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Analysis of how Serum Ferritin and the Aspartate Aminotransferase-to-Platelet Ratio Index (APRI) are correlated to Hepatic MRI T2* Findings in Children with Beta-Thalassemia Major

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Abstract

Background: Iron overload is a major complication in patients with beta-thalassemia major. Excessive iron accumulation leads to organ dysfunction. The regular assessment of iron is crucial to effectively manage iron overload in these patients. This study evaluated the correlation between serum ferritin levels, the Aspartate Aminotransferase-to-Platelet Ratio Index (APRI), and hepatic MRI T2* findings in children with beta-thalassemia major.

Materials and Methods: This retrospective analytical study was conducted at Shahid Sadoughi Hospital in Yazd in 2023. The research population comprised all the children under the age of 15 with beta-thalassemia major who had undergone multiple blood transfusions (at least ten units of blood).

Results: The participants in this study were 70 children with beta-thalassemia major, including 35 males and 35 females. Their mean age was 3.52 ± 10.76 years. The mean relaxation time of liver MRI T2* was 4.42 ± 4.91 ms. The participants also had the APRI score of 0.55 ± 1.90, the aspartate aminotransferase (AST) level of 23.82 ± 36.29, and the serum ferritin level of 285.01 ± 3244.04 (ng/mL). Based on MRI T2* results, 30% of the patients had a severe liver iron overload, 27.1% had a moderate overload, 21.4% had a severe overload, 18.6% had a mild overload, and 2.9% had near-normal iron levels.The AST level demonstrated a significant association with the type of chelation treatment (P = 0.003). The duration of blood transfusion (in years) showed a strong positive correlation with the patients' age (Pearson's coefficient = 0.996).

Conclusion: This study indicates the elevated serum ferritin levels and APRI scores in patients with beta-thalassemia major, most of whom have abnormal MRI T2* findings. However, no significant correlation was observed between the APRI score, serum ferritin level, and MRI T2* results.

Keywords: Thalassemia, Blood transfusion, Iron overload, MRI T2*

Introduction

Beta-thalassemia is a prevalent inherited anemia worldwide, particularly in regions known as the thalassemia belt, including the Mediterranean, the Arabian Peninsula, parts of Africa, Iran, Turkey, India, and Southeast Asia (1). In these areas, the prevalence of the disease-associated gene ranges from 5.5% to 15%, while it is significantly lower in Western countries. In Iran, the number of thalassemia patients exceeds 18,000, with a higher concentration in regions surrounding the Persian Gulf, the Caspian Sea, and the Sea of Oman.

The prevalence of beta-thalassemia in Isfahan and central Iran has reached approximately 8% (2-4).

Beta-thalassemia is characterized by inadequate or zero production of the beta chain of hemoglobin, resulting in imbalanced hemoglobin structure and the premature destruction of red blood cells. The symptoms of the disease include anemia, change of physical appearances, skeletal abnormalities, weakness, and growth retardation (5).

Severe forms of beta-thalassemia are characterized with varying degrees of anemia in early childhood, necessitating

regular and frequent blood transfusions starting from the first year of life (6). While blood transfusion is life-saving, it inevitably leads to complications such as excessive iron accumulation in vital organs such as the heart, endocrine glands, and liver, resulting in complications associated with organ-specific iron deposition, particularly in the liver (7). Iron deposition-related complications include liver cirrhosis, cardiac disorders, diabetes, hypothyroidism, and hypogonadism (7, 8). Preventing the complications related to iron deposition requires iron chelation therapy, typically administered subcutaneously. The deposition of iron in vital organs, especially the liver, is of particular concern (9).

The liver serves as the primary organ for iron storage in the body. Iron accumulation and reactive oxygen species can cause hepatocellular damage and contribute to the development of liver fibrosis (10). Iron overload, hepatitis C infection, and frequent blood transfusions are the factors that can accelerate the progression of liver fibrosis (11).

Serum ferritin levels are commonly used to assess and monitor iron overload in thalassemia patients, as they correlate with hepatic iron concentration (12). However, it is essential to note that liver biopsy is the gold standard for the diagnosis of liver fibrosis despite its invasiveness and cost (13). Therefore, alternative, less invasive, more cost-effective, and reliable methods are needed to evaluate iron overload in the body and liver. One such method is the Aspartate Aminotransferase-to-Platelet Ratio Index (APRI), which is a novel criterion (13).

Additionally, measuring iron levels, such as serum ferritin, can be influenced by various diseases and inflammatory conditions, making it challenging to diagnose increased iron loads because this marker also increases in many other disorders (14, 15). Liver biopsy, as previously mentioned, is invasive and has specific disadvantages. In recent years, cardiac and liver magnetic resonance imaging (MRI) has been used to assess iron stores in patients. MRI is a safe non-invasive method that provides a more accurate evaluation of body iron stores, as it directly measures iron accumulation without being affected by other factors (16). The interaction between ferritin, or hemosiderin, and water molecules can decrease the transverse magnetic velocity, resulting in a decrease in T2 and T2* relaxation times in iron-rich tissues. This phenomenon appears as a decrease in the signal intensity and makes the tissue look darker on magnetic resonance imaging (MRI) (16, 17).

Given the significance of early diagnosis and treatment of increased iron overload, which can reduce morbidity and mortality in patients with beta-thalassemia major, this study aims to investigate how ferritin levels and APRI scores correlate to MRI T2* findings in children with beta-thalassemia major.

Materials and Methods

This retrospective analytical study was conducted at Shahid Sadoughi Hospital in Yazd, Iran, from 2023 to 2024. The study aimed to investigate the relationship between certain factors and beta-thalassemia major in pediatric patients under 15 years of age who had undergone multiple blood transfusions (at least ten units). The research population included all the eligible patients who met the inclusion criteria. Several exclusion criteria were also applied, including non-cooperation of the patients and their parents, history of aplastic anemia, history of blood cancer or leukemia, history of hepatitis, type 1 diabetes, or cancer, and history of congenital liver diseases.

The Aspartate Aminotransferase-to-Platelet Ratio Index (APRI) was calculated using the following formula:
APRI = (AST / Upper Limit of Normal AST) × 100 / Platelet count (10^9/L).
Additionally, a collaborating radiologist quantitatively assessed the Relaxation Time factor in liver MRI T2*. The data were collected and subsequently analyzed with the SPSS software version 26. The qualitative data were presented as frequencies and percentages, while the quantitative data were reported as means and standard deviations. The Pearson correlation test assessed the relationships between the variables, with R and P values reported. Furthermore, the variables that were significantly correlated to the three dependent factors were subjected to multivariate analysis to determine their adjusted effects. A significance level of less than 0.05 was considered statistically significant.
Ethical considerations were strictly followed throughout the study. The research protocol received approval from the Ethics Committee of Shahid Sadoughi University of Medical Sciences in Yazd, Iran, with the ethics code of IR.SSU.MEDICINE.REC.1402.103. To ensure the patients' data confidentiality, all the personal information was treated with utmost discretion. Also, informed consent was obtained from all the participants before their involvement in the study.

Results

Out of the 70 patients included in the study, 35 were male (50%) and 35 were female (50%). The average age of the study participants was 10.76 ± 3.52 years. The youngest patient was three years old, while the oldest one was 15. None of the patients had undergone splenectomy.
Among the participants, 71.4% received treatment with Deferasirox tablets, 10% were treated with a combination of Desferal injections and Deferasirox tablets, 8.6% received a combination of Desferal injections and Deferiprone tablets, 5.7% took Desferal injections alone, and 4.3% were treated with Deferiprone tablets alone.
For the studied patients, the mean relaxation time in liver MRI T2* was 4.91 ± 4.42 ms. The recorded parameters included the APRI index (1.90 ± 0.55), platelet count (350.64 ± 134.26 × 10^3/mL), aspartate aminotransferase (AST) level (36.29 ± 23.82), ferritin level (3244.04 ± 285.01 ng/mL), and duration of blood transfusion (9.71 ± 3.49 years).
Regarding the extent of liver iron overload, 30% of the patients (21 cases) were classified as having a severe overload, 27.1% (19 cases) had a moderate overload, 21.4% (15 cases) had a very severe overload, 18.6% (13 cases) had a mild overload, and 2.9% (2 cases) had a close-to-normal condition.
There were no significant differences in terms of the relaxation time variables in liver MRI T2*, the APRI index, AST level, and the duration of blood transfusion based on the patients' gender (P = 0.299, P = 0.108, P = 0.460, and P = 0.432, respectively). However, the platelet count and ferritin level showed significant associations with gender, as determined by the Independent Samples Test (P = 0.047 and P = 0.006, respectively) (Table I).
Moreover, there was no significant difference in the liver MRI T2* relaxation time, APRI, platelet level, ferritin level, and the duration of blood transfusion concerning the type of chelator treatment (P = 0.382, P = 0.108, P = 0.983, P = 0.866, respectively). Similarly, no significant difference was found between the AST levels in the patients with regard to the type of chelator treatment (P = 0.631 and P = 0.379). However, a significant relationship was observed between the AST level and the type of chelator treatment (P = 0.003). A significant difference was also noted between the

group receiving Deferiprone tablets alone and the group receiving Deferasirox alone (P = 0.016), as well as between the group receiving Deferiprone tablets and Desferal injections and the group receiving Deferasirox alone (P = 0.023). Furthermore, there was a significant difference between the group receiving Deferasirox alone and the group receiving Deferasirox together with Desferal (P = 0.001). Lastly, there was a significant difference between the group receiving Deferiprone tablets along with Desferal and the group receiving Deferasirox tablets along with Desferal (P = 0.009).

The ferritin level and APRI did not significantly correlate with MRI T2*. However, a significant correlation was found between the duration of blood transfusions and the patients' age; the Pearson coefficient in this case was 0.996 (P = 0.0001) (Table II).

Table I: Mean quantitative variables in patients with beta-thalassemia major

Variable	Gender	Mean	Standard deviation	P Value
Liver MRI T^2* (ms)	Male	4.94	3.68	0.299
	Female	4.88	4.97	
APRI	Male	0.75	2.67	0.108
	Female	0.35	0.40	
Platelet level (10^3/ml)	Male	349.06	121.66	0.047
	Female	352.03	147.56	
AST level (U/L)	Male	34.69	24.00	0.460
	Female	37.89	23.88	
Ferritin level (ng/mL)	Male	2632.26	2171.85	0.006
	Female	3855.83	3316.53	
Duration of blood transfusion (years)	Male	9.80	3.42	0.432
	Female	9.63	3.62	

Table II: Correlation results of the investigated variables in patients with beta-thalassemia major

Variable		Liver MRI T^{2}* (ms)	APRI	Platelet level (10^{3}/ml)	AST level (U/L)	Ferritin level (ng/mL)	Duration of blood transfusion (year)	Age (year)
MRI T2* (ms)	Pearson correlation coefficient	1	-.044	.258	-.345	-.434	.081	**.082**
	P value		.716	.031	.003	.000	.503	**.502**
	Number	**70**	**70**	**70**	**70**	**70**	**70**	**70**
APRI	Pearson correlation coefficient	-.044	1	-.159	.071	.093	-.017	**-.015**
	P value	.716		.189	.558	.446	.890	**.902**
	Number	**70**	**70**	**70**	**70**	**70**	**70**	**70**
Platelet level (10^{3}/ml)	Pearson correlation coefficient	.258	-.159	1	-.239	-.389	-.207	**-.213**
	P value	.031	.189		.046	.001	.086	**.076**
	Number	**70**	**70**	**70**	**70**	**70**	**70**	**70**
Aspartate aminotransferase **(AST) level (U/L)**	Pearson correlation coefficient	-.345	.071	-.239	1	.581	-.176	**-.158**
	P value	.003	.558	.046		.000	.144	**.191**
	Number	**70**	**70**	**70**	**70**	**70**	**70**	**70**
Ferritin level (ng/mL)	Pearson correlation coefficient	-.434	.093	-.389	.581	1	.191	**.186**
	P value	.000	.446	.001	.000		.113	**.122**
	Number	**70**	**70**	**70**	**70**	**70**	**70**	**70**
Duration of blood transfusion (year)	Pearson correlation coefficient	.081	-.017	-.207	-.176	.191	1	**.996**
	P value	.503	.890	.086	.144	.113		**.000**
	Number	**70**	**70**	**70**	**70**	**70**	**70**	**70**
Age (year)	Pearson correlation coefficient	.082	-.015	-.213	-.158	.186	.996	**1**
	P value	.502	.902	.076	.191	.122	.000	
	Number	70	70	70	70	70	70	**70**

Discussion

Thalassemia is a hereditary blood disorder characterized by the production of abnormal hemoglobin (18). Blood transfusion is the primary treatment method used to prolong the lifespan of thalassemia patients (19). However, blood transfusion poses certain risks, including iron overload, infection, antibody formation against red blood cells, hyperactive reactions, and cholecystitis (20).

Excessive transfusion-induced iron overload can damage the heart, liver, and endocrine systems, including glands responsible for hormone production and regulation in the body. The accumulation of iron deposits characterizes this damage. Without proper treatment and the use of iron chelators, iron in nearly all patients with beta-thalassemia will increase to fatal levels (21). Consequently, vital organs such as the liver, heart, and endocrine glands gradually lose their function due to excessive iron accumulation.

Cardiac siderosis, which occurs as heart failure, arrhythmia, myocarditis, pericarditis or heart attack, is the leading cause of death (71%) in patients with thalassemia major (22).

In thalassemia major, liver fibrosis is directly correlated to age, the number of the blood transfusions received, and liver iron concentration. Therefore, regular assessment of iron status is crucial to effectively manage the iron overload in patients with beta-thalassemia major. Although liver biopsy along with the biochemical measurement of liver iron concentration is considered as the gold standard for evaluating the total body iron reserves, it is an invasive technique and not practical for routine use (23).

The present study aimed to investigate the correlation of serum ferritin levels and APRI criteria to liver T2* MRI findings in 70 children diagnosed with beta-thalassemia major. The mean duration of liver T2* MRI in the studied patients was 4.91 ± 4.42 ms, with an APRI index of 1.90 ± 0.55. The platelet levels were found to be 350.64 ± 134.26 (x10^3/mL), while the AST levels were 36.29 ± 23.82. The serum ferritin levels were found to be 3244.04 ± 285.01 (ng/mL), and the average duration of blood transfusion was 9.71 ± 3.49 years. Based on the MRI T2* results, 30% of the patients had severe liver iron overload, 27.1% had a moderate overload, 21.4% had a severe overload, 18.6% had a mild overload, and 2.9% had iron levels close to normal. The platelet and ferritin levels showed a significant relationship with the gender of the patients, with lower levels observed in the boys than in the girls with the same conditions. Additionally, there was a strong positive correlation between the duration of blood transfusion (in years) and the age of the patients, as indicated by a Pearson coefficient of 0.996.

Hashemieh et al. (24) studied 120 patients and indicated a moderate correlation between MRI T2* relaxation time and serum ferritin levels. However, they concluded that serum ferritin levels are not a reliable predictor of kidney iron overload.In the present study, there were elevated levels of ferritin and MRI T2* relaxation time which surpassed the normal range. Unlike the findings of Hashemieh and his team, no significant correlation was found between these two variables in this study. It is worth noting that the correlation reported in their research was deemed moderate.

In a study conducted on 154 patients, Pipaliya et al. (25) found a significant correlation between the liver fibro scan results andthe MRI T2* findings, while no significant relationship was observed between the tissue elastography results and the serum ferritin levels. Similarly, in the present study, there were elevated levels of APRI, ferritin, and MRI T2*, aligning with the results of the study by Pipaliya's team. However, ferritin and APRI levels were not significantly correlated to MRIT2*.

Keikhaei and Niazpoor (26) examined 80 patients with tuberculosis anemia and demonstrated a significant negative correlation between ferritin and liver MRI T2*. Furthermore, Ghazizadeh et al. (27) studied 31 patients and revealed that individuals with abnormal MRI T2* results for their liver often had abnormal spirometry. The present study noted that over 97% of the patients with beta-thalassemia major had abnormal T2* MRI findings in the liver.

Conclusion

The findings of this study indicate the elevated levels of ferritin and APRI in patients with beta-thalassemia major compared to normal levels. Additionally, the majority of these patients exhibit abnormal MRI T2* results. However, no

significant correlation was observed between APRI index, ferritin serum level, and MRI T2*. It is recommended for future studies to use larger sample sizes and control other variables to validate or refute the findings of the present study.

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None

Authors' contribution
Roohollah Edalatkhah was in charge of formal analysis, investigation, methodology,
validation, writing the original draft, reviewing and editing.
Marjan Kargar undertook the data collection and writing the original draft.
Maryamalsadat Yazdanparast contributed to methodology, validation, and writing the original draft.

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Ethical considerations
The research protocol received approval from the Ethics Committee of Shahid Sadoughi University of Medical Sciences in Yazd, Iran (ethics code: IR.SSU.MEDICINE.REC.1402.103).

Conflict of interest
The authors declare no conflict of interest.

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