

# Electrochemical Determination of Uric Acid in Human Urine Using Nickel Hexa-Cyano Ferrate Modified Carbon Paste Electrode

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## Abstract

*Nickel hexa-cyano ferrate modified carbon paste electrode for selective determination of Uric acid was used. The electrochemical behavior of uric acid at the nickel hexa-cyano ferrate modified electrode was investigated by cyclic and differential pulse voltammetric methods. It was found that the anodic peak current increased upon the addition of the modifier in the paste. The electrochemical reaction was investigated in phosphate buffer solution of pH 9 using the present electrode. The observed behavior was found irreversible in nature, involving the transference of two electrons at sensor/ analyte interface. Optimization of the parameters required for both techniques have been made at the best electrode composition of 20% nickel hexa-cyano ferrate, 55% graphite and 25% paraffine oil. The proposed method is simple and rapid with its excellent selectivity and sensitive within the linear range of 2–12  $\mu\text{M}$  uric acid with a detection limit of  $1.8 \times 10^{-7}$  M and correlation coefficient  $R^2 = 0.9976$ . The recovery results of uric acid obtained on the application of standard addition technique in the urine samples, nickel hexa-cyano ferrate modified carbon paste sensor has been successfully utilized for determination of uric acid in human urine.*

**Key words:** Carbon paste electrode, Voltammetry, Modified Electrode, Uric acid, Nickel hexa-cyano ferrate, Human urine.

## 1. Introduction

The development of voltammetric sensors for the determination of uric acid (UA) in human fluid such as urine and serum has received considerable interest in recent years. The electrochemical techniques received much interest, as they are more selective and less time consuming than those based on other colorimetric or spectrophotometric methods. Electrodes, modified with polymers /pretreatment were successfully used for the determination of UA [9]. Although these methods showed good selectivity and sensitivity, but these are mainly based on adsorption phenomena, so these either need pre-concentration of UA before or the electrode surface

renewal after, each measurement. These suffer the interference effects of other electro-active components' and the oxidation requires a high over potential [10]. Electrochemical methods offer an analytical platform which can exhibit a higher selectivity and sensitivity than the other commonly employed methods and have the inherent advantage of lower cost and rapid sensing time. Various methods developed and applied for qualitative and quantitative determination of UA, such as chromatography [7], electrophoresis, mass spectroscopy; colorimetric methods [4] like on and have been reported in literature widely.

Recently electrochemical sensors and biosensors have attracted considerable interests because of their high sensitivity, low instrumentation costs, a ready capacity for miniaturization, and direct electronic readout. Electrochemical detection of UA can provide inexpensive and rapid screening techniques which support conventional techniques for both laboratory level and field-work analysis. Various electrochemical sensors and biosensors, such as ion-exchange membrane coated electrode, chemically modified electrode [5], [12] or enzyme-modified electrode [6], Poly (N,N-dimethylaniline) film-coated GC electrode[15], Glassy carbon electrode coated with paste of multi-walled carbon nanotubes and ionic liquids [3], Cysteine modified glassy carbon electrode [2], and so on, have been developed for the sensitive determination of UA. These analyses are generally performed at centralized laboratories, requiring extensive labor and analytical resources which often result in a lengthy turn-around time. The official method for the determination of uric acid in clinical laboratory is with the use of spectrophotometer. Uric acid is oxidized by uricase to produce allantoin and hydrogen peroxide. The hydrogen peroxide reacts with 4-aminoantipyrine and 3, 5-dichloro-2-hydroxybenzene sulfonate in a reaction catalyzed by peroxidase to produce a colored product. The change in absorbance is directly proportional to the concentration of uric acid in the sample. This paper has presented a modified carbon paste sensor using nickel hexacyano ferrate (NHCF) modifier to carry out the voltammetric study of uric acid.

## **2. Experimental Part**

### **2.1 Reagents and Chemicals:**

The reagents and chemicals used were uric acid (Spain), Graphite powder (BDH, England), paraffin oil (Nice, India), di-sodium hydrogen orthophosphate anhydrous (BDH, England), sodium dihydrogen orthophosphate (Nice,India), NaOH (Scharlau, Spain), HCl (Nice,India), Potassium chloride (Nice,India),Nickel chloride (Nice, India) and potassium hexacyano ferrate III (KiraLight, India) were used in the experiment. All chemicals were of analytical grade. The entire present experimentation was carried out at room temperature using de-ionized water for the preparation of all required solutions.

## 2.2 Apparatus:

The electrochemical experiments were carried out in a three-electrode system containing Ag/AgCl as a reference electrode, platinum wire as a counter electrode and unmodified carbon paste electrode (UCPE) or NiHCF modified carbon paste electrode as working electrode. The experiment and processing of data were made using BAS 50W voltammetric analyzer, which was connected to Dell Pentium personal computer. The pH of the buffer solution was measured with a 353 ATC digital pH meter with combination glass electrode. 1ml Syringe (Plastipak, Spain) and Whatman filter paper were used for the preparation of the working electrode. During the measurements, the solution in the cell was not stirred.

## 2.3 Preparation of Solutions:

Supporting electrolyte of phosphate buffers in different pH ranging from 4-10 was prepared from 0.1M  $\text{NaH}_2\text{PO}_4$  and 0.1M  $\text{Na}_2\text{HPO}_4$  in distilled water. The pH of the solutions was adjusted by adding drops of 0.1M HCl and 0.1M NaOH. Stock solution of uric acid was prepared by dissolving 11.2 ml in 0.1 M Phosphate buffer solution. The required concentration of uric acid solutions were prepared by diluting the stock solution with the supporting electrolyte (PBS).

## 2.4 Preparation of Nickel (II) Hexacyanoferrate (III)

Nickel (II) Hexacyanoferrate (III) was prepared by mixing a 0.25M potassium hexacyanoferrate (III) solution and a 0.5 M Nickel (II) chloride solution with Ni/Fe atomic ratio of 1:2 followed by precipitation. The precipitate was filtered in a whatman filter paper, washed with distilled water several times and dried at room temperature for 4 days.

## 2.5 Working Electrode preparation

Preparation of carbon paste electrode (CPE):

100 mg bare carbon paste was prepared by mixing graphite powder with Paraffin oil. The composition of the paste was 75 % graphite powder and 25 % paraffin oil. The mixture was homogenized with mortar and pestle for 30 minutes, and then the homogenized paste was packed in to the tip of a plastic syringe. A copper wire was inserted from the back side of the syringe to provide electrical contact. Then the surface of the electrode was smoothed against a filter paper. Carbon paste electrode is ready.

Preparation of modified electrode (NiHCFE):

Modified carbon paste was prepared by carefully mixing the dispersed graphite powder with NiHCF at varying ratio and subsequently added to 0.250 g of paraffin oil (25%). The mixture was homogenized with mortar and pestle for 30 minutes. The modified carbon paste was packed into an electrode body, consisting of plastic syringe equipped with copper wire serving as an

electric contact. Appropriate packing was achieved by pressing the electrode surface against a whatman filter paper.

## 2.6 Real sample preparation for analysis

Urine samples were collected from four different volunteers using the technique of 'per 24-hour urine sample and stored in the refrigerator. Five ml of each urine sample were transferred in to separate 100 ml volumetric flask. This volume is completed to the mark using phosphate buffer (pH 9) by shaking thoroughly to dissolve. The differential pulse voltammograms were recorded in the potential range between -200 and +1200 mV Vs Ag/AgCl at a scan rate 60 mV/s. The percentage content or concentration of uric acid in these urine samples was determined from the calibration curve.

## 3. Result and Discussions

### 3.1 Electrochemical properties of NiHCFE

Figure.1 shows the cyclic voltammogram of NiHCF modified carbon paste electrode in phosphate buffer solution (pH 9). According to the cyclic voltammogram of NiHCF/CPE in buffer solution, a redox peak is observed between -200 and 1200 mV (vs. Ag/AgCl). This is the potential range where oxidation/reduction of uric acid takes place.

### 3.2 Electrochemical Behaviors of uric acid on NiHCFE

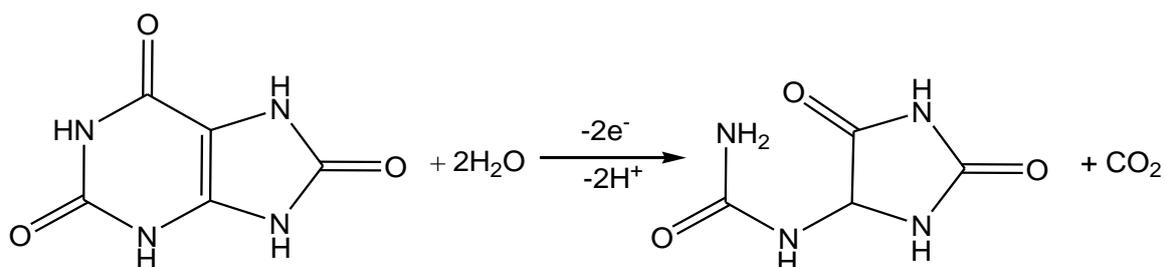
The cyclic voltammograms of a NiHCFE in phosphate buffer at pH 9 in the presence (a) and absence (b) of 2mM uric acid were shown in Figure 2. With the addition of uric acid, the oxidation peak current increased significantly from  $-2.430 \times 10^{-4} \text{A}$  to  $-2.85 \times 10^{-4} \text{A}$ , when compared with that obtained at the modified carbon paste electrode in the absence of uric acid. The peak separation ( $\Delta E$ ) from the cyclic voltammogram of modified electrode in the presence of uric acid was found to be 364 mV, which shows the irreversible characteristics of the electrode process. The electrochemical properties of uric acid at CPE without NiHCF have been examined using cyclic voltammetry, and the result was shown in Figure 3. At unmodified CPE, 2mM Uric acid yields a very low oxidation peak at 545.9 mV (vs. Ag/AgCl) in PBS of pH 9. Cyclic voltammograms of UA at the NiHCF modified electrode and the bare electrode are shown in Figure 6 (curve a) and figure 7, which shows that the current response of UA at the bare electrode is weak,  $i_{pa} = -0.23 \mu\text{A}$  and the current response of UA at the NiHCF/CPE is much better,  $i_{pa} = -285 \mu\text{A}$ . Oxidation peak current of UA at the modified electrode is almost 200 times of the current response at the bare electrode. The peak potential shifts towards less positive potential of 179 mV in comparison with the unmodified CPE. This peak current enhancement and the fall of oxidation over potential indicates that the NiHCF can significantly catalyze the UA oxidation process and the electron transfer rate of UA in NiHCF is much faster, NiHCF/CPE greatly improves the determining sensitivity of uric acid.

### 3.3 Effect of Electrode Composition

The voltammetric response was highly influenced by the composition of the working electrode in the determination of analyte. The effect of the amount of NiHCF in the carbon paste on the voltammetric response of the modified carbon paste electrode was studied by varying the amount of NiHCF between 5% and 25 % as presented in figures 4 & 4A. The peak currents increased with increasing amount of NiHCF up to 20% (w/w). For NiHCF amounts higher than 20% (w/w) the peak currents decreased significantly. This occurs due to a decrease in the graphite content in the paste and, consequent reduction of the conductive electrode area. The best carbon paste composition was found for an electrode composition of 20% (w/w) NiHCF, 55% (w/w) graphite and 25% (w/w) paraffin oil.

### 3.4 Effect of the pH of the supporting electrolyte

The effect of the pH of the supporting electrolyte on the anodic peak current and peak potential of UA at NiHCF modified carbon paste electrode was studied over a large pH range between 4 up to 10 in solution containing 2 mM of uric acid in 0.1 M PBS as supporting electrolyte at a scan rate of 100 mV/s. As seen in Figure 5 both the peak current and peak potential varied with changes in the pH of the solution. Cyclic voltammograms in figures 5 & 5A represent the electrochemical behavior of uric acid at different pH of the buffer in the range 4 to 10. Anodic oxidation peak current first increases with increasing pH and attains maxima at pH 9 thereafter, it decreases as pH value continue to increase. This indicates that the uric acid (UA) oxidation reaction at the interface involves protons in it. The better sensitivity and shape of the voltammogram was obtained at pH 9. Therefore, pH 9 of working buffer solution was chosen. The electrochemical oxidation of uric acid at NiHCF modified carbon paste electrode is generally pH dependent. The graph of anodic peak potential versus pH was plotted and the result shows that the anodic peak potential was shifted linearly towards less positive side with increasing in the pH values. The anodic peak potential of UA was shifted from 597 mV to 362 mV with respect to the pH change from 4 to 10 (figure 5 B). This linearity indicates that equal number of protons and electrons were involved in the electrochemical oxidation of UA at NiHCF/CPE [28]. Based on this finding, the most probable reaction mechanism for the oxidation of UA at NiHCF/CPE is shown below:



### 3.5 Effect of Scan rate on the peak current and peak potential of UA at NiHCFE

The influence of the scan rate on the electrochemical response of UA at modified electrode was investigated by cyclic voltammetry. The effect of scan rate on the oxidation peak current of 2mM uric acid using NiHCF modified electrode at PBS (PH 9) was studied by varying the scan rate from 20-100 mV/s. The resulting voltammogram (figure.6), the graph showing the relation between  $i_p$  versus  $v$  and  $v^{1/2}$  were drawn in figure.6-A& B, respectively.

The oxidation peak potential was observed to shift positively with the increase in scan rate in the range from 20 mV/s to 100 mV/s. The oxidation peak current increased linearly as the scan rates. The linear equation is:

$i_{pa} (A)=108.4+1.97v (mV/s)$  ( $R^2=0.9982$ );and the oxidation peak current increased linearly as the square root of the scan rate, $v^{1/2}$ ,ranges from 4 to 10 ( $R^2=0.9987$ ). These results indicate that the oxidation of UA at NiHCF modified electrode is a diffusion-controlled process, which is the typical characteristic of irreversible reactions. A scan rate 100 mV/s was chosen for the further studies.

### 3.6 Determination of Kinetic Parameters

The electron transfer coefficient ( $\alpha$ ) can be calculated from the slope of the resulted curve of  $E_p$  vs.  $\log v$  using equation:

$$E_{pa} = K + \frac{2.3RT \log v}{2(1-\alpha)n_a F} \quad (1)$$

Slope =

$$\frac{2.3RT}{2(1-\alpha)n_a F} \quad (2)$$

Where  $\alpha$  is transfer coefficient,  $n_a$  is the number of electrons involved in the rate-determining step,  $v$  is scan rate,  $R$  is gas constant,  $E_{pa}$  is peak potential. Based on Figure 7 and Eqn. (2), the value of transfer coefficient ( $\alpha$ ) was calculated as,

$$0.121 = \left( \frac{2.3RT}{2(1-\alpha)n_a F} \right) \quad (3)$$

The value of transfer coefficient ( $\alpha$ ) from this calculation is 0.756. Higher value of transfer coefficient ( $\alpha$ ) indicates deviation from reversible system. By calculating  $\alpha$  from the slope of  $E_{pa}$  vs.  $\log v$  curve,  $k$  can be obtained from equation (4).

$$K = E^\circ + \frac{RT}{(1-\alpha)n_a} \times \left( 0.78 + \frac{2.3}{2} \log \left( \frac{(1-\alpha)n_a F D}{(K_{(s,h)})^2 RT} \right) \right) \quad (4)$$

Where  $\alpha$  is transfer coefficient,  $n_\alpha$  is the number of electrons involved in the rate-determining step,  $E^\circ$  is formal electrode potential  $E^\circ = (E_{pa} + E_{pc})/2 = 0.355$  V;  $k$  is heterogeneous electron transfer rate constant;  $D$  is diffusion coefficient. Based on Figure 7 and Eqn. (2), the value of  $\alpha$  was calculated as 0.756; and using the equation:

$i_p = (2.99 \times 10^5) n (\alpha n_\alpha)^{1/2} A C D^{1/2} v^{1/2}$ , and  $D = 2.79 \times 10^{-3} \text{ cm}^2 \text{ s}^{-1}$ ,  $A = 0.701 \text{ cm}^2$ , and  $n = 2$ . The experimental intercept of Eq. (1),  $K$  was obtained as  $0.342 \text{ cm}^{-1}$ . By substituting the above values in Eq. (4), we found that the heterogeneous electron transfer rate constant  $k_{s,h} = 4.29 \times 10^{-4} \text{ cm s}^{-1}$ .

### 3.7 Optimization of Differential Pulse Voltammetric Conditions

The modified carbon paste electrode gave larger differential voltammetric peak compared to unmodified carbon paste electrode, as shown in the figure.8. Besides the increments' of peak current, the oxidation peak potential also shifts towards less positive value indicating that the NiHCF modified CPE accelerates the electron transfer reaction at the electrode surface. This was confirmed earlier by cyclic voltammetry investigation part. Hence, NiHCFE was further systematically studied by differential pulse voltammetry for the determination of uric acid in the potential range from -200 to 1200 mV.

Effect of Scan rate:

As shown below in Figure 9 the differential pulse voltammograms of 1mM uric acid at the NiHCFE was run at different scan rates starting from 10 to 60 mv/s at 0.1 M phosphate buffer solution of pH 9. The peak current increased with increasing of scan rate up to 60 mV/s but it decreased afterwards. Therefore, a scan rate 60 mV/s was chosen for subsequent experiments.

Effect of Pulse Amplitude:

The peak current increases with increasing magnitude of the differential pulse amplitude within the studied range i.e. between 60–240 mV as shown in the figures 10&10A. A well resolved and sharp peaked voltamogram obtained at 240 mV amplitude. Therefore, pulse amplitude of 240 mV is chosen for subsequent experiments.

### 3.8 Effect of Concentration and Detection Limits

Based upon the optimum conditions, the effect of varying uric acid concentration on the differential pulse voltammetric peak current response of uric acid was studied at NiHCFE. The plot of differential pulse voltammetric peak current versus concentrations of uric acid has been found to be linear in the range of  $2 \times 10^{-6}$  M to  $12 \times 10^{-6}$  M with a correlation coefficient of  $R^2 = 0.9976$  ( $n = 6$ ) and a standard deviation ( $\delta$ ) of 0.42367 (figure 11A). The linear regression equation obtained as:

$$I_p (\mu\text{A}) = 36.86 + 1.11429C (\mu\text{M})$$

The magnitude of detection limit calculated by using the formula;  $LoD = 3\delta/m$ ; of  $1.14 \times 10^{-6} M$ . In this formula ‘ $\delta$ ’ represent the standard deviation and ‘ $m$ ’ represents the slop of curve between anodic oxidation peak current at each concentration of uric acid and concentration of uric acid in the studied range of it from figure 11A i.e. from the standard calibration curve. The magnitude of detection limit and the working concentration range of this study have been compared with some recent studies reported in literature along with modifier material and the technique employed and is presented in table-1.

Table-1: Comparison between the results of the present study and the studies recently reported in literature.

Electrode	Modifier used	Method	Linear range (molL <sup>-1</sup> )	Detection limit (molL <sup>-1</sup> )
CPE	Fe <sup>3+</sup> doped zeolite	SWV	0.3-700 $\mu$	$0.8 \times 10^{-7}$
GCE	Carbon-coated nanoparticle	SWV	0.5-20 $\mu$	$1.5 \times 10^{-7}$
CPE	Nafion	CV	0-50 $\mu$	$2.5 \times 10^{-7}$
GCE	Poly(N,N-dimethylaniline)	DPV	1.25-68.75 $\mu$	$1.25 \times 10^{-6}$
GCE	Fe <sup>3+</sup> doped zeolite/graphite	SWV	2-80 $\mu$	$2.32 \times 10^{-7}$
GCE	Graphene	CV	$6.0 \times 10^{-7}$	$2.0 \times 10^{-6}$
CPE	NiHCF	DPV	2-12 $\mu M$	$1.14 \times 10^{-6} M$

### 3.9 Quantitative determination of uric acid

The validity of the proposed modified electrode for the determination of uric acid using differential pulse voltammetry has been proved on quantification of uric acid in the four different urine samples, collected on the basis of ‘per 24-hour urine sample’ without spiking these samples and also on spiking these with two different known concentrations of standard uric acid in these urine samples. The recovery results of uric acid obtained by using DPV technique with present sensor, for all spiked and nonspiked samples have been presented in table-2. The percentage of recovered quantity of uric acid varies from 95.58% to 98.50%. These observations from this table have showed that the proposed method is suitable for determination of uric acid in human urine using NiHCFE and its utilization can be exploited for testing of uric acid in the country side area laboratories as well as in small cities, for the welfare of humanity.

Table 2: Quantity of uric acid in human urine samples, collected per 24-hour urine, spiked with varying concentrations of standard uric acid, using DPV method.

Sample	Quantity of the Uric Acid ( $\mu\text{M}$ )			Recovery (%)
	Spiked UA	Expected UA	<sup>a</sup> Found UA	
U-1	0.0	----	$03.34 \pm 0.49$	----
	4.0	07.34	$07.13 \pm 1.23$	98.50
	9.0	12.34	$12.00 \pm 1.04$	97.23
U-2	0.0	----	$03.17 \pm 1.10$	----
	4.0	07.17	$06.94 \pm 0.89$	96.78
	9.0	12.17	$11.93 \pm 0.94$	98.06
U-3	0.0	----	$03.69 \pm 0.52$	----
	4.0	07.69	$07.37 \pm 0.47$	95.82
	9.0	12.69	$12.33 \pm 1.61$	97.14
U-4	0.0	----	$03.21 \pm 0.23$	----
	4.0	07.21	$06.97 \pm 0.67$	96.67
	9.0	12.21	$11.69 \pm 0.96$	95.91

<sup>a</sup>Mean value  $\pm$  Standard deviation, for five repetitions' in each case.

#### 4. Conclusion

In the present work, we have introduced a new electrochemical sensor based on nickelhexacyanoferrate modified carbon paste electrode. The electrochemical oxidation of uric acid was successfully studied by CV and DPV with this modified electrode. Several voltammetric parameters have been optimized and their influence on the peak current and peak potential have been studied. The voltammogram resulted from the study showed that an irreversible process at electrode/analyte interface occur involving the transference of two electrons per molecule of uric acid. The detection limit has been much improved to allow a sensitive detection of uric acid. The very low detection limit and its high sensitivity suggest that the modified carbon paste electrode can act as a useful electrode material for the development of electrochemical sensor for uric acid. The linear scan rate dependence showed that the system undergoes diffusion controlled electrode process. The anodic transfer coefficient and diffusion coefficient were determined. The proposed electrode was used in determination of uric acid in human urine with satisfactory recovery.

## 5. Acknowledgement

The authors are grateful to Department of Chemistry, College of Natural and Computational Sciences, Mekelle University.

## 6. Figures

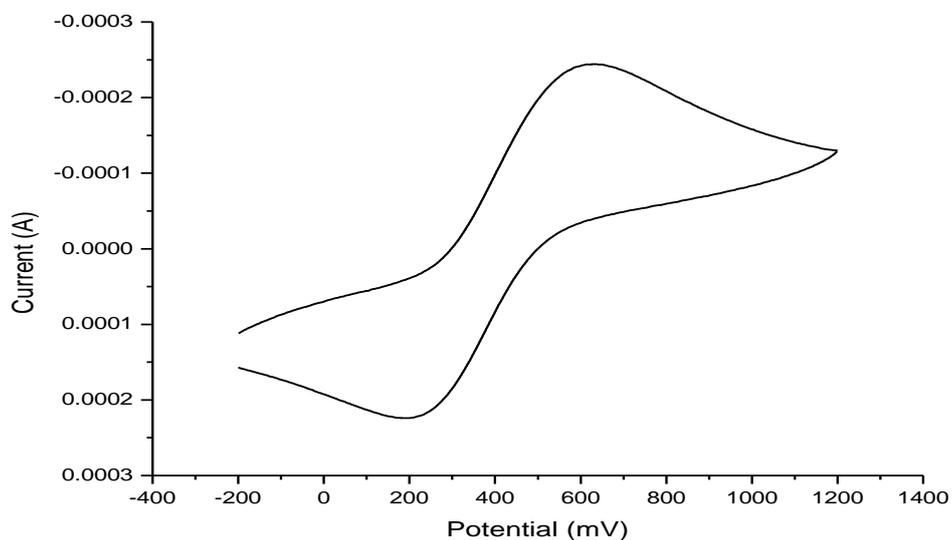


Figure- 1: Cyclic voltammogram of NiHCF/CPE in 0.1M PBS (pH 9) and scan rate 100 mV/s.

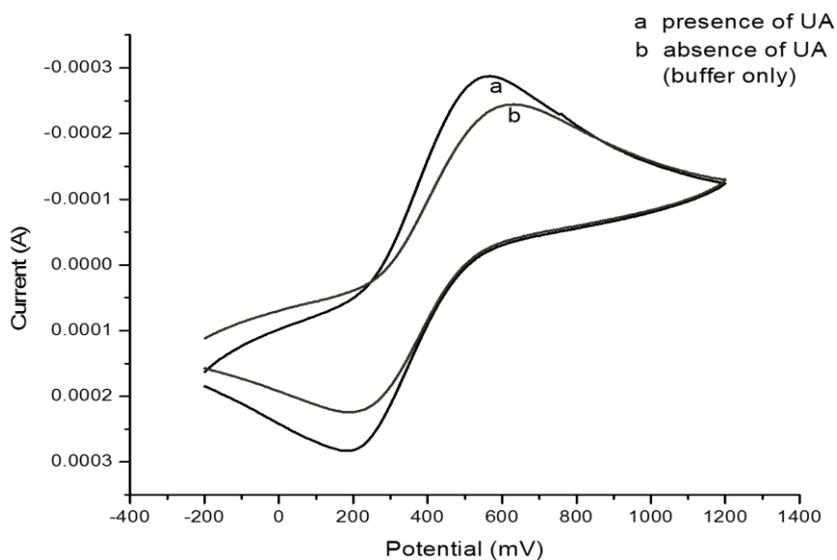


Figure- 2: Typical cyclic voltammograms of NiHCF/CPE in 0.1M PBS (pH 9) at a scan rate 100 mV/s in the presence (a) and absence (b) of 2 mM uric acid.

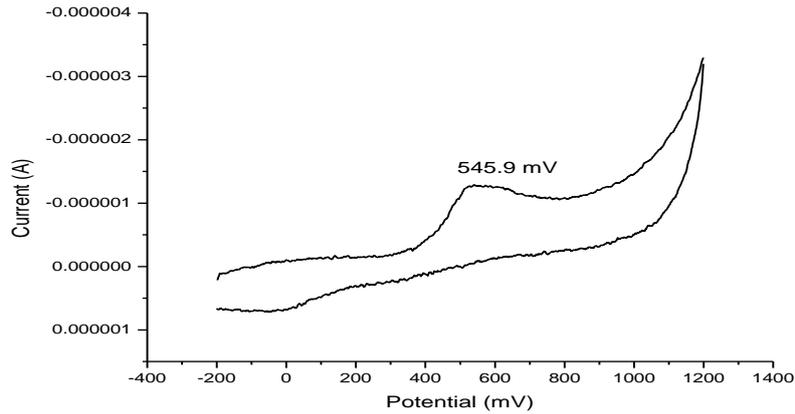


Figure- 3: The electrochemical behavior of 2 mM uric acid at the unmodified carbon paste electrode in 0.1 M phosphate buffer solution of pH 9, at a scan rate 100 mV/s.

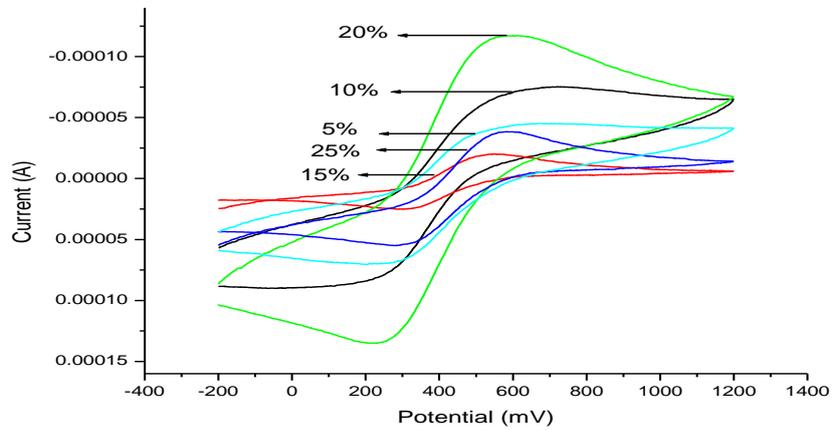


Figure-4: Cyclic voltammograms of different amounts of NiHCF in presence of 2 mM uric acid and 0.1 M phosphate buffer (pH 9) at scan rate 100 mV/s.

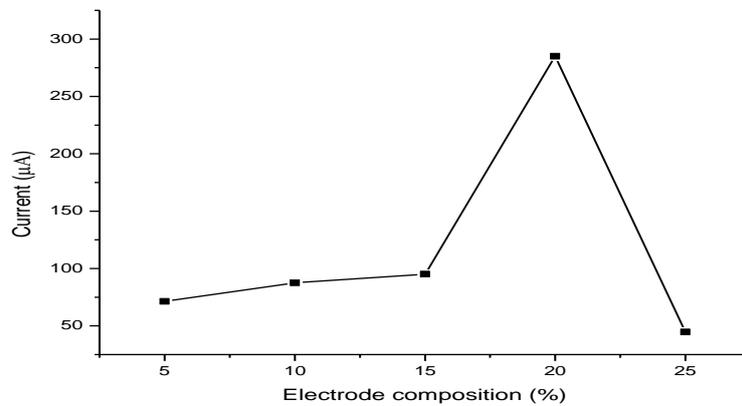


Figure- 4A: Effect of electrode composition on anodic peak current in 2 mM uric acid at 0.1 M PBS of pH 9, ranging from 5 to 25 % NiHCF modifier at a scan rate 100 mV/s.

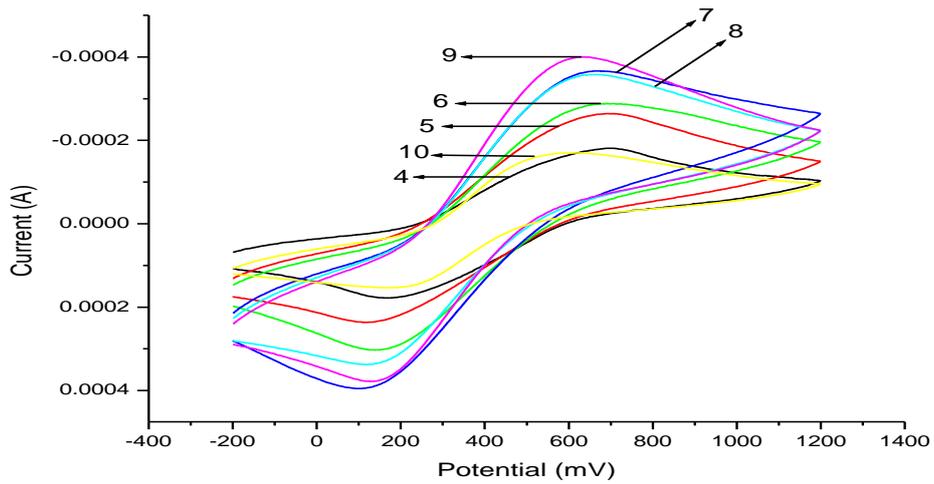


Figure- 5: Cyclic voltammogram of 2mM uric acid at different pH in 0.1 M PBS solution at scan rate 100 mV/s.

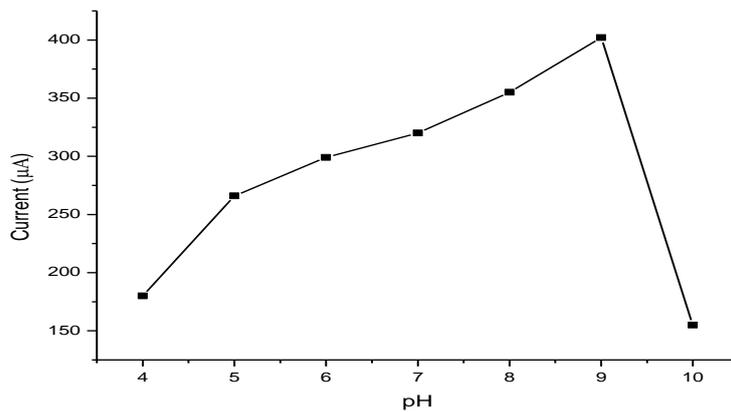


Figure- 5A: Dependence of the peak current for 2 mM UA on the pH of the supporting electrolyte at NiHCF modified carbon paste electrode. Scan rate is 100 mV/s.

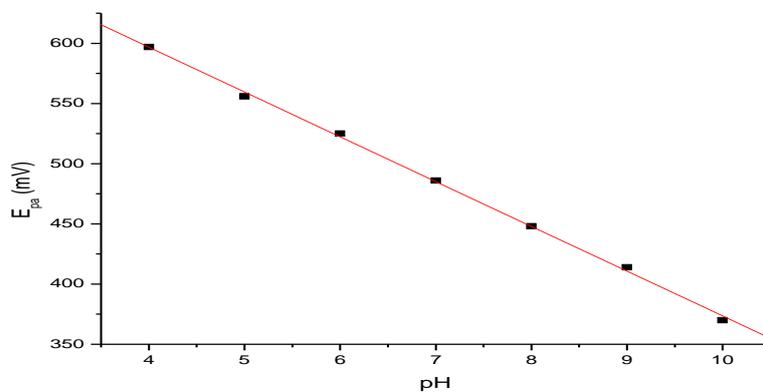


Figure 5B: Plot of peak potential versus the pH of the supporting electrolyte

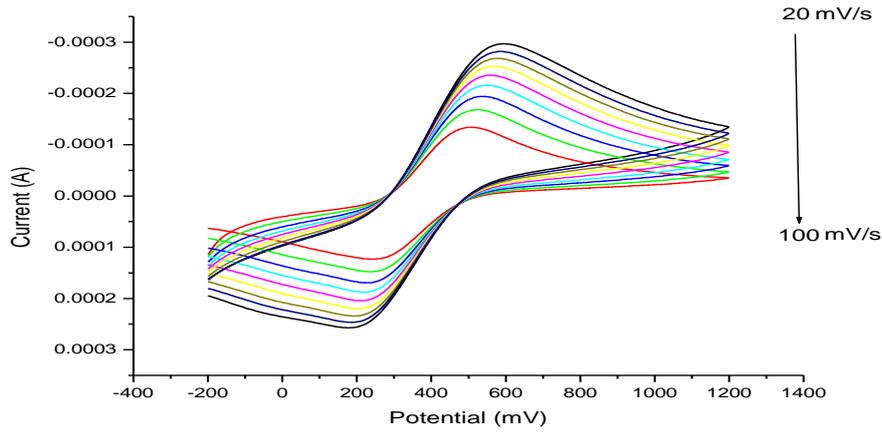


Figure-6: Cyclic voltammograms for 2mM uric acid in 0.1 M PBS (pH 9) at modified electrode at different scan rates (20,30,40,50,60,70,80,90 and 100 mV/s).

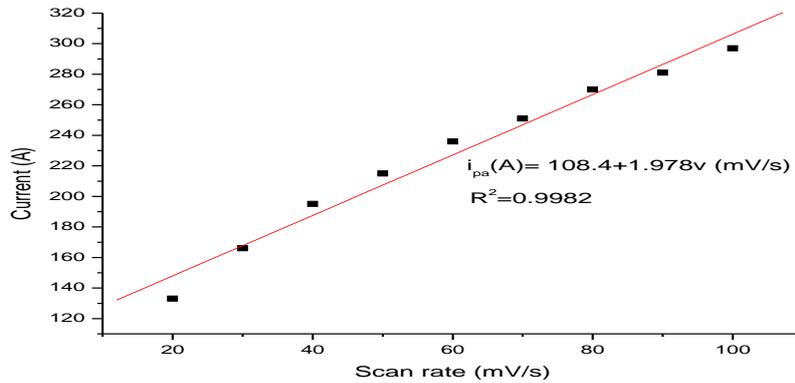


Figure-6A: Effect of variation of scan rate on the anodic peak current of 2 mM uric acid in 0.1 M PBS (pH9), Scan Rate: 20-100 mV/s.

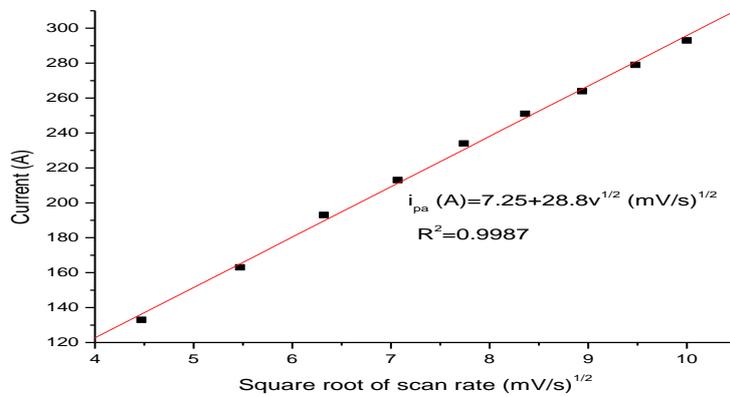


Figure-6B: Effect of square root of scan rate on cyclic voltammetric peak currents of 1mM uric acid in 0.1 M PBS of pH 9.

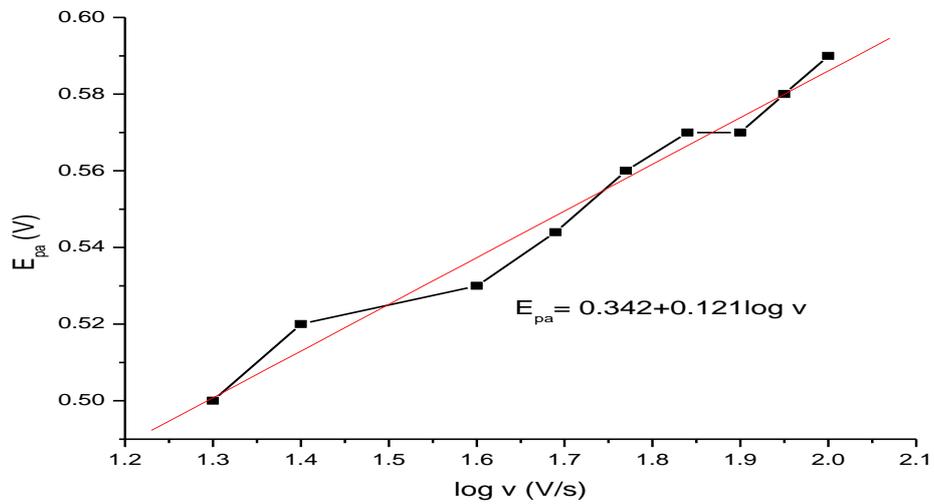


Figure 7: Plot of E<sub>pa</sub> versus log v

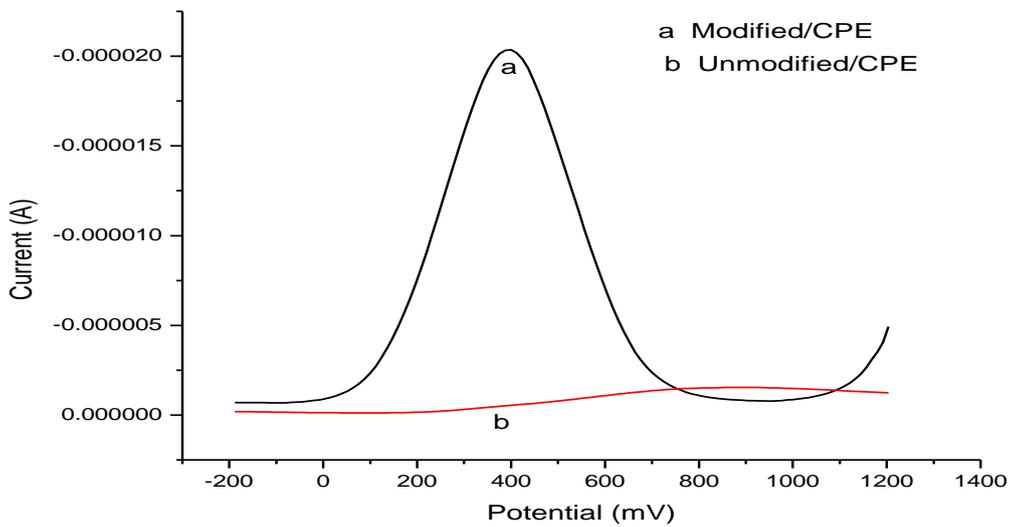


Figure-8: Differential pulse voltammetry of 1 mM uric acid at (a) NiHCF/CPE and (b) unmodified carbon paste electrode and in 0.1 M PBS (pH 9) at a scan rate of 60 mV/s pulse amplitude of 240 mV.

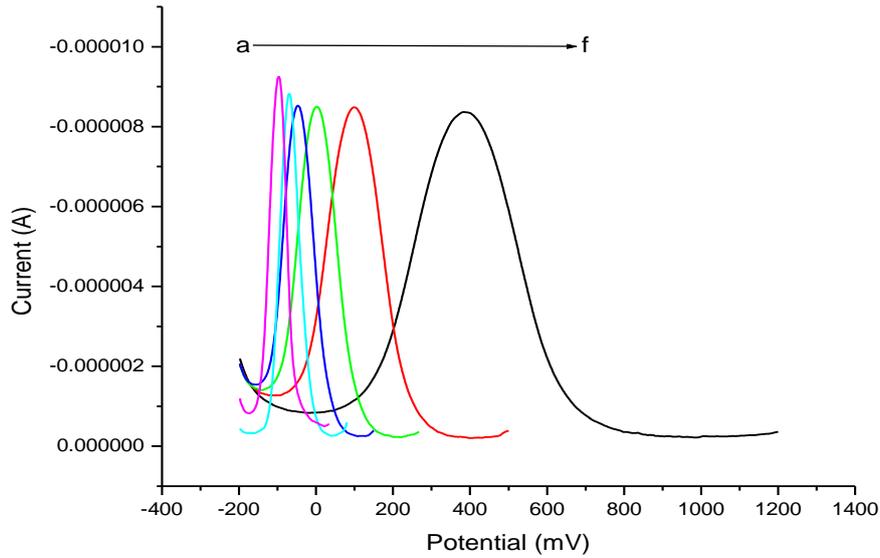


Figure-9: Differential pulse voltammograms of 1 mM uric acid at NiHCF/CPE in 0.1 M PBS of (pH 9) at a scan rates of: (a) 60; (b) 50; (c) 40; (d) 30; (e) 20 and (f) 10 mV/s using pulse amplitude of 240 mV.

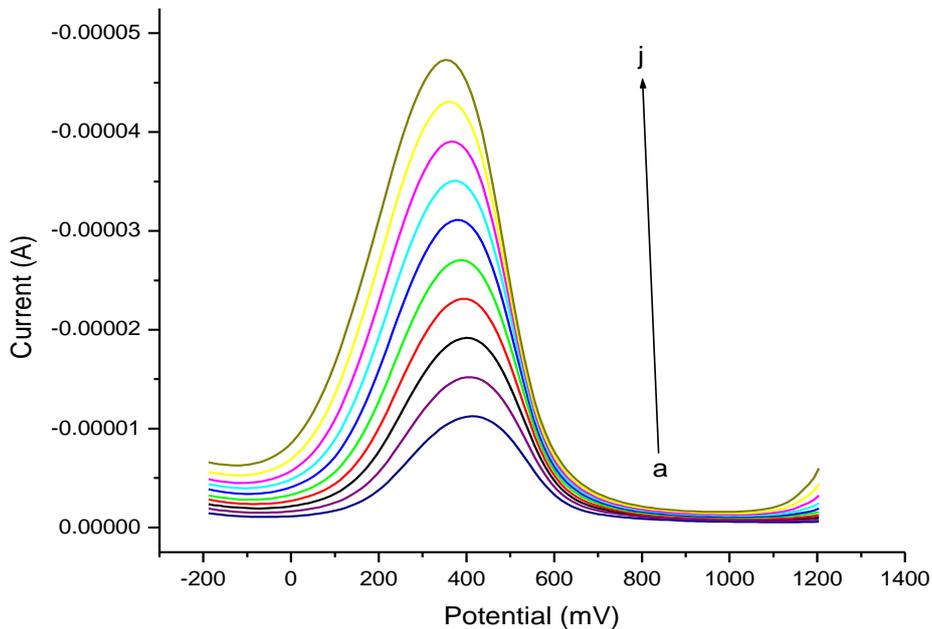


Figure-10 : Differential pulse voltammogram of 1 mM uric acid in 0.1 M PBS (pH 9) at NiHCF/CPE at a scan rate of 60 mV/s and different pulse amplitudes of (a) 60; (b)80; (c) 100; (d) 120; (e) 140; (f) 160; (g) 180; (h) 200; (i) 220 and (j) 240 mV.

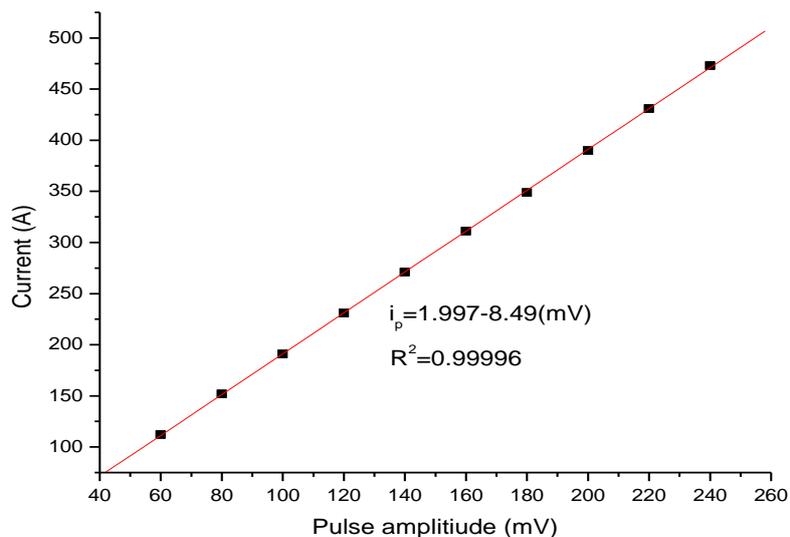


Figure-10 A: Plot of peak currents of 1 mM uric acid versus pulse amplitude

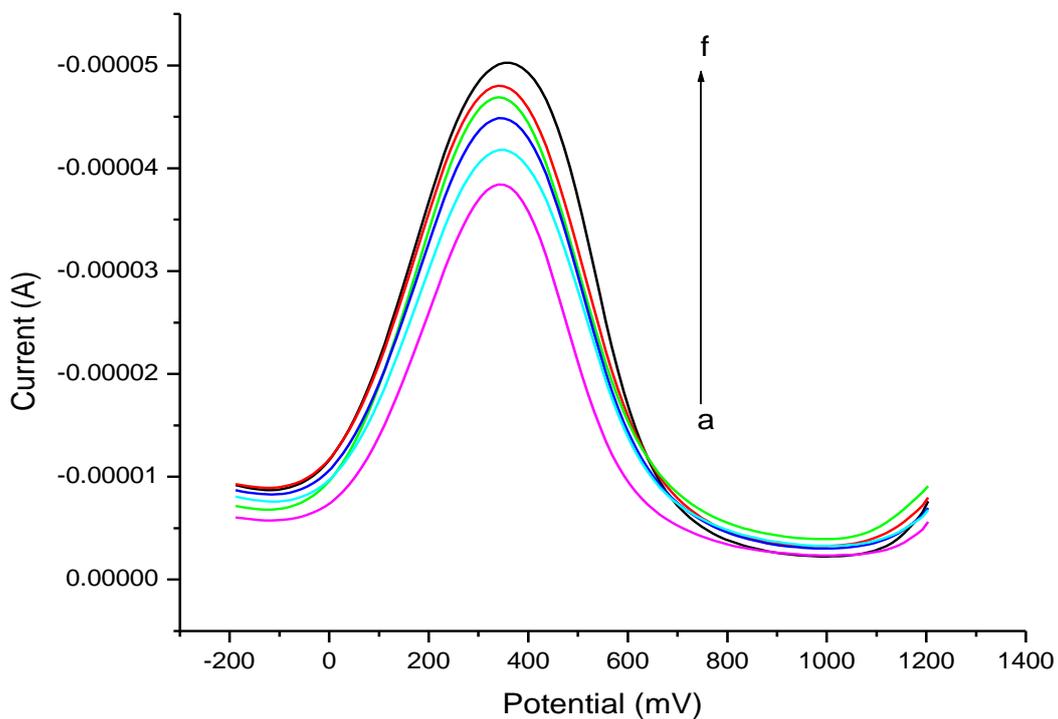


Figure 11 : DPV at different concentrations of uric acid i.e, (a) 2; (b) 4; (c) 6; (d) 8; (e) 10 and (f) 12  $\mu$ M in 0.1 M PBS of pH 9 at scan rate of 60 mV/s and pulse amplitude of 240 mV.

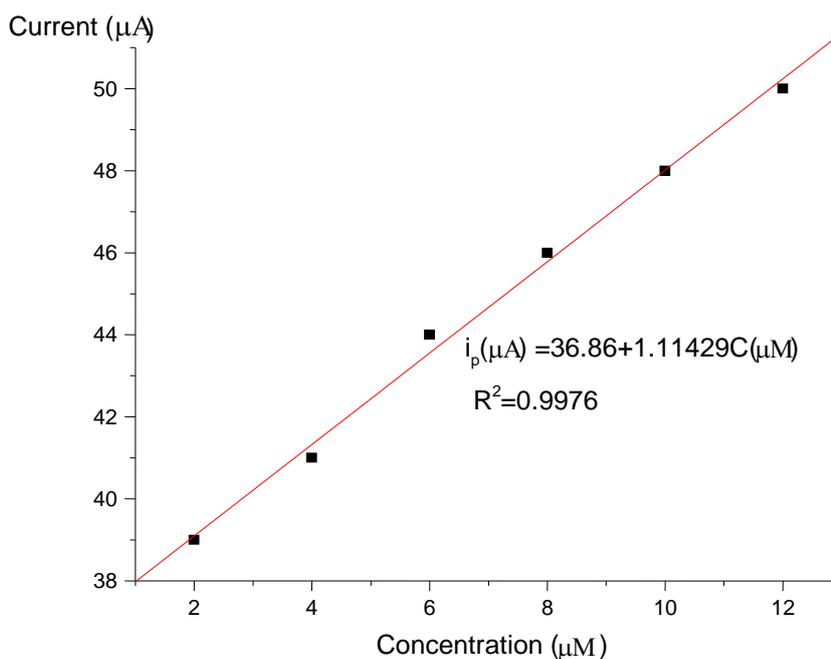


Figure 11 A: Plot of peak current versus concentration.

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