Estimation of the concentrations of complement components, alph-1-antitrypsin and alph-2-macroglobulin in patients with B-thalassemia

Alaa J.H.; Shakir, H.M.; Zina, H.M.; Raheel, T.O., and Mohamed, H.A.

1,2,5 University of Babylon, College of Science, Biology Department
3, 4 Babylon Health Directorate

Abstract:

An immunological study were performed to estimation the concentrations of complement components (C3 and C4), alph-1-antitrypsin and alph-2-macroglobulin in sera of patients with B-thalassemia in Babylon province. The concentrations of these immunological parameters were determined by single radial immunodiffusion assay (SRID). This study revealed decreases in levels of complement components (C3 and C4) in patients which reached 89.2 and 19.9 mg/dl respectively, while it was 96.7 and 21.9 in control group, furthermore, this study revealed decreases in levels of alph-1 antitrypsin in patients in comparison with the healthy subjects, while there was no differences in mean of concentrations of alph-2 macroglobulin between these two groups. The results illustrated that decreases in levels of complement components as well as alph-1 antitrypsin in patients compared with healthy subjects in Babylon province.

Keywords: Complement, Alph-1 antitrypsin, Alph-2 macroglobulins, Patients, B-Thalassemia.

Introduction:

The increased frequency of infections associated with B-thalassemia major seems to be related to abnormalities of the immune system (Lombardi et al., 1994; Uguccioni et al., 1993), and the predisposition to autoimmune diseases is under the control of immune response genes, which play a central role in the presentation of antigens to the immune system (Bottazzo et al., 1986). B-thalassemia major itself is not a true hemochromatosis, it is a secondary hemochromatosis, which is based on the combination of chronic hemolytic anemia, iron intestinal hyperabsorption, and multiple blood transfusions that the patients receive (Jessup and Manno, 1998). Similarities between microbial antigens and self HLA molecules may result in autoimmune reaction after infections (Abbas et al., 1997). B-thalassemia is caused by defect of synthesis the beta-globulin chains of the hemoglobin tetramer (Cao, 2010; Luca et al., 2010), as a result the patients suffer from chronic anemia due to a process called ineffective erythropoiesis with massive splenomegaly and dramatic iron overload (Libani et al., 2008; Luca et al., 2001). The serum complement levels were found in some studies to be normal (Virgin et al., 1997; Corry et al., 1981), and in some patients a deficiency in the hemolytic activity of the classical (ConstantouLakis et al., 1978) or the alternative complement pathways was detected (Corry et al., 1981). Romeo et al. (1983) investigated a possible relationship between alph-1 antitrypsin phenotypes and liver disease in transfusion dependent thalassemia subjects. Alpha-2 macroglobulin is a large plasma protein found in the blood, and it is produced by the liver, as well as is a major component of the alpha-2 band in protein electrophoresis, also this protein is able to inactivate an enormous variety of proteinases (including serine, cysteine, aspartic and...
metalloproteinases), and it functions an inhibitor of coagulation by inhibiting thrombin, and inhibitor for fibrinolysis by inhibiting plasmin and kallikrein (Do Boer et al., 1993; Anderson et al., 1995).

The aim of this study to evaluate the changes in some humoral parameters of immune system in B-thalassemia patients in babylon governorate.

Materials and Methods:

1- Patients and Control:

Twenty five patients (15 males and 10 females) with B-Thalassemia their ages ranged between 10 - 20 years were studied at the thalassemic center in maternity and pediatrics hospital (Babylon province). All patients were living in babylon province and pre-diagnosed by physician. Fifteenth apparently healthy subjects (8 males and 7 females) with no symptoms of thalassemia disease were selected as control group.

2- Blood Samples:

The blood samples were drawn from each patients and controls (5 ml) by vein puncture using disposable syringes. The blood was placed in disposable tube, kept to clot at room temperature, and then centrifuged at 3000 r.p.m (Bishop et al., 1985) for 10 minutes. Sera were carefully transferred to appendrof tubes and stored in a liquotes at deep freezing at -20 C until used.

3- Immunological Tests:

The concentrations of complement components (C3 and C4), alpha-1 antitrypsin and alpha-2 macroglobulin were estimated according to Mancini et al., (1965) to which and manual procedure of linear chemical procedure to which company of Al macen Joaquim. Costa, Motgat, Barcelona, Spain.

4- Statistical Analysis:

The results were analyzed using completely randomized design (Donald, 1989).

Results:

The means of complement components C3 and C4 in patients were lower compared with control group, which reached 89.2 and 19.9 mg/dl respectively, while it was 96.7 and 21.9 mg/dl consecutively in control group as illustrated in table (1)

Table (1): The concentrations of complement components C3 and C4 (mg/dl) in patients with B-thalassemia.

<table>
<thead>
<tr>
<th>Nature Of treatment</th>
<th>Conc. Of C3 (mg/dl) Mean + SE</th>
<th>Conc. Of C4 (mg/dl) Mean + SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>89.2+ 7.4</td>
<td>19.9+2018</td>
</tr>
<tr>
<td>Control</td>
<td>96.7+ 8.6</td>
<td>21.9+ 2.8</td>
</tr>
</tbody>
</table>

Meantime, the means of concentrations of alpha-1 antitrypsin (mg / dl) were slightly lower in patients as compared with control group, while there was no significant difference in concentration of alpha-2 macroglobulin between patients and control group as revealed in table (2)
Table (2) : The concentrations of alph-1 antitrypsin and alph-2 macroglobulin (mg/ dl) in patients with B- thalassemia .

<table>
<thead>
<tr>
<th>Nature of treatment</th>
<th>Conc. of alph-1 antitrypsin (mg/dl) Mean ± SE</th>
<th>Conc. of alph-2 macroglobulin (mg/dl) Mean ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>112.5 ± 10.8</td>
<td>157.5 ± 12.8</td>
</tr>
<tr>
<td>Control</td>
<td>120.2 ± 10.5</td>
<td>158.6 ± 16.2</td>
</tr>
</tbody>
</table>

Discussion :

The patients with B- thalassemia showed lower values of serum complement components C3 and C4 . The donation is responsible for autoantibodies to the antigens involved which are derived from the donors because almost are HLA incompatibilities . The component of the immune system includin natural killer cells , were so depressed in multiple transfusion as it happened in aquired immunodeficiency virus infected people ( Pedro et al . , 1992) , also Rund and Rachmilewitz (2005) reported that immunological abnormalities in patients with B- thalassemia included : decreased opsonization and granulocyte phagocytosis as well as alteration in B and T cell number and function. ( Kapadia et al . , 1980 ; Munn et al . , 1981) . Factors such as splenectomy , iron overload ( Kapadia et al . , 1980 ) , repeated exposure to foreign antigens at the time of blood transfusion ( Gascon and Young , 1984) , and the use of the chelating agent ( deferoxamine ) ( Lederman et al . , 1984 ) have profound effects on the immune system . Iron overload has been a major cause of immunological disturbance in thalassemia , the mechanisms suggested include toxic effects of high iron levels on lymphocyte function ( Bryan and leech , 1981 ) . Ahmed et al. (2005) illustrated that serum levels of complement components C3 and C4 were lower in patients with B- thalassemia in comparison with healthy subjects . Splenectomized patients had no differences in values of C3 and C4 than non-splenectomized patients . Serum complement components C3 and C4 were consistently reduced in our patients , and this can be either due to reduced synthesis or increased consumption ; the latter is more probable with the rate of infection in patients which seems to be high . Our observation of significantly lower C3 and C4 levels early in life in thalassemic patients , however , points to the possibility of deficient complement synthesis may also act as a contributing factor to immune deficiency . The controversy concerning alteration of serum complement levels in B- thalassemia patients may be due to marked heterogeneity of the patients in different studies , and this heterogeneity concerns race , socioeconomic class , nutritional status and environmental factors ( Vergin et al . , 1997 ).

The table(2) results revealed increases in alph -1 antitrypsin levels in B-thalassemic patients , while there is no differences between patients and control groups in values of alph-2 macroglobulin . Romeo et al . , (1983) illustrated that the mean concentration of alph-1 antitrypsin was significantly increased in thalassemic patients . Acommon variant ( polymorphism ) , of alph-2 macroglobulin leads to increased risk of Al zheimer's disease , also its levels are increased in nephritic syndrome ( Blacker et al . , 1998 ; Kavacs , 2000) . Individuals inheriting two B- thalassemic alleles experience deficit in beta – chain production , which lead to excess production of alph- globulin ( Luca et al . , 2010) . Furthermore , excess alpha – globulin chains in B- thalassemia is
creating tetramers of hemoglobin that accumulate and precipitate in the erythroid progenitors, forming inclusion bodies that cause oxidative membrane damage within the RBCs and immature developing erythroblasts in the bone marrow (Libani et al., 2008).

Conclusions:

These results can be due to continuous exposure to antigens, repeated infections, chronic liver disease and splenectomy. The only probable cause of humoral immune deficiency found in these patients is a defect in serum complement levels, while there is an increase in alpha-1 antitrypsin level.

References:


