Talanta 82 (2010) 106–112

Contents lists available at ScienceDirect

Talanta

journal homepage: www.elsevier.com/locate/talanta

Electrochemical determination of some antidiabetic drugs for type 2 diabetic patients

Waheed A. Badawy[a,*], Mohammed A. El-Ries[b], Inas M. Mahdi[b]

[a] Chemistry Department, Faculty of Science, University of Cairo, Gamma Street, 12613 Giza, Egypt
[b] Pharmaceutical Chemistry Division, National Organization for Drug Control and Research, Dokki, Cairo, Egypt

ARTICLE INFO

Article history:
Received 2 January 2010
Received in revised form 4 April 2010
Accepted 6 April 2010
Available online 14 April 2010

Keywords:
Cyclic voltammetry
Differential pulse voltammetry
Glimepiride
Glyburide
Pioglitazone
Rosiglitazone

ABSTRACT

Quantitative determination of rosiglitazone, pioglitazone, glimepiride and glyburide as antidiabetic drugs for type 2 diabetic patients was performed conveniently and economically using cyclic voltammetry (CV) and differential pulse voltammetry (DPV). Carbon paste (CPE) and glassy carbon (GCE) electrodes were successfully used as sensors for these drugs in Briton–Robinson (B–R) as buffer solution. The preparation of CPE and the GCE as ion selective electrodes is based on the construction of 10% standard drug ion pair with reineckate or tungstophosphate imbedded as electroactive material. Working standards were freshly prepared just before the assay by dilution from a 10^{-2} mol L^{-1} drug stock solution. At a scan rate of 100 mV s^{-1} the cyclic voltammograms showed a well defined anodic peak with high selectivity. The DVP gave a reproducible well defined diffusion controlled peak for each drug at a scan rate of 10 mV s^{-1}. The oxidation peaks were used to determine the tested drug concentrations. The quantitative determination of the four drugs in their pharmaceutical preparations by the proposed electrochemical technique was found to be identical with the values obtained by the standard HPLC method. A mean % recovery of 100 ± 1 was obtained and the % relative standard deviation was 1.62 indicating the high precision of the method and the confidence in its repeatability. The proposed electroanalytical technique using either the CPE or the GCE is economic, selective and can be applied for both the qualitative and quantitative determination of the drugs in their pharmaceutical preparations, without special drug separation.

1. Introduction

Rosiglitazone [5-((4-(2-(methyl-2-pyridinylamino) ethoxy) phenyl) methyl)-2,4-thiazolidinedione], pioglitazone [5-((4-(2-(5-ethyl-2-pyridinyl) ethoxy) phenyl) methyl)-,(+-)-2,4-thiazolidinedione], glimepiride [3-ethyl-N,N-bis (3-ethyl-4-methyl-2-oxo-5H-pyrrol-2-yl)-4-methyl-2-oxo-5H-pyrrole-1-carbox-amide] and glyburide [5-chloro-N-[2-[4-(cyclohexyl-carbamoyl-sulfamoyl) phenyl] ethyl]-2-methoxy-benzamide] are well known compounds that clinically used in the treatment of type 2 diabetes mellitus. The structural formula of each drug is presented in Fig. 1.

Rosiglitazone is a thiazolidinedione antihyperglycemic agent which works by increasing insulin sensitivity in target tissues, as well as decreasing hepatic gluconeogenesis [1]. It is extensively metabolized by cytochrome P450 $2C_8$ and so may have some utility as an in vivo probe for this enzyme. Pioglitazone is also an oral thiazolidinedione antihyperglycemic agent and is given as pioglitazone hydrochloride. Its doses are expressed in terms of the base, which act primarily by reducing insulin resistance. The effects of pioglitazone on serum lipid concentrations appear to differ from those of rosiglitazone [2]. Glimepiride and glyburide are the potent second generation oral sulfonylurea antihyperglycemic agents that widely used for the treatment of type 2 diabetes mellitus [3,4]. Glyburide works by inhibiting ATP-sensitive potassium channels in pancreatic beta cells. This inhibition causes cell membrane depolarization, opening of voltage-dependent calcium channels, thus triggering an increase in intracellular calcium into the beta cell that stimulates insulin release.

Several analytical methods have been reported for the determination of rosiglitazone, pioglitazone, glimepiride and glyburide based on high performance liquid chromatography (HPLC) [5–10] and high performance thin layer liquid chromatography (HPTLC) [11,12]. Electrochemical methods were proved to be useful for sensitive and selective determination in pharmaceutical compounds. These methods do not require tedious pre-treatment and involve limited pre-separation, and consequently reduce the cost of analysis [13,14]. The use of bulk modified carbon-paste electrodes in the electromeric determination of many hazardous compounds and pharmaceutical preparations is an interesting subject for intensive investigations [15–17]. The aim of the present paper is to investigate the voltammetric oxidation behavior of rosiglitazone,

* Corresponding author. Tel.: +20 2 3567 6558; fax: +20 2 3568 5799.
E-mail addresses: wbadawy@cu.edu.eg, wbadawy50@hotmail.com (W.A. Badawy).

0039-9140/$ – see front matter © 2010 Elsevier B.V. All rights reserved.
doi:10.1016/j.talanta.2010.04.006

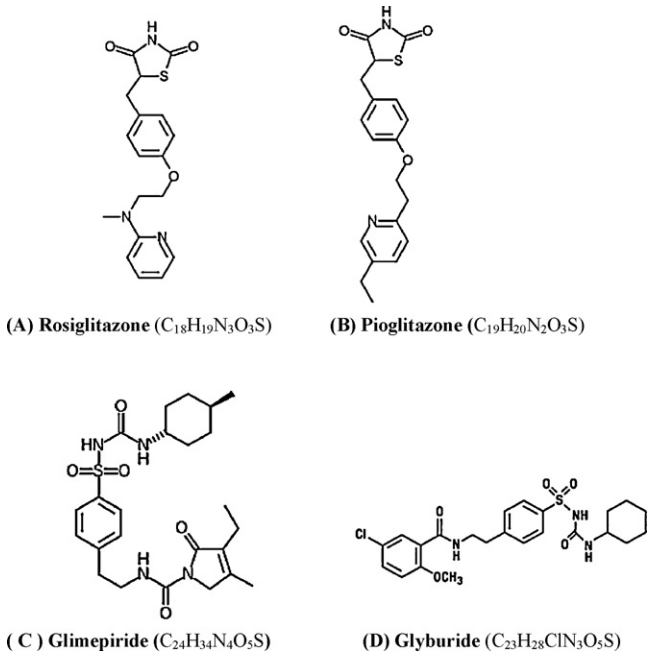

Fig. 1. Structural formula of each drug.

pioglitazone, glimepiride and glyburide at carbon paste-, (CPE) and glassy carbon (GCE) electrodes using cyclic and differential pulse voltammetry. It is aimed at economic, accurate and fast quantitative determination of the tested drugs as standards or in their pharmaceutical formulations.

2. Experimental

2.1. Reagent

Rosiglitazone, pioglitazone, glimepiride and glyburide were supplied from Galaxo Smith Kline, Eli Lilly, Sanofi-Aventis and Aventis, respectively. The pharmaceutical dosage forms of these drugs in Egypt are from Avandia®, Glustin, Amaryl® and Daonil®, respectively. Standard stock solutions (10^{-2} mol L^{-1}) were prepared by dissolving appropriate weight of each drug standard powder (rosiglitazone, pioglitazone, glimepiride and glyburide 0.179 g, 0.178 g, 0.245 g and 0.247 g, respectively), in 50.0 mL methanol under continuous stirring until complete dissolution of the drug. The standard solution was then kept in a refrigerator.

Working standards were freshly prepared just before assay by dilution of the standard stock solution using an appropriate amount of Britton–Robinson (B–R) buffers in the range from pH 2.0 to 10.0, which served as supporting electrolyte. Unless otherwise stated, all solutions were prepared using doubly distilled water and analytical grade reagents. Also, all potentials were measured against and referred to the Ag/AgCl reference electrode ($E^0_{Ag/gCl(NHE)} = 0.222$ V).

2.2. Apparatus

Cyclic voltammetry (CV) and differential pulse voltammetry (DPV) were performed using an electrochemical workstation. A three compartment electrochemical cell, incorporating the working electrode (GCE or CPE) was used. The carbon paste of uniform graphite particles was mixed with a paraffin binder (for use in aqueous media). The reference electrode was the Ag/AgCl (3 mol L^{-1} KCl) and a Pt-wire was used as an auxiliary electrode. The operating conditions for the DPV were 50 mV pulse amplitude, 30 ms pulse width and a scan rate of 10 mV s^{-1}. The working procedures and preparation conditions of both the GCE and CPE were always the same.

2.3. Procedure

To ensure perfect charge transfer and reproducible data, the working electrodes were subjected to a constant cleaning procedure before each experiment. The GCE or CPE electrode was polished with 0.5 mm diameter alumina powder wetted with bi-distilled water on a smooth polishing cloth. The electrode was rinsed with ethanol then with water, dried and fitted to the electrochemical cell. A cyclic voltammetric cleaning step in 0.1 mol L^{-1} $HClO_4$ was applied. The potential scan was carried out between 0.2 and 1.5 V at a scan rate of 20 mV s^{-1} for a large number of scans. Generally, 10 min of potential cycling were enough to produce a reproducibly clean surface. The cell was then filled with 5 mL B–R buffer as the blank solution and the potential scan started from 0.0 to +1.5 V. After recording the voltammetric data of the blank solution, an appropriate amount of the material to be tested (rosiglitazone, pioglitazone, glimepiride or glyburide) was added and the voltammetric response at the working electrode was recorded. The measurements were carried out at constant room temperature of 25 ± 1 °C and the peak heights were evaluated by the tangent procedure [18]. The pH of the solution was found to have a significant effect on the voltammetric drug response.

2.4. Preparation of drug specimens and calibration curves

The preparation of standards and test samples is summarized in Table 1. Generally, duplicate samples of 5 drug tablets were weighed and finely powdered in a mortar. An average weight equivalent to the individual drug units was accurately weighed, then transferred to an amber glass volumetric flask. The mass was then dissolved in 0.1 mol L^{-1} $HClO_4$ using an ultrasonic bath for 15 min. An additional amount of water was added to adjust a constant volume. The whole solution was shaken until full mixing and 10.0 mL portion was centrifuged at 30 rounds per min (rpm) at room temperature until a clear supernatant solution was obtained, that was used for the voltammetric measurements. A calibration curve was made by the method of standard additions in which known concentrations of each drug in a small volume (0.5 mL, 10^{-2} mol L^{-1}) of concentrated standard were added to a large volume (50.0 mL) of the sample solution to minimize the effect of dilution. The results of the voltammetric measurements at both the GCE and CPE working electrodes were compared with reference HPLC data for rosiglitazone [19], pioglitazone, glimepiride [7] and glyburide [20]. Details of the experimental procedures and preparations are described elsewhere [21].

3. Results and discussion

3.1. Cyclic voltammetric determination of different drugs

The prepared samples were subjected to a series of investigations to optimize the determination conditions of the pharmaceutical preparations. Different supporting electrolytes, namely HCl, H_2SO_4, acetic acid/sodium acetate, ammonium chloride/aqueous ammonia, KCl, phosphate buffer and Briton–Robinson (R–B), buffer were investigated. From all those supporting electrolytes the B–R buffer solution was found to give the best and most reproducible results and was used in all investigations.

3.1.1. Effect of pH

The voltammetric behavior of the tested drugs was found to be affected by the solution pH and the type of the supporting electrolyte. The effect of the above mentioned supporting electrolytes

Table 1
Samples and standard solutions preparation for determination of rosiglitazone, pioglitazone, glimepiride and glyburide.

Sample concentration per tablet (mg) Avandia	Sample type (nominal tablet strength) (mg) Rosiglitazone	Volume of standard stock (mL) Stock standard solution (80 mg L^{-1})	Volume of working standard (mL)	No. of tablets and average of one tablet	Flask volume for tablet (mL)	Concentration (mg/mL)
4.0	4			5 1	250 50	0.08
2.0	2	25	50	5 1	250 50	0.04
1.0	1	25	100	5 1	250 50	0.02
Sample concentration per tablet (mg) Glustin	**Sample type (nominal tablet strength) (mg) Pioglitazone**	**Volume of standard stock (mL) Stock standard solution (standard mg/diluting solvent mL)**	**Volume of working standard (mL)**	**No. of tablets and average of one tablet**	**Flask volume for tablet (mL)**	**Concentration (mg/mL)**
30.0	30	30	100	5 1	500 100	0.3
15.0	15	15	100	5 1	500 100	0.15
Sample concentration per tablet (mg) Amaryl	**Sample type (nominal tablet strength) (mg) Glimepiride**	**Volume of standard stock (mL) Stock standard solution (200 mg L^{-1})**	**Volume of working standard (mL)**	**No. of tablets and average of one tablet**	**Flask volume for tablet (mL)**	**Concentration (mg/mL)**
3.0	3	30	100	5 1	250 50	0.06
2.0	2	20	100	5 1	250 50	0.04
1.00	1	10	100	5 1	250 50	0.02
Sample concentration per tablet (mg) Daonil	**Sample type (nominal tablet strength) (mg) Glyburide**	**Volume of standard stock (mL) Stock standard solution (200 mg L^{-1})**	**Volume of working standard (mL)**	**No. of tablets and average of one tablet**	**Flask volume for tablet (mL)**	**Concentration (mg/mL)**
5.00	5	50	100	5 1	250 50	0.1

and the pH of the solution on the peak current of the tested drugs were recorded. The different investigations were then carried out in the best supporting electrolyte, i.e. the B–R buffer over the pH range 2.0–10.0 with different scan rates. At a scan rate of 100 mV s^{-1} the cyclic voltammograms showed a well defined anodic peak with high selectivity. Typical cyclic voltammograms of 1×10^{-6} mol L^{-1} drug in the B–R buffer of pH 5 recorded with a 100 mV s^{-1} scan rate at the GCE are presented in Fig. 2. Example of the effect of scan rate on the peak current is presented as insert in this figure. Similar results were obtained using the CPE.

Rosiglitazone was found to give a well defined anodic peak at pH 5, whereas the other drugs (pioglitazone, glimepiride and glyburide) show their clear peaks at pH 6. In all cases, no cathodic peak in the reverse scan was recorded, which means that the oxidation of the tested drug is irreversible. The optimum concentration of the B–R buffer at the respective pH was found to be 0.04 mol L^{-1}. It was observed that the peak potential shifts to the positive direction when the solution pH was decreased. On both the CPE and GCE electrodes, the peak potential shows a semi-linear relation between pH 2 and pH 6 for rosiglitazone and between pH 2 and pH 8 for the other drugs. As the pH increases the peak current decreases. At pH $\geq$ 8 the drug precipitates and no well defined peak can be recorded. The decrease of the peak current with the increase of the solution pH is attributed to the fact that the electroactive species of the drug occur in the basic form and the drug molecules are exchanging protons during the redox process. The rate of formation of the basic form of the drug conjugate acid it reaches its diffusion controlled limiting value at higher pH, where the rate of transformation of the conjugate acid to the basic form reaches its maximum.

3.1.1.1. Mechanistic data for the tested antidiabetic drugs. All tested drugs posses NH ionizable group depending on the solution pH. By the consideration of the electrostatic attraction, one could expect that the cationic form exists at pH 5 for rosiglitazone and at pH 6 for pioglitazone, glimepiride and glyburide.

3.1.2. Effect of scan rate

The scan rate was found to affect both the peak potential and peak current. By increasing the scan rate, the peak potential shifts in the anodic direction. The positive shift is accompanied by an increase in the peak current. The peak current (i_p) increases with the scan rate (ν), as logarithmic function, in the range between 10 and 300 mV s^{-1}. Considering a reduction process at potentials well positive from the redox potential, there is no faradic reaction in response to the pulse, so the difference current is zero. At potential around the redox potential, the difference current reaches a maximum, and decreases to zero as the current becomes diffusion controlled [22].

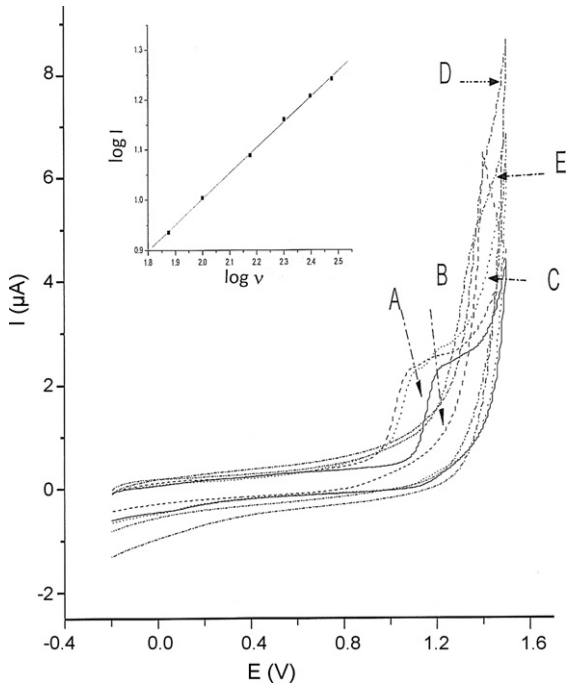

Fig. 2. Cyclic voltammogram of the different drugs recorded on GCE surface at pH 5.0 with a scan rate of 100 mV s^{-1}. (A) Rosiglitazone (—); (B) pioglitazone (- - -); (C) glimepiride (···); (D) glyburide (-·-·-·-); (E) blank solution (··—··—). (Insert): log i_p versus log ν for the determination of glimepiride recorded at GCE.

A typical example of the variation of the peak current of glimepiride with the scan rate is presented as insert in Fig. 2. The relation between i_p and ν is formulated as:

$$\log i_p = A + B \log \nu$$

where A is the intercept and B is the slope of the linear relation [23,24]. The experimental results of these measurements and the calculated values of A and B for each drug on both the CPE and GCE are presented in Table 2. The regression (R) is ≥0.992 and the precision (P) is ≤0.0001. An increase in the scan rate is accompanied by a positive shift in the peak potential and an increase in the peak current. The increase of i_p with ν is obeying the above log–log relation. The value of the slope of the obtained linear relations is around 0.5 which implies that the participating species are transported by a diffusion process [24]. This means that the electrode surface is, immediately, completely covered with the electroactive species. From the different investigated scan rates, the 100 mV s^{-1} gave the best voltammograms and higher selectivity.

Table 2
The calculated values from regression line equation for the diffusion current of each drug [N = number of runs and SD = standard deviation].

Drug	Electrode	N	SD
Rosiglitazone	CPE $A = -0.20 \pm 0.11$ $B = 0.51 \pm 0.05$	6	0.02
	GCE $A = -0.80 \pm 0.06$ $B = 0.56 \pm 0.03$	7	0.02
Pioglitazone	CPE $A = -0.20 \pm 0.13$ $B = 0.54 \pm 0.06$	6	0.03
	GCE $A = -0.40 \pm 0.09$ $B = 0.59 \pm 0.04$	6	0.02
Glimepiride	CPE $A = -0.20 \pm 0.04$ $B = 0.47 \pm 0.02$	6	0.01
	GCE $A = -0.03 \pm 0.04$ $B = 0.52 \pm 0.02$	6	0.01
Glyburide	CPE $A = -0.40 \pm 0.06$ $B = 0.45 \pm 0.03$	6	0.01
	GCE $A = -0.26 \pm 0.03$ $B = 0.49 \pm 0.01$	6	0.01

3.2. Differential pulse voltammetric determination of the drug samples

A scan rate of 10 mV s^{-1} gave well defined DPV peaks and reproducible results. The peak current increases with the successive additions of the drug. The differential pulse voltammograms for the determination of different concentrations of the four tested drugs at the CPE and GCE surfaces are recorded. Standard measurements were carried out and a calibration curve was constructed for each drug. Typical examples of the differential pulse voltammograms of rosiglitazone, and pioglitazone, recorded at the CPE and GCE are presented in Figs. 3 and 4a and b, respectively. The corresponding calibration curve is presented as insert in the respective voltammograms.

The regression data were obtained according to the Mircocol Origin software [25] and are summarized in Table 3. The regression (R) is ≥0.992 and the precision (P) is ≤0.0001. The peak current of any unknown drug concentration is measured and the concentration can be extrapolated from the corresponding calibration curve.

The lower limits of detection (LOD) and lower limits of quantization (LOQ) were calculated according to the following equations [26]:

$$\text{LOD} = 3 \times \text{SD/slope}$$

$$\text{LOQ} = 10 \times \text{SD/slope}$$

where SD is the standard deviation obtained from 5 different runs. The calculated values for each drug at both the CPE and GCE are presented in Table 3.

After having the calibration curve, the pharmaceutical preparations of the drugs presented in Table 1, were then measured by the DPV method. The electrochemical data were then compared with the data obtained by HPLC as a standard reference method [27,28]. There were no significant differences between the electrochemical method based on the CPE and GCE electrodes and the reference HPLC method. The comparison was made by calculating the % recovery of each drug in its pharmaceutical preparation and the standard error in the measurements by both techniques. The % recovery was obtained by the standard addition technique, where different levels of standards were added to previously analyzed samples. The amount of the measured drug concentration is then plotted against the amount of the added standard. The intercept of the plot gives the amount of the drug per sample. The percentage recovery is given by the ratio between the extrapolated value and the practical value of the pharmaceutical preparation. The standard error was calculated for at least five runs. The % recovery was calculated and a mean % recovery of 100 ± 1 was obtained. The standard deviation was found to be ±1.64 with a relative standard deviation of 1.62.

The data of these experiments and calculations are presented in Table 4. From the data presented in this table it is clear that the proposed electro-analytical method applied in this work for the determination of the investigated drugs is convenient, economic, less time consuming and can be used for the quality control in drug

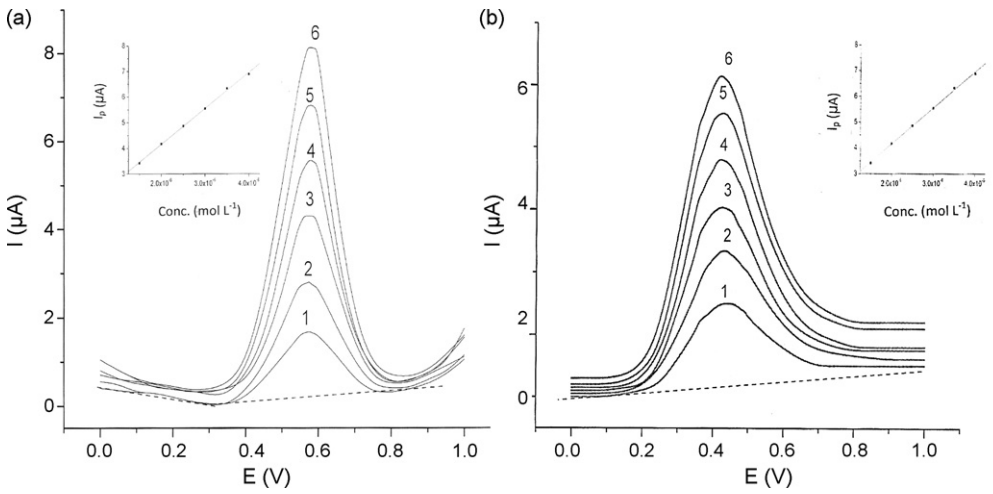

Fig. 3. (a) Differential pulse voltammograms for the determination of rosiglitazone at the CPE as a function of concentration of the drug: pulse amplitude = 50 mV, scan rate = 10 mV s^{-1}. The dotted line represents the blank solution. (1) 1.5×10^{-6} mol L^{-1}; (2) 2.0×10^{-6} mol L^{-1}; (3) 2.5×10^{-6} mol L^{-1}; (4) 3.0×10^{-6} mol L^{-1}; (5) 3.5×10^{-6} mol L^{-1}; (6) 4.0×10^{-6} mol L^{-1}. (Insert): calibration curve of the variation of the anodic peak current with the concentration of rosiglitazone at the CPE surface. (b) Differential pulse voltammograms for the determination of rosiglitazone at the GCE as a function of concentration of the drug: pulse amplitude = 50 mV, scan rate = 10 mV s^{-1}. The dotted line represents the blank solution. (1) 1.5×10^{-6} mol L^{-1}; (2) 2.0×10^{-6} mol L^{-1}; (3) 2.5×10^{-6} mol L^{-1}; (4) 3.0×10^{-6} mol L^{-1}; (5) 3.5×10^{-6} mol L^{-1}; (6) 4.0×10^{-6} mol L^{-1}. (Insert): calibration curve of the variation of the anodic peak current with the concentration of rosiglitazone at GCE surface.

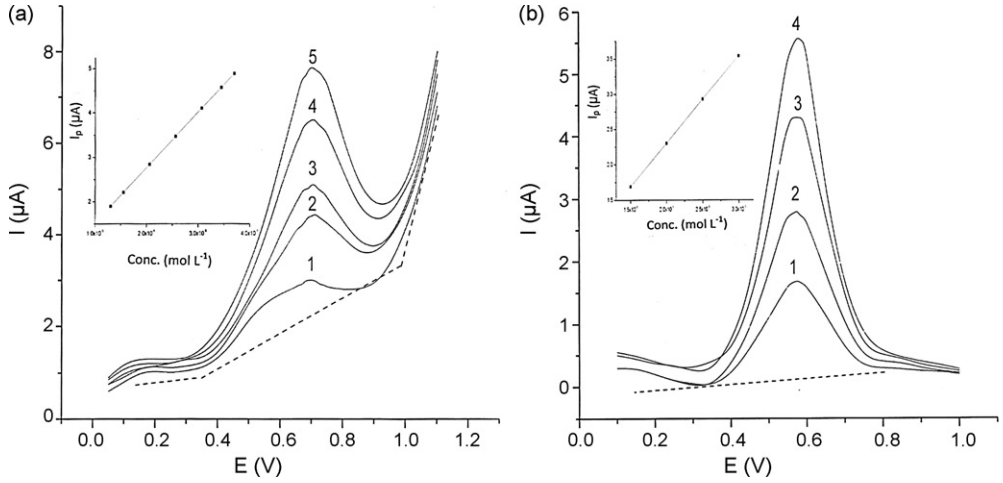

Fig. 4. (a) Differential pulse voltammograms for the determination of pioglitazone at the CPE as a function of concentration of the drug: pulse amplitude = 50 mV, scan rate = 10 mV s^{-1}. The dotted line represents the blank solution. (1) 1.5×10^{-6} mol L^{-1}; (2) 2.0×10^{-6} mol L^{-1}; (3) 2.5×10^{-6} mol L^{-1}; (4) 3.0×10^{-6} mol L^{-1}; (5) 4.0×10^{-6} mol L^{-1}. (Insert): calibration curve of the variation of the anodic peak current with the concentration of pioglitazone at the CPE surface. (b) Differential pulse voltammograms for the determination of pioglitazone at the GCE as a function of concentration of the drug: pulse amplitude = 50 mV, scan rate = 10 mV s^{-1}. The dotted line represents the blank solution. (1) 1.5×10^{-6} mol L^{-1}; (2) 2.0×10^{-6} mol L^{-1}; (3) 3.0×10^{-6} mol L^{-1}; (5) 4.0×10^{-6} mol L^{-1}. (Insert): calibration curve of the variation of the anodic peak current with the concentration of rosiglitazone at GCE surface.

analysis. The reproducibility of the measurements was also investigated and for five independent experiments at a concentration of 1.0×10^{-6} mol L^{-1} drug solution. Fairly reproducible data, which leads to more confidence in the method, were obtained.

3.3. Validation of the method

The specificity of the method was confirmed by investigation of the voltammograms of both the standards and the drugs' test

Table 3
Regression data of the calibration lines for the quantitative determination of rosiglitazone, pioglitazone, glimepiride and glyburide at CPE and GCE surfaces by DPV technique [*N* = number of runs, *R* = regression, SD = standard deviation, *P* = precision, LOD = lower limit of detection, LOQ = lower limit of quantization, Rosi: = rosiglitazone, Pio = pioglitazone, Gli = glimepiride, Gly = glyburide].

Electrode type	Drug	Linearity range (mol L^{-1})	Slope (μA (mol L^{-1})$^{-1}$) ± SE	Intercept (μA ± SE)	SD	*N*	LOD (mol L^{-1})	LOQ (mol L^{-1})
CPE	Rosi.	1.5×10^{-6}–4×10^{-6}	$3 \times 10^{6} \pm 2 \times 10^{4}$	$-2 \pm 6 \times 10^{-2}$	0.04	6	5×10^{-8}	2×10^{-7}
	Pio.	1.5×10^{-6}–4×10^{-6}	$1 \times 10^{6} \pm 7 \times 10^{4}$	$0.3 \pm 2 \times 10^{-1}$	0.11	5	3×10^{-7}	9×10^{-7}
	Gli.	1.5×10^{-6}–3×10^{-6}	$1 \times 10^{6} \pm 6 \times 10^{3}$	0.1 ± 10^{-1}	0.06	4	2×10^{-7}	5×10^{-7}
	Gly.	1.5×10^{-6}–4×10^{-6}	$2 \times 10^{6} \pm 4 \times 10^{4}$	$^{-}0.6 \pm 10^{-1}$	0.08	6	1×10^{-9}	3×10^{-7}
GCE	Rosi.	1.5×10^{-6}–4×10^{-6}	$1 \times 10^{6} \pm 3 \times 10^{4}$	$1 \pm 7 \times 10^{-2}$	0.05	6	1×10^{-7}	4×10^{-7}
	Pio.	1.5×10^{-6}–3×10^{-6}	$1 \times 10^{6} \pm 6 \times 10^{3}$	$^{-}0.2 \pm 10^{-2}$	0.01	4	2×10^{-8}	2×10^{-8}
	Gli.	1.5×10^{-6}–4×10^{-6}	$1 \times 10^{6} \pm 1 \times 10^{5}$	$0.7 \pm 3.3 \times 10^{-1}$	0.2	5	6×10^{-7}	2×10^{-6}
	Gly.	1.5×10^{-6}–3×10^{-6}	$2 \times 10^{6} \pm 2 \times 10^{5}$	$^{-}1.8 \pm 3.7 \times 10^{-1}$	0.2	4	3×10^{-7}	9×10^{-7}

Table 4
Assay for rosiglitazone, pioglitazone, glimepiride and glyburide in pharmaceutical preparations by the electrochemical and the standard HPLC techniques [SE = standard error].

Sample	Claimed (mg)	Recovery% ± SE of CPE	Recovery% ± SE of GCE	Recovery% ± SE of HPLC
Avandia tablets (rosiglitazone)	4.0	99 ± 0.6	101 ± 0.3	100 ± 0.4
	2.0	99 ± 0.4	100 ± 0.6	98 ± 0.5
	1.0	99 ± 0.4	98 ± 0.7	100 ± 0.6
Glustin tablets (pioglitazone)	30.0	100 ± 0.6	98 ± 0.6	99 ± 0.5
	15.0	101 ± 0.5	99 ± 0.4	99 ± 0.4
Amaryl tablets (glimepiride)	3.0	102 ± 0.3	101 ± 0.6	100 ± 0.6
	2.0	100 ± 0.3	99 ± 0.6	101 ± 0.4
	1.0	100 ± 0.6	100 ± 0.4	101 ± 0.5
Daonil tablets (glyburide)	5.0	101 ± 0.5	99 ± 0.6	102 ± 0.4

Table 5
Mean percent recovery of different samples of different concentrations for the four tested drugs (three runs for each).

Set no.	Standard	Concentration added of standard solution ($\mu g\,mL^{-1}$)	Concentration found of standard solution ($\mu g\,mL^{-1}$)	%Found	Mean %found
I		4.0	3.6	91	
II	Rosiglitazone	8.0	8.3	104	97 ± 0.5
III		12.0	12.4	104	
I		15.0	15.2	101	
II	Pioglitazone	30.0	30.6	102	101 ± 0.4
III		45.0	44.7	99	
I		3.0	2.9	97	
II	Glimepiride	6.0	6.4	107	102 ± 0.2
III		9.0	9.2	102	
I		5.0	4.6	93	
II	Glyburide	10.0	10.4	101	100 ± 0.6
III		15.0	15.4	103	

solutions. Identical voltammograms were obtained. The addition of the standard antidiabetic drugs' solutions to the corresponding test solutions did not change the characteristics of the differential pulse voltammogram of each drug and the same calibration curve was obtained. No significant differences between the proposed electrochemical method and the reference HPLC method could be found.

The accuracy of the method for the determination of the four tested drugs in their tablets' forms was performed by the addition of the standard of each drug to its sets of solutions containing the formulated adjuvant. Three sets of different concentrations of standards for each of the tested drugs were measured and the percent recovery was calculated. The results of these experiments are summarized in Table 5. The mean percent recovery presented in Table 5 indicates the accuracy of the method.

3.4. Precision and repeatability

Each determination either for the standards or the test solutions of the four drugs has been carried out at least three times. The relative standard deviation (RSD) was calculated to be 1.62 indicating the high precision of the method and the confidence in its repeatability.

3.5. Robustness

The robustness of the proposed method is evaluated by the constancy of the peak area values with the deliberated small changes in the experimental parameters, which was realized by the method. The time between the preparation of the solutions and the measurement gives an indication about this factor. As described in the procedure section for the determination of the standards ($15\ \mu g\,mL^{-1}$), the measurements were carried out at 30 min intervals over a period of 2 h. The % recovery was calculated and a mean % recovery of 100 ± 1 was obtained. The standard deviation was calculated and found to be ±1.64 with a relative standard deviation of 1.62.

The above measurements show that the prepared solutions are stable and could be measured within a period of 2 h after preparation without any effect on the accuracy and precision of the method. This is indicated by the mean % recovery and RSD, respectively.

4. Conclusions

Rapid and accurate determination of active drugs for type 2 diabetic patients either as standards or in their pharmaceutical preparations was achieved electrochemically. The electromeric method used involves two different working electrode sensors, the CPE and GCE, for the determination of rosiglitazone, pioglitazone, glimepiride and glyburide. The differential pulse voltammetry, DPV, was found to be fast, sensitive and selective without any significant difference to the reference HPLC method. The method, beside its low cost, can be applied for the drug analysis in any form without special separation or sample preparations. It is very selective without any interference from solution constituents.

References

[1] The Martindale Extra Pharmacopeias, 32nd ed., Royal Pharmaceutical Society, London, 1999, p. 331.
[2] A.J. Kiayias, D.E. Vlachou, E. Theodosopoulou, E. Lakka, Diabetes Care 25 (2002) 1251.
[3] R.C. Turner, C.A. Cull, V. Frghi, R.R. Holman, JAMA 281 (1999) 2005.
[4] N. Jones, T. Jones, L. Menci, X. Jane, M. Freed, M. Kreider, Diabetologia 44A (2001) 904.
[5] J.D. Dai, D.J. Jin, Y.L. Liu, Yaou Fenxi Zazhi 21 (2001) 36.
[6] A.M. Muxlow, S. Fowles, P. Russell, J. Chromatogr. B 752 (2001) 77.
[7] S.X. Wei, Z.Q. Zhang, Yaowu Fenxi Zazhi 21 (2001) 365.
[8] C.E.C.A. Hop, P.R. Tiller, L. Romanyshyn, Rapid Commun. Mass Spectrom. 16 (2002) 212.
[9] B.L. Kolt, B.B. Raut, A.A. Deo, M.A. Bagool, D.B. Shinde, J. Chromatogr. A 37 (2003) 788.
[10] R.N.V.S. Mamidi, B. Benjamin, M. Ramesh, N.R. Srinivas, Biomed. Chromatogr. 17 (2003) 417.

[11] L. Kolt, B.B. Raut, A.A. Deo, M.A. Bagool, D.B. Shinde, J. Chromatogr. A 42 (2004) 27.
[12] B. Fried, J. Sherma, Thin Layer Chromatography, Chromatographic Science Series, 4th ed., Marcel Dekker, Inc., New York, 1999, p. 81.
[13] F. Sayin, S. Kir, J. Pharm. Biomed. Anal. 25 (2001) 153.
[14] S.A. Özkan, B. Uslu, P. Zuman, Anal. Chim. Acta 457 (2002) 265.
[15] X.E. Jiang, I.P. Guo, X.G. Du, Talanta 61 (2003) 247.
[16] A. Salimi, V. Alizadeh, Hadadzadeh, Electroanalysis 16 (2004) 1984.
[17] N. Fay, E. Dempsey, T. Maccormac, J. Electroanal. Chem. 574 (2005) 359.
[18] X.L. Wang, H.Y. Zahr, H.Y. Lin, G.C. Liu, J.N. Fang, B.K. Vhen, Electroanalysis 20 (2008) 1055.
[19] W.R. Heineman, P.T. Kissinger, J. Am. Lab. 60 (1982) 29.
[20] R.T. Sane, S.N. Menon, S. Inamdar, M. Mote, G. Gundi, Chromatographia 59 (2004) 451.
[21] W.A. Badawy, M.A. El-Riesb, I.M. Mahdi, Anal. Sci. 25 (2009) 1431.
[22] A.J. Bard, L.R. Faulkner, Electrochemical Methods—Fundamentals and Applications, John Wiley & Sons, New York, 1980 (Chapter 5).
[23] Pharmacopeias European Monograph, 2003, p. 0718.
[24] D.K. Gosser, Cyclic Voltammetry, Simulation and Analysis of Reaction Mechanism, VCH, New York, 1993, p. 43.
[25] Origin pro7, Scientific Analysis and Graphing Software, http://www.OriginLab/TechnicalSupport.
[26] J.C. Miller, J.N. Miller, Statistics for Analytical Chemistry, Ellis Hardwood Series, PTR Prentice Hall, New York/London, 1993, p. 119.
[27] W.Z. Zhong, M.G. Williams, J. Pharm. Biomed. Anal. 14 (1996) 645.
[28] P. Kovaiková, J. Klima, J. Dohnalb, L. Tisovskab, J. Pharm. Biomed. Anal. 36 (2004) 205.