

Prevalence of Red Blood Cell Alloantibodies in Blood Donors of Zanjan Province; the Preliminary Report of the North West of Iran

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Red blood cell (RBC) alloantibodies, especially IgG class, are clinically significant because of their association with hemolytic disease of newborns (HDN), hemolytic transfusion reaction (HTR) and a significant reduction in the lifespan of transfused red blood cells. The frequency of RBC alloantibody was variously reported due to difference in selected population and laboratory test sensitivity from less than 0.5% to up to 60%. In the presence of these mentioned alloantibodies, preparation of antigen-negative blood products for transfusion is necessary. In this cross-sectional study, 75 donors (73 males and 2 females) participated. ABO blood group typing was performed via two methods (cell type using anti-A, anti-B and anti-D and also the reverse or serum type using defined A and B cell suspensions). The serum samples were subjected to antibody screening test utilizing commercially available kits containing three vials of RBC with known surface antigen. The results were recorded and for donors with positive results, antibody identification test was also performed using a panel of 11 RBC with known surface antigen. In this study, of 75 donors, 73 (97.33%) were male and 2 (2.66%) were female. Participated donors age range was between 19 and 66 years. The frequency of blood groups in this population were A (52%), O (29.33%), B (14.66%) and AB (4%). and also Rh⁺(97.33%) and Rh⁻(2.67%). Alloantibodies were detected in serum of 6 donors (8%). Identified alloantibodies were against K, c and e antigens. These antibodies were only identified in samples of men may be due to small sample size of women. No significant correlation between donor's age and type of alloantibodies were detected. The prevalence of alloantibodies in Zanjanian population was 8%. Therefore, detection and identification of these antibodies in the donor population could be essential to prevent adverse hemolytic reaction in case of blood transfusion.

Key words: Alloantibody, Blood donor population, Antibody screening, Antibody identification.

When incompatible blood transfusions due to red blood cells (RBCs) injected to stimulate an immune response in the person receiving the blood and immune antibodies are produced. The antibodies that are produced in response to foreign antigens is called alloantibodies^{1,2}. Red blood cell

(RBC) alloantibodies, especially IgG class, are clinically significant because of their association with hemolytic disease of newborns (HDN), hemolytic transfusion reaction (HTR) and a significant reduction in the lifespan of transfused red blood cells. The frequency of RBC alloantibody was variously reported due to difference in selected population and laboratory test sensitivity from less than 0.5% to up to 60%^{3,4}. The presence

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of red blood cells (RBCs) antibodies in sera, preparation of antigen-negative blood products for transfusion is necessary⁵. Properly blood transfusion can save lives of patient's, but if the guidelines transfusion is not regarded, it can cause serious complications and even be lethal. The frequency of these side effects has been reported in 5.3% of recipients of blood⁶. Identification antibodies in individual who are at the diagnosis of the physician needs to recipient the blood, helps with the selection of suitable and antigen -negative products for the transfusion, the risk of complications of incompatible blood transfusions greatly reduced⁷.

Antibody screening test is a simple and inexpensive method to identification a large number of unwanted and important antibodies; which are generally against minor blood group antigens. In this method, the recipient serum samples with 2-5% suspension 11 different O blood group of red blood cells, that are display antigen of important blood groups, is incubated. These antigens include blood group D, C, E, c, e, M, N, S, s, P1, Lea, Leb, K, k, Fya, Fyb, Jka and Jkb. Identification of available antigens and compatible blood transfusions may help prevent alloimmunization⁸.

Because of the importance and prevalence of antibodies against minor blood groups in individuals and have not statistics of the prevalence of antibodies in the normal population, we decided to examine frequency of alloantibodies in donor blood transfusion.

MATERIALS AND METHODS

In this cross-sectional study, 75 donors (73 males and 2 females) participated. ABO blood group typing was performed via two methods (cell type using anti-A, anti-B and anti-D and also the reverse or serum type using defined A and B cell suspensions). The serum samples were subjected to antibody screening test utilizing commercially available kits containing three vials of RBC with known surface antigen. For antibody screening, the serum from the clot was separated by centrifugation at 3000 RPM for 10 minutes, and then in three separate tubes added two drops of serum and a drop of cell suspension added them. The tubes were centrifuged for 30 seconds and the agglutination and hemolysis was checked and

2 drops of 22% albumin added to the tubes and the tubes have incubated for 30-15 minutes at 37 C. Then the tubes were centrifuged at 3000 RPM for 30 seconds and the agglutination and hemolysis were checked. Then the tubes washed 3 times with saline and completely dried up, and each tube 2 drops of Anti-Human Globulin commercially available added. The tubes were centrifuged at 3000 RPM for 30 seconds and the agglutination and hemolysis were checked. The results were recorded and for donors with positive results, antibody identification test was also performed using a panel of 11 RBC with known surface antigens (D, C, E, c, e, f, V, C^w, M, N, S, s, Lu^a, Lu^b, P1, Le^a, Le^b, K, k, Fy^a, Fy^b, Jk^a and Jk^b). In the end, the results obtained were analyzed.

RESULTS

In this study, of 75 donors, 73 (97.33%) were male and 2 (2.66%) were female. Participated donors age range was between 19 and 66 years. The frequency of blood groups in this population were A (52%), O (29.33%), B (14.66%) and AB (4%). and

Table 1. Prevalence of Alloantibody in Zanzanian population

Alloantibody	Total
K	4 (%66.66)
c	1 (%16.66)
e	1 (%16.66)

Table 2. Gender distribution in Zanzanian population

Gender	Total
Male	73 (%97.33)
Female	2 (%2.66)

Table 3. The frequency of blood groups in Zanzanian population

Blood Groups	Total
A ⁺	37 (%49.33)
A ⁻	2 (%2.66)
B ⁺	11 (%14.66)
AB ⁺	3 (%4)
O ⁺	20 (%20.66)
O ⁻	2 (2.66)

also Rh⁺(97.33%) and Rh(2.67%). Alloantibodies were detected in serum of 6 donors (8%). Identified alloantibodies were against K, c and e antigens. These antibodies were only identified in samples of men may be due to small sample size of women. No significant correlation between donor's age and type of alloantibodies were detected.

DISCUSSION

The prevalence of alloantibodies in Zanjanian population was 8%. In a study conducted in Minnesota region on normal population between 1975 and 1995, the rate of prevalence of alloantibody against Red cell antigens was less than one percent. the most prevalent antibody was against E, Le and K antigens⁹. In another study, in Kuwait, the prevalence of alloantibodies was % 0.49. The prevalence of alloantibodies in the population of Kuwaiti women was higher than men¹⁰. In our study, alloantibodies were only identified in samples of men may be due to small sample size of women. In a study conducted in Tehran (Iran) between 1997 and 1998, No significant correlation between gender and the incidence of alloantibodies not found¹¹. In a study conducted in the blood bank of University Kebangsaan Malaysia Medical Centre in 2010, the prevalence of alloantibodies was %0.76. The most prevalent antibody was Anti-Mai (%30.4). Anti-E and anti-c were the most common combination of multiple alloantibodies¹². In another study, in Eastern India, in 2015, the prevalence of alloantibodies was %5.6. The most prevalent antibody was against c antigen (%28.75)¹³. In Iran, study about the prevalence of alloantibodies in the normal population has not been done. In a study conducted in Tehran between 1997 and 1998, the rate of prevalence of alloantibodies in patients referred to the hospital for surgery was %0.97. In this study, the most prevalent antibody was against Rh (Anti-E and Anti-c) and Kell (Anti-K) antigens¹¹. In a study conducted in Imam Khomeini hospital on outpatient surgery in 2013, the rate of prevalence of alloantibody against Red cell antigens was % 0.9¹⁴. In a study conducted in southwestern Iran in 2013, in patients with thalassemia major and Intermediate, the rate of prevalence of alloantibodies was % 18.7 and the rate of prevalence of autoantibodies was %12.7. In

addition to the alloantibodies, autoantibodies are also a source of danger and must be evaluated. In this study, the most prevalent antibody was against Rh (%55) and Kell (%33) antigens¹⁵. In a study conducted in northeastern Iran in 2009 on 133 in patients with thalassemia disease is detected 12 alloantibodies in 9 patient that these antibodies were against Rh (D, C and E) antigens. The most commonly detected antibodies were against D (88/88%) antigens¹⁶. The most prevalent alloantibody was against K (% 66.66), c (% 16.66) and e (% 16.66) antigens in Zanjanian population (our study). In a study conducted in Ardabil, the most prevalent of alloantibody was against K (%30), E (%15) and c (%15) antigens¹¹. In a study conducted in southeastern Iran in patients with thalassemia major, the rate of prevalence of alloantibody was %17.9 that these antibodies were against Rh and Kell antigens¹⁷. In a study conducted at four hospitals in Tehran, in 3092 patients referred to the hospital for surgery have been identified 30 alloantibodies Identified. Alloantibodies were Anti-Kell (% 23.53), Anti-E (% 20.59), and Anti-c (% 17.56)¹⁸. In a study conducted in Yazd (Iran) Iran in patients with beta thalassemia, the rate of prevalence of alloantibody was %4 that these antibodies were against Kell, C and D antigens¹⁹. In another study from northwest of Iran, in 2016, in patient with beta thalassemia major, the rate of prevalence of alloantibody was %16.32 that these antibodies were against Kell, E and c antigens²⁰.

CONCLUSION

The prevalence of alloantibodies in Zanjanian population was 8%. Therefore, detection and identification of these antibodies in the donor population could be essential to prevent adverse hemolytic reaction in case of blood transfusion.

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