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Detection of Weak Rh D (DU) Phenotype among Blood Donors.

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ABSTRACT

Background: The Rh blood group system is one of the most polymorphic systems in human. Following the discovery of the ABO blood group systems, the greatest breakthrough in transfusion medicine was the discovery of the Rh antigen. . The most common and immunogenic are D, C, E, c and e. The RhD blood group antigen has been shown to be subject to many phenotypic variations. The "weak D" actually refers to red cells with the aberrant Rh-D protein expressing reduced membrane surface D antigen. There is one misconception that individuals with weak phenotypes cannot make anti-D in contrast to partial D because they have low levels of complete D antigen. **Methods:** Commercially available monoclonal anti D sera was used to detect Rh-D factor status. Individuals found negative with saline anti-D were further investigated and confirmed for weak D antigen by using anti human globulin serum (Indirect Coomb's technique). **Results:** During this study 19,347 healthy blood donors were tested for Rh-D factor status. Among these 17,295(89.4%) were Rh-D factor positive while 2052(10.6%) donors were Rh-D factor negative. Among the Rh-D factor negative individuals, 4(0.19%) were weak D positive. **Conclusion:** It is important to detect and determine all RH negative individuals by saline method for the detection of weak D status to reduce the chances of alloimmunization.

Keywords: RH-D factor, D-antigen, blood donors, transfusion, phenotype.

INTRODUCTION

The Rh blood group system is one of the most polymorphic systems in human. It plays major role in incompatible RBC transfusion conflicts and in the hemolytic disease of the newborn due to the maternal-fetal blood group incompatibility.

Following the discovery of the ABO blood group systems, the greatest breakthrough in transfusion medicine was the discovery of the Rh antigen by Levine and Stetson in 1939.Rh system is a complex blood group system having 49 different antigens. The most common and immunogenic are D, C, E, c and e. The D antigen is encoded by the RhD gene while RhC, RhE, Rhc and Rhe antigens are encoded by the RHCE gene. Out of all these, D antigen is the most significant. D antigen has more than 30 distinct epitopes, more than 100 known haplotypes with similar phenotypes of different alleles. D is often called the Rh antigen and the terms Rh positive and Rh negative refer to presence or absence of D antigen respectively. The RhD antigen has been reported to consist of a mosaic of at least 9 D epitopes (epD1-epD9). The RhD blood group antigen has been shown to be subject to many phenotypic variations.[1,2]

Recent testing shows a large number of monoclonal anti-D (MAb-D) reagents has suggested the presence of a minimum of 30 different epitope structure distributed along the extracellular portions of the RhD protein.[4] Thus a change/changes, in the amino acid sequence of RhD may not ablate the entire D antigen but can cause epitope loss, giving rise to variant forms of D antigen. In Weak D" RBCs demonstrate reduced quantities of the D antigen. As a result, weak or no agglutination reaction is demonstrated by these RBCs with the anti D reagents at the immediate spin phase.

In "Partial D" RBCs, the RHD protein is mutated in an exofacial loop, eliminating at least one D-specific epitope. However, the numbers of RhD antigens on the RBC surface are normal. Weaker variants of D (Du), have fewer than normal D antigen per red cell but have all the epitopes. Weak D is weakly immunogenic and requires detection by antihuman globulin.[6] Partial D antigen lack one or more epitopes on red blood cells8. Thus RBCs having partial D antigen are agglutinated distinctively by some but not all monoclonal anti D reagents. The frequency of weak D phenotype varies with the method used, the reagent used, and the racial mix

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tested (Division of Medical Laboratory Science, 2001). Weak D phenotypic expression is known to arise from three mechanisms:-In one of these mechanisms, referred to as gene interaction, there is a suppressive effect of the C gene when in trans to the D gene (e.g.,D-ce/Ce).
The second is when part of the D antigen is missing (partial D)
Thirdly, the presence of an aberrant form of D (eg. At the molecular level) would result in weak phenotypic expression.[8] The "weak D" actually refers to red cells with the aberrant Rh-D protein expressing reduced membrane surface D antigen. There is one misconception that individuals with weak phenotypes cannot make anti-D in contrast to partial D because they have low levels of complete D antigen.

Aim of the study

This study was undertaken with the objective to detect the prevalence of weak RhD antigen among healthy blood donors.

MATERIALS AND METHODS

Study Centre

This retrospective study was performed at Blood Bank, Teerthanker Mahaveer Hospital and Research Centre, Moradabad.

Data Collected

3 years data was analyzed retrospectively. (from 2014-2017)

Assessment

Commercially available monoclonal anti D sera was used to detect Rh-D factor status.
Individuals found negative with saline anti-D were further investigated and confirmed for weak D antigen by using anti human globulin serum (Indirect Coomb's technique).

Statistical Analysis

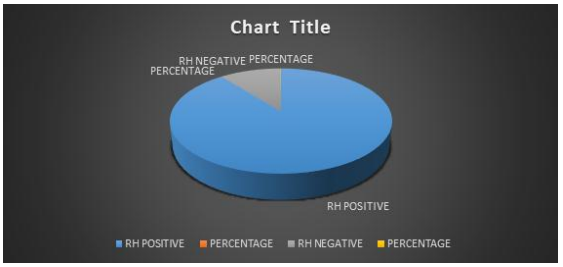
Data was analyzed using percentage of RH-D Positive, Negative & Weak D positive among donors.

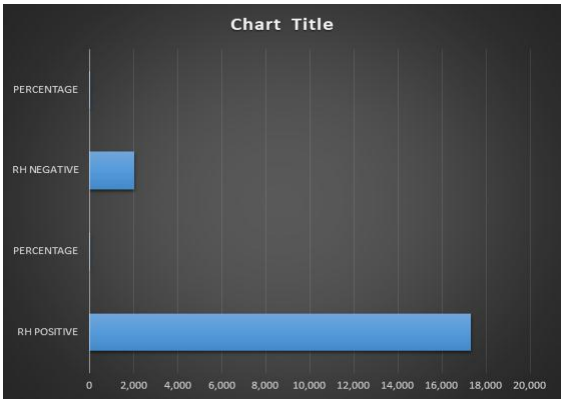
RESULTS

Table:-1 Weak D prevalence in blood donors

RH POSITIVE	PERCENTAGE	RH NEGATIVE	PERCENTAGE	WEAK D POSITIVE
17,295	89.3%	2052	10.06	4(0.19%)

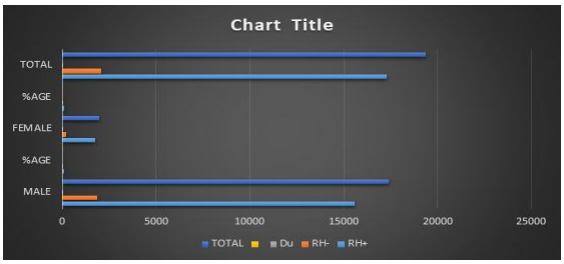
- During this study 19,347 healthy blood donors were tested for Rh-D factor status.
- Among these 17,295(89.4%) were Rh-D factor positive while 2052(10.6%) donors were Rh-D factor negative.
- Among the Rh-D factor negative individuals, 4(0.19%) were weak D positive.

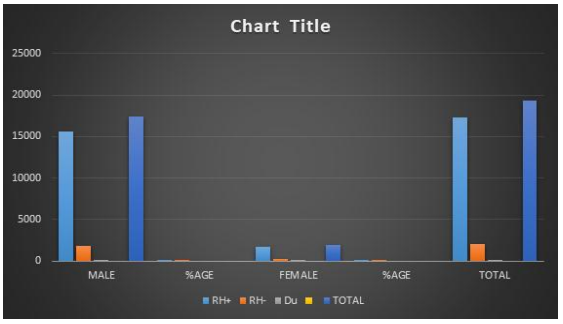




DISTRIBUTION OF RH-D ANTIGEN IN MALES AND FEMALES

	MALE	%AGE	FEMALE	%AGE	TOTAL
RH+	15566	89.39	1729	89.4	17295
RH-	1844	10.59	204	10.55	2048
Du	3	0.017 of total males 0.16% of RH- males	1	0.05of total females 0.49% of RH- females	4
TOTAL	17413		1934		19347





DISCUSSION

Weak D is a phenotype with either a qualitative or quantitative difference in the RhD moiety resulting in a weakened expressed of D antigen. The D antigen is highly immunogenic, if Rh-D positive blood is transfused to a Rh-D negative recipient, the recipient is likely to develop anti-D alloantibodies. Another important fact is that if sensitized Rh negative women conceives an Rh D positive fetus, the passage of anti D antibodies across the placenta to the fetus results in hemolytic disease of newborn

The clinical significance of weak D antigen is that if transfused to RhD sensitized subject, can result in hemolytic transfusion reaction. The incidence of weak D varies worldwide, and it ranges from 0.2 to 1% in Caucasian individuals. In present study frequency of weak D antigen observed is 0.19% which is similar to the study conducted in Dr. Muhammad Usman et al,[3] and a study conducted in Moroccan population4 The frequency is also similar to that found in Albanian population (0.14).[5]

Whereas in a study conducted by Dr. Geeta et all the prevalence was slightly on the higher side (0.49%) On the contrary high prevalence of weak D individuals is seen in Africans (10%).[6,7] Among Indians 0.09%-0.189% prevalence has been reported8. According to American Association of Blood Banks (AABB) it is mandatory to detect the weak D/partial D status of the donor but the recipient can be safely considered as RhD negative. women of child bearing age and efficacy of anti-human gamma globulin in detecting weak D antigen is well accepted.

CONCLUSION

Frequency of weak D antigen is observed in our study is 0.19%. Several research studies proved that weak D antigen is immunogenic and can produce alloimmunization if transfused to Rh-D negative subjects. Thereby we conclude that it is important to detect and determine all RH negative individuals by saline method for the detection of weak D status to reduce the chances of alloimmunization.

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