A Study of Serum Magnesium, Zinc, Copper and Glycohemoglobin In Children With Type 1 Diabetes Mellitus

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Abstract:

Magnesium and some trace elements known to act as antioxidants or to be related directly to glucose metabolism may be altered in diabetes mellitus. The aim of the present work is to study the state of these elements in relation to disease duration, control and complications.

The study included 25 children with type 1 diabetes and 13 apparently healthy controls studied for serum magnesium, zinc, copper and blood HbA1c. Serum magnesium, zinc and copper were significantly lower in diabetic children than in controls. Serum magnesium was significantly lower in cases with longer duration of disease (≥5years). Serum magnesium level negatively correlated with the duration of diabetes. Serum zinc was significantly lower in uncontrolled than in controlled and in complicated than in uncomplicated cases. Serum zinc level correlated negatively with the degree of disease control (HbA1c). It is to be concluded that serum magnesium, zinc and copper are deficient in diabetic children. This deficiency may be related to disease control and/or complications. Supplementation of diabetic children with these nutrients may help in control of the disease and its complications.

Introduction:

Diabetes mellitus is a heterogeneous disease characterized by an absolute or relative deficiency of insulin as well as insulin resistance. Trace element status may be altered in diabetic patients.[1] Locally produced reactive oxygen intermediates (ROI's) are involved in the effector mechanism of pancreatic β cell destruction[2] and they are known to disrupt the antioxidant enzymes by releasing trace elements.[3,4] Some trace elements like copper and zinc may act as antioxidants and prevent membrane peroxidation, others e.g. magnesium act directly on glucose metabolism through its role as a cofactor in the phosphorylation of glucose.[5] Previous studies,[6-9] reported derangements of these metals in diabetic patients with cardiovascular complications.

The present study aims at evaluating the changes of serum magnesium, zinc and copper in children with type 1 diabetes in relation to the disease duration, control and complications.

Subjects and Methods:

This study included 25 children with insulin dependent diabetes mellitus (14 males and 11 females, mean age 10.5±3.7 years). Also, 13 apparently healthy children (7 males and 6 females, mean age 10.2±3.4 years) were enrolled in the study as controls. Consent was taken from the parents to enroll their children in the study. All patients were recruited from the emergency unit of the pediatric department, Assiut University Hospital. Controls were coming for clinical evaluation and blood count before some minor operations. Full history and clinical examination were done. A part of blood samples obtained for the initial need of patients and controls was utilized for the biochemical determinations of the present study.

Serum zinc determination:

It was done colorimetrically without deproteinization by using the kits of SENRINEL CH. Milan, Italy.

Principle: Zinc, in a pH 8.6 buffer system, forms with complexant 5-Br-PAPs, a stable colored complex, the color intensity of which is proportional to the amount of zinc in the sample. The interferences, due to oligoelements present in the sample, are eliminated using particular reaction and specific masking agent.

Serum copper determination:

It was done colorimetrically without deproteinization by using the kits of SENRINEL CH -Milan, Italy.

Principle: Copper, liberated in a pH 4.7 buffer system from ceruloplasmin, to which the metal is bound, forms specific complexant 3.5-DiBr-PAESA a stable colored chelate, the color intensity of which is proportional to the amount of copper in the sample.

Serum magnesium determination:

It was done by direct colorimetric method with xylidil Blue-1.

Principle: Magnesium ions form, with xylidil blue-1, in a hydro-alcoholic solution, a blue-violet colored complex, the color intensity of which is proportional to the amount of magnesium in the sample.

Glycosylated hemoglobin (HbA1c) determination: One cubic milliliter of blood in heparinized tube was used. Ion exchange chromatography kits supplied by BIOMEDI Toulouse-France were used.

Principle: After preparing the hemolysate, where the labile fraction is eliminated, hemoglobins are retained by a cationic exchange resin. HbA1C is specifically eluted after washing away the HbA1a+b fraction and is quantified by direct photometric reading at 4/5 nm.

$$\text{HbA1C \%} = \frac{A(\text{HbA1c})}{3 \times A\ (\text{HbA1a+b})} \times 100$$

where A= Absorbance

Normal value 4.2-6.2% (Blisse and Abraham 1985).[10]

Statistical Analysis:

This was done by using student t-test. Values were expressed as mean±standard deviation. Pearson correlation coefficient was used for numeric and homogenous parameters. P value <0.05 was considered significant.

Results:

Table I :Some descriptive clinical criteria of children with type 1 diabetes.

Character	Number	%
• Duration > 5 years	18/25	72
• Duration < 5 years	7/25	28
• Females	11/25	44
• Males	14/25	56
• History of mumps	8/25	32
• Family H of diabetes	9/25	36
• Diabetes control: - Controlled (HBA_{1c}<9%) - Uncontrolled (HbA1c >9%)	 12/25 13/25	 48 52
• Uncomplicated cases	16/25	64
• Complicated cases	9/25	36
* Neuropathy	2/9	22.22
* Nephropathy	2/9	22.22
* Hypertension	1/9	11.11
* Cataract and vitreous hge	1/9	11.11
* Goitre	1/9	11.11
* Dermatitis	1/9	11.11
* Palpitation	1/9	11.11

Table I shows some pertinent descriptive clinical criteria of children with IDDM. Table II shows comparison of serum magnesium, zinc, copper and HbA1c in diabetic cases and controls.

Table II: Serum magnesium, zinc, copper and HbA1c in diabetic cases and controls.

Group	Magnesium mg/dl	Zinc µg/dl	Copper µg/dl	HbA1c %
Diabetics (n= 25)	1.96 ±0.26	103.28 ±15.74	74.56 ±9.15	9.26 ±3.65
Controls (n= 13)	2.22 ±0.22	129.55 ±31.48	166.10 ±38.54	5.18 ±0.64
P	<0.01	<0.05	<0.001	<0.001

Table III: Serum magnesium, zinc, copper and HbA1c in IDDM cases in relation to duration of disease.

Group	Magnesium mg/dl	Zinc µg/dl	Copper µg/dl	HbA1c %
IDDM cases duration ≥5ys (n=18)	1.87 ±0.18	102.09 ±18.17	74.12 ±8.87	9.49 ±3.74
IDDM cases duration <5 ys (n=7)	2.20 ±0.28	106.33 ±6.35	75.69 ±10.47	8.66 ±3.61
P	0.01	N.S	N.S	N.S

Table III shows comparison of serum magnesium, zinc, copper and HbA1c in diabetic children in relation to duration of disease.

Table IV: Serum magnesium, zinc, copper and HbA1c in IIDDM cases <5 years of disease duration and controls.

Group	Magnesium mg/dl	Zinc µg/ dl	Copper µg/ dl	HbA1c %
IDDM <5 years of dis. duration (n=7)	2.20 ±0.28	106.33 ±6.35	75.69 ±10.47	8.66 ±3.61
Controls (n=13)	2.22 ±0.22	129.55 ±31.48	166.10 ±38.54	5.18 ±0.64
P	N.S	<0.05	<0.001	<0.05

Table V: Serum magnesium, zinc, copper and HbA1C in IDDM children ≥5 years of diagnosis versus controls.

Group	Magnesium mg/dl	Zinc µg/dl	Copper µg/dl	HbA1c %
IDDM ≥5years of dis. duration (n=18)	1.87 ±0.18	102.04 ±18.17	74.12 ±8.87	9.49 ±3.74
Controls (n=13)	2.22 ±0.22	129.55 ±31.48	166.10 ±38.54	5.18 ±0.64
P	<0.001	<0.01	<0.001	<0.001

Tables IV and V show serum magnesium, zinc, copper and HbA1c in IDDM cases <5years and ≥ 5 years of disease duration in comparison with controls.

Table VI: Serum magnesium, zinc, copper and HbA1c in controlled versus uncontrolled IDDM cases.

IDDM Group	Magnesium mg/dl	Zinc µg/dl	Copper µg/dl	HbA1c %
Controlled (n=12) (HbA1c<9%)	2.03 ±0.24	110.68 ±17.70	77.86 ±9.29	5.99 ±0.27
Uncontrolled (n=13) (HbA1c>9%)	1.899 ±0.27	96.44 ±10.13	71.52 ±8.26	12.27 ±2.46
P	N.S	<0.05	N.S	<0.001

Table VII: Serum magnesium, zinc, copper and HbA1c in complicated versus uncomplicated IDDM cases.

IDDM Group	Magnesium mg/dl	Zinc µg/dl	Copper µg/dl	HbA1c %
Complicated IDDM (n=9)	1.92 ±0.27	94.24 ±11.09	70.62 ±9.73	13.62 ±1.42
Uncomplicated IDDM (n=16)	1.99 ±0.26	108.36 ±15.94	76.78 ±8.299	6.80 ±1.52
P	N.S	<0.05	N.S	<0.001

Table VI shows comparison of serum magnesium, zinc, copper and HbA1c in controlled versus uncontrolled IDDM cases. Table VII shows the studied parameters in complicated versus uncomplicated IDDM cases.

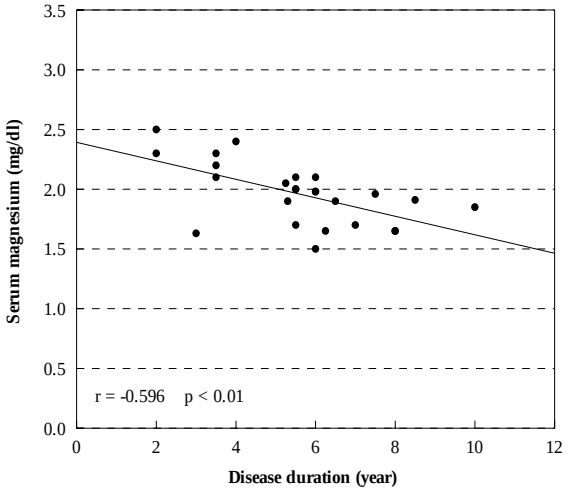

Fig. 1. Correlation between disease duration and serum magnesium level

Figure 1 shows the significant negative correlation between serum magnesium level and known duration of diabetes (r=-0.596, p<0.01).

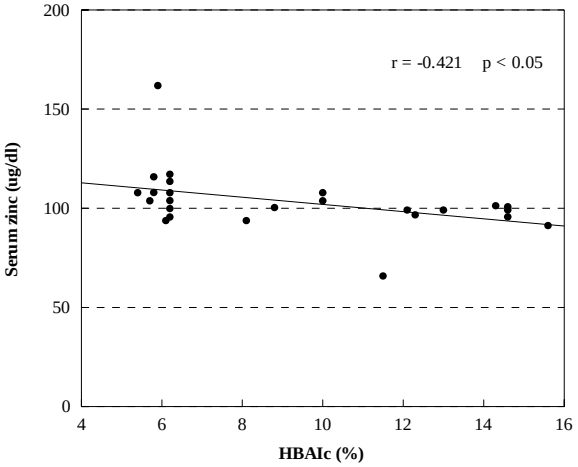

Fig. 2. Correlation between HbA1c and serum zinc in diabetic patients

Figure 2 shows the significant negative correlation between disease control (HbA1c) and serum zinc levels (r= -0.421, p<0.05).

Discussion:

The interrelationship between diabetes and various minerals is characterized by a high degree of reciprocity. Chronic uncontrolled hyperglycemia can cause significant alterations in the status of these nutrients, and conversely some of these substances can directly modulate glucose homeostasis.[11]

In the present study, as regards serum magnesium, children with type 1 diabetes showed significantly lower levels than controls (P<0.01). Patients with disease duration after diagnosis ≥5ys showed significantly lower serum magnesium level than those less than 5 years and than controls (P<0.01 and P<0.001 respect). Such a difference was not noticed on comparing cases less than 5 years of disease duration and controls. These findings are consistent with others,[7,12,13] who found serum and plasma magnesium to be lower in diabetic patients than in controls. Magnesium levels may decrease due to urinary loss by osmotic action, glucosuria and hyperglycemia. Jain and his colleagues,[14] in 1976, showed that hypomagnesemia is common among diabetics and is due to glucosuria causing excessive urinary loss of magnesium. Also, Isbir et al.,[15] in 1994, reported that insulin dependent diabetic patients showed significantly lower magnesium levels than controls (P<0.01). In line with our results, Ewald et al.,[16] in 1983, found an inverse correlation between serum magnesium levels and duration of diabetes in children.

Magnesium is important in maintaining the electrical potential in nerve and muscle membranes and is also involved in glucose homeostasis. It is a cofactor in various enzymatic pathways involved in glucose oxidation.[17] Mooradian and Morley,[11] in 1987, reported that magnesium imbalance has been implicated in diabetes mellitus both as a cause and a consequence.

In the present study, serum zinc was significantly lower in diabetic children than in controls regardless of the duration after diagnosis. It was significantly lower in uncontrolled than in controlled and in complicated than in uncomplicated cases. Serum zinc correlated negatively with glycohemoglobin (HbA1c). In line with our results, Isbir et al,[15] in 1994, reported that serum zinc is significantly lower in patients with insulin dependent diabetes mellitus (IDDM) than levels measured in matched controls. Also, Quilliat et al.,[18] in 2001, reported that diabetes due to chronic pancreatitis was associated with decreased plasma zinc concentration. Furthermore, Sitasawad et al.,[1] in 2001, showed that zinc was deficient in serum, leukocytes and hemoglobin of IDDM subjects than in controls.

Zinc is an essential constituent of enzymes in many major metabolic pathways and is found in nucleic acids and bone. The role of zinc in carbohydrate metabolism has been the subject of considerable

interest. Approximately 0.5% of crystalline insulin is zinc.

Zinc deficiency has been associated with reduced insulin secretion and increased tissue resistance to insulin action.[19] Also, zinc enhances the binding of insulin to hepatocyte membranes[20] and potentiates the lipogenic effect of insulin in adipocytes.[21] Severe zinc deficiency may cause glucose intolerance, but its role in the pathogenesis of diabetes is not proven.[22] The lower serum zinc in diabetic subjects is probably the result of diabetes related hyperzincuria and impaired intestinal absorption of zinc.[23] Mocchegiani et al.,[24] in 1989, reported that IDDM subjects have reduced levels of thymulin which is a biomarker of zinc biological activity.

As regards serum copper level in this study, it was significantly lower in diabetic children than in controls regardless of the duration of diabetes (P<0.001) and these levels are not affected by the state of control or complications of diabetes. These data are in line with Williams et al.,[25] in 1995, who proved that copper and zinc were reduced in diabetic children providing evidence of a role of these antioxidants in this disease. Sjogren et al.,[26] in 1986, reported that skeletal muscle content of copper in type 1 diabetes may be reduced. Craft and Failla,[27] in 1983, and Lau and Failla,[28] in 1984, found that in streptozocine-induced diabetic rats, intestinal copper absorption and urinary excretion are increased which is reversed by insulin therapy.

Conversely, our results are inconsistent with those of Noto et al.,[29] in 1983; Walter et al,[9] in 1991 and Zargar et al,[30] in 1998 who reported hypercupremia in diabetic patients compared to controls. However, they relate this hypercupremia to older age and to use of contraceptives by some diabetic women studied or to disease complications such as retinopathy and hypertension.

It is noteworthy that in the present study, all children are below eleven years of age and only one patient showed hypertension and another one showed vitreous hemorrhage. To the best of our knowledge, the pathogenetic implications of the reported hypercupremia are not clear because copper deficiency, not excess, is associated with elevated serum cholesterol levels.[31] Furthermore, copper may be consumed in the oxidation-reduction reactions because it has a dominant role in diverse proteins such as cytochrome oxides, superoxide dismutase and cerulopasmin.[22]

Glycohemoglobin (HbA1c) is significantly higher in diabetic children than in controls and is not affected by the duration of diabetes. It is also significantly higher in uncontrolled than in controlled and in complicated than in uncomplicated cases (P<0.001 for each, tables VI and VII).

HbA1c represents the fraction of hemoglobin to which glucose has been non-enzymatically attached in the blood stream[32] and is a reliable index of long term control of diabetes. Its measurement reflects the average blood glucose in the preceding 2-3 months, consequently, as an indicator of long term glycemic control. Sperling [32] in 2000, reported that the more consistently low the HbA1c level, the better the metabolic control and the less severe and delayed onset of the microvascular complications.

In the present study, HbA1c was 5.18±0.64% in healthy subjects, 5.99±0.27% in controlled diabetics and 12.27±2.46% in uncontrolled diabetics. This is in line with Behrman and his colleagues,[32] in 2000, who reported that in healthy individuals, HbA1c is less than 7%, while values of 6-9% represent good metabolic control, values 9-12% represent fair control and values >12% signify poor control. Also, HbA1c is negatively correlated with serum zinc levels (r=-0.421, P<0.05. fig. 4). In line with this, Holecek et al.,[33] in 1995, reported that exogenous multivitamin and zinc raise plasma zinc, decrease HbA1c and reduce the probability of diabetic complications.

Conclusions:

1. Serum magnesium, zinc and copper are deficient in diabetic children.
2. Magnesium deficiency is more pronounced with longer duration of the disease.
3. Zinc deficiency is more pronounced in uncontrolled and complicated cases.

Recommendations:

Supplementation of children with type 1 diabetes with some micro-and macronutrients may help in control of their blood sugar and in decreasing diabetic complications.

References:

1. Kamata K, Kobayashi T. Changes in superoxide dismutase mRNA expression by streptozocin induced diabetes. Br J Pharmacol 1996; 119: 463-66.

2. Lenzen S, Drinkgern J, Tiedge M. Low antioxidant enzyme gene expression in pancreatic islets compared with various other mouse tissues. Free Rad Biol Med 1996; 20: 463-66.
3. Bond JS, Failla ML, Unger DF. Elevated manganese concentration and arginase activity in livers of Streptozotocin-induced diabetic rats. J Biol Chem 1983; 258: 8004-9.
4. Seelig MS, Heggtveit HA. Magnesium interrelationship in ischemic heart disease: a review. Am J Clin Nut 1974; 127: 59-79.
5. McNair P,Christiansen C, MadsbadS et al. Hypomagnesemia, a risk factor in diabetic retinopathy. Diabetes 1978; 127: 1075-7.
6. Ceriello A, Guingliano D, Dello Russo P, Passariello N. Hypomagenesemia in relation to diabetic retinopathy. Diabetes Care 1980; 5: 558-9.
7. Walter RM, Uriu-Hare JY, Oline KL et al. Copper zinc, manganese, magnesium status and complicationS of diabetes mellitus. Diabetes Care 1991; 14: 1050-6.
8. Blissé E, Abraham ECJ. HbAlc Kit Code: HbA1c8, HbAlc50. Chromatog 1985; 344: 81-91.
9. Mooradian AD, Morley JE. Micronutrient status in diabetes mellitus. Am J Clin Nutr 1987; 45: 877-895.
10. Fujji S, Takemurs MW, Akai T, Okuda K. Magnesium levels in plasma, erythrocyte and urine in patients with diabetes melllitus. Horm Metabol Res 1982; 14: 161-2.
11. Sjogren A, Floren CH, Nilsson A. Magnesium, potassium and zinc deficiency in subjects with type II diabetes mellitus. Acta Med Scand 1988;224: 461-5.
12. Jain AP, Gupto NN, Kumar A. Some metabolic effects of magnesium in diabetes mellitus. J Assoc Phys India 1976; 24: 827-31.
13. Isbir T, Tamer L, Taylor A, Isbir M. Zinc, Copper and magnesium in isulin-dependent diabetes. Diabetes Res 1994; 26(1):41
14. Ewald U, Gebre-Medhin M, Tuvemo T. Hypomagnesemia in diabetic children. Acta Paediatr Scand 1983; 72: 367-71.
15. Yajnik CS, Smith RF, Hockaday TDR, Ward NI. Fasting plasma magnesium concentrations and glucose disposal in diabetes. Br Med J 1984; 288: 1032-34.
16. Quilliot D, Dousset B, Guerci B, Dubois F, Drouin P, Ziegler O. Evidence that diabetes mellitus favors impaired metabolism of zinc, copper & selenium in chronic pancreatitis. Pancreas 2001;22(3): 299- 306.
17. Sitasawad S, Deshpande M, Katdare M, Tirth S, Parab P. Beneficial effect of supplementation with copper sulphate on STZ-diabetic mice (IDDM). J Trace Elements in Medicine and Biology 2001; 52 (2): 77-84.
18. Hotta M, Tashiro F, Ikegami H, Niwa H, Ogihara T, Yodoi , Miyazaki J. Pancreatic B-cell expression of thioredoxin, an antioxidative and antiapoptotic protein, prevents autoimmune and streptozotocin induced diabetes. J Exp Med 1998; 188: 1445-51.
19. Kirchgessner M, Roth HP, Weigand E. (1976): Biochemical changes in zinc deficiency. In Prasad AS, Oberleas D (eds): Trace Elements in Human Health and Disease. Zinc and copper. Vol 1. New York Academic Press 1976: 189-266.
20. Arquilla ER, Packer S, Tarmas W, Miyamoto S. The effect of zinc on insulin metabolism. Endocrinology 1978; 103: 1440-9.
21. Coulston L, Dandona P. Insulin-like effect of zinc on adipocytes. Diabetes 1980; 29: 665-7.
22. Mooradian AD, Failla M, Hoogwerf B, Maryniuk M, Walie-Rosset J. Selected vitamins and minerals in diabetes. Diabetes Care 1994; 17 (5): 464-79.
23. Song MK, Mooradian AD. Intestinal zinc transport. Influence of streptozocine-induced diabetes, insulim and arachidonic acid. Life Sci 1988; 42: 687- 94.
24. Mocchegiani E, Boemi M, Fumelli P, Fabris N. Zinc-dependent low thymic hormone level in type 1 diabetes. Diabetes 1989; 38: 932- 7.
25. Williams NR, Rajput-Williams J, West JA et al. Plasma, granulocyte and mononuclear cell copper and zinc in patients with diabetes mellitus. Analyst 1995; 120 (3): 887-90.
26. Sjogren A, Edvinson L, Floren CH, Abdulla M. Zinc and copper in striated muscle and body fluids from subjects with diabetes mellitus type 1. Nutr Res 1986; 6: 147- 54.
27. Craft NE, Failla ML. Zinc, iron and copper absorption in the streptozocin diabetic rat. Am J Physiol 1983; 244: E122- 28.
28. Lau AL, Failla ML. (1984): Urinary excretion of zinc,copper and iron in the streptozocin-diabetic rat. J Nutr 1984; 114: 224-33.
29. Noto R, Alicata R, Sfoglian L, Neri S, Bifarella M. A study of cupremia in a group of elderly diabetics. Acta Diabet Lat 1983; 20: 81-5.
30. Zargar AH, Shah NA, Massod SR et al. Copper, zinc and magnesium level in non-insulin dependent diabetes mellitus. Postgrad Med J 1998; 74: 665-8.
31. Klevay LM, Inman L, Johnson LK et al. (1984): Increased cholesterol in plasma in a young man during experimental copper depletion. Metabolism 1984; 33: 1112-18.
32. Sperling MA. Diabetes mellitus. In Behrman RE, Kliegman RM, Jenson HB (eds): Nelson Textbook of Pediatrics, 16thed. Philadelphia, Pennsylvania: WB Saunders Co 2000: 1782-85.
33. Holecek V, Racek J, Jerabek Z. Administration of multivatamin combinations and trace elements in diabetes. Cas Lek Cesk 1995; 134 (3): 80-3.