Original Article

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Correlation of Metal Ions in Diabetic Patients

Biochemistry Section

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ABSTRACT

Introduction: Diabetes Mellitus (DM) presents with insulin deficiency results in raised blood sugars. Metals are the inorganic elements present in very small amounts in the living tissues and play an important role at cellular level and hence are important for life. Macro metals like Magnesium (Mg) are present in large quantity and hence are called as macro nutrients.

Aim: To evaluate the correlation of metal ions in Type I and Type II DM.

Materials and Methods: It was a prospective study, carried out at Acharya Vinoba Bhave Rural Hospital, JNMC, Sawangi (Meghe), Wardha, Maharashtra, India from January 2017 to January 2019. Total 260 patients were enrolled in the study. The participants were divided in two groups, diabetic and non-diabetic, based on blood sugar levels. The blood samples were analysed for Calcium, Magnesium, Phosphorous and Iron. The comparison was done using student's t-test.

Results: The mean values of Iron (Fe) were 162.17±13.27 µg/dL, Calcium (Ca) 8.21±0.34 mg/dL, Mg 1.49±0.07 mEq/L, and Phosphorous (P) 2.92.±0.49 mg/dL for the whole study population. The various metal levels when compared with the levels of fasting and post prandial blood sugar in Non-diabetic group and Diabetic group the difference was statistically significant for Fe, Ca and P and was not significant for Mg.

Conclusion: Fe levels were raised and Ca, Mg and P were depleted in patient suffering from DM.

Keywords: Calcium, Macronutrients, Magnesium, Micronutrients, Phosphorus

INTRODUCTION

Long-term vascular complications of DM represent a major cause of morbidity and mortality in patients with DM. Additionally, other biochemical disorders are associated with vascular complications like hyperlipidaemia and oxidative stress [1], these observations suggest that additional factors may be involved in the acceleration of diabetic vascular disease.

Metals are the inorganic elements present in very small amounts in the living tissues and play important role at cellular level and hence are important for life [2]. The requirement of macro nutrients is around 100 mg/day and they are present in high levels in various tissues [4]. On the contrary, some metals like copper (Cu), zinc (Zn), iron (Fe) and manganese (Mn), chromium (Cr), are required in small quantities are called as micro nutrients or trace elements [5]. Metals are involved in various physiological processes [6]. Many metals function as metalloproteinase or metalloenzymes [6,7]. The proteins without metal containing prosthetic groups are not able to perform such physiological functions [8]. Metals enable various functions like muscles contraction, relaxation, and ions help to transmit impulses through the nerves. Biofluids containing metals in soluble salt forms and proper metabolic functioning of these metals depend on their normal levels in various body tissues [2], it's because of such diverse metabolic characters and functions some metals mainly Mg, Cu, Zn, Mn, Fe and Cr are considered as essential for normal human body and health [1].

Metal ions play an essential roles in about one third of enzymes and enzymatic reactions [1]. These ions control the enzyme catalysed reactions by modifying electron flow in a substrate or enzyme and without such appropriate metal ion any biochemical reaction catalysed by a particular metalloenzyme would progress very slowly [1].

Metal ions play an important role in growth, development and the metabolism in living system. Impaired metabolism of trace elements is observed in diabetic patients. It has been reported that the urinary excretion of Ca is increased in both the types of diabetes mellitus causing a decrease in blood levels of these elements [9,10]. Another study reported that the level of Mg was significantly lower in serum of patients with Insulin Dependent Diabetes Mellitus (IDDM) [11]. The vascular complications are more common in poorly controlled diabetes patients and are because of tissue hypoxia secondary to paradoxical metabolic imbalance in inorganic phosphate, leading to reduction in high energy phosphate and tissue hypoxia [12]. Exocrine pancreatic function is altered in diabetes; in poorly controlled diabetes amylase levels are raised [13,14]. The importance of metal ions in enzymatic reactions and impairment in metabolism of such trace elements in DM, it is important to study the correlation of these metal ions in diabetic versus non-diabetic patients, considering it, the study was aimed to evaluate the correlation of Fe, Ca, Mg and P in patients suffering with DM.

MATERIALS AND METHODS

It was a case control study, carried out on 260 patients at Acharya Vinoba Bhave Rural Hospital, JNMC, Sawangi (Meghe), Wardha, Maharashtra, India from January 2017 to January 2019. The study was approved by IEC DMIMS (DU) wide approval letter DMIMS (DU)/IEC/2013-14/315.

The consent was obtained from all the participants before enrolment. The participants were divided in two groups: A total of 130 patients suffering with DM that were already diagnosed and on treatment and 130 Non-diabetic patients with normal blood sugar. For diabetic patients fasting >126 mg/dL and post prandial >200 mg/dL were considered cut-off values for analysis. The levels of metal ions (Fe, Ca, Mg and P) were estimated and compared with Non-diabetic population (fasting blood sugar <126 mg/dL and post prandial blood sugar <200 mg/dL). The blood samples were collected first in the fasting state and later 2 hours post meal. All the metal ions were also analysed twice, first with fasting and second time with post prandial samples to avoid reporting and analytical bias.

The various metals were analysed using wet chemistry reagents and reported in units Fe (µg/dL), Ca (mg/dL), Mg (mEq/L), P (mg/dL), respectively. The various tests were done using following methods: GOD-POD method for Plasma Glucose, Arsenazo method for Ca, Calmagite method for Mg, Molybdate UV method for P, Ferrozine

method for Fe.

STATISTICAL ANALYSIS

The statistical analysis was done considering the values of Blood Sugar (BS), fasting BS and Post prandial BS. The corresponding values of Fe, Ca, Mg and P with fasting and post prandial BS were analysed. The mean values of metal ions in the groups after matching the groups were analysed after applying test of significance within the group, mean, Standard Deviation of mean (SD) and Standard Error of Mean (SEM) and student's t-test were calculated using SPSS software.

RESULTS

The mean values of Fe were 162.17±13.27 µg/dL, Ca 8.21±0.34 mg/dL, Mg 1.49±0.07 mEq/L, and P 2.92±0.49 mg/dL [Table/Fig-1].

	Mean	SD	SEM	Median
Iron (µg/dL)	162.18	13.28	6.64	164.545
Calcium (mg/dL)	8.21	0.35	0.173	8.12
Magnesium (mEq/L)	1.49	0.079	0.039	1.495
Phosphorous (mg/dL)	2.92	0.49	0.24	2.865

[Table/Fig-1]: The values of Fe, Ca, Mg and P levels.

The mean values of fasting Fe was statistically higher in diabetic group than the non-diabetic group, while the Ca, Mg and P levels were lower [Table/Fig-2].

Parameters	Non-diabetic group (N=130) Fasting BS <126 mg/dL	Diabetic group (N=130) Fasting BS >126 mg/dL	p-value
Iron (µg/dL)	156.23±1.50	172.86±8.24	p<0.0001
Calcium (mg/dL)	8.28±1.32	7.93±0.49	p=0.0050
Magnesium (mEq/L)	1.58±0.49	1.46±1.038	p=0.2336
Phosphorous (mg/dL)	3.22±1.53	2.51±1.19	p<0.0001

[Table/Fig-2]: The values of Blood sugar in Nondiabetic and Diabetic Groups and corresponding Iron, Ca, Mg and P levels.

The mean values of post prandial Fe was statistically higher in the diabetic group. Ca, Mg and P levels were lesser in the diabetic group than the non-diabetic [Table/Fig-3].

Parameters	Non-diabetic group (N=130) PP BS <200 mg/dL	Diabetic group (N=130) PP BS >200 mg/dL	p-value
Iron (µg/dL)	146.24±6.29	173.38±7.90	p<0.0001
Calcium (mg/dL)	8.67±1.40	7.96±1.89	p=0.0007
Magnesium (mEq/L)	1.53±0.89	1.40±1.99	p=0.4971
Phosphorous (mg/dL)	3.46±1.33	2.49±1.67	p<0.0001

[Table/Fig-3]: The values of Blood sugar in Nondiabetic and Diabetic Groups and corresponding Iron, Ca, Mg and P.

DISCUSSION

It was a prospective study conducted to evaluate the correlation of Fe, Ca, Mg and P in patients suffering with DM. The levels of these metal ions were evaluated in patients suffering with DM and were compared with the control group.

Fe levels were found to be statistically higher while P levels were statistically lower in the diabetic group. When post prandial BS and metal levels were compared, it was found that the results were in linear correlation to that of fasting BS levels and the status of corresponding metal ions. Pancreatic islets are affected and cause diabetes because of imbalance of some essential metals [15].

Iron

In this study, Fe was increased in patients with raised fasting and postmeal BS, in patients with DM. The mean values of Fe were 156.23±1.4990 in non-diabetic group and 172.86±8.2387 in diabetic group and the difference was statistically significant (p<0.0001) in fasting sample group and it was 146.24±6.2846 in non-diabetic group and 173.38±7.8964 in diabetic group and the difference was statistically significant (p< 0.0001) in post prandial group.

Fe is essential for synthesis of haemoglobin, myoglobin and elastin production [17]. Transferrin transports free Fe into cells. Newly diagnosed diabetics have increased ferritin, which stores free Fe [18,19].

A study by Jiang R et al., reported higher level of ferritin in diabetics as compared to the non-diabetic subjects [20]. This study had also similar findings where the Fe levels are raised in diabetic population and the difference of Fe levels was statistically significant (p<0.0001). Another study found a positive correlation between serum ferritin and Fe deposition in tissues, and that, it increases linearly with the duration of diabetes [21]. Haemochromatosis is a condition where Fe overload occurs and an elevated serum Fe is an index for this [22]. Several studies showed association between haemochromatosis and type II diabetes [21,22]. Oxidation of biomolecules such as nucleic acids, proteins and lipids by an elevated level of Fe may contribute to development of Type II DM by decreasing insulin secretion from pancreatic beta cells and also by concomitant increase of insulin resistance [23]. Sharifi F et al., observed a linear correlation of serum Fe levels and insulin resistance [24].

Calcium

A frequent dyselectrolytemia and other electrolyte disturbances like ketoacidosis, nonketotic hyperglycaemic hyperosmolar syndrome are associated with depleted levels of Ca, Mg, Potassium and Phosphate. This is because of the levels of intracellular Ca triggers insulin release. In this study, the difference of serum Ca was statistically significant (p=0.0050 and p=0.0007). But it is significant to note that the values observed were within normal reference ranges, though the difference was statistically significant. This may be due to minor variation in the reported range. El Rahman Hassan SA et al., reported, Serum Ca 8.87±0.08 mg/dL in diabetic patients and 9.51±0.07 in control group with a statistically significant difference among Sudanese population [25]. Other studies also showed that the mean of serum Ca level was lower in patients with type2 DM than in general population [26,27].

Another study by Suh S et al., observed that Ca level changes are positively correlated with HbA1c and fasting plasma glucose levels [28]; it was concluded that elevated serum Ca levels are associated with Type II DM.

Magnesium

Mg is the most abundant macro-nutrient. About 300 enzymes are dependent on Mg, which are involved in glucose homeostasis, nerve transmission, DNA and RNA production [29,30].

In this study, difference of Mg levels was statistically not significant (p=0.2336) and (p=0.4971). Mg deficiency may cause decrease in insulin-mediated glucose uptake. Diabetics were shown to have low blood Mg and raised urinary Mg [31]. Animal model studies found that, Mg supplementation prevented insulin resistance and reduced development of diabetes [32].

A study reported by Ramadass S et al., wherein 50 patients of type II DM were analysed for fasting and post prandial glucose, HbA1c and Mg and then divided into patients with good, poor control and those who need intervention [33]. It was found that Mg levels were inversely proportional to the HbA1c levels and also with the duration of type II DM. It was concluded that lower levels of Mg are associated with uncontrolled Type II DM and the duration of disease. In the index study also, it was evident that Mg levels were depleted in patients suffering with DM [34].

Phosphorus

Phosphorus is important for storage, transfer, and liberation of

energy in the organism as well as in the intermediate metabolism of carbohydrates, fat, and proteins. Inorganic phosphate is an important component of DNA and RNA and also involved in glycolysis and oxidative phosphorylation and hence in Adenosine Triphosphate (ATP) formation [35].

In this study, P was statistically lower in the diabetic group. Similar reports were observed by Linyan F and Xueping L, where 224 were enrolled and observed that the serum level of phosphate in type II diabetic group was significantly lower than that in the control group ($p<0.05$) [36]. It was concluded that levels of P was depleted in Type II DM which is indicative of P metabolism disorders.

Limitation(s)

The renal excretion and associated co-morbidities like nephropathy, hypertension and other diseases were not considered in this study. The duration and treatment was not taken into account during the analysis.

CONCLUSION(S)

It is concluded from the study that the normal levels of metals like Fe, Ca, Mg and P are disturbed in patients with DM. It was observed that Fe levels were raised and Ca, Mg and P were depleted in patient suffering with DM. The difference of levels of Fe, Ca and P in diabetic and non-diabetic groups was statistically significant; however Mg was not significant.

It is recommended to study the levels of these metal ions considering the duration of DM, creatinine clearance and diabetic control and dietary supplements of these elements.

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