

# Bone Mineral Density in $\beta$ Thalassemia Major and Intermedia, Correlation with Biochemical and Hormonal Profiles

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Grant sponsor: Iran University of Medical Sciences, Grant number: 555

## Abstract

**Background:** Expansion of bone marrow cavity and decreased cortical and trabecular bone tissues and osteoporosis are resulted from beta-thalassemia. The aim of this study was to assess bone mineral density (BMD) in patients with  $\beta$  thalassemia major and intermedia, and to determine their biochemical and hormonal profiles that may affect BMD.

**Materials and Methods:** In a cross sectional study from October 2004 to April 2006, 305 patients (273 thalassemia major [137 males and 136 females] and 32 thalassemia intermedia [13 males and 19 females]) were evaluated for BMD. Dual x-ray absorptiometry was performed at 3 sites: spine (L2-L4), femoral neck, and radius. Z score < -2.5 was considered as osteoporosis, and between -1 and -2.5 as osteopenia. Z-scores were calculated according to bone density values based on age and sex. Patients were grouped according to age 3-6, 6-10, 10-13, 13-16, and over 16 years old. The stage of puberty was determined according to Tanner staging and its progression was followed. Biochemical and hormonal profiles of patients were recorded.

**Results:** In thalassemia major, mean age was  $14 \pm 6.5$  years, and mean BMD Z-score of spine, radius and hip were  $-2.3 \pm 0.9$ ,  $-2.8 \pm 1.2$ , and  $-1.9 \pm 1.4$ , respectively. Mean age of patients with thalassemia intermedia was  $13.4 \pm 6.2$  years, and the mean Z-score of spine, radius, and hip were  $-2.1 \pm 0.9$ ,  $-2.0 \pm 1.3$ , and  $-2.3 \pm 1.3$ , respectively. Hypogonadism was detected in 36% of thalassemia major and 35% of thalassemia intermedia; but hypothyroidism, diabetes mellitus, and hypoparathyroidism were detected only in thalassemia major with frequency of 2.8%, 1.8%, and 1.2%, respectively. BMD in spine and radius were significantly lower in patients with hypogonadism than in patients with normal puberty ( $P=0.039$  and  $P=0.015$ , respectively). Height Standard Deviation Score (HSDS) was not significantly different in groups of osteoporosis and normal. Osteoporosis was seen in all age groups, and was more common in males than females at spine and radius bones ( $P<0.001$ ). It was less common in patients with hypothyroidism, hypoparathyroidism, and diabetes mellitus. BMD Z-Score had significant correlation with serum ferritin only in radius area ( $P=0.04$ ), and it had no significant correlation with serum Ca, P, Mg and Zn.

**Conclusion:** Our results showed that the patients with thalassemia major and intermedia had low BMD. Patients with hypogonadism and males had lower BMD. Young children also had low bone mass, so early attention is essential.

**Keywords:** Beta-thalassemia, Bone mineral density, Hormones, Osteoporosis.

## Introduction

Beta-thalassemia is a hereditary disease due to unbalanced globin chain synthesis with ineffective erythropoiesis and increased peripheral hemolysis. Expansion of bone marrow cavity and decreased cortical and trabecular bone tissues and osteoporosis are resulted.<sup>1,2</sup> Bone disease in thalassemia is manifested by diffuse bone pain, scoliosis, spinal deformities, nerve compression, spontaneous fractures, and severe osteoporosis.<sup>3-5</sup> Untreated patients with thalassemia major (TM) are presented with severe bone deformities very early in life.<sup>6</sup> In contrast, osteopenia and osteoporosis are major causes of morbidity in patients who survive longer as a result of improved treatment. The aim of treatment in thalassemia is to prevent anemia and bone marrow hyperplasia by regular blood transfusion. With advance in blood transfusion management, there has been a marked reduction in classical thalassemic bone changes. However, well transfused patients require adequate iron chelation therapy, such as desferrioxamine (DFO). DFO has been shown to have a direct toxic effect on bone growth, possibly due to its anti-DNA effects.<sup>7,8</sup> It also chelates trace metals other than iron, most significantly zinc, which is a co-factor for alkaline phosphatase. On the other hand, suboptimal transfusion causes bone marrow hyperplasia which results in thinning of the bone cortex.<sup>9</sup> Iron overload may result in end organ damage, especially in endocrine glands.<sup>10</sup> The etiology of bone disease in thalassemia is multifactorial. Factors such as hormonal deficiency especially gonadal failure, bone marrow expansion, increased iron stores, DFO toxicity, and calcium/vitamin D deficiency all seem to have a serious impact on the impaired bone metabolism in the disease.<sup>9</sup> Studies have found associations between osteoporosis and male gender, hypogonadism, and diabetes mellitus.<sup>5</sup> Several sensitive techniques are now available for quantity assessment of the degree of osteoporosis and total bone mass. Bone density measurement by dual X-ray absorptiometry (DXA) of the lumbar spine, femoral neck, and forearm is recommended as one of the most reliable and non-invasive techniques.<sup>3,11</sup> This study was performed to assess BMD in TM and thalassemia intermedia (TI), and to determine

biochemical and hormonal changes that may affect BMD, and to help prevent progressive disease in patients at young ages.

## Materials and Methods

In a cross sectional study, three hundred and five patients with TM and TI between the ages of 3 and 40 years who came to hematology and endocrinology clinics from October 2004 to April 2006 were enrolled into the study. In TM, most patients received transfusion every 3-4 weeks in order to maintain pretransfused Hb concentration above 9 g/dl. Chelation therapy included DFO 30-40 mg/kg, 5-6 times a week started at the age of 3 years to maintain serum ferritin below 1000 ng/dl. In TI, 26.7% received transfusion periodically (during infection or pregnancy because of low Hb), 26.7% received regular transfusion (because of early phases of bone face deformity or failure to thrive), and 46.7% never received blood transfusion.

The current study protocol was approved by the research deputy of the Iran University of Medical Sciences and Health Services. Informed consent was obtained from all participants. Patients were divided into two groups of TM and TI according to the age of diagnosis, the need of transfusion, and the results of hemoglobin electrophoresis, and follow up of the patients. Patients were also grouped into 3-6, 6-10, 10-13, 13-16, and >16 years old age groups. All patients were examined by one hematologist and one endocrinologist. Puberty was assessed according to Marshall and Tanner staging. Girls older than 13 and boys older than 14 years who did not have any secondary sex characteristics were assessed for delayed puberty. The progression of puberty was noted during follow up period, periodic laboratory tests were done (LH, FSH, estradiol, and testosterone), and questions regarding menarche and ejaculation were asked from the patients who had entered puberty to determine arrested ones. Low dose testosterone for 3 to 6 months was administered to trigger delayed puberty with 6 month intervals of observation and standard deviation scores for height (HSDS) were calculated according to sex and age of CDC 2000. Hypothyroidism with different types and severity was evaluated in the study subjects.

BMD was measured using DXA, Osteocor II France. Results were expressed as grams per square centimeter, Z-score were calculated based on BMD of normal age and sex-matched Caucasian population provided by the manufacturer of the DXA device. BMD was done by one radiologist at 3 sites: lumbar spine (L2-L4), femoral neck, and forearm mid radius (radius). BMD-Z score  $< -2.5$  was considered as osteoporosis, and between  $-1$  and  $-2.5$  as osteopenia. Venous blood samples were obtained to determine serum calcium (Ca), phosphorus (P), alkaline phosphatase (ALP), Zinc (Zn), Magnesium (Mg), ferritin, fasting blood sugar (FBS), T3 (RIA), T4 (RIA), TSH (IRMA), and parathyroid hormone (if needed) for all patients. Analysis of data was performed using SPSS version 13. T-test, Pearson correlation, and Chi-square were used for analysis. Tests were considered significant if P-value was  $< 0.05$ .

## Results

General characteristics of study participants are illustrated in table 1. There was no significant difference in the age of two groups. Mean serum ferritin in TM and TI was  $1618 \text{ ng/ml} \pm 123.8$ , and  $574 \pm 193.6$ , respectively. The frequency of endocrine disorders is shown in table 2. From 175 patients who were at the age of puberty, 63

patients (36%) had hypogonadism which was documented as the most common endocrine disorder. The youngest age of patients with hypogonadism was 13 years.

Seven patients, all from the TM group, had hypothyroidism (4 central and 3 subclinical type). There was significant correlation between serum ferritin and BMD Z-Score in radius area ( $P=0.04$ ), but not in other sites. BMD Z-Score of spine, radius, and femoral neck were lower than normal in both types of thalassemia and in both genders. However, it was significantly lower in TM and male gender at the radius area ( $P=0.048$ ) (table 4). There was no significant difference in BMD Z-Score between males and females in TI. In TM, BMD Z-Score was significantly lower in males than females at spine and radius areas ( $P < 0.001$ ), but in femoral neck it was significantly lower in females ( $P=0.05$ ) (table 4). BMD Z-Score was even lower in younger children showing a negative correlation with age ( $P < 0.001$ ) (table 3, figures 1-3). The youngest child with osteoporosis was 4 years old.

Totally the frequency of osteoporosis and osteopenia was 43% and 50%, respectively. Only 7% of patients had normal BMD. In patients with hypogonadism 30% had osteoporosis, and 62% had osteopenia. In these patients, BMD Z-Score in spine and radius was  $-2.2 \pm 1$  and  $-2.7 \pm 1$ , respectively. It

**Table 1.** Characteristics of study groups.

	TM*	TI
Gender		
Male No. (%)	137 (50.2)	13 (40.6)
Female No. (%)	136 (49.8)	19 (59.4)
Total No.	273	32
Age (mean $\pm$ SD) years	3 – 40 ( $14 \pm 6.5$ )	4 – 28 ( $13.4 \pm 6.25$ )

\* TM: thalassemia major; TI: thalassemia intermediate

**Table 2.** Endocrine disorders in study subjects.

	TM No+/total No (%)	TI No+/total No (%)	Total No+/total No (%)
Hypogonadism	57/158 (36)	6/17 (35)	63/175 (36)
Hypothyroidism	7/243 (2.8)	0/22 (0)	7/265 (2.6)
Hypoparathyroidism	3/245 (1.2)	0/30 (0)	3/275 (1.1)
Diabetes mellitus	5/273 (1.8)	0/32 (0)	5/305 (1.6)

**Table 3.** Z score (mean±SD) of BMD in different age groups.

Age (Year)	Radius	Spine	Femoral Neck	No. of patients (%)
3-6	-3.1±1	-2.6±0.6	-3.5±0.6	45 (14.7)
6-10	-3.1±1	-2.8±0.6	-2.7±1	48 (15.7)
10-13	-3.2±1	-2.5±0.7	-2.4±1.4	51 (16.7)
13-16	-2.7±1	-2.4±0.9	-1.4±.2	62 (20.3)
>16	-2.2±1	-1.8±0.9	-1.02±1	99 (32.4)
all	-2.7±1.2	-2.3±0.9	-2±1.4	305 (100)

**Table 4.** BMD Z Score in two types of thalassemia according to gender.

	Gender	TM	TI	P value*
Spine	Male	-2.6 ±0.8	-2.3±0.9	NS
	Female	-2.1 ±0.9	-2.0±0.8	NS
	Total	-2.3±0.9	-2.1±0.9	NS
P value**		≤0.001	NS	
Radius	Male	-3.3 ±1.1	-2.6±1.5	0.048
	Female	-2.3 ±1	-2.1±1.2	NS
	Total	-2.8±1.2	-2.3±1.3	0.034
P value**		≤0.001	NS	
Femoral neck	Male	-1.8 ±1.6	-2.2±1.8	NS
	Female	-2.2 ±1.3	-1.9±0.9	NS
	Total	-1.9±1.4	-2.0±1.3	NS
P value**		≤0.001	NS	

TM: thalassemia major, TI: thalassemia intermedia.

\* P-value between two types of thalassemia, \*\* P-value between males & females.

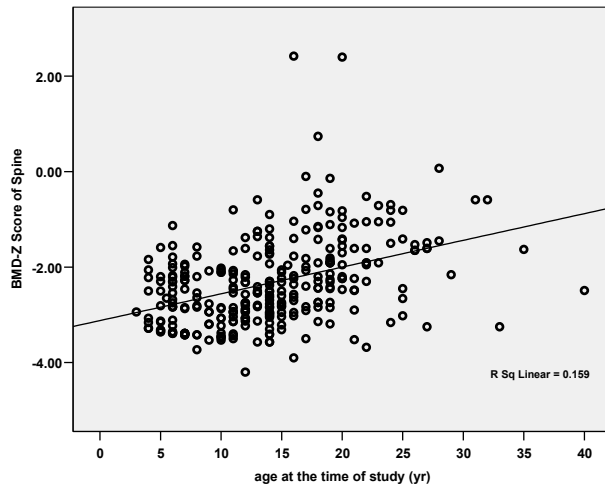
was significantly lower than those who did not have hypogonadism (-1.9±0.9 and -2.3±1) (P=0.04 and 0.015). In hip it was -1.3 in patients with hypogonadism and -1.17 in others showing no significant difference.

Mean HSDS in 3 groups of osteoporosis, osteopenia, and normal patients was -1.20 ± 0.17, -1.28 ± 0.15, and -1.23 ± 0.29, respectively. There was not any significant difference between these 3 groups (P=0.9). There was not any correlation between BMD and serum Ca, P, Mg, Zn, T3, T4, TSH, and ferritin. There was only correlation between serum ferritin and BMD Z-score in radius area (P=0.04). There was not any significant difference between TM and TI in biochemical profiles.

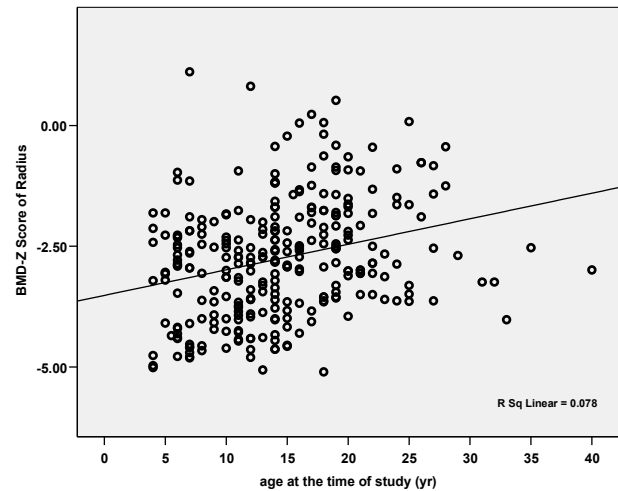
## Discussion

In this study, 305 thalassemia patients (279 TM and 33 TI) were evaluated. We used DXA which still remains the most commonly used method. The advantages of this technique include relatively low radiation exposure, worldwide availability, and short duration of the procedure.

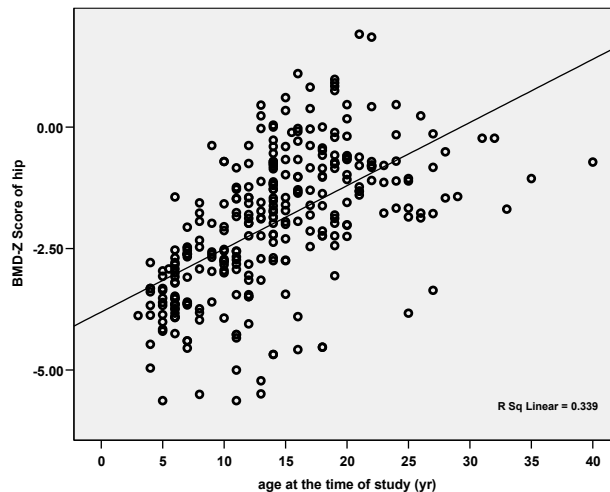
According to this study patients with TM and TI had low bone mineral density. Although the mean BMD value in the TM group was lower than those with TI, there was not a significant difference. In a similar study performed by Mahachoklertwattana et al. 48 children and adolescents with TM and TI were evaluated. Mean BMD values in the TM group were lower than those in the TI group, but they were not significantly different.<sup>12</sup> In another study,



**Figure 1.** Correlation of bone mineral density Z-Score of spine with age.



**Figure 2.** Correlation of bone mineral density Z-Score of radius with age.



**Figure 3.** Correlation of bone mineral density Z-Score of hip with age.

there were 49 TM and 57 TI patients, and both groups showed low BMD.<sup>13</sup>

In our study children with lower age had lower bone density; the same has been reported by others.<sup>14-16</sup> Perhaps it is due to bone marrow expansion in small children with low bone volume. In contrast to our study, in a recent study performed by Christoforidis et al. 35 young thalassemic patients were evaluated. All BMD Z-scores were within normal range with a mean Z-score of 0.42 for girls and -0.41 for boys.<sup>17</sup>

Comparing three sites in our study, radius showed more significant reduction of bone density in both TM and TI. It was in concordance with

Voskaridou study.<sup>15</sup> In most studies, only spine and femoral neck were evaluated, and the most affected site was spine.<sup>8,12-14,18</sup> In our study, there was more reduction in spine bone density compared to femoral neck.

In TM there was significant difference in bone density between males and females; in males it was lower in spine and radius and higher in femoral neck. Christoforidis et al. reported that mean Z-score was lower in boys.<sup>17</sup> Some differences were reported in Jensen et al and Benign et al studies,<sup>5,19</sup> but in other studies no difference was reported.<sup>13,15</sup>

In our study, there was not any correlation between BMD and serum ferritin, Ca, P, ALP, Mg, and Zn.

Some previous studies mentioned suboptimal transfusion, which resulted in chronic hypoxia, delayed puberty, and greater bone marrow expansion as the main reason for low BMD.<sup>12,13</sup> In our patients, there was not any significant difference between patients with pretransfusion Hb>9 gr/dl and Hb less than 9 gr/dl (P-value=0.1). The mean HSDS in 3 groups of osteoporosis, osteopenia, and normal TM and TI patients was not significantly different (P=0.9).

In recent studies, scientific interest has been focused on genetic factors affecting bone mass acquisition.<sup>20-22</sup> Although certain genes have been implicated in the pathogenesis of postmenopausal osteoporosis, they have not been studied thoroughly in thalassemia-induced osteoporosis.<sup>17</sup> We think reduction of bone mass is multifactorial and it needs more evaluation.

## Conclusion

In this study, a large group of patients were evaluated. We studied patients older than 3 years old to find the earliest time of bone density reduction. Our results showed that patients with both thalassemia major and intermedia had low bone mass. In comparison of 3 sites, radius showed more reduction and is a good site for evaluation of BMD. Young children had low bone mass, and need early attention. We suggest annual evaluation of BMD in all thalassemia patients from the age of 3 years old.

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