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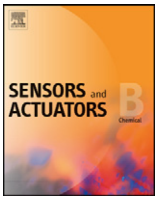

Amperometric sensing of nitrite based on electroactive ferricyanide–poly(diallyldimethylammonium)–alginate composite film

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ABSTRACT

Poly(diallyldimethylammonium chloride) (PDDA) and sodium alginate (AlgNa) were mixed to obtain a water-insoluble interpenetrating polymer composite via electrostatic interaction and then cast on an Au electrode surface, followed by robust immobilization of $K_3Fe(CN)_6$ to yield a $K_3Fe(CN)_6$–PDDA–Alg/Au electrode of stable electroactivity. The interaction processes and electrode modification are studied by visual inspection and electrochemical methods. Under optimized conditions, the $K_3Fe(CN)_6$–PDDA–Alg/Au electrode can efficiently electrocatalyze the reduction of nitrite in acidic aqueous solution and give a linear nitrite-detection range of 5.0 μM to 9.7 mM with a sensitivity of 83.7 $\mu A\,cm^{-2}\,mM^{-1}$, which is amongst the best results reported for nitrite sensing. The electrode was also used for nitrite electroanalysis in real sample matrices with satisfactory results.

1. Introduction

The ferri-/ferrocyanide ($[Fe(CN)_6]^{3-/4-}$) redox couple with excellent electrochemical properties has been so widely used in both fundamental and applied electrochemistry [1–3]. However, $[Fe(CN)_6]^{3-/4-}$ is highly water-soluble, which limits its uses as a surface-confined redox mediator for the development of mediated electrochemical sensors [4–7]. It is well known that the electrochemical sensors involving solution-state redox mediators may suffer from the limitations from higher costs, lower efficiency, and higher environmental burden. Although several methods have been previously reported to confine $K_3[Fe(CN)_6]$/$K_4[Fe(CN)_6]$ on an electrode surface [8–19], most of which are based on electrostatic interaction of cationic species with anionic $[Fe(CN)_6]^{3-/4-}$, the stable electrode-confinement of $[Fe(CN)_6]^{3-/4-}$ to develop high-performance integrative electrochemical sensors remains a challenge.

Poly(diallyldimethylammonium chloride) (PDDA) is an excellent cationic polyelectrolyte of wide applications such as in waste water treatment as a primary organic coagulant [20]. The properties of PDDA such as high charge density, water-solubility, chemical stability, biocompatibility, and environmental friendship as well as low cost also make it a good material in fabricating electrostatic assembly to immobilize macromolecules and/or nanomaterials for sensing and catalytic applications [20–23], loading target DNA molecules for gene transfection [24,25], forming polyelectrolyte hydrogels for protein purification [26,27], and even acting as a bifunctional agent to prepare soluble graphene nanosheets [28]. On the other hand, the carboxyl-containing polysaccharide alginate (Alg) as a well-known biopolymer has found wide scientific and industrial applications [29,30]. Very recently, we reported that mixing PDDA and sodium alginate (AlgNa) can form a robust water-insoluble interpenetrating polymer composite film for enzyme immobilization and amperometric biosensing [31]. On the other hand, the sensitive determination of nitrite is of high significance due to its environmental toxicity and its suspected carcinogenicity in human body [32,33]. Among various nitrite sensors reported, the electrochemical ones have attracted much attention because of their quick response, high sensitivity, and favorable convenience to be miniaturized. For amperometric methods, the electroreduction of nitrite is more favorable than the oxidation due to the high overpotential necessary for nitrite oxidation, which tends to invite interferences from coexisting electroactive substances [34].

Herein, we report a new method for stably confining $K_3[Fe(CN)_6]$ redox mediator onto PDDA–Alg composite to form an efficient electroactive transducer for integrative electrochemical sensing applications. The strategy demonstrated here is essentially based on the interaction of PDDA and $K_3[Fe(CN)_6]$ [8], after reinforcing the film by adding Alg. The prepared $K_3[Fe(CN)_6]$–PDDA–Alg/Au electrode can efficiently electrocatalyze the reduction of nitrite

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and exhibit good analytical performance for amperometric nitrite sensing.

2. Experimental

2.1. Instrumentation and chemicals

All electrochemical experiments were conducted on a CHI660C electrochemical workstation (CH Instrument Co.), and a conventional three-electrode electrolytic cell was used. An Au disk electrode with 2.0-mm diameter (area 0.031 cm^2) served as the working electrode (WE). The reference electrode (RE) was a KCl-saturated calomel electrode (SCE) and a Pt wire (CE) served as the counter electrode. All potentials here are cited versus SCE. A computer-interfaced HP4395A impedance analyzer was employed in the quartz crystal microbalance (QCM) experiments. AT-cut 9-MHz piezoelectric quartz crystals (PQCs) with 12.5-mm wafer diameter (Model JA5, Beijing Chenjing Electronics Co., Ltd.) were used. The Au electrode with 6.0-mm diameter (key-hole configuration, 0.29 ± 0.01 cm^2 area) on one side of the PQC was exposed to the solution and served as the working electrode (WE), while that on the other side faced air. The solution pH measurements were carried out on a pHS-3C pH meter equipped with a composite pH glass electrode (Shanghai Leici Sci. Instr. Inc., China), after careful pH-calibration procedures. AlgNa and PDDA (20 wt%, MW 400,000–500,000) were Aldrich products. $K_3[Fe(CN)_6]$ was purchased from Tianjin Rainda Chemical Co., Ltd. (Tianjin, China). Aqueous solutions at pH 0.0–8.0 were prepared from H_2SO_4, KOH, H_3PO_4 and/or its salts. All other chemicals were of analytical grade or better quality and used as received. Milli-Q ultrapure water (Millipore, $\geq 18\ M\Omega\ cm$) was used throughout. All the experiments were done at room temperature (around 25 °C).

2.2. Procedures

To clean the Au disk electrode surface, the bare gold disk electrode was abraded with fine SiC paper and polished carefully with 0.3 and 0.05 μm alumina powder and then sonicated in water and absolute ethanol, respectively. Prior to the electrochemical experiments, the Au disk electrode was subjected to continuous potential cycling (0–1.5 V, 30 $mV\ s^{-1}$) in 0.20 M aqueous $HClO_4$, until the cyclic voltammogram became reproducible.

$K_3[Fe(CN)_6]$–PDDA–Alg/Au electrodes were prepared as follows. 300 μL PDDA (0.20 wt% aqueous) and 200 μL AlgNa (1.0 wt% aqueous) were mixed thoroughly. After 13,000-rpm centrifugation for 5 min, the isolated PDDA–Alg composite was thrice washed with ultrapure water [31]. 1.0 μL PDDA–Alg was cast on the gold disk electrode surface. After solvent evaporation, the electrodes (denoted as PDDA–Alg/Au) were first immersed into the aqueous solution of 2.0 mM $K_3[Fe(CN)_6]$ for 15 min, then taken out from the solution, and finally rinsed with ultrapure water to obtain the $K_3[Fe(CN)_6]$–PDDA–Alg/Au electrodes. We found that the adsorption of $K_3[Fe(CN)_6]$ onto the PDDA–Alg/Au electrodes was a relatively quick process, as discussed later. The prepared PDDA–Alg/Au and $K_3[Fe(CN)_6]$–PDDA–Alg/Au electrodes were thoroughly rinsed with ultrapure water, dried at ambient temperature, and stored in a refrigerator at 4 °C while not in use.

During amperometric sensing of nitrite, the prepared electrode was potentiostated at 0.2 V in stirred 0.10 M K_2SO_4 + 0.50 M H_2SO_4 to detect reduction of nitrite. The response current was marked with a change value between the steady-state current after adding nitrite and the initial background current without nitrite.

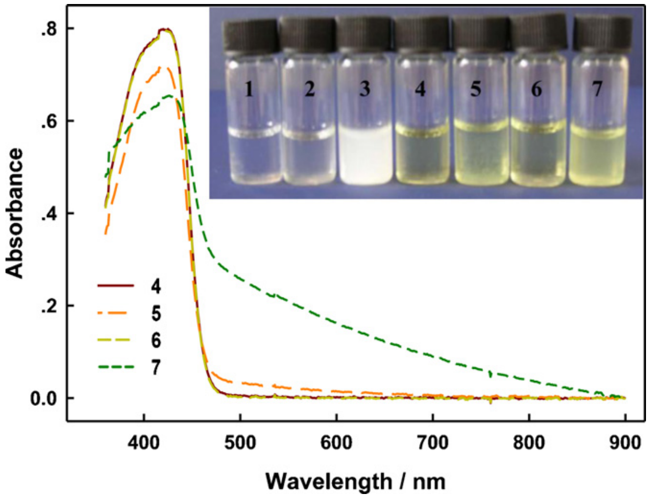

Fig. 1. Digital image of 1 mL 1.0 wt% AlgNa (1), 1 mL 0.20 wt% PDDA (2), 0.4 mL 1.0 wt% AlgNa + 0.6 mL 0.20 wt% PDDA (3), 2 mM $K_3Fe(CN)_6$ solution (4), 0.9 mL 2 mM $K_3Fe(CN)_6$ solution + 0.1 mL 0.20 wt% PDDA (5), 0.9 mL 2 mM $K_3Fe(CN)_6$ solution + 0.1 mL 0.20 wt% Alg (6), and 0.9 mL 2 mM $K_3Fe(CN)_6$ solution + 0.1 mL of the mixture shown in bottle #3 (7). UV–vis spectra of solutions 4 and 6 as well as supernatants of dispersions 5 and 7 after quiescence are also shown.

3. Results and discussion

3.1. Fabrication and characterization of the sensing electrodes

First, we examined the interaction of PDDA with Alg, as well as $K_3[Fe(CN)_6]$ with PDDA, Alg and PDDA–Alg by visual inspection, respectively. As shown in Fig. 1, mixing of colorless PDDA (1) and light yellow AlgNa (2) yielded a milky dispersion of PDDA–Alg hydrogel precipitates (3) within seconds. Adding $K_3[Fe(CN)_6]$ (4) into PDDA immediately yielded yellow $K_3[Fe(CN)_6]$–PDDA precipitates (5), into Alg yielded a transparent $K_3[Fe(CN)_6]$–Alg solution (6), and into PDDA–Alg immediately yielded yellow $K_3[Fe(CN)_6]$–PDDA–Alg precipitates (7). These experiments demonstrate the easy occurrence of $K_3[Fe(CN)_6]$–PDDA interaction in either the presence or the absence of Alg. UV–vis spectra of $K_3[Fe(CN)_6]$ solution as well as supernatants of $K_3[Fe(CN)_6]$–PDDA, $K_3[Fe(CN)_6]$–Alg, and $K_3[Fe(CN)_6]$–PDDA–Alg mixtures after quiescence are also shown in Fig. 1. The maximum absorbance of $K_3[Fe(CN)_6]$ at 422 nm follows the order of $K_3[Fe(CN)_6] \approx$ Alg–$K_3[Fe(CN)_6]$ > $K_3[Fe(CN)_6]$–PDDA > $K_3[Fe(CN)_6]$–PDDA–Alg, implying that both the PDDA and the PDDA–Alg composite can enrich $K_3[Fe(CN)_6]$ but Alg cannot enrich $K_3[Fe(CN)_6]$, and the enrichment efficiency of PDDA–Alg is higher.

We also examined the interaction of solution-state $K_3[Fe(CN)_6]$ and added PDDA by cyclic voltammetry (CV) at bare Au disk electrode. As shown in Fig. 1S in the Supplemental Data, nearly reversible redox peaks of the $[Fe(CN)_6]^{3-/4-}$ redox couple were observed in the potential range of 0.15–0.25 V in the absence of PDDA. In general, the $K_3[Fe(CN)_6]$ ions dissolved in aqueous solutions exhibit redox peaks in this potential range [34–36], though a notable acidification of the solution can positively shift the peak potentials. The shape and size of the CV responses changed notably with gradually adding PDDA in the 0.1 M PBS buffer (pH 7.4) containing 2 mM $K_3[Fe(CN)_6]$. The shape and size of the redox peaks at 0.15–0.25 V decreased and finally disappeared, while another redox peaks appeared in the lower potential region (at 0.05–0.15 V). The peak currents of the new redox peaks increased with the increase of PDDA concentration at first, and then became stable when PDDA concentration reached at 0.05 wt%. The observations are similar to those reported previously [8].