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Short communication

Stripping voltammetric analysis of organophosphate pesticides using Ni/Al layered double hydroxides as solid-phase extraction

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ABSTRACT

A sensitive electrochemical stripping voltammetric biosensor is designed for organophosphate pesticides (OPs) based on solid-phase extraction (SPE) using Ni/Al layered double hydroxides (LDHs) modified glassy carbon electrode (labeled as Ni/Al-LDHs/GCE). The Ni/Al-LDHs as the host are highly efficient to capture OPs, which dramatically facilitates the enrichment of nitroaromatic OPs onto their surface and realizes the stripping voltammetric detection of OPs. The stripping voltammetric performances of methyl parathion (MP) intercalated into LDHs were evaluated by cyclic voltammetric and square-wave voltammetric (SWV) analysis. The combination of the host–guest supramolecular structure, SPE, and stripping voltammetry provides a fast, simple, and sensitive electrochemical method for detecting nitroaromatic OPs by using MP as a model. The stripping analysis is linear over the MP concentration ranges of 0.001–0.1 and 0.2–1.0 μg mL^{-1} with a detection limit of 0.6 ng mL^{-1} (S/N = 3). The developed biosensor exhibits good reproducibility and acceptable stability. This study offers a new promising protocol for OPs analysis.

1. Introduction

Owing to the high toxicity, extensive use of organophosphates pesticides (OPs) for pest control has raised serious public concern regarding the healthy, environment and food safety (Arduini et al., 2006; Li et al., 2007a,b). They disrupt the cholinesterase enzyme that regulates acetylcholine, which often causes respiratory paralysis and death (Kim et al., 2005). OP residues in crop, livestock, and water pose a severe threat to human life. Therefore, rapid determination and reliable quantification of a trace level of OP compounds have become increasingly important for public security and health protection. Traditional analysis of OPs is routinely carried out by gas/liquid chromatography or mass spectroscopy (Chen and Huang, 2006; Rotiroti et al., 2005; Leandro et al., 2006). These methods often require complicated pretreatment steps, extensive labor resources, and are not applicable for on-site determination. Over the past few years, enzyme-based biosensors have emerged as a promising alternative to detect pesticides (Vakurov et al., 2004; Sotiropoulou et al., 2005; Jeanty and Marty, 1998; Sadik et al., 2003; Li et al., 2007a,b). For instance, inhibition or noninhibition biosensor systems, respectively, based on the immobilization of acetylcholinesterase (AChE) or organophosphates pesticide hydrolyse (OPH) onto various electrochemical transducers, have been extensively developed for pesticides analysis (Mulchandani et al., 1998; Wang et al., 1999; Schulze et al., 2003; Kok and Hasirci, 2004; Liu and Lin, 2006; Gong et al., 2009). Nevertheless, the operational conditions are mostly limited by the denaturation of enzymes. To overcome this problem, stripping voltammetric analysis combined with preconcentration of OPs appears to be an ideal and highly sensitive technology without using enzyme. In particular, the efficient preconcentration of OPs onto a certain substrate is critical for stripping analysis. As is well known, solid-phase extraction (SPE) is a most popular method for sample preparation and separation and often used in the field of separation science (Hennion, 2000; Shi et al., 2006). During SPE, analytes are generally extracted and concentrated by an adsorbent. Stripping voltammetry combined with SPE will obviously well meet the requirements of decentralized point-of-care tests or field detections of OPs in the environment with portable instruments and simple operations (Liu and Lin, 2005a,b; Du et al., 2008).

Layered double hydroxides (LDHs), known as hydrotalcite-like materials, are a class of two-dimensional nanostructured anionic clays. The positively charged layers contain edge-shared metal M(II) and M(III) hydroxide octahedral, with charges neutralized by A^{n-} anions located in the interlayer spacing or at the edges of the lamella. LDHs may be represented by the general formula $[M^{II}_{1-x}M^{III}_{x}(OH)_2]^{x+}$ $(A^{n-})_{x/n}\cdot mH_2O$, where M^{2+} and M^{3+} are di- and tri-valent metal cations, respectively, the value of x is in the range between 0.22 and 0.33 (Braterman et al., 2004). Recently, LDHs have received much attention as host materials to construct host–guest supramolecular structures. They offer great possibilities for fabricating tailored functional materials with novel properties.

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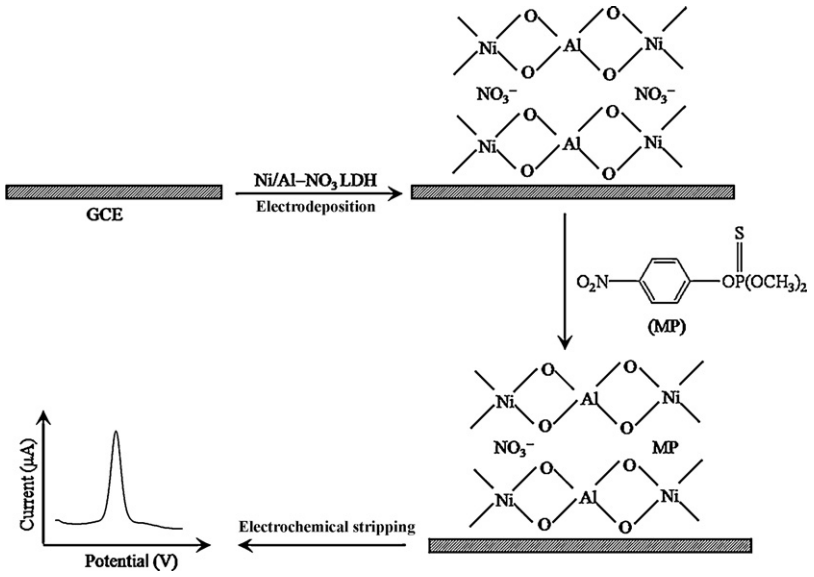


Scheme 1. Schematic illustration for electrochemical sensing nitroaromatic OP compound. (A) Electrodeposition of a Ni/Al-NO_3-LDHs film onto a GCE surface; (B) MP intercalated into the interlay space of LDHs onto a GCE surface; and (C) electrochemical stripping detection of MP.

Through appropriate intercalation processes, the host–guest hybrid materials based on LDHs have been used in catalysis, scavengers, adsorbents, photochemistry, electrochemistry, and pharmaceutics (Wei et al., 2008; Zhang et al., 2008; Scavetta et al., 2007a,b; Choudary et al., 2002; Inacio et al., 2001; Hernández et al., 2007; Scavetta and Tonelli, 2005; Bruna et al., 2006; Chao et al., 2008). Previous studies on the adsorption of some hazardous organic polar molecules like phenolic compounds, or pesticides by LDHs have demonstrated their feasibility as the scavenger to remove undesirable toxicants in the environment (Inacio et al., 2001; Bruna et al., 2006; Chao et al., 2008). Since organic polar molecules can be selectively hosted in the interlayer space, this feature inspires us to use LDHs for solid-phase extraction (SPE) of OPs. It is expected to dramatically facilitate the enrichment of OPs and realize their stripping voltammetric detection. Among OP compounds, nitroaromatic OPs including methyl parathion (MP), paraoxon, and fenitrothion, exhibit good redox activities at an electrode surface.

In this paper, we describe an electrochemical stripping analysis of nitroaromatic OPs based on SPE at a Ni/Al-LDHs-modified electrode (Scheme 1). The new LDHs-based electrochemical sensing protocol involves simple one-step electrosynthesis of a thin Ni/Al-LDHs film onto a glassy carbon electrode surface (A), subsequent intercalation of MP as a model (B), and final electrochemical stripping detection of adsorbed MP (C). Nowadays, LDHs-based sensors are mainly focused on electrochemical analysis of ethanol, methanol and glucose, where M(II) in LDHs as the electron mediator undergoes a redox reaction (Scavetta et al., 2007a,b). To the best of our knowledge, this is the first report on the determination of OPs by utilizing LDHs as the host of OPs. Encouragingly, the combination of the host–guest supramolecular structure, SPE, and stripping voltammetry opens up new opportunities for fast, simple and sensitive analysis of OPs.

2. Experiments

2.1. Reagents

Methyl parathion (MP) was obtained from Treechem Co. (Shanghai, China). The Ni/Al-LDHs were electrosynthesized starting from freshly prepared solutions of Ni and Al nitrates. All other chemicals were of analytical-reagent grade and used without further purification. Doubly distilled water was used. The 0.1 M phosphate buffer solutions (PBS) of different pH values were prepared for use.

2.2. Apparatus

Electrochemical measurements were performed on a CHI 660A electrochemical workstation (CHI, USA) with a conventional three-electrode system comprising a platinum wire as an auxiliary electrode, a saturated calomel electrode (SCE) as reference, and the modified or unmodified glass carbon electrode (GCE) as a working electrode. All experiments were carried out at ambient temperature. Semi-empirical PM3 optimizations were carried out by using Gaussian 98 (A.7) (Gaussian Inc.).

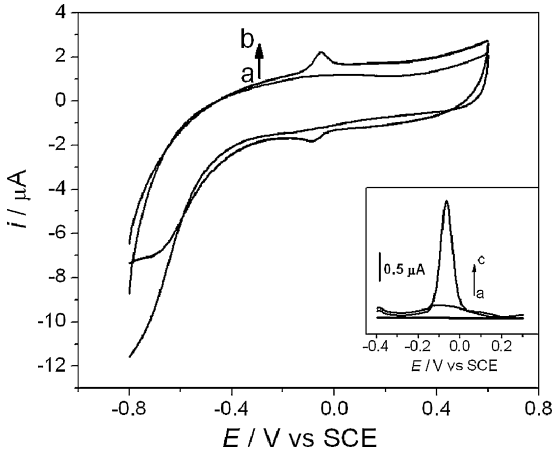


Fig. 1. Cyclic voltammograms of Ni/Al-LDHs/GCE (a) methyl parathion intercalated onto Ni/Al-LDHs/GCE (b) in 0.1 M PBS (pH 5.7). Scan rate: 100 mV/s. Inset: stripping voltammograms obtained at Ni/Al-LDHs/GCE in the absence of MP (a), the solution containing 0.1 μg mL^{-1} MP without preconcentration (b), and after SPE in 0.1 μg mL^{-1} MP (c). SWV conditions: scanning potential range, −0.4 to 0.3 V; frequency, 25 Hz; potential increment, 4 mV; amplitude of the square-wave, 20 mV. Electrolyte: 0.1 M PBS pH 5.7.

2.3. Electrode preparation

Prior to modification, the basal GCE was polished to a mirror finish using 1.0, 0.3 and 0.05 μm alumina slurries. After each polishing, the electrode was sonicated in ethanol and doubly distilled water for 5 min, successively, in order to remove any adsorbed substances on the electrode surface. Finally, it was dried under nitrogen atmosphere ready for use. A Ni/Al-LDHs film was electrochemically generated on the cleaned GCE at a constant potential of −0.9 V vs. SCE for 60 s in an aqueous solution containing 0.0225 M Ni $(NO_3)_2$, 0.0075 M $Al(NO_3)_3$ and 0.3 M KNO_3 (Scavetta et al., 2007a,b), according to the following reactions:

$$NO_3^- + H_2O + 2e \rightarrow NO_2^- + 2OH^-$$

$$(1-x)Ni^{2+} + xAl^{3+} + 2OH^- + xNO_3^- + mH_2O \rightarrow [Ni_{1-x}Al_x(OH_2)]^{x-}(NO_3)_x \cdot mH_2O$$

After modification, the electrode (labeled as Ni/Al-LDHs/GCE) was thoroughly rinsed with water and kept at room temperature for further use.

2.4. Measurement procedure

The Ni/Al-LDHs-modified GCE was first immersed into a stirred sample solution containing the desired MP concentration for a given time, and rinsed with water. Then, it was transferred into the blank electrolyte (0.1 M, pH 5.7 PBS) for SWV measurements. Multiple successive SWV scanning was used to remove the bound MP until the anodic stripping response disappeared. Also, the regeneration of the sensor surface was achieved. Before electrochemical measurements, the electrolyte solution was purged with nitrogen gas for 10 min.

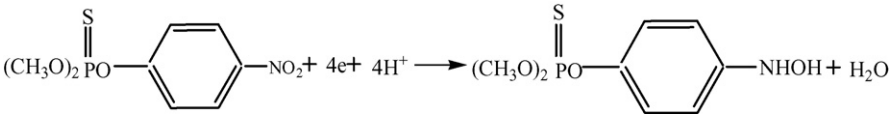
3. Results and discussion

3.1. Electrochemical characterization of Ni/Al-LDHs/GCE

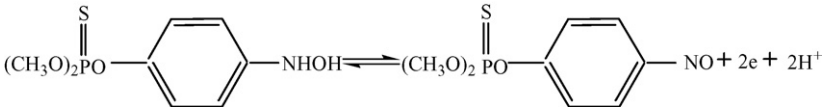
A typical CV curve recorded at Ni/Al-LDHs/GCE in 0.1 M NaOH at a scan rate of 20 mV/s is shown in Fig. S1b. Compared with the bare GCE (Fig. S1a), the cathodic and anodic peaks, centered at potentials of 0.603 and 0.422 V vs. SCE, respectively, can be ascribed to the redox couple of Ni(II)/Ni(III) in LDHs. The result agrees well with the previous reports on the redox behavior of Ni(II)/Ni(III) (Scavetta et al., 2007a,b), indicating that the Ni/Al-NO_3-LDHs film has been successfully electrosynthesized onto the surface of GCE.

3.2. Effect of methyl parathion on response of Ni/Al-LDHs/GCE

Since the interlayer space of LDHs can host the nitroaromatic OPs, it provides a facile approach to attach OPs onto an electrode surface. Fig. 1 displays the CV of MP intercalated on Ni/Al-LDHs/GCE in 0.1 M PBS (pH 5.7). During the potential range from −0.8 to 0.5 V, no obvious redox peak appeared in the absence of MP (Fig. 1a). However, a pair of well-defined redox peaks (E_{pa}, −0.04 V; E_{pc}, −0.08 V) and an irreversible reduction peak (E_{pc}, −0.67 V) were observed (Fig. 1b) with the intercalation of MP onto Ni/Al-LDHs/GCE. The irreversible reduction peak corresponds to the reduction of the nitro group to the hydroxylamine group (reaction (1)). The reversible redox peaks are attributed to a two-electron-transfer process (reaction (2)), shown as below. These profiles are consistent with those described elsewhere for nitroaromatic OPs (Liu and Lin, 2005a,b):



(1)



(2)

SWV analysis has a higher sensitivity than that of other electrochemical techniques. As shown in the inset of Fig. 1, no anodic stripping peak was observed at Ni/Al-LDHs/GCE in blank PBS (curve a). Evidently, a very sharp and well-defined stripping peak at a potential of about 0.06 V vs. SCE (curve c) appeared with the intercalation of MP into LDH. Compared with the direct measurement in MP sample solution (curve b), the stripping peak current at MP intercalated Ni/Al-LDHs/GCE was greatly enhanced. This may be attributed to the specific host–guest interaction between Ni/Al-LDHs and MP. Furthermore, the dipole moment of MP was calculated to be 4.9885 D by semi-empirical PM3 optimization. The bigger value of dipole moment, the easier a molecule is intercalated into the interlayer space of LDHs. Obviously, the presence of a Ni/Al-NO_3-LDHs film plays a great role, providing an advantageous and high-performance platform for the preconcentration of MP. The SPE through the intercalation of MP into LDHs leads to the enrichment of MP onto the surface of the electrode, and is therefore responsible for the significant enhancement of the current response.

3.3. Effects of extraction time and pH on the response of AChE-Au-PPy/GCE

We further optimized the experimental parameters to get high-performance stripping analysis of OPs. It was found that extraction time was one of the most influential parameters in pesticide analysis. The peak currents increased rapidly with an increase of immersion time, and then tended to be stable at ~20 min. This indicates that the intercalation of MP into LDHs reaches saturation (Fig. S2a).

The response of MP was also dependent on pH of adsorption media. The stripping signal was enhanced with an increase of pH up to 5.7, but decreased at higher pH (Fig. S2b). In an acidic solution (pH < 5), the partial dissolution of LDHs matrix onto the substrate is likely to occur by acidic hydrolysis (Inacio et al., 2001). Since basic media would result in the degradation of OP compounds and H^+ was involved in the irreversible reduction of MP (as shown in the reactions of (1)–(2)), a pH value of 5.7 was selected for measurements in this study (Wang et al., 2001).

3.4. Analytical performance for the detection of methyl parathion

Fig. 2 displays the SWV response of adsorbed MP by the SPE process at Ni/Al-LDHs/GCE. Well-defined peaks, proportional to the concentration of the corresponding MP, were observed in two

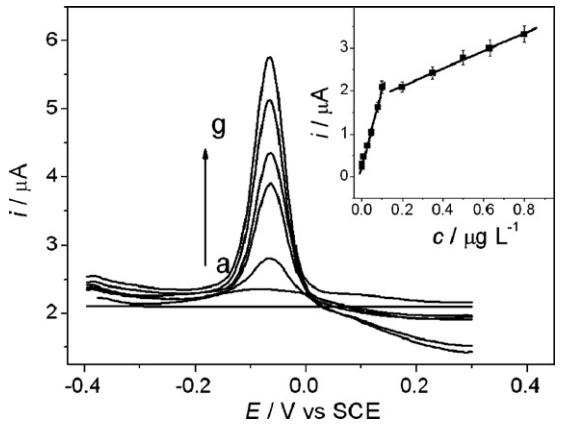


Fig. 2. Stripping voltammograms of increasing MP concentrations (from bottom to top, 0, 1, 10, 80, 100, 500, and 800 ng mL^{-1}, respectively). The inset shows the calibration curve. Electrochemical stripping detection conditions are the same to those in Fig. 1.

ranges, 0.001–0.1 and 0.2–1.0 μg mL^{-1}. The linear equations were $i/\mu A = 0.237 + 17.88\, c/\mu g\, L^{-1}$, and $i/\mu A = 1.712 + 2.04\, c/\mu g\, L^{-1}$, with the correlation coefficients of 0.9971 and 0.9982, respectively (inset of Fig. 2). A detection limit of 0.6 ng mL^{-1} was obtained with the calculation based on signal-noise ratio equal to 3. This detection limit is believed to be the lowest one reported so far. It is significantly lower than 13.2 ng mL^{-1} at a carbon-paste electrode by using stripping analysis (Liu and Lin, 2005a,b), lower than 1.0 ng mL^{-1} at ZrO_2-nanoparticles modified electrodes (Liu and Lin, 2005a,b; Du et al., 2008), and those reported with enzyme-based inhibitor electrochemical sensors (Schulze et al., 2003; Kok and Hasirci, 2004; Liu and Lin, 2006; Gong et al., 2009). Our results demonstrated that the proposed Ni/Al-LDHs composite film was reliable for the determination of OPs. The relative standard deviation was 5.2% for 10 replicate determinations of 0.1 μg mL^{-1} MP, indicating acceptable reproducibility.

The stability of the LDHs-modified electrode could be maintained by being stored at 4 °C in a dry condition. No obvious decrease in the response of MP was observed in the first 10-day storage. After a 30-day storage period, the sensor retained 75% of its initial current response. No obvious interferences were observed from the other electroactive nitrophenyl derivatives such as nitrobenzene, nitrophenol and other oxygen-containing inorganic ions (PO_4^{3-}, SO_4^{2-}, NO_3^-) with the peak currents of MP varied slightly. It is likely due to the strong host–guest interaction between Ni/Al-LDHs and MP. By semi-empirical PM3 optimization, the dipole moment of MP (MP_{dm} = 4.9885 D) is larger than those of other nitrophenyl derivatives (for example, $nitrophenol_{dm}$ = 3.6329 D; $nitrobenzene_{dm}$ = 4.1636 D). A molecule with a larger dipole moment is easier to intercalate into the interlayer space of LDHs. Obviously, the intercalation capability of MP to LDHs is much stronger than other nitrophenyl derivatives. Furthermore, the stripping peak potential of MP is about 0.06 V, whereas the oxidation potentials of other phenol compounds and electroactive species are more than 0.35 V. Such a low stripping potential of 0.06 V could avoid the interference efficiently.

To further demonstrate the practicality of the proposed method, the recovery tests were carried out by adding different amounts of MP into garlic samples (summarized in Table S1). The recoveries were from 97.5% to 105.0%. The results indicated that the proposed method was highly accurate, precise and reproducible. It can be used for direct analysis of relevant samples.

4. Conclusions

This work has successfully demonstrated a simple and efficient strategy for stripping analysis of OPs at Ni/Al-LDHs-modified GCE. As a typical host material, LDH films were able to extract nitroaromatic OPs from the solution, and facilitated the preconcentration of MP onto the surface with the stripping current response greatly enhanced. The fast, sensitive and selective determination of MP was realized. The resulting biosensor showed both good reproducibility and ideal stability. We believe that the combination of SPE and LDHs-based host–guest hybrid materials as well as SWV will pave the new way for electrochemical detection of OPs.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bios.2009.07.008.

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