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Original Article

Pattern of Transfusion Transmissible Infections, ABO and RH Blood Groups, and Hemoglobin Electrophoresis Phenotypes in Voluntary Non-Remunerated Blood Donors in Ibadan, Nigeria

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Background: Voluntary Non-Remunerated Donors (VNRD) are the safest source of blood and blood products with lower seroprevalence rates of transfusion transmission infections (TTIs); however, they constitute the minority of donors in Nigeria. This study aimed to describe the pattern of TTIs, ABO and Rh blood groups, and hemoglobin electrophoresis among VNRD.

Methods: This was a 7-year retrospective review of VNRD data on demographic, seroprevalence of TTIs, ABO, and Rh blood groups, and hemoglobin electrophoresis over 7 years in a tertiary health facility in Nigeria.

Results: A total of 6003 VNRD donated over the study period. The overall seroprevalence (95% CI) of hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency virus (HIV), and Syphilis were 3.7% (3.2, 4.2), 1.2% (1.0), 1.6% (1.3, 1.9), and 0.5% (0.3, 0.7), respectively. Males' seroprevalence rates were significantly higher than females for HCV (1.4% vs. 0.5%; $P = 0.032$) and HIV (1.8% vs. 0.7%; $P = 0.013$). The prevalence of blood groups O, A, B, and AB were 55.1%, 21.2%, 20.0%, and 3.9%, respectively, while hemoglobin electrophoretic phenotypes A, A, Haemoglobin AS (AS), and Haemoglobin AC (AC) were 72.4%, 21.7%, and 5.9%, respectively.

Conclusions: This study revealed that HBV has the highest seroprevalence, and the TTI seroprevalence is higher among male than female donors.

Key words: VNRD, ABO blood group, HIV, transfusion transmission infections

INTRODUCTION

Voluntary non-remunerated blood donation (VNRBD) forms the foundation for safe blood transfusion globally, and thus the World Health Organization (WHO) has recommended that every unit of blood and blood components should be produced from VNRBD. [1] The number of VNRD in Nigeria is, however, negligible, as most donations are from two categories, namely paid and family replacement donors. [2–5] Blood units from VNRD have been reported to be safe and have lower prevalence rates of transfusion transmissible infections (TTIs) when compared with other groups of donors. [5,6] It is

believed that VNRD's level of awareness of the TTIs associated risk behavior is high, and their resolve to continue to donate blood makes them live a healthy lifestyle. Efforts to ensure safe blood should be geared toward formulating policies to retain VNRD and find ways to recruit more of them.

ABO and Rh D blood groups, the two most immunogenic among the blood group systems, typing is recommended pretransfusion to ensure blood compatibility between the donor and the recipient. [7] However, the antigen frequency of ABO/Rh blood groups varies from one population to another. Generally, blood group O has been reported to be the most common ABO blood group, followed by A, B, and AB. Also, Rh D positivity is more prevalent than Rh D negativity. [8-10] It has been documented that AB blood units are not usually requested for in the blood banks, while it is also quite difficult to recruit Rh D-negative individuals for blood donation. [11,12]

The hemoglobin phenotype of the donor is also an important consideration in blood transfusion services because people living with sickle cell disease, for example, are not meant to be transfused with blood with sickle cell trait (AS), as this may worsen their clinical conditions. HbA is reported to be the most common hemoglobin phenotype, followed by AS and AC in blood donors. [13-15] Hemoglobin electrophoresis is important to determine the pool of blood units that might be needed for persons living with sickle cell disease when in need of blood transfusion.

This study, therefore, aimed to determine the prevalence rate of TTIs (hepatitis B virus [HBV], hepatitis C virus [HCV], human immunodeficiency virus [HIV], and Syphilis), ABO and RhD blood groups, and hemoglobin electrophoresis patterns in VNRD.

METHODS

Study Design and Setting

This is a retrospective review of voluntary non-remunerated blood donors who donated between 2011 and 2017 at the University College Hospital Blood Bank, Ibadan. The hospital is a tertiary referral center serving southwestern Nigeria. This study was carried out to determine the prevalence of TTIs, ABO, and Rh blood groups, and hemoglobin electrophoresis phenotypes in the donors. These tests are routinely done for all blood donors in the hospital.

Study Population and Sample Size

All VNRD who met the blood donation criteria according to WHO guidelines and donated over the past 7 years constituted the study population. Data from repeat blood donors who were identified by their unique donor identification numbers were analyzed once to avoid duplication of data. There were 6003 VNRD during this period, of which 5989 (99.77%), 5753 (95.84%), and 5246 (87.39%) had complete TTIs, ABO/Rh blood grouping, and hemoglobin electrophoresis results, respectively, and were analyzed. Flow diagram of cases selection (**Figure 1**).

ABO/Rh blood grouping was performed by the microwell technique according to a method described by Rowley et al. [16].

Data Collection

The data were extracted from the Voluntary Blood Donor Register in the blood bank. Information extracted included sociodemographic (age, gender), hemoglobin phenotype, ABO, Rh D blood groups, and results of TTIs (HBV, HCV, HIV, and Syphilis).

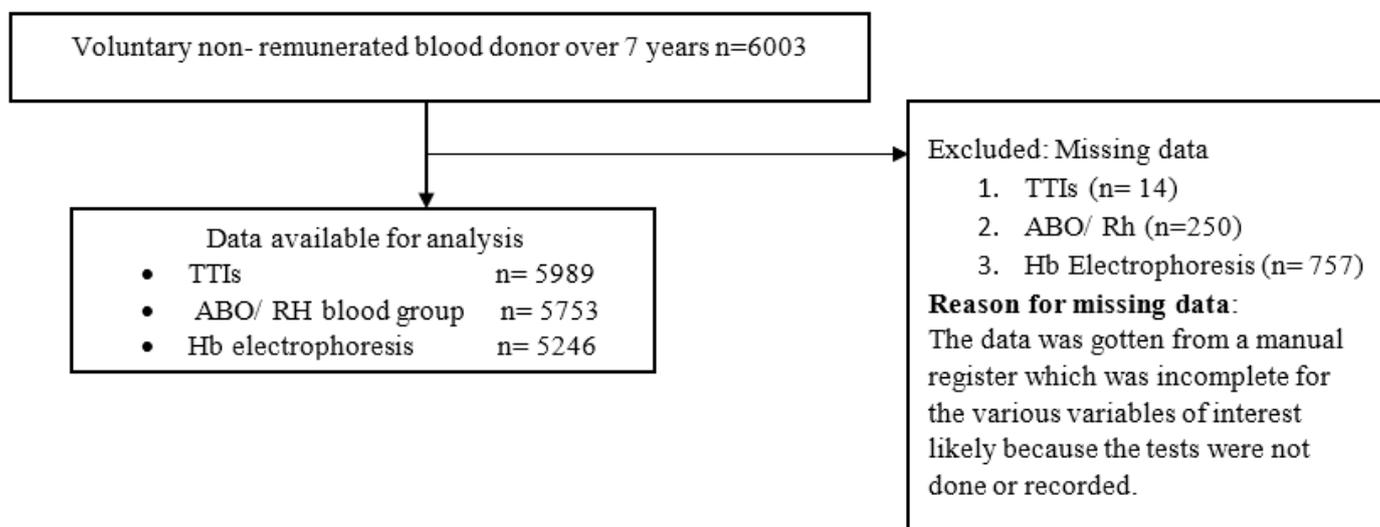


Figure 1: Flow diagram showing total records of voluntary non-remunerated blood donors (n = 6003), exclusion, and final sample sizes for TTIs, ABO/Rh blood groups, and hemoglobin electrophoresis. The missing data for each category were different because the blood samples were run on different benches and recorded by different personnel.

Hemoglobin electrophoresis was performed by alkaline cellulose electrophoresis (pH: 8.4) using an electrophoresis tank manufactured by Junyi Electrophoresis Co., Ltd, China. Hemoglobin phenotype was read from ponceau S-stained paper compared with control samples A, S, and C. TTIs were carried out by the ELISA method according to the standard recommended by the Nigerian Federal Ministry of Health.

Data Analysis

Data were analyzed using the Statistical Package for the Social Sciences (SPSS), IBM version 25. Continuous variables (age, weight, and hemoglobin) were normally distributed (Kolmogorov-Smirnov test of normality) and were summarized as means and standard deviation, and bivariate analysis was performed using the independent sample *t*-test. Categorical variables were summarized as percentages, and multivariable analysis was performed by Pearson's Chi-square and Fisher's Exact Test where applicable. Age group, gender, and year of testing were explored as predictors of positive TTIs using Logistic regression analysis. All tests were two-sided, and statistical significance was set at a probability value of *P* < 0.05.

Ethical Consideration

Approval was obtained from the University of Ibadan/ University College Hospital Health Research Ethics Committee (UI/EC/25/01019). Data obtained were stored without identifiers in a password-protected computer that was only accessible to the researchers.

RESULTS

A total of 6003 voluntary non-remunerated blood donors were recorded over the 7 years.

Demographic Characteristics of the Donors

The mean age of the donors was 31.6 ± 10.3 years. The majority (78.4% [4893/5928]) of the donors were under 40 years of age. The male constituted 81.6% (4888) of the donors, and the male-to-female ratio was 4.5:1. There was a significant difference in the mean between both genders (male, 32.0 ± 10.1 vs. female, 29.6 ± 11.1 years; mean diff = 2.3 years; SE = 0.34; *t* = 6.91; *P* < 0.0001; **Table 1**).

Prevalence of Transfusion-Transmissible Infection in the Donors

The analysis of the frequency of TTIs shows the prevalence estimates (95% CI) of HBV, HCV, HIV, and Syphilis were 3.7% (3.2, 4.2), 1.2% (1.0), 1.6% (1.3, 1.9), and 0.5% (0.3, 0.7), respectively, over the 7 years.

Hemoglobin Electrophoresis Pattern of the Donors

The hemoglobin electrophoresis distribution pattern of the donors, as shown in **Table 1**, reveals that hemoglobin A accounted for the majority (72.3%, 3928/5246), and hemoglobin variant AC was the least 322 (5.9%, 322/5246).

Table 1: Age, gender, hemoglobin electrophoresis, and blood group distribution among voluntary non-remunerated blood donors.

Variables	Mean ± SD	Frequency	Percentage (%)	Estimate (%)	95% CI
Age (years)		(n = 5928)			
17–39		4649	78.4		
≥40		1280	21.6		
Average	31.6 (10.3)				
Gender		(n = 5990)			
Female		1100	18.4		
Male		4890	81.6		
Hb		(n = 6003)			
Electrophoresis					
AA				0.723	0.711, 0.735
AS				0.217	0.206, 0.228
AC				0.059	0.053, 0.066
Blood group		(n = 6003)			
O Rh positive				0.509	0.496, 0.522
A Rh-positive				0.199	0.189, 0.209
B Rh positive				0.188	0.178, 0.198
AB Rh positive				0.037	0.032, 0.048
O Rh negative				0.042	0.037, 0.048
A Rh-negative				0.013	0.010, 0.016
B Rh negative				0.012	0.009, 0.015
AB Rh negative				0.002	0.001, 0.003

95% CI: 95% confidence interval (lower, upper).

ABO and Rh Blood Group Distribution Among the Donors

The frequency distribution of the blood donors was presented in **Table 1**. Blood group O constituted the majority (55.1%, 2925/5753), while the least common was AB (3.9%, 221/5754). Rh-positive blood type was the most common (93.19%, 5362/5754).

Bivariate Analysis of TTIs and Gender, Age Group, and Year of Blood Donation

We explored the rates of the transfusion-transmissible infections among males and females using the chi-square test of association. The result revealed that male donors were significantly more likely to test positive for HCV and HIV infection ($\chi^2 = 6.36, P = 0.032$ and $\chi^2 = 06.20, P = 0.013$, respectively). However, similar patterns were observed for HBV and syphilis. Age groups and years of testing were not found to be associated with TTIs positivity. Chi-square for trend showed that the rate of HBV positivity decreased from 2011 to 2013, and then a gradual marginal increase was observed thereafter ($\chi^2 = 10.68, P = 0.026$). HCV, HIV, and Syphilis, though, showed a similar pattern, but these did not reach statistical significance (**Tables 2-5**).

Logistic Regression for the Predictors of TTIs Positivity

There was no identifiable predictor of TTIs among age group, gender, and year of donation for the rates, as the Omnibus Tests of Model Coefficients and Wald statistics were not significant ($P > 0.05$), and ORs all contained 1.

DISCUSSION

The provision of safe blood in Sub-Saharan Africa has largely been negatively impacted by the low rate of voluntary blood donation and the relatively high prevalence rate of TTIs. [5,17-19] High population prevalence of Sickle cell trait (1 in 4) and sickle cell disease is also a cause of/for concern in the blood supply because of the need to provide HbA blood units for persons with Sickle cell disease. This study was designed to determine the prevalence of TTIs and population distribution of ABO, Rh Blood groups, as well as Hemoglobin electrophoresis pattern in voluntary non-remunerated blood donors.

In this index study, the majority of the VNRD were between 18 and 40 years. This pattern is consistent with similar findings in the Nigerian and African populations. [3,20-23] This

Table 2: Pattern of transfusion transmissible infections among voluntary non-remunerated blood donors.

Variable TTIs	Year of donation							
	2011 n = 901 n (%)	2012 n = 860 n (%)	2013 n = 1060 n (%)	2014 n = 566 n (%)	2015 n = 215 n (%)	2016 n = 1024 n (%)	2017 n = 1370 n (%)	Total n = 6003 n (%)
Syphilis								
Negative	901 (99.4)	857 (99.7)	1058 (99.8)	566 (100)	214 (99.5)	1017 (99.3)	1361 (99.2)	5974 (99.5)
Positive	5 (0.6)	3 (0.3)	2 (0.2)	0 (0)	1 (0.5)	7 (0.7)	10 (0.7)	28 (0.5)
HCV								
Negative	891 (98.2)	851 (99.0)	1053 (99.3)	566 (100)	214 (99.5)	1012 (98.8)	1351 (98.5)	5938 (98.8)
Positive	16 (1.8)	9 (1.0)	7 (0.7)	0 (0)	1 (0.5)	12 (1.2)	21 (1.5)	66 (1.2)
HBV								
Negative	879 (97.0)	825 (95.9)	1030 (97.2)	566 (100)	207 (96.3)	975 (95.2)	1312 (95.6)	5794 (96.3)
Positive	27 (3.0)	35 (4.1)	30 (2.8)	0 (0)	8 (3.7)	49 (4.8)	60 (4.4)	209 (3.7)
HIV								
Negative	890 (98.2)	847 (98.5)	1050 (99.1)	565 (99.8)	210 (97.7)	1001 (97.8)	1352 (98.5)	5915 (98.4)
Positive	16 (1.8)	13 (1.5)	10 (0.9)	1 (0.2)	5 (2.3)	23 (2.2)	20 (1.5)	88 (1.6)

HCV: hepatitis C virus; HBV: hepatitis B virus; HIV: human immunodeficiency virus; TTIs: transfusion-transmissible infections.

Table 3: Rates of transfusion transmissible infections by years of study.

TTI	Year of donation								P	P for trend
	1 2011	2 2012	3 2013	4 2014	5 2015	6 2016	7 2017			
No. in group	906	860	1060	566	215	1024	1312			
Percentage of infection										
HBV	3.0	4.1	2.8	2.7	3.7	4.8	4.4	0.090	0.026**	
HCV	1.8	1.0	0.7	1.1	0.5	1.2	1.5	0.272	0.810	
HIV	1.8	1.5	0.9	1.2	2.3	2.2	1.5	0.294	0.534	
Syphilis	0.6	0.3	0.2	0.2	0.5	0.7	0.7	0.402	0.134	

HCV: hepatitis C virus; HBV: hepatitis B virus; HIV: human immunodeficiency virus; TTIs: transfusion-transmissible infections.

Chi-square linear-by-linear association.

** P value < 0.05.

Table 4: Association between gender and age group characteristics and transfusion transmissible infections.

Variable	TTI							
	HBV (n=5988)		HCV (n=5989)		HIV (n=5989)		Syphilis (n=5988)	
	Pos	Neg	Pos	Neg	Pos	Neg	Pos	Neg
	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
Gender								
Male	178 (3.6)	4710 (96.4)	62 (1.3)	4827 (98.7)	81 (1.7)	4808 (98.3)	23 (0.5)	4862 (99.5)
Female	29 (2.6)	1066 (97.4)	4 (0.4)	1096 (99.6)	7 (0.6)	1093 (99.4)	5 (0.5)	1095 (99.5)
	$\chi^2 = 2.72$	$p = 0.099$	$\chi^2 = 6.74$	$p = 0.009^{**}$	$\chi^2 = 6.46$	$p = 0.011^{**}$	$\chi^2 = 0.23$	$p = 0.891$
Age group (years)								
17–39	165 (3.6)	4481 (96.4)	50 (1.1)	4597 (98.9)	66 (1.4)	4580 (98.6)	18 (0.4)	4628 (99.6)
≥40	43 (3.4)	1237 (96.6)	15 (1.2)	1265 (98.8)	21 (1.6)	125 (98.4)	10 (0.8)	1270 (99.2)
	$\chi^2 = 0.11$	$p = 0.741$	$\chi^2 = 0.09$	$p = 0.770$	$\chi^2 = 0.36$	$p = 0.562$	$\chi^2 = 3.31$	$p = 0.069$

HCV: hepatitis C virus; HBV: hepatitis B virus; HIV: human immunodeficiency virus; TTIs: transfusion-transmissible infections.

Pearson Chi-square test of association.

** $P < 0.05$.

Pos: positive; Neg: negative.

Table 5: Predictors of transfusion transmissible infections among voluntary blood donors.

Variables	Syphilis		HCV		HBV		HIV	
	OR (95% CI)	P-value						
Age group (years)								
17–39	0.49 (0.23, 0.07)	0.073	0.92 (0.51, 1.64)	0.774	1.08 (0.76, 1.52)	0.675	0.86 (0.53, .418)	0.563
≥40	1		1		1		1	
Gender								
Female	0.96 (0.37, 2.59)	0.939	0.29 (0.10, 0.79)	0.016*	0.71 (0.48, 1.67)	0.097	0.38 (0.18, 0.83)	0.015*
Male	1		1		1		1	

OR: odds ratio; 95% CI: 95% confidence interval; HCV: hepatitis C virus; HBV: hepatitis B virus; HIV: human immunodeficiency virus.

* $P < 0.05$.

age group is the most accessible during blood drives, and they belong to vibrant social groups/organizations that are favorably disposed to voluntary blood donation, such as the Red Cross and JCI. Similarly, most of the donors are males. [3,20,24–26] This finding is also consistent with previous studies done in Nigeria and other African countries. The low proportion of female donors could be because of the myth that women are not supposed to donate blood, and because of other physiological demands on them, such as menstruation, pregnancy, and lactation.

Transfusion Transmissible Infections

The prevalence of TTIs in VNRD investigated in this study revealed 3.7%, 1.2%, 1.6%, and 0.5% for HBV, HCV, HIV, and syphilis, respectively. In a study among VNRD in Tanzania, Mremi et al. reported a prevalence rate of HBV (4.8%), HCV (1.1%), HIV (1.8%), and syphilis (1.7%). [27] Among donors generally, prevalence rates are documented to be higher. Ogbenna et al. in Lagos reported rates of 5.79%, 2.23%, 1.34%, and 0.88% for HBV, HCV, HIV, and syphilis, respectively. [5] In other regions of Nigeria, HBV, HCV, HIV, and syphilis range between 10.5% to 18.6%, 2.4% to 6.0%, 2.0% to 3.1%, and 1.1% to 2.7%, respectively, as reported in Osogbo and Calabar. [18,28] The donors in these studies

were not only VNRD but also included other types of blood donors. Although the prevalence rates of TTIs vary from one region to another, all the studies show a pattern that HBV is the most common and syphilis is the least. We opine that these differences in prevalent rates may be due to the fact that these studies were conducted in different regions of Nigeria and used different sample sizes and methodologies. Furthermore, VNRD have lower prevalence rates than family replacement and paid donors, which may be due to their awareness of the risks associated with TTIs, their willingness to disclose their medical history during screening, and their willingness or resolve to continue to donate blood make them to live a healthy lifestyle.

We also observed that male gender is significantly associated with positivity for HCV and HIV infection. Several studies have shown that there is a higher prevalence of TTIs among male blood donors in Nigeria. A study conducted in Nigeria showed that the donor pool was male-dominated (98.7%) and had a higher prevalence (4.2%) of TTIs than female donors (0.0%). Another study found that the overall prevalence of blood transfusion-transmitted infections was higher among male blood donors (4.63%) than among female blood donors. Similarly, a study conducted on voluntary blood donors in Nigeria found that almost one-third of all male and 29.3%

of all female donors were positive for at least one of the four blood-borne pathogens. [29–36] The reason for the higher prevalence of TTIs among male donors could be due to several factors, such as lifestyle, occupation, and exposure to risk factors associated with TTIs. [35,37]

ABO and Rh Blood Groups

In this investigation, blood group O was the most common ABO phenotype (55.1%), followed by A (21.2%), B (20.0%), and AB (3.9%). Previously in Nigeria, Damulak et al. [38] and Aliyu et al. [39] reported a slightly lower O (49.0% and 44.1%) and A (19.7), higher B (26.6%) and AB (4.7% and 5.9%), while Enosolease and Bazuaye [40] reported similar frequencies with our findings. The prevalence of the O blood group in Africa ranges from 41.3% in Ethiopia to 52.0 % in Uganda. [12,41,42] RhD positive is the most encountered phenotype in the index study (93.19%). This is slightly lower than the previously reported prevalence of 97.7% by Etura et al. [43] but still follows the usual pattern. The study prevalence of Rh negative (6.9%) is like previous studies in Nigeria by Adeyemo et al., which reported 6.0%. [44] This data is in tandem with the already confirmed genetic variation in the inheritance of ABO and Rh blood antigens across races and geographical entities.

Hemoglobin Phenotypes

The prevalence of HbA, HbAS (sickle cell trait), and AC in the index study as 72.4%, 21.7%, and 5.9%, respectively. Ajayi et al. in 2022 [45] reported a similar HbA pattern of 72.4%, AS 22.6%, and AC of 1%. Sickle cell trait prevalence is less than the values previously reported in Benin, 23.19%, and Lagos, 23.3%. but similar to Akinboro et al., who reported a prevalence of 21.59% in 2012. [46,47] This general pattern of hemoglobin phenotype suggests a stable equilibrium of the sickle cell gene in Nigeria, and it has consequences for the control policy of sickle cell disease. Efforts to entrench genetic counselling and neonatal screening programs as components of the standard of care must be intensified to reduce morbidity and mortality associated with Sickle Cell Disease (SCD) in Nigeria.

One limitation of this study is the use of only cellulose acetate alkaline electrophoresis to determine the donor's hemoglobin phenotype, as this method might have missed some hemoglobin variants that co-migrate in this medium.

CONCLUSIONS

This study revealed that HBV seroprevalence is the highest compared with HCV, HIV, and Syphilis among VNRD and that the TTI seroprevalence is higher among male donors. Therefore, efforts to improve blood safety should include bringing more women into the donor pool and reducing the prevalence of TTIs by increasing immunization coverage for HBV, as well as screening of blood donors at all blood transfusion service centers.

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AUTHORS' CONTRIBUTION

Each author has made a substantial contribution to the present work in one or more areas, including conception, study design, conduct, data collection, analysis, and interpretation. All authors have given final approval of the version to be published, agreed on the journal to which the article has been submitted, and agree to be accountable for all aspects of the work.

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CONFLICT OF INTEREST

None.

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