STUDY THE RELATIONSHIP BETWEEN HEPcidIN HORMONE CONCENTRATION, INTERLUKINE LEVEL 2,6 AND SOME PHYSIOLOGICAL PARAMETERS IN PATIENTS WITH B- THALASSEMIA MAJOR IN WASIT PROVINCE/ IRAQ.

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ABSTRACT

beta thalassemia major is a hereditary hemolytic anemia caused by a defect in the ability of erythroblasts to synthesize the beta chain of patients hemoglobin. Our study had included 40 patient's blood sample from both beta thalassemia major and beta thalassemia major with HCV had been compare with 20 control's blood sample. Where the interleukin-2 ,interleukin-6 and hepcidin hormone had been measured. We observed a significant increasing (p ≤ 0.05) in the value of immunological test involved IL-2 & IL-6 and hormonal test involved hepcidin hormone in patients compared with control. In addition observed a significant increasing (p≤ 0.05)in neutrophils activity by using nitro blue tetrazolum stain(NBT).The results of correlation indicated that there was a significant correlation between hepcidin hormone and each of IL-6, ferritin and total iron binding capacity TIBC, also a positive correlation between it and urea and negative with creatinine and the absence of a relationship between it and calcium ions. There is a significant correlation between Interleukin -2 with each of Interleukin-6 and ferritin , and there is a significant negative correlation between IL-6 and total iron binding capacity. There was no significant effect of interleukin 2&6 with each of urea, creatinine and calcium .It was found that there was a significant correlation between the neutrophils activity and interleukin 2&6 , and a highly significant correlation with hepcidin hormone.

Key word Beta-thalassemiamajor , hepcidin hormone, IL-2, IL-6 , thalassemia with hepatitis C virus, Neutrophil activity by using NBT stain.

I. INTRODUCTION

Thalassemia refer to a group of inherited blood disorders that lead to the formation of major public health problems throughout the world, especially in developing countries(Weiss et al., 2019). Thalassemia is a term that came from Greek origin , the wordthalssa means (sea), and the word hemia mean (blood) the term refer to a defect in the production of globin unit (alpha and beta) in hemoglobin as a result of the presence of gene mutations responsible for the amino acid sequence in the protein (patric and Eliezer, 2011). This disease spread in the Mediterranean region in the Middle East, Southeast Asia and Africa (Bhandari et al., 2018).Dr. Thomscooley was the first to describe thalassemia clinically in 1925, so it was called by the name of cooley's anemia ,where it was attributed to anemia Mediterranean because of it's discovery in Mediterranean region and then called it thalassemia major with a better understanding of genes, and the first cases of this disease were recorded in Italy and the united states ,and the word thalassemia was used for the first time in 1932(Johnect et al., 2013).Patients with thalassemia vary great , there are individuals with mild to silent mutations and then to mild hypochromic anemia , while other individuals may suffer from moderate to severe anemia dependent on blood transfusion (Khan et al., 2021). Blood transfusion was consider the first place for the treatment of global beta thalassemia major , this is done by transfusion of blood every two weeks in severe cases, which affect the patient's life and sometimes causes frequent infection and immune reactions in addition to an increase iron into vital organs and this is reasons for the increase number of death, especially bin Elderly thalassemia (Porter and Garbowski, 2014). Most of the Mediterranean anemia patients who depend on frequent blood transfusion appear to have serious clinical symptoms that affect their health due to excess iron. And the accumulation in the body in this case with need iron chelation therapy in order to normalize their iron level (transferrin saturation is less than 50%). These substance are given to apatient whose ferritin rises to 1,000 ng/ml , as the iron chelator works to
remove excess iron from plasma and cells by binding to chelable iron, it is easily excreted through urine and feces (Haghpanahet et al., 2014). The accumulated iron in patients with thalassemia can be treated by chelation therapy. This treatment improved the life expectancy of patients with thalassemia major (Neufeld, 2010). There are patients who are treated by taking hydroxyurea or thalidomide, and sometimes both for those who do not respond well to blood transfusion (Shah et al., 2020). The liver secretes peptide hormone that helps regulate iron in the body called hepcidin, and level of this hepcidin in patients with beta thalassemia major are very low, the concentration of hepcidin in beta thalassemia patients who receive blood transfusions are much higher than the non-transfused patients and this due to the increase in iron load as they have complications that include abnormalities in liver cells, cardiomyopathy and endocrine disorders resulting from iron overload, which often leads to the death of the patient (Pasricha et al., 2013). Since iron overload is one of the complications of blood transfusions suffered by patients with thalassemia, the hormone hepcidin regulates iron balance by inhibiting iron absorption and remove heavy metal to control on its toxicity (Saliba et al., 2015). Recently, the possibility of conducting a prenatal diagnosis is more safe and cheaper, where the genetic material derived from the emerged fetus (cell or cell –free DNA) is obtained from the mother’s blood and tested, and its does not pose any risk to the fetus (Giambona et al., 2016). Thalassemia patients who receive regular blood transfusions are at a risk of contracting hepatitis virus after a blood transfusion (Al-Farmawy et al., 2012). Hepatitis C virus is the major cause of infection among thalassemia patients and can lead to severe liver infections accompanied by all the long-term illness such as symptoms of disability, cirrhosis and hepatocellular carcinoma (Ataei et al., 2012). Its prevalence among thalassemia patients has expectation and the amount of blood transfusion were associated with an increased risk of infection with this virus (Alavianet et al., 2010).

II. MATERIALS AND METHODS

These study include 60 patients with both gender, 20 blood sample of patients with beta thalassemia major and 20 samples of beta thalassemia major patients with hepatitis C virus were collected and compare with 20 apparently healthy as a control group during the period from (November 2020 to April 2021) in the thalassemia center in Wasit province. Collecting samples from patients before the blood transfusion and after the centrifugation process, then distributing the serum to perform the necessary laboratory analysis. These include: hormonal test involved hepcidin hormone and immunological test such as Interlukin-2 (IL-2), and Interlukin-6 (IL-6). And examination of neutrophil activity by using neurul blue tetrazolium stain

Data entered and managed by using SPSS. Descriptive statistics (frequencies ± standard deviation with tables, Shapiro Wilk test for normality and Tukey's rule identify outliers ) and inferential statistics (Independent t-test-ANOVA, Kruskal-Wallis Test and correlation coefficient) were used P-value of ≤ 0.05 considered statistically significant.

III. RESULT AND DISCUSSION

In table (1) the result showed a significant increasing (P ≤ 0.05) in the hepcidin hormone for the patients compared to control. Where the mean value of hepcidin (mean ± SD) was in thalassemia patients (12.40 ± 7.26) pg/ml, and for thalassemia with viral hepatitis C was (34.4 ± 16.2) pg/ml, and for control was (6.1 ± 1.6) pg/ml.

In table (2) the results showed a significant increasing (P ≤ 0.05) for Interlukin-2 and Interlukin-6 in patients and compared with control. Where the result of Interlukin-2 (mean ± SD) in thalassemia patients was (39.7 ± 13.4) pg/ml, and for thalassemia with viral hepatitis C was (48.1 ± 17.1) pg/ml, and for control was (23.0 ± 7.75) pg/ml. The same table (2) showed the result of Interlukin-6 (mean ± SD) in thalassemia patients was (84.2 ± 19.7) pg/ml, and in thalassemia with viral hepatitis C was (83.3 ± 16.8) pg/ml, and the control was (40.3 ± 16.8).

In Table (3) the result showed a significant increasing (P ≤ 0.05) for Neutrophils activity by using Nitro Blue Tetrazolium Stain (NBT) for patients compared with health controls.

In table (4) the result showed correlation between hepcidin hormone and IL-2& IL-6 and some biochemical parameters in patients.

Table (1). Change in level of hormone hepcidin in thalassemia patients and thalassemia with viral hepatitis C compared with healthy control.
In table(1) indicated that was a significant increase in hepcidin hormone in thalassemia patients and thalassemia with viral hepatitis C compared to healthy control. Hepcidin hormone is an important regulator of iron homeostasis that contributed to the different metabolism (Meier-Ewert, et al., 2000). Where the data showed a significant increase in the level of hepcidin hormone in patients with thalassemia major and thalassemia with hepatitis C virus compared to health control. The reason for this increase was inflammatory condition (Alsaidiand, Zubair 2016).This conditions is due to antigen stimulation to repeat blood transfusions for patients. These results showed that serum hepcidin is elevated in people with thalassemia patients and thalassemia with
viral hepatitis C, and these elevation is more clearer in thalassemia patients who suffer with severe iron overload (Kaddahet et al, 2017). These result reported with (Kearney et al., 2007), Thalassemia patients with hepatitis C virus also contributed to a large increase of the production of hepaticidin, through the inflammation agent especially interleukin -6. Hepcidin hormone has antibacterial activity properties (Krause et al., 2000).

The results in table (2) showed that there was a significant increase in the immunoglobulin interleukin-2,6 in the group of thalassemia patients and thalassemia with viral hepatitis C, compared to health controls as in the above table. These results consistent with, (Morcos et al., 2010). As interleukin-2 has biological and clinical importance in thalassemia major patients, where interleukin-2 plays role in the mechanism of activation and proliferation of the T-Cell that give natural reaction against microbes. Elevated high level of IL-2 have been observed in patients with immune deficiency disease, infections, inflammatory. IL-2 also has been used in the treatment of the viral infections, vaccine manufacturing, and treatment of immune deficiency disease, (Haremmanet al., 2013).

In the same table (2) showed a significant increase in interleukin-6 for compared to healthy controls, as IL-6 has a role in a variety of chronic inflammatory disease, interleukin-6 has several functions, including stimulating the immune system against the disease as it is an anti-inflammatory, responsible for the activation and synthesis of protein in the acute phase that companies infections. (Tanaka et al., 2014). The current study showed an increase in IL-6 levels in both groups of patients compared to healthy controls, and the cause of increase levels of IL-6 may be the inflammation that occurs after repeated blood transfusions. Increase in IL-6 is attributed to the continuous stimulation of transfusion-related antigen and iron overload with phagocytosis activity (Yaylim et al., 2001). Most acute phase proteins are produced the liver and these proteins increase in concentration in blood plasma and decrease in response to inflammation. Also, the increase in interleukin-6 levels in patients with thalassemia and thalassemia with hepatitis C virus may the formation of an increase production, a defect in iron metabolism and an increase in IL-6 was documented in patients with beta thalassemia major (El-Rasheidyet et al., 2016). These results are agreement with (El-Rashidy et al., 2016).

Table (3) showed a significant increase in Neutrophil activity by using nitro blue tetrazolium stain (NBT) in patients and compared with healthy control. The result was in agreement with (Javad et al., 2008) in Iran they showed a significant increase in the phagocytic activity of patients compared with control. Another study was done by (Cantinicauzet et al., 1990) in Brussels, where they noticed that the activity of polymorphous phagocytic cells, has experienced situations due to Iron overload, in another study conducted by the same researcher in 1999, the action of sulfur-specific in neutrophils was impaired as a result of cytokines due to excess iron. All thalassemia patients suffer from chemical defects in neutrophils and all of them have a positive history of purulent infections (Matzner et al., 1993). Also limited (Kutukuceret al., 1996) chemical impairment in neutrophils and described the defect was found that might be caused by the iron overload after blood transfusion. Also (Ozturk et al., 2001) showed the increase in macrophages and neutrophils in patients is caused by the increase in cytokines inflammatory disease, especially interleukin-6.

In table (4) showed the correlation between hepcidin hormone with each of IL-6, ferritin and TIBC where it's believed the interleukin-6is the dominant catalyst of hepcidin during inflammation (Rodriguez et al., 2014). There is a significant positive correlation between hepcidin hormone with ferritin & TIBC which confirm the role of hepcidin in increasing the iron concentration in the body, these results agreed with (Peters et al., 2010) and (Sadiaa et al., 2012). There is also an insignificant positive correlation between hepcidin and urea and an insignificant negative with creatinin (Christensen and Brin, 1997). An insignificant correlation found between hepcidin and calcium (Kradith et al., 2016). The result showed a highly significant positive correlation between hepcidin and neutrophils activity (Peyssonnaux et al., 2006). There is a significant correlation between IL-2 and both IL-6 & ferritin, the reason due to the stimulatory effect of cytokines on hepaticidin production especially interleukin -6 (Salwaet al., 2010). While we notice a loss of correlation between IL-2 and both of TIBC & creatinin. A highly significant negative correlation was observed between IL-6 and TIBC, and these result agreed with (Hentzeet al., 2010), (Wessling –Resnick 2010), Interleukin -2 &6 were also insignificant correlation with urea, creatinine, and calcium, except IL-2 loss correlation with creatinine. Noted significant correlation between Interleukine 2&6 with neutrophils activity.

REFERENCES:


