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Electrochemical Determination of Hydralazine Hydrochloride by Preanodised Screen Printed Carbon Electrode

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The electrochemical determination of Hydralazine hydrochloride (Hy-Hcl) at preanodised screen printed carbon electrode (PSPCE) has been reported. Preanodisation was carried out by keeping the fixed potential at 2.0 V for 120 s by cyclic voltametry (CV) in pH 7 phosphate buffer solution contain KCl. Scanning electron microscopy and electrochemical impedance spectroscopy were used to characterize the preanodised screen printed carbon electrode (SPCE). The electrochemical determination of (Hy-Hcl) has been examined by linear sweep voltametry. In CV, (Hy-Hcl) has showed more oxidation current at PSPCE when compared with only pristine SPCE. Further, Hy-Hcl has the linear range of detection from 0.1 µM to 29.57 µM with a low detection limit was founded as 0.02 µM. Moreover, Hy-Hcl at PSPCE has exhibited an acceptable reproducibility, repeatability with good sensitivity and storage stability. In addition, as a future prospective this method could be used for accurate detection of Hy-Hcl in pharmaceutical industries.

Keywords: Screen printed carbon electrode, preanodised Screen printed carbon electrode, Hydralazine hydrochloride, electrochemical determination, cyclic voltametry, linear sweep voltametry.

1. INTRODUCTION

Hydralazine hydrochloride (Hy-Hcl) is the drug which is widely prescribed antihypertensive for hypertension is available in both the forms orally and intravenously [1-2]. The main action of this medicine is relaxing and dilation of the peripheral arteries, due to the blood pressure. The drug is used to cure the hypertension to reduce the blood pressure to avoid heart attacks. The Hy-Hcl has been analyzed by various traditional methodologies such as, titrimetry (3-4), HPLC [5], spectrophotometry [6-7], fluorimetry [8] and electrochemical methods [9-10]. Among various methods, electrochemical methods are very simple and user friendly when compared with other traditional methods [11-14].

Moreover, electrochemical analysis is widely used to analyze pharmaceutical drugs with high precision and accuracy [15]. Besides that, SPCE has used for various analytical applications in electrochemistry with and without modifications [16-17]. Various forms of pretreatment methods have been reported in literature to achieve the better sensitivity at the electrode surfaces [18]. Among various methods, electrochemical pretreatment is less extensive and user friendly when comparing with other methods. The previous literature reports proved that, the preanodised SPCE (PSPCE) has more electron conductivity and also has higher sensitivity to detect the biomolecules. However, only very few papers have been reported for Hy-Hcl detection or another drugs by electrochemical methods [19-24]. But, the detailed electrochemical determination of Hy-Hcl at PSPCE has not been reported yet.

In this paper we have used simple electrochemical preanodisation technique[25-26] at SPCE to detect the Hy-Hcl in the lab and real samples. Moreover, PSPCE shows enhanced oxidation peak for the Hy-Hcl, when compare to only bare SPCE. Additionally, the oxidation peak potential was found in PSPCE at lower potential than that of bare SPCE. The proposed anodized SPCE shows good sensitivity and selectivity for the Hy-Hcl. Further, Hy-Hcl has the wider linear range of detection from 0.1 µM to 29.57 µM with a good sensitivity. Moreover, Hy-Hcl at PSPCE has holds an acceptable repeatability, reproducibility with good storage stability.

2. EXPERIMENTAL

2.1 Chemicals

Hy-Hcl was purchased from Sigma Aldrich. Potassium chloride (KCl) was obtained from Sigma Aldrich. The supporting electrolyte used for all experiments was pH 7 phosphate buffer solution (PBS), which was prepared by using 0.05 M Na_2HPO_4 and NaH_2PO_4 solutions. All other chemicals were of analytical grade and used without further purifications.

2.2 Apparatus

CHI 750A and 1205 electrochemical workstations (CH Instruments) were employed for the cyclic voltammetry and amperometric studies. Surface morphological studies were carried out using Hitachi S-3000 H scanning electron microscope (SEM). A Conventional three-electrode system consisting of a modified screen printed carbon electrode (SPCE) was used as the working electrode (active surface area = 0.4 cm^{-2}), an Ag/AgCl electrode (Sat. KCl) as the reference electrode and a platinum wire with 0.5 mm diameter as the counter electrode was employed for electrochemical experiments. All measurements were carried out at room temperature in an anaerobic condition.

2.3 Fabrication of preanodised SPCE

Prior to modification, the SPCE was sonicated in ethanol and hydrochloric acid followed by double distilled water. After successive sonication the electrode was rinsed with double distilled water and allowed to dry at room temperature. Subsequently, the SPCE electrochemically preanodised at 2.0 V for 120 s in 0.05 M PBS (pH 7) contain KCl as reported by previous literature [10].

The fresh Hy-Hcl solution was prepared by using PBS (pH 7) and stored in refrigerator when not in use. The preanodised SPCE was used for further experiments and stored in 4ºC under dry condition when not in use.

3. RESULTS AND DISCUSSION

3.1 Surface characterization of electrochemically preanodised SPCE

The surface morphology of electrochemically preanodised SPCE was characterized by SEM. From the figure 1, we can clearly observe some cracked surface with large pores at the preanodised electrode. Moreover, only SPCE we can't see such a cracked images at the electrode surface (figure not shown). This is confirmed that, after the pretreatment the electrode has more surface area and it is effectively used for the detection of Hy-Hcl.

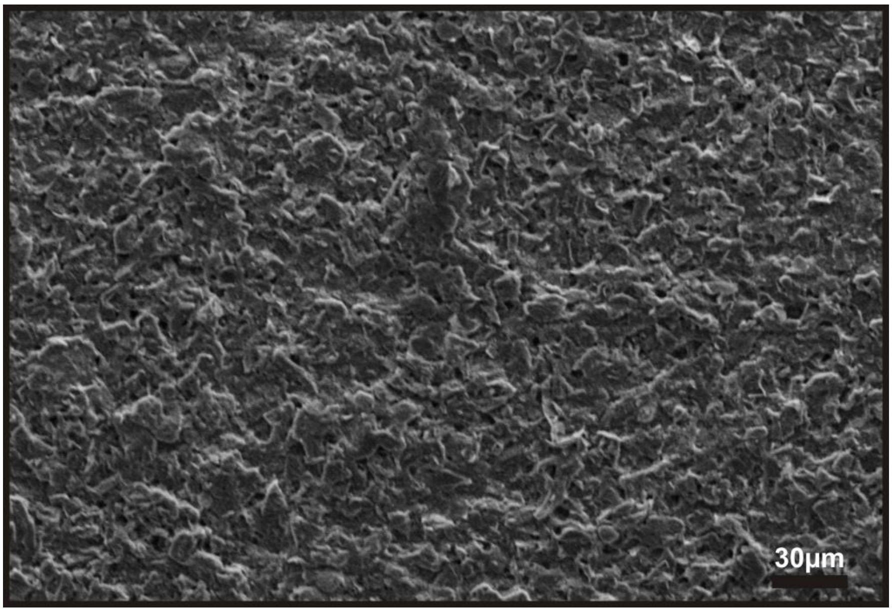

Figure 1. SEM image of Preanodised screen printed carbon electrode.

3.2 Electrochemical impedance spectroscopy (EIS) studies

The electrochemical impedance behavior at different modified electrode surfaces has been investigated by using EIS. This gives us useful information of the impedance changes on the electrode surface between each step. Fig. 2 shows the results of two different modified electrodes (bare SPCE, and PSPCE), in PBS containing 5 mM $Fe(CN)_6^{3-/4-}$. The Randles equivalent circuit model has been provided in the Fig. 2 inset, where the equivalence circuit parameters such as Ret, Rs, Cdl and Zw represents the electron transfer resistance, solution transfer resistance, double layer capacitance and Warburg impedance, respectively. In the present study, EIS of bare SPCE exhibits a small semicircle part with (Ret = 1.15 KΩ) as shown in Fig. 2a. The semicircular part diameter is equivalent to the electron transfer resistance. The linear part at lower frequencies corresponds to the diffusion process. But, a larger semicircle is observed at the PSPCE (Fig. 2b). From the EIS results confirmed that, the hindrance electron transfer occurs at PSPCE, when compared with only bare SPCE. The reason due to the electrostatic repulsion between charged surface of PSPCE with probe molecule of $Fe(CN)_6^{3-/4}$ [15].

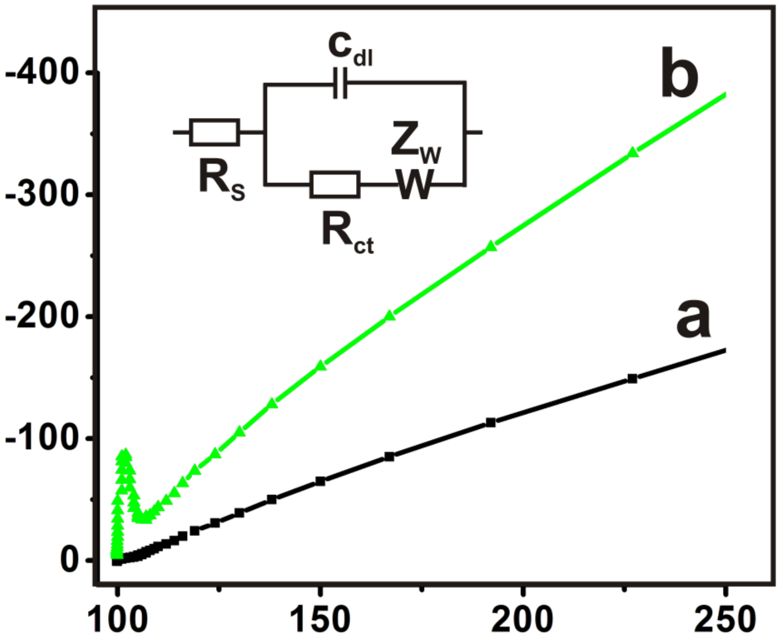

Figure 2. EIS of bare SPCE (a) and PSPCE (b) in 5 mmol L^{-1} $Fe(CN)_6^{3-/4-}$ containing 0.1 M KCl. Inset is the Randles equivalent circuit. R_s, C_{dl}, R_{ct} and Z_w represent the resistance of the electrolyte solution, double layer capacitance, charge–transfer resistance and the Warburg impedance, respectively. The frequency range is from 0.1 Hz to 100 kHz.

3.2 Electrochemistry of Hydralazine Hcl at preanodised SPCE

Cyclic voltammogram was employed to evaluate the electrochemical activity of Hy-Hcl at different modified electrodes. The electrochemistry of Hy-Hcl at PSPCE was examined in deoxygenated PBS (pH 7). Fig. 3 shows the cyclic voltammograms (CV) obtained at bare SPC (a) and preanodised SPC film modified electrodes in the presence of 10 μM Hy-Hcl in deoxygenated PBS at the scan rate of 50 mV s^{-1}. Cyclic voltammograms were recorded in the potential range of −0.2 to 0.5 V. From that figure, the oxidation peak appeared at 0.185 V for Hy-Hcl at PSPCE. However, for only SPCE the oxidation peak was appeared at 0.365 V for Hy-Hcl. Moreover, the peak potential of PSPCE towards Hy-Hcl is lower than that of SPCE. However, the oxidation peak current of Hy-Hcl at PSPCE had fifteen times higher than that of SPCE. The reason for the effective detection may be due to presence of COOH at the PSPCE after pretreatment with SPCE. From the results confirmed that, PSPCE can be effectively used to detect Hy-Hcl at lower potential with high sensitivity.

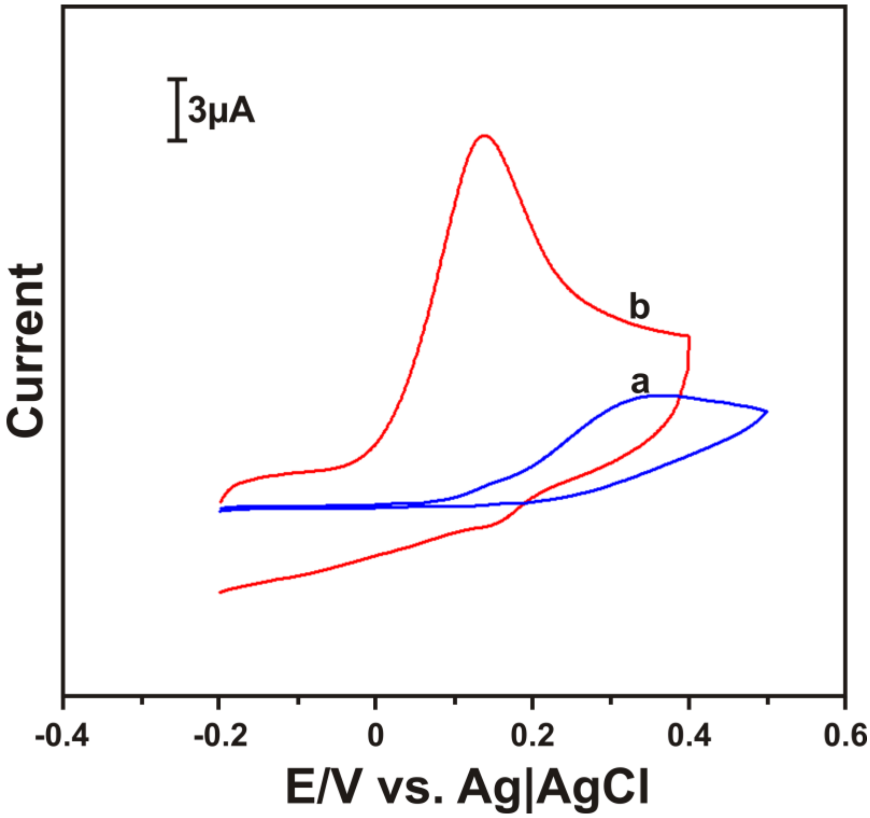

Figure 3. Cyclic voltammograms of bare SPCE (a) and PSPCE (b) in 10 µM Hy-Hcl in deoxygenated PBS at 50 mV s^{-1} scan rate.

3.3 Effect of scan rate and pH

The effect of scan rate on the voltammetric response of 10 µM Hy-Hcl at PSPCE in deoxygenated PBS also has been studied as shown in Fig. 4. Upon increasing the scan rates, the oxidation peak current increased linearly. Meanwhile oxidation peak also shifted towards the positive side. The oxidation peak current linearly increased with the increasing the scan rate between 0.05 and 0.5 Vs^{-1} as shown in Fig. 4 (inset). These results confirmed that, the oxidation of Hy-Hcl at PSPCE was a typical adsorption-controlled process [16]. The effect of pH also played an important role, which is determined to calculate the number of electron or protons transferred to the electrode surface. Fig. 5 shows the effect of pH on PSPCE in 10 µM Hy-Hcl contain various buffer solutions (pH 4 to 11). In each pH solution, a pair of stable and well-defined reversible redox peaks was observed for Hy-Hcl. In addition, both anodic and cathodic peak potentials are shifted to negative potentials with increasing pH 7-11. Furthermore, in the case of pH to 4-5 solutions, both anodic and cathodic peak potentials were shifted towards the positive potential side. Moreover, the utmost current response is observed at pH 7. The formal potential of each cathodic and anodic peak of Hy-Hcl exhibits a linear dependence with pH 4-11 with a slope of −60.4 mV/pH, with the correlation coefficient is 0.9778. This slope value is closer to the theoretical value of Nernstian equation for equal number of proton and electron transfer process [27].

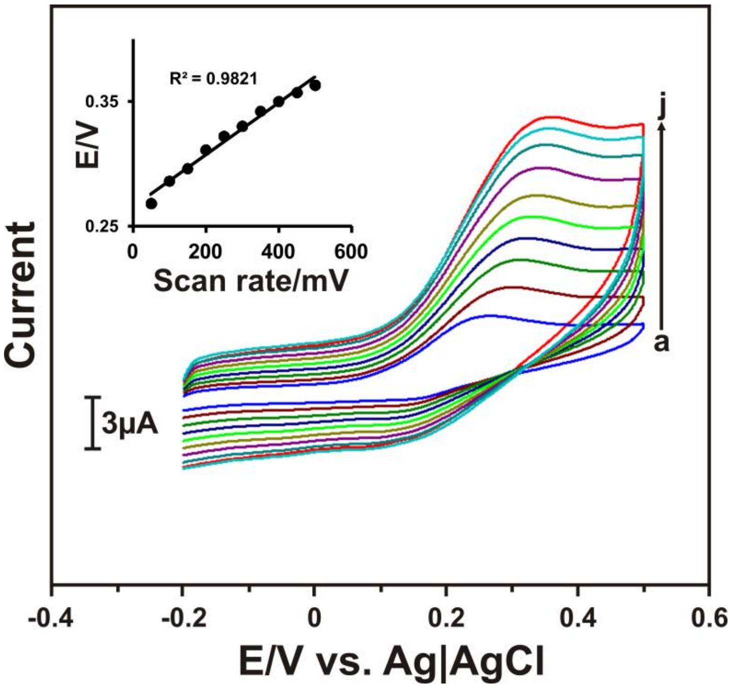

Figure 4. Cyclic voltammograms recorded at PSPCE in 10 μM Hy-Hcl in deoxygenated PBS at different scan rates. The scan rates from inner to outer are: 50 to 500 mV s^{-1}. Inset shows the linear dependence of I_{pa} vs scan rate (50 to 500 mV s^{-1}).

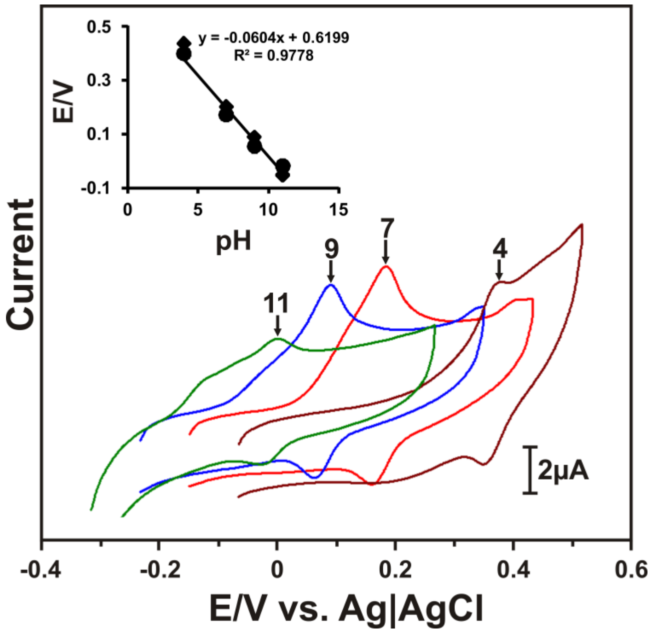

Figure 5. Cyclic voltammograms obtained at PSPCE in 10 μM Hy-Hcl in deoxygenated various buffer solutions (pH 4–11) at the scan rate of 50 mV s^{-1}. Inset shows the influence of pH on E_{pa} and E_{pc} of 10 μM Hy-Hcl at PSPCE.

3.4 Linear Sweep voltametry

Fig. 6 shows the linear sweep voltammograms of PSPCE in the presence of different concentration of Hy-Hcl in deoxygenated PBS (pH 7). In the presence of Hy-Hcl, a good oxidation peak appeared at 0.185 V at the electrode surface. Upon increasing the concentration, the oxidation current also increased gradually up to 29.57 µM. Moreover, the oxidation peak current has increased lineally with successive addition of different concentration Hy-Hcl (a-l). Furthermore, the calibrations curve from LSV, the different addition of Hy-Hcl concentrations are having the linear ranging from 0.1 to 29.57 µM with a correlation coefficient of 0.9955 (Fig. 6 inset). The LOD was found as a 0.023 µM. These results validates that the PSPCE has possess better electrocatalytic activity towards Hy-Hcl with the sensitivity of 1.07 µA µM^{-1}.

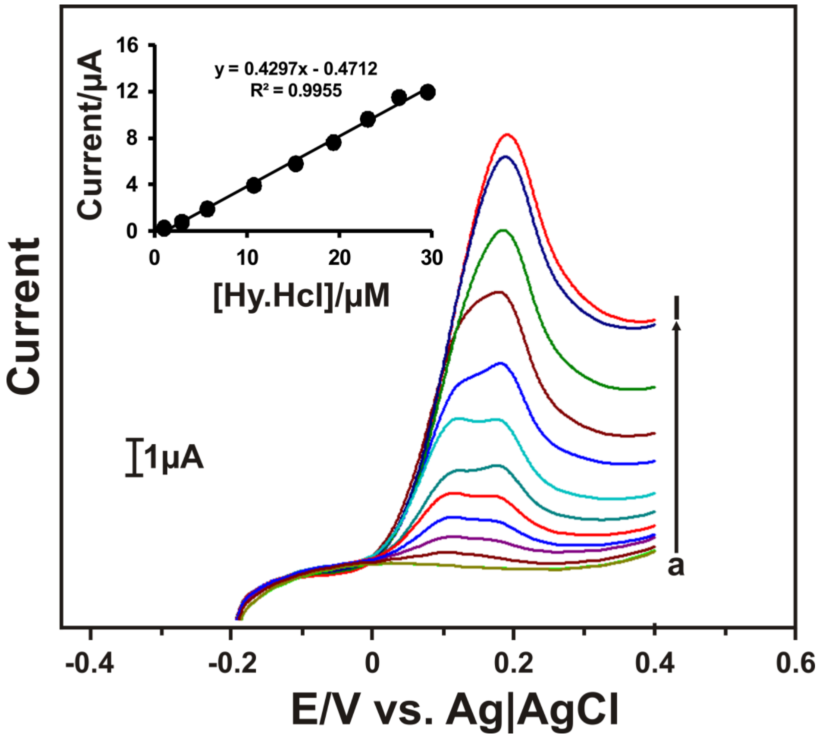

Figure 6. Linear sweep voltammogram of PSPCE (a) without and (b-l) with 0.1 – 29.57 µM of Hy-Hcl in deoxygenated PBS at 50 mV s^{-1} scan rate. Inset plot shows the linear dependence of I_{pa} vs. concentration of Hy-Hcl.

3.5 Real sample analysis

The real sample analysis is very important to evaluate the practicability of the modified electrode activity towards the analytes with an acceptable recovery. For real sample analysis, commercially available Hydralazine hydrochloride tablets (25 mg LC) were used for the analysis. Prior to the analysis, the tablets were crushed to fine powder with mortar and pestle. For the preparation of 0.001 M Hy-Hcl real sample, the crushed tablet powder was transferred into 10 ml volumetric flask, 5 ml of pH 7 (0.05 mM PBS) solution was added followed by ultrasonication in cold water bath about 20 minutes, finally the solution was made up to 10 ml mark with the buffer solution. The real sample analyses were carried out by LSV analysis. The analytical performances of Hy-Hcl at PSPCE are summarized in table 1.

Table 1. Determination of Hy-Hcl in commercially available Hy-Hcl tablets by LSV.

Sample labeled	Added (µM)	Found (µM)	Recovery (%)
1	5	4.92	98.4
2	10	10.12	101.2
3	15	14.86	99.06

From Table 1, it was evident that, the modified electrode has possessed good recovery results towards Hy-Hcl tablets, and it could be used in pharmaceutical industries in near future.

3.6 Stability, repeatability and reproducibility

In order to examine the storage stability of the modified electrode towards Hy-Hcl, it was stored in N_2 saturated PBS (pH 7) containing 10 µM of Hy-Hcl at 4 °C, and the response current was monitored every thirty minutes up to three hours using CV study. After three hours the modified electrode retained about 86 % of Hy-Hcl initial sensitivity at 4 °C, representing the modified electrode has good storage stability towards the Hy-Hcl detection. The repeatability and reproducibility of the proposed sensor were evaluated by CV studies. The three electrodes fabricated independently showed an acceptable reproducibility of 4.73 % and also it has acceptable repeatability about 4.2 % for 10 successive measurements for 10 µM Hy-Hcl. These results validating that, the modified electrode has the good repeatability and reproducibility for Hy-Hcl detection.

4. CONCLUSIONS

In summary, the simple electrochemical technique was used to determine the Hy-Hcl at PSPCE. The Preanodisation was carried out by keeping the fixed potential at 2.0 V by cyclic voltammetry (CV) in pH 7 phosphate buffer solution which consisting KCl. The SEM results shows, the PSPCE has cracked surface at the preanodised electrode surface. The Hy-Hcl has the linear range of detection from 0.1 µM to 29.57 µM with a LOD of 0.02 µM. Besides that, Hy-Hcl at PSPCE has possessed an acceptable reproducibility, repeatability with good sensitivity and storage stability. The real sample analysis also validates that, the modified electrode has good recovery results towards Hy-Hcl contains pharmaceutical tablets. This method could be used for accurate detection of Hy-Hcl in pharmaceutical industries in near future.

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