Frequency and specificity of red blood cell alloantibodies among blood transfusion recipients in Specialist Hospital, Sokoto

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Abstract

Background: Blood transfusion usually results in production of alloantibody against one or more foreign red blood cell antigens which may complicate subsequent transfusions. The probability of alloimmunization depends on number and frequency of transfusion, antigen immunogenicity, recipient immune response and ethnicity. Studies have demonstrated varied frequency of alloimmunization after multiple blood transfusions. The paucity of information on the foregoing in our environment prompted this study.
Methods: A cross-sectional descriptive survey of the frequency and specificity of alloantibodies among transfused patients. Using their clinical records and blood transfusion history, data were analyzed with reference to sex, date of birth, history of transfusion, surgery and pregnancy.
Results: A total of 150 transfused participants, in Specialist Hospitals, Sokoto were studied. Overall alloantibody positivity was 17.3% with females accounting for 96% and males 4%. The three most frequent alloantibodies were anti-K (34.62%), anti-E (23.08%) and anti- C (15.39%). Most common clinically significant alloantibody identified in men and women were anti-K and anti-E, respectively. The most common causes of alloimmunization for men and women were surgery and pregnancy related blood transfusion respectively.
Conclusions: Alloimmunization against RBC antigens among blood transfused patients is common. Most common alloantibodies identified were anti-K and anti-E in females and anti-K in male and female respectively. The testing of blood donors for red cell antigens and antibody screening of recipients will be rewarding.

Key words: Transfusion, RBC, Alloantibody, Sokoto

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Introduction

Red blood cell (RBC) transfusion is a life-saving therapy for severe anaemia of varied aetiologies, but the risk of RBC alloimmunization remains a major cause for concern in patients receiving blood transfusion[1]. Alloimmunization occur because of antigen disparity between donor and recipient as no two individuals except identical twins have the same genetic make-up. Most studies on patients who had repeated blood transfusion found that alloantibodies significantly posed difficulties in cross-matching of blood during transfusion[2]. It is therefore imperative to know sufficiently about these alloantibodies prior to transfusion of RBC[3]. Although the general public is worried about the infection risks of transfusion, haemovigilance reports show that antigen-antibody reactions are responsible for the enormous majority of acute and delayed transfusion reactions[4].
Alloantibody test should be considered as additional testing in all transfusion as a routine to prevent possible incidence of alloimmunization[5,6]. It would also be cost-effective if such screening is carried out before transfusion in order to provide compatible blood for alloimmunized patients. With the increase in life expectancy associated with technological developments, increases in the number of chronic-degenerative disease and more complex surgeries and increase in complications of pregnancies, higher numbers of blood transfusions have been observed. In a study conducted by Erabor[7], to find out the participation of female gender in blood donation in Sokoto, observed that about 14,965 units of blood were donated for transfusion in a tertiary hospital in a period of two and half years with a high cross matched to transfusion ratio[8]. The frequency of RBC alloantibodies other than the ABO system was also reported to be on the increase[9]. This often results in difficulties in finding compatible blood and a higher risk of delayed haemolytic transfusion reactions[10]. However, in most blood transfusion services particularly in sub-Saharan Africa, phenotyping for clinically significant red cell antigens on the donor's red cells are not routinely done and the facilities for alloantibodies testing are often not available[11].

There is paucity of data on the prevalence of clinically significant alloantibodies among patients in whom red cell transfusion is indicated[7]. Family replacement blood donation continues to thrive in Sokoto putting especially women potentially at risk of developing alloantibody to antigens present in donor

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from their spouses and in-laws[11]. This potentially puts her future pregnancies at risk of haemolytic disease of newborn. There is increasing public awareness of the possible complications of blood transfusion with intense media attention focusing particularly on the risk of transfusion transmitted infections[12]. The result is that the public recognize that blood transfusion is becoming more and more unsafe, whereas the reality is that blood transfusion has probably never been safer[13]. This public concern is also misdirected, as the majority of potential blood recipients are aware of possible infectious complications but oblivious to the risks of incompatible transfusion[14].

Alloimmunization to clinically significant red cell antigen is a major complication observed among pregnant women and multiple-transfused patients making it difficult to source compatible blood units for transfusion[15]. Transfusion of incompatible RBCs may lead to an immune-mediated haemolytic transfusion reaction, and to avoid such cases, pre-transfusion compatibility testing is rewarding. To the best of our knowledge *there has been no previous study on frequency & specificity of RBC alloantibodies as consequence of transfusion of whole blood among blood recipients in Sokoto.* This study aims to determine the prevalence and specificity of alloantibodies that has the potential to cause haemolytic disease of newborn and haemolytic transfusion reaction in patients in whom red cell transfusion is indicated. The rate of RBC alloimmunization depends on the characteristics of the population being studied[16]. This study, apart from adding to knowledge, will provide valuable information for management of transfusion reactions in this environment. The data generated could justify the advocacy for the implementation of a programme on routine antenatal prophylaxis for Rh-negative pregnant women[17]in Sokoto, North Western Nigeria.

Materials and Methods

It was a hospital-based descriptive cross-sectional survey carried out between May and October 2015 among one hundred and fifty heterogeneous patients of different age groups, gender and educational status, attending specialist hospital Sokoto. An ethical clearance was obtained from the specialist hospital ethical committee with the assurance that all information gathered would be treated with utmost confidentiality. Four miles of blood samples was collected from each patient after observing the standard technique. The data was analyzed using a statistical package for social sciences (SPSS) version 20. Frequencies and percentages of results were expressed in mean and standard deviation. Differences in values based on socio-demographic variables of subjects was determined and compared statistically. A p-value of < 0.05 was considered as significant in all statistical comparisons.

Results

A total of 150 blood recipients aged between 1-80 years were studied, comprising of 19 (12.7%) male and 131 (87.3%) female with the mean age of 31.73±12.98. All the 26 (17.33%) who were positive for red blood cell alloantibodies had clear specificity, (Table1) (96.15) females and (3.85%) male.

Table 1: Prevalence of alloantibodies among transfusion recipient of different age groups

Age(years)	No recipients tested	No. Positive (Percent of total)
1-10	5	0 (0)
11-20	20	4 (15.4)
21-30	69	8 (30.8)
31-40	33	9 (34.6)
41-50	8	1 (3.85)
51-60	9	1 (3.85)
61-70	5	3 (11.5)
71-80	1	0 (0)
>80	0	0 (0)
Total	150	26 (100)

$X^2 = 4.89$, P value= 0.17

Table 2: Relation of various factors with production of alloantibody in study patients

Related factor	Presence of alloantibody n=150	Negative n=124	Positive n=26	P value for each factor
Gender	Male (n=19)	18 (95)	1 (5)	0.11
	Female (n=131)	106 (81)	25 (19)	
Transfusion history	Negative (n=73)	67 (92)	6 (8)	0.04
	Positive (n=77)	57 (74)	20 (26)	
Transfusion reaction history	Negative(n=130)	110 (85)	20 (15)	0.74
	Positive (n=20)	14 (70)	6(30)	
Surgery history	Negative(n=102)	88(86)	14(14)	0.21
	Positive (n=48)	36(75)	12(25)	
Pregnancy history	Negative (n=25)	24 (96)	1 (4)	0.03
	Positive (n=125)	100 (80)	25 (20)	
Abortion history	Negative(n=141)	116 (82)	25 (18)	0.61
	Positive (n=9)	8 (89)	1 (11)	
Miscarriage history	Negative (n=86)	74 (86)	12 (14)	0.20
	Positive (n=64)	50 (78)	14 (22)	
Stillbirth history	Negative (n=98)	82 (84)	16 (16)	0.61
	Positive (n=52)	42 (81)	10 (19)	

**Values in parentheses are percentages, Gender $\chi^2 = 2.45$, Transfusion history $\chi^2 = 8.24$, Transfusion reaction history $\chi^2 = 0.12$, Surgery history $\chi^2 = 1.53$, Pregnancy history $\chi^2 = 8.45$, Abortion history $\chi^2 = 0.25$, Miscarriage history $\chi^2 = 1.60$, Stillbirth history $\chi^2 = 0.25$,

Patients between the ages of 21-30 years formed the majority representing 69(46%). Seventy seven (51.33%) had positive history of blood transfusion while 73 (48.67%) had no history of transfusion (Table 2). Of the subject with no history of transfusion, 6 (8%) were positive for alloantibodies, while 20 (26%) with transfusion history were positive for alloantibodies (Table 3).

Table 3: Distribution of RBC alloantibodies (26) among alloantibody positive patients (n=26)

Alloantibody	Male (%) (n=1)	Female (%) (n=25)	Total (%) (n=26)
K	1 (100)	8 (32.00)	9 (34.62)
E	0	6 (24.00)	6 (23.08)
C	0	4 (16.00)	4 (15.39)
Leb	0	6 (24.00)	6 (23.08)
Fya	0	1 (4.00)	1 (3.85)
Total	1 (3.85)	25 (96.15)	26 (100)

(n=26) 'n' represents no. of patients with a type of antibody
Key: K:Kell antibody, E: Epsilon antibody, C: Kidd antibody, Le^b: Lewis antibody, Fy^a: Duffy antibody

The study also showed a significant difference in respect of development of alloantibodies among the study subject with history of transfusion and pregnancy as the p value was<0.05 which was not the case with the history of miscarriage, transfusion reaction, abortion and stillbirth (p>0.05) (Table 3).

A total of 26 RBC alloantibodies were identified in 26 patients, (3.85% in male, and 96.15% in female). Of these, 21 (81%) had one alloantibody and 5 (19%) had two antibodies. Five alloantibodies were detected with four having clinically significant RBC alloantibodies, listed by specificity and frequency as anti-K with 34.62%, anti-E with 23.08 %, anti-c with 15.39 %.

Overall, the most frequently identified alloantibody in men, as a percentage of total male antibodies, was anti-K with 3.85 %; and the most frequently identified alloantibody in women, as a percentage of total female antibodies, was anti-K with 32%, followed by anti-E and anti-Ly_b with 24% each.

Discussion

RBCs are highly polymorphic, vested with a multitude of surface antigens that are known to cause immune transfusion reactions of varied magnitude in different individuals. Thus it is a common practice that pre-transfusion compatibility testing is done to prevent these undesirable effects of transfusion[3]. Antibody screening and identification is usually carried out to determine the specificity of antibodies detected atypically, thus enabling choice of donor RBC units most suitable for the recipients[4,18]. Alloimmunization to clinically significant red blood cell antigen is a major complication associated with immunohaematological services in developing countries. The daunting challenges militating against effective immunohaematological service include absence of phenotype units for multiply transfused patients, reliance on family blood replacement donors, commercially remunerated blood donors, absence of universal alloantibody testing amongst ante-natal women and suboptimal management of clinically significant antibodies in pregnancy that is associated with haemolytic disease of the new born. Therefore, to avoid ABO-incompatibility in blood transfusions and unwarranted exposure to Rh D positive RBCs, determination of ABO group, Rh D typing, antibody detection, and antibody identification, and cross-matching are strongly advocated[3].

In this study, the overall prevalence of alloantibody was 17.3% in which 3.85% of patients who demonstrated alloantibodies were males, while 96.15 % were females. In other words, 1 out of 19 males demonstrated presence of alloantibodies and 25 out of 131 females showed presence of alloantibodies in their sera. The obtained results indicated a higher preponderance of RBC alloimmunization in females compared to males. The results of this study indicting history of pregnancy and transfusion history as some of the most probable predispositions for the alloimmunization in females and males, agreeing with India study, where such factors were similarly indicted[19]. There was statistical significant difference (p 0.03) between alloantibodies in patients with pregnancy history compare to those without. Of the 125 participants with positive history for pregnancy 25 (20 %) were positive for the demonstration of alloantibodies, thus indicating that pregnancy is an important factor for alloimmunization and development of alloantibodies. There was also statistical significant difference (p 0.04) in the demonstration of alloantibodies in participants with transfusion history than those without. Since 51% of patients with alloantibodies had transfusion history, it seems that transfusion has an important role as an alloimmunization factor. In this study the most frequently shown alloantibody was anti-K followed, by anti-E and anti-Leb, This was in close agreement with the result of another study in Europe by Pahuja, *et al*[19], where the most frequent alloantibodies in alloimmunized patients were alloantibodies against Rh and Kell antigens.

In another study conducted by Winters, *et al*[20], in the USA, anti-E and anti-K were the most frequently observed antibodies in alloimmunized patients, further supporting the result of this study with regards to anti-K. Use of different enhancement methods with regards to the choice of panel of cells and optimization of screening procedures could be considered as an explanation for

these observed differences. In studies using methods that enhance detection of antibodies to Rh specificities, anti-E is detected more frequently[21,22]. This study demonstrated that anti-E was the second most common alloantibody among females, followed by anti-Le_b and anti-c.The high frequency of Lebin women during pregnancy observed in this study was comparable to what has been shown by other studies[20]. During pregnancy, Lewis antigens become weaker, which occurs most often through associated appearance of anti-Leb.

Conclusion

This study has shown that the rate of alloimmunization against RBC antigens among transfused patients in specialist hospital is indeed high. The most common alloantibodies identified in the sera of these patients were mostly of the anti-K and anti-E specificity. In addition to the routine determination of ABO group, Rh D typing, antibody screening and identification, should be included as part of the routine cross-matching procedure. A policy that obligates transfusion of ABO, and Rh D-compatible blood for fertile women should be advocated in State and private hospitals. ABO, Rh grouping and alloantibody screening test for every woman of child bearing age should be mandatory. Intensive compatibility testing to rule out other RBC antigens in a bid to avoid development of RBC antibodies in patients who chronically received blood transfusion is strongly advocated.

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