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# Mechanism Investigation and Electrochemical Assay of Dextromethorphan based on (CuO-CNPs) Composite

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**Abstract**- A novel type of modified electrode was developed to elucidate how dextromethorphan (DM) undergoes oxidation and to measure it with very high sensitivity using voltammetry. To make the sensor, a mixture containing copper oxide and carbon nanoparticles (CuO/CNPs) was prepared and then deposited as a thin layer on the surface of a glassy carbon electrode (GCE). The CuO nanoparticles were synthesized via a straightforward and environmentally friendly hydrothermal method. Comprehensive characterization using scanning electron microscopy and electrochemical impedance spectroscopy confirmed the successful fabrication and enhanced properties of the CuO/CNPs/GCE. The synergistic combination of the large surface area of the CNPs and the superior electrocatalytic activity of the CuO NPs resulted in a remarkable amplification of the DM oxidation signal. Compared to a bare GCE, the modified electrode showed more than 30 times higher peak current, and the oxidation overpotential was reduced by 70 mV, which means the reaction happens more easily. After carefully adjusting the experimental conditions, the way DM oxidizes at CuO/CNPs/GCE was examined. The sensor turned out to be very effective for measuring DM, with a linear response in the concentration range of 0.07-20.0 µM and a detection limit as low as 53 nM. To see if it works in real-life situations, the CuO/CNPs/GCE was also used to measure DM in commercial pharmaceutical products. The results were accurate and reliable, showing that this method can be useful for practical analysis.

**Keywords-** Dextromethorphan, Copper oxide; Carbon Nanoparticle; Glassy Carbon Electrode; Voltammetric; Analysis

## 1. INTRODUCTION

Allergies represent a global health concern and are among the most common reasons for individuals to seek medical care. These conditions are frequently triggered by aeroallergens, which adversely affect the respiratory system and initiate allergic responses [1]. Typical manifestations include symptoms resembling the common cold and chronic cough, often associated with influenza. These respiratory ailments are predominantly linked to viral infections in the upper airways and are particularly prevalent in pediatric populations [2,3]. Standard therapeutic regimens often involve the administration of antihistamines, decongestants, or a combination of both. However, the misuse of pharmaceutical drugs, including legally available over-the-counter options, remains a persistent global public health challenge [4].

Therapeutically, Dextromethorphan (DM) is known in medicine as an effective cough suppressant resulting from throat and bronchial irritation. Doctors usually prescribe it when someone has a dry cough that doesn't bring up any mucus, which often happens when the throat or airways get irritated [5]. It acts centrally by suppressing the cough reflex within the medulla oblongata of the brain, thereby effectively alleviating cough and associated respiratory symptoms [6]. DM lacks opioid-like analgesic properties and addiction potential. Its favorable safety profile and efficacy have established it as a replacement for codeine in over-the-counter cough and cold products for over four decades [7]. Nevertheless, when ingested in substantially higher than therapeutic doses, DM can induce serious side effects. In such high doses, it starts to act like hallucinogenic drugs, comparable to those of phencyclidine and ketamine [8, 9]. The toxicity of DM is notably dose-dependent, characterized by distinct "plateaus" of effects. These range from mild stimulation at the first plateau, to states resembling alcohol and marijuana intoxication at the second, and progress to full dissociative experiences at the third and fourth plateaus, similar to recreational doses of ketamine [3]. This unique psychoactive profile, coupled with its low cost and legal status, has made DM an attractive substance of abuse for adolescents and young adults.

From an analytical perspective, various methods have been reported for determining DM. These include several electrochemical approaches, as well as techniques such as chromatography [10-12], conductometry [13], potentiometry [14,15], and spectrophotometry [16,17]. While many of these methods are sensitive and selective, yet they suffer from notable shortcomings that limit their practical application, such as complex sample preparation, the requirement of highly trained personnel, expensive instrumentation, and time-consuming procedures [18]. These limitations restrict their utility for routine analysis. In contrast, electrochemical methods present a compelling alternative. They are cheap, easy to use, and fast. They can pick up multiple targets at once with great accuracy, and they actually work outside the lab-on real samples [19].

Over the past ten years, electrodes modified with carbon nanoparticles (CNPs) have emerged as a fundamental tool in the field of electroanalysis [20]. This widespread adoption is attributed mainly to the ability of CNP-based platforms to facilitate cost-effective, rapid, highly sensitive, sufficiently selective, and versatile quantification methods for a diverse range of analytes [21]. Their exceptional suitability as an electrode modification material can be traced back to a few important characteristics that complement each other. They conduct electricity well, they resist chemical changes, and they offer plenty of surface area for reactions [22]. Furthermore, CNPs exhibit significant resistance to fouling and possess a strong adsorption capacity for various target molecules, collectively enhancing their analytical performance [23].

For this research, we aimed to develop an electrochemical sensor capable of detecting DM sensitively in aqueous environments. To achieve this, we fabricated a modified electrode by coating a glassy carbon electrode (GCE) with a nanocomposite film consisting of copper oxide and carbon nanoparticles (CuO/CNPs). The incorporated CuO/CNPs nanocomposite was able to facilitate electron transfer kinetics at the electrode interface significantly, thereby enhancing the oxidation peak current of DM. To our knowledge, the present work represents the first account of an electrochemical sensor utilizing the CuO/CNPs nanocomposite for DM determination. Key experimental parameters influencing the oxidation signal were systematically investigated and optimized. Under these optimized conditions, the proposed sensor demonstrated satisfactory performance for quantifying DM across a wide concentration range.

## 2. EXPERIMENTAL SECTION

### 2.1. Materials and reagents

Dextromethorphan hydrobromide came from TEMAD Co. (Iran). Carbon nanoparticles (CNPs, Emperor 2000, Cabot Corporation) with an average diameter of 9–18 nm were consumed as received, without any purification. The buffer solution (Britton-Robinson (BR, 0.04 M) was made by combining glacial acetic acid, boric acid, and orthophosphoric acid. From this stock solution, the pH of working buffers was adjusted by adding appropriate amounts of 0.2 M NaOH. For the analysis of real samples, dextromethorphan HBr tablets (15 mg per tablet, Pursina Pharmaceutical Co.) were procured from local pharmacies in Tehran.

### 2.2. Apparatus

Electrochemical experiments were conducted via a SAMA 500 potentiostat/galvanostat (I. R. Iran). A conventional three-electrode cell was employed throughout this work. This included a GCE (2.0 mm diameter, Azar Electrode Co., Iran) as working electrode, a calomel reference electrode (saturated, SCE), and a platinum wire counter electrode. A digital pH meter (Milwaukee MI 180, Romania) was employed to measure and adjust the pH of solutions.

Furthermore, the surface morphology of the synthesized copper oxide nanoparticles and the modified electrode was characterized by scanning electron microscopy using a Philips XL30 instrument (Netherlands).

### 2.3. Fabrication of the CuO/CNPs/GCE

The fabrication of the CuO/CNPs/GCE involved several steps. First, the bare working electrode was polished on a soft cloth using 0.1 μm alumina slurry. It was then rinsed with water, immersed in deionized water, and sonicated for 2.0 minutes to ensure a clean surface. Separately, a homogeneous dispersion of CNPs was prepared by adding 4.0 mg of CNPs to 4.0 mL of DMF and subjecting the mixture to ultrasonic agitation for approximately 25 minutes. This produced a uniform black suspension. An appropriate volume of the suspension of CNPs was then drop-cast onto the cleaned GCE surface. The electrode was subsequently placed in an oven at 60 °C to dry. Following this, the CNPs-modified GCE was immersed in a 1.0 mM $CuCl_2$ solution, and a constant potential of -0.4 V was applied for 10 seconds to electrochemically deposit copper onto the electrode surface. Then, the electrode was several times rinsed with distilled water, transferred to an alkaline solution (0.1 NaOH), and 20 multiple cycles of voltammetry (CV) tests were recorded across a potential span extending (-0.1 - 1.0 V), at a constant sweep speed of 100 mV/s. During CV cycling in alkaline solution in the positive potential, the metallic copper on the CNPs/GCE surface was oxidized and converted to $Cu^{2+}$ ion, which reacts immediately with the hydroxyl ions adjacent to the electrode surface and is converted to copper hydroxide/oxide and covered the surface of the CNPs/GCE [24]. Before the main experiment, successive cycling of the GCE modified with CuO/CNPs, was performed in a buffered solution at pH 7.5, with the potential scanned between -0.5 V and +1.0 V at a rate of 50 mV/s, which helped to gain more reproducible voltammetric results.

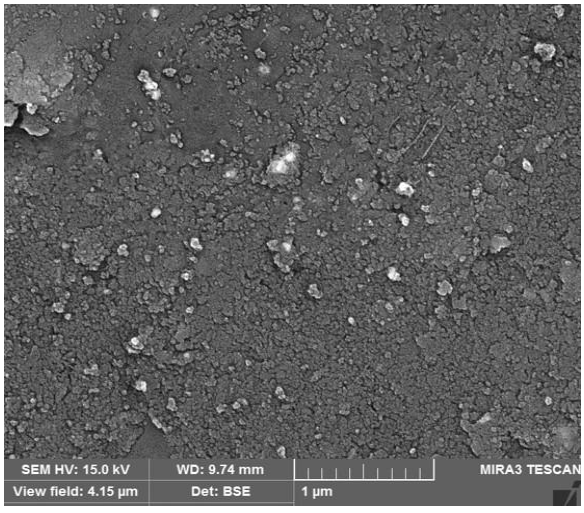


**Figure 1.** SEM images of CuO/CNPs/GCE

## 3. RESULTS AND DISCUSSION

### 3.1. Surface morphology of the CuO/CNPs/GCE

The surface features of the fabricated CuO/CNPs/GCE was characterized via scanning electron microscopy (SEM). Figure 1 shows that the CuO/CNPs nanocomposite forms a uniform film on the GCE surface. This homogeneous distribution creates a textured, rough topography that noticeably increase the electroactive surface area and provides a high density of active sites for electrochemical processes.

### 3.2. Electrochemical behavior of DM at CuO/CNPs/GCE

The electro-oxidation of DM on the surface of various electrodes was evaluated via cyclic voltammetry (CV, 100 mV/s) in a BR buffer solution of pH 7.5. Figure 2 shows CVs recorded by the bare GCE and modified GCE with different nano-structures, including CNPs and CuO/CNPs recorded in the 10 µM DM solution (buffer, pH 7.5). The electro-oxidation of DM takes place through an irreversible process at the bare and modified GCEs. The oxidation peak potential ($E_p$) of DM was appeared at 0.900, 0.863, and 0.829 V at GCE, CNPs/GCE, and CuO/CNPs/GCE, respectively. The oxidation peak current ($I_p$) of DM was equal to 0.45, 11.89, and 13.54 µA at bare GCE, CNPs/GCE, and CuO/CNPs/GCE, respectively. An enhancement of peak current together with the over-potential reduction of the DM electro-oxidation was observed after GCE surface modification. These evidences reveal the electro-catalytic effect of the CNPs and nano CuO that is mainly due to increment of the conductivity and efficient area of the electrode surface, and acceleration of the electron transfer between the electrode and DM molecules.

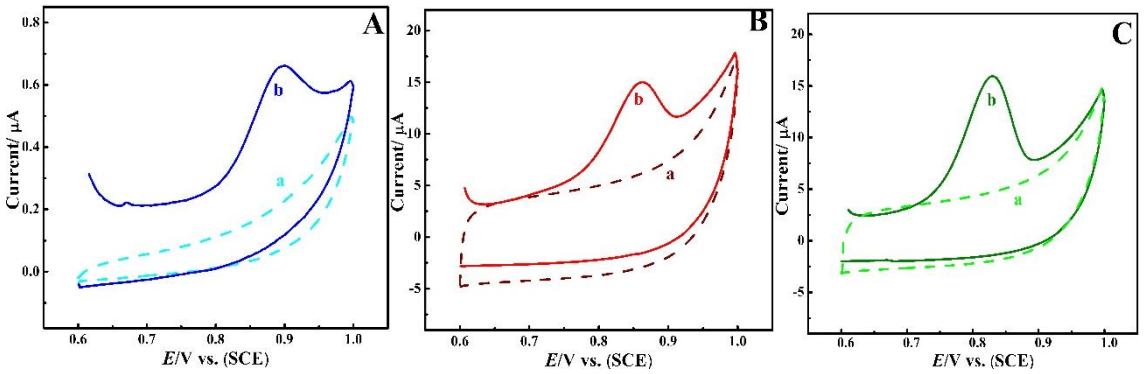


**Figure 2.** CVs of (A) bare GCE, (B) CNPs/GCE, and (C) CuO/CNPs/GCE in pH 7.5 BR buffer (0.04 M) without (a) and with (b) 10 µM DM

Previous studies indicated that the DM oxidation is affected by the pH of the supporting electrolyte [1, 25-30]. Hence, the role of pH value affecting voltammetric response of 10 µM DM was evaluated in the pH span of 5.0 to 10.0 by CV, Fig. 3A. The anodic peak current

gradually enhanced following the change of pH values from 5.0 to 7.5 and then reduced, Fig. 3B. The anodic potential was displaced toward less positive potentials according to the Eq (1), Fig. 3B. The obtained slope of 66 mV/pH, revealed that DM oxidation involved the same number of electrons and protons at CuO/CNPs/GCE. In further experiments, the pH of the supporting electrolyte was set at 7.5 to gain the highest sensitivity.

$$E_p = -0.066\ pH + 1.34 \qquad (R^2=0.993) \qquad (1)$$

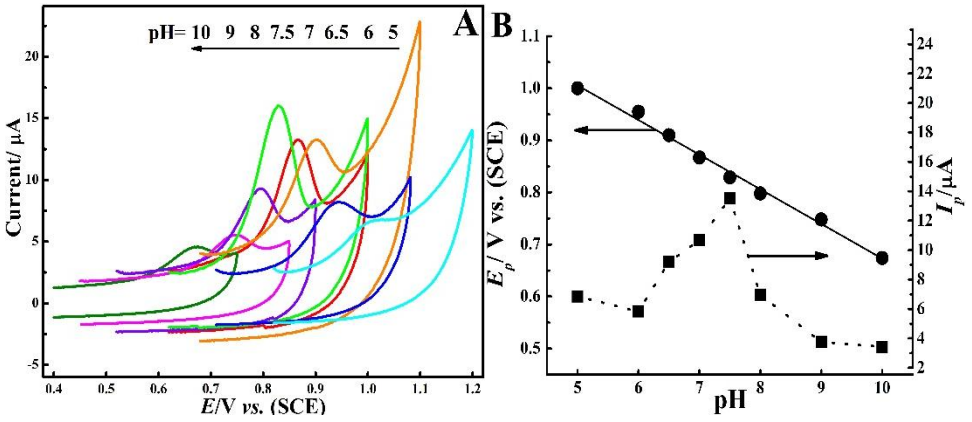


**Figure 3.** (A) CVs of 10 μM DM at CuO/CNPs/GCE in pH 5.0-10.0 buffer. (B) (●) Effect of pH on $E_p$ and (■) $I_p$

### 3.3. Influence of potential sweep rate

To investigate the kinetics of the DM electro-oxidation process, the influence of the potential scan rate was examined. Figure 4A presents cyclic voltammograms of 100 μM DM in a 0.04 MM BR buffer (pH 7.5) at sweep rates of 5 - 200 mV/s. The oxidation peak current increased at a constant rate with increment of the sweep rate (Eq. (2), Fig. 4B). In addition, the plot of Log($I_p$) vs. Log(υ) yielded a straight line with a slope of 0.8 (Eq. (3), Fig. 4C). This slope value suggests that a mixture of adsorption and diffusion phenomenon controls the DM oxidation process. This behavior is attributed to the initial adsorption of DM molecules onto the CuO/CNPs/GCE surface, facilitated by л-л interactions, followed by diffusion through the porous nanocomposite film.

$$I_p = 0.07\ \upsilon + 0.81 \qquad (R^2=0.994) \qquad (2)$$

$$\text{Log}\ I_p = 0.80\ \text{Log}\ \upsilon - 0.68 \qquad (R^2=0.998) \qquad (3)$$

Furthermore, a positive shift in the peak potential was resulted after increment of sweep rate, and a linear dependence was observed between $E_p$ and log υ, revealing that the DM oxidation is limited kinetically and obeys of an irreversible behavior.

The polarization curves (log *I vs. E* plots, Eq. (4)) for the DM electro-oxidation at CuO/CNPs/GCE were plotted at different potential scan rates. The a in Eq. (4) is a constant

value while b is equal to $(1-\alpha)n_aF/2.303RT$). Here, the mean amount of 0.69 was obtained for α revealing the activation free energy curve for DM electro-oxidation is non-symmetrical.

$$\text{Log } I = a + b\,E \qquad (I\text{: A, } E\text{: V}) \qquad (4)$$

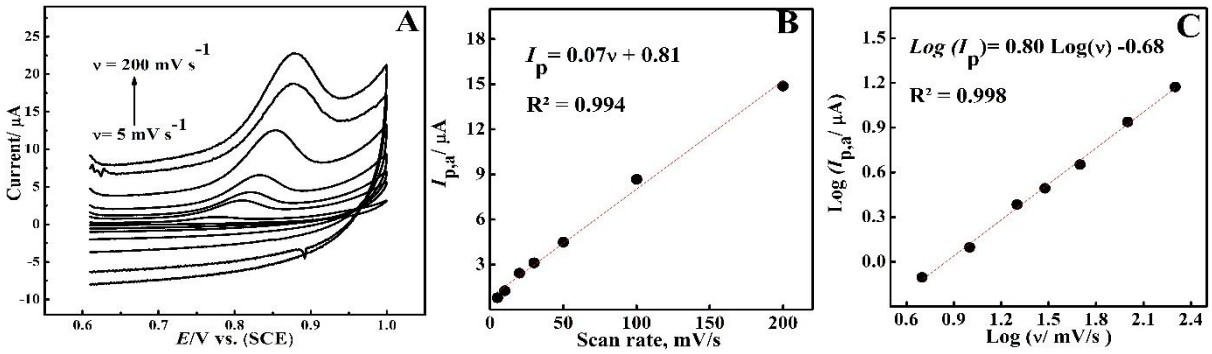


**Figure 4.** (A) CVs of 100 µM DM at CuO/CNPs/GCE in pH 7.5 buffer at different scan rates. The plots of (B) $I_p$ *vs.* scan rate and (C) Log ($I_p$) *vs*. Log (ν)

### 3.4. Optimization of the effective variables

As mentioned the GCE modified with CuO/CNPs composite improved the DM electro-oxidation. To achieve best performance for DM analysis, the volume of CNPs suspension used for the GCE surface modification and the pre-concentration time of DM at the electrode surface were optimized. At first, DM oxidation response was tested at the GCE modified with various volumes of CNPs suspension having a concentration of 1.0 mg/mL, Fig. 5A.

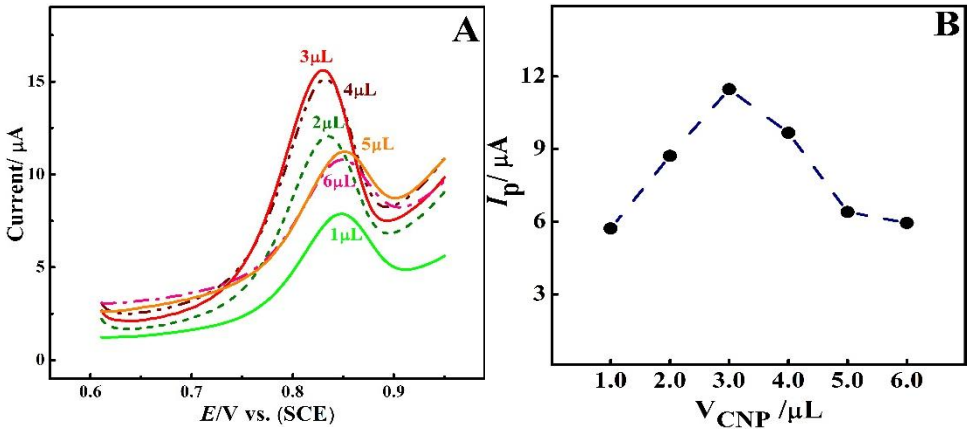


**Figure 5. (A)** LSVs of 10 µM DM at CNPs/GCE made with different volumes (1-6 µL) of CNPs suspension (pH 7.5 buffer. No accumulation). (B) Plot of $I_p$ versus drop size of CNPs suspension

The recorded LSVs showed an increment of DM peak current after covering the GCE surface with a greater volume of CNPs suspension up to 3.0 µL, followed by the reduction of peak current with further addition of CNPs, Fig. 5B. Hence, 3.0 µL of the CNPs suspension was used for the GCE modification.

Evaluation of the voltammograms revealed the adsorption of DM molecules on the CuO/CNPs/GCE surface that led to the enhancement of peak current. Therefore, the CVs of 10 µM DM solution were recorded under various pre-concentration times up to 700 s, Fig. 6A. The graph of peak currents of DM versus accumulation time showed a sharp current increment up to 400 s followed by a slow change with further increasing the accumulation time, which reached to the almost steady amount after 600 s, Fig. 6B. Hence, the pre-concentration step was set at 600 s in subsequent experiments.

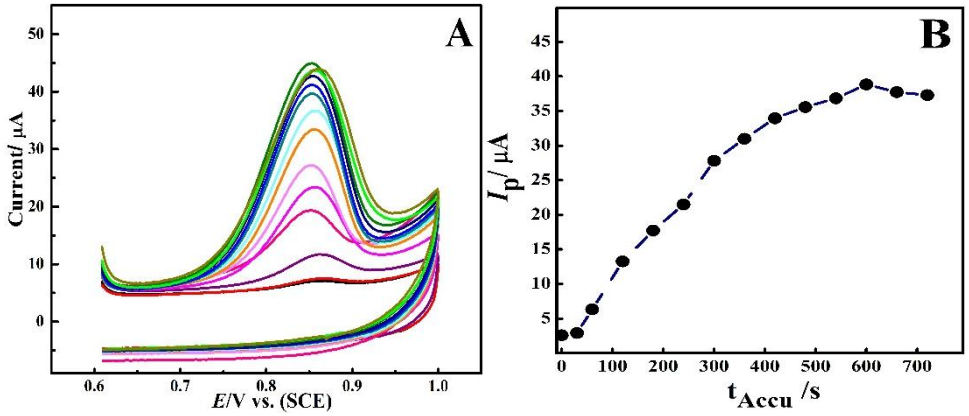


**Figure 6.** (A) CVs of 10 µM DM at CuO/CNP/GCE in pH 7.5 buffer recorded under various accumulation times. (B) $I_p$ *vs.* accumulation time plot

### 3.5. Analytical characteristics

Figure 7A displays the LSVs of various concentrations of DM recorded by CuO/CNPs/GCE. In these study the anodic peak current of DM showed a linear relation with the corresponding concentrations in the range of 0.07 - 20.0 µM, according to Eq. 5, Fig. 7B. The LOD of 53 nM was resulted for DM quantification based on 3σ/m.

$$I_p\,(\mu A) = 3.42\ C_{DM}\,(\mu M) + 3.21 \qquad (R^2 = 0.996) \qquad (5)$$

Analytical response properties of the CuO/CNPs/GCE were compared with previous electrochemical reports. As can be seen in Table 1, in comparison to most of the developed modified electrodes, the modified CuO/CNPs/GCE obviously showed a wider linear response range and a smaller limit of detection for DM analysis. CuO/CNPs/GCE has some advantages such as antifouling behavior, nontoxicity, high stability, and good repeatability besides its fast and simple fabrication.

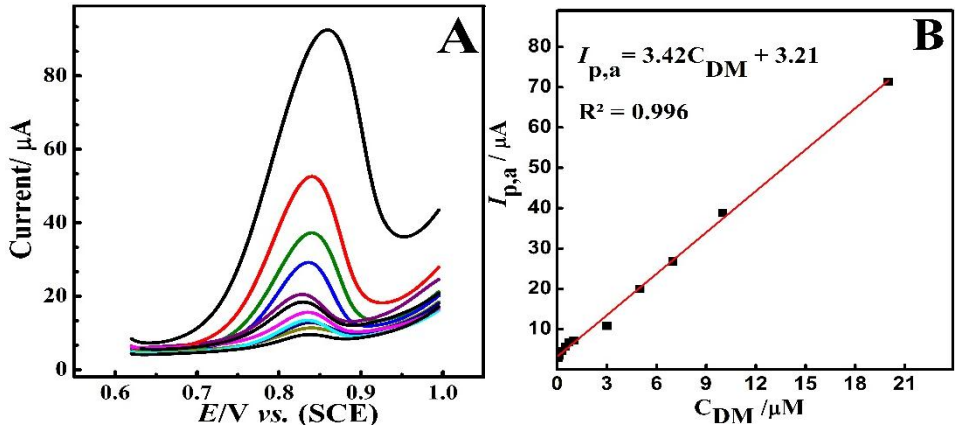


**Figure 7.** (A) LSVs of 0.07-20 µM DM at CuO/CNP/GCE (pH 7.5 buffer, accumulation 600 s, scan speed 100 mV/s). (B) Calibration plot of $I_p$ *vs* MD concentration

The stability of the CuO/CNPs based sensor was evaluated via LSV analysis of 1.0 µM DM. Successive LSV analysis in one day and also repeated measurements in consecutive days were performed, which showed less than 2.9 and 4.9 % reduction in the peak current of the first recorded LSV, respectively, with the freshly prepared modified electrode. These results confirmed the acceptable stability and repeatability of the peak currents CuO/CNPs nano-composite-based sensor developed for DM measurement.

**Table 1.** Comparison of the performance of the modified electrodes for DM analysis

| Sensor | Method | Liner range (µM) | LOD (nM) | Ref. |
|---|---|---|---|---|
| CNT-CµPIL[1] | CV | 30-3300 | 8800 | [27] |
| PDDA/MWCNT/CD/PGE[2] | DPV | 2-600 | 1900 | [29] |
| RGO/SPCE[3] | DPV | 0.0135-4.05 | 5 | [25] |
| CNP/CPE[5] | DPV | 8-800 | 2900 | [5] |
| SPCE | DPV | 2.97-198.75 | 1000 | [30] |
| MnHCF/CS/SPCE[6] | SWV | 0.062-240 | 9 | [1] |
| LINGE[7] | DPV | 2.5-400 | 1800 | [19] |
| CuO/CNPs/GCE | LSV | 0.07-20 | 53 | This Work |

[1]CNT-CµPIL: Carbon nanotube-carbon micro-particle-ionic liquid composite
[2]PDDA/MWCNT/CD/PGE: Poly diallyl-dimethyl ammonium chloride/multiwall carbon nanotubes/carbon quantum dots/pencil graphite electrode.
[3]RGO/SPCE: Reduced graphene oxide/screen-printed carbon electrode
[4]BNNS: Bird nest-like nano-structure
[5]CNP/CPE: Carbon nanoparticles/carbon paste electrode
[6]MnHCF/CS/SPCE: Manganese hexacyanoferrate/chitosan/SPCE
[7]LING: Laser-induced nitrogen-doped graphene electrode.

### 3.6. Interference studies

The analytical accuracy of the sensor, particularly in complex matrices, is of paramount importance for clinical applications. To assess the selectivity of the proposed sensor, the

potential interference from common inorganic ions and biomolecules was investigated. The results demonstrated excellent selectivity, as a 100-fold excess of ions such as $NO_3^-$, $Cu^{2+}$, $K^+$, $Na^+$, $Cl^-$, $Li^+$, and $SO_4^{2-}$ caused no significant interference in the determination of DM. Furthermore, the sensor's capability for simultaneous analysis was confirmed in a mixture containing 1.0 mM of ascorbic acid (AA), dopamine (DA), and uric acid (UA), and 10 µM DM, where three well-resolved peaks were observed in the LSV measurements, Fig. 8. This distinct separation of voltammetric signals confirms the high selectivity of the CuO/CNPs/GCE and its strong potential for the accurately analyzing DM in complex, real-world clinical samples.

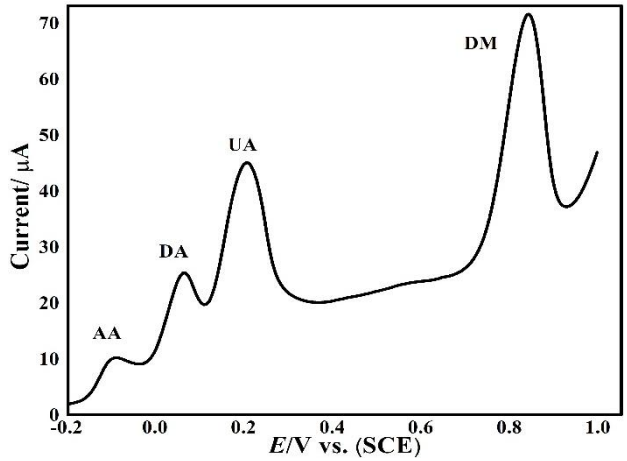


**Figure 8.** LSVs of 10 µM DM in the presence of 1.0 mM AA, DA, and UA in 0.04 M BR buffer solution (pH 7.5); accumulation time 600 s, scan rate 100 mV/s

### 3.7. Quantification of DM in pharmaceutical preparations

The real sensing ability of CuO/CNPs/GCE was evaluated via the analysis of DM in pharmaceutical product using standard addition method. The stock solution of DM HBr tablet was prepared using an adequate amount of fine powder of pre-weighted and ground DM HBr tablets. Then the given diluted DM solutions were prepared and spiked with DM standard solution in the span of 0.4–10 µM. The LSVs of the final DM solution were recorded and the plot of $I_p$ *vs.* spiked DM concentrations was drawn. The average amount of 14.3 mg/tablet was resulted for DM HBr content with a RSD of 6.2 % (n= 3) showing the accurate real sensing ability of the developed sensor.

## 4. CONCLUSION

Combination of the nanoparticles of carbon and copper oxide led to preparation of a powerful composite useful in electrochemical sensor construction. The GCE surface was modified by a thin layer of CuO/CNPs for which SEM characterization confirmed the good

dispersion of nanostructures on the electrode surface without significant electron transfer resistance. The CuO/CNPs modifier increased the effective surface area of the GCE. The CuO/CNPs-modified GCE enabled the fast and reliable determination of DM. Effective adsorption of DM molecules on the CNPs through л-л interaction effectively enhanced the DM oxidation response. Employing the LSV technique, the CuO/CNPs/GCE exhibited excellent performance for DM analysis over the concentration range of 0.07–20 μM, with an extremely low LOD of 53 nM, satisfactory selectivity, and recovery in pharmaceutical preparations.

**Acknowledgments**

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**Declarations of interest**

The authors declare no conflict of interest in this reported work.

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