

RESEARCH PAPER

Preparation and Characterization of Nanofibers Composed of Fullerene, Gold Nanoparticles, PANi, and PVA for Medical Biosensor Fabrication Using the Electrospinning Method

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ABSTRACT

The aim of this study is to prepare nanofibers from fullerenes and gold nanoparticles at concentration of (0, 0.4, 1.9 and 3.6) ppm with PANi and PVA, for medical biosensor fabrication using the electrospinning method and to study the effect of both gold nanoparticles and fullerenes on the structural, morphological, XRD and Cyclic Voltammetry properties of nanofibers. The gold nanoparticles are causing pressure in the crystal lattices of the carbon materials (C₈₀, C₇₀, and C₆₀ polymers), Also These systematic displacements are not due to mere physical mixing, but rather are conclusive evidence of a real interaction at the atomic/crystal scale between gold and carbon, nanofibers with sizes ranging from 21.99 nm to 80.5 nm appeared, as did small, the sensor based on AuNP/CNF/PANi/PVA on a gold electrode exhibits a clear dose – dependent electrochemical response, this medical biosensor capable of reliably detecting CA II in the 0-125 ng/ml range.

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INTRODUCTION

In recent years, biosensors have become a crucial focus in research related to medical diagnostics and environmental monitoring, due to their ability to deliver precise and sensitive results for detecting a wide range of biomolecules. However, significant challenges remain in improving the sensitivity and stability of these sensors. Medical biosensors, in particular, require continuous performance enhancement to ensure accuracy and reliability [1]. While nanomaterials such as gold and carbon can significantly enhance the sensitivity of biosensors, their high cost

and the intricacies of manufacturing remain substantial hurdles [2]. Gold nanoparticles exhibit unique properties, such as high electrical conductivity and a large surface area, which make them highly effective in improving the sensitivity for detecting biomolecules [3]. Carbon nanotubes, on the other hand, offer exceptional mechanical and electrical properties that enhance the stability and extend the operational lifespan of sensors [4]. Additionally, Poly Aniline (PANi), a conductive polymer, is known for its ability to interact with ions and biomolecules, enhancing the sensitivity of sensors while contributing to

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improved stability [5]. Furthermore, Poly Vinyl Alcohol (PVA) is a biodegradable polymer that can be used as a support material in medical sensor fabrication, reducing costs while promoting environmental sustainability [6]. The integration of gold nanoparticles and carbon nanotubes with PANi and PVA offers several potential benefits for improving the performance of biosensors. This combination enhances both electrical conductivity and sensitivity, allowing for more accurate detection of biomolecules [7, 5]. The addition of carbon nanofibers to PANi contributes to long-term sensor stability, making these devices more reliable across various environments [8, 9]. Using PVA as a supporting material provides an economical solution, reducing manufacturing costs, as it is a biodegradable polymer that minimizes environmental waste [10, 11].

In terms of potential applications, these enhanced sensors could significantly improve the early detection of tumors, increasing the effectiveness of early-stage disease diagnosis [12]. The flexibility of PANi and PVA also makes them suitable for developing wearable medical devices, providing continuous monitoring of health parameters with greater comfort and safety [13, 14]. Additionally, PVA can be used to create implantable sensors that offer long-term disease monitoring while safely degrading within the body over time [15].

Nevertheless, several challenges must still be

addressed, such as improving the biocompatibility of the materials to ensure they the body. Achieving long-term stability in diverse biological and chemical environments also requires further refinement of material formulations [16,17]. Additionally, the manufacturing processes need to be optimized to be more cost-effective and scalable, while ensuring high-quality sensor performance [18].

MATERIALS AND METHODS

In this study several instruments and materials Suppliers: Carbon electrodes from dray batteries, gold, Distilled Deionized Water (DDW), Alcohol, 96% Acetone. To prepare the colloidal solution of carbon according to a previous study [19] to produce nanoparticles of gold and fullerene, the carbon was placed in 4 beakers, each beaker containing 50 ml of deionized water, and each one was bombarded with 3000 laser shots, then the gold was also bombarded with a number of laser shots: (0, 1000, 2000, 3000) respectively, with a laser beam using an Nd -YAG pulsed laser, its Parameter Value: Wavelength 1064 nm, the diameter of the spot 2 mm, the laser energy was 100 mJ, the pulse duration 6 Hz and laser pulse width 10 nm. The distance between the surface of solution and the laser lens was 12 cm Fig. 1.

After preparing the 50 ml colloidal solution, put it in the syringe of the electrospinning device Fig. 2. The electrospinning device was used with specific

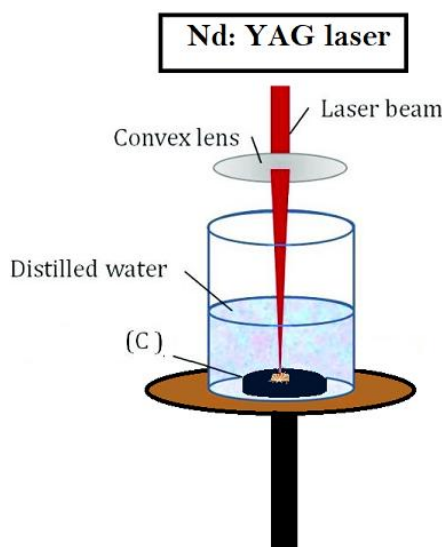


Fig. 1. Schematic illustration of the laser ablation method in liquid.

parameters. The distance between the needle and the sample in the rotating cylinder was 10 cm, the high voltage between them was 23.5 KV,

the cylinder rotated around its axis at a speed of 3000 rpm, and the colloidal fluid flow rate in the injection system was equal to 0.1 ml per hour, the colloidal solution exits from the nozzle of the syringe head, which is the positive pole, to the opposite rotating cylinder, that negative pole.

RESULTS AND DISCUSSION

By analysis of X-ray diffraction (XRD) patterns (x-ray Anode Material Cu Generator Settings radiation and accelerated voltage of 40 kV and 30 mA). Williamson-Hall relation was [20] applied to estimate the crystalline size

$$D_{hkl} = \left(\frac{A\lambda}{\cos\theta \cdot B_{hkl}} \right) + (4\epsilon \sin\theta) \quad (1)$$

FE-SEM images achieved via field emission scanning electron microscope (FE-SEM, INSPECT-550) have been used to evaluate the morphological properties of the prepared film surfaces, addition to the energy dispersive x-ray

(EDX) microanalysis to estimate elemental analysis associated with the prepared films, as well Raman spectra were achieved to detect their vibrational and rotational.

XRD patterns have been analyzed based on COD 96-900-1992, ICSD 98-041-2242, ICDD 01-089-8491, ICSD 98-005-6668, and ICSD 98-009-6620 cards. Pure carbon films showed a polycrystalline structure with the dominance of C_{70} phases at 2θ of approximately 20.33° , 22.83° , 29.04° , and 39.66° . The dominant phase was observed at about 29.04° (242) with crystallite size found at about 89.22 nm. Likewise, the presence of three main phases, C_{80} , C_{70} and C_{60} - Polymer but with lower attributions as shown in Fig. 3.

Experimental location of the first reference curve (without gold)

In the first curve, the reference C_{80} peak is the peak at 13.5547° . We note that the second curve (0.4 ppm) at 15.9907° appears to be shifted significantly to the right ($+2.436^\circ$). Curve 3 (1.9 ppm) at 16.0038° appears to be shifted significantly to the right ($+2.449^\circ$). Curve 4 (3.6 ppm) at 15.8213° appears to be shifted significantly to the

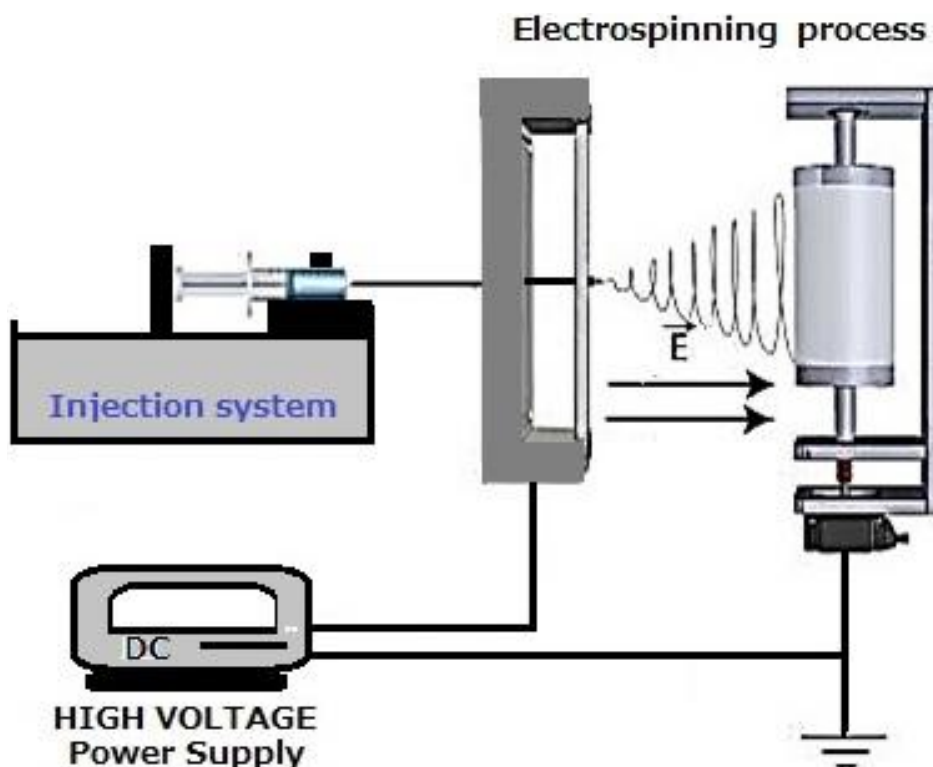


Fig. 2. Electrospinning device used in this work

right (+2.266°). We conclude that the addition of gold caused a systematic and significant shift of the C_{80} peaks toward higher (right) angles. C_{70} peaks, which is the second indicator in the tables - the most important peak, the reference is the peak 20.3254°, where we notice curve 2 (0.4 ppm): 20.3161° shows a very slight shift to the left (-0.0093°). As for curve 3 (1.9 ppm): 20.5815°, there is also a clear shift, but to the right (+0.2561°). In curve 4 (3.6 ppm): 20.3503°, also a shift to the right (+0.0249°). We conclude that the behavior is more complex. At low concentration (0.4 ppm), the position is almost unchanged, but with increasing concentration, a shift to the right occurs, which is more pronounced at medium concentration (1.9 ppm). Polymer C_{60} peaks, which is index 9 in the tables, the first reference curve is the peak 73.1248° and the second curve (0.4 ppm): 73.5587° appears to be shifted to the right by (+0.4339°), and in the third curve (1.9 ppm also the peak): 73.3975° shifted to the right by (+0.2727°), and the fourth curve (3.6 ppm): 73.4005° in the peak shifted to the right (+0.2757°).

The shift in diffraction angle (2θ) is directly related to the change in the distance between crystal planes (d-spacing) according to Bragg's law:

$$n\lambda = 2d \sin\theta$$

Rightward shift (increasing θ): This means a decrease in the distance (d) between crystal planes. Physical Interpretation: This often indicates compressive strain within the crystal lattice. The introduction of smaller gold atoms or particles (or their formation in interstitial sites) can compress and bring the crystal planes closer together, leading to a decrease in the value of d and thus an increase in the angle θ .

Leftward shift (decreasing θ): This means an increase in the distance (d) between crystal planes. This indicates a tensile strain within the lattice. This very slight displacement was observed in one case (C_{70} at 0.4 ppm), and may be due to the presence of impurities or an initial surface interaction causing a slight expansion.

1. The dominant effect is pressure: The dominant and clear displacement direction in your data is to the right, which means that the gold nanoparticles are causing pressure in the crystal lattices of the carbon materials (C_{80} , C_{70} , and C_{60} polymers). This pressure leads to a decrease in the distances between the atomic planes.

2. Evidence of interaction: These systematic displacements are not due to mere physical mixing, but rather are conclusive evidence of a real interaction at the atomic/crystal scale between

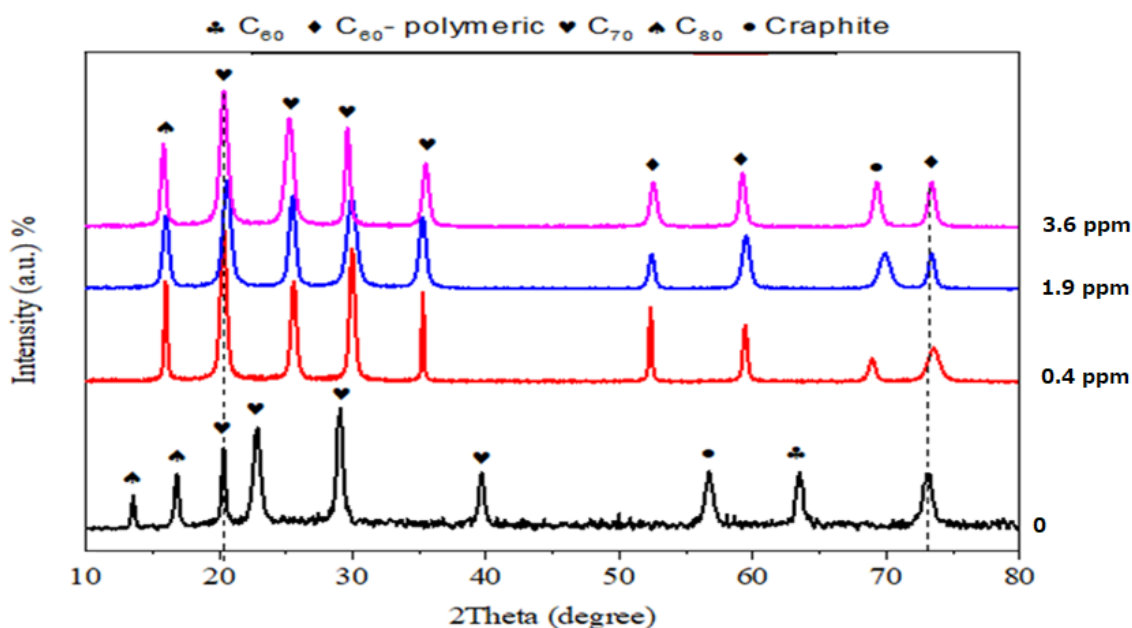


Fig. 3. XRD patterns of Fullerene-Au films

gold and carbon. The gold particles are not just sitting on the surface; they permeate the structure and create internal stress.

3. Concentration and structure dependence: The magnitude of the response (amount of displacement) varies depending on the type of carbon molecule (C_{80} was most affected) and the gold concentration, indicating that the reaction mechanism and the extent of each structure's influence may differ.

The rightward shift of the peaks is direct experimental evidence that nanogold exerts pressure on the carbon crystal structures, altering their fundamental physical properties. This is critical to the performance of the hybrid material in various applications such as catalysis and electronics.

Fig. 4 represents FE-SEM images that mainly give morphological information about the surface of the prepared Fullerenes and AuNPs-fullerene nanofiber with 0, 0.4, 1.9, and 3.6 ppm.

A clear separation was observed between the different particles, especially the nanofibers, which are believed to be C_{60} polymers with an average diameter of approximately 39 nm, as shown in Fig. 4. The addition of gold nanoparticles led to a clear change in the stages of nanofiber formation. The addition of gold at concentrations of 0, 0.4, 1.9, and 3.6 ppm resulted in a variety of formations,

consisting of scattered spherical particles with diameters ranging from 17.86 to

35.73 nm in the absence of gold. In addition, hairs with a thickness of approximately 29 nm to 42 nm and nanofibers with a diameter of approximately 53 nm also appeared. At a concentration of 0.4 ppm, nanofibers with sizes ranging from 21.99 nm to 80.5 nm appeared, as did small, few-numbered spheres with diameters ranging from 24.56 nm to 42.43 nm, which are believed to be either gold or a by-product of the nanofibers due to the high speed of the collecting drum. At a concentration of 1.9 ppm, holes appeared in the tissue with diameters ranging from 37.9 nanometers to 120.6 nanometers. Increasing the concentration of gold nanoparticles to 3.6 ppm led to the appearance of large random clusters, while the spheres had diameters ranging from 224.6 nanometers to 422.7 nanometers, as shown in Fig. 4.

Cyclic Voltammetry test

Here, the results of cyclic voltammetry (CV) analysis of nanofibers fabricated as biosensors are presented, using a mixture of carbon nanoparticles and gold nanoparticles at varying concentrations of carbon and carbon-gold (0, 0.4, 1.9 and 3.6) ppm.

The electrodes were fabricated by depositing gold on a square piece of glass with a mask, then

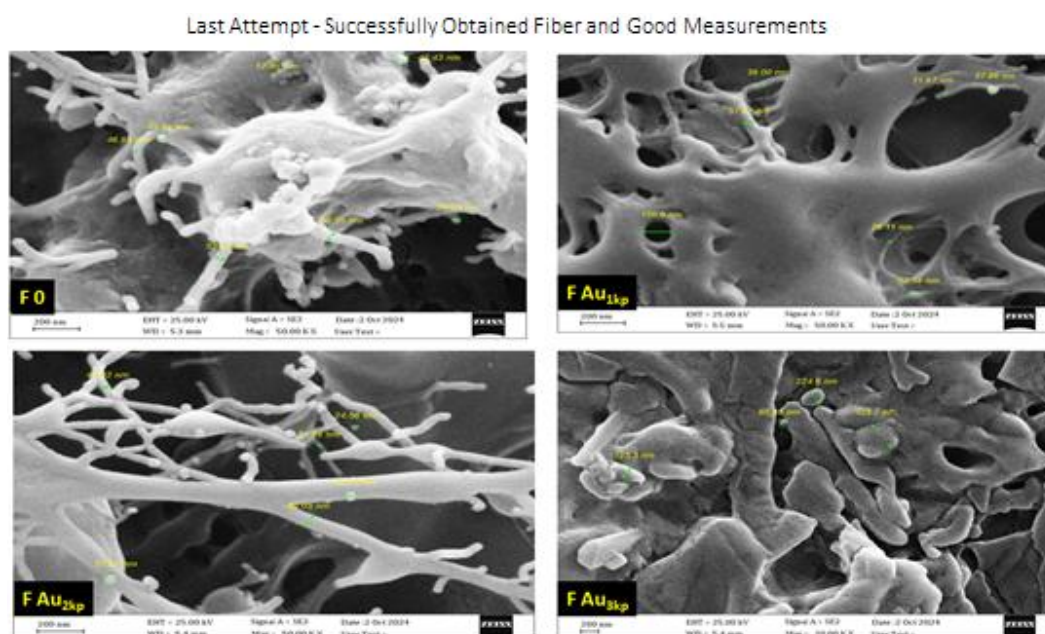


Fig. 4. FE-SEM images of the prepared fullerenes and AuNPs-

depositing the aforementioned mixture onto the nanofiber electrodes using an electrospray. The biosensor was supplied with a voltage ranging from -5 V to +5 V. The voltage between the electrodes and the current flowing through them were measured after applying different concentrations of CAII to each experiment over four cycles for subsequent study and evaluation of the sensor. The current intensity often increases with increasing CAII concentration. Redox signals are considered an indicator and diagnostic of fullerene interactions with tumor [21]. The inclusion of gold electrodes also improved the currents, due to the nature of gold nanoparticles, which provides a large surface area that enhances

the interaction, which is crucial for enhancing the sensitivity of chemical sensing. The large surface area of gold nanoparticles allows for greater adhesion of reagents, which promotes increased interaction between the analyte and the biological receptor, thus amplifying signal transmission. Gold nanoparticles (AuNPs) are considered an ideal support for the development of sensors, due to their properties, most notably good conductivity and high surface area [22, 23, 24].

0 ng/mL (Blank)

In the absence of CA II, the electrode surface remains fully active, resulting in a relatively high oxidation peak current (≈ 0.76 nA) and a

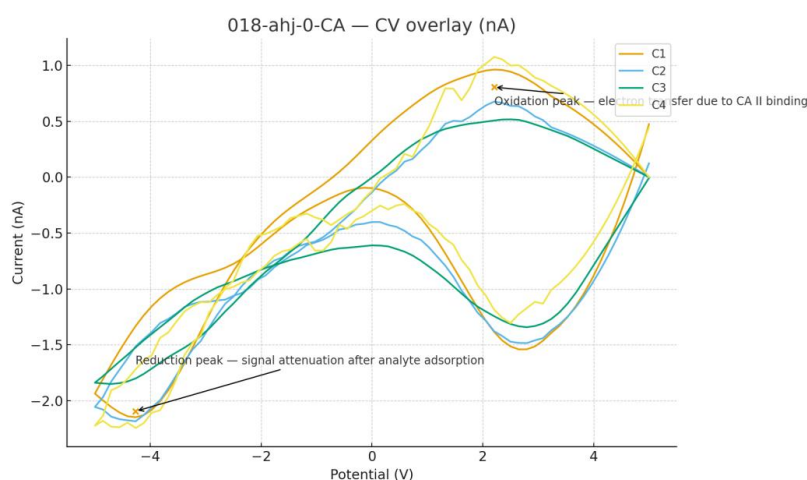


Fig. 5. CV curve for 0 ng/mL (Blank) concentration

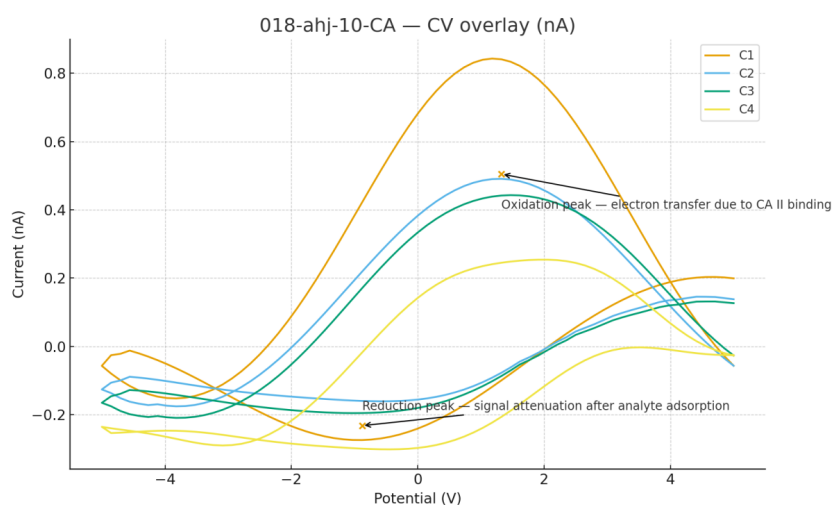


Fig. 6. CV curve for 10 ng/mL concentration

total charge of ≈ -18.4 nC. This reflects efficient electron transfer through the AuNP/CNT/PAni/PVA film layer. This condition represents the baseline response, which is used for comparison with the other tested concentrations.

10 ng/mL

At this low concentration, CA II molecules begin to bind to the active sites on the electrode surface. Consequently, the oxidation peak current decreases to ≈ 0.59 nA, and the total charge decreases to ≈ -15.7 nC. This reduction indicates

the formation of a thin protein layer that partially blocks electron transfer pathways, producing a clear and measurable response relative to the blank.

30 ng/mL

As the concentration increases, more CA II molecules bind to the surface, yet the oxidation peak current slightly increases to ≈ 0.77 nA while the charge continues to decrease to ≈ -14.2 nC. This dual behavior suggests protein reorganization on the electrode surface, where the adsorbed

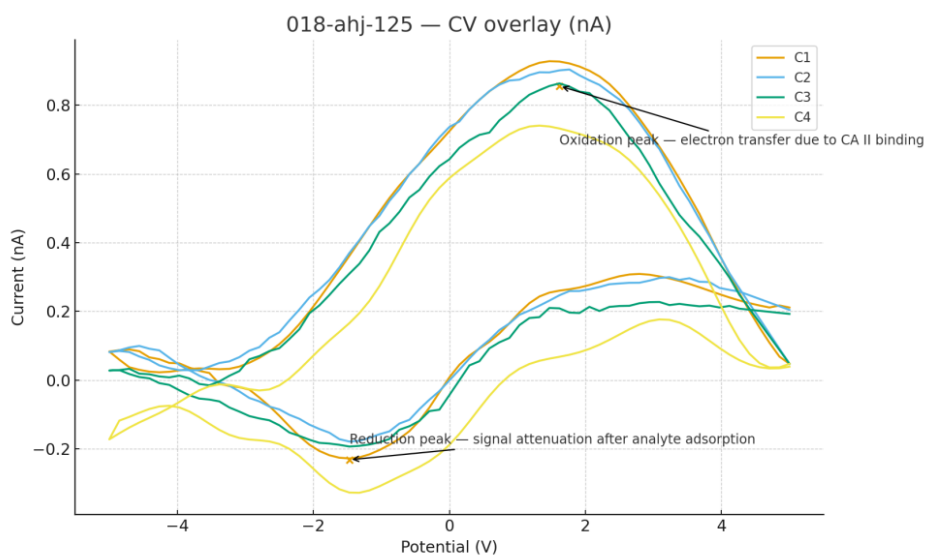
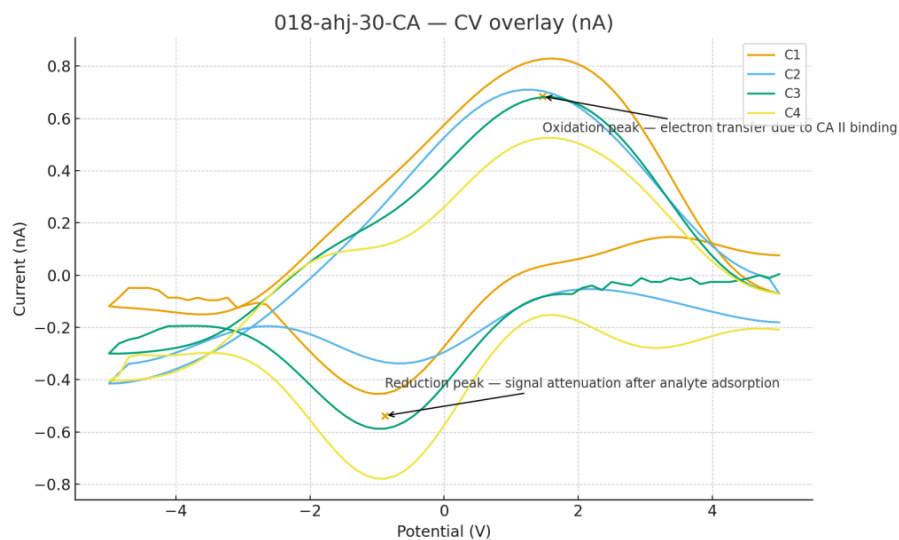


Table 1. Key Analytical Metrics

Parameter	Observed Value	Interpretation
Q trend (Charge Behavior)	Decreasing with concentration	Confirms signal-off behavior and analyte adsorption
RSD% (Reproducibility)	< 10%	Stable and repeatable response across four cycles
R ² (Calibration Linearity)	≈ 0.97	Excellent linear correlation across 0–125 ng/mL
LOQ (Limit of Quantification)	≈ 15–20 ng/mL	Accurate quantification region starts above 10 ng/mL
LOD (Limit of Detection)	≈ 5–7 ng/mL	Sensitive enough to detect below the lowest tested concentration (10 ng/mL)

layer becomes more compact while still leaving limited electron transport channels available. This stage is commonly observed in biosensors near pre-saturation binding levels.

125 ng/mL

At high CA II concentration, dense protein accumulation occurs on the electrode surface. This leads to a reorganization effect that creates localized electron transfer pathways, which is reflected by an increase in the oxidation peak current to ≈0.83 nA and a rise in charge to ≈ −24.4 nC. This behavior is known as the surface reorganization effect, confirming that the sensor response is concentration-dependent and arises from specific interaction with the target protein.

Electrochemical Sensor Evaluation for CA II Detection

Based on the cyclic voltammetry (CV) analysis and extracted charge (Q) values, current (Ip) from four consecutive cycles at, CA II concentrations ranging from 0 to 125 ng/mL, the AuNP/CNT/PAni/PVA composite electrode deposited on a gold substrate exhibited a clear dose–response behavior. Both the total charge (Q) and the oxidation current decreased progressively with increasing CA II concentration, indicating effective electron-transfer modulation due to specific antigen binding at the electrode surface (Table 1).

CONCLUSION

The AuNP/CNF/PAni/PVA-based electrochemical sensor demonstrates reliable sensitivity and selectivity toward the CA II biomarker. The clear dose-dependent signal suppression, high linearity ($R^2 \approx 0.97$), and low LOD (~5–7 ng/mL) indicate that the sensor effectively detects CA II within the studied range (0–125 ng/mL). The low RSD (<10%) further supports film stability and consistent electrochemical response. Hence, this sensor can be considered a promising platform for cancer biomarker detection using CV techniques.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

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