





Article

Detection of Levofloxacin Using a Simple and Green Electrochemically Polymerized Glycine Layered Carbon Paste Electrode

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Abstract: The easy and rapid analytical tool, electrochemically polymerized (EP) glycine (GN) layered carbon paste electrode (LCPE), was used for the analysis of levofloxacin (LN) using cyclic voltammogram (CV) and differential pulse voltammetry (DPV). The surface features and activities of the electrochemical sensors EPGNLCPE and bare carbon paste electrode (BCPE) were analyzed using electrochemical impedance spectroscopy (EIS), field emission scanning electron microscopy (FE-SEM), CV and DPV methods. The modified sensor (EPGNLCPE) offers a vibrant and sensitive electrochemical LN oxidation peak by controlling overpotential and the electrode material fouling effect unlike BCPE. Under improved experimental conditions, the DPV method was used to analyze LN on EPGNLCPE by varying its concentration in 0.2 M phosphate buffer solution from 30 to 90 μM , resulting in a good linear relationship (between peak current and concentration), lower limit of detection (LOD: 8.436×10^{-7} M) and lower limit of quantification (LOQ: 2.812×10^{-6} M). Finally, real-time application of the sensor was tested by analyzing LN in medicinal samples, and good LN recovery was observed.

Keywords: levofloxacin; glycine; carbon paste; electrochemical analysis



Citation: Bhimaraya, K.; Manjunatha, J.G.; Moulya, K.P.; Tighezza, A.M.; Albaqami, M.D.; Sillanpää, M.

Detection of Levofloxacin Using a Simple and Green Electrochemically Polymerized Glycine Layered Carbon Paste Electrode. *Chemosensors* **2023**, *11*, 191. <https://doi.org/10.3390/chemosensors11030191>

Academic Editors: Rocco Cancelliere, Laura Micheli and Giuseppina Rea

Received: 6 February 2023

Revised: 3 March 2023

Accepted: 8 March 2023

Published: 13 March 2023



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1. Introduction

The antibiotic drug LN can prevent Gram-negative and Gram-positive bacterial infections in respiratory and urinary systems. LN is normally recommended for bacterial infections of the skin, kidneys and bladder, as well as for pneumonia, gastroenteritis and sinuses [1–4]. LN overdose can cause harmful effects in the human body, including Stevens–Johnson syndrome, heart-related problems, irreversible peripheral neuropathy and muscle rupture [5–9]. The major problem with LN is that it is not metabolized completely in the body, meaning about 87% of LN is found in urine. Antibiotics are considered an embryonic toxin in the ecosystem and are commonly found in sewage, posing a high risk to aquatic life and other living organisms (animals and humans) [10,11]. Thus, the detection and quantification of LN are important to the health of living organisms and the ecosystem.

Based on this viewpoint, many attempts have been made to analyze LN using high-performance liquid chromatography [12], capillary electrophoresis [13], enzyme-based immunosorbent analyses [14], spectrofluorimetry [15]. The electrochemical analysis involved a number of difficulties. Furthermore, they require experts to handle the instruments, require high quantities of chemicals and solvents, need time to obtain results and have comparatively high instrument costs.

Electrochemical measures are suitable for the analysis and quantification of biologically/electrochemically active compounds in complex medicinal samples. These approaches have user-friendly qualities such as simple instrument handling, a low power requirement, low-cost instrumentation, quick results, low LOD, good stability, high sensitivity and low-chemical-quantity requirements [16–20]. Electrochemical methods need some key working electrode materials such as carbon-based materials, nanomaterials, metal–organic frameworks, polymer-based composites and surface-active agents. These methods have previously been used for the preparation and modification of electrochemical sensors and, in the current study, carbon paste electrodes are used as a key material in the preparation of electrochemical sensors due to their outstanding catalytic action, electrical conductivity, low background current, mechanical potency, thermal and chemical steadiness and good electronic properties. Furthermore, CPEs, in particular, have hydrophobic surfaces and possess good hydrophobic and electronic associations with polymerized compounds and analytes [21,22]. However, CPEs have the drawback of being predisposed to mechanical damage during use. Mercury has historically been used as an electrode material, primarily as a sphere-shaped drop formed at the end of a glass capillary through which the liquid metal is allowed to flow. Monomers such as amino acids and dyes were used to formulate polymer-layered electrochemical sensors. These electropolymerized films comprise various functional groups, which play a main role in electrocatalytic activity and are responsible for the improved electrochemical activity of the analytes [23–25]. Since these features are helpful, in the current work, we selected carbon as an active base electrode material for LN detection. Electrochemical methods also offer good conductivity, sensitivity and stable voltametric signals for bioactive molecules on the surface of amino-acid-based modified electrode materials. Electro-polymerization amino-acid-modified electrodes require consideration and are applied as biological modifier agents due to their high sensitivity, stability, reproducibility, repeatability and strength relative to the electrode surface [26]. Hence, a poly (GN)-modified film has been developed for LN detection on the CPE surface. Previous modifications of LN electrochemical sensors include boron-doped diamond electrode (BDDE), poly(o-aminophenol) multiwalled carbon nanotubes composite film modified electrode (PoAP/MWCNT), molecularly imprinted polypyrrole–graphene–gold nanoparticle-modified electrode (MIP/G-Au NPs/GCME), PDDA functionalized reduced graphene oxide decorated with gold nanoparticles(Au-NPs/PDDA/rGO/GCE), single-stranded DNA polymers from single-walled carbon nanotubes/gold electrodes modified with nanohybrid (ssDNA/SWCNT/Au), carbon paste enhanced with nano-sized fumed silica (NSF-Si/CPE), hydrochloride with multiwalled carbon nanotubes polymeric alizarin film modified electrode (HCl/MWCNTs PAR-ME), cathodic reduction electrodeposition of a binder-free cobalt-doped Ni-MOF film (BF-Co/Ni-MOF/GCE), reduced graphene oxide modified with antimony and copper nanoparticles (CuNPs/rGOM/GCE), silver vanadate nanorods with silver nanoparticles decorated with nitrogen-doped reduced graphene oxide (Ag-NPs/AgVO₃/N-rGO/SPCE) and multi-walled carbon nanotubes modified with stannic oxide (MWCNT/M-SnO₂). These electrode materials provide acceptable and comparable results when it comes to LOD sensitivity, selectivity and stability in the detection of LN with respect to the current carbon-based electrochemical sensor materials.

The current study tests a simple and green EPGNLCPE for the sensitive electrochemical analysis and quantification of LN in medicinal samples. This sensor is used for the first time for the detection of LN. The interference effect of the modified electrode on LN is also tested in the presence of certain metal ions.

The equipped electrochemical sensor holds potential for electroanalytical application in future analysis of various bio/electroactive and significant components in several resources, including water, food, forensics and medicine, as well as biological, agricultural and environmental materials. Additionally, it may help with energy storage applications such as supercapacitors, solar energy and flexible electronics.

2. Materials and Methods

2.1. Apparatus and Chemicals

The chemicals used were of scientific grade. LN (97.42% purity) and glycine were bought from Merck, India. The stock solutions were diluted with distilled water. Graphite powder (90% purity) and silicone oil—binder materials—are responsible for holding the active material particles within the electrode together to maintain a strong connection between the electrode and the analyte. These binding materials are normally inert (99.99% purity) and were bought from Nice Chemicals, India. The additional synthetic substances were of analytical grade (>98%) and were used without pre-treatment. Phosphate buffer solution (PBS, pH ranging from 3.0 to 6.0) was prepared by blending NaH_2PO_4 (99% purity) and Na_2HPO_4 (99.5% purity). Carbon powder (90% purity and with particle size of 150 mesh) and KCl (99.5% purity) were bought from Nice Chemicals, India. Where needed, the reagents were adjusted using strong acid or strong base. For confirmation, pH values were verified using a pH meter.

2.2. Instrumentation

CV, DPV and EIS estimations were conducted using CH Instruments (CHI 6038E). The three electrodes comprised EPGNLCPE as the working electrode, saturated calomel electrode as the reference electrode and platinum electrode as the auxiliary electrode. The parameters related to the kinetics of EPGNLCPE electrode responses were investigated at various scan rates at room temperature using CV. As the estimations were generally completed in a relatively concentrated electrolyte (0.1 molL^{-1} KCl) with a relatively low concentration of the electroactive substance (1 mmolL^{-1} ferrocyanide) at 100 mV s^{-1} , the impact of ohmic misfortunes was not addressed. CV was used for the assessment of the impact of EPGNLCPE on electrodeposition of the polymer film. BCPE of the finest composition was obtained by mixing 70% carbon powder and 30% silicone oil for about 10–15 min in an agate mortar. A grinder was used to produce a complete and homogeneous paste. The obtained homogeneous CP was closely packed into a Teflon tube with a 3.0 mm inner width, and the electrode surface was smoothed using tissue paper and washed with distilled water. ZEISS instrument (DST-PURSE Laboratory, Mangalore University, India) was used to generate field emission-scanning electron microscopy (FE-SEM) data from all the prepared electrode materials.

2.3. Preparation of BCPE

BCPE was prepared using 70% carbon powder and 30% silicone oil, which were mixed for 10–15 min in an agate mortar. A grinder was used to produce a homogeneous paste. The obtained homogeneous CP was slowly added into a Teflon tube with a 3.0 mm inner width, and the electrode surface was smoothed using tissue paper and washed with distilled water. The obtained material is called BCPE. A copper wire was inserted to provide an electrical connection and was connected to the instruments.

2.4. Preparation of EPGNLCPE

EPGNLCPE was prepared through electrochemical polymerization of GN in 0.2 M PBS of pH 7.0 on the surface of CPE, and complete cycling proceeded of 10 CV cycles at a scan rate of $0.1 \text{ V}\cdot\text{s}^{-1}$ and a potential gap from -1.0 V to 2.0 V . After 10 CV cycles, the modified electrode surface was washed with distilled water to develop a new electrode surface called EPGNLCPE. This modified electrode was used for all analyses.

3. Results and Discussion

3.1. Surface Morphology of EPGNLCPE and BCPE

FE-SEM images show the morphological qualities of the BCPE and EPGNLCPE obtained using FE-SEM. Figure 1 shows the FE-SEM images of the bare and modified electrode samples. Figure 1a shows incomplete irregularly shaped fragments with large cavities, which show the prepared BCPE surface. Meanwhile, Figure 1b shows tiny layers of EPGNLCPE.

CPE that equivalently cover the outside of the incomplete irregularly shaped fragments, which indicate that the carbon paste is covered with EPGNLCPE films.

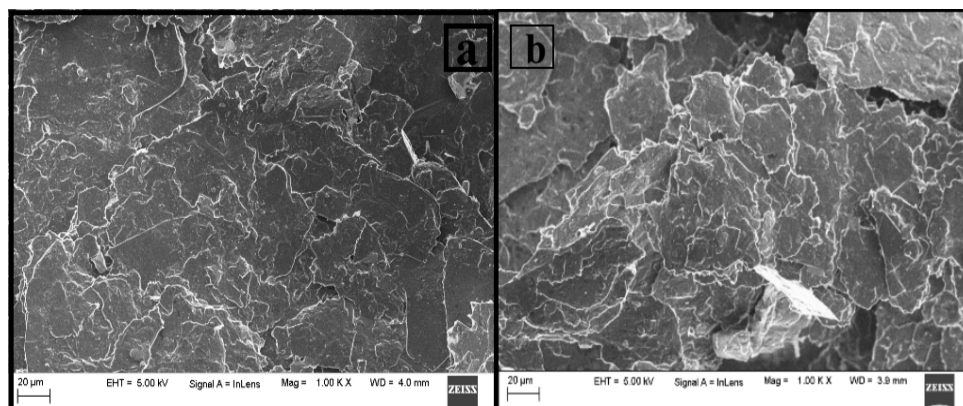


Figure 1. FE-SEM images of (a) BCPE and (b) EPGNLCPE.

3.2. Electrochemical Impedance Analysis of EPGNLCPE and BCPE

Nyquist plots of EPGNLCPE (curve b) and BCPE (curve a) as outlined in Figure 2 show that BCPE yields a bigger semicircle while EPGNLCPE yields a smaller semicircle. The size of the semicircle is directly proportional to the R_{ct} ; hence, the modified electrode EPGNLCPE has less R_{ct} and high conductivity compared with BCPE. Considering the continuous decrease in the electron transfer resistance (R_{ct}) values of EPGNLCPE, one might say that electron transfer between the $[\text{Fe}(\text{CN})_6]^{3-}/[\text{Fe}(\text{CN})_6]^{4-}$ redox probe and the sensor surface is quicker due to the EPGNLCPE. This demonstrates that modification of the polymer improved the conductivity of the sensor, resulting in easy electron transfer.

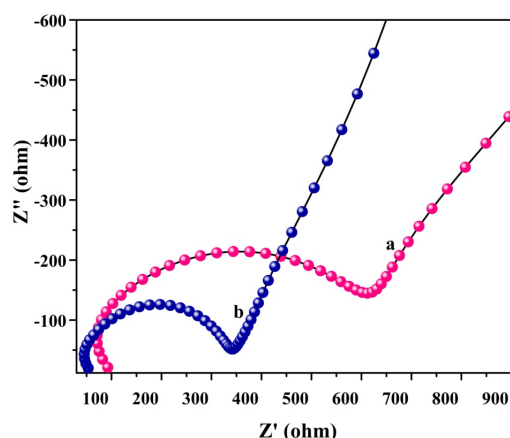


Figure 2. Nyquist plots of BCPE (a) and EPGNLCPE (b).

3.3. Electrochemical Behavior of $\text{K}_4\text{Fe}(\text{CN})_6$ on the Modified Electrode

The electrochemical behavior of $\text{K}_4\text{Fe}(\text{CN})_6$ in 1 mL of 0.1 M KCl at a scan rate of 0.1 V s^{-1} was investigated for both electrodes. Increased peak current was observed for EPGNLCPE (curve b), which was highly improved compared to the signal obtained for BCPE (curve a), as shown in Figure 3. EPGNLCPE displayed a high oxidation peak current. Conversely, a low peak current was seen with BCPE under similar conditions. This shows that the presence of the modifier promotes charge transfer reactions, with greater electron movement and, consequently, higher conductivity.

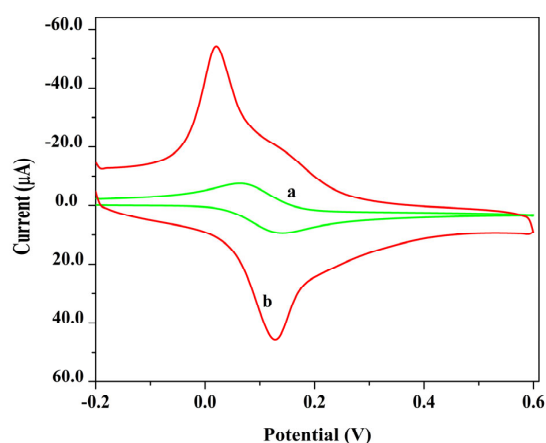


Figure 3. CV of 1.0 mM $K_4Fe(CN)_6$ in a probe of 0.1 M KCl on EPGNLCPE (a) and BCPE (b).

EPGNLCPE and BCPE surface areas were assessed using a solution of 1.0 mM $K_4Fe(CN)_6$ in a probe of 0.1 M KCl. To follow the reversible interaction, the Randles–Sevcik equation [18] was applied as follows:

$$I_{pa} = 2.69 \times 10^5 n^{3/2} A C_0 D^{1/2} \nu^{1/2} \quad (1)$$

The surface area of the sensor was calculated from the above relationship. The sensor surface areas of EPGNLCPE and BCPE were 0.195 and 0.04 cm², respectively. EPGNLCPE had a larger surface area than BCPE.

3.4. Electro-Polymerization of EPGNLCPE on BCPE

In the fabrication of electrochemically polymerized glycine layered on a BCPE surface (EPGNLCPE), the number of electrochemically polymerized voltammogram cycles was optimized to achieve maximum electrochemical activity for LN. To that end, we varied the CV cycles: 5, 10, 15 and 20. Here, 10 CV cycles displayed the maximum current response against LN (Figure 4B); hence, 10 CV cycles were chosen as the best for EPGNLCPE and were used throughout the experiment. Electrochemical polymerization of 1 mM GN was conducted using the CV method and the plot is shown in Figure 4A. The cell comprises 0.2 M PBS, pH 7.0, as supporting media with a sweep rate of 0.1 Vs⁻¹ on BCPE. The potential gap ranges from −1.0 V to 2.0 V. Here, we can see the linear reliance of peak current on the polymerization cycle, which provides some insight into the development of a polymeric film on the BCPE surface. After the electrochemical polymerization process, the obtained electrode is washed with distilled water and used for further analysis.

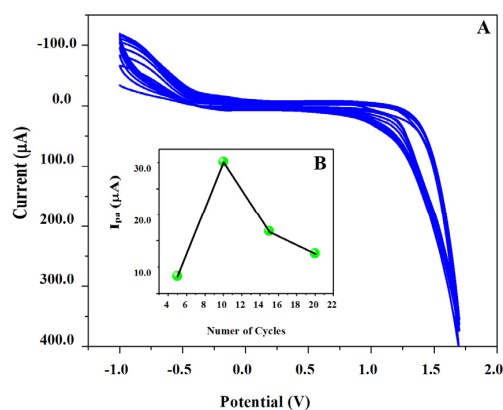


Figure 4. Cyclic voltammograms of 0.1 M glycine layered on carbon paste electrode with 10 electropolymerized cycles with a 0.1 Vs⁻¹ scan rate (A). Plot of the number of CV cycles vs. I_{pa} (B).

3.5. Electrocatalytic Oxidation of LN on EPGNLCPE and BCPE

Figure 5 represents the CV reactions of the electrochemical oxidation of 0.1 mM LN in 0.2 M PBS, pH 4.0, on the surface of the electrodes, BCPE (curve a) and EPGNLCPE (curve c). As can be seen in the figure, the anodic peak potential levels for LN oxidation on EPGNLCPE and BCPE are 101 and 106 mV, respectively. These outcomes show that the peak potential for LN oxidation at the EPGNLCPE sensor shifts by 5 mV toward negative potential when contrasted with BCPE. Nonetheless, EPGNLCPE shows higher anodic peak current for the oxidation of LN compared with BCPE, demonstrating that the polymer-fabricated sensor has essentially enhanced the performance of the sensor toward LN oxidation. The anodic peak was not observed in the absence of LN, as revealed by curve b.

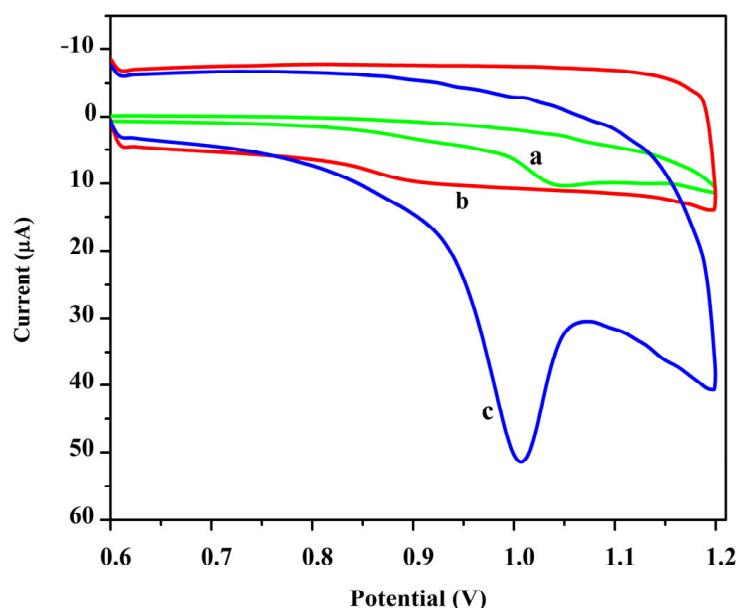


Figure 5. CVs observed for 0.1 M of LN on BCPE (a), EPGNLCPE (c) and a blank (b).

3.6. Effect of pH

The effect of pH was examined as it is significant in determining the electrochemical performance of the analyte and the sensor because it affects the mechanism of the electrochemical response of the analyte. In this analysis, the effect of pH was evaluated using PBS of different pH—ranging from 3.0 to 6.0—in voltametric studies (Figure 6a). The results show that raising the pH increased the negativity of the peak potential. This indicates the association of H^+ in the electrochemical response. Increasing the pH from 3.0 to 4.0 and peak current also increases. (Figure 6b). Hence, we chose 4 as the ideal pH for the electrochemical analysis, and all remaining analyses were performed at this pH. Anodic peak potentials changed linearly with pH (Figure 6c) as per the below linear regression equation: $E_{pa}(V) = 1.1790 \pm 436 \text{ pH} (R^2 = 0.99)$

The slope observed at 0.0436 V shows the association with pH^{-1} and shows that the number of electrons and H^+ ions involved in the reaction are equal.

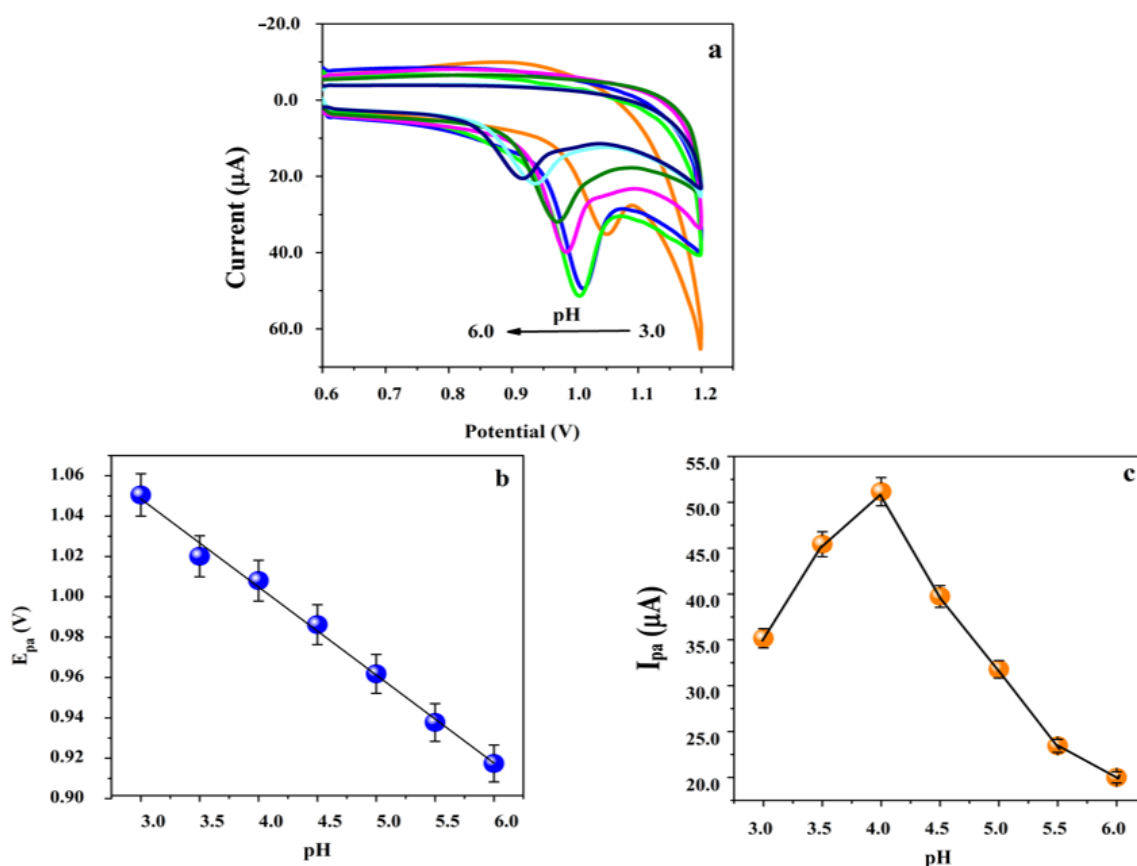


Figure 6. (a) CVs of 0.1 mM LN in 0.2 M PBS with pH ranging from 3.0 to 6.0 measured at 0.1 Vs⁻¹ scan rate. Plots of (b) E_{pa} vs. pH and (c) I_{pa} vs. pH.

3.7. Influence of Scan Rate

The effect of scan rate on the oxidation reaction of 0.1 mM LN in 0.2 M PBS, pH 4.0, onEPGNLCPEwas electrochemically analyzed to determine the reliability of the oxidation peak current and peak potential during variations in the scan rate. The peak current is directly proportional to the scan rate of 0.1 Vs⁻¹ (Figure 7).The examination was conducted in a potential of 0.6–1.2 at a scan rate of 0.1 Vs⁻¹. The plot of $\log I_{pa}$ as a function of $\log v$ (Figure 7b) represents the diffusion-controlled electrode process since its slope of 0.475 is near to the theoretical value of one, suggesting that LN oxidation on the surface of EPGNLCPE is dominated by surface diffusion-controlled processes [27] associated with the accompanying equation: $\log I_{pa}(A) = 0.475 \log v (V s^{-1}) - 3.94$; $R^2 = 0.99$. The plot of E_{pa} versus \log scan rate (Figure 7c) shows that peak potential increased in positivity, with an increase in the scan rate indicating the irreversibility of the electrode process. The relationship between peak potential and the log of scan rate was $E_{pa} (V) = 0.065 \log v (V s^{-1}) - 1.0818$; $R^2 = 0.99$. The Laviron equation [28] can be used to calculate the number of electrons required to make the electrode process irreversible.

$$E_{pa} = E^0 + \frac{2.303RT}{(1-\alpha)nF} \log v + \frac{2.303RT}{(1-\alpha)nF} \log \left(\frac{(1-\alpha)nF}{RTK^0} \right) \quad (2)$$

The linear plot of E_{pa} vs. $\log v$ was obtained using the above equation. From the slope of the plot αn , α is the transfer coefficient, and n is the number of electrons transferred, which was determined to be 1.9686(2). For an irreversible electrode response, α was thought to be 0.5; consequently, n was determined to be 2. Hence, the same number of electrons and protons was involved in the reaction, and the electrode response was determined to be 1. The probable electrode transfer response in the oxidation reaction process was

determined. The surface coverage concentration (Γ) of LN at EPGNLCPE was calculated using the following relative Equation (3).

$$\Gamma = \frac{Q}{nFA} \quad (3)$$

The calculated value of the surface coverage concentration of LN on EPGNLCPE was found to be 1.035 nm/cm^2 . The probable electrochemical oxidation reaction of LN is shown in Figure 8.

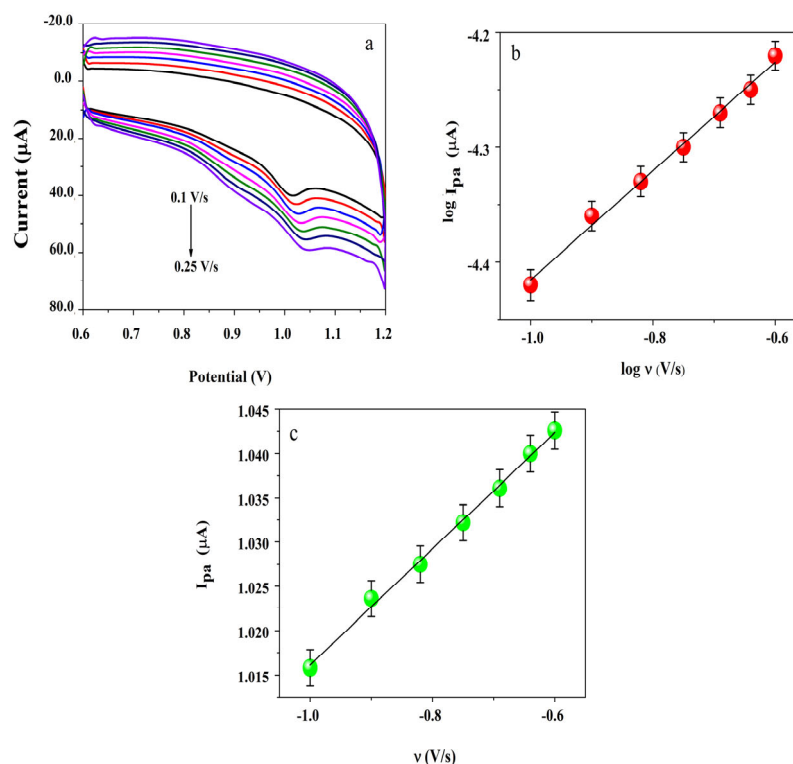


Figure 7. (a) CVs of 0.1 M LN at optimal pH with various scan rates ($0.1\text{--}0.25 \text{ Vs}^{-1}$). Plots of (b) $\log I_{pa}$ vs. $\log v$ and (c) E_{pa} vs. $\log v$.

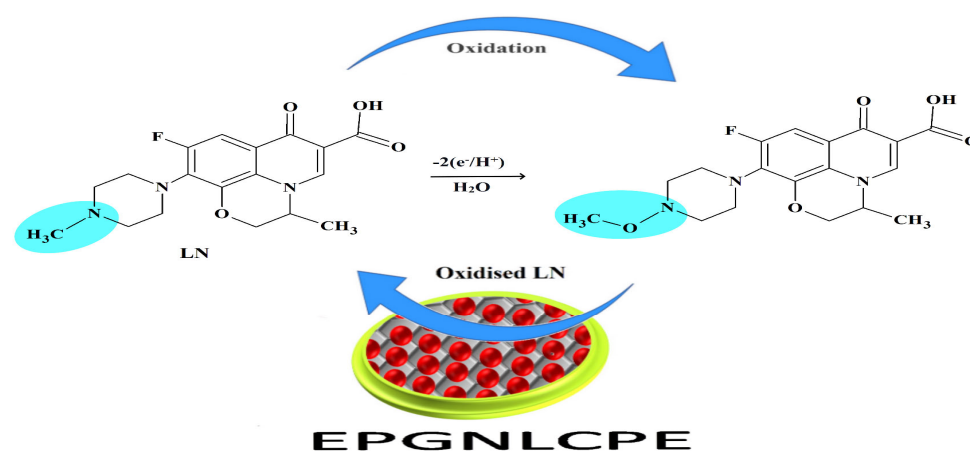


Figure 8. Probable electrochemical redox reaction of LN on EPGNLCPE surface.

3.8. Interference Analysis

The selectivity of the planned electrode sensor for LN in the presence of certain metal ions and organic molecules in 0.2 M PBS, pH 4.0, was examined at a scan rate of 0.1 Vs^{-1} .

These metal ions are Al^{3+} , Cu^{2+} , K^+ , Mg^{2+} , Na^+ , and Pb^{2+} , and the organic molecules RF, DA, AA, and TZ—present at a concentration ratio of 1:1—using CV methods. The corresponding results are shown in Figure 9. The RSD values of the anodic peak current acquired for the analytes were under 0.9%, showing that these compounds do not exert any effect on LN. The comparative error of the valuation of LN in the presence of these interferences is below $\pm 5\%$. This indicates that the proposed sensor has acceptable selectivity and low interference.

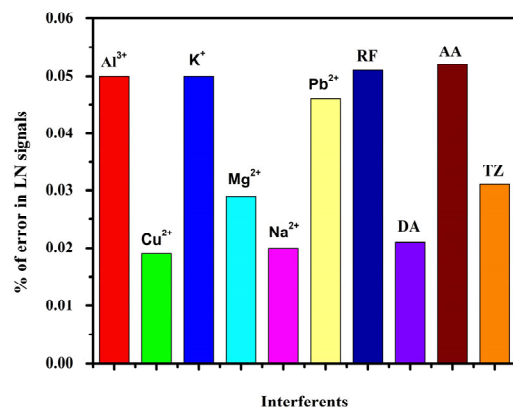


Figure 9. Plot of interferences vs. % of error in LN signal and cyclic voltammograms of different metal ions in the presence of LN.

3.9. Limit of Detection Using EPGNLCPE

The LOD and LOQ of EPGNLCPE were determined by changing the concentration of LN in 0.2 M PBS, pH 4.0. The use of the DPV strategy in the field of electrochemical examination offers a significant option for the detection of different bioactive species. The point of this study was to determine the limit of detection (LOD) of LN in 0.2 M PBS, pH 4.0, using EPGNLCPE. LOD is the lowest concentration that produces an electrical signal that is at least more than three times the amplitude of the background current. Here, voltammograms in 0.2 M PBS, pH 4.0, were recorded from 0 to 1.1 V. The analysis focused on the anodic oxidation of LN over 30 to 90 μM as the graph in Figure 10a shows. To assess LOD using EPGNLCPE, the calibration plot of the oxidation current versus concentration of LN was charted, as shown in Figure 10b.

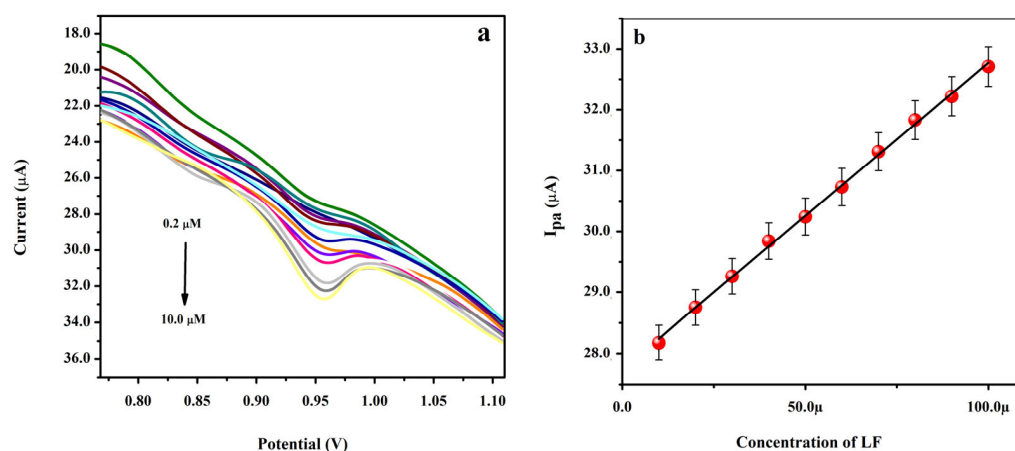


Figure 10. (a) Differential pulse voltammetry of LN with concentrations varying from 30.0 μM to 90.0 μM in 0.2 M phosphate buffer on EPGNLCPE measured at a 0.1 Vs^{-1} scan rate. (b) Plot of LN vs. I_{pa} .

LOD and LOQ were determined to be $8.436 \times 10^{-7} \text{ M}$ and $2.812 \times 10^{-6} \text{ M}$, respectively, using the following equations: $\text{LOD} = 3 \times \sigma/S$ and $\text{LOQ} = 10 \times \sigma/S$, where “ σ ” is the

standard deviation of the intercept and “S” is the slope of the curve. Table 1 shows comparisons of LN assurances from a few reports mentioned in the literature. Compared with the different reports, our study had the lowest LOD [28–39].

Table 1. Comparisons of present LOD of LN in different electrodes, and methods used in previous LN reports.

Electrodes	Method	Linear Range (mM)	LOD (mM)	References
BDDE	SWV	10–80.9	2.88	[28]
	CV	48–100	10.01	
PoAP/MWCNT	DPV	3.0–200	1.00	[29]
MIP/G-Au NPs/GCME	DPV	1–100	0.53	[30]
Au-NPs/PDDA/rGO/GCE	LSV	10.0–800	3900.0	[31]
dsDNA-CPE	DPV	0.5–5.0	100.0	[32]
ssDNA/SWCNT/Au	SWV	1.0–10	0.0752	[33]
NSF-Si/CPE	DPV	2–1000	0.09	[34]
HCl/MWCNTs PAR-ME	LSV	5–100	0.4	[35]
BF-Co/Ni-MOF/GCE	LSV	0.1–500.0	0.022	[36]
CuNPs/rGOM/GCE	DPV	0.1–2.5	10.00	[37]
Ag-NPs/AgVO ₃ /N-rGO/SPCE	DPV	0.09–671	0.00792	[38]
MWCNT/M-SnO ₂	DPV	1.0–9.9	0.20	[39]
EPGNLCPE	DPV	30–90	0.678	Present Work

The electrochemical mechanism of LN and oxidation reactions is shown in Figure 11. Here, LN molecules are oxidized by losing one proton and one electron, generating oxidized LN products in the presence of an electrocatalyst (modified electrode material). This reaction proceeds through a heterogenous electrocatalytic reaction.

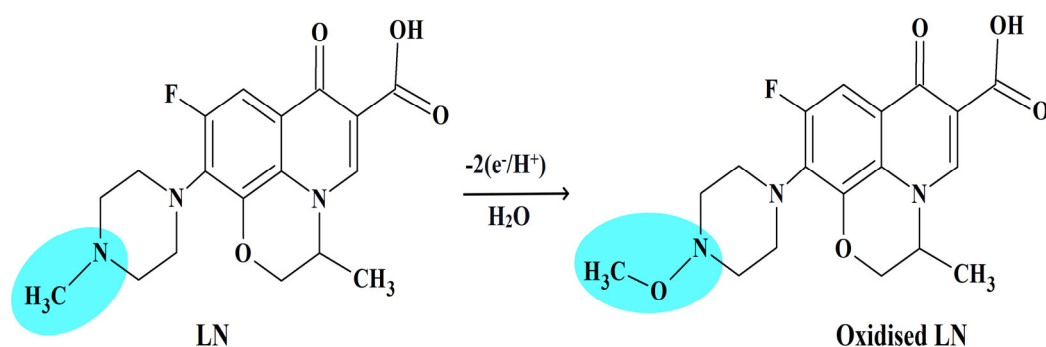


Figure 11. LN reactions [18].

3.10. Reproducibility, Repeatability and Stability Studies

The reproducibility, repeatability and stability of the projected electrochemical sensor EPGNLCPE for the LN redox reaction in 0.2 M PBS, pH 4.0, were studied using CV methods at a scan rate of 0.1 Vs⁻¹. The fabricated sensors were stored at room temperature to analyze their stability. The sensor was used to measure LN assurance using CV. The results showed that the EPGNLCPE current response saved 98.93% of its underlying reaction after 15 days. The repeatability of the fabricated sensors was determined by analyzing five replicates of a similar electrolyte containing LN using a CV system, and the relative standard deviation (RSD) was calculated and found to be 1.748%. Moreover, the reproducibility of the EPGNLCPE sensor was analyzed using five EPGNLCPE independently prepared

following the same preparation process, and the RSD was determined to be 0.11%. The results showed that EPGNLCPE had great reproducibility in LN assurance.

3.11. Application to a Pharmaceutical Preparation

The proposed analysis of 0.2 M PBS, pH 4.0, using the CV technique was effectively applied in the assurance of LN in LN tablets. Good recoveries, \pm RSD% (99.80 ± 0.991), were acquired. The legitimacy of the proposed strategy was evaluated using the standard addition procedure, with great recovery— \pm RSD% (99.98 ± 0.828). EPGNLCPE resulted in good LN recovery from LN tablet samples, ranging from 98.9% to 99.4%, and the results are shown in Table 2.

Table 2. Recovery data of LN in tap tablet samples.

Sample	Trial	Added (μ M)	Found (μ M)	Recovery %
Tablets	1.	50	49.46	98.9
	2.	60	59.5	99.2
	3.	70	69.6	99.4

4. Conclusions

In the present study, electropolymerized glycine was successfully produced on the CPE layer, and this electrode was used to detect LN. Electrochemical oxidation on the surfaces of EPGNLCPE and BCPE materials was successfully studied. The developed electrochemical sensor was used, for the first time, for the quantification of LN in both commercial and tablet samples. The BCPE and EPGNLCPE materials were prepared successfully using a modest, easy and low-cost procedure. Additionally, these materials were characterized successfully using CV, DPV, FE-SEM and EIS, and the results were compared. The effect of the scan rate shows that EPGNLCPE is diffusion-controlled for the LN oxidation reaction with the transfer of one electron and one proton. Furthermore, the effect of pH on the electrochemical oxidation of LN was studied and the results showed that the electrochemical oxidation potential and current are pH-dependent, with the transfer of the same electron and proton number (1:1). The efficacy of EPGNLCPE for LN was studied using CV and DPV methods, with a good linear range and low LOD were obtained. The EPGNLCPE material showed a clear-cut anti-interference capability with fine selectivity for LN detection in the presence of certain metal ions, and this result was confirmed based on the error percentage of the LN signal (less than $\pm 5\%$). All these aspects associated with the proposed sensor and methodology make this the best device for the analysis of various electroactive compounds in food, environmental, biological and medicinal samples.

Author Contributions: Methodology, J.G.M.; Software, K.B., J.G.M. and M.S.; Validation, K.B.; Formal Analysis, K.B. and J.G.M.; Data Curation, K.B., J.G.M., K.P.M. and A.M.T.; Visualization, M.D.A. All authors have read and agreed to the published version of the manuscript.

Funding: This work is funded by the SC/ST Fellowship of Mangalore University (grant number MU/SCT RF/CR5/2019-20/SCT-1) and the Researchers Supporting Project of King Saud University, Riyadh, Saudi Arabia (grant number RSP-2023R267).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: 1. Kanthappa is grateful to the financial support from the SC/ST Fellowship (Fellowship No. MU/SCT RF/CR5/2019-20/SCT-1), Mangalore University. 2. Ammar M. TIGHEZZA and Munirah D. Albaqami are grateful for the Researchers Supporting Project Number (RSP-2023R267) King Saud University, Riyadh, Saudi Arabia.

Conflicts of Interest: The authors declare no conflict of interest with any organization or reviewers.

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