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Nerve conduction velocity and electromyography study in children with transfusion-dependent beta-thalassemia

Aliaa Mohammed Diab[1], Elham Abdelghafar Nawar[1], Shaimaa Alaa Mahmoud*[1], Khaled M. Abdel Azim Eid[2], Aziza Ibrahim Ali Ibrahim[1], Samar Mahmoud Elbahy[1]

[1] Pediatric Department, Faculty of Medicine, Benha University, Benha, Egypt

[2] Pediatric Hematology Department, Faculty of Medicine, Cairo University, Cairo, Egypt

Corresponding author: Shaimaa Alaa Mahmoud

Email: shaimaaalaa434@gmail.com

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Abstract:

Background: Beta thalassemia is the most prevalent chronic hemolytic anemia with multiple recognized complications. Neuropathic changes have been recorded more frequently these days due to longer life expectancies. We conducted this work to evaluate the neurophysiological status of transfusion-dependent thalassemic children and to investigate the impact of different clinical factors and iron overload status on the findings.

Methods: We conducted a non-randomized convenient study including 50 beta thalassemic children on regular blood transfusion, in addition to iron chelation therapy. All participants underwent a comprehensive clinical assessment, laboratory tests, and neurophysiological studies; nerve conduction and electromyography studies. Data were collected, including the most recent cardiac and hepatic MRI findings.

Results: Out of the 50 children included, 12 patients (24%) had abnormal sensory nerve conduction (peripheral neuropathy), and all children had normal motor nerve conduction. Only four patients (8%) showed myopathic changes by electromyography. These changes were strongly correlated with hepatic iron overload.

Conclusions: Children with transfusion-dependent beta-thalassemia on regular iron chelation therapy are at risk of neurological changes, as peripheral neuropathy and myopathy, necessitating regular follow-up of these patients.

Keywords: Nerve conduction study, Electromyography, Transfusion-dependant beta-thalassemia, peripheral neuropathy, myopathy.

Introduction

Beta-thalassemia is a genetic disorder that lowers the body's ability to make the hemoglobin beta-globin chain. People of Asian, Mediterranean, and Middle Eastern ancestry have the greatest frequency of beta-thalassemia mutations. The beta-globin gene has over 200 known mutations that cause thalassemia, which results in a great deal of variation in the disease's symptoms and genotype **(1).**

The life expectancy of thalassemic patients has been greatly increased thanks to blood transfusions and chelation therapy, turning thalassemia from a fatal childhood disease into a chronic condition that can be managed for a long time **(2)**. Subsequently, patients develop complications such as heart failure, liver cirrhosis, and multiple endocrine disorders that result from elevated iron deposition, which is caused by enhanced iron absorption and repeated transfusions during a person's lifetime **(3,4).** Thus, a crucial part of the medical therapy of thalassemia patients is assessing iron accumulation in various organs **(5).**

Multiple studies have shown that patients with transfusion-dependent thalassemia (TDT) have neurological involvement, most often in the form of cognitive impairment **(6,7).** Previous investigations have also revealed the neurotoxic effects of iron chelation therapy, namely in terms of ophthalmic and auditory nerve problems **(8)**. Moreover, adults and teenagers with TDT were reported to have peripheral neuropathy **(9).** Iron overload, chronic hypoxia, and deferoxamine-induced neurotoxicity are among the causes that have been linked to these consequences. However, the frequency of peripheral neuropathy in TDT children and its correlation with other disease-related variables is still undetermined **(9).**

This study aimed to evaluate the presence of peripheral neuropathy and myopathy in children with TDT and their correlation with different clinical and laboratory variables, including cardiac and hepatic iron overload measured by magnetic resonance imaging (MRI).

Methods:

This study is a non-randomized convenient study conducted on fifty thalassemic children regularly attending the Pediatric Hematology Clinic at Benha University and Benha Children Hospitals for treatment and follow-up, from February 2022 to February 2023. The children that were included belonged to the age group of 10 to 18 years old, had a clinical diagnosis of beta-thalassemia, which was verified using hemoglobin (Hb) electrophoresis, were receiving regular iron chelation therapy and blood transfusions for a minimum of seven years. Individuals with any condition affecting the nervous system or those with a history of neurological illness were excluded.

A full history was taken from all cases, especially the history of any neurological symptoms, duration of blood transfusion and chelation therapy, and previous surgery such as splenectomy. All cases were assessed clinically for neurological signs, and were subjected to laboratory tests as complete blood picture, serum ferritin levels, serum aspartate aminotransferase level (AST), and alanine aminotransferase level (ALT). The most recent cardiac and hepatic MRI results for iron overload status were collected.

All cases were subjected to neurophysiological studies (nerve conduction studies and electromyography) performed by Trutrace Deymed Diagnostic device at the neurophysiological clinic at the Pediatric Department of Benha University Hospitals by a single Pediatric Neurologist.

For motor nerve conduction, the median nerve in the upper limb and the tibial nerve in the lower limb were assessed. For the sensory nerve conduction, the superficial peroneal nerve was assessed. Latency, amplitude, and nerve conduction velocity were measured. Electromyography was performed on the biceps muscle in the upper limb & tibialis muscle in the lower limb to assess the presence of myopathy.

Ethical considerations

Approval of the Research Ethics Committee at Benha University, Faculty of Medicine (MoHP No. 0018122017/ Certificate No: 1017), Study No. MS 16-2-2022) was granted prior to commencing the study in accordance with the 1964 Declaration of Helsinki's ethical standards. Before participating in the study, all participants' guardians were asked to provide written informed consent outlining the study's purpose and the procedures that would be carried out.

Statistical analysis

Version 20.0 of the IBM Statistical Program for the Social Sciences (SPSS) software was used for data analysis. Qualitative data was presented in the form of percentages and numbers. A normal distribution was confirmed using the Kolmogorov-Smirnov test. Mean and standard deviation were used to describe quantitative data. Multivariate and univariate binary logistic regression were used to identify the predictors of sensory neuropathy and myopathy. Significant results are indicated by a P value ≤ 0.05.

Results:

Fifty children (25 males and 25 females) with TDT were included in our study, with a mean age of 14.76 ± 2.38 years. Blood transfusion was given regularly every 3-4 weeks, the mean pre-transfusion Hb was 9.1 ± 0.69 gm/l, and the least duration of blood transfusion was 9.5 years with a mean of 14.43 ± 2.4 years. Splenectomy was performed in 13 patients (26%). The mean serum ferritin level was 3071.38 ± 2380.97 ng/dl. Chelating agents were given when serum ferritin level was above 1000 ng/dl. In our study, the least duration of chelation therapy was 9.2 years, with a mean of 13.63 ± 2.26. Different chelating agents were used, including deferasirox, deferiprone, and deferoxamine **(table 1)**.

Table (1): Clinical characteristics, laboratory findings, cardiac and hepatic MRI data of the study participants

Parameter	Mean	±SD (range)
Clinical characteristics		
Age (years)	14.76	±2.38 (10-18)
Sex, No. (%)		
Male	25	(50%)
Female	25	(50%)
Splenectomy, No. (%)	13	(26%)
Duration of blood transfusion (years)	14.43	±2. 4 (9.5-17.7)
Duration of chelation (years)	13.63	±2.26 (9.2-17)
Chelating agent, No. (%)		
Deferasirox and & deferiprone	24 (48%)	
Deferasirox	17 (34%)	
Deferasirox and deferoxamine	4 (8%)	
Deferoxamine and deferiprone	4 (8%)	
Laboratory data		
Hb (g/dl)	9.1	±0.69 (7.6-10.5)
Platelets (× 10^9/L)	314.28	±112.01 (160-600)
TLC (× 10^9/L)	9.17	±3.44 (4.56-19.5)
AST(U/l)	49.38	±44.87 (15-215)
ALT(U/l)	49.22	±63.58 (12-292)
Serum ferritin (ng/dl)	3071.38	±2380.97 (390-11318)
Cardiac and hepatic MRI		
LIC (mg/g)	8.32	±6.34 (0.24-22.7)
T2* (ms)	30.59	±13.29 (10-67)

SD: Standard deviation, Hb: Hemoglobin, TLC: Total leucocytic count, AST: Aspartate transaminase, ALT: Alanine transaminase, LIC: Liver iron concentration.

On assessing iron overload status by cardiac and hepatic MRI, it was noted that hepatic iron deposition was more evident than cardiac iron deposition, as 80% of the patients showed hepatic iron deposition ranging from light to severe, compared to only 20% of patients showing cardiac iron deposition **(figure 1).** Data of cardiac and hepatic MRI are presented in **(table 1).**

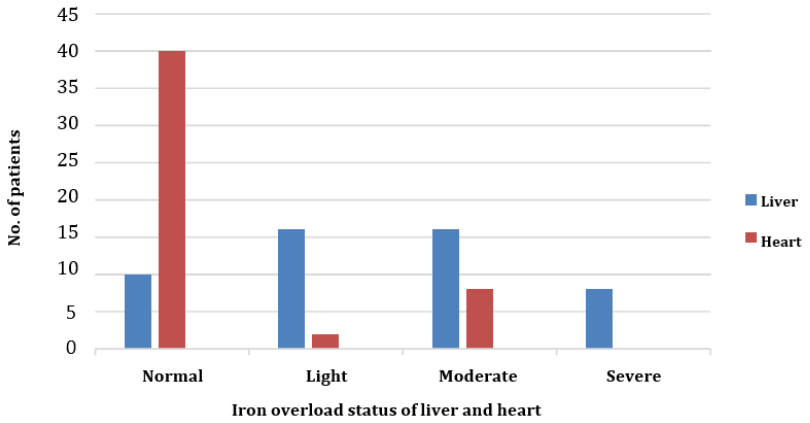

Figure (1): Iron overload status of the participants' heart and liver assessed by MRI

It was noted that liver iron concentration (LIC) was positively correlated with serum ALT level, which was, in turn, positively correlated with serum ferritin. Serum ferritin was negatively correlated with pretransfusion Hb. However, no correlation was observed with duration of blood transfusion, age, heart T2*, or LIC.

All participants had no neurological symptoms, and normal neurological examination. However, by neurophysiological studies, neuropathic changes were reported in 14 patients (28%). Nerve conduction studies showed that 12 patients (24%) had chronic axonal sensory neuropathy, while 38 patients (76%) had normal sensory nerve conduction, and all patients had normal motor nerve conduction **(table 2)**. Electromyography results showed that only four patients (8%) had small amplitude, polyphasic early recruitment, and patchy myopathic changes, and 46 patients (92%) had no abnormality detected **(figure 2).** Only two patients out of 50 (4%) had combined sensory neuropathy and myopathy.

Table (2): Nerve conduction study of the study participants

Examined nerve	Parameter	Mean	±SD (range)
Sensory nerve conduction			
Superficial peroneal nerve	**Amplitude (mv)** Normal range >6 mv	19.17	±22.54 (2.9-99.4)
	Latency (ms) Normal range <4.4 ms	2.95	±1.32 (0.6-6.2)
	CV(m/s) Normal range >40 m/s	58.79	±45.58 (25-246)
Motor nerve conduction			
Median nerve	**Amplitude (mv)** Normal range >4 mv	7.41	±2.84 (4-14.3)
	Latency (ms) Normal range <4.4	2.93	±1.26 (1.9-3.6)
	CV(m/s) Normal range >49 m/s	59.58	±7.68 (52-70)
Tibial nerve	**Amplitude (mv)** Normal range >4 mv	8.66	±2.65 (4-13.2)
	Latency (ms) Normal range <5.8 ms	3.34	±0.74 (1.9-4.8)
	CV(m/s) Normal range >41 m/s	55.30	±24.05 (42-214)

SD: standard deviation, CV: conduction velocity.

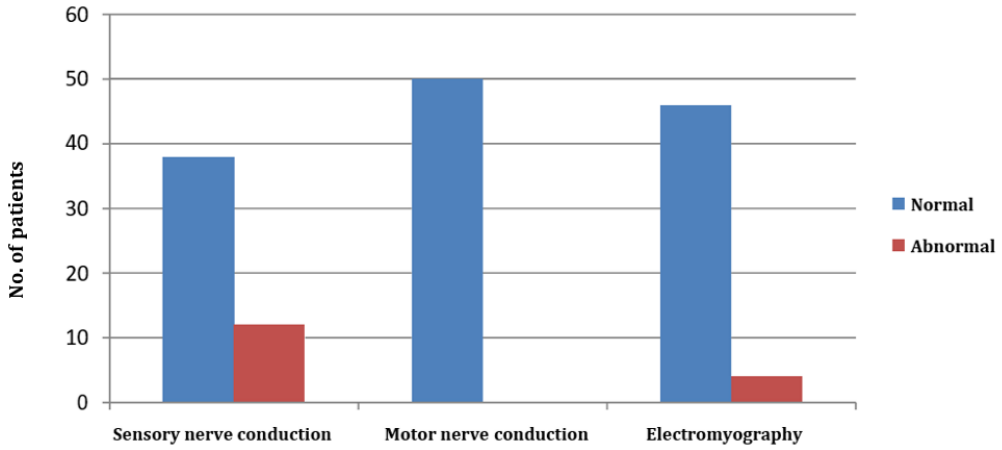

Figure (2): Neurophysiological status of the study participants

On performing univariate logistic regression analysis, older age, splenectomy, lower pretransfusion Hb, higher platelet counts, higher ALT levels, and increased LIC were significantly associated with sensory neuropathy. Only high ALT and increased LIC levels were significantly associated with myopathy **(table 3).** Duration of blood transfusion, chelation therapy, serum ferritin, and heart T2* were not risk factors for peripheral neuropathy or myopathy changes.

Table (3): Univariate binary logistic regression of clinical and laboratory parameters to predict sensory neuropathy and myopathy

Parameter	Sensory neuropathy				Myopathy			
	p-value	OR	CI		p-value	OR	CI	
			Lower	Upper			Lower	Upper
Age	.025*	1.923	1.463	1.949	. 103	1.718	. 896	3.297
Sex	.192	2.471	.634	9.625	. 998	$0.31^{*}10^{9}$	. 000	
Duration of disease	.153	.808	0.603	1.08	. 349	1.26	. 780	2.02
Duration of chelation therapy	.260	.840	.629	1.14	. 784	1.07	. 672	1.69
Splenectomy	.037*	1.886	1.056	1.913	. 274	. 314	. 040	2.500
Hb	.016*	0.558	0.933	0.062	. 550	. 645	. 153	2.721
TLC	.717	1.038	.850	1.267	. 515	. 883	. 606	1.285
Platelets	.017*	1.016	1.003	1.028	. 077	. 979	. 957	1.002
AST	.084	.988	.975	1.002	. 106	1.013	. 997	1.029
ALT	.021*	1.996	1.987	3.005	. 018*	1.015	1.003	1.027
Serum ferritin	.464	1.000	1.000	1.000	. 719	1. 000	.999	1. 000
Hepatic LIC (mg/g)	.002*	1.315	1.11	1.523	. 003*	1.264	1.041	1.304
Cardiac T2* (ms)	.514	.984	.938	1.033	. 632	. 980	. 900	1.066

OR: Odds ratio, CI: confidence interval, Hb: Hemoglobin, TLC: Total leucocytic count, AST: Aspartate transaminase, ALT: Alanine transaminase, LIC: Liver iron concentration.

Multivariate logistic regression analysis was performed to detect independent predictors of sensory neuropathy. High platelet counts, high ALT levels, and increased LIC were found to be significant **(table 4).** It was not possible to perform multivariate logistic regression analysis on myopathic patients due to their small number.

Table (4) Multivariate binary logistic regression of clinical and laboratory parameters to predict sensory neuropathy

Parameter	p-value	OR	CI	
			lower	Upper
Age	.141	1.006	.401	1.139
Splenectomy	.159	4.992	.534	46.706
Hb	.398	2.138	.367	12.457
Platelets	.045*	1.019	1.000	1.038
ALT(SGPT)	.048*	1.025	1.002	1.036
hepatic LIC (mg/g)	.022*	1.904	1.829	1.986
Constant	.522	.003		
R = 0.557 = p <0.001				

OR: Odds ratio, CI: confidence interval, Hb: Hemoglobin, ALT: Alanine transaminase, LIC: Liver iron concentration

Discussion

In the current study, neurophysiological studies were performed on a random sample of 50 thalassemic children with no neurological symptoms or signs. There were neuropathic changes in 14 patients (28%) in the form of chronic axonal sensory neuropathy, myopathy, or combined changes.

Literature is sparse regarding polyneuropathy in TDT patients. Neurological involvement is usually discovered during a neurophysiological or neuroimaging examination and does not immediately manifest with relevant signs and symptoms (i.e., subclinical) **(10).** The reported incidence of neuropathy in TDT patients ranged from 22 to 78% **(11).**

In this study, none of the patients reported neurological complaints or aberrant neurological clinical signs. However, after performing sensory nerve conduction examinations on the superficial peroneal nerve, it was found that 12 patients (24%) had chronic axonal sensory neuropathy, and all patients had normal motor conduction study. On performing electromyography, only four patients (8%) showed a short motor unit action potential duration, with small amplitude, polyphasic early recruitment, and patchy myopathic changes.

A study conducted by **Sawaya et al.** with 30 thalassemic patients found that while motor distal latencies were unaffected, sensory distal latencies in the upper and lower limb nerves exhibited a significant delay. Possible early sensory peripheral polyneuropathy was noted in 78% of thalassemic patients with delayed sensory latencies and 54% with decreased sural amplitudes. This is quite high compared to our results; this could be attributed to the fact that 40% of their patients reported symptoms, including symmetrical and distal lower extremity paresthesia and numbness, along with an older age range of 14 to 35 years. They emphasized that neuropathy correlated with older age and was worse in non-TDT, while blood transfusion and desferrioxamine appeared to be correlated with better nerve function. No correlation was found between neuropathy and hemoglobin level or splenectomy **(8)**.

Papanastasiou et al., in their study of 53 beta-thalassemia patients with a mean age of 17.9 **±** 7.9, found that 22% of thalassemic patients had characteristics of a mild peripheral sensorimotor neuropathy, both clinically and electrophysiologically. There was a strong positive association between the mean conduction velocity of the tibial and peroneal nerves and the mean hematocrit value, suggesting that this age-related polyneuropathy, which often manifests in the second and third decades of life, was caused by chronic hypoxia **(12).** Likewise, in **Nemtsas et al.** study, who studied 36 patients (age range, 16-57 years), 50% of patients with beta-thalassemia had either polyneuropathy or myopathy. 38.9% of patients had polyneuropathy, and 27.8% had myopathy, with one-third of the patients having an overlap of the two conditions **(7).**

In two different studies performed on 34 and 40 beta-thalassemia patients to assess nerve functions, sensory neuropathy was reported in 21% and 25% of patients, respectively **(13,14).** While in **Negi et al**. study, which included children with thalassemia (age range, 5-18 years), There were no significant differences between the patients and controls in the distal latency, amplitude, and nerve conduction velocity of the three motor and sensory nerves that were evaluated **(4).**

Kaushik et al. examined fifty TDT children (age range, 5-15 years), and none showed any sign of peripheral neuropathy, either clinically or electrophysiologically **(9)**. Similarly, **Işıkay et al.** examined 154 thalassemic children (age range, 1-17 years) and compared them to controls, found no evidence of large-fiber neuropathy or neurological problems in any of the subjects. This could be attributed to the young age of the included children in these studies **(15)**.

In the study by **Risha et al.,** neurological symptoms were present in 31.3% of the investigated cases with beta-thalassemia. Of the individuals exhibiting neurological symptoms, 25.5% had aberrant results from electrophysiological studies of the motor and sensory nerves; all of them were older than 16 years, indicating a positive correlation between neuropathy and age, which could be attributed to longer exposure to ischemia **(11).**

The disparity between the studies could be attributed to various factors, including variations in sample sizes, the age range of the patients under investigation, and the availability of advanced supportive care **(11).** According to reports, neurological consequences have been associated with several causes, such as prolonged hypoxia, iron overload, deferoxamine neurotoxicity, and bone marrow expansion; nevertheless, the exact etiology of these complications remains unclear **(16).**

On investigating the predicting factors of peripheral neuropathy in the study participants by univariate logistic regression analysis in our study, it was noted that these changes were significantly correlated with increased age, which is consistent with previous studies **(8,11,12,17),** indicating that these changes build up in older patients as a long-term complication of the disease.

Moreover, low hemoglobin level was associated with an increased risk of neuropathy in this study; this was similarly found in previous studies **(12,17),** indicating that chronic hypoxia plays an important role in the development of polyneuropathy. Nerve ischemia was discovered to be a well-known risk factor, which was more pronounced in sensory than in motor nerve conduction investigations **(18).** Also, in this study, splenectomy and higher platelet count were significantly correlated with neuropathy; this could suggest that microthrombi formation contributes to neuropathic changes in thalassemic patients.

One of the main factors contributing to the development of polyneuropathy and myopathy in TDT patients is iron overload. Iron overload was accused of many of the complications of thalassemia, as iron excess has been described as a contributor to free radical-induced cellular damage and neurotoxicity. It has also been shown previously that in vitro oxidative stress causes neuronal death in response to elevated ferric iron concentrations **(15)**. Furthermore, certain proteins, such as beta and alpha-synuclein, linked to effects on the central nervous system, can congregate due to iron buildup and oxidative stress. However, the peripheral nervous system's iron homeostasis remains a mystery **(19,20).**

In this study, neuropathy and myopathy changes were strongly correlated with increased hepatic iron overload as assessed by MRI and increased ALT, but no correlation between serum ferritin and neuropathy was detected in our study, this was confirmed by univariate and multivariate regression analysis. **Nemestas et al.** study revealed an important correlation between neurophysiological study results and iron overload assessed by MRI, but no correlation was found with serum ferritin; this result implies that serum ferritin is not a reliable indicator of iron overload and its associated consequences in all patients with thalassemia. They suggest that iron overload status, as assessed by MRI, could predict nerve and muscle injury **(7)**.

Contradictory results were found on the correlation between neuropathic changes and serum ferritin, suggesting that it could not be used as a dependable marker to detect iron overload status and predict related complications. Similar to our results, serum ferritin levels were very high in thalassemia patients in **Işıkay et al.** study, but they had no bearing on neuropathy **(15);** also, no significant correlation was detected between neuropathy and serum ferritin level in other studies **(11,12).** On the contrary, according to **Wong et al.,** there was a substantial correlation between serum ferritin levels and subclinical toxicity to the peripheral or central nervous systems **(13)**. Similarly, **El-Tagui et al.** found a strong association between high serum ferritin levels and motor neuropathy, as evaluated by electrophysiological studies. Serum ferritin level above 2000 ng/ml was a predictor of those at higher risk **(21). Negi et al.** found that as serum ferritin levels rose, distal latency and nerve conduction velocity slowed for both sensory and motor nerves **(4)**.

MRI is now the preferred tool for early detection, quantification, and monitoring of the cardiac and hepatic iron load. We recommend using regular MRI studies as a marker to predict and evaluate the presence of peripheral neuropathy and myopathy in TDT patients and subsequently aid in preventing and treating this problem.

Our study has limitations, such as the limited sample size, the young age of participants, and the fact that no control group was included. We recommend further studies that could assess neurophysiological studies on a larger sample of different age groups and compare them to healthy controls.

Conclusions:

In this study, transfusion-dependent thalassemic children were found to be susceptible to peripheral neuropathy, especially sensory neuropathy and patchy myopathy, which was more evident with increased age. Hepatic iron overload status, as assessed by MRI, was an important predictor of neuropathic and myopathic changes. Therefore, It is crucial to employ neurophysiologic monitoring to discover impairments in neuronal pathways early on and manage them appropriately.

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