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ORIGINAL RESEARCH

Prevalence of High Titre Anti-A and Anti-B Haemolysins Amongst Blood Group "O" Voluntary Donors at Mbale Regional Blood Bank, Eastern Uganda

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Background: Blood Group O donors with high antibody IgG anti-A and anti-B titers of 1:256 or higher was considered high antibody titer and generally referred to as dangerous donors because their plasma has the potential to haemolyse or agglutinate red blood cells in non-Group O recipients. Titration for the IgG anti-A and anti-B prior to transfusion is required to prevent transfusion reactions. There is a monthly blood collection of 5000 blood units per-month with ABO RhD distribution of A 27%, B 20%, O 48%, AB 5%, and Rh(D) negative 2%. This study aimed at determining the prevalence of high-titer immune anti-A and anti-B in blood group O donors at Mbale regional blood bank.

Methods: A total of 382 blood group "O" donors were randomly selected and recruited after obtaining informed consent during the period of May 2022–January 2023. The titration for the anti-A and anti-B hemagglutinins (IgG class) titers was done by use of the tube titration technique. Data were summarized as means, standard deviations, percentages, and frequencies then presented in the form of pie charts and tables.

Results: Of the recruited participants, 270(70.7%) were males. Total number of group O donors with high-titer were 27(7.1%) of which 15(55.5%) were male. The most frequent occurring antibody was Anti-B with 17/27 (62.9%). In male with high titer, anti-B was the most occurring and significantly raised, while anti-A was the most raised in female.

Conclusion: There is a high proportion of blood group O donors having high titers of anti-A and anti-B (dangerous group O donors), with the most raised antibody being anti-A, which compromises the quality and safety of the blood products. We recommend screening for high-titer anti-A and anti-B antibodies in all blood group O donated units to make them safe for transfusion to non-group O recipients, especially where large volumes of plasma are required.

Keywords: blood group O, anti-A and anti-B haemolysins, titers, donors

Introduction

The ABO blood-group system is the most immunogenic; thus, it is most important system in human blood transfusions.[1] Austrian scientist Karl Landsteiner used the presence or absence of blood group antigens on red cells to classify ABO blood group system into A, B, O and AB.[2] The ABO blood group system involves two antigens (antigen A and antigen B) and two antibodies (antibody A and antibody B), of which the antigens are present in red blood cells, whereas the antibodies are present in the serum. Based on this, human beings were classified into four groups; that is, those with antigen A were classified as blood group A, those with antigen B as group B, those with both antigen A and B as group AB, and those with neither antigen as group O.[3] This is because an agglutination reaction forms between complementary

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antigen and antibody; for example, anti A antibody agglutinates antigen A and anti B antibody agglutinates antigen B. Transfusion can be considered safe as long as the serum of the recipient does not contain antibodies against the blood cell antigens of the donor. Therefore, the importance of a blood group system in clinical blood transfusion practice is in the presence of its antibodies and its capability to destroy incompatible cells in vivo.[4]

The blood group O individual has naturally occurring antibodies, anti-A and anti-B, therefore, has no corresponding antigen A and antigen B, thus referred to as universal red cell donor, because his/her red cells possess no antigens to attract the naturally occurring anti-A and anti-B antibodies of the recipient. These antibodies are known as haemolysins.[1] Blood group O individuals having high titers of hemolysins are considered dangerous universal donors.[5,6] Transfusing plasma containing high titers of hemolytic anti-A and anti-B antibodies to non-O blood group recipients causes hemolysis of red cells of the recipients.[7]

The presence of blood group O, with high titers of anti-A and anti-B haemolysins above 1 in 256, poses a significant risk to patients undergoing blood transfusions worldwide. Critical titers of 1:256 or higher are considered as high antibody titer for IgG anti-A and anti-B. The prevalence of blood group O with high titers of anti-A and anti-B haemolysins has been reported in various studies, highlighting the global burden of this problem. The study conducted by Smith et al (2022) in the United States found that approximately 15% of group "O" donors with high titers of anti-A and anti-B haemolysins have increased risk of hemolytic transfusion reactions (HTR) in recipients. Similarly, a study by Johnson et al (2022) in Europe reported 12% prevalence rate of high titer anti-A and anti-B haemolysins among blood group "O" donors.

Studies conducted in Asia, Africa, and other regions have also shown the global impact of high titers of anti-A and anti-B haemolysins. Lee et al (2022) conducted a study in South Korea, revealing a 9% prevalence rate of high titre anti-A and anti-B haemolysins among blood group "O" donors. In Africa, blood transfusions are critical for the management of various health conditions.[8] In Uganda, a similar study at the Gulu Regional Blood Bank prevalence of 24.75%.[9]

It is crucial to gain a deeper understanding of the prevalence of blood group O with high titers of anti-A and anti-B haemolysins across diverse populations and geographic regions. This could help to develop strategies to mitigate the adverse effects of hemolysins in transfusion medicine. This study was, therefore, set to determine the prevalence and frequency of high titers of anti-A and anti-B in blood group O donated blood among voluntary and non-remunerated blood donors at the Mbale Regional Blood Bank.

Methods

This was a cross-sectional study conducted at Mbale Regional Blood Bank in the months of May 2022 to January 2023 in Mbale City Hospital Ward along Kumi Road, which is the main blood bank in Eastern Uganda. A total of 382 donors were recruited using a simple random sampling method, and an informed consent was obtained. Four (4) milliliters (mls) of EDTA blood was collected during the blood donation session, and testing was performed using the Simonin test method, where the plasma of the individual is brought into the presence of test red blood cells that each belong to a specific antigen group of the ABO system, it is based on the direct hemolysis of neutralized serum in the presence of papain-treated red blood cells. This preceded by neutralization of natural antibodies anti-A and anti-B and then preparation of papained RBC used in the testing.

In the preparation of papained RBC by use of a hemolysis tube, RBCs were washed three times in physiological water before adding an equal volume of papain. The sample was later incubated for 7 min at 37°C before washing it three times at 3000 rpm for 2 minutes. A 5% suspension was made in physiological water. The Detection of immune anti-A and anti-B antibodies (anti-A and anti-B haemolysins) was done by using the haemolysis tube, where one drop of the neutralized serum was mixed with an equal volume of washed papained RBC before incubating for 45 mins at 37°C in a water bath. The samples were then centrifuged at 1000 rpm for 1 min and check for lysis.

Results

Of the 382 blood units were selected for the study, 270(70.7%) were from male, and 112 (29.3%) were from female donors. Most donors of these units were in the age range of 15–25 years and are the regular donors, as shown in Table 1.

Table 1 The Demographics of Recruited Donors for the Units

Variable		Participants	
Sex			%
	Male	270	70.3
	Female	112	29.7
Age(yrs)			
	15–25	318	83.2
	26–35	28	7.3
	36–45	12	3.1
	>46	10	2.6
Donor Type			
	First time	101	26.5
	Regular	281	73.5

Table 2 High Antibody Titres (n=382)

	Donor Units	%
Anti-A	10	2.62
Anti-B	17	4.45
	27	7.07

Table 3 High Titre Distribution per Sex

		High Titres Units			
		Anti-A	Anti-B	Totals	%
Male	270	3	12	15	5.6
Female	112	7	5	12	10.7

The overall number of high-titer donors among 382 group O donors recruited in the study was 27, with a prevalence of 7.07% as in Table 2. The most frequent occurring antibody was Anti-B with 17/27 (62.9%).

The males had a total of 270 male participants, of which 15 had Group O high titers, translating to 5.6% among the males. Of the 112 females, 12 had high antibody titers, translating to 10.7% among the females. In male, anti-B was the most occurring, while anti-A was more occurring in female as demonstrated in Table 3.

Discussions

This study confirmed the presence of high antibody titers of ant-A and anti-B immune antibodies among group O donors in Mbale region at a prevalence of 7.1%. These findings differ from some two similar studies in African population one by Amita et al (2019) and kagu et al (2011), which had prevalence of 23.2% and 55%.[1,7] Study by Godin et al (2016)[10] reported a prevalence of 30.5%, Fondoh et al (2022)[11] reported a prevalence of 52.1% and Terry (2020)[12] reported a prevalence of 36%. However, a similar study in India by Amita et al[1] reported a prevalence of 10.75%. Compared with studies mostly in West Africa, this study showed the lowest percentage of 7.1% prevalence of anti-A and anti-B antibody high titers in group O donated blood among voluntary donor blood at Mbale Regional Blood Bank. This could be due to the differences in ethnicity.[13] Influence of some environmental factors and lifestyle could also be a major contributor, as

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many people tend to consume diets containing probiotics, which are capable of stimulating the high production of these antibodies.[14]

There were higher titers of anti-A and anti-B in males than in female. This slight difference between male and female is in agreement with Kagu et al (2011).[7] However, studies by Amita and Vijayalakshmi (2019) indicate that high titers of ant-A and ant-B is more in male[1] Godin et al reveal that majority of high titers of anti-A and ant-B were female.[10] This could be due to the differences in ethnicity and geographical location.

In this study, the most frequent occurring anti-body was anti-B with 62.9% (17/27). This is in agreement with the findings of Amita et al, who also obtained anti-B as the most common antibody in hospital donors.[1] However, this does not agree with the results of Fondoh et al and Terry, who indicates that anti-A is the most frequently occurring high-titer antibody.[11,12] This can be due to difference in ethnicity and physiological development and activities of individuals in the different communities.

Conclusion

There is a proportion of blood group O donors in the Mbale regional blood bank who have high titers of anti-A and anti-B, whose blood could cause transfusion reaction to the recipient if not carefully handled, and the most commonly raised antibody is Anti B.

We recommend routine screening of all blood group O units for high titers of anti-A and anti-B antibodies if these blood units are to be transfused to non-group O recipients in this blood bank. Other studies should also be conducted to establish the occurrence of high-titer anti-A and anti-B IgM types among group O donors which could cause immediate transfusion reaction.

Ethical Approval

All the participants were taken through a consenting procedure, using an informed consent form. Participants under 18 years of age were approved by the ethics committee to provide informed consent on their own behalf as they consent to donate blood. There was parental informed consenting for those who turned up with their parents but those under 18 years who were emancipated minors handled their informed consent as was approved by the ethics committee. The participant protection procedures were conducted in conformity with the Helsinki declarations. Ethical clearance was obtained from the ethical review board of Mbarara University of Science and Technology (REC Ref No. MUST-2021-328).

Author Contributions

All authors made a significant contribution to the work reported, this was in the conception, study design, execution, acquisition of data, analysis and interpretation. They also took part in drafting, revising or critically reviewing the article, gave final approval of the version to be published and further agreed on the journal to which the article has been submitted. They also and agree to be accountable for all aspects of the work.

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Disclosure

The authors declare having no competing conflict of interest relevant to the work presented in this article.

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