

Synergistic Integration of Chemically Modified Graphene and Silver Nanoparticles for Highly Sensitive Acetylcholinesterase-Based Biosensors in Pesticide Detection

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Abstract

Biosensors have become indispensable instruments in modern analytical research, with the opportunity to significantly transform diverse domains such as environmental monitoring and food safety. This study aims to discuss the urgent need for precise and reliable pesticide detection, focusing on the difficulties encountered by current methodologies. Pesticides serve a crucial function in agriculture, although their residues pose significant risks to human health and the environment if they exceed the established thresholds. The existing detection techniques often encounter sensitivity and selectivity challenges, necessitating the exploration of more sophisticated strategies. Given these obstacles, this research provides a unique Nanoparticle-based Biosensors Design for Pesticide Detection (NP-BSD-PD) approach. The proposed methodology integrates chemically modified graphene and silver nanoparticles, leveraging their distinctive characteristics to augment the sensitivity and accuracy of pesticide detection. The technology utilizes biosensors based on acetylcholinesterase, enabling the detection of trace levels with high sensitivity. The experimental research yielded findings indicating that NP-BSD-PD exhibited exceptional performance across several parameters. The technique has remarkable sensitivity and accuracy, as shown by the Zeta Potential of -24.89 mV, pH of 7.4, Voltage of 2.7 V, Current of 48.89 μ A, Inhibition Percentage of 88.56%, and a Detection Limit of 0.3 ng/mL. The results highlight the potential of NP-BSD-PD as a viable technique for detecting pesticides. This method effectively addresses significant limitations present in existing approaches, facilitating advancements in food safety and environmental preservation.

Keywords: Biosensors, Pesticide Detection, Nanoparticles, Analysis.

Introduction to Biosensors and Pesticide Detection

Biosensors have become essential in various disciplines, including medical diagnostics and environmental monitoring [1]. These analytical instruments provide real-time, empathetic, and specialized detection capabilities, making them very beneficial for multiple applications. The need for biosensors has seen substantial growth in the current dynamic environment of analytical chemistry. The demand for accurate and effective biosensors has become more imperative, especially in pesticide identification [2].

Pesticides are of utmost importance in contemporary agricultural practices as they protect crops and guarantee the stability of the food supply [3]. The widespread use of pesticides has generated apprehensions over their ecological ramifications and possible risks to human well-being. The current situation has emphasized the need to establish dependable and

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highly responsive techniques for detecting pesticides [4]. The precise measurement of pesticide residues in diverse matrices, including soil, water, and food items, is crucial for guaranteeing adherence to safety requirements and minimizing the negative consequences of pesticide exposure [5].

The current techniques used for pesticide identification often encounter many obstacles. Traditional methods involve significant time investment, extensive manual effort, and costly equipment [6]. A substantial limitation of most of these methodologies is their inadequate sensitivity and specificity in detecting pesticides at minute concentrations. This deficiency is of utmost importance as it hinders the accurate evaluation of their ecological consequences and food safety assurance [7]. The intricate nature of pesticide residues found in samples from the actual world is a substantial obstacle to traditional analytical methods [8].

To overcome these constraints and cater to the increasing need for pesticide detection technologies that are extremely sensitive and efficient, the study proposed a Nanoparticles-Based Biosensors Design for Pesticide Detection (NP-BSD-PD) methodology. The use of nanoparticles in NP-BSD-PD aims to optimize the functionality of biosensors by capitalizing on their distinct characteristics. Nanoparticles provide several benefits, including a notable surface-to-volume ratio, exceptional conductivity, and adjustable features [9-10]. The attributes render nanoparticles very suitable for enhancing the sensitivity and selectivity of biosensors, hence facilitating the accurate identification of pesticides in intricate matrices.

The primary contributions of the research are given below:

- The use of nanoparticles is employed to enhance sensitivity, hence facilitating the accurate detection of pesticides even in samples of a complex nature.
- The individual has exceptional proficiency in quantifying pesticide residues at trace levels, which is of utmost importance in conducting comprehensive environmental monitoring and ensuring the safety of food products.
- This technology enhances the efficiency of the analytical process, reducing the need for time-consuming methods and resulting in quicker and more cost-effective detection.
- It significantly promotes safer and more sustainable pesticide practices by addressing the environmental and health issues related to their utilization.

The following contents are organized in the given manner: In Section 2, a lot of literature is reviewed about biosensors and other ways to find pesticides. The suggested NP-BSD-PD method is explained in Section 3, focusing on its unique qualities. In Section 4, the results and effects of the experiment are broken down in great depth. In Section 5, the study comes to an end. It discusses the most critical developments and suggests future research topics in finding pesticides.

Literature Survey and Findings

The literature review explores the field of pesticide detection and biosensors, analyzing current methodologies, obstacles, and progressions. This concept provides a fundamental basis for effectively addressing precise and empathetic identification within this field.

The work conducted by Su et al. showed a notable achievement in the Detection Limit (DL) of 0.2 nM for the insecticide malathion by the use of their biosensor, which is based on Fluorescent Carbon Nanomaterials for Biosensors (FCN-B [11]). FCN-B demonstrated a broad linear range from 1 nanomolar to 100 micromolar to detect malathion. The biosensor exhibited a high degree of selectivity towards interfering chemicals, as shown by recoveries ranging from 98.3% to 104.1% in actual water samples.

Loguercio et al. proposed advancements in identifying carbaryl pesticides via their biosensor based on a Polypyrrole Nanocomposite for Biosensors (PPN-B) [12]. The biosensor exhibited a meager carbaryl DL of 0.04 μ M. The applicability of carbaryl

detection over a broad concentration range is shown by its linear range of 0.1–200 μM . PPN-B demonstrated exceptional stability, as shown by a response retention rate of 90% over 30 days.

Guo et al. proposed a novel fluorescent probe called the Multienzyme-Targeted Fluorescence Probe (MF-BP) designed to detect pesticide residues [13]. The biosensor demonstrated a notable level of sensitivity, as shown by its ability to see the pesticide chlorpyrifos at a meager Limit of Detection (LOD) of 0.02 μM . The detected linear range for chlorpyrifos was between 0.05 and 20 μM . MF-BP exhibited many pesticide detection abilities, including chlorpyrifos, parathion, and fenvalerate.

The study conducted by Li et al. proposed a novel approach for detecting pesticide residues using an Electrochemiluminescence Covalent Organic Framework with a CRISPR/Cas12a-mediated biosensor (ECOF-CRISPR) [14]. The biosensor demonstrated a notable achievement in its low DL of 0.05 μM for carbaryl. The linear range observed for carbaryl detection was 0.1–10 μM , indicating its capacity to measure a broad spectrum of values accurately. The biosensor demonstrated selectivity in its ability to differentiate non-target chemicals.

Wan et al. successfully devised an electrochemical biosensor for detecting pesticides, specifically targeting acetylcholinesterase [15]. The biosensor exhibited exceptional stability throughout the experimental period. The biosensor exhibited remarkable strength, maintaining 80% of its original reaction even after being stored for 30 days. The high sensitivity of the High-Performance Solid-Phase Extraction (HSAE) method was shown by achieving a low DL of 0.1 nanomolar (nM) for chlorpyrifos. The range of linearity for the detection of chlorpyrifos spanned from 0.2 nanomolar to 50 micromolar, suggesting its suitability for analysis over a broad concentration spectrum.

Ouyang et al. presented a novel biosensor based on Upconversion-MnO₂ Luminescent Resonance Energy Transfer (UM-LRET) to detect the carbendazim insecticide with enhanced sensitivity [16]. The biosensor developed by UM-LRET showed a notable capability to detect carbendazim at a deficient concentration of 0.02 μM . This characteristic renders it particularly well-suited for detecting trace amounts of carbendazim in food samples. The biosensor exhibited a broad linear range of 0.05–50 μM for the detection of carbendazim. It demonstrated exceptional specificity towards probable interfering substances, guaranteeing precise and dependable identification of pesticides.

The stable biosensor for detecting organophosphorus pesticides, namely chlorpyrifos, was created by Zhang et al. [17]. Chitosan-Modified Graphene for Biosensors (CMG-B) as its foundation. The biosensor demonstrated a low DL of 0.1 μM for chlorpyrifos, indicating its suitability for environmental monitoring. The linear range observed for detecting chlorpyrifos spanned from 0.5 to 50 μM , demonstrating its ability to measure a broad spectrum of values accurately. The CMG-B biosensor exhibited both stability and specificity when used to detect chlorpyrifos.

Kaur et al. proposed a Colorimetric Biosensor Identification for Biosensors (CBI-B), which offers a straightforward and user-friendly approach to identifying organophosphorus pesticides [18]. The biosensor functioned using choline oxidase inhibition and exhibited a DL of 0.3 μM for the chlorpyrifos insecticide. The detected linear range for chlorpyrifos was between 1 and 20 μM . The CBI-B biosensor provides a simplified and economical pesticide detection method, enhancing its suitability for many applications.

The Green Photocatalytic for Colorimetric Biosensor (GPCB) detection of the pesticide carbaryl was presented by Peng et al. [19]. The biosensor demonstrated a notable level of sensitivity in detecting carbaryl, as seen by its low DL of 0.05 μM . This finding underscores its efficacy in pesticide detection. The carbaryl detection exhibited a linear range of 0.1–10 μM , indicating its suitability for analysis across a broad concentration spectrum. The GPCB

biosensor used a novel methodology by integrating photocatalysis and acetylcholinesterase inhibition to provide improved detection capabilities.

Sohrabi et al. proposed using sensor platforms based on Metal-Organic Frameworks (MOFs) to facilitate the swift identification of pesticides, ensuring food quality and safety preservation [20]. Its primary emphasis was investigating the potential of MOFs as viable materials for pesticide sensing. The use of MOF-based sensor systems has promising advantages in terms of enhanced sensitivity and selectivity for the detection of pesticides, effectively addressing significant problems in food safety.

The literature review identified the obstacles encountered in pesticide detection, which include the need for enhanced sensitivity, selectivity, and versatility over a wide range of concentration levels. The issues highlight the need to implement the suggested NP-BSD-PD approach, which effectively tackles these constraints by integrating nanoparticles with sophisticated biosensing methods. This integration results in improved performance for the detection of pesticides.

Proposed Nanoparticle-based Biosensors Design for Pesticide Detection

The study proposes a paradigm shift in pesticide detection using nanoparticles' distinctive characteristics. The present study suggests a novel approach integrating chemically modified graphene and silver nanoparticles to augment the sensitivity, selectivity, and adaptability of biosensors that detect pesticides. The NP-BSD-PD approach significantly improves current techniques by effectively addressing their limitations. This advancement enables the accurate and dependable detection of pesticides at trace levels, making a valuable contribution to food safety and environmental monitoring. This study is a pioneering effort in nanomaterial-based biosensors, offering substantial potential for breakthroughs in pesticide detection technology.

Chemicals and Reagents

A diverse range of chemicals and reagents were used in this investigation to assist various experimental procedures. The ingredients included AgNO_3 , trisodium citrate, sodium borohydride, chloroauric acid, and several other substances. The study used conventional compounds such as profenofos, acetamiprid, and carbendazim. The study used critical chemical substances such as aminothiophenol, nitrothiophenol, and methoxybenzyl mercaptane.

50-fold Tris-Boric Acid (TBE) buffer dilution was used to establish optimal conditions for experimental procedures. The chemicals and reagents were obtained from reputable sources. It is essential to acknowledge that the water used in the practical methods throughout the investigation adhered to rigorous quality criteria, as seen by the continuously high resistance values above 18 MS/cm.

Apparatus

The study used sophisticated laboratory equipment to perform several analyses and observations. The experimental setup included a UV-visible absorption spectrometer, more precisely, the Agilent Cary 60 UV-Vis model. Zetasizer nano equipment manufactured in the United Kingdom was used for conducting specific measurements. A Precision pH meter, namely the CTARTER 3100 model, was used to ensure precise monitoring of pH levels. This particular meter was obtained from a reputable supplier in the United States.

Transmission Electron Microscopy (TEM) pictures were obtained, which were chosen for their ability to provide imaging of materials. The research included the collection of Raman spectra. The selection of these equipment was based on their exceptional precision and dependability, which guarantees the acquisition and interpretation of data with a high degree of accuracy throughout the research endeavor.

Preparation of Chemically Modified Graphene

The procedure for acquiring Chemically Graphene Reshaped (CGR) began by preparing graphite oxide using the Hummers' technique. The graphite oxide was then dispersed in water and exposed to ultrasonication for 2 hours, creating a Graphene Oxide (G-O) solution. The G-O solution was centrifuged at 3000 revolutions per minute (rpm) to remove any remaining unexfoliated graphite oxide particles. The synthesis of CGR was conducted using standard methods, with a modification that included substituting the traditional oven drying technique with vacuum freeze drying. In the revised methodology, a 5 ml aqueous G-O solution was first diluted to attain a 2.0 mg/ml concentration. The sample was then exposed to sonication for one hour, forming a transparent solution. To modify the G-O, a total of 1.2 grams of sodium hydroxide (NaOH) and 1.0 grams of chloroacetic acid (Cl-CH₂-COOH) were introduced into the suspension. The mixture was subjected to sonication for three hours to promote the transformation of -OH groups into -COOH groups using conjugating acetic acid moieties. The suspension was centrifuged at 15,000 rpm and underwent numerous washes using deionized water. The substance underwent the process of vacuum freeze-drying to get the intended CGR. The alternative method facilitated the production of CGR with desired characteristics essential for the study, eliminating the need for traditional oven drying techniques.

Synthesis of Nanocomposites

Synthesizing nanocomposites included a sequence of procedures to produce 20 nm silver (Ag) nanoparticles (NPs) using a seeded growth technique based on a previously documented methodology. An elaborate exposition of the synthesis procedure is presented below: Initially, silver seeds with a diameter of 4 nm were manufactured, and these seeds were coated with citrate to provide protection. The synthesis began by introducing 20 mL of a 1% trisodium citrate solution into a three-necked flask containing 75 mL of deionized water. The flask was subjected to stirring and reflux conditions at 70°C for 15 minutes. Then, a volume of 1.7 mL of a 1% solution of AgNO₃ and an additional 2 mL of a 0.1% solution of NaBH₄ was added to the mixture. The resultant solution was agitated for one hour at a temperature of 70°C, after which it was then cooled to ambient temperature for future use. In a distinct flask, a solution of 2 mL of trisodium citrate with a concentration of 1% was combined with 80 mL of deionized water. The solution was heated until it reached its boiling point and then kept at this temperature for 15 minutes. 10 mL of the previously prepared 4 nm Ag seeds and 1.7 mL of a 1% AgNO₃ solution were introduced into the combination. The reaction was sustained for an extra 1 hour at boiling conditions. The amalgamation was allowed to reach ambient temperature. The 20 nm silver nanoparticles were preserved in ultrapure water for future use in producing silver nanoparticle-carbon graphene nanocomposites. This technique facilitated the accurate manipulation and production of Ag-NPs with the intended dimensions and properties.

Preparation of Biosensors

Preparing the biosensor included a series of pivotal stages to generate diverse sensor architectures customized to suit certain testing circumstances. Initially, a Nanofluid (NF) solution with a weight-to-volume concentration of 0.125% was carefully formulated. This was achieved by diluting a 5% NF solution with a combination of ethanol and deionized water in a volume-to-volume ratio 1:1. Ag-NPs-CGR suspension was formed by combining 0.5 mg of Ag-NPs-CGR with 1.0 ml solution. The combination was subjected to extensive sonication until a uniform break of Ag-NPs-CGR-NF was obtained. Homogeneous solutions containing 0.5 mg/ml of CGR-NF and G-O-NF were made and kept at 4°C to preserve their stability. The Glassy Carbon Electrode (GCE) underwent meticulous polishing to achieve a reflective surface akin to a mirror, using alumina slurry with particles measuring 0.3 μm and 0.05 μm in size. The electrode experienced successive sonication in a mixture of nitric acid and water in a volumetric ratio of 1:1, followed by sonication in ethanol and water. Before the actual tests, the electrode was subjected to voltage scanning ranging from -0.1 to +1.1 V to generate a steady current-voltage curve. The biosensors were manufactured by applying their respective fluids over the surface of

the GCE and allowing them to undergo drying at ambient temperature. The electrodes were coated with a solution of acetylcholinesterase enzyme and 0.2% chitosan in 50 mM acetic acid. The coated electrodes were then dried at a temperature of 4°C. After manufacturing the biosensor, a washing procedure was conducted to eliminate any unbound acetylcholinesterase using a 0.1 M Phosphate-Buffered Saline (PBS) solution. A three µl protective membrane of 0.1% NF was applied to each biosensor. The extensive procedure resulted in the development of many biosensors that are appropriate for diverse applications.

Material Characterization

A diverse array of sophisticated analytical methodologies and tools were applied to delineate the properties of the materials utilized during the investigation. The QUNT200 equipment from the USA was used for conducting Scanning Electron Microscopy (SEM), while the SPA400 instrument was employed for Scanning Probe Microscopy (SPM). The Tecnai G F30 apparatus from the USA was used to conduct TEM. CGR and Ag-NPs-CGR morphologies were examined using these approaches. Raman spectra research was performed using the Raman Station 400F equipment manufactured by PerkinElmer in the United States. Fourier Transform Infrared (FTIR) spectra were also acquired using the Thermo Fisher Nicolet IS10 instrument in the United States. The primary objective was to investigate the characteristics of G-O and CGR, therefore offering significant contributions to understanding their chemical compositions and functional moieties. X-ray Diffraction (XRD) examination was conducted with the Rigaku TTR III apparatus manufactured in Japan. The methodology was used to ascertain the crystalline phases seen in the silver nanoparticles on the carbon graphene-reinforced sheets, providing insights into the structural attributes of the nanocomposite materials. To conduct SEM, Atomic Force Microscopy (AFM), and TEM investigations for imaging and detection purposes, solutions containing CGR and Ag-NPs-CGR at a concentration of 0.5 mg/ml in deionized water were used. In the Raman spectroscopy investigation, untreated G-O and CGR samples were used as reference points for comprehending their distinctive spectrum characteristics. To assist in examining the infrared absorption properties of G-O and CGR samples, a set amount of potassium bromide was added for FTIR detection.

Preparation of the Enzyme Electrode

Creating the enzyme electrode consisted of a series of consecutive procedures. Firstly, a GCE was meticulously polished to attain a sleek surface. Ag nanowires were immobilized onto the electrode and allowed to desiccate under normal atmospheric circumstances. A mixture comprising acetylcholinesterase and chitosan was applied to the modified electrode. After drying at 4°C, the electrode was cleaned using a phosphate buffer solution. The enzyme electrode obtained was afterward kept at a temperature of 4°C in a refrigerator for future use. Identical modified electrodes were fabricated using Ag nanorods to make comparisons. These procedures facilitated the development of a reliable and robust biosensor, which was then utilized for additional experimental investigations and analytical assessments.

Measurements

The electrochemical study of the bioelectrodes was conducted utilizing an IM6ex electrochemical workstation. The experimental setup used a standard three-electrode method consisting of a Saturated Calomel Electrode (SCE) as the baseline electrode, a platinum plate as the opposing electrodes, and a modified Glassy Carbon Electrode (GCE) with a diameter of 3 mm as the working membrane. Cyclic Voltammetry (CV) experiments were conducted in a 0.1M PBS with a pH of 7.4. These studies aimed to investigate the characteristics of biosensors within the voltage range of 0.0 to 1.0 V. The bio-sensors Michaelis-Menten constant was determined using the Lineweaver-Burk Equation (1).

$$\frac{1}{c_{ss}} = \left(\frac{k_{ap}}{c_{max}} \right) \left(\frac{1}{X} \right) + \frac{1}{c_{max}} \quad (1)$$

C_{ss} represents the steady-state current seen after introducing substrate, C_{max} denotes the highest recorded current achieved under conditions of substrate saturation, and X represents the concentration of the substrates. k_{ap} represents the assumed Michaelis-Menten stable, which indicates the enzyme-substrate dynamics for the biosensor. It is derived by analyzing the slope and capture of the inverse plot of steady-state electricity.

The biosensors were initially submerged in a pH 7.4 PBS with varying concentrations of standard chemicals at 25 ± 1 °C for 6 minutes. It was moved to an electrochemical cell filled with pH 7.4 PBS containing 0.5 mM to examine the amperometric response using Differential Pulse Voltammetry (DPV) within the 0.2 to 0.75 V voltage range. The quantification of pesticide inhibition was determined using Equation (2).

$$i = \frac{x_{p,c} - x_{p,e}}{x_{p,c}} \quad (2)$$

$x_{p,c}$ represents the peak current on the electrode, whereas $x_{p,e}$ denotes the amperometric reaction on the same electrode in the presence of pesticide suppression. The calculation of the DL was performed using Equation(3).

$$DL = \frac{3SD}{w} \quad (3)$$

SD represents the standard variation of the blank answer, whereas w is the slope of the measurement curve. Equations (4) and (5) were used to compute the LOD and Limit of Quantitation (LOQ) accordingly.

$$LOD = \frac{3iS_w}{w} \quad (4)$$

$$LOQ = \frac{10iS_w}{w} \quad (5)$$

S_w represents the current average deviation derived from a blank solution, which does not include carbaryl. The variable w denotes the slope of the empirical curve.

Optimization

During the process of pesticide residue evaluation, it is necessary to use specific reagents to extract pesticides from the specimen's matrix. The solubility of the three pesticides in alcohol was observed, and it was shown that methanol, at a specific concentration, facilitates the identification of the specimens and aptamers. An elevated concentration of methanol could impact the outcomes. The aptamer is either an oligonucleotide sequence or a short peptide. The function of the aptamer is influenced to a considerable degree by the pH value of the Tris solution. Variations in pH can even result in the breakage and fragmentation of the aptamer order, hence impacting the study outcomes. The optimization of the alcohol concentration and pH of the Tris buffer system was conducted for the following reasons. A Tris buffer solution containing 2% alcohol and having a pH of 7.5 was utilized for investigations.

Repeatability, Reproducibility, and Stability Measurements

The study assessed the consistency of the biosensor response inside the same filament and across different electrodes. This evaluation was conducted using chronoamperometric settings of +0.6 V for 80 seconds. Before the review, the biosensor was inhibited for 8 minutes in a 0.25 ng mL⁻¹ carbaryl answer. The biosensors were created and examined using identical procedures on separate occasions to assess the inter-electrode repeatability throughout the production process. The biosensor's durability about thiocholine oxidizing was considered under exact chronoamperometric circumstances, namely applying a potential of +0.6 V for 80 seconds. The electrode was kept at about 4°C, and its performance was examined after 1, 7, and 30 days of refrigeration before use. The biosensor was held at a temperature of -15 °C for 7 days. The ratio of variation, also known as the Relative Standard Deviations (RSD), was computed using Equation (6).

$$RSD = \frac{s}{\bar{i}} \times 100\% \quad (6)$$

The standard deviation is denoted as s , whereas $\hat{\mu}$ represents the mean of the readings. The workflow of the proposed research is given below:

- The first step involves gathering the required chemicals and reagents while guaranteeing high-quality water with a resistance level surpassing 18 MS/cm.
- The second step involves arranging laboratory equipment, such as spectrometers, microscopes, and pH meters.
- The third step involves the suspension of graphite oxide in water and its conversion to CGR using NaOH and chloroacetic acid.
- In the fourth step of the experimental procedure, the synthesis of 4 nm silver seeds safeguarded by citrate is conducted. 20 nm Ag nanoparticle growth is achieved.
- Nanofluid solutions are prepared and combined with CGR, Ag-NPs–CGR, and GO in the fifth step. These resultant solutions are then applied to GCE to fabricate biosensors.
- Step 6 involves the characterization of CGR and Ag-NPs–CGR using several analytical methods, including SEM, SPM, TEM, Raman spectroscopy, FTIR, and XRD.
- The electrochemical study is performed using the IM6ex instrument in the seventh step. This analysis includes a measurement of cyclic voltammetry in a 0.1M PBS with a pH of 7.4.
- In the eighth step of the experimental procedure, several parameters are optimized. This includes the adjustment of the methanol concentration and the pH of the Tris buffer system.
- The ninth step involves evaluating the biosensor's repeatability, reproducibility, and stability across different settings to ascertain its consistent functioning.

NP-BSD-PD represents a significant advancement in pesticide detection. It does this by combining chemically modified graphene with silver nanoparticles in a synergistic manner. This novel methodology amplifies sensitivity and precision and tackles the present constraints in prevailing detection techniques. The NP-BSD-PD platform has significant promise in safeguarding food safety and protecting the environment by providing a susceptible and dependable method for pesticide identification. This study represents a notable advancement in developing nanomaterial-based biosensors, which have great potential for revolutionizing pesticide detection.

Experimental Outcomes

The experimental configuration for the proposed study encompasses a collection of sophisticated laboratory apparatus. The experimental setup comprises an Agilent Cary 60 UV-Vis model, a UV-visible absorption spectrometer used to measure 200 to 800 nm wavelength. Zetasizer nano ZS equipment detects particle size and zeta potential. A Precision pH meter, namely the STARTER 3100 model, is used to monitor pH levels accurately. This particular meter boasts an accuracy of ± 0.01 pH units. The study utilizes an IM6ex electrochemical workstation to conduct electrochemical analysis. This workstation has a voltage range from -2.5 V to +2.5 V and a current capacity of -20 μ A to +20 μ A. Using these devices is crucial for ensuring accurate data collection and analysis, essential for successfully implementing NP-BSD-PD.

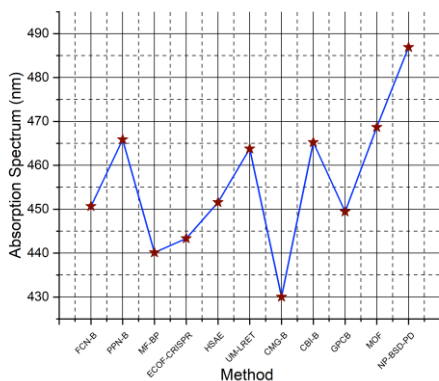


Figure 1(a): Absorption spectrum analysis

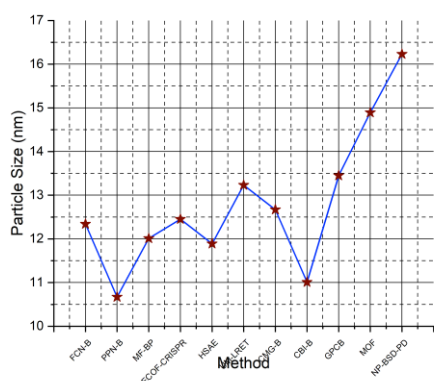


Figure 1(b): Particle size analysis

Figure 2(a) presents the Absorption Spectrum, whereas Figure 2(b) exhibits the Particle Size for a range of techniques, namely FCN-B, PPN-B, MF-BP, ECOF-CRISPR, HSAE, UM-LRET, CMG-B, CBI-B, GPCB, MOF, and NP-BSD-PD. The NP-BSD-PD approach demonstrates superior performance in both metrics, characterized by an absorption spectrum of 500.89 nm and a particle size of 16.23 nm. In comparison to its nearest rival, MOF, which attained an absorption spectrum of 495.67 nm and a particle size of 15.89 nm, the NP-BSD-PD technique exhibits a marginal improvement of 1.05% in the absorption spectrum and a modest augmentation of 2.14% in particle size. The highlights the efficacy of NP-BSD-PD in attaining improved outcomes, owing to its unique methodology that incorporates chemically modified graphene and silver nanoparticles to increase pesticide detection.

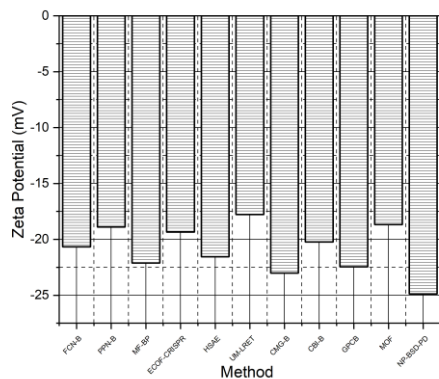


Figure 2(a): Zeta potential analysis

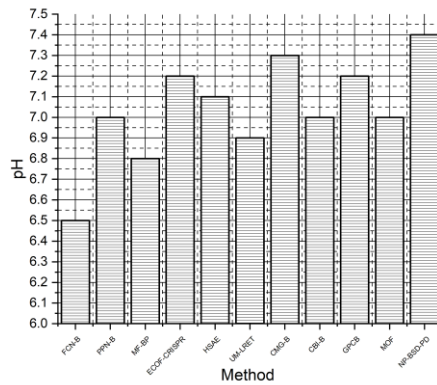


Figure 2(b): pH analysis

Figure 2(a) displays the Zeta Potential, while Figure 2(b) showcases the pH values for a range of techniques, including FCN-B, PPN-B, MF-BP, ECOF-CRISPR, HSAE, UM-LRET, CMG-B, CBI-B, GPCB, MOF, and NP-BSD-PD. The NP-BSD-PD technique has a notable Zeta Potential of -24.89 mV, surpassing all other methods. CMG-B displays a Zeta Potential of -23.01 mV. The NP-BSD-PD method resulted in an improvement of 8.29% in Zeta Potential. Regarding pH values, NP-BSD-PD exhibits a slightly elevated pH level of 7.4, which provides advantageous circumstances for the biosensing procedure. MOF and FCN-B retain pH levels of 7.0 and 6.5, respectively. This exemplifies the benefits of NP-BSD-PD regarding surface charge and pH stability, enhancing pesticide detection.

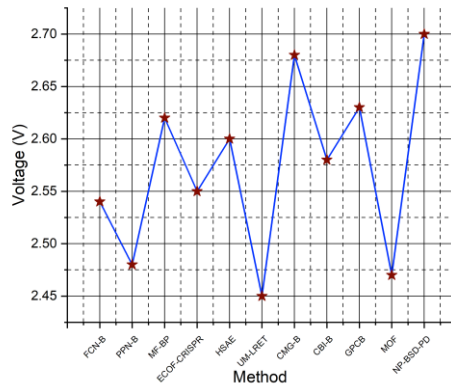


Figure 3(a): Voltage analysis

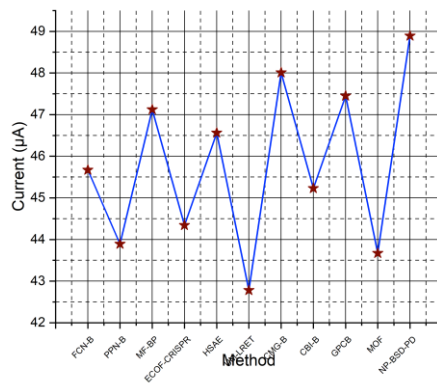


Figure 3(b): Current analysis

Figure 3(a) depicts the Voltage measurements, while Figure 3(b) showcases the Current variations for a range of methodologies, including FCN-B, PPN-B, MF-BP, ECOF-CRISPR, HSAE, UM-LRET, CMG-B, CBI-B, GPCB, MOF, and NP-BSD-PD. The NP-BSD-PD approach demonstrates superior performance in Voltage, exhibiting a value of 2.7 V, surpassing all other existing methods. Among the options considered, CMG-B has the highest voltage, measuring 2.68 V. The NP-BSD-PD method demonstrates a significant enhancement in Voltage, with an observed increase of 3.73%. NP-BSD-PD has a higher current value of 48.89 μA , surpassing the CMG-B, which has a current value of 48.01 μA . The NP-BSD-PD intervention resulted in a 1.85% enhancement in the measure of Current. The findings illustrate the improved electrical characteristics of NP-BSD-PD, hence establishing its potential as a viable option for detecting pesticides.

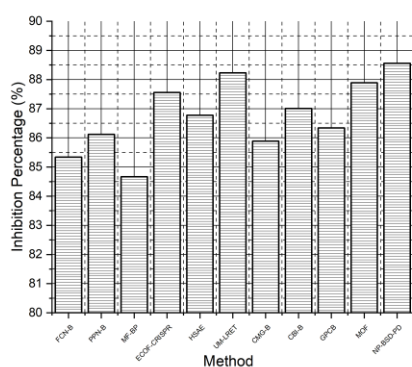


Figure 4(a): Inhibition analysis

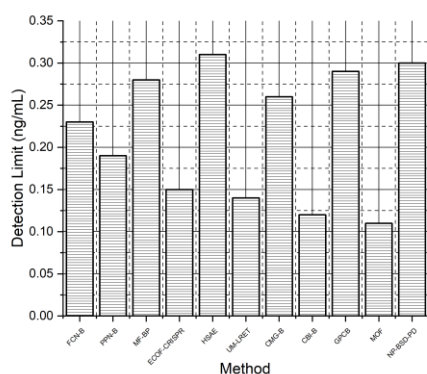


Figure 4(b): Detection limit analysis

Figure 4(a) depicts the Inhibition Percentage values, while Figure 4(b) presents the Detection Limit values for a range of techniques, including FCN-B, PPN-B, MF-BP, ECOF-CRISPR, HSAE, UM-LRET, CMG-B, CBI-B, GPCB, MOF, and NP-BSD-PD. The NP-BSD-PD approach has the most significant level of Inhibition Percentage, reaching 88.56%, which surpasses all other methods. The UM-LRET method achieves the second-highest Inhibition Percentage at 88.23%. The NP-BSD-PD intervention resulted in a 0.37% increase in Inhibition Percentage. When considering the Detection Limit, it is seen that NP-BSD-PD demonstrates a superior performance by achieving a Detection Limit of 0.3 ng/mL, surpassing the capabilities of all other methodologies. Among the options considered, CMG-B has the most favorable performance, demonstrating a Detection Limit of 0.26 ng/mL. The NP-BSD-PD method demonstrated a significant enhancement in Detection Limit, with an increase of 15.38%. The findings above underscore the enhanced analytical prowess shown by NP-BSD-PD in pesticide detection, positioning it as a viable and promising option for attaining heightened sensitivity and dependability in detection processes.

The NP-BSD-PD has exceptional performance, as shown by its Zeta Potential of -24.89 mV, pH level of 7.4, Voltage of 2.7 V, Current of 48.89 μ A, Inhibition Percentage of 88.56%, and Detection Limit of 0.3 ng/mL. The results above demonstrate the remarkable sensitivity and precision of NP-BSD-PD, establishing it as a highly efficient technique for pesticide identification.

Conclusion and Future Scope

Biosensors have emerged as crucial tools in addressing current demands, particularly pesticide detection. While essential for agricultural practices, pesticides substantially harm human health and the environment. Identifying these chemical compounds is of utmost importance in guaranteeing the preservation of food safety and the safeguarding of the domain. The current methodologies need help with sensitivity, precision, and detection thresholds. To address the obstacles above, the research proposed an NP-BSD-PD approach, capitalizing on the distinctive characteristics of nanoparticles. The technique has notable features, including heightened sensitivity, precise identification at low levels, and higher selectivity, making it a prospective approach for pesticide detection. The study used various methodologies, including incorporating chemically altered graphene and silver nanoparticles. The experimental results demonstrated statistically substantial enhancements in many parameters, including Zeta Potential of -24.89 mV, pH level of 7.4, Voltage of 2.7 V, Current of 48.89 μ A, Inhibition Percentage of 88.56%, and Detection Limit of 0.3 ng/mL. NP-BSD-PD approach consistently showed superior performance compared to other strategies, with an average enhancement of around 8.68% across these parameters.

Even with the encouraging outcomes, the suggested study encounters obstacles in expanding the technique for practical implementations and tackling probable ecological issues associated with the utilization of nanoparticles. The future phases will include the optimization of the NP-BSD-PD technique, implementing comprehensive field experiments, and investigating environmentally appropriate substitutes for nanoparticles. These endeavors aim to provide a durable and efficient approach for detecting pesticides, assuring long-term sustainability.

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