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Voltammetric Determination of Uric Acid In Kidney and Chronic Kidney Disease Urine Samples Relies on a Mercury Electrode

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ABSTRACT

A voltammetric method has been described that is characterized by speed and high accuracy, and at the same time it is considered a sensitive and fast method compared to traditional methods for determining uric acid in urine. The method was determined that relies on the use of a mercury electrode as the basic substrate for the working electrode through the use of two different techniques at the same time for the purpose of uric acid determination. Uric acid based on uric acid reduction wave. In the presence of acetate buffer solution with a pH of 7.0, the optimal conditions that affect the method were studied, such as the pH, equilibrium time, and volume of the reducing agent. The size of the model through the sequence of addition of samples against the reference electrode (Ag/AgCl) in the presence of (KCl) with a concentration of (3) molar at a clear reduction wave at a voltage of (-0.6) volts and gave results with regular linear relationships for all the methods used and they were applied and compared. By traditional methods of measurement using Spectrophotometric.

1. Introduction

The human body contains a variety of significant biological substances, including uric acid. Being a crucial sign, it is utilized in the medical identification of numerous illnesses, including renal disease. In a study, the amount of uric acid in urine was measured by electrochemical, and the results were compared to those from standard spectrophotometry methods. (S.P.Kounaves, 2023),(B. Larijani et al., 2017),(K. Swamy et al., 2020).

The ultimate result of purine metabolism in the human body, uric acid is the end product of purine metabolism where diuresis occurs. The structural formula of uric acid is depicted in Figure 1.

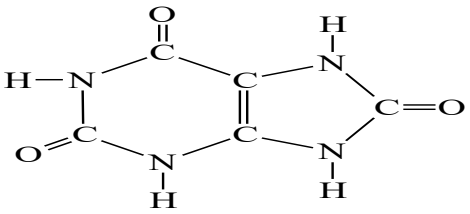

Figure 1. Uric acid Structure ,Chemically uric acid is 2, 6, 8 trihydroxypurine.

Uric Acid (UA) or 2,6,8-trihydroxypurine is a white crystalline powder that is soluble in water and an electroactive biomolecule. It is one of the major byproducts of purine metabolism in the human body. The concentration of uric acid in the blood ranges from 120 mM to 450 mM, while in healthy individuals it is approximately 2 mM in urine. Humans in good health typically eliminate 400–700 mg through urine daily(M. Rafiee et al., 2021). Unusual uric acid levels can indicate a number of physiological conditions.Excess serum uric acid buildup in the

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blood can result in gout or arthritis. High uric acid levels create hyperuricemia (R. Priefer et al., 2020),(Linzhi Li etal 2024)

Low uric acid levels cause hyperuricemia and other illnesses such heavy hepatitis and Lesch-Nyhan syndrome (Anna Ricapito etal.,2024). Additionally, uric acid is a marker for the renal failure. Its quantitative determination in body fluids is necessary for the treatment of diseases. Traditional techniques are often used to estimate the amount of uric acid based on spectroscopic methods.

It was noted with the development of science that many techniques were introduced for the purpose of measuring uric acid in different ways, such as electrical methods, which were characterized by changes and renewal of the methods through either changing the type of technology used or changing the working electrodes used, such as different solid electrodes such as used Zirconium oxide-porous carbon to detect uric acid (Hosna Cheraghi etal 2023) or voltammetric method depend on gold electrod or mercury electrode to measured uric acid concentration (AL- Jawadi et al., 2019),(RiyantoRizki ,Ariadi Tama 2020) , coulometric techniques, etc.(I. F. Abdullin et al.) and used High-performance liquid chromatography (HPLC) (Yanjie Chen etal.,2021) , Muhammad Yani Zamzam etal. Used Thin Layer Chromatography Method to identification of medicinal compound in uric acid (Zamzam etal.,2023).

2. Methodology

2.1.Preparation of Sample

Measurement procedure with sample(AL-Jawadi et al., 2018) , (AL- Jawadi et al., 2019) it use for two technique's (DPASV and DPP) as the following :

with DPASV technique:

1. A 5 ml pH solution (pH = 7.0) was added to the dry, clean measuring cell.
2. After transferring the nitrogen gas for ten minutes, the reference (Blank) measurement is completed.
3. After adding 0.02 mL of the sample and five minutes of nitrogen gas, measure.

Method of measurement with sample using DPP technique:

1. (5) ml of acetate buffer (pH = 7.0) is placed in a glass measuring cell (clean and dry).
2. The passage of nitrogen gas is checked for (15) minutes of dissolved oxygen.
3. The appropriate voltage is highlighted and then the result is plotted representing the reference (blank) polarogram.
4. Successive cells of uric acid are added at a concentration of (10-3) molar by using a micropipette until the measurement.
5. Nitrogen gas is passed for a period of (3) after adding each measurement procedure.

3. Results and discussion

3.1. Conditions optimization

The ideal circumstances for DPASV / DPP

By adding 5 milliliters of acetate buffer with a pH of 7.0 and adding 0.1 of uric acid, the ideal conditions were found for the analysis of the uric acid voltammogram for both techniques proposed by the research for measurement , as the following of table.1 , which produced the maximum value of the propagation current (Ip) and the best form of the uric acid reduction wave.

Table.1: The ideal circumstances for DPASV / DPP

DPASV		DPP	
Optimisation of the Conditions	**Values**	**Optimisation of the Conditions**	**Values**
Drop time	1 Sec	Deposition Time	5 Sec
Pulse amplitude	100 mV	Conditioning Time	20 Sec
		Equilibration Time	10 Sec
Scan rate	100 mV/sec	Scan Rate	100 mV/sec
Initial potential		- 0.1 V	
Final potintial		- 0.8 V	
pH		7.0	

3.2.Electrochemical responses of Uric acid

A comprehensive voltammetric study was carried out with both techniques used for the purpose of comparing them with the conventional methods later.As Figure .2(A. & B.) shows the volatammograms obtained for the electrochemical response of Uric acid by Differential pulse plots (DPP) and were recorded by adding graded amounts of uric acid at different concentrations after determining the optimal conditions listed in Table 1.

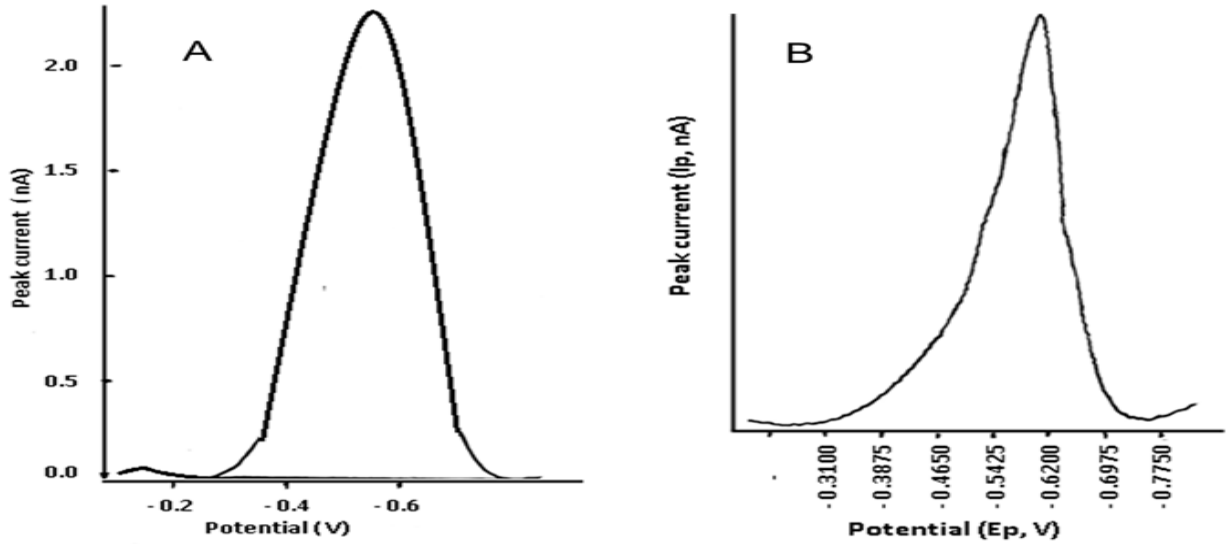

Figure 2 . shows the volatammograms obtained for the electrochemical response of Uric acid by (B.) Differential pulse plots (DPP) and (A.)DPASV.

The accuracy and sensitivity of the method in voltammetric measurement based on mercury electrode as working electrode versus reference electrode (Ag/AgCl) were observed through the electrochemical waves of both techniques.

3.3.Effect of adding concentration of Uric Acid without /with Addition of Urine

The effect of increasing the concentration of uric acid was studied after the optimal conditions were achieved by taking (5) milliliters of the pH solution (pH 7.0) and the measurements by adding successive quantities of solution (10-3) molar uric acid and

concentrations ranging as shown in figure (3,4).The correlation coefficient value (R) is 0.9912 (for DPP) & 0.9910 (for DPASV) without adding urine ,and The correlation coefficient value (R) is 0.9941 (for DPP) & concentrations ranging with and without adding urine.

0.99512 (for DPASV) without adding urine. These results indicate that the reduction process was regular and the figure.2 show this voltammogram was measured with the DPP/DPASV excretion of uric acid with

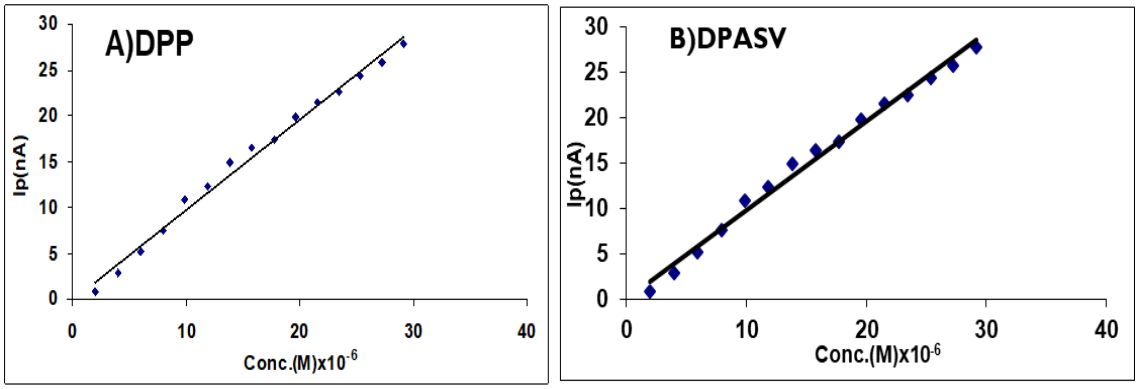

Figure 3 . the relationship between the concentration of uric acid and the current(without urine) ,for (A)DPP , (B)DPASV.

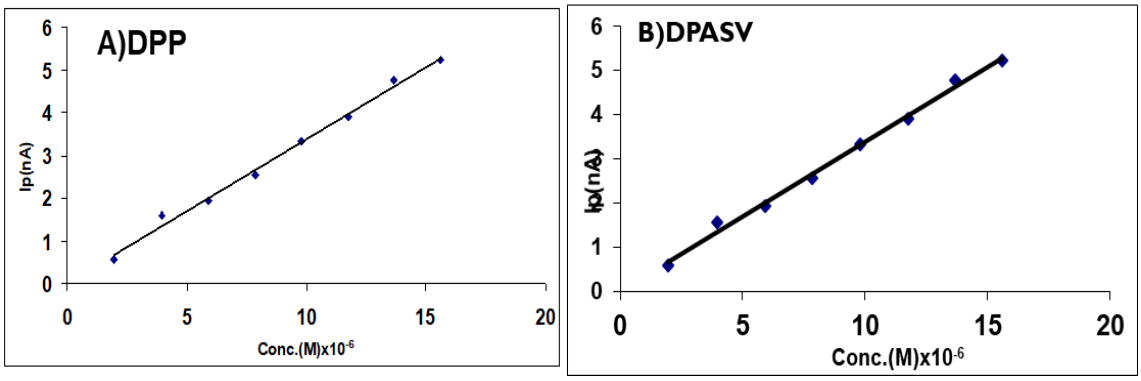

Figure (4): The relationship between the concentration of uric acid and the current(with urine) ,for (A)DPP , (B) DPASV.

It was noted through the standard curve in Figure (3 and 4) that the correlation relationship was linear and regular, which proves the accuracy of the method, which was reinforced by the value of the correlation coefficient for the curves mentioned above.

3.4.Clinical Application of Uric Acid by DPP/ DPASV

The amount of uric acid (mg / 24hr) was estimated in the administration of healthy and sick people using the (DPP)/ (DPASV), to measure uric acid in urine samples A reduction of uric acid was observed in the urine sample as shown in (Figure 5) For the concentration of uric acid in both natural and pathological condition.

Through the comparative study between the voltammetric methods based on the mercury electrode and the traditional method using spectroscopy (based on color) shown in Figure 5 above, which is the first comparison of urine samples to estimate uric acid by the above methods, it was noted that there is high accuracy and agreement between the new methods and the traditional methods, which provides a new method for comparison with high accuracy.

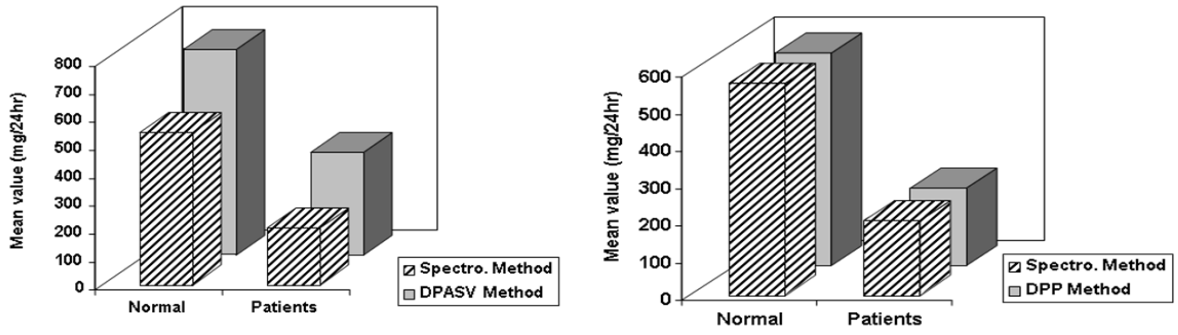

Figure 5 . shows the comparison between the suggested method of voltmeter (DPP/DPASV) and the color method.

For this purpose, and to prove the accuracy of the comparison between the proposed method and the color method to estimate the concentration of uric acid, an equation is used that is considered the correction equation between the proposed methods and the traditional methods through the two equations (Equation 1: based on the comparison between the traditional methods and the method and Equation 2: for the comparison between the traditional methods and the method) as shown in Below:

DPASV method = [(203.0606) + (0.985511× colorimetric method)] …1

DPP method = [(0.980524) + (1.081453× colorimetric method)] ...2

The proposed equations above, both equations (1 and 2), were applied to different models of the same disease and gave a high degree of agreement with the measurements conducted by traditional methods and their application by voltametric methods, which proves the accuracy and success of the proposed method for measurement.

4. Conclusions

One of the most current methods for estimating uric acid in mg/24 h in healthy individuals with kidney disease and chronic kidney disease is the use of DPP/DPASV, Among the most popular techniques for measuring uric acid in milligrams per day in healthy people with kidney disease and chronic kidney disease is DPP/DPASV, which is based on using the mercury electrode as a working electrode in a voltammetric cell for the first time in comparison to the conventional method.

When the results of this method were compared to the colorimetric method, which is commonly used in pathological analyses, and normal, healthy cases were used as a reference for measurement during the diagram shown in the conclusion, it was found that the suggested voltammetric method produced results that were consistent with the results of theIt is noted from the comparison between the results of this method and the results of the colorimetric method frequently used in pathological analyses and taking normal healthy cases as a reference for measurement during the diagram shown in the conclusion and based on the comparison of the results of The two methods, it was discovered that the proposed voltammetric method produced results consistent with the results of the colorimetric method, with the difference from the first method in the following areas: economy, solution properties, interactions, and sensitivity.

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