

UTILIZATION OF GLYCOSYLATED HAEMOGLOBINAND MICROALBUMIN IN THE MANAGEMENT OF UNCONTROLLED DIABETES MELLITUS PATIENTS ATTENDING KIBAGABAGA DISTRICT HOSPITAL, RWANDA

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ABSTRACT

Background: Diabetes Mellitus is a clinical syndrome described by hyperglycemia because of the total or relative lack of insulin. One of chronic complication of poorly controlled diabetes is diabetic nephropathy which may prompt end-stage renal disease. **Objective:** To establish a correlation between glycosylated haemoglobin and microalbumin in the management of uncontrolled diabetes mellitus patients attending Kibagabaga District Hospital. **Materials and Methods:** Hospital-based cross-sectional design was used. Data was collected from 246 participants that attended the diabetic outpatient diabetic clinic at Kibagabaga District Hospital from March to June 2019. Spot urine was collected to measure microalbumin. Microalbumin was used as urinary albumin-to-creatinine ratio (ACR) > 3 mg/mmol. The Whole blood samples were collected in Ethylenediamine tetraacetic acid (EDTA) tube for glycosylated haemoglobin (HbA1c) measurement. A cut-off value of HbA1c > 6.5 % was used as uncontrolled diabetes mellitus. Data were analyzed by statistical software" SPSS" version 21. **Results:** Out of 246 patients, 178 (72.36%) were female

and 68 (27.64%) were male. The youngest and oldest had 31 and 81 years old respectively and the average age was 57.9 years. The duration of diabetes varies from 1 to 19 years with an average of 6.5 years. 113 (31.5%) patients with (HbA1c <6.5%) were excluded from the study during screening and 246 (68.5%) patients with uncontrolled diabetes (HbA1c > 6.5%) were included in the study. 80 (32.9%) of study subjects had microalbumin (ACR>3mg/dl) and 166 (67.1%) had normoalbuminuria (ACR<3mg/dl) and no case of macroalbuminuria (ACR>100 mg/dl) recorded. ACR was significantly positively correlated with HbA1C and duration of diabetes. **Conclusions:** Our study revealed that around 68.5% of the diabetic patients were uncontrolled and 32.9% of them had microalbumin. The significant positive correlation between ACR and HbA1c results among uncontrolled diabetes patients helped us to propose the access to glycosylated haemoglobin and microalbumin testing at the district hospital level. This will also assist to predict the kidney complication among uncontrolled diabetes patients.

Key Words: *diabetes nephropathy, glycosylated haemoglobin, microalbuminuria*

INTRODUCTION

Diabetes mellitus is a heterogeneous arrangement of metabolic deregulation depicted by hyperglycemia (Fac & Naiss, 2005). In 2011 the estimate has risen to 366 million, and the projection for 2030 is 552 million. The biggest increment (92%) has been anticipated for countries in the lowest income group (Babu Kondaveeti, Kumaraswamy, Mishra, Aravind Kumar, & Anand Shaker, 2013). World Health Organization, 2016 reported 1.5 million deaths diabetes alone and 2.2 million deaths due to diabetes complications in 2012. Forty-three percent of these 3.7 million passings happen before the age of 70 years. Hyperglycemia is a typical

impact of uncontrolled diabetes mellitus is related with the development of various inconveniences, the fundamental ones being nephropathy, neuropathy, retinopathy, and cardiovascular diseases (Kinmonth, Wareham, & Williams, 2010).

Diabetic nephropathy influences around 15 to 25% of type 1 diabetic patients and 30 to 40% of patients with type 2 diabetes (Schrijvers, De Vriese, & Flyvbjerg, 2004). In Rwanda, the prevalence of diabetic nephropathy is not known. However, the hospital-based study conducted by Rudasingwa et al., 2012 on diabetes complications have found diabetes mellitus microvascular neuropathy (53%), retinopathy (23%), and nephropathy (20%), and macrovascular complications (4%) diagnosed based on clinical findings due to lack of reagent to test glycosylated haemoglobin and microalbumin. Jamison et al., 2006 have confirmed that very many countries in Sub Sahara cannot afford screening and treatment for DM complications. The fasting blood sugar (FBS) is used by district hospital laboratories has been appeared to be inconsistent because of the many reasons which are known to influence glucose levels in the blood (ie. age, time of day, push, suppers, and so forth.). Also, it has been observed that diabetic patients may fleetingly enhance their consistency preceding facility visits and in this way information in light of center checking may yield one-sided appraisals of their level of glucose control (“World Health Organization Laboratory Diagnosis and Monitoring of Diabetes Mellitus,” 2002). Timely and accurate glycemic control may prevent the anticipation or delay diabetic nephropathy. The regular glycemic control likewise decreases the occurrence of micro and macroalbuminuria by 39% and 54%, respectively (Dounousi et al., 2015). The current study covered the laboratory test gap for uncontrolled diabetes mellitus monitoring to evaluate the treatment efficacy and early kidney complication diagnosis at Kibagabaga District Hospital. In addition, the obtained results helped to establish the correlation between HbA1C and microalbumin in the management of uncontrolled diabetic patients attending Kibagabaga District Hospital.

MATERIALS AND METHODS

This study was a hospital-based cross-sectional study. Ethical approval was granted by the Institutional Research and Ethics Committee of the University of Rwanda (college of medicine and health sciences). All participants were explained about the research and only who were willingly signed the consent form were recruited. 359 type 2 diabetes mellitus patients attended internal medicine consultation at Kibagabaga District Hospital during the study period from March 2019 to June 2019 were recruited. 113 (31.5%) patients with (HbA1c <6.5%) were excluded from the study during screening and 246 (68.5%) patients with uncontrolled diabetes (HbA1c > 6.5%) were included in the study. In all study patients, a demographic data was captured (gender, sex and diabetes duration) before sample collection. 5 ml of fasting venous blood was collected from the antecubital vein of each study subject in EDTA tube for glycosylated haemoglobin levels. A random urine sample was collected in the sterile labeled container for determination of microalbumin using the albumin-to-creatinine ratio method. The specimens were labeled using permanent marker to assign participant the research unique identifier. The participants were advised that the only who will have microalbumin will repeat after 3 months for confirmation. Specimens

were transported from Kibagabaga District Hospital to the processing laboratory at the University of Rwanda in the College of Medicine and Health Sciences in the laboratory of Biomedical Laboratory Sciences on ice packed cool boxes within 1 hour. Urine samples were aliquoted in duplicate vials. All specimens were then stored at 2-8oC awaiting laboratory testing. Albumin, creatinine (in urine) and HbA1c (whole blood) was measured by an immunoturbidimetric method using an automatic analyzer (COBAS C111, Roche Diagnostics GmbH, Mannheim, Germany). Microalbumin was calculated using the ACR ratio method of random urine spot.

An HbA1c > 6.5 % was considered to be uncontrolled while ACR >3.0 mg/L was considered as microalbumin. The data were analyzed using SPSS version19.0. Pearson correlation coefficient was calculated to find the linear relation between HbA1C and microalbuminuria. P value was taken as significant at 5 percent confidence level (P<0.05). Significance level represented as; (*) less significant (p<0.05) (**) significant (p<0.01) (***) highly significant (p<0.001).Data were presented in the form of tables, graphs, and charts.

RESEARCH RESULTS

A total of 359 participants were enrolled in this study. After testing the HBA1c levels 113 patients (HBA1c <6.5%) were excluded from the study. 246 patients with uncontrolled diabetes mellitus were followed up between March to June 2019.

Table 1: Demographic Data

Gender	Number (n)	Percentages	Age		Diabetes duration
Male	68	27.6	Mean	54.69	4.99
			95% CI	52.61-56.77	4.07-5.90
			Std	8.589	3.767
			Min	38	1
			Max	81	14
			Range	43	13
			IQR	11	4
			Female	178	72.4
			95% CI	57.77-60.57	6.33-7.84
			Std	9.468	5.102
			Min	31	1
			Max	79	19
			Range	48	18
			IQR	13	10

Out of 246 patients, 178 (72.36%) were females and 68 (27.64%) were males. Mean \pm SD of ages in years for males and females were 54.69 ± 8.589 and 57.77 ± 9.468 , respectively. Mean \pm SD of the duration of diabetes in years for males and females was 4.99 ± 3.767 and 7.08 ± 5.102 , respectively.

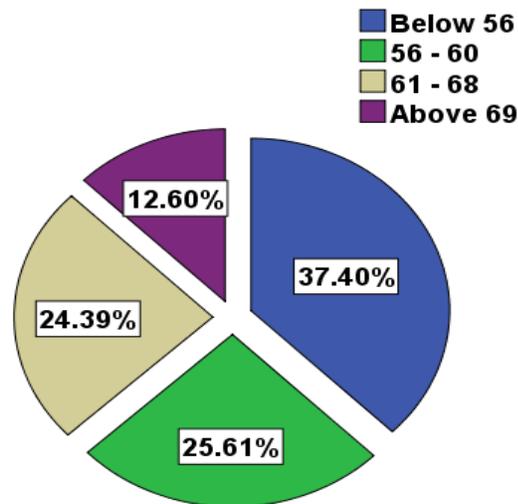


Figure 1: Age Category (years)

The above chart shows that the ages are categorized into below 56, 56-60, 61-68 and above 68. The majority of study subjects were aged below 56 years (37.4%).

Table 2: Age Category versus Diabetes Duration (Years) Category

Age Category	Diabetes Duration (Years) Category				Total
	1 – 2	3 – 4	5 - 7	8 – 19	
Below 56	28 (11.3%)	25 (10.1%)	25 (10.1%)	14 (5.69%)	92 (37.3%)
56 – 60	17 (6.91%)	10 (0.40%)	14 (5.69%)	22 (8.94%)	63 (25.6%)
61 – 68	18 (7.31%)	7 (2.84%)	8 (8.25%)	27 (10.9%)	60 (24.3%)
Above 69	4 (1.62%)	0 (0.00%)	3 (1.21%)	24 (9.75%)	31 (12.60%)
Total	67 (27.23%)	42 (17.0%)	50 (20.3%)	87 (35.3%)	246

Age and diabetes duration were classified into 4 categories: aged below 56 (37.39%), age ranged between 56 to 60 (25.60%), age grouped from 61 to 68 (24.39%), aged over 69 years (12.60%) and the diabetes duration age ranged from 1-2 (27.23%), 3-4(17.0%), 5-7(20.3%), and 8-19 (35.3%). This was statically significant since p-value < 0.05.

Table 3: Microalbumin levels in the Study Population

	Normoalbumin <3 mg/dl N= (%)	Microalbumin >3mg/dl N= (%)	Total %	p-value
Age category				.000
Below 56	32.9%	4.5%	37.4	
56 – 60	17.5%	8.1%	25.6	
61 – 68	13.4%	11.0%	24.4	
Above 69	3.3%	9.3%	12.6	
Total	67.1%	32.9%	100	
Gender				0.004
Male	22.4%	5.3%	27.7	
Female	44.7%	27.6%	72.3	
Total	67.1%	32.9%	100	
Diabetes duration years				.000
1 – 2	27.2%		27.2	
3 – 4	17.1%		17.1	
5 – 7	19.1%	1.2%	20.3	
8 – 19	3.7%	31.7%	35.4	
Total	67.1%	32.9%	100	

Of the study population, 67.1% had normalbumin while 32.9% had microalbumin (27.6% females and 5.3% males) and no case of macroalbumin was recorded. Normalbumin and microalbumin cases had diabetes duration ranging from 1 to 19 and 5-19 years respectively. This was statistically significant (p-value <0.05).

Table 4: Age Category VS Glycosylated Haemoglobin (Hba1c) (%) Category

Count		Glycosyalated Haemoglobin (Hba1c) (%) Category				Total
		6.5-7.08	7.08-7.95	7.96-11.55	Above 11.55	
Age Category	Below 56	25 (10.16%)	20 (8.13%)	38 (15.44%)	9 (3.65%)	92 (37.40%)
	56– 60	15 (6.09%)	9 (3.65%)	22 (8.94%)	17 (6.91%)	63 (25.60%)
	61 – 68	12 (4.88%)	10 (4.06%)	15 (6.09%)	23 (9.35%)	60 (24.40%)
	Above 69	3 (1.21%)	2 (0.81%)	4 (1.63%)	22 (8.95%)	31 (12.60%)
Total		55 (22.35%)	41 (16.67%)	79 (32.11%)	71 (28.87%)	246

Out of the 246 uncontrolled diabetic patients 55 (22.35%), 41(16.66%), 79 (32.11%), and 71(28.86%) had HbA1c 6.5-7.08 %, 7.08-7.95%, 7.96-11.55 and above 11.55 respectively. Majority had age below 56 years old (37.40%) and HbA1c ranging between 7.96-11.55 (32.11%). This was statistically significant since p-value <0.05.

Table 5: Gender Vs Glycosylated Haemoglobin (Hba1c) (%) Category

Count		Glycosylated Haemoglobin (Hba1c) (%) Category				Total
		6.5-7.08	7.08-7.95	7.96-11.55	Above 11.55	
Gender	Male	27 (10.9%)	12 (4.87%)	21 (8.53%)	8 (3.25%)	68 (27.6%)
	Female	28 (11.38%)	29 (11.79%)	58 (23.58%)	63 (25.61)	178 (72.3%)
Total		55 (22.35%)	41 (16.66%)	79 (32.1%)	71 (28.8%)	246

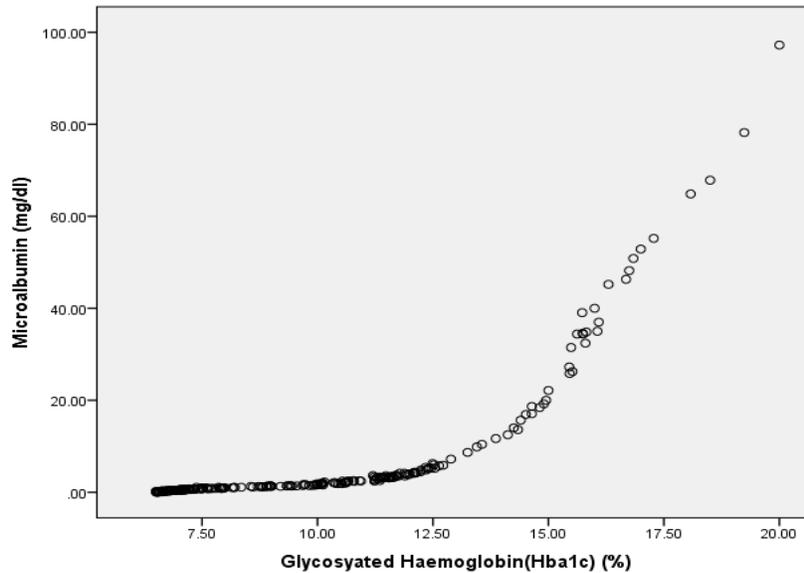
The above table shows that out of 246, 178 (72.3%) were females and 68 (27.6%) were males. Majority of females had HbA1c above 11.55 (25.61%) while for males HbA1c ranging from 6.5-7.08 (10.9%). This was statistically significant since p-value = 0.000<0.05.

Table 6: Correlation between Glycosylated Haemoglobin, microalbumin and diabetes duration among the Study Population

		Microalbumin (mg/dl)	Hba1c (%)	Diabetes Duration (Years)
Microalbumin (mg/dl)	Pearson Correlation	1	.800**	.664**
	Sig. (2-tailed)		.000	.000
	N	246	246	246
Glycosyated Haemoglobin (HbA1c) (%)	Pearson Correlation	.800**	1	.869**
	Sig. (2-tailed)	.000		.000
	N	246	246	246
Diabetes Duration (Years)	Pearson Correlation	.664**	.869**	1
	Sig. (2-tailed)	.000	.000	
	N	246	246	246

** . Correlation is significant at the 0.01 level (2-tailed).

Table 6 shows a positive correlation between Microalbumin and glycosylated haemoglobin HbA1c (r=0.800), microalbumin and diabetes duration (r=0.664), glycosylated haemoglobin and diabetes duration (r=0.869). The correlation was significant at 0.01 (<0.05).



Graph 2: Relationship between Glcosylated (HBA1c) and Microalbumin among the Study Population

A positive correlation was found between the microalbumin and glycosylated haemoglobin at the level of significance (0.05) and this was evidenced by Pearson correlation coefficient ($r=0.800$) ($P<0.05$) was considered statistically significant (Table 6).

DISCUSSION

Diabetic nephropathy is one of the long-term complications of diabetes mellitus. Shah, Acharya, Shrestha, & Shrestha, 2017 classified diabetes patients under-treatment based on aggravating glycemic control into normal control ($HbA1c < 6.5\%$) and uncontrolled group ($HbA1c > 6.5\%$). The purpose of this study was to test the microalbumin level among uncontrolled diabetes patients. 246 diabetic patients with $FBS \geq 126$ mg/dl (7.0mmol/l) were recruited. This study revealed 77.7 % ($n=191$) had uncontrolled diabetic Mellitus ($HBA1c > 7\%$). This observation was similar to that made by (Nduati, 2016) Mathari National Teaching Hospital (Kenya) and Ngassa Piotie, Van Zyl, & Rheeder, 2015 in Tshwane district hospital (South Africa) where more than 75 % of the diabetic patients had uncontrolled diabetes mellitus. There was a female predominance in this study participant of 72.4% over males (27.6%) which may be a reflection of health-seeking behavior amongst men and women at the site. The population survey may help to explain this predominance. The overall prevalence of microalbumin was 32.9% in 246 known uncontrolled diabetes mellitus patients. Deferent epidemiological and cross-sectional studies have revealed different variations in the prevalence of microalbumin in diabetic patients. Ngassa Piotie, Van Zyl, & Rheeder, 2015 reported 23% of microalbuminuria and 10% macroalbuminuria cases among 798 diabetic patients who had attended the Kalafong Diabetic Clinic (South Africa) in 2012. (Wanjohi, Otieno, Ogola, & Amayo, 2002 reported a prevalence of 26% in 100 types 2 diabetic patients in Kenya. Boelter et al., 2015 reported the prevalence of microalbuminuria at 29% in 149 diabetic patients in hospital based-research attending the

diabetes clinic at Kilimanjaro Christian Medical Centre in northern Tanzania. Molefe-Baikai, Molefi, Cainelli, & Rwegerera, 2018 reported a prevalence of 44.6% among 289 patients with type 2DM in a tertiary clinic in Gaborone, Botswana. The discrepancy in the prevalence of microalbumin can be attributed to several factors such as the difference in population, the sample size, the definition of microalbumin, and the measurement methods of microalbumin and urine collection tested. The all reported prevalence is with a range of 30-40% revealed by Noubiap, 2015 in the systematic review of diabetic nephropathy in the sub-Saharan Africa region.

In the present study, there was a statistically significant positive correlation between microalbuminuria and HbA1c. Ngassa Piotie, Van Zyl, & Rheeder, 2015 reported also a significant positive correlation between HbA1c levels and microalbumin among 754 diabetes patients, attending a diabetes clinic at the Kalafong Hospital in Pretoria, South Africa. A similar correlation was reported by (Subramani & Prabhusamy, 2016) in Velammal medical college hospital, Madurai, a tertiary care hospital (India). Microalbuminuria cases were always confirmed by a second urine specimen in the present study (Estimated et al., 2015).

The present study has highlighted a strong positive correlation between microalbuminuria with a higher level of HbA1c level ($r=0.800$, $p<0.05$) among study participants ($r=0.664$, $p<0.05$). Uncontrolled hyperglycemia is responsible for the pathogenesis of diabetic nephropathy. The presence of advanced glycosylation end products (AGEs) is the basic etiology of many important diseases. The diabetes mellitus is frequently escorted with hyperglycemia and oxidative stress, an increased rate of AGE-formation is observed. This reaction causes morbidity of diabetes, end-stage kidney and heart diseases (Nass, Simm, & Oh, 2009).

Our study agreed with a study done by Muraliswaran et al., 2016 in India that found a significant positive correlation between microalbumin and glycosylated haemoglobin in uncontrolled glycemic patients. The positive correlation between microalbuminuria and HbA1c in uncontrolled diabetes mellitus was reported by Khan, Khan, & Ahmad, 2012 and in our study, a significant positive correlation between duration of diabetes and HbA1c. A positive correlation between microalbuminuria and duration of diabetes was reported by other previous studies done by Khan et al., 2012 in Pakistan, Kamuhabwa & Charles, 2014 in Tanzania and (Idowu, Ajose, Adedeji, Adegoke, & Jimoh, 2017)

This study is in contrast with Omar, Musa, Osman, & Adam, 2018 have shown the longer duration of diabetes was not associated with poor glycemic control. These variations are probably related to the different distributions of patients' ages in different studies.

We recognize that the lack of a non-diabetic control population is a challenge of our study; however, this study was not designed to determine the effect of diabetes on the kidney, but rather to explore the correlation between microalbumin and hba1c among uncontrolled diabetes mellitus.

In summary, microalbumin levels showed significant correlations with glycosylated haemoglobin monitoring failure and diabetes duration as a risk factor of diabetes complications and the trigger of diabetes nephropathy. We also found that all patients with microalbuminuria were found to have uncontrolled glycemia. The proportion of uncontrolled glycemic increased with diabetes duration. A significantly high proportion of microalbumin was observed in patients who had uncontrolled diabetes and had the disease from 8 to 19 years after diagnosis.

Our study has some potential strength. First, this study has used new testing methodologies on a hospital-based study among uncontrolled diabetic patients. Second, this study had helped clinicians to prevent or to delay DN among diabetic patients for a better health outcome. However, we should recognize a number of limitations of this study. First, we enrolled one district hospital in this study; their findings are not applicable to other settings. Second, our statistical analyses are preliminary to confirm the correlation of MA with other risk factors. Nonetheless, further studies should deal with challenges highlighted in the present study.

CONCLUSION

Our study showed that around 68.5% of the diabetic patients were uncontrolled and 32.9% of them had microalbumin. Our results prove to increase accessibility to microalbuminuria and HbA1c testing for all the diabetic patients at district hospitals would improve monitoring and reduce the complications of diabetes mellitus. The present study emphasizes partnership of government through the Ministry of Health, hospitals, diabetic association and health policymakers to implement the strict glycaemic control and testing for microalbuminuria which is an early indicator of diabetic nephropathy.

RECOMMENDATIONS

The present study suggests glycosylated haemoglobin testing is a monitor of glycemic control and microalbumin test in the spot urine sample for an indicator of renal involvement in diabetic subjects give a confident method for early diagnosis and help the physician to intervene. The prevention, delay or reverse the progression of diabetic nephropathy can be attained only by perfect long term metabolic control.

We recommended that HbA1c assay equipment and reagents should be provided for all district hospitals, microalbumin should be carried out at regular intervals.

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