

Study of Some Purine Metabolic Enzymes in Sera of Patients with Renal Failure

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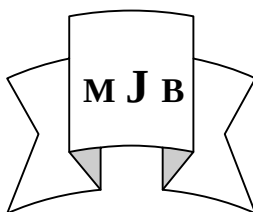
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Abstract

one hundred seven patients (51 male and 56 females) suffering from renal failure admitted to Mirjan Teaching Hospital were included . Twenty healthy individuals were included as control group. Blood samples were collected and the sera were separated . It was found that renal failure was more predominant among the patients age group rang from 40 – 70 years old. Besides , it was found out that the mean values of some biochemical parameters importance for the detection of the disease which were investigated in this study were higher than the normal . It was found that urea and creatinine mean value for all patients were highly significant if compared to control group. The mean value ($M \pm SD$) of Adenosine deaminase activity was decreased significantly (14.30 ± 10.47) in all patients with renal failure if compared to the control group (63.80 ± 22.98). On the other hand ,the mean of xanthine oxidase activities also reduced but not significantly (4453.98 ± 694.86) if compared with control group (4669.45 ± 811.80) .

دراسة لبعض الانزيمات المؤيضة للبيورينات لمرضى الفشل الكلوي

الخلاصة

تضمنت هذه الدراسة , (107) مريض (51 من الذكور و 56 من الاناث) يعانون من عجز الكلية والذين تراجعون مستشفى مرجان التخصصي للطب الباطني . كما شملت (20) شخص من الاصحاء كمجموعة سيطرة .

فقد وجد ان مرض عجز الكلية كان سائدا بدرجة كبيرة بين المرضى للاعمار بين (40 – 70) سنة تم التحري عن بعض المؤشرات البايوكيميائية لمرضى العجز الكلوي ومقارنة ذلك بالاصحاء (مجموعة السيطرة) وقد وجد ان تركيز اليوريا والكرياتنين قد سجلا معدلات معنوية عاليه عند جميع المرضى كذلك درست فعالية انزيم (ادينوسين دي اميناز) في مصل المرضى وقد لوحظ ان فعالية هذا الانزيم قد تضاعلت بدرجة كبيرة وبشكل معنوي في حين لا توجد هناك فروقات معنوية من دراسة فعاليته لدى المرضى موزعة حسب الجنس .

اضافة الى ذلك تم دراسة فعالية انزيم (زانثين اوكسيديز) في مصول مرضى العجز الكلوي وقد وجد ان فعالية الانزيم قد تضاعفت بدرجة غير معنوية كما وجد ان ليس هناك اية فروقات عند قياس فعالية الانزيم لدى الاناث والذكور من مرض العجز الكلوي .

Introduction

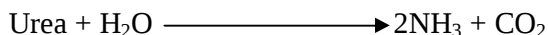
Renal Failure is a loss of renal function ;characterized by uremia, and the retention of other nitrogenous wastes in the blood. Renal failure can broadly be divided into two categorie [1].

The type of renal failure (acute and chronic) is determined by the trend in the serum creatinine. Other factors which may help differentiate acute and chronic kidney disease include the presence of anemia and the kidney size on ultrasound . Long –standing .,i.e. chronic kidney disease generally leads to anemia and small kidney size [2].

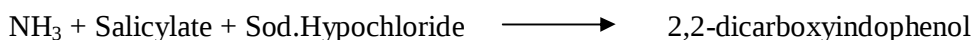
Purine metabolic enzymes such as xanthine oxidase (EC.1.1.3.22) and adenosine deaminase (EC.3.5.4.4) play a major role in degradation of purines such as adenine and guanine [3]. These enzymes are widely distributed in small intestine , kidney and liver [4].

No previous studies are known about the relationship between renal failure and nucleotide metabolic

Urease



Nitro prusside



Green complex

PH > 12



37 °C

enzymes , So this study is carried out to show the role of some nucleotide metabolic enzymes in renal failure such as adenosine deaminase activity and xanthine oxidase activity in renal failure (ADA and XOD).

Materials and Methods

Patients

In this study , 107 patients (51 male and 56 female) suffering from renal failure admitted to Mirjan Teaching Hospital unit of artificial kidney were included. Also , 20 healthy persons were included and distributed accordingly as a control group . Blood samples were obtained and the sera of them were subjected for testing.

Methods

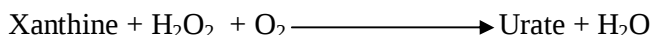
Determination of urea and creatinine

Urea and creatinine were estimated using kits provided by biomerieux company , France .

Principle of urea and creatinine

Determination of Xanthine Oxidase activity [5]

Xanthine oxidase activity (the oxidase form) was determined by the method of Ackerman and Bril (50) in



Determination of Adenosine deaminase Activity[6]

The adenosine deaminase assay is based on the enzymatic deamination of adenosine to inosine , which is converted to hypoxanthine by purine nucleoside phosphorylase (PNP) .Hypoxanthine is then converted to uric acid and hydrogen peroxide (H₂O₂) by xanthine oxidase (XOD). H₂O₂ is further reacted with N-Ethyl -N-(2-hydroxyl -3- sulfopropyl) -3-methylaniline (EHSPT) and 4-aminoantipyrine (4-AA) in presence of peroxidase (POD) to generate guinone dye which is monitored kinetically.

Table 1 Mean and standard deviation of adenosine deaminase in patients with renal failure and control group .

Test	Mean ± SD		P values
	Patients	Control	
Adenosine Deaminase (ADA) U/L	14.30 ± 10.47	63.80 ± 22.98	0.001

ADA was expressed in the cytosol of all cells as deficiency of ADA results in accumulation of adenosine.This would result in the excretion of some amount of adenosine in the urine [7].

Xanthine Oxidase activity

sera of control subjec and patients with renal failure. This method depends on the enzymatic oxidation of xanthine which is followed spectrophotometrically by measuring uric acid formation at (293) nm. [5]

Results and Discussion

Adenosin deaminase activity :

Few studies had previously pointed out findings about the level of adenosine deaminase enzyme in uremic patients especially those suffering from renal failure. In this study , it was seen that adenosine deaminase levels decreased in all patients (n=107) with renal failure (with the value 14.30 U/L respectively) . This result was highly significant in renal failure cases when compared with the control group (p<0.01), as shown in Table (1) .

Xanthine oxidase activity is one of the most important enzyme in nucleotide metabolism . The enzyme activity was investigated in all patients of renal failure and in the control group. It was found that the mean value of enzyme activity in the sera of patients was (4453.9 U/L) and in the control group was (4669.45 U/L) .

According to the results above , there was no significant difference between the enzyme activity of patients and in

the control group ($P > 0.05$) , as shown in Table (2).

Table 2 Mean and standard deviation of xanthine oxidase in patients with renal failure and control group.

Test	Mean $\pm$ SD		P. Value
	Patients	Control	
Xanthine Oxidase Activity U/L	4453.98 $\pm$ 694.86	4669.45 $\pm$ 811.80	0.260

$P < 0.05$ Significant

$P > 0.05$ not Significant.

This mild decrease of xanthine oxidase in uremic patients may be attributed to the effect of high levels of urea in the sera of those patients which might inhibit for this enzyme [8]. Also, when the enzyme activity is reduced , xanthine will accumulate in human tissues and then excreted in urine resulting xanthineurea [7,9].

Relationship between urea , creatinine and nucleotide metabolic enzymes

Statistical analysis was performed to show whether there was relationship between the presence of urea and serum creatinine at high levels and the activities of xanthine oxidase and adenosine deaminase . The results obtained in this study reveal that there is no relationship between urea when present at high concentrations and the decrease in xanthine oxidase ($r = - 0.001$; $P > 0.05$).

However , urea acts as inhibitor for XOD , and it is suggested that urea acts by reversible attachment at the substrate binding site . So, its effect on XOD may have occurred by indirect mechanism [10]. Thus, there was no direct relationship between urea and enzyme level in the sera of the patients .In addition to that , it was observed that high levels of creatinine had no relationship with the reduction of XOD levels in the sera of the patients. ($r = - 0.016$; $p > 0.05$).

On the other hand , it was found that urea had no relationship with the decrease occurred in adenosine deaminase activity ($r = - 0.092$; $p > 0.05$). Additionally, creatinine elevation in renal failure patients had no relationship with the reduction in the adenosine deaminase activity ($r = - 0.117$; $p >$

0.05). Although urea plays a role as an inhibitor to xanthine oxidase (and

adenosine deaminase [10] but its effect on the enzymes activities has little importance since there is increase in xanthine in the sera of the patients or adenosine [12] respectively. However, the main cause that results in decreasing such enzymes comes from haemodialysis procedure which can effect chiefly adenosine deaminase activity but not at the same degree on xanthine oxidase activity [11]. Some studies have showed that the high reduction in adenosine deaminase activity (4.5 folds decrease if compared to control) had stemmed from the effect of haemodialysis on the transcription of the gene encoding this enzyme [13] , leading to decrease of ADA and increase of the concentration of adenosine. . It has been seen that a deficiency in ADA activity causes moderate to complete lack of immune function .Therefore , most patients with renal failure suffer from weakening in immune system especially those on dialysis process [14].

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