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Incidence of Diabetic Nephropathy in Southern Nigeria

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Diabetic nephropathy is a progressive kidney disease caused by angiopathy of capillaries in the kidney glomeruli. This study was designed to find out the frequency of occurrence of renal complications in diabetic patients. This was achieved by examining for the presence of microalbuminuria using Albumin Creatinine Ratio (ACR) in a spot urine. A total of 95 asymptomatic diabetic patients and 19 non-diabetic controls were used for the study. These patients were grouped into two: those with analbuminuria and others with microalbuminuria using the ACR = 30 mg g^{-1}, which approximates 24 h urinary albumin excretion in mg. Results obtained showed mean significant increases in the ACR in the diabetic group (48.58±4.14 mg g^{-1}) when compared to the control (22.76±5.14 mg g^{-1}; $p<0.05$). There were also mean significant increases in ACR (48.58±4.14 mg g^{-1}; $p<0.05$) with increase in glycosylated haemoglobin (5.90±0.26%; control: 3.72±0.13%) as well as with duration of diabetes (4.17±0.63 years) amongst the diabetic patients. Our analysis thus, indicated that there was a significantly higher ACR (54.09±8.87 mg g^{-1}; $p<0.05$) in men than in women (45.65±4.24 mg g^{-1}). The study also showed that the incidence of nephropathy amongst diabetics in Southern Nigeria is 72.63%.

Key words: Diabetic Nephropathy (DN), Albumin Creatinine Ratio (ACR), Body Mass Index (BMI), Glycosylated Haemoglobin (HbA$_1$c), Southern Nigeria diabetics

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INTRODUCTION

One of the most important clinical features of diabetes is its harmful effect on the kidney's small blood vessels (microangiopathy) which may progress to diabetic nephropathy (American Diabetes Association Inc., 2004). Diabetic nephropathy is a clinical syndrome characterized by persistent albuminuria, a relentless decline in Glomerular Filtration Rate (GFR), intra-renal hypertension and increased relative mortality from cardiovascular diseases (Rehman and Hamayun, 2004). Microalbuminuria is a strong indicator for the development of microvascular and macrovascular disease in patients with type-1 as well as type-2 diabetes mellitus (Stehouwer *et al.*, 1992). Once proteinuria develops irreversible deterioration in renal function and renal failure occurs (Gaibaltt and Adoga, 2006). Diabetic nephropathy occurs in approximately 1/3rd of individuals with type-1 and type-2 DM (Rehman *et al.*, 2005). Genetic susceptibility contributes significantly to the risk of developing nephropathy in type-1 diabetes mellitus (Chowdhury *et al.*, 1997). Similarly, familial predisposition to essential hypertension increases the risk of diabetic nephropathy (Fagerudd *et al.*, 1998). This familial clustering of diabetic renal disease susceptibility clearly suggests that in addition to the inherited and environmental factors that produce hyperglycaemia, the predisposition to nephropathy is under genetic control. Risk factors for development of Diabetic Nephropathy (DN) include hyperglycaemia, hypertension and smoking (Timothy and Peter, 2000). Patients with reduced renal functional reserve capacity may be more prone to microalbuminuria when exposed to conditions such as hyperglycaemia or hypertension (Hovind *et al.*, 2004). Microalbuminuria is a reversible state and can be resolved by glycaemic control and specific medications like angiotensin converting enzyme inhibitors and angiotensin receptor blockers. These drugs help to reduce microalbuminuria and a combination of these two types of drugs may be the best to avoid going to the stage of proteinuria (Ghazalli and Meng, 2003).

Screening for Diabetic Nephropathy (DN) involves monitoring at least yearly for urinary albumin excretion greater than 30 mg day^{-1} (Timothy and Peter, 2000). Although not done routinely, recent evidence suggests that the type of protein excreted, whether high molecular weight (Immunoglobulins G and M) or low molecular weight (alpha-1 and beta-1 microglobulins) correlate with the severity of the renal histologic lesion and may predict the outcome and the response to the therapy (Brenner *et al.*, 2001). The present study was carried out to provide baseline information on Diabetic Nephropathy (DN) regarding frequency of occurrence and the need to reduce the risk or slow progression of diabetic nephropathy.

MATERIALS AND METHODS

Data collection: In the Government Specialist Hospital, Benin City, Edo State, Nigeria, 2007, a total of 128 diabetic patients were interviewed for data collection out of which 95 patients (volunteers) gave their consent for the study. Specific questionnaire was used which included variety of questions such as present age, duration of diabetes (in years), familial history regarding the same or other diseases (heart disease, kidney disease) and information about the socio-economic position (occupation), education and life style of the patients. Parameters such as height (m) and weight (kg) of patients were analysed to check their link with disease prevalence and incidence.

Nineteen apparently healthy normal subjects were also interviewed to serve as control subjects. These controls were not known to suffer from diabetes, diabetic nephropathy, cardiovascular disease, hypertension or any renal disorder.

Sample collection

Blood: Subjects/volunteers were fasted overnight and 7.0 mL of venous blood were collected from the cubita fosa in the morning on or about 8.00 am and dispensed into 3 different specimen bottles, 2.0 mL into fluoride oxalate (for glucose estimation), 2.0 mL into ethylenediamine tetrachloroacetic acid (for glycosylated haemoglobin estimation) and 3.0 mL into lithium heparin (for urea estimation).

Urine: A spot urine sample (mid-stream) was also collected into a sterile universal bottle for urine albumin and creatinine estimation. This is more convenient for the patients than a timed (24, 4 h or overnight) urine collection.

Sample preparation: Blood samples were immediately centrifuged at 3000 g for 5 min to separate plasma from the cellular components. The plasma was aspirated and used for the different assays. Urine was assayed immediately for urinary albumin and creatinine.

Analysis: Glucose levels were estimated by the glucose oxidase method as outlined by Bartham and Trinder (1972), glycosylated haemoglobin (HbA$_1$C) by the fast ion exchange separation method as outlined by Nuttall (1998), urea by the Urease-Berthelot method as outlined by Weatherburn (1967), creatinine by the modified Jaffe's method as outlined by Spierto *et al.* (1979) and urinary albumin by the Lowry *et al.* (1951) method.

RESULTS

This study was based on 69 (72.63%) patients diagnosed with Diabetic Nephropathy (DN) out of the 95 patients who consented to the study. Of the 69 patients 25 (36.23%) were males and 44 (63.77%) were females. Mean pooled/aggregate age was 55.80 ± 1.03 years while the duration of diabetes in both sexes was 4.17 ± 0.63 years.

The mean urinary albumin levels from the study was 197.46 ± 39.59 mg dL^{-1} for control group and 380.41 ± 12.55 mg dL^{-1} for the diabetic group ($p<0.05$) while plasma creatinine concentration was 0.44 ± 0.04 mg dL^{-1} for control and 0.90 ± 0.04 mg dL^{-1} for the diabetic group (Table 1). Results obtained showed that the ACR was 22.76 ± 5.14 and 48.48 ± 4.14 mg g^{-1} for control and diabetics, respectively ($p<0.05$) while plasma urea level was 23.89 ± 0.63 mg dL^{-1} for control group and 35.06 ± 1.87 mg dL^{-1} for the diabetic group (Table 1; $p<0.05$).

The mean urinary albumin, plasma creatinine, urea and ACR were found to be significantly ($p<0.05$) higher in the diabetic group than in the control.

Results also show that the ACR, Fasting Plasma Glucose (FPG) and HbA_1c were significantly elevated ($p<0.05$) in males (ACR, 54.09 ± 8.87 mg g^{-1}, FPG 158.48 ± 15.55 mg dL^{-1} and HbA_1c $6.45\pm0.57\%$) than in females (ACR 45.65 ± 4.24 mg g^{-1}, FPG 144.05 ± 7.73 mg dL^{-1} and HbA_1c $5.61\pm0.26\%$) within the diabetic group (Table 2).

Table 3 shows gender consideration in control and diabetics of mean urinary albumin, ACR, BMI and BP measurements. In the diabetic group the urinary albumin and ACR are significantly elevated ($p<0.05$) in males (385.82 ± 17.81 mg dL^{-1} and 54.09 ± 8.87 mg g^{-1}, respectively) than in females (377.53 ± 16.8 mg dL^{-1} and 43.50 ± 4.20 mg g^{-1}, respectively). However, the BMI, diastolic and systolic blood pressures are significantly higher ($p<0.05$) in females (26.9 ± 0.62 kg m^{-2}, 110.00 ± 1.38 and 210.00 ± 3.43 mmHg, respectively) when compared to their male counterparts (24.47 ± 0.80 kg m^{-2}, 81.97 ± 2.11 and 134.09 ± 3.01 mmHg, respectively).

In diabetic patients with microalbuminuria (ACR = 30 mg g^{-1}), HbA_1c and ACR are slightly elevated though significant ($p<0.05$) in the males ($6.86\pm0.72\%$ and 59.06 ± 8.9 mg g^{-1}) than in females ($5.53\pm0.31\%$ and 58.13 ± 5.9 mg g^{-1}, respectively (Table 4). However, in

Table 1: Urinary albumin, plasma creatinine, plasma urea and ACR levels in control (non-diabetic) and diabetic patients

Groups	No.	Mean urinary albumin	Mean plasma creatinine	Mean ACR	Mean plasma urea
		----- (mg dL^{-1}) -----			
Control (non-diabetic)	19	197.46 ± 39.59^a	0.44 ± 0.04^a	22.76 ± 5.14^a	23.89 ± 0.63^a
Diabetic patients	95	380.41 ± 12.55^b	0.90 ± 0.04^b	48.58 ± 4.14^b	35.06 ± 1.87^b

Values in the same column with different letters differ significantly ($p<0.05$); ACR: Albumin creatinine ratio; No.: No. of subjects

Table 2: ACR, FPG and HbA_1c levels, gender consideration in control (non-diabetics) and diabetic patients

Levels	Control (non-diabetic) (n = 19)			Diabetic patients (n = 95)		
	Male (n = 8)	Female (n = 11)	Total (n = 19)	Male (n = 33)	Female (n = 62)	Total (n = 95)
ACR (mg g^{-1})	20.83 ± 9.02^a	24.16 ± 6.35^a	22.76 ± 5.14^a	54.09 ± 8.87^b	45.65 ± 4.24^{bc}	48.58 ± 4.14^b
FPG (mg dL^{-1})	84.56 ± 3.71^a	87.40 ± 3.14^a	86.05 ± 2.47^a	158.48 ± 15.55^b	144.05 ± 7.73^{bc}	149.06 ± 7.38^b
HbA_1c (%)	5.49 ± 0.48^a	6.26 ± 0.57^a	3.72 ± 0.13^a	6.45 ± 0.57^b	5.61 ± 0.26^{bc}	5.90 ± 0.26^b

Values in the same row with different letters differ significantly ($p<0.05$); ACR: Albumin creatinine ratio; FPG: Fasting plasma glucose; HbA_1c: Glycosylated haemoglobin

Table 3: Urinary albumin, ACR, BMI and BP gender consideration in control (non-diabetic) and diabetic patients

Measurements	Control (non-diabetic) (n = 19)			Diabetic (n = 95)		
	Male (n = 8)	Female (n = 11)	Total (n = 19)	Male (n = 33)	Female (n = 62)	Total (n=95)
Urinary albumin (mg dL^{-1})	122.73 ± 34.71^a	251.8 ± 59.66^a	197.46 ± 39.59^a	385.82 ± 17.81^b	377.53 ± 16.8^{bc}	380.41 ± 12.55^b
ACR (mg g^{-1})	20.83 ± 9.02^a	240.16 ± 6.34^a	22.76 ± 5.14^a	54.09 ± 8.87^b	45.65 ± 4.2^{bc}	48.58 ± 4.14^b
BMI (kg m^{-2})	25.08 ± 0.94^a	21.9 ± 1.5^a	23.24 ± 0.98^a	24.47 ± 0.8^b	26.9 ± 0.62^{bc}	26.04 ± 0.51^b
Diastolic BP (mmHg)	80.00 ± 5.00^a	84.00 ± 2.67^a	76.84 ± 2.54^a	81.97 ± 2.11^b	110.00 ± 1.38^{bc}	84.53 ± 1.17^b
Systolic BP (mmHg)	116.67 ± 11.06^a	131.00 ± 6.74^a	120.53 ± 3.01^a	134.09 ± 3.70^b	210.00 ± 3.43^{bc}	137.68 ± 2.59^b

Values in the same row with different letters differ significantly ($p<0.05$); ACR: Albumin creatinine ratio; BMI: Body mass index; BP: Blood pressure

Table 4: Distribution of sex and HbA_{1C} in diabetic volunteers with and without microalbuminuria

Disrtibutions	ACR (mg g^{-1})	Age (years)	HbA_1C (%)
With microalbuminuria (ACR ≥30)			
Male (n = 25)	59.06 ± 8.9^a	55.48 ± 2.21^a	6.86 ± 0.72^a
Female (n = 44)	58.13 ± 5.9^b	55.51 ± 2.21^b	5.53 ± 0.31^b
Total (n = 69)	58.98 ± 5.24	56.38 ± 1.24	5.94 ± 0.33
Without microalbuminuria (ACR <30)			
Male (n = 8)	23.18 ± 1.51^a	55.00 ± 2.38^a	5.41 ± 0.87^a
Female (n = 18)	20.09 ± 1.84^b	57.78 ± 2.57^b	5.83 ± 0.49^b
Total (n = 26)	21.35 ± 1.24	56.92 ± 1.90	5.69 ± 0.42

Values in the same column with different letters differ significantly ($p<0.05$); ACR: Albumin creatinine ratio; HbA_1c: Glycosylated haemoglobin
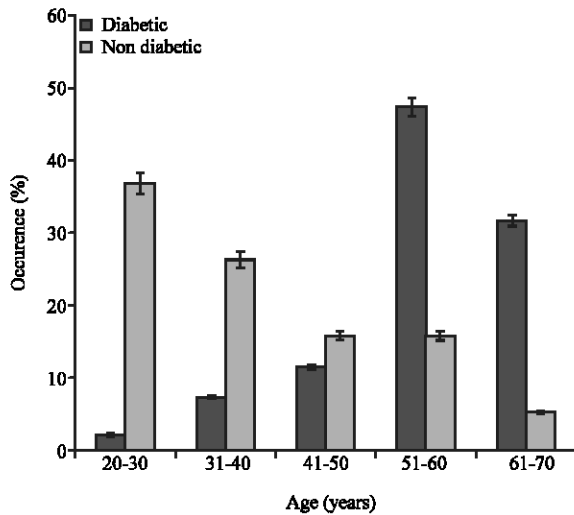
Fig. 1: Age distribution of diabetic and non diabetic (control) patients (no. in parenthesis indicate % occurrence)
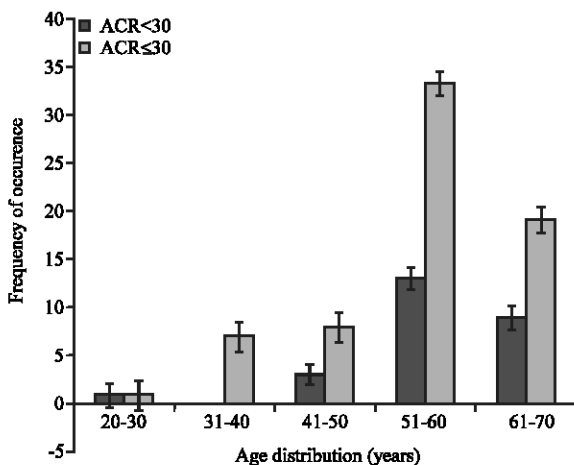

Fig. 2: Albumin Creatinine Ratio (ACR) distribution among various age groups of diabetic volunteers. (No. in parenthesis indicates frequency of occurrence)

Fig. 3: Relationship between duration of diabetes and ACR
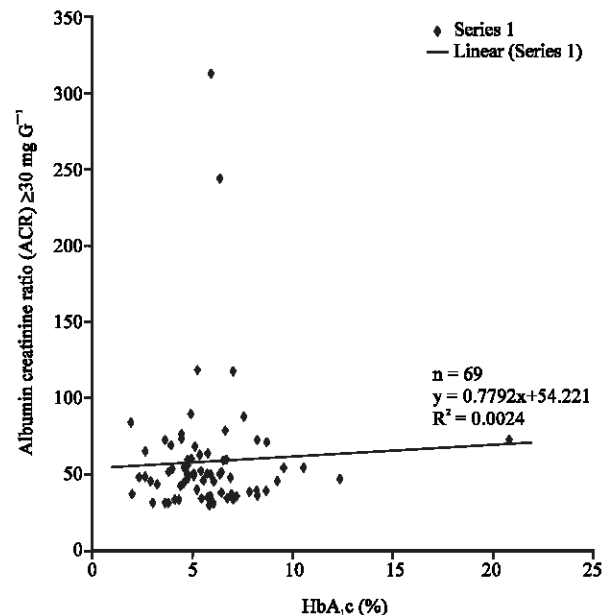
Fig. 4: Relationship between Glycosylated haemoglobin (HbA1c) and ACR ≥ 30

diabetic patients with normoalbuminuria (ACR <30 mg g^{-1}), the mean HbA_1c was significantly elevated ($p<0.05$) in the females ($5.82\pm0.49\%$) when compared to the male subjects (5.41 ± 0.87). Similarly, with microalbuminuria, the ACR in males (23.18 ± 1.51 mg g^{-1}) was significantly higher ($p<0.05$) than that in the female subjects (20.09 ± 1.84 mg g^{-1}).

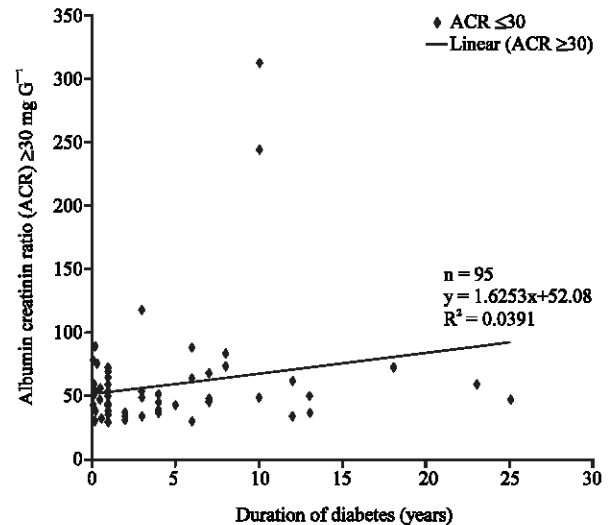Figure 1 shows the frequency of age distribution among the control (non-diabetic) and diabetic patients with greater prevalence within the age range of 51 to 60 years. The number of diabetic patients with microalbuminuria were also prevalent within the same age range (Fig. 2).

Figure 3 shows the relationship between ACR and duration of diabetes in diabetics with microalbuminuria (ACR = 30 mg g^{-1}). There was a positive correlation between microalbuminuria and duration of diabetes ($R^2 = 0.04$; $p<0.05$), as well as between microalbuminuria and HbA_1c ($R^2 = 0.0024$; $p<0.05$, Fig. 4).

DISCUSSION

Diabetic Nephropathy (DN) is a clinical syndrome characterized by persistent albuminuria, a relentless decline in Glomerular Filteration Rate (GFR), raised arterial blood pressure and increased relative mortality from cardiovascular diseases (Rehman and Hamayun, 2004).

Microalbuminuria refers to the appearance of low but abnormal levels = 30 mg day^{-1} or 20 µg min^{-1} of albumin in the urine or an ACR = 30 mg g^{-1} in a spot urine. Patients with microalbuminuria are referred to as having incipient nephropathy. Without specific intervention, 20- 40% of type-2 diabetic patients with microalbuminuria progress to overt nephropathy and by 20 years after onset of overt nephropathy, approximately 20% would have progressed to end stage renal disease (American Diabetes Association Inc., 2004). According to Timothy and Peter (2000), once overt nephropathy has developed, treatment is a delaying rather than a preventive measure.

In a study by Rehman and Hamayun (2004), 35.58% of the total patients examined were diagnosed with Insulin Dependent Diabetes Mellitus (IDDM) nephropathy. The mean age at diagnosis was 52.01±0.41 years while the duration of diabetes in IDDM nephropathy patients was 13.19±0.21 years after which they were diagnosed with DN. The mean Body Mass Index (BMI) in DN patients was 26.49±0.23 kg m^{-2}. The present study showed the incidence of microalbuminuria in diabetic patients aged 20-70 years as 72.63%. Out of the 95 patients who consented to the study, 69 (72.63%) had significant ACR = 30 mg g^{-1}. This is much higher than the earlier results of Rehman and Hamayun (2004) although, they considered only patients with IDDM in their study. However, in this study, patients with IDDM and Non-Insulin Dependent Diabetes Mellitus (NIDDM) were considered.

The present study also show that the average age of duration of diabetes with microalbuminuria (nephropathy) is 4.17±0.63 years. This again is lower than that obtained by Rehman and Hamayun (2004) where the duration of diabetes in IDDM nephropathy patients was 13.19±0.21 years. In this study however, IDDM and NIDDM patients were considered.

According to the American Diabetes Association Inc. (2004), microalbuminuria rarely occurs with short duration of type-1 diabetes, therefore screening in individuals with type-1 diabetes should begin after 5 years of disease duration. The increase in ACR with age suggests increased risk of nephropathy with age. Our finding that microalbuminuria correlated positively (R^2 = 0.04; p<0.05, Fig. 3) with duration of diabetes corroborated earlier studies of Williams and Pickup (1999), who suggested that the incidence of microalbuminuria increases with increase in duration of diabetes. The higher mean value of ACR obtained in men when compared with that in women from this study also corroborates earlier reports of Mogensen (1984), who found lower urinary albumin excretion rate in women than in men.

According to Williams and Pickup (1999) hypertension is about twice as common in diabetic patients as in the non-diabetic population. Analysis from our studies show that 59% of the diabetic patients were hypertensive and that 57.97% of the microalbuminuric diabetics were hypertensive which supports earlier findings (Williams and Pickup, 1999).

The establishment from our studies of a positive correlation (R^2 = 0.002, p<0.05, Fig. 4) between HbA$_1$c and ACR suggests that poor glycemic control is associated with progression of microalbuminuria. The Microalbuminuria Collaborative Study Group UK (1993) had emphasized correlating HbA$_1$c levels with the progression of the disease.

To reduce the risk and slow the progress of DN, blood glucose control should be optimized. Attention therefore, should be directed towards prevention rather than to treatment of diabetic nephropathy. Once overt nephropathy is present, progression cannot be avoided but only delayed.

CONCLUSION

This study showed that the incidence of nephropathy amongst diabetics in Southern Nigeria was 72.63%. Our study also indicated that ACR increases with age and duration of diabetes and it was higher in men than in women amongst diabetic patients.

ACKNOWLEDGMENTS

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