Clinical Evaluation of Some Biochemical and Hematological Variables and Their Relationship to Serum Ferritin Levels of Major Beta Thalassemia Patients in Maysan Governorate - Iraq.

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Abstract

Thalassemia is a recessive genetic disease resulting from a disorder and defect in hemoglobin production. The current study was designed to evaluate some hematological and biochemical variables and their relationship to serum ferritin (iron stock) levels in Thalassemia patients and study them in terms of housing, age and blood types in Maysan Governorate. This study was conducted during the
period from December 2020 to the end of October. The results of the current study recorded a highly significant relationship (P<0.000) for all hematological parameters with a decrease in the levels of (HGB) (RBC), (MCV) for patients and an increase in the level of white blood cell count (WBC) and platelet count (PLT) compared to the control. The current study recorded a highly significant relationship (P<0.000) and high levels of all liver enzymes (ALP), (ALT), (AST), (TSB), (TP) compared to the control group. The present study showed a slight decrease in urea levels and an increase in creatine levels compared to the control. The results of linear regression analysis showed a correlation between ferritin and hematological parameters, where the relationship was significant with RBC, HGB, MCV and insignificant with WBC, PLT, while there was a correlation between ferritin levels and liver and kidney enzymes, where the relationship was significant and positive with ALP. (AST) (TSB), and non-significant positive for (ALT), (TP), (Urea), (Creatinine).

Keywords: Beta-Thalassemia, Hematological, Biochemical, Ferritin, Pearson's correlation

1. Introduction

Genetic disorders in human hemoglobin that fall under the category of single-gene disorders are among the most common disorders, affecting approximately 5% of the total world population who carry one or more mutations in the genes responsible for hemoglobin production [1], [2]. One of the most common genetic blood disorders affecting humans is thalassemia (Mediterranean anemia). This is because hemoglobin is primarily composed of two types of protein, alpha-globin and beta-globin, which bind with the heme pigment to form the entire molecule of hemoglobin. And that any genetic defect that prevents the body from producing enough of these proteins for one or both proteins will result in blood cells being unable to transport enough oxygen, resulting in childhood
anemia. The oxygen requirements of the human body necessitate the continuous circulation of blood containing a high concentration of hemoglobin (Hb) and lasts throughout life. The amount of Hb influences oxygen transport, but so does the protein’s affinity for the gas, which can be optimized for environmental conditions by varying the concentration of effectors such as hydrogen ions, chloride, and CO₂ [3].

The portion of the hemoglobin molecule is made up of two distinct chains, alpha and beta, and either can be impacted. More than 200 different mutations can cause thalassemia [4], [5]. Because each is distinguished by the presence of a small number of common mutations and a large number of rare mutations, these mutations are not evenly distributed, but rather have a geographic and racial origin. Thalassemia is a hereditary disorder, which means that at least one of your parents must be a carrier. It is caused by a genetic mutation or the deletion of specific key genetic components [6], [7]. Thalassemia is classified into two types: alpha and beta-thalassemia. Each of them is classified into several types based on the degree of the mutation. Alpha and beta thalassemia are caused by reduced or absent production of Beta-globin chains [8]. Beta-thalassemia is more common in people of Mediterranean, African, and Southeast Asian ancestry, whereas alpha thalassemia is more common in people of African and Southeast Asian ancestry [9].

Mutations in the HBB gene on chromosome 11 result in autosomal recessive beta thalassemia. The nature of the mutation determines the severity of the illness [10]. The cornerstones of treatment for -thalassemia are red blood cell transfusions and iron chelation therapy, with allogeneic hematopoietic stem cell transplantation and gene therapy providing additional disease-management options for eligible patients. With up to 90% of severe cases of Beta-thalassemia occurring in resource-constrained countries, and estimates indicating that 22,500 deaths occur each year as a direct result of inadequate transfusion.
The primary treatment for thalassemia is blood transfusion. However, multiple blood transfusions can result in iron overload. As a result, increased erythropoiesis intensifies dietary iron absorption from the gastrointestinal tract, resulting in a severe form of iron overload [11], [12]. Iron deposition in the liver, heart, muscles, and kidneys can cause severe damage to a variety of organs. The most serious complication of iron overload that requires chelation is cardiotoxicity [13]. The liver is also the primary iron storage organ and the only site of ferritin synthesis. Iron is normally bound to protein in the liver, and free iron is extremely toxic. Iron, in its unbound form, stimulates the production of free radicals, which cause toxicity and liver and Renal damage [14], [15]. In other studies, the relationship between serum ferritin and hepatic iron concentration in several blood borne thalassemia patients [16]. has been reported in several blood borne thalassemia patients. There is, however, a scarcity of data on the relationship between iron overload and hematological disorders, as well as liver and kidney damage in thalassemia patients.

2. Materials and Methods

2.1 Subject and Study Design

This study was conducted in Center of Maysan Governorate for patients with beta thalassemia who visit the Genetic Blood Disease Center to obtain appropriate treatment after clinical confirmation of their infection. (100) patients and (70) as a control group, their ages ranged between (1-50 years) and they were divided into four age groups: first age group (1-10), second age group (11-19 years). The third age group (20-29 years) and the fourth age group (30 years) and more. Patients' information was collected through a questionnaire prepared in advance for this purpose, which involved taking a lot of information about each patient through face-to-face meeting with clinically diagnosed patients and their parents. Including name, gender, race, residence and in general a detailed and comprehensive history
of the disease was presented and then questions were asked about the family history, which included the kinship between the number of parents of affected and unaffected children, and the number of deceased siblings of any of the relatives with thalassemia and blood types. Figure (1) shows the design of the study.

![Figure (1) general design of the research]

### 2.2 Collection of Blood samples

Five ml of intravenous blood samples were collected from patients and control regimen of healthy subjects after obtaining parental consent. Blood was drawn from patients before periodic transfusion. The collected blood was roughly evenly divided (2.5 ml) into two types of tubes, an EDTA tube to prevent coagulation and a gel tube to obtain serum after leaving the blood in this tube for (30) minutes until the blood clotted and then centrifuged for 5 minutes and the serum was isolated with Taking into account the lack of hemolysis at 4000 rpm, collecting serum in Eppendorf tubes, then using blood serum in biochemical tests, and using blood in EDTA tubes to study and evaluate hematological parameters.
2.3 Hematological Analysis:

The blood parameters were measured using an automatic chemical analyzer, which is a device (Sysmex XS-1000i). The device calculates the volumes of red blood cells (RBC) and platelets (PLT) using hydrodynamic focus-enhanced electronic resistance detection. Hemoglobin (HGB) is converted to SLS-hemoglobin and read optically. White blood cell counts are evaluated using semiconductor laser flow cytometry using scattered light and fluorescence to determine differences in cell size, complexity, and RNA/DNA content.

2.4 Biochemical Analysis:

Functional liver enzymes alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), total bilirubin (TSB) and total protein (TP) were analyzed using the Cobas Integra 400 Analyzer automatically. Ferritin concentration and analysis were measured using Cobas Integra e 411 Analyzer automatically.

2.5 Statistical Analysis:

All results of the current study were studied and expressed as the mean standard deviation of all patients with beta thalassemia major. At the same time, significant differences were found between all beta thalassemia patients and groups of healthy individuals using the statistical program for social sciences (SPSS) version 22. Calculation of Pearson correlation (r). The P value was less than 0.05 for the smallest level of significance.

3. Results

3.1 Region of Residence:
Figure (2); Geographical distribution of beta thalassemia patient samples in Maysan Governorate and its regions

The study sample included (100) patients, and the result showed that city center had the largest percentage of infection then the percentage (12%) belonging to the district of Almajar, the percentage (7%) to the Kumait district, the percentage (5%) to the district of Al-Maimuna the percentage (6%) for the Qeleat salih, percentage (4%) belongs to each of the districts of Al-Musharrah and the percentage (3%) Ali Al-Shargi, and the percentage (2%) to the district of Ali Al-Gharbi respectively As in Figure (2)

3.2 Age-specific results and their effect on hematological and biochemical parameters

Figure (3); (A): A pie chart displays the Age results for the Control group. (B): A pie chart displays the Age results for the Thalassemia group, G1: The first age group is less than or equal
to (10) years, **G2**: Second age group (11-19) years, **G3**: Third age group (20 - 29) years, **G4**: Third age group more or equal to (30) years.

The age groups in this study were divided into four groups, and the study showed that the highest percentage of the study sample for patients was in the age group between (11-19) years (36%) and then (28%). for ages. (≤ 10), these percentages begin to decrease with age until they reach (24%) for age between (20 - 29) years and (12%) for age (≥ 30), as well as for healthy people, it was the highest percentage of the same age group for patients, which are between (11-19) years, and at a percentage almost equal to patients, which is (35.71%). then these percentages start decreasing with age as well (32.85%) for age between (20 - 29) years and (11.42%) for age (≥ 30), Figure (3).

**Table (1)** Hematological and biochemical characteristics of patients according to Age

<table>
<thead>
<tr>
<th>Parameters</th>
<th>≤10 year</th>
<th>11-19 year</th>
<th>20-29 year</th>
<th>≥ 30 year</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemoglobin (HGB)</td>
<td>10.51±0.22</td>
<td>10.44±0.19</td>
<td>10.39±0.31</td>
<td>10.44±0.42</td>
<td>0.237</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>4.22±0.09</td>
<td>4.10±0.07</td>
<td>4.12±0.08</td>
<td>4.14±0.13</td>
<td>0.804</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>8.05±2.69</td>
<td>10.47±2.14</td>
<td>10.36±2.40</td>
<td>21.37±3.76</td>
<td>0.035*</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>289.91±120.74</td>
<td>469.90±96.03</td>
<td>369.67±107.63</td>
<td>413.93±168.37</td>
<td>0.698</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>32.38±3.89</td>
<td>24.89±3.10</td>
<td>26.23±3.47</td>
<td>21.41±5.43</td>
<td>0.334</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>142.77±11.84</td>
<td>170.32±9.41</td>
<td>123.61±10.55</td>
<td>130.07±16.51</td>
<td>0.008**</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>38.88±38.88</td>
<td>33.30±3.28</td>
<td>33.24±3.68</td>
<td>30.13±5.75</td>
<td>0.588</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>0.94±0.161</td>
<td>1.00±0.128</td>
<td>1.36±0.14</td>
<td>0.88±0.225</td>
<td>0.131</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>7.14±0.08</td>
<td>7.27±0.07</td>
<td>7.37±0.07</td>
<td>7.17±0.122</td>
<td>0.221</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>2028.66±1085.59</td>
<td>3873.12±683.43</td>
<td>1925.63±967.75</td>
<td>1464.34±1513.79</td>
<td>0.324</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>23.15±1.19</td>
<td>25.88±0.95</td>
<td>24.35±1.06</td>
<td>23.56±1.66</td>
<td>0.296</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>0.50±0.90</td>
<td>0.54±0.72</td>
<td>1.71±0.80</td>
<td>2.72±1.26</td>
<td>0.361</td>
</tr>
</tbody>
</table>

*P < 0.05, ** P < 0.001

The study showed, as shown in Table (1), the study of the effect of different age groups on biochemical and hematological parameters. The study did not indicate significant differences except for two parameters, where the concentrations were statistically significant at the probability level (p < 0.05) for
all age groups. The results of the current study showed statistically significant differences in the level of white blood cell counts (WBC) for all age groups, where the average concentrations of (WBC) were (8.05 ± 2.69), (10.74 ± 2.14), (10.36 ± 2.40), (21.37 ± 3.76). for age groups (10 years) (11-19 years old), (20-29 years old) (30 years old) respectively. The differences were also highly statistically significant for alkaline phosphatase (ALP) and for all age groups, where the mean was (142.77 ± 11.84), (170.32 ± 9.41), (123.61 ± 10.55), (130.07 ± 16.51) and for the age groups. (10 years) (11-19 years) (20-29 years) ((30 years) respectively

3.3 Blood groups and their effect on hematological and biochemical parameters

Table (2): shows Blood Groups parameter for both control and thalassemia group.

<table>
<thead>
<tr>
<th>ABO Groups</th>
<th>A</th>
<th>B</th>
<th>AB</th>
<th>O</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N</td>
<td>Ratio</td>
<td>N</td>
<td>Ratio</td>
<td>N</td>
</tr>
<tr>
<td>Healthy</td>
<td>17</td>
<td>24.28%</td>
<td>30</td>
<td>42.85%</td>
<td>6</td>
</tr>
<tr>
<td>Thalassemia</td>
<td>26</td>
<td>26%</td>
<td>41</td>
<td>41%</td>
<td>4</td>
</tr>
</tbody>
</table>

Table (2) the distribution of blood groups for males and females in thalassemia patients and comparing them with the control group. where it was found that the highest percentage is group (B) with 41%, followed by group (O) with 29% group (A) 26% and AB 4% while The percentages in the control group were (42.85%) for group B, followed by 24.28% for both (O) and (A) followed by the lowest percentage of 8.57% for group AB. The study showed no significant differences between men and women for patients and healthy people, considering that these results may be due to sampling opportunities or the number of samples.

Table (3): Hematological and biochemical characteristics of patients according to blood groups
The results of the current study, as shown in Table (3) showed the effect of blood groups on the biochemical and hematological variables. The study indicates that there are differences with statistical significance for two biochemical variables, where the differences were significant and statistically significant in the average concentration of alkaline phosphatase enzyme at the probability level. \( p < 0.05 \) and mean concentration \((127.53 \pm 11.38), (162.18 \pm 8.76), (98.72 \pm 23.55), (148.10 \pm 11.14)\) for ALP, according to blood groups (A, B, AB, O) respectively. And highly statistically significant differences in the mean creatinine concentrations with a probability level \( p < 0.05 \) as follows \((1.52 \pm 0.82), (0.569 \pm 0.635), (7.16 \pm 1.70)\) for each blood group (A, B, AB, O) respectively. There are no statistically significant differences for the rest of the chemical and hematological variables. Significant differences in creatinine levels, whether low or high, indicate the presence of kidney dysfunction, the increase may be caused by eating a large amount of protein from eating meat and nutritional supplements, or acute kidney inflammation, or the decrease in creatinine level may be caused by age or poor Nutrition or the effect of tumors in the kidneys or the effect of some medications, and significant differences in ALP concentrations indicate the presence of bone disorders or liver damage.
3.4 Evaluation of the level of hematological parameters of patients and control group

Table (4): The level of hematological characteristics in study subjects.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Healthy (N= 70)</th>
<th>Patients (N= 100)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total Mean ± SD</td>
<td>Total Mean ± SD</td>
<td></td>
</tr>
<tr>
<td>RBC x 10^6/µL</td>
<td>4.88 ± 0.51</td>
<td>3.41 ± 0.64</td>
<td>P=0.000 **</td>
</tr>
<tr>
<td>HGB (g/dL)</td>
<td>13.19 ± 1.50</td>
<td>8.08 ± 1.28</td>
<td>P= 0.000 **</td>
</tr>
<tr>
<td>MCV(FL)</td>
<td>83.93 ± 6.90</td>
<td>76.18 ± 6.89</td>
<td>P= 0.000 **</td>
</tr>
<tr>
<td>WBC X 10^3/µL</td>
<td>8.94 ± 12.45</td>
<td>13.44 ± 19.07</td>
<td>P=0.086 *</td>
</tr>
<tr>
<td>PLT x 10^3/µL</td>
<td>276.61 ± 76.61</td>
<td>501.27 ± 952.28</td>
<td>P= 0.051*</td>
</tr>
</tbody>
</table>

In the table (4) the results of the study showed that there were statistically significant differences in the hematological parameters between thalassemia patients and the healthy ones. In terms of changes in the number of blood cells (WBC, RBC, PLT), the study showed that there was a significant increase in the number of white blood cells in patients, which amounted to (13.44 ± 19.07) compared to the members of the control group, which amounted to (8.94 ± 12.45), where the value was (P = 0.086), which indicates a statistically significant difference. The study also showed that the level of red blood cell count (RBC) significantly decreased in patients (3.41 ± 0.64) compared to the control group (4.88 ± 0.51), where the results indicated a highly significant difference with statistical significance (P=0.000). The results of the current study for thalassemia patients indicated a higher level of platelet count (PLT) which reached (501.27 ± 952.28) compared to the control group (276.61 ± 76.61), with significant differences (P = 0.051) as well. The results of the study showed a decrease in the Mean corpuscular volume (MCV) which means that the size of red blood cells is smaller than the normal size, as the average size of the red blood cells was (76.18 ± 6.89) compared to the healthy ones (83.93 ± 6.90), and the study showed The
presence of highly significant differences that were statistically significant (P = 0.000). The current study indicated a decrease in hemoglobin concentration in thalassemia patients (8.08 ± 1.28) compared to the control group (13.19 ± 1.50), with a highly significant difference that was statistically significant in thalassemia patients (8.08±1.28) (P = 0.000).

### 3.5 Evaluation of the Level of Biochemical Parameters of Patients and Control Group

Table (5): indicates the distribution of the level of concentrations of liver function parameters patients and healthy people according to the effect of some biochemical variables for the province of Maysan and its districts and sub-districts.

**Table (5): The level of biochemical characteristics in study subjects.**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Healthy (N= 70)</th>
<th>Patients (N= 100)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total Mean ± SD</td>
<td>Total Mean ± SD</td>
<td></td>
</tr>
<tr>
<td>ALT (U/L)</td>
<td>21.74 ± 7.74</td>
<td>31.92 ± 30.45</td>
<td>P= 0.007 **</td>
</tr>
<tr>
<td>ALP (U/L)</td>
<td>82.50 ± 14.27</td>
<td>210.51 ± 98.74</td>
<td>P= 0.000 **</td>
</tr>
<tr>
<td>AST(U/L)</td>
<td>20.01 ± 6.17</td>
<td>48.71 ± 32.29</td>
<td>P= 0.000 **</td>
</tr>
<tr>
<td>T.S. Bilirubin (mg/dL)</td>
<td>0.55 ± 0.25</td>
<td>1.56 ± 1.28</td>
<td>P= 0.000 **</td>
</tr>
<tr>
<td>Total Protein (g/dL)</td>
<td>7.07 ± 0.33</td>
<td>7.40 ± 0.66</td>
<td>P= 0.000 **</td>
</tr>
<tr>
<td>Ferritin (ng/mL)</td>
<td>30.46 ± 19.70</td>
<td>5169.94 ± 8705.13</td>
<td>P= 0.000 **</td>
</tr>
<tr>
<td>Urea( mg/dL)</td>
<td>25.72 ± 8.00</td>
<td>23.07 ± 6.89</td>
<td>P= 0.022 *</td>
</tr>
<tr>
<td>Creatinine mg/dL</td>
<td>0.70 ± 0.20</td>
<td>1.45 ± 7.25</td>
<td>P= 0.384</td>
</tr>
</tbody>
</table>

*P < 0.05, ** P < 0.001

In the table (5) the results of liver function tests for all liver enzymes were shown, where there was a significant (P < 0.007) increase in ALT concentration in
patients (31.92 ± 30.45) compared with healthy subjects (21.74 ± 7.74). The AST level also increased significantly as it was (P<0.000) in patients (48.71 ± 32.29) compared with healthy people (20.01 ± 6.17), also the current study indicated a highly significant increase in the level of ALP concentration in patients (210.51 ± 98.74) compared to the control group. (82.50 ± 14.27). A significant increase in the levels of total bilirubin was also observed, where it was (P<0.000) in patients (1.56 ± 1.28) compared with healthy subjects (0.55 ± 0.25). The study also indicated an increase in the levels of total protein in patients (7.40 ± 0.66), compared to healthy controls (7.07 ± 0.33) with high significance and statistical significance (P = 0.000). The study also indicated a highly significant and statistically significant increase (P = 0.000) for ferritin levels among patients (5169.94 ± 8705.13) compared to healthy controls (30.46 ± 19.70). The study also showed a significant increase (P = 0.022) in urea levels for patients (23.07 ± 6.89) compared to healthy controls, which was (25.72 ± 8.00). Also, there was an increase in creatinine levels for patients (1.45 ± 7.25) compared to the control group (0.70 ± 0.20), but with a non-significant difference (P = 0.384).

3.6 Results of the correlation between ferritin levels and hematological parameters
Figure (4) A: Scatter plot showing a significant (inverse) correlation between serum ferritin levels and red blood cell (RBC) levels in thalassemia patients., B: Scatter plot showing a significant negative (weak) correlation between serum ferritin levels and hemoglobin (HGB) in thalassemia patients.

Figure (4). C: Scatter plot showing a significant (inverse) correlation between serum ferritin levels and mean corpuscular volume (MCV) in thalassemia patients., D: Scatter plot showing a non-significant (inverse) correlation between serum ferritin levels and white blood cell count (WBC) in thalassemia patients.
Figure (4). E: Scatter plot showing a significant (positive) correlation between serum ferritin levels and platelet count (PLT) in thalassemia patients.

3.7 Results of the Correlation Between Ferritin Levels and Biochemical parameters.

Figure (5) A: Scatter plot showing a non-significant positive correlation between serum ferritin and ALT levels in thalassemia patients., B: Scatter plot showing a significant positive correlation between serum ferritin and ALP levels in thalassemia patients.
Figure (5) C: Scatter plot showing a significant positive correlation between serum ferritin and AST levels in thalassemia patients. D: Scatter plot showing a significant positive correlation between serum ferritin and Total Bilirubin (TSB) levels in thalassemia patients.

Figure (5). E: Scatter plot showing a non-significant positive correlation between serum ferritin and Total protein (TP) levels in thalassemia patients. G: Scatter plot showing a non-significant positive correlation between serum ferritin and creatinine levels in thalassemia patients.
Figure (5) F: The scatter plot shows a non-significant positive correlation between serum ferritin levels and Urea levels in thalassemia patients.

4. Discussion

The study indicated the spread of beta thalassemia disease in all regions of Maysan governorate. We conclude from this that thalassemia is a genetic and recessive genetic disease that spreads in all regions at different rates. The study showed that the most vulnerable age group to infection was (11-19 years), and this percentage decreases with age, as well as the increase in percentages and numbers of patients in the older age groups, indicating that blood transfusion and chelation therapy extend the life of these patients [17]. The study showed statistically significant differences in white blood cell count and alkaline phosphatase enzyme concentration. The current study indicates obtaining different percentages of blood groups for males and females for patients and for control. Bearing in mind that these results may be due to sampling opportunities or number of samples. This study agreed with the study [18] with a slight difference in the proportions where genetic disorders, especially beta-marine anemia, were studied in India. It was found that blood group (B) was the most common with (47.2%) followed by group B (O) with 32.9% and in another study conducted by researcher [19]. on 933
patients suffering from blood disorders and beta thalassemia mainly in North India, the highest percentage of patients with blood type (O) (36.55%) and blood type (B) was found in the second place (35.78%) This study agreed with the study [20]. with a small difference in the proportions where genetic disorders, especially beta-thalassemia, were studied in India. It was found that blood group (B) was the most common with (47.2%) followed by group (B), (O) with 32.9% and in another study conducted by researcher [21]. on 933 patients suffering from blood disorders and beta thalassemia mainly in North India, the highest percentage of patients with blood type (O) (36.55%) and blood group (B) was found in second place (35.57%).

The study showed that there were statistically significant differences in alkaline phosphatase (ALP) and creatinine and significant differences in creatinine levels. whether low or high, indicates the presence of kidney dysfunction, and significant differences in ALP concentrations indicate the presence of bone Liver disorders or damage. The current study indicates highly significant differences with statistical significance resulting from the effect of blood parameters. This increase may be due to the general conditions of the disease and the hyperactivity of the immune system. as these patients receive regular treatment and different blood donors. Red blood in and out of the bone marrow, which stimulates orthopoietin secretion by the kidneys, which then stimulates the bone marrow to produce and form red and white blood cells [22].

A decrease in the level of red blood cell (RBC) counts has been observed due to genetic mutations in the genes responsible for the synthesis of hemoglobin protein chains that lead to a disturbance in the biosynthesis of globin chains and a loss of homeostasis in hemoglobin synthesis [23]. This may also affect the number, shape, and size of red blood cells during the stages of their formation in the bone marrow. As a result, red blood cells are small in size and therefore do not contain the same normal size as red blood cells, [24]. It was noted that there is an increase
in the number of platelets as a result of the secondary infection that patients are exposed to as a result of continuing blood transfusions, which leads to a deterioration in the patient's health status, such as parasitic diseases, viral hepatitis and acquired immunodeficiency virus. We cannot forget that splenectomy leads to a significant increase in the number of platelets in patients with beta thalassemia, and the increase may reach four times the normal percentage after splenectomy, due to the body's inability to do so. Get rid of it. of extra cells in the blood [25].

The current study showed significant differences and statistical significance for the enzymatic and non-enzymatic parameters, except for creatinine. The enzymatic and non-enzymatic parameters are spread in a number of body organs such as the liver, heart, kidneys, pancreas and skeletal muscles. Alanine aminotransferase (ALT) is concentrated in the liver. In the results of our study, there was an increase in the levels of this enzyme compared to members of the control group. The increase in the concentration of this enzyme is evidence of liver damage and disorder, as liver diseases are associated with blood transfusion, which leads to increased levels of iron in all parts of the body [26] The presence of aspartate aminotransferase (AST) is concentrated in the liver, heart and skeletal muscle. A noticeable increase in the levels of this enzyme was also observed, and this indicates a defect in the tissues in which it is present. There was also a significant increase in the levels of alkaline phosphatase (ALP) compared to the control group, and the explanation for this increase is that most of the activity of this enzyme comes from (bone tissue and liver). The enzyme enters the blood circulation and then increases its level and effectiveness in the blood [27]. that there is a significant increase. Statistically significant for both total bilirubin and total protein when compared with the levels of members of the control group and that the cause of the increase in total bilirubin (T.B.S) is the result of the breakdown of red blood cells and there (T B.S) in bile in the liver, and the healthy
liver gets rid of bilirubin from the body. Increasing its concentration in the blood serum is an indication of liver injury and damage. If damage occurs, bilirubin is excreted from the liver into the bloodstream, causing what is known as jaundice, which is the result of iron deposition in the body's organs.

Bilirubin and ALT may act synergistically or independently in promoting chronic liver disease and thus cell damage. During the study, a significant increase in the levels of total protein (albumin, globulin) and total protein produced by the liver and the immune system was also observed. and waste removal [28]. Thus the results of this study are consistent with those of other studies conducted by him [29]. and it was found that liver function is increased three to four times in patients with beta thalassemia compared to normal individuals [30]. The results of our study showed that there is a significant decrease in urea levels with significant differences compared to the control group and that the decrease in concentrations is due to liver damage, that is, the inability of the liver to form it, as urea is the metabolic product from the conversion of proteins into amino acids in the liver and then into ammonia. Then convert it to urea. As for the causes of increased urea concentration, it is an indication of kidney disease. In the case of kidney disease, it is not excreted with urine, so its concentration in the blood increases. One of the most important causes of kidney disease is the deposition of iron in large quantities in the kidney tissues, which loses its function, which is renal filtration. The presence of large amounts of iron will result in the formation of a hydroxyl radical from a known reaction, (Fenton reaction)

\[ \text{Fe}^{2+} + \text{H}_2\text{O}_2 = \text{OH}^- + \text{OH}^- + \text{Fe}^{3+} \]

These free radicals are among the most damaging free radicals to cells, so we see them attack all biomolecules such as lipids, proteins and DNA, leading to cell death and other effects [31]. It was also observed that there was a significant increase in creatinine concentrations in thalassemia patients compared to the
The rate of iron absorption from the GI tract is about 3-4 times higher in beta-thalassemia patients than the normal rate in non-transmitted thalassemia severe patients, iron absorption is abnormal from the GI tract. It leads to an accumulation of iron about 2-5 grams per year, which in turn depends on the expansion of erythrocytes. In this way, iron is stored in thalassemia patients by transfusion-related iron overload as well as increased iron absorption from the gastrointestinal tract [33]. Iron is stored primarily as ferritin in hepatocytes. Ferritin is a 450 kDa protein made up of 24 subunits (types L and H), which are found in almost all types of cells. Within the ferritin shell, iron ions combine with phosphate and hydroxide ions to form a crystalline complex that can store about 4,500 iron ions (Fe $^{+3}$) [34]. Serum ferritin is secreted as iron-free to a level of about 3000 μg/L from macrophages, but above this value, iron-laden ferritin leaks from hepatocytes increase. Infection or Cancer [35]. To estimate the level of iron toxicity, critical values of blood ferritin vary from 1000-3000 mcg/L in different studies Furthermore, standard values of blood ferritin level also vary to a wide range in males (10 -220 μg/L) and females (10-85 μg/L) under normal conditions. Serum iron and ferritin levels correlate well, [36]. For this reason, serum ferritin concentration is generally used to estimate iron overload in beta-thalassemia patients. In the current study, the mean serum ferritin level (5,169.94 ± 8705.13 ng/mL) is much higher than the peak value (1000 mcg/L) indicating a significant rise in iron due to multiple transfusions in thalassemia patients Our result is in agreement with the work of [37].

Those who found serum ferritin and iron levels in thalassemia patients were much higher than those in the control group. Linear regression relationship between ferritin and hematological and biochemical parameters. Our study aimed to find an association between hematological and biochemical parameters and
higher levels of serum ferritin in the thalassemia patients under study who undergo regular blood transfusions and chelators used to remove iron. The current study showed that there was a non-significant positive correlation between the level of ferritin in the blood and (ALT) in our study population \((r = + 0.128, p = 0.096)\). The reason tends to be that abnormal amounts of liver enzymes are likely due to liver damage caused by excess iron in thalassemia patients who are receptive to different blood. In the current study, we found elevated levels of \((r= 0.192, p = 0.012\) for AST), \((r = 0.466, p=0.000\) for ALP) and serum bilirubin (TSB) as well. \((p = 0.010, r = 0.197\) for TSB) and the results of our study also showed a positive and insignificant association between serum ferritin and \((p = 0.277, r = 0.084\) for total protein). Liver, especially ALT, AST and ALP, there was a positive correlation between serum ferritin and liver enzymes \((p = 0.000, r = 0.303\) for HGB), \((p = 0.000, r = 0.335\) for RBC), \((p = 0.003, r = 0.228\) for MCV), while the correlation with ferritin was not significant for white blood cells \((p = 0.577, r = 0.043\) for WBC), and also not significant for platelets \(( p = 0.766, r = 0.023\) for PLT).

**Conclusions**

The current study indicates the presence of disturbance and significant changes and statistical significance of biochemical and hematological parameters, and this could be the result of red blood cell breakage and excess blood transfusion and the resulting increase in iron concentration and thus deposition in the liver, kidneys and other organs where there was a significant correlation Positive between the two periods, which represents iron stores and hematological and biochemical parameters. This study shows the need to intensify efforts to follow up and investigate the causes of the disease and reduce them.

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References


6- Cao, A., & Galanello, R. [2010]: Beta-thalassemia. Genetics in medicine: official journal of the American College of Medical Genetics, 12(2), 61–76.


10- Goldman, L. & Schafer, A. I. [2010]: Goldman-Cecil Medicine, Elsevier Health Sciences.


22- Ponticelli, C.; Musallam, K.M.; Cianciulli, P.andCappellini, M.D. [2010]: Renal complications in transfusion-dependent beta thalassaemia, a review. Blood Reviews; 24: 239–244


