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The cutoff of ferritin for evaluation of osteoporosis in patients with Thalassemia Major: A cross-sectional analytic study

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Abstract

Background: This study aimed to assess cutoff of ferritin for evaluation of osteoporosis in patients with Thalassemia Major (TM).

Materials and Methods: This analytic cross-sectional study was conducted in17 Shahrivar children's hospital, Rasht, Iran, from November 2017 to November 2018. The inclusion criteria were indicated as the presence of TM in patients aged 12-19 years old with records of their regular visits. The exclusion criteria were noted as the presence of any chronic bone diseases such as osteomalacia or osteogenesis imperfecta, delayed puberty, hypothyroidism, parathyroid dysfunction, renal failure, liver failure, and growth hormone deficiency. Ferritin level was assessed, and bone densitometry was performed for all patients with TM.

Results: In this study, 53 females (54.6%) and 44 males (43.4%) were enrolled. Results showed that 36 (37.1%), 49 (50.5%), and 12 (12.4%) patients had a normal bone density, osteopenia, and osteoporosis, respectively. Comparing these three groups showed that despite higher mean serum level of ferritin in TM patients with osteoporosis than patients with osteopenia and normal bone density, no significant statistical difference was noted in these three groups ($P > 0.05$). Besides, the mean ferritin level in patients with abnormal bone densitometry (osteopenia and osteoporosis) was higher than in patients with normal ones. A significant difference was noted between abnormal and normal densitometries ($p=0.03$). The Area under the Curve for ferritin was 0.708, and the cutoff point was indicated for ferritin was 2006 ng/ml.

Conclusion: Regarding the results, there was a high frequency of osteoporosis and osteopenia in teenagers with TM. As bone density abnormality formation is time-consuming, and prevention is the primary strategy for management, it is highly recommended to assess bone mineral density regularly starting from early childhood.

Keywords: Ferritin, Osteoporosis, Thalassemia Major

Introduction

Thalassemias are heterogeneous genetic disorders due to decreased synthesis of alpha or beta chains of hemoglobin (Hb) (1). Beta thalassemia results from point mutations in the beta-globin gene. Based on the zygosity of the beta-gene mutation, it has three categories: beta-thalassemia minor, Beta thalassemia major (TM), and beta-thalassemia intermedia. TM is caused by a homozygous mutation of the beta-globin gene (2). Individuals with TM are usually assessed between ages 6 and 24 months; they subsequently require regular red blood cell (RBC) transfusions to survive. Transfusion iron overload is one of the most important complications of TM. Complications of iron overload in children include growth retardation and failure of sexual maturation; in adults, liver fibrosis and cirrhosis, involvement of the endocrine glands, osteoporosis, and cardiac diseases with dilated cardiomyopathy and arrhythmias (3, 4). Although removal of the excess iron in the body is impossible due to lack of the physiological mechanism in the human body, the prognosis has dramatically improved over the past decades with the advent of new methods to measure organ iron before the appearance

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of clinical symptoms, new chelators, and increased blood safety measures (3). Deferoxamine (DFO) is the first approved iron chelator shown to be effective in TM patients. It is effective, but its infusions are time-consuming, expensive, and painful in children. Deferiprone (DFP) is the proper choice for patients who showed an inadequate response to Deferasirox (DFX) and DFO. DFP is the second choice treatment in TM patients when DFO is unavailable. Combination therapy with subcutaneous or intravenous DFX and oral DFP is recommended in patients with severe cardiac dysfunction. DFX is an oral iron chelator used daily and considered the first choice in many countries. (5). Moreover, osteopenia and osteoporosis are among the most common causes of morbidity in adult patients with TM. They are accounted for approximately 40–50% of the increased risk of fractures in this population (6). The pathogenesis of bone abnormality in patients with TM is complex and multifactorial. It may be altered by nutritional status, lifestyle, and type of iron chelators, increased osteoclast maturation, decreased osteoblast activity, and genetic factors (7-11). Researchers recently found that osteoporosis in TM patients occurs due to imbalanced receptor–activator of nuclear factor-kappa B ligand (RANKL)/ RANK, osteoprotegerin, and l as Noggin (12). Reduction of bone mass in patients with TM may also be related to diverse acquired factors including, hepatitis, expansion of bone marrow, growth hormone deficiency, insulin growth factor-(IGF)-1 deficiency, delayed sexual maturation, diabetes, hypothyroidism, parathyroid gland dysfunction, and direct iron toxicity on osteoblasts.They seem to happen due to iron overload (13-14). Meanwhile, iron chelators can effectively reduce the iron burden and improve the aforementioned problems, but they may conversely induce growth failure, bone abnormalities, and cartilage alterations. Previous investigations showed controversial results regarding the effect of administering high-dose DFO (15-16). On the other hand, it was demonstrated that anemia per se may be related to low bone mass in non-transfused E/β- thalassemia patients (17-18). Since ferritin is an easily accessible and relatively reliable laboratory, method especially in developing countries, in this study, investigators aimed to define the cutoff level of ferritin, which may put them at the risk of osteopenia or osteoporosis,s as well as assessing the frequency of bone density abnormality in teenage patients with TM.

Materials and Methods

Patients and settings:

This analytic cross-sectional study was conducted in17 Shahrivar children's hospital, Rasht, Iran, from November 2017 to November 2018. The inclusion criteria were indicated as the presence of TM in patients aged 12-19 years old with records of their regular visits. The exclusion criteria were noted as the presence of any chronic bone diseases such as osteomalacia or osteogenesis imperfecta, delayed puberty, hypothyroidism, parathyroid dysfunction, renal failure, liver failure, and growth hormone deficiency.

Data gathering:

For all patients, bone densitometry was performed. Bone mineral density (BMD) of the lumbar spine (L1-L4) and femur neck was measured by using dual X-ray energy absorptiometry technique (DXA; Hologic, Inc., Bedford, MA, USA). The Z-scores of BMD at the hip and lumbar spine were categorized and recorded in three subgroups: normal: Z score > -1, osteopenia: Z score between -1 and -2.5, and osteoporosis: Z score < -2.5 (19). Demographic characteristics include age, sex, height, weight, body mass index(BMI), along with other variables such as type of iron chelators (DFO, DFX, and DFP), duration of chelation therapy, laboratory results (hemoglobin (Hb), creatinine (cr), phosphorus (p), calcium (Ca) and vitamin D), and status of puberty based on tanner staging were recorded. Mean ferritin levels

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during the previous six months were recorded. Moreover, investigators did not assess ferritin levels during acute diseases routinely.

Statistical Analysis:

Data were reported by descriptive statistics (mean, number, standard deviation, and percent). Kolmogorov- Smirnov test assessed the normality of quantitative data. Normally and non_normal distributed quantitative variables were assessed using the one-sample T-test and the Mann-Whitney U test. Pearson and Spearman correlation coefficients, respectively, assessed the correlation of normal and non-normal distributed quantitative variables. The receiver operating curve (ROC) was designated to indicate the cutoff of ferritin in SPSS version 19. P-value < 0.05 indicated statistical significance.

Ethical considerations:

This study was approved by the ethics committee of the vice-chancellor of research at Guilan University of Medical Sciences, Rasht, Iran (Code= IR.GUMS.REC.1397.459, Date= 2019-02-23). Informed consent was obtained from all participants or guardians.

Results

In this study, ninety-seven patients were enrolled with a mean age of 17.28 ± 2.2 years. Most of them (53 patients) were female (54.6%). The mean weight (kg), height (cm), and BMI (Kg/m2) were 51.7±8.82, 154.7 ±8.31, and 21.23±1.57, respectively. Results showed that the mean level of serum calcium (mg/dL), vitamin D (ng/mL), and phosphorus (mg/dL) were 8.5 ± 0.57, 21.35 ± 8.7, and 4.4 ± 0.44, respectively. Also, the mean serum ferritin during the previous six months was 1613/8 ± 1362 ng/mL. Results showed that 36 (37.1%), 49 (50.5%), and 12 (12.4%) patients had a normal bone density, osteopenia, and osteoporosis, respectively. Comparing these three groups showed that despite higher mean serum level of ferritin in TM patients with osteoporosis than patients with osteopenia and normal bone density, no significant statistical difference was noted in these three groups (P >0.05). Besides, the mean ferritin level in patients with abnormal bone densitometry (osteopenia and osteoporosis) was higher than in patients with normal ones. A significant difference was noted between abnormal and normal densitometries (p=0.03). Furthermore, there was no significant difference between groups in terms of other laboratory parameters (p>0.05) (Table 1). Results showed no significant difference in types of iron chelators, duration of chelation therapy, and status of puberty between patients with abnormal and normal bone densitometry (p>0.05). Assessing age of puberty (years) and duration of chelation therapy (years) showed that the mean age of puberty in patients with normal and abnormal densitometry was 12.75±1.34 and 12.86±1.47, respectively (p=0.569). Also, the duration of iron chelation in the normal densitometry group was 14.19±1.2 and in the abnormal densitometry group was 14.26±1.92 years (p=0.51). In this study, among 54 patients using DFO, 18 patients (33%) had normal, and 36 patients (67%) had abnormal densitometry. Furthermore, among 35 patients who received DFX, 16 (45%) abnormal and 19 (55%) normal densitometry results were reported. DFP was administered for the 8 remained patients and showed 5 (62.5%) abnormal and 3 (37.5%) normal densitometry results. Comparing groups showed no significant relationship between the type of iron chelators and the densitometry status (p=0.98). Table 2 shows a significant linear correlation between the mean ferritin level and score status (p=0.03).The ROC curve was designated to define the diagnostic profile of serum ferritin in identifying abnormal bone densitometry among patients with thalassemia.

The Area under the Curve (AUC) for ferritin was 0.708, and the cutoff point was indicated for ferritin was 2006 ng/ml (sensitivity= 58.3%, specificity=82.4 %) (Figure1)

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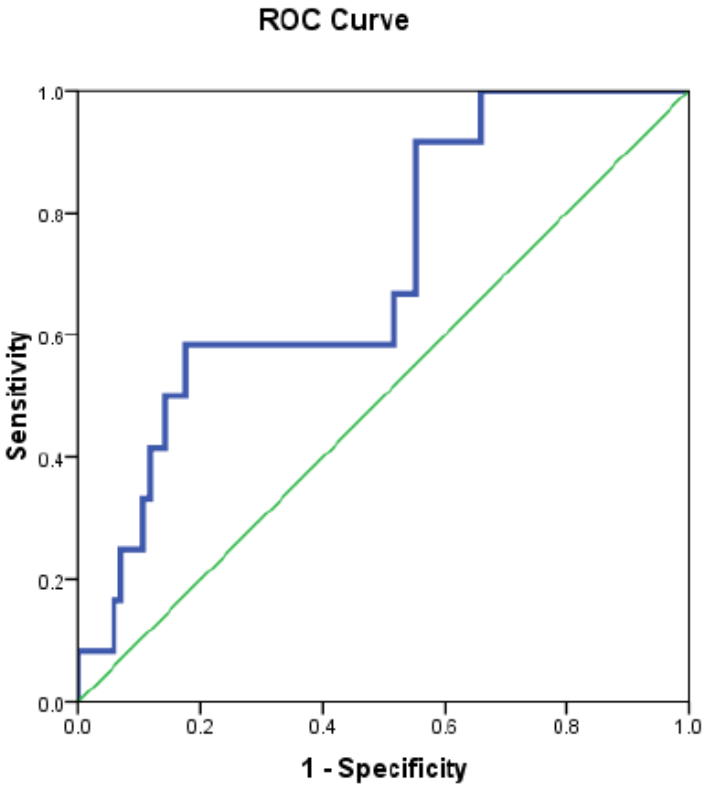

Figure 1. Receiver operating characteristic (ROC) curve of different ferritin cutoffs

Table I: Comparing laboratory results based on densitometry status

Laboratory results	Densitometry status	mean±SD	P-value*
Ferritin (ng/ml)	abnormal	1697.9± 1519	0.03
	normal	1348.8± 910	
Hb (g/dl)	abnormal	8.68± 0.9	0. 86
	normal	8.63± 0.67	
Cr (mg/dl)	abnormal	0.67±0.14	0.441
	normal	0.65±0.09	
Ca (mg/dl)	abnormal	8.55±0.59	0.307
	normal	8.42±0.52	
P (mg/dl)	abnormal	4.08±0.57	0.828
	normal	4.07±0.49	
Vitamin D (ng/ml)	abnormal	21.31± 7.71	0.751
	normal	21.42± 8.76	

*Independent t-test

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Table II: Comparing ferritin level in patients based on Z-score

index	Status	number	mean± SD (ng/ml)	p-value
Z score index	Osteoporosis (<-2.5)	12	2423.7±1924	0.03
	Osteopenia (-1 - -2.5)	49	1644.1±1472	
	Normal (>-1)	36	1348.8±910	

Discussion

In this study, results showed two following important points. First, a high prevalence of BMD abnormality was noted in teenagers with TM, and second, a higher ferritin level was associated with this abnormality. It was mentioned that the yield of developing abnormal bone density raised significantly by reaching the ferritin cut-off point (2006 ng/ml). The estimated frequency of osteoporosis and osteopenia in the present study was 62.8 %. It was a little lower than the study by Shamshirsaz et al. (20) and higher than Skordis et al.(6) and Leung et al. (19) but was well placed in the range reported by Tzoulis et al. (22). Regardless of the study design and implicated factors, the present study, in agreement with previous studies, showed a high frequency of BMD abnormality in TM, even in young patients. Since prevention is the main strategy of the treatment program for osteoporosis, early diagnosis in patients with TM, which should be started since early childhood, is strongly endorsed (23).

Osteoporosis is a major cause of morbidity in TM patients with a complex pathophysiology. It may be altered by nutritional status, lifestyle, and type of iron chelators, increased osteoclast maturation, decreased osteoblast activity, and genetic factors (7-11). On the other hand, it is a frequent problem in disorders characterized by iron overloads, such as thalassemia and hereditary hemochromatosis (24). The relative contribution of these factors is uncertain (25). Although De Sanctis et al. emphasized the effect of TM de novo on bone health without the intervening factors of transfusions, iron intoxication, and chelation (26), the role of iron overload in bone density abnormality in these patients is the subject of increasing interest (25). However, the exact role of iron in the development of osteoporosis in this disorder is not established yet. Excess iron, an inevitable consequence of recurrent blood transfusion, has many direct and indirect harmful effects on bone metabolism. The disturbed balance between bone formation and resorption is reported, which may occur by iron overload and results in bone weakening. A murine model reported a significant correlation between increased reactive oxygen species, serum tumor necrosis factor-α, interleukin-6, and the severity of iron overload in mice. The severity of iron overload in this model has a dose-dependent effect on bone composition trabecular and cortical thinning of bone which may be related to increased bone resorption. These findings lead to bone microarchitecture changes leading to bone loss (24). Evidence suggested that both increased bone resorption and decreased bone formation are involved in pathological bone loss in iron overload conditions (27). Furthermore, Iron induced hypoparathyroidism, GH- IGF axis disturbance, and hypogonadism are among the most studied implicating factors in bone density abnormality in TM patients

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(6, 28, and 29). Despite the limitations of determining ferritin level as an indicator of total body iron burden (30), it is still used worldwide, especially in developing countries, to adjust iron chelation protocol in patients with TM. Previous studies have shown a significant correlation between ferritin level with several severe complications in patients with TM, such as cardiac disease (31), diabetes (32), hypothyroidism (33), Hypoparathyroidism (34), and osteoporosis (35). In this study, investigators found that a ferritin level above 2006 ng/ml put the patients at the risk of a decrease in bone density. In a previous study, investigators found 1953 ng/ml as the ferritin cut-off for hypothyroidism development in patients with TM (36). Together, these findings showed that a ferritin cut-off level of about 2000 ng/ml may be a helpful indicator of improper iron chelation. They indicated the need for more intensified protocols to decrease the risk of some severe complications of TM, including osteoporosis. Regarding the limited sample size and the retrospective type of the present study, the findings such as the relation between BMD based on age and sex needs to be confirmed by further multi-center prospective studies with a larger sample size.

Conclusion

Most patients had abnormal BMD and a higher ferritin level in this study. The yield of developing abnormal bone density raised significantly by reaching the ferritin cut-off point (2006 ng/ml). Regarding the results, there was a high frequency of osteoporosis and osteopenia in teenagers. As bone density abnormality formation is time-consuming, and prevention is the primary strategy for management, it is highly recommended to assess BMD regularly starting from early childhood.

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Conflict of interest

The authors declared no conflict of interest.

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