EVALUATION OF ENVIRONMENTAL SENSORS DESIGNING TO MEASURE PARAXON BY MODIFIED ELECTRODES OF GLASSY CARBON USING NANO-PARTICLES OF MAGNESIUM OXIDE AND ACETYLCHOLINE ESTERASE

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ABSTRACT

Since, nowadays the measurement of acetylcholine esterase (AChE) is done in medical method and measurement method is time-consuming and costly, so manufacturing a sensor for fast and easy measurement with low cost is a necessity in our society due to excessive use of pesticides in agriculture for having more products. Biosensors can be replaced the current analytical methods. Enzyme-based electrochemical biosensors are appropriate that are identified as a promising alternative to pesticides, because of their high sensitivity, fast response, miniature size. The possibility of paraxon toxin concentration detection and measurement has been provided from electrochemistry by modifying glassy carbon electrode surface with Magnesium oxide nanoparticles and acetylcholinesterase and discussed the electrochemical behavior of this protein structure. Magnesium 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 Magnesium oxide nanoparticles has shown the absorption in 380nm area. So, the synthesized nanoparticles has quantum property and the property is because of an increase in the size of the nanoparticles to the content.

Keywords: Magnesium Oxide Nanoparticles, Paraxon, Carbon Paste Electrode and Acetylcholinesterase

INTRODUCTION

The researchers have done the evaluation of paraxon measurement using modified carbon paste electrode with Magnesium oxide nanoparticles and acetylcholine esterase.so, in this section, a detailed discuss would be provided for the main material of the research.

Carbon Electrodes

The carbon-based electrodes are widely used for electrochemical analysis. This is due to the wide range of their potential to lower the cost of inaction on chemical, sensory applications and suitable for diagnostic. In contrast, the electron transfer rate observed in the surface carbon is often less than the rate observed in the metallic electrodes. The electron transfer activity is affected by the origin and history of the carbon surface (Mccreey, 1996).

Glassy Carbon Electrodes

Glassy carbon (or glass-like) is known due to its good mechanical properties and a wide array of potential ineffectiveness of the chemical (solvent resistance) and relatively repeatable performance (Bokros, 1977). Its Ingredient controlled by a carefully heat and produced by previous modeled polymer resin (phenol Frmalvyyd) in atmosphere (Wang, 1984). The structure of glassy carbon include delicate interwoven strips of crosslinked graphene sheets. Because of the high density and small pores doesn't have special procedures to fill the pores.

However, pretreatment of the glassy carbon electrode used to create and increase efficiency Manal and repeatable analysis (Kong, 2003).

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

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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).

Purposes of Research

1. To identify and measure the paraxon concentration by the modified electrode with Nano materials and to analyze the chemical behavior of acetylcholinesterase on this electrode

2) Using a modified glassy carbon electrode with nanoparticles of Magnesium oxide and acetylcholinesterase to measure the paraxon toxic

MATERIALS AND METHODS

Required Materials

Cetyl trimethyl ammonium bromide (CTAB) as the surfactant, Magnesium sulfate 0.03 M (CdSO4), 0.09 M sodium hydroxide (NaOH), acetic acid 0.06 (CH3COOH), toluene, 70% ethanol, acetylcholinesterase enzyme, paraoxon, phosphate buffer (PBS), includes sodium phosphate solution(NaH2PO4 and Na2HPO4), 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), 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 $^{\circ}$ 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

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

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recorded in Excel and then has been used. Voltammetry peak in this part, has been used to characterize the activity of acetyl cholinesterase. Figure (8-3) ha shown of acetyl cholinesterase behavior/ Magnesium 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: $10^{-7.5}M$

 10^{-10} M, 10^{-9} M, 10^{-8} M, 10^{-7} M, 10^{-7} M, 10^{-7} M, 10^{-7} M, 10^{-10} M, 10^{-1

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

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

Where $^{ip, control}$ is the peak of AChE Biosensor / Magnesium oxide nanoparticle / glass carbon electrode and $^{ip, exp}$ is the peak of AChE Biosensor / Magnesium oxide nanoparticle / glass carbon electrode with paraxon.

Acetyl cholinesterase / Magnesium 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.

Magnesium Oxide Nanoparticles have a very high surface to volume ratio and this factor has made a big impact on the exchange of electrons between the nanoparticles to glassy carbon electrode. In this figure Voltagram a is related to row carbon electrode and expresses the fact that there is no redox peaks for a Voltagrama. In this figure, Vltagram b is related to carbon nanoparticles of magnesium and represents a redox peaks due to improved electron transport with regard to the use of nanoparticles modified electrode surface.

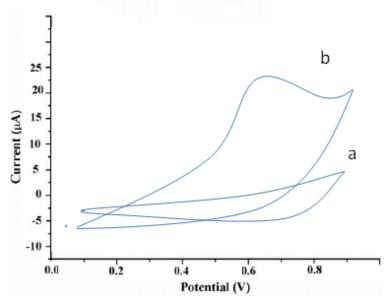


Figure 1: The acetyl cholinesterase / Magnesium oxide nanoparticle / glass carbon electrode behavior after adding paraxon, an organophosphate pesticide

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In next part, there would be investigation on the electron transport properties of acetylcholinesterase on carbon paste electrode with nano-magnesium oxide. In Figure 2 (B & A), a linear dependence is observed between the anodic scan rate so that the redox peak currents increased linearly with scan rate. The oxidation peak current (ipa) is proportional to the scan rate. Scanning speeds were used, respectively, from low to high mountain peaks, respectively, 100, 200, 300, 400, 500, 600, 700 and 800 mV on second. The correlation coefficient is equal to 0.9971 for the anodic peak. This phenomenon refers to the fact that the process of redox species is controlled adsorbed on the electrode surface and is the expression of stable acetyl cholinesterase on the electrode surface.

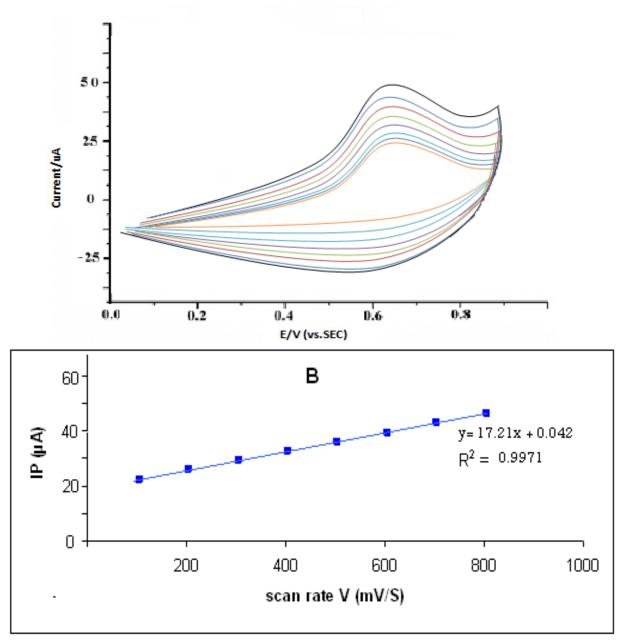


Figure 2: (A) sweep speeds from low to high peak to peak, respectively, 100, 200, 300, 400, 500, 600, 700 and 800 mV per second; and (B) the linear dependence anodic peak (blue line) with the scan rate



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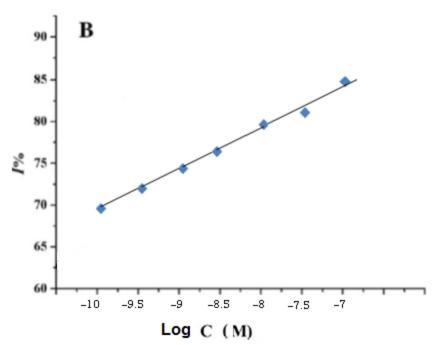


Figure 3: Linear equation of the curve in the range of inhibition by paraoxon concentration in moles equal to I% = (6.453c + 67.46)%

Conclusion

In this study, the effect of pH on electron transfer between the enzyme acetylcholinesterase immobilized carbon paste electrode was investigated and magnesium oxide nanoparticles was synthesized by chemical methods on the fixed surface carbon and increases the rate of electron transfer between thio colin and Carbon electrodes. Formation of nanoparticles depends on the surfactant and organic solvent and like the surfactant help to adhere nanoparticles synthesized stems on the surface. So by this operation, the stabilization of the particles and the formation or growth of the core particles to achieve a high degree of uniformity. Nanoparticles synthesized were studied in bulk and morphological by using ultraviolet spectrometer, X-ray diffraction and scanning electron microscopy. 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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