The Diagnostics, 1(3): 25-33, August 2018

Full Length Research Paper

Serum minerals and lipid profiles of type 2 diabetic and hypertensive out-patients at a University Teaching Hospital in Nigeria

[1]Ekwunoh, P.O., [2]Edeogu, S.C., [1]Onochie, A.U.,
[1]Alaebo, P.O., [3]Ezeigwe, O.C and [2]Mbadugha, N.N
[1]Department of Biochemistry, Chukwuemeka Odumegwu Ojukwu University Uli.
[2]Department of Chemical Pathology, Nnamdi Azikiwe University, Nnewi Campus
[3]Department of Applied Biochemistry, Nnamdi Azikiwe University, Awka.

Corresponding author's e-mail address: peterekwunoh@gmail.com

ABSTRACT

Diabetes and hypertension are important clinical conditions which are also independent risk factors for other diseases. Some anthropometric indices, plasma lipid profile and mineral levels were compared amongst 150 Out-patients with type 2 diabetes (DM), hypertension (HBP), and hypertensive diabetes (HBP+DM), and 50 apparently diabetes- and hypertension-free (controls) at Nnamdi Azikiwe University Teaching Hospital, Nnewi. Total cholesterol (TC), triglyceride (TG), high density lipoprotein cholesterol (HDL-C) and low density lipoprotein cholesterol (LDL-C) were determined using standard methods. Trace elements (chromium, copper, magnesium and zinc) were determined by atomic absorption spectrophotometry (AAS), while sodium and potassium were determined by flame photometry. Results showed that the DM (28.28±8.99 kg/m^2) and HBP+DM (27.38±4.41 kg/m^2) patients had non-significantly higher BMI levels compared to the HBP (26.00±4.05 kg/m^2) and controls (26.18±3.88 kg/m^2), while SBP and DBP were significantly higher in the HBP (153.02±28.42 and 94.04±12.40 mmHg) and HBP+DM (133.24±11.04 and 87.36±10.58 mmHg) patients than in DM (111.20±8.75 and 71.40±8.30 mmHg) and controls (116.68±11.56 and 76.60±1.80 mmHg). Also, TC and LDL-C concentrations of the DM (4.19±0.87 and 2.48±0.73 mmol/L), HBP (4.45±0.63 and 2.73±0.58 mmol/L) and HBP+DM (4.60±0.98 and 2.72 mmol/L) patients were significantly higher than those of the control subjects (3.72±0.37 and 1.56±0.53 mmol/L), while the patients' HDL-C and TG levels were non-significantly elevated compared to the control group values. Na concentrations of the HBP+DM group was significantly ($p<0.000$) higher, while that of Cu was lower than those of the DM, HBP and control subjects. Age and gender did not significantly ($p>0.05$) affect the levels of the parameters studied in the patient groups. The results call for early diagnosis and monitoring of dyslipidaemia in these group of patients for better prognosis.

Keywords: Diabetes mellitus, hypertension, dyslipidaemia, minerals, metabolic disorders

INTRODUCTION

Diabetes mellitus and hypertension have become clinical conditions of public health importance especially in developing countries due to their high mortality and morbidity. Diabetes is a condition in which the body does not produce enough or properly respond to insulin, a hormone produced in the pancreas. Insulin enables cells to absorb glucose and so when non-functional or insufficient, glucose tend to accumulate in the blood and lead to various complications. Many medical conditions are associated with diabetes mellitus and these include high blood pressure, elevated cholesterol levels,

polycystic ovary syndrome, chronic pancreatitis among others (Tierney *et al.*, 2002; Rother, 2007).

Hypertension, on the other hand, is a chronic medical condition in which both systolic and diastolic blood pressures are elevated above 140 and 90 mmHg respectively. It is one of the risk factors for stroke, heart attack, heart failure and is a leading cause of chronic renal failure (Pierdomenico *et al*., 2009); even moderate elevation of arterial blood pressure leads to shortened life expectancy.

Information on plasma lipid concentrations and prevalence of dyslipidaemia among patients with type 2 diabetes mellitus and or hypertension is important. Several reports have confirmed that diabetes and hypertension are independently associated with dyslipidaemia among Nigerians (Igwe *et al*., 2017; Nwankwo *et al*., 2017). Furthermore, the occurrence of diabetes mellitus\ hypertension is associated with a number of socio-demographic factors such as age and gender (Valliyot *et al.*, 2014).

Trace elements are inorganic molecules which are essential for life. In states of absolute deficiency, death results and in limited intake, biological functions are impaired (Burtis and Asswood, 2003). Direct association of trace amounts of macro elements and even derangement in micronutrient concentrations with diabetes have been reported by Nourmohammadi *et al*. (2000). There is accumulating evidence that metabolism of several essential elements is altered in diabetes and several other diseases, and that these nutrients might have specific roles in the pathogenesis and progress of such diseases (Murray *et al*., 2000; Vincent, 2000).

Although, there is a growing body of information on metabolic changes in diabetes mellitus\hypertension, there is currently insufficient data on such changes in combined occurrence of diabetes and hypertension in Nigeria. Moreover, no study has specifically assessed the effect of age and gender differences on lipid profile and minerals in hypertensive diabetics in Anambra State. This study was therefore designed to determine and compare the changes in some anthropometric indices, lipid profile and minerals of type 2 diabetics, hypertensive and hypertensive diabetics attending Nnamdi Azikiwe University Teaching Hospital in Anambra State. The study will provide data that will help to achieve adequate diagnostic and therapeutic control of dyslipidaemia and mineral derangement in diabetic and hypertensive patients.

MATERIALS AND METHODS

Study population

About 372 out-patients attending the Nnamdi Azikiwe University Teaching Hospital Nnewi at the commencement of this study constituted the study population.

Sample size (SS) was computed using Cochran formulae: $SS = \frac{(Z\text{-score}^2)(P)(1-P)}{(M)^2}$ (Cochran 1963). Where, Z-score is 1.645 at 90% confidence interval with study population (SP) of 372; P is population proportion (expressed as decimal assumed to be 0.5 (50%); and M is margin of error at 5% (0.05).

Substituting these values,

$SS = \frac{(1.645^2)(0.5)(1-0.05)}{(0.05)^2} = 514$, and Adjusted SS (ASS) according to Barlett *et al*. (2001).

$$\text{is} = \frac{SS}{1+\left(\frac{SS-1}{SP}\right)} = \frac{514}{1+\left(\frac{514-1}{372}\right)} = \frac{514}{1+\left(\frac{513}{372}\right)}$$

$$\text{ASS} = \frac{514}{1+1.3790} = 216$$

Eventually, 200 subjects comprising 50 diabetic normotensive, 50 hypertensive non-diabetics, and 50 hypertensive diabetics, together with 50 apparently normal subjects who gave informed consent fully participated in the study.

Ethnical clearance was from the Ethics Committee of Nnamdi Azikiwe University Teaching Hospital Nnewi.

Inclusion and exclusion criteria

Patients on oral hypoglycaemia drugs or whose diagnosis of diabetes was made at the age 40 years and above with no record of ketosis were considered to have type 2 diabetes mellitus. Diabetic subjects on insulin injection and those not up to 40 years

(grouped as type 1 diabetics) were excluded from the study. Subjects with systolic pressure greater than 140 mmHg and or diastolic blood pressure greater than 90 mmHg measured using standard procedures were included as hypertensives.

Determination of blood pressures, heights, weights and BMI of subjects

Blood pressure of each subject was measured using Mercury sphygmomanometer (Desk 605P, Suzuken Company Japan) according to standard protocol. The body weights of the subjects were measured with the aid of a mechanical weighing scale (SALTER 200, England) to the nearest 0.5 kg, with the subjects wearing light clothing and without shoes. Heights of barefooted subjects were measured using a measuring tape to the nearest 0.1m. Body mass index (BMI) was calculated as weight (kg) of the subjects divided by their corresponding squared height (m^2).

Blood sample collection

Eight milliliters (8ml) of blood was collected from each subject, out of which about 2ml was dispensed into fluoride oxalate bottle and spun for glucose estimation using glucose oxidase method of Barham and Trinder (1972). The remaining blood sample was allowed to clot, spun and the serum separated for lipid profile, sodium, potassium and trace elements estimations.

Determination of lipid profile

Total cholesterol (TC) was determined by enzymatic end point method, while high density lipoprotein cholesterol (HDL-C) and low density lipoprotein cholesterol (LDL-C) were determined by precipitation enzymatic method as described by Groove (1979) and Burstein *et al*. (1980). Triglyceride (TG) concentration was estimated by colorimetric method of Fossati and Prencipe (1982).

Determination of the elements

Sodium and potassium concentrations were determined using an atomic emission spectrophotometer (Model AVI572A and Agilent Technology Model 4100MP-AES, England) while the trace elements (magnesium, zinc, copper and chromium) were evaluated with the aid of an atomic absorption spectrophotometer (Surgifield Medical Model AA320N, England).

Data analysis

Data generated were presented as means with standard error. They were analyzed in MS Excel version 2013 and shown in bar charts with error bars indicating standard error at 5% level of significance.

RESULTS

The BMI, SBD, DBP, FBS, serum lipids and mineral levels of DM, HBP and HBP+DM of out-patients and control subjects studied are presented in Figure 1(a-e).

The DM (28.28±8.99 kg/m^2) and HBP+DM (27.38±4.41 kg/m^2) out-patients had non-significantly higher BMI levels compared to the HBP (26.00±4.05 kg/m^2) and control (26.18±3.88 kg/m^2) subjects (Figure 1a).

In Figure 1b, there were significantly higher levels of SBP and DBP in the HBP (153.02±28.42 and 94.04±12.40 mmHg) and HBP+DM (133.24±11.04 and 87.36±10.58 mmHg) patients than in DM (111.20±8.75 and 71.40±8.30 mmHg) and control (116.68±11.56 and 76.60±1.80 mmHg) subjects. On the other hand, the FBS concentrations of the DM (15.18±5.88 mmol/L) and HBP+DM (13.56±4.29 mmol/L) patients were significantly higher than those of the HBP (6.68±3.56 mmol/L) and control (4.62±0.80 mmol/L) groups. The TC and LDL-C concentrations of the DM (4.19±0.87 and 2.48±0.73 mmol/L), HBP (4.45±0.63 and 2.73±0.58 mmol/L) and HBP+DM (4.60±0.98 and 2.72 mmol/L) patients were significantly higher than those of the control subjects (3.72±0.37 and 1.56±0.53 mmol/L), while the patients' HDL-C and TG levels were non-significantly elevated compared to the control group values (Figure 1c).

In Figure 1d, the Na concentrations of the HBP+DM (144.90±5.90 mmol/L) group was significantly ($p<0.000$) higher than those of the DM, HBP and control subjects (136.30±3.40, 139.60±5.30 and 137.50±3.60

mmol/L respectively). However, the K concentrations of the patients were non-significantly lower than that of the control group (4.50±0.3 mmol/L). Also, the Mg level of the HBP+DM subjects at 0.30±0.10 mmol/L was significantly lower than those of the DM (0.90±0.10 mmol/L), HBP (0.90±0.90 mmol/L) and the control (1.00±0.10 mmol/L) subjects.

There was no significant difference between the copper levels of the control (135.80±14.8 mmol/L) and DM (138.30±16.90 mmol/L) groups, while those of the HBP (103.70±15.40 mmol/L) and HBP+DM (70.90±14.60 mmol/L) were significantly reduced (Figure 1e). The zinc levels of the patients were significantly lower than that of the control (102.30±5.60 mmol/L) group, with HBP+DM having the significantly least level of 48.10±6.70 mmol/L, followed by that of the DM (60.30±8.40 mmol/L) and HBP (80.50±8.60 mmol/L) groups. The HBP patients had the significantly highest chromium level (0.70±0.01 mmol/L), while control group showed the lowest value at 0.009±0.01 mmol/L.

Figure 2 is the distributions of FBS, TG, TC, HDL-C and LDL-C concentrations of the DM, HBP and HBP+DM patients by age. The values of the various parameters did not vary significantly amongst the patient groups with age but the DM group had consistently higher FBS values but lower TG, TC, HDL-C and LDL-C concentrations when compared with those of the HBP and HBP+DM subjects.

Figure 3 presents the distribution of values of BMI, SBP, DBP, lipid profile parameters and minerals of the DM, HBP and HBP+DM patients studied by gender.

The results show that gender did not significantly affect the levels of BMI, SBP and DBP as well as the concentrations of the lipid profile parameters and most of the minerals. However, there was observed significantly higher values of Mg and Cr in the female diabetics (0.94±0.11 and 0.07±0.02 mmol/L) than the male diabetics (0.86±0.14 and 0.06±0.02 mmol/L). Similarly, female hypertensives had significantly higher Cr level (0.07±0.01 mmol/L) than their male counterparts. Similarly, female diabetic hypertensives had significantly higher Cu value (74.62±15.99 µg/dl) than the males of the same group (65.42±10.28 µg/dl).

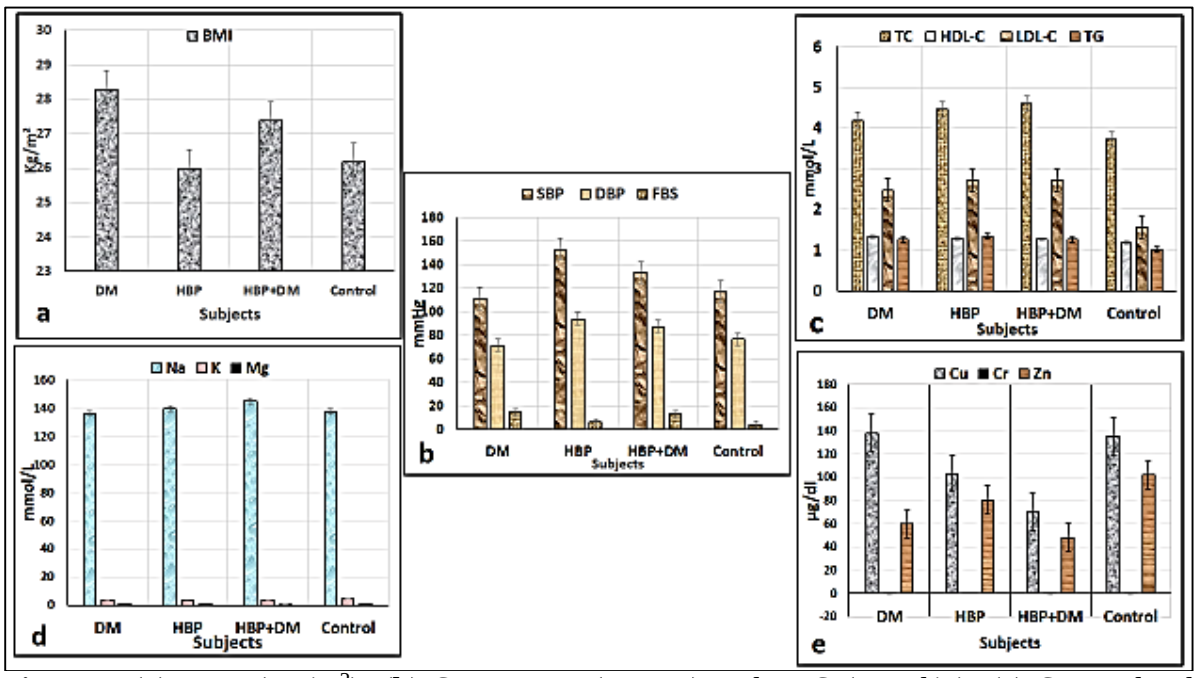

Figure 1:(a) BMI (Kg/m^2), **(b)** SBD, DBP (mmHg) and FBS (mmol/L), **(c)** Serum lipids (mmol/L), **(d)** Na, K, Mg (mmol/L), and **(e)** Cu, Cr, and Zn (µg/dl) amongst DM, HBP, and HBP+DM Out-patients and Control subjects.

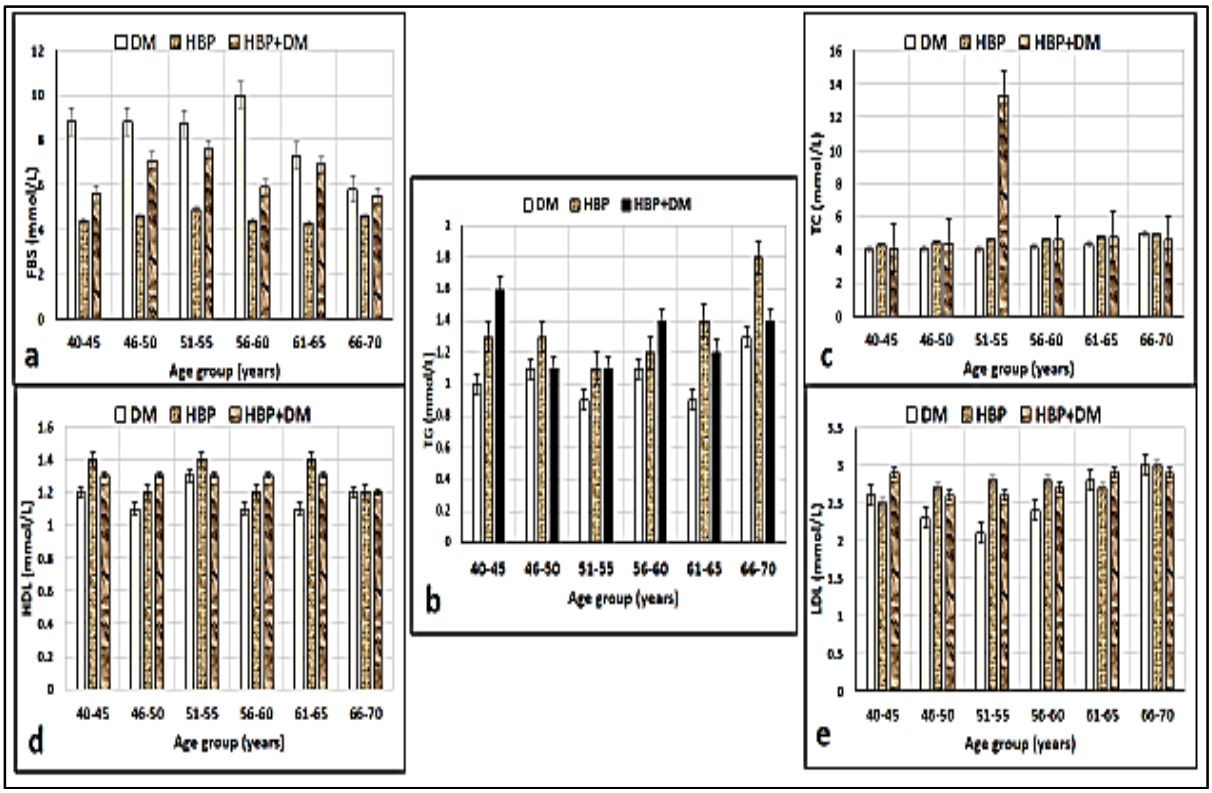

Figure 2: Distribution (mmol/L) of **(a)** FBS **(b)** TG, **(c)** TC, **(d)** HDL, and **(e)** LDL among DM, HBP, and HBP+DM Out-patients by age-groups.

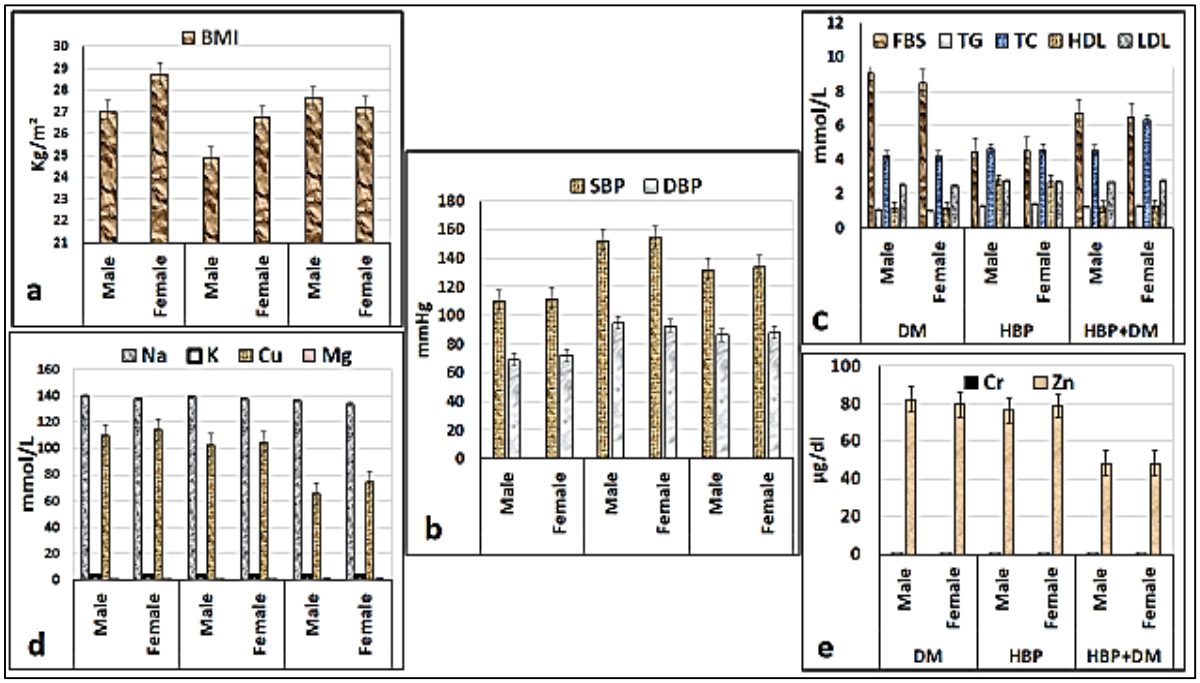

Figure 3: Distribution of (a) BMI (kg/m²), (b) SBP and DBP (mmHg), (c) Lipids (mmol/L), (d) Minerals (mmol/L; µg/dl) among DM, HBP, and HBP+DM by gender.

DISCUSSIONS

High levels of body mass index (BMI) and blood pressures have been identified as hallmarks of diabetes and hypertension respectively. This is in tune with our observation of significantly higher levels of BMI in the studied diabetic (DM) and diabetic hypertensive (HBP+DM) subjects when compared with the hypertensive (HBP) and normal subjects (Figure 1a).

A similar trend was observed among the subjects for fasting blood sugar (FBS). On the other hand, there were significantly higher levels of systolic and diastolic blood pressures in the HBP and HBP+DM subjects than the DM patients and the controls (Figure 1b).

Systolic and diastolic blood pressures make up the total blood pressure in the body. Generally, in hypertensives, there is elevated blood pressure. This assertion is in consonance with the observation in the present study in which the hypertensive and hypertensive diabetic patients had significantly higher blood pressures than those with only diabetes and the control subjects. Systolic and diastolic blood pressures have been related to risk of coronary heart disease in a linear fashion: the higher the level of blood pressure, the greater the incidence of coronary heart disease.

A study of the lipid profile levels of the diabetics, hypertensives and diabetic hypertensives showed that there were no significant differences observed among them (Figure 1c). Perhaps due to the fact that the patients were on medications. However, their lipid profile levels were significantly higher than those of the control subjects. Diabetic and hypertensive patients are generally at high risk of dyslipidaemia characterized by hyper-cholesterolaemia, hyper-triglyceridaemia, and hyper-lipoproteinaemia (mostly very low density lipoproteins [LDL]), and low levels of high-density lipoprotein cholesterol (HDL-C) and a shift in LDL distribution towards small, dense particles (Igwe *et al.*, 2007). Our result corroborates the findings of Isezuo *et al.,* (2003) who reported that total cholesterol, high density lipoprotein cholesterol, low density lipoprotein cholesterol and triglyceride levels do not differ significantly among patients with hypertension, diabetes and concurrent hypertension and diabetes.

Increase in blood glucose level increases blood osmolality, which directly increases blood pressure. Similarly, increase in blood lipids such as cholesterol promoted by increase in low density lipoprotein leads to thickening of the arterial walls causing a further rise in blood pressure. This agrees with the study carried out in Saudi Arabia in 2008 which showed that hypertensive individuals have significantly higher mean levels of total cholesterol and low density lipoprotein cholesterol compared with controls (Afridi *et al*., 2008).

People with increased BMI are overweight and are predisposed to hypertension and diabetes. Thus, increased BMI can lead to increased blood pressure which invariably causes significantly higher levels of cholesterol and low density lipoprotein. This finding is in line with previous similar study by Nasri and Yasdni (2006) on correlation between lipid levels and blood pressure values in patients with type 2 diabetes, which reported a positive correlation (r = 0.196, p = 0.031) between LDL-cholesterol and systolic blood pressure values.

It was seen in Figures 1d and 1e that all the minerals analyzed in this study (sodium, magnesium, chromium, copper, and zinc) differed significantly between the patients

and control subjects except for potassium, which were not significantly different in all the groups studied. Although, the potassium values were non-significantly different within the groups, but there was a lower mean potassium level in the hypertensives compared to the controls. The hypertensive individuals had significantly higher serum sodium levels than the other groups. When sodium levels are persistently elevated, the body loses potassium and retains water, making blood pressure rise (Afridi *et al*., 2008). This is inconsonance with the earlier report that hypertensive individuals have significantly higher serum sodium, chloride and calcium levels but a significantly lower potassium level when compared to normotensives (Al-Muhana *et al.*, 2006). Potassium functions closely with sodium and chloride to maintain fluid distribution and pH balance and to augment nerve-impulse transmission, muscle contraction, and regulation of heartbeat and blood pressure. Marginal change in potassium concentration can induce an increase in fluid volume and lead to an impairment of blood pressure regulating mechanisms which can result in hypertension in susceptible individuals (Jerry, 2000).

Magnesium levels were lower in the patients than the control group, with the hypertensive diabetics having the lowest magnesium level (Figure 1d), perhaps due to the co-existence of the two conditions. Magnesium helps relaxation of blood vessels, promoting normal blood pressure. It also plays an important role in carbohydrate metabolism by regulating blood sugar levels. Low magnesium levels have been implicated as an important factor in most disorders of metabolic syndrome X, a group of health disorders that include insulin resistance plus high blood pressure, elevated cholesterol, obesity, and certain age-related conditions (Resnick, 1992; Resnick *et al*., 1992). This agrees with the deduction from the examination of 22 research papers on type 2 diabetes, in which nearly half of all patients had depleted magnesium levels and a third more had suboptimal levels (Rosolova *et al*., 1997; De Lenardis *et al*., 2000). Also, in a double-blind, placebo-controlled study, people who took 411 to 548 milligrams of magnesium daily achieved a reduction in both systolic and diastolic blood pressure (Itoh *et al*., 1997).

The blood copper levels of the patients differed significantly, with the diabetics having the highest level of copper among the patients which is not significantly different from that of the controls. On the other hand, the patients had significantly lower zinc concentration than the control subjects, with hypertensive diabetics having lowest levels of both copper and zinc. Meanwhile, in all the subjects studied copper levels were significantly higher than their respective zinc concentrations. Copper helps prevent cardiovascular problems such as high blood pressure, while zinc has been associated with normal immune function. Elevated copper level has been correlated with low zinc level in diabetics. Excess Zn antagonizes Cu levels (Zagar and Shah, 1998). Low zinc level in diabetics generally is due to physiological losses of zinc from the body due to hyperglycaemia. Thus, the lowest zinc level observed amongst the hypertensive diabetics might be due to complications caused by the loss in zinc. Insufficient blood zinc level causes oxidative stress which may damage the cells irreversibly leading to some classic complications of diabetics of which hypertension is one of them (Naila *et al*., 2008).

Results in Figures 2 and 3 showed that age and gender did not significantly affect the

levels of BMI, blood pressures, minerals and lipid profile parameters of the studied DM, HBP and HBP+DM patients. However, the female patients had generally higher levels of BMI, SBP and DBP as well as the lipid profile parameters and minerals studied. This corroborated earlier observation from the studied area that, although female diabetic patients are less prone or exposed to DM-associated lifestyle changes, yet they had significantly higher BMI, systolic and diastolic blood pressures, FBS and lipid profile parameters than male diabetics (Nwankwo *et al.,* 2017; Igwe *et al.,* 2017).

CONCLUSION

Results of the present study buttressed the role of increase in BMI and blood pressures in the diagnosis and management of DM and HBP. The study also highlighted the need for monitoring of changes in lipid profile parameters of DM, HBP and HBP+DM patients. However, the lipid profile results did not indicate significantly negative outcome for HBP+DM patients compared to those with diabetes or hypertension alone. The observed lower levels of minerals, especially the microelements, in the patients reiterates the need for micronutrient supplementation in metabolic disease patients. Age and gender did not significantly affect the levels of the parameters studied in the patient groups. Thus, age- and gender-based interventions may not be very necessary amongst the patients. However, the need for closer monitoring, early diagnosis and treatment of dyslipidaemia amongst these group of patients is a necessity for better patient outcome.

Conflicts of Interest
Authors declared no conflicts of interest.

Contributor's Statement
Multiple contributors were required to complete this study, and each author read and approved the final manuscript before submission.

REFERENCES

Al-Muhana, F.A., Larbi, E.B, Al-Ali, A.K, Al-Sultan, A., Al-Ateeeq, S., Soweilem, L., Goa, L., Bahnassy, A.A., Al-Rubaish, A and Abdulmohsen, M.F. (2006). Haematological, lipid profile and other biochemical parameters in Normal and Hypertensive subjects among the population of the Eastern Province of Saudi Arabia. *East Afri Med J.,* 83(1):44-48.

Afridi, H.I., Kazi, T.G., Kazi, N., Jamali, M.K., Arain, M.B., Jalbani, N., Sarfaraz, R.A., Shah, A., Kandhro, G.A., Shah, A.Q and Baig, J.A. (2008). Potassium, calcium, magnesium, and sodium levels in biological samples of hypertensive and non-hypertensive diabetes mellitus patients. *Biol Trace Elem Res.,* 124(3):206-224.

Barham, D and Trinder, P. (1972). An improved colour reagent for the determination of blood glucose by the oxidase system. *Analyst*, 97:142.

Bartlett, J.E., Kotrlik, J.W and Higgins, C.C. (2001). Organizational Research: Determining Appropriate Sample Size in Survey Research. *Learning and Performance Journal*, 19:43-50.

Burstein, M., Scholnick, H.R and Mofin, R. (1980). Rapid method for the isolation of lipoproteins from human serum by precipitation with polyanions. *Scand J Clin Lab Invest.,* 40:583-595.

Burtis, A.C and Ashwood, R.E. (2003). Trace elements. In: *Tietz Fundamentals of Clinical Chemistry.* 5th ed. Saunders, India. Pp. 570-575.

Cochran, W.G (1963). Sampling Techniques, 2nd Ed., New York: John Wiley and Sons, Inc.

De Lenardis, M., Schindler, R and Classen, H.G. (2000). Hypomagnesemia and suboptimal plasma. Mg levels in diabetes mellitus: frequencies and consequences. *Magnes Bul.,* 22:53-59.

Fossati, P and Prencipe, L. (1982). Serum triglycerides determined colorimetrically with an enzyme that produces hydrogen peroxide. *Clin Chem.,* 28(10):2077-2080.

Grove, T.H. (1979). Effect of reagent pH on determination of high-density lipoprotein cholesterol by precipitation with sodium phosphotungstate-magnesium. *Clin Chem.,* 25(4):560-564.

Igwe, C.U., Duru, L.A.D., Ukwamedua, H and Ikaraoha, C.I. (2007) Prevalence of

hyperlipidaemia amongst insulin dependent and non-insulin dependent diabetics in Delta State, Nigeria. *Tropical Doctor,* 37(2):120-121.

Igwe, C.U., Emejulu, A.A., Nwankwo, C.N and Onwuliri, V.A. (2017). Alterations in Lipid Profile and Atherogenic Predictor Indices of Type 2 Diabetes Mellitus Patients in Awka, Anambra State, Nigeria. *Biochemistri*., 29(2):44-53.

Isezuo, S.A., Badung, S.L.H and Omotoso, A.B.O. (2003). Comparative analysis of lipid profiles among patients with type 2 diabetes mellitus, hypertension and concurrent type 2 diabetes, and hypertension: a view of metabolic syndrome. *J Natl Med Assoc*., 95(5):328-334.

Itoh, T., Matsumoto, M., Nakamura, M., Okada, A., Shirahushi, N., Hougaku, H., Hashimoto, H., Sakaguchi, M., Handa, N., Takeshita, T., Merimoto, K and Hari, M. (1997). Effects of daily alcohol intake on the blood pressure difference depending on an individual's sensitivity to alcohol: Oriental flushing as a sign to stop drinking for health reasons. *Journal of Hypertension,* 5(11):1211-1217.

Jerry, LN. (2000). Diabetes and Magnesium: The Emerging Role of Oral Magnesium Supplementation. *The Magnesium Report* Clinical, research, and Laboratory News for Cardiologists Third Quarter.
www.mgwater.com/diabetes.shtml

Murray, R.K., Granner, P.A and Rodwell, V.W. (eds.) (2000). Metabolism of carbohydrates. *Harpers Biochemistry* 25th ed. Appleton and Lange New York. Pp 190-195

Naila, M., Ghulam, H.B., Rafi, A.G., Iqbal, A.M., Muhammad, A.M and Muhammad, S.M. (2008). Serum Zinc and Magnesium in Type 2 diabetic patients. Department of Medicine, Liaquat University of Medical and Health sciences (LUMHS), Jamshoro, Sindh,
www.emro.who.int/imemrf/jcpsp/jcpcp.

Nasri, H and Yazdani, M. (2006): The relationship between serum LDL-cholesterol, HDL-cholesterol and systolic blood pressure in patients with type 2 diabetes. *Kardiol Pol.,* 64(12): 1364-1368.

Nourmohammadi, I., Shalmani, I.K., Shaabani, M and Gohari, L. (2000). Zinc, Copper, Chromium, Manganese and magnesium levels in serum and hair of insulin independent diabetics. *Arch Iranian Med.,* 2:88-91.

Nwankwo, C.N., Igwe, C.U., Emejulu, A.A and Onwuliri, V.A. (2017). Demographic and Anthropometric Indices of Male and Female Type 2 Diabetes Mellitus Patients in Anambra State, Nigeria. *FUTOJNLS,* 3(2):126-138.

Pierdomenico, S.D., Di Nicola, M and Esposito, A.L. (2009). Prognostic value of Different Indices of Blood Pressure Variability in Hypertensive Patients. *American Journal of Hypertension,* 10:10-38.

Resnick, L.M. (1992). Cellular calcium and magnesium metabolism in the pathophysiology and treatment of hypertension and related metabolic disorders. *Am J Med.,* 93(2A): 11S-20S.

Resnick, L.M., Gupta, R.K., Bhargava, K.K., Hgruenspan, H., Alderman, M.H and Laragh, J. (1992). Cellular ions in hypertension, diabetes and obesity. A nuclear magnetic resonance spectroscopic study. *Hypertension* 17:951-957.

Rosolova, H., Mayer, O and Reaven, G. (1997). Effect of variations in plasma magnesium concentration on resistance to insulin-mediated glucose disposal in non-diabetic subjects. *J Clin Endocrinol Metab.,* 82:3783-3785.

Rother, K.I. (2007). Diabetes Treatment – Bridging the Divide. *N Engl J Med.,* 356(15):1499-1501.

Tierney, L.M., Phee, M.C and Papadakis, S.J. (2002). Diabetics. *Current Medical Diagnosis & Treatment.* 48th ed. Lange Medical Books/McGraw-Hill, New York. P120.

Valliyot, B., Sreedharan, J., Valliyot, S.B and Jayakumary-Muttappallymyalil, J. (2014). Demographic parameters associated with type 2 diabetes mellitus in North Kerala, India. *American Journal of Research Communication,* 2(9):39-48.

Vincent, J.B. (2000). Quest for the molecular mechanism of chromium action and its relationship to diabetes. *Nutr Rev.,* 58:67-72.

Zagar, A.H and Shah, N.A. (1998). Copper, zinc and magnesium levels in non-insulin dependent diabetes mellitus. *Postgrad Med J.,* 74:665-668.