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CODEN: IJBABN **Original Research Article**

A study on various red cell antibodies in admitted patients and its gender wise distribution in a tertiary care centre

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Abstract

Background: Clinically significant red cell antibodies are usually of IgG type reacting at 37^0C associated with haemolytic transfusion reactions. IgG type antibodies against red cell antigens are more prevalent in admitted patients, as this group has higher chances of red cell alloimmunization due to higher chances of prior red cell transfusion.
Aims and objectives: 1) To detect various irregular red cell antibodies in the admitted patients. 2) To study the gender wise distribution of various antibodies.
Materials and methods: We prospectively studied samples from 14753 admitted patients who required blood transfusion by performing antibody screening and identification**.** Red cell transfusion history, clinical conditions, blood grouping & antibody screening results were also studied.
Results: Total 276 (3.5%) cases of red cell antibodies were detected with 78 cases in 6359 males and 198 cases in 8394 female patients. Antibodies were grouped as single antibodies (187), multiple antibodies (21), antibodies against low frequency antigens (4), antibodies against high frequency antigens (29), both auto and alloantibodies (3) and no discernible pattern seen (5). The difference in rate of alloimmunization between males and females was found be statistically significant with a p-value < 0.001.
Conclusion: Our study recommends performing phenotyping for Rh system antigens and K antigen for all patients with planned surgeries and patients not needing emergency transfusions. Extended red cell phenotyping should be done for almost all chronically transfused patients and phenotypically matched blood units should be transfused specially in female patients.
Keywords: Admitted patients, Alloantibodies, Alloimmunization

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1. Introduction

Blood transfusion although life saving is associated with inherent risk of alloimmunization to red cell antigens. Red cell transfusion can result in the development of alloantibodies against one or more red cell antigens. The risk of alloimmunization is high in patients receiving multiple blood transfusions. The most important determination for any transfusion is to exclude the presence of clinically significant alloantibodies in the patient's blood before selecting RBC for transfusion. Usually, blood banks provide only ABO and Rh antigens matched blood, so the risk of alloimmunization to minor blood group antigens is very high.

1.1 Aims and objectives

1) To detect various irregular red cell antibodies in the admitted patients
2) To study the gender wise distribution of various antibodies.

2. Materials and Method

This was a prospective study. This study was carried out at the red cell serology laboratory of Department of Immunohematology and Blood Transfusion, BJMC Ahmedabad, over a period of six months from June 2014 to December 2014, this study was approved by institutional ethics committee.

Every day on an average between 200-300 blood samples from both OPD and admitted patients came in blood bank for blood grouping and pretransfusion testing but everyday about 60-150 patients had transfusion. Our study targeted admitted patients requiring blood transfusion, blood samples were collected in pilot tubes (EDTA and Plain) from 14753 admitted patients (n=14753), whose blood samples were sent in the department either for crossmatch or simply for blood grouping for an anticipated blood transfusion. These samples were subjected to blood grouping and antibody screening. Out of 14753 patients 29 patients were readmitted during the period of study with 09 patients having history of blood transfusion during first time admission.

EDTA samples were first centrifuged at 2000 rpm then subjected to blood grouping in the fully automated immunohematology system then again screened for irregular antibodies in the fully automated immunohematology system "Qwalys-3 (Manufacturer "Diagast")". If on first run antibody screening result was positive then it was subjected to second run after centrifugation as false positive results are very common with fully automated antibody screening system [Figure 1].

Figure 1-Flowchart Ab screening final

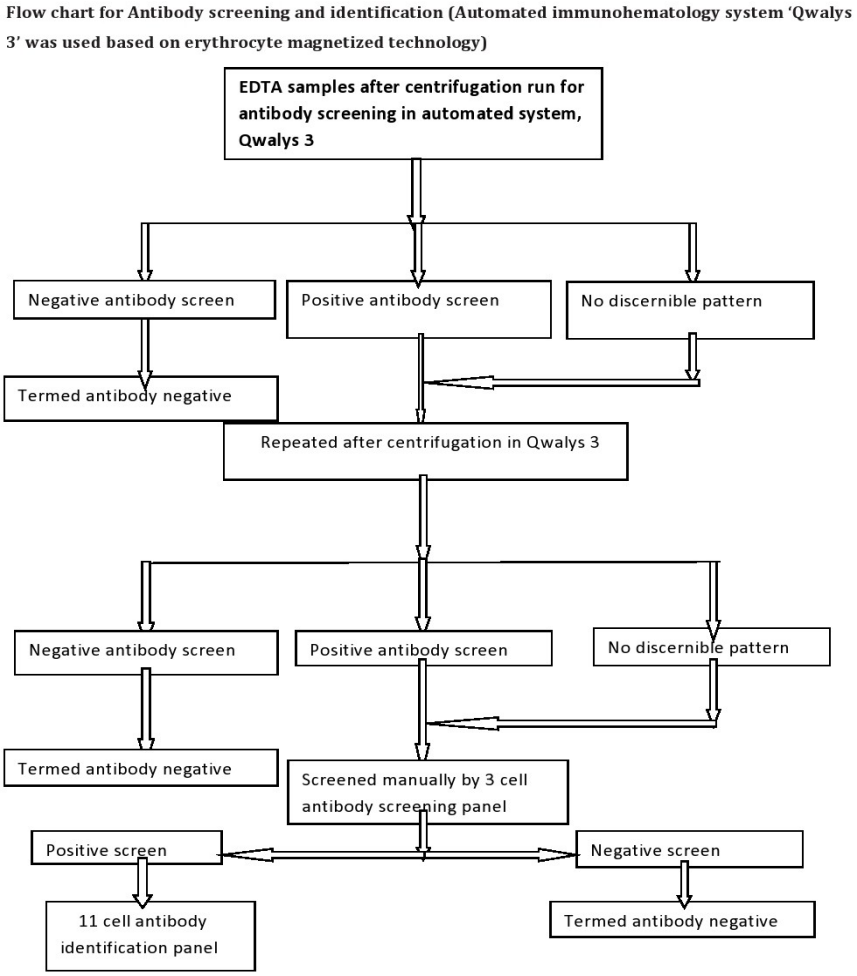
	Rh-hr		Spender Donor Donneur Donatore Donante Dador	Rh-hr						Kell						Duffy		Kidd		Lewis		P	MNS				Luth.		Xg		Spez. Antigene Special types Antigènes part. Antigeni particolari Otros Antígenos Tipos especiais	
				D	C	E	c	e	C^w	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P$_1$	M	N	S	s	Lu^a	Lu^b	Xg^a	♂ ♀		
1	CCC^{w}D.ee	$R_1{}^wR_1$	894587	+	+	0	0	+	+	0	+	0	+	nt	nt	+	0	+	0	0	+	0	+	0	0	+	0	+	+			1
2	CCD.ee	R_1R_1	053203	+	+	0	0	+	0	+	+	0	+	nt	nt	0	+	0	+	+	0	+	+	0	0	+	+	+	0			2
3	ccD.EE	R_2R_2	235024	+	0	+	+	0	0	0	+	0	+	nt	nt	0	+	0	+	0	+	0	0	+	0	+	0	+	nt			3
4	Ccddee	r′r	002739	0	+	0	+	+	0	0	+	0	+	nt	nt	0	+	+	0	0	0	0	+	+	0	+	0	+	+			4
5	ccddEe	r″r	415472	0	0	+	+	+	0	0	+	0	+	nt	nt	+	0	+	0	0	+	+	+	+	0	+	0	+	nt			5
6	ccddee	rr	856916	0	0	0	+	+	0	+	+	0	+	nt	nt	0	+	+	0	0	0	+	+	0	+	0	0	+	+			6
7	ccddee	rr	303739	0	0	0	+	+	0	0	+	0	+	nt	nt	+	0	0	+	0	+	+	+	+	0	+	+	+	0		Co(b+)*	7
8	ccD.ee	R$_o$r	767559	+	0	0	+	+	0	0	+	0	+	+	nt	0	0	+	0	0	0	+	+	+	0	+	0	+	+			8
9	ccddee	rr	174579	0	0	0	+	+	0	0	+	+	+	nt	nt	0	+	0	+	0	+	0	0	+	0	+	0	+	nt			9
10	ccddee	rr	855041	0	0	0	+	+	0	0	+	0	+	nt	nt	0	+	+	+	+	0	+	0	+	+	0	0	+	0			10
11	ccddee	rr	349033	0	0	0	+	+	0	0	+	0	+	nt	nt	+	0	+	0	0	+	+	0	+	0	+	0	+	+			11
	Patient / Patient / Patient / Paziente / Paciente / Paciente																													Eigenkontrolle / Autocontrol / Autocontrôle / Autocontrollo / Autocontrol / Auto-controlo	

The samples giving positive result even after second run were subjected to manual antibody screening by column agglutination technology (Gel card method) in commercially available 3-cell antigen panel (ID Dia I, II, III; Dia Med ID microtyping system by Biorad) with an autocontrol. Samples giving positive antibody screen on three cell screening panel were subjected to antibody identification by extended eleven cell panel [Figure 2] [Figure 3].

Figure 2- 3 cell panel

	Rh-hr		Spender Donor Donneur Donatore Donante Dador	Rh-hr						Kell						Duffy		Kidd		Lewis		P	MNS				Luth.		Xg	
				D	C	E	c	e	C^w	K	k	Kp^a	Kp^b	Js^a	Js^b	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P$_1$	M	N	S	s	Lu^a	Lu^b	Xg^a	♂ ♀
I	CCC^{w}D.ee	$R_1{}^wR_1$	129948	+	+	0	0	+	+	0	+	0	+	nt	nt	+	0	+	0	+	0	0	0	+	0	+	0	+	0		
II	ccD.EE	R_2R_2	612928	+	0	+	+	0	0	0	+	0	+	nt	nt	0	+	0	+	0	+	+	+	0	+	0	0	+	+		
III	ccddee	rr	119450	0	0	0	+	+	0	+	+	0	+	nt	nt	0	+	+	+	0	+	+	+	+	0	+	0	+	+		

Figure 3- 11 cell panel



3. Results

Of the total 14753 samples 276 (1.87%) samples had antibodies with 6359 males and 8394 females. Previous red cell transfusion was most common cause (120) (43.5%), followed by Pregnancy (94)(34.1%) for antibody production. 78 antibody positive samples (28.26%) were from 6359 male patients (47.8%) and 198 samples (71.7%) from 8394 females patients (52.1%). The difference in rate of alloimmunization between males and females was found to be statistically significant with a p-value < 0.001. Alloimmunization due to pregnancy can be considered to be the main attributing factor for the higher rate of alloimmunisation in females.

Figure 4: Gender wise distribution of various alloantibodies detected

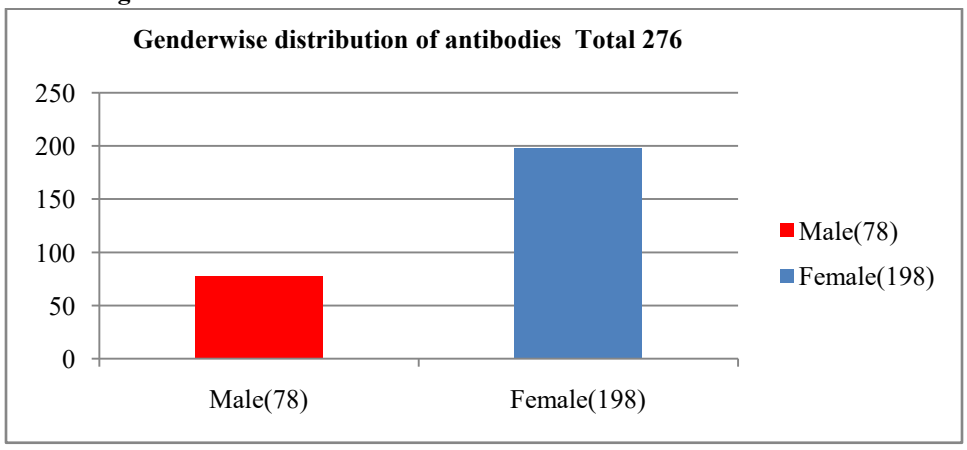


Table 1: Gender wise distribution of auto and alloantibodies

Total antibodies	Male	Female	Total
Autoantibodies (27)	10	17	27
Alloantibodies (249)	91	158	249
Total	101	175	276

Table 2: Various types of alloantibodies encountered in the study

Various types of alloantibodies	Number of cases	In male patients	In female patients
Single Antibodies	187	66	121
Multiple Antibodies	21	7	14
Antibody against low frequency antigens	4	3	1
Antibody against high frequency antigens	29	11	18
Both auto and alloantibodies	3	1	2
No clear pattern seen	5	3	2
Total	**249**	**91**	**158**

Table 3: Gender wise distribution of various types of single alloantibodies

Various types of single alloantibodies	Males	Females
Anti-D	2	71
Anti-c	7	18
Anti-C	4	10
Anti-E	12	24
Anti-e	0	1
Anti-C^w	1	0
Anti-M	5	2
Anti-N	3	1
Anti-s	2	1
Anti-S	3	2
Anti-Fy^a	2	1
Anti-K	9	5
Anti-Jk^a	1	0
Total	**51**	**136**

4. Discussion

In a study it was found that female gender could be a risk factor for red cell alloimmunization following transfusion [1]. Antibodies may be induced by pregnancy. Foetal anaemia, hyperbilirubinaemia, and even hydrops foetalis death may result from maternal RBC alloimmunization. Female gender as well as increasing age may be a risk factor in itself for red cell alloimmunization [2].

Jagadeesan *et al* found in a study that female gender had more prevalence of AIHA in comparison to male gender [3], our study also had more prevalence of autoantibodies against red cell antigens in female gender. Similar to our study Makroo *et al* found in an study the high prevalence of anti-D with single antibody to gender wise ratio more for the female gender in comparison to male gender [4].

In another study by Patel *et al* anti-D was the most common antibody [5] and Rh system antibodies as well MNS system antibodies were found to be very common in another study [6]. Our study population had a large number of patients having been transfused previously or had history of any sort of exposure, antibodies against Rh antigens predominated in our study, a similar study also demonstrated high rate of alloimmunization and predominance of antibodies against Rh antigens [7].

Our study population had large number of pregnant women, anti-D antibody was most prevalent antibody in our study as pregnancy is a risk factor for anti-D formation, and a similar study also demonstrated high prevalence of anti-D amongst both Rh positive and negative women [8]. In another study a higher rate of alloimmunization was found in multiply transfused patients, our study population also had large number of multiply transfused patients [9].

In another study anti-D as well as other red cell antibodies alloimmunization was found in pregnant women so irregular antibody screening was mandatorily recommended [10]. High rate of anti-D alloimmunization was also observed in a study by *Kahar* [11] and other study demonstrated non Rh alloimmunization in pregnant females [12].

5. Conclusion

As per results observed in our study, it's recommended to perform phenotyping for Rh system and K antigen at least and provide phenotypically matched red cells for all patients with planned surgeries not needing emergency transfusions. Extended red cell phenotyping should be done for almost all chronically transfused patients and female patients of child bearing age and phenotypically matched blood units should be transfused.

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