Int. J. Electrochem. Sci., 16 **(2021)** Article ID: 210242, doi: 10.20964/2021.02.32

International Journal of
ELECTROCHEMICAL
SCIENCE
www.electrochemsci.org

Voltammetric Behavior of Trimetazidine Hydrochloride at Pre-anodized Carbon Paste Electrode and Its Determination in Pharmaceutical Formulations

Sen Fan[1], *Lei Ji*[2], *Guoliang Mao*[2], *Huan Wang*[2], *Yongkui Fang*[2], *Jiguo Duan*[2], *Yuanhai Zhu*[2,*] *and Hua Song*[2]

[1] College of Petroleum Engineering, Northeast Petroleum University, Daqing 163318, P.R. China
[2] College of Chemistry and Chemical Engineering, Northeast Petroleum University, Daqing 163318, P.R. China
[*]E-mail: yuanhaizhu@163.com

Received: 23 October 2020 / *Accepted:* 4 December 2020 / *Published:* 31 December 2020

A functional pre-anodized carbon paste electrode (PACPE) was prepared by 35 successive cycles of scans between 1.0V-1.8V in 0.2 mol L^{-1} NaOH solution. The electrode reaction mechanism of trimetazidine hydrochloride (TMZ) was carefully studied by CV method and was determined as a two-electron/two-proton process, which is put forward for the first time. Experiment showed that the electrostatic attraction between the protonated TMZ and the negatively charged PACPE surface, both were affected by pH, was the dominant factor influencing the adsorption and the peak current. The PACPE exhibited excellent electrocatalytic effects towards the oxidation of TMZ. In the optimum conditions, the oxidation peak current was linear with TMZ concentration in the range of 5.0×10^{-7}—5.0×10^{-5} M (r=0.998), and the detection limit was estimated to be about 1.5×10^{-7} M. This method can be successfully applied to the determination of TMZ in pharmaceutical formulation.

Keywords: Trimetazidine hydrochloride; pre-anodized carbon paste electrode; cyclic voltammetry; pharmaceutical analysis

1. INTRODUCTION

Trimetazidine dihydrochloride (TMZ), (1-(2,3,4-Trimethoxbenzyl) piperazine) (Fig. 1), is an antianginal drug which can regulate ion exchange across cell membrane and prevents cellular edema caused by ischemia and anoxia[1]. TMZ has no negative inotropic effects or vasodilatory characteristics and can be combined with other antianginal drugs as a complementary therapy mostly for patients with companion diseases such as left ventricular dysfunction and diabetes mellitus[2].The interest in pharmacokinetic mechanism and the demand for quality control have promoted the development of trimetazidine analysis methods[3]. Some analytical methods have been established for the determination of trimetazidine in pharmaceutical formulations and biological fluids, including HPLC[4,5], LC-MS[6], GCMS[7,8], HPTLC[9,10], spectrophotometry[11], chemiluminescence[3] and voltammetry[12]. The methods listed above are mainly traditional chromatography or related hyphenated chromatographic techniques. Although these techniques are quite reliable and sensitive, they are limited by expensive instrumentation, high cost and complicated pre-treatment process. Compared with these methods, electrochemical determination has the advantages of low cost, high sensitivity, simple operation and in-situ monitoring. Investigations on direct electrochemical detection of TMZ, however, are quite limited.

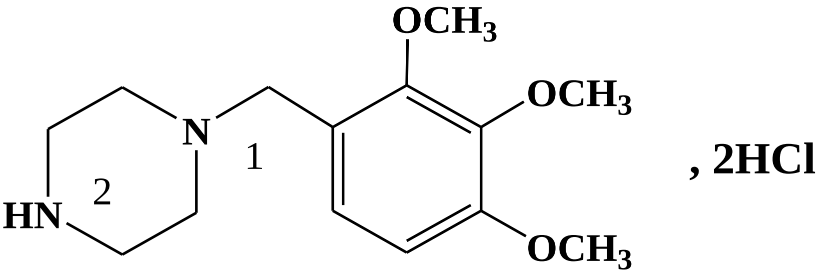


Figure 1. Chemical structure of trimetazidine hydrochloride

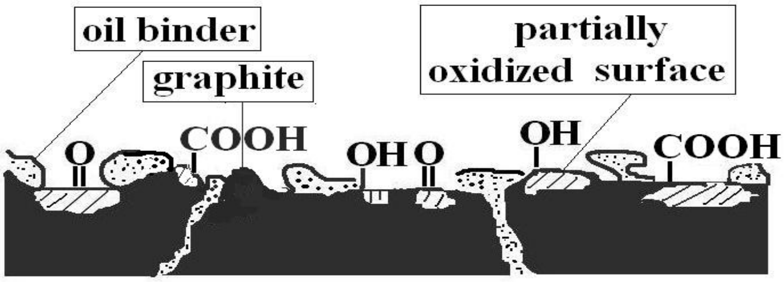


Figure 2. A schematic view of anodic activation of the carbon paste surface.

Carbon paste electrode (CPE) has been widely used due to the advantages of easy preparation, low cost, wide electrochemical window and easy chemical modification [13–17]. The pre-anodized carbon paste electrode (PACPE) is a chemically modified electrode obtained by anodizing the carbon paste electrode in dilute sodium hydroxide solution. By means of a simple anodized pre-treatment, many oxygen-containing functional groups such as carboxyl (-COOH), carbonyl (-C=O), phenolic hydroxyl (-OH), lactones, and ethers, can be easily modified on the surface of CPE (Fig. 2)[18,19], which make the PACPE demonstrate excellent electro-catalytic activities[18]. However, PACPE only has catalytic activity to some special substances, and it is very difficult to predict for which compounds the voltammetric behavior can be improved in this way [20]. This may be the reason why the PACPE has not been widely used. In this work, it was accidentally found that PACPE showed good catalytic effect on the electrochemical oxidation of TMZ, and the voltammetric behavior of TMZ on PACPE and its detection in pharmaceutical formulations is investigated.

2. EXPERIMENTAL

2.1. Reagents and Apparatus

Standard TMZ was obtained from Chinese National Institute for Food and Drug Control and was prepared into a stock solution of 1×10^{-3} M with doubly distilled water. Other chemicals except TMZ were of analytical grade. All experiment of cyclic voltammetry was performed on an electrochemical workstation of CHI 660e (Chenhua Co., Shanghai, China) with a three-electrode system. The working electrode was a Teflon-tube filled with carbon paste. A Pt wire with enough large surface area was used as the counter electrode, and a saturated camel electrode (SCE) served as the reference electrode. A Shanghai Leici pHS-25 digital pH meter was employed to carry out all pH-metric measurements.

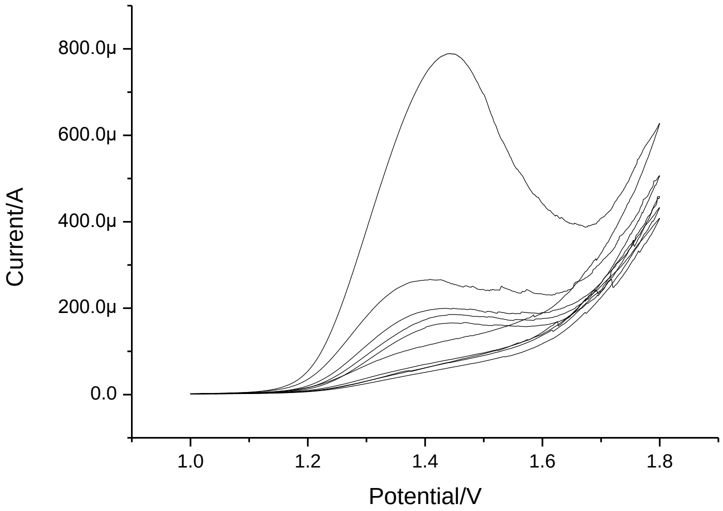


Figure 3. The first 5 successive cycles in the process of anodization of CPE in 0.2 M NaOH solution.

2.2. Preparation of the PACPE

A proper amount of graphite powder and paraffin oil were put into a mortar and repeatedly ground to obtain a homogeneous carbon paste, which was filled into the cavity of Teflon-tube with inner a diameter of 2 mm, and then the CPE surface was polished shiny on the weighing paper. The polished carbon paste electrode was placed in 0.2 M NaOH solution and scanned between 1.0 and 1.8 V at a rate of 100 mVs^{-1}. Experiment showed that the oxidation sensitivity of TMZ reached the best at 35 cycles, which were selected to anodize the CPE. Fig. 3 is cyclic voltammograms of the first 5 successive cycles of pre-anodization.

2.3. Preparation of Sample Solutions

The sample solution was prepared as follows. TMZ tablets were ground into powder and dissolved into certain amount of secondary distilled water, and then the obtained mixture was sonicated and filtrated through a 0.45-μm nylon filter to remove the insoluble substances. Thus obtained sample solution can be diluted with the 0.04 M Britton-Robinson (B-R) buffer according to the concentration range determined.

2.4. Procedure for the Determination of TMZ in the Samples

In this work, 0.04 M B-R buffer solution (pH 4.0) was selected as the supporting electrolyte for TMZ determination. A certain amount of sample solution was injected into a small electrolytic cell containing the buffer. After accumulation for 60s at open circuit, the voltammograms from 0.40 V to 1.3 V were recorded. The concentrations of the sample were calculated according to peak currents measured at 1.01v. Before each measurement, the PACE was renewed by repeated potential scans from 0.40 V to 1.3 V in the blank buffer at a rate of 100 mVs^{-1}.

3. RESULTS AND DISCUSSION

3.1. Electrochemical performance of PACPE

Fig. 3, first reported here, shows that within the potential range from 1.0 V to 1.8 V the oxidation peaks of hydroxyl radical (OH^-) appear near 1.4 V at the CPE, and the peak currents gradually decrease with the increase of scanning cycles, which may be attributed to the adsorption of the produced gases which prevent the electron transfer. The voltammetric curves finally stabilize and maintain unchanged owing to the establishment of adsorption-diffusion steady state. The previous work [21] indicates the following electrode reaction may account for the oxidation peak in Fig.3:

$$C(s) + OH^- \rightarrow C(s)OH(ads) + e \qquad (1)$$

The observed gas evolved on the surface of the CPE during pre-anodization was explained by the following reaction [21]:

$$4C(s)OH(ads) \rightarrow 4\{C\} + 2H_2O + O_2 \qquad (2)$$

Where C(s) implies an intact carbon lattice and {C} indicates carbon material etched from the graphite particles.

The above chemical reaction(2) proposed , which includes the production of carbon, was based on the discovery of carbon fragments in the solution of the working electrode during the study of electrochemical oxidation of carbon fibers in NaOH solutions [21]. However, in anodizing the CPE, no obvious formation of carbon debris in the vicinity of the electrode was found. Moreover, reaction (2) failed to explain how the surface oxygen-containing functional groups were formed. Therefore, there should be a more reasonable mechanism to explain not only the formation of gas, but also the formation of carbon oxygen functionalites on the PACPE surface. After the electrode oxidation (1), the hydroxyl ions (OH^-) were actually be oxidized into hydroxyl free radicals ($OH^\bullet$) and adsorbed on the electrode surface. Hydroxyl free radicals ($OH^\bullet$) were considered as one of the most powerful oxidizing species [22-24], which could readily undergo electrophilic addition reaction with the unsaturated bonds on the graphite surface introducing a lot of -C-OH groups on it as they did with carbon nanotubes [25]. The -C-OH groups could be easily oxidized into quinone groups –C=O or further converted into –COOH groups by hydroxyl radicals $OH^{\bullet}$ [25]. The gas produced on the PACPE should be CO_2 coming from the decarboxylation reaction on the electrode surface [25]:

$$\text{PACPE–COOH} + OH^{\bullet} \rightarrow \text{PACPE}^{\bullet} + CO_2 + H_2O \qquad (3)$$

It was impossible for the above reaction to completely consume all the carboxyl groups on the surface of the PACPE, because they were produced constantly as the pre-anodization continued, hydroxyl (-OH), carboxyl (-COOH) or carbonyl (-C=O) groups always coexisted as shown in Fig 2. Whether there is reaction (2) in the preanodization of CPE needs further study.

Oxygen-containing functional groups, such as hydroxyl (-OH), carboxyl (-COOH), and carbonyl (-C=O) on the CPE are polar and hydrophilic, they repel the hydrophobic molecules of the oil binder on the CPE turning the lipophilic CPE surface to hydrophilic (Fig. 2), and many clean and micro-reactive sites are produced in etching process, which make anodization an effective way of enhancing the electrode sensitivity [18].

Fig. 4 is the voltammetric responses of ferricyanide/ ferrocyanide at the CPE and PACPE before and after anodization. Not only the peak currents are remarkably enhanced, but also the redox peak potential separation decreases from 230 mV to 78 mV, indicating that a reversible electron transfer process occurs at the PACPE similar to that on a clean GCE.

Investigations have proved that the repulsive effect of the negatively charged carboxylates on $Fe(CN)_6^{3-/4-}$ anions can be ignored for the pre-anodized GCE and the PACPE, however, their repelling forces on organic small molecule anions such as ascorbic acid are obvious[19, 21-27]. The experiment also showed that a pair of reversible redox peaks also appeared on the electrode modified with carboxylated carbon nanotubes for $Fe(CN)_6^{3-/4-}$ reaction in 0.1M KCl solution. The repulsive force of PACPE to $Fe(CN)_6^{3-/4-}$ anion always exists, which is only balanced by the favorable effect of the newly generated active sites and neutral groups on the pre-anodized carbon surface. Evidently, mechanism of the reaction of ferricyanide/ferrocyanide at anodized carbon material electrode is complex and needs further study.

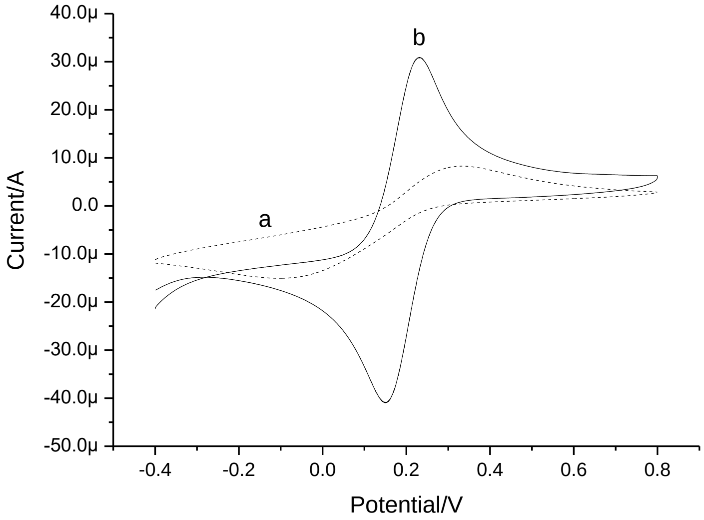


Figure 4. Cyclic voltammograms of $K_4[Fe(CN)_6]$ at different electrodes: (a) CPE, (b) PACPE; $c(K_4[Fe(CN)_6])=1.0\times10^{-3}$ M; supporting electrolyte: 0.1 M KCl; scan rate: 100 mVs^{-1}.

3.2. Voltammetric Response of TMZ at PACPE

It is well known that organic amine groups are electroactive in certain conditions. The structure of TMZ (Fig. 1) indicates that nitrogen atoms at sites 1 or 2 all may be oxidized, but the real oxidation site should be at site 1, because the basicity or ability to donate electrons of the tertiary amine is greater than that of the secondary. Moreover, as is introduced in the following context, the oxidation transition state at site 1 is more stable and the product does not destroy the stable hexatomic piperazine ring. Voltammograms of 5.0×10^{-5} M TMZ at CPE and PACPE in B-R buffer solution (pH=4) are showed together in Fig. 5 for comparison, obviously the peak current at PACPE is greatly enhanced and the peak potential is decreased.

The peak current has a positive relation with the adsorption amount and electron transfer rate. Similar to “like dissolves like” principle, hydrophilic interface is easy to adsorb hydrophilic substances, while hydrophobic interface is easy to adsorb hydrophobic substances. TMZ is soluble in water because of the two protonated hydrophilic amine groups in the molecule, thus it is difficult for them to adsorb on the hydrophobic surface of the CPE. At the CPE, a small amount of adsorbed TMZ molecules tend to be oriented with trimethoxbenzyls to the hydrophobic surface and piperazine groups to aqueous solution. Such an adsorption state is obviously unfavorable to the electron transfer of the oxidation site 1, therefore, a broad and weak oxidation peak appears at about 1.10V in the voltammogram (Fig. 5). However, after pre-anodization in mild NaOH solution, the surface of CPE will change from hydrophobicity to hydrophilicity (as shown in Fig. 2). Protonated and positively charged piperazines in TMZ molecules are easily adsorbed on the negatively charged hydrophilic PACPE surface through hydrogen bond and electrostatic attraction, while the hydrophobic part of the TMZ molecule may lie on the graphite. Such a molecule orientation is favorable for the adsorption and the electron transfer, thus results in an increase in peak current and a decrease in overvoltage. Fig. 5 shows that TMZ exhibits a sharp and well-defined oxidation peak at 1.0 V, suggesting the oxidation of

TMZ is significantly catalyzed at PACPE.

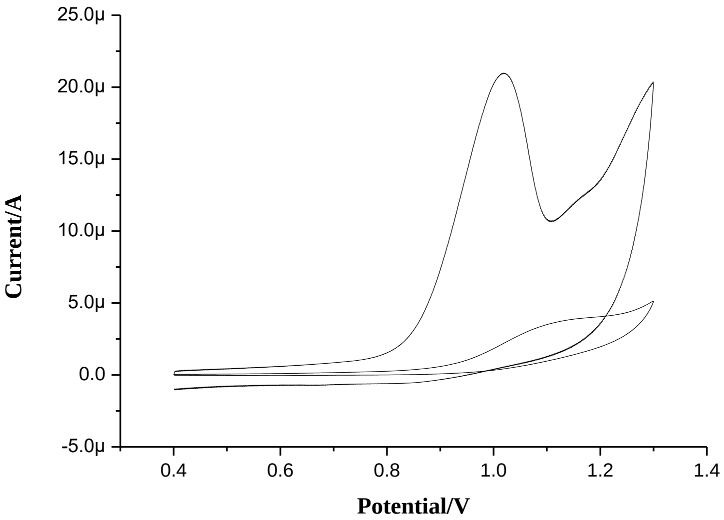


Figure 5. Voltammograms of 5.0×10^{-5} M TMZ in B-R buffer solution (pH=4): a) at CPE b) at PACPE. Accumulation time was 60 s (open circuit); scan rate was $0.1Vs^{-1}$.

3.3. The pH dependence of the peak potential and peak current

Fig.6 shows the experimental results of the relationship between the peak potential and peak current of different pH values, and the peak potential decreases linearly with the pH from 2 to 7 with a slope of -0.061 V/pH (r=0.988) (inset of Fig. 6), which is very close to -0.059 V/pH, indicating that protons and electrons lost are the same in the process of TMZ oxidation at PACPE.

Many factors may affect the peak current, such as the amount of adsorption, the rate of electron transfer and so on. Fig. 6 shows that the peak currents first increase with the acidity of the buffer, and then begin to decrease after reaching a maximum value at about pH=4. Referring the pK_a of benzoic acid (4.21), in the range of pH>4 the surface of PACPE may be negatively charged due to the ionization of carboxyl groups. The number of protonated TMZ molecules and the electrostatic attraction to the PACPE surface increase with the acidity of the B-R buffer, therefore, the amount of adsorption and the oxidation peak current of TMZ increase accordingly. However when pH<4 the ionization of the carboxyl groups will be inhibited and the carbonyl groups begin to protonate, such changes will weaken the electrostatic attraction between the electrode surface and positively charged TMZ molecules, resulting in a decrease in peak currents. On the other hand, the increase in protonation has an opposite effect to decrease the peak current, because protonation can increase solubility of TMZ in water, which may lead to a decrease in amount of adsorption. Evidently for the system studied here, the electrostatic attractions between negatively charged PACPE surface and the highly protonated TMZ molecules are dominant factor affecting the peak currents of TMZ. In this work, pH=4 of the B-R buffer solution was employed for the determination of TMZ.

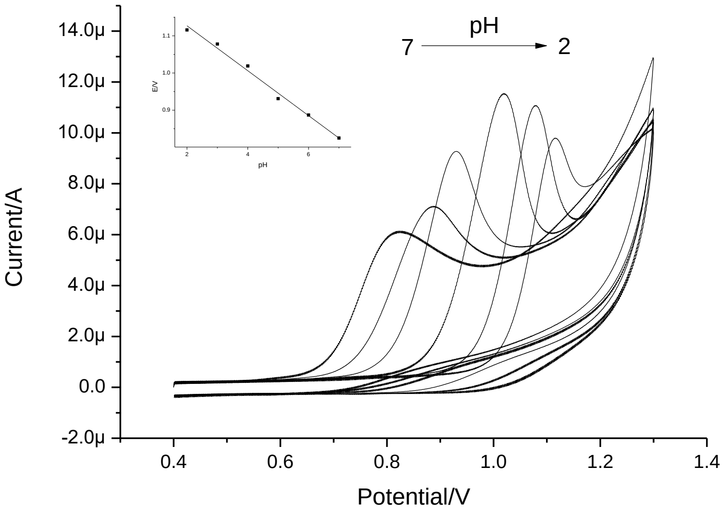


Figure 6. Cyclic voltammograms of 3.0×10^{-5} M TMZ in 0.04 M B-R buffer solution at different pHs, 2, 3, 4, 5, 6, 7, scan rate 0.1V s^{-1}, the insert is the plot of E_p vs pH. Accumulation time: 60s.

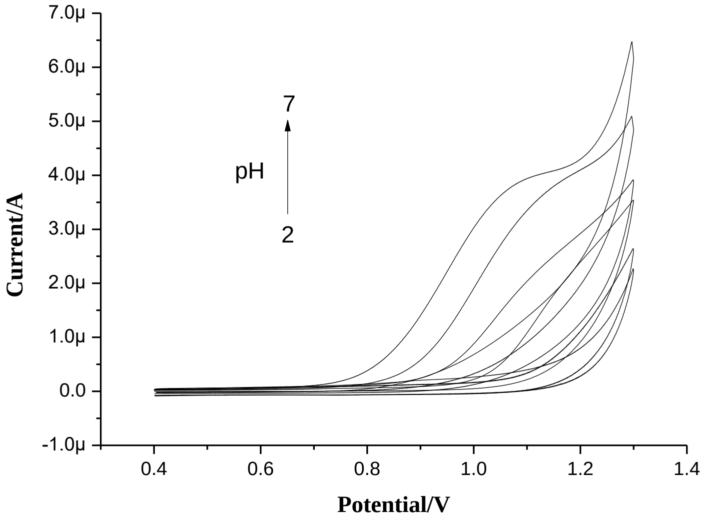


Figure 7. Cyclic voltammograms of 2.0×10^{-5} M TMZ in 0.04 M B-R buffer solution at different pHs, 2–7, scan rate was 0.1V s^{-1}.

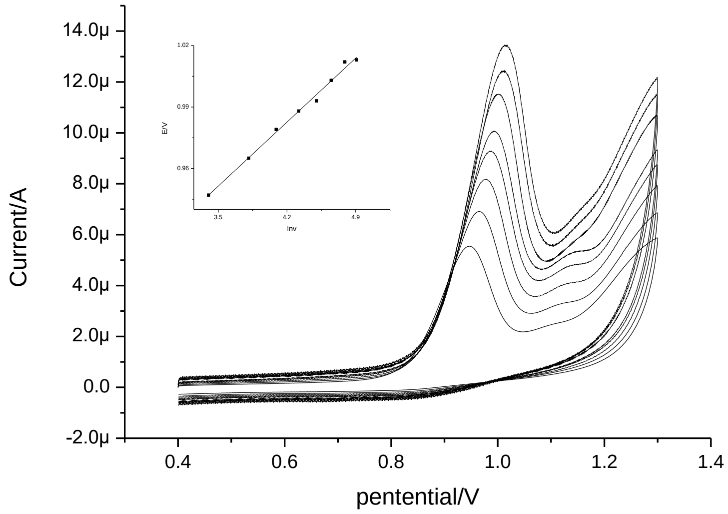


Figure 8. Influence of scan rate on the anodic peak of 2.0×10^{-5} M TMZ in 0.04 M B-R buffer (pH =4.0). Scan rate: 30, 45, 60, 75, 90, 105, 120, 135 mVs^{-1} (from bottom to top). Accumulation time: 60s. The insert is a plot of E_p vs. ln v

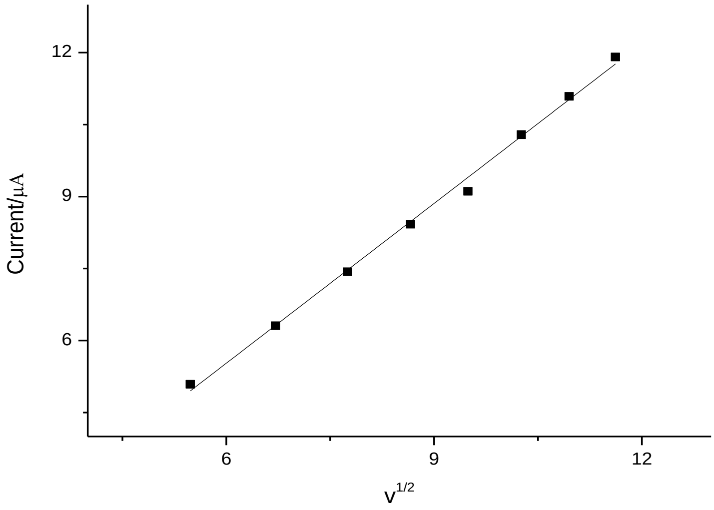


Figure 9. Plot of the anodic current vs.the square root of the scan rates: 30, 45, 60, 75, 90, 105, 120, 135 mVs^{-1}. Other conditions are same as in Fig.8

It is a common phenomenon that the oxidation peak current of some small molecules decreases with the decrease of acidity at the anionic carbon electrode in a certain pH range, and a few researchers believe that at the PACPE such a phenomena should be attributed to the decrease in the amount of oxygen reduced at the auxiliary electrode (AE) because of lack of H$^+$ at high pH [28-30]. In other words, the control step is transferred to the AE, which is obviously different from our explanation in this work. Actually in order to study the potential-current relationship on the working electrode the surface area of the AE in the three-electrode system must be made large enough and nonpolarizable, or else the data obtained would be meaningless. All our experimental results were acquired with the area of AE 100 times more than that of working electrode to keep the reaction fast enough, and the only role of the AE was conducting current, thus it was impossible for the reaction on the AE to evolve into a control step at high pH and to decide the size of the peak current.

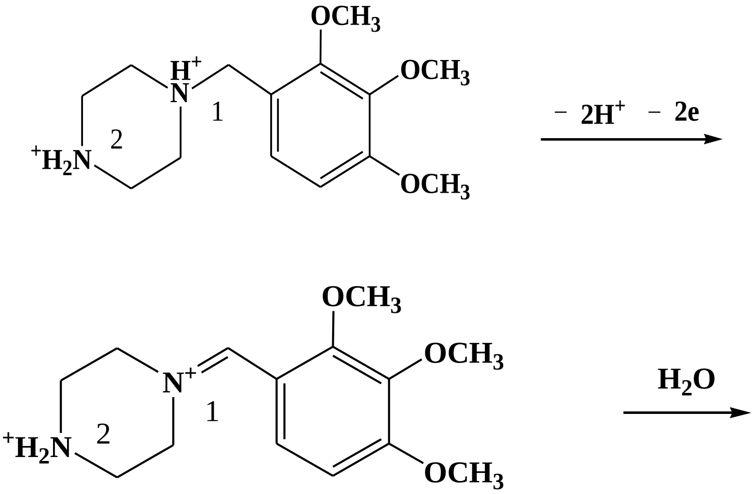
It is predicted that the oxidation peak current at bare CPE should increase with the increase of pH value due to the decrease in solubility of TMZ (Fig. 7). The solubility of organic amine in water decreases with the pH increase because of the reduced protonation, which is often used to separate amine from aqueous solution at high pH. Similar results are also obtained for another organic amine drug phentolamine Mesylate [31].

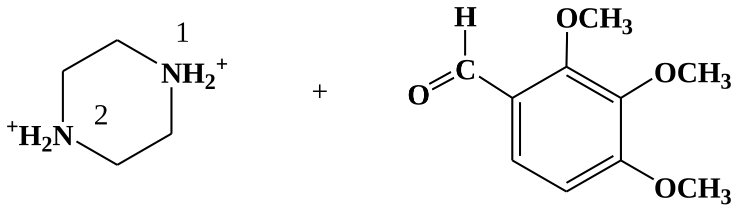
3.4. Mechanism of the Electrode Reaction of TMZ

In 2×10^{-5} M TMZ solution (pH=4), the effects of scan rate on the oxidation peak current and peak potential at the PACPE by cyclic voltammetry (Fig. 8) was studied. It was found that there was a linear relationship between the peak currents and the square root of the scan rates from 30 to 135 mVs^{-1}, $I_\mathrm{p} = 1.11\mathrm{v}^{0.5}\text{-}1.13$ (r=0.996, Fig. 9), which indicated that the oxidation process of TMZ at the PACPE was controlled by diffusion. The insert of Fig. 8 shows that E_p varies linearly with lnv from 30 to 135 mVs^{-1}, and the fitting relation is E_p=0.7953+0.0446lnv (r=0.993). As for an irreversible diffusion-controlled process, the peak potential E_p can be expressed by the following equation [32]:

$$E_\mathrm{p} = E^{0'} + \frac{RT}{(1-\alpha)nF}\left\{0.780 + \ln\left(\frac{D_R^{1/2}}{k^0}\right) + \ln\left[\frac{(1-\alpha)nFv}{RT}\right]^{1/2}\right\}$$

where α is the electron transfer coefficient, k^0 is the standard heterogeneous rate constant, n is the electron transfer number involved in the oxidation, v is the scan rate, and $E^{0'}$ is the formal potential, and other symbols have their usual meaning. According to the slope of fitting relation, (1-α) n is calculated to be 0.87 (taking T = 298). Assume α to be 0.5, and n is estimated as 1.74~2. Combined with the previous conclusions, we know that the electroxidation of TMZ is a two-electron/two-proton process, which can be expressed as follows.





Trimetazidine is biprotonated both in crystal and in acidic solution, therefore, it participates in the electrode reaction in the form of bivalent positive ions at pH=4.

As stated above, oxidation occurs at N1 rather than N2 mainly because the former has higher basicity, however, if N1 was directly bound to benzene ring as in ciprofloxacin and norfloxacin, the case would be totally different. This time the lone-pair electron of sp3 hybrid of the N1 atom contained more p orbital and formed a stable conjugate system with the adjacent benzene ring, which greatly reduced its basicity, so the oxidation occurred on the stronger basic N2 not the N1 atom [23, 34]. The oxidation of bromhexine at platinum electrodes modified with nickel functionalized multi-walled carbon nanotubes had similar mechanism [35].

3.5. Chronocoulometry

The diffusion behavior of TMZ was studied by chronocoulometry. The total charge Q in the electrolysis of TMZ can be expressed by the integrated Cottrell equation [32]: $Q=2nFAcD_R^{0.5}t^{0.5}\pi^{-0.5}+Q_{\mathrm{dl}}+Q_{\mathrm{ads}}$, where the symbols and meaning in the equation are usual in electrochemistry and will not be explained one by one here. The slope of Q vs. $t^{0.5}$ was calculated to be 8.15 $\mu Cs^{-0.5}$, and according to this, the diffusion coefficient D_R of TMZ was determined as 1.52×10^{-4} cm^2s^{-1} with the known parameters A=3.14cm^2, c=10^{-4}M and n=2.

3.6. The Optimum of Accumulation Time

The influence of accumulation time on the peak current was studied at the TMZ concentration of 1×10^{-5}M. It was found that at open circuit the peak current increased with the accumulation time within 60 s and then maintained nearly unchanged due to the saturation of TMZ adsorption. For higher sensitivity and efficiency, 60 s of the accumulation was used for quantification of TMZ in this work.

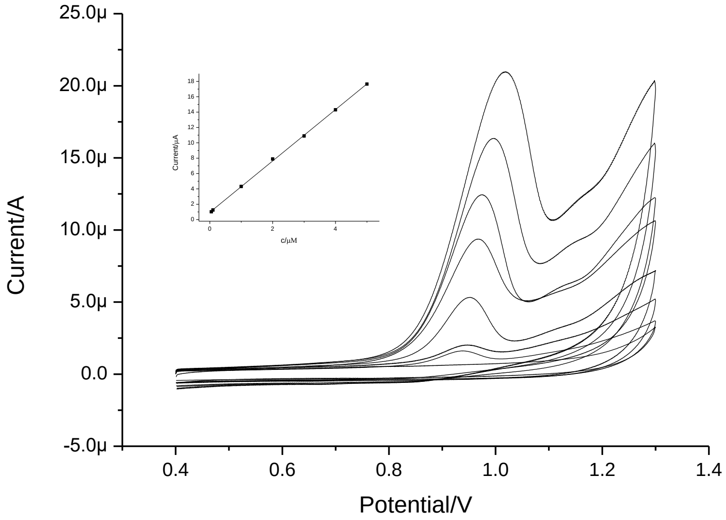


Figure 10. Cyclic voltammograms of TMZ of different concentrations at PACPE at pH = 4. Curves from the bottom to the top: 0, 5×10^{-7}, 1.0×10^{-6}, 1×10^{-5}, 2×10^{-5}, 3×10^{-5}, 4×10^{-5}, 5×10^{-5}M respectively. The insert is a linear plot between the peak current and the concentration of TMZ. Accumulation time: 60 s, scan rate: 0.1Vs^{-1}.

3.7. Electrochemical Detection of TMZ in Real Samples

In optimized condition, cyclic voltammetry was used to find the quantitative relationship between the current response and different concentrations of TMZ. Being accumulated at open circuit in pH=4 buffer solution for 60 s, voltammograms were recorded with the potential varied from 0.4 to 1.3 V (Fig. 10). The peak current increased linearly with the TZM concentration from 5.0×10^{-7} to 5.0×10^{-5} M (inset of Fig. 10), which can be expressed as I_p /μA=0.334c/μM+0.952 (r=0.998 for n=8) with a detection limit about 1.5×10^{-7} M (S/N=3). The reproducibility of the PACPE was examined by measuring 1.0×10^{-5} M TMZ solution repeatedly for 7 times with the same electrode, and the relative standard deviation of about 4.7% indicates excellent reproducibility.

Interferences of some foreign substances on the detection of 2.0×10^{-5} M TMZ were examined. At pH=4 the electrode surface , as discussed above, is negatively charged like a Nafion film, which can reduce the interference of some negatively charged organic molecules and improve the selectivity of the electrode. It was found that at least 50-fold concentrations of vitamin B_1, vitamin B_6, vitamin A, glucose, lactose, ascorbic acid, uric acid, 100-fold of K^+, Na^+, NH_4^+, Ca^{2+}, Fe^{2+}, Cu^{2+}, 200-fold of Cl^-, NO_3^- , CO_3^{2-} and SO_4^{2-}did not interfere the current response of TMZ oxidation at PACPE (signal changes below 5%), suggesting that the proposed method was suitable for TMZ quantification in real samples.

The feasibility of the proposed method in analysis of pharmaceutical formulations was verified by recovery test. Certain amount of standard TMZ solution was pipetted into the sample solution of known concentration, then the response of peak current before and after adding standard solution was measured, and the recovery was calculated. Data of the TMZ detection in tablets were listed in table 1 and the results were consistent with the label. The experimental data of recovery range from 96.44% to 99.04%, suggesting that this new sensor for TMZ is accurate and feasible.

Table 1. Determination of TMZ in tablets

original found[a] （$\times 10^{-5}$ M）	standard added （$\times 10^{-5}$ M）	total found[a] （$\times 10^{-5}$M）	R.S.D (%)	Recovery (%)
1.04			3.3	
	1.00	2.17	4.7	106.4
	2.00	2.99	4.2	98.5
	3.00	3.80	3.6	94.1

[a] average value of three parallel measurements

4. CONCLUSION

The reaction mechanism of TMZ at PACPE was studied by cyclic voltammetry in detail, and it was found that the electrostatic attraction between the protonated TMZ and the negatively charged PACPE surface was the dominant factor affecting the adsorption and the peak current. This explanation is different from that reported in the literature. Investigation showed that oxidation of TMZ at PACPE was a two proton/two electron process, and the PACPE can be used to determine TMZ in pharmaceutical formulation. The proposed method has advantages of simplicity, low cost, high sensitivity and a low enough detection limit. The renewal of the PACPE is efficient, which ensures the reproducibility of individual measurements. The proposed method could possibly be adopted for the pharmacokinetic studies, as well as quality control laboratories.

ACKNOWLEDGEMENTS
We thank doctor Deqiang Ji for assistance with the experiments.

CONFLICT OF INTEREST
The authors declared that they have no conflicts of interest exist in the submission of this manuscript.

References

1. Khedr, M. M. Sheha, I. A. Darwish, *J. Chromatogr*. B, 856(2007) 337.
2. Dezsi, A.Csaba , *Am. J. Ther*. 23(2016), e871.
3. J. Li, J. Quan, J. Du, M. Liu, *Spectrochim. Acta, Part* A, 114(2013), 33.
4. M. K. Jeoung, K. S. Kim, C. Kim, N. H. Kim, Y. B. Chung, J. T. Hong, D. C. Moon, J. Liq. *Chromatogr. R. T.*, 28(2005), 1299.
5. F. Hasin, A. Amin, M. I. Hussain, T. Jahan, N. A. Hama, *World J. Pharm. Res*., 6(2017), 275.
6. L. Ding, B. Gong, X. Chu, J. Hu, H. Zheng, *J. Pharm. Biomed. Anal*., 44(2007), 526.
7. L. Fay, G. Michel, P. Goupit, C. Harpey, M. Prost, *J. Chromatogr.* B, 490(1989), 198.
8. T. S. Belal, T. Awad, C. R. Clark, *J. AOAC Int*., 97(2014), 1514.
9. S. O. Thoppil, R. M. Cardoza, P. D. Amin, *J. Pharm. Biomed. Anal*., 25(2001), 15.
10. S. Agrawal, P. Gurjar, B. Katheriya, *Curr. Pharm. Anal*., 13(2019), 243.
11. Y. M. Issa, F. M. Abouattia, F. M. Abdelgawad, S. M. Abdelhamid, *Anal. Lett*., 35(2002), 451.
12. M. M. Ghoneim, P.Y. Khashaba, A.M. Beltagi, *J. Pharm. Biomed. Anal*., 27(2002), 235.
13. M.B. Gelbert', D.J. Curran, *Anal. Chem*., 58(1986), 1028.
14. K. Ravlchandran, R. P. Baldwin, *Anal. Chem*., 55(1983), 1586.

15. J.E. Huo, E.B. Shangguan, Q.M. Li, *Electrochim. Acta*, 89(2013), 600.
16. A.Pahlavan, V. K. Gupta, A. L. Sanati, F. Karimi, M. Yoosefian, M. Ghadami, *Electrochim. Acta*, 123(2014), 456.
17. Bahrami, A. Besharati-Seidani, A. Abbaspour, M. Shamsipur, *Electrochim. Acta*, 118(2014), 92.
18. Svancara, K. Vytras, J. Barek, J. Zima, *Crit. Rev. Anal. Chem*., 31(2001), 311.
19. R. L. McCreery, *Chem. Rev.*, 108(2008): 2646.
20. X. Cai, K. Kalcher, C. Neuhold, Fresenius *J. Anal. Chem*., 348(1994), 660.
21. G. K. Kiema, G. Fitzpatrick, M. T. Mcdermott, *Anal. Chem*., 71(1999), 4306.
22. H. Bai, W.T. Jiang, G.P. Kotchey, W. A. Saidi, B.J. Bythell, J.M. Jarvis, A.G. Marshall, R.A. S. Robinson and A. Star, *J. Phys. Chem*. C, 118 (2014), 10519.
23. X.J. Zhou, Y. Zhang, C. Wang, X.C. Wu, Y.Q. Yang, B. Zheng, H.X. Wu, S.W. Guo and J.Y. Zhang, *ACS Nano*, 6 (2012), 6592.
24. Y. Liu, R. Wang, J. Lang and X. Yan, *Phys. Chem. Chem. Phys*., 17 (2015), 14028.
25. W.Li, Y. Bai, Y. Zhang, M. Sun, R. Cheng, X. Xu, Y. Chen and Y. Mo, *Synth. Met*., 155 (2005), 509.
26. R. C. Engstrom, V. A. Strasser, *Anal. Chem*., 56 (1984), 136.
27. A. L. Beilby, A. Carlsson, *J. Electraanal. Chem*., 248(1988), 283.
28. J. Li, F. Gao, E. Shangguan, Q. Li, *Electrochim. Acta*, 136(2014), 377.
29. L. Ma, Z. Wang, Q. Li, *Analyst*, 137(2012), 432.
30. J. Li, E. Shangguan, D. Guo, F. Gao, Q. Li, X. Z. Yuan, H., *Electrochim. Acta*, 186(2015), 209.
31. S. Fan, L. Ji, G. Mao, X. Sui, H. Wang, Y. Zhu, H. Song, *Int. J. Electrochem. Sci*., 15(2020), 4148.
32. A. J. Bard, L. R. Faulkner, Electrochemical Methods Fundamentals and Applications, John Wiley and Sons, New York, 2001.
33. K.J. Huang, X. Liu, W.Z. Xie and H.X. Yuan, *Colloids Surf.*, B, 64 (2008), 269.
34. L. Fotouhi, M. Alahyari, *Colloids Surf*., B, 81 (2010), 110.
35. A. Kutluay, M. Aslanoglu, *Sens. Actuators*, B, 192 (2014), 720.