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Original Research

Evaluation of glycated albumin and microalbuminuria as early risk markers of nephropathy in type 2 diabetes mellitus

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Abstract

Introduction: Since glycated albumin (GA) reflects with short term variations and these glycated protein shows degree of hyperglycemia, the objective of the study is to find GA and microalbuminuria as an early risk markers along with the duration of uncontrolled diabetes mellitus in type 2 diabetic nephropathy.
Materials and Methods: The present cross-sectional study included randomly selected uncontrolled type 2 DM [n=75], controlled type 2 DM [n=75] and healthy controls [n=75]. The fasting venous blood was obtained for GA and serum creatinine, while their morning urine sample was obtained for detection of microalbuminuria. Statistical analysis was done using SPSS version 16.0. One-Way ANOVA was performed. All p-values < 0.05 were considered as statistically significant.
Results: The mean GA, microalbuminuria and serum creatinine were the highest in uncontrolled DM when compared with controlled DM respectively. Microalbuminuria and GA had a significant correlation with duration of diabetes ($p<0.0001$).
Conclusion: The present study identifies that the risk of microalbuminuria increases with poor glycemic control. Persistent increase in GA and microalbuminuria may be considered as risk markers in diabetic nephropathy. Therefore, regular screening for microalbuminuria and GA estimation can help in clinical management to prevent complications.

INTRODUCTION

Metabolic derangement syndrome is one of the cause of diabetes mellitus (DM) and it is frequently associated with permanent and irreversible functional and structural changes in the cells of the body, particularly the vascular system changes leading in turn to development of well defined clinical entities called the complications of diabetes mellitus effecting eye, the kidney, microvascular and nervous system. During abnormal glucose homeostasis body fails to produce insulin due to DM which is characterized by hyperglycemia and impairment in all metabolisms due to deficiency of insulin secretion [1]. In type 2 DM >80% were diagnosed with ESRD which is the single most end stage renal disease. Some of the western studies shows out of 44% of ESRD >80% suffering from type 2 DM [2]. Some of the Indian studies show the prevalence of microalbuminuria ranging from 19.7% to 28.5% in type 2 DM [3]. Diabetic

nephropathy is a common consequence of prolong DM which appears involvement and complex interaction between genetic and environmental factors [4]. The pathology basis of elevated urinary albumin excretion due to the protein glycosylation with advanced glycated end products and their deposition results the hypertrophy of glomerular and renal system, which in turn leads to the leakage of low molecular weight proteins (albumin) [5]. The continuous persistent leakage of these proteins in urine results overt diabetic nephropathy results gradual development of ESRD and cardiovascular complications [6]. As testing for glycated albumin (GA) levels has slowly becoming an established practice in different labs. More and more scientific and clinical research has pointed to the GA as associated with diabetes as a direct cause of several significant areas of diabetes complications. It has become clear that GA plays a dual role: as an indicator or marker of intermediate glycation, and as a causative agent of the damage of diabetes complications The present study was carried out to evaluate microalbuminuria in relation to GA and duration of diabetes. Microalbuminuria and GA were measured as risk markers of renal damage and glycemic control respectively.

MATERIALS AND METHODS

The objective of the present study was to determine the prevalence of microalbuminuria in relation to GA and associated risk factors among type 2 diabetic patients. Ethical clearance was obtained from the institutional ethical committee (Regd. No. MAPIMS /958/PO/ac/09/CPCSEA) as well as the oral informed consent from the subjects. The present study was conducted from January 2009 to December 2011.

Inclusion criteria

Among 345 diabetic patients who visited diabetic out patient clinic of tertiary hospital of Kancheepuram and MAPIMS&R, 225 subjects aged between 40-60 years of either sex with known history of type 2 DM chosen [based on the screening recommendation by American Diabetes Association (ADA)]. The study had 3 groups: Group A consisted of patients on default antidiabetic treatment [uncontrolled DM (n=75)], Group B comprised of patients on regular antidiabetic treatment [controlled DM (n=75)] and Group C included age-matched healthy controls (n=75).

Exclusion criteria

Diabetic patients suffering from any other medical problems such as infections chronic kidney disease, hypertension, angina and acute coronary syndrome, coronary bypass surgery or percutaneous coronary interventions were excluded from the study.

Purposive random sampling technique was used for data collection. Venous blood samples were collected after 12 hours fasting into two test tubes; with no anticoagulant for serum creatinine, and with anti coagulant for FBG analyzed in Olympus AU 400 auto analyzer. Serum creatinine was analysed by alkaline picrate, Jaffe's Method (Biocon® Kit, Germany) Plasma GA levels were measured by an enzymatic method using albumin specific protease, ketoamine oxidase and albumin assay reagent on the Hitachi Autoanalyser (Lucica GA-L, Asahi Kasei Pharma Corp, Tokyo, Japan) [8,9]. GA was hydrolyzed to amino acids by albumin specific proteinase and then oxidized by ketoamine oxidase to produce hydrogen peroxide, which was measured quantitatively. The GA value was calculated as the percentage of GA relative to total albumin, which was measured with bromocresol purple method. Twenty four hour urine sample was collected in a container (without preservative) for analysis of albumin. Microalbuminuria was estimated by ion-exchange high performance liquid chromatography (HPLC, sigma Aldrich Ascentis® Chennai)

Statistical analysis

The statistical analysis was done by using SPSS version 16.0. One-Way ANOVA method was applied to observe association of microalbuminuria with GA and duration of diabetes. P value < 0.0001 was considered as statistically significant.

RESULTS

Among the 150 Type 2DM patients studied [controlled and uncontrolled groups], 45% had a family history of diabetes and male:female ratio was 1.17:1. In Table 1, the glycemic control of Group A, Group B and Group C were compared between diabetic patients and healthy controls with serum creatinine levels (The recommended reference level for glycated albumin was by ADA) [7]. Microalbuminuria was compared between controlled and uncontrolled diabetic patients in Table 2. Based on Table 1 and Table 2, the Microalbuminuria increased significantly with poor glycemic control and correlated with elevated serum creatinine levels indicating renal damage ($p<0.0001$). The parameter of the studied groups according to duration of diabetes was summarized in Table.3. In type 2 DM patients, microalbuminuria and glycemic control have shown a significant linear correlation with duration of diabetes ($p<0.0001$)

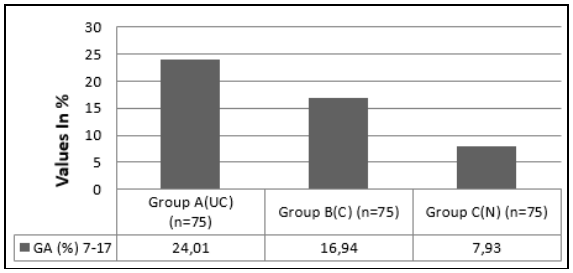

Figure 1. Comparison of glycated albumin levels between Group A, Group B and Group C.

GA: F ratio – 392.41, degree of freedom- 2, p value < 0.0001.
Group A (UC) = uncontrolled blood sugar level in type 2 DM with increased GA levels
Group B (C) = controlled blood sugar level in type 2 DM with moderate increase GA levels
Group C (N) = normal subjects with normal GA levels

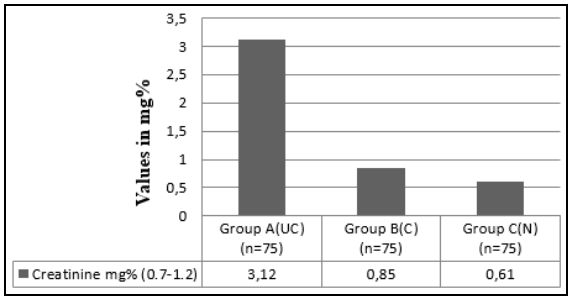

Figure 2. Comparison of serum creatinine levels between Group A, Group B and Group C.

Serum Creatinine: F ratio – 65.19, degree of freedom - 2, p value < 0.0001.
Group A (UC) = uncontrolled type 2 DM subjects with altered creatinine levels
Group B (C) = controlled type 2 DM subjects with normal creatinine levels
Group C (N) = normal subjects with creatinine levels with in the range

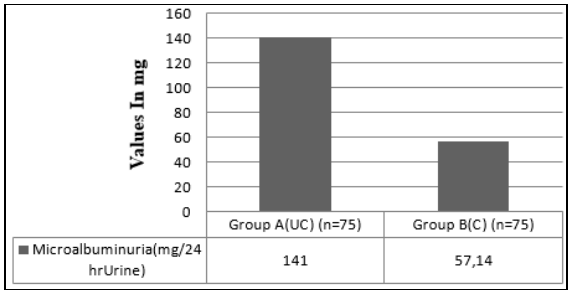

Figure 3. Comparison of microalbuminuria between Group A (uncontrolled) and Group B (controlled)

Microalbuminuria: F ratio – 67.82, degree of freedom - 1, p value < 0.0001.
Group A (UC) = uncontrolled type 2 DM subjects showing higher percentage of microalbuminuria
Group B (C) = controlled type 2 DM subjects showing microalbumunuria with in the range
Group C = Microalbuminuria was not seen, they were normal subjects

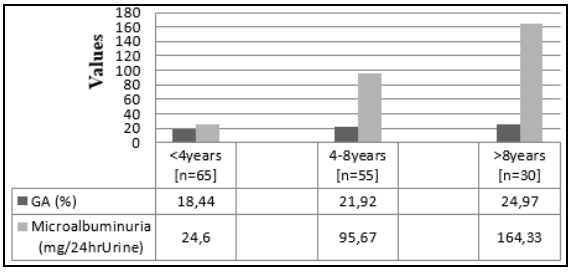

Figure 4. Duration of uncontrolled type 2 DM with microalbuminuria and glycated albumin

F ratio – 408.50, degree of freedom – 2, p value < 0.0001.
Group 1 =levels of GA and the prevalence of microalbuminuria in uncontrolled type 2 DM less than 4 years
Group 2 = levels of GA and the prevalence of microalbuminuria in uncontrolled type 2 DM between 4-8 years
Group 3 = n levels of GA and the prevalence of microalbuminuria in uncontrolled type 2 DM more than 8 years

DISCUSSION

Our study presents data on prevalence and associations of microalbuminuria with altered glycated albumin levels in type-2 diabetes mellitus. The prevalence of microalbuminuria in our study is 37%, higher in to the study by Ghai *et al.* where prevalence was reported at 25%, when compared [10]. The prevalence of microalbuminuria in the present study is high because of the fact that most of the patients were on irregular treatment with poor glycemic control and the small sample size. It shows that the good glycemic control is the strong influencing factor playing a key role in transition of normicroalbumunuric subjects to microalbuminuric. Present study has shown statically significant linear relationship of degree of albuminuria with altered GA levels, because of factors like duration of diabetes and glycemic control. GA plays a double role in diabetes complications. In addition to being a marker for glycation, glycated albumin has been directly implicated for a role in several major complications of diabetes, including atherosclerosis, nephropathy, retinopathy and cognitive function [11]. Several recent studies have confirmed that point measurements of GA and glycated hemoglobin (HbA1c) are closely correlated, and that values for glycated albumin accurately represent the equivalent values for HbA1c. Levels of GA change more rapidly over time in response to changes in treatment (as reflected by changes in fasting plasma glucose) than do levels of HbA1c [12,13]. The normal cut off value for GA in our population was derived using control group and it was 15% (range 7-17%). Our cut off value was within the range reported in the Japanese population. The reference interval of GA was 12.3-16.9% according to Japan Diabetes Society. The normal GA % range in US population (11.6±1.6%) was slightly

lower than our cut off value [14]. Protein glycation is both a marker for diabetes complications and an underlying cause of those complications. The purpose of diabetes monitoring is to help diabetics control glycation. Diabetes is currently monitored by a combination of daily testing of short term monitoring of blood glucose (SMBG) and long-term testing (HbA1c). A monthly diabetes monitoring test based on GA has the potential to provide better information for monitoring glycation [15].

Our study shows negative correlation of microalbuminuria with creatinine clearance which is insignificant statistically. And all the subjects in our study found with normal creatinine clearance and serum creatinine levels. Depending up on the renal hemodynamics and systemic blood pressure, urinary findings, and susceptibility to therapeutic interventions diabetic nephropathy categorized into different stages. There will be elevated glomerulation with no albuminuria in initial renal hyperfusion stage. In the second stage called clinical latency stage there will be increased glomerular filtration with no albumin in urine. Third stage called incipient nephropathy with normal glomerular filtration and presence of microalbuminuria which used to occur in cases suffering with DM more than five years. There will be decrease in glomerular filtration, macroproteinuria and other clinical manifestations of nephropathy in the subsequent stage finally ends up with ESRD with suppressed glomerular filtration and very high albumin levels in urine [6]. By with the above discussion we can conclude that microalbuminuria may not associate with abnormal creatinine or creatinine clearance.

Present study has shown positive correlation of microalbuminuria with duration of diabetes mellitus with altered GA levels which is in accordance with many previous reports. There is a direct relation between the duration of diabetes and with the development of microalbuminuria, because of prolonged exposure to hyper glycemia as well as deposition of advanced glycated end products. The regular treatment for controlling diabetes will also play a crucial role in the development of type 2 diabetic nephropathy [16-18].

CONCLUSION

Our study indicates that increased levels of GA and microalbuminuria reflect a quicker response to short-term changes in diabetes treatment and best glycemic index in uncontrolled diabetes mellitus. This could be due to default treatment therefore regular screening of GA and microalbuminuria should be performed at every one month in addition to HbA1c. There is a demonstrated need for an intermediate glycation index to monitor diabetes. A test based on GA can provide a stable monthly index of glycemic control.

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SOURCE OF SUPPORT

Nil

CONFLICT OF INTEREST

None declared

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