Seroprevalence of HIV and frequencies of haemoglobin genotypes, ABO and Rh blood groups, among premarital couples in Port Harcourt, Nigeria

Jeremiah ZA, PhD, MSc, AMLSCN
Department of Medical Laboratory Sciences, Rivers State University of Science and Technology, Port Harcourt, Nigeria
Okon IA, MSc, AMLSCN
Department of Haematology & Blood Transfusion, University of Port Harcourt Teaching Hospital, Port Harcourt, Nigeria
Jeremiah TA, MSc, AMLSCN
Department of Medical Laboratory Sciences, Rivers State University of Science and Technology, Port Harcourt, Nigeria

Correspondence to: Dr ZA Jeremiah, E-mail: zacjerry@yahoo.com

Abstract

Background
Premarital screening is fast gaining ground as a prerequisite for the solemnisation of holy matrimony by many faith-based organisations in Nigeria, yet there is scanty literature on the subject.

Methods
Fifty (50) premarital heterosexual couples (50 males and 50 females) were screened for the presence of human immunodeficiency virus (HIV), haemoglobin genotypes, and ABO and Rh blood groups using standard laboratory procedures.

Results
The prevalence of HIV in this study was found to be 2.0%. The frequencies of the haemoglobin genotypes were as follows: HbAA (72%), HbAS (26%) and HbAC (2.0%), while that of ABO and Rh blood groups were: group A (22%), B (14%), AB (0%), 0 (64%), Rh 'D' positive (96%) and Rh 'D' negative (4.0%). The distribution of these variables between the sexes was statistically significant (χ^2 = 21.630, p< 0.01). The frequencies of the haemoglobin genotype combinations of the intending couples were as follows: HbAA/AA (52%), AA/AS (36%), AA/AC (4.0%) and AS/AS (8.0%). This distribution pattern was also found to be highly statistically significant (χ^2 = 38.957, p < 0.0001). The mean age of the participants was 35.48 ± 12.26 years for men and 26.32 ± 7.31 years for women (t = 10.538, p < 0.05).

Conclusion
This study observed a low prevalence of HIV among the premarital couples and a relatively high frequency of HbAS/AS couples. The ABO and Rh blood groups were found to be stable and consistent with previous reports. For the prevention of hereditary blood diseases, haemolytic disease of the newborn and transmission of HIV to offspring, premarital testing should be encouraged

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Introduction

A premarital test is defined as a test in which intending couples are tested for genetic, infectious and blood-transmitted diseases to prevent any risk of transmitting the disease to their children. It provides the baseline assessment of prospective marriage couples with the aim of reducing the unproductive genetic risk and also reduces the incidence of babies born with common haemoglobinopathies and infectious diseases.[1]

In 1994, the World Health Organization (WHO), through its control programme of haemoglobin disorders, recommended that public information and education, population screening for carriers, definition of local mutations, treatment and genetic counselling and prenatal diagnosis must be confirmed together for preventing hereditary blood diseases.[2]

Acquired Immune Deficiency Syndrome (AIDS) was discovered in Nigeria in 1986. With over 3 million people already affected with the virus,[3] coupled with several other factors such as hereditary diseases, especially sickle cell disease (SCD), public awareness about the dangers of raising children with SCD, mandatory premarital counselling by many faith-based organisations and the dire need to curb the HIV/AIDS pandemic in Africa, there has been a high demand for premarital screening as a prerequisite for marriage by faith-based organisations.

Although premarital screening by faith-based organisations has been going on for about five years, few published reports on the process are to be found in southern Nigeria,[4,5] and the few publications that are available centre mainly on HIV and premarital couples, with little or no regard for other genetic and infectious diseases. In this report, we present the prevalence of human immunodeficiency virus (HIV), haemoglobin genotypes and ABO and Rh blood groups among intending couples in Port Harcourt, Nigeria on the basis of the faith-based referral approach.

Materials and Methods
Subjects

The study population consisted of one hundred (100) premarital men and women (i.e. 50 couples) who attended the voluntary premarital counselling of some faith-based organisations and who were referred to the medical laboratory for premarital testing. Institutional ethical approval was received prior to the commencement of the study. All the subjects were offered confidential pre-test HIV and genetic counselling by the marriage committee, headed by a medical officer, and thereafter informed consent was obtained for blood sample collection. The researchers returned the results of the tests directly to the marriage committee. The marriage committee then disclosed the results of the tests to the intending couples during their counselling sessions. The stages involved in the counselling process fall beyond the scope of this study. All the participants were apparently healthy individuals with no clinical evidence of any disease. The mean age of the males was 35.48 ± 12.26 years, while that of the females was 26.32 ± 7.31 years.

Two millilitres of venous blood were collected into potassium ethylenediaminetetracetic acid (EDTA) salt and used for blood grouping and genotype, while the remaining clotted sample (serum) in the syringe was used for determining the presence of human immunodeficiency virus (HIV).

Methods

The antibodies to human immunodeficiency virus (HIV) in the serum of the intending couples were tested using a double ELISA test: determine ™ HIV 1 & 2 (Abbott, London) and Immuno comb II HIV 1 & 2 Bispot (Orgenics, Israel). The tests were performed in accordance with the manufacturers' standard operating procedures. HIV seropositivity was defined as a reactive result on both ELISA tests, as recommended by WHO/UNAIDS for asymptomatic patients.[6] The positive samples tested for HIV-1 only.

Haemoglobin electrophoresis was performed on each sample with cellulose acetate paper at pH 8.9. The procedures followed were those described by Brown.[7] The ABO and Rh blood groups of the intending couples were determined by the haemagglutination technique (tube method) with anti-A, anti-B, anti-A,B and anti-D monoclonal anti-bodies (Biotec, Suffolk, UK). Red cells were phenotyped according to standard procedures as described by Judd[8] and Brecher.[9] All laboratory tests were performed at the University of Port Harcourt Teaching Hospital Haematology Laboratory.

Statistics

The data were analysed using the Statistical Package for Social Sciences (SPSS) for Windows (version 9.0). The frequency distributions of the ABO, Rh and haemoglobin genotypes and HIV prevalence were analysed using the chi-square (χ^2) test, while the mean age of the study participants was computed using the one-tailed t-test. The statistically significant level was set at alpha 0.05 (p < 0.05).

Results

Premarital screening tests were performed on 50 couples (50 males and 50 females) as directed by the marriage committee of the referral churches and with the advice of a medical officer. The frequencies of the variables are shown in Table I. The following results were obtained: group A (22%), B (14%), AB (0%), HbAA (72%), HbAS (26%) and HbAC (2.0%). The prevalence of HIV in this study was found to be 2.0%. The distribution of the variables between the sexes was subjected to a chi-square (χ^2) test and found to be significant (χ^2 = 21.630, p< 0.01).

Table I: Seroprevalence of HIV and frequencies of the ABO and Rh blood groups, haemoglobin genotypes and HIV among premarital couples in Port Harcourt

	ABO				Rh 'D'		Genotype			HIV	
	A	B	AB	0	Rh Pos	Rh Neg	AA	AS	AC	Pos	Neg.
Sex	%	%	%	%	%	%	%	%	%	%	%
Males	6	8	0	36	50	0	32	16	2	2	48
Females	16	6	0	28	46	4	40	10	0	0	50
Total	22	14	0	64	96	4	72	26	2	2	98
Chi-Square (χ^2) = 21.630*											

*   = P < 0.01 (significant)
%   = Per cent

Table II: Distribution of the genotype combinations among the intending couples

Genotype combination	Frequency (%)
AA/AA couples	52.0
AA/AS couples	36.0
AA/AC couples	4.0
AS/AS couples	8.0
Chi-square (χ^2) test = 38.957*	

* = p < 0.0001 (significant)

Table III: Mean age of study participants

t-test	Males n = 50	Females n = 50
Mean (years)	35.48	26.32
SD	12.26	7.31
t-stat = 10.5378*		
t-critical (one tail) = 1.7108		

• = P ≤ 0.05 (significant)
n = number of samples

Table IV: Sex differences in the observed and expected counts of the haemoglobin genotypes of the study participants

Hb genotypes	Males		Females	
	Ob	Ex	Ob	Ex
HbAA	32.0	41.2	40.0	30.8
HbAS	16.0	14.8	10.0	11.2
HbAC	2.0	1.2	0	0.8
Chi-square (χ^2) test = 21.630*				

* = p < 0.01 (significant)
OB = Observed count
EX = Expected count

Table II shows the distribution of the genotype combinations among the intending couples. The following results were obtained: AA/AA (52%), AA/AS (36%), AA/AC (4.0%) and AS/AS (8.0%). This distribution pattern was also found to be highly statistically significant (χ^2 = 38.957, p < 0.0001). The mean age of the premarital couples is shown in Table III. The mean age of the male participants was 35.48 ± 7.31 years. This difference was found to be statistically significant when subjected to a t-test (t = 10.538, p < 0.05). The difference between the observed and expected values of the haemoglobin genotypes among premarital male and female participants was subjected to chi-square (χ^2) analysis, as shown in Table IV. There was a statistically significant difference in values in relation to the sex of the participants (χ^2 = 21.630, p < 0.31).

Discussion
Since the first case of AIDS was reported in Nigeria in 1986, over 3 million people have been infected with the virus.[3] The following trend in the prevalence of HIV in Nigeria has been reported: 1.8% in 1991, 3.8% in 1993, 4.5% in 1996, 5.4% in 1999, 5.8% in 2001 and 5.0% in 2003.[3] Similarly, an HIV prevalence of 6.6% was reported among antenatal women in Rivers State.[3] However, few reports on premarital HIV testing demanded by faith-based organisations could be found in Rivers State, Nigeria. The earliest report was by Akani *et al.*[5] In their report, an alarming rate of 27.4% HIV prevalence was recorded among intending couples from faith-based organisations. Contrastingly, an HIV prevalence of 2.0% was recorded among premarital couples in this study, and this 2.0% prevalence was only among males. The high prevalence recorded by Akani *et al.* is comparable only to reports from some parts of South Africa, where greater than 30% was observed among pregnant women. A similarly high prevalence of 25.6% was recorded for 621 South Malawian pregnant women.[10] A study by Ejele *et al.* recorded a prevalence of as low as 3.6% among unemployed females in Port Harcourt.[11] The present study is in agreement with a study by Peterson and White, in which the HIV prevalence among female and male premarital couples in the United States of America was found to be 0.0 to 0.4% and 0.0 to 1.1% respectively.[12] This study also confirms the report by UNAIDS that HIV infection is much more common among men (estimated at 60% versus 20% in women).[13]

The rationale for the premarital screening policy is that, if one or both partners test HIV positive, they will take this into account when considering whether to have children or not. However, the results were neither intended to discourage the couples from getting married nor to impose any decision on them regarding whether they should marry or not. The aim rather was to provide adequate knowledge about HIV and genetic disorders to intending couples to enable them to live productive and happy family lives. The decision to proceed to the final stage of marriage after the counselling sessions is entirely the responsibility of the intending couples. During the period of the study, the church never interfered in their decisions, thus the fundamental human rights of the individual couples were never violated. When compared with the HIV prevalence of 6.1% among premarital couples in the South-east of Nigeria, the result of this study appears very low.[4] It is possible that the moral values inculcated in the believers helped in bringing down the prevalence of HIV in church members.

Sickle cell anaemia is a common phenomenon in Africa. Earlier reports on haemoglobinopathies in Nigeria centred mostly on pregnant women, children and the general population.[14,15,16] There is a paucity of information on the haemoglobinopathies among premarital couples. In this study, the overall frequencies of the haemoglobin genotypes were as follows: HbAA (72%), HbAS (26%) and HbAC (2.0%). This report was similar to

earlier reports by Nwafor and Banigo, in which HbAA accounted for 73% while HbAS and HbSS accounted for 0.56%.[14] These values, except those for HbSS, were found to be consistent with the values obtained in this study.

Sickle cell disease (SCD) has remained an unresolved health problem in Nigeria, as in most of sub-Saharan Africa. However, Nigeria appears to be in a special position as it probably has the largest sickle cell gene pool in the world, with more than 85 000 HbSS births per year.[17] Despite the large and increasing burden of the disease, little has been done to reduce it, perhaps because of a lack of political will, or a lack of sufficient funds due to the deteriorating economy. In spite of this, the number of HbSS patients who survive to adulthood in Nigeria is increasing, especially among the elites, who have access to good health care. This is not surprising, as research has shown that socioeconomic status has a modifying influence on morbidity in sickle cell disease.[18] With more HbSS patients surviving into adulthood, the chances of transferring the gene to their offspring through marriage become greater. This is even more reason why premarital screening for haemoglobinopathies should be encouraged in order to offer the intending couples the opportunities to know their haemoglobin genotype status and also to weigh the disadvantages before trying the nuptial knot.

An analysis of the haemoglobin genotype combination among intending couples revealed a highly statistically significant pattern (χ^2 = 38.957, p < 0.0001). Fifty-two per cent (52%) of the couples were AA/AA, 36% were AA/AS, while AA/AC and AS/AS accounted for 4.0% and 8.0% respectively. For a single AS/AS combination, there is a one in four chance of having a sickler. This implies that 8.0% of the couples in this study population will produce about four homozygous HbSS children (ratio of 1:25). If this ratio is compared with the general population, then the frequency of Hb AS/AS couples obtained in this study could be regarded as high. When subjected to chi-square analysis, the observed and expected count of the inherited genes between sexes showed a statistically significant pattern (χ^2 = 21.630, p < 0.01). This implies that, in future, there will be a modulation of genes, with a tendency for HbAS to increase in the female population. However, gene frequencies were not calculated using the Hardy-Weinberg equation, so it is difficult to state categorically how the shift in the Hardy-Weinberg equilibrium will affect the gene flow in the population. A detailed study with a larger sample size will be needed to offer explanations for this observation.

The distribution of the ABO and Rh blood groups among the premarital couples in this study was as follows: group A (22%), group B (14%), group AB (0%) and Group O (64%). Rh 'D' positive accounted for 96%, while 4% of the study population was Rh 'D' negative. These results are in agreement with previous reports in the literature.[16, 19] The clinical significance of testing for blood groups is maternal alloimmunisation, which occurs when a woman's immune system is sensitised to foreign erythrocyte surface antigens, stimulating the production of antibodies, the most common route being blood transfusion, foetomaternal haemorrhage associated with delivery, trauma, spontaneous or induced abortion, ectopic pregnancy or invasive obstetrical procedures. In the event of alloimmunisation during pregnancy, these antibodies can cross the placenta and, if the foetus is positive for erythrocyte surface antigens, lead to haemolysis of foetal erythrocytes and anaemia, a condition known as haemolytic disease of the newborn (HDN).[20] HDN can also be caused by maternal anti-A, anti-B, anti-A,B IgG isoagglutinins, which are mostly present in type O mothers. Although a type O mother of a type A or B infant occurs in 15% of pregnancies, the frequency of HDN due to ABO isoagglutinins is only one in 150 births.[20]

In conclusion, the prevention of hereditary blood diseases is possible by preventing the birth of children that carry these illnesses. For this purpose, premarital haemoglobin electrophoresis use should be widespread and compulsory. Also, the spread of the overwhelming AIDS pandemic in sub-Saharan Africa can be reduced by a collective effort of individuals and organisations, including churches. The overall effect will be an improvement in the quality of family life.

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Figure 1: Possible offspring from an AS/AS marriage
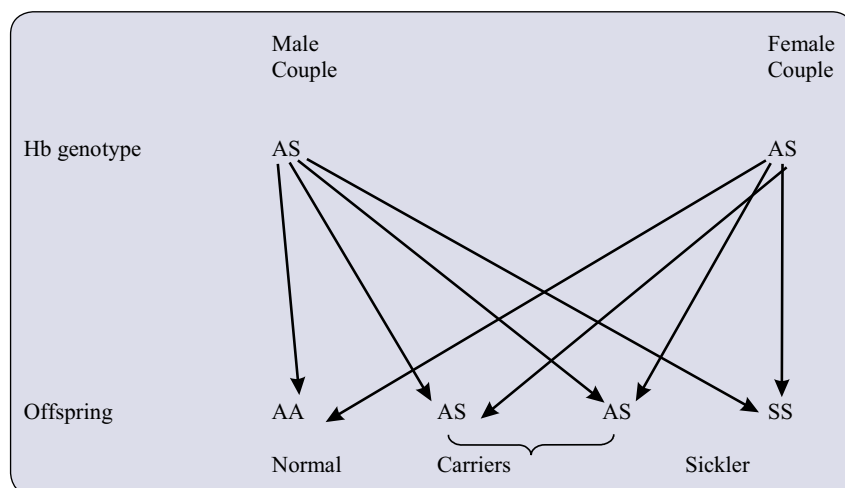
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