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Clinical Manifestation of Pulmonary Dysfunction in β -Thalassemia Major

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Respiratory function tests and arterial blood gas analysis were performed on 59 patients with β -thalassemia major (27 M, 22 F, age range: 18-45 years). All investigations were performed 24 h before the patients received a blood transfusion or when they were in a stable state hematologic condition. Echocardiography was performed in all patients and the ejection fraction was employed as a measure of cardiac function. No patient had clinical signs of pulmonary dysfunction. Pulmonary function tests, however, showed a reduction of all main parameters (TLC, FEV1, FEV1/FVC, FEF 25-75% and RV) in most patients with β -thalassemia major, indicating a restrictive type of dysfunction. Arterial blood gas values were within the normal range. There was no evidence that the observed abnormalities in pulmonary function were secondary to congestive heart failure. The low hemoglobin concentration and a fall in the diffusing capacity of the alveolar-capillary membrane, together with the dependence of the reduced pulmonary diffusing capacity on age and serum ferritin levels, as well as of the entity of restrictive disease on age, suggests that pulmonary dysfunctions in patients with TM are due mainly to lung fibrosis and/or interstitial edema related to iron overload. Also iron deposition due to repeated blood transfusions may play a central role in determining lung alterations although the majority of patients are well chelated, suggesting that more than one causal mechanism could be involved.

Key words: β -thalassemia, pulmonary dysfunction, restrictive ventilatory failure

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INTRODUCTION

β -thalassemia major is a transfusion-dependent, inherited, chronic anemia caused by deficient production of β -globin chains that combine to form hemoglobin; consequently, free α -chains precipitate within red blood cells and most erythroid cells die in the bone marrow. Iran, a country 1,648,000 km² wide, has a large number of thalassemia major patients like many other countries in the region (Rahim and Abromand, 2008). β -thalassemia is very rare in Iran. The gene frequency of β -thalassemia, however, is high and varies considerably from area to area, having its highest rate of more than 10% around the Caspian Sea and Persian Gulf. The prevalence of the disorder in other areas is between 4-8%. In Isfahan, a city built around the river Zayandeh-Rood in the central part of Iran, the frequency rises again to about 8%. In the Fars Province, in southern Iran, the gene frequency is also high and reaches 8-10% (Rahim and Abromand, 2008). The increasing workload dictates increasing automation, which may necessitate the use of automated robotic platforms to prepare samples and reactions and the use of automated platforms to perform the actual detection. The complex mutational spectrum of the hemoglobinopathies, especially relevant in a multi-ethnic community, requires a method with the capacity to scan the β (and/or α) globin genes rapidly and accurately for all mutations (Rahim *et al.*, 2007). Reduced hemoglobin synthesis, ineffective erythropoiesis and short erythrocyte survival in patients with β -thalassemia major lead to severe anemia and tissue hypoxemia which can, however, be partially corrected by regular transfusions aimed at maintaining the mean Hb level around 11-12 g dL⁻¹. However, in β -thalassemia major transfusion treatment increases the iron load thereby determining hemosiderosis in major organs such as the heart, liver and endocrine glands. Iron chelation with desferrioxamine (DFO) has become standard therapy to reduce these complications in patients with thalassemia major. Since patients' survival has greatly improved over the last 10 years, multi-organ impairment due to hemosiderosis often occurs. The lungs may also be involved. Although pulmonary dysfunction is not the most significant clinical manifestation of thalassemias, or indeed does not produce any symptoms, a certain reduction of pulmonary volumes has been reported to occur in most subjects with β -thalassemia (Bacalo *et al.*, 1992; Bosi *et al.*, 2003; Factor *et al.*, 1994; Luyt *et al.*, 1993; Nanas *et al.*, 2008; Priftis *et al.*, 2006; Sritippayawan *et al.*, 2005). The aim of this study was to evaluate and classify pulmonary dysfunction in patients with β -thalassemia major in order to determine the predominant lung dysfunction in these disorders.

MATERIALS AND METHODS

Patient's evaluation: We measured pulmonary function in 59 patients with β -thalassemia major (27 M, 22 F, age range: 18-45 years) enrolled from the Research Center of Thalassemia and Hemoglobinopathies in Ahwaz, Iran during Jan. 2007 to Jan. 2008. To be enrolled into the study patients were required to be at least 18 year old, to be able to perform pulmonary function tests and not to be in overt cardiac failure. The diagnosis of thalassemia was based on hematologic data and family studies as we described in earlier study (Rahim *et al.*, 2007). The thalassemia genotype was defined in all patients. To maintain the Hb level at or above 11 g dL⁻¹, patients with β -thalassemia major were treated with regular blood transfusions and subcutaneous DFO treatment. Fifteen out of the 59 patients had a splenectomy. Nine subjects had a positive family history for respiratory diseases or allergic symptoms; one had a history of allergic bronchial asthma in infancy; 4 were smokers. Serum iron, serum ferritin levels and transferrin saturation were measured every three months using routine tests. The pretransfusion hemoglobin level ranged from 8.0 to 10.1 g dL⁻¹ (Mean \pm SD). The β -thalassemia major patients received 2-3 units of blood every 2-4 weeks (approximately 20 mL kg⁻¹ of packed red blood cells).

All patients with β -thalassemia received daily chelation therapy with subcutaneous injection of DFO (20-40 mg/kg/day). During the entire study period no patients had any symptoms of acute disease of the respiratory tract; at the time of the study all patients were in a stable condition. All patients underwent an examination of the upper respiratory tract. All patients had a chest X-ray on entry to the study. These were performed using a body plethysmograph. Bacalo *et al.* (1992) described a significant reduction in the Forced Expiratory Volume in 1 sec (FEV1) and Forced Vital Capacity (FVC) following blood transfusion and therefore all tests were performed in the morning, 24 h before the patient received the planned blood transfusion. FVC, FEV1, Total Lung Capacity (TLC), Residual Volume (RV), FEV1/FVC and Forced Expiratory Flow at 25 to 75% of FVC (FEF25-75%) were recorded using a pneumotacograph: the best of three technically acceptable values was selected. Values are reported in liters and as percentages of predicted normal values (Polgar and Promadhat, 1971; Rosenthal *et al.*, 1993) corrected for body temperature, atmospheric pressure and saturation with water vapor. Restrictive failure was classified as mild when TLC values were between 70-79% of predicted, as moderate between 60-69% of predicted and severe when < 60% of predicted. The pO₂ and pCO₂ were determined by arterial blood gas analysis. Cardiac

function was evaluated by ECG and echocardiogram: echocardiographic ejection fraction, calculated from M-mode recordings or two-dimensional M-mode studies, was employed to assess cardiac function.

Statistical analysis: Comparisons with normal values were made using Student's unpaired t-test; results were considered statistically significant when $p < 0.05$; linear regression was used to analyze the joint effects of several variables. Summarized data are presented using correlation coefficients and Means \pm SD for group data.

RESULTS

The study included 59 patients (10-45 years) who had been given a diagnosis of β -thalassemia major were randomly selected from the institution (Research Center of Thalassemia and Hemoglobinopathies, Ahwaz Jondishapour University of Medical Sciences, Ahwaz, Iran), whose physical characteristics are reported in Table 1. Of the patients with β -thalassemia major, one had nasal polyposis and one suffered from chronic sinusitis; 7 reported seasonal allergic rhinitis but had no symptoms and were negative when tested for bronchial hyperresponsiveness. No clinical signs of pulmonary dysfunction or evidence of heart failure were found. Chest X-ray was normal in 30 patients with β -thalassemia major, while in the remaining a reticulo-nodular pattern was described. These findings did not correlate with results of the pulmonary function tests (PFTs): in fact, all the patients with the more severe restriction had a normal chest X-ray. Arterial oxygen saturation was normal in all patients (mean value 98%). Table 2 shows the main results of pulmonary function in β -thalassemic patients. Results are expressed as Mean \pm SD. TLC was below the mean predicted value for age and height in 12 of 59 patients (20.3%) with β -thalassemia major. FVC, FEV1 and RV were also reduced indicating restrictive lung dysfunction. Of the β -thalassemic major patients, 2 had mild, 10 moderate and 17 severe restrictive diseases. When corrected for hemoglobin concentration, diffusing capacity of lung for carbon monoxide (D_{LCO}) values approached the lower limits of normal in 16 of subjects with β -thalassemia major (mean value: 24.29 \pm 4.94 mL/min/mmHg, $p = 0.077$). The mean serum ferritin levels were 1594 \pm 1,800 ng m⁻² (range 510-8,629; normal values: 30-320 ng mL⁻¹) in β -thalassemia major. No arrhythmia or signs of congestive heart failure or pulmonary hypertension were found. All the enrolled patients had an ejection fraction >60% (mean value: 67.3 \pm 5.54%). Table 3 shows the main characteristics

Table 1: Anthropometric characteristic and spirometric results

Variables	Female patients (29)	Male patients (30)
Age (years)	20.03 \pm 9.23	19.75 \pm 6.56
Height (m)	157.76 \pm 16.66	152.12 \pm 14.74
Weight (kg)	46.46 \pm 13.79	45.87 \pm 10.78
FVC (predicted) (%)	78.07 \pm 15.54*	75.81 \pm 19.26*
FEV1 (predicted) (%)	79.00 \pm 14.23*	77.42 \pm 16.48*
FEV1/FVC (predicted) (%)	107.00 \pm 7.16	104.15 \pm 10.63
FEF25-75 (%) (predicted) (%)	70.03 \pm 17.56*	67.42 \pm 15.17*
RV (predicted) (%)	73.19 \pm 12.56	69.48 \pm 11.96
TLC (predicted) (%)	77.29 \pm 13.65	74.49 \pm 12.56

FVC: Forced Vital Capacity; FEV1: Forced Expired Volume in one second; FEF25-75%: Forced Expiratory Flow at 25 to 75% of FVC; TLC: Total Lung Capacity and RV: Residual Volume. Values represent Means \pm SEM. Significantly different from normal ($p < 0.001$)

Table 2: Pulmonary function in the patients enrolled in the study

Variables	Observed	Predicted	Percentage (Mean \pm SD)
TLC	3.72 \pm 0.87 [‡]	5.23 \pm 0.75	3.19 \pm 12.56
FVC	3.12 \pm 0.85 [‡]	3.86 \pm 0.65	77.29 \pm 13.65
FEV1	2.72 \pm 0.67 [‡]	3.25 \pm 0.49	78.49 \pm 12.96
RV	0.98 \pm 0.27*	1.34 \pm 0.15	71.69 \pm 24.56
FEF 25-75 (%)	2.54 \pm 0.74 [‡]	3.42 \pm 0.51	70.52 \pm 17.82
FEV1/FVC	0.84 \pm 0.31 ^{NS}	1.25 \pm 0.21	108.20 \pm 9.87

Statistical difference between observed and predicted values: *Significant ($p < 0.05$); [‡]Highly significant ($p < 0.01$); ^{NS}Non-significant. Definition of abbreviation; FVC: Forced Vital Capacity, FEV1: Forced Expired Volume in one second; FEF25-75%: Forced Expiratory Flow at 25 to 75% of FVC; TLC: Total Lung Capacity and RV: Residual Volume

Table 3: Severity of respiratory dysfunction, age and ferritin levels (ng dL⁻¹) of patients expressed as mean values

Respiratory function	No. of detected patient	Age	Ferritin level
Normal	30	27.5 \pm 3.8	1342 \pm 698
Mild	2	26.0 \pm 1.5	993 \pm 312
Moderate	10	20.0 \pm 6.1	3763 \pm 874
Severe	17	23.0 \pm 1.6	1279 \pm 315

of patients in terms of pulmonary function and serum ferritin levels expressed as a mean of values over the previous year. Finally, there were negative correlations between FVC, FEV1, FEV1/FVC (FEV1%), FEF25-75% and ferritin of patients (Fig. 1). As shown in Fig. 1 only FEF25-75% has significant negative correlation with ferritin ($r = -0.26$ $p(r) = 0.04$).

DISCUSSION

Several investigators have studied pulmonary function in β -thalassemia patients (Cooper *et al.*, 1980; Keens *et al.*, 1980; Hoyt *et al.*, 1986; Azarkeivan *et al.*, 2008; Isma'eel *et al.*, 2008; Freedman *et al.*, 1990; Piatti *et al.*, 1999, 2006; Santamaria *et al.*, 1994; Voskaridou *et al.*, 2007; Grant *et al.*, 1986) but their results are conflicting and variably small airway obstruction or a restrictive pattern of lung disease have been described. Table 4 shows the main data reported in literature concerning pulmonary function in

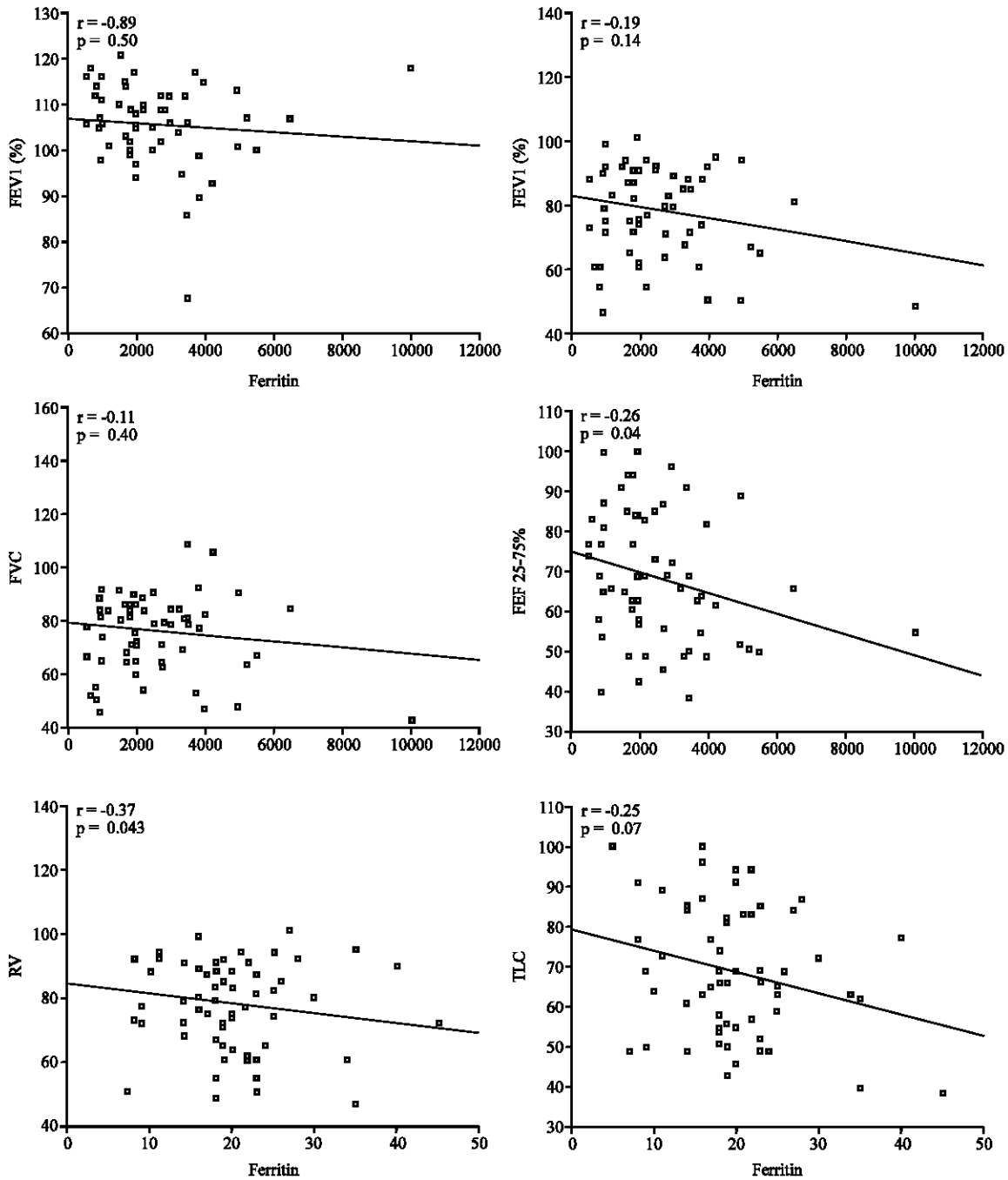


Fig. 1: Relationships between ferritin and percentage predicted Forced Vital Capacity (FVC), Forced Expired Volume in one second (FEV1), Forced Expiratory Flow at 25 to 75% of FVC (FEF25-75%), total lung capacity (TLC) and Residual Volume (RV) in 59 transfusion-dependent, thalassemia major patients. The slope of each relation is indicated together with correlation coefficient and level of significance. Solid symbols represent data from patients

β -thalassemic patients. In this study pulmonary function in 59 β -thalassemia patients has been evaluated, by means of spirometry, lung volumes, diffusion capacity and

arterial blood gas analysis. A restrictive failure pattern was the predominant observation in β -thalassemia major patients. There was no correlation between the degree of

Table 4: Available data reported in literature concerning pulmonary function in β -thalassemia patients

No. of patients	Age range	PO ₂	Pulmonary function	D _{LCO}	Reference
17	6- 18	Hypoxemia in 15/17	Restriction in 8/17	Reduced in 13/15	Cooper <i>et al.</i> (1980)
12	18.4±2.6 [#]	Hypoxemia in 10 /12	Small airway obstruction in 11/12	Normal	Keens <i>et al.</i> (1980)
19	10-29	-	Small airway obstruction	Normal in 16/19	Hoyt <i>et al.</i> (1986)
8	14-24	Hypoxemia in 5/8	Restriction	Reduced but increased	Grant <i>et al.</i> (1986)
35	8-33	Hypoxemia in 85%	Restriction in 24/35	-	Grisaru <i>et al.</i> (1990)
10	7-23	-	Normal	-	Lands <i>et al.</i> (1991)
17	6-17	Hypoxemia in 2/17	Restriction in 7/17	Reduced in 57%	Bacalo <i>et al.</i> (1992)
15	5-18	Hypoxemia in 6/13	Restriction	Reduced	Luyt <i>et al.</i> (1993)
29	6-40	Hypoxemia in 1/29	Restriction	Reduced in all 7/29	Factor <i>et al.</i> (1994)
12	13.4±3.9 [#]	Increased	Restriction	Increased	Santamarina <i>et al.</i> (1994)
19	18-35	-	Restriction in 55.5%	Reduced in all 6/19	Patti <i>et al.</i> (1999)
30	25.5 [*]	-	Restriction in 86.6%	Reduced in all 30	Arora <i>et al.</i> (2001)
18	25-51	-	Restriction in 38.8%	Reduced in 5/18	Patti <i>et al.</i> (2006)
84	18-55	Pulmonary hypertension in 33%	-	-	Voskaridou <i>et al.</i> (2007)
139	21.1 [*]	-	Restriction in 101/139	-	Azarkeivan <i>et al.</i> (2008)
59	18-45	-	Restriction in 29/59	Reduced in 16/59	Present research

[#]Values represented as Mean±SD, ^{*}Values represented as Mean

restriction and serum ferritin levels, chelation treatment or number of transfusions. The reasons for these respiratory alterations may be manifold: foremost, deposition and tissue accumulation of iron may be critical for the development of a restrictive pattern of lung dysfunction in β -thalassemia. Iron deposition in the lung may theoretically be correlated with serum ferritin values or iron deposits in the liver. This study did not, however, find relationships between restrictive lung disease and serum ferritin levels, desferrioxamine dose, or liver iron concentration (data not shown). It is well known that serum ferritin, although being a common parameter used for monitoring chelation therapy, does not accurately reflect the total iron burden as documented in different reports (Camaschella *et al.*, 1996 ; Hamdy *et al.*, 2007). The relationship between altered pulmonary function tests and iron deposition in the lung remains unclear.

Landing *et al.* (1987) and Morris *et al.* (2006) found that some patients had pulmonary hemosiderosis, ferrugination of connective tissue, alveolar septa and blood vessels and interstitial fibrosis: all these derangements could predispose to the development of restrictive lung disease.

Conversely, Grisaru *et al.* (1990) examining autopsy specimens of 6 subjects with β -thalassemia major, found normal alveolar septa and only one case of increased hemosiderin deposits; in one other case, small recent thrombi were found in some small branches of the pulmonary arteries.

Cooper *et al.* (1980) evaluated lung autopsy specimens from 8 patients with thalassemia and found no sign of fibrosis. In an autopsy series, 44% of thalassemia patients had evidence of pulmonary arterial obstruction (Sonakul *et al.*, 1980) in spite of the fact that only a limited number had recurrent chest pain, hypoxemia and right ventricular hypertrophy. Grisaru *et al.* (1990) highlights the occurrence of right ventricular dysfunction and abnormal pulmonary function in thalassemia patients. The

patients in this study were specifically selected for having normal cardiac function as assessed by physical examination, ECG and echocardiography; one can therefore speculate that the lung function derangements found in our patient reflect a primary lung pathologic condition.

Lands *et al.* (1991) studied 10 thalassemic subjects pre- and post-diuresis in order to evaluate the role of possible fluid overload in altering pulmonary function: baseline function was normal and no change occurred following diuresis. Luyt *et al.* (1993) measured free radical production by polymorphonuclear cells to identify a potential relationship with tissue damage in the lungs, but the results failed to indicate any correlation with pulmonary function parameters.

Piatti *et al.* (1999) performed PFT, chest X-ray and D_{LCO} in 19 β -thalassemia major patients. Their results showed 6 abnormal chests X-ray, 29 restricted pattern in PFT, 6 restrictive patterns according to D_{LCO} and normal arterial oxygen in all patients. Arora *et al.* (2001) examined PFT in 30 β -thalassemic patients and found restrictive abnormality in PFT among 86.6% of subjects and restrictive lung disease in all 30 patients taking into account decreased D_{LCO}.

Recently, Piatti *et al.* (2006) conducted a study on PFT in asymptomatic, non-smoking β -thalassemia major patients. Their reported restrictive pattern in 55.5% and 5 subjects with decreased D_{LCO}. Voskaridou *et al.* (2007) observed pulmonary hypertension in 84 Hemoglobin S/ β -thalassemic patients and found 33% of disorder in study subjects. Azarkeivan *et al.* (2008) studied 139 transfusion-dependent β -thalassemic patients and found abnormal lung pattern in 39 with respect to chest X-ray and 101 restricted patterns according to changes in PFT.

Hepatosplenomegaly may contribute to a lung restrictive pattern by reducing chest wall compliance while increases in vital capacity and expiratory reserve volume are observed in patients following splenectomy.

Only 6 patients in our population had mild hepatosplenomegaly, 15 had been splenectomized. In conclusion, as hypothesised by Santamaria, 29 lung dysfunctions in β -thalassemia may be multifaceted: in fact, the existence of an a priori situation is possible which, irrespective of the transfusional regimens, could worsen the consequences of iron deposition.

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