

# Oxidative and anti-oxidative effect of anesthesia compounds (ketamine and xylazine) on the rabbit blood sample using electrochemical method

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## ABSTRACT

Ketamine (KN) and xylazine (XZ) are one of the famous anesthetics using for the animal. The electrochemical properties of the KN and XZ compounds in the rabbit blood samples were studied by cyclic voltammetric (CV) technique using glassy carbon electrode (GCE). The study was included different concentrations, pH, scan rates, interferences between them (KN and XZ) and reliability (stability). The oxidation-reduction current peaks was studied of the KN and XZ alone and both (mixed) in the blood medium to determine the effect of these compounds on the blood components.

It was found the oxidation- reduction current peaks for mixed of KN and XZ in blood medium at 0.1 and -0.8 V respectively, and the oxidation peak was enhanced in acidic pH (3), while the reduction peaks was enhanced in alkaline pH (8). Also, the mixing of KN and XZ in blood medium was acted to decrease the oxidation peak and enhance the reduction peak, so it can be said that mixing is best to work in the animal state.

**Keywords:** cyclic voltammetry, ketamine, xylazine, rabbit, blood, redox process

## INTRODUCTION

Initial studies using electrochemical analysis to finding the oxidation – reduction properties have been made for different pharmaceutical materials and their effects on the blood composition [1-5].

Xylazine may be inhaled or the drug will be used as a veterinary anesthetic. The recommended dose varies between species given intravenously, intramuscularly, subcutaneously or orally, either by itself or in combination with other anesthetics, such as ketamine which is a drug primarily used to initiate and maintain anesthesia [6,7]. The chemical structure of xylazine and ketamine illustrated in Figure 1 and 2 respectively.

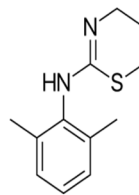


FIGURE 1. Structure of xylazine

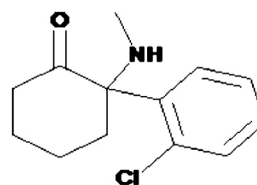


FIGURE 2. Structure of ketamine

The electrochemical behavior of ketamine is studied through the use of cyclic voltammetry (CV) in global buffers with a pH ranging from 2.0-12.0. Kinetic parameters, such as transport coefficients, propagation coefficients, and heterogeneous constant values of the forward rate were evaluated using these techniques. The CV was used to estimate ketamine in selected drug combinations [8].

Analytical electrical sensor ketamine was first reported using [bpy 3] 2+ as an electrochemical sensor probe. The niche of this electrolysis method is the absence of any sample before treatment, as well as being cost-effective and time-consuming. The methodology showed that it is useful for measuring low levels, ng / ml, and ketamine not only in buffered solutions but also in alcoholic beverages [9].

Ketamine is one of the most widely abused drugs in the world and poses a serious threat to human health and social stability; therefore, the ability to monitor the substance accurately in real time is essential. However, there are still many problems facing this goal, such as the generally low concentration of disturbed target molecules in complex samples that are subject to analysis during criminal investigations. In this work, careful and selective detection of ketamine by a molecularly printed electrochemical sensor [10].

The first time, an electrochemical study of xylazine in water and organic media suggested a quantitative method using a glass carbon electrode. A simple and accurate quantitative method was performed using a pulse differential voltage measurement. Under improved experimental conditions, a linear analytic curve was obtained for xylase concentrations ranging from 0.5 to 256 L with, with a maximum detection of 120 nmol/l. The proposed method for determining xylazine was applied in pharmaceutical preparations [11].

Electrochemical techniques were used to estimate xylazine hydrochloric acid (XLZ) in bulk powder, drug manufacturing and human serum. Electrical oxidation of XLZ in multilayer carbon nanotubes (MWCNT), methyl fluorophosphates ion crystal 1-n-butylinidium (BMH) and sodium dodecyl sulfate (SDS) MWCNT-BMH-SDS with pH = 7.0, studied in many of different dielectric structures and pH values. Effective experiments

and parameters were improved to adhere to the evaluation of XLZ, and detection limit was observed as 4.80 nm. The accuracy and accuracy of the method recognized was tested by retrieval studies with good frequency and reproducibility of the estimated method. The drop method was successfully practiced in the form of a dose and an oblique serum [12,13].

In the current research, the effect of KN and XZ compounds on the rabbit blood samples by electrochemical analysis using cyclic voltammetric method to determine the redox current peaks of the Anesthesia Compounds (Ketamine and Xylazine).

## EXPERIMENTAL

### Materials

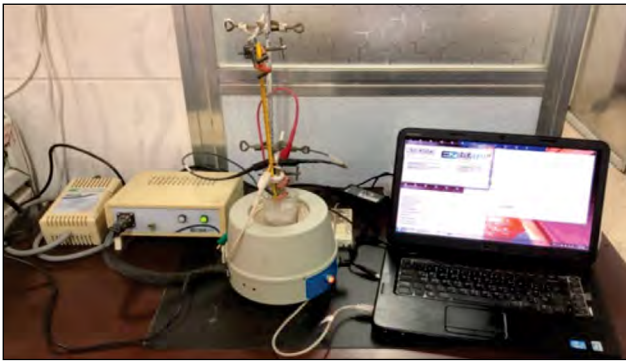
Anesthesia materials used to anesthetize animals such as ketamine 10% from Alfasan company (Holland), xylazine 2% from Alfasan (Holland), blood samples received from rabbit type (*Oryctolagus cuniculus*), Normal saline (0.9% NaCl W/V) from Alcon Parenterals (India) Ltd, 0.1 M HCl and 0.1 M NaOH solutions.

### METHOD

Cyclic voltammetry (CV) device contain a potential station type EZstat (Potentiostat / Glvanostat) from NuVant Systems Company (USA). The cell of CV made from quartz size of 15 ml within the electrolyte solution (blood samples) is placed inside the blood of the working glass-carbon electrode (GCE), the reference electrode type silver/silver chloride and an auxiliary electrode as platinum wire. All three electrodes immersed in the blood sample of the CV cell, and these three electrodes connected to the potentiostat and with personal computer to determine the voltammogram of the results as shown in Figure 3 [14].

## RESULTS AND DISCUSSION

In this study, a cyclic voltammetric technique was used to determine the oxidative and reductive peaks of each of ketamine and xylazine compositions as well as both in rabbit blood samples. The study included different concentrations, scan rates, pH, and reliability (stability).



**FIGURE 3.** Cyclic voltammetry set up

### Calibration study

From the results the effect of using different concentrations of both ketamine (KN) and xylazine (XZ) as well as both of them was studied in the rabbit blood samples to identify the reaction of oxidation – reduction current peaks.

### Effect of the ketamine compound on the blood medium

Figure 4 shows the cyclic voltammogram of ketamine 10% at different concentrations in rabbit blood medium which indicate the oxidation – reduction current peaks at potential of 1 and -0.8 V respectively.

Through the relationship between the electrical current and the concentrations of ketamine in the rabbit blood sample, Figure 5 showed that the effect of the concentration of ketamine in the blood

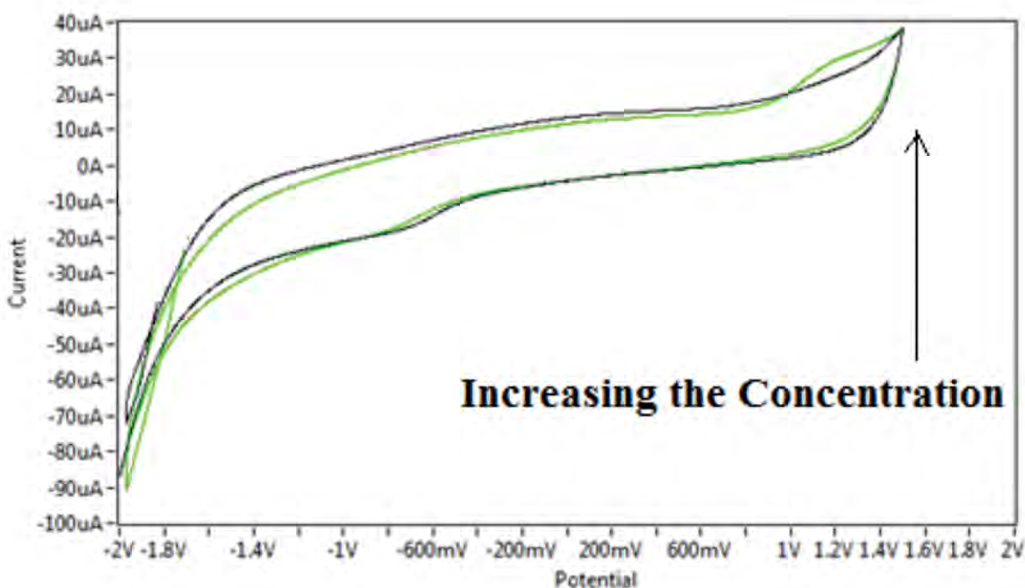
on the oxidation peak in the cyclic voltammogram which illustrated a leaner line by equation  $y = 39x + 25.68$ , with good sensitivity  $R^2 = 0.8973$  [15]. The oxidation current peak of ketamine compound in the blood sample was increased, also the same behavior in the reduction peak has increased to the different concentrations of the ketamine according to the equation:  $y = 31.721x + 19.894$ , with good sensitivity  $R^2 = 0.8864$  as shown in Figure 6 [16].

Effect the mix of ketamine and xylazine on the blood medium

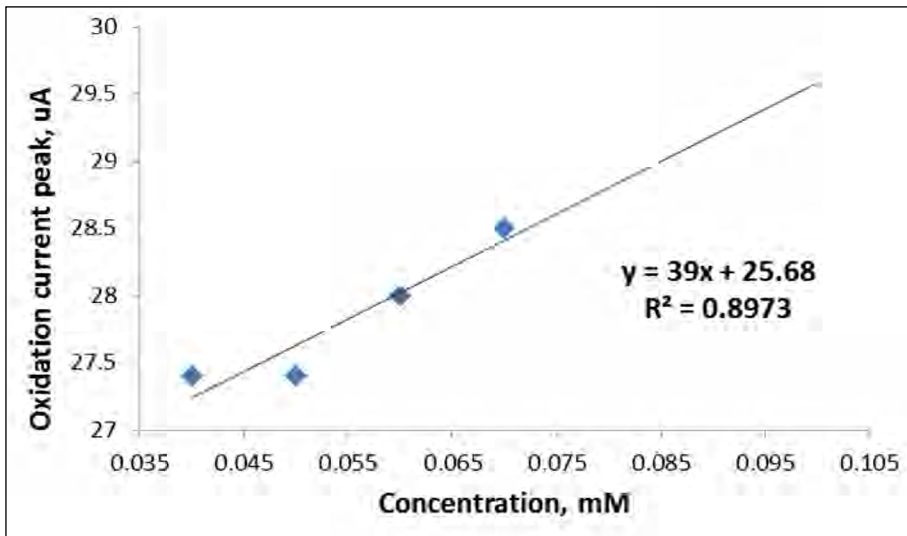
New phenomena has been found for the effect of the mix of ketamine and xylazine on the blood components at different concentrations as shown in Figure 7 which illustrated the cyclic voltammogram of the oxidation current peak was decreased with the relationship in Figure 8 by equation:  $y = -769.09x + 32.42$  with good sensitivity of  $R^2 = 0.8667$ , while the reduction peak was enhanced with relationship of Figure 9 by equation of  $y = 120.3x + 20.767$  with good sensitivity  $R^2 = 0.8233$ , so the mix of KN and XZ acts as anti-oxidative anesthesia in blood medium [17].

### Effect different scan rates

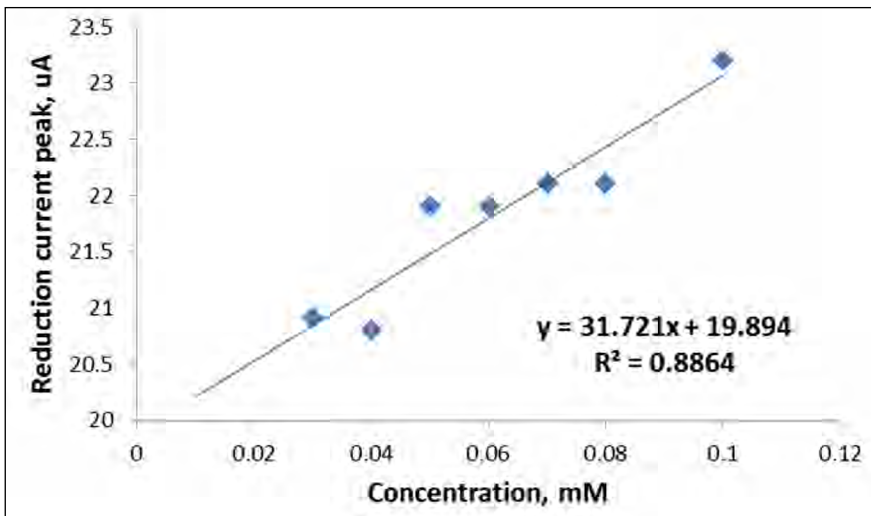
Figure 10 illustrated the cyclic voltammogram of mix of XZ and KN in blood sample at different scan rates which enhanced the redox peaks against to increasing the scan rates. Figure 11 shows the current of oxidation-reduction peaks linearly in-



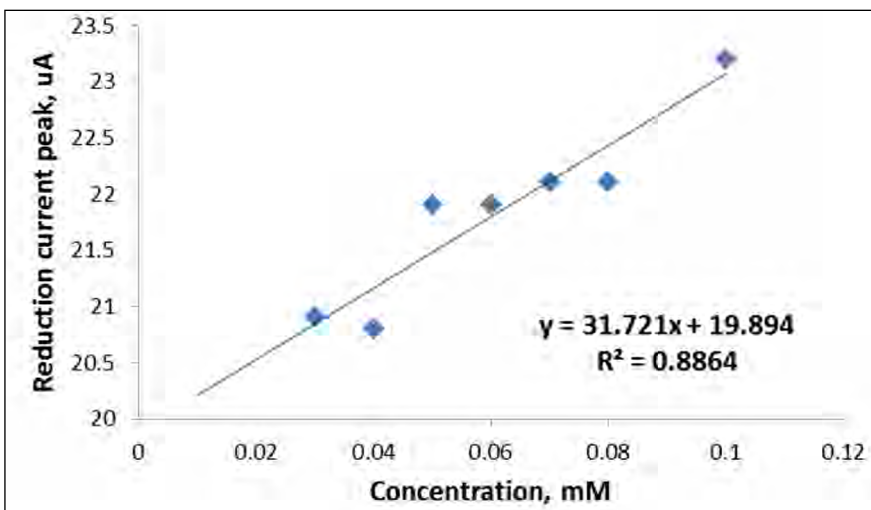
**FIGURE 4.** Cyclic voltammogram of different concentrations of ketamine 10% in blood medium using GCE versus Ag/AgCl reference electrode at scan rate of 0.1 Vsec<sup>-1</sup>



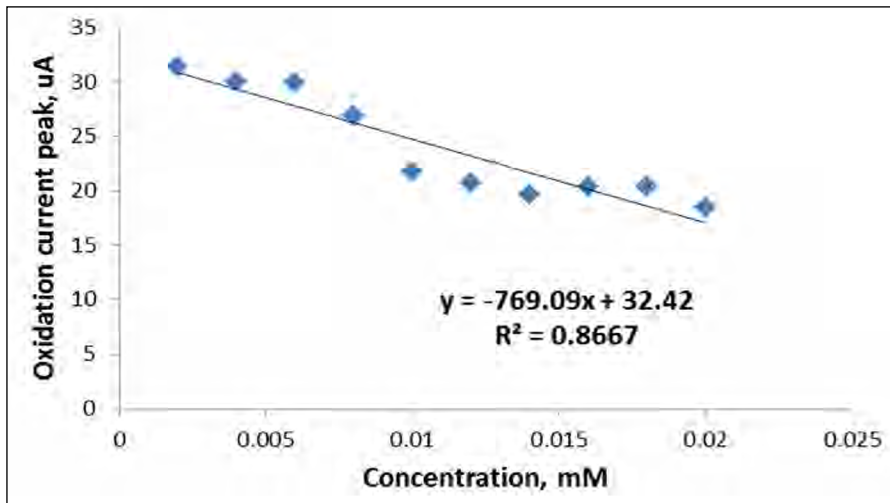
**FIGURE 5.** Relationship between oxidation current peak and different concentration ketamine 10% in rabbit blood sample



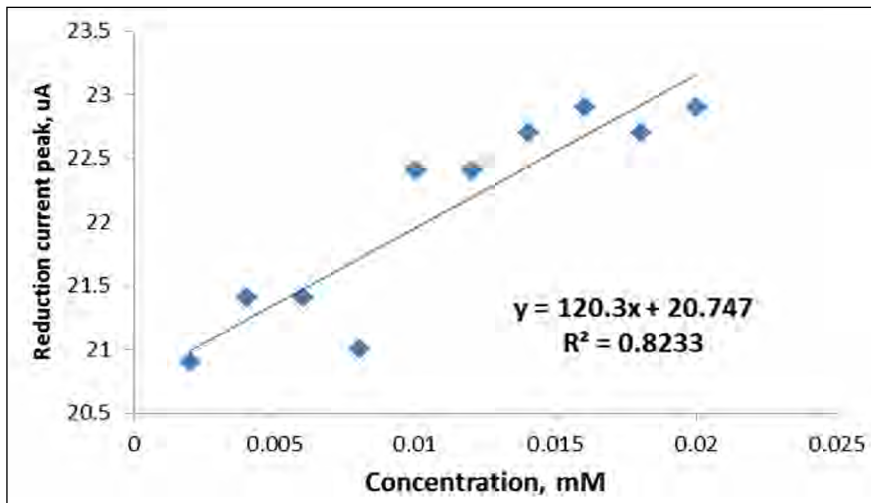
**FIGURE 6.** Relationship between reduction current peak and different concentration of ketamine 10% in rabbit blood sample



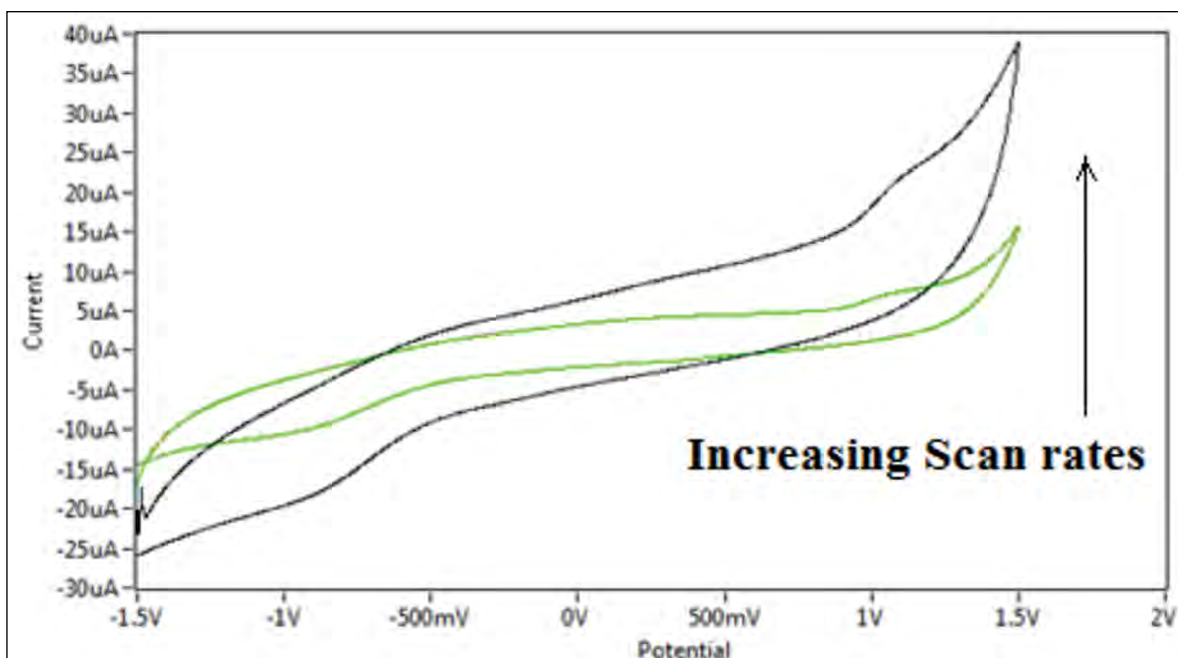
**FIGURE 7.** Cyclic voltammogram of different concentrations of Xylazine 2% in 0.1 mM ketamine 10% in blood medium using GCE versus Ag/AgCl reference electrode at scan rate of 0.1 Vsec-1



**FIGURE 8.** Relationship between oxidation current peak and different concentration of mix of ketamine 10% and xylazine 2% in rabbit blood sample

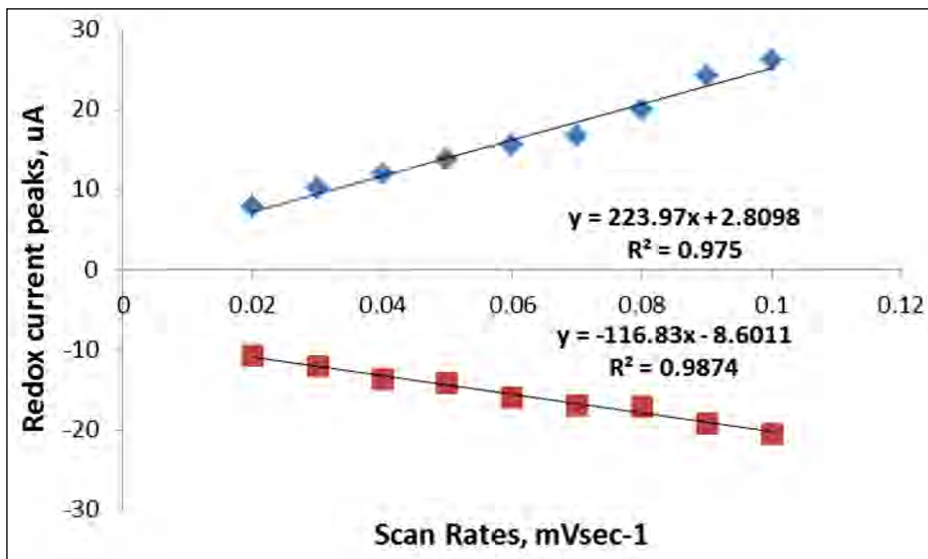


**FIGURE 9.** Relationship between reduction current peak and different concentration of mix of ketamine 10% and xylazine 2% in rabbit blood sample

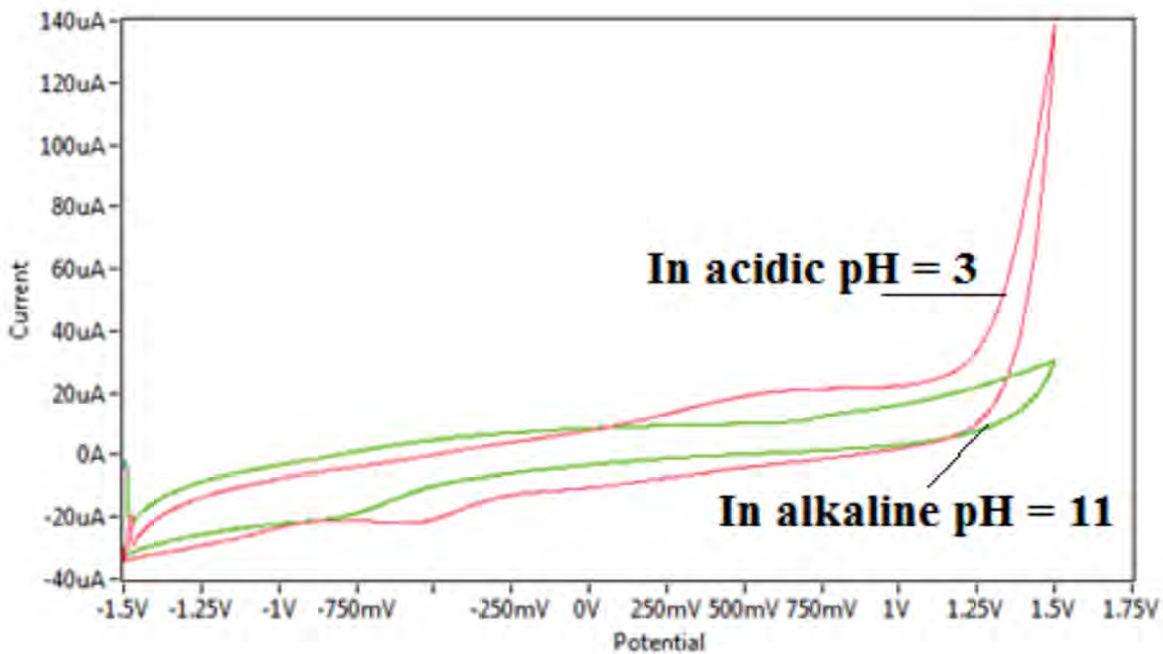


**FIGURE 10.** Cyclic voltammogram of Xylazine 2% and ketamine 10% in blood medium using GCE versus Ag/AgCl reference electrode at different scan rate of 0.01 - 0.1 Vsec<sup>-1</sup>





**FIGURE 11.** Relationship between redox current peaks and different scan rates (0.01-0.1 mVsec<sup>-1</sup>) of mix of Xylazine 2% and ketamine 10% in rabbit blood medium



**FIGURE 12.** Cyclic voltammogram of Xylazine 2% and ketamine 10% in blood medium at different pH using GCE versus Ag/AgCl reference electrode at scan rate 0.1 Vsec<sup>-1</sup>

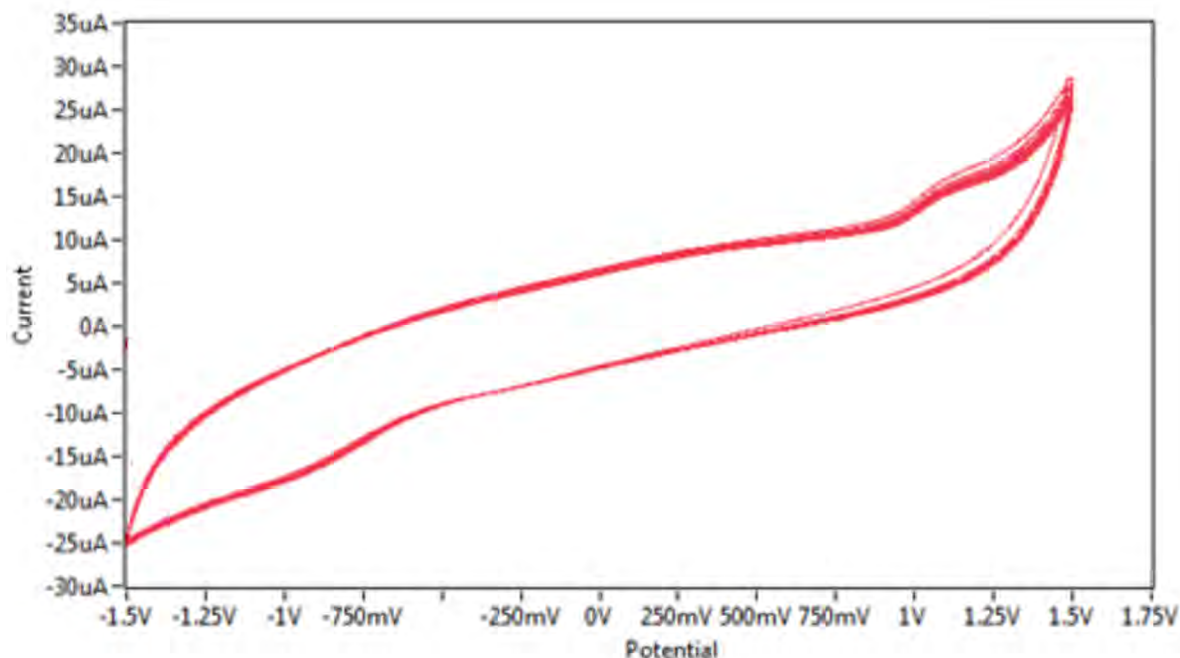
creases  $I_{pa} = 223.97 SR + 2.8098$  with good sensitivity  $R^2 = 0.975$  and  $I_{pc} = -116.83 SR - 8.6011$  with good sensitivity of  $R^2 = 0.9874$ , respectively. The electrochemical process of the mix of XZ and KN in blood medium on the GCE was suggested as adsorption controls [18].

Also, it can be determined the diffusion coefficient ( $D_f$ ) values of the oxidation - reduction reaction of mix XZ and KN compound in blood medi-

um which depended on the components Randles-Seveik equation [19,20]:

$$I_p = (2.69 \times 10^5) n^{3/2} AC D_f^{1/2} V^{1/2}$$

Where  $I_p$  the current peak,  $n$ ,  $C$  and  $A$  indicate the number of electron moles that transferred in this reaction, charge transferred (Colomb) and the electrode area (cm<sup>2</sup>), respectively. The  $D_f$  and  $V$  indicate the diffusion coefficient (cm<sup>2</sup> / s) and potential, respectively.



**FIGURE 13.** Cyclic voltammogram of Xylazine 2% and ketamine 10% in blood medium at ten times using GCE versus Ag/AgCl reference electrode at scan rate 0.1 Vsec<sup>-1</sup>

When applying Randies equation to calculate the diffusion coefficient of oxidation – reduction reaction of the mix of XZ and KN in blood medium, it was found a good values of oxidation  $D_{fa}$  is  $5.12 \times 10^{-6}$  and of reduction  $D_{fc}$  is  $3.5 \times 10^{-6}$ .

#### Effect different pH (acidic and alkaline)

In this study, each of XZ and KN have different electrochemical properties in different pH medium (acidic and alkaline), also, when used mix of XZ and KN in blood medium have different properties. Figure 12 shows that the oxidation peak of the mix of XZ and KN was disappeared in alkaline pH, while the reduction peak enhanced in this pH, so, the anesthesia (mix XZ and KN) acts as anti-oxidative reagent in alkaline blood medium [21].

#### Study reliability and stability

To obtain the best results in this study, the glassy carbon electrode was used for effect mix of XZ and

KN in the blood medium which has overlapping cyclic voltammogram for ten times as shown in Figure 13.

## CONCLUSIONS

In practical veterinary anesthesia, the better method is using a mix of xylazine (XZ) and ketamine (KN), which can ensure the safety in the blood of the animals. So, the using of electrochemical analysis by cyclic voltammetry a good method for oxidation-reduction peaks of the mix in the blood medium. The mix of anesthesia was found anti-oxidative of the blood medium comparing with using it as a single from the mix. Also, the mix of anesthesia of XZ and KN in the blood medium acts as an anti-oxidative reagent in alkaline pH, while the mix of anesthesia (XZ and KN) act as oxidative stress for the blood components in acidic pH.

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