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Serum Trace Elements (Zinc, Copper and Magnesium) in Iraqi Patients with Thalassemia Major Receiving Desferrioxamine and its Relation with Growth State

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Abstract

Background Patients with β-thalassemia major (TM) require periodic blood transfusion and iron-chelating therapy for all their life and they frequently show complications like trace elements abnormalities.

Objectives To evaluate the levels of zinc, copper and magnesium in these patients and to study the relation of these elements with growth state.

Methods Fifty-four patients with TM, with mean age (15.4 years) and age range (7-31) years, and thirty healthy subjects as controls, their mean age (14.7 years) and age range (8-30 years) were included in this case – control study. Each group divided into children and adult groups. Serum levels of zinc, copper and magnesium of all groups were measured calorimetrically. Serum levels of ferritin were measured depending on ELFA technique, in addition to height and weight of all patients and subjects.

Results Mean serum levels of ferritin were significantly elevated in TM patients as compared to controls, while mean serum levels of zinc and magnesium of both patients groups were significantly decreased as compared with control subjects. Mean serum levels of both patients groups were significantly elevated compared to control subjects groups.

Conclusion Patients with TM were suffering from hypozincamia and hypomagnesmia and required additional adjustment, the patients also showed hypercupremia, and there was obvious growth defect in which may be a result of hypozincamia.

Keywords Thalassemia, zinc, height, short-stature

Introduction

Beta thalassemia major is an inherited disease resulting from reduction or total lack of beta globin chains, thalassemic patients need repeated blood transfusion for survival [(1)], and recurrent blood transfusion lead to accumulation of excess iron in the body tissues [(2)] causing progressive organ dysfunction that is fatal without chelation therapy [(3)]. Desferrioxamine (DF) has been the major iron-chelating treatment [(4)]. DF has a low general toxicity, perhaps because of its low lipid solubility [(5)]. Different mechanisms of possible DF toxicity include blockade of critical iron-dependent enzymes and reduction in critical trace elements other than ferric (copper, zinc, magnesium and calcium) [(6)].

Short stature, low weight and sex development delay are common in children with beta-thalassemia, this may be related to iron overload [(7)]. DF-induced dysplasia is associated with height reduction and can be seen in patients receiving DF chelation therapy at doses less than 50 mg/kg/day [(8)]. Iron chelation has been correlated with growth failure and bone abnormalities, and high DF dosage has been associated with cartilage alterations [(9)].

Zinc is one of the essential micronutrients in human and plays a particular role in human growth and development [10]. Patients with thalassmia had shown chronic zinc deficiency. Zinc deficiency may cause hyperzincuria, high ferritin levels, hepatic iron overload and hepatic dysfunction [11]. Short stature, low body weight, aneroxia and hypogonadism were found in the zinc deficient patients and also in most of the thalassemic patients [12]. It is well known that somatomedins mediate growth by contributing to the effect of growth hormone and they require zinc to be synthesized in the liver [13].

The serum concentration of copper in patients with TM depends on several factors including the amount of copper intake in daily diet, intestinal uptake of copper, iron accumulation, kidney function, copper to zinc ratio and administration of DF [14]. Magnesium may be reduced in a number of anemias, including β-thalassemia [15], hypomagnesemia may occur due to hypoparthyoidism [16]. In this study we aimed to evaluate the levels of trace elements (zinc, copper and magnesium) in Iraqi patients with TM and to study the relation between these minerals levels and growth state in these patients. In this study CDC growth charts were regarded as reference for evaluation of growth status [17], as these charts are recommended at inherited blood diseases center at AL-Karamma Teaching Hospital for evaluation of growth status of each patient. By using these charts, the measurements like height, weight can be compared with that of person of the same gender and age. Each chart has smoothed curves or lines that represent growth percentiles, these curves (percentiles) serves as a reference for comparison. The height or weight will be plotted on the grid and then compared to these percentiles [17].

Method

Fifty-four patients with TM [34 male (62.9%), 20 female (37%)] with mean age (15.4 years) and age range (7-31) years who were attending the inherited blood diseases center at AL-Karamma Teaching Hospital periodically, and 30 apparently healthy subjects (18 males, 12 females) with mean age (14.7 years) and age range of (8-30) years as controls were included in the study after obtaining their informed consents. Twenty patients (37.1%) were below 12 years age (children); thirty-four patients (60.9%) were above 12 years age (adult). For control subjects 12 subjects (40%) were below 12 years age (children) and 18 subjects (60%) were above 12 years age (adult). TM diagnosis was based on the clinical, hematological and hemoglobin electrophoresis profiles.

All patients were on periodic blood transfusion (every fourteen days) and daily desferrioxamine at dose of 40 mg/kg body weight given subcutaneously. Exclusion criteria included patients with hepatitis and patients receiving minerals supplements. Blood drawing was done at tenth day of blood transfusion. Eight ml blood samples were collected from each patient and control by vein puncture after 12 hours fast, transferred to 10ml sterile plane tube, allowed to clot for 30 minutes at room temperature and centrifuged at 3000 rpm for 5 minutes to obtain serum. Serum aliquots (about 2 ml) were divided into four 1ml eppendroffs tubes for ferritin, zinc, copper and magnesium estimation. Height of each patient and control subject was evaluated at standing position without head or foot gear with a stadiometer instrument (measuring board with a movable headboard), while weight was calculated with beam balance scale.

Body mass index (BMI) had been estimated from person's weight and height; it was calculated by dividing weight (in kilograms) by height (in square meters). Assessment of growth was made depending on CDC growth charts, which were also used in this center to evaluate growth of the patients, as shown on figure 4, figure 5 and figure 6. Child (patient or control subject) with height for age percentile below third were classified as having short stature, and those with weight for age percentile below third were classified as having underweight [17]. For adult groups, patient or control subject with BMI for age percentile below third were regarded as underweight. Serum ferritin level was measured

by kit from bioMérieux (France) which depends on assay combines a one-step enzyme immunoassay sandwich method with a final Fluorescent detection (ELFA), using VIDAS instrument. Serum zinc and copper were measured by kit from LTA Company (Italy). Serum magnesium was measured by kit from LiNEAR Chemicals Company (Spain).

Statistical analysis

Data were translated into a computerized database structure .An expert statistical advice was sought for statistical analysis using SPSS version 12 computer software. Data in this study were presented as mean ± standard deviation (mean ±SD). ANOVA and student's t-test were used to compare the group means. The linear regression test was applied for the correlation between different parameters, and the significance of the r value was checked using t-test. The P value <0.05 was considered to be statistically significant.

Results

Table 1 shows the demographic characteristics of the subjects. There was non-significant difference between the control subjects and patients regarding gender, age. Mean age of patients group was (15.4 years) and age range (7-31) years, mean age of children patients group was (8.95 years), while mean age of adult patients group was (19.2 years). Mean age of control subjects group was (14.7 years) and age range (8-30 years), mean age of children controls group was (9 years), while mean age of adult controls group was (18.5 years). Serum analysis showed significantly (P <0.05) elevated levels of ferritin in patient group (1725.5±558.1 ng/ml), as compared with that of the control subjects 126.8 ±43.5 ng/ml.

Table 1. Demographic data of the studied groups

Characteristic	Patients group			Controls group		
	children	Adult	Total	Children	Adult	Total
Number	20	34	54	12	18	30
Gender F/M	6/14	14/20	20/34	4/8	6/12	12/18
Age (years)	8.95±1.1	19.2±5.8	15.4±2	9±0.85	18.55±5.6	14.7±6.4
Ferritin (ng/ml)	-	-	1725.5± 558.1	-	-	126.8± 43.5

Serum zinc levels were significantly decreased (P <0.05) in children and adult patients groups (60.25±15.1 µg/dl, 55.17±12.5 µg/dl) as compared to children and adult control subjects (91.6±9.03 µg/dl, 90.6±8.13 µg/dl), respectively. Serum copper levels were significantly elevated (P <0.05) in children and adult patients groups (135.7±45.5 µg/dl) (139.3±46.8 µg/dl) as compared to children and adult control subjects (89.8.8±11.8 µg/dl, 95.2±9.17 µg/dl, respectively). Serum magnesium levels of patients were significantly decreased (P <0.05) in children and adult patients groups (1.69±0.31 mg/dl, 1.77±0.38 mg/dl), as compared to children and adult control subjects (2.03 ±0.19 mg/dl, 2.04±0.21 mg/dl, respectively).

For adult groups, mean of BMI of patients group (19.9±6.2 kg/m^2) was non-significantly lower than that of control subjects (21.76±3 kg/m^2). Twenty from thirty-four patients (85%) having BMI for age percentile below third, compared to none of control subjects.

For children groups, height of patients was non-significantly decreased (113.3±21.4cm), as compared to control subjects (122.8±4.76cm). Sixteen out of 20 patients (80%) having height for age percentile below third, compared to 2 from 12 control subjects (16.6%). Weight of patients were non-significantly decreased (22.9±3.98 kg), as compared to control subjects (25±2.08 kg). Weight for age and weight for height percentiles of all control subjects were above third compared in 7 from 20 patients (35%) and 1 from 20 patient (5%) patients, respectively (Table 2).

Table 2. Age, serum levels of zinc, copper and magnesium, height, weight and BMI of patients groups and control subjects groups.

Parameter	Children			Adults		
	Patients	Control	P value	Patients	Control	P value
Age	8.95±1.14	9±0.85	0.897	19.2±5.8	18.5±5.6	0.701
Zn (µg/dl)	60.2±15.1	91.6±9.03	0.000	55.1±12.5	90.6±8.1	0.000
Cu (µg/dl)	135.7±45.5	89.8±11.8	0.003	139.3±46.8	95.2±9.1	0.000
Mg (mg/dl)	1.69±0.31	2.03±0.19	0.002	1.77±0.38	2.04±0.2	0.009
Weight (kg)	22.9±3.98	25±2.08	0.103	-	-	-
Height (cm)	113.3±21.4	122.8±4.7	0.144	-	-	-
BMI (kg/m^2)	-	-	-	19.9±6.2	21.7±3	0.247

Zn: zinc, Cu: copper, Mg: magnesium, BMI: body mass index

The results of the present study revealed a significant correlation among age, height and weight in children patients and control group (P <0.05). Also there is significant correlation among age and BMI in adult patients and control group (P <0.05).

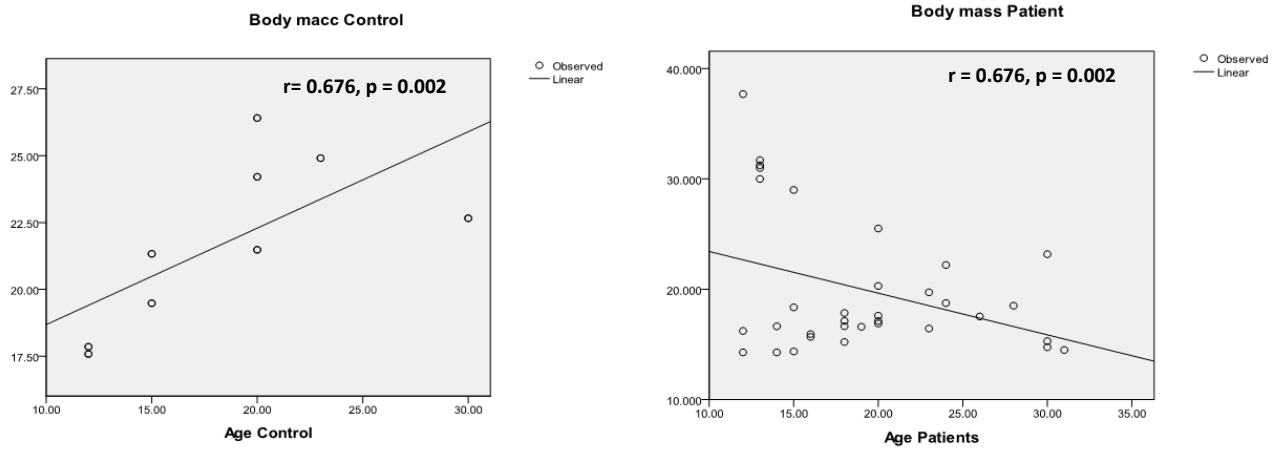

Figure 1. Correlation between body mass index and age in adult controls (left), adult patients (right)

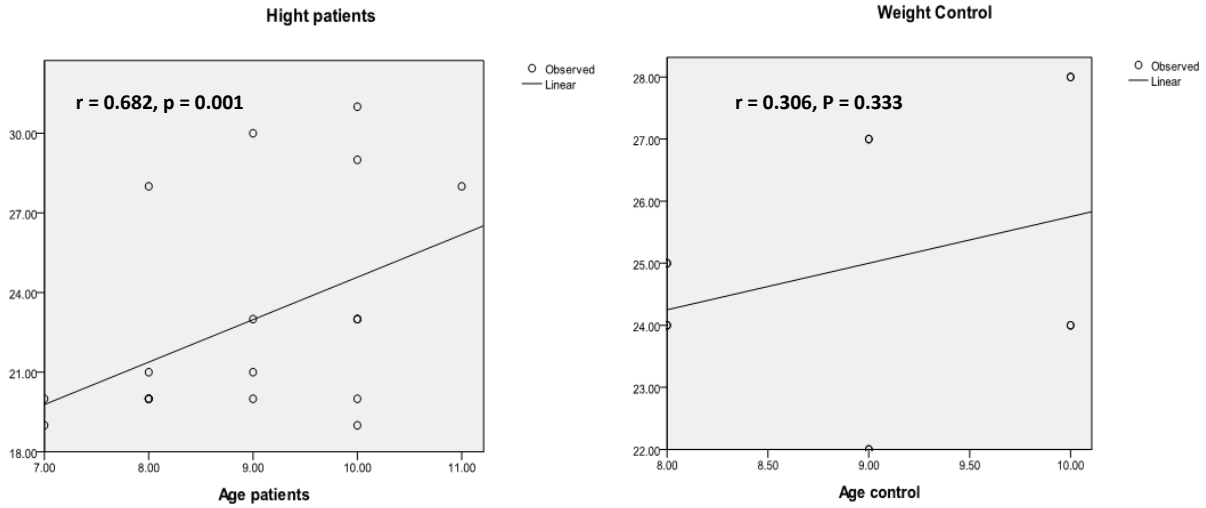

Figure 2. Correlation between weight and age in children patients (left), and children controls (right)

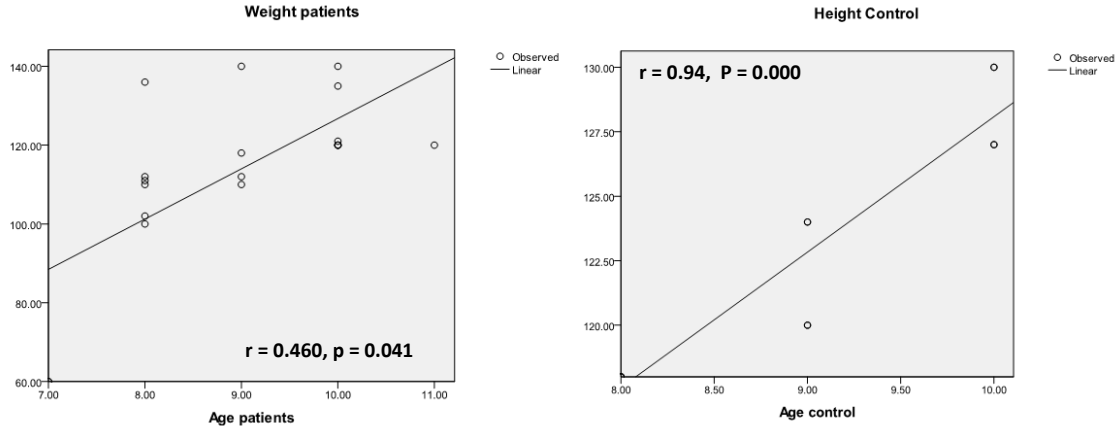

Figure 3- correlation between height and age in children patients (left), and children controls (right)

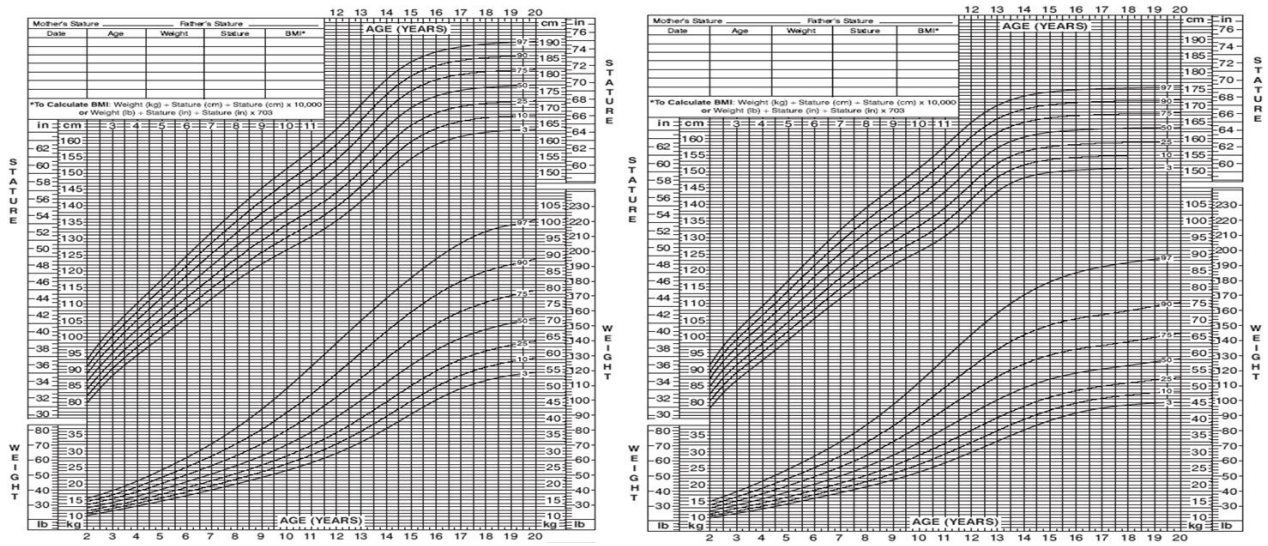

Figure 4. CDC growth chart -age-to weight and age –to stature percentiles (2-20 years) for boys (left), for girls (right).

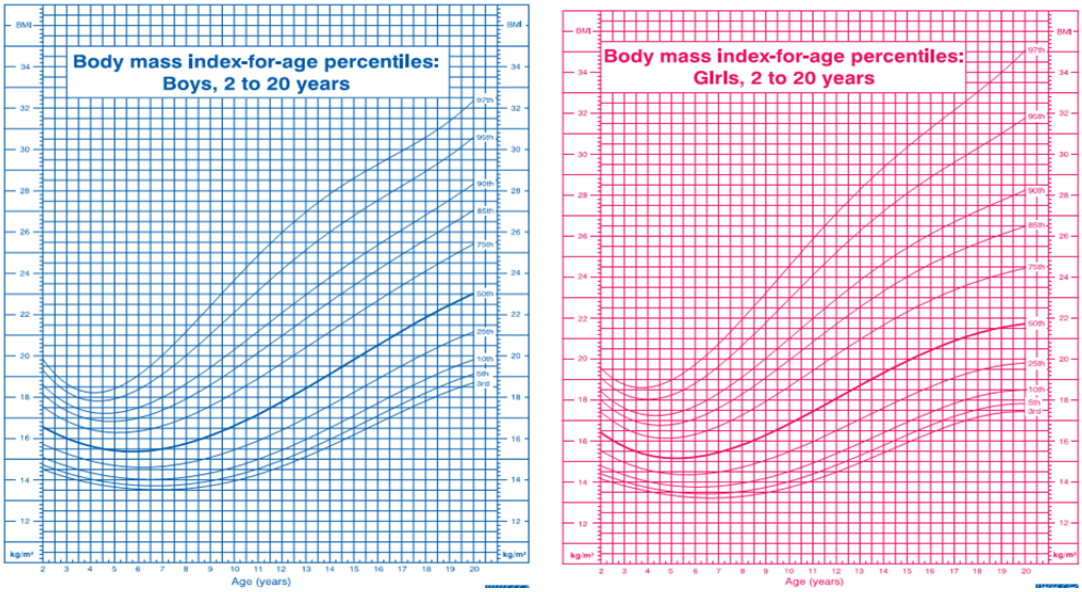

Figure 5. CDC growth - chart body mass index-for - age percentiles (2-20 years) for boy (left), girl (right).

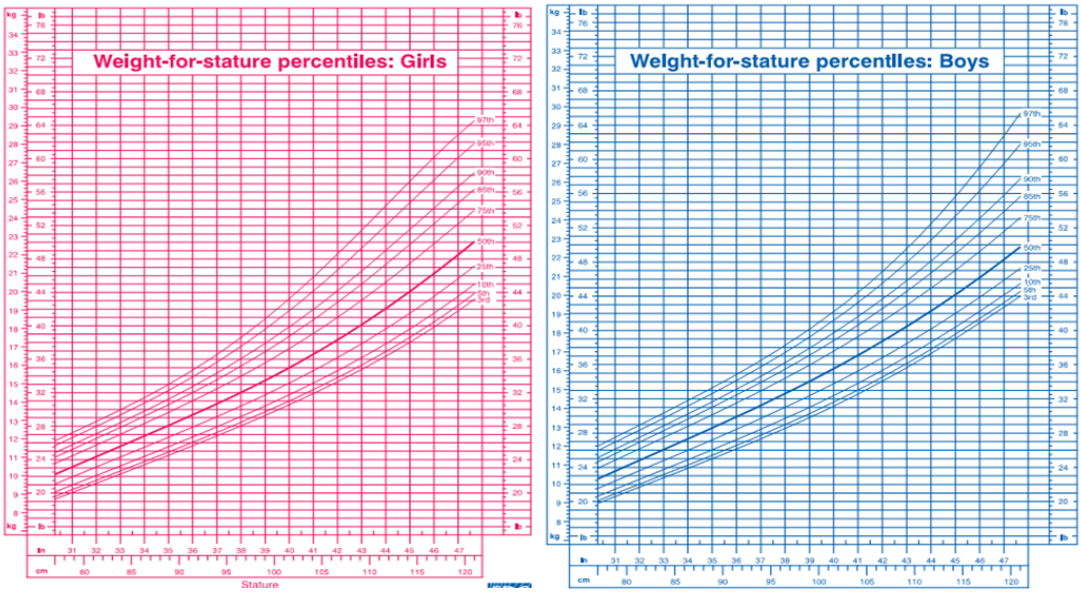

Figure 6. CDC growth chart – weight-for-stature percentiles (2-20 years) for girls (left), for boys (right).

Discussion

In this study, patients with TM showed lower levels of serum zinc as compared to that of control subjects ($p<0.05$), which indicates that most of patients had hypozincemia; this may be related to dietary insufficiency of zinc in those patients in addition to the effects of disease and desferrioxamine administration without dose adjustment for each patient. This result correlates with results of many previous studies [11,14,18,19]. Mehdizadeh et al have reported that mean serum zinc level was significantly higher in thalassemia patient group, and concluded that zinc deficiency was rare in thalassemia [20], Kosarian *et al* reported normal serum zinc level in major thalassemia patients and control, thus they were not affected by zinc deficiency [21]. Previous studies have shown that zinc deficiency is a growth-limiting factor in thalassemia since linear growth in the patients who received zinc supplementation is equal to that of normal healthy children [22]. The causes of zinc deficiency in thalassemia patients may be related to insufficient amount of zinc in daily meals, abnormality in urinary excretion of zinc, kidney dysfunction, disturbance in zinc metabolism and higher level of zinc excretion in sweat [23]. In this study zinc deficiency and iron overload as observed from the elevated levels of serum ferritin of patients can be regarded among factors led to the observed short stature (height percentile below third) and low body weight (weight percentile below third) among children patient and decreased BMI among adult patients, this is similar to that of other studies where short stature was more prevalent in patients above the age of 10 years [24], also in other study in which patients older than twenty years, 75% of girl and 62% of boy having height percentile below third [25].

The levels of serum copper of TM patients were significantly higher than that found in control subjects which correlate with the results of many other studies [16,26], while the study by Kassab-Chekir showed no change in serum copper concentration in thalassemia patients [19]. Bekheirnia [27], Tabatabae [28] and Naser [18] revealed reduction in serum copper concentration. Al-Samarrai et al concluded that the etiology of hypercupremia is hemochromatosis which is principal complication of thalassemia [16]. Hypercupremia in another study in patients with beta thalassemic explained to result from defective erythropoieses and excess denaturation of α and

β globin chains that generate free radicals which lead to oxidative damage [29].

The levels of serum magnesium of patients were significantly decreased when compared to that of control subjects, the decrease in magnesium levels may result from dietary insufficiency of magnesium and the inappropriately high dose of desferrioxamine (40 mg/kg). Previous study showed that magnesium levels were within normal levels [30]. In another study, magnesium depletion in thalassemia patients was documented by low serum magnesium levels, abnormal magnesium tolerance tests, and low symptoms responsive to magnesium therapy [17]. We suggest for further work to estimate the levels of these minerals in patients receiving new classes of treatment used recently in this center.

From the above results we conclude that thalassemic patients suffer from trace minerals abnormalities as they showed hypozincemia, hypomagnesemia and hypercupremia and these abnormalities associated growth defect.

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References

1. Shazia Q, Mohammad Z T, Rahman T, et al. Correlation of oxidative stress with serum trace element levels and antioxidant enzyme status in beta thalassemia major patients: A Review Article. Anemia. Review 2012; Available online at http://www.hidawi.com/in.the press/.
2. Khan FU, Khan MH, Tariq A, et al. Frequency of complications in beta thalassemia major in DI Khan. Biomedica. 2007; 23(6): 31-3.
3. Cohen AR. Management of iron overload in the pediatric patient. Hematol Oncol Clin North Am. 1987; 1(3): 521-44.
4. Daar S, Pathare AV. Combined therapy with desferrioxamine and deferiprone in beta thalassemia major patients with transfusional iron overload. Ann Hematol. 2006; 85(5): 315-9.
5. Porter JB, Huehns ER. The toxic effects of desferrioxamine. Baillieres Clin Hematol. 1989; 2(2): 459-74.
6. Olivieri NF, Buncic JR, Chew E, et al. Visual and auditory neurotoxicity in patients receiving subcutaneous deferoxamine infusion. N Eng J Med. 1986; 314: 869-73.
7. Huang YL, Liu S, Xia T, et al. Relationship between growth disorders and iron overload in children with beta- thalassemia. Zhongguo Dang Dai Er Ke Za Zhi. 2008; 10(5): 603-6.
8. Chan YL, li CK, Pang LM, et al. Deferrioxamine-induced long bone changes in thalassemic patients-radiographic features, prevalence and relations with growth. Clin Radiol. 2000; 55(8): 610-4.
9. Di Stefano M, Chiabotto P, Roggia C, et al. Bone mass and metabolism in thalassemic children and adolescents treated with different iron-chelating drugs. J Bone Mineral Metab. 2004; 22: 53-7.
10. Mahyar A. The preventive role of zinc from communicable and non-communicable diseases in children. NCD Malaysia. 2005; 4(2): 21-6.
11. Arcasoy A, Canata D, Sinav B, et al. Serum zinc levels and zinc binding capacity in thalassemia. J Trace Elem Med Biol. 2001; 15(2-3): 85-7.
12. De Sanctic V. Growth and puberty and its management in thalassemia. Hor Res. 2002; 58(suppl 1): 72-9.
13. Faranoush M, Rahiminejad MS. Zinc supplementation effect on linear growth in transfusion dependent beta-thalassemia. IJBC. 2008; 1: 29-32.
14. Kajanachumpol S, Tatu T, Sasanakul W, et al. Zinc and copper status of thalassemia children. South Asian J Trop Med Public Health. 1997; 28(4): 877-80.
15. Hyman CB, Ortega JA, Costin G, et al. The clinical significance of magnesium depletion in thalassemia. Ann N Y Acad Sci. 1980; 344: 436-43.
16. AL-Samarrai AH, Adaay MH, AL-Tikritti KA, et al. Evaluation of some essential element levels in thalassemia major patients In Mosul district, Iraq. Saudi Med J. 2008; 29(1): 94-7.
17. Center for Disease Control and Prevention (CDC). National Center for Health Statistics (NCHS). National Health and Nutrition Examination Survey Data. Hyattsville MD: United States. Department of Health and Human Services, Center for Disease Control and Prevention, 2008. Vital and Health Statistics Series 11, Number 246. 2000 CDC Growth Charts for the United States: Methods and Development.
18. Nasr MR, Ali S, Shaker M, et al. Antioxidant micronutrient in children with thalassemia in Egypt. East Mediter Health J. 2004; 8(4-5): 490-5.
19. Kassab-Chekir A, Laradi S, Ferchichi S, et al. Oxidant /anti-oxidant status and metabolic data in patients with beta-thalassemia. Clin Chim Acta. 2003; 338(1-2): 71-86.
20. Mehdizadeh M, Zamani G, Tabatabaee S. Zinc status in patients with major beta-thalassemia. Pediatr Hematol Oncol. 2008; 25(1): 49-54.

21. Kosarian M, Valaee N, Mahdyyanea D. Do the desferal receiver thalassemic patients have zinc deficiency? J Mazandaran Univ Med Sci. 2000; 26(10): 1-6.
22. Yazdiha MS, Franoush M. Assessment of serum zinc concentration in thalassemia major children. Pajonesh Dar Pezeshki. 2003; 27(1): 23-5.
23. Quirolo K, Vichinnky E. Hemoglobin disorders. In: Behraman RE. eds. Nelson Textbook of Pediatrics. 18th ed. Philadelphia: Saunders; 2007. p. 2033-9.
24. Hamidah A, Rahmah R, Azmi T, et al. Short stature and truncal shortening in transfusion dependent thalassemia patients: results from a thalassemia center in Malaysia. South Asian J Trop Med Public Health. 2001; 32(3): 625-30.
25. Kwan EW, Lee AW, Li AC, et al. A cross-sectional study of growth, puberty and endocrine function in patients with thalassemia in Hong Kong. J Pediat Child Health. 1995; 31(2): 83-7.
26. Claster S, Wood JC, Noetzli L, et al. Nutritional deficiencies in iron overloaded patients with hemoglobinpathies. Am J Hematol. 2009; 84(6): 344-8.
27. Bekheirnia R, Shamshiraz AA, Kamgar M, et al. Serum zinc and its relation to bone mineral density in beta-thalassemia adolescents. Boil Trace Elem Res. 2004; 97(3): 215-24.
28. Tabatabei M, Kamkar M, Habibzadeh MR. Metabolic and endocrine complications in beta-thalassemia major; a multicenter study in Tehran. Boshehr Med J. 2003; 5(1): 72-3.
29. Vives-Corrous JL, Miguel-Garcia A, Pujades MA, et al. Increased susceptibility of microcytic blood cells to in vitro oxidative stress. Eur J Haematol. 1995; 55(5): 327-31.
30. Arcasoy A, Cavdor AO. Changes of trace minerals (serum iron, zinc, copper and magnesium) in thalassemia. Acta Haematol. 1975; 53: 341-4.

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