Surface Functionalized Prussian Blue-coated Nanostructured Nickel Oxide as a New Biosensor Platform for Catechol Detection

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An amperometric biosensor has been developed for highly efficient and sensitive detection of catechol using Prussian blue (PB)-coated nickel oxide (NiO) nanoparticles (NPs) as a matrix for the immobilization of tyrosinase enzyme. The NiO NPs were synthesized by sol-gel method using sodium dodecyl sulphate as anionic surfactant and the surface of the synthesized NiO NPs was modified with PB to enhance electrocatalytic activity and to prevent surface aggregation. After confirmation of successful synthesis of the PB-NiO NPs from transmission electron microscopy (TEM) and energy dispersive X-ray spectroscopic (EDS) studies, the prepared NPs were deposited onto a working electrode of a commercially available screen printed carbon electrode (SPCE) substrate. The tyrosinase enzyme was covalently immobilized onto the PB-NiO deposited SPCE for selective detection and estimation of catechol through electrochemical methods via cyclic voltammetry (CV) and chronoamperometric techniques. The functionalization of tyrosinase on the electrode surface was verified by atomic force microscopy (AFM) and scanning electron microscopic (SEM) techniques and the electrochemical response studies of the proposed biosensor showed high sensitivity of 0.954 μA/μM for catechol in a wide linear range (1 – 50 μM) with low detection limit (LOD) of 0.087 μM. The developed sensor also exhibited a fast response time of 27 s and decent selectivity for catechol detection.

Keywords Electrochemical biosensor, amperometry, catechol, tyrosinase, NiO nanoparticles, Prussian blue

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Introduction

The detection of phenolic compounds is a major concern nowadays because of their toxic effects on human health and the environment and the significance of accurate detection of such type of compounds increases rapidly due to their growing use in consumer products. Among these, catechol or 1,2-dihydroxybenzene is a potential hazardous phenolic compound that is widely used in cosmetics, textiles, pharmaceuticals and tanning industries.¹ The presence of catechol in the industrial effluent and discarded consumer products causes severe environmental pollution.² Moreover, catechol is regularly used during the preparation of pesticides, photographic developer, plastics, paper, dyes and textile products and releases directly into the ground and surface water.³ As a result, there is a high chance of catechol exposure to human health that can cause serious adverse effects by damaging the functions of the kidney, liver, lungs, gastrointestinal and genitourinary track.⁴⁵ Due to such potential risk on human health and the environment, quick and precise on-site detection of catechol is urgently required. The most frequently used analytical techniques for catechol detection are based on spectrophotometry,⁶ chromatography,⁷ electrochemiluminescence,⁸ and capillary electrophoresis⁹ methods. Unfortunately, most of these procedures are time consuming, highly sophisticated, costly, cumbersome and difficult to implement in handheld format for on-site detection. In this context, biosensors can provide a better solution for on-site, real time and cost-effective determination of catechol due to having advantageous features such as simple operation, ease in miniaturization, minute reagent consumption and rapid detection, etc. Besides this, the presence of specific biological recognition elements in biosensors also helps in highly selective and sensitive detection of catechol. During an electrochemical process, catechol can be selectively oxidized to benzoquinone in the presence of the tyrosinase enzyme (EC 1.14.18.1) and at lower potential, benzoquinone is further reduced to form catechol. Therefore, an amperometry-based electrochemical biosensing can be an efficient way for catechol detection because of the ease of integrating the highly selective enzymatic detection process and due to having electrochemical activity of the catechol itself. However, the performance of a biosensor greatly depends on the proper immobilization of the enzymes, especially on the matrix, and the method used for the immobilization. The nanoparticles provide an effective platform for the enzyme immobilization owing to higher surface area, high electrocatalytic activity, efficient charge transfer abilities and high surface energy.¹⁰ Among the various nanoparticles, nickel oxide nanoparticles offer an excellent platform for tyrosinase immobilization due to easy mobility of the electrons, low cost, high chemical stability, biocompatibility and easy availability of the raw materials.¹¹ Moreover, the efficiency of a NiO NPs-based amperometric
biosensor can be further enhanced with the introduction of a mediator that can accelerate electron mobility from the biological detection molecules to the electrode surface. In this context, the surface of NiO NPs can be modified with the conventional electron mediator Prussian blue. Prussian blue or ferric hexacyanoferrate is a widely known electron mediator for amperometric biosensors due to having high electrocatalysis and excellent electrochemical behaviors. Moreover, the surface modification of NiO nanoparticles with Prussian blue helps in the restriction of surface aggregation and oxidation, resulting in higher loading of the enzymes on the electrode surface. Apart from that, the stable and firm immobilization can increase activity of the enzyme. In this regard, the covalent immobilization technique is highly effective because of strong binding and minimal leakage of the enzymes from the electrode surface, and the covalently attached enzyme can be sustained in the harsh environment of different pH and ionic strength.

Hence, for highly selective and sensitive detection of the environmental pollutant catechol, an amperometric biosensor has been developed using Prussian blue (PB)-modified nickel oxide (NiO) nanoparticles (NPs) as immobilization matrix and tyrosinase enzyme as biological detection element. In the following sections, the experimental techniques related to the synthesis of PB-NiO NPs and the covalent immobilization of tyrosinase onto PB-NiO modified screen printed carbon electrodes (SPCE) are described. After verification of successful synthesis of the nanoparticles and the formation of electrodes through different characterization techniques, the fabricated tyrosinase/PB-NiO/SPCE was used for electrochemical detection and estimation of catechol. The response parameters of the developed tyrosinase/PB-NiO/SPCE biosensor have been calculated and compared with previously reported literature.

**Experimental**

**Reagents and chemicals**

The reagents and chemicals 1,2-dihydroxybenzene (C₆H₆O₂), tyrosinase (EC 1.14.18.1 from Agaricus bisporus) with activity of 1000 U mg⁻¹ of solid), N-ethyl-N′-(3-dimethylaninopropyl) carbodiimide hydrochloride (EDC), N-hydroxysuccinimide (NHS), hexamethylenediamine [NH₂(CH₂)₆NH₂], citric acid (C₆H₈O₇) and iron(II) chloride (FeCl₂) were obtained from Sigma-Aldrich (India). Nickel nitrate hexahydrate [Ni(NO₃)₂·6H₂O], sodium dodecyl sulfate (C₁₂H₂₅O₄SNa) and potassium ferricyanide (K₃[Fe(CN)₆]) were purchased from Alfa Aesar (India). All other reagents were of analytical grade and used without any further purification. The as-prepared tyrosinase/PB-NiO/SPCE electrodes were used for amperometric detection and estimation of catechol. Before enzyme immobilization, carboxylic groups were activated on the PB-NiO/SPCE electrode and immobilized owing to formation of the amide bond between the activated carboxylic groups with the amine group on the electrode surface. After washing and removal of unbound particles, the working electrode surface was functionalized with an aqueous solution of NaOH as anionic surfactant. After proper dispersion, the resultant solution was added dropwise to the NiO NPs with K₃[Fe(CN)₆] solution under continuous stirring at 1500 rpm for 3 h prior to conducting the experiment.

**Apparatus**

The as-synthesized NiO and PB-NiO NPs were characterized using transmission electron microscopy and energy-dispersive X-ray spectroscopic (TEM and EDS, JEOL JEM-200FS) studies. The surface morphology of the developed electrodes was investigated by atomic force microscopy (AFM, Bruker dimension icon with Scan Asyst) and scanning electron microscopic (SEM, JEOL JSM-6010LA) techniques. The electrochemical measurements were carried out using a potentiostat (DropSens μStat400) with the fabricated tyrosinase/PB-NiO/SPCE electrodes in phosphate buffer saline (PBS, 50 mM, pH 6.5, 0.9% NaCl) as electrolyte.

**Synthesis of PB-coated NiO NPs**

The synthesis of NiO NPs was conducted by a wet chemical sol-gel method according to a previous report. Briefly, 20 mM of [Ni(NO₃)₂·6H₂O] and 40 mM of NaOH were separately dissolved in deionized water (DW) and 10 mM of sodium dodecyl sulfate (SDS) was added to the [Ni(NO₃)₂·6H₂O] solution as anionic surfactant. After proper dispersion, an aqueous solution of NaOH was added dropwise to the Ni(NO₃)₂·6H₂O solution to obtain a pH level of up to 11.5 - 12 and kept under continuous stirring for another 2 h at 25°C. During the process, a light green precipitate of Ni(OH)₂ was produced and separated by centrifugation. The precipitate was then washed with DW and ethanol 3 - 4 times each and then dried at 70°C for 24 h, followed by calcination at 300°C for 3 h to form NiO NPs.

Surface modification of the prepared NiO NPs with PB was carried out using a chemical method. For that, 1 mM K₃[Fe(CN)₆] and 0.5 mM citric acid were added to the homogenous aqueous dispersion of NiO NPs (0.5 mg mL⁻¹) and kept under stirring for 30 min for proper mixing. Separately, 1 mM FeCl₃ and 0.5 mM citric acid were dissolved in DW and the resultant solution was added dropwise to the previous NiO NPs with K₃[Fe(CN)₆] solution under continuous stirring at 25°C. The formation of PB-modified NiO NPs were indicated after the immediate appearance of a blue color in the solution.

**Bio-electrode preparation**

In the present work, a commercially available screen printed carbon electrode (SPCE) with graphite working electrode, Ag/AgCl reference electrode and graphite counter electrode was used as a substrate to develop the catechol biosensor. First, the working electrode of the SPCE was functionalized with an amine group using hexamethylenediamine solution (10% w/v in phosphate buffer, pH 7.5) and after drying and washing of unbound particles, 10 μL of well dispersed PB-NiO NPs solution was deposited. The PB-NiO NPs were covalently attached on the working electrode surface due to having citrate ions-induced carboxylic groups, which form strong covalent bonds with the amine group on the electrode surface. After washing and removal of unbound particles, the working electrodes were functionalized with tyrosinase enzyme molecules that can selectively detect catechol. Before enzyme immobilization, carboxylic groups were activated on the PB-NiO/SPCE surface by using freshly prepared solutions of 0.4 M N-ethyl-N’-(3-dimethylaninopropyl) carbodiimide hydrochloride (EDC) as a coupling agent and 0.1 M N-hydroxysuccinimide (NHS) as an activator. Subsequently, 10 μL of freshly prepared tyrosinase enzyme solution (1 mg mL⁻¹ in PBS, pH 6.5) was uniformly spread on the carboxylic group functionalized working electrode surface. The tyrosinase was covalently immobilized owing to formation of the amide bond between the activated carboxylic groups on the PB-NiO/SPCE electrode and the amine terminal of the enzymes. After characterization, the as-prepared tyrosinase/PB-NiO/SPCE electrodes were used for the detection and estimation of catechol by electrochemical method.

**Biosensor studies**

Cyclic voltammetry (CV) measurements of the bare SPCE,
PB-NiO/SPCE and tyrosinase/PB-NiO/SPCE were recorded in PBS (50 mM, pH 6.5, 0.9% NaCl) at 50 mV/s scan rate in the potential range of –0.5 to +1 V. The oxidation potential of catechol for the developed tyrosinase/PB-NiO/SPCE biosensor was determined with 100 μM catechol solution via CV studies. The response of the tyrosinase/PB-NiO/SPCE biosensor was denoted in the presence of 0.1 – 250 μM catechol solution by chronoamperometry technique using the oxidation potential of catechol. The sensing parameters of catechol for the fabricated tyrosinase/PB-NiO/SPCE biosensor were evaluated from the calibration curve of response studies. The selectivity studies of the developed biosensor were performed in the presence of ascorbic acid (100 μM), uric acid (100 μM) and dopamine (100 μM), while maintaining a constant catechol (100 μM) concentration.

Results and Discussion

Material characterization of the synthesized nanoparticles

The structural morphology of the synthesized NiO and PB-NiO NPs were investigated through transmission electron microscopy (TEM) and energy dispersive X-ray spectroscopic (EDS) studies. Figure 1(a) reveals the TEM image of NiO NPs, showing the presence of randomly oriented and semi-spherical nanoparticles with an average particle size of 10 nm. The existence of the clearly visible lattice fringes indicates the formation of highly crystalline nanostructures. However, after surface modification with PB, the NiO NPs show (Fig. 1(b)) a little enhancement in particle diameter, ranging from 15 to 17 nm. The particles also display the same semi-spherical shape with less visible lattice fringes and a dark layer at the nanoparticle surface arises due to PB. The synthesis of NiO NPs followed by surface modification with PB were further verified from EDS studies. The elemental analysis from EDS studies shows the consistent peaks for Ni and O for NiO NPs (Fig. 2(a)). After modification with PB, the additional peaks of Fe together with Ni and O are obtained for PB-NiO NPs (Fig. 2(b)), confirming the presence of PB on the NiO NPs surface. In both EDS measurements, the peaks of C and Cu are detected from the substrate and the absence of any further peaks indicates purity of the synthesized samples.

Surface morphological studies of the fabricated electrodes

The formation of a thin film of PB-NiO NPs on the SPCE working electrode surface and further immobilization of the tyrosinase enzyme were confirmed from atomic force microscopy (AFM) and scanning electron microscopic (SEM) studies. The AFM micrograph of the bare SPCE taken in 5 × 5 μm scan area (Fig. 3(a)) shows the rough surface of the working electrode with porous morphology due to the presence of graphite microparticles. After deposition of PB-NiO NPs on
the SPCE surface, the scan area of the PB-NiO/SPCE has been compacted to $1 \times 1 \mu m$ to closely show the morphology of the surface due to the presence of PB-NiO nanoparticles (Fig. 3(b)). Here, uniform distribution of the NPs were observed with nanoporous granular morphology. However, after enzyme immobilization (Fig. 3(c)), additional elevated structures were detected on the surface of the PB-NiO/SPCE due to the presence of tyrosinase enzymes. This AFM micrograph of the tyrosinase/ PB-NiO/SPCE has been displayed with $1 \times 1 \mu m$ scan area to minutely show the presence of the enzyme, forming the additional elevated structures on the PB-NiO/SPCE surface. The nanoporous granular morphology of the PB-NiO NPs on the SPCE surface perhaps facilitates the higher enzyme loading on the electrodes.

The surface morphologies of the SPCE, PB-NiO/SPCE and tyrosinase/PB-NiO/SPCE were further examined with SEM studies. Figure 4(a) shows the SEM image of the blank SPCE ($5 \mu m$ scale), indicating the presence of graphite microparticles with rough and mesoporous morphology. After deposition of PB-NiO NPs on the blank SPCE, the SEM image of the PB-NiO/SPCE (Fig. 4(b)) has been zoomed out to $10 \mu m$ scale to confirm the existence of nanoparticles with rough and nanoporous morphology. Finally, after enzyme immobilization, the SEM image of the tyrosinase/PB-NiO/SPCE (Fig. 4(c)) has been further zoomed out to $50 \mu m$ scale, which shows the presence of smooth and globular morphology of the macromolecular structure of the enzymes and confirms uniform enzyme immobilization over the working electrode surface of the developed electrode.

**Electrochemical characterization**

The cyclic voltammogram of the blank SPCE (Fig. 5, curve (a)) shows no specific peak due to the absence of any redox reaction in the mediator free electrolyte solution. However, the cyclic voltammogram of the PB-NiO/SPCE (Fig. 5, curve (b)) shows particular redox peaks due to a reversible redox reaction between Fe(II) and Fe(III) ions of Prussian blue, and the NiO NPs facilitate direct electron transfer due to having high electrocatalytic activity and higher surface area. After immobilization of the enzyme, the cyclic voltammogram of tyrosinase/PB-NiO/SPCE (Fig. 5, curve (c)) reveals no particular change in redox peaks except a slight shift to the higher potential. This may be due to the conjugation of the macromolecular structure of the enzyme, and the insulating nature of biomolecules perhaps hinders electron migration from the electrode surface to the surrounding electrolyte. Though after addition of 100 $\mu M$ catechol into the electrochemical cell, the cyclic voltammogram for the blank SPCE (Fig. 5, curve (d)) shows no distinct detection peaks for catechol owing to less conductive activities of the bulk graphite surface. However, in the presence of 100 $\mu M$ catechol solution, the cyclic voltammogram of the PB-NiO/SPCE (Fig. 5, curve (e)) displays
well-defined oxidation and reduction peaks for catechol and the high electrocatalytic activity of PB-NiO NPs helps in improved electron movement to the substrate. The cyclic voltammogram of the tyrosinase/PB-NiO/SPCE with 100 μM catechol solution (Fig. 5, curve (f)) also demonstrates definite redox peaks for catechol. The enzyme tyrosinase can oxidize the substrate catechol into benzoquinone and at lower potential benzoquinone can be reduced to the corresponding catechol again. It has been observed that the redox mechanism of catechol involves a quasi-reversible electron transfer process. The first redox couple was obtained for redox reactions of catechol, where catechol was oxidized at 0.45 V to benzoquinone and the benzoquinone again reduced to form catechol at 0.018 V. But some of the benzoquinone was transformed into the more easily oxidizable intermediate product leucochrome. The second redox couple at lower potentials (0.033 and –0.135 V) was observed due to the oxidation and reduction of the intermediate products. However the oxidation peak for catechol has been detected at 0.45 V for the developed tyrosinase/PB-NiO/SPCE and the peak current intensifies rapidly owing to fast electron transfer from active sites of the enzyme to the electrode, while the presence of PB-NiO NPs accelerates electron migration.

**Amperometric response studies of the developed biosensor**

Response studies of the developed tyrosinase/PB-NiO/SPCE biosensor for catechol were conducted using chronoamperometry with increasing concentrations of catechol (0 – 250 μM) in PBS (50 mM, pH 6.5, 0.9% NaCl). It has been observed (Fig. 5, curve (d)) that catechol is oxidized at 0.45 V with the fabricated tyrosinase/PB-NiO/SPCE, thus the detection potential for catechol in chronoamperometric studies was fixed to 0.45 V. The response time of the developed biosensor was determined with the highest concentration of catechol solution (250 μM) through chronoamperometry measurement. A response of 90% was acquired within 27 s, indicating this as the response time of the developed biosensor. Hence, in the successive response studies of the tyrosinase/PB-NiO/SPCE for catechol, the amperometric current responses were denoted at 27 s. As shown in Fig. 6(a), the amperometric current responses become enhanced with increasing concentrations of catechol. This can be attributed to the transfer of a large volume of electrons with higher concentrations of substrate during enzymatic reaction. The PB-NiO NPs act as a good acceptor of the electrons for transfer to the electrode via Fe(I)/Fe(II) redox couple, resulting in enhanced electrochemical response. From the calibration curve of the response studies (Fig. 6(b)), it can be observed that the tyrosinase/PB-NiO/SPCE biosensor exhibits linearity in a range of 1 to 50 μM with high sensitivity of 0.954 μA/μM for catechol detection. The low detection limit (LOD) and standard deviation have been determined as 0.087 and 0.089 μA, respectively, with a linear regression coefficient of 0.965. The following equation has been obtained for the calibration curve in the linear range (Eq. (1)).

\[
\delta[i(\mu A)] = 0.954[(\mu A)(\mu M)^{-1}] \times \text{concentration} [\mu M] \tag{1}
\]

The calibration curve of the entire concentration range (0.1 – 250 μM) for catechol has been fitted with the Boltzmann equation (Eq. (2)) with regression coefficient of 0.901 and Chi²/DoF value of 28.845.

\[
\delta[i(\mu A)] = A_2 + \frac{(A_1 - A_2)}{\left[1 + \exp((\text{concentration} [\mu M] - X_0)/dx)\right]}, \tag{2}
\]

where \(A_1 = -5304.43, A_2 = 270.89, X_0 = -525.09\) and \(dx = 176.17\). During response studies of the tyrosinase/PB-NiO/SPCE (Fig. 6(a)), a broad hump with a very small amplitude has been observed at 80 s in the spectrum of 250 μM catechol solution. This can be attributed to the sensor drift. Since it appeared after saturation (around 40 s), it indicates there was no certain effect in sensor measurements. Similarly, the negligible sensor drift at
110 s for 100 μM catechol solution has no definite impact in sensor studies as it appeared long after saturation of sensor response. Nevertheless, a comparative table containing catechol sensing parameters of the developed biosensor along with other reported literature is given in Table 1. As compared to other reported results, such as by Singh et al.16 and Kim et al.17 sensitivity of the fabricated biosensor increases several fold with considerably decreased low detection limit for catechol in a sufficiently comparable detection range.

Selectivity studies of the developed tyrosinase/PB-NiO/SPCE for catechol

The selectivity of the developed tyrosinase/PB-NiO/SPCE biosensor for catechol over ascorbic acid (AA), uric acid (UA) and dopamine (DA) was analyzed in PBS (50 mM, pH 6.5, 0.9% NaCl) by chronoamperometry technique. At first, the response of the biosensor was observed for blank, AA (100 μM), UA (100 μM), DA (100 μM) and catechol (100 μM) individually. The results show the usual current response of the control sample (100 μM catechol solution) and marginal increase in current response for AA and UA solutions, while the DA solution reveals a significant increase in the current response. This can be attributed to the closer oxidation potentials of catechol and dopamine and the use of the enzyme tyrosinase, which has common selectivity for both the substrates. Apart from that, an interference study was executed for further investigation of the selectivity of the developed biosensor for catechol in the presence of AA, UA and DA. The measurements were taken using the chronoamperometry technique with the solution containing equal amounts (1:1) of catechol (100 μM) and interferents (100 μM each), such as AA, UA and DA. The changes in amperometric current response for catechol in the presence of interfering agents were measured with respect to the control sample (100 μM catechol) and presented in Table 2. As shown in Table 2, no major changes in amperometric current response were observed in the presence of interferents during detection of catechol with the fabricated tyrosinase/PB-NiO/SPCE biosensor. This may be due to the execution of catechol detection at a fixed potential of 0.45 V at which oxidation of catechol took place for the developed tyrosinase/PB-NiO/SPCE, while the use of the tyrosinase enzyme made the detection procedure highly selective and specific. However, DA can be a moderate interferent in this detection method.

Conclusions

In conclusion, we have successfully established a highly sensitive and selective detection method for the environmental pollutant catechol using PB-modified NiO NPs and tyrosinase enzyme conjugates. The developed biosensor exhibits high sensitivity (0.954 μA/μM) in a linear range of detection 1 - 50 μM and the measurement can be completed within 27 s. Moreover, the sensor shows a very low detection limit of 0.087 μM, a broad detection range of 0.1 to 250 μM, and selective detection of catechol in the presence of ascorbic acid, uric acid and dopamine. The significant improvement in sensing parameters exhibited by the developed sensor has been made possible due to the stability and high electrocatalytic activity provided by the PB-NiO NPs immobilization matrix, and the enzyme tyrosinase helps in selective and specific detection of catechol. Furthermore, the proposed sensing methodology features a rapid, low-cost and easy-to-fabricate process that could be used efficiently in real time and for on-site detection of other phenolic pollutants in soil and water.

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Table 1 Comparison of sensing parameters of the fabricated tyrosinase/PB-NiO/SPCE for catechol along with those reported in literature

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<tbody>
<tr>
<td>Tyrosinase/agarose-guar gum composite/GCE</td>
<td>Differential pulse voltammetry</td>
<td>60 - 800</td>
<td>—</td>
<td>6</td>
<td>14</td>
</tr>
<tr>
<td>Tyrosinase/polyethylene/palladium electrode</td>
<td>Potentiometry</td>
<td>1 - 16</td>
<td>10 mV/μM</td>
<td>1</td>
<td>15</td>
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<tr>
<td>Tyrosinase/sol-gel Au NPs/IrOx</td>
<td>Amperometry</td>
<td>1 - 6</td>
<td>0.01 μA/μM</td>
<td>3</td>
<td>16</td>
</tr>
<tr>
<td>Tyrosinase/p-type Si matrix</td>
<td>Conductometry</td>
<td>50 - 100</td>
<td>—</td>
<td>50</td>
<td>2</td>
</tr>
<tr>
<td>Tyrosinase/sol-gel silicate/nafion/GCE</td>
<td>Amperometry</td>
<td>5 - 100</td>
<td>0.2 μA/μM</td>
<td>0.35</td>
<td>17</td>
</tr>
<tr>
<td>Tyrosinase/PB-NiO/SPCE</td>
<td>Amperometry</td>
<td>1 - 50</td>
<td>0.954 μA/μM</td>
<td>0.087</td>
<td>Present work</td>
</tr>
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Table 2 Selectivity of the developed tyrosinase/PB-NiO/SPCE biosensor for catechol over ascorbic acid (AA), uric acid (UA) and dopamine (DA)

<table>
<thead>
<tr>
<th>Sample</th>
<th>Value of amperometric current response/μA</th>
<th>Standard variation/±SD</th>
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<tbody>
<tr>
<td>Blank</td>
<td>1.197</td>
<td>0.084</td>
</tr>
<tr>
<td>AA (100 μM)</td>
<td>7.067</td>
<td>0.464</td>
</tr>
<tr>
<td>UA (100 μM)</td>
<td>5.838</td>
<td>0.383</td>
</tr>
<tr>
<td>DA (100 μM)</td>
<td>48.404</td>
<td>3.095</td>
</tr>
<tr>
<td>Catechol (100 μM)</td>
<td>118.224</td>
<td>8.254</td>
</tr>
<tr>
<td>Catechol (100 μM) + AA (100 μM)</td>
<td>109.382</td>
<td>7.598</td>
</tr>
<tr>
<td>Catechol (100 μM) + UA (100 μM)</td>
<td>101.868</td>
<td>7.198</td>
</tr>
<tr>
<td>Catechol (100 μM) + DA (100 μM)</td>
<td>140.608</td>
<td>9.405</td>
</tr>
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References