The Pharma Innovation Journal 2018; 7(10): 18-21

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.03
TPI 2018; 7(10): 18-21

www.thepharmajournal.com
Received: 21-08-2018
Accepted: 05-09-2018

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Study of 24 hours urinary proteins, creatinine, & serum total protein and creatinine in diabetic and hypertensive patients

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Abstract
Objective: The aim of the study is early possible detection of any abnormal biochemical parameter will help in diagnosis of renal diseases which could be helpful in prevention of complications and permanent damage of kidney. The aim of the present study is taken up to assess the severity of the renal damage in diabetic and hypertensive subjects using biochemical parameters such as Creatinine, Total protein and Albumin in Serum and urine which are also estimated by standard procedures.
Method: The present study is carried out in Department of Biochemistry, SIMS. The relevant data is gathered from the Department of medicine, SIMS and hospital during the year 2016. In the present study, there are 4-groups namely control (Normal subjects), Diabetic Group, Hypertensive Group, Diabetic and Hypertensive Group. 20 patients in each group and age varies between 42 to 65 years in all the subjects of all groups. All the cases were selected based on the disease. The study was done in patients who are admitted in Department of medicine.
Results: There is an increase in Urinary total protein, Urinary Albumin and Serum Creatinine. All the patients of three diseased groups showed increased levels of Urinary Total Protein and Albumin. Showing much significance statistically when compared with when normal group. P-Values for all the urinary parameters are highly significant as they are less than 0.05. P-value is significant for serum creatinine and serum albumin in diabetic and Hypertensive group. The results indicate the extent of damage of the glomerular membrane. Due to damage of glomerular membrane, loss of total protein and albumin is higher in diabetic and hypertensive group compare to normal P Values of diabetes with hypertension are more significant than only diabetes or hypertension.
Conclusion: The results concluded that, estimation of Urinary creatinine, total protein and albumin in 24-hours urine gives valuable information in early diagnosis can lead to prognostic assessment and suitable treatment as early as possible which can prevent further damage to gloumerular Membrane in particular and other systems of diabetic and hypertensive patient.

Keywords: Diabetes, hypertension, albumin, creatinine, protein

Introduction
The patients with Diabetes mellitus are at a high risk of suffering from renal damage. Persistent proteinuria is the hallmark of diabetic nephropathy [1]. In Diabetes mellitus, the vascular permeability increases and albuminuria appears when the metabolic regulation is poor, because of glycosylation and a loss of negative charges on the glomerular membrane. Diabetic nephropathy and deterioration of the renal function in Diabetes mellitus are preventable by the diagnosis of proteinuria at an early stage.
In recent years, dipsticks have been developed for the detection of proteinuria, which are claimed to be more convenient and sensitive as compared to the qualitative tests. It has been found by many researchers that dipstick tests are too insensitive to evaluate the diabetic patients for microproteinuria. In these situations, the quantitative assessment of the urine protein is necessary [3, 4]. The most accurate method for the detection of microproteinuria is the measurement of the protein content in 24 hours urine samples.

Materials and Methods
The present study was carried out in Department of Biochemistry, SIMS Hyderabad. The relevant data is gathered from the Department of medicine, SIMS and hospital during the year 2016.

Study design
The present study include 20 diabetic cases, 20 hypertensive cases, 20 diabetic and hypertensive cases and 20 normal individuals who serves as control group they are all above 42 years. All the cases were selected based on the disease
The following bio-chemical parameters are estimated and compared with normal persons of the same age group (above 42 years).

Exclusion criteria
The exclusion criteria which were considered for the selection of the diabetic patients were hypertension, pregnancy, emotional or physical stress, and smoking, exposure to extremes of climate, urinary tract infections, any intercurrent illness, strenuous exercise and menstruation, to rule out any proteinuria which had occurred from other causes.

Collection of sample
The patients and the controls were instructed to collect untimed spot urine samples. The urine samples were collected at room temperature, without adding any preservatives. Immediately after their collection, the urine samples were analyzed for protein and creatinine. Venous blood was collected from the subjects by venipuncture of the antecubital vein in a fluoride bulb. The serum was then separated by centrifugation at 3000 rounds per minute, for 15 minutes, for the estimation of the fasting serum glucose concentrations.

Biochemical analysis
Heller's nitric acid test, the heat coagulation test and the sulfosalicylic acid test were used qualitatively for the detection of the urinary protein [5]. A semi quantitative estimation of the urinary protein was done by the urinary dipstick method. The quantitative estimation of the urinary protein was performed by the colorimetric Biuret method [6, 7]. The colourimetric estimation of urinary creatinine was done by the modified Jaffe's method [7, 8, 9]. The estimation of fasting serum glucose was done by the GOD – POD (Glucose oxidase – Peroxidase) method [10].

Results
In the present study, Total Number of cases was 80, out of which 20 were normal persons. The remaining 60 were suffering from diabetes and hypertension. The age group varied from 42 to 65 years. The study was done in patients who are admitted in Department of medicine.
The cases were discussed under four groups; control subjects, Diabetic group, Hypertensive group and Diabetic and Hypertensive group. In the present analysis the mean values were compared with mean values of control subjects. The mean value of Urinary total protein and Urinary Albumin are significantly raised in study population compared with control subject. There is an I significant ncrease in Urinary total protein, Urinary Albumin and Serum creatinine in both diabetic and diabetic & hypertensive compared with normal control subjects.

Table 1: Comparable values of 24 hrs serum and urine parameters between control and Diabetic group

Parameter	Control group	Diabetic group	P value
Urinary creatinine	0.96±0.41	0.61±0.45	0.004
Urinary total protein	0.15±0.03	0.69±0.62	0.002
Urinary albumin	0.08±0.04	0.22±0.19	0.010
Serum creatinine	0.99±0.14	1,25±0.35	0.008
Serum total protein	6.75±0.10	6.81±0.22	0.15
Serum albumin	3.97±0.24	3.99±0.27	0.78

Table 2: Comparable values of 24 hrs serum and urine parameters between control and Hypertensive group

Parameter	Control group	Hypertensive group	P value
Urinary creatinine	0.96±0.41	1.11±0.56	0.38
Urinary total protein	0.15±0.03	0.52±0.36	0.0003
Urinary albumin	0.08±0.04	0.21±0.15	0.001
Serum creatinine	0.99±0.14	1,16±0.20	0.0009
Serum total protein	6.75±0.10	6.86±0.24	0.085
Serum albumin	3.97±0.24	3.89±0.29	0.40

Table 3: Comparable values of 24 hrs serum and urine parameters between control and Hypertensive and diabetic group

Parameter	Control group	Diabetic & Hypertensive group	P value
Urinary creatinine	0.96±0.41	1.39±0.63	0.021
Urinary total protein	0.15±0.03	0.54±0.41	0.001
Urinary albumin	0.08±0.04	0.35±0.38	0.005
Serum creatinine	0.99±0.14	1.4±0.57	0.0002
Serum total protein	6.75±0.10	6.68±0.23	0.25
Serum albumin	3.97±0.24	3.73±0.40	0.026

Discussion
Diabetic nephropathy occurs in one third of the patients who suffer from Diabetes mellitus. Proteinuria is considered to be the hallmark of the incipient nephropathy in these patients [11]. So, quantifying the urine protein accurately and precisely is vital for monitoring the disease progression in patients who suffer from Diabetes mellitus. An early detection of proteinuria in diabetic patients may help in preventing the irreversible renal damage in these patients [12].
A well-documented test for the diagnosis of proteinuria has been the timed collection of urine over 24 hours [13]. The use of the 24-hours urine collection is necessitated due to variations in the protein excretion throughout the day, since the urinary protein excretion follows a circadian rhythm.

However, the 24-hours urine collection is cumbersome, inconvenient and often incomplete in outpatients [14]. To our knowledge, this is the first study to show that the protein: creatinine ratio measured in spot morning urine specimens tightly correlates with 24 urinary protein excretion rate and accurately predicts rate of decline of glomerular filtration rate and risk of progression to end stage renal failure in non-diabetic patients with protein uria and chronic renal disease In the present study, there are 4-groups namely control (Normal subjects), Diabetic Group, Hypertensive Group, Diabetic and Hypertensive Group. The age varies between 42 to 65 years in all the subjects of all groups. All the parameters like urinary creatinine, total protein, albumin and serum creatinine, serum proteins, and albumin were estimated in all the groups. About 20 cases were studied in the control group who showed normal levels of all the parameters estimated, so they served good controls. 20 cases each were studied under the Diabetic Groups, Hypertensive Group and Diabetic and Hypertensive Group. Urinary Total Protein, Urinary Albumin and Serum Creatinine showed elevated levels. Control Group Values of all parameters are in normal range. Diabetic group compared to normal urinary creatinine value is less. Serum Creatinine is slightly raised. Urinary Total Protein and Albumin is elevated. Serum Total protein and Albumin value is insignificant. The results were shown in table-1. Mean and SD values are high compared to normal of all 3-Urinary parameters are significant. Hypertensive group compared to normal urinary creatinine is less, serum creatinine is slightly raised, and serum total protein and albumin value is insignificant. Urinary Total Protein and Albumin is elevated. The results are shown in table-2. In the present study, all three urinary parameters are significant. Diabetic and hypertensive group compared to normal serum creatinine is slightly raised, serum total protein and albumin value is insignificant. Urinary total protein and llbumin is elevated. The results were shown in table-3. The present results revealed that all Urinary parameters are significantly raised.

All the patients of three groups showed increased levels of Urinary Total Protein and Albumin. Showing much significance statistically when compared with when normal group. All the values are compared with normal. P-Values for all the urinary parameters are highly significant as they are less than 0.05. P-Value is significant for Serum Creatinine and Serum Albumin in Diabetic and Hypertensive Group.

The results indicate the extent of damage of the glomerular Membrane. Due to damage of Glomerular membrane, loss of total protein and albumin is higher in diabetic and Hypertensive group compare to normal. P Values of Diabetes with Hypertension are more significant than only Diabetes or Hypertension. Today, the results of many studies [1, 15, 16&17] indicate that proteins filtered through the glomerular capillary may have intrinsic renal toxicity which, together with other independent risk factors such as hypertension, can have a contributory role in the progression of renal damage [18, 19]. In the present series, evidence of an highly significant correlation between baseline 24hour urinary protein excretion rate and the rate of the glomerular filtration rate during follow up corroborates this hypothesis. On the other hand, evidence that the protein: creatinine ratio even more accurately than 24 hour urinary protein excretion predicted the rate of decline suggests that the ratio as compared with 24 hour urinary protein is a more precise indicator of the kidney traffic of plasma proteins.

Conclusion

The statistical analysis of above data showed that there was significant elevation of Urinary total protein and albumin levels in all three group of Diabetic and Hypertensive patients indicating damage of gloumerular membrane. Estimation of urinary creatinine, Total Protein and Albumin in 24-hours urine gives valuable information in early diagnosis can lead to prognostic assessment and suitable treatment as early as possible which can prevent further damage to gloumerular membrane in particular and other systems of diabetic hypertensive patient.

Acknowledgement

The author thankful to Department of laboratory medicine in SIMS provide the facilities to carry out the research work.

Conflict of Interest

The author declare that they have no conflict of interest

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