Red Cell Alloantibodies in Multi - Transfused Patients of Beta Thalassaemia Major

Madeeha Rehan*, Atifa Shuaib**, Lubna Zafar*
*Department of Pathology, Foundation Medical College, Rawalpindi; **Department of Pathology, Rawalpindi Medical College, Rawalpindi

Abstract

Background: To study the frequency and specificity of various red cell alloantibodies in multi-transfused patients of beta thalassaemia major.

Methods: In this comparative study two hundred diagnosed cases of Beta Thalassemia Major, of all ages and both genders, who received at least ten blood transfusions, were included. Patients with known alloantibodies, or with known autoimmune diseases, patients with known renal or liver disorders and with viral infections were excluded. Five ml venous blood was taken in a clear glass tube. Sera were separated and blood grouping and direct and indirect anti-globulin tests were performed on all samples. Samples positive for indirect anti-globulin test were stored in aliquots at -20 °C, till the tests were performed in batches. After thawing, antibody screening and antibody identification tests were performed in batches of ten on all indirect anti-globulin test (IAT) positive samples employing 11 cell panel of DIA-MED using standardized blood bank methods. All the results were recorded. The alloantibodies detected and their specificity was determined.

Results: Out of 200 patients six patients (3%) were diagnosed to have alloantibodies which belonged to Rh system i.e. Anti-E and Anti-D.

Conclusion: Rate of alloimmunization is low in our setup in multi-transfused patients of beta thalassaemia major. The most common alloantibody found in our patients is Anti-E.

Key Words: Beta Thalassaemia Major, Transfusion dependent anaemias, Red cell alloantibodies.

Introduction

Thalassaemias are heterogeneous group of genetic disorders of hemoglobin synthesis which result from a reduced rate of production or absence of one or more of the globin chains of hemoglobin. Beta thalassaemia major is a severe transfusion dependent, inherited anemia in which there is a profound defect of beta chain production. Excess alpha chains precipitates and accumulate in the red cell precursors in the bone marrow resulting in ineffective erythropoiesis. There are many mutations which causes thalassaemia and related disorders. These mutations can affect every step in the pathway of globin gene expression, transcription, processing of the mRNA precursor, translation of mature mRNA, and preservation of post translational integrity of the Beta chain. More than 200 mutations in beta chain have been described. The curative standard treatment for Beta Thalassaemia Major is stem cell transplant which is highly specialized and expensive procedure thus not affordable by most of the patients. The main and commonly practicing treatment modality for homozygous Beta-Thalassaemia remains regular blood transfusion along with iron chelation to remove the antecedent transfusional iron overload.

Alloimmunization of red blood cells in multiple transfused patients of beta thalassaemia major complicates the transfusion therapy in these patients. Alloimmunization of red blood cells is dependent on genetic and acquired patient related factors, number of transfusion received and immunogenicity of the antigen. Heterogeneity of population, lack of better matched donor for thalassaemic patients and use of post storage blood may contribute to high alloimmunization rate. RBC alloimmunization has a significant negative impact on laboratory and in institutional resources, associated with a need for increased laboratory testing difficulties in identification and procurement of compatible blood units for transfusion, and with the evaluation and management of transfusion reactions. Antibody screening tests are not routinely performed before transfusion in patients of thalassemia. As a result undetected antibodies at the time of new transfusion can cause hemolytic transfusion reactions.

Patients and Methods

This descriptive study was carried out at Thalassaemia treatment centre, Holy Family Hospital Rawalpindi. Two hundred diagnosed cases of Beta Thalassemia Major, of all ages and both genders, who received at
least ten blood transfusions, were included. Patients with known alloantibodies, or with known autoimmune diseases, patients with known renal or liver disorders and with viral infections were excluded. Total number of transfusions received, transfusion interval and annual transfusion requirement were noted. Five ml venous blood was taken in a clear glass tube. Sera were separated and blood grouping and direct and indirect anti-globulin tests were performed on all samples. Samples positive for indirect anti-globulin test were stored in aliquots at -20 °C, till the tests were performed in batches. After thawing, antibody screening and antibody identification tests were performed in batches of ten on all indirect anti-globulin test (IAT) positive samples employing 11 cell panel of DIA-MED using standardized blood bank methods. All the results were recorded. The alloantibodies detected and their specificity was determined.

**Results**

Out of 200 patients of Beta-Thalassaemia Major 107 (53.5%) were females and 93 (46.5%) were males. The age range was from 2 years to 22 years with the mean age of 8.1 year. Maximum number of patients in this study belonged to 6-10 years of age Haemoglobin concentration ranged from 4.3 to 10.2 gm/dl with a mean concentration of 7.0 gm/ dl. In total of 200 cases studied, all IAT positive cases were subjected to alloantibody screening and identification test. Red cell alloantibodies were detected in 6 (3%) patients (Table 1). Anti-E was detected in 4 patients and Anti-D was present in 2 patients. (Table 2)

**Table 1: Age and sex distribution of alloantibodies**

<table>
<thead>
<tr>
<th>Age</th>
<th>No. of Patients</th>
<th>Alloantibodies</th>
<th>Males</th>
<th>Females</th>
</tr>
</thead>
<tbody>
<tr>
<td>Upto 5 years</td>
<td>42</td>
<td>None</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td>5-10 years</td>
<td>119</td>
<td>4</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>10-15 years</td>
<td>31</td>
<td>2</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>15-20 years</td>
<td>6</td>
<td>None</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td>&gt;20 years</td>
<td>2</td>
<td>None</td>
<td>None</td>
<td>None</td>
</tr>
</tbody>
</table>

**Table 2: Type and Specificity of alloantibody detected**

<table>
<thead>
<tr>
<th>Type of antibody detected</th>
<th>Frequency of antibody detected</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anti E</td>
<td>2%</td>
</tr>
<tr>
<td>Anti D</td>
<td>1%</td>
</tr>
</tbody>
</table>

**Discussion**

According to present study, the frequency of red cell alloimmunization in thalassaemic patients receiving multiple blood transfusions in our set up is low i-e, only 3%. Our results are in agreement with a study conducted by Bhatti F, et al. at AFIT Rawalpindi. They reported RBC alloimmunization as 4.97% in 161 patients which mainly belonged to Rh system and Kell system.10, 13 Our data contrast with a study conducted by Hassan K et al, which showed higher rate of alloimmunization in patients of Beta thalassaemia major. Alloantibody was detected in 22.7% patients out of total 75 patients, and the most frequent alloantibody was found to be Anti-Kpa.12 A study at Aga Khan University Karachi by Bilwani F, et al, revealed a rate of alloimmunization of 9.2% in 97 patients of thalassaemia major. Anti-k was the commonest followed by non-specific and anti-D.11 In southern Iran and Shiraz 711 thalassaemic patients were studied by Karimi M, et al and in these patients 5.3% were found to have alloantibodies. The most common alloantibodies were Anti-kell>Antih-Rh (D) >Antih-Rh (E).14 Wang LY, et al, in Taiwan revealed that of the 30 thalassaemic patients included in the study 11 (37%) were found to have clinically significant alloantibodies, and most common antibody detected was Anti-E.6 In a study carried out in Saudia Arabia the development of alloantibodies were determined in 68 multi-transfused patients and the overall frequency of alloantibody formation in our patients is 22.06%.15 However in another study carried out in India the overall incidence of RBC alloimmunization in transfused patients was 3.4% with anti-c being the most common specificity (38.8%), followed by anti-E (22.2%), anti-M (11.1%), anti-Le(a) (11.1%), anti-D (5.6%), anti-Jk(a) (5.6%) and anti-Le(b) (5.6%).16

If prevalent transfusion practices continue the problem of alloimmunization can be significantly increased.12 Alloantibodies against red blood cells should not be overlooked in patients of Thalassaemia receiving multiple blood transfusions.11 Ideally genotyping and cross matching of red blood cells of major and minor blood groups should be done in patients receiving multiple blood transfusions.9

**Conclusion**

1. Rate of red blood cell alloimmunization in multiply transfused patients of Beta-Thalassaemia major is low in our setup. The commonest alloantibody found in thalassaemic patients in our setup is Anti-E.
2. The alloantibodies screening in transfusion dependent thalassaemias is imperative in cases with
difficulties in cross matching, having haemolytic transfusion reactions and in those who failed to maintain a desirable haemoglobin level.

References