-ABA in 0.1 M PBS at pH 6 on bare Au electrode. p-ABA was oxidized to free radical at the surface of the electrode, the radicals then combined together rapidly, and the product reacted as follows (Figure 1).

As a result of cyclic voltammetric studies, oxidation peak of p-ABA was observed at potential range -200 and -400 mV. Then conducting polymer modification on the bare Au electrodes were achieved in a three-electrode single-compartment cell containing 1x10^{-3} M p-ABA dissolved in 0.01 M HCl [34]. The p-ABA films were grown on bare gold electrode at -250 mV constant potential with bulk electrolysis technique. Films were grown for 150 s unless stated otherwise. Modified and prepared electrodes were washed several times with ultra-pure water before using. Then they were used for voltammetric experiments in a three-electrode single-compartment cell containing electrolyte solution.

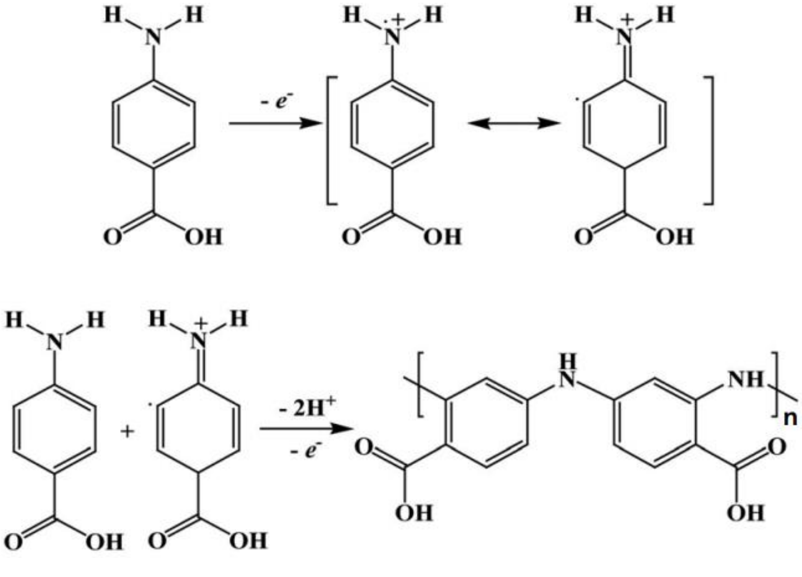

Figure 1. *The electrochemical polimerization mecanizm of p-ABA [33]*

3.2. Detect of the Oxidation Potential of BPA

The oxidation of BPA in aqueous solutions on solid electrode has been showed (Figure 2).

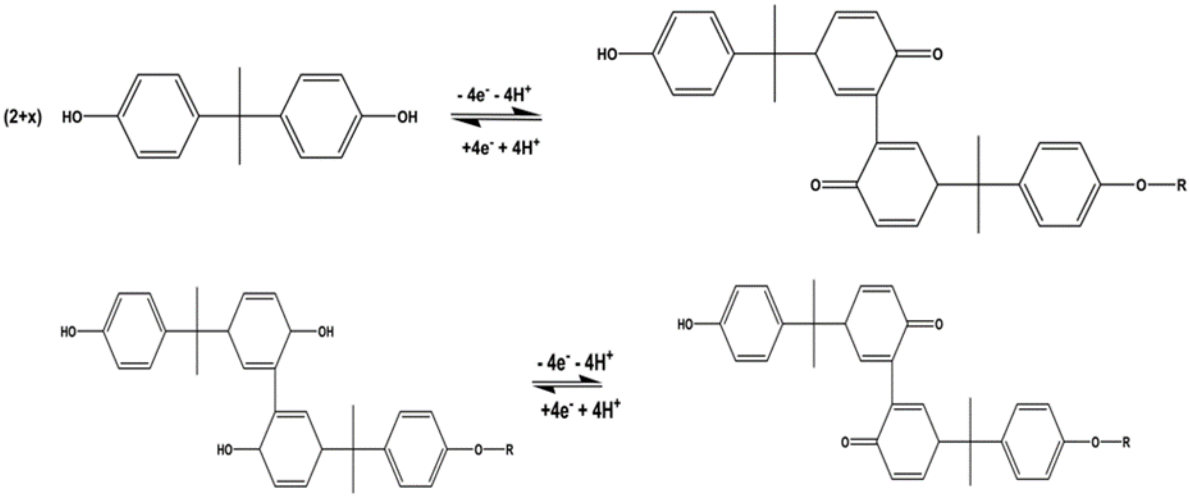

***Figure 2.** Mechanism for BPA electro-oxidation on solid electrode surface [35]*

As show in Figure 3.1, the polymerization of the p-ABA were carried out at about -250 mV potential. In Figure 3.2, it was noted that the cyclic voltammetric responses were obtained in 0.1 M PBS pH = 6 on bare AuE (Figure 3.2a), in 0.1 M PBS pH 6 containing 0.33 mM BPA on bare AuE (Figure 3.2b) and in 0.1 M PBS pH=6 containing 0.33 mM BPA on p(p-ABA) modified AuE (Figure 3.2c). As can be seen from this voltammogram, the oxidation peak of BPA was observed in the potential range of 600-700 mV both on bare AuE and modified AuE.

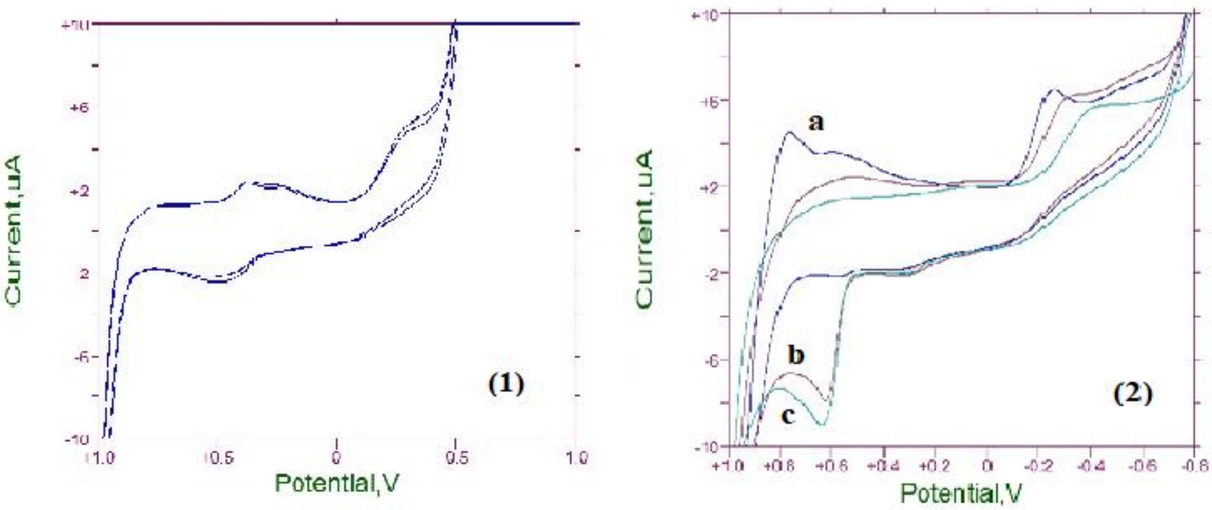

***Figure 3.** CVs of (1) The electropolimerization of 1x10^{-3} M p-ABA in 0.01 M HCl on bare AuE, (2) 0.1 M PBS, pH= 6 (a) 0.33 mM BPA in 0.1 M PBS on bare AuE, pH=6 (b), 0.33 mM BPA in 0.1 M PBS, pH=6 on p(p-ABA) modified AuE (c)*

3.3. Effect of the Electrolyte Type

The effect of the support electrolyte type was examined using the DPV method in various supporting solutions (Na_2SO_4, NaCl, KCl, KNO_3, $NaClO_4$, CH_3COONa, LiCl and PBS) each with a concentration of 0.1 M (Figure 4). The highest peak current for 0.33 mM BPA was obtained for 0.1 M PBS solution (Figure 5).

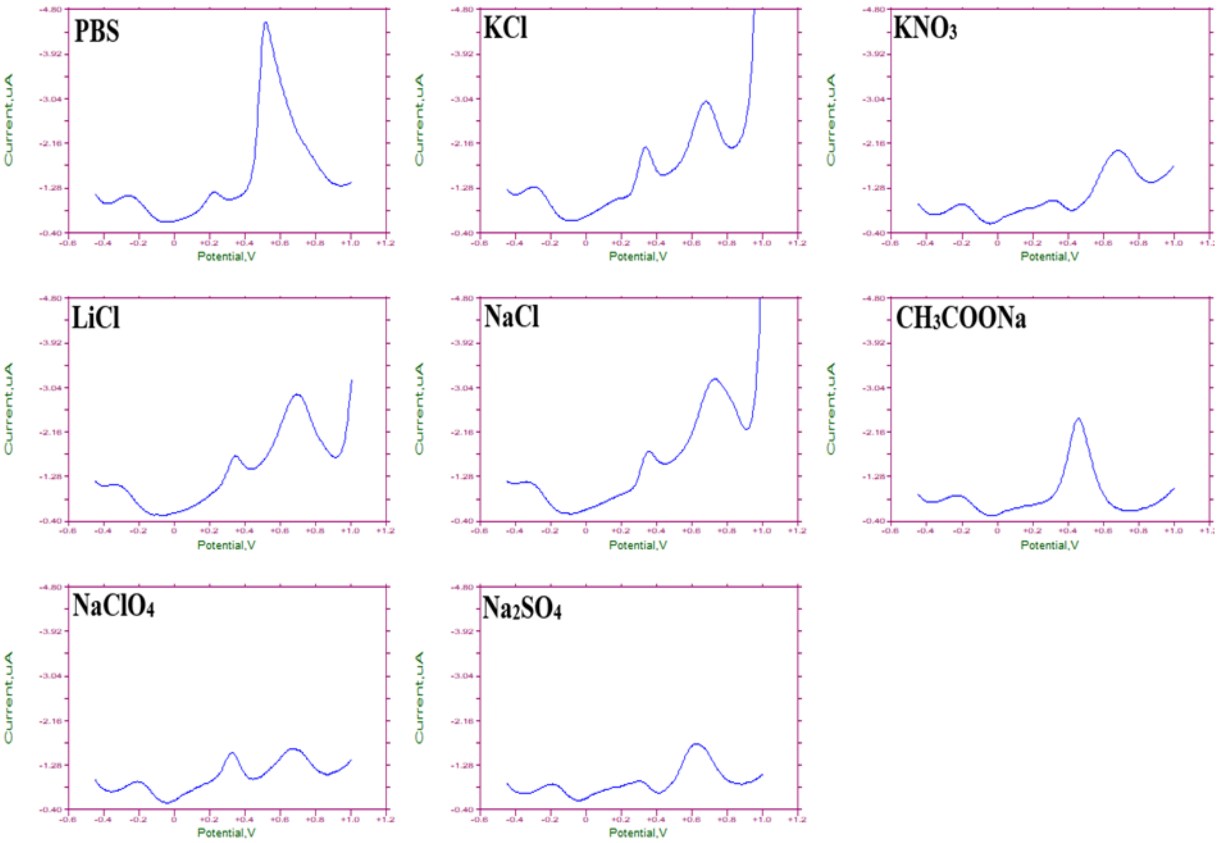

Figure 4. *DPVs of 0.33 mM BPA in 0.1 M different electrolyte types on p(p-ABA) modified AuE*

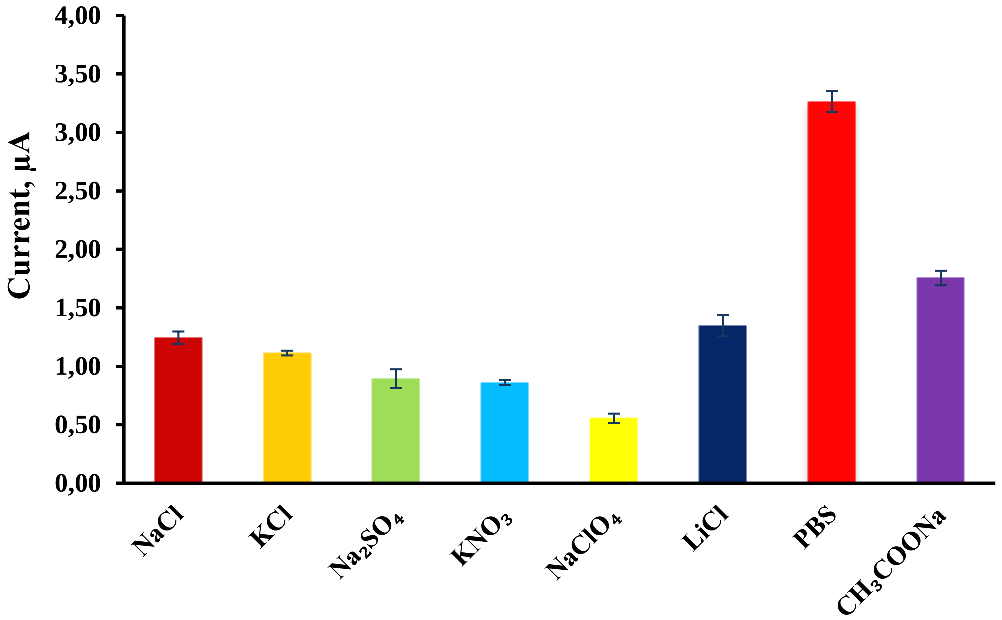

Figure 5. *The DPV peak currents for BPA in different electrolytes on p(p-ABA) modified AuE*

3.4. Effect of Electrolyte pH

The influence of the pH on the voltammetric response of 0.33 mM BPA in 0.1 M PBS electrolyte was investigated in the pH 4, 5, 6, 7 and 8. The maximum oxidation peak current was found to be for 0.1 M PBS at pH 6 (Figure 6a). The influence of pH on oxidation peak potential was illustrated in Figure 6b.

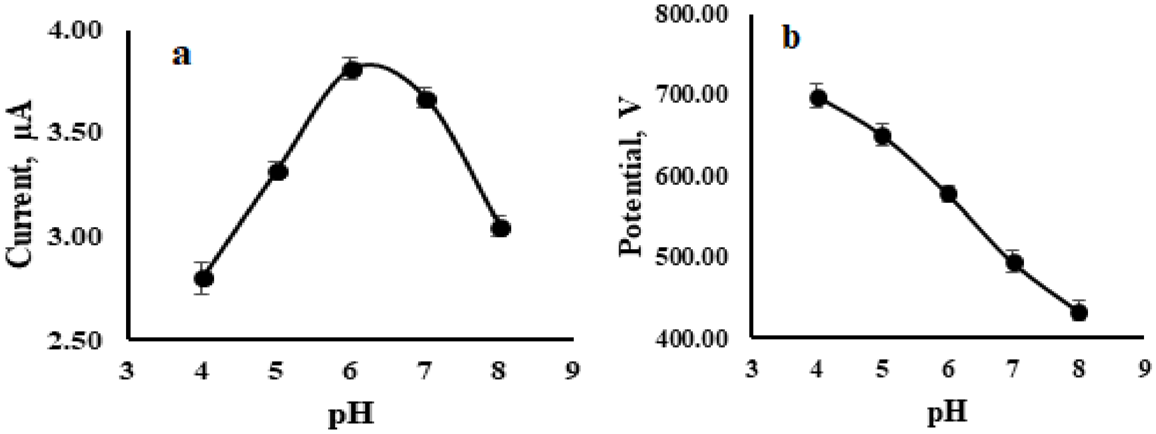

Figure 6. *The relationship electrolyte between pH and oxidation peak current (a), peak potential (b) of 0.33 mM BPA on p(p-ABA) modified AuE in 0.1 M PBS*

3.5. Effect of Scan Rate

The effect of the scan rate on the DPV response for BPA in the range 20-100 mV/s was examined. The maximum response was found to be for 60 mV/s scan rate in 0.1 M PBS at pH 6 (Figure 7).

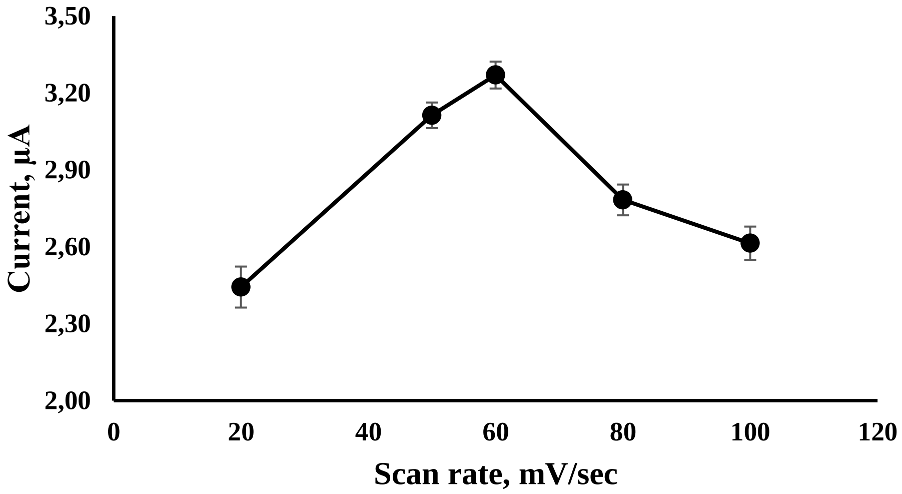

Figure 7. *The relationship between scan rate and oxidation peak current for BPA on p(p-ABA) modified AuE in 0.1 M PBS*

3.6. Effect of Polimerization Time

The relationship between charge and time during electropolymerization with BE was investigated. As shown in Figure 8, it was observed that after from 150 seconds, the charge did not increase due to the electrode reaching saturation.

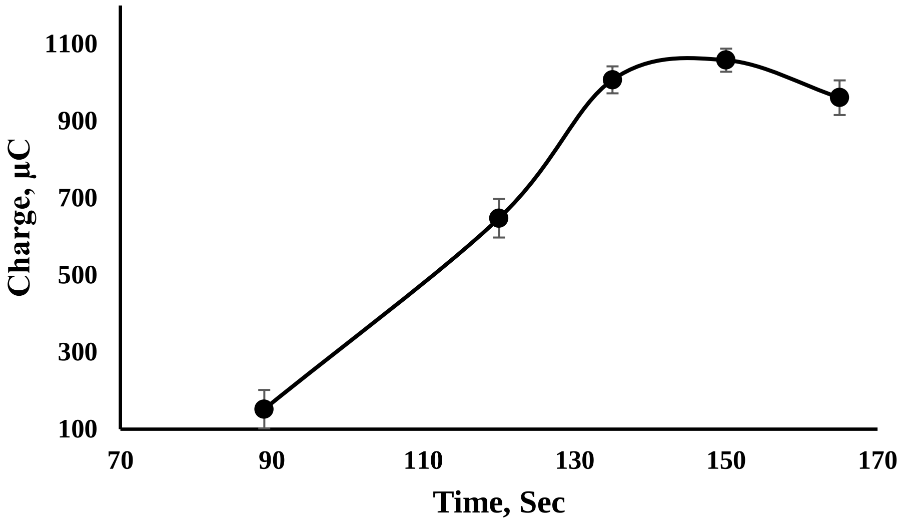

Figure 8. *The relationship between charge and time during electropolymerization with BE on bare AuE in 0.1 M PBS, pH=6*

After the Au electrodes modification with p(p-ABA), it was obtained voltammograms of the BPA on this electrode in 0.1 M PBS at pH 6. As shown in Figure 8 and 9, it was also observed that after from 150 seconds, both the oxidation peak current of 0.33 µM BPA and charge decrease. Therefor it was decided that the best conductive polymeric membrane film was made at 150 s.

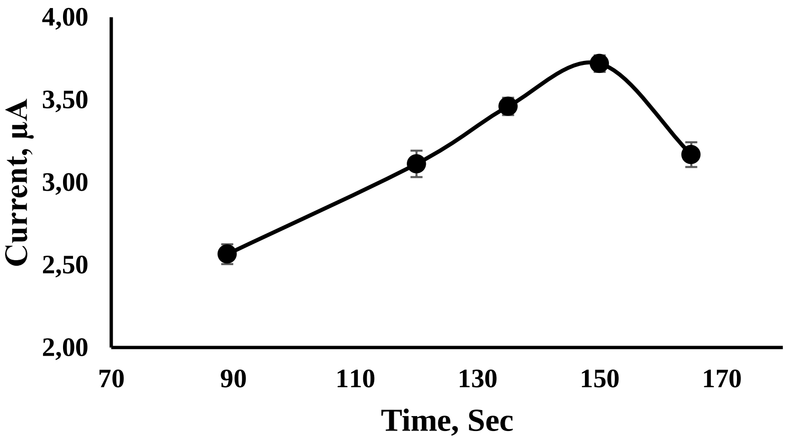

Figure 9. *The relationship between polimerization time and voltammetric response of 0.33 µM BPA on p(p-ABA) modified AuE in 0.1 M PBS*

3.7. Calibration Curves and Detection Limit for the Determination of BPA

In this study, for the quantitative electrochemical detection of BPA, DPV technique is used. DPV responses of 2, 5, 8, 10, 20, 30, 50, 70, 100 µM (Figure 10) and 1.6, 2.4, 3.2, 4.0, 4.8, 5.6, 6.4, 7.2, 8.0, 8.8, 9.6 µM BPA were obtained under optimum conditions that have been appropriated above.

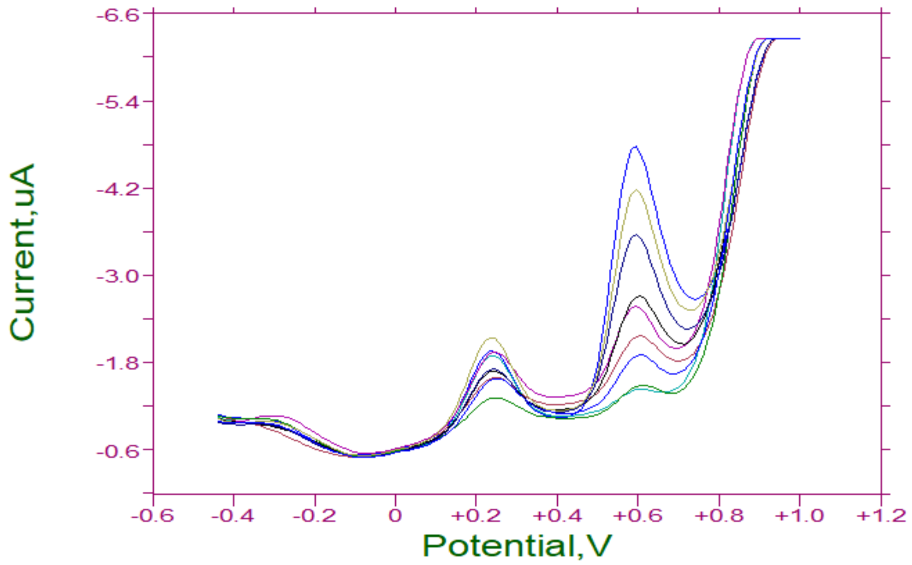

Figure 10. *Effect of the BPA concentration on p(p-ABA) modified AuE in PBS pH=6, scan rate 60 mV/s*

Calibration plots of high concentration (Figure 11a) and low concentration (Figure 11b) of BPA were also obtained from mentioned DPV data. The developed electrochemical sensor has linear response to BPA in the ranges of 2-100 µM and 1.6-9.6 µM. LOD value of prepared modified electrode for BPA measurement was calculated as 0.26 µM. The p(p-ABA) modified AuE showed a very high R^2-value (0.999) according to Figure 11b.

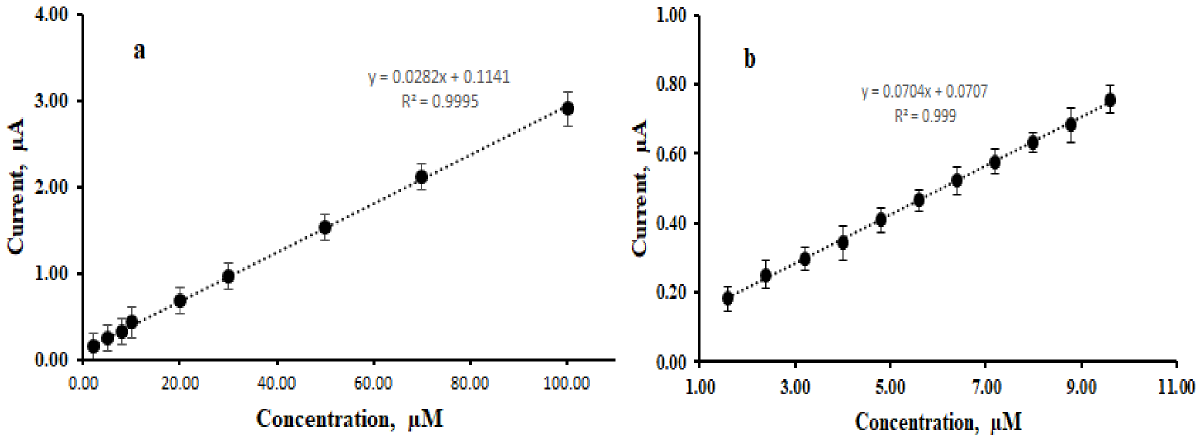

Figure 11. *Calibration plots for BPA determination on p(p-ABA) modified AuE in 0.1 M PBS, scan rate 60 mV/s.* ***a)*** *high concentration* ***b)*** *low concentration*

3.8. Stability of the Proposed Sensor

Figure 12 demonstrated the stability of the p(p-ABA) modified Au electrode. The magnitude and reproducibility of the DPV responses obtained with thirteen measurements showed the stability of the p(p-ABA) electrode for 1.6 µM BPA in 0.1 M PBS.

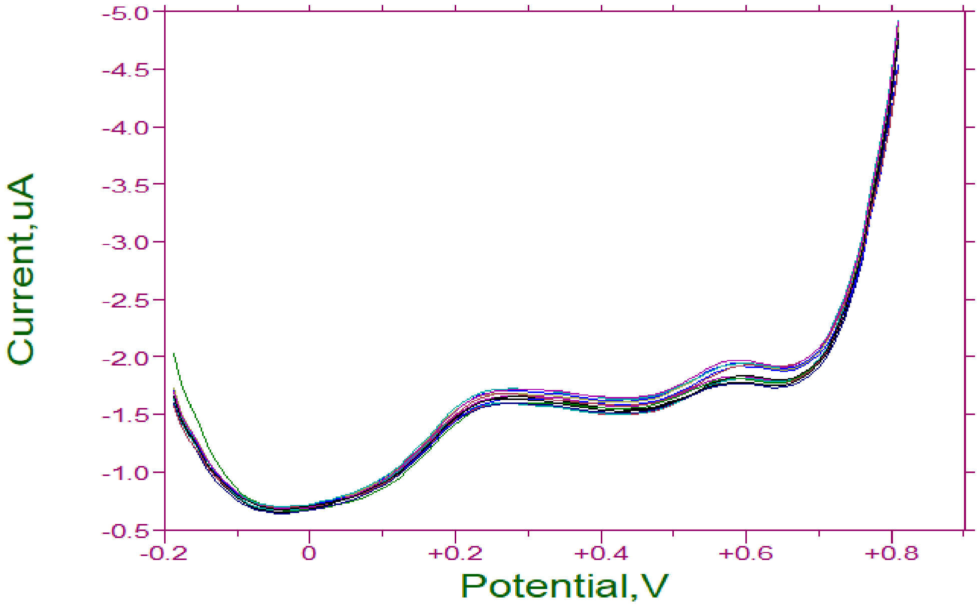

Figure 12. *Reproducibility of the DPV responses of 1.6 µM BPA on p(p-ABA) modified AuE in 0.1 M PBS n=13)*

3.9. Determination of the BPA in Drinking Water in the PET Bottle

Same brand plastic bottled water samples were purchased from a supermarket in Turkey. Water samples were classified in three groups. The first group of these samples were kept at room temperature for twenty days. The other group was kept in the car for twenty day and the last group was also kept in the freezer for twenty days. The standard addition method was applied to determine the content of BPA in these samples. The contents of BPA in real samples and the recovery results are listed in Table 1. pH 6 PBS were prepared both with bottled drinking water and ultra pure water. In the initial detection DPV signals were not observed in PBS solutions prepared in both ways. In this case, BPA standard solution (70 µM of final concentration) were also spiked to the two PBS and measured with DPV method by the sensor. The contents of BPA in real samples and the recovery results are listed in Table 1. The recovery values of the modified electrode prepared were obtained by comparing the calculated concentrations with a calibration curve with the spiked concentration. According to the results in Table 1, prepared Au modified electrodes are suitable for measuring BPA in real water samples.

Table 1. *Determination of BPA spiked bottled drinking water by the proposed method, (n=3)*

Samples	Original	Added Concentration, µM	Detected Concentration, µM	RSD%	Recoveries%
Drinking water kept at room temperature	nd	70 µM	71.63	2.65	104.68
Drinking water kept in the car	nd	70 µM	74.25	3.84	106.06
Drinking water kept in the freezer	nd	70 µM	71.08	4.50	102.22

4. Conclusion

Due to some toxic effects, analysis of BPA in water, food, cosmetics and daily used materials is very important. However, in BPA analysis, time consuming and costly techniques are often used. New techniques with practical, sensitive and fast must be developed to make these analyzes. This situation is especially a requirement in eco-toxicological studies and drinking water. Drinking water pollution is very important for human health and environment. Therefore, there is a need to develop sensitive methods for the determination of toxic compounds such as BPA.

In this study, AuE was electrochemically modified with p(p-ABA) for the selective, practical, fast and sensitive measurement of BPA in bottled water samples. Prepared modified AuE have short response time, good sensivity and high selectivity. As a result of this study, an electrochemical sensor with higher sensitivity and lower detection limit was developed compare with some published studies in literature (Table 2).

Table 2. *Comparison of some characteristics of the different electrodes for the determination of BPA*

Electrode	Mode	Linear Range (*m*ol/L)	LOD (mol/L)	Electrolyte pH	R^2	Ref.
MWCNTs/PCV/GCE	LSV	$5x\ 10^{-8}$- $1x\ 10^{-4}$	$1x10^{-8}$	PBS 6.5	0.998	[6]
SH-β-CD/NPGL/GE	SWV	$3x10^{-7}$- $1x10^{-4}$	$6x10^{-8}$	PBS 7	0.9928	[35]
AuNp@MOF/CPE	DPV	$2.0x10^{-4}$- $1.0x\ 10^{-3}$	$3.7x10^{-5}$	PBS 7	0.998	[36]
P-*p*ABA/AuE	DPV	$1.6x10^{-6}$- $1x\ 10^{-4}$	$2.6\ x10^{-7}$	PBS 6	0.999	This work

SH-β-CD/NPGL/GE: thiolated beta-cyclodextrin/nanoporous gold leaf/gold electrode
MWCNTs/PCV/GCE: multi-walled carbon nanotubes/poly crystal violet/glassy carbon electrode
AuNp@MOF/CPE: gold nanoparticles@metal organic framework/carbon paste electrode

When Table 2 is examined, it can be stated that analytical parameters of the developed sensor, which involves no adsorption, deposition or incubation of the analyte on the electrode, is compatible with the data reported in literature. Compared to the literature, proposed sensor has low detection limit and wide linear range. This sensor has many important advantages such as rapid response time, low detection limit, low cost and high reproducibility for determination of BPA.

Finally, this developed method for the determination of BPA in bottled drinking water can be applied to different types of water such as seawater and lake water.

CONFLICTS OF INTEREST

No conflict of interest was declared by the author.

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