

# Indirect Electroanalysis of 3-Methyl-4-Nitrophenol in Water Using Carbon Fiber Microelectrode Modified with Nickel Tetrasulfonated Phthalocyanine Complex

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## Abstract

Electrochemical detection of 3-methyl-4-nitrophenol (MNP) in direct phenol oxidation occurs at high potentials and generally leads to progressive passivation of the electrochemical sensor. This study describes the use of a carbon fiber microelectrode modified with a tetrasulfonated nickel phthalocyanine complex for the detection of MNP at a lower potential than that of direct phenol oxidation. The MNP voltammogram showed the presence of an anodic peak at  $-0.11$  V vs SCE, corresponding to the oxidation of the hydroxylamine group generated after the reduction of the nitro group. The effect of buffer pH on the peak current and SWV parameters such as frequency, scan increment, and pulse amplitude were studied and optimized to have better electrochemical response of the proposed sensor. With these optimal parameters, the calibration curve shows that the peak current varied linearly as a function of MNP concentration, leading to a limit of detection (LoD) of  $1.1$   $\mu\text{g/L}$ . These results show an appreciable sensitivity of the sensor for detecting the MNP at relatively low potentials, making it possible to avoid passivation phenomena.

## Keywords

3-Methyl-4-Nitrophenol, Carbon Fiber Microelectrode, Nickel Tetrasulfonated Phthalocyanine, Indirect Electroanalysis, Square Wave Voltammetry

## 1. Introduction

Organophosphorus (OPs) compounds are commonly used pesticides to control insects and improve crop yields. In Burkina Faso, fenitrothion (FT), an organophosphate insecticide, is widely used in cotton cultivation and during locust invasions [1] [2]. Like other organophosphates, FT acts by inhibiting acetylcholinesterase, which disrupts nerve impulse transmission [3] [4]. Although FT degrades quickly and does not accumulate much in the environment, its degradation product, 3-methyl-4-nitrophenol (MNP), is highly persistent in the environment, particularly in water. MNP is toxic and can cause harm to biological systems as an endocrine disruptor [2] [5]. Therefore, it is essential to develop analytical methods for detecting these environmental pollutants. While various analytical methods have been used to determine OPs, electroanalytical techniques are more attractive due to their simplicity, high sensitivity, and good selectivity [6]. Several studies have reported the development of electrochemical sensors for quantifying OPs pesticides and/or their metabolites [7]-[14]. For example, Ensafi *et al.* [15] modified a glassy carbon electrode with polysilicon for the electrochemical analysis of fenitrothion in water and fruit samples. A nano-silver/dodecane modified glassy carbon electrode was developed by Kumaravel *et al.* [16] and used to detect fenitrothion in potato samples. Han *et al.* [17] recently developed an electrochemical sensor based on nitrogen-sulfur co-doped holey graphene (NS-HG) to detect fenitrothion in environmental samples. However, to our knowledge, very few studies report the electrochemical detection of MNP, the most stable metabolite of fenitrothion. Thus, in a previous work [1], we described an electrochemical sensor based on an ultramicroelectrode modified with a nickel tetrasulfonated phthalocyanine complex to determine MNP in the direct oxidation of the phenol group. In another work [2], we reported electrode passivation during direct electrooxidation of MNP, and we proposed an anti-passivation strategy to mitigate this issue. However, this strategy poses a challenge for the durability of the sensor.

In this work, we report the indirect detection of 3-methyl-4-nitrophenol by monitoring the oxidation peak of the hydroxylamine group generated following the reduction of the nitro group using square wave voltammetry as an analytical technique.

## 2. Experimental

### 2.1. Reagents and Solutions

3-methyl-4-nitrophenol (MNP), sodium phosphate monobasic ( $\text{NaH}_2\text{PO}_4$ ), sodium phosphate dibasic ( $\text{Na}_2\text{HPO}_4$ ), nickel tetrasulfonated phthalocyanine (NiTSPc), potassium ferrocyanide ( $\text{K}_4\text{Fe}(\text{CN})_6$ ), potassium ferricyanide ( $\text{K}_3\text{Fe}(\text{CN})_6$ ), potassium chloride (KCl), sodium hydroxide (NaOH), sulfuric acid ( $\text{H}_2\text{SO}_4$ ) and absolute ethanol were all purchased from Sigma-Aldrich and were used without further purification. Phosphate buffer solution was prepared by mixing the appropriate amount of  $\text{NaH}_2\text{PO}_4$  and  $\text{Na}_2\text{HPO}_4$  and was used as the supporting

electrolyte. A solution for electrochemical cleaning of naked carbon fiber microelectrode (CFME) was prepared by doing an equi-volume mix of H<sub>2</sub>SO<sub>4</sub> 0.5 M and absolute ethanol. Solution of NiTSPc 2 mM was prepared by dissolving 98 mg of this compound in 50 mL of NaOH 0.1 M solution. All aqueous solutions were prepared with distilled water.

## 2.2. Apparatus

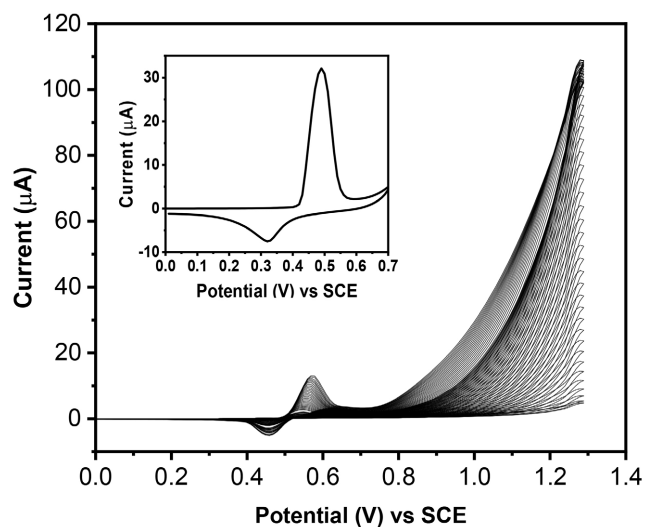
All voltametric measurements were realized using a potentiostat DY 2300 (Digi-IVY Instruments, USA) controlled by the DY 2300 EN software via a computer. CFME (diameter  $\Phi = 12 \mu\text{m}$ , length = 5 mm) and modified carbon fiber microelectrode by NiTSPc film (CFME/p-NiTSPc) were used as working electrodes while saturated calomel electrode (SCE) and platinum electrode (diameter  $\Phi = 250 \mu\text{m}$ ) were used respectively as reference and counter electrodes. Electrochemical impedance spectroscopy (EIS) experiments were performed using Orygaflex potentiostat purchased from Orygalys company (Lyon, France) controlled by a computer using OrigaMaster 5 as software. The frequency range of 1 kHz to 100 MHz. Scanning electron microscopy (SEM) and energy dispersive X-ray spectroscopy (EDX) experiments were achieved on CFME and CFME/p-NiTSPc with a JEOL type JSM-6301F (SCIAM, Angers university). SEM images were obtained from secondary electrons under 3 - 5 keV with magnifications situated between 3000 and 5000. EDX spectra were obtained using a beam with an energy of 20 keV.

## 2.3. Analytical Procedure

Cyclic voltammetry was used to modify CFME and to investigate the electrochemical behavior of MNP on CFME and CFME/p-NiTSPc. A solution of 5 mM [Fe(CN)<sub>6</sub>]<sup>3-/4-</sup> (1:1) and containing 0.1 M KCl was used for EIS experiments. Square wave voltammetry has been used for quantitative analyzes in aqueous samples by following the indirect oxidation peak of MNP.

## 2.4. Electrochemical Modification of CFME

Before using a CFME, it is important to clean it for removing any impurities on the carbon fiber. This cleaning is done electrochemically in a mixture of H<sub>2</sub>SO<sub>4</sub> and absolute ethanol (1:1, v/v) by sweeping in cyclic voltammetry at a scan rate of 50 mV/s and for 10 cycles from -1.5 V to 1.5 V. Then, the cleaned CFME is modified by p-NiTSPc following the modification protocol already reported in previous works [18]. **Figure 1** shows the obtained voltammograms for the electrodeposition of p-NiTSPc on CFME and the electrochemical revelation (insert) of modified CFME/p-NiTSPc. The p-NiTSPc film thickness was evaluated from the last cyclic voltammogram of p-NiTSPc electrodeposition. Under these experimental conditions, the p-NiTSPc film electrodeposited presents a recovery of approximately  $1.28 \times 10^{-8} \text{ mol/cm}^2$  determined from integrating the oxidation peak, corresponding to a film thickness of 128 nm, as previously reported [19].



**Figure 1.** 75 successive cyclic voltammograms of CFME in a 0.1 M NaOH aqueous solution containing 2 mM of NiTSPc at the potential scan rate of 100 mV/s. Insert show 1 cyclic voltammogram of CFME/p-NiTSPc in a 0.1 M NaOH aqueous solution at the potential scan rate of 100 mV/s.

### 3. Results and Discussions

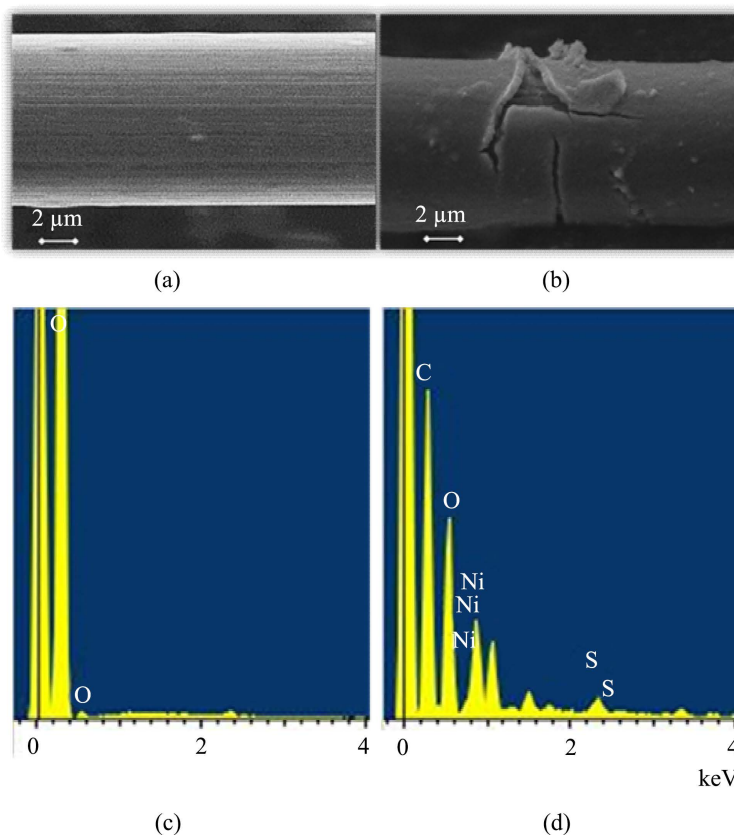
#### 3.1. SEM and EDX Characterization of Working Electrodes

**Figure 2(a)** and **Figure 2(b)** shown SEM images of unmodified and modified CFMEs respectively. By comparing these figures, there is a clear difference in the state of naked and modified carbon fibers. Indeed, **Figure 2(a)** shows a fiber with evenly distributed streaks, while **Figure 2(b)** shows a fiber covered with a matter which is the nickel phthalocyanine film. Moreover, EDX analyzes were carried out on unmodified and modified CFMEs and the obtained results are presented in **Figure 2(c)** and **Figure 2(d)**.

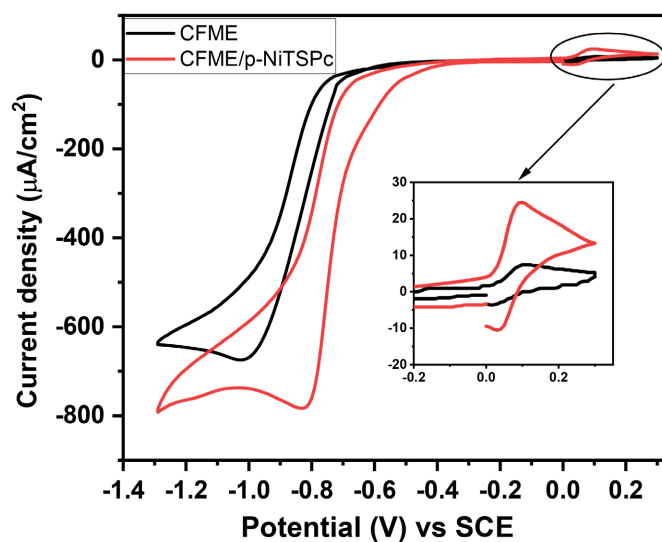
We can note on the two spectra the presence of carbon which can be attributed to the fiber. The presence of oxygen on the unmodified fiber is due (**Figure 2(a)**) to the electrochemical cleaning solution which contains both  $\text{H}_2\text{SO}_4$  and EtOH. However, in the case of the modified fiber, the increase in the quantity of oxygen (**Figure 2(b)**) compared to that of the unmodified fiber can be attributed to the sulfonate group of tetrasulfonated nickel phthalocyanine. Furthermore, on the EDX spectrum of the modified CFME (**Figure 2(d)**), we also observe the presence of nickel and sulfur (which are found in NiTSPc) and which are absent from the spectrum of the unmodified fiber (**Figure 2(c)**). These results are approve of the electrode modification by the p-NiTSPc film.

#### 3.2. Comparison of Voltammetric Behavior of 3-Methyl-4-Nitrophenol at Naked and Modified Carbon Fiber Microelectrodes

The cyclic voltammogram of MNP at a concentration of 30 mg/L in a pH 7 phosphate buffer with a naked CFME and CFME/p-NiTSPc is shown in **Figure 3**.



**Figure 2.** SEM images of (a) CFME and (b) CFME/p-NiTSPc. EDX spectra of (c) CFME and (d) CFME/p-NiTSPc.



**Figure 3.** Cyclic voltammogram of 30 mg/L MNP in PBS pH 7 at unmodified CFME (black) and CFME/p-NiTSPc (red) versus SCE at the scan rate  $v = 50$  mV/s.

An examination of this voltammogram indicates the presence of three peaks with the two tested electrodes: one irreversible peak and two reversible peaks. For the unmodified CFME, the irreversible peak is observed around  $-0.90$  V and

can be attributed, according to previous reports, to the direct reduction of the nitro group to a hydroxylamine group after a transfer of  $4 e^-$  [2]. In addition, reversible peaks observed around  $-0.11$  V and  $0.02$  V are respectively due to the oxidation of the hydroxylamine group previously generated to the nitroso group and the reduction of the nitroso group to the hydroxylamine group as already reported [6]. Moreover, when CFME/p-NiTSPc is used, we observed an increase in the intensity of all peaks and a shift of reduction and oxidation peaks to the anodic and cathodic potential respectively. These results reflect that the p-NiTSPc film has an electrocatalytic effect on the indirect oxidation and direct reduction reactions of MNP. Thus, CFME presents better sensitivity in the presence of the p-NiTSPc film.

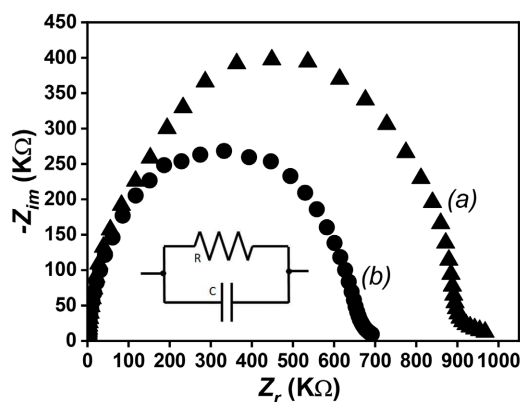
### 3.3. Characterization of CFME and CFME/p-NiTSPc by Electrochemical Impedance Spectroscopy

**Figure 4** show the EIS plot of CFME and CFME/p-NiTSPc realized in  $5$  mM  $[\text{Fe}(\text{CN})_6]^{4-/3-}$  solution containing  $0.1$  M KCl.

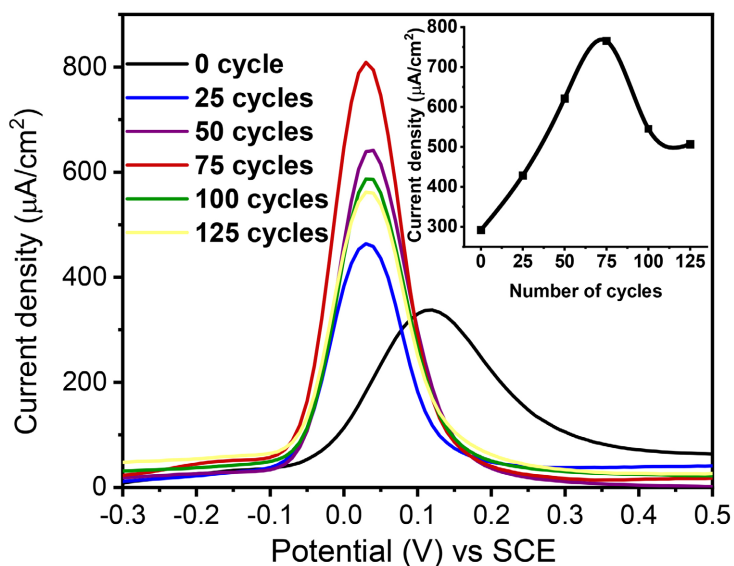
It can be seen on the **Figure 4** that the Nyquist diagram of the unmodified and modified CFME are different. Indeed, the charge transfer resistance ( $R_{ct}$ ) of the CFME is  $930$  k $\Omega$ , while that of CFME/p-NiTSPc is reduced to  $680$  k $\Omega$ . These results show on the one hand, the change on the electrode surface and then its modification, and on the other hand, show the enhancement of the electron transfer rate of the system  $[\text{Fe}(\text{CN})_6]^{3-/4-}$  and then the electrocatalytic properties of the p-NiTSPc film.

### 3.4. Effect of Electrodeposition Cycle Number of p-NiTSPc on CFME

In order to obtain a sensitive sensor, the modification of the CFME has been optimized through the number of cycles of electrodeposition of the p-NiTSPc film. The voltammograms obtained using modified CFMEs at different cycles of electrodeposition are shown in **Figure 5**.



**Figure 4.** Nyquist plots of (a) CFME and (b) CFME/p-NiTSPc obtained in  $5$  mM  $[\text{Fe}(\text{CN})_6]^{3-/4-}$  ( $1:1$ ) solution containing  $0.1$  M KCl. The frequency range was from  $100$  KHz to  $100$  MHz.



**Figure 5.** SWV of MNP at 500 mg/L ( $3.3 \times 10^{-3}$  M) in a PBS pH = 7 with CFME and CFME/p-NiTSPc as working electrodes. SWV parameters: frequency = 50 Hz; pulse amplitude = 50 mV; potential increment = 10 mV.

This figure shows a variation in peak intensity with the number of nickel phthalocyanine electrodeposition cycles. Indeed, we first notice that as the number of electrodeposition cycles increases, the current density increases to reach a maximum at 75 cycles. However, beyond 75 cycles, there is a decrease in current density showing a diffusion limiting process of the film deposited. Indeed, an increase in the thickness of the electrodeposited p-NiTSPc film could affect its nanoporosity and therefore limit the diffusion of MNP towards the surface of the electrode similarly observed by Pontié *et al.* [20]. In addition, we notice that the oxidation peak shift towards cathodic values. This shift may be attributed to the electrocatalytic effect of the p-NiTSPc film, which was previously reported in Section 3.2 and depicted in Figure 3. The electrocatalytic effect of the p-NiTSPc film enhances the rate of this reaction by providing an efficient pathway for electron transfer, leading to a significant increase in the electrochemical activity of the system.

### 3.5. Optimization of pH

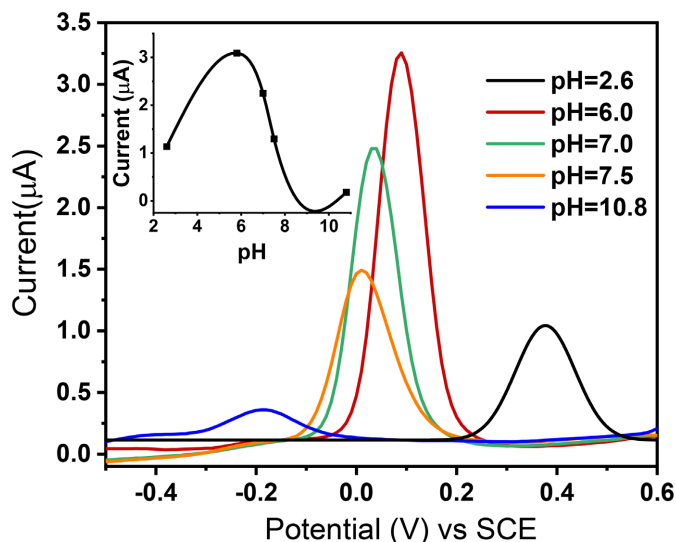
Figure 6 shows the voltammograms of MNP obtained at different pHs of the PBS.

It can be seen from Figure 6 that as the pH increases, the oxidation peak potential shifts towards the cathodic values. This result could be explained by the deprotonation of hydroxylamine, generating anionic species rich in electrons and, therefore, more easily oxidizable. In addition, the representation of the peak intensity as a function of pH (inset of Figure 6) also shows a variation of the peak intensity with a maximum at pH = 6. This value pH = 6 has been retained as the optimal value in further experiments.

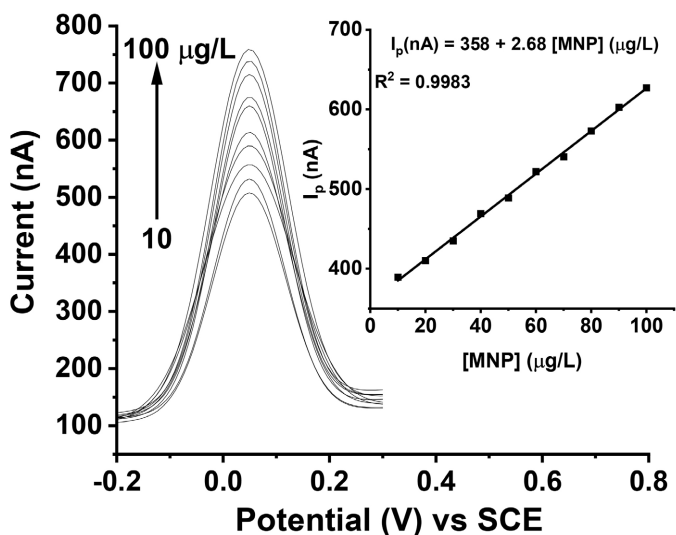


### 3.6. Quantitative Analysis of MNP

Taking into account the optimized number of cycles of electrodeposition of the p-NiTSPc film and the optimized analysis pH of the MNP, the parameters of the SWV were also optimized. The optimal values obtained are a frequency of 50 Hz, a pulse amplitude of 70 mV and a step potential of 50 mV. Thus, these optimal values were used for the quantitative analysis of MNP. **Figure 7** shows the square wave voltammograms for different concentrations of MNP standard on CFME/p-NiTSPc.



**Figure 6.** SWV of MNP at 500 mg/L in a phosphate buffer at different pH at a CFME/p-NiTSPc. Inset: plot of peak intensity versus pH of PBS. SWV Parameters: frequency = 50 Hz; pulse amplitude = 50 mV; step potential = 10 mV.



**Figure 7.** SWV of MNP at different concentrations (10, 20, 30, 40, 50, 60, 70, 80, 90, and 100 μg/L) in phosphate buffer (pH = 6) at a CFME/p-NiTSPc. Inset: plot of peak intensity versus concentration of MNP. SWV parameters: frequency = 50 Hz; pulse amplitude = 70 mV; potential increment = 50 mV.



**Figure 7** shows that the peak current varies linearly with the concentration of MNP in the range of 10 - 100 µg/L with a regression equation of  $I_p$  (nA) = 2.68C (µg/L) + 358 and a limit of detection (3 S/m) of 1.1 µg/L. Although this value is very similar to the detection limit obtained in our previous work (previous LoD = 0.75 µg/L) [1] when detecting MNP in direct oxidation of the phenol group, analysis in indirect oxidation could be attractive to exploit because it is without risk of passivation. Thus, considering the proximity of the detection limit values, indirect MNP oxidation analysis could be an alternative to direct MNP oxidation detection without altering its sensitivity through any electrode passivation.

#### 4. Conclusion

MNP was detected using SWV and by measuring the indirect oxidative peak current of the hydroxylamine group to nitroso group at modified CFME with p-NiTSPc. The pH of the analysis solution and the SWV parameters have been optimized, and the optimal values are as follows: pH = 6, frequency 50 Hz, scan increment 50 mV, and pulse amplitude 70 mV. In the concentration range of 10 µg/L to 100 µg/L, the peak current varied linearly. The lowest limit of detection (LoD) calculated for MNP on CFME/p-NiTSPc is 1.1 µg/L. These results demonstrate that CFME/p-NiTSPc has high sensitivity and can be used to detect MNP in water without electrode passivation affecting sensitivity.

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#### Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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