Brief Report

Prevalence of 'Weak D' Phenotype in Patients in A Specialized Hospital

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ABSTRACT

Background: The Rhesus (Rh) blood group is the most immunogenic blood group in humans among all blood groups. It is one of the complex systems in immunohematology containing 56 different antigens. The Rh blood group shows variable expression of D antigen due to gene polymorphism. The variable expressions are weak D, partial D, and DEL.

Materials and Methods: A retrospective observational study was conducted at Evercare Hospital Chattogram from April 2021 to March 2023. Blood samples obtained for ABO and RhD blood grouping were tested by column agglutination technique. RhD negative samples were further tested for Du testing to identify weak-D phenotype.

Results: A total of 5700 blood samples were taken from patients. Out of these 5496 (96.5%) were RhD positive while 204 (3.5%) were RhD negative. Among Rh D negative cases, 4 were weak-D positive constituting 1.9% among Rh D negatives and 0.07% among total patients.

Conclusion: All RhD negative samples should be checked for Du testing. We must take caution on application of the Du test. It may be used to decrease the burden of RhD negative transfusion.

Key words: *Weak D Phenotype; Blood donors; Immunogenic.*

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INTRODUCTION

Rhesus (Rh) blood antigens were first described by Levine and Stetson in 1939 which is one of the major blood groups after ABO blood group. The Rh system consists of over 56 antigens, with D being the major antigen expressed by Rh D protein. Among 56 antigens of Rh system, five (D, C, c, E, and e) are detected that causing haemolysis of RBC. Rh system shows variable expression of D antigen. It is one of the complex systems in immunohematology. Weak D antigen is a phenotype where the D antigen is weakly expressed on red blood cells and this antigen cannot be detected by routine spin methods[1]. Weak D RBCs demonstrate less number of the D antigen showing weak or no agglutination reaction by these RBCs with the anti D reagents at the immediate spin phase. Various types of D variants like as weak D, partial D, and DEL are due to gene polymorphism. It may cause RhD-positive person to behave like RhD negative which could result in alloimmunization.

MATERIALS AND METHODS

This retrospective observational study was conducted at the Department of Transfusion Medicine of a tertiary care hospital of Bangladesh from April 2021 to March 2023. EDTA blood samples were taken from patients. Blood samples obtained for ABO and RhD blood grouping were tested by column agglutination technique using Ortho BioVue ABD forward and reverse cassettes (Ortho Clinical Diagnostics, USA). Du-testing was performed in all RhD negative samples to identify weak-D phenotype. Weak D testing was performed with Ortho BioVue AHG polyspecific cassettes, where 10µl of 5% cell suspension, 40 µl anti-D (Monoclonal, IgM + IgG, Tulip Diagnostic P, and LTD, India) and 50µl Ortho BLISS solution were kept in Ortho BioVue Heat Block at 37°C for 10 min. Then, it was centrifuged for 5 min and the results were noted and compared with positive and negative control.

RESULTS

A total of 5700 blood samples were taken from patients. Out of these 5496 (96.5%) were RhD positive while 204 (3.5%) were Rh D negative. Among RhD negative cases, 04 were weak-D positive constituting 1.9% among Rh D negative and 0.07% among total patients (Table 1). Blood group O was found to have the maximum number of weak D antigen samples with male predominance (table 2 and 3)

Table.1: Distribution of RhD antigen

Blood group	No of patients	Frequency
RhD positive	5496	96.5%
RhD negative	204	3.5%
Total	5700	100%

Table.2: Distribution of RhD antigen in males and females

Blood Group	No of patients	Frequency
Male	3	75%
Female	1	25%
Total	4	100%

Table.3: Distribution of weak D antigen in the specific blood groups

Blood group	No of patients	Frequency
A	0	0
B	1	25%
O	3	75%
AB	0	0
Total	**4**	**100%**

DISCUSSION

In certain individuals, D antigen expression on red blood cells is diminished, resulting in Weak D, a form of D phenotype seen in human beings. The usual procedures (spin tube method) utilized at the blood center are unable to identify this antigen. As a result, numerous ways are employed to illustrate this weakly expressed antigen. The most popular approaches are extended incubation, and the use of the Antihuman Globulin Indirect Coombs Test. Antibody responses can be elicited in responder Rh-Negative people after exposure to 0.1 ml of Rh (D) antigen positive cells. Only red blood cells have Rh antigens. The beginning of Rh antibody synthesis is irreversible, and circulating antibodies may persist for years[2]. Among the RhD-negative individuals, approximately 80% will develop anti-D after the first exposure to RhD-positive blood and only 7%–8% will remain non responsive[3]. Polymorphism of the immune response genes affects an individual's ability to produce antibodies[4]. HLA-antigens are crucial for cell to cell interaction in antibody production and high anti-D titers, have been linked to certain HLA-DR types.[5] Thus an individual's genetic predisposition may prevent the one from producing antibodies if exposed to a given antigen[6]. This is of concern in RhD negative pregnant females, as it can cause alloimmunization if accidentally weak D antigen-positive blood is transfused. The identification of D antigen is particularly useful in patients with frequent and high transfusion requirements such as patients with HIV/AIDS, chronic renal failure, malignancies, aplastic anemia and other bone marrow failures, sickle cell anemia (SCA) and other cases of chronic anemias [7,8]. The incidence of Rh negativity worldwide varies between 3% and 25% and that of weak D antigen from 0.2% to 1%[9]. In my study revealed that out of 5,700 patients, 204(3.5%) patients were RhD negative and of negative patients, 04 (1.9%) expressed the weak D antigen. In another study in Bangladesh showed that out of 177,702 patients, 7359 (4.1%) patients were RhD negative and of negative patients, 14 (0.19%) expressed the weak D antigen[10]. In other study in Bangladesh found that Rh positive were 97.54% and negative were 2.45% and weaker D reaction found in 7 (0.013) %[11].

Makroo et al. Found 7.19% RhD negativity in the Indian population and weak D in 0.01%[12]. Another study done by Krishna et al. reported the prevalence of weak D antigen to be 0.06% in the Indian population[13]. In the Pakistani population, the prevalence of RhD negative was found to be 13.7% and among them, 1% were weak D antigen-positive in Lahore, Punjab[14]. A study conducted in Africa by Okrah found 7.75% RhD negative and 6.45% weak D positive in blood donors which is slightly high[15]. Xhetani et al. reported 10.86% RhD-negative donors out of which 0.14% were weak D positive in the Albanian population[16]. The prevalence of weak D antigen is 0.5% in Europe, 3% in USA, and 0.8% in Brazil[16,17,18]. In this study molecular genotyping could not be done and also from these data, we cannot generalize the prevalence of weak D as it was done in a metropolitan area and more rural areas need to be included.

CONCLUSION

This study has found that 1.9% of patients who were typed as Rhesus negative are actually weak D positive. Due to strong immunogenicity of RhD antigen, cells need to be appropriately tested.

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