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Voltammetric Behavior of Isoniazid and Its Electroanalytical Determination at Carbon Nanotubes-CTAB Modified Glassy Carbon Electrode

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Abstract- An electrochemical sensor for sensitive determination of isoniazid (INH) using multi-walled carbon nanotubes-cetyltrimethyl ammonium bromide (CTAB) surfactant composite on the surface of a glassy carbon electrode was developed. The peak current increased greatly compared with bare GCE and MWCNT/GCE electrode. The oxidation process was irreversible over pH range studied and exhibited a diffusion controlled behavior. The current response was found to be directly proportional to the concentration of INH in the concentration range of 1.0×10^{-7} M to 1.2×10^{-6} M with a limit of detection (LOD) of 2.14×10^{-9} M. The modified electrode showed good selectivity and reproducibility. The developed analytical method was successfully applied to isoniazid determination in human serum and urine samples. This method can be employed in clinical analysis, quality control and routine determination of drugs in pharmaceutical formulation.
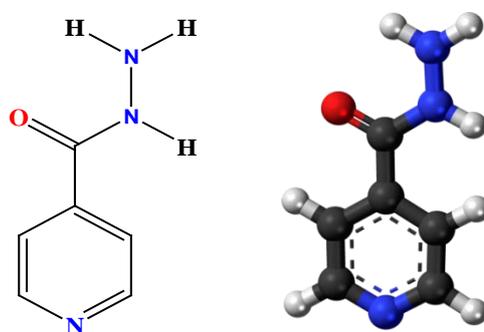
Keywords - Isoniazid, Multi-walled carbon nanotubes, CTAB, Diffusion controlled, Real samples

1. INTRODUCTION

Drug analysis is an important tool for drug quality control. Therefore, the development of simple, sensitive, rapid and reliable method for determination of drugs still continues to be of great importance. Isoniazid (pyridine-4-carboxylic acid hydrazide or isonicotinyl hydrazine

(INH) Scheme 1.) is an important bacteriostatic drug used for the chemotherapy of tuberculosis caused by mycobacteria [1,2]. In the over dosage of INH drug during the metabolism, causes heptatoxicity and sometimes fatal liver damage resulting in death [3]. Therefore it necessitates developing a suitable, selective and specific technique for the determination of INH in clinical chemistry. Owing to its therapeutic importance and widespread use, considerable attention has been paid to develop sensitive and rapid analytical techniques for the determination of INH either in pure form or in pharmaceutical preparations or in biological fluids. The various analytical methods reported for the determination of INH are based on spectrophotometry [4,5], spectrofluorometry [6,7], high performance liquid chromatography [8,9], colourimetric and titrimetric analysis [10,11]. The main problems encountered in using such methods are either the need for derivatization or the need for time-consuming extraction procedures.

Voltammetric methods satisfy many of the requirements for such tasks, particularly owing to their inherent specificity, rapid response, high sensitivity, low cost, simplicity, and relatively short analysis time for the determination of organic molecules, including drugs and related molecules in pharmaceutical dosage forms and biological fluids. Electrochemical techniques using modified electrodes can be considered for the determination of drugs in pharmaceutical laboratories. Determination of isoniazid by using bare electrodes [12,13] and different modified electrodes have been developed [14-16].

Scheme 1. Structural formula of Isoniazid

Multi-walled carbon nanotubes [17,18] (MWCNTs) possess superior electrical conductivity. This is especially most advantageous in the electronic industries. Because of their special physico-chemical properties, MWCNTs are expected to play a major role in numerous applications. In the field of electrochemical sensing, nowadays MWCNTs are used as sensors because of their usefulness in analytical determinations. Surfactants, even in trace quantities, can exert a strong effect on the electrode process. In recent days surfactants have been of tremendous research importance because of their many useful applications in antiseptics, cosmetics, detergents, material fabrication and drug delivery as it forms micelles

with aggregation in aqueous solutions. Successive addition of the cationic surfactant CTAB enhances greatly the anodic peak current which is attributed to the adsorption of surfactant onto electrode surface forming a positively charged hydrophilic film with the polar head group pointing to the bulk of the solution. The CTAB could increase the particle growth rate by increasing the reduction rate of MWCNTs and its uniform dispersion increases surface area. The electrostatic force between CTAB and INH facilitates reaching of INH to the electrode surface faster, and as a consequence, the reaction becomes easier. Other surfactants, anionic Sodium dodecyl sulfate and non-ionic surfactant Triton X-100 did not enhance the peak current significantly. Here we have developed an electrochemical method for the determination of INH using multi-walled carbon nanotube with cationic surfactant, cetyltrimethyl ammonium bromide (CTAB) modified glassy carbon electrode. This proposed method is sensitive and fast response technique with low detection limit and good selectivity and sensitivity.

2. EXPERIMENTAL

2.1. Reagents and chemicals

MWCNTs (N90%, O.D.: 10–15 nm, I.D.: 2–6 nm, length: 0.1–10 μm) and Isoniazid were purchased from Sigma–Aldrich, Bangaluru, Karnataka, India. The supporting electrolyte used for experiments was phosphate buffers prepared from KH_2PO_4, Na_2HPO_4, Na_2PO_4, H_3PO_4 and Cetyltrimethyl ammonium bromide were purchased from SD Fine chemicals, Mumbai, India. pH 3.0–10.2 and were prepared according to the method developed by Christian and Purdy [19]. All solutions were prepared with Millipore water. All other chemicals were of analytical grade.

2.2. Instrumentation

Electrochemical measurements were carried on a CHI 630D electrochemical analyzer (CH Instruments Inc., USA). The voltammetric measurements were obtained in a 10 mL single compartment three-electrode glass cell with Ag/AgCl (3.0 M KCl) as a reference electrode, a platinum wire as auxiliary electrode and modified MWCNT-CTAB/GCE as working electrode. pH of solution was measured with an Elico LI120 pH meter (Elico Ltd., India). All experiments were carried out at an ambient temperature of 25 °C±0.1 °C.

2.3. Preparation of modified electrode

The Glassy carbon electrode was carefully hand-polished with 0.05 μM and 0.3 μM (particle size) alumina slurry on a Buehler polishing cloth, then washed with Millipore water and pretreated with NaOH (10%), HNO_3: H_2O (1:1, v/v) and acetone. Finally the electrodes were rinsed with millipore water. The electrode modification [20] by electro deposition

method was carried out by the solution containing 0.3 g/L MWCNT and 0.2 g/L CTAB, which was sonicated for 1 h. The ultrasonication of MWCNT and CTAB lead to the dispersion of nanotubes, and embedded the surfactants on the surface of carbon nanotubes [21] CTAB is the cationic surfactant which makes the nanotubes positively charged and through simple electro deposition process or casting, they form a thin layer at the electrode surface.

The coated electrodes (GCE-MWCNT/CTAB) were immersed in the bicarbonate solution (0.01 M) for 30 min in order to extract the residual surfactants from the surface of electrode. The solvent was allowed to evaporate at room temperature in the air and modified electrode was characterized by Scanning electron microscopy (SEM), Atomic Force Microscope (AFM) topographical scan and the Fourier transform infrared (FT-IR) spectroscopy which are shown in Fig. 1. SEM and AFM are the most appropriate and accurate characterization tools for investigation of surface morphology structure and shape estimation.
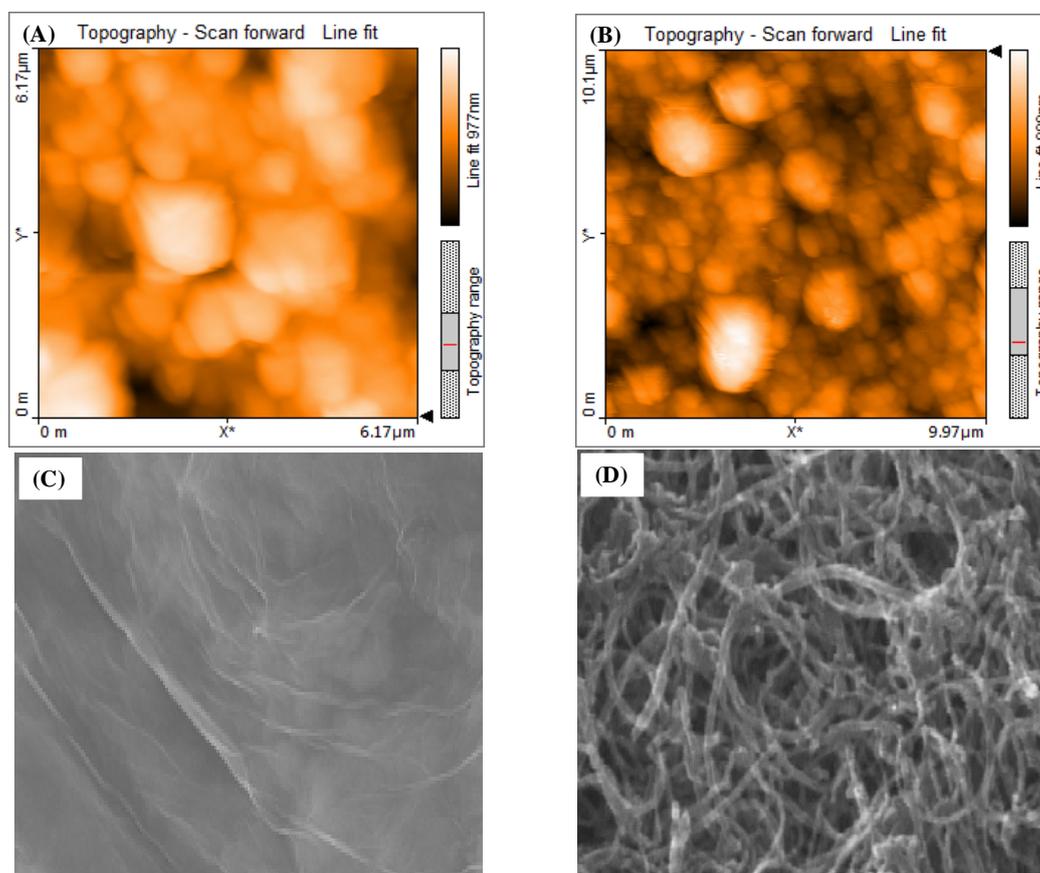

Fig. 1. Modified electrode characterized images of **(A)** AFM topography of MWCNT; **(B)** AFM topography of MWCNT with surfactant CTAB; **(C)** SEM topography of MWCNT; **(D)** SEM topography of MWCNT with surfactant CTAB

SEM image showed before the modification were very smooth and after modification were

rough which increases surface area and facilitated electrode deposition easily. AFM morphology confirms the more adsorption of MWCNT on the glassy carbon electrode in the presence of the CTAB. Therefore modified electrode surface is rough and shows good conductivity. The Fourier transform infrared (FT-IR) spectroscopy of MWCNT and MWCNT-CTAB modified electrodes were measured with a Thermo Nicolet Avatar 360 FT-IR Spectrometer within the range of 500–4000 (cm^{-1}). The addition of cationic surfactant (CTAB) for charging and dispersing the nanotubes affected the peaks that appeared in the FT-IR spectra. The broad peak at 3430 cm^{-1} is attributed to the O-H stretching bond which is due to the water absorption of the carbon material in the atmosphere. The weak peak observed in all samples at 1725 cm^{-1} is related to the C=O bond which is probably due to the impurities of carbon and MWCNTs. The peaks at around 2923 and 2853 cm^{-1} are related to the symmetric alkane stretching of C-H bond, the intensity of which is higher in the MWCNT-CTAB/GCE due to the presence of the surfactant. Also the peaks appearing at around 1468 and 1256 cm^{-1} were attributed to the CH_2 (alkane bending) and C-N amine bond exclusively observed in the MWCNT-CTAB/GCE sample due to the existence of the surfactants.

2.4. Area of the electrode

The electro active area of the MWCNT-CTAB-modified electrode and the bare GCE were obtained by cyclic voltammetry using 1.0 mM $K_3Fe(CN)_6$ as a probe at different scan rates. For a reversible process, the Randles–Sevcik formula has been used [22].

$$I_{pa} = (2.69 \times 10^5)n^{3/2}AD_0^{1/2}C_0v^{1/2} \tag{1}$$

where, I_{pa} refers to the anodic peak current, 'n' is the number of electrons transferred, 'A' is the surface area of the electrode, 'D_0' is diffusion coefficient, 'v' is the scan rate and 'C_0' is the concentration of $K_3Fe(CN)_6$, respectively. For 0.001 M $K_3Fe(CN)_6$ in 0.1 M KCl electrolyte T=298 K, R=8.314 J $K^{-1}mol^{-1}$, F=96,480 C mol^{-1}, n=1, D_0=7.6×10^{-6} cm^2s^{-1} then from the slope of the plot of I_{pa} v/s $v^{1/2}$, the electrode surface area was calculated. In our experiment the slope was 0.645×10^{-4} µA $(V/s)^{-1/2}$ and the area of electrode was calculated to be 0.128 cm^2. The area of the unmodified glassy carbon electrode was calculated to be 0.0397 cm^2. The modified electrode provides active surface area three times higher than the bare glassy carbon electrode.

2.5. Plasma sample preparation

Human blood samples were collected in dry and evacuated tubes (which contained saline and sodium citrate solution) from a healthy volunteer. The samples were handled at room temperature and were centrifuged for 10 min at 1500 rpm for the separation of plasma within 1 h of collection. The samples were then transformed to polypropylene tubes and stored at-20

°C until analysis. The plasma samples, 0.2 mL, were deprotonised with 2 mL of methanol. After vortexing for 15 min, the mixture was then centrifuged for 15 min at 6000 rpm, and supernatants were collected.

The MWCNT-CTAB/GCE was first activated in phosphate buffer pH=4.2 (0.2 M), by cyclic voltammetric sweeps between 0.0 and 1.0 V until a stable cyclic voltammogram was obtained. Then electrodes were transferred into another 10 ml of phosphate buffer solution containing proper amount of isoniazid. After open-circuit accumulation for 100 s with stirring and quiet for 20 s, potential scan was initiated and cyclic voltammograms were recorded between 0.0 and 1.0 V, with a scan rate of 50 mV s^{-1}.

3. RESULTS AND DISCUSSION
3.1. Electrochemical behavior of isoniazid (INH)

The electrochemical behavior of isoniazid at MWCNT-CTAB/GCE was studied by cyclic voltammetry at pH=4.2. The cyclic voltammograms obtained for 1.0 mM isoniazid solution at a scan rate of 50 mVs^{-1} and potential ranges from +0.0 to +1.0 V exhibits a well defined anodic peak at about +0.424 V. The results are shown in Fig. 2. The voltammograms corresponding to the first cycle was generally recorded. GCE, MWCNT/ GCE electrodes show oxidation peak current weaker and wider, sensitivity was poor compared to the MWCNT-CTAB/GCE and exhibits high intensity and sharp oxidation peak which indicate that modified electrode has good catalytic activity for oxidation of INH. On the reverse scan, no corresponding reduction peak was observed indicating that the electrode process of isoniazid is an irreversible one.
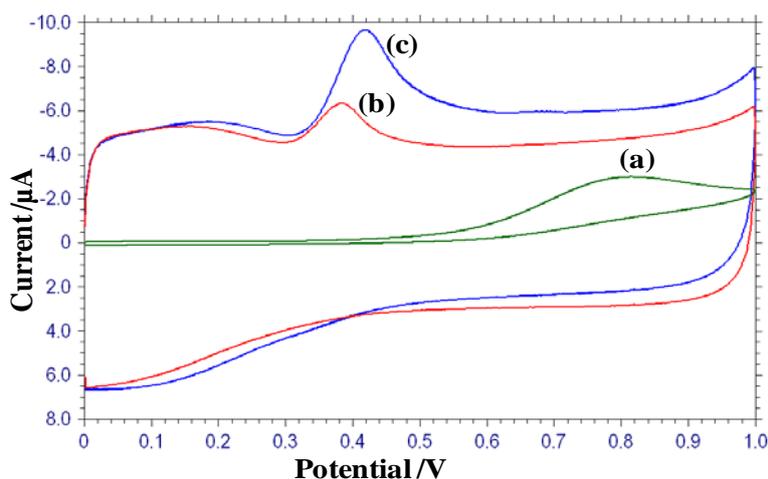

Fig. 2. Cyclic voltammogramm obtained for 1.0 mM INH at phosphate buffer (0.2 M) solution with scan rate 50 mVs^{-1} at **(a)** INH+GCE; **(b)** INH+MWCNT; **(c)** INH+MWCNT/CTAB

3.2. Effect of MWCNT-CTAB composition

We examined the effect of MWCNT-CTAB suspension amount on the electrochemical behavior of INH. The results suggested that amount of MWCNT-CTAB suspension influenced the current responses of INH. Fig. 3 demonstrates the relationship between the oxidation peak current of INH and the amount of MWNT-CTAB suspension used for coating GC electrode. As can be seen, the peak current gradually increased with increasing the amount of MWNT-CTAB suspension from 0 to 10 μL, owing to the increased effective electrode surface area for INH oxidation. Further increasing the amount of MWCNT-CTAB suspension, the peak current almost keeps constant. However, when it exceeds 14 μL, the peak current slightly decreases as shown in Fig. 3. When the coating film is too thick, the film no longer adheres tightly to the glassy carbon, reducing conductivity and part of the MWCNT-CTAB leaves the electrode surface. More excessively coated amount of MWCNT-CTAB suspension leads to less adherent film. Accordingly, 10 μL of MWCNT-CTAB suspension solution providing the maximum current response was used in further experiments.
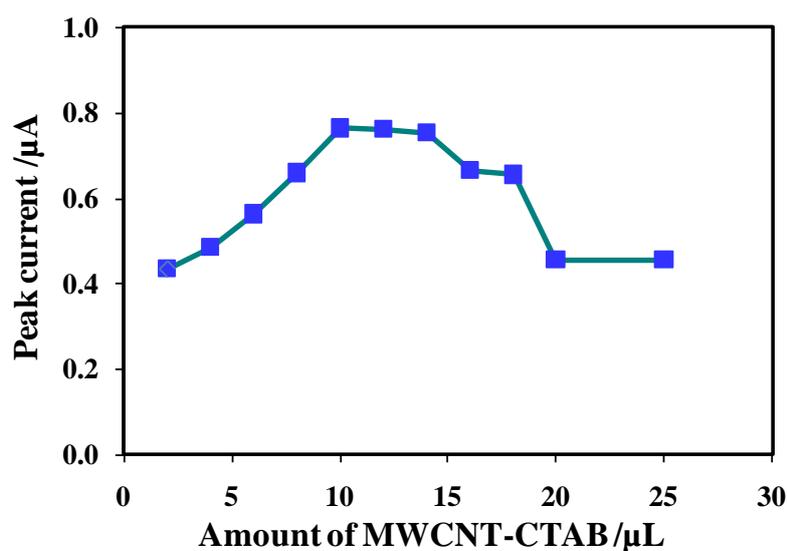

Fig. 3. Effect of MWCNT-CTAB suspension amount on the electrochemical behavior of INH

3.3. Effect of pH

The electrode reaction might be affected by supporting electrolyte and the pH of medium which are essential parameters to analyze the electrochemical reaction. There are four types of supporting electrolytes; Phosphate buffer solution (PBS), Britton–Robinson buffer solution (B-R BS), Acetate buffer solution and borate buffer solution which are commonly used in the voltammetric studies. In this experiment we have chosen the PBS which gave a sharper and better sensitive peak current. Electro-oxidation of 1.0 mM INH was studied over the pH

range of 3.0-10.4 in phosphate buffer solution by cyclic voltammetry which is as shown in Fig. 4a. The pH of solution influenced the peak current and peak potential considerably. With the increase in pH of the solution, peak potential shifted to less positive values, (Fig. 4b) and obeys the following equation: E_p (V)=0. 598-0. 042pH; (r=0.973)

The slope of the plot of E_p versus pH was found to be 42.0 mV/ pH which is close to the theoretical value of 30.0 mV which indicates the involvement of unequal number [23,24] of electrons and protons transfer in electrode reaction.

The plot of I_{pa} versus pH (Fig. 4c) clearly shows the peak current is affected by the pH value. INH showed the highest peak current at pH=4.2, then as the pH increases peak intensity decreased continuously. So pH 4.2 was selected for further studies.
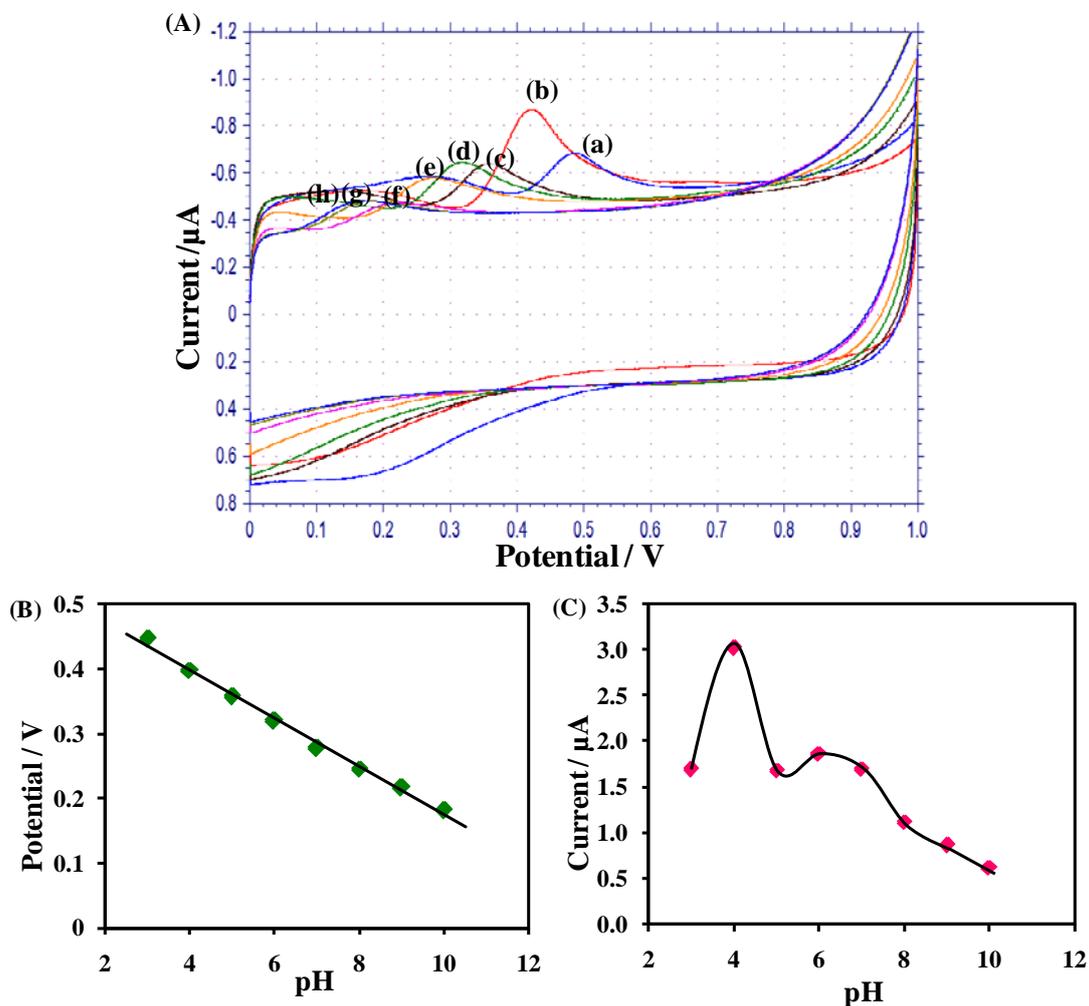

Fig. 4. (A) Cyclic voltammogram of 1.0 mM INH at different pH variation (a) 3.0, (b) 4.2, (c) 5.0, (d) 6.0, (e) 7.0, (f) 8.0, (g) 9.2 (h) 10.4. **(B)** Influence of pH on the peak potential of 1.0 mM INH on MWCNT-CTAB/GCE at a scan rate of 50mV s^{-1} in phosphate buffer. **(C)** Variation of peak current with pH of 1.0 mM INH on MWCNT-CTAB/GCE at scan rate of 50 mV s^{-1}

3.4. Influence of scan rate

Useful information involving an electrochemical mechanism generally can be acquired from the relationship between peak current and scan rate. The influence of scan rate on the electrochemical behavior of 1.0 mM INH was shown in Fig. 5a.   In the range from 25 to 300 mV s^{-1}, the oxidation peak current increased gradually with the increase in the scan rate, indicating that direct electron transfer between INH and modified electrode surface.

This was also supported by the slope value of the plot log I$_{pa}$ *vs.* log scan rate (*v*) as 0.843 as shown in Fig. 5b, which is nearly equal to a theoretical value of 1.0 for adsorption controlled process [25].

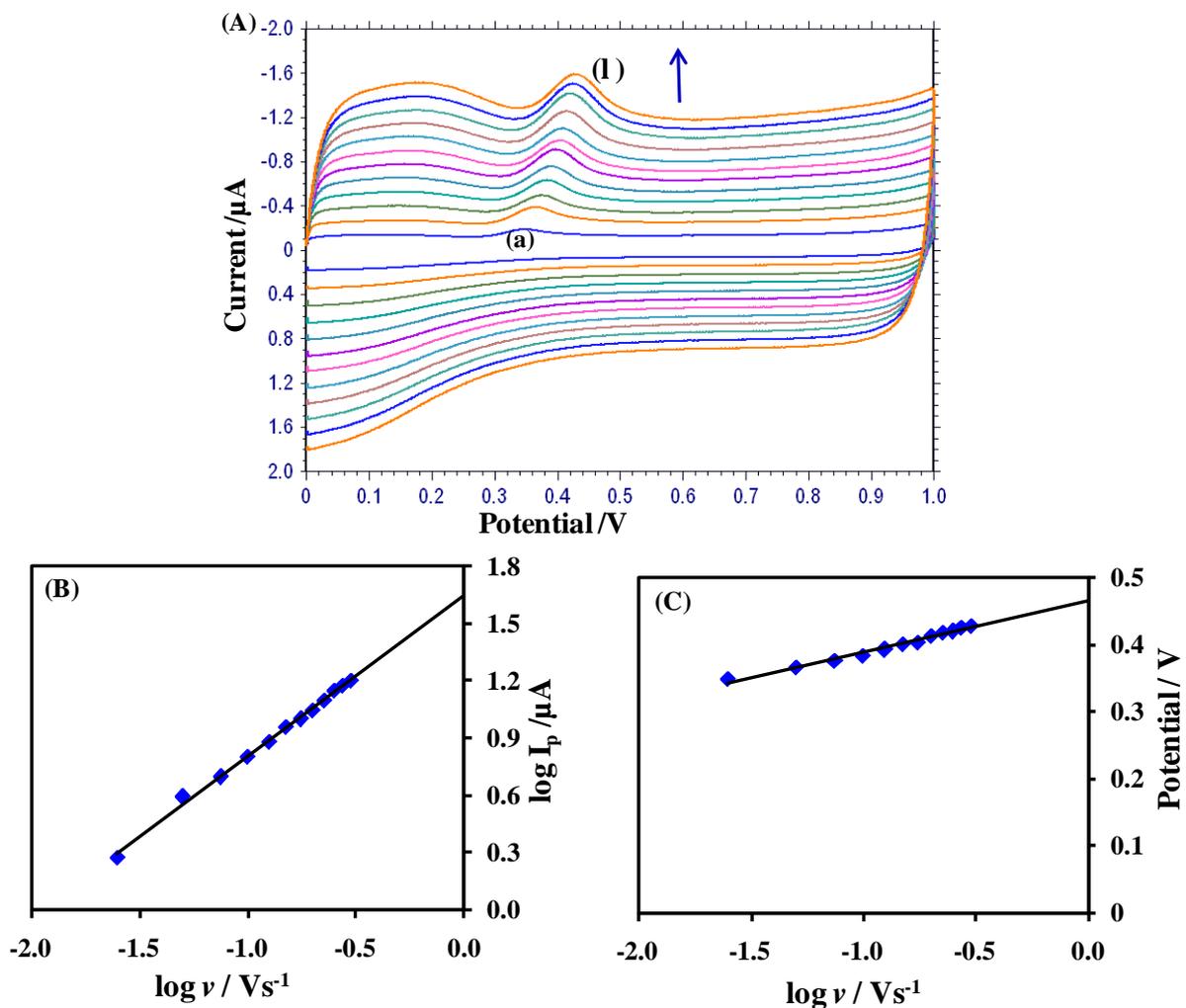$$\log I_p \ (\mu A)=0.843 \log v \ (Vs^{-1})+1.647 \quad (r=0.996) \tag{2}$$

Fig. 5. **(A)** Cyclic voltammograms of 1.0 mM INH at different scan rates (**a to *l*** : 25 to 300 mV s^{-1} respectively) in 0.2 M phosphate buffer solution; **(B)** The plot of log peak current versus log scan rate; **(C)** Relationship between the peak potential and the logarithm of the scan rate

The INH oxidation peak was shifted to more positive potentials with the increase in the scan rate, an expected behavior of irreversible electrochemical reactions. A linear relationship was observed between E_p and log v in the range from 25 to 300 mV s^{-1} (Fig 5c) represented by a linear equation; $E_{pa}(V)=0.075$ log v (Vs^{-1})+0.464 (r=0.982) .

Such behavior reveals the irreversible nature of the electrochemical process for INH. According to the Laviron theory [26] for an irreversible electrode process, E_p is defined by the following equation. (eq. 3)

$$E_P = E^{0'} + \left(\frac{2.303RT}{\alpha nF}\right) log \left(\frac{RTk^0}{\alpha nF}\right) + \left(\frac{2.303RT}{\alpha nF}\right) log v \qquad (3)$$

Where α is the charge transfer coefficient, k^0 is the heterogeneous electron transfer rate constant, n is the number of electrons transferred, v is the scan rate, and $E^{0'}$ is the formal redox potential. Other symbols have their usual meanings. This relationship allows n to be calculated from the slop of E_p versus log v. Considering T=298 K, R=8.314 J K^{-1} mol^{-1}, and F=96485 C mol^{-1}, the value of αn can be easily calculated.

For an irreversible electrode process the transfer coefficient "α" value 0.5 and from the slope of E_p *vs*. log v (0.075), the "αn" value calculated to be 0.788. Thus, the number of electron transferred in the irreversible electrode process was found to be 1.57≈2.0 in the oxidation reaction. If $E^{0'}$ is known, the value of k^0 can be estimated. $E^{0'}$ in the above equation can be obtained from the ordinate intercept of E_p *vs* v at curve $v=0$. Thus, intercept of E_p *vs*. log v plot is 0.464. Using this information and Eq. (1), the k^0 value obtained was 9.17×10^2 s^{-1}.

3.5. Mechanism

The electrochemical oxidation of INH was found to contain two electrons and four protons transfer. The electro oxidation mechanism may be proposed as in Scheme 2 which is based on earlier report [27].
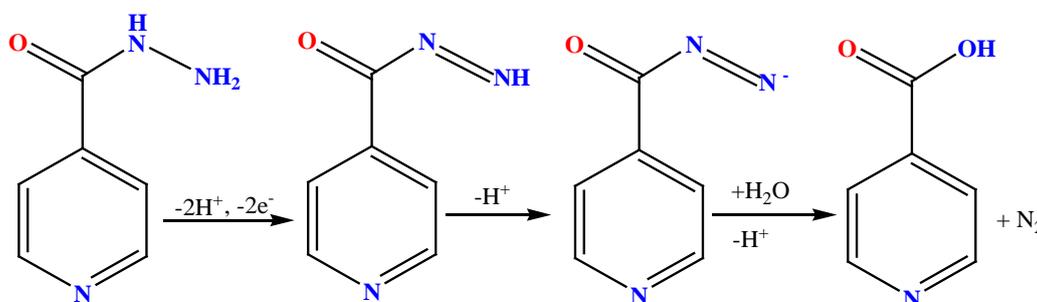
Scheme 2. Mechanism of electro oxidation of INH

3.6. Calibration curve of isoniazid determination

Differential pulse voltammetric (DPV) method was used for quantification of INH since DPV gave much higher current sensitivity and better resolution than cyclic voltammetry. According to the obtained results, it was possible to apply this technique to the quantitative determination of INH. The phosphate buffer solution of pH=4.2 was selected as the supporting electrolyte for the quantification of INH as it gave a maximum peak current. Differential pulse voltammograms obtained with increasing amounts of INH showed that the peak current increased linearly with increasing concentration, as shown in Fig. 6a. Using the optimum conditions described above, linear calibration curve was obtained for INH in the range of 1.0×10^{-7} to 1.2×10^{-6} M, there after deviation was observed as shown in Fig. 6b. The linear equation of calibration plot is;

$$I_{pa}(\mu A) = 1.82[INH]\ M + 0.936;\ (\ r = 0.976\ ) \tag{4}$$

Deviation from linearity was observed for more concentrated solutions, due to the adsorption of oxidation product of INH on the electrode surface. Related statistical data of the calibration curves were obtained from the five different determinations. The limit of detection (LOD) and quantification (LOQ) were 2.14×10^{-9} M and 7.15×10^{-9} M respectively. The LOD and LOQ were calculated using the equations:

$$LOD = 3s/m \tag{5}$$

$$LOQ = 10s/m \tag{6}$$

where 's' is the standard deviation of the peak currents of the blank (five replicates), and 'm' is the slope of the calibration curve..

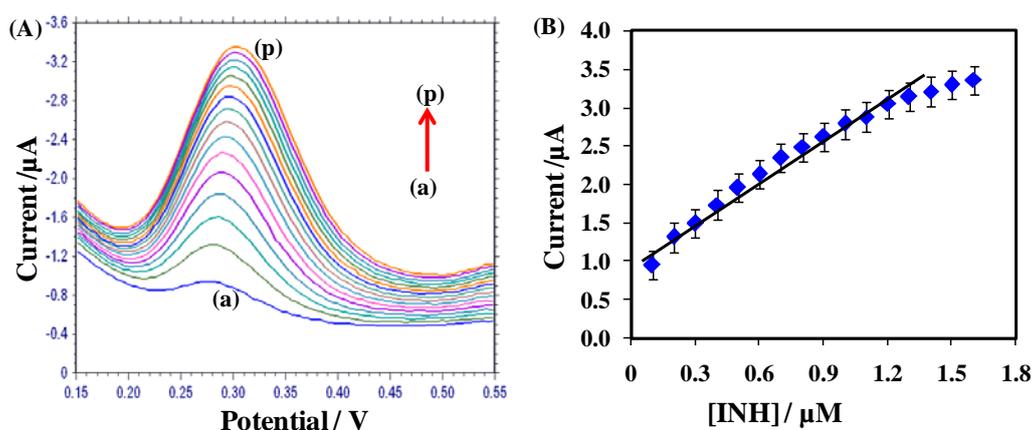**Fig. 6. (A)** Differential-pulse voltammograms with increasing concentration of INH from 1.0×10^{-7} M to 1.6×10^{-6} M at pH 4.2 of phosphate buffer solution on a MWCNT-CTAB/GCE; **(B)** Voltammetric peak current versus concentration of INH

The comparison of concentration range and detection limit for INH using different methods are tabulated in Table 1. The proposed method was compared with the previous literature methods [28-35]. The proposed method is more sensitive and selective with good precision and low detection limit

Table 1. The comparison of linearity range and detection limits for INH drug using different electrodes

Methods	Linearity range	LOD	Ref.
Carbon Paste Electrode	6.1×10^{-8} to 2.0×10^{-7} M	5.0×10^{-8} M	[12]
Gold Electrode	5.0×10^{-5} to 2.0×10^{-4} M	9.69×10^{-8} M	[13]
Functionalized Multiwalled Carbon Nanotube Modified Electrode	1.0×10^{-6} to 7.0×10^{-5} M	2.8×10^{-7} M	[27]
A Novel Molecularly Imprinted Electro Chemilumine Scence Sensor	1.0×10^{-6} to 1.1×10^{-4} M	8.0×10^{-8} M	[28]
Multi-walled Carbon Nanotube Paste Electrode	1.0×10^{-6} to 1.0×10^{-3} M	5×10^{-7} M	[29]
Electrochemically Reduced Graphene Oxide Modified Electrode	2.0×10^{-6} to 7.0×10^{-5} M	1.7×10^{-7} M	[30]
Fe(tmphen)$_3^{2+}$ -exchanged Nafion®-Modified Electrode.	5.0×10^{-6} to 2.0×10^{-4} M	1.3×10^{-5} M	[31]
Ordered Mesoporous Carbon Modified Electrode.	1.0×10^{-7} to 3.7×10^{-4} M	8.4×10^{-8} M	[32]
Influence Of Cobalt Content On Nano structured Alpha-Phase-Nickel hydroxide Modified Electrode.	1.0×10^{-6} to 1.0×10^{-3} M	5.19×10^{-8} M	[33]
Over Oxidized Polypyrrole Glassy Carbon Modified Electrode.	4.0×10^{-6} to 2.0×10^{-3} M	3.6×10^{-6} M	[34]
Multi-Walled Carbon Nanotubes Composite With CTAB Modified Glassy Carbon Electrode	1.0×10^{-7} to 1.2×10^{-6} M	2.14×10^{-9} M	Present Work

3.7. Interference study

The possible analytical application of several interfering substances on the determination of 10.0 µM INH was investigated. The tolerance limit was defined as the maximum concentration of the interfering substance that caused an error less than ±5% for determination of isoniazid. Effect of the interfering ions was verified by comparing the anodic current of INH after 100-fold excess addition of Glucose, citric acid, dextrose, gum

acacia, lactose, oxalic acid, sucrose, starch, ascorbic acid, tartaric acid, uric acid, epinephrine, norepinephrine, benserazide, levodopa, carbidopa, isoproterenol and methyldopa. The experimental results showed that foreign substances did not interfere with the voltammetric signals listed in Table 2. Thus, the procedures were able to assay INH in the presence of excipients, and hence it can be considered specific.

Table 2. Effect of the interfering substances on voltammetric response of INH

Excepients(1 .0 mM) + INH (10.0 µM)	Potential (V)	(%) Signal Change
Only INH	0.418	-
INH+Glucose	0.420	0.476
INH+Citric acid	0.423	1.182
INH+Dextrose	0.416	- 0.480
INH+Gum Acacia	0.424	1.415
INH+Lactose	0.415	-0.722
INH+Oxalic acid	0.412	-1.456
INH+Sucrose	0.421	0.712
INH+Starch	0.412	-1.456
INH+Ascorbic acid	0.413	-1.121
INH+Tartaric acid	0.422	0.947
INH+Uric acid	0.423	1.182
INH+Levodopa	0.424	0.966
INH+Carbidopa	0.422	0.947
INH+Methyldopa	0.415	0.480

3.7. Reproducibility and selectivity of the modified electrode

The MWCNT-CTAB/GCE intra-day reproducibility, modified electrodes were fabricated in five days and were tested in 1.0 mM INH solution and maximum current was determined. The relative standard deviation (RSD) was 3.6%, revealing that this method possesses good reproducibility. The modified electrode was stored for one week and there is less decrease of oxidation peak current of INH. Therefore this electrode has long term stability under ambient conditions.

3.8. Real sample analysis

The developed differential pulse voltammetric method for the INH determination was applied to urine samples. The recoveries from urine were measured by spiking drug free urine

with known amounts of INH. The urine samples were diluted 100 times with the phosphate buffer solution before analysis without further pretreatment. A quantitative analysis can be carried out by adding the standard solution of INH into the detect system of urine samples. The calibration graph was used for the determination of spiked INH in urine samples with a recovery range of 98.8 to 100.9%. The results of three urine samples obtained are listed in Table 3.

The determination of INH in a human blood plasma sample was investigated. Methanol was added to plasma sample and then centrifuged. Sample was diluted with PBS pH=4.2 and spiked with solutions of INH by standard addition method. Differential pulse voltammetric method was applied to the analysis of samples. The recovery determined was in the range from 99.95 to 100.8%, and the RSDs are listed in Table 3. Thus, satisfactory recoveries of the analyte from the real samples make the developed method applicable in clinical analysis.

Table 3. Determination of INH in urine and plasma samples

Sample	Added (10^{-5}M)	Found[a] (10^{-5}M)	(%) Recovery	RSD (%)	Bias (%)
Urine Sample 1	1	0.988	98.84%	0.347	-1.16
Urine Sample 2	3	2.995	99.86%	0.643	-0.14
Urine Sample 3	5	5.025	100.50%	0.751	0.50
Plasma Sample 1	2	1.984	100.83%	1.210	-0.8
Plasma Sample 2	4	4.005	100.12%	0.404	0.125
Plasma Sample 3	6	5.993	99.91%	1.299	-0.25

[a]Average of five determinations

4. CONCLUSIONS

GCE modified with MWCNT and surfactant CTAB showed high electrocatalytic activity. The suspension of multiwalled carbon nanotubes-cetyl trimethyl ammonium bromide increased the surface-to-volume ratio effectively. The INH undergoes two electrons and four protons transfer and is a diffusion controlled process. MWCNT-CTAB/GCE shows an interesting feature such as good sensitivity and dynamic measuring range, lower limit of detection 2.14×10^{-9} M and high stability towards INH determination. Therefore, this highly stable and renewable electrode can be introduced as a good electrochemical sensor in different systems.

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