Portugaliae Electrochimica Acta 26/4 (2008) 315-324

PORTUGALIAE ELECTROCHIMICA ACTA

Cathodic Stripping Voltammetric Determination of Losartan in Bulk and Pharmaceutical Products

I.H.I. Habib[1,*], S.A. Weshahy[2], S. Toubar[3] and M.M.A. El-Alamin[3]

[1]Analytical Chem. Lab., Applied Organic Chem. Dept., National Research Centre, Dokki, Cairo, Egypt
[2]Analytical Chemistry Department, Faculty of Pharmacy, Cairo University, Egypt
[3]Analytical Chemistry Department, Faculty of Pharmacy, Helwan University, Egypt

Received 14th November 2007; accepted 29th January 2008

Abstract

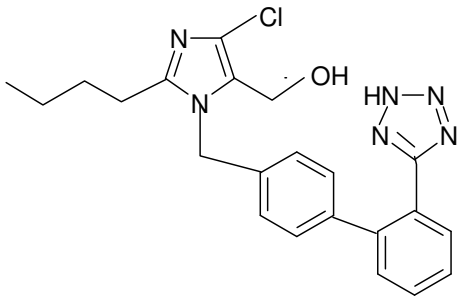
Cathodic stripping voltammetric determination of losartan using hanging mercury drop electrode HMDE was described. The method was based on adsorptive accumulation of the species at HMDE and at pH 7, followed by alternating current AC sweep. The behavior of adsorptive stripping response was studied under various experimental conditions, e.g. type of supporting electrolyte, pH, accumulation time, scan rate and mode of sweep (direct current DC, differential pulse DP, square wave SW and AC). In Britton-Robinson buffer solution, pH 7, a quasi-reversible reaction took place. The reduction response was more sensitive than the oxidation one and it was linear over the concentration range of 0.16-1.2 μg/mL. The determination of the cited compound in oral dosages was achieved using the standard addition method. The average of determinations obtained by square wave adsorptive voltammetric method with its standard deviation was 100.1±3%.

Keywords: losartan, adsorptive stripping voltammetry, HMDE, stability.

Introduction

Losartan (*2-n-butyl-4-chloro-1-[p-(o-1H-tetrazol-5-ylphenyl)benzyl]-imidazole-5-methanol mono potassium salt,* shown below) is the first member of a new chemical class of a highly selective, orally active, non-peptide angiotensin II receptor antagonist with an improved safety and tolerability profile. It is prescribed alone or in combination with hydrochlorothiazide for the treatment of moderate to sever hypertension [1].

* Corresponding author. E-mail address: ihihabib@yahoo.com



Scheme 1

Limited methods for analysis of the cited species have been reported. These include high performance liquid chromatography HPLC [1-10], high performance thin layer chromatography HPTLC [11-12], capillary electrophoresis [13-16], supercritical fluid chromatography [16], fluorimetry [17], UV-spectrophotometry [4,18-21] and colourimetry [22].
In the present work, the electrochemical behavior of losartan on a hanging mercury drop electrode HMDE was thoroughly investigated and the optimum conditions for the quantification analysis and for applying as stability-indicating method were studied.

Experimental

Apparatus

All different modes of adsorptive stripping voltammograms (direct current DC, differential pulse DP, square wave SW, and alternating current AC) were recorded using Metrohm 693 VA processor (Switzerland) and VA 694 stand equipped with three electrodes Ag/AgCl-3 M KCl, a platinum electrode and hanging mercury drop electrode (HMDE). The pH measurements were carried out with digital pH-meter Metrohm.

Reagents

All chemicals used are analytical grade. Twice distilled water was used throughout all experiments. Four pharmaceutical products, either in single form, namely Cozaar (MSD Pharm. Ind., Netherlands), Losar (Unipharm, Egytp), containing 100 and 50 mg losartan per tablet, respectively, or in mixture, namely Hyzaar (MSD Pharm. Ind, Netherlands) and Losarmepha-Plus (Sigma, Egypt), containing 50 mg losartan and 12.5 mg hydrochlorothiazide per tablet were used.
A stock solution of losartan (0.4 mg/mL) was prepared by dissolving in bidistilled water. Modified Britton-Robinson BR buffer solutions over the range pH 3-11 were also prepared.

Procedure

An aliquot of losartan solution (1.6-12 μg/mL) was placed in a 10 mL-measuring flask containing 5 mL of modified Britton-Robinson buffer pH 7 (0.04 M) and 0.1 mL 0.1 M Na EDTA. The flask was then completed with distilled water to the mark. The solution is then transferred to the electrode cell and de-aerated by

pure nitrogen gas at 1 atmosphere for 2 min. Adsorption was carried at –700 mV for 30 s with continuous stirring at speed of 2000 rpm. The stirrer was stopped and the solution was allowed to rest for 10 s, then voltammogram was recorded using a HMDE with a drop size of ~0.15 mm^2, a pulse amplitude "ΔE" of 30 mV, frequency 25 Hz for AC, a scan rate "ν"of 20 mV/s over a reduction potential range from –700 to –1400 mV. The mean of triplicate measurements of content in the sample was calculated using standard addition method at room temperature (*ca*. 27 °C). To assay losartan in pharmaceutical products, a tablet was placed in a beaker containing distilled water and then ultrasonated for 15 min. The beaker was transferred to a 100 mL measuring flask and completed to the mark with water without any filtration. Aliquot of the solution containing the nominated range concentration of losartan was added into a 10 mL-measuring flask and the procedure completed as described above. UV first derivative spectrophotometry was performed for losartan as a reference method [18].

Results and discussion

The electrochemical behavior of losartan on hanging mercury drop electrode HMDE was carefully studied in the modified Britton-Robinson BR buffer solution. Losartan exhibited one distinct and well-defined cathodic peak at a potential around -1.2 V as depicted by cyclic voltammogram given in Fig. 1. A broad and weak peak could also be observed in the anodic scan. The ratio of cathodic and anodic peak heights is greater than one and the distance between both peaks is in the range 60-200 mV, suggesting the quasi-reversible nature of the electrode process.

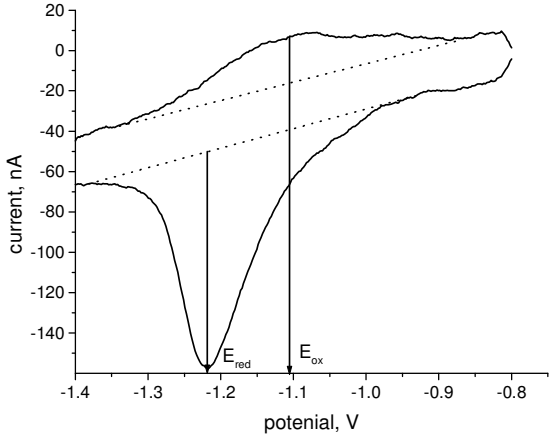


Figure 1. Cyclic voltammogram showing quasi-reversible reaction of losartan with a reduction peak at -1.2 V and an oxidation peak at -1.1 V.

At pH values lower than 5, no reduction peak was noticeable. The cathodic peak current was increased gradually from pH 5 to 7 and then remained constant up to 11 as shown in Fig. 2. Parallel to this behavior is noticed a peak potential which was shifted toward less negative values up to pH 8. This voltammetric behavior may indicate that the electrode process could involve a strong adsorption

phenomenon on HMDE [23]. The pH effect study revealed that protons participate directly in the reduction process and that the proton-transfer reaction precedes the electrode process proper [24]. A good linear relation was got in the pH range 5-7 with a slope 0.160 and correlation coefficient r=0.983. It can be concluded from the expression $\Delta E_p/\Delta$pH = 0.059 $p/\alpha n$ that the ratio between protons p and fractional electrons transferred αn is 2.71. For quasi-reversible electrode reaction, the value of αn is found to be 0.72 obtained from the relation $(E_p-E_{p/2})=0.048/\alpha n$ at different pH, where $E_{p/2}$ is a potential at half peak current [24].

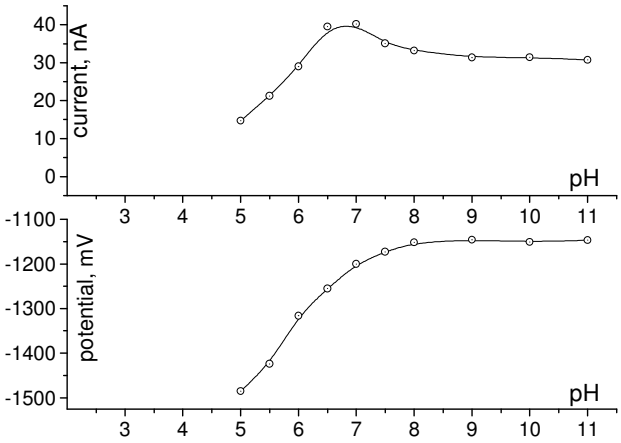
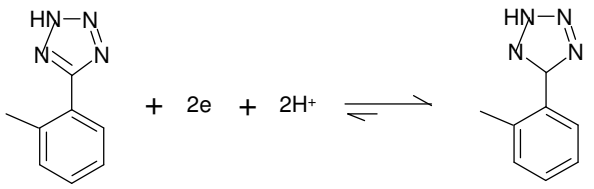


Figure 2. Relation between different pH and current and relation between different pH and potential, at 4 µg/mL of losartan , ν = 60 mV/s, ta = 30 s and ΔE = 50 mV.

It can be concluded, from these results, that the number of electrons transferred n is 2 (electron transfer coefficient α is selected arbitrary between 0.45-0.30 as a result of electron transfer slowness relative to mass transfer), while that of protons participated p is 2.

From the structure of losartan, as shown in Scheme 1, the molecule possessed imidazole group and tetrazolyl group. The imidazole group cannot polarographically reduce in aqueous solution [25], but it can be reduced drastically at ca. -1.77 V in organic media [26]. On the other hand, the halide group in most organic compounds can be reduced only in alkaline media [27]. Accordingly, the mechanism of electrode reaction can be assigned to proton transfer reactions coupled to electron transfer to reduce azomethine group C=N in tetrazolyl group into saturated bond by consuming 2 electrons and 2 protons, as shown below:



Variation of ionic strength at 0.02, 0.04, 0.06, 0.08 and 0.1 M supporting electrolyte BR at pH 7, has no distinct effect on the peak current and potential indicating the reduction is predominantly diffusion controlled. Addition of 0.1 mL 0.1 M Na EDTA was necessary to eliminate the interfering effect of zinc on the measurement of losartan.

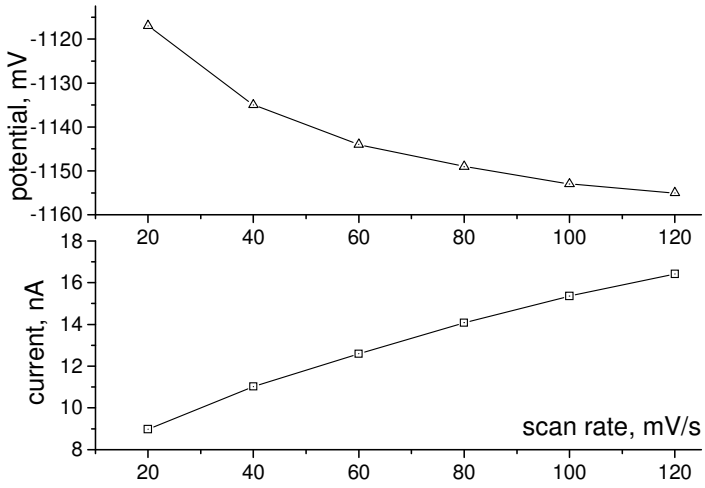


Figure 3. Effect of scan rate on 0.8 µg/mL losartan, at ta = 30 s, ΔE = 50 mV and pH 7 (0.04 M).

The variation of scan rate from 20 to 120 mV/s, revealed that the peak current is increased while the potential is shifted to more negative when the scan rate is increased, as shown in Fig. 3, confirming the quasi-reversible nature of the reduction process where the rate of electron transfer is slower than the scan rate.

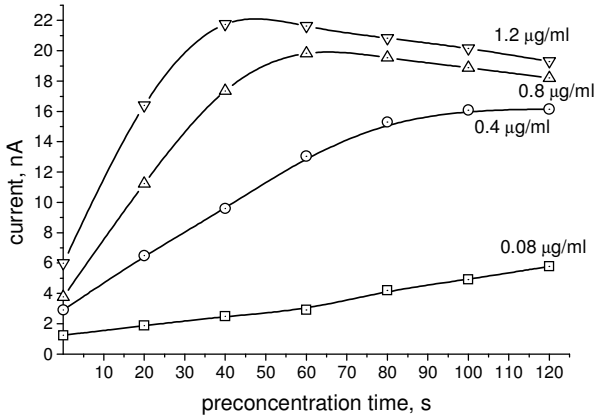


Figure 4. Effect of accumulation time ta at different concentrations of losartan at: ν = 60 mV/s, ΔE = 50 mVand pH 7 (0.04 M).

Plotting log I versus log ν gave up a straight line with a slope = 0.35 (r = 0.999) which deviates from the value expected, 0.5, for an ideal reaction of diffused

species [28] confirming the existence of adsorption component. The interfacial adsorptive character of the drug on the HMDE was also identified from the peak current dependence, upon preconcentration of the drug in BR buffer at pH 7. The peak current is increased linearly, as shown in Fig. 4, with increasing the accumulation time up to t_a =40 s for all concentrations studied (0.08-1.20 µg/mL). Above that time, the deviation from the linearity is observed because of the formation of multilayer on the surface of the electrode.

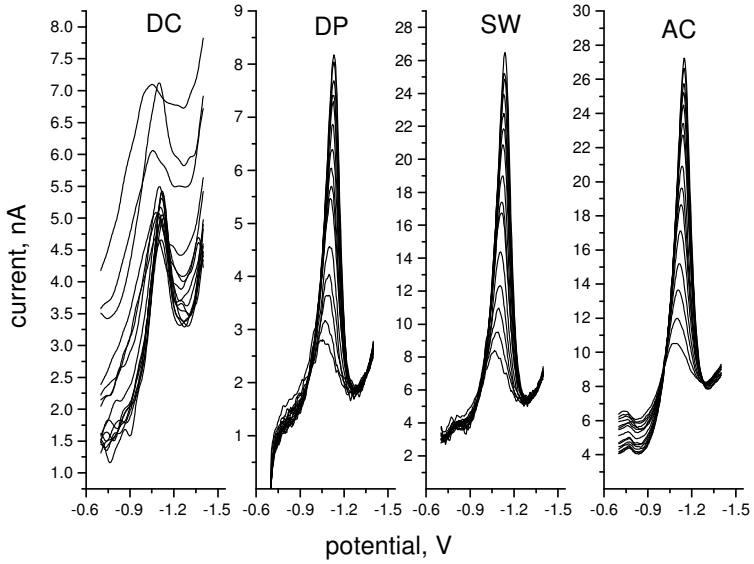


Figure 5. Effect of different modes of scan over the concentration range of 0.08-1.2 µg/mL with ta = 30 s at ν = 20 mV/s, ΔE = 30 mVand pH 7 (0.04 M).

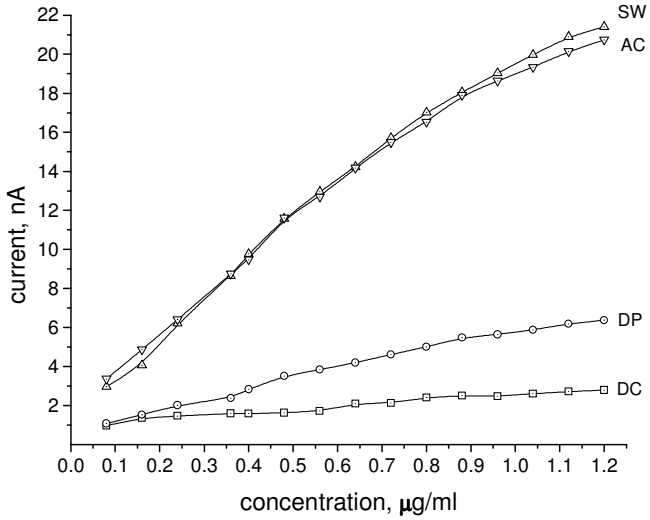


Figure 6. Effect of calibration curves using different modes of scan over the concentration range of 0.08-1.2 µg/mL with ta = 30 s at ν = 20 mV/s, ΔE = 30 mVand pH 7 (0.04 M).

Based on the optimized parameter mentioned above, the calibration curves are constructed using different modes of sweep, viz. direct current DC, differential pulse DP, square wave SW and first harmonic alternating current AC, for the comparison purpose, over the concentration range 0.16-1.20 μg/mL losartan. As shown in Fig. 5 and 6, the AC and SW modes gave highest proportionality compared with other modes and lowest detection limit at 0.12 μg/mL as explicit in table 1. Worth mentioning that hydrochlorothiazide HCZ could be determined polarographically only at pH 9 [29]. In the present medium, pH 7, no peak due to HCZ has been detected.

Table 1. Regression parameter for determination of losartan using different modes of sweeps.

LOSARTAN	DC	DP	SW	AC
Slope	1.5±0.1	4.8±0.2	16.7±0.7	16.1±0.6
Intercept	1.1±0.1	1.0±0.1	2.8±0.5	3.1±0.4
Regression coef.	0.990	0.993	0.989	0.993
L.O.D.(μg/mL)	0.15	0.13	0.15	0.12
Conc. range (μg/mL)	0.16-1.2	0.16-1.2	0.16-1.2	0.16-1.2

Repeatability and reproducibility

The intra- and inter-day precision was evaluated by assaying freshly prepared solutions in triplicate on the same day and on three successive days, respectively using the proposed method. The repeatability of the results obtained by means of the proposed AC voltammetric procedure was examined by performing five replicate measurements for 0.88 μg/mL losartan following pre-concentration for 30 s. Mean recoveries of 99.80±0.30 and 99.21±1.01 % (n = 5) on the same day and on three successive days were achieved, respectively, indicating high precision of the proposed procedure and that it is suitable for quality control of losartan.

The optimized parameters were also applied for determining the cited compound in Egyptian pharmaceutical products containing 100 and 50 mg losartan per tablet alone, viz. Cozaar and Losar, respectively, or 50 mg losartan in combination with 12.5 mg hydrochlorothiazide HCZ per tablet, viz. Hyzaar and Losarmepha-plus, using standard addition technique as given in Table 2. Hydrochlorothiazide and additives did not observe any interference on measuring losartan. The average value measured by AC adsorptive stripping voltammetry with its standard deviation was 100.1±3% for losartan in nominated drugs showing that they were in good agreement with those obtained by the uv first derivative spectrophotometric method [18]. The results show that the calculated t- and F- values did not exceed the theoretical values at 95% confidence level [30].

Stability

Stability testing of losartan was performed under various stress conditions in order to assure the selectivity and provide an indication of the stability-indicating

properties of the proposed voltammetric method. Thus, the acid effect on the stability of losartan is depicted in Fig. 7 where 20% degradation of losartan proceed by dimerization slowly within the first day in 5 M HCl at 85 ºC and a plot of log concentration versus degradation time exhibits a first order kinetic relationship with a slope -4.2×10^{-2} (R=0.999). The sample preparations did not exhibit any degradation peaks that could interfere with the reduction peak of losartan. Furthermore, no degradation was seen in alkaline degradation.

Table 2. Determination of losartan in pharmaceutical tablets using the alternating current method.

Trade name, mg losartan/tablet	Found*, mean ± sd%	
	Proposed	Reference
Cozaar, 100 mg	99.8±3	95.9±1
Losar, 50 mg	102.5±2	96.1±3
Hyzaar, 50 mg	101.5±2	100.3±2
Losarmepha-Plus, 50 mg	100.0±3	96.4±4
Mean ± sd	100.1±3	97.7±2
t-test (t_{th} = 2.20 at α = 0.05)	2.01	
F-test (F_{th} = 2.98 at α = 0.05)	1.18	

*Average of three determinations100.25 ± 2.04

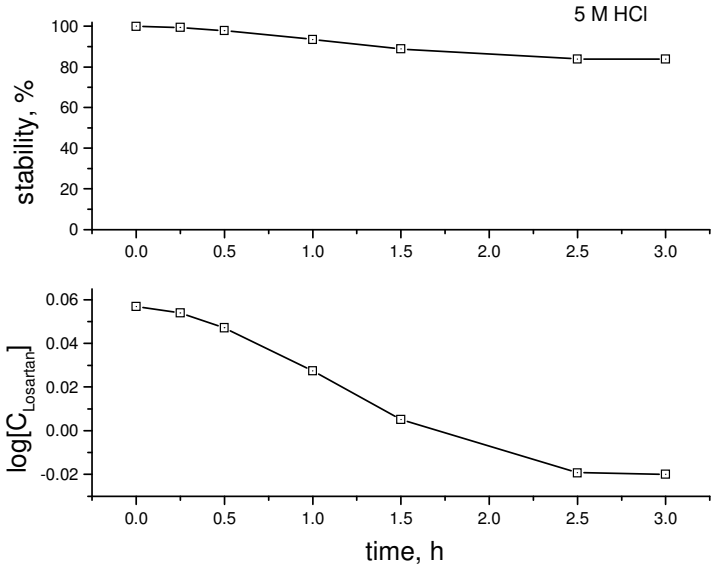


Figure 7. Effect of time on the stability of losartan solution in 5 M HCl at 85 ºC (above) and good first order kinetic correlation with the concentration (below).

The TLC chromatogram of the acid-degraded spots for losartan showed the degraded spots at R_f 0.35 and 0.57, respectively, using acetonitrile-methanol-0.1% acetic acid solution (35:25:40) as a mobile phase which is in accordance with the results given in the literature [31-34].

Conclusion

The present results show that by careful choice of the operating parameters, the adsorptive stripping voltammetry enhanced significantly the sensitivity and selectivity for the analysis of losartan as bulk drug and in pharmaceutical formulations. The result obtained is in good agreement with that obtained by uv first derivative spectrophotometric technique. The proposed method may be extended to study the degradation kinetics of losartan under various conditions.

Acknowledgements

Thanks are due to Volkswagen Stiftung, Kastanienallee 35, 30519 Hannover, Germany, for the financial support of purchasing the Metrohm model 693 VA processor and 694 VA stand.

References

1. J. Nie, M. Zhang, Y. Fan, B. Xiang, Y.Q. Feng, *J. Chromatogr.-B* 828 (2005) 62-69.
2. S. Ulu , S. Saglik, *Turk. J. Pharm. Sci*. 1(2004) 165-175.
3. E. Dinc, O. Ustundag, A. Ozdemir, D. Baleanu, *J. Liq. Chromatogr. Relat. Technol.* 28 (2005) 2179-2194.
4. N. Erk, *J. Pharm. Biomed. Anal*. 24 (2001) 603-611.
5. C.A. Mueller, W. Weinmann, S. Dresen, A. Schreiber, M. Gergov, *Rapid. Commun. Mass-Spectro.* 19 (2005) 1332-1338.
6. L. Kristoffersen, E.L. Qiestad, M.S. Opdal, M. Krogh, E. Lundanes, A.S. Christopherrrsen, *J. Chromatogr.-B* 850(2007) 147-160.
7. M. Polinko, K. Riffel, H.C. Song, M.W. Lo, *J. Pharm. Biomed. Anal*. 33 (2003) 73-84.
8. L. Gonzalez, RM. Alonso, RM. Jimenez, *Chromatographia* 52 (2000) 735-740.
9. D.L. Hertzog, J.F. McCafferty, X.G. Fang, R.J. Tyrrell, R.A. Reed, *J. Pharm. Biomed. Anal*. 30 (2002) 747-760.
10. L. Gonzalez, J.A. Lopez, R.M. Alonso, R.M. Jimenez, *J. Chromatogr. A* 949 (2002) 49-60.
11. R.T. Sane, M. Francis, S. Pawar, *Indian Drugs* 39 (2002) 32-35.
12. S.A. Shah, I.S. Rathod, B.N. Suhagia, S.S. Savale, J.B. Patel, *J. AOAC-int* 84 (2001) 1715-1723.
13. S. Hillaert, W. Van-den-Bossche, *J. Pharm. Biomed. Anal*. 31 (2003) 329-339.
14. S. Hillaert, W. Van-den-Bossche, *J. Chromatogr. A* 979 (2002) 323-333.
15. M. Zhang, F. Wei, Y.F. Zhang, J. Nie, Y.Q. Feng, *J. Chromatogr. A* 1102 (2006) 294-301.
16. R.C. Williams, V.L. Alasandro, V.L. Fasone, R.J. Boucher, J.F. Edwards, *J. Pharm. Biomed. Anal.* 24 (1996) 1539-1546.

17. E. Cagigal, L. Gonzalez, R.M. Alonso, R.M. Jimenez, *Talanta* 54 (2001) 1121-1133.
18. O.C. Lastra, I.G. Lemus, H.J. Sanchez, R.F. Perez, *J. Pharm. Biomed. Anal.* 33 (2003) 175-180.
19. C. Vetuschi, A. Giannandrea, *Anal. Lett.* 36 (2003) 1051-1064.
20. N.R. Lande, B.M. Shektar, S.S. Kadam, S.R. Dhaneshwar, *Indian-Drugs* 37 (2000) 577-581.
21. H.K. Jain, A.K. Singhai, R.K. Agrawal, *Indian-Drugs* 37 (2000) 239-242.
22. A.H. Prabhakar, R. Giridhar, J. Pharm. Biomed. Anal. 27 (2002) 861-866.
23. E. Laviron, J. Electroanal. Chem. 52 (1974) 355-393.
24. P. Zuman, The Elucidation of Organic Electrode Processes, Academic Press, New York, (1969) 21.
25. A.A. Samarkandy, *J. King Abdulaziz University: Science* 19 (2007) 23-40.
26. N.O. Pekmez, M. Can, A. Yildiz, *Acta Chim. Slov.* 54 (2007) 131–139.
27. A.A. Gazy, H. Mahgoub, E.F. Khamis, R.M. Youssef, M.A. El-Sayed, *J. Pharm. Biomed. Anal.* 41 (2006) 1157-1163.
28. E. Laviron, *J. Electroanal. Chem.* 112 (1980) 1-23.
29. M.E. Martin, O.M. Hernandez, A.I. Jimez, J.J. Arias, F. Jimenez, *Anal. Chim. Acta* 381 (1999) 247-256.
30. J.C. Miller, J.N. Miller, Statistics for Analytical Chemistry, 3rd Ed , Ellis Horwood , Chichester 53 (1993).
31. K.E. McCarthy, Q. Wang, E.W. Tsai, R.E. Gilbert, D.P. Ip, M.A. Brooks, *J. Pharm. Biomed. Anal.* 17 (1998) 671-677.
32. M. Lusina, T. Cindric, J. Tomaic, M. Peko, L. Pozaic, N. Musulin, *Int. J. Pharmaceutics* 291 (2005) 127–137.
33. Z. Zhao, Q. Wang, E.W. Tsai, X. Qin, D. Ip, *J. Pharm. Biomed. Anal.* 20 (1999) 129-136.
34. R.A. Seburg, J.M. Ballard, T.L. Hwang, C.M. Sullivan, *J. Pharm. Biomed. Anal.* 42 (2006) 411-422.