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Hematological and biochemical evaluation of β-thalassemia major (βTM) patients in Gaza Strip: A cross-sectional study

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ABSTRACT

Objectives: In Gaza Strip, Palestine, β-thalassemia is a major public health problem where more than 300 β-thalassemia major (βTM) patients are currently being managed at governmental hospitals. We set up to evaluate the hematological and biochemical aspects of our βTM patients at the Gaza European hospital and their correlation with iron overload.

Methods: Our study included 65 transfusion-dependent βTM, as well as 37 apparently healthy subjects as control group. The hematological and biochemical evaluations included complete blood count, coagulation profile liver and kidney function tests, fasting blood sugar, lipid profile, and serum ferritin.

Results: Deteriorated hematological and biochemical statuses were reported in both males and females of βTM patients as compared to the control group. Statistical comparisons showed no significant differences between males and females βTM patients in all parameters except for total cholesterol. The results concerning the splenectomized versus non-splenectomized patients revealed significantly higher values in splenectomized patients for white blood cell (WBC), platelet, aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, albumin, total protein, cholesterol, and potassium concentration compared to the non-splenectomized patients. Patients infected with hepatitis C virus and/or hepatitis B virus showed significant decrease in WBC count as compared to infection free patients, while for serum urea and creatinine, the virally infected βTM patients revealed significantly higher values compared to infection free patients.

Conclusion: This study justified the necessity for strengthening the efforts for regular evaluation and follow-up of the βTM patients which could be used to improve or modify the management protocols and thus ameliorating their deteriorated hematological and biochemical status.

Keywords: Chelation therapy, iron overload, splenectomy, transfusion, β-thalassemia major

Introduction

The thalassemias are the global most common human monogenic disease; they are a family of hereditary anemias that occur because of mutations in the hemoglobin (Hb) gene clusters that impair the rate of synthesis of one or more of the globin chain subunits of the Hbs tetramer.[1] Thalassemias have been encountered practically in every racial group and geographic location of the world; however, they are most common in the broad belt extended from sub-Saharan Africa, through the Mediterranean region, Middle East and Arabian Peninsula to the Indian subcontinent, and India and South-eastern Asia.[2] The high frequencies of thalassemia are found in those areas historically afflicted with endemic malaria.[3,4]

Thalassemias are principally classified according to the individual globin gene or genes affected (i.e., α-, β-, γ-, δ-, δβ-, and γδβ--thalassemias) with α- and β-thalassemias being the most commonly encountered types worldwide.[5] Furthermore, the severity of thalassemia diseases is usually subclassified according to whether the mutation results in totally absent or only partially reduced globin chain synthesis. Totally absent globin synthesis is designated with an "^{o}" superscript, while partially reduced synthesis is designated with "$^{+}$" superscript.[6] Nevertheless, the β-thalassemia is the most important type of thalassemias, it is so common, widely distributed and result in severe anemia in the homozygous and compound heterozygous states.[6,7] The basic molecular defect in β-thalassemia results in either absence (β^{o}) or reduced (β^{+}) beta-chain production;

however, α-chain synthesis proceeds at a normal rate.[8] The hallmark of the β-thalassemia disease is the imbalanced globin chain synthesis, in which α-chain synthesis proceeds at a normal rate, and hence, there is an excess of α-chain in the erythrocytes which are unstable and precipitate in the bone marrow red cell precursors, giving rise to a large intracellular inclusions and leading to ineffective erythropoiesis and a group of subsequent pathophysiological mechanisms.[4,8] As a consequence of reduced β-chain production, the quantity of the produced adult Hb (α_2 β_2) is negatively affected as well.[3,9] More than 200 different genetic variants have been identified in the β-globin (*HBB*) gene and confirmed as responsible mutations for the development of the varied severities of the β-thalassemia disease. Most types of β-thalassemia are due to point mutations, and large deletion mutations are found in rare cases.[3,10]

The severity of clinical syndromes likely depends on the type of mutation in the *HBB* gene, and previous studies have revealed relationships between hematological/clinical phenotype and the type of β-thalassemia mutation.[11,12] In Gaza Strip, Palestine, β-thalassemia is a major public health problem where the screening studies estimated the prevalence of β-thalassemia gene carriers between 2.6% and 4.3%, with some significant differences in different locations of the Gaza Strip. The relatively high consanguinity rate, as well as the remarkable prevalence of the defects in *HBB* gene, resulted in more than 300 βTM patients currently being managed through blood transfusions and iron chelation at the government hospitals of the Gaza Strip.[13-15] Therefore**,** we designed the present work to evaluate hematological and biochemical aspects of our βTM patients at the European Gaza Hospital and their correlation with iron overload. The outcomes of our evaluation study could be used to improve or modify the management protocols, such as blood transfusions and iron chelation, for our patients in Gaza Strip.

Materials and Methods

This work was performed according to the cross-sectional descriptive study design and targeted toward the βTM patients who are currently being transfused and managed for the clinical symptoms and manifestations of the disease at the European Gaza Hospital, Gaza Strip. Our study included 65 transfusion-dependent βTM patients (32 males and 33 females) aged 12–42 years old, as well as 37 apparently healthy subjects (18 males and 19 females) of the same age range as control group. Cases and controls blood samples were collected from January to April 2018. The present study was performed in accordance with the ethical standards laid down in the 1964 and 1975 Declarations of Helsinki including the latest modifications of 2008, and it was approved by the Helsinki Ethical Committee of the Palestinian health research with approval number PHRC/HC/298/17. Informed written consent was obtained from the subjects (patients and controls) or their parents in case of children for enrollment in the present study.

About 5 ml of venous blood were collected from all patients and divided equally (2.5 ml) into K_3-EDTA tubes and serum tubes. The blood in the EDTA tubes was used to perform complete blood count using a Cell Dyne 1700 electronic counter (Sequoia-Turner Corporation, California, USA), as well as to separate the plasma for the coagulation profile that included prothrombin time-international normalized ratio and prothrombin percent activity determination. While the blood in serum tubes was used to determine the followings parameters using commercially available kits : aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), serum bilirubin, urea, creatinine, uric acid, total protein, albumin, globulins, fasting blood sugar, lipid profile (total cholesterol and triglycerides), serum ferritin, serum potassium, serum phosphorus, and serum calcium. It is worthwhile mentioning that all blood withdrawals of the β-thalassemic patients were performed just before the scheduled blood transfusion regimen.

The data of the study were statistically analyzed using SPSS program (version 20, IBM Corporation, Somers, NY). Comparisons between the mean values of the different parameters were performed using the independent *t*-test analysis and one-way analysis of variance. Any two-tailed $P < 0.05$ was considered statistically significant result.

Results

This work was performed at European Gaza Hospital where more than one-third of the Palestinian βTM patients in the Gaza Strip are currently being transfused and managed for the clinical symptoms and manifestations of the disease. In the current work, 65 transfusion-dependent βTM patients including 32 males, mean age of 20.3 years and 33 females mean age of 20.8 years were included for the hematological and biochemical evaluations. In this result section, we presented the evaluation results in three comparisons: Males and females βTM patients versus the corresponding control group, splenectomized versus non-splenectomized, and finally, βTM patient with viral infections (hepatitis C [HCV] or hepatitis B [HBV]) versus infection-free patients.

Table 1 shows the hematological and biochemical evaluation in βTM male and female patients as compared to the corresponding control group. Deteriorated hematological and biochemical statuses were reported in both males and females of our βTM patients. Severe anemia (7.4 ± 0.8 g/dL and 7.36 ± 1.57 g/dL) and severe iron overload (7162.4 ± 3297.3 and 7068.7 ± 3826.0 ng/ml) were reported in the males and females βTM patients, respectively, which are associated with significantly worsened liver (AST 83.3 ± 38.6 and 74.0 ± 34.4 IU/L; ALT 82.6 ± 40.0 and 74.5 ± 41.5 IU/L; and ALP 421.3 ± 169.7 and 352.1 ± 150.9 U/L, respectively, in males and females, $P < 0.001$) and kidney (urea 29.7 ± 18.8 and 25.7 ± 9.2 mg/dL; serum creatinine 0.59 ± 0.3 and 0.48 ± 0.18 mg/dL; and uric acid 6.8 ± 2.1 and 6.0 ± 1.7 mg/dL, respectively, in males and females,

Table 1: Hematological and biochemical evaluation in β-thalassemic male and female patients as compared to control group

Parameters	Males				Females			
	Cases	Controls	Independent samples test		Cases	Controls	Independent samples test	
	N=32	*N*=18	*t*	*P*	*N*=33	*N*=19	*t*	*P*
Age (years)	20.3±5.0	22.6±3.8	−1.685	0.098	20.8±6.7	22.5±2.7	−1.099	0.277
Blood counts								
WBC×10^9/L	39.4±26.3	7.4±1.5	5.149	<0.001	40.3±24.7	7.1±1.2	5.834	<0.001
Hb (g/dL)	7.4±0.8	13.9±0.7	−29.131	<0.001	7.36±1.57	12.6±1.2	−17.105	<0.001
PLT×10^9/L	603.6±208.6	261.0±37.2	6.875	<0.001	571.2±253.8	270.1±41.6	5.112	<0.001
Ferritin (ng/mL)	7162.4±3297.3	84.7±28.1	9.066	<0.001	7068.7±3826.0	45.1±39.9	7.969	<0.001
Liver function tests								<0.001
AST (IU/L)	83.3±38.6	23.3±7.3	6.502	<0.001	74.0±34.4	24.1±5.0	6.270	<0.001
ALT (IU/L)	82.6±40.0	24.2±7.6	6.177	<0.001	74.5±41.5	24.4±6.5	5.196	<0.001
ALP (U/L)	421.3±169.7	194.8±109.5	5.085	<0.001	352.1±150.9	189.1±39.2	5.304	<0.001
Total bilirubin (mg/dL)	2.1±0.8	0.5±0.2	7.819	<0.001	1.9±1.1	0.4±0.2	6.121	<0.001
Direct bilirubin (mg/dL)	0.57±0.22	0.2±0.1	7.917	<0.001	0.52±0.27	0.2±0.1	6.374	<0.001
Albumin (g/dL)	3.9±0.6	3.8±0.3	0.291	0.773	3.7±0.8	3.8±0.4	0.304	0.762
Total protein (g/dL)	8.0±0.6	6.7±0.6	7.270	<0.001	7.6±1.6	7.2±0.7	2.629	0.011
Kidney function test and electrolytes								
Urea (mg/dL)	29.7±18.8	19.3±6.2	2.270	0.028	25.7±9.2	17.7±5.9	3.391	0.001
Serum creatinine (mg/dL)	0.59±0.3	0.3±0.2	3.510	0.01	0.48±0.18	0.3±0.2	3.492	0.001
Uric acid (mg/dL)	6.8±2.1	3.5±0.6	6.158	<0.001	6.0±1.7	3.3±0.7	6.526	<0.001
Na (mEq/L)	138.1±2.3	138.0±4.2	0.008	0.994	139.7±3.0	138.5±2.6	1.460	0.151
K (mEq/L)	4.7±0.5	4.0±0.3	5.846	<0.001	4.7±0.6	4.0±0.2	5.120	<0.001
Ca (mg/dL)	3.1±3.8	9.2±0.6	−6.080	<0.001	4.4±4.3	9.3±0.7	−4.985	<0.001
Sugar and lipid								
Fasting glucose (mg/dL)	112.5±81.8	103.6±11.3	0.455	0.651	91.6±38.1	94.2±11.3	0.291	0.772
Cholesterol (mg/dL)	106.8±20.6	142.7±20.4	−5.923	<0.001	121.8±23.7	145.8±19.4	−3.737	<0.001
Triglycerides (mg/dL)	188.3±105.1	81.3±24.0	4.239	<0.001	206.4±61.2	70.5±14.1	9.489	<0.001
Coagulation profile								
Prothrombin time (s)	14.6±1.1	13.2±1.0	4.090	<0.001	14.3±1.3	12.9±0.7	4.591	<0.001
International normalized ratio	1.2±0.1	1.0±0.1	6.859	<0.001	1.2±0.1	1.0±0.1	5.111	<0.001
Prothrombin % activity	69.9±12.3	90.7±6.7	−6.614	<0.001	74.5±13.2	87.0±6.9	−3.843	<0.001

P<0.05 is statistically significant. AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline phosphatase, WBC: White blood cell, PLT: Platelet, Hb: Hemoglobin

$P < 0.05$) function tests. Statistical comparisons showed no significant differences ($P > 0.05$) between males and females βTM patients in all parameters except for total cholesterol where data from females (121.8 ± 23.7 mg/dl) revealed higher mean value compared to males (106.8 ± 20.6 g/dl) with $P < 0.05$. However, both male and female cholesterol means were within the normal reference range.

Analyses using the Pearson correlation coefficient revealed a significant correlation between serum ferritin as a biomarker of iron overload and liver function tests in terms of AST and ALT (Pearson correlation [r] of 0.462 and 0.452, respectively).

The results concerning the splenectomized versus non-splenectomized βTM patients are presented in Table 2. Significantly higher abnormal values ($P < 0.05$) were reported in splenectomized patients for white blood cell (WBC) (44.0 ± 23.6 vs. 6.1 ± 2.4 × 10^9/L), platelet (PLT) (630.9 ± 205.3 vs. 233.4 ± 53.2 × 10^9/L), AST (81.9 ± 37.0 vs. 52.3 ± 18.4 IU/L), ALT (82.0 ± 41.1 vs. 50.4 ± 23.6 IU/L), ALP (397.6 ± 164.8 vs. 291.3 ± 116.8 IU/L), albumin (3.9 ± 0.7 vs. 3.3 ± 1.1 g/dL), total protein (7.9 ± 1.2 vs. 7.1 ± 1.0 g/dL), cholesterol (116.8 ± 23.0 vs. 93.7 ± 13.5 mg/dL), and potassium concentration (4.8 ± 0.5 vs. 4.3 ± 0.2 mEq/L) compared to the non-splenectomized patients. While patients infected with HCV or HBV showed significant decrease in WBC count (29.6 ± 17.2 vs. 42.5 ± 26.5 × 10^9/L) as compared to infection free patients, however, for serum urea (35.6 ± 28.0 vs. 25.7 ± 8.2 mg/dL) and creatinine (0.72 ± 0.41 vs. 0.49 ± 0.16 mg/dL), the virally infected βTM patients revealed significantly higher values [Table 3].

Table 2: Hematological and biochemical evaluation in splenectomized versus none splenectomized β-thalassemic patients

Parameters	Controln *N*=37	Splenectomized *N*=58	None splenectomized *N*=7	*F*	AVOVA *P* value	*Post hoc* test *P* value		
						Splenectomized versus none splenectomized	Splenectomized versus control	None splenectomized versus control
Age (years)	22.5±3.2	20.5±5.8	21.0±6.5	1.895	0.156	0.790	0.055	0.464
Blood counts								
WBC×10^9/L	7.2±1.4	44.0±23.6	6.1±2.4	52.896	<0.001	<0.001	<0.001	0.880
Hb (g/dL)	13.2±1.2	7.4±1.3	7.0±0.7	399.679	<0.001	0.249	<0.001	<0.001
PLT×10^9/L	265.6±39.2	630.9±205.3	233.4±53.2	68.713	<0.001	<0.001	<0.001	0.621
Ferritin ng/mL	64.3±39.6	7283.8±3613.0	5746.0±2745.1	74.513	<0.001	0.176	<0.001	<0.001
Liver function tests					<0.001			
AST (IU/L)	23.7±6.1	81.9±37.0	52.3±18.4	46.702	<0.001	0.011	<0.001	0.017
ALT (IU/L)	24.3±7.0	82.0±41.1	50.4±23.6	37.029	<0.001	0.015	<0.001	0.05
ALP (IU/L)	191.9±80.2	397.6±164.8	291.3±116.8	29.420	<0.001	0.034	<0.001	0.070
Total bilirubin (mg/dL)	0.5±0.2	2.0±1.0	1.8±0.8	47.330	<0.001	0.486	<0.001	<0.001
Direct bilirubin (mg/dL)	0.2±0.1	0.55±0.25	0.53±0.24	50.260	<0.001	0.691	<0.001	<0.001
Albumin (g/dL)	3.8±0.3	3.9±0.7	3.3±1.1	5.279	0.007	0.002	0.269	0.012
Total protein (g/dL)	7.0±0.7	7.9±1.2	7.1±1.0	30.103	<0.001	<0.001	<0.001	0.746
Kidney function test and electrolytes								
Urea (mg/dL)	18.5±6.0	28.7±15.3	20.0±6.1	8.227	<0.001	0.08	<0.001	0.768
Serum creatinine (mg/dL)	0.3±0.2	0.55±0.26	0.44±0.1	12.485	<0.001	0.279	<0.001	0.136
Uric acid (mg/dL)	3.4±0.7	6.3±1.9	7.3±2.1	41.444	<0.001	0.099	<0.001	<0.001
Na (mEq/L)	138.3±3.4	138.9±2.8	138.6±2.3	0.505	0.605	0.780	0.319	0.810
K (mEq/L)	4.0±0.3	4.8±0.5	4.3±0.2	37.122	<0.001	0.004	<0.001	0.140
Ca (mg/dL)	9.2±0.6	3.6±3.9	5.1±4.9	34.118	<0.001	0.245	<0.001	0.003
Sugar and lipid								
Fasting glucose (mg/dL)	98.8±12.1	104.1±67.7	85.4±7.6	0.451	0.639	0.371	0.630	0.533
Cholesterol (mg/dL)	144.3±19.7	116.8±23.0	93.7±13.5	26.714	<0.001	0.008	<0.001	<0.001
Triglycerides (mg/dL)	75.8±20.0	199.9±88.4	176.3±61.8	36.137	<0.001	0.398	<0.001	0.002
Coagulation profile								
Prothrombin time (s)	13.0±0.8	14.4±1.2	14.8±1.1	19.658	<0.001	0.299	<0.001	<0.001
International normalized ratio	1.0±0.1	1.2±0.1	1.2±0.1	36.083	<0.001	0.354	<0.001	<0.001
Prothrombin % activity	88.8±7.0	72.3±13.2	71.1±10.2	26.149	<0.001	0.790	<0.001	<0.001

P<0.05 is statistically significant. AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline phosphatase, WBC: White blood cell, PLT: Platelet, Hb: Hemoglobin, ANOVA: Analysis of variance

Discussion

β-thalassemia is a major health problem in Palestine, with a considerable prevalence of the *HHB* gene thalassemic variants among the Palestinian population of the Gaza Strip. The previous studies conducted in the Gaza Strip showed a significant prevalence (2.6–4.3%) of β-thalassemia gene in this population where more than 300 β-thalassemia patients are currently being managed through blood transfusions and iron chelation at the government hospitals of the Gaza Strip.[14,16,17] Although of the tremendous success of the national obligatory premarital testing program for β-thalassemia in Gaza Strip [14] however, the approaches towards providing the appropriate medical care for the β-thalassemia patients should be continued and improved for promoting the wellness and health of the patients. In this cross-sectional study, we assessed the hematological and biochemical aspects of our βTM patients at the European Gaza Hospital and their correlation with iron overload that could highlight the necessity for suggesting the future management approaches and protocols.

Our βTM patients from both sexes are suffering from worsened clinical situation revealed from their hematological and biochemical test parameters, which are mainly secondary to their severely anemic status and the overloaded iron due to chronic blood transfusion regimen associated with inadequate chelation therapy. The Hb levels of the male and female βTM patients were decreased to 53% and 58%, respectively, of the normal levels reported in their counterparts of the control group. Despite the chelation therapy provided to our male

Table 3: Hematological and biochemical evaluation in virally infected versus virally free β-thalassemic patients

Parameters	Control *N*=37	Virally infected *N*=13	Virally free *N*=52	*F*	ANOVA *P* value	*Post-hoc* test *P* value		
						Virally infected versus virally free	Virally infected versus control	Virally free versus control
Age (years)	22.5±3.2	24.7±7.3	19.5±5.0	8.289	<0.001	0.001	0.167	0.004
Blood counts								
WBC×10^9/L	7.2±1.4	29.6±17.2	42.5±26.5	33.837	<0.001	0.039	0.001	<0.001
Hb (g/dL)	13.2±1.2	7.4±0.8	7.3±1.3	393.822	<0.001	0.830	<0.001	<0.001
PLT×10^9/L	265.6±39.2	534.9±231.6	600.8±231.3	35.913	<0.001	0.256	<0.001	<0.001
Ferritin (ng/mL)	64.3±39.6	7924.6±4368.0	6909.4±3320.6	73.870	<0.001	0.250	<0.001	<0.001
Liver function tests								
AST (IU/L)	23.7±6.1	81.2±51.2	78.0±32.4	40.701	<0.001	0.724	<0.001	<0.001
ALT (IU/L)	24.3±7.0	78.4±43.7	78.6±40.3	31.972	<0.001	0.983	<0.001	<0.001
ALP (U/L)	191.9±80.2	411.2±148.3	379.9±167.2	26.145	<0.001	0.569	<0.001	<0.001
Total bilirubin (mg/dL)	0.5±0.2	1.8±0.9	2.0±0.9	48.810	<0.001	0.160	<0.001	<0.001
Direct bilirubin (mg/dL)	0.2±0.1	0.53±0.23	0.55±0.25	50.383	<0.001	0.597	<0.001	<0.001
Albumin (g/dL)	3.8±0.3	3.6±0.4	3.8±0.8	2.009	0.14	0.053	0.226	0.323
Total protein (g/dL)	7.0±0.7	8.1±0.7	7.7±1.3	21.989	<0.001	0.209	<0.001	<0.001
Kidney function test and electrolytes								
Urea (mg/dL)	18.5±6.0	35.6±28.0	25.7±8.2	10.459	<0.001	0.009	<0.001	0.007
Serum creatinine (mg/dL)	0.3±0.2	0.72±0.41	0.49±0.16	17.578	<0.001	0.003	<0.001	<0.001
Uric acid (mg/dL)	3.4±0.7	6.3±1.7	6.4±2.0	38.953	<0.001	0.925	<0.001	<0.001
Na (mEq/L)	138.3±3.4	139.2±3.2	138.8±2.7	0.579	0.562	0.636	0.328	0.434
K (mEq/L)	4.0±0.3	4.7±0.6	4.7±0.5	30.065	<0.001	0.855	<0.001	<0.001
Ca (mg/dL)	9.2±0.6	3.5±3.9	3.8±4.2	33.039	<0.001	0.784	<0.001	<0.001
Sugar and lipid								
Fasting glucose (mg/dL)	98.8±12.1	124.4±92.9	96.3±54.3	1.605	0.206	0.081	0.124	0.825
Cholesterol (mg/dL)	144.3±19.7	112.5±27.5	114.7±22.4	21.560	<0.001	0.741	<0.001	<0.001
Triglycerides (mg/dL)	75.8±20.0	226.4±125.6	190.0±72.2	38.017	<0.001	0.092	<0.001	<0.001
Coagulation profile								
Prothrombin time (s)	13.0±0.8	14.7±1.3	14.4±1.2	19.483	<0.001	0.362	<0.001	<0.001
International normalized ratio	1.0±0.1	1.2±0.1	1.2±0.1	35.341	<0.001	0.952	<0.001	<0.001
Prothrombin % activity	88.8±7.0	70.3±12.7	72.7±13.0	26.446	<0.001	0.500	<0.001	<0.001

P<0.05 is statistically significant. AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline phosphatase, WBC: White blood cell, PLT: Platelet, Hb: Hemoglobin, ANOVA: Analysis of variance

and female βTM patients, the overloaded iron increased dramatically and reached, respectively, 84- and 157-fold of the normal levels seen in their counterparts of the control group. The same trend was noticed in the indicator of liver and kidney function tests where the βTM patients showed increased level reaching in some tests 3.5-fold of the values of the control group.

Excess iron is extremely toxic to all body tissues, leading to significant morbidity and mortality among β-thalassemic patients as well as other iron-overload conditions where it causes serious and irreversible biological damage, such as cirrhosis, liver fibrosis, heart disease, and endocrine abnormalities.[18] Although cardiomyopathy is the leading cause of death in most of the transfusion-dependent βTM patients,[3] liver dysfunction resulting from severe iron overload and/or chronic viral infections is becoming a more important cause of mortality.[19-21] Nevertheless, the new trends and development in chelation therapy routes, monotherapy, or combined therapy, together with appropriate monitoring and continuous health education programs can protect or ameliorate such consequences and improve thalassemia patients survival and their quality of life.[22-25] Previous works have emphasized on the deleterious effects of iron overload on the liver function, indicated by the liver enzymes activities and when serum ferritin level exceeds 1000 ng/ml and the number of transfusions are more than 30, derangement in liver enzymes starts occurring in the transfusion-dependent β-thalassemia patients.[26]

The spleen is a substantial organ in the modulation of immunity, inflammation, and thrombosis. Systemic consequences and dysfunctions are expected in asplenic individuals.[27] In this cohort and to improve the clinical condition of our βTM

patients, splenectomy was performed in almost 90% of the 65 transfusion-dependent thalassemic cases due to hypersplenism (cytopenia caused by splenomegaly) or huge splenomegaly that causing pain and discomfort for the patient.[28] Compared to non-splenectomized βTM patients, our splenectomized βTM patients showed significantly higher abnormal values that reflect more deterioration in their hematological and biochemical indicators. For example, the WBC increased to about 7-fold, PLT to about 3-fold, and liver function indicators to about 2-fold. In general, considerable fluctuations have been reported in blood cell counts post-splenectomy. These include leukocytosis with a tendency toward lymphocytosis and monocytosis and thrombocytosis.[29,30] Splenectomized patients were found to have a greater degree of susceptibility to infections and increased risk of septic complications associated with a high mortality rate than non-splenectomized patients,[31-34] which could explain the dramatic increases in the WBC count in our splenectomized βTM patients.

Regarding the thrombocytosis in our splenectomized patients, the thrombocytosis persists indefinitely after splenectomy. This usually appears to be a consequence of continuing anemia with a hyperplastic marrow.[35,36] Although a reactive thrombocytosis is not usually associated with thromboembolic problems, the high PLT count may have contributed to the serious and sometimes fatal episodes of pulmonary embolism and deep vein thrombosis that has occurred following splenectomy in some of our deceased βTM patients. Therefore, anti-PLT therapy is mandatory given to our splenectomized patients to avoid thromboembolic problems. Splenectomy and transfusion naivety have been considered as an important risk factor for hypercoagulability and thromboembolic events in thalassemic patients.[3,37] The study of Ammara *et al*., 2014, about the long-term follow-up after splenectomy in thalassemia patients revealed thrombocytosis and the risk of thromboembolism. In addition, splenectomized patients with thalassemia intermedia were shown to have high PLTs count and at high risk to develop thromboembolic events than their non-splenectomized counterparts.[37] Moreover, splenectomy has been considered as an important risk factor for infections;[38] therefore, proper pre-operative vaccination can reduce the risk of overwhelming post-splenectomy infections.[39]

Our splenectomized patients showed higher but not significantly different Hb level compared to the non-splenectomized patients. Splenectomy was found to improve anemia but does not reduce iron burden or the requirement for blood transfusion in the transfusion-dependent disorders.[39,40]

βTM patients are most likely to develop several transfusion-related infections and complications included, but not limited to, HBV and HCV.[18] Of our βTM patients at the European Gaza Hospital, 20% acquired HBV or HCV infection from blood transfusion. It is worthwhile to mention that the mean age of the βTM patients with HBV or HCV is significantly higher than the viral free βTM patients. The older βTM patients who acquired the viral infection may receive the transfusion regimen when routine screening of donated blood is inappropriate due to insufficient laboratory techniques, procedures, or instruments. Nevertheless, the younger βTM patients representing 80% of our cohort are infection free which emphasizing on the improvement of blood donor screening programs with effective screening techniques that considered essential to prevent spreading of serious viral infection among βTM patients. Therefore, it is very much imperative for the health-care providers and blood banks to implement stringent serological pretransfusion screening of blood which will bring down the occurrence of transfusion-transmitted viral infections in βTM patients.[41]

Conclusion

The present cross-sectional study revealed deteriorated hematological and biochemical statuses of our βTM patients from both sexes. The presences of βTM patients infected with HCV or HBV advocate for implementing stringent serological pretransfusion screening of blood to bring down the occurrence of transfusion-transmitted viral infections in βTM patients. This study justified the necessity for strengthen the efforts for regular evaluation and follow-up of the βTM patients with the stress on iron chelation therapy uses, which could be strongly advisable to improve or modify the management protocols and thus improving their clinical picture, as stem cell transplantation is not feasible in our country.

References

1. Cappellini MD, Porter JB, Viprakasit V, Taher AT. A paradigm shift on beta-thalassaemia treatment: How will we manage this old disease with new therapies? Blood Rev 2018;32:300-11.
2. Weatherall DJ. The evolving spectrum of the epidemiology of thalassemia. Hematol Oncol Clin North Am 2018;32:165-75.
3. Taher AT, Weatherall DJ, Cappellini MD. Thalassaemia. Lancet 2018;391:155-67.
4. Weatherall DJ. The role of the inherited disorders of hemoglobin, the first "molecular diseases," in the future of human genetics. Annu Rev Genomics Hum Genet 2013;14:1-24.
5. Rooks H, Clark B, Best S, Rushton P, Oakley M, Thein OS, *et al*. A novel 506kb deletion causing epsilongammadeltabeta thalassemia. Blood Cells Mol Dis 2012;49:121-7.
6. Mettananda S, Higgs DR. Molecular basis and genetic modifiers of thalassemia. Hematol Oncol Clin North Am 2018;32:177-91.
7. Viprakasit V, Ekwattanakit S. Clinical classification, screening and diagnosis for thalassemia. Hematol Oncol Clin North Am 2018;32:193-211.
8. Thein SL. Molecular basis of β thalassemia and potential therapeutic targets. Blood Cells Mol Dis 2018;70:54-65.
9. Oikonomidou PR, Rivella S. What can we learn from ineffective erythropoiesis in thalassemia? Blood Rev 2018;32:130-43.
10. Giardine B, van Baal S, Kaimakis P, Riemer C, Miller W, Samara M, *et al.* HbVar database of human hemoglobin variants and thalassemia mutations: 2007 update. Hum Mutat 2007;28:206.
11. Danjou F, Anni F, Galanello R. Beta-thalassemia: From genotype to phenotype. Haematologica 2011;96:1573-5.

12. Asadov C, Abdulalimov E, Mammadova T, Gafarova S, Guliyeva Y, Aliyeva G, *et al.* Genotype-phenotype correlations of β-thalassemia mutations in an Azerbaijani population. Turk J Haematol 2017;34:258-63.
13. Sirdah M, Bilto YY, el Jabour S, Najjar K. Screening secondary school students in the gaza strip for beta-thalassaemia trait. Clin Lab Haematol 1998;20:279-83.
14. Sirdah MM. Consanguinity profile in the Gaza Strip of Palestine: Large-scale community-based study. Eur J Med Genet 2014;57:90-4.
15. Reading NS, Sirdah MM, Tarazi IS, Prchal JT. Detection of nine mediterranean β-thalassemia mutations in palestinians using three restriction enzyme digest panels: A reliable method for developing countries. Hemoglobin 2014;38:39-43.
16. Tarazi I, Al Najjar E, Lulu N, Sirdah M. Obligatory premarital tests for beta-thalassaemia in the Gaza Strip: Evaluation and recommendations. Int J Lab Hematol 2007;29:111-8.
17. Sirdah MM, Sievertsen J, Al-Yazji MS, Tarazi IS, Al-Haddad RM, Horstmann RD, *et al.* The spectrum of β-thalassemia mutations in Gaza Strip, Palestine. Blood Cells Mol Dis 2013;50:247-51.
18. Origa R. B-thalassemia. Genet Med 2017;19:609-19.
19. Li CK, Chik KW, Lam CW, To KF, Yu SC, Lee V, *et al.* Liver disease in transfusion dependent thalassaemia major. Arch Dis Child 2002;86:344-7.
20. Hashemizadeh H, Noori R, Kolagari Sh. Assessment hepatomegaly and liver enzymes in 100 patients with beta thalassemia major in Mashhad, Iran. Iran J Ped Hematol Oncol 2012;2:171-7.
21. Salama KM, Ibrahim OM, Kaddah AM, Boseila S, Ismail LA, Hamid MM, *et al.* Liver enzymes in children with beta-thalassemia major: Correlation with iron overload and viral hepatitis. Open Access Maced J Med Sci 2015;3:287-92.
22. Kwiatkowski JL. Current recommendations for chelation for transfusion-dependent thalassemia. Ann N Y Acad Sci 2016;1368:107-14.
23. Di Maggio R, Maggio A. The new era of chelation treatments: Effectiveness and safety of 10 different regimens for controlling iron overloading in thalassaemia major. Br J Haematol 2017;178:676-88.
24. Kini PA. Management of ß-thalassemia – Consensus and controversies. Pediatr Hematol Oncol J 2017;2:94-7.
25. Colah KI, Gorakshakar A. Burden of thalassemia in India: The road map for control. Pediatr Hematol Oncol J 2018;2:79-84.
26. Rameshwar L, Suman AS, Meena P, Goyal S. Correlation of liver enzymes with serum ferritin levels in β-thalassemia major. Int J Res Med Sci 2016;4:3271-4.
27. O'Neal HR Jr., Niven AS, Karam GH. Critical illness in patients with asplenia. Chest 2016;150:1394-402.
28. Ricerca BM, Di Girolamo A, Rund D. Infections in thalassemia and hemoglobinopathies: Focus on therapy-related complications. Mediterr J Hematol Infect Dis 2009;1:e2009028.
29. Lipson RL, Bayrd ED, Watkins CH. The postsplenectomy blood picture. Am J Clin Pathol 1959;32:526-32.
30. Horowitz J, Leonard D, Smith J, Brotman S. Postsplenectomy leukocystosis: Physiologic or an indicator of infection? Am Surg 1992;58:387-90.
31. Rahav G, Volach V, Shapiro M, Rund D, Rachmilewitz EA, Goldfarb A, *et al.* Severe infections in thalassaemic patients: Prevalence and predisposing factors. Br J Haematol 2006;133:667-74.
32. Sakran W, Levin C, Kenes Y, Colodner R, Koren A. Clinical spectrum of serious bacterial infections among splenectomized patients with hemoglobinopathies in Israel: A 37-year follow-up study. Infection 2012;40:35-9.
33. Sinwar PD. Overwhelming post splenectomy infection syndrome - Review study. Int J Surg 2014;12:1314-6.
34. Sari TT, Gatot D, Akib AA, Bardosono S, Hadinegoro SR, Harahap AR, *et al.* Immune response of thalassemia major patients in Indonesia with and without splenectomy. Acta Med Indones 2014;46:217-25.
35. Khan PN, Nair RJ, Olivares J, Tingle LE, Li Z. Postsplenectomy reactive thrombocytosis. Proc (Bayl Univ Med Cent) 2009;22:9-12.
36. Perisano C, Marzetti E, Spinelli MS, Calla CA, Graci C, Maccauro G. Physiopathology of bone modifications in beta-thalassemia. Anemia 2012;2012:320737.
37. Taher AT, Musallam KM, Karimi M, El-Beshlawy A, Belhoul K, Daar S, *et al.* Splenectomy and thrombosis: The case of thalassemia intermedia. J Thromb Haemost 2010;8:2152-8.
38. Leone G, Pizzigallo E. Bacterial infections following splenectomy for malignant and nonmalignant hematologic diseases. Mediterr J Hematol Infect Dis 2015;7:e2015057.
39. Ammara SA, Zahranc MA, Embaby M. Splenectomy for patients with b-thalassemia major: Long-term outcomes. Egypt J Surg 2014;33:232-6.
40. Zhou YL, Zhang XH, Liu TN, Wang L, Yin XL. Splenectomy improves anaemia but does not reduce iron burden in patients with haemoglobin H constant spring disease. Blood Transfus 2014;12:471-8.
41. Kiani RA, Waheed U, Asad MJ, Abbasi S, Zaheer HA. Epidemiology of transfusion transmitted infection among patients with β-thalassaemia major in Pakistan. J Blood Transfus 2016;2016:5.