# TRACE METALS STATUS OF DIABETIC PATIENTS RESIDENT IN ABAKALIKI, SOUTH EAST, NIGERIA

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**ABSTRACT**

***Available evidence shows that there are abnormalities in the metabolism of several trace elements in diabetes mellitus which may have contributed to the pathogenesis and progression of the disease. This study is aimed at investigating the levels of selected trace metals (zinc, copper, calcium, magnesium and lead) in the serum of type I and II diabetes mellitus patients. Thirty non-insulin-dependent diabetes mellitus (NIDDM), 30 insulin-dependent diabetes mellitus (IDDM) and 30 apparently healthy non-diabetic control subjects participated in the study. After an overnight fast, blood samples of all the subjects were collected and the serum concentrations of glucose and these trace elements were measured. Fasting blood sugar (FBS) was determined by glucose oxidase method to confirm the status of the patients and controls while the levels of the trace metals were measured by atomic absorption spectrophotometer (AAS). The mean levels of FBS for the diabetic patients were significantly higher (P<0.05) than those of the control group. Similarly, the mean serum concentrations of copper and lead were significantly higher (P<0.05) in the diabetic patients as compared to the controls. However, the mean values for zinc, calcium and magnesium were significantly lower (P<0.05) in the sera of diabetic patients when compared with the controls. These findings suggest that the altered metabolism of these elements may play a role in the development, pathogenesis and progression of diabetes mellitus.***

**Keywords:** Diabetes mellitus, Fasting blood sugar, Trace metals, Abakaliki

## INTRODUCTION

Diabetes mellitus is a metabolic disease that results from absolute or relative deficiency of insulin secretion and/or insulin action, characterized by disorder in metabolism and persistent rise in blood glucose levels above normal (hyperglycaemia) and its attendant complications (Grundy *et al.,* 1999; Tierney *et al.,* 2002). Some of the characteristic symptoms of diabetes mellitus are excessive urination (polyuria), excessive thirst, increased fluid intake (polydipsia), blurred vision and weight loss. These symptoms, however, may not be present if the blood glucose level is only mildly elevated (Lionel, 2007). The population of people with diabetes mellitus sometimes known as "the silent killer" is increasing at an alarming rate yearly, suggesting that environmental factors may be involved in its aetiololgy (Jones *et al.,* 2005). Nearly two decades ago, it was recognized that diabetes was neglected and that many patients were being denied specialist care. Several community studies have shown that these patients suffer a disproportionate burden of ill health and are responsible for increased economic and social demands on society (Alberti, 1991).

Mineral elements constitute a minute part of the living organism. However, they play essential roles in both growth and metabolism

(Aggett and Cornerford, 1995; Cohen *et al.,* 1997). Abnormalities in trace metals metabolism have been demonstrated in many human diseases. In particular, diabetes mellitus has been reported to be associated with abnormalities in the metabolism of zinc, copper, calcium and lead (Walter *et al.,* 1991). Reports have shown that as a group, diabetics excrete more zinc, calcium and magnesium in the urine than non-diabetics, causing a decrease in the plasma levels of these elements from these patients (Walter *et al.,* 1991). Studies by Isbir *et al.* (1994) has shown that the levels of zinc and magnesium were significantly lower while that of copper was significantly higher in the serum of patients with insulin-dependent diabetes mellitus. It was reported by Rao *et al*. (1987) that zinc has an insulin-like effect on the manifestation of diabetes. Zinc supplements have been shown to lower blood sugar levels in people with type I diabetes (Rao *et al.,* 1987) though some evidence indicates that zinc supplementation in people with type II diabetes does not improve their ability to process sugar (Niewoehner *et al.,* 1986).

Family studies suggest that zincuria could be under polygenic control (Pidduck *et al.,* 1970). Urinary zinc excretion appears to be controlled by alleles at a number of loci and diabetics might possess a different assortment of these alleles as compared to non-diabetics (Pidduck *et al.,* 1970). The exact cause of hyperzincuria is not known. However, disturbed metabolism of zinc metalloenzymes and abnormal binding of zinc to tissue proteins have been suggested as possible causes. The low plasma zinc levels in diabetics suggest that the hyperzincuria is of renal origin. Renal tubular defect in handling zinc and glucose-induced osmotic diuresis are other possibilities (Pidduck *et al.,* 1970).

Lead is a heavy metal dangerous to most organs and systems in the human body. It interferes with body metabolism and cellular functions such as the regulation of oxygen transport and energy generation. It is a cumulative metabolic poison with affinity for the sulphur-containing amino acids. Exposure to even low levels has been associated with health hazards especially in infants, pregnant women and nursing mothers (Baghurst *et al.,* 1987; Needleman and Gatsonis, 1990). The effects of lead poisoning in diabetes subjects have also been recognized over three decades ago (Wedeen *et al.,* 1975). According to reports, lead produced damaging effects in the haematopoetic, hepatic, cardiac, renal, reproductive and gastro-intestinal systems (Stoiek and Stoczynska, 2003). Lead levels have been investigated in many pathological conditions and it has been reported to be high in diabetes mellitus (Akinloye *et al.,* 2010).

Changes in the plasma concentrations of several trace metals have been reported in patients suffering from diabetes mellitus. It has also been suspected that these alterations may contribute to some of the metabolic dysfunctions observed in these patients. The present study is therefore aimed at evaluating trace metal status in two types of diabetes mellitus patients in this environment.

## MATERIALS AND METHODS

**Subjects:** Thirty patients suffering from non-insulin-dependent diabetes mellitus (NIDDM) and 30 patients suffering from insulin-dependent diabetes mellitus (IDDM) who had been clinically diagnosed for at least 12 months prior to the study, attending outpatient clinic of the Department of Medicine, Ebonyi State University Teaching Hospital (EBSUTH) were recruited after their oral consent was obtained. These subjects suffering from both types of diabetes were free from other diseases at the time of the study. Again, 30 ages matched apparently healthy volunteers served as the controls. The non-diabetic controls were selected after medical examination and laboratory tests to determine their fasting blood glucose levels. The clinical characteristics of the study subjects are shown in Table 1.

**Sample Collection:** About 10 mls of fasting venous blood were drawn from the antecubital vein of each subject using a sterile needle and syringe. The collected blood was dispensed into a clean and dry plain tube. Each sample was allowed to clot undisturbed for 15 minutes and centrifuged at 4000 rpm for 10 minutes to separate serum from the clot. The serum was dispensed into another clean and dry plain tube

and some aliquots collected for fasting sugar determination. The remaining sample was stored at –20$^{o}$C until trace metals analysis was performed.

**Biochemical Analysis:** The serum glucose concentration was measured using glucose oxidase spectrophotometric method (Trinder, 1969). Serum levels of copper, zinc, magnesium, lead and calcium ions were estimated by atomic absorption spectrophotometry (Kaneko, 1999).

**Statistical Analysis:** The SPSS software package for windows version 18.0 was used for statistical analysis and means were considered significantly different where $p<0.05$. All data are expressed as mean ± standard deviation. Statistical difference between each parameter for the diabetic and control groups was performed using analysis of variance (ANOVA).

## RESULTS

The mean age, body mass indices (BMI) and fasting blood glucose levels of all the subjects indicated that the mean age of the diabetic patients and the controls are statistically similar. There was, however, a significant difference ($p<0.05$) in the mean BMI of the diabetic patients when compared to the control. As expected, the fasting blood glucose levels of the diabetic patients was significantly higher ($P<0.05$) than those of the control group (Table 1).

The mean serum zinc, calcium and magnesium concentrations were significantly lower ($P<0.05$) in both types of diabetics in comparison to the control. Again, the mean serum concentrations of zinc, calcium and magnesium were significantly decreased ($P<0.05$) in NIDDM subjects when compared to IDDM subjects (Table 2). Furthermore, the mean levels of copper and lead were significantly higher ($P<0.05$) in the sera of both types of diabetes when compared to the control. Additionally, the mean serum concentrations of copper and lead were significantly increased ($P<0.05$) in NIDDM patients when compared to IDDM patients.

## DISCUSSION

Some trace metals are known to act as antioxidants by preventing membrane peroxidation, while others play their roles as metal-containing enzymes (metalloenzymes) where they help in metabolism of macromolecules such as glucose. It has been suggested that imbalance in the levels and abnormalities in the metabolism of some trace elements are common findings in many human diseases, particularly diabetes mellitus (Triptych *et al.,* 2004). Tallman and Taylor (1999) reported that dysfunctional neuroendocrine-endocrine interactions can contribute to this disturbance in trace metal metabolism thereby leading to severe complications seen in diabetic patients.

In this study, serum zinc, calcium and magnesium were observed to be significantly lower in both types of diabetes mellitus when compared to the control. These results are in agreement with those of Abou-Seif and Youssef (2004) that investigated the levels of these metals in type I and type II diabetes mellitus. The findings that the levels of these metals were lower in the sera of NIDDM patients than in IDDM patients may suggest that oxidative stress is more in NIDDM than in IDDM (Abou-Seif and Youssef, 2001). Since diabetics excrete more of these metals than non-diabetics, the lower levels of zinc, calcium and magnesium in the sera of both types of diabetics may be attributed to impaired absorption and/or excessive loss in the urine of diabetics (Brown *et al.,* 1999).

Reports have shown that zinc has insulin-like effects since it causes or enhances glucose uptake. It has been proposed that zinc enhances the uptake of glucose by inhibiting glycogen synthetase thereby blocking the conversion of glucose to glycogen (Rao *et al.,* 1987). As such, the decreased level of zinc in the plasma of diabetic patients may be another contributory factor to the high blood glucose observed in them.

Magnesium is involved in glucose homeostasis at multiple levels. It catalyses the various enzymes involved in the phosphorylation of glucose both at its anaerobic metabolism and oxidative decarboxylation in the citric acid cycle.

**Table 1: Fasting blood glucose and anthropometric indices of diabetic patients resident in Abakaliki, South East, Nigeria**

| Parameters | Control(n = 30) | NIDDM(n = 30) | IDDM(n =30) |
|---|---|---|---|
| **Sex (M/F)** | 17/3 | 21/9 | 18/12 |
| **Age (years)** | 45.0 ± 6.40 | 48.0 ±7.12 | 46.0 ± 5.61 |
| **Weight (kg)** | 78.8 ± 4.85 | 80.6 ±7.91 | 81.3 ± 8.15 |
| **Height (cm)** | 158.1 ± 16.51 | 161.0 ± 9.66 | 160.3 ±12.08 |
| **Fasting blood glucose (mmol/L)** | 3.87± 0.25 | 12.67 ± 2.98 | 13.12 ± 2.02 |

**Table 2: Concentrations of trace metals in the sera of diabetic patients resident in Abakaliki, South East, Nigeria**

| Groups | Zinc (µg/dl) | Calcium (mg/dl) | Copper (µg/dl) | Magnesium (mg/dl) | Lead (µg/dl) |
|---|---|---|---|---|---|
| **Control** | 91.11 ± 3.67 | 4.96 ±0.51 | 99.72 ±2.36 | 2.10 ±1.24 | 21.0± 5.11 |
| **IDDM** | 71.35 ± 4.01 | 3.04 ±3.11 | 121.05 ±1.66 | 1.30 ±0.98 | 30.00 ± 2.55 |
| **NIDDM** | 50.91 ± 3.42 | 2.33 ±2.24 | 133.14 ±2.08 | 0.40 ±0.31 | 38.00 ±3.02 |

It is a co-factor in the glucose transport system of hepatocyte plasma membranes and it also regulates hepatocyte mitochondrial functions (Srivastava *et al.,* 1993). Reports have also shown that magnesium also plays a role in the release of insulin, the maintenance of the pancreatic β-cell cycle, as well as increasing the affinity and number of insulin receptors (Elamin and Tuvemo, 1990).

Magnesium depletion has been reported to have atherogenic potential. Hypomagnesaemia has been postulated as a possible risk factor in the development and progression of diabetic retinopathy (McNair *et al.,* 1978; Zarger *et al.,* 2002). Studies have shown that patients with myocardial infarction had reversal of abnormal lipoprotein patterns to normal on administration of magnesium (Seelig and Heggtyeit, 1974). Pham *et al.* (2009) reported that the lower serum magnesium levels observed in type II diabetics could be associated with more rapid decline in renal function in such patients.

The higher levels of copper and lead in the sera of both types of diabetes noted in this study are consistent with the findings of Akinloye *et al.* (2010) and Abou-Seif and Youssef (2004). The increased levels of copper ions observed in diabetes mellitus might be attributed to hyperglycaemia that may stimulate glycation and release of copper ions from copper-containing enzymes. This argument has been supported by Lin (1996) who reported elevation in the concentrations of both lenticular copper ions and the protein-unconjugated copper ions than that of protein conjugated copper ions. According to Lin (1996), this increase resulted in the decrease in the reactivity of copper-containing enzymes such as superoxide dismutase and ceruloplasmin in the lens of diabetic patients.

It has been speculated that higher levels of lead in the blood of diabetics may be a contributory factor to the decline in renal function noted in these patients. Many studies have associated high blood lead levels to decline in kidney function (Staeseen *et al.,* 1992; Tsaih *et al.,* 2004). Again, since one of the signs of lead toxicity is impaired renal function and one of the most prominent complications of diabetes mellitus is kidney damage (Mailloux, 2007), it is likely that the higher blood lead observed in diabetic patients may be associated with the kidney damage complication of diabetes mellitus. Studies have also demonstrated toxic effects due to exposure to lead and have suspected high blood lead levels as a contributory factor to the progression of diabetic complications in diabetic patients (Tsaih *et al.,* 2004).

Studies have shown that the toxic effects due to lead can be reduced by the presence of other trace metals. Micronutrients such as zinc

and calcium affect lead toxicity by interacting with lead at its primary site of action (Peraza *et al*., 1988; Bogden *et al.,* 1992; Babalola *et al.,* 2007). Zinc for instance has been reported to influence both tissue accumulation of lead and susceptibility to lead toxicity (Dioka *et al.,* 2004). Available evidence suggests that as dietary zinc increases, lead absorption and its subsequent toxicity decreases (Peraza *et al.,* 1988; Dioka *et al.,* 2004). Therefore, the impairment in renal function attributed to the high levels of lead in the blood of the diabetics can be decreased by increasing the available of these micronutrients in the diet. Foods such as fish, cereals, fruits and vegetables are known to be rich in these micronutrients.

In conclusion, this study has shown that there is disturbed trace metals metabolism in diabetes mellitus. Abnormalities in the metabolism of trace elements may induce the secretion of pancreatic amylase, another diabetic complication in diabetes mellitus. The findings in this study suggest that increased concentrations of lead and copper and decreased levels of zinc, calcium and magnesium may be a contributory factor to the pathogenesis and progression of diabetes mellitus.

## REFERENCES

ABOU-SEIF, M. A. M. and YOUSSEF, H. (2001). Oxidative stress and male IGF-I, gonadotropin and related hormones in diabetic patients. *Clinical Chemical Laboratory Medicine,* 39(7): 618 – 623.

ABOU-SEIF, M. A. and YOUSSEF, A. (2004). Evaluation of some biochemical changes in diabetic patients. *Clinica Chimica Acta,* 346: 161 – 170.

AGGETT, P. J. and CORNERFORD, J. G. (1995). Zinc and human health. *Nutrition Review*, 53: 16 – 22.

AKINLOYE, O., OGUNLEYE, K. and OGUNTIBEJU, O. O. (2010). Cadmium, lead, arsenic and selenium levels in patients with type II diabetes mellitus. *African Journal of Biotechnology,* 9(32): 5189 – 5192.

ALBERTI, K. G. M. M. (1991). The health of the nation: role of diabetes. *British Medical Journal,* 303: 769 – 772.

BABALOLA, O. O., OJO, L. O. and AKINLEYE, A. O. (2007). Status of the levels of lead and selected trace elements in type II diabetes mellitus patients in Abeokuta, Nigeria. *African Journal of Biochemistry Research,* 1(7): 127 – 131.

BAGHURST, P. A., ROBERTSON, E. F. and WIGG, N. R. (1987). The port study. Lead effects on pregnancy outcome and early childhood development. *Neurotoxicology,* 8: 395 – 405.

BOGDEN, J. D., GERTNER, S. B., CHRISTAKOS, S., KEMP, F.N., YANG, Z., KATZ, S. R. and CHU, C. (1992). Dietary calcium modifies concentrations of lead and other metals and renal calbindin in rats. *Journal of Nutrition,* 122: 1351 – 1360.

BROWN, I. R., MCBAIN, A. M., CHALMERS, J., BROWN, E. R., CAMPBELL, I. W. and LEWIS, M. J. (1999). Sex difference in the relationship of calcium and magnesium excretion to glycaemic control in type I diabetes mellitus. *Clinica Chimica Acta,* 283: 119 – 128.

COHEN, R. M., FIELDMAN, G. M. and FERNANDETZ, P. C. (1997). The balance of acid, base and electrolytes in health and disease. *Kidney International,* 52: 287 – 293.

DIOKA, C. E., ORISAKWE, O. E., ADENIYI, F. A. A., MELUDU, S. C. and AKEN, E. E. (2004). Biochemical assessment of lead overload and the protective effect of zinc in guinea pigs. *Journal of Biomedical Investigation,* 2(2): 76 – 81.

ELAMIN, A. and TUVEMO, T. (1990). Magnesium and Insulin-dependent diabetes mellitus. *Diabetes Research and Clinical Practice,* 10: 203 – 209.

GRUNDY, S. M., BENJAMIN, I. and BURK, J. (1999). Diabetes and cardiovascular disease. A statement for health care professionals. *American Heart Association Circulation,* 10: 1134 – 1146.

ISBIR, T., TAMER, L., TAYLOR, A. and ISBIR, M. (1994). Zinc, copper and magnesium in insulin-dependent diabetes. *Diabetes Research,* 26(1): 41 – 45.

JONES, C. A., KROLEWSKI, A. S. and ROGUS, J. (2005). Epidemic of end-stage disease in

people with diabetes in the United States population: do we know the cause? *Kidney International,* 67: 1684 – 1688.

KANEKO, J. J. (1999). *Clinical Biochemistry of Animal.* 4th edition. KANEKO, J. J. (Editor). Academic Press Incorporated, New York.

LIN, J. (1996). The association between copper ions and peroxidative reaction in diabetic cataract. *Nippon Gakkai Zasshi,* 100(9): 672 – 679.

LIONEL, O. P. (2007). Metabolic syndrome. *Circulation,* 115: 32 – 35.

MAILLOUX, L. (2007). Upper date dialysis in diabetic nephropathy. *Nephropathy,* 9: 2 – 5.

MCNAIR, P., CHRISTIANSEN, C., MADSBAD, S., LAURITZEN, E., FABER, O., BINDER, C. AND TRANSB, I. J. (1978). Hypomagnesaemia - a risk factor in diabetic retinopathy. *Diabetes,* 2(7): 1075 – 1077.

NEEDLEMAN, H. I. and GATSONIS, C. A. (1990). Low level lead exposure and 1Q of children. *Journal of American Medical Association,* 263: 673 – 678.

NIEWOEHNER, C. B., ALLEN, J. I. and BOOSALIS, M. (1986). Role of zinc supplementation in type II diabetes mellitus. *American Journal of Medicine,* 81: 63 – 68.

PERAZA, A., MAJORED, F., AYALA, F., DAVID, S. and LEONARD, T. R. (1988). Effects of micronutrients on metal toxicity. *Environmental Health Perspective,* 106: 203 – 215.

PHAM, P. C., PHAM, P. M., PHAM, P. T., PHAM, S. V. and PHAM, P. A. (2009). The link between lower serum magnesium and kidney function in patients with diabetes mellitus type II deserves a closer look. *Clinical Nephrology,* 71(4): 375 – 379.

PIDDUCK, R. G., WREN, P. J. and EVANS, D. A. P. (1970). Hyperzincuria of diabetes mellitus and possible genetically implications of this observation. *Diabetes,* 19: 240 – 247.

RAO, K. V. R., SESHIAH, V. and KUMAR, T. V. (1987). Effect of zinc sulfate therapy on glucose and lipids in type I diabetes. *Journal of Association of Physicians India*, 35: 52 – 53.

SEELIG, M. S. and HEGGTYEIT, A. (1974). Magnesium interrelationship in ischemic heart disease. American Journal of Clinical Nutrition, 27:59 – 79.

SRIVASTAVA, V. K., CHAUHAN, A. K. and LAHIRI, V. L. (1993). The significance of serum magnesium in diabetes mellitus. *Indian Journal of Medical Sciences,* 47(5): 119-123.

STAESSEN, J. A., LAUWERYS, R. R., BUCHET, J. P., BULPILT, C. J., RONDIA, D. and VANRENTERGHEN, Y. (1992). Impairment of renal function with increasing blood lead concentration in the general population. The cadmibel study group. *New England Journal of Medicine,* 327(3): 151 – 156.

STOIEK, E. and STOCZYNSKA, A. (2003). Lead effect on vascular endothelium. *Medical Practice,* 54: 87 – 93.

TALLMAN, D. L. and TAYLOR, C. G. (1999). Potential interactions of zinc in the neuroendocrine- endocrine disturbances of diabetes mellitus type II. *Canadian Journal of Physiology and Pharmacology,* 77: 919 – 933.

TIERNEY, L. M.. MCPHEE, S.J. and PAPADAKIS, M. A. (2002). *Current Medical Diagnosis and Treatment.* International Edition, Lange Medical Books/McGraw-Hill, New York.

TRINDER, P. (1969). Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. *Annals of Clinical Biochemistry*, 6: 24 – 27.

TRIPTYCH, S., SUMATHI, S. and RAJ, G. B. (2004). Minerals nutritional status of type II diabetic subjects. *International Journal of Diabetes in Developing Countries,* 24: 27 – 28.

TSAIH, S. W., KORRICK, S., SCHWARTS, S., AMARASIRIWARDENA, C., ARO, A., SPARROW, D. and HU, H. (2004). Lead, diabetes, hypertension and renal function: the normative aging study. *Environmental Health Perspective,* 112(11): 178 – 182.

WALTER, R. M., URIU-HARE, J. Y., OLIN, K. L., OSTER, M. H., ANAWALT, B. D., CRITCHFIELD, J. W. and KEEN, C. L . (1991). Copper, zinc, manganese and

magnesium status and complications of diabetes mellitus. *Diabetes Care,* 14(11): 1050 – 1056.

WEDDEN, R. P., MAESAKA, J. K., LIPAT, G. A. and VITALE, L. F. (1975). Occupational lead nephropathy. *American Medical Journal,* 59(5): 630 – 641.

ZARGER, A. H., BASHIR, M. T., MASSODI, S. R., LAWAY, B. A., WANI, A.T., KHAN, A. R. and DAR, F. A. (2002). Copper, zinc and magnesium levels in type I diabetes mellitus. *Saudi Medical Journal,* 23(5): 539 – 542.