A Study on Pulmonary Pressure and Serum Ferritin in β-Thalassemic Children

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Abstract

Background: Among the sequelae associated with a diagnosis of β-thalassemia, pulmonary arterial hypertension remains a concern in patients with β-thalassemia major.

Objectives: To evaluate the pulmonary pressure and systolic left ventricular function and detect their relationship with the iron status in β-thalassemic patients.

Patients and Methods: A cross-section study on 50 β-thalassemia major children > 5 years old diagnosed by CBC and hemoglobin electrophoresis and on regular blood transfusion/1-2 months was done. The patients were studied by echocardiography regarding their pulmonary pressure level measurement and left ventricular systolic function estimation (FS%). Serum ferritin level was also measured.

Results: β-thalassemic children developed systolic and, to a lesser extent, diastolic pulmonary hypertension (36% and 4% respectively). There was a significant positive correlation between serum ferritin level and systolic and diastolic pulmonary pressure. A left ventricular systolic dysfunction was detected in 18% of patients**.** However, there was no correlation between serum ferritin level and left ventricular systolic dysfunction.

Conclusion: Pulmonary hypertension and not the left ventricular systolic dysfunction was related to the serum ferritin level in β-thalassemic children.

Keywords: β thalassemia, Pulmonary pressure, Serum ferritin.

Abbreviation: RBCs: Red blood cells; TRmax: Tricuspid Regurgitant jet Velocity Maximum; PR: pulmonary regurgitation; RAP: Right Atrial Pressure; ELISA: Enzyme-Linked Immunosorbent Assay; PAH: Pulmonary Arterial Hypertension.

Introduction:

Cardiac disease is one of the major causes of death in thalassemia, as myocardial dysfunction and heart failure were considered common causes of morbidity and mortality in children with β-thalassemia major, which were related mainly to chronic hemolysis and iron overload. However, PAH has emerged as a major risk factor for impaired right ventricular function in such patients. [(i)]

Prevalence rates of PAH in β-thalassemic patients exceeded 50% in some studies. Although the exact mechanisms implicated in the pathogenesis of PAH in β-thalassemia remain unclear, its association with several risk factors. [(ii) (iii)]

Iron overload is implicated in the pathogenesis of several vascular disorders, including microangiopathic hemolytic

anemia, vasculitis, atherosclerosis, arterial thrombosis, and reperfusion injury. It is evaluated by measuring serum ferritin level as an indirect cardiac iron. [(iv)]

Serum ferritin has been used to predict iron overload status in clinical practice due to its strong correlation with hepatic iron, representing an indirect index for estimating the total body iron stores. Serum ferritin has been shown to have a positive relationship with the amount of blood transfusion in β - thalassemia patients. Furthermore, it has been shown that a serum ferritin level greater than 1800 μg/L was associated with an increased cardiac iron concentration. That serum ferritin greater than 2500 μg/L was associated with the increased prevalence of cardiac events. [(4)]

Patients and Methods

Study Site:

Assiut University Children's Hospital (Hematology Unit, Hematology Clinic and Cardiology Clinic).

Study Population:

Cross section study on 50 children (older than 5 years) that were diagnosed as β thalassemia major confirmed by complete blood count and electrophoresis, and they were on regular blood transfusion every 1-2 months of an average amount of 150-250 cc of packed RBCs at each transfusion.

Ethical Considerations:

Approval of the study protocol was obtained by the Ethical Scientific Committee of the Faculty of Medicine - Assiut University and informed consent was obtained from the parents before enrollment.

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Inclusion Criteria:

All β thalassemia major children older than 5 years.

Exclusion Criteria:

Patients with other types of thalassemia.

Patients younger than 5 years

Children with congenital or rheumatic heart disease.

Acute infections, inflammatory diseases, collagen diseases, hepatic diseases, and malignancy cause an increase in serum ferritin levels.

History and Clinical Examination

A detailed history and thorough clinical examination were done on all children. Detailed data were recorded on demographics, the age of onset of the disease, the frequency of blood transfusion, the age of onset, the type of iron chelation therapy, and any symptoms of pulmonary congestion, low cardiac output, or heart failure. Systemic, abdominal, and cardiac examination findings were recorded in detail.

Serum levels of ferritin and

echocardiographic assessment were done for all included children.

Echocardiography:

All patients were referred for echocardiography using M-mode, 2D, Doppler, and color Doppler echocardiography to estimate the pulmonary pressure and determine the diameter of cardiac chambers.

Estimation of Pulmonary Artery Systolic Pressure:

Continuous wave Doppler of the tricuspid regurgitation trace was used to measure the pressure difference between the right ventricle and right atrium. The simplified Bernoulli equation (P = 4 [TRmax]2) was used to calculate this pressure difference using peak tricuspid regurgitation velocity or by obtaining peak systolic gradient of tricuspid regurgitation and adding the value of right atrial pressure. A value of ≤ 2.8 m/s suggests low probability of systolic pulmonary hypertension (≤ 30 mmHg), a value of 2.9–3.4 m/s indicates intermediate probability (mild to moderate) (30-45 mmHg), and a value > 3.4 m/s (> 45 mmHg) suggests a high probability for systolic pulmonary hypertension (severe). Pulmonary arterial hypertension is a mean pulmonary artery pressure > 25 mmHg in all age groups. [(5)]

Estimation of Pulmonary Artery Diastolic Pressure:

Pulmonary artery diastolic pressure was measured by pulmonary regurge-end velocity. End pulmonary regurge velocity was measured at the time of pulmonary valve opening; pulmonary artery diastolic

pressure is calculated from the following equation: 4 (PR-end velocity)2 + RAP. [(v)]

Fractional Shortening (FS %) Measurement:

Fractional shortening was measured as an indicator of left ventricular systolic function using the M mode modality. Left ventricular end-systolic (ESD) and end-diastolic diameters (EDD) were measured, and then FS% was calculated using the formula FS = EDD − ESD/EDD × 100. Normal FS% is > 28%. [(vi)]

Laboratory Investigations:

Three mL of venous blood samples were drawn from each subject under an aseptic condition. ELISA measured serum ferritin. Normal Serum ferritin level is from 12 to 300 ng/mL). [(vii)]

Statistical Analysis:

The data were tested for normality using the Anderson-Darling test and for homogeneity variances before further statistical analysis. Categorical variables were described by **number and percent** (N, %), whereas continuous variables were described by mean and standard deviation **(Mean±, SD)**. The chi-square and Fisher exact tests were used to compare categorical and continuous variables using the t-test and the Independent-Samples T-tests. A two-tailed $p < 0.05$ was considered statistically significant. We used Pearson and Spearman correlation to show the association between variables. All analyses were performed with the **IBM SPSS 20.0** software.

Results

Table 1: The Demographic data of the studied patients

Demographic data	(n=50)	%
Age		
5-10 y	17	34.0
>10 y	33	66.0
Sex		
Male	31	62.0
Female	19	38.0
Residence		
Rural	43	86.0
Urban	7	14.0

Table 2: Systolic and diastolic pulmonary pressure and systolic left ventricular function (Fractional shortening; FS %) estimated by echocardiography in thalassemic patients studied

Echocardiography	(n=50)	%
Systolic Pulmonary pressure		
Normal (<30 mmHg)	32	64.0
Abnormal (>30 mmHg)	18	36.0
Diastolic Pulmonary pressure		
Normal (<15 mmHg)	48	96.0
Abnormal (>15 mmHg)	2	4.0
Fractional shortening (FS %)		
Normal (>28%)	41	82.0
Abnormal (<28%)	9	18.0

Table 3: The demographic data in relation to systolic pulmonary hypertension

(n = 50)	Systolic Pulmonary Hypertension	
	With	Without
5-10 y (n = 17)	1 (5.8%)	16 (94.2%)
>10 y (n = 33)	17 (51.5%)	16 (48.5%)
Male (n = 31)	9 (29%)	22 (71%)
Female (n = 19)	9 (47.4%)	10 (52.6%)
Rural (n = 43)	14 (32.6%)	29 (67.4%)
Urban (n = 7)	4 (57%)	3 (43%)

Table 4: A comparison between patients with normal and high serum ferritin levels in relation to pulmonary pressure (systolic and diastolic) and left ventricular systolic function (FS%)

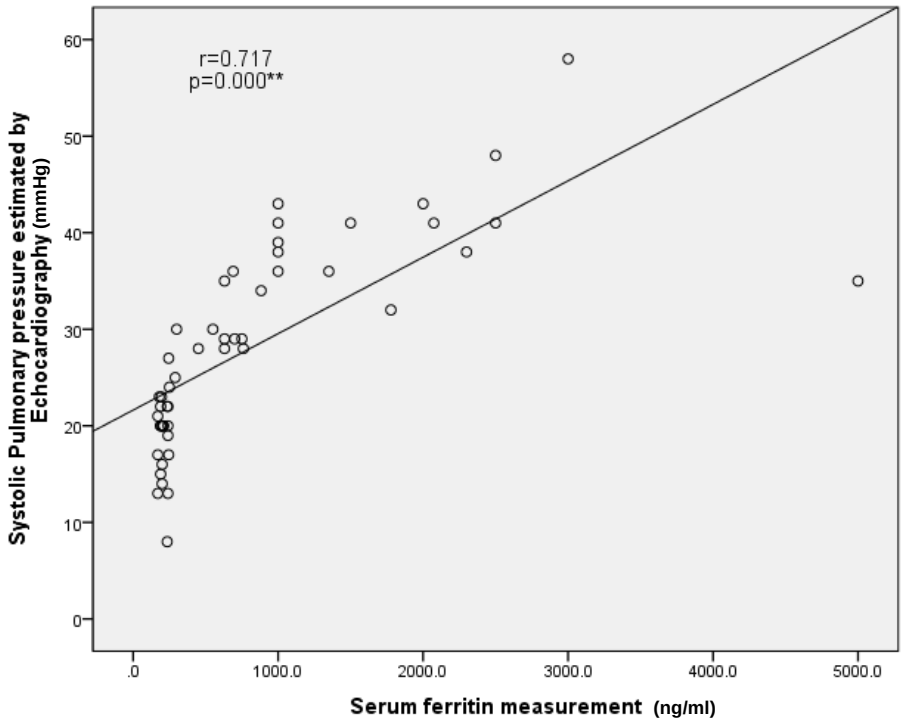
Serum Ferritin Level	Pulmonary Pressure Range (Mean±SD) mmHg		FS% Range (Mean±SD) %
	Systolic	Diastolic	
Normal (< 300ng/ml) (n= 23)	8-27 18.96±4.31	0-17 2.35±4.59	24-52.3 37.26±6.77
High (> 300 ng/ml) (n= 27)	25-58 35.96±7.33	0-48 9.04±9.40	21-57 38.32±8.21
P. value	<0.001	<0.003	0.625

Table 5: Iron status and chelating agent used in relation to systolic pulmonary pressure in the thalassemic patients studied

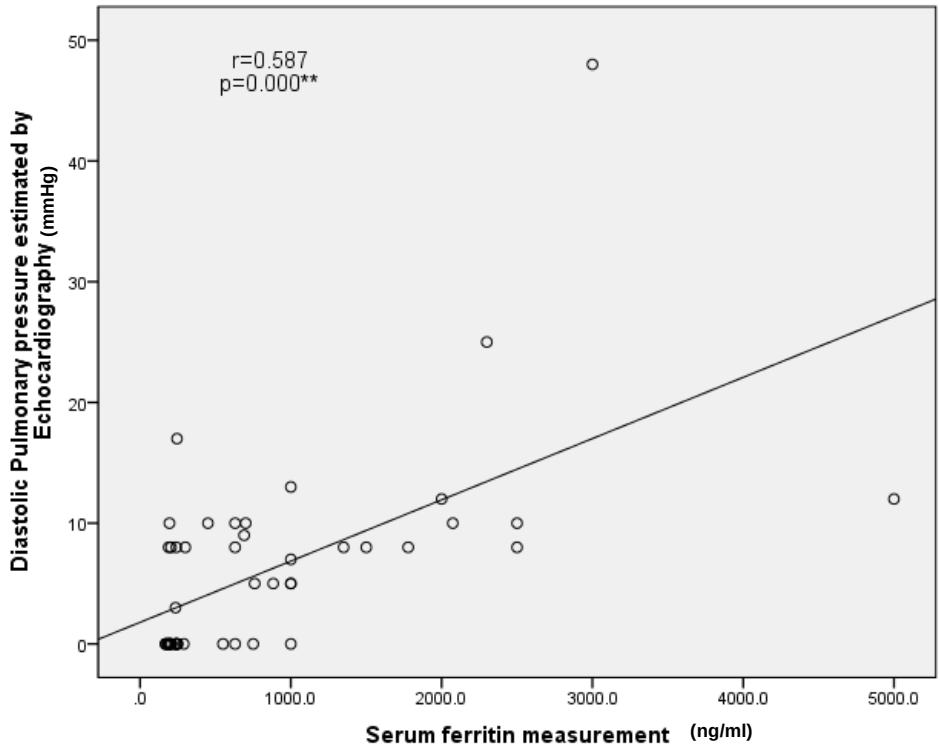
	(n=50)	%	Systolic Pulmonary Hypertension	
			With	Without
Serum Ferritin Level				
Low (< 300 Ng/Ml)	23	46.0	0 (0.0%)	23 (100%)
High (> 300 Ng/Ml)	27	54.0	18 (66.6%)	9 (33.4%)
Using Iron Chelating Agents				
Regular oral	23	46.0	3 (13%)	20 (87%)
Irregular oral	27	54.0	15 (55.5%)	12 (45.5%)

Table 6: The correlation between systolic and diastolic pulmonary pressure, FS %, and serum ferritin level

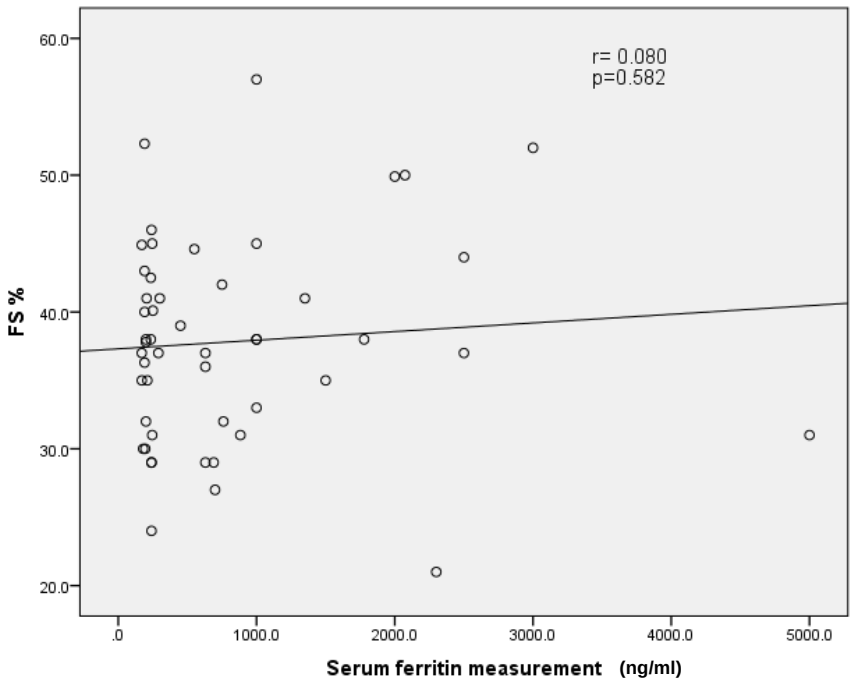
		Systolic Pulmonary pressure	Diastolic Pulmonary pressure	FS %
Serum ferritin	r	0.717	0.587	0.080
	p	0.000**	0.000**	0.582



(Fig. 1) Systolic pulmonary pressure in correlation with serum ferritin level



(Fig. 2) Diastolic pulmonary pressure in correlation with serum ferritin level



(Fig. 3) Left ventricular systolic function (FS%) in correlation with serum ferritin level

Table 6 and Fig. 1,2, and 3 show the correlation between systolic and diastolic pulmonary pressure, left ventricular systolic function (FS %), and serum ferritin level. Significant positive correlations exist between systolic, diastolic, and serum ferritin levels (r= 0.717, p= 0.000), (r= 0.587, p= 0.000), respectively. There is no correlation between left ventricular systolic function (FS %) and serum ferritin level (r=0.080, p= 0.582).

Discussion

PAH is one of the most common causes of morbidity and mortality in patients with hemolytic disorders and is a frequent finding in echocardiographic screening of patients with β thalassemia. It is a pathological hemodynamic condition defined as an increase in mean pulmonary arterial pressure ≥ 25 mmHg at rest and assessed by echocardiography or right heart catheterization. [(viii)]

In the present study, the mean of systolic and diastolic pulmonary pressure was 28.14±10.5 mmHg (8-58 mmHg) and 5.96±8.22 mmHg (0-48 mmHg). Normal systolic pulmonary pressure was detected in 64% of patients, while systolic pulmonary hypertension was detected in 36%. Normal diastolic pressure was detected in 96% of patients, while diastolic pulmonary hypertension was detected in (2/50) 4% (Table 2).

Although all our patients were on regular blood transfusions, they developed systolic pulmonary hypertension in 36% of them. Controversy, **Karimi et al.** [(ix)]recorded that PAH frequently occurs in non-transfused thalassemia patients and may be an indication for transfusion therapy. He mentioned that the risk factors for developing PAH in thalassemic patients include non or irregular blood transfused (3.5-times); hydroxyurea treatment (2.6-times), nucleated red blood cell count ≥300×10(6)/l (2.59-times) or a previous history of thromboembolic events (3.7-times).

Thirteen percent of our patients on regular oral iron chelating agents and 55.5% on irregular use developed systolic PAH (table 5). **Karimi et al.** [(9)] recorded that PAH develops 2.3 times in non or irregular iron chelation patients.

In the present study, the age of our thalassemic patients ranged from 5-10 years in 34% and was >10 years in 66% of them. Males were 62% and females were 38%. Eighty-six percent of patients were from rural areas, while 14 % were from urban areas (Table 1). At the same time, 5.8% of 5-10 years old patients and 51.5% of >10 years old patients had systolic pulmonary hypertension. Twenty-nine percent of males, 47.4% of females, 32.6% of rural patients, and 57% of urban patients developed systolic pulmonary hypertension as well (table 5). Against **Tantiworawit et al., [x]** who reported that neither age nor gender were associated with PAH.

Guidotti et al. [xi] recorded that no thalassemic patients who were presented with such complaints developed evidence of pulmonary hypertension. He attributed that to pulmonary dysfunction in his study, which is responsible for these symptoms rather than pulmonary hypertension.

Left ventricular systolic function (FS%) ranged from 21-57% (mean±SD) 37.83±7.52 %. Normal left ventricular systolic function was in 82% of patients, while left ventricular systolic dysfunction was 18% (table 4). This is in agreement with **Pal et al., [xii]** who detected a spectrum of myocardial involvement, including left ventricular systolic dysfunction, dilatation, failure, PAH, and right ventricular dilatation in his study and explained that the presence of excess iron, which is unbound, and able to redox cycle between Fe+2 and Fe+3. It can generate reactive oxygen species (ROS), resulting in myocardial cell death.

The present study shows that serum ferritin ranged from 170 - 5000ng/ml (mean ± SD; 821.90±951.22). Normal serum ferritin levels were detected in 46% of patients, and abnormally high levels were detected in 54%. Forty-six percent of patients were on regular oral iron chelating agents, while 54% were on irregular usage. About 66.6% of patients with high serum ferritin levels developed systolic pulmonary hypertension. On the other hand, 55.5% of patients with irregular oral intake of iron-chelating agents developed systolic pulmonary hypertension. Eighty-seven percent of patients on regular oral chelating agents recorded normal systolic pulmonary pressure (table 9). There were significant positive correlations between systolic pulmonary pressure, diastolic pulmonary pressure, and serum ferritin level (r= 0.717, p= 0.000), (r= 0.587, p= 0.000) respectively (Table 6 and fig.2 and 3).

The presence of non-transferrin-bound iron in the sera of β-thalassemia patients can cause oxidative vessel injury and endothelial activation. Moreover, iron overload could lead to hepatic disease and subsequent alterations in coagulation factor levels, thus worsening the hypercoagulable state implicated in these patients as a risk factor for developing PAH.[xiii] [xiv]

Silvilairat et al. [xv] reported increased serum ferritin value and iron deposition in the myocardium; thus, the diastolic dysfunction grade increased.

Against, the study that demonstrated that serum ferritin is not suitable for its use as a predictive indicator of myocardial iron deposition due to its lack of relationship with cardiac iron. [xvi]

Conclusion

Beta thalassemia children develop systolic and, to a lesser extent, diastolic pulmonary hypertension, even though they are receiving regular blood transfusions and regular oral iron chelating therapy. The left ventricular systolic dysfunction could also be detected in about 18% of patients.

A significant positive correlation was found between serum ferritin level and systolic and diastolic pulmonary pressure. However, there was no correlation between serum ferritin level and left ventricular systolic dysfunction.

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