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ESTIMATION OF URINARY PROTEIN CREATININE INDEX IN DIABETES MELLITUS PATIENTS WITH MORE THAN 10 YEARS OF DIABETIC HISTORY

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Abstract: **Diabetes is a group of metabolic diseases in which there are high blood sugar levels over a prolonged period. The chronic hyperglycemia of diabetes is associated with long-term damage, dysfunction, and failure of different organs, especially the eyes, kidneys, nerves, heart, and blood vessels.[1] Proteinuria has been recognized as one of the earliest signs of renal function deterioration in diabetes mellitus. In the present study, an attempt has been made to establish the PCI in random urine samples, as a convenient, quick and reliable method for the estimation of proteinuria in diagnosing and monitoring diabetic nephropathy. An attempt has also been made to determine the optimal cutoff value of PCI for the prediction of significant proteinuria. This study signifies the role of urinary PCI in detecting even a minor increase in the protein excretion in a random urine specimen. Total of 50 samples were collected from the IPD and OPD patients, 25 patients aged 25 to 65 years, who were diagnosed as diabetics and were confirmed by the estimation of fasting serum glucose (>126 mg/dl). 25 normal healthy subjects, age and sex matched with the diabetic patients, were selected as the Controls. The comparison of urinary protein, urinary creatinine and urinary protein:creatinine index(PCI) between control and study group was analyzed using unpaired "t"-test. The urinary protein levels were significantly increased, urinary creatinine levels were decreased and urinary protein: creatinine index was significantly higher in diabetic patients as compared to the control group. The present study suggests that random urinary PCI can be a good predictor of significant proteinuria in long term suffering patients of diabetes mellitus during diabetic nephropathy. This test could be a reasonable alternative to the 24-hour urine sample collection for the detection of significant proteinuria in diabetes mellitus patients.**

Keywords: **Diabetes mellitus, Protein, Proteinuria.**

1. INTRODUCTION

Diabetes mellitus, commonly referred to as diabetes, is a group of metabolic diseases in which there are high blood sugar levels over a prolonged period.[2] Symptoms of high blood sugar include frequent urination, increased thirst, and increased hunger. If left untreated, diabetes can cause many complications.[3] Acute complications include diabetic ketoacidosis and non ketotic hyperosmolar coma.[4] Serious long-term complications include cardiovascular disease, stroke, kidney failure, foot ulcers and damage to the eyes.[3] Diabetes is due to either the pancreas not producing enough insulin or the cells of the body not responding properly to the insulin produced.[5] Damage to the kidneys, known as diabetic nephropathy, can lead to tissue scarring, urinary protein loss, and eventually chronic kidney disease. Proteinuria has been recognized as one of the earliest signs of renal function deterioration in DM. Proteinuria occurs due to alterations in the glomerular

permeability and later, due to a failure in the reabsorption of filtered protein by the tubular cells. Normally, most of the healthy adults excrete 20-150 mg of protein in urine over 24 hours.[6] In DM, 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.[7] In an attempt to fulfill the need for a reliable and quick measurement of the urinary protein, various researchers have proposed the calculation of ratios such as the Urinary Protein/Urinary Creatinine (UP/UC), the Urinary Albumin/Urinary Creatinine (UA/UC) and the PCI on spot urine samples.[8] These parameters take into account the fact that the creatinine excretion remains fairly constant in the presence of a stable Glomerular Filteration Rate (GFR), thus, eliminating the variations in the urinary protein concentration during the day. Good correlation has been found between the results of proteinuria which were obtained from these parameters and those which are calculated from the 24-hour urine samples.[9] Proteinuria is conventionally detected by qualitative tests, e.g. the sulfosalicylic acid test, Heller's nitric acid test and the heat coagulation test. But, these tests are not sensitive enough to detect the microproteinuria which is seen during the initial stages of diabetic nephropathy.[6] In this study, an attempt has been made to establish the PCI in random urine samples, as a convenient, quick and reliable method for the estimation of proteinuria in diagnosing and monitoring diabetic nephropathy in newly diagnosed diabetes mellitus patients. An attempt has also been made to determine the optimal cut off value of PCI for the prediction of significant proteinuria. This study signifies the role of urinary PCI in detecting even a minor increase in the protein excretion in a random urine specimen.

2. MATERIAL AND METHODS

2.1 Ethical clearance:

This study was carried out in the Department of Biochemistry in collaboration with the Department of Medicine NIMS Medical College and Hospital, Shobha Nagar, Jaipur, Rajasthan. The institutional ethical clearance was obtained from Ethical Committee of the college.

2.2 Study population:

A total of 50 subjects were enrolled into this study after obtaining informed consent. Among the subjects recrited, 25 were diagnosed with diabetes mellitus based on fasting serum glucose (> 126 mg/dl) for at least two occasions. Rest of the subjects was non-diabetics who served as an age matched control group. All the participants were enrolled from out-patient department of Medicine, NIMS Medical College and Hospital. All the relevant demographic data and clinical history were obtained by verifying patient records. Exclusion criteria involved, Surgical patients**,** Pregnant women**,** ICU admitted patients**,** Chronic hypertensive patients, Children**,** Urinary tract infections**,** Emotional or physical stress**,** Strenuous exercise. 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.

2.3 Quantitative determination of total urinary protein:

Protein reacts in acid solution with pyrogallol red and molybdate to form a coloured complex. The intensity of the colour formed is proportional to the protein concentration in the sample.[9]

2.4 Estimation of urinary creatinine:

The assay is based upon the reaction of creatinine with the sodium picrate as described by Jaffe. Creatinine reacts with alkaline picrate forming a red complex.The time interval chosen for measurements avoids interferences from other constituents of the specimen (urine sample). The intensity of colour formed is proportional to the creatinine concentration in sample.[10]

2.5 Calculation of the Protein Creatinine Index (PCI):

[11] The urinary PCI will be calculated by the following equation:

$$\text{Urinary PCI} = \frac{\text{Urinary protein (mg/L)}}{\text{Urinary creatinine (mmol/L)}} \times 10$$

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2.6 Statistical analysis:

Statistical analysis was done, using IBM SPSS 21 for Windows software Microsoft Excel 2007 and scientific calculator. Student t-test was applied on the data of the case history taken from the patients from the NIMS HOSPITAL and correlation of both the urinary protein and urine creatinine was studied. The normal range of the urinary PCI was calculated from the data which was obtained from the urine samples from normal healthy subjects. The Student's't' test was used to compare the PCIs of the normal healthy controls and the diabetic patients. The results were expressed as Mean ± Standard Deviation (SD). The comparison of urinary protein, urinary creatinine and urinary protein:creatinine index(PCI) between control and study group was analyzed using unpaired "t"-test. Statistical significance was considered to be significant at a p value of <0.05.

3. OBSERVATIONS AND RESULTS

Table1. Comparison of study variables between control and diabetic patient group (More than 10 years of diabetic history)

Parameter	Control Group (n=25)	Study Group (n=25)	P-value
Urinary Protein (mg/dl)	**9.89 ± 3.92**	**50.47 ± 28.06**	**<0.001 ***
Urinary Creatinine (mmol/dl)	**0.82 ± 0.42**	**0.53 ± 0.19**	**<0.05 ***
PCI	**136.43 ± 46.52**	**909.19 ± 250.06**	**< 0.001***

PCI; Protein:Creatinine Index ,SD; Standard deviation, Values are mean ± SD * = significant.

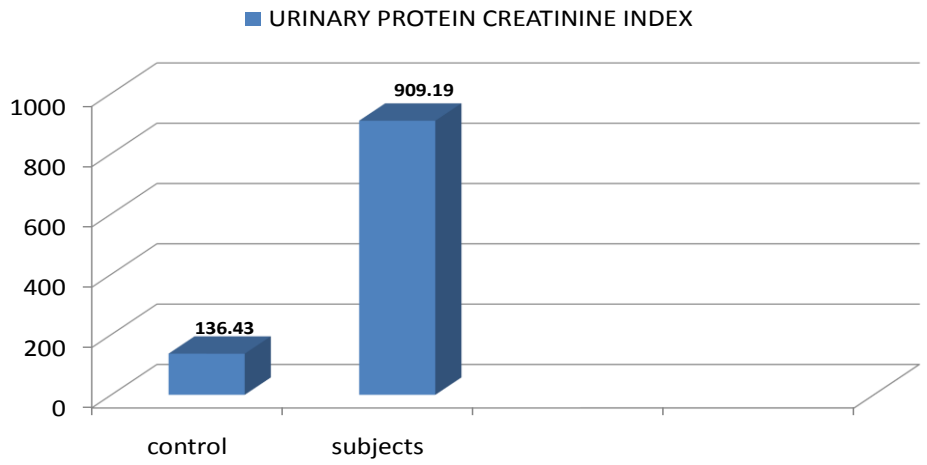

Fig.1- Comparison of urinary PCI (Protein:Creatinine Index) between control and diabetic group (More than 10 years of Diabetic history) by Students` t-test.

4. DISCUSSION

In the present study the normal range of urinary PCI which was obtained from the control group is 63 -200 and the mean urinary PCI obtained for this group was found 136.43 ± 46.52. The level of urinary protein, urinary creatinine and protein creatinine index (PCI) among the study group showed significant difference when compared with control group ($p<0.05$). The mean urinary protein concentration which was found in the diabetic group (More than 10 years of Diabetic history) was (50.47 ± 28.06), and in the control group, it was (9.89 ± 3.92 mg/dl). The protein excretion in the spot urine samples in the diabetic group was found to be significantly higher in comparison to that in the control group, with a p value of < 0.001. The amount of creatinine which was excreted in urine in diabetes mellitus patients (0.53 ± 0.19) was significantly low as compared to that excreted in urine in the control subjects (0.82 ± 0.42 mmol/dl), with p-value <0.05. A significantly increased value of the PCI was observed in diabetic patients (More than 10 years of Diabetic history) (909.19 ± 250.06) as compared to that in the control group, where the PCI was (136.43 ± 46.52) ($p<0.001$).

Biradar et al, reported that the urinary protein excretion was significantly elevated in type 1 and type 2 diabetes mellitus patients. The mean values of the 24-hour urinary protein which were obtained in their study was 1.6 ± 1.7 gm/day, which correlated well with the P:C ratio of 1.27 ± 1.55.[12]

Anoop Kumar et al. conducted a similar study in diabetic patients irrespective of the duration of disease and found that the normal range of the PCI was 60 to 220. Significantly higher amount of proteins were found to be excreted in urine in diabetic patients (25.37 ± 12.51 mg/dl) as compared to those in normal subjects (8.93 ± 3.54 mg/dl). On comparison of the PCI between the controls and the diabetic subjects, it was found to be significantly elevated in the Diabetes mellitus patients (controls = 114.65 ±47.97 and in the diabetic patients =373.04 ± 98.53) ($p < 0.001$).[13]

5. CONCLUSION

The present study suggests that random urine PCI can be a good predictor of significant proteinuria in diabetic nephropathy. This test could be a reasonable alternative to the 24-hour urine sample collection for the detection of significant proteinuria in diabetes mellitus patients with more than 10 years of disease history. It is recommended that the PCI should be specially employed for the assessment for microproteinuria in diabetic patients, when in a few instances a negative result may be obtained by the semi-quantitative dipstick test. The simplicity, accuracy and the lower cost of the PCI justifies its preferential diagnostic use.

REFERENCES

[1] Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Report of the Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Diabetes Care 1997; 20: 1183– 1197.[PMC-NCBI].cited2015.

[2] Diabetes Programme. [World Health Organization]. About diabetes. [Updated 4 April 2014. Cited 2015].

[3] Diabetes Fact sheet No.312. [World Health Organization]. Diabetes [Updated January 2015. Cited2015].

[4] Kitabchi, Umpierrez. Hyperglycemic crises in adult patients with diabetes. Diabetes Care 32 (7); 2009.

[5] Shoback, edited by David G. Gardner, Dolores (2011). Greenspan's basic & clinical endocrinology (9th ed.). New York: McGraw-Hill Medical. p7-8.

[6] Marshall WJ, Bangert SK. Clinical Biochemistry Metabolic and Clinical Aspects. 2nd Ed. Churchill Livingstone Elsevier L.; 2008. p. 329.

[7] Abuelo JG. Proteinuria: Diagnostic principles and procedures. Annals of Internal Medicine.1983;98:186–91.

[8] Keave WF, Eknoyan G. Proteinuria, albuminuria, risk, assessment, detection, elimination (PARADE).A position paper of the National Kidney Foundation. Am J Kidney Dis. 1999;33:1004–10.

[9] Matrix HJ, Chi-Yuan HSU. Use of albumin/creatinine ratio to detect microalbuminuria: implications of sex and race. J Am SocNephrol. 2002;13:1034–39.

[10] Keave WF, Eknoyan G. Proteinuria, albuminuria, risk, assessment, detection, elimination (PARADE).A position paper of the National Kidney Foundation. Am J Kidney Dis. 1999;33:1004–10.

[11] Mc Murray JR.In. Varley's Practical Clinical Biochemistry. 6th Ed. Butterwath- Heinemann Ltd 2006;436-51.

[12] Biradar SB, Kallaganad GS, Rangappa M, Kashinakurti SV, Retnakaran R. Correlation of spot urine protein-creatinine ratio with 24-hour urinary protein in type 2 diabetes mellitus patients : a cross sectional study *JRMS* 2011.16(5):6334-39.

[13] Kumar Anoop, Kapoor Sandeep and Gupta R.C; Journal of clinical and diagnostic research,ISSN-0973-709X; Comparison of Urinary Protein: Creatinine Index and Dipsticks for Detection of Microproteinuria in Diabetes Mellitus Patient; Published online Apr 1, 2013; 7(4): 622–626.