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Significance of Antibody Screening and Identification in Pretransfusion Testing-A Retrospective Study

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Abstract: ***Pretransfusion compatibility testing has gone through many changes. To provide safe blood for transfusion does not merely imply thorough testing for infectious markers, but also protection from haemolytic transfusion reactions resulting from alloimmunization against red cell antigens. Continual efforts to improve blood safety have led to introduction of regular screening protocols for detection of unexpected immune antibodies at the various transfusion centre across the globe. The ultimate goal is to determine the exact specificity of the antibody and to provide blood that lacks the corresponding antigen to the patient4. The frequently detected unexpected Blood cell alloantibodies in daily transfusion practice are directed towards the Rh (D, C, E, c & e) and Kell (K) antigens ,followed by other blood group antigens of the Duffy, Kidd, MNS & other minor blood group system5. These unexpected antibodies are responsible for acute and delayed haemolytic transfusion reactions as well as haemolytic disease of foetus and newborn5,6. To determine the frequency of unexpected red cell antibodies, data of antibody screening from Jan 2014 to Dec 2014 were retrieved from case records at department of transfusion medicine, Sakra world hospital, Bangalore and assessed for the presence of alloantibodies. All cases underwent antibody screening and if found positive were subjected to antibody characterization /identification. All antenatal women and only autoantibodies were also included in the study. Antibody screening was performed using commercial cell panel (ID Diacell of Bio-Rad) by Anti human globulin method. In case of a positive antibody screen, further testing was done using extended cell panel, to precisely characterise the unexpected antibody (ies).***

Keywords: Auto -Antibodies, Compatibility Testing

1. Introduction

Pretransfusion compatibility testing has gone through many changes. In early 1900s, when blood transfusion was initially practiced, major crossmatches (that is donor's erythrocytes tested against the recipients serum) as well as minor crossmatches (that is donor serum tested against the recipients erythrocytes) were considered necessary. In the 1950s, anti-human globulin reagent was being used in immune haematological tests, and knowledge about red blood cell antigens proliferated. Furthermore, as increasing numbers of individuals around the world were receiving transfusions, the body of knowledge about transfusion therapy and clinical significance of crossmatching is also increased. There was a growing consensus that compatibility testing could be simplified without increasing the risk of transfusing incompatible blood [1]

In 1981, during a meeting of the Blood Products Advisory Committee of the Food and Drug Administration (FDA) of the United States, studies were presented that showed a 1 in 17000 chance of missing a clinically significant antibody when antibody screening was incorporated into the compatibility tests[2].As a result of meeting ,the FDA's Office of Biologics Research and Review issued a memorandum allowing the major cross match step to be eliminated ,provided that the recipient's serum to be tested for unexpected alloantibodies by an "equally sensitive method that demonstrates clinically significant antibodies reactive at 37 deg centigrade[3.]

To provide safe blood for transfusion does not merely imply thorough testing for infectious markers, but also protection from haemolytic transfusion reactions resulting from alloimmunization against red cell antigens. Continual efforts to improve blood safety have led to introduction of regular screening protocols for detection of unexpected immune antibodies at the various transfusion centre across the globe. The ultimate goal is to determine the exact specificity of the antibody and to provide blood that lacks the corresponding antigen to the patient[4]. The frequently detected unexpected Blood cell alloantibodies in daily transfusion practice are directed towards the Rh(D,C,E,c & e) and Kell (K) antigens ,followed by other blood group antigens of the Duffy,Kidd ,MNS & other minor blood group system[5]. These unexpected antibodies are responsible for acute and delayed haemolytic transfusion reactions as well as haemolytic disease of foetus and newborn[5,6]. Alloimmunization occur when a foreign antigen introduced in an immune-competent host evokes an immune response. Thiscommonly occurs following transfusion of blood or in pregnancy, when red cells that bear antigen absent from the individuals own blood enter the circulation[7].

2. Material and Methods

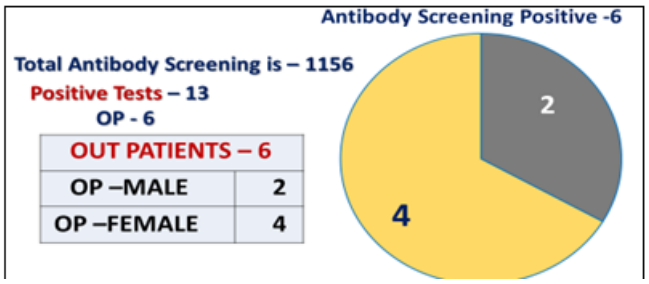
To determine the frequency of unexpected red cell antibodies, data of antibody screening from Jan 2014 to Dec 2014 were retrieved from case records at department of transfusion medicine,Sakra world hospital,Bangalore and assessed for the presence of alloantibodies.All cases underwent antibody screening and if found positive were subjected to antibody characterization /identification. All

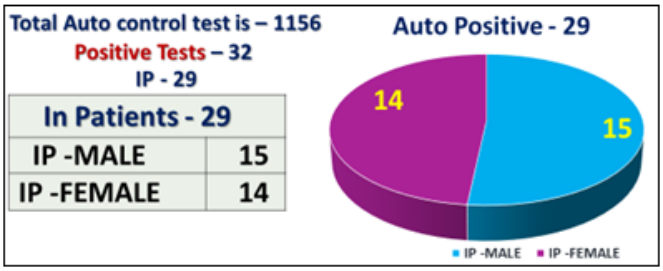
antenatal women and only autoantibodies were also included in the study.

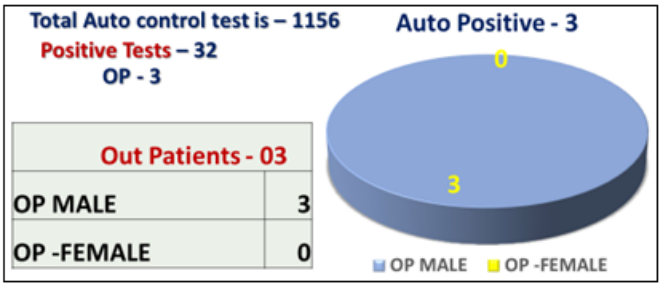
Antibody screening was performed using commercial cell panel (ID Diacell of Bio-Rad) by Anti human globulin method. In case of a positive antibody screen, further testing was done using extended cell panel, to precisely characterise the unexpected antibody (ies).

3. Result

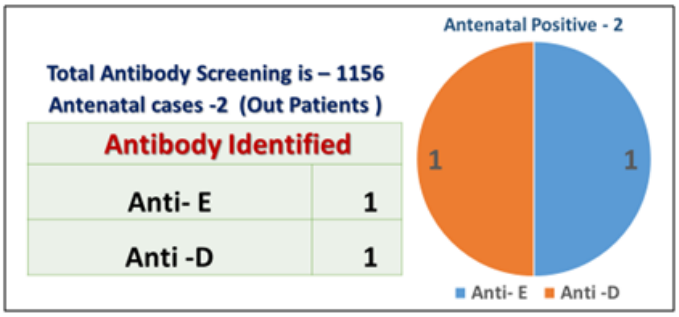
It is a retrospective study in which evaluation of 1912 cases (870:50% males and 1042:54.4%females) done. All samples (patient and antenatal) were screened for the presenceof unexpected antibodies. Antibody screening was positive in 19 patients (0.99%). In the serum samples of 37 patients only autoantibodies were identified, 4 cases revealed autoantibody also with underlying alloantibody. The total alloimmunisation rate was 0.99%, alloimmunisation in antenatal females was 0.15%.Among the antenatal females anti D was the most common (4 cases) Anti E and Anti K one each.

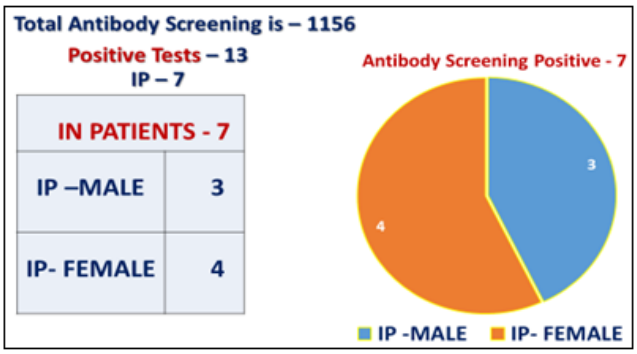




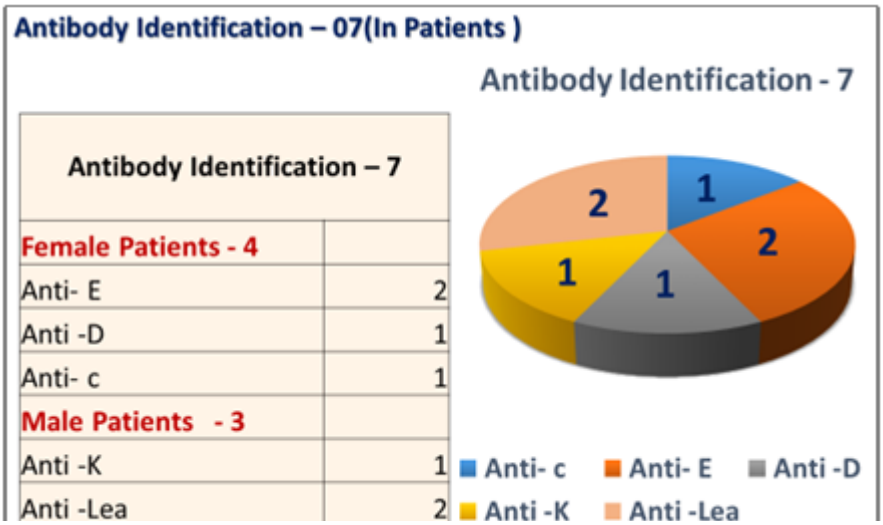


Total Antibody Screening sample – 1156	
IP PATIENTS - 875	
Male	495
FEMALE	380
OUT PATIENTS – 277	
Male	31
FEMALE	246
EMR - FEMALE	1
PHC - FEMALE	3





Study	Year	Total patients tested	Positive	Rate (%)	Most common allo antibody detected
Ko et al[8]	2012	22436	340	1.52	Le
Shin et al [9]	2009	15014	234	1056	Anti E
Lee et al[10]	2000	23735	109	0.46	Anti E+c
Chaudhary & Agarwal[11]	2011	2026	26	1.28	Anti E
Tormey et al[12]	2008	18750	450	2.4	Anti K
Present study	2015	1912	19	0.99	Anti D



4. Discussion

RBC alloimmunization is a result of genetic disparity of red cells between donor and recipient or between mother andfoetus[7].In india information available on alloimunizaion is limited to select patient populations like multitransfused[8], or pregnant women[9] and data on general patients is very limited.The standard pretransfusion testing protocols require detection and identification of clinically significant antibodies reacting in antihuman globulin (AHG) phase after incubation at 37 deg centigrade.

In present study, the overall alloimmunization rate was 0.99 % which was low when compared with a study done by Ko et al[10], Shin et al[11], Chaudhary & Agarwal[13]and Tormey et al[14] .This difference could be due to varied study populations and that our study population comprised general hospital patients and not the high risk groups like the multitransfused. In a similar study by Lee et al[12], prevalence of alloimmunization reported was 0.46 % which was less than our study. Female patients had higher rate of alloimmunization than male in our study. A systematic review by verduin et al[15]also showed that women have slightly higher rate of alloimmunization than men .Higher rates of alloimmunization in females may be attributed to antigenic exposure during pregnancy in addition to transfusions which is the only source of exposure in men. Alloantibodies development can significantly complicate transfusion therapy and result in difficulties in cross-matching of blood. Clinically significant antibodies are capable of causing mild or severe adverse events following transfusion, such as haemolytic transfusion reactions or haemolytic disease of the foetus and newborn[16]. Thus, knowledge of such alloantibodies is essential not only in the multitransfused patients but in all hospital patients who require or may require transfusion. This not only helps in selecting appropriate RBC products for transfusion but also avoid unnecessary delays in provision of blood in case of emergencies or surgical complications.

As it is evident from present study and existing literature [5,6] the most common alloantibodies found were against the common Rh AND Kell antigens(D,C,c,E,e,K). These findings emphasize on the role of extended antigen typing for recipients as well as donors, and the importance of being able to provide Rh and Kell matched blood.

The current practice for providing compatible blood to patients in cases of alloimmunization in many Indian centres is still reliant upon random cross-matching of available units in the inventory.But this practice is neither safe nor cost-effective ,as clinically significant antibodies are frequently detected in our patient population and cross-matching is not fully effective as a procedure of ensuring absence of the antigen to which the patient is immunized[13].Hence antibody screening and identification should be done along with phenotyping donors ,in order to follow safe transfusion practices .

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