Original Article *Iran J Ped Hematol Oncol. 2023, Vol 13, No 1, 52-63*

A survey of intracranial blood flow velocity in thalassemia intermedia in Khuzestan Province, Iran

Shahram Rafie[1], Bijan Keikhaei[2], Hourshid Sarshad[1*]

1. Department of Neurology, School of Medicine, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran
2. Thalassemia & Hemoglobinopathy Research center, Health research institute, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran.
*Corresponding author: Dr. Hourshid Sarshad, Department of Neurology, School of Medicine, Ahvaz Jundishapur University of Medical Sciences, Ahvaz. Email: drhsarshad1@gmail.com. ORCID ID: 0000-0002-1551-0682

Received: 17 November 2021 **Accepted:** 11 January 2022

Abstract

Background: Beta-thalassemia intermedia (BTI) is a type of hemoglobinopathy with an increased risk of cerebrovascular accidents, and transcranial cerebral Doppler ultrasonography (TCD) through determining the mean cerebral blood flow velocity (CBFV) can serve to predict the risk of a developing stroke. This study aims to compare patients with beta-thalassemia intermedia and healthy individuals in terms of the cerebral blood flow velocity.

Materials and Methods: This research was a case control study on 35 BTI patients and 25 healthy subjects. The patients were categorized into three age groups including 7-10, 11-15 and 16-20 years old. The mean CBFVs were compared between the two groups. The factors of age, gender, serum ferritin level, hemoglobin level, spleen size, thrombocytosis, and thalassemia genotypes were evaluated for their effects on CBFVs.

Results: Mean CBFVs were significantly higher in all the intracranial arteries of BTI patients compared to normal subjects (p-value < 0.05). The hemoglobin levels showed a negative correlation between the left and right vertebral arteries of BTI patients in terms of blood flow velocity (p-value < 0.05). The mean CBFVs in the left vertebral and basilar arteries were negatively correlated to age in BTI. There was no correlation among ferritin level, thrombocytosis, splenomegaly, splenectomy, XmnI polymorphism, and cerebral blood flow velocity in the BTI patients group (p-value > 0.05).

Conclusion: This study showed that cerebral blood flow velocities of BTI patients were higher than normal control group. In addition, CBFVs were not affected by factors such as gender, serum ferritin, platelet count, size of spleen and XmnI genotype, however, there was negative correlation between age and hemoglobin level with CBFVs.

Keywords: Blood flow velocity, Cerebrovascular accident, Doppler ultrasound, Thalassemia intermedia

Introduction

Beta thalassemia intermedia (BTI) is a type of hereditary hemoglobinopathy caused by disorders in the structure of beta hemoglobin chains (1, 2). The severity of the clinical features depends on the destructed globin and the structural changes in other chains and the co-inheritance of the other abnormal globin chains (3-5). BTI patients have a higher incidence of thromboembolic events (about 4%) than the general population. These patients are exposed to silent infarction (6-8). The main factors in this regard include nitric oxide synthesis deficiency, vascular endothelial dysfunction, chronic anemia, presence of conditions for the hypercoagulable state in the secondary thrombocytosis, cardiac and hepatic dysfunctions, hypothyroidism, splenectomy, and irregular transfusions (9-12). In patients with sickle cell anemia, Transcranial Doppler (TCD) ultrasonography is used to estimate the risk of stroke based on the flow rate of intracranial vessels (13). TCD is a simple method for achieving accurate and reproducible results without side effects (14, 15). Since BTI and sickle cell anemia are profoundly different in terms of pathophysiology, the results based solely on measuring the flow rate of intracranial

[DOI: 10.18502/ijpho.v13i1.11633]

vessels are not reliable. Because of the dearth of studies in this field, some studies conducted only on sickle cell anemia are reported here.

According to STOP criteria, time averaged mean of maximum blood flow (TAMM) is classified as usual (TAMM ≤170 m/s), conditional (170-200 m/s), and abnormal (TAMM ≥200 m/s) in middle cerebral or distal carotid arteries. It is recorded at two steps with an interval of at least two weeks and a transducer of 2 MHz. As STOP criteria suggests, there is a risk of stroke at values higher than 200 m/s; however, transfusion therapy can reduce the risk in more than 90% of cases (16, 17). Transcranial ultrasound can predict the risk of stroke in sickle cell anemia by measuring the speed of blood flow in the cerebral arteries. Despite the high prevalence of thalassemia and with the assumption that cerebral blood flow velocity in thalassemia is similar to sickle cell anemia, the vascular assessment of Thalassemia intermedia patients seems reasonable to prevent stroke and vascular accidents (12). Indeed, coping with the debilitating effects of stroke and the long-term impacts of silent infarctions on the life quality of thalassemia patients, such as functional and cognitive decline and loss of IQ, is necessary through preventive and curative measures (18).

Despite the confirmed association between TCD findings and stroke in patients with sickle cell anemia (19), no study has been done to determine the consequences of high blood flow rates in thalassemia intermedia patients. Therefore, this study aims to compare patients with beta-thalassemia intermedia and healthy individuals in terms of the cerebral blood flow velocity in and to investigate the effects of factors such as age (20), gender, spleen size or spleen resection, blood markers, and genetic patterns on the cerebral blood flow velocity in those patients.

Materials and Methods

The present study is of a case control type conducted in a hematologic outpatient clinic in southern Iran. The subjects were 35 patients from 7 to 20 years of age with the confirmed disease. The exclusion criteria were transfusion history, diabetes mellitus, congestive heart failure, vascular thrombotic events, and the use of contraceptive pills or hydroxyurea capsules in the past six months. The control group consisted of 25 healthy people with a normal cell count and no evidence of a blood disorder. The required data were obtained from the patients and the existing files. All the blood samples were taken in the laboratory of Shafa Hospital at 8:00 am, and all the subjects had stable hemodynamics and were tested in a supine position.

An examiner performed Doppler sonography with a French-made LOOKI device (190198), a 2-MHz probe for intracranial arteries, and an 8-MHz probe for internal carotid arteries. The standard protocol was performed for all contributers at rest and stable condition (21) through transtemporal, transorbital, transforaminal, and submandibular bone windows (22).

Statistical analysis

The data analysis was carried out using the Kolmogorov-Smirnov test for the normality of distribution. After the distribution of intracranial blood flow velocity was normalized, the effect of platelet count on velocity was examined with an independent t-test.

Depending on the normality of distribution, a series of tests were performed. The impacts of splenectomy and genetic map as well as the effect of splenomegaly and Xmnl gene were interpreted according to the non-parametric Mann-Whitney U test and the non-parametric Kruskal-Wallis test, respectively. The connection between age, serum ferritin and hemoglobin level on one hand and the blood flow velocity in intracranial arteries was investigated using Spearman's correlation coefficient. ANOVA, i.e., the one-way analysis of

[DOI: 10.18502/ijpho.v13i1.11633]

variance, was also used to compare the speed variations in the age groups. Moreover, LSD follow-up tests served to compare the differences among the age groups in terms of blood flow velocity. A regression test was used to study the synchronous effects of age and sex variables on cerebral blood flow velocity. P-values less than 0.05 were considered significant.

Ethical consideration

Ethical approval was obtained from the Ethical Committee of Ahvaz Jundishapur University of Medical Sciences. The ethical certificate number is IR.AJUMS.REC.1394.328.

Results

The results of the experiments revealed that the cerebral blood flow velocities in all the intracranial arteries, including middle, anterior, posterior cerebral, and basilar, and vertebral arteries were significantly higher in the BTI patients than in the control group (Figure 1). The effects of gender and age on the cerebral blood flow velocity, as found in the current study, are reported in Table I. It was observed that males and females were not different in this regard. The effect of age on the cerebral blood flow velocity in the BTI patients yielded significant negative spearman's correlation coefficients of -0.457 for the basilar artery and -0.445 for the left vertebral artery. To gain more accurate results, the effects of age and gender on cerebral blood flow velocity were evaluated through regression analyses. The results were significant on the right vertebral, basilar, and left vertebral arteries. The coefficient of determination (R^2) was 24% for the basilar artery, 27% for the right vertebral artery, and 23% for the left internal carotid artery. These percentages addressed age and gender variables together (Table II). To compare the cerebral blood flow velocity changes in different age groups, the patients were categorized into the three groups of 7-10, 11-15and 16-20 years of age. There were significant differences between the left and right vertebral arteries and between the basilar and left internal carotid arteries. More details are presented in Figure 2.

The effects of hemoglobin and serum ferritin levels are reported in Table III. As Spearman's coefficients in the table suggest, regarding their effects on the cerebral blood flow velocity in the patient groups, the hemoglobin levels were negatively correlated in the left and right vertebral arteries. Spearman's correlation coefficients referred to the point about those two vessels. There was no relationship between the serum ferritin level and the patients' cerebral blood flow velocity. Table IV presents the effects of the spleen status on the cerebral blood flow velocity in the patients. Among 35 BTI patients, 17 (48.6%) had splenomegaly, 14 (40%) were splenectomized, and 4 had a standard spleen size. The size of the spleen and splenectomy had no relationship with cerebral blood flow velocity. A comparison of cerebral blood flow velocities between patients with and without thrombocytosis is shown in Figure 3. Sixty percent of the patients had platelet counts of less than 500000. In the corresponding experiments, thrombocytosis was defined as a platelet count above 500,000 (23). In the current study, the platelet count did not significantly influence the cerebral blood flow velocity.

The effect of XmnI polymorphism on the cerebral blood flow velocity was evaluated too (Table V). Twenty patients were analyzed for their genetic XmnI patterns. Nine of them were negative for the gene, some were +/-, and the rest were +/+. No association was found between different genetic XmnI patterns and cerebral blood flow velocity. Table VI compares the cerebral blood flow velocities for B0/B0 and B0/B+ beta-thalassemia genotypes. Of the 25 patients participating in the genetic study, 19 were B0/B0 and 6 were B0/B+.

[DOI: 10.18502/ijpho.v13i1.11633]

The evaluation of the two beta-thalassemia genotypes (B0/B0 and B0/B+) and their cerebral blood flow velocities showed a significant difference in the right internal carotid artery, and the velocity was higher in the B0/B0 group.

Table I. Effects of gender and age association on cerebral blood flow velocity

*Arteries	Female	Male	P-value	Age Spearman's correlation coefficient	
Rt MCA	72.81 ± 30.89	65.15 ± 25.37	0.426	0.098	0.612
Lt MCA	88.75 ± 31.74	76.37 ± 23.47	0.194	-0.059	0.736
Rt ACA	57.87 ± 16.02	58.21 ± 24.07	0.962	-0.167	0.337
Lt ACA	65.31 ± 18.01	68.11 ± 31.13	0.754	-0.186	0.334
Rt PCA	51.25 ± 21.37	59.63 ± 21.24	0.255	-0.206	0.234
Lt PCA	61.81 ± 20.01	59.42 ± 28.13	0.778	-0.253	0.142
Rt ICA	57.75 ± 19.86	55.11 ± 18.86	0.689	-0.153	0.38
Lt ICA	61.37 ± 20.09	55.58 ± 30.34	0.519	-0.250	0.148
BA	71.25 ± 20.43	64.89 ± 27.67	0.453	-0.457	0.006
Rt VA	59.37 ± 23.59	47.73 ± 21.25	0.134	-0.277	0.107
Lt VA	55.69 ± 22.81	51.96 ± 19.53	0.605	-0.445	0.007

*ACA: anterior cerebral artery, BA: basilar artery, BTI: beta-thalassemia intermedia, ICA: internal carotid artery, MCA: middle cerebral artery, PCA: posterior cerebral artery, VA: vertebral artery, Lt: left, Rt: right.
The association between age and CBF velocity determined by Spearman's rank overall correlation coefficient

Table II. Simultaneous effects of age and gender on cerebral blood flow velocity

Arteries		Rt MCA	Lt MCA	Rt ACA	Lt ACA	Rt PCA	Lt PCA	Rt ICA	Lt ICA	BA	Rt VA	Lt VA
Regression	R Square	0.059	0.070	0.243	0.037	0.066	0.062	0.100	0.229	0.238	0.268	0.163
Unstandardised coefficient	sex	13.692	11.28	-5.32	2.119	-7.058	17.064	-2.141	6.448	1.137	11.409	-0.313
Unstandardised coefficient	Age	-0.689	-5.646	-5.353	-6.473	-4.91	-5.2	-10.972	-16.282	-17.327	-15.399	-10.049
	P-value	0.378	0.313	0.377	0.584	0.338	0.359	0.187	0.016	0.013	0.007	0.058

*ACA: anterior cerebral artery, BA: basilar artery, BTI: beta-thalassemia intermedia, ICA: internal carotid artery,, MCA: middle cerebral artery, PCA:posterior cerebral artery, VA: vertebral artery, Lt: left, Rt: right. The evaluation was conducted through regression analysis.

[DOI: 10.18502/ijpho.v13i1.11633]

Table III. Relationship of hemoglobin and serum ferritin level with cerebral blood flow velocity

	Arteries	Rt MCA	Lt MCA	Rt ACA	Lt ACA	Rt PCA	Lt PCA	Rt ICA	Lt ICA	BA	Rt VA	Lt VA
BTI group	Pearson's coefficient	-0.078	-0.251	0.037	0.157	0.089	-0.002	0.092	0.064	-0/274	365 -0/	-0/353
	P-value	0.657	0.146	0.835	0.367	0.61	0.927	0.527	0.713	0.112	0.031	0.038
Control group	Pearson's coefficient	-0.069	0.102	0.224	0.156	-0.125	-0.006	-0.162	-0.272	-0.015	-0.188	-0.147
	P-value	0.742	0.628	0.282	0.455	0.551	0.976	0.439	0.188	0.944	0.368	0.483
	P-value of two groups	0.97	0.19	0.49	0.9	0.43	0.99	0.36	0.22	0.34	0.49	0.42

*ACA: anterior cerebral artery, BA: basilar artery, BTI: beta-thalassemia intermedia, ICA: internal carotid artery, MCA: middle cerebral artery, PCA: posterior cerebral artery, VA: vertebral artery, Lt: left, Rt: right.

Table IV. Comparison of splenectomized and non-splenectomized patients as well as three groups of patients (normal size spleen, splenomegaly, splenectomized) in terms of cerebral blood flow velocity

Arteries*	Non-splenectomized (21 patients)	Splenectomized (14 patients)	P-value	Normal size (4 patients)	Splenomegaly (17 patients)	Splenectomized (14 patients)	P-value
Rt MCA	68.4 ± 31	69 ± 22	0.49	55.5 ± 23	71.4 ± 32	69.0 ± 22	0.70
Lt MCA	83.7 ± 27	79.4 ± 29	0.81	75.7 ± 23	85.6 ± 28	79.4 ± 29	0.69
Rt ACA	58.2 ± 23	57.7 ± 15	0.61	48.7 ± 10	60.4 ± 25	57.7 ± 15	0.85
Lt ACA	66.3 ± 30	67.5 ± 16	0.17	63.5 ± 11	66.9 ± 33	67.5 ± 16	0.18
Rt PCA	57.7 ± 21	52.8 ± 21	0.43	60.7 ± 13	57.0 ± 23	52.8 ± 21	0.57
Lt PCA	59.9 ± 27	61.3 ± 19	0.55	42.2 ± 15	64.1 ± 28	61.3 ± 19	0.98
Rt ICA	59.6 ± 20	51.3 ± 15	0.37	48.7 ± 19	62.1 ± 20	51.3 ± 15	0.20
Lt ICA	61.7 ± 31	53 ± 15	0.87	50.0 ± 6	64.4 ± 33	53.5 ± 15	0.70
BA	74.6 ± 28	57.5 ± 11	0.10	58.5 ± 25	78.5 ± 28	57.5 ± 11	0.09
Rt VA	58.4 ± 26	44.9 ± 13	0.16	44.5 ± 22	61.7 ± 26	44.9 ± 13	0.09
Lt VA	59.3 ± 23	45.0 ± 11	0.13	48.2 ± 16	62.0 ± 25	45.0 ± 11	0.09

*ACA: anterior cerebral artery, BA: basilar artery, BTI: beta-thalassemia intermedia, ICA: internal carotid artery, MCA: middle cerebral artery, PCA: posterior cerebral artery, VA: vertebral artery, Lt: left, RT:right.
The effects of splenectomy and spleen size on CBF velocity were determined via a non-parametric Mann-Whitney U test and a non-parametric Kruskul-Wallis test, respectively.

[DOI: 10.18502/ijpho.v13i1.11633]

Table V. Comparison of three types of XmnI polymorphism in terms of cerebral blood flow velocities

Arteries*	XmnI+/+(2)	XmnI+/-(9)	XmnI-/-(9)	P-value
Rt MCA	67.5 ± 10.6	64.33 ± 26.25	69.4 ± 25.96	0.72
Lt MCA	112.5 ± 53.03	77.11 ± 36.01	76 ± 18.17	0.42
Rt ACA	80.1 ± 8.38	53.67 ± 16.69	53.21 ± 6.53	0.15
Lt ACA	56 ± 10	63.44 ± 21.42	58.4 ± 11.67	0.57
Rt PCA	56.5 ± 20.5	61.22 ± 26.48	49.2 ± 17.65	0.69
Lt PCA	87 ± 2.83	57.67 ± 28.31	50.8 ± 10.8	0.37
Rt ICA	50.5 ± 0.71	56.22 ± 20	58 ± 15.31	0.62
Lt ICA	59 ± 15.56	49.22 ± 12.11	54.6 ± 17.59	0.73
BA	72 ± 32.53	56.78 ± 18.25	53 ± 6.16	0.81
Rt VA	69.5 ± 38.9	46 ± 17.94	40.2 ± 10.18	0.46
Lt VA	62.5 ± 14.85	47.56 ± 16.35	45.8 ± 8.79	0.35

*ACA: anterior cerebral artery, BA: basilar artery, BTI: beta-thalassemia intermedia, ICA: internal carotid artery, MCA: middle cerebral artery, PCA: posterior cerebral artery, VA: vertebral artery, Lt: left, RT: right. The results were achieved via a non-parametric Kruskul-Wallis test.

Table VI. Comparison of B0/B0 and B0/B+ patients in terms of cerebral blood flow velocity

Arteries*	B0/B0(19)	B0/B+(6)	P-value
Rt MCA	70.21 ± 32.32	52.67 ± 11.02	0.239
Lt MCA	83.74 ± 32.28	67.67 ± 14.84	0.265
Rt ACA	61.05 ± 19.87	54.67 ± 10.5	0.504
Lt ACA	68.11 ± 30.96	62.67 ± 16.33	0.824
Rt PCA	60.37 ± 24.19	52.17 ± 16.22	0.567
Lt PCA	61.42 ± 28.81	58.13 ± 6.91	0.99
Rt ICA	66.31 ± 20.05	42.5 ± 6.22	0.007
Lt ICA	66.21 ± 31.19	49.73 ± 16.41	0.226
BA	66.05 ± 27.19	60.17 ± 11.16	0.949
Rt VA	55.26 ± 26.34	39 ± 9.47	0.308
Lt VA	55.73 ± 22.32	43.83 ± 10.91	0.339

*ACA: anterior cerebral artery, BA: basilar artery, BTI: beta-thalassemia intermedia, ICA: internal carotid artery, MCA: middle cerebral artery, PCA: posterior cerebral artery, VA: vertebral artery, Lt: left, RT: right. The assay was done according to the non-parametric Mann-Whitney U test.

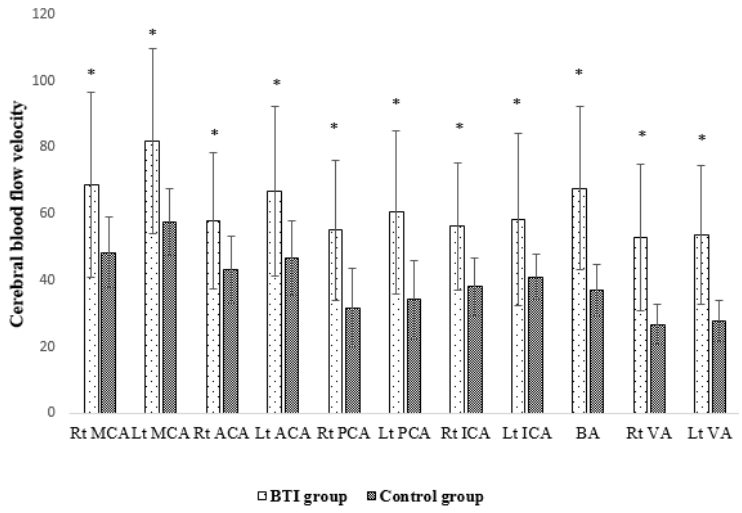

Figure 1. Comparison of intracranial flow velocities in BTI and healthy cases according to a parametric independent student t-test (*: significant difference of $p < 0.05$ between the groups. ACA: anterior cerebral artery, BA: basilar artery, BTI: beta-thalassemia intermedia, ICA: internal carotid artery, MCA: middle cerebral artery, PCA: posterior cerebral artery, VA: vertebral artery, Lt: left, Rt: right)

[DOI: 10.18502/ijpho.v13i1.11633]

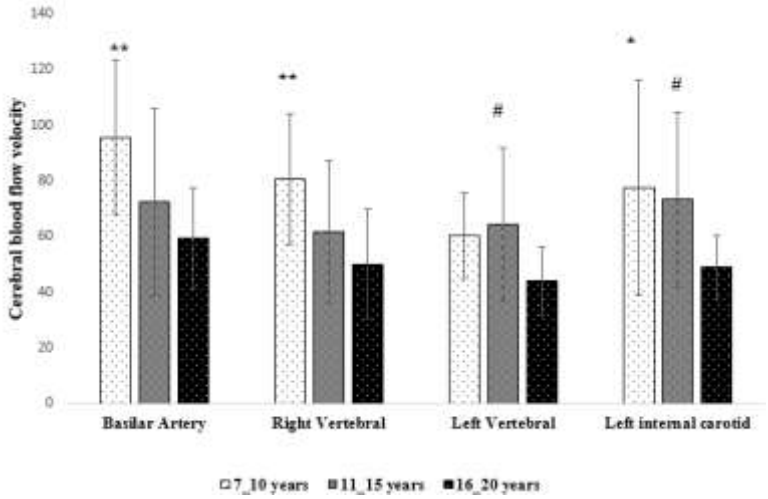

Figure 2. One-way ANOVA for the comparison of age groups in terms of cerebral blood flow velocity (*: significant difference of $p < 0.05$ between 7-10 and 16-20 age groups, **: significant difference of $p < 0.01$ between 7-10 and 16-20 age groups, #: significant difference of $p < 0.05$ between 11-15 and 16-20 age groups, ##: significant difference of $p < 0.01$ between 11-15 and 16-20 age groups)

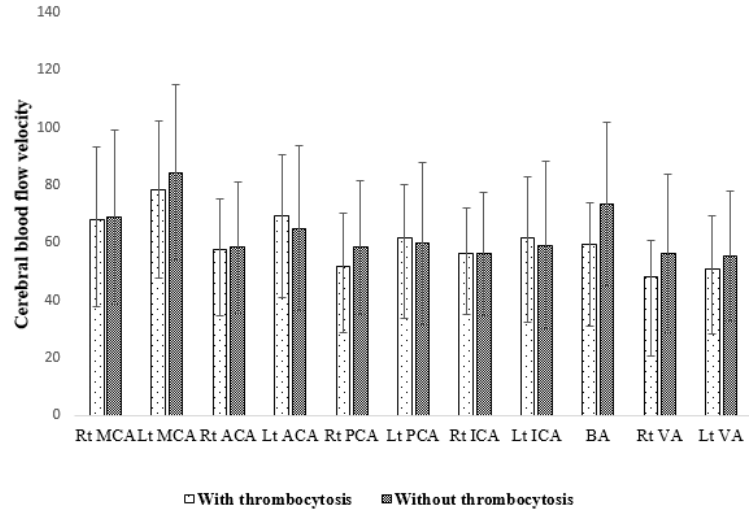

Figure 3. Comparison of cerebral blood flow velocities between the patient groups with and without thrombocytosis according to a parametric independent student T-test (*ACA: anterior cerebral artery, BA: basilar artery, BTI: beta-thalassemia intermedia, ICA: internal carotid artery, MCA: middle cerebral artery, PCA: posterior cerebral artery, VA: vertebral artery, Lt: left, RT: right)

[DOI: 10.18502/ijpho.v13i1.11633]

Discussion

This study used the Doppler ultrasound technology to evaluate the hemodynamic and intracranial flow velocities in BTI patients as compared to a control group of healthy people. The results generally showed that the blood flow velocities in all intracranial arteries, including middle, anterior, posterior cerebral and basilar arteries, as well as vertebral arteries were significantly higher for the BTI patients than for the control group. Due to various contradictory factors, the evaluation of atherosclerosis in patients with BTI has always been a field of ongoing research. Lowering the serum cholesterol and blood viscosity through lowering the hematocrit and arterial blood pressure is a task that can reduce the incidence of atherosclerosis. So, anemia is an independent risk factor for cardiovascular diseases, and a severe iron overload is a risk factor for atherosclerosis (24-26). In a study by Nassef et al. (26), atherosclerosis in adult patients with thalassemia intermedia was assessed by the measuring of the conduit artery flow velocity and wall thickness using color Doppler ultrasound. It emerged that a combination of laboratory tests of cholesterol and ferritin panels might provide a clearer view of atherosclerosis. Similarly, transcranial Doppler findings showed that mean flow velocities and pulsatility indices were higher in the patients than in the controls.

In splenectomized patients, hypercoagulability rises due to the prevalence of phosphatidylserine-positive red blood cells, thrombin generation, and microvesicles (27). Nassef et al. (26) compared splenectomy and nonsplenectomy patients and showed that the total leukocyte count, platelet count, lactate dehydrogenase, ferritin, PSV, left MFV, and MCA were all significantly higher in the splenectomy cases. This is in contrast to the results of the present study; there was no significant difference between the BTI males and females in this regard. As Nassef and his colleagues stated, more than one parameter should be applied to assess atherosclerosis in beta-thalassemia intermedia. They also found some evidence for an increased risk of central rather than peripheral ischemia in the patients. Age is another known factor that affects cerebral blood flow velocity (20, 28, 29). According to the results of this study, the average blood flow velocity in the age group of 7-10 years was higher than that in the age group of 16-20 years, which is consistent with the results of some other studies that reported an increase in the blood flow velocity of healthy people under 10 (30, 31). In the present study, there was no significant relationship between gender and blood flow velocity in the cerebral arteries of the thalassemia intermedia patients. A study by Tarumi et al. (20) showed that increasing age is correlated with increased cerebral blood flow pulsatility after midlife. It was also observed that diastolic cerebral blood flow was lower but steady-state cerebral blood flow was higher in women than in men of similar age. Next, age and gender-related variations in cerebral blood flow pulsatility were found to be independently correlated with carotid pulse pressure. In two studies on healthy subjects, increased blood flow velocity in both middle cerebral and basilar arteries was reported in girls aged 4-8 (32) and in girls aged 10-16 (33).One of the most influential factors in increasing the blood flow velocity of cerebral arteries is hemoglobinopathy (34, 35). In an experiment, the effect of hemoglobin level on the cerebral blood flow velocity of the patient was negatively correlated to the blood velocity in the left and right vertebral arteries. Although the present study and a similar study by Ashjazadeh (36) found no evidence of stroke among thalassemia intermedia patients, some studies have reported silent infarctions in thalassemia intermedia patients (10, 37-39). Increased blood flow velocity and severe anemia with the risk of ischemic stroke have also been proved in patients

[DOI: 10.18502/ijpho.v13i1.11633]

with sickle cell anemia (40-43). Most brain strokes in BTI patients are reported as small vessel ischemic strokes, but TCD prediction power in this case is restricted because it only examines large vessels (37, 39). This study showed no significant relationship between blood flow velocity and serum ferritin levels in thalassemia intermedia patients. However, in the patients with sickle cells studied by Santiago (9), a direct correlation was found between ferritin levels and cerebral blood flow velocity. In this study, the platelet count in the thalassemia intermedia patients was under 500,000 in 21 people (60%) and more than 500,000 in 14 people (40%). Following splenectomy, there was no significant relationship between platelet counts and thrombocytosis with intracerebral blood flow velocity. As another finding in this research, the size of the spleen and the history of splenectomy did not correlate to the blood flow velocity. Kawasaki (44) and Karimi (45) reported similar results on major thalassemia patients. However, an increased incidence of asymptomatic stroke in patients with thrombocytosis splenectomy thalassemia intermedia was identified in MRI studies by Musallam (37). In the present study, XmnI polymorphism genes were searched in 20 patients. Of them, 9 were heterozygote, 2 were homozygote, and the remaining 9 had no gene. There were no differences in the cerebral blood flow velocity between the three groups. In BTI patients, the appearance of the XmnI gene as the level of fetal hemoglobin rises is an optimal factor in the disease phenotype and a strong predictor of the disease severity (46, 47). Of the 35 patients, 26 underwent genotypic analyses for beta gene mutation, but 16 lacked the IVS-1-6 gene. The non-gene group show increasing blood flow velocity in the right internal carotid which is consistent with previous studies that disease phenotype improves in the presence of IVS-1-6 gene (48, 49). The limited size of the sample in this study can be a shortcoming for the assessment of the effects of variables. So, there is a need for prospective studies on larger populations. Although transcranial Doppler sonography is a non-invasive method of determining CBFVs, the examiner's skill, high sensitivity, and positive predictive value of TCD for screening arterial stenosis are its major limiting factors. It is recommended that the relationship of cerebral blood flow velocity and brain infarctions, both silent and symptomatic, be verified through the brain MRI of thalassemia intermedia patients. If there is any relationship between cerebral blood flow velocity and cerebrovascular accidents, transcranial Doppler sonography can be used as a primary stroke prevention method in thalassemia intermedia.

Conclusion

This study showed that cerebral blood flow velocities of BTI patients were higher than normal control group. In addition, CBFVs were not affected by factors such as gender, serum ferritin, platelet count, size of spleen and XmnI genotype , however, there was negative correlation between age and hemoglobin level with CBFVs.

Acknowledgment

This study is based on a thesis by Hourshid Sarshad. It was financially supported by the deputy of research affairs at Ahvaz Jundishapur University of Medical Sciences. The authors would like to thank all those who contributed to the study. Ethical approval was obtained from the Ethical Committee of Ahvaz Jundishapur University of Medical Sciences. The ethical certificate number is IR.AJUMS.REC.1394.328.

Conflict of interest

The authors declare no conflict of interest.

References

1. Haddad A, Tyan P, Radwan A, Mallat N, Taher A. beta-Thalassemia

[DOI: 10.18502/ijpho.v13i1.11633]

Intermedia: A Bird's-Eye View. Turk J Hematol 2014;31(1):5-16.
2. Karimi M, Cohan N, De Sanctis V. Thalassemia Intermedia; Folic Acid and Vitamin B12 Supplementation. What We Know and What is Needed? SSU 2017;7(1):57-62.
3. Karimi M, Zarei T, Haghpanah S, Azarkeivan A, Kattamis C, Ladis V, et al. Evaluation of Endocrine Complications in Beta-Thalassemia Intermedia Patients: A Cross Sectional Multi-Center Study. Blood 2018;132:2343.
4. Vasseur C, Domingues-Hamdi E, Ledudal K, Le Corvoisier P, Barau C, Ghaleb B, et al. Red blood cells free α-haemoglobin pool: a biomarker to monitor the β-thalassemia intermedia variability. The ALPHAPOOL study. Br J haematol 2017;179(1):142-153.
5. Maia de Oliveira da Silva JP, Brugnerotto AF, S. Romanello K, KL Teixeira K, Lanaro C, S. Duarte A, et al. Global gene expression reveals an increase of HMGB1 and APEX1 proteins and their involvement in oxidative stress, apoptosis and inflammation pathways among beta-thalassaemia intermedia and major phenotypes. Br J haematol 2019;186(4):608-619.
6. Abboud MR, Maakaron JE, Khoury RA, Tamim HM, Shehab M, Haddad F, et al. Intracranial blood flow velocities in patients with sickle cell disease and beta-thalassemia intermedia. Am J Hematol 2013;88(9):825.
7. Sherwani P, Arora M, Sharawat IK, Panda PK, Ranjan R, Saxena S. Congenital absence of internal carotid artery and complex cerebral vascular abnormalities with thalassemia intermedia: A novel association? Ann Indian Acad Neurol 2020;23(6):817.
8. Ghadiri F, Sahraian MA, Naser Moghadasi A. Beta-thalassemia intermedia with ischemic transients misdiagnosed as multiple sclerosis: A case report. Curr J Neurol 2021:185-187.
9. Santiago RP, Vieira C, Adanho CSA, Santana SS, Guarda CC, Figueiredo CVB, et al. Laboratory and Genetic Biomarkers Associated with Cerebral Blood Flow Velocity in Hemoglobin SC Disease. Dis Markers 2017;2017:6359871.
10. Abdulhai F, Jaffa MA, Elias J, Zakka P, Hotait M, Bou-Fakhredin R, et al. Digital thermography and vascular involvement in β-thalassemia intermedia. Ann Hematol 2021;100(10):2471-2477.
11. El-Masry HMA, El-Shanshory MR, Mahmoud A-EAH, Tolba OA, Elsharkawy HAMA-E. Comparative therapeutic effect of the L-arginine and sildenafil on beta-thalassemia children with increased tricuspid regurgitant jet velocity. J Pediatr Hematol Oncol 2020;5(2):25-29.
12. Iranmanesh F, Hashemi A, Jenabzade A, Akhavanghalibaf M, Dehesh T, Abutorabi-zarchi M. Cerebral hemodynamic in patients with major β-Thalassemia using transcranial Doppler sonography. SSU 2021;11(3):192-197.
13. Kamal AK, Rehman H, Mustafa N, Ahmed B, Jan M, Wadivalla F, et al. Diagnostic TCD for intracranial stenosis in acute stroke patients: experience from a tertiary care stroke center in Karachi, Pakistan. BMC Res Notes 2015;8:341.
14. Bathala L, Mehndiratta MM, Sharma VK. Transcranial doppler: Technique and common findings (Part 1). Ann Indian Acad Neurol 2013;16(2):174-179.
15. Nag DS, Sahu S, Swain A, Kant S. Intracranial pressure monitoring: Gold standard and recent innovations. World J Clin cases 2019;7(13):1535.
16. Kwiatkowski JL, Granger S, Brambilla DJ, Brown RC, Miller ST, Adams RJ, et al. Elevated blood flow velocity in the anterior cerebral artery and stroke risk in sickle cell disease: extended analysis from the STOP trial. Br J Haematol 2006;134(3):333-339.
17. Liu H, Wang J, Xu Z. Prognostic utility of serum 25-hydroxyvitamin D in patients with stroke: A meta-analysis. J Neurol 2020; 267(11):3177-3186.

[DOI: 10.18502/ijpho.v13i1.11633]

18. Ansari S, Baghersalimi A, Azarkeivan A, Nojomi M, Hassanzadeh Rad A. Quality of life in patients with thalassemia major. SSU 2014;4(2):57-63.
19. Alexandrov AV. Cerebrovascular ultrasound in stroke prevention and treatment. 2nd ed. Chichester, West Sussex: Wiley-Blackwell 2011; 280 -285
20. Tarumi T, Zhang R. Cerebral blood flow in normal aging adults: cardiovascular determinants, clinical implications, and aerobic fitness. J Neurochem 2018;144(5):595-608.
21. Smith KJ, Ainslie PN. Regulation of cerebral blood flow and metabolism during exercise. Exp Physiol 2017;102(11):1356-1371.
22. Montrief T, Alerhand S, Jewell C, Scott J. Incorporation of transcranial doppler into the ED for the neurocritical care patient. The AJEM 2019;37(6):1144-1152.
23. Long SS, Nyquist A-C. CHAPTER 288. Laboratory Manifestations of Infectious Diseases. In: Long SS, editor. Principles and Practice of Pediatr Infect Dis J (Third Edition). Edinburgh: W.B. Saunders; 2008; 1368-1381.
24. Ahmad Ibrahim O, Ahmad AB, Nigm DA, Hussien AN, Mohammad Ibrahim WH. Subclinical atherosclerotic predictive value of inflammatory markers in thalassemia intermedia patients. Expert Rev Hematol 2021;14(7):669-677.
25. Fonseca AC, Silva DP, Infante J, Ferro JM. Cerebrovascular Complications of Anemia. Curr Neurol Neurosci Rep 2021;21(10):1-11.
26. Nassef S, El Shenoufy M, Rawi R, El Demerdash D, Hassan M, Mustafa H, et al. Assessment of Atherosclerosis in Peripheral and Central Circulation in Adult β Thalassemia Intermedia Patients by Color Doppler Ultrasound: Egyptian Experience. J Vasc Res 2020;57(4):206-212.
27. Muhammad Isa M, Thayeb A, Yani A, Hutagalung M. Post total splenectomy outcome in thalassemia patients. BMJ 2019;8:947-949.
28. Alwatban MR, Aaron SE, Kaufman CS, Barnes JN, Brassard P, Ward JL, et al. Effects of age and sex on middle cerebral artery blood velocity and flow pulsatility index across the adult lifespan. J Appl Physiol 2021;130(6):1675-1683.
29. MacDonald ME, Williams RJ, Rajashekar D, Stafford RB, Hanganu A, Sun H, et al. Age-related differences in cerebral blood flow and cortical thickness with an application to age prediction. Neurobiol Aging 2020;95:131-142.
30. Chen JJ, Rosas HD, Salat DH. Age-associated reductions in cerebral blood flow are independent from regional atrophy. Neuroimage 2011;55(2):468-478.
31. Demirkaya S, Uluc K, Bek S, Vural O. Normal blood flow velocities of basal cerebral arteries decrease with advancing age: a transcranial Doppler sonography study. Tohoku J Exp Med 2008;214(2):145-149.
32. Tontisirin N, Muangman SL, Suz P, Pihoker C, Fisk D, Moore A, et al. Early childhood gender differences in anterior and posterior cerebral blood flow velocity and autoregulation. Pediatrics 2007;119(3):e610-615.
33. Vavilala MS, Kincaid MS, Muangman SL, Suz P, Rozet I, Lam AM. Gender differences in cerebral blood flow velocity and autoregulation between the anterior and posterior circulations in healthy children. Pediatr Res 2005;58(3):574-578.
34. Zamani S, Borhan Haghighi A, Haghpanah S, Karimi M, Bordbar MR. Transcranial Doppler screening in 50 patients with sickle cell hemoglobinopathies in Iran. Pediatr. Hematol oncol J 2017;39(7):506-512.
35. Aydin S, Yeral H, Oktay G, Çoban Y, Akbaş Y, Köker A, et al. Associations Between Transcranial Doppler Flow Velocities, Laboratory Parameters, and Clinical Features in Turkish Children With Sickle Cell Disease. Pediatr Hematol Oncol J 2021;43(7):249-253.

[DOI: 10.18502/ijpho.v13i1.11633]

36. Ashjazadeh N, Emami S, Petramfar P, Yaghoubi E, Karimi M. Intracranial Blood Flow Velocity in Patients with beta-Thalassemia Intermedia Using Transcranial Doppler Sonography: A Case-Control Study. Anemia 2012;2012:798296.
37. Musallam KM, Taher AT, Karimi M, Rachmilewitz EA. Cerebral infarction in beta-thalassemia intermedia: breaking the silence. Thromb Res 2012;130(5):695-702.
38. Musallam KM, Beydoun A, Hourani R, Nasreddine W, Raad R, Koussa S, et al. Brain magnetic resonance angiography in splenectomized adults with beta-thalassemia intermedia. Eur J Haematol 2011;87(6):539-546.
39. Teli A, Economou M, Rudolf J, Tzovaras F, Gourtsa V, Kondou A, et al. Subclinical central nervous system involvement and thrombophilic status in young thalassemia intermedia patients of Greek origin. Blood Coagul Fibrinolysis 2012;23(3):195-202.
40. Adams RJ. Prevention of stroke in sickle cell anemia. J Law Med Ethics 2014;42(2):135-138.
41. Hyacinth HI, Adams RJ, Voeks JH, Hibbert JM, Gee BE. Frequent red cell transfusions reduced vascular endothelial activation and thrombogenicity in children with sickle cell anemia and high stroke risk. Am J Hematol 2014;89(1):47-51.
42. Mynarek M, Bettoni da Cunha Riehm C, Brinkmann F, Weissenborn K, Tell-Luersen M, Heuft HG, et al. Normalized transcranial Doppler velocities, stroke prevention and improved pulmonary function after stem cell transplantation in children with sickle cell anemia. Klin Padiatr 2013;225(3):127-132.
43. Al Hashmi A, Aaron S. Acute Ischemic Stroke in Sickle Cell Disease Challenges for Thrombolysis. Dubai Med J 2020;3(44):173-177.
44. Kanavaki A, Kattamis A, Delaporta P, Papassotiriou I, Spengos K. Evaluation of Intracranial Cerebral Blood Flow Velocities in Splenectomised and Non-Splenectomised Patients with beta-Thalassemia Intermedia Using Transcranial Doppler Sonography. In Vivo 2015;29(4):501-504.
45. Karimi M, Haghpanah S, Ashjazadeh N, Shariat A, Nazeri M, Emami S, et al. Cerebral artery velocity determined by transcranial doppler ultrasonography in patients with beta-thalassemia intermedia compared to beta-thalassemia major. Clin Appl Thromb Hemost 2013;19(4):367-373.
46. Bashir S, Mahmood S, Mohsin S, Tabassum I, Ghafoor M, Sajjad O. Modulatory effect of single nucleotide polymorphism in Xmn1, BCL11A and HBS1L-MYB loci on foetal haemoglobin levels in β-thalassemia major and Intermedia patients. JPMA 2021;71(5):1394-1398.
47. Hariharan P, Gorivale M, Sawant P, Mehta P, Nadkarni A. Significance of genetic modifiers of hemoglobinopathies leading towards precision medicine. Sci Rep 2021;11(1):1-10.
48. Asadov C, Alimirzoeva Z, Mammadova T, Aliyeva G, Gafarova S, Mammadov J. beta-Thalassemia intermedia: a comprehensive overview and novel approaches. Int J Hematol 2018;108(1):5-21.
49. Chen B, Huang P, Yi S, Chen Q, Tang Y, Zhang Q, et al. First Detection of a Splice Site beta-Thalassemia Mutation, IVS-I-6 (T > C) (HBB: c.92 + 6T > C) in a Chinese Family. Hemoglobin 2015; 39(3):207-208.

[DOI: 10.18502/ijpho.v13i1.11633]