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A Study to Determine the Prevalence of Weak D in Donors and Patients at a Tertiary Care Teaching Hospital in Telangana

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Authors' contributions

This work was carried out in collaboration among all authors. Author MM contributed to the conception and design of the work; contributed significantly to the acquisition, analysis and interpretation of data; and drafted the work. Author VBS contributed significantly to the conception and design of the work; and contributed significantly to the analysis and interpretation of data and substantially revised the work. Authors JA and DE contributed to drafting, revising and submission of the article. All the authors have read and approved the final manuscript.

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ABSTRACT

Introduction: The Rhesus (Rh) blood group system is considered the next most important after ABO. It is of clinical significance in regard to transfusion and pregnancy. The Weak D phenotype (Du)is a weakened form of D antigen that cannot be detected by routine grouping (using immediate spin tube methodology).

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Aims:
1) To determine the prevalence of weak D.
2) To assess the implications in terms of alloimmunization.
3) To provide knowledge on the weak D status and enhance the importance of weak D determination in the donor and patient population.
Study Design: A retrospective study.
Place and Duration of Study: Licensed blood bank in Malla Reddy Narayana Multispecialty Hospital under Department of Pathology, data from blood bank records was collected from 1st of August 2018 to 31st of July 2019.
Methodology: It was a one-year study at a tertiary care centre and study group included both donors and patients who were admitted and those attending outpatient departments who were identified for blood grouping test at the blood bank. Data was collected from the blood bank records.
Results: A total of 4054 blood samples were analysed out of which of 3405 were Rh D positive and 649 samples were RhD negative. On further testing 12 out of 649 Rh D negative, were found to be weak D positive.
Conclusion: For a safer transfusion and to prevent alloimmunisation it is recommended to develop protocols for weak D testing for the individuals who are Rh D negative on routine testing in regions showing significant prevalence.

Keywords: Weak D; alloimmunisation blood transfusion; Rh D; prevalence.

1. INTRODUCTION

The major discovery in blood group systems was associated with the discovery of the ABO blood groups in 1900 by Karl Landsteiner. This was followed by the description of Rh blood group system by Levine and Stetson in 1939. The Rhesus system discovery divided the human race into two, those who were Rh D antigen (D positive and Rh D negative) [1]. Rhesus blood group system is of clinical significance next to ABO system. It is also highly immunogenic with numerous polymorphisms [2]. By the year 2015, 58 Rh antigens have been identified and documented. Rh system has RhD antigens and RHCE types. RhD has the D antigen while RHCE has D, C, E, c and e [3]. However the most immunogenic is the RhD antigen regarding transfusion and pregnancy [4]. In 1946, Stratton described a weakly reacting D antigen [5]. The weak D phenotype is a weakened form of D antigen that in routine D antigen testing will react with some anti-D but not with others (when 37 C incubation or an immediate spin is given). Weak D RBC has D antigen but fewer in number as compared to normal Rh D-positive red cells. Weak D phenotypic expression arises by three mechanisms. Suppressive effect of C gene when in trans to D gene, when the part of D antigen is missing (partial D) or due to presence of aberrant form of D [6]. Demonstration of this weakly expressed antigen requires evaluation by prolonged incubation and use of antihuman globulins, enzymes, extended phenotyping and genotyping [7].

In most instances presence of serological weak D phenotypes are suspected when a weak reaction (≤2+) is encountered while performing RhD typing. Problems in immunohematological testing tend to occur when blood donors are wrongly typed as RhD negative in spite of having a trace amount tof RhD antigen which can cause alloimmunization [8].

2. MATERIALS AND METHODS

A cross-sectional study for a period of one year was carried out with prior approval from institutional ethical committee at licensed blood bank in Malla Reddy Narayana Multi-specialty Hospital under Department of Pathology from 1st of August 2018 to 31st of July 2019 catering to patients from in and around Suraram. The study group included all donors and patients both admitted as well as attending outpatient departments. 2 ml blood sample was collected in EDTA vacutainers and tested for Rh typing and ABO forward and reverse grouping by conventional slide and tube agglutination methods.

The samples that turned out to be Rh negative by conventional slide and tube methods (using IgM monoclonal Anti D reagent of Tulip company) were subjected further to confirmation by gel card method (Matrix™ Gel System). The weakD (D^u) test was performed using 1% red cell suspension in Matrix Diluent 2(LISS), that was prepared by dispensing 1ml of Matrix diluents2 (LISS) into clean labelled test tube and adding

10µl of packed red cells and mixed gently. 50µl of patients red cell suspension was pipetted into a labelled microgel tube and 25µl of Matrix™ Anti- D IgG was added and incubated at 37˚C for 15min in Matrix Card Warmer. The gelcards were then centrifuged in Matrix™ Card Centrifuge for 1cycle(10min) and results interpreted by two observers independently.

3. RESULTS

A total of 4054 blood samples were analyzed during the study period for ABO and Rh D antigen grouping. A total of 3405 were Rh D positive and 649 samples were RhD negative when tested by conventional slide and spin tube techniques. The blood samples which were negative on routine typing were further tested for presence of weak D and 12 of these 649 samples were found to be weak D or D^u positive.

In the present study, 84% (3405 out of 4054 samples) were Rh positive, 15.71% (637 out of 4054) were Rh negative and 0.29% were found to be weak D positive. The study also revealed that weak D was more common in blood group B (5 in 12) compared with blood group A (4in 12) and O group (3 in 12) while AB blood group phenotype was not associated with RhD phenotype in as shown in the Table 1.

Table 1. Distribution of weak D(DU) among blooddonors and patients

Blood Group	Rh D positive	Rh D negative	Weak D
O	1285	215	03
A	860	136	04
B	991	232	05
AB	269	54	00
Total	3405	637	12
Percentage	84 %	15.71%	0.29%

4. DISCUSSION

“The RHD gene polymorphism leads to phenotypic polymorphism of D variants including weak D, Del and partial D” [9]. “Weak D red cells have fewer D antigens per cell than normal Rh-positive cells. (110 to 9000 per red blood cell).In Weak D one or more amino acid substitutes are found in the region that are presumed to be in or below the membrane and may interfere with the assembly of Rh complexes” [10]. The importance of detecting weak D lies in the fact that transfusion of red cells from a person with ‘weak D phenotype to a ‘D Negative’ person may result in alloimmunization and subsequent exposure to such ‘D Positive’ red cell can lead to fatal hemolytic reaction or hemolytic disease of newborn in a sensitized pregnant female. “Even 0.5 ml of Rh D antigen exposure in Rh negative individual can induce antibody response” [11]. Considering the risk of immunogenicity, the persons with weak D phenotype are typed based on whether the person is donor or the recipient. The recipients with weak D should be considered D negative and must be transfused with D negative blood and as donors they are considered as D positive.

“The incidence of Rh negativity worldwide varies between 3%-25% and that of weak D antigen ranges from 0.2%-1%” [12]. The variation may be because of lack of set standards for performing the tests, type of reagents used (monoclonal,polyclonal,blended), objective and subjective variation in interpretation of test results. Further, it has been adequately documented that D epitopes distribution differs with different geographic locales and ethnicities of the populations [13]. In our study weak D constituted 0.29% of the whole study sample and 1.85% of all Rh negative samples screened. It shows the prevalence of weak D among the population in and around Suraram village, Telangana India.

In a recent Indian study by AR Srivastava et al. [14] at a tertiary hospital in Maharashtra ,out of a total of 17,262 samples, 15,400 (89.2%) tested to be Rh Positive and 1,866 (10.8%) tested Rh-negative. Weak D was Positive in 52 (0.027%) samples out of the RhD negative samples.

Aslam et al. [15], had conducted a study and found that the frequency of Rh-positive was 86.3% and Rh-negative 13.7% (close to our study) and 1% individual were weak D-positive (high compared to present study). A hospital based Indian study by Anshu gupta et al. [16], had shown that out of 3619 cases, 3502 cases (96.7%) were Rh positive,117 cases (2.98%) Rh negative and 9 cases (7.2% of total Rh negatives and 0.25% out of total samples) were weak D positive (close to present study). Makroo et al. found 7.19% RhD negativity in the Indian population and weak D in 0.01%. When compared to the other studies, except for the study done by Aslam A et al. [15] stated above, the prevalence of weak D% was found to be high in our study as represented in the Table 2, which may be attributed to geographic and ethinic factors.

Table 2. Distribution of Weak D percentage in other studies

S. No	Author, year	Region	Weak D%	Rh positive	Rh negative
1.	Makroo et al 2010	India	0.01%		7.19%
2.	Aslam A et al, 2015	Lahore	0.9%	86.3%	12.6%
3.	Anshu Gupta et al, 2015	East Delhi	0.25%	96.7%	2.98%
4.	AR. Srivastava et al, 2021	India	0.027%	89.2%	10.8%
5.	Our study	India	0.29%	84%	15.71%

5. CONCLUSION

Rh D antigen is highly immunogenic. It is associated with immunological responses (immunization) once introduced to those who are RhD negative via pregnancy, transfusion or Transplantation. Therefore, it is essential to screen all donors and recipients for the presence of RhD antigens as well as weak D phenotype to ensure safe transfusion and prevention of hemolytic disease of the fetus and the newborns (HDFN). Detection of RhD and the weak phenotypes in all clinical settings (national blood services, hospital blood banks and laboratories) is vital towards the reduction of alloimmunisation at the time ensuring safer transfusion practices.

6. IMPLICATIONS OF THE STUDY

The study will provide information on the prevalence of weak D among the donor and patients population in the tertiary care teaching hospital in Telangana.

It will also support in the development of testing protocols as well as guidelines for weak D determination both in donors and patients.

CONSENT

As per international standard or university standard, patient(s) written consent has been collected and preserved by the author(s).

ETHICAL APPROVAL

The institutional Ethics committee has given approval for the study during IEC meeting. Ref: Project No: MRMCWIEC/AP/70/2021.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Kumar H, Mishra DK, Sarkar RS, Jaiprakash M. Difficulties in immunohaematology: The weak D antigen. Medical Journal Armed Forces India. 2005 Oct 1;61(4):348-50.
2. Krishna GD, Babu KS, Arun R, Jothibai DS. A study on Rh incompatibility and frequency of weak D among blood donors and patients at a tertiary care referral teaching hospital in Tirupati, Andhra Pradesh. Journal of Clinical and Scientific Research. 2015;4(4):281-4.
3. Flegel WA. The genetics of the Rhesus blood group system. Blood Transfus. 2007 Apr;5(2):50–7.
4. Westhoff MC. The structure and functiocomplex. Semin Hematol 2007;44(1):42–50.
5. Avent ND, Ridgwell K, Tanner MJ, Ans Tayyab M, Malik AR, Khan AS. D u Phenotype-A REVIEW. JAMC. 2000;12(3).
6. Tippet P. Subdivisions of the Rh Antigen D, review. Medical Laboratory Scientist Journal1988; 45(1):88-91.
7. Acharya S, Kumar R, Acharya R, Kudesia S, Kishore S. Weak D antigen Revisited. Indian Medical Gazette. 2011;342-45.
8. Afroz T, Rahman M, Naznin B, Islam MA, Saleh AJ. Evaluation of prevalence of weak D antigen among rhesus-negative patients in tertiary care hospitals in Bangladesh: A multicenter study. Global Journal of Transfusion Medicine. 2021 Jul 1;6(2):146.
9. Wafi ME, Housse HE, Nourichafi N, Bouisk K, Benajiba M, Habti N. Prevalence of weak D phenotype among D negative C/E+ blood donors in Morocco. Int J Blood Transfus Immunohematol. 2016;6:3-7.

10. Flagel WA. How I manage Donors and patients with weak D phenotype. In: Victor Hoffbrand, Daniel Catovsky, Edward GD, Tuddenham, editors. Post Graduate Haematology. 5th ed. Oxford: Blackwell Publishing. 2008;70.
11. Makroo RN, Raina V, Chowdhry M, Bhatia A, Gupta R, Rosamma NL. Weak D prevalence among Indian blood donors. Asian Journal of Transfusion Science. 2010 Jul;4(2):137.
12. Kumar H, Mishra DK, Sarkar RS, Jaiprakash M. Difficulties in immunohaematology: The weak D antigen. Medical Journal Armed Forces India. 2005 Oct 1;61(4):348-50.
13. Kulkarni SS, Gupte SC, Vasantha K, Mohanty D, Ghosh K. Varied distribution of RhD epitopes in the Indian population. National Medical Journal of India. 2007 Jul 1;20(4):169.
14. Srivastava AR, Dhote SW, Singh I. A retrospective study on the prevalence of weak D antigen (Du) in a blood bank in a tertiary care hospital in Maharashtra, India. MGM Journal of Medical Sciences. 2021 Oct 1;8(4):410.
15. Aslam A, Azmi R, Sheikh MZ, Javaid I. Frequency of weak expression of D ALLELE" among healthy blood donors. Pakistan Journal of Physiology. 2015; 11(3):22-24.
16. Gupta A, Mirza S, Khurana S, Singh R, Chaturvedi S, Singh B. Enigmatic weak D antigen: An experience in a Tertiary Care Hospital of East Delhi. Journal of Clinical and Diagnostic Research: JCDR. 2016 Jun;10(6):EC12.

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