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Original Research Article.

Serum Zinc Level in Persons with Prediabetes and it's Relation with Glycemic Status Attending Tertiary Care Hospital in Bangladesh

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ABSTRACT

Background: Type 2 diabetes mellitus is becoming one of the major health problems worldwide. Especially in South East Asia, type 2 diabetes has gained critical significance. As prediabetes prevalence is increasing worldwide, it has become an important concern to prevent diabetes at an early stage in Bangladesh.

Objectives: Estimation of serum zinc level and establishment of its relation with glycemic status in individuals with pre-diabetes.

Materials and Methods: This cross-sectional study encompassed 126 (age: 35.09±9.96 years, mean ± SD; Sex: 16/110, M/F) subjects with prediabetes and 126(age: 29.08±9.28 years, mean ± SD; Sex: 22/104, M/F) healthy nondiabetic controls from the out-patient department of Endocrinology, BSMMU consecutively. Serum zinc was measured by using Atomic Absorption Spectrophotometry. Height, weight, waist circumference, acanthosis nigricans, hypertension, SGPT& serum creatinine were recorded as confounding variables.

Results: Serum zinc level in persons with prediabetes was lower than that in control (0.76±0.01 vs. 0.78±0.01mg/L, M±SEM, p=0.28). There was statistically significant difference for zinc level in gender groups (M vs. F: 0.84±0.02 vs. 0.75±0.01 mg/L, M±SEM, p<0.001) and monthly family income groups (p=0.02). Also zinc level was statistically similar among glycemic status groups apart from zinc level in between control and combined glucose intolerance (CGI) groups (control vs. CGT: 0.78±0.01 vs. 0.72±0.02mg/L, M±SEM, p=0.03). Among cases comparisons between groups with or without risk factors like: smoking (0.72±0.03 vs. 0.76±0.10 mg/L, p=0.42), smokeless tobacco (0.73±0.03 vs. 0.76±0.01mg/L, p=0.46), hypertension (0.80±0.03 vs. 0.75±0.01 mg/L, p= 0.14), family history of DM (0.75±0.02 vs. 0.77±0.02mg/L, p=0.52), family history of CVD (0.74±0.02vs.0.77±0.01mg/L,p=0.28), over-weight (0.76±0.01 vs.0.74±0.05mg/L, P=0.59), waist circumference (0.75±0.01 vs. 0.79±0.04mg/L, p=0.40) and acanthosis nigricans (0.75±0.02 vs. 0.76±0.02mg/L, p=0.70), showed no statistically significant difference. None of the variables like age (r= -0.02, p=0.19), BMI (r= 0.14, p=0.12), FPG (r= -0.05, p=0.60) and PG 2h after 75g glucose (r=0.10, p=0.28), HbA1c (r=0.04, p=0.64), serum creatinine (r=0.01, p=0.87) showed significant relationship with the level of zinc except SGPT which showed significant relation with zinc among cases (r= 0.28, p=0.002) and among all participants (r=0.17, p=0.008) but not in control group (r=0.07, p=0.43).

Conclusion: It is concluded that persons with prediabetes had serum zinc level within normal limit and there was found no statistically significant relationship between HbA1c and zinc.

Keywords: Diabetes Mellitus, Serum Zinc Level, Prediabetes, Glycemic Status.

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INTRODUCTION

Diabetes Mellitus is a major public health problem for both developed and developing countries.[1] Considering the differences in risk factors across countries, it is estimated that the number of diabetics will be more than double by the year 2030 compared to 2000.[2,3] Prediabetes is an intermediate stage in the transition from normal glucose tolerance (NGT) to overt type 2 diabetes mellitus and is characterized by impaired fasting glucose (IFG) or impaired glucose tolerance (IGT) or impaired HbA1c.[4] Pre-diabetes

individuals have a six fold increased risk of developing type 2 diabetes compared with those with normal glucose values.[5] In a meta-analysis evaluating the progression of pre-diabetes to diabetes was found to be 4% to 6% for isolated IGT, for isolated IFG 6% to 9% and for both IFG and IGT was 15% to 19%.[6] In the Taranomon Hospital Health Management Center Study the incidence of diabetes was reported as 7% in the group with an HbA1c 5.7% to 6.4%.[7]

The rate of progression to diabetes depends on the degree of insulin resistance and deficiency of insulin secretion as well as other diabetes risk factors, such as age, family history, overweight or obesity or history of gestational diabetes or polycystic ovary syndrome. Unfortunately the prevalence of both diabetes and pre-diabetes is increasing worldwide due to population growth, aging, urbanization, unhealthy food habits and increasing prevalence of obesity and physical inactivity.[8]

Moreover pre-diabetes is a condition that increases the risk of developing macrovascular and microvascular diseases. Moreover diabetes is one of the largest global health emergencies in the 21st century due to considerable cost of appropriate disease control and management of chronic complications.[9] Lifestyle interventions could reduce the incidence of T2DM by 58% over 3 years as Diabetes Prevention Programme Research Group.[10] According to International Diabetes Federation, people living with pre-diabetes are expected to rise from 318 million in 2015 to 481 million by 2040. Among them about 70% lives in low and middle income countries. In Bangladesh, the prevalence of IGT was estimated at 1.71% in 2013. Overall the age adjusted prevalence of diabetes and pre-diabetes was 9.7% and 22.4% respectively.[11] Monitoring blood glucose is an essential part of diagnosis of pre-diabetes and prevention of type 2 diabetes mellitus. However routinely used blood glucose level in patients has a limited value. Since it gives information only about the glucose concentration at the time of sampling and is influenced dramatically by diet. An accurate index of persons mean blood glucose level over a period of 6-8 weeks can be provided by measurement of specific type of glycated haemoglobin (called HbA1c) concentration in blood.[13] Hence HbA1c% provides index of blood glucose control in cumulative sense. It is a reliable indicator of long term hyperglycemia.[12] The role of trace elements for improvement of disturbed metabolic conditions like pre-diabetes and diabetes has been gaining attention.[13]

Zinc is the second most trace-element profusely distributed in the body after iron.[14]In several studies on humans as well as animals have established that zinc plays a major role in the synthesis, storage, release and action of insulin. Metabolically inflammation has been proposed as a key step in the pathogenesis of type 2 diabetes mellitus.[15] In people with IFG hepatic insulin resistance is moderate on the other hand people with IGT have moderate to severe muscle insulin resistance and impaired plasma insulin responses to glucose load.[16,7,8]

The metabolism of several minerals has been reported to be altered in diabetes mellitus and these elements might have specific role in the pathogenesis and progression of the disease – notably zinc.[17] Previous studies have reported reduced concentration of serum zinc in pre-diabetes and diabetes mellitus.[15]

Recently there has been a growing interest in the role of zinc signaling in type 2 diabetes based on the fact that reduced levels of serum zinc have been observed in patients with diabetes[18] The clinical relevance of zinc in type 2 diabetes is further emphasized by recent developments in understanding of dysregulation of zinc partitioning in this disease.[19] Moreover ZNT8 expression responds to variation in zinc and lipid levels in human beta cell, with repercussions on insulin secretion.[20] On the basis of these existing data, it is reasonable to hypothesis that low serum zinc may predispose to insulin resistance and insulin secretion defect and consequently, that zinc supplementation in pre-diabetes may prevent or delay the progression to type 2 diabetes mellitus. This study therefore, set out to determine if there is a difference in serum zinc concentration between in persons with pre-diabetes & normal individuals and to assess its relation with glycemic status in a tertiary care hospital of Bangladesh.

MATERIALS & METHODS

The present case control study was undertaken on newly diagnosed subjects with pre diabetes in the department of Endocrinology, Bangabandhu Sheikh Mujib Medical University (BSMMU) from March 2015 to July 2017. After getting the approval from International Review Board (IRB) of BSMMU, 126 subjects with prediabetes, non-pregnant adults (Age ≥18 years) were enrolled as case and same number of voluntarily participants, who apparently healthy matching with age & sex were taken as control of this study.

Patients suffering from malabsorption syndrome, endocrine disorders, malignancy, liver and chronic kidney diseases were excluded. As well as patients receiving zinc supplementation or drugs that can modify zinc metabolism were not included in this study.

According to Americans Diabetes Association *(*ADA*)* 2016(Standards of Medical Care in Diabetes, 2016) Subjects whose, HbA1c:5.7-6.4% or fasting plasma glucose: 5.6-6.9 mmol/L or Two-hour plasma glucose: 7.8-11 mmol/L during an OGTT were diagnosed as prediabetes. Based on the ADA guidelines 2016 participants attending Endocrinology OPD, BSMMU, with OGTT &HbA1c reports were categorized as Normal glucose tolerance (NGT): FPG< 5.6 mmol/L or 2-h PG in 75-g OGTT <7.8 mmol/L or HbA1c< 5.7%, Impaired fasting glucose (IFG): FPG 5.6 to 6.9 mmol/L, Impaired glucose tolerance (IGT): 2-h PG in 75-g OGTT 7.8 to 11 mmol/L, Combined glucose intolerance (CGI): IFG + IGT, Impaired HbA1c: HbA1c 5.7% to 6.4% .BMI of the subjects were calculated using standard formula, BMI=Weight (kg) / [Height $(m)^2$] and classified as follows: Under-weight: BMI < 18 kg/m^2 , Normal body-weight: BMI ≥ 18 to 22.9 kg/m^2, Over-weight: BMI ≥ 23 to 27.4 kg/m^2 and Obese- BMI: ≥ 27.5 kg/m^2. Waist circumference (WC) was measured at the area midpoint between the lower border of rib cage and the iliac crest. Normal value of WC according to International Diabetes Federation (IFD) criteria: Men: <90cm, Women<80cm in South Asian population. Data was collected in questionnaire after completion of history and physical examination. Then 5 ml blood was taken from each subject in a test tube maintaining all aseptic precaution, Serum was separated and stored at Endocrinology department at -20◦ C for zinc assay. Serum Zinc was measured using Atomic Absorption Spectrometric Analysis. According to Todd et al. 1979 normal serum zinc level was taken as: 0.55 – 1.50 mg/ L. Data were analyzed using computer based SPSS program (version 22.0. P values ≤ 0.05 considered as significant.

Table 1: Demographic Characteristics of the Participants

Parameter	Case(n=126)	Control(n=126)	Total(N=252)	p-Value
Age(Years M±SD)	35.09±9.96	29.08±9.28	32.08±10.06	0.00
Gender [n (%)]				
Male	16(12.7%)	22(17.5%)	38(15.1%)	0.29
Female	110(87.3%)	104(82.5%)	214(84.9%)	
Occupation [n (%)]				
Service holder	18(14.3%)	22(17.5%)	40(15.9%)	
Housewife	81(64.3%)	63(50%)	144(57.1%)	
Business	8(6.3%)	5(4%)	13(5.2%)	0.00
Unemployed	7(5.6%)	4(3.2%)	11(4.4%)	
Others	12(9.5%)	32(25.4%)	44(17.5%)	
Monthly family income (In thousands)				
01-30	91(72.2%)	96(76.2%)	187(74.2%)	
31-60	30(23.8%)	30(23.8%)	60(23.8%)	0.16
61-90	1(.8%)	0	1(0.4%)	
91 and above	4(3.2%)	0	4(1.6%)	
F/H DM [n (%)]	68(54%)	59(46.8%)	127(50.4%)	0.26

Table 2: Clinical and Laboratory Characteristics of the Participants

Characters	Case (n=126) Mean ± SD	Control (n=126) Mean ± SD	Total (N=252) Mean ± SD	p-Value
BMI(kg/m²)	28.77±5.06	26.41±4.84	27.59±5.08	0.00
WC (cm)	98.87±12.40	90.12±12.88	94.49±13.36	0.00
AN [n (%)]	59 (46.8%)	24 (19%)	83 (32.9%)	0.00
SBP	121.63±12.87	114.44±12.65	118.4±13.23	0.00
DBP	79.76±8.89	74.13±9.76	76.95±9.73	0.00
FPG	5.33±0.63	4.66±0.42	4.99±0.64	0.00
PG 2 h GL	8.32±1.39	6.11±0.84	7.22±1.59	0.00
HbA1c	5.79±.36	5.23±.30	5.51±.43	0.00
S. Creatinine	0.73±0.16	0.69±0.15	0.71±0.16	0.07
S. SGPT	28.10±11.68	22.25±8.60	25.18±10.65	0.00

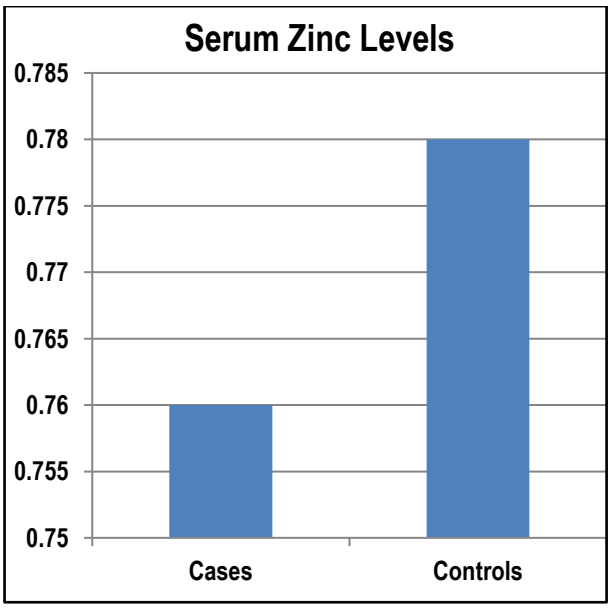

Fig 1: Distribution of serum Zinc levels among Participants (Analyzed by Student's t-test)

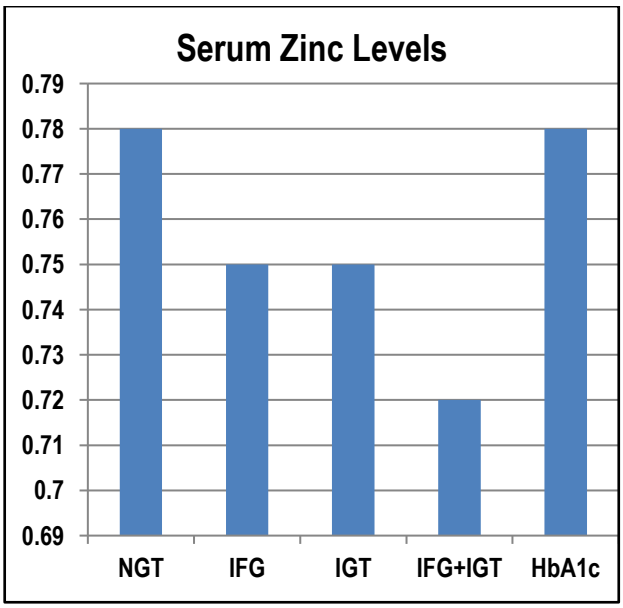

Fig 2: Serum Zinc levels among controls and subgroups of cases (Analyzed by Student's t-test, NGT= Normal glucose tolerance, IFT= Impaired fasting tolerance, IGT= Impaired glucose tolerance, HbA1c= Hemoglobin A1c)

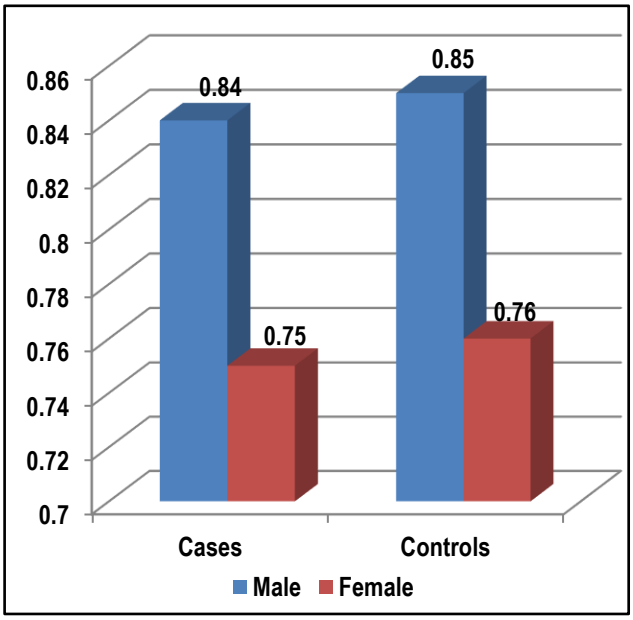

Fig 3: Gender wise distribution of serum Zinc levels among Participants (Analyzed by Student's t-test)

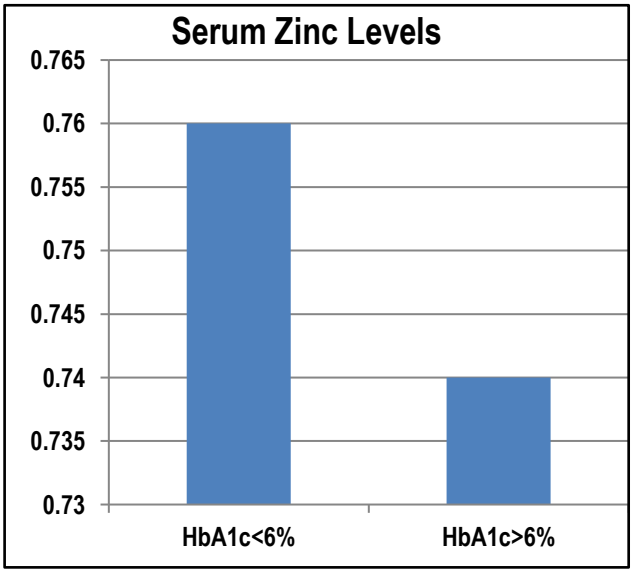

Fig 4: Distribution of serum Zinc levels among cases in relation to HbA1c levels (Analyzed by Student's t-test, HbA1c= Hemoglobin A1c)

RESULTS

The present study measured serum zinc level in 252 participants, among them 126 cases whose mean age was 35.09± 9.96 years and that in control group was 29.08± 9.28 years, (mean± SD) (p=0.00). Female participants were 84.90%. Significant differences in the groups were seen in age and occupation (p-value 0.000 and 0.010 respectively). Cases had mean serum zinc concentration 0.76±0.01mg/L (M±SEM) and that in controls (NGT) was 0.78±0.01mg/L. Zinc level was lower in cases than controls though not statistically significantly different (p= 0.28). Prediabetes individuals were subdivided into IFG, IGT, CGI (IFG+IGT) and only impaired HbA1c. Serum zinc level was 0.75±0.01mg/L in IFG, 0.75±0.01mg/L in IGT, 0.72±0.01mg/L in CGI and 0.78±0.02mg/L in prediabetes individuals with only impaired HbA1c. It was observed that mean zinc value showed decreasing trend with subclasses of increasing intensification of dysglycemia except the subgroup of people with prediabetes with only impaired HbA1c which expressed a similar value (0.78mg/L) to that of control. However zinc level in CGI was lower than that in NGT group and the difference was statistically significant (p=0.03). Here one group having HbA1c≤ 6% had zinc level 0.76±0.01mg/L and another group with HbA1c> 6% 0.74±0.01mg/L (M±SEM). Zinc level was higher in the subgroup of case with HbA1c≤ 6% than that of another group of case with HbA1c> 6% though the difference was not statistically significant (p=0.41). . None of the variables as age, BMI, waist circumference, FPG, Plasma glucose 2 hours after glucose load, HbA1c, Serum creatinine with in the total participants or prediabetes or control group showed statistically significant relation with serum zinc level except SGPT which correlated with zinc in prediabetes group (r= 0.28, p=0.002) and when all subjects combined together (r=0.17, p=0.008) but not in control group (r=0.07, p=0.43).

DISCUSSION

In this study individuals with prediabetes were investigated for serum zinc concentration. It was found that zinc level was statically similar in case and control groups and both the groups had their zinc level within normal range. The present findings also revealed that males had significantly higher level of zinc than that in females irrespective of glycemic status. This is in agreement with the observation of other studies done in subjects with prediabetes.[21, 16] In contrast to the present study, reported that respondents with prediabetes as well as diabetes had lower zinc levels than normal participants in a cross-sectional study among 280 Bangladeshi adults aged ≥ 30 years.[22] It is evident that serum zinc level among persons with prediabetes and diabetics is variable. Participants of this study aged 18 years and above which might have an impact on the disparity of findings. In the present study serum zinc level was found 0.78mg/L in NGT, 0.75mg/ in IFG, 0.75 mg/L in IGT, 0.72 in CGI (IFG+IGT) and 0.78mg/L in pre-diabetes individuals with only impaired HbA1c. The observed serum zinc level showed decreasing trend with subclasses of increasing intensification of dysglycemia including IFG, IGT, CGI except the subgroup of people with pre-diabetes with only impaired HbA1c. However the zinc level in cases with CGI was lower than that in non-diabetics subjects and was statistically significant (p=0.001). It was also observed that subgroups of prediabetes individuals with HbA1c≤ 6% had zinc level 0.76mg/L and that of the group with HbA1c>6% had zinc value 0.74mg/L which was statically similar (p=0.41). Again present study found positive correlation between zinc and HbA1c (r=0.02). This was in accordance with the study done among the diabetes population.[16] The zinc status of Bangladeshi women is unknown; however, 33% of women residing in rural areas are underweight (National Institute of Population Research and Training, Mitra and Associates, and Macro International, 2009)[23], indicative of low energy intakes and presumably proportionally low zinc intakes. In this study majority of participants were female, most of them were in obese & overweight BMI groups and came from urban areas. For this reason their zinc level might be within normal level. Serum zinc concentration was used to assess body zinc status in this study. Zinc status has been measured in a number of tissues such as serum or plasma, different blood cells, hair, and nails.[16] However, serum zinc concentration is viewed as the most

appropriate indicator for evaluating individual's zinc status,, although the sensitivity and specificity of the serum zinc level might be limited by the responsiveness to various confounding factors such as acute stress, infection, On the other hand, it was reported that the mineral level in white blood cell can reflect whole-body mineral status more effectively and seems to be one of the best determinants of mineral status. In addition, we could not find significant relation in between zinc level and glycemic status because we did not measure intracellular zinc which influenced more on glucose metabolism.[24] In this study zinc measurement was done using atomic absorption spectrophotometry (AAS) rather than inductively coupled plasma mass spectrometry (ICP-MS). However, studies showed that ICP-MS provided much lower detection limit, high precision, high accuracy and reliable isotopic analysis compared to other methods for measuring trace elements.[18] So zinc assay might influence our study result. Moreover, the zinc level was considered in light of the reference normal range. It was found that virtually all the subjects had normal zinc level. It may be due to normal range of serum zinc in this region yet not established. People randomly take vitamins and minerals without knowing their names. Drinking of tube well water may also be a factor interfering zinc level. Thus, the factors responsible for the contradictory findings by different investigators remain to be explored further. Regarding gender, zinc level was 0.84mg/L in male participants and 0.75mg/L in female subjects that was statically significant (p= 0.001). That was in agreement with the observation of diabetes subject.[25] But the present study findings were contradictory to the observations.[13] However, Islam et al observed that the level of zinc was lower in diabetic and prediabetes subjects than that for apparently healthy individuals. In relation to different age groups, the level of zinc was not statically significant that went along the findings done by Nsonwu et al studied among subjects with diabetes.[25] In conclusion, zinc level in prediabetes subjects was lower than that in non-diabetic controls though not statistically significant. However the zinc level in cases having both IFG &IGT was lower than that in non-diabetic controls and was statistically significant. Male subjects had higher zinc level irrespective of glycemic status.

CONCLUSION

Individuals with pre-diabetes had serum zinc level within normal limit. Zinc level in pre-diabetes subjects was lower than that in non-diabetic controls though not statistically significant. However the zinc level in cases with both IFG & IGT was lower than that in non-diabetic controls and was statistically significant. Gender and socioeconomic status difference for zinc level were statistically significant. But level of zinc was found to be statistically similar in groups regarding age, BMI or glycemic status (HbA1c).

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