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Original Article

Bone Mineral Density, Osteoporosis Prevalence and Influential Factors in Osteogenesis in Patients with Beta Thalassemia Major: A Cross-Sectional Study

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Abstract

Background: Osteoporosis and osteopenia considered as severe problems in Beta thalassemia major (BTM) that can lead to bone fractures. We aimed to investigate bone mineral density status and the laboratory parameters related to bone metabolism in BTM patients and compare the statically differences between the mentioned parameters in the patients with or without bone fractures.

Methods: The results of laboratory parameters including; Hemoglobin (Hb), Calcium (Ca), Vitamin D (Vit D), Phosphorus (P), Magnesium (Mg), Alkaline Phosphatase (ALP), Ferritin (FER), Serum Iron (SFe), Thyroid Stimulating Hormone (TSH), T3, T4 and Parathyroid Hormone (PTH), and BMD (by using Dual-Energy X-ray Absorptiometry (DEXA) method) were investigated in 143 BTM patients from thalassemia center of Dezful University of Medical Sciences in 2023.

Results: Seventy-two women and 71 men with confirmed BTM diagnosis with age range (32.4 ± 9.7) were entered in the study. Laboratory parameters including TSH, T3, T4, ALP, FER 247) and SFe showed a significant difference between fracture and non-fracture groups respectively ($P \leq 0.05$). In this study, Z-Score between -1.1 and -2.4 was considered osteopenia, and below -2.5 was osteoporosis. Among the studied patients 36.6%, 39.1% and 24.2% had normal bone density, osteopenia and had osteoporosis respectively.

Conclusion: Osteopenia and osteoporosis had a high frequency among the studied patients that can prone them to bone fractures. Continuous examinations of laboratory tests, especially T3, T4, TSH, FER, SFE, can be helpful for faster diagnosis and therapeutic interventions to prevent the occurrence of osteopenia and pathological fractures.

Keywords: Bone mineral density; Osteoporosis; Osteopenia; Dual-energy x-ray absorptiometry method; Beta thalassemia major

Introduction

Thalassemia is an inherited disorder resulting from defective production of globin chains, leading to ineffective erythropoiesis and hemolysis. Each year, 300,000 newborns worldwide are diagnosed with thalassemia major (TM) (1). Thalassemia affect about 5.2% of the global population (2). Beta thalassemia major (BTM) designates patients who experience severe anemia and often require blood transfusions to sustain life (3). Excessive iron accumulation, particularly in heart, liver, and endocrine glands can result from frequent blood transfusions (4). Hemoglobinopathies, such as BTM, can be fatal during childhood. Nevertheless, since the optimization of the blood transfusion and iron removal program in 1970, the quality of life for these patients has improved; yet they continue to face other obstacles, such as growth and development (1, 5).

Bone health has become a prominent concern for the patients in recent decades (6). The incidence of osteoporosis is approximately 40%-50% among the well-treated patients (7). The relationship between bone health and iron homeostasis is intricate (8). The precise mechanisms of bone disorders in thalassemia have been remained incompletely understood (9). In patients diagnosed with BTM, the occurrence of anemia and inefficient hematopoiesis causes bones widening and enlargement and leads to a thinning of their peripheral area, which stands as the principal reason for bone fragility (10, 11). Other factors that contribute to the hastening of osteoporosis including; postponement of puberty, hypothyroidism, under-functioning parathyroid glands, insufficient growth hormone secretion, diabetes, hypogonadism, iron buildup in bone tissue, vitamin D deficiency, and lack of activity. Osteopenia and osteoporosis are the most common bone disorders in BTM which cause bone fractures and sometimes death (10-13).

We aimed to investigate bone mineral density (BMD) status and the laboratory parameters related to bone metabolism in BTM patients and compare the statically differences between the mentioned parameters in the patients with or without bone fractures.

Materials and Methods

In this cross-sectional descriptive study, 143 BTM patients confirmed with their disease via CBC and electrophoresis tests from thalassemia center of Dezful University of Medical Sciences in 2023 were selected. Blood samples for laboratory parameters were obtained 1 week following transfusion. CBC, TSH, T3, T4, PTH, Vitamin D, Ca, P, ALP, AST, ALT, SFe, FER, and Mg levels were measured. Furthermore, the patients' BMD was measured using DEXA.

Statistical Analysis

The data was then described using frequency and percentage for qualitative variables, and mean and standard deviation for quantitative variables. To examine the data, independent *t*-tests (Mann-Whitney in the absence of normality), Chi-square tests, and Fisher's exact tests were utilized. The Shapiro-Wilk test was used to establish normality of the data. SPSS ver. 16 (Chicago, IL, USA) software was employed for all analyses. The level of significance for this study was set at ≤0.05. Binary regression was used to calculate the odds ratio between fracture incidence and ferritin and serum iron levels of patients.

Ethics approval and consent to participate

This study was conducted in accordance with the Declaration of Helsinki. Ethical approval was obtained from Institutional Review Board and the Ethics Committee of DUMS (IR.DUMS.REC.1401.070). The aims and the methods of the study were verbally explained to the participants. Participation in the study was voluntary, and participants could withdraw from the study at will. They were assured of the confidentiality of their data. Written informed consent was obtained from all the participants prior to their inclusion within the study.

Results

Characteristics of the patients

Between 165 BTM patients after excluding 22 patients from the study due to their unwillingness to participate 143 cases, comprising 71 male (49.5%) and 72 females (50.5%) were evaluated Patient ages ranged from 5 to 63 yr, patients' demography was shown in (Table 1).

Table 1: Comparing Characteristics and history of the patients as (Mean±SD) in total BTM. Abbreviations: SD, standard deviation; BMI, body mass index

Characteristics of the patients			
		Frequency	Percent / Std
Sex	Men	71	49.5%
	Women	72	50.5%
Age ± SD (year)		32.4	± 9.7
Mean height ± SD (cm)	Men	162.2	± 6.86
	Women	155.43	± 5.44
Mean weight ± SD (kg)	Men	59.09	± 9.68
	Women	52.49	± 8.02
Mean BMI ± SD (kg/m^2)	Men	22.51	± 3.72
	Women	21.69	± 2.91
Smoker		19	11.80 %
Alcoholism		8	4.96 %
HIV infectious / hepatitis		0	0

Laboratory findings

Laboratory investigations revealed hypothyroidism in 35 patients, hypoparathyroidism (PTH less than 13pg/ml) in 52 patients, and elevated ALP levels (more than 140 U/L) in 124 patients. The average number of units of packed cells transfused per year showed for the total patients, fracture group, and non-fracture group in Table 2.

The mean FER was 1115.2 ± 183.57µg/l. There was a significant difference in SFe levels between the fracture and non-fracture groups ($P \leq 0.05$). All patients received vitamin D and calcium supplements. The mean level of vitamin D was 31.3 ± 21.1ng/ml and 35.7 ± 16.2 ng/ml for the fracture and non-fracture groups, respectively. There was no statistically significant difference between the two groups ($P \leq 0.05$).

Overall, 53 cases with a history of fractures in the past were recorded in all patients. The fracture group had an average Z-score of -1.41 ± 0.928 for the spine and -1.81 ± 0.731 for the femoral neck. An analysis showed a statistically significant relationship between the patients' fracture history and their TSH, T4, T3, FER, SFe, and ALP levels ($P \leq 0.05$) (Table 2). Binary regression for calculation of odds ratio between fracture risk and laboratory parameters showed ferritin and serum iron levels had a statistically significant difference between fracture occurrences (Table 3).

The mean ± SD spine Z-score and thigh Z-score for all patients were -1.803 ± 1.078 and -1.183 ± 0.918, respectively. Fifty-three participants (37.1%) had a fracture risk, determined by a spine Z-score of -2.5 or less, and 62 out of 143 participants (38.5%) had low BMD, classified by a spine Z-score ranging from -1.1 to -2.4 (Table 4).

Table 2: Comparing of laboratory parameters (Mean±SD) in total BTM, fracture and non-fracture groups

Variable		*Fracture (n = 53)*	*No fracture (n = 90)*	*P-value*	*BTM (n = 143)*
CBC	WBC ($10^3/\mu L$)	9.6 ± 3.4	11.1 ± 5.8	0.328	10.3 ± 4.1
	RBC ($10^6/\mu L$)	3.3 ± 0.4	3.31 ± 0.3	0.0831	3.18 ± 0.3
	PLT ($10^3/\mu L$)	512± 201	459± 225	0.216	485± 213
	Hb (g/dL)	8.5 ± 1.05	8.8 ± 0.94	0.063	8.7 ± 1.0
Transfused units of packed cell /year		13.8 ± 1.2	14.1 ± 1.4	0.708	13.9 ± 1.1
FER (µg/dl)		1200± 122	831 ± 247	0.022*	1115.2 ± 183.57
SFe (µg/dl)		145± 82.5	126 ± 50.7	0.038*	135.8 ± 65.6
TSH (mU/l)		3.87 ± 1.64	3.29 ± 2.19	0.012*	3.58 ± 1.90
T4 (ng/dL)		9.89 ± 1.98	11.56 ± 1.10	0.032*	9.97 ± 1.54
T3 (ng/dL)		1.27 ± 2.88	2.15 ± 0.44	0.039*	2.07 ± 1.66
PTH (pg/ml)		33.6 ± 1.92	36.12 ± 4.5	0.528	34.6 ± 3.2
Vit D3 (ng/ml)		31.3 ± 21.1	35.7 ± 16.2	0.239	33.0 ± 18.6
Ca (mg/dl)		9.2± 0.6	9.3 ± 0.6	0.245	9.2 ± 0.6
P (mg/dl)		4.4 ± 0.7	4.6 ± 1.0	0.712	4.6 ± 1.4
Mg (mg/dl)		2.1± 0.3	2.0 ± 0.2	0.620	2.0 ± 0.2
ALT (U/L)		35.0 ± 22.3	31.5± 19.8	0.598	33.0 ± 21.0
AST (U/L)		32.5 ± 21	33.4 ± 22.9	0.0512	32.4 ± 23.4
ALP (U/L)		536± 85.0	487 ± 98.0	0.020*	511.3 ± 80.8

Abbreviation: WBC, white blood cell; RBC, red blood cell; PLT, platelet; Hb, hemoglobin; FER, ferritin; SFe, serum Iron; TSH, thyroid stimulating hormone; T4, Thyroxine; T3, Triiodothyronine; PTH, parathyroid hormone; Vit D3, 25-Hydroxyvitamin D3; Ca, calcium; P, Phosphorous; Mg, Magnesium; ALT, alanine transaminase; AST, aspartate aminotransferase; ALP, alkaline phosphatase

Table 3: Variable(s) entered on step 1: SFe and Ferritin

		B	*S.E.*	*Wald*	*df*	** Sig.*	*** Exp(B)*	*95.0% C.I.for EXP(B)*	
	Variable							Lower	Upper
Step 1a	Ferritin	.001	.000	21.213	1	.000	1.001	1.001	1.002
	Constant	-1.764	.329	28.725	1	.000	.171		
Step 1a	sFe	.194	.026	53.566	1	.000	1.214	1.153	1.279
	Constant	-22.575	3.049	54.813	1	.000	.000		

The spread of the odds ratio is also displayed. At higher levels of ferritin or serum iron, fractures will occur with an increased risk of 1.001 and 1.214, respectively (**); Significant (*)

Table 4: Comparing of Spine BMD Z-score and Femoral BMD Z-score (Mean±SD) in total BTM, fracture and non-fracture groups. Abbreviation: BMD, bone mineral density; DXA, dual-energy bone densitometry

Variable		*Fracture (n = 53)*	*No fracture (n = 90)*	*P-Value*	*BTM (n = 143)*	*P-value*
Age (yr)	Male	22.14 ± 6.86	29.26 ± 10.62	0.233	25.7 ± 8.78	0.782
	Female	29.10 ± 5.02	31.05 ± 10.94		30.07 ± 9.48	
Sex	Male	23 (40.4%)	50 (55.5%)	0.546	71 (49.5 %)	0.693
	Female	30 (59.6)	40 (44.5%)		72 (50.0 %)	
Spine BMD Z-score		-1.41 ± 0.928	-2.38 ± 0.705	0.0001 *	-1.79 ± 1.065	0.029*
Femoral BMD Z-score		-1.81 ± 0.731	-0.64 ± 0.698	0.0001 *	-1.16 ± 0.908	0.018*

Discussion

Individuals with TM experience decreased BMD and growth during childhood, which impedes their growth and development (14).

Patients with TM experience several hormonal consequences and bone disorders (15, 16). 63.3% of the examined patients have been affected with osteopenia (39.1%) and osteoporosis (24.2%). Previous research studies also have been shown a decrease in bone density in various skeletal regions ranging from 11% to 83.9%, consistent with the findings of this study (17-21).

Considering the endocrine disorders and metabolic complications in TM patients, it is necessary to use appropriate treatment to reduce the complications (22). Medical records revealed that 75 BTM cases experienced puberty disorders, while 34 cases demonstrated delayed puberty (the absence of breast development in girls beyond 13 yr of age and absence of testicular growth to at least 2.5 cm in length in boys beyond 14 yr of age). Among the disorders investigated in this study, diabetes (9.31%), hypothyroidism (6.83%) and hypoparathyroidism (5.59%) were the most commonly observed endocrine disorders.

ALP is recognized as a marker for bone turnover. Its high levels suggest increased bone cellular activity and when value exceeds >700-750 IU/L, in the absence of liver disease, it is associated with osteopenia even if it may not be clinically apparent (23). In the study, serum ALP levels were measured and a significant difference was observed between the fracture and non-fracture groups ($P<0.05$). Serum ALP levels greater than four times the standard adult levels have been recognized as an indicator of bone disease in the absence of any liver disease (11).

In this study, 53 patients (37.1%) had a history of fractures within the past years, which is a significantly higher prevalence compared to Vogiatzi et al.'s study (12.1%) (6). Our study showed a higher level of FER or SFe, increased risk of fracture with odd ratio 1.001 and 1.214 respectively. A FER greater than 1800 µg/L presented a higher risk for endocrine disorders in TM although the level of FER in our study exceeded the normal range; there was no significant difference in ferritin levels between groups with and without fracture. Bone disorders are directly related to increased blood iron levels, and iron overload disrupts osteoblast function, induces apoptosis in these cells and stimulates osteoclast production (24). On the contrary, hormonal disorders in thalassemic patients have indirect impacts on the ossification or formation of bones, and the functionality of cells that generate them. Osteoblast differentiation is stimulated and osteoclast cells are inhibited by using iron chelators (9, 25, 26).

Our study discovered a statistically significant difference in the Z-scores between the fracture and non-fracture groups. It is necessary to moni-

tor the BMD status of thalassemic patients receiving blood transfusion (27). To predict the risk of fractures in such patients, evaluating BMD periodically and determining both clinical and laboratory parameters is crucial.

Conclusion

Osteoporosis and osteopenia are among the most incapacitating complications of TM, resulting in skeletal pain and pathological fractures in affected patients. Iron supplementation, which causes oxidative stress reactions, leads to osteoblast apoptosis and damage. Consequently, early detection and timely intervention aid in preventing the development of pathological fractures in these patients. Using antioxidants in conjunction with iron-chelating medications appears to be extremely beneficial in promoting bone formation in TM patients. Combination of iron chelators like, DFO (deferoxamine), DFP (deferiprone), and DFX (iron deferasirox), with bisphosphonate drugs are commonly administered to prevent osteoporotic conditions. The monoclonal antibody (Denosumab) as an anti-RANKL agent, inhibits the production of osteoclasts and bone destruction. Moreover, it is currently undergoing phase 2b clinical trials for patients over three years of age and with a Z-score of less than 2.5. This treatment may help to reduce bone-related complications in these patients.

Journalism Ethics considerations

Ethical issues (Including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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Conflict of interest

The authors declare that there is no conflict of interest.

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