A Sensitive Voltammetric Sensor Based on Synergistic Effect of Graphene–Polyaniline Hybrid Film for Quantification of Calcium Antagonist Lercanidipine

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ABSTRACT: A chemically modified sensor based on the synergistic effect of graphene and polyaniline for electrochemical sensing of calcium antagonist lercanidipine (LCP) has been developed. Scanning electron microscopy, electrochemical impedance spectroscopy, square-wave voltammetry, and cyclic voltammetry were utilized to characterize the morphology and electroanalytical performance of the fabricated sensor. Under optimized conditions, reduction peak current was linear over the wide concentration range from 5 to 125 ng mL^{-1} with correlation coefficient of 0.9998. The limit of detection and the limit of quantification were found to be 1.94 and 5.89 ng mL^{-1}. The developed sensor also exhibited good reproducibility and long-term stability. In addition, the proposed method was successfully applied to the determination of LCP in pharmaceutical formulation which is proved by recovery studies. Graphene–polyaniline composites are expected to be promising material for biosensing applications because of the ease of fabrication, excellent electrochemical performance, and high electroactive surface area. © 2014 Wiley Periodicals, Inc. J. Appl. Polym. Sci. **2014**, *131*, 40959.

INTRODUCTION

Electrochemical sensors are appealing as they have many potential applications in various fields, including environmental monitoring of trace metals, clinical testing of toxic metals, medical diagnosis, and routine analysis in food and pharmaceutical industries.[1–5] Various nanomaterials have been explored such as graphene, carbon nanotubes, conducting polymers, and metal oxides for the modification of electrodes to enhance the sensitivity and selectivity of the electrochemical sensor.[6–12]

In particular, polymer nanocomposites based on the conductive polymers have been extensively studied. Out of various conductive polymers, polyaniline (Pani) has been considered for many potential applications owing to its wide range of conductivity, film-forming ability, low cost, facile synthesis, biocompatible nature, and environmental resistance.[13–16] On the other hand, graphene has also been become a material of choice owing to its unique electronic properties, large surface-to-volume ratio, high conductivity, and small band gap. The structure of graphene is based on a single-atom-thick sheet of carbon atoms those are densely packed in a honeycomb structure; its perfect two-dimensional structure provides a large detection area. In addition, graphene-based electrochemical sensors can also have

a much higher sensitivity because of the low electronic noise from thermal effect.[17–23] Graphene–polyaniline (Grp-Pani) nanocomposite recently has been applied as electrochemical sensors to realize the potential benefits of lower overvoltages, higher currents, and enhanced electron transfer rates.[24–26]

Lercanidipine (Scheme 1) [2-[(3,3-diphenylpropyl) methylamine]-1,1-dimethylethyl methyl 1,4-dihydro-2,6-dimethyl-4-(3-nitrophenyl)−3,5pyridine carboxylic ester] (LCP) is a novel dihydropyridine calcium antagonist which is used in the treatment of hypertension, angina pectoris, and Raynaud's syndrome based on its selectivity and specificity on the smooth vascular cells.[27]

The review of the literature revealed that few methods have been reported for the determination of the LCP in human plasma and pharmaceutical formulation namely capillary electrophoresis (CE), high-performance liquid chromatography (HPLC), and liquid chromatography–mass spectrometry.[28–30] However, these methods are expensive and require sophisticated instrumentation, highly skilled personnel, and time-consuming extraction procedures which restrict their use in routine analysis. Among the most commonly used instrumental techniques, electroanalytical approach is one of the choices owing to its
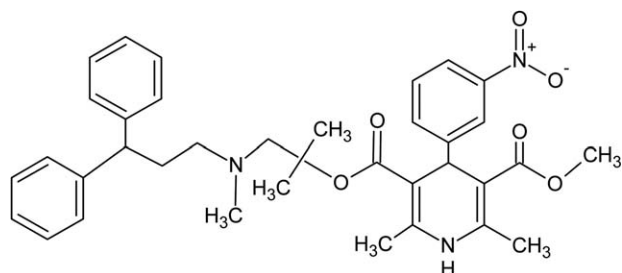
Scheme 1. Chemical structure of LCP.

certain advantages, such as relatively low operational cost, moderate or no sample preparation, no time consumption, high sensitivity with selectivity, and its dynamic range offer.[31–33] Electro-oxidation of LCP using voltammetric techniques has been reported.[34,35] However, there is no report available on the study of electroreduction of LCP at Grp–Pani hybrid film sensor. In the present voltammetric method, the detection limit is found to be at the nanogram level and makes it more sensitive than already reported methods.

EXPERIMENTAL

Chemicals and Reagents

Graphene (12 nm) was procured from Graphene Laboratories, USA. All other chemicals used were of analytical reagent grade. Aniline was distilled under reduced pressure prior to use and other reagents were employed without further purification. LCP (99% purity) was obtained from Glenmark Generics Ankleshwar, India, and was used as received. Tablets containing LCP (Lotensyl) labeled 10 mg were obtained from commercial sources. KCl (1.0 mol L^{-1}) solution was prepared in double-distilled water and used as supporting electrolyte. A stock solution of LCP (1.0 mg mL^{-1}) was prepared in *N,N*-dimethylformamide (DMF). The solutions for recording voltammograms were prepared by mixing appropriate volumes of stock solution, buffers, and 1.0 mol L^{-1} of KCl.

Apparatus and Measurements

Electrochemical measurements were performed using a µ AUTOLAB TYPE III (Eco-Chemie B.V., Utrecht, The Netherlands) potentiostat–galvanostat with 757VA computrace software. The Grp–Pani/glassy carbon electrode (GCE) was used as working electrode, Ag/AgCl as reference electrode, and graphite as auxiliary electrode. All the solutions examined by electrochemical techniques were purged for 15–20 min with purified nitrogen gas after which a continuous stream of nitrogen was passed over the solutions during the measurements. All the pH-metric measurements were made on a Decible DB-1011 digital pH meter fitted with a glass electrode, which was previously standardized with buffers of known pH.

Synthesis of Pani Nanopowder

The Pani nanopowder was prepared by interfacial polymerization method using ammonium persulfate as an oxidizing agent. Typical synthesis consists of the following steps: (i) 1 mL of pre-distilled aniline was dissolved in 50 mL of CCl_4 and labeled it as Solution A. (ii) In brief, 450 mg of ammonium persulfate was dissolved in 0.1*M* of camphor sulfonic acid and stirred

until a homogenous solution is obtained and labeled it as Solution B. (iii) Then Solution A was added into the Mixture B with vigorous stirring for 10 min by keeping it at temperature (4–5°C) for 18–20 h. Polymerization reaction occurred at interfacial layer between two immiscible solvents. (iv) Then prepared nanopowder was extracted by centrifugation, washed with deionized water and ethyl alcohol for several times, and left it overnight for drying at room temperature.[36–38]

Fabrication of Grp–Pani/GCE Hybrid Film Sensor

Prior to modification, the GCE was polished with 0.05–0.1 µm of alumina powder on soft microcloth pads and then rinsed thoroughly with double-distilled water. Then, the electrode was sonicated in deionized water, ethanol for 5 min to remove adsorbed alumina particles on the electrode surface. Pani and Grp (ratio, 1 : 2) were dispersed in DMF and sonicated for 60 min to give a suspension of 1 mg mL^{-1}. In total, 8 µL of the above suspension was casted by micropipette onto the surface of freshly polished GCE and dried at room temperature for ∼20–30 min to obtain the Grp–Pani/GCE sensor. Similar suspensions of individual 1 mg mL^{-1} of Pani and Grp were prepared in DMF to obtain Pani/GCE, Grp/GCE sensors and were used for comparative study. After each measurement, the modified sensors were regenerated by thoroughly washing the electrode with double-distilled water.

## RESULTS AND DISCUSSION

Characterization of the Sensor

Scanning Electron Microscopy. Figure 1 shows the changes in morphology obtained by scanning electron microscopy (SEM) for Pani, Grp, and Grp–Pani films. Individually, the layers of Grp and Pani were observed as sheet and rod forms, respectively [Figure 1(A, B)]. The surface morphology of the Pani film changed considerably with the addition of Grp. Figure 1(C) shows that the sheets of Grp appeared to be deposited within the Pani layer. The Pani rods seem to be incorporated within the edges of the Grp matrix.

Particle Size Analysis. The specific surface area and pore structure of the Pani nanopowder were determined by using surface area and pore size analyzer (Micromeritics). The specific surface area was calculated by Brunauer–Emmett–Teller (BET) equation.[39] It was found that the BET surface area, pore volume, and pore radius of the Pani nanopowder were 95.062 m^2/g, 1.659 cc/g, and 15.271 Å, respectively.

Cyclic Voltammetry. The cyclic voltammetric analysis with redox reactions of $Fe(CN)_6^{3-}/Fe(CN)_6^{4-}$ was used to evaluate the electrochemical behavior of the bare GCE and modified electrodes that is Pani/GCE, Grp/GCE, and Grp–Pani/GCE in 1.0 m*M* of $K_3Fe(CN)_6$ solution at a scan rate of 100 mV s^{-1}. It is inferred that on scanning the potential from −0.2 to +0.8, Grp–Pani/GCE electrode shows the remarkable increment in current value as compared to the Grp/GCE, Pani/GCE, and bare GCE electrodes (Figure 2). The significant increment in peak current with decreasing potential clearly demonstrates that Grp–Pani nanocomposite acts as an efficient electron mediator for the study of redox species. The effective surface area of the
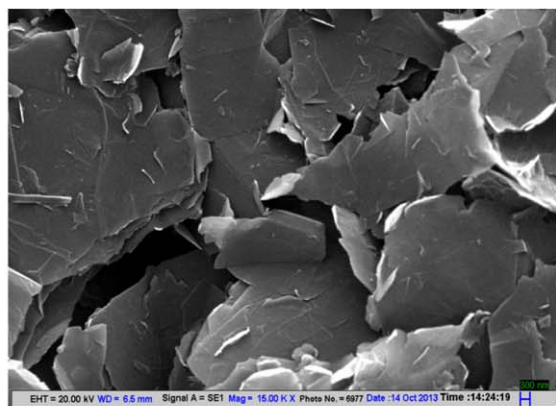
(A)
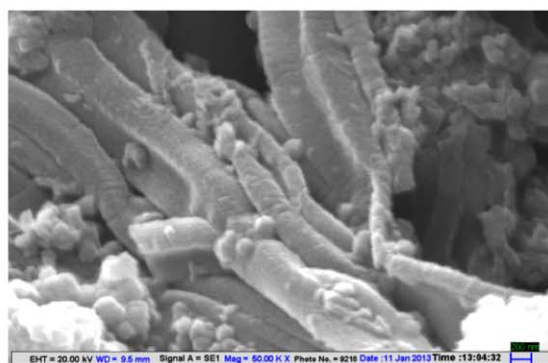
(B)
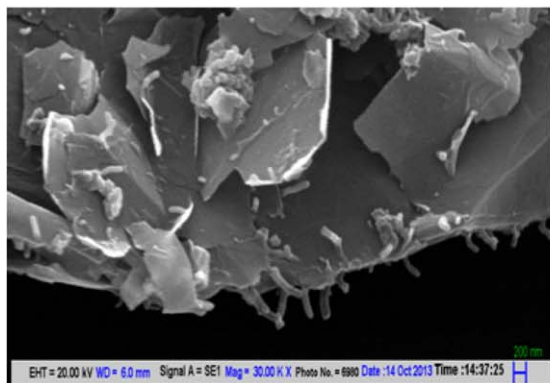
(C)

Figure 1. Scanning electron micrographs of (A) Pani, (B) Grp, and (C) Grp–Pani nanocomposite. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

modified electrodes can be determined using the Randles–Sevcik equation.[40]

$$I_p = (2.69 \times 10^5) \; n^{3/2} \; A \; C_0 D_0^{1/2} v^{1/2} \tag{1}$$

For $K_3Fe(CN)_6$, $n = 1$ and $D = 7.6 \times 10^{-6} \; cm^2 \; S^{-1}$. From this equation, the effective surface area (A) is proportional to the value of $I_p/v^{1/2}$. The effective surface areas of the bare and modified GCEs were calculated from the Randles slope and are summarized in Table I. It clearly demonstrates that the high
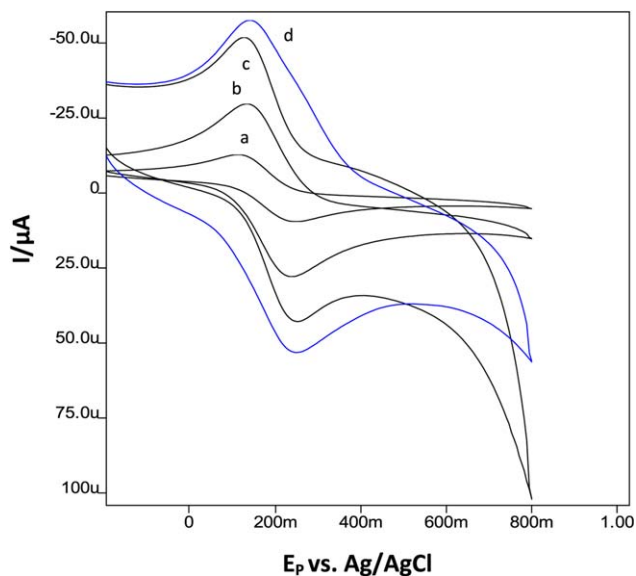

Figure 2. Comparison of the cyclic voltammetric behavior at modified electrode in 1.0 mM of $K_3Fe(CN)_6$ (A) GCE, (B) Pani/GCE, (C) Grp/GCE, and (D) Grp–Pani/GCE. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

sensitivity of Grp–Pani/GCE is owing to the combined influence of Pani and Grp present in the matrix.

Electrochemical Impedance Spectroscopy. Electrochemical impedance spectroscopy study was performed to evaluate the charge-transfer resistance (R_{CT}) at the interface of electrodes after surface modification. The electronic transfer properties of the bare and modified GCEs were measured at AC amplitude of 0.01 V and scanning frequency from 0.1 to 100,000 Hz. Figure 3(A) shows the Nyquist plots of the bare GCE, Pani/GCE, Grp/GCE, and Grp–Pani/GCE in 1.0 mM of $K_3Fe(CN)_6$ containing 0.1M of KCl. By fitting the data using an appropriate equivalent circuit [Figure 3(B)], the values of charge-transfer resistance were determined (Table I). It clearly demonstrates that the lower charge transfer resistance of Grp–Pani/GCE hybrid film can be attributed to the excellent conductivity and the large surface area. The decrease in the charge-transfer resistance can be related to the electrode coverage and is given by the following equation:

Table I. Characterization and Performance Parameters of the Fabricated Grp–Pani/GCE Hybrid Film Sensor

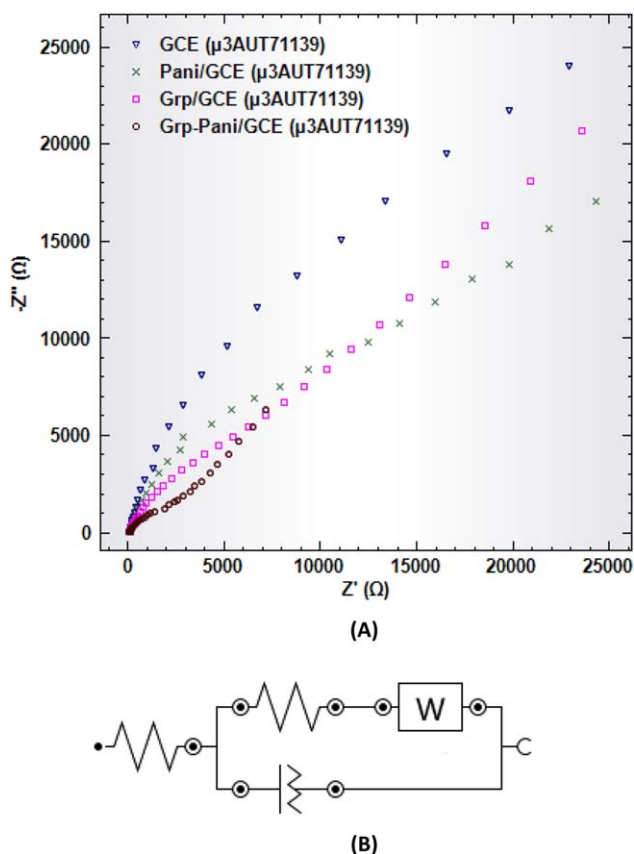| Working electrode	(1 mM $K_3Fe(CN)_6$)		I (μA) (80 ng mL^{-1} LCP)	
	R_{CT} (kΩ)	A (cm^2)	SWV	CV
Bare GCE	27.9	1.0×10^{-3}	8.60	2.17
Pani/GCE	13.4	1.2×10^{-3}	11.7	3.23
Grp/GCE	8.35	1.5×10^{-3}	14.2	3.86
Grp–Pani/GCE	2.32	1.9×10^{-3}	17.5	6.48

Figure 3. (A) A Nyquist plots of 1.0 mM of $K_3Fe(CN)_6$ at GCE (A, triangle), Pani/GCE (A, cross), Grp/GCE (A, square), Grp–Pani/GCE (A, circle), and (B) corresponding equivalent circuit. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

$$(1\text{-}\emptyset) = \frac{R_{CT}^0}{R_{CT}} \tag{2}$$

Where ø is the apparent electrode coverage and R_{CT}^0 and R_{CT} are the charge-transfer resistances measured at bare and modified GCE.[41] In the equivalent circuit R_s, R_{ct}, W, and Q represent the solution resistance, charge-transfer resistance, the Warburg impedance, and the constant phase element. A constant phase element was used instead of the pure capacitance because of the depression in the high-frequency semicircle and again the line that appeared at low frequency is because of diffusion of the electroactive species in the bulk of the solution.

Electrocatalytic Reduction of LCP at Grp–Pani/GCE

To investigate the voltammetric performance of Grp–Pani/GCE hybrid film sensor, electrocatalytic reduction of LCP was carried out by square-wave and cyclic voltammetry. Figure 4 shows square wave voltammograms of LCP at bare GCE, Pani/GCE, Grp/GCE, and Grp–Pani/GCE. To accomplish this purpose, the electrochemical response of a solution of LCP (80 ng mL^{-1}) was examined in the potential range from -0.2 to -1.6 V versus Ag/AgCl. LCP in Britton–Robinson (BR) buffer (pH = 4.5) exhibits a well-defined cathodic peak. The electrochemical response is irreversible as no anodic peak is observed during the reverse scan. It is inferred that the LCP currents obtained at Grp–Pani/GCE are larger as compared to Pani/GCE, Grp/GCE,

and bare GCE, indicating that the coexistence of Pani and Grp in a hybrid film enhances the voltammetric performance of the sensor. As the square-wave voltammetry (SWV) is more sensitive than cyclic voltammetry, the detailed studies of LCP reduction are carried out using SWV.

Effect of Electrolyte and pH. The choice of the supporting electrolyte is an important parameter in electroanalytical studies because its composition and pH affect the properties of the solution as well as the electrode–solution interface.[42] In this study, various buffers such as BR, acetate, and phosphate were used as supporting electrolytes for the reduction of LCP at Grp–Pani/GCE hybrid film sensor. LCP gave one well-defined cathodic peak with respect to the signal enhancement and the symmetric peak profile in BR-buffered solution. Effect of pH on the reduction of LCP was studied in the range of 2.5–12.0 at a constant concentration (70 ng mL^{-1}) with same sensor. Optimum peak current was achieved at pH 4.5; with the rise in pH, the peak current decreases up to pH 6.5, afterward peak become vanished which indicates the involvement of protons in the electrode process, and finally dislocated in neutral as well as alkaline pH owing to lower number of available protons [Figure 5(A)]. Thus, BR buffer of pH 4.5 was selected for the study of LCP reduction. The increase in pH shifted the LCP reduction potential toward negative side which reveals that the pH of the supporting electrolyte exerted a significant influence on the electroreduction of LCP. The linear dependence of reduction peak potential on pH can be expressed by the following relationship:

$$\frac{E^p}{V}\left(vs.\ \frac{Ag}{AgCl}\right) = -0.0562\ pH - 0.3755;\ r^2 = 0.9966 \tag{3}$$

The observed slope of 0.056 V/pH clearly indicates that an equal number of electrons and protons are involved in the reduction of LCP.

Effect of Material Loading. The electrochemical behavior of Grp–Pani-modified electrodes is capable of improving the
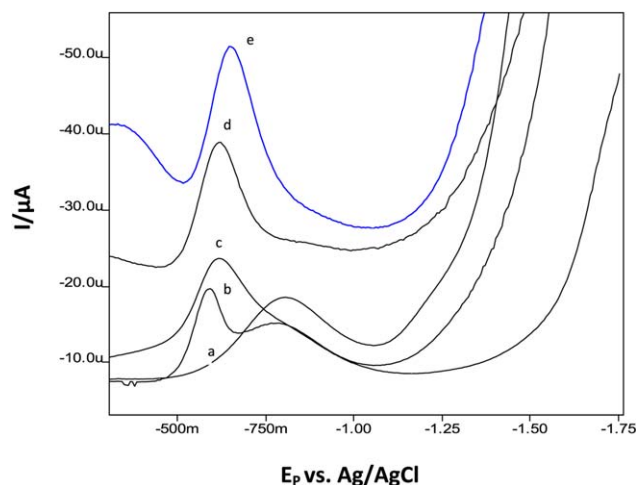

Figure 4. Electrocatalytic effect of fabricated sensor toward the LCP reduction. Square wave voltammograms, blank (curve a), at bare GCE (curve b), Pani/GCE (curve c), Grp/GCE (curve d), and Grp–Pani/GCE (curve e). [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]
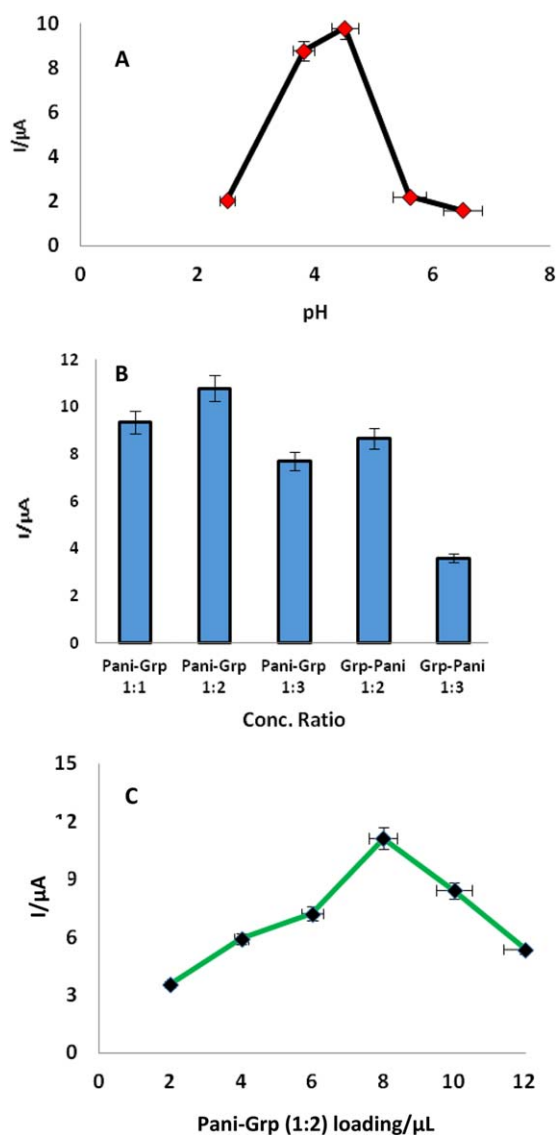
Figure 5. Optimization of experimental conditions for the reduction of LCP at Grp–Pani/GCE sensor including 5% of error bar graphs on three replicate analyses. (A) Effect of pH. (B) Optimization of Pani–Grp concentration ratio. (C) Optimal loading of Pani–Grp nanocomposite at GCE. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

kinetics of the electrode processes and sensitivity of the measurements. These improvements are accompanied with a considerable decrease of capacitive current, which causes the enhancement of the detection limit in electroanalytical measurements. For this purpose, square-wave voltammograms were recorded at a fixed concentration of LCP (70 ng mL^{-1}) with different concentrations ratio of Grp–Pani to prepare modified Grp–Pani/GCE hybrid film sensor [Figure 5(B)]. As shown in Figure 5(B), at concentration ratio of 1 : 2 (Pani–Grp) a relatively high cathodic current is obtained.

Furthermore, the effect of different loadings of Pani–Grp hybrid film on LCP reduction current was determined by the volume of Pani–Grp (1 : 2) suspension casted onto GCE surface [Figure

5(C)]. The reduction peak current, (I_p) of LCP increases significantly on increasing the loading of Pani–Grp suspension from 2 to 8 µL which confirms the enhancement of the kinetics of the electrode process. However, the I_p decreased when the amount of Pani–Grp exceeded 8 µL. This is probably owing to the increased thickness of the hybrid film which may block the mass transport of LCP onto the Pani–Grp hybrid film. Thus, 8 µL is the optimized loading of Pani–Grp for the study of LCP reduction. The resulting enhancement in the cathodic peak current with the concentration of Grp–Pani suspension for the LCP reduction makes the Grp–Pani hybrid film sensor very suitable for the detection of the trace amounts of LCP.

Calibration Curve and Limit of Detection

Under the optimized conditions, applicability of the proposed voltammetric procedure for the determination of LCP was examined by measuring the cathodic peak current as a function of concentration of LCP at Grp–Pani/GCE hybrid film sensor. Figure 6(A) shows the systematic increase in the peak current
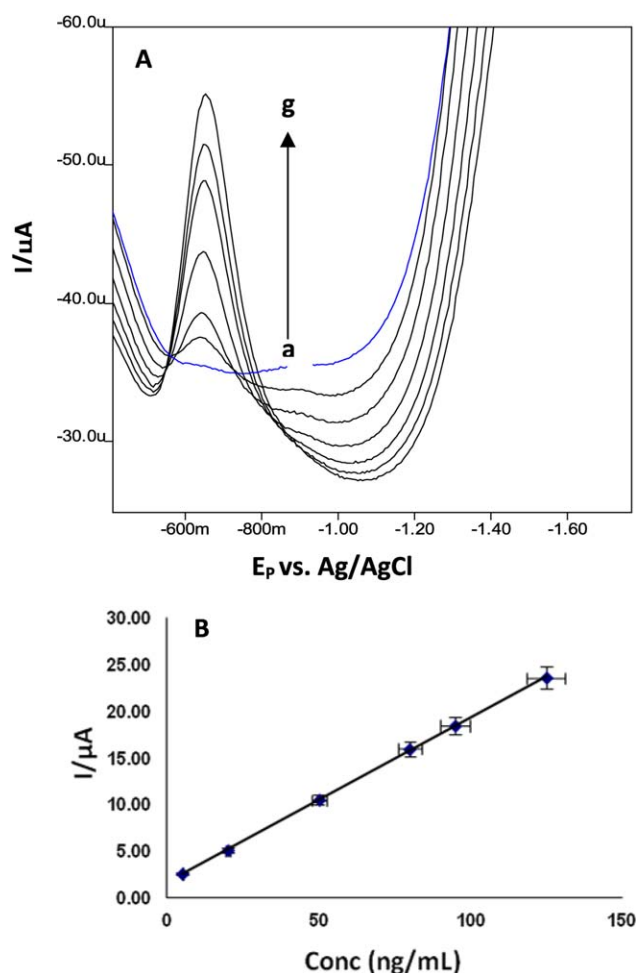

Figure 6. (A) Overlapped square-wave voltammograms of LCP at different concentrations: (a) blank solution, (b) 5 ng mL^{-1}, (c) 20 ng mL^{-1}, (d) 50 ng mL^{-1}, (e) 80 ng mL^{-1}, (f) 95 ng mL^{-1}, and (g) 125 ng mL^{-1}. (B) Linearity curve including 5% of error bar graph on three replicate analysis of LCP at different concentrations. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

Table II. Comparison of Detection Limit of the Proposed Method with the Other Reported Methods

Method	Detection limit (M)	References
HPLC with UV detector	9.3×10^{-7}	Lueje et al.[29]
HPLC with EC detector	7.5×10^{-7}	Lueje et al.[29]
Capillary electrophoresis	1.6×10^{-9}	Christians and Holzgrabe[28]
Differential pulse voltammetry with dropping mercury electrode	9.3×10^{-6}	Lueje et al.[34]
SWV with boron-doped diamond electrode	1.4×10^{-10} (in supporting electrolyte) 6.9×10^{-10} (in serum)	Altun et al.[35]
SWV with Pani–Grp/GCE hybrid film sensor	3.1×10^{-12}	This article

values with an increase in the concentration of LCP in the range from 5 to 125 ng mL^{-1} (8.17×10^{-12}–$2.04 \times 10^{-10}M$) and the peak current versus concentration plot [Figure 6(B)] presents a good linearity for LCP reduction. Linear relationship between peak current and concentration of LCP can be expressed at modified sensor by the following equation:

$$I_p(\mu A) = (0.1763)C + 1.6980; \quad r^2 = 0.9998 \quad (4)$$

where C is the concentration (ng mL^{-1}) of LCP having correlation coefficients of 0.9998. The detection and quantification limits for modified sensor were calculated by using the formula $3\sigma/b$ and $10\sigma/b$, respectively, where σ is the standard deviation and b is the slope of regression line, and they were found to be 1.94 ng mL^{-1} ($3.17 \times 10^{-12}M$) and 5.84 ng mL^{-1} ($9.62 \times 10^{-12}M$). Table II compares the detection limit of the proposed method with the other reported methods. [28,29,34,35] As summarized in Table II, it is clear that the proposed method is more sensitive as compared to the previously reported methods.

Reproducibility, Stability, and Interference Study
The reproducibility of the sensor for the study of LCP reduction was evaluated by measuring the electrocatalytic activity with same concentration of LCP with five individual sensors. The relative standard deviation was found to be 3.36%, indicating a good reproducibility of the proposed method. Stability of sensor was examined by measuring the voltammetric current response at intervals of 1–2 days, at fixed concentration of 70 ng mL^{-1} LCP over a period of 1 week. The electrode was used daily and stored at room temperature. The results showed that the current response was 82% of its initial value after 1 week, which indicated that the stability of the sensor was satisfactory for the study of LCP reduction.

To examine the interference of excipients and specificity of the proposed method in the optimized condition for the estimation of LCP, the recovery experiments were carried out in the presence of excipients such as colloidal silicon dioxide, lactose anhydrous, and stearic acid or alternatively lactose, starch, silica precipitated, talc, and magnesium stearate. For this purpose, voltammograms were recorded with six replicates of LCP standard in the presence of excipients and interference of the excipients was also studied by comparing the obtained peak current values under optimized conditions. Recovery experiments for LCP were performed in the range from 15 to 115 ng mL^{-1}

(Table III). The average recovery was found to be 98.94% which indicates that the excipients have no significant interference with the voltammetric determination of LCP. Thus, the proposed procedure can be considered specific.

Analytical Application of Grp–Pani/GCE to Pharmaceutical Formulation
To demonstrate analytical application, the Grp–Pani/GCE hybrid film sensor was used for the voltammetric determination of LCP in pharmaceutical formulation. To develop a voltammetric method for determining LCP, the SWV was selected, because the peaks are sharper and better defined at lower concentration of LCP than those obtained by cyclic voltammetry, with a lower background current, resulting in improved resolution. Voltammograms of LCP showed a very well defined cathodic peak at BR buffer (pH = 4.5). There was no need for any extraction step prior to the drug study. The mean recovery of the drug in LCP tablets was found as 98.94%, showing no significant excipient interference, and thus the developed procedure is able to quantify LCP in the presence of excipients and hence it can be considered the specific method. Thus, the present investigation revealed that the proposed method is simple, specific, sensitive, and effective for the determination of LCP at Grp–Pani/GCE hybrid film sensor.

CONCLUSIONS

In this study, a Grp–Pani hybrid film electrochemical sensor has been developed. The proposed sensor exhibited an excellent electrochemical performance toward the LCP reduction and remarkably enhancing the reduction peak current of LCP in comparison with Grp/GCE, Pani/GCE, and bare GCE. The developed sensor was characterized by SEM, electrochemical impedance spectroscopy, square wave, and cyclic voltammetry. The obtained results showed that Grp–Pani/GCE hybrid film

Table III. Recovery Studies of LCP in the Presence of Excipients

Amount added[a] (ng mL^{-1})	Amount found (ng mL^{-1})	Recovery (%)	Relative standard deviation[b] (%)
15	14.82	98.81	1.82
55	54.51	99.12	1.05
115	113.73	98.90	0.61

[a] Synthetic solutions of LCP with excipients.
[b] Number of replicates = 6.

sensor exhibited excellent electrocatalytic activity, better reproducibility, and low detection limit which can be attributed to its larger effective surface area and greater electron transfer rate. In addition, the high percentage of recovery also showed that the compound is completely extracted from tablet formulation and the results indicated that the proposed method is sensitive, accurate, precise, and cost effective as well as time saving for routine analysis in quality control laboratories. The large surface area along with increased number of electron-transfer points of Grp–Pani/GCE hybrid film sensor may offer a new approach for developing novel types of highly sensitive and stable electrochemical biosensors.

ACKNOWLEDGMENTS

REFERENCES

1. Song, M. J.; Kim, J. H.; Lee, S. K.; Lim, D. S.; Hwang, S. W.; Whang, D. *Electroanalysis* **2011**, *23*, 2408.

2. Kimmel, D. W.; Blanc, G. L.; Meschievitz, M. E.; Cliffel, D. E. *Anal. Chem.* **2012**, *84*, 685.

3. Lopez, B. P.; Merkoci, A. *Trends Food Sci. Technol.* **2011**, *22*, 625.

4. Prakash, S.; Chakrabarty, T.; Singh, A. K.; Shahi, V. K. *Biosens. Bioelectron.* **2013**, *41*, 43.

5. Yang, W.; Ratinac, K. R.; Ringer, S. P.; Thordarson, P.; Gooding, J. J.; Braet, F. *Angew. Chem.* **2010**, *49*, 2114.

6. Jain, R.; Rather, J. A. *Colloids Surf. B* **2011**, *83*, 340.

7. Jain, R.; Sharma, R. *J. Appl. Electrochem.* **2012**, *42*, 341.

8. Sun, A.; Zheng, J.; Sheng, Q. *Electrochim. Acta* **2012**, *65*, 64.

9. Shan, D.; Zhang, J.; Xue, H. G.; Zhang, Y. C.; Cosnier, S.; Ding, S. N. *Biosens. Bioelectron.* **2009**, *24*, 3671.

10. Yang, Y.; Wang, Z.; Yang, M.; Li, J.; Zheng, F.; Shen, G.; Yu, R. *Anal. Chim. Acta* **2007**, *584*, 268.

11. Li, X.; He, G.; Han, Y.; Xue, Q.; Wu, X.; Yang, S. *J. Colloid Interface Sci.* **2012**, *387*, 39.

12. Jain, R.; Rather, J. A.; Dwivedi, A.; Vikas *Electroanalysis* **2010**, *22*, 2600.

13. Wang, F.; Wang, W.; Liu, B.; Wang, Z.; Zhang, Z. *Talanta* **2009**, *79*, 376.

14. Xu, L.; Zhu, Y.; Tang, L.; Yang, X.; Li, C. *J. Appl. Polym. Sci.* **2008**, *109*, 1802.

15. Jia, Q.; Shan, S.; Jiang, L.; Wang, Y. *J. Appl. Polym. Sci.* **2010**, *115*, 26.

16. Tiwari, D. C.; Jain, R.; Sharma, S. *J. Appl. Polym. Sci.* **2008**, *110*, 2328.

17. Jain, R.; Tiwari, D. C.; Pandey, P. *Electroanalysis* **2013**, *25*, 1363.

18. Jain, R.; Dhanjai *Colloids Surf. B* **2013**, *105*, 278.

19. Kuilla, T.; Bhadra, S.; Yao, D.; Kim, N. H.; Bose, S.; Lee, J. H. *Prog. Polym. Sci.* **2010**, *35*, 1350.

20. Shrivastava, R.; Sharma, R.; Satsangee, S. P.; Jain, R. *J. Electrochem. Soc.* **2012**, *159*, B795.

21. Kang, X.; Wang, J.; Wu, H.; Liu, J.; Aksay, I. A.; Lin, Y. *Talanta* **2010**, *81*, 754.

22. Wang, Y.; Li, Y.; Tang, L.; Lu, J.; Li, J. *Electrochem. Commun.* **2009**, *11*, 889.

23. Choi, B. G.; Park, H. S.; Park, T. J.; Yang, M. H.; Kim, J. S.; Jang, S. Y.; Heo, N. S.; Lee, S. Y.; Kong, J.; Hong, W. H. *ACS Nano* **2010**, *4*, 2910.

24. Sahoo, S.; Bhattacharya, P.; Hatui, G.; Ghosh, D.; Das, C. K. *J. Appl. Polym. Sci.* **2012**, *128*, 1476.

25. Liu, S.; Xing, X.; Yu, J.; Lian, W.; Li, J.; Cui, Huang, M. *J. Biosens. Bioelectron.* **2012**, *36*, 186.

26. Fan, Y.; Liu, J. H.; Yang, C. P.; Yu, M.; Liu, P. *Sens. Actuat. B* **2011**, *157*, 669.

27. McClellan, K. J.; Jarvis, B. *Drugs* **2000**, *60*, 1123.

28. Christians, T.; Holzgrabe, U. *Electrophoresis* **2000**, *21*, 3609.

29. Lueje, A. A.; Pujol, S.; Squella, J. A.; Vergara, L. J. N. *J. Pharm. Biomed. Anal.* **2003**, *31*, 1.

30. Chen, K.; Zhang, J.; Liu, S.; Zhang, D.; Teng, Y.; Wei, C.; Wang, B.; Liu, X.; Yuan, G.; Zhang, R.; Zhao, W.; Guo, R. *J. Chromatogr. B* **2012**, *899*, 1.

31. Jain, R.; Dwivedi, A.; Mishra, R. *Langmuir* **2009**, *25*, 10364.

32. Goyal, R. N.; Gupta, V. K.; Chatterjee, S. *Biosens. Bioelectron.* **2009**, *24*, 3562.

33. Yilmaz, S; Uslu, B.; Ozkan, S. A. *Talanta* **2001**, *54*, 351.

34. Lueje, A. A.; Vergara, L. J. N.; Pujol, S.; Squella, J. A. *Electroanalysis* **2002**, *14*, 1098.

35. Altun, Y.; Uslu, B.; Ozkan, S. A. *Anal. Lett.* **2010**, *43*, 1958.

36. Xu, L.; Zhu, Y.; Tang, L.; Yang, X.; Li, C. *J. Appl. Polym. Sci.* **2008**, *109*, 1802.

37. Hopkins, A. R.; Sawall, D. D.; Villahermosa, R. M.; Lipeles, R. A. *Thin Solid Films* **2004**, *469–470*, 304.

38. He, Y. *Mater. Sci. Eng. B* **2005**, *122*, 76.

39. Brunauer, S.; Emmett, P. H.; Teller, E. *J. Am. Chem. Soc.* **1938**, *60*, 309.

40. Song, M. J.; Hwang, S. W.; Whang, D. *J. Appl. Electrochem.* **2010**, *40*, 2099.

41. Sabatani, E.; Rubinstein, I. *J. Phys. Chem.* **1987**, *91*, 6663.

42. Agostinho, S. M. L.; Villamil, R. F. V.; Neto, A. A.; Aranha, H. *Quim. Nova* **2004**, *27*, 813.