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Albuminuria and urinary albumin-creatinine ratio of hypertensives attending a tertiary hospital at Enugu, Nigeria

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ABSTRACT

Background: People with high blood pressure (hypertensives) are at high risk of nephropathy and some studies point to increased albuminuria and albumin-creatinine ratio (ACR) as early indices of this condition. **Objective:** This study evaluated the presence of albuminuria and determine urine ACR of hypertensives attending the University of Nigeria Teaching Hospital Enugu, Nigeria. **Materials and Methods:** The study was carried out on eighty-nine (89) volunteers (50 tests and 39 controls) aged 21-77 years. Subjects' anthropometry was measured. Blood and urine samples were collected and analyzed for serum creatinine (SCr), urine creatinine (UCr) and urine albumin using Jaffe modified kinetic method and Enzyme-Linked Immunosorbent Assay (ELISA), respectively. **Results:** Of the 89 subjects, there was a significant difference in the height, SCr, eGFR, and UCr of both sexes. Of which, BMI was lower in males than the females. However, there was no significant difference between systolic blood pressure (SBP), diastolic BP (DBP), pulse, weight, urine albumin and ACR with the sex of both subjects. The test subjects had significantly higher SBP, DBP, weight, BMI, SCr, urine albumin, and ACR but lower eGFR and UCr when compared with the control subjects. Besides, there was a significant difference in the SBP, height, SCr and eGFR between the sexes of test subjects. Of which, males had relatively higher values. **Conclusion:** Findings from this study revealed that the hypertensives were at high risk of kidney injuries. Furthermore, albuminuria was observed to be much lower than the current threshold employed to define microalbuminuria and was associated with lower eGFR.

Keywords: Hypertensive nephropathy, Albuminuria, Nigeria, Blood pressure

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INTRODUCTION

Hypertension is a pathologic condition where the pressure in the arteries is persistently high (1). Hypertension is one of the major chronic non-communicable diseases of global health significance (2). Of which, it was shown that one in every four individuals is affected (3). Furthermore, this is expected to increase to 29% by 2025 (4).

Hypertension can be primary (essential/ idiopathic) or secondary. Primary hypertension is due mainly to genetic disposition and non-specific lifestyle while secondary hypertension is caused by identifiable pathology such as chronic kidney diseases, endocrine disorder and narrowing of arteries (5). Thus, the causes of hypertension are multifactorial, and its consequences include increased risk of brain damage, cardiac disease, renal disease, and retinal complications (6).

Albuminuria (the presence of albumin in the urine) is an early marker of damage to the glomerular membrane, and it is classified into A1, A2 and A3 depending on the amount of albumin present; A1 occurs in normal physiologic condition value < 29mg/L, A2 is termed microalbuminuria with a value between 30-300mg/L and A3 is termed macroalbuminuria with value > 300mg/L(6). Albuminuria has been prognostic for most cardiovascular disorders (7) and it is equally considered a marker for end-organ damage and endothelial dysfunction (8). Besides, albuminuria can result from hyperfiltration, nephrosclerosis, endothelial dysfunction and glomerular basement membrane abnormalities that can't be detected by dipstick technology but by ELISA (9).

Albumin-creatinine ratio (ACR) can be used for urine albumin estimation to detect microalbuminuria and it is expressed in milligram (mg) of albumin per gram (g) of creatinine (9). Due to variation in the urine concentration and flow rate of albumin, excreted albumin must be adjusted to creatinuria which is 30-300mg albumin/g creatinine equivalent to 3.4-33.9 mg albumin/mmol creatinine. This is considered positive for microalbuminuria (10).

Hypertensive nephropathy could be asymptomatic and delays early diagnosis (6). Consequently, its effect on the kidney can be devastating (11). Microalbuminuria has been indicated as an index for endothelial dysfunction and end-organ damage and can be used to predict the risk of kidney damage in hypertensive patients. However, there is a paucity of published data on the incidence of albuminuria and the albumin-creatinine ratio of hypertensive persons in Southeast Nigeria. Hence, the present study evaluated the presence of albuminuria and determine urine ACR of hypertensives attending the University of Nigeria Teaching Hospital Enugu, Nigeria.

MATERIALS AND METHODS

Study Area

The study was carried out in the University of Nigeria Teaching Hospital (UNTH), Enugu South-East Nigeria. The hospital is located at 6.3014° N, 7.4619° E and 22 kilometres from Enugu the capital along the Enugu Port Harcourt expressway and covers an area of 200 acres and serves mainly the rural and urban areas of Enugu, as well as referrals from the neighbouring states of Anambra, Abia, Ebonyi, Imo, Delta, Benue and Kogi.

Study Population

The study was carried out on a total of 89 volunteers (50 test and 39 control groups) from the University of Nigeria Teaching Hospital Ituku-Ozalla aged from 21-77 years.

Study Design

This was a case-control study.

Data Collection

A structured questionnaire was used to obtain medical history as well as other relevant data from the participants, including informed consent. Absolute confidentiality of participants' data was maintained.

Subject Selection and Criteria

Samples were collected randomly from hypertensive subjects attending the University of Nigeria Teaching Hospital Ituku-Ozalla, Enugu while the control samples were collected from apparently healthy individuals that are not hypertensive.

Sample Collection and Preparation

About 15 millilitres (ml) of early morning spot urine was collected into a clean, chemically free wide neck transparent container and the urine was stored at 4^0C in a plain tube for micro-albumin and urine creatinine assay. Blood was collected from the participants using a sterile 2ml syringe which was transferred into a sterile labelled plain vial. The blood sample in the plain tubes was centrifuged at 5000 revolutions per minute for 5minutes and the serum was dispensed into a serum container and stored at 4^oC for 2days for serum creatinine assay.

Laboratory analytical procedure

Serum creatinine

Biochemical analyses were performed on the serum specimen using Humalyzer 2000 chemistry semi auto-analyzer (Human GmBH Diagnostica Worldwide, Wiesbaden, Germany). The modified Jaffe kinetic method was used as modified by the Cromatest kit. Results were interpreted as described by the kit's manufacture instructions.

Urine Micro-albumin

Biochemical analysis was performed on the urine using Mindray 96A Enzyme-Linked Immunosorbent Assay (ELISA) Microplate Reader was used (Mindray Bio-Medical Electronics Company, Shenzhen, China). The concentration of micro-albumin in the samples was then determined by comparing the optical density of the samples to the standard curve using Creative Diagnostic ® ELISA kit (Cat.No: DEIA2299) (New York, USA). All the assay was conducted following the kit manufacturer's instructions.

Determination of Glomerular Filtration Rate

Serum creatinine values were used to estimate glomerular filtration rate (GFR) following the Chronic Kidney Disease Epidemiology Equation (CKD-EPI) formula (12, 13).

Anthropometric Measurements

The following anthropometric data were measured: height, weight, and blood pressure. Standing height was measured to the nearest centimetre (cm) using a stadiometer with footwear removed. Weight was measured to the nearest kilogram (kg) with a manual Seca 761 scale (Vogel and Halke, Hamburg, Germany) after participants have removed outer garments, cell phones, and footwear. Blood pressure (BP) and pulse rate were measured with an automated sphygmomanometer (OMRON HEM705CP, Omron Matsusaka Co, Matsusaka city, Mie-Ken, Japan) using appropriate cuff size after participants have sat undisturbed for at least 5 minutes. Three consecutive readings were taken one minute apart, and the mean of the three readings was used for the analysis. Body mass index (BMI) was calculated using the metric body mass index formulae weight (kg)/ (height $(m)^2$.

Statistical Analysis

Data obtained from this study were analyzed using the statistical package for social sciences (SPSS) version 22 of Windows (IBM Inc. Chicago, IL, USA). Data were

presented as means and standard deviations. The student's t-test was used to determine the mean differences between the two groups. All *p* values <0.05 were considered statistically significant.

RESULTS

Of the 89 subjects, the mean±SD age was 50.81±14.73 years. Findings from the study of this subject showed that males had significantly higher SCr, eGFR, height and UCr but lower BMI (1.71±0.08m, 102.11±13.38 µmol/L, 87.16±15.74 ml/min/1.73m^2, 13.25±2.30 mmol/L, and 26.39±4.44 Kg/m^2 respectively) when compared with the females (1.63±0.08m, 88.00±14.06 µmol/L, 78.66±19.54 ml/min/1.73m^2, 10.94±1.03 mmol/L, and 30.59±5.45 Kg/m^2) respectively ($p<0.05$) (Table 1). However, there was no significant difference between systolic blood pressure (SBP), diastolic BP (DBP), pulse, weight, urine albumin and ACR with the sex of subjects.

The test subjects had significantly higher SBP, DBP, weight, BMI, SCr, urine albumin, and ACR but lower eGFR and UCr (139.96±22.14 mmHg, 82.68±13.69 mmHg, 83.00±14.82 Kg, 29.80±4.70 Kg/m^2, 99.02±14.42 µmol/L, 13.83±5.44 mg/L, 1.25±0.48 mg/mmol, 73.58±14.93 ml/min/1.73m^2, and 11.06±1.37 mmol/L, respectively) when compared with the control subjects (113.46±9.93 mmHg, 72.36±7.25 mmHg, 74.44±13.18 Kg, 26.75±5.74 Kg/m^2, 90.15±15.03 µmol/L, 11.09±5.02 mg/L, 13.45±2.17 mmol/L, 94.97±14.49 ml/min/1.73m^2, and 13.45±2.17 mmol/L respectively) (Table 2). However, there was no significant difference between the pulse rate and height of both groups ($p>0.05$).

Besides, there was a significant difference in the systolic blood pressure, height, serum creatinine and eGFR between the sex of test subjects where males had relatively higher values compared to the females (Table 3) ($p < 0.05$).

Table 1: Anthropometric, Demographic and Biochemical Parameters by Sex of Participants

	All (n=89) Mean ± SD	Male (n=45) Mean ± SD	Female (n=44) Mean ± SD	*p* value
Age (year)	50.81±14.73	48.76±14.98	52.91±14.33	0.185
Systolic BP (mmHg)	128.35±22.12	130.18±22.46	126.48±21.91	0.434
Diastolic BP (mmHg)	78.16±12.39	78.31±13.34	78.00±11.50	0.907
pulse (BPM)	73.75±9.03	73.80±9.88	73.70±8.20	0.961
Weight (Kg)	79.25±14.68	77.49±14.24	81.05±15.07	0.256
Height (M)	1.67±0.09	1.71±0.08	1.63±0.08	0.001*
BMI((Kg/m^2)	28.47±5.37	26.39±4.44	30.59±5.45	0.001*
Serum creatinine(µmol/L)	95.13±15.38	102.11±13.38	88.00±14.06	0.001*
eGFR(ml/min/1.73m^2)	82.96±18.13	87.16±15.74	78.66±19.54	0.026
Urine albumin(mg/L)	12.63±5.41	13.26±4.84	11.99±5.92	0.271
Urine Creatinine(mmol/L)	12.11±2.12	13.25±2.30	10.94±1.03	0.000*
Urine ACR (mg/mmol)	1.07±0.49	1.05±0.44	1.01±0.53	0.595

*Significant difference determined by two-tailed Student t-test at 95% confidence interval

Table 2: Comparison of Anthropometric, Demographic and Biochemical Parameters of Study groups

	All (n=89) Mean ± SD	hypertensive (n=50) Mean ± SD	Control (n=39) Mean ± SD	*p* value
Age (year)	50.81±14.73	57.40±9.73	42.36±15.81	0.000*
Systolic BP (mmHg)	128.35±22.14	139.96±22.14	113.46±9.93	0.000*
Diastolic BP (mmHg)	78.16±12.39	82.68±13.69	72.36±7.25	0.000*
pulse (BPM)	73.75±9.03	74.66±9.16	72.59±8.85	0.286
Weight (Kg)	79.25±14.68	83.00±14.82	74.44±13.18	0.006*
Height (M)	1.67±0.09	1.67±0.10	1.67±0.08	0.683
BMI((Kg/m^2)	28.47±5.37	29.80±4.70	26.75±5.74	0.007
Serum creatinine(µmol/L)	95.13±15.38	99.02±14.42	90.15±15.03	0.006*
eGFR(ml/min/1.73m^2)	82.96±18.13	73.58±14.93	94.97±14.49	0.000*
Urine albumin(mg/L)	12.63±5.41	13.83±5.44	11.09±5.02	0.017
Urine Creatinine(mmol/L)	12.11±2.12	11.06±1.37	13.45±2.17	0.000*
Urine ACR (mg/mmol)	1.07±0.49	1.25±0.48	0.84±0.40	0.000*

*Significant difference determined by two-tailed Student t-test at 95% confidence interval

Table 3: Comparison of Anthropometric, Demographic and Biochemical Parameters by Sex of Test Participants

	All (n=50) Mean ± SD	Male (n=22) Mean ± SD	Female (n=28) Mean ± SD	*p* value
Age (year)	57.40±9.73	57.27±9.41	57.50±10.15	0.963
Systolic BP (mmHg)	139.96±22.14	148.18±17.88	133.50±23.30	0.018
Diastolic BP (mmHg)	82.68±13.69	86.77±12.95	79.46±13.61	0.060
pulse (BPM)	74.66±9.16	72.73±10.92	76.18±7.35	0.189
Weight (Kg)	83.00±14.82	84.91±14.68	81.50±15.03	0.425
Height (M)	1.67±0.10	1.73±0.09	1.62±0.07	0.001*
BMI((Kg/m^2)	29.80±4.70	28.43±4.49	30.89±4.65	0.066
Serum creatinine(µmol/L)	99.02±14.42	103.91±15.96	95.18±12.03	0.032*
eGFR(ml/min/1.73m^2)	73.58±14.93	80.73±16.42	67.96±10.97	0.002*
Urine albumin (mg/L)	13.83±5.44	14.39±4.64	13.40±6.04	0.528
Urine Creatinine (mmol/L)	11.06±1.37	11.42±1.62	10.78±1.08	0.104
Urine ACR (mg/mmol)	1.25±0.48	1.27±0.42	1.24±0.53	0.840

*Significant difference determined by two-tailed Student t-test at 95% confidence interval

DISCUSSION

In this study, a comparison of results was carried out in three phases: between males and females of the study population, between tests (hypertensive) and controls (non-hypertensive), finally between male tests and female tests. Our findings showed that there was a significant difference in the height, BMI, serum creatinine, urine creatinine and eGFR in males and females of the study population. This is in concordance with the literature and other studies that estimated these variables in the adult population (14, 15, 16).

Males are known to be generally taller than women which give them a higher muscle mass and there is a direct relationship between muscle mass and creatinine production. The bigger the muscle mass the higher the level of creatinine produced and vice versa. The higher muscle mass of males leads to the significant difference seen in the value of eGFR, concentration of serum creatinine and concentration of urine creatinine. Women are generally known to have more fat cells when compared to the male counterpart this is due to hormonal differences giving rise to higher BMI in women than in males.

Significant statistical difference was observed in the age, systolic pressure, diastolic pressure, weight, BMI, serum creatinine, urine creatinine, urine albumin, urine albumin-creatinine ratio and eGFR between the test and control subjects which is in agreement with the findings from other studies (17, 18, 19, 20). The higher serum creatinine levels observed in the test subjects can be adduced to the destruction of the filtration barrier leading to decreased filtration causing serum creatinine levels to be higher in the test group when compared with controls that have an intact functional filtration barrier. Glomerular membrane destruction in the test subjects could be the reason urine creatinine value is higher in the control subject when compared to the test subject, thereby confirming the inverse relationship between serum creatinine and urine creatinine.

The transmission of elevated blood pressure to the glomeruli leads to increased hydraulic pressure and increased glomerular filtration (21) the elevated pressure causes low molecular weight albumin to pass through the glomerular filtration barrier thereby supporting the statistical significance observed in urine albumin with test having higher mean. Changes in size and charge selectivity of the glomerular membrane (21) can equally be attributed to being a cause of increased albumin level seen in the test subject; this will result in the loss of the negatively charged albumin. Increased albumin-creatinine ratio observed in test subjects can be attributed to alterations in the glomerular capillaries which lead to increased albumin excretion due to its low molecular weight (22).

The urinary creatinine value decrease could be due to the destruction of the filtration barrier giving rise to high albumin-creatinine value when standard formulae are used in the calculation of the albumin-creatinine ratio. Weight gain has been associated with the development of hypertension and cardiovascular complication while weight loss has been implicated to reduce the risk for hypertension (17).

Also, a significant difference was observed in systolic blood pressure, height, serum creatinine and eGFR between the test male and test female. This is in support of the report by Mathieson (14) which showed that creatinine excretion varies between individuals according to the muscle mass which has been attributed to an individual height. The significant difference in the systolic pressure reported in the males could

be adduced to higher basal metabolic rate and variability among the research subjects because some had normal blood pressure when their blood pressure reading was taken while others had hypertensive value.

CONCLUSION

Findings from this study revealed that the hypertensives were at high risk of kidney injuries. Furthermore, albuminuria was observed to be much lower than the current threshold employed to define microalbuminuria and was associated with lower eGFR.

Declarations

i. Funding: none received
ii. Conflicts of interest/Competing interests: None declared by authors
iii. Ethics approval: Ethical approval was obtained from the ethical committee of the University of Nigeria Teaching Hospital, Enugu, Nigeria.
iv. Consent to participate: Informed written consent was obtained from all participating subjects before recruiting into the study, following the standards of human experimentation and with the Helsinki Declaration of 1975.
v. Consent for publication: not applicable
vi. Availability of data and material: Available upon request through the corresponding author
vii. Code availability: not applicable

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