

The Role Of Du Testing In Scaling Down The Burden Of Rhesus-D Negative Transfusion In Northern Nigeria

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Citation

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Abstract

The low frequency of RhD negativity in Nigeria coupled with a sub-optimal national blood transfusion service has resulted in scarcity of RhD negative blood in Nigerian blood banks. This study was conducted to determine the frequency of D^u antigen among RhD negative patients scheduled for transfusion with the aim of scaling down the burden of RhD negative transfusion in northern Nigeria. The RhD groups of 2978 patients that were scheduled for transfusion were determined. Samples that were RhD negative were further tested for the D^u antigen. A total of 2897 (97.3%) samples were RhD positive while 81 (2.7%) samples were RhD negative. Out of the 81 RhD negative samples, 4 (4.9%) were D^u positive while the remaining 77 (95.1%) samples were D^u negative. Therefore, the D^u test may be used to scale down the burden of RhD negative transfusion by 4.9% in northern Nigeria.

INTRODUCTION

The rhesus (Rh) system was the fourth blood group system to be discovered, yet in terms of clinical importance it is only second to that of the ABO system, because majority of RhD negative individuals become immunized after exposure to RhD positive red cells, and such antibodies can cause severe haemolytic transfusion reactions and haemolytic disease of the new born [1]. Individuals may have red cells that express fewer D sites per red cell than normal and are referred to as D^u positive individuals [1]. The D^u is classified into high-grade, low-grade and very low-grade, which has the lowest expression of the D antigen [1]. D^u positive cells have a weak expression of the D antigen and may be misclassified as D negative cells in routine Rh grouping procedures [1]. However, the more elaborate indirect antiglobin test (IAT) is capable of detecting all grades of D^u with the exception of the very low-grade types [1]. In the Caucasians, the frequency of RhD negativity is about 15% [2]. In contrast to the Caucasian figures, the frequency of RhD negativity in the Nigerian population ranged from less than 1% to about 6% in different ethnic population groups spread across the country. The highest frequency RhD negativity was found in the southern part of the country where frequencies of 6.01% and 5.46% were reported in the south central and south western regions respectively [3,4]. The frequency of RhD negativity is even lower in the northern regions where frequencies of 1.44%

and 0.6% were reported in the north eastern and north western regions respectively [5,6]. This has resulted in RhD negative blood scarcity in Nigerian blood banks, the scarcity being more severe in the northern part of the country.

In clinical practice it usually not necessary to determine whether a RhD negative patient is D^u positive because no harm results from transfusing Rh-negative blood into such patients even if they are D^u positive. This approach is particularly more appropriate for the Caucasian populations in which the frequency of RhD negativity is relatively high and the well developed national blood transfusion services in such countries greatly facilitate the procurement and distribution of RhD negative blood. In contradistinction to the Caucasian nations, the low frequency of RhD negativity in Nigeria is accentuated by low donor drive and sub-optimal national blood transfusion service, which has virtually relegated the daunting responsibilities of donor recruitment and blood collections to individual hospital blood banks [7,8]. Consequently, there is a severe scarcity of RhD negative blood in Nigerian blood banks, a situation that has practically made any request for RhD negative blood a nightmare for both clinicians and blood bank staff. It is therefore rational to attempt to scale down the number of requests for RhD negative blood by subjecting all RhD negative patients who require transfusion to IAT in order to detect those that are D^u positive so that they may be transfused with RhD positive blood. This study was

conducted to determine the frequency of D^u antigen among RhD negative patients scheduled for transfusion with the aim of scaling down the burden of RhD negative transfusion in northern Nigeria.

MATERIALS AND METHODS

This study is a prospective study in which all patients that were scheduled for blood transfusion at the Federal Medical Centre (FMC), Birnin Kudu, northwestern Nigeria and University of Maiduguri Teaching Hospital (UMTH), Maiduguri, northeastern Nigeria were grouped for RhD antigen and those who tested negative for the D antigen were subsequently tested for D^u by the IAT method; making sure that every patient is studied only once. The study was conducted during the years 2004 and 2005 at Birnin Kudu and from January 2005 to December 2006 at Maiduguri. Rhesus grouping and D^u testing were carried out using manual techniques and in accordance with reagent manufacturer’s instructions [9,10]. One drop of saline-reacting polyclonal IgG anti-D blended with monoclonal IgM anti-D (Biotec UK) was mixed with 1 drop of 2% saline suspension of patient red cells in a glass test tube, the mixture was incubated for 15 minutes at room temperature and read macroscopically and macroscopically for agglutination after centrifugation at 500g for 15 seconds. Both positive and negative controls using known D positive and D negative cells were used in each case. Patients’ cells that were not agglutinated were further tested with IAT for detection of D^u. A mixture of 2 drops of anti-D and 1 drop of 3% saline suspension of patient red cells were incubated at 37 ° C for 45 minutes, washed 4 times with 3 ml of saline and in each cycle, 2 drops of anti-human globulin (anti-IgG, Biotec UK) were added after the last wash, the tests were read macroscopically and microscopically after centrifugation at 500g for 15 seconds. Negative controls using known D negative red cells were run in parallel; these tubes must be free of agglutination otherwise the whole procedures for tests and controls must be repeated. Sensitized red cells were used to confirm the validity of all negative tests; agglutination must be visible after addition of sensitized red cells and centrifugation otherwise the test result is invalid and the procedures must be repeated.

RESULTS

The blood samples of a combined total of 2978 patients that were scheduled for transfusion were analyzed at the blood banks of the FMC, Birnin Kudu, northwest Nigeria (896 patients) and UMTH, Maiduguri, northeast Nigeria (2082

patients). Out of the 2978 samples tested with anti-D, 2897 (97.3%) samples were positive while the remaining 81 (2.7%) samples were negative as shown in Table 1.

Figure 1

Table 1: Frequency of RhD Antigen in Patients for Transfusion

BLOOD GROUP	No. OF DONORS	FREQUENCY (%)
RhD Positive	2897	97.3
RhD Negative	81	2.7
Total	2978	100

Out of the 81 RhD negative samples tested for the D^u antigen, 4 (4.9%) samples were positive while the remaining 77 (95.1%) samples were negative as shown in Table 2.

Figure 2

Table 2: Frequency of D Antigen in RhD negative Patients

BLOOD GROUP	No. OF DONORS	FREQUENCY (%)
D ^u Positive	4	4.9
D ^u Negative	77	95.1
Total	81	100

DISCUSSION

The frequency of RhD negative patients in this study was 2.7%, which was higher than the frequencies of 1.44% and 0.6% we reported in earlier studies among blood donors in UMTH, Maiduguri and FMC, Birnin Kudu respectively [5,6]. This apparent disparity was thought to be a reflection of the heterogeneity of the patient populations in the two hospitals. The towns in which the two hospitals are located are cosmopolitan and many of the patients are non-indigenous civil servants and business persons who originated from the southern part of the country where the frequency of RhD negative group is relatively higher. The resultant effect was an elevated frequency of RhD negativity among our patients in comparison to the frequencies that were earlier reported among blood donors, which were almost entirely derived from indigenous northerners with low frequency of RhD negativity [5,6]. Nonetheless, this study showed that 4.9% of our RhD negative patients were actually D^u antigen positive. The identification of D^u positive patients has triple clinical advantage in our environment where RhD negative blood is scarce. Firstly, the major advantage is that such patients can be transfused

with RhD positive donor blood without any risk of sensitization since they do possess the D antigen, which they expressed at lower levels on their red cells [1]. Secondly, valuable and scarce RhD negative blood would be conserved to be used for patients that are genuinely RhD negative. This is particularly important since donor blood is always in short supply and inadequate with respect to clinical requirement of patients in Nigeria [7,8]. Thirdly, female patients that are identified as D^u positive will not be subjected to unnecessary and costly anti-D immunoglobulin injections if they deliver RhD positive babies [11]. This is important in Nigeria where such injections are always imported and hence expensive and not easily affordable by many patients.

The identification of D^u antigen is particularly useful in patients with frequent and high transfusion requirements such as patients with HIV/AIDS, chronic renal failure, malignancies, aplastic anaemia and other bone marrow failures, sickle cell anaemia (SCA) and other cases of chronic anaemias [12,13]. With a population of over 140 million, Nigeria is the most populous black nation and carries a heavy disease burden due to SCA, which affects about 2% of the general population [13]. From global perspective, Nigeria has more SCA patients than any other country in the world [13]. It is therefore imperative that patients with SCA and other frequently transfused patients that are RhD negative should be tested for D^u antigen so that their high transfusion requirements may be easily met with the use of RhD positive blood if they are found to be D^u positive. It must however be appreciated that even though the D^u test has the potential of easing out transfusion requirements if an RhD negative patient is found to be D^u positive, the test must be carried out with caution under rigorously controlled laboratory and reagent conditions so as to avoid the possibility of false positive errors that may lead to wrongful administration of RhD positive blood to RhD negative patients with severe immunological and clinical consequences [1,9].

CONCLUSION

The frequency of D^u antigen in our RhD negative patients was 4.9%. Therefore cautious and controlled application of the D^u test may be used to scale down the burden of RhD negative transfusion by 4.9% in northern Nigeria.

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