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***Research Article***

# INVESTIGATION ON THE ENVIRONMENTAL SENSORS DESIGNING TO MEASURE PARAXON BY MODIFIED ELECTRODES OF GLASSY CARBON THROUGH NANO-PARTICLES OF CADMIUM OXIDE AND ACETYLCHOLINE ESTERASE

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## ABSTRACT
Regarding this fact that the measurement of acetylcholine esterase (AChE) is done by medical method and this method is time-consuming and costly, so creating a sensor for fast and easy measurement with low cost is essential in our society due to excessive use of pesticides. It can be say that biosensors could be replaced tby the present analytical methods. Enzyme-based electrochemical biosensors because of their high sensitivity, fast response, and miniature size are appropriate as a promising alternative to pesticides. Through modifying glassy carbon electrode surface with cadmium oxide nanoparticles and acetylcholinesterase and discussed the electrochemical behavior of this protein structure the possibility of paraxon toxin concentration detection and measurement has been provided from electrochemistry. Cadmium oxide nanoparticles were synthesized by chemical methods in the laboratory and this claim that this synthesized Nano particles are cadmium oxide, was confirmed by using X-ray (XRD). Spectrum Uv -vis of cadmium oxide nanoparticles has shown the absorption in 380nm area. So, the synthesized nanoparticles have quantum property and the property is because of an increase in the size of the nanoparticles to the content.

***Keywords:*** *Cadmium Oxide Nanoparticles, Paraxon, Carbon Paste Electrode and Acetylcholinesterase*

## INTRODUCTION
We have done the evaluation of paraxon measurement using modified carbon paste electrode with cadmium oxide nanoparticles and acetylcholine esterase. So, in this section, a detailed discuss would be provided for the main material of the research.

### *Physical and Chemical Properties of Cadmium*
Cadmium is more considering because of creating environmental pollution. So that just 2 grams is enough to kill people. On the other hand, no effective biological role has been known for cadmium.
However, it has many applications in industry. Cadmium is a soft and bluish-white metallic element. It produced from zinc clarification and refining and most of its feature is similar to Zink. Cadmium and its compounds are highly toxic. Naturally, about 25,000 tons of cadmium enters into the environment, annually (Derning, 2005).

### *Cadmium Oxide*
Carbonates and nitrates are formed by Burning the metal in air or as a result of laser. This oxidation is provided of cadmium in smoke from. Cdo color changes from greenish yellow to brown and finally almost black according the accessed heated. These colors are associated with different types of crystal lattice defects. This metal hydroxide is produced from increased alkali to salty solution and all will be in deposit form. Different method should be applied to produce cadmium oxide Nano-structures in different shapes and sizes. So, current and conventional methods were formed to produce metal oxide nanostructures for spreading nanotechnology and still there is an attempt to design new and more complete and complicated methods. Other applications of metal oxide include its applying as the inorganic antibacterial agents in the tile industry that is because of the higher thermal resistance of the agents at the applied temperatures in the industry (Shukla *et al.*, 2012).

***Modified Chemical Electrodes with Nanoparticles***

There has been emerged a new approach in electrode system through modified Electrodes with nanoparticles. These electrodes relates to the placing a reagent on the surface, aimed to use its behavior in a modified surface. Therefore, such a deliberate change could eliminate many of the dialytic electrochemistry problems analysis eliminates many of the problems and provide a basis for new dialytic applications and different sensory devices. As a whole, modifier group should spread on electrode surface to prepare a modified electrode (Huang *et al.,* 2007).

***Acetyl Cholinesterase***

Acetyl cholinesterase (AChE), has two binding and connecting site for acetylcholine. The substrate would bind with enzyme in anionic site, and choline would release in esterification site and the enzyme acetylated. Then the water would be absorbed by the acetylated enzyme and the choline base would change and convert to acetic acid and the enzyme would free.

Acetyl cholinesterase enzyme is bound to choline from its ionic site and would attach to the acetyl from its easter and cause degradation of acetylcholine to choline and acetic acid.

***Paraxon***

Paraxon is a dangerous pesticides, relates to organophosphate group.

Paraxon or O, O diethyl O-4- phosphate Nitrophenyl is the result of parathion oxidation metabolism in the liver. One of the most toxic Pesticides is considered by the provided conversion (P = SP = O). Like other organophosphate, paraxon would inhibit the cholinesterase activity through phosphorylation of serine in the active site and is not able to degrade acetylcholine .Increased acetylcholine would highly stimulate nicotinic and muscarinic receptors, that eventually led to seizures and brain lesions in dramatic situation and at last to death. Paraxon is methabolized in the body by the available paraxonise in plasma and liver (Zhang, 2005).

## MATERIALS AND METHODS

***Materials***

Cetyl trimethyl ammonium bromide (CTAB) as the surfactant, cadmium sulfate 0.03 M ($CdSO_4$), 0.09 M sodium hydroxide (NaOH), acetic acid 0.06 ($CH_3COOH$), toluene, 70% ethanol, acetylcholinesterase enzyme, paraoxon, phosphate buffer (PBS), includes sodium phosphate solution($NaH_2PO_4$ and $Na_2HPO_4$), all applied and used solutions and materials were purchased from Sigma-Aldrich. Deionized distilled water was used to produce all solutions.

***Instruments***

Three electrodes have been used in this research that includes working electrode (glassy carbon by the enzyme acetylcholinesterase), reference electrode (saturated calomel electrode), and a counter electrode (platinum electrode) with a diameter of 4 mm.

Cyclic Voltammetry tests were performed by potentiostat galvanostat Dutch Palm sense. It is connected to the computer that has the software GPES 4/9. This software will record the input data and plots the considered and desired peaks. Each experiment is done 3 times in related concentrations and each time has been a complete tricycle.

TEM and SEM images have been taken by model DSM 960A microscope and CEM 902A of Zeiss company. X-ray diffraction XRD using a Siemens device and X-ray wavelength of CuK (equivalent to A 1. 54056 were analyzed too. UV- Vis spectroscopy device was provided by spectrophotometer Shimadzu UV 160 and spectrum UV – VIS. The required Samples were prepared at the temperature of 25 ° C. a Fixed bed flow and a ceramic boat (length, width, height.8 (1.5 x1)) was used to prepare nanoparticles of cadmium oxide.

## RESULTS AND DISCUSSION

***Results***

The obtained cycled voltammogram was saved by the software Echem stored and transferred to the Excel program. The needed edition done on the charts and then its parameters were extracted (anodic peak area, peak area of the cathode, the anode and cathode). UV- spectroscopy spectrum obtained from software and recorded in Excel and then has been used. Voltammetry peak in this part has been used to characterize the activity of acetyl cholinesterase. Figure 1 has shown of acetyl cholinesterase behavior/ cadmium oxide nanoparticle / glass carbon electrode after adding paraxon, an organophosphate pesticide. After incubation, Biosensor was placed in paraxon standard solution in phosphate buffer (PH = 7) for 12 minutes and then transfer it to a voltammetry cell containing 5 mm thiosulfate acetyl choline chloride Voltamogram than normal courier free of paraoxon at a fixed concentration of acetyl choline chloride thiosulfate showed shorter peak that shows the inhibition of acetylcholinesterase by paraxon and reducing the amount of its enzymatic activity. Paraxon at different concentrations of paraxon was like this**:** $10^{-10}$ M ، $10^{-9}$ M ، $10^{-8.5}$ M ، $10^{-8}$ M ، $10^{-7.5}$ M ، $10^{-7}$ M ، as it is shown in figure 1, it is clear that the current peak decrease more by increasing in paraxon concentrations, since paraxon as a pesticide with intense toxicity would cause irreversible inhibition of acetylcholinesterase and as a result, enzyme activity, would decrease.

The Efficiency of enzyme inhibition by paraxon is obtained from the following equation:

$$\text{inhibition}\ (\%) = \frac{i_{P,\text{control}} - i_{P,\text{exp}}}{i_{P,\text{control}}} \times 100$$

Where $i_{P,\text{control}}$ is the peak of AChE Biosensor / cadmium oxide nanoparticle / glass carbon electrode and $i_{P,\text{exp}}$ is the peak of AChE Biosensor / cadmium oxide nanoparticle / glass carbon electrode with paraxon.

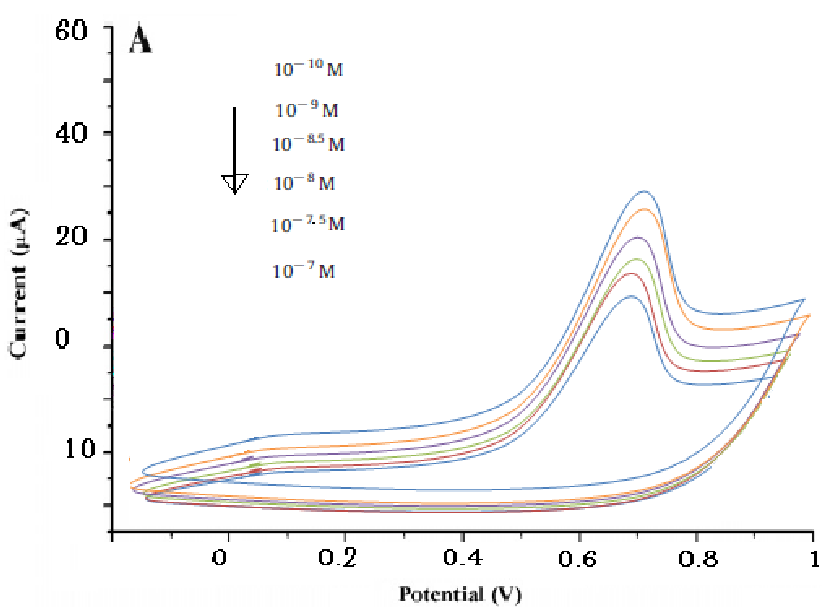


**Figure 1: It has shown the acetyl cholinesterase / cadmium oxide nanoparticle / glass carbon electrode behavior after adding paraxon, an organophosphate pesticide**

As shown in Figure 2, the equation of the linear part of the enzyme inhibition curve by paraxon in the molar concentration of 10(-10) to 10(-7) is I% = (6.453c + 67.46) % .The correlation coefficient is equal to 0.9956. Detection limit was calculated $2.69 \times 10^{-10}$ M .

Acetyl cholinesterase / cadmium oxide nanoparticle / glass carbon electrode is a simple method for the determination of paraxon value and amount due to significant changes in the voltammetry signal.

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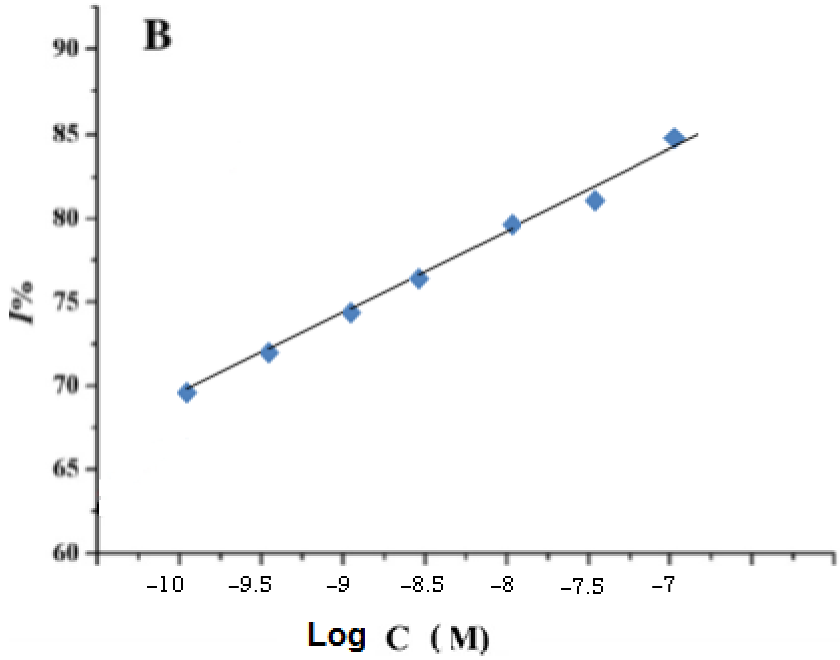


**Figure 2: The linear equation of enzyme inhibition curve by paraxon in concentration area in10 (-7) to 10(-10) moles are equal to I% = (6.453c + 67.46) %**

***Conclusion***

We have synthesized the cadmium oxide nanoparticles, chemically in the lab and In the next research stage, this Nano-particles using a ray diffraction X (XRD) confirmed this claim that this synthesized nanoparticles are of cadmium oxide. Spectrum Uv - vis of cadmium oxide nanoparticles showed the absorption in 380nm area. So the synthesized nanoparticles have quantum property and this property is due to the increase of the size of the nanoparticles to its contents. Direct electrochemistry of acetylcholinestrase is easily provided in glassy carbon electrode and analyzed and clear peaks appear. This electrochemical process will be applied in phosphate buffer solution (PBS) in pH =7.

This Biosensor can be used to detect paraxon Because of easy preparation and appropriate property and characteristics. The proposed approaches for research on this topic are using a gold electrode or electrode graphite or carbon paste electrode rather than a glassy carbon electrode. Other modifier like Nickel Oxide nanoparticles, carbon nanotubes, copper oxide and Zinc Oxide can be used that are appropriate in many ways for the application. Changing the scan rate or the buffer used can be discussed and analyzed.

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