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Original Article

Liver iron overload and hepatic function in children with thalassemia major

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Abstract

Background Routine blood transfusions and increased intestinal iron absorption lead to iron accumulation in various organs, especially the liver. To date, T2-star magnetic resonance imaging (T2*MRI) is a valuable tool to evaluate iron level in organs.

Objective To assess the degree of liver iron overload among children with thalassemia major (TM) and its possible correlations with hepatic function laboratory values.

Methods This cross-sectional study was conducted in Cipto Mangunkusumo Hospital. The degree of liver iron overload was evaluated by T2*MRI. Assessments of serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), albumin, and bilirubin levels were done to evaluate liver function.

Results A total of 291 TM children were included in this study. The mean age of subjects was 12 years. Most of the subjects were diagnosed as β-thalassemia homozygote (54.6%) and β-thalassemia/HbE (41.2%). Deferiprone (DFP) was the most commonly used iron chelator. Less than 10% of the subjects had normal liver iron deposition. The AST and ALT values increased proportionally with the severity of liver iron overload, with significant, moderately negative correlation coefficients (r=-0.388 and -0.434, respectively). However, albumin level decreased proportionally with the severity of liver iron overload, with a significant, moderately positive correlation coefficient (r=0.323). Liver T2* MRI had no significant correlations with direct, indirect, and ratio of direct/total bilirubin levels.

Conclusion Most of the children with TM have mild to severe liver iron overload. Liver T2* MRI has significant, moderate correlations with AST, ALT, and albumin values. Bilirubin level has no correlation with T2* MRI. Our findings suggest that monitoring of AST, ALT, and albumin levels is important because they may reflect the severity of liver iron overload. However, they should not be used as the only predictors of iron overload. **[Paediatr Indones. 2018;58:233-7; doi: http://dx.doi.org/10.14238/pi58.5.2018.233-7].**

Keywords: *thalassemia; liver iron overload; MRI; hepatic function test*

Thalassemia is an inherited blood disorder characterized by decreased or absent globin chains. It is the most common single gene disorder worldwide and is mostly inherited in an autosomal recessive pattern. There are two main types of thalassemia, α- and β-thalassemia. The combination of thalassemia and hemoglobin variant has a high prevalence in the population.[1]

Thalassemia major (TM) is the most severe form of thalassemia. The two main treatments for TM are routine blood transfusions and iron chelation therapy. The iron from blood transfusions accumulates in organs. A state of chronic anemia causes increased iron absorption in the gastrointestinal system. Subsequently, these two conditions lead to iron overload in various organs, which may induce cell damage.[2]

Among various organs, the liver has the highest capacity to store excess iron in the body and is very prone to damage by iron toxicity.[3] Therefore,

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evaluation of iron deposition in the liver is crucial in TM patients. One of the best techniques to evaluate liver iron deposition is T2* MRI because it is non-invasive, reproducible, and accurate.[4] In this study, we aimed to evaluate the degree of liver iron overload in pediatric thalassemia major patients, as well as its correlation with laboratory values of hepatic function.

Methods

This cross-sectional study was done in Cipto Mangunkusumo Hospital. Iron deposition in the liver was assessed by T2* gradient echo (GRE) sequence MRI 1.5 Tesla (Siemens Avanto, Germany). Liver T2* value was acquired after scanning the center of the liver at 12 different echo times (1.3-23 ms) and analyzed using *CMRtoolsTM* software (Thalassemia-Tools, London, United Kingdom). The degree of liver iron overload was determined based on T2* MRI value: normal >6.3 ms, mild 2.7-6.3 ms, moderate 1.4-2.7 ms, and severe <1.4 ms.5 Blood specimens were collected to evaluate laboratory values of liver function: AST, ALT, albumin, direct bilirubin, and indirect bilirubin. The normal reference laboratory values for these assessments were as follows: AST <27 U/L; ALT <23 U/L; albumin 3.2-4.4 g/dL; direct bilirubin <0.3 g/dL; and indirect bilirubin 0.3-0.9 g/dL.

Results

A total of 291 TM children were included in this study. The mean age of subjects was 12 (SD 1.64) years. There was no significant difference between the number of male and female subjects. Most subjects were diagnosed with β-thalassemia (54.6%) and β-thalassemia/HbE (41.2%). Deferiprone (DFP) was the most common iron chelator used, followed by deferasirox (DFX), and a combination of DFP-DFX. None of the subjects used deferoxamine (DFO) as a single iron chelator. More than 90% of subjects had mild to severe liver iron overload (**Table 1**).

Table 2 shows the laboratory values of hepatic function in different stages of liver iron overload.

Table 1. Demographic characteristics of subjects

Variables	(N=291)
Mean age, years (SD)	12 (1.64)
Sex, n (%)	
Male	156 (53.6)
Female	135 (46.4)
Type of thalassemia, n (%)	
α-thalassemia	10 (3.4)
β-thalassemia	159 (54.6)
β-thalassemia/HbE	120 (41.2)
α-β-thalassemia/HbE	2 (0.8)
Iron chelator, n (%)	
Monotherapy	
DFO	0 (0)
DFP	191 (65.6)
DFX	40 (13.7)
Combination therapy	
DFO+DFP	17 (5.9)
DFO+DFX	5 (1.7)
DFP+DFX	34 (11.7)
No chelation	4 (1.4)
Degree of liver iron overload, n (%)	
Normal	27 (9.3)
Mild	182 (28.2)
Moderate	01 (34.7)
Severe	81 (27.8)

The AST tended to increase proportionally with the degree of liver iron deposition. The same result was observed for ALT values. However, mean albumin level in normal liver was the highest (4.66 g/dL), and decreased with increasing severity of iron overload. The mean direct and indirect bilirubin values were lower in mild and moderate liver iron overload, compared to normal liver. However, these mean values increased in the severe iron overload group. There was only a slight difference in direct bilirubin between the mild and moderate liver iron overload groups. The mean of direct/total bilirubin ratio was almost equal for all degrees of liver iron overload (0.29-0.33).

Table 3 and **Figure 1** show significant moderate correlations between liver T2* MRI and AST, ALT, and albumin ($P<0.05$ for all). The ALT value showed the strongest correlation ($r=-0.434$), followed by AST ($r=-0.388$), and albumin ($r=0.323$). However, the former two were negative and the third was a positive correlation. There was no significant correlation between liver T2* MRI and bilirubin level.

The scatterplots of the liver function test values are presented in **Figure 1**.

Table 2. Liver function indicators among different degrees of liver iron overload

Variables	Degree of liver iron overload			
	Normal (n=27)	Mild (n=82)	Moderate (n=101)	Severe (n=81)
Mean AST (SD), U/L	29.55 (11.44)	28.89 (18.04)	33.10 (16.30)	44.72 (22.35)
Mean ALT (SD), U/L	20.85 (17.74)	21.44 (22.04)	32.26 (23.60)	46.58 (37.93)
Mean albumin (SD), g/dL	4.66 (0.36)	4.55 (0.28)	4.46 (0.34)	4.32 (0.33)
Mean direct bilirubin (SD), mg/dL	0.53 (0.14)	0.42 (0.15)	0.43 (0.16)	0.59 (0.43)
Mean indirect bilirubin (SD), mg/dL	1.53 (1.03)	1.08 (0.55)	0.91 (0.39)	1.21 (0.52)
Mean direct/total bilirubin ratio (SD)	0.31 (0.08)	0.29 (0.06)	0.33 (0.05)	0.32 (0.08)

Table 3. Correlation coefficient between liver T2* MRI and liver function tests

Indicator	r	P value
AST	- 0.388	0.001
ALT	- 0.434	0.001
Albumin	0.323	0.002
Direct bilirubin	0.032	0.735
Indirect bilirubin	0.109	0.248
Direct/total bilirubin ratio	- 0.146	0.120

Discussion

The liver is the most important organ for iron metabolism in the body. It has three essential functions: 1) it is the primary site for production of iron-binding proteins, including transferrin, which is the major serum iron-binding protein that maintains systemic iron balance; 2) it is also the major storage site for iron excess, facilitated by liver ferritin that can store up to 4,500 atoms of iron; and 3) it controls the mobilization of iron from storage site to circulation for metabolism.[3]

Liver contains about 70% of total body iron. Therefore, liver iron concentration may reflect the total iron body contents and be useful for monitoring response to therapy. This fact gives rise to another consequence: the liver is the organ most damaged by iron, compared to other organs in the body.[6] Accumulated iron in cells leads to the production of reactive oxygen species (ROS), primarily by Fenton reaction. Ferrous iron catalyzes the decomposition of hydrogen peroxide, forming hydroxyl radicals, which are the most toxic ROS. Hydroxyl radicals target both carbohydrate, protein, and nucleic acids. Long term iron toxicity leads to cell death, fibrosis, and carcinogenesis.[7,8]

Several biochemical markers are widely known to reflect hepatic function: AST, ALT, albumin, and serum bilirubin. In our study, increased AST and ALT may reflect the severity of liver iron overload. The AST and ALT are cytoplasmic enzymes that catalyze the transaminase reaction in liver. Any kind of hepatocellular injury may disrupt cellular membrane permeability and cause leakage of transaminase enzymes into the extracellular compartment. Subsequently, elevated enzyme activity can be detected in the blood.[9,10]

Another interesting finding in this study was that mean AST values were observed to be within normal range in all iron overload groups, whereas mean ALT values were normal only in the normal and mild liver iron overload groups. This finding may have been due to ALT's greater specificity for liver than that of AST. The highest concentration of AST is found in heart, compared to other organs such as liver, skeletal muscle, and kidney. However, ALT is primarily found in liver, compared to other organs.[9,11]

Our study demonstrated that albumin synthesis decreased as the severity of liver iron overload increased. The coefficient correlation between liver T2* MRI value and albumin was positive, unlike the transaminases. During albumin metabolism, only small portions of synthesized albumin are stored in liver, while the majority is released into the bloodstream. Any hepatocyte injury may cause decreased production of albumin, which can be then detected in blood.[12,13]

We did not find any significant correlations between liver T2* value and direct, indirect, or direct/total bilirubin ratio. Almost all subjects in this study had abnormal increased total bilirubin value (>1.2 mg/dL), with direct/total bilirubin ratio of 0.3. This finding

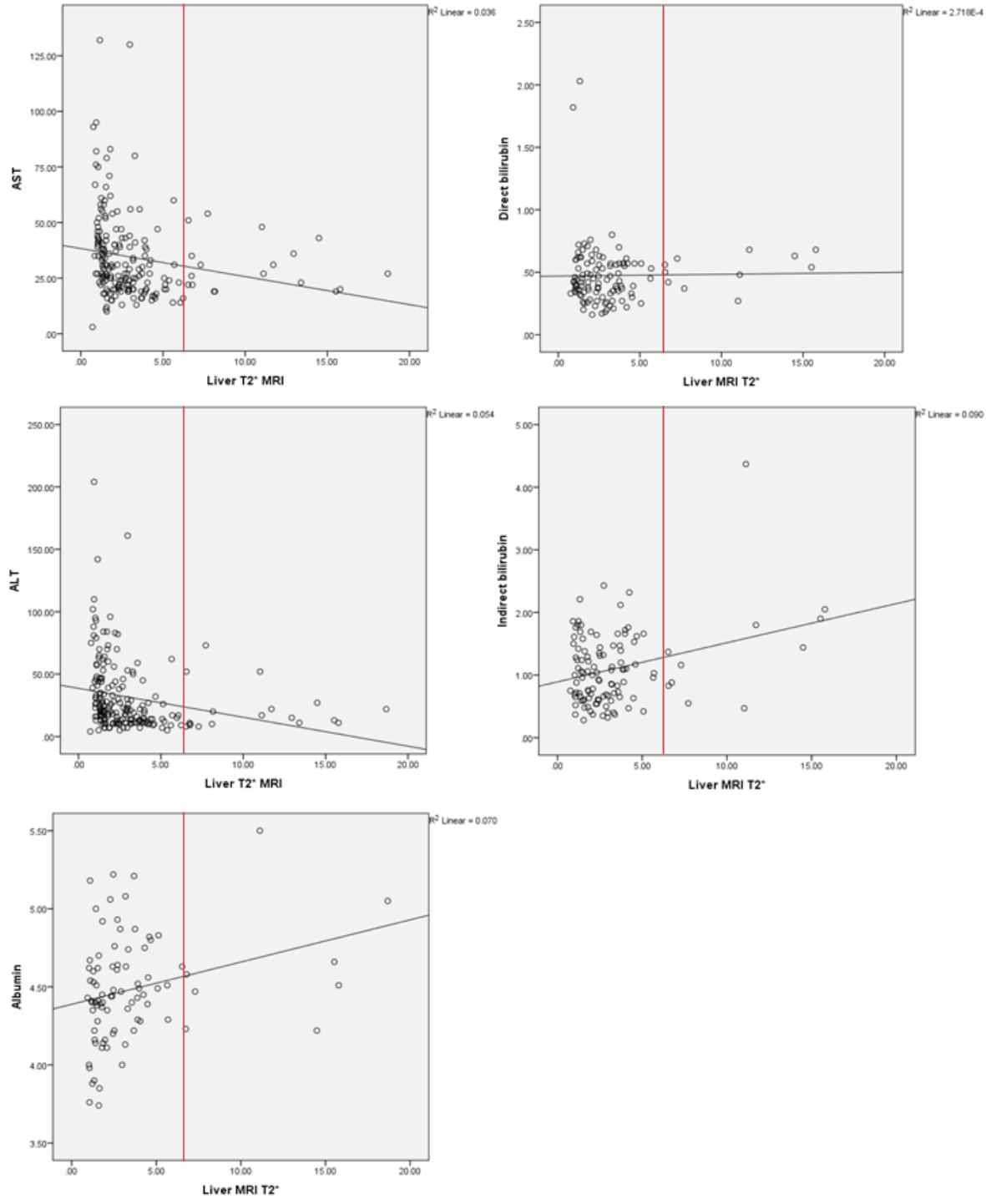

Figure 1. Scatterplots between liver MRI T2* value vs. hepatic function laboratory values (T2* value on the right side of red line is categorized as normal)

means that most of the subjects were in a conjugated hyperbilirubinemic state, regardless of the degree of iron overload. Conjugated hyperbilirubinemia in thalassemia subjects may be caused by hepatocellular damage due to iron toxicity.[9,14]

This study demonstrates that most children with TM have mild to severe liver iron overload. There are significant, negative moderate correlations between liver T2* values and AST and ALT, as well as a significant, positive moderate correlation with albumin. Bilirubin level has no correlation with T2* value. Our findings suggest that monitoring of AST, ALT, and albumin levels is important because they may reflect the severity of liver iron overload. However, they should not be used as the sole predictors of liver iron overload.

Conflict of interest

None declared.

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