



Tikrit Journal of Pure Science

ISSN: 1813 – 1662 (Print) --- E-ISSN: 2415 – 1726 (Online)

Journal Homepage: <http://tjps.tu.edu.iq/index.php/j>



Evaluation the correlation between Inulin and Cystatin C with kidney function tests and GFR in patients with renal disorders

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DOI: <http://dx.doi.org/10.25130/tjps.25.2020.045>

ARTICLE INFO.

Article history:

-Received: 7 / 1 / 2020

-Accepted: 8 / 3 / 2020

-Available online: / / 2020

Keywords: GFR , Chronic renal failure, diabetic nephropathy, Inulin Clearance, Cystatin C

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ABSTRACT

Background: Kidneys are important organs in the human body because of their great importance in assisting the body to remove waste products that resulting from food metabolism through renal filtration of blood plasma from unwanted substances and excreted through diuresis and recover substances that body needs to the blood, relative effectiveness of the kidney can be estimated by measuring the efficiency of glomerular filtration rate (GRF), represented by amount of inulin filtrate into renal tubules for a specified period of time, any difference in normal value indicates the presence of a disorder in the kidney, which affects on performance of its functions and occurrence of imbalance in internal environment of the body, and leads to disrupting functions of other parts of the body, measuring rate of renal filtration is one of the best ways to know function of the kidneys and diagnose various kidney diseases and prevent their development.

Materials and Methods : The study included measuring rate of inulin clearance in the blood plasma, and estimating level of cystatin C and a number of biochemical variables in serum and evaluation the effect of body mass index BMI. 50 sample was collected from patients with chronic renal failure, and 50 sample of patients with diabetic nephropathy and 50 sample as control group.

Results: The results indicate that a significant decrease in the rate of inulin clearance in plasma and serum albumin, with a significant increase in level of serum cystatin C, glucose, urea and creatinine, and a non significant differences in level of serum uric acid in patients with chronic renal failure and diabetic nephropathy as compared to control group.

Introduction

The kidney is a double organ whose functions are remove the waste products of metabolic processes from the blood and regulate amount of body fluids, and has an important role in maintenance of normal body functions, the basic structural units of kidneys are nephrons which works to purify and filter the blood and remove waste and harmful toxins in form of diuresis, kidneys represent body's urinary system along with bladder, two ureters and one stream of diuresis called urethra[1] , glomerular filtration rate GFR is one of the functions that keeps kidneys functioning normal, and represents the net rate of turnover of plasma which is filtered through hundreds or thousands of glomeruli in quantities at Separate time periods, measuring or estimating GFR is the

basis for knowing all renal function and predicting the risk of illness before reaching final stage of kidney disease, as well as diagnosing and classifying stages of acute and chronic renal failure , GFR can be measured and estimated using standard endogenous markers such as cystatin C, urea, creatinine and exogenous markers such as inulin[2].

Renal disorders are among the most important causes that leading to death in many countries, renal disorders can be classified into two main categories: acute renal failure ARF in which kidneys stop working suddenly and completely but may regain their function and chronic renal failure CRF is a gradual loss of function in a large number of nephrons which causes a decrease in kidney function

usually due to complications of kidney disease and that causes a continuous reduction in GFR and accumulation harmful chemicals substance such as creatinine and urea in the blood[3].

Most common causes of chronic kidney disease are diabetes, therefore there is an increasing proportion of patients with diabetic nephropathy[4], diabetes causes a number of changes in metabolism in the body and blood circulation these changes lead to kidney damage and that leading to excreted albumin in urine called Albuminuria[5], as diabetic nephropathy progresses the structure of glomeruli known as glomerular filtration barrier will become increasingly damaged, and it is responsible for the selective filtration of blood that enters in glomerulonephritis and usually do not allow passage of water molecules and small proteins[6].

knowing rate of renal filtration is very important to maintain normal functioning of the kidneys, one of the most important ways to estimate efficiency of the kidneys is inulin clearance test, inulin filtered freely through glomerulus capillaries and not reabsorbed into the renal tubules, does not metabolize or bind to plasma proteins and does not affect on rate of renal filtration, inulin clearance test is done by injecting inulin in blood and measuring amount that excreted with urine during a specified period [7].

Aim of the study :

This study aimed to evaluate the correlation between Inulin and Cystatin c with Albumin ,Glucose,Uric acid ,Urea and Creatinine levels in blood of patients with renal disorders .

Materials and methods

Study design: This study was conducted on 150 samples of blood , 50 samples as control group, and

50 samples as patients group with chronic renal failure and 50 samples for patients with diabetic nephropathy male and female were included in this study, their ages ranged between (20-65) years; all samples were collected from Tikrit Teaching Hospital, Salahalddin General Hospital and a group of public laboratories in Salahalddin for the period from October 2018 to February 2019.

Inulin clearance was determined by using Elisa kit ready (American company Biopal) and cystatin C determined by using diagnostic kit (Korean company Boditech).

Statistical Analysis

The results of this study were statistically analyzed by using variance analysis software (IBM SPSS Statistics for Windows, Version 23.0., NY: IBM Corp.), and ANOVA test was used to analyze the variance between three groups at the probability level ($P \leq 0.05$), the correlation coefficient was found using Pearsons correlation coefficient to find the relationship between inulin clearance and other biochemical variables in this research.

Results and discussion

The results included statistical values of measurement inulin clearance in the blood plasma and measurement of serum (Cystatin C, Albumin, Glucose, Uric acid, Urea, Creatinine) in the blood of patients with renal failure, diabetic nephropathy compared with the control group for both sexes, a significant differences were found in this study. In this study the mean $\pm$ SD of inulin clearance in the blood plasma , Cystatin C, Albumin, Glucose, Uric acid, Urea, Creatinine in the blood serum where Summarized in table (1).

Table 1: The mean $\pm$ SD of Inulin, Cystatin C, Albumin, Glucose ,Uric acid, Urea, Creatinine for groups under investigation

Parameter	Control	Diabetic nephropathy	Renal Failure	P Value
	Mean $\pm$ S.D N=50	Mean $\pm$ S.D N=50	Mean $\pm$ S.D N=50	
Inulin(ml/min)	119.8 $\pm$ 14.3	97.1 $\pm$ 9.8	22.4 $\pm$ 7.2**	0.05 $\leq$
Cystatin C(mg/L)	0.8 $\pm$ 0.12	1.2 $\pm$ 0.3	3.7 $\pm$ 0.27	0.05 $\leq$
Albumin (g/dl)	4.9 $\pm$ 1.7	3.0 $\pm$ 2.9	2.4 $\pm$ 1.5	0.05 $\leq$
Glucose (mg/dl)	96.1 $\pm$ 11.4	162.5 $\pm$ 18.5**	123.3 $\pm$ 9.6	0.05 $\leq$
Uric acid (mg/dl)	5.3 $\pm$ 1.4	5.6 $\pm$ 1.2	5.8 $\pm$ 3.5	N.S
mg/dl) (Urea	24.2 $\pm$ 13.0	58 $\pm$ 16.9	82.7 $\pm$ 27.1**	0.05 $\leq$
Creatinine(mg/dl)	0.9 $\pm$ 0.4	1.3 $\pm$ 0.6	6.8 $\pm$ 2.4**	0.001 $\leq$

The results indicates that rate of inulin Clearance was (22.4 $\pm$ 7.2ml / min) in the blood plasma of patients with chronic renal failure, while (97.1 $\pm$ 9.8 ml / min) in patients with diabetic nephropathy and (119.8 $\pm$ 14.3 ml/ min) in control group, the results showed a significant decrease at the probability level ($P \leq 0.05$) in patients with chronic renal failure and diabetic nephropathy as compared to control group.

This results was agreement with STANLEY et al [8] and STERNER et al [9], the causes of the reduction in chronic renal failure and diabetic nephropathy refer to damage filtration units leads to a decrease in

filtration process and lower glomerular filtration rate of the kidneys which leads to decreased renal clearance of inulin in the plasma during the unit of time, inulin is completely filtered through the glomeruli and rate of renal clearance directly proportional to glomerular filtration rate and thus lead to a lower normal level of filtered rate in the blood plasma and urine [10, 11].

The results showed a significant increase at the probability level ($P \leq 0.05$) in serum cystatin C of patients with chronic renal failure and diabetic nephropathy as compared to control group(3.7 $\pm$ 0.27

mg / L), (1.2 ± 0.3 mg / L), (0.8 ± 0.12 mg / L) respectively, this result was agree with many studies (Moreira et al) [12], (Wong et al) [13], (Mumtaz et al) [14], and the reason for increase is due to lower in glomerular filtration rate which lead to damage of the kidneys, causing accumulation of waste, toxic substances and excess water in the body[15], cystatin C is closely linked to glomerular filtration rate by renal glomeruli, as well as the low rate of absorption by tubule as a result concentration of cystatin C is increase in blood and the fact that cystatin C is not affected by age, sex and nutritional status and because it constant concentration in the body and small molecular weight gives an accurate diagnosis of glomerular filtration rate [16].

The results indicates that level of serum albumin was (2.4 ± 1.5 g / dl) of patients with chronic renal failure, while it was (3.0 ± 2.9 g / dl) in patients with diabetic nephropathy, and (4.9 ± 1.7 g / dl) in control group, the results showed a significant decrease at the probability level ($P \leq 0.05$) in the blood of patients with chronic renal failure and diabetic nephropathy as compared to control group.

This results was agreement with the result of (Lang J et al) [17], (Junlin et al) [18], the causes of reduction refer to the kidney damage Resulting from chronic renal failure and diabetic nephropathy, which lead to loss of albumin through damaged filtration units and glomeruli, and the amount of albumin increases in the urine and there is a decrease in the concentration of albumin in the blood, which ultimately leads to the status of hypoalbuminemia [19], or possible cause is due to malnutrition and the increase in the body's need for albumin [20].

The results indicates that level of serum glucose was (123.3 ± 9.6 mg / dl) of patients with chronic renal failure, while it was (162.5 ± 18.5 mg / dl) in patients with diabetic nephropathy, and (96.1 ± 11.4 mg / dl) in control group, the results showed a significant increase at the probability level ($P \leq 0.05$) in the blood of patients with chronic renal failure and diabetic nephropathy as compared to control group.

This results in agreement with the result of many studies (Maher et al) [21], (Radica Z et al) [22], where it was found that high blood sugar leads to diabetic nephropathy, and leads to several dangerous factors that contributed to the development of diabetes nephropathy and its complications which may gradually cause diabetic retinopathy, increase levels of blood sugar continuously lead to chronic renal failure and kidney disease [23], and makes kidney working harder to do filtration function and that lead to damaged kidney [24].

Regarding to the results of uric acid levels showed in table (1) the results showed a non significant differences at the probability level ($P \leq 0.5$) in the blood of patients with chronic renal failure and diabetic nephropathy as compared to control group. (5.8 ± 3.5 mg / dl), (5.6 ± 1.2 mg / dl), (5.3 ± 1.4 mg / dl) respectively and this may refer to the all patients

that their samples were studied taking Zyloric (Allopurinol), an effective drug for lowering uric acid and inhibiting its synthesis in the body, and used to treat inflammatory diseases in the body and is a disincentive to purine through inhibition of the enzyme xanthine oxidase and responsible for converting the oxidation hypoxanthine to xanthine [25],[26], uric acid depends in its composition on effectiveness of the enzyme xanthine oxidase Which is produced in the liver, and then blood transfer of uric acid to the kidneys and approximately (75%) are filtered and reabsorbed one more time through small renal tubules[27], these results are agree with the results of many previous studies (Marian et al)[28],(Richard J et al)[29],(Amirhesam et al)[30] which proved the effectiveness of this drug in preventing and reducing the percentage of uric acid production in patients with chronic renal failure and diabetic nephropathy.

The results indicates that level of serum urea was (82.7 ± 27.1 mg / dl) of patients with chronic renal failure, while it was (58.5 ± 16.9 mg / dl) in patients with diabetic nephropathy, and (24.2 ± 13.0 mg / dl) in control group, the result showed a significant increase at the probability level ($P \leq 0.05$) in the blood of patients with chronic renal failure and diabetic nephropathy as compared to control group.

This results was in agreement with the result of (AlJumaili) [31], the causes of increase level of urea refer to chronic renal failure and diabetic nephropathy which may cause a reduction of excretion urea in the urine, renal diseases leads to a low excretion of urea resulting in accumulation and high concentration of urea in the blood, and second reason for increase urea is the lack of commitment Patients with diet and intake large amount of proteins and that leads to increase urea concentration in the body [32], and it was found that the level of urea is close to the level of creatinine in the blood, increase in urea means presence a disorder of the renal filtration function [33].

The results indicates that level of serum creatinine was (6.8 ± 2.4 mg / dl) of patients with chronic renal failure, while it was (1.3 ± 0.6 mg / dl) in patients with diabetic nephropathy and (0.9 ± 0.4 mg / dl) in control group, the results showed a significant increase at the probability level ($P \leq 0.05$) in the blood of patients with chronic renal failure and diabetic nephropathy as compared to control group.

This results was agreement with the result of (Alhbab) [34], (Philip et al) [35], the causes of increase level of creatinine in the blood refer to the fact that creatinine is a waste metabolic nitrogen metabolic processes, which excreted with urine in normal state, but in presence of a disorder in the kidney may lead to a gradual loss of kidney function and insufficiency in filtration and that leads to increase creatinine concentration in the body, the patients with chronic renal failure and diabetic nephropathy have a low value of glomerular filtration

rate, and the concentration of creatinine inversely proportional to the glomerular filtration rate, any simple reduction in GFR leads to elevation in concentration of creatinine in the blood [36], increase in level and formation of creatinine are usually associated with muscle mass, high amounts of proteins, and thyroid disorders [37], and second reason for increase creatinine may be due taking patients some drugs that cause an increase in the proportion of creatinine, which contains a high proportion of substances that are deposited in the kidneys [38].

The effect of body mass index (BMI) of the patients and control groups was studied, as shown in the table (2).

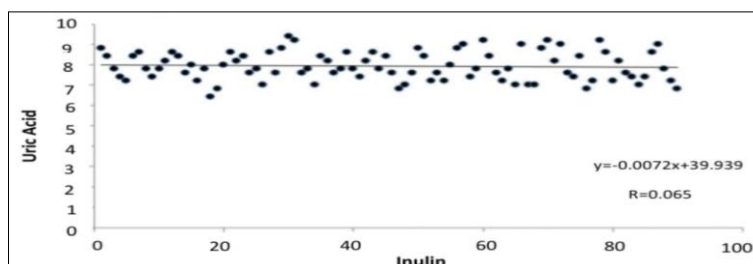
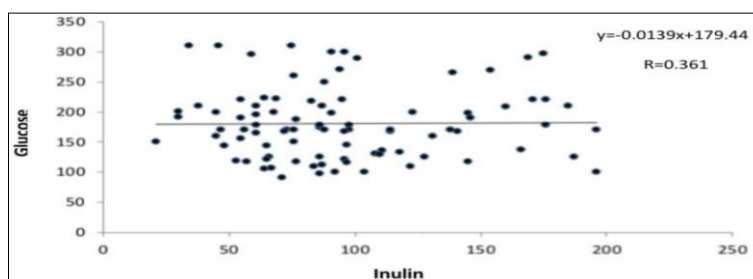
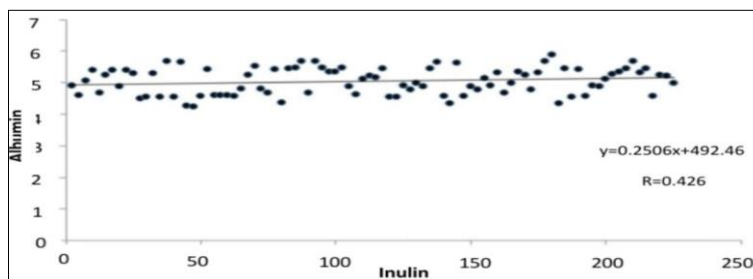
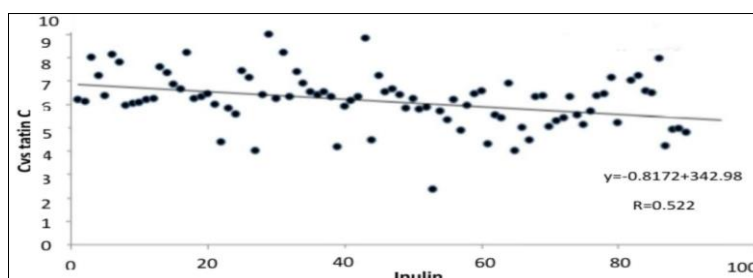
Table 2: represents the standard deviation rate of BMI in patients and control

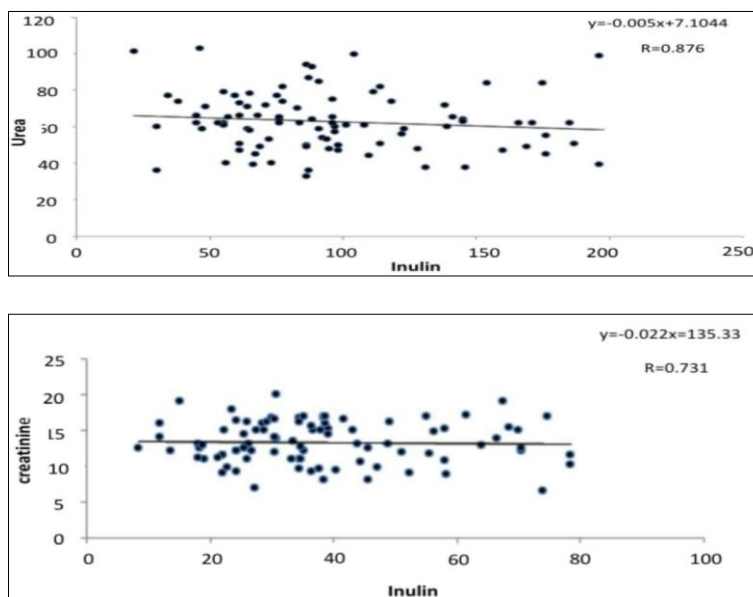
Groups	Mean± SD (kg/m ²) N=50	P Value
Renal Failure	17.3 ± 2.4	0.05 ≤
Diabetic nephropathy	20.6 ± 1.9	
Control	26.5 ± 2.7	

The results indicates that a significant decrease in the BMI level at the probability level ($P \leq 0.05$) and level was ($17.3 \pm 2.4 \text{ kg / m}^2$) in patients with chronic renal failure, while the body mass index ($20.6 \pm 1.9 \text{ kg/m}^2$) in patients with diabetic nephropathy compared to the control group ($26.5 \pm 2.7 \text{ kg / m}^2$), this result agree with the study of (Maruja et al) [38], where it was found that the level of BMI decreases in patients with chronic renal failure and Diabetic nephropathy, due to the loss of large amounts of protein and the destruction of fatty tissue during the disease, which leads to a decrease in body mass significantly.

Correlation between inulin clearance and other biochemical parameters

Correlation values r were measuring between the rate of inulin Clearance and biochemical variables, as shown in the following figures: -





Figures represents Coefficient of correlation of inulin clearance with biochemical variables

Conclusion

The results showed a significant decrease in the rate of inulin clearance in the blood plasma, and a decrease in the level of albumin, and a significant increase in the level of cystatin C, glucose, urea and

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تقييم العلاقة بين الاينولين و السستاتين سي مع اختبارات وظائف الكلية و معدل الترشيح الكلوي لدى مرضى القصور الكلوي

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الملخص

الخلفية: تعد الكلية من الاعضاء المهمة في جسم الإنسان لما لها من أهمية كبيرة في مساعدة الجسم في التخلص من الفضلات الناتجة عن أيض المواد الغذائية وذلك عن طريق الترشيح الكلوي لبلازما الدم من المواد غير المرغوب فيها والموجودة في الدم بطرحها في الإدرار مع استرجاع المواد التي يحتاجها الجسم الى الدم و الكلى و يمكن تقدير فعالية الكلية النسبية من خلال قياس كفاءة الترشيح الكبيبي ممثلا بكمية الاينولين المترشح الى جوف النيببات الكلوية لمدة زمنية محددة وإن أي اختلاف عن القيمة الطبيعية يدل على وجود خلل في الكلية مما يؤثر على قيامها بوظائفها و حدوث خلل في البيئة الداخلية للجسم ، فتختل بذلك وظائف بقية أعضاء الجسم الأخرى، يعد قياس معدل الترشيح الكلوي من افضل الوسائل لمعرفة وظيفة الكليتين و تشخيص امراض الكلية المختلفة و منع تطورها .

المواد و طرق العمل : هذه الدراسة تضمنت قياس معدل التخلص من الاينولين في بلازما الدم وتقدير مستوى السستاتين سي و عدد من المتغيرات الكيموحيوية في مصل الدم و دراسة تأثير مؤشر كتلة الجسم لجميع العينات التي تمت دراستها لمرضى العجز الكلوي المزمن (50 عينة) ، و مرضى اعتلال الكلية السكري (50 عينة) ، و مجموعة السيطرة (50 عينة) .

النتائج : اظهرت النتائج وجود انخفاض معنوي في معدل تنقية الدم من الإينولين من قبل الكلية، و انخفاض في مستوى الألبومين ، و ارتفاع معنوي في مستوى السستاتين سي ، الكلوكرز ، اليوريا و الكرياتينين و عدم وجود فروق معنوية في مستوى حامض اليوريك في مصل الدم لدى المرضى المصابين بالعجز الكلوي المزمن و اعتلال الكلية السكري مقارنة مع مجموعة السيطرة عند مستوى الاحتمالية ($P \leq 0.05$) .