

WEAK RHD (D^u) PHENOTYPIC EXPRESSION AMONG THE BLOOD DONORS AND RECIPIENTS IN PESIMSR, KUPPAM

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ABSTRACT

BACKGROUND

Rh blood group system is based on the presence or absence of the Rh antigen, often called the Rh factor, on the cell membranes of the red blood cells (erythrocytes). Some red cells possess the D antigen but it is expressed so weakly that the cells are not agglutinated directly by anti-D sera. An indirect antiglobulin test is necessary to identify patients with the Weak D (formerly known as D^u) phenotype.¹ Detecting the Rh blood group as well as D^u testing among the Rh-negative individuals is important as the antibodies against Rh antigen can cause transfusion reaction and haemolytic anaemias.²

MATERIALS AND METHODS

This Cross-sectional study was done on 6346 individuals who were blood donors and recipients of blood bank of PESIMSR, Kuppam over a period of 6 months. They were grouped for ABO Rh D antigen and those negative for Rh D antigen were subjected to D^u test by IAT (indirect antiglobulin test).

RESULTS

Among 6346 individuals who were blood donors and recipients subjected to Rh typing, 422(6.64%) individuals were found to be Rh negative and 5924 (93.36%) individuals were found to be Rh positive. Out of 422 Rh negative samples, 1(0.24%) was found to be D^u positive, while the remaining 421(99.76%) samples were D^u negative.

CONCLUSION

Detection of weak D^u positive blood donors and recipients would remarkably reduce the incidence of transfusion reactions and save on the scarce O negative blood for emergencies.

KEYWORDS

D^u Antigen, Indirect Antiglobulin Test, Transfusion reactions.

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BACKGROUND

ABO blood group system was discovered by Landsteiner in 1901 and then along with Weiner a major breakthrough was the discovery of Rh antigen in 1939 which further led to description of haemolytic disease of newborn by Levine and Stetson.³

The Rh system was first discovered in Rhesus monkeys. Rh gene lies on chromosome 1. RHD and RHCE are the two homologous genes which codes for D protein and C,c,E,e proteins respectively. Rh antigen especially D are highly immunogenic. Consequently, based on the presence or absence of D antigen on the surface of RBCs, individuals are categorized as Rh Positive and Rh Negative respectively.³

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Individuals who express fewer D sites per red cell than normal are considered as D^u positive.¹ The D^u is classified into high-grade, low-grade and very low-grade depending on the expression of weak antigens on the surface of red blood cells. Individuals with D^u positive cells with weak expression of D antigens on them may be misclassified as D negative cells in routine Rh grouping procedures and therefore it is necessary to perform D^u test (IAT) on them. The antibody in Rh system is called anti-D antibody and is produced when Rh negative individual receives the Rh-positive blood. These antibodies develop insidiously in the first encounter, but rapidly in the subsequent encounters. 5-12% of the Indian population is found to be Rh-negative.^{4,5} An extensive 10-year study done in India shows that 7.19% of blood donors were Rh negative and the weak D variant among them was 0.01%.³ Slightly lower values have been reported in a Korean study and higher values among Caucasians.^{6,7} It has also been noted that the prevalence of ABO and Rh blood group varies from region to region. Therefore, we intend to find the frequency of ABO and Rh-negative individuals as well as D^u positive individuals among the blood donors and recipients in PESIMSR, Kuppam.

Consequently, it reduces the incidence of transfusion reactions. In unforeseen medical emergencies where Rh negative blood is required, if the recipient is found to be D^u positive, Rh positive blood can be transfused to them instead of Rh negative blood with scarce availability of Rh negative blood among the donors.

Aims and Objectives-

1. To determine the prevalence of ABO blood group among the blood donors and recipients.
2. To determine the prevalence of Rh negative individuals among the blood donors and recipients.
3. To determine the prevalence of D^u antigen among the blood donors and recipients who were found to be Rh negative.

MATERIALS AND METHODS

A written informed consent was taken from all the subjects. The blood donors and recipients put together 6346 individuals of PES Institute of Medical Science and Research, Kuppam, Andhra Pradesh, were subjected for ABO blood grouping as well as for Rh D antigen and those negative for Rh D antigen were subjected to D^u test by IAT (indirect antiglobulin test).

Inclusion Criteria-

Blood donors and blood recipients of PESIMSR, Kuppam who are willing to participate in the study.

Exclusion Criteria-

Subjects not willing to participate in the study.

Procedure- D^u testing was done using Standard operating procedure for Matrix™ Gel system. 0.8% red cell suspension in Matrix™ diluents 2 (LISS) was prepared by dispensing 1ml (1000µl) of Matrix™ diluents 2 (LISS) into a clean and labeled test tube to which 25µl of packed cells were added and mixed gently. The card was observed for appearance of agglutination and was labeled with the donor's/recipient's details. 50µl of red cell suspension (0.8%) was pipetted into the labeled microtube. Then 25 µl of AGTROL or Ercyclone Anti D IgG was added to the microtube. This was incubated at 37°C for 30 mins in Matrix Card incubator. The card was centrifuged in Matrix™ card centrifuge for 1 cycle (10 mins) and the reaction was read.⁸

Data was extrapolated in MS- Excel 2007 version and was analyzed for percentage.

RESULTS

Among 6346 individuals (3809 were females and 2537 were males) who were blood donors and recipients subjected to Rh typing, 422(6.64%) individuals were found to be Rh negative and 5924 (93.36%) individuals were found to be Rh positive as shown in Table 1.

Blood Group	No. of Donors and Recipients	Frequency (%)
RhD positive	5924	93.36
RhD negative	422	6.64
Total	6346	100

Table 1. Frequency of RhD Antigen among Blood Donors and Recipients

Out of 422 Rh negative samples, 1 (0.24%) was found to be D^u positive, while the remaining 421 (99.76%) samples were D^u negative as shown in Table 2.

Blood Group	No. of Donors and Recipients	Frequency (%)
D ^u positive	1	0.24
D ^u negative	421	99.76
Total	422	100

Table 2. Frequency of D antigen in RhD negative blood donors and recipients

Among 422 Rh negative blood groups, 188 (44.55%) were O negative, 138 (32.7%) were B negative, 74 (17.54%) were A negative and 22 (5.21%) were AB negative as shown in Figure 1.

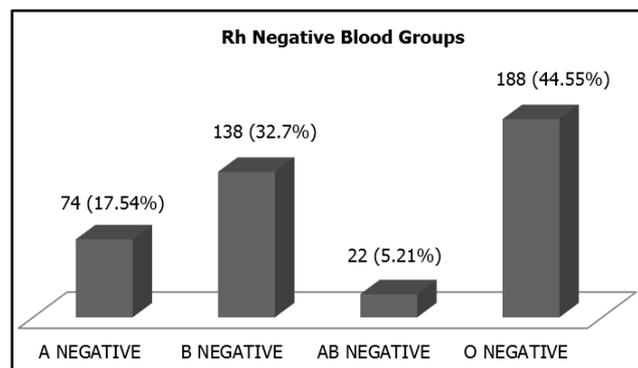


Figure 1. Graph Illustrating Percentage Distribution of Various ABO Blood Group Systems with Rh Negativity

Among 5924 Rh positive blood groups, 2409 (40.66%) were O positive, 1914 (32.31%) were B positive, 1218 (20.56%) were A positive and 383 (6.47%) were AB positive as shown in Figure 2.

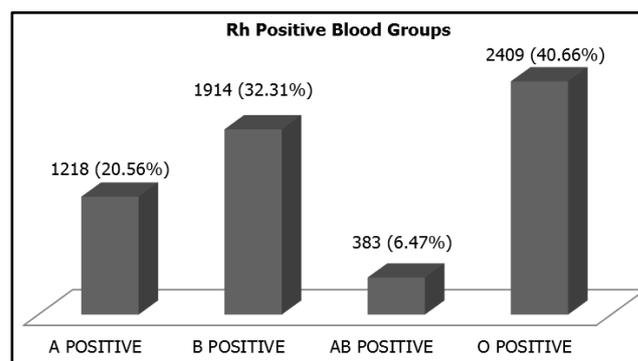


Figure 2. Graph Illustrating Percentage Distribution of Various ABO Blood Group Systems with Rh Positivity

Among 74 A negative individuals, 31 were males and 43 were females, out of 188 O negative individuals 73 were males and 115 were females, among 138 B negative individuals 57 were males and 81 were females and out of 22 AB negative individuals 8 were males and 14 were females as shown in Table 3.

Blood Group	Males	Females	Total
A negative	31	43	74
O negative	73	115	188
B negative	57	81	138
AB negative	8	14	22

Table 3. Male and Female Distribution of Various Rh negative Blood Groups

And we noted that out of 81 B negative females one was found to be D^u positive.

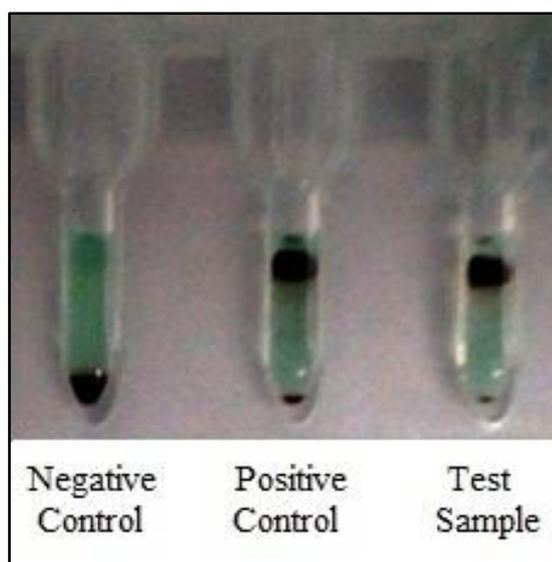


Figure 3. Photograph with Positive, Negative Control and Test sample of a B Negative Individual the Test Sample Shows Agglutination

Among 1218 A positive individuals, 512 were males and 706 were females, of 2409 O positive individuals, 968 were males and 1441 were females, of 1914 B positive individuals, 750 were males and 1164 were females and among 383 AB positives, 138 were males and 245 were females as shown in Table 4.

Blood Group	Males	Females	Total
A positive	512	706	1218
O positive	968	1441	2409
B positive	750	1164	1914
AB positive	138	245	383

Table 4. Male and Female Distribution of Various Rh Positive Blood Groups

DISCUSSION

To be able to donate or receive blood, an individual should know his or her blood group. This is essential in unforeseen medical emergencies.

The frequency of Rh negative blood donors and recipients in this study was found to be 6.65% which is higher than the frequencies of 3.30%, 3.9% and 1-6% reported in Rewa region of Madhya Pradesh, among Kenyans and Nigerians respectively.^{2,9} 0.24% of Rh negative blood donors and recipients was found to be D^u positive in our study which is a little below 0.43%, 0.59% and 0.8% prevalence reported among Rewa region of Madhya Pradesh, Europe and Brazil respectively.^{2,10,11} On the other hand, 0.24% is a little high in comparison with 0.14% in Albanias.¹² The variable results of our study in comparison to several other studies is because of variable demographic profile and social milieu.

Identification of D^u positive donors and recipients can prevent transfusion reactions and reduce the incidence of unnecessary Rh-negative blood transfusions which is scarce. Rh antibodies may develop in any Rh-negative individual in the event of Rh positive blood transfusion eventually leading to Haemolytic transfusion reactions.¹³ Therefore, weak D positive patients should always receive Rh positive blood. Identification of D^u positive individuals is also useful in patients with frequent transfusion requirements such as patients with HIV/AIDS, Chronic renal failure, advanced malignancies, aplastic anaemia, bone marrow failures, sickle cell anaemia and other causes of chronic anaemias.¹⁴ However false positive errors that may lead to administration of RhD positive blood to RhD negative patients which can cause severe immunological and clinical consequences must be taken care of by meticulously and rigorously controlled laboratory and reagent conditions.

Blood groups are genetically determined and the incidence of ABO and Rh genes and their phenotypes vary widely across races and geographical boundries.¹³ The most prevalent ABO phenotype among the individuals visiting blood bank of PESIMSR was found to be O (40.92%) followed by B (32.34%), A (20.36%) and AB (6.38%). Studies from South India show similar distribution with predominance of O blood group followed by B, A and AB.^{15,16} In contrast, studies done in North India and Pakistan shows the predominance of blood group B followed by O, A and AB.^{17,18}

Blood transfusion service is an integral part of modern health care system for efficient medical care. Blood transfusion service aims in providing adequate, effective and safe blood and blood products to meet the patient’s needs.

CONCLUSION

Our study concluded that the incidence of Rh negative blood group was 6.64% and D^u positive was 0.24% among the blood donors and recipients of PESIMSR. D antigen is most immunogenic and plays an important role in immunohaematology and blood banking. Detection of weak D^u positive blood donors and recipients would remarkably reduce the incidence of transfusion reactions and save on the scarce O negative blood for emergencies.

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