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Risk factors for heart disease in transfusion-dependent thalassemia: serum ferritin revisited

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Abstract

Heart disease remains a leading cause of morbidity and mortality in transfusion-dependent thalassemia (TDT), which can be attributed to several factors but primarily develops in the setting of iron overload. This was a retrospective cohort study utilizing Webthal® patient data from five major centers across Italy. Patients without heart disease were followed-up for 10 years (2000–2010) and data were collected for demographics, splenectomy status, serum ferritin and hemoglobin levels, and comorbidities associated with heart disease. Among 379 patients analyzed (mean age 22.9 ± 5.1 years, 47.8% men), 44 (cumulative incidence: 11.6%) developed heart disease during the period of observation. Splenectomy ($p = 0.002$) and serum ferritin level ($p < 0.001$) were the only risk factors with significant association with heart disease. A serum ferritin threshold of ≥ 3000 ng/mL was the best predictor for the development of heart disease (86.4% sensitivity and 92.8% specificity, AUC: 0.912, 95% CI 0.852–0.971, $p < 0.001$). On multivariate analysis, only a serum ferritin level ≥ 3000 ng/mL remained significantly and independently associated with increased risk of heart disease (HR: 44.85, 95% CI 18.85–106.74), with a 5- and 10-year heart disease-free survival of 58 and 39%. The association between iron overload and heart disease in patients with TDT is confirmed, yet a new serum ferritin level of 3000 ng/mL to flag increased risk is suggested.

Keywords Thalassemia · Heart disease · Iron overload · Serum ferritin

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Introduction

Heart disease remains a major cause of morbidity and mortality in transfusion-dependent thalassemia (TDT) patients [1]. It has been attributed to several underlying disease factors, including chronic anemia and associated organ morbidity (endocrine and liver disease), although iron overload from repeated transfusions remains the primary source of cardiac tissue damage [2]. The introduction of magnetic resonance imaging (MRI cardiac T2*) for the assessment of myocardial iron content provided a novel advancement to tailor iron chelation in patients showing significant siderosis, considering the established association with the development of heart disease [3–5]. The use of cardiac T2* has been associated with improved outcomes in TDT patients [6]. However, the availability and feasibility of routine assessment may not be widely applicable especially in resource-poor countries where the disease is highly prevalent. Thus, understanding the role of risk factors for heart disease that can be readily identifiable

from a well-conducted clinical examination or routine laboratory testing remains of high merit to allow tailoring of necessary intervention. With this background, we analyzed data from a large group of TDT patients in Italy followed for 10 years, to evaluate the role of clinical and laboratory risk factors in the development of heart disease.

Patients and methods

This was a retrospective cohort study utilizing Webthal® TDT patient data from five major thalassemia treatment centers across Italy. The Webthal® project and database were previously described [2]. Briefly, it is a computerized clinical record for thalassemia patients attending the participating centers, with specialized and comprehensive domains for data collection on key aspects of the disease. A Secure Socket Layer system and passwords are used to ensure data safety and confidentiality. At each center, an Ethics Committee approval is obtained, and written informed consents for data collection and use are retrieved from patients (according to the Italian law, ART. 79 D.Lgs. 196/02).

For this study, we collected data on all patients treated at participating centers spanning the period 01/01/2000–01/01/2010. We excluded patients who already had heart disease at the start of observation. Retrieved data on the analyzed patients included demographics, splenectomy status, system-generated mean serum ferritin and hemoglobin levels across the period of observation (recorded at an average of 6–12 times per year), presence or absence of the following comorbidities: hypothyroidism, diabetes mellitus, liver cirrhosis (screened for at least once per year and confirmed histologically through liver biopsy or by biopsy-validated thresholds on transient elastography), and the development of heart disease. The list of a priori-defined potential risk factors was kept to a minimum, and restricted to measures readily and commonly available, to allow for meaningful interpretation and applicability in routine clinical practice. In the Webthal® database, heart disease is documented as per any of the following criteria:

- Patients receiving cardioactive drugs as defined in previous work [7].
- Patients being hospitalized for congestive heart failure.
- Patients with a left ventricular ejection fraction < 50%, as measured by echocardiography four-chamber view, annually.
- Patients with dysrhythmia except isolated extrasystolic premature ventricular (PVC) and supraventricular contractions.
- Patients with pulmonary hypertension.

Statistical analysis

Data were described with mean ± standard deviation and percentages. Univariate analyses were conducted using the independent samples t test for continuous variables and the Chi-squared test for categorical variables. A receiver operating characteristic (ROC) curve analysis was used to identify the best serum ferritin threshold to predict heart disease, using the highest sum of sensitivity and specificity within the area under the curve (AUC). Heart disease-free survival analysis was performed by constructing Kaplan–Meier curves, and p values from the log-rank test were reported. Cox multivariate regression analysis was performed to identify independent roles for risk factors on the development of heart disease, by considering risk factors with a $p < 0.1$ on univariate analysis (conservative approach). All p values are two sided with the level of significance set at < 0.05.

Results

A total of 379 patients were included in this analysis and observed for 10 years. The mean age at the start of observation was 22.9 ± 5.1 years (range 12.3–39.9), including 181 (47.8%) men. Patients' characteristics are summarized in Table 1. Forty-four patients developed heart disease during the period of observation, giving a cumulative 10-year incidence rate of 11.6% (95% CI 8.8–15.2). Among the 44 patients with heart disease, 11 (25%) had left ventricular ejection fraction < 50% as measured by echocardiography four-chamber view, 38 (86.3%) were receiving cardioactive drugs, 11 (25%) were hospitalized for congestive heart failure, and 22 (50%) had dysrhythmia (except isolated extrasystolic PVC and supraventricular contractions). None of the patients had documented pulmonary hypertension. None of the patients died during the period of observation.

Table 1 Patients' characteristics ($n = 379$)

Parameter	Value
Age at baseline in years, mean ± SD (range)	22.9 ± 5.1 (12.3–39.9)
Male, n (%)	181 (47.8)
Splenectomized, n (%)	170 (44.9)
Serum ferritin in ng/mL, mean ± SD (range)	2343.3 ± 1987.4 (11–14,142)
Hemoglobin[a] in g/dL, mean ± SD (range)	9.4 ± 0.4 (7.7–11.2)
Hypothyroidism, n (%)	39 (10.3)
Diabetes mellitus, n (%)	22 (5.8)
Liver cirrhosis, n (%)	3 (0.8)

[a]Pre-transfusion
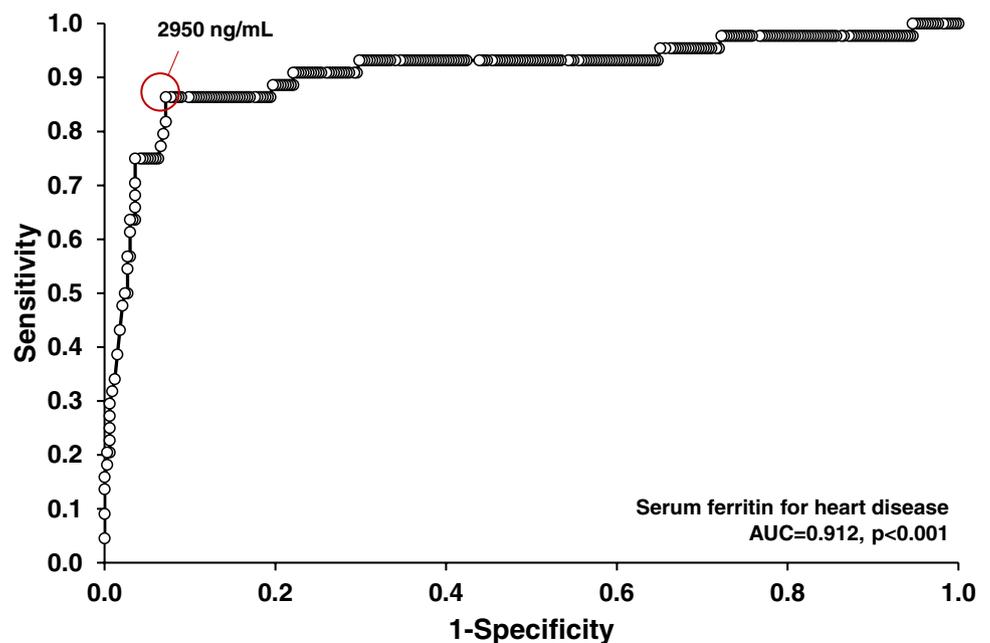
Univariate analysis

Among the assessed risk factors, mean age and gender distribution were comparable between patients who developed and those who did not develop heart disease. Similarly, there was no statistically significant difference in the proportion of patients who had hypothyroidism, diabetes mellitus, or liver cirrhosis between the two groups. The mean pre-transfusion hemoglobin level was also comparable in both patient groups, noting that 90% of the whole evaluated population had pre-transfusion hemoglobin levels above the recommended value of ≥ 9.0 g/dL [8]. A higher proportion of splenectomized patients was observed in patients who developed heart disease compared with those who did not (65.9 vs 42.1%, $p = 0.002$). Similarly, the mean serum ferritin level was higher in patients who developed heart disease compared with those who did not (4728.6 ± 1928.4 vs 1575.5 ± 1002.8, $p < 0.001$) (Table 2).

Identifying a serum ferritin threshold

To select a serum ferritin threshold to carry forward for further analysis, we applied a ROC curve analysis to establish the best threshold that can predict the development of heart disease. On ROC curve analysis, the serum ferritin value of 2950 ng/mL is the best predictor for the development of heart disease (86.4% sensitivity and 92.8% specificity, AUC: 0.912, 95% CI 0.852–0.971, $p < 0.001$) (Fig. 1). Hence, the level of ≥ 3000 ng/mL was used in further analysis.

Survival analysis and multivariate regression

The cumulative heart disease-free survivals for all patients are 90 and 88% at 5 and 10 years, respectively. In a Cox regression analysis including risk factors with a $p < 0.1$ on univariate analysis, splenectomized patients have a 2.85 (95% CI 1.33–4.64) increased risk of developing heart disease compared to non-splenectomized

Table 2 Comparisons of demographics, laboratory, and clinical parameters in patients who developed or did not develop heart disease

Parameter	Heart disease ($n = 44$)	No heart disease ($n = 335$)	p value
Age at baseline in years, mean $\pm$ SD	24.2 ± 5.0	22.7 ± 5.1	0.051
Male, n (%)	22 (50.0)	159 (47.5)	0.437
Splenectomized, n (%)	29 (65.9)	141 (42.1)	0.002
Serum ferritin in ng/mL, mean $\pm$ SD	4728.6 ± 1928.4	1575.5 ± 1002.8	< 0.001
Hemoglobin[a] in g/dL, mean $\pm$ SD	9.4 ± 0.4	9.4 ± 0.4	0.615
Hypothyroidism, n (%)	5 (11.4)	34 (10.1)	0.484
Diabetes mellitus, n (%)	4 (9.1)	18 (5.4)	0.244
Liver cirrhosis, n (%)	1 (2.3)	2 (0.6)	0.310

[a]Pre-transfusion

Fig. 1 Receiver operating characteristic (ROC) curve of the sensitivity and specificity of serum ferritin to predict heart disease. The serum ferritin value of 2950 ng/mL is the best predictor for the development of heart disease (86.4% sensitivity and 92.8% specificity)

patients. Patients with a serum ferritin level of ≥ 3000 ng/mL ($n = 62$) have a considerably increased risk of 46.94 (95% CI 19.78–111.41) compared with patients having serum ferritin levels < 3000 ng/mL ($n = 317$). On multivariate analysis, only serum ferritin level ≥ 3000 ng/mL remains significantly and independently associated with increased risk of heart disease (HR 44.85, 95% CI 18.85–106.74) (Table 3). The 5-year heart disease-free survival is 98% for patients with serum ferritin level < 3000 ng/mL and 58% for patients with serum ferritin ≥ 3000 ng/mL. The 10-year heart disease-free survivals are 98 and 39%, respectively. Figure 2 illustrates survival curves for both serum ferritin groups, with a log-rank test Chi-square value of 223.799, $p < 0.001$.

Discussion

Our study confirms the role of iron overload in the development of heart disease in TDT patients. Although cardiac iron loading has been shown to lag behind other organs (e.g., liver and pancreas), patients eventually reach considerable myocardial iron concentrations that have been associated with cardiac dysrhythmias, failure, and death [3, 4, 9]. Initial exposure to harmful iron species such as non-transferrin-bound iron has also been suggested as a means for earlier tissue damage before significant tissue iron loading develops [10].

Serum ferritin remains a widely used marker for the assessment of iron overload. Although spot measurements may be misleading if patients have ongoing infections, inflammation, or liver disease, interpretation of a

Table 3 Cox regression analysis for the development of heart disease within 10 years

Parameter	Univariate HR (95% CI)	p value	Multivariate HR (95% CI)	p value
Age for every 1 year increase	1.053 (0.997–1.113)	0.065	1.008 (0.956–1.063)	0.778
Splenectomized	2.485 (1.332–4.636)	0.004	1.888 (0.939–3.794)	0.074
Serum ferritin ≥ 3000 ng/mL	46.939 (19.776–111.411)	< 0.001	44.854 (18.847–106.744)	< 0.001

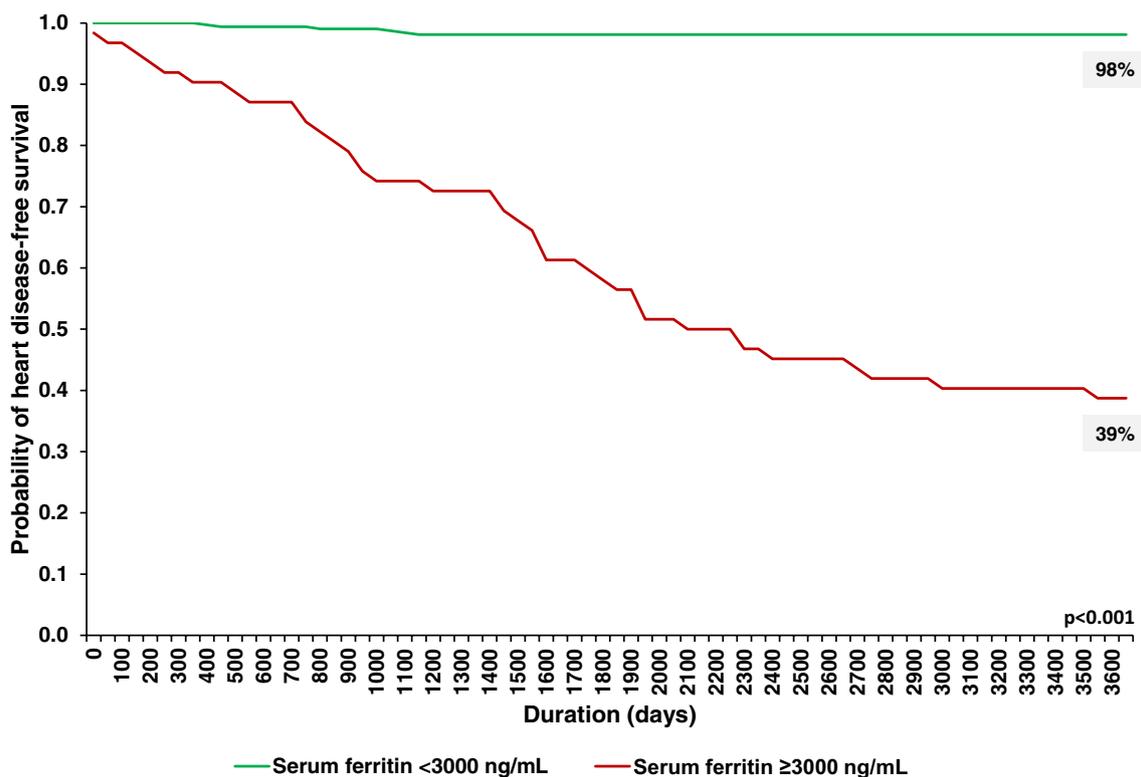*HR* hazard risk, *CI* confidence interval

Fig. 2 Kaplan–Meier heart disease-free survival for serum ferritin ≥ 3000 and < 3000 ng/mL

series of measurements can give a good reflection of systemic iron overload status [8, 11, 12]. A serum ferritin level of > 2500 ng/mL has been classically used to flag patients at risk of heart disease and call for optimization or change in management [8, 13]. This threshold is mainly based on a study by Olivieri and colleagues, who evaluated risk factors for heart disease in a group of 97 patients with TDT followed for a median of 12 years, latest until June 1991 [14]. Similar to the observation in this study, serum ferritin was independently and significantly associated with the risk of heart disease although at a lower threshold than the one we identified (2500 vs 3000 ng/mL). It should be noted, however, that the study by Olivieri and colleagues pre-selected and tested the threshold while we identified it from a ROC curve analysis [14].

Our study covered a time period where important changes in practice occurred, mainly the introduction of cardiac T2* MRI and oral iron chelators with significant efficacy in removing heart iron [6]. Thus, the probability of heart disease associated with high serum ferritin levels appeared lower in the later years of our observation period compared with the initial years when subcutaneous deferoxamine had been the predominant chelator. Differential effects on different iron parameters have been reported for the different chelators [15], and patients may have been experiencing a decrease in heart iron despite lower rate of serum ferritin decline, thus, leading to a picture of persisting high serum ferritin levels yet at a lower incidence of heart disease. As such, it should be noted that as much as high serum ferritin can be a good predictor of heart disease risk in this patient population, the effect of lowering serum ferritin level versus other more direct estimates of heart iron such as cardiac T2* MRI on heart disease incidence should be carefully interpreted.

The introduction of oral chelation therapy has transformed the ability to control cardiac iron overload and disease in TDT patients. High-dose deferiprone was superior to low-dose deferoxamine for myocardial iron removal in patients with mild–moderate baseline cardiac T2* [16], while the addition of deferiprone to deferoxamine enhanced myocardial iron removal [17]. Combination therapy with deferiprone and deferoxamine has been associated with the ability to reduce severe myocardial iron siderosis and improve cardiac function in patients' left ventricular heart failure [18] although more recent work indicates that the effects do not surpass those of deferoxamine monotherapy [19]. Deferasirox was shown to continually improve cardiac T2* and myocardial iron concentration over 3 years in patients with TDT irrespective of myocardial iron overload severity at baseline [20]. Compared to deferoxamine, deferasirox therapy is associated with a higher proportion of patients achieving normalization of their cardiac T2* after 1 year [21]. In clinical trials on patients without heart disease at baseline, no patients with myocardial siderosis developed heart failure while on deferasirox [20, 21], while recent data from observational studies show potential for improvement in heart function [22].

Our study has limitations. The large sample of patients and long period of observation may have precluded us from collection of other risk factors relevant to heart disease in TDT patients. However, these contributing factors would have been relatively rare opposed to the effects of iron overload and related complications, and if existing, the chances that they would have a confounding effect on the role of serum ferritin would be low in the absence of a mediating effect on iron overload. Similarly, the role of protective factors such as pharmacologic therapy in patients with preclinical signs or symptoms of heart disease could not be accounted for although these would have been more likely utilized in patients with high serum ferritin value, and could only underestimate the risk of heart disease apparent in the high serum ferritin group. Lastly, a similar analysis evaluating the association between heart disease and baseline or serial trends of serum ferritin overtime (compared to observation average) is warranted in future studies.

In conclusion, our study has confirmed the association between iron overload and heart disease in patients with TDT and identified a new serum ferritin level of 3000 ng/mL to signal risk from heart disease and need for management optimization accordingly. The effects of long-term iron control and heart disease prevention by specific iron chelators lowering serum ferritin levels below this threshold merit evaluation in future trials, especially taking into consideration parallel changes in cardiac T2* MRI. The need for further serum ferritin reduction extends beyond protection from heart disease, and lower levels may still be needed to ensure prevention of other morbidities TDT patients may suffer from, especially with faster rates of iron loading and damage noted in other organs compared to the heart.

Author contributions Study concept design: GD and GLF. Data collection and assembly: all authors. Data analysis and interpretation: GD and GLF. Manuscript drafting: GD and GLF. Manuscript critical revision for intellectual content: all authors. All authors approved the manuscript prior to submission.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Statement of human and animal rights At each center, an Ethics Committee approval is obtained.

Informed consent Written informed consents for data collection and use are retrieved from patients (according to the Italian law, ART. 79 D.Lgs. 196/02).

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