ORIGINAL ARTICLE

Prevalence of haemolysins in blood donors in Nnamdi Azikiwe University Teaching Hospital, Nnewi, Anambra State, Nigeria

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ABSTRACT

Background: The presence of high titres of haemolysins (lytic antibodies) in the sera of donors could predispose to adverse blood transfusion reactions.

Objective: To evaluate the prevalence of haemolysins among blood donors at the Nnamdi Azikiwe University Teaching Hospital, Nnewi, Anambra State.

Methodology: A total of 1,370 donors were randomly selected for the study between April and September 2014. Each donor had 3mls of blood collected; 2mls was dispensed into plain bottles and serum extracted for haemolysin testing, while 1ml was dispensed into ethylene diaminetetracetic acid (EDTA) bottle for blood grouping. Haemolysin testing was done by reacting freshly prepared standard A, B, AB and O red cell suspensions with sera from each participant while blood grouping was done using commercial anti-sera kits. Ethical approval was obtained from the institution's Review Committee, and all participants gave informed consent.

Results: The general prevalence of haemolysins was 220/1370 (16.06%), while alpha, beta and alpha plus beta haemolysins were detected in 70 (5.11%), 90 (6.5%) and 60 (4.38%) of donors, respectively. The number of donors with blood groups A, B and O were 265, 165 and 940, while the distribution of haemolysins was 18.87%, 18.18% and 14.89%, respectively. Of the 220 donors in whom haemolysins were detected, 18.2% (40/220), 27.3% (60/220), 40.9% (90/220) and 13.6% (30/220) were aged 18-27years, 28-37years, 38-47years and 48-57years, respectively.

Conclusion: There is a high prevalence of haemolysins in our donors, particularly in those with blood group A and in the age range of 38-47years.

***Keywords**: Blood groups, lytic antibodies, South-East Nigeria, transfusion*

INTRODUCTION

Allogenic blood has never been more in demand than it is today, particularly, in the developing countries. One of the biggest challenges to transfusion science is accessing safe and adequate quantities of blood and blood products. One strategy geared towards the optimum utilization of scarce donor units is the use of blood and blood products against the ABO blood group barrier.

Globally, approximately 80million units of blood are donated each year.[1] Out of these, only 2million units are donated in sub-

Saharan Africa, where the need for blood transfusions is great and supply limited because directed donation (also referred to as family replacement) is often the rule.[2] In a bid to meet the increasing transfusion needs, red cell and plasma products are transfused against the ABO blood group barrier. Unfortunately, the occurrence of anti-A (alpha-haemolysin) and anti-B (beta-haemolysin) in group O donors was reported to be high in African and Asian populations compared to that of Caucasians.[3,4,5,6,7] The higher frequency of haemolysin in these populations has been attributed to higher loads of malaria parasitaemia and intestinal parasitic infections, and is postulated to account for the high frequency of ABO-haemolytic disease of the newborn (HDN) in Africans.[8,9]

Various prevalence rates have been reported in Asia, ranging from 28% to 62.8% as noted in two medical colleges in India.[10,11] In the same vein, several works have been carried out in Nigeria, aimed at determining the prevalence rates of haemolysin among blood donors across the various geographical zones. A prevalence rate of 23.2% was reported in Ilorin in South-West zone of Nigeria.[12] In Calabar, South-South zone, a prevalence rate of 28.5% was observed, while in Sokoto, North-West, 10% was reported.[13,14] In North-Central, Nigeria (Jos and Zaria) prevalence rates of 38.1% and 32.3%, were noted.[6,15]

Correspondingly, in North-East Nigeria, a collaborative study involving two tertiary Hospitals; the University of Maiduguri Teaching Hospital and Aminu Kano Teaching Hospital, a prevalence rate of 55.4%, was reported.[16]Notwithstanding the high frequency reported in different regions of Nigeria, blood group O remains the most common and most prescribed blood group type, particularly when blood and blood product transfusion go against ABO group barrier in Nigeria.[16]

There are no reports on the prevalence rates of serum haemolysins in South–East Nigeria in published literature. This observation, therefore, necessitated this study, which was aimed at determining the prevalence of serum haemolysins among blood donors in a tertiary health institution in Nnewi, South-East Nigeria.

METHODOLOGY

Study Area: The site for this study was Nnamdi Azikiwe University Teaching Hospital (NAUTH) Nnewi, Anambra State, South-East Nigeria. It is a tertiary facility that serves the entire State with a population of four million, one hundred and seventy-seven thousand, eight hundred and twenty-eight (4,177,828) according to the 2006 census (National Bureau of Statistics, 2009). It also serves its neighbouring States; Delta, Imo and Enugu. Nnewi is a metropolitan city encompassing two Local Government Areas; Nnewi North and South.

Study Population / Sampling Frame: The study population included healthy blood donors who presented at the Nnamdi Azikiwe University Teaching Hospital blood bank for blood donation. A total of one thousand, three hundred and seventy donors were recruited by simple random sampling (1 out of every 3 presenting donor) between the months of April and September 2014. Only donors who satisfied the criteria for donor recruitment, and gave consent were enrolled.[17] Those with a history of auto-immune disorders, on long term immunosuppression or on medications such as penicillin antibiotics, rifampicin, methyldopa or thiazide diuretics were excluded from the study.

Ethical Consideration: Ethical Approval for this research was obtained from the Review Committee of the hospital.

Method of Sample Collection: The blood donors had 3ml of blood collected following standard protocol; 2ml was dispensed into plain containers and allowed to clot undisturbed. The expressed serum was separated by centrifugation and dispensed into clean test tubes for haemolysin testing, while the remaining 1ml was dispensed into ethylene diaminetetracetic acid (EDTA) bottle for blood grouping.

Study Protocol: Washed pooled standard A, B, AB and O red cell suspensions of 5% concentration respectively, were prepared

every morning for the testing, following standard protocol.[18] Donor sera were adsorbed using the method of Dacie and Lewis.[18] The ABO blood groups were determined using known (commercial) antisera (*Carper Laboratories, UK*). The reactions of anti-A, anti-B, and anti-AB sera were checked using known A cells, B cells, AB cells and O cells. Both the controls and tests were run together and results recorded in standard plus system format.

Statistical Analysis: All data analyses were done using SPSS version 20 computer software (SPSS Inc., Chicago IL, USA), and the prevalence of haemolysins across gender, blood and age groups were presented as percentages.

RESULTS

The study investigated a total of 1370 blood donors for the presence of lytic antibodies (haemolysins). Out of the 1370 blood donors screened, 220 donors tested positive to haemolysin, giving a prevalence rate of 16.1%.

The reactivity distribution pattern for the various lytic antibodies were; 70 (5.11%) for alpha (α) haemolysin, 90 (6.57%) for beta (β) haemolysin while alpha plus beta (α+β) haemolysin was 60 (4.38%), *Table 1*. The distribution of haemolysins among the various donor blood groups were 265 (18.87%), 165 (18.18%) and 940 (14.89%) for blood groups A, B, and O, respectively, as shown in *Table 2*.

Table 1. The distribution of haemolysins among blood donors

Alpha Haemolysin (α-haemolysin)		**Beta Haemolysin (β-haemolysin)**		**Alpha + Beta Haemolysin (α+β-haemolysin)**		**Number of Donors with Haemolysin**		**Total number of donors screened**
Present	%	Present	%	Present	%	Present	%	
70	5.11	90	6.57	60	4.38	220	16.1	1370

Table 2. The distribution of alpha and beta haemolysins by blood groups

Blood group	**Blood group Frequency**	**Total No of donors with Haemolysin present**		**Alpha+BetaHaemolysin**		**Alpha Haemolysin**		**Beta Haemolysin**	
		Present	(%)	Present	(%)	Present	(%)	Present	(%)
A	265	50	18.87	0	0	0	0	50	18.87
B	165	30	18.18	0	0	30	18.18	0	0
O	940	140	14.89	60	6.38	40	4.26	40	4.26
TOTAL	1370	220	16.06	60	4.38	70	5.11	90	6.57

In this study, there were 1060 male donors and 310 females; giving a male:female ratio of 3.4:1. Female donors recorded 22.6% (70/310) positivity for haemolysins while it was 14.1% (150/1060) in male donors, *see Table 3*.Of the 220 donors in whom haemolysins were detected, 18.2% (40/220), 27.3% (60/220), 40.9% (90/220) and 13.6% (30/220) were ages 18-27years, 28-37 years, 38-47 years and 48-57years, respectively and none in age groups 48-57years and ≥ 58 years, respectively (Table 4).

Table 3. Frequency of alpha and beta haemolysin by gender

		Total positive for Haemolysin		Alpha+Beta Haemolysin		Alpha Haemolysin		Beta Haemolysin	
Gender	**Donors**	**Present**	**(%)**	**Present**	**(%)**	**Present**	**(%)**	**Present**	**(%)**
Male	1060	150	14.14	30	2.83	40	3.77	80	7.55
Female	310	70	22.58	30	9.68	30	9.68	10	3.23
Total	1370	220	16.06	60	4.38	70	5.11	90	6.57

Table 4. Distribution of the blood donors positive for haemolysin by age groupings

Age (years)	**18-27**	**28-37**	**38-47**	**48-57**	**≥58**
Frequency	40/220 (18.18%)	60/220 (27.27%)	90/220 (40.91%)	30/220 (13.64%)	0/22 (0%)

DISCUSSION

One strategy geared towards the optimum utilization of scarce donor units is the use of blood and blood products against ABO blood group barrier; especially in areas where blood donors are scarce such as in Nigeria. The non-availability of donor blood of all ABO groups at all times necessitates the transfusion of non-identical ABO donor blood to certain recipients. In this situation, a number of blood banks fail to screen for the presence of lytic antibodies (haemolysins) before they release units for transfusion against ABO blood group barrier.

The blood group distribution in this study shows that blood group O (69%) is the most prescribed and most utilized blood group type in our environment. This is consistent with the report of Kagu, *et al,* in North-East Nigeria, and appears to mirror the observation that blood group O has the highest distribution in the general population.[16,19]

This study observed a high prevalence of haemolysins among the donors (16.1%). The production of haemolysins could follow events such as ABO heterospecific pregnancy, mismatched blood transfusion or sensitization by A or B-like substances.[20] Our finding is comparable to reports from Ilorin (23.3%), higher than was reported in Sokoto (10%), and lower than reports from Calabar (28.5%), Jos/Zaria (38.1% and 32.3%, respectively), and Kano / Maiduguri (55.4%).[6,12,13,14,15,16] These regional differences could be as a result of geographical variations in the intensity of some infectious diseases. Indeed, studies have shown that the serum levels of haemolysin is dependent on malaria parasitaemia, as well as, intestinal helminthiasis.[8,9]

Our study showed that blood group A donors had the highest prevalence of haemolysins (18.87%), *see Table 2*. This is in contrast to the reports of Emeribe and Anyanwu, *et al,* in which prevalence was noted to be higher in donors with blood group O.[21,22] These observations generally underscore the potential dangers inherent in transfusing patients against ABO blood barriers, without first screening such units for haemolysins.

We had a higher number of male donors compared with females in this study, *see Table 3*. This is in agreement with earlier studies which alluded to a male dominated donor pool in Nigeria and the West African sub-region.[23,24,25] The female donors, however, showed a higher positivity for haemolysin compared to males (70/310; 22.6% vs. 150/1060; 14.1%). Previous reports on the

relationship between serum haemolysin and gender have been conflicting. While the report of Milison was in agreement with our observation, that of Olawumi and Anyanwu, *et al*, showed no change and higher serum levels in males, respectively.[12,22,26]

A number of studies had reported that serum haemolysin levels increased with age.[22,26] On the contrary, Olawunmi, *et al* reported that haemolysin levels remained the same, irrespective of age, among blood donors in Ilorin, South-West Nigeria.[12] Our observation in this study re-iterated the former reports; donors within the 38-47year age group had the highest positivity (40.9%; 90/220), of haemolysins as shown in *Table 4*.

CONCLUSION

A high prevalence rate of serum haemolysin was noted in this study, particularly in blood group A donors and the 38-47year age group. This observation calls for concern, and we therefore, recommend that as much as possible, group-specific donor units should be selected for cross-matching for patients. In situations where this is not practicable, however, such units must be screened for α and β haemolysins, in order to mitigate the adverse transfusion reaction that could follow such transfusions.

LIMITATION OF THE STUDY:

This study did not quantify the titres of haemolysin in the donors.

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