DOI: 10.7860/JCDR/2014/9758.4613

Original Article

Biochemistry Section

Prevalence of Microalbuminuria among Rural North Indian Population with Diabetes Mellitus and its Correlation with Glycosylated Haemoglobin and Smoking

JUHI AGGARWAL[1], MAYUR KUMAR[2]

ABSTRACT

Introduction: Macrovascular and microvascular complications of diabetes mellitus are a consequence of metabolic derangement mainly hyperglycemia. Diabetic nephropathy being one of them causes end stage renal disease. Hence, to detect renal involvement, microalbuminuria can be considered as an early marker.

Aim: To study mean albumin creatinine ratio (ACR) in type 1 and type 2 diabetes mellitus with respect to HbA1c, duration of diabetes and smoking.

Materials and Methods: Two hundred cases of type 1 and type 2 diabetes mellitus and 100 controls, age and sex matched were included in this study and measured for spot urinary albumin, spot urinary creatinine, fasting plasma glucose and HbA1c.

Results: It was observed that mean ACR was significantly elevated in type 1 and type 2 diabetes mellitus as compared to the controls. Mean ACR increases in diabetics with poor glycemic control, duration of diabetes and smoking.

Conclusion: The early detection of microalbumin in diabetics can significantly reduce the progression of renal complications and before the development of proteinuria.

Keywords: ACR, Diabetes mellitus, Diabetic nephropathy, HbA1c, Microalbuminuria

INTRODUCTION

Diabetes mellitus is one of the most important metabolic disorder whereby morbidity is associated with serious complications which involve macrovascular disease (Coronary artery disease, Cerebrovascular disease and Peripheral Arterial Disease) as well as microvascular disease (retinopathy, nephropathy and neuropathy) [1]. The risk of chronic complications in patients of diabetes mellitus depends on the duration of hyperglycemia and they usually become manifest in the second decade of the development of Diabetes mellitus [2].

Increased intracellular glucose leads to formation of advanced glycosylation end products (AGEs) via non-enzymatic glycosylation of cellular proteins. These advanced glycosylation end products crosslink proteins i.e. collagen, extracellular matrix proteins and hence, promote glomerular dysfunction [3].

In the capillaries of the renal glomeruli, albumin binds to the glycated basement membrane which leads to increased basement membrane thickening and is a characteristic of diabetic microangiopathy [4]. Hence, the development of diabetic nephropathy is initiated. Increased intraglomerular pressure, loss of negatively charged glycosaminoglycans in the basement membrane and increased basement membrane pore size contribute to albuminuria [5].

Also, hyperglycemia increases the expression of transforming growth factor beta (TGFβ) in the glomeruli and of matrix proteins. TGFβ contributes to both cellular hypertrophy and enhanced collagen synthesis, which is seen in diabetic nephropathy [6].

In random spot collection technique, normal albumin excretion is less than 30 µg/mg of creatinine; microalbuminuria is defined as albumin excretion of 30-299 µg/mg of creatinine. Albumin excretion 300 µg/mg of creatinine or higher is called macroalbuminuria. Microalbuminuria is used as a screening test for the presence of Diabetes related kidney disease [7].

Diabetes mellitus causes progressive changes to the kidneys and ultimately results in diabetic nephropathy [8]. This complication progresses over years and may be delayed by aggressive glycemic control. Glycosylated hemoglobin (HbA1c) is measured as a marker of glycemic control [9]. High levels of HbA1c are associated with increased risk of development of microangiopathy in diabetics. Microalbumin measurements are useful to assist in diagnosis at an early stage and before the development of proteinuria [10]. An annual assessment of kidney function by the determination of urinary albumin excretion and of HbA1c for glycemic control is recommended for diabetes patients [11].

The present study evaluates the prelevance of albumin creatinine ratio (ACR) in patients of diabetes mellitus, along with its relation with HbA1c, duration of diabetes and effect of smoking in rural North Indian population, which previously has not been studied.

MATERIALS AND METHODS

The present cross-sectional study was conducted from July 2011 to June 2012 in the Department of Biochemistry, Santosh Medical College & Hospital, Ghaziabad, Uttar Pradesh, India. This study was approved by ethical committee of the institution and informed consent was obtained from all the subjects. Purposive random sampling technique was used for data collection. Patients included in the study were having history of Diabetes mellitus for 10y or more and were undergoing treatment i.e. Oral Hypoglycemics for Diabetes and were normotensive.

Cases having proteinuria which was detectable by dipstick tests and having any evidence of urinary tract infection were excluded from the study.

For statistical significance and comparison, all these cases were divided into three different groups. Their age was ranging from 20-50 years. Urinary albumin, urinary creatinine, fasting plasma glucose and HbA1c levels were measured in all the subjects.

Duration of DM (in yrs.)	No. of Patients (n=90)	ACR (µg/mg of creatinine)	HbA1c (%)	p-value	Remarks
Group1 10-14	58	58.83	7.04	<0.001	S
Group 2 15-19	28	74.96	7.89	<0.001	S
Group 3 ≥20 years	14	92.33	8.45	<0.001	S

[Table/Fig-1]: Comparison of ACR, duration of diabetes and glycosylated hemoglobin (HbA1c) in cases of type 1 diabetes mellitus

Duration of DM (in yrs.)	No. of Patients (n=110)	ACR (µg/mg of creatinine)	HbA1c (%)	p-value	Remarks
Group1 10-14	58	64.45	7.04	<0.001	S
Group 2 15-19	28	86.37	7.89	<0.001	S
Group 3 ≥20 years	14	144.5	8.45	<0.001	S

[Table/Fig-2]: Comparison of ACR, duration of diabetes and glycosylated hemoglobin (HbA1c) in cases of type 2 diabetes mellitus

Group	Habit	No. of Patients	ACR (µg/mg of creatinine)	p-value	Remarks
Type 1 Diabetes mellitus	Smokers	42	89.0	<0.001	S
	Non-smokers	48	25.04		
Type 2 Diabetes mellitus	Smokers	52	125	<0.001	S
	Non-smokers	58	26.38		

[Table/Fig-3]: Comparison of ACR with smoking in cases of type 1 and type 2 diabetes mellitus (s= significant)

Group- I: The Control Group

Hundred normal healthy individuals comprised of 59 males and 41 females which were selected from teaching faculty and para medical staff at Santosh Medical College & Hospital.

Group-II: Type I Diabetes Mellitus

Ninety cases of type I Diabetes mellitus comprised of 47 males and 43 females.

Group-III: Type II Diabetes mellitus

Hundred and ten cases of type 2 Diabetes mellitus comprised of 62 males and 48 females.

The diabetic cases were selected from the OPD patients attending the Diabetes Clinic, Department of Internal Medicine, Santosh Medical College & Hospital, UP, India.

Urine sample was collected in clean, sterile plastic container. Collected urine sample was tested by dipsticks for the presence of frank proteinuria. Dipstick negative urine samples were used for quantitation of albumin and creatinine within 4h of voiding. Urinary albumin was analysed by ion exchange high performance liquid chromatography (HPLC) method [12]. Urinary creatinine was estimated by alkaline picrate, Jaffe's Method [13]. In this study, randomly selected patients went for spot urinary albumin estimation and spot urinary creatinine estimation to detect microalbuminuria by measuring albumin creatinine ratio and HbA1c level estimation was done to analyse the glycemic control in diabetic patients.

Blood samples were collected after 10-12h of fasting under all aseptic precautions. Approximately 2ml of venous blood was drawn by a dry and sterilized syringe, to measure fasting plasma glucose by enzymatic method [14] and HbA1c by immune-inhibition method [15].

STATISTICAL ANALYSIS

Data analysis was done using the Statistical Package for Social Sciences (SPSS) for Windows version 10. Chi-square test and Logistic Regression were used to determine the correlation between ACR with glycosylated hemoglobin, duration of diabetes and smoking. p-value (<0.001) was considered as statistically significant.

RESULTS

Among 200 cases of diabetes mellitus, 35% patients had a familial history of diabetes mellitus. [Table/Fig-1,2] shows the comparison of albumin creatinine ratio (ACR) with glycosylated hemoglobin and duration of diabetes in type 1 and type 2 diabetes mellitus patients respectively. Based on the analysis, it was observed that albumin creatinine ratio (ACR) increased significantly with poor glycemic control and with duration of diabetes ($p < 0.001$).

[Table/Fig-3] shows that albumin creatinine ratio (ACR) was significantly elevated in type 1 and type 2 diabetes mellitus patients with smoking habits as compared to type 1 and type 2 diabetics with no history of smoking.

DISCUSSION

Diabetes mellitus is a global problem. The increased morbidity and mortality in Diabetes mellitus lead to search for marker for early detection of renal complications.

It was reported that out of 135 patients with long standing type I Diabetes mellitus (> 30 years duration) 24.4% patients developed microalbuminuria during 7y follow up period [16]. In a study, 277 patients of type I Diabetes mellitus were followed for a median period of 18y (range 1-21.5y). They found that 33% patients developed microalbuminuria [17].

The DEMAND study found that the overall global prevalence of microalbuminuria was 39% [18]. It has been reported the prevalence of microalbuminura in Japanese type 2 patients was 32% [19].

In a study conducted in Saudi Arabia, the overall prevalence of microalbuminuria was reported in both type I and type II Diabetes mellitus as 49.3% [20]. It was also observed that the prevalence of microalbuminuria was found to be 52.04% among all diabetic patients [21].

It was reported that the risk of developing microalbuminuria in patients with known duration of Diabetes 10-14 years was 4.11 times higher compared with patients with known duration of Diabetes 0-4 years [22].

The level of mean blood glucose, effectiveness of treatment and risk of development of possible long term chronic complications are associated with poor glycemic control. It was stated that urinary microalbumin and HbA1c levels were significantly higher in the diabetic cases [23].

Microalbumin levels were linearly correlated to the duration of diabetes and HbA1c. In our study we observed that elevated levels of HbA1c and microalbuminuria are related with increase in the duration of the disease and was found statistically significant ($p<0.001$). In a recent study it was observed that microalbuminuria has a highly significant correlation with HbA1c ($p<0.05$) [24].

It was also reported that microalbuminuria is related to hyperglycemia and control of blood glucose level prevents the development of nephropathy in type 1 & 2 diabetes [25].

In our study, type I and type II Diabetes mellitus cases who were smokers, the mean albumin creatinine ratio was found to be 89 and 125µg/mg of creatinine respectively while in type I and type

II Diabetes mellitus who were non-smokers, the mean albumin creatinine ratio was found to be 25.04 and 26.38 µg/mg of creatinine respectively [Table/Fig-3].

When the difference of mean albumin creatinine ratio was compared between smoker and non-smoker type I and type II diabetic patients, it was found to be statistically significant ($p<0.001$).

In accordance with our study, it was concluded that smoking is associated with high amounts of carboxyhaemoglobin and decreased oxygen delivery to tissue; albuminuria may be caused by hypoxia in the renal microcirculation [26].

It was found that smoking increases the risk of progression from microalbuminuria to overt proteinuria [27] and that in patients of type I Diabetes mellitus, loss of renal function was slower in those patients who stopped smoking [28]. It was concluded that in patients of type 2 Diabetes mellitus, smoking increases the risk of developing microalbuminuria and they progressed to end stage renal disease twice as quickly when compared with non-smokers [29].

It was identified that smoking was as an important risk factor for the development and progression of microalbuminuria in type 2 Diabetes mellitus patients [30].

CONCLUSION

Our study indicates that increased levels of microalbuminuria have a direct correlation with HbA1c and smoking in diabetic patients. It is also associated with duration of diabetes because of prolonged exposure of hyperglycemia and formation of advanced glycosylation end products. Microalbuminuria can be considered as an early marker of renal involvement in diabetes mellitus and its early detection can significantly reduce the progression of renal complications associated with it. Additionally, there is an urgent call for regular screening of microalbuminuria and HbA1c in both newly diagnosed and already diagnosed diabetes.

REFRENCE

[1] Klein R. Hyperglycemia and microvascular and macrovascular disease in diabetes. *Diabetes Care*. 1995;18: 258-68.
[2] Joshi SR, Parikh HM. India- Diabetes capital of the world: New heading towards hypertension. *J Assoc Physicians India*. 2007; 55: 323-24.
[3] Pavenstadt H, Kriz W, Kretzler M. What is the mechanism of microalbuminuria in Diabetes? *Physiol Rev*.2003; 83: 253-307.
[4] Barnett AH. Origin of the microangiopathic changes in diabetes. *Eye*.1993; 7: 218-22.
[5] Mason RM, Wahab NA. Extracellular matrix metabolismin diabetic nephropathy. *J Am Soc Nephrol*. 2003;14:1358-73.
[6] Reeves WB, Andreoli TE. Transforming growth factor beta contributes to progressive diabetic. 97(14):7667-9, doi: *Nephropathy*. 10:1073/ pnas: 97.14.7667.
[7] Bishop L, Fody P, Schoeff E. Clinical Chemistry- Techniques, Principles, Correlations. 2010;6:326.
[8] Kaushik B, Kutty AVM, Shetty HV. Glycemic control modifies the association between microalbuminuria and C-reactive protein in type 2 Diabetes mellitus. *Indian Journal of Clinical Biochemistry*. 2007;22(2)53-9.
[9] Use of Glycated Haemoglobin (HbA1c) in the Diagnosis of Diabetes Mellitus, WHO/NMH/CHP/CPM/11.1 Abbreviated Report of a WHO Consultation (2011).
[10] Basu S, Chaudhari S, Bhattacharyya M, Chatterjee TK, Todi S, Majumdar A. Microalbuminuria: An inexpensive, non invasive bedside tool to predict outcome in critically ill patients. *Indian Journal of Clinical Biochemistry*. 2010;25(2):146-52.
[11] Naveen P, Kannan N,Vamseedhar A, Bhanu PG, Aravind KR. Evaluation of Glycated hemoglobin and microalbuminuria as early risk markers of nephropathy in type 2 Diabetes mellitus. *Int J Biol Med Res*. 2012;(2):1724-26.
[12] Comper WD, Osicka TM, Clark M, MacIsaac RJ, Jerums G. Earlier detection of microalbuminuria in diabetic patients using a new urinary albumin assay. *Kidney Int*. 2004;65:1850-55.
[13] Bowers LD. Kinetic serum creatinine assays: The role of various factors in determining specificity. *Clin Chem*. 1980; 26:551.
[14] Teitz NW. Fundamentals of clinical chemistry (243), W.B. Saunders and Co. Philadelphia PA 1976.
[15] Jeppson J, Uwe K, John B, Andreas F, Wieland H, Tadao H, et al. Approved IFCC reference method for the measurement of glycosylated hemoglobin in human blood. *Clin Chem Lab Med*. 2002;40(1):78-89.
[16] Chankramath S Arun, James Stoddart, Paul Mackin, Jean M Nadeod, John P New, Sally M Marshal. Significance of microalbuminuria in long duration type 1Diabetes. *Diabetes Care*. 2003;26:2144-49.
[17] Peter H, Lise T, Peter R, Berit RJ, Malene G, Inge T, et al. Predictors for the development of microalbuminuria and macroalbuminuria in patients with type 1Diabetes: inception cohort study. *BMJ*. 2004;328:1105.
[18] Parving HH, Lewis JB, Ravid M, Remuzzi G, Hunsicker LG. DEMAND investigators: prevalence and risk factors for microalbuminuria in a referred cohort of type II diabetic patients: a global perspective. *Kidney Int*. 2006;69:2057-63.
[19] Hiroki Y, Koichi K, Masashi K. Microalbuminuria is common in Japanese Type 2 diabetic patients: A nationwide survey from the Japan Diabetes Clinical Data Management Study Group (JDDM 10). *Diabetes Care*. 2007;30(4):989-92.
[20] Khalid SAG. Microalbuminuria among patients with diabetes type I and type II at the armed forces hospital in Jubail. *Annals of Saudi Medicine*. 2001;21(3-4): 236-40.
[21] Vivek A, Ranjit KS, Parduman S, Arora MM, Somani BL. Incidence of microalbuminuria in hypertensive patients. *Indian J Med Biochem*. 2004;8(2):57-61.
[22] Bahman PT, Abdul SAK, Liza LI, Catherine MZ, William HH. Does microalbuminuria predict Diabetic Nephropathy. *Diabetes Care*. 2001;24:1560-6.
[23] Kundu D, Roy A, Mandal T, Bandyopadhyay U, Ghosh E, Ray D. Relation of microalbuminuria to glycosylated hemoglobin and duration of type 2 diabetes. *Niger J Clin Pract*. 2013;16(2):216-20. doi: 10.4103/1119-3077.110159.
[24] Shehnaz AS, Jawed AB, Tehseen I, Tahseen K, Muhammad B, Syed SH. Prevalence of microalbuminuria with relation to glycemic control in type-2 diabetic patients in Karachi. *J Ayub Med Coll Abbottabad*. 21(3): 83- 86.
[25] Giunti S, Barit D, Cooper ME. Mechanism of diabetic nephropathy. Role of hypertension. *Hypertension*.2006;48:519–26.
[26] Redehill A Fianco, Cerecedo A, Hemseu A, Stensdotter M, Pernow J, Lundberg JM. Cigarette smoking induced elevation of plasma neuropeptide Y levels in Humans. *Clin Physiol*. 1989;9:243-48.
[27] Rossing P, Hougard P, Parving HH. Risk factors for development of incipient and overt diabetic nephropathy in type 1 diabetic patients: a 10 years prospective observational study.*Diabetes Care*. 2002;25:859-64.
[28] Sawicki PT, Didjurgeit U, Muhlhauser I, Bender R, Heinemann L, Berger M; Smoking is associated with progression of diabetic nephropathy. *Diabetes Care*.1994;17:126-31.
[29] Biesenbach G, Grafinger P, Janko O, Zazgonik J. Influence of cigarette smoking on the progress of clinical diabetic nephropathy in type 2 diabetic patients.*Clin Nephrol*.1997;48:146-50.
[30] Gambaro G, Bax G, Fusaro M. Cigarette smoking is a risk factor for nephropathy in type 1 Diabetes. *Diabetes Nutr metab*. 2001;14:337-42.

PARTICULARS OF CONTRIBUTORS:
1. Assistant professor, Department of Biochemistry, Santosh Medical College and Hospital, Ghaziabad, India.
2. Specialist, ESI Hospital, Okhla, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Dr. Juhi Aggarwal,
C-78, Mahendru Enclave, Gate no. 4, Near Hans Cinema, Delhi-110033, India.
Phone : 9990104307, E-mail : jaggarwal38@gmail.com

FINANCIAL OR OTHER COMPETING INTERESTS: None.

Date of Submission: **Apr 25, 2014**
Date of Peer Review: **May 28, 2014**
Date of Acceptance: **Jun 29, 2014**
Date of Publishing: **Jul 20, 2014**