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HMTLV-1 among Pregnant Women and Blood Donors in a Resource Poor Setting, Southeast, Nigeria

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ABSTRACT

Aim: *Blood donation and transfusion is very common in Obstetrics and Gynecology practice with large volumes transfused daily. Many diseases can be transmitted through blood transfusion and vertically from mother to her unborn child including Human T-cell lymphoma/Leukemia virus type 1(HMTLV-1). This study was done in Abakaliki, Ebonyi State, Southeast Nigeria where screening for this virus is not done routinely. To determine the cost-effectiveness of routine screening of pregnant women and blood donors for the virus.*

Methods: *This was a prospective, cross-sectional study. Two hundred and fifety participants were recruited 150 donors and 100 pregnant. Social demographic characteristics and history of blood donation were noted. Data were collated, tabulated and analyzed using simple percentages and tables.*

Results: *The 250 participants recruited had zero prevalence of HMTLV-1 antibody. Among the 150 donors, 10 were females and 93.3% had donated while only 4% had received blood in the past. Among the pregnant women 10% had received while 6% had donated. The mean age was 28.2 ± 9.1 (between 18 – 55 years). The mean parity for the female participants was 2.8 ± 1.2 (p1 – 6).*

Conclusion: *The zero prevalence in this study may make its inclusion in routine screening non-cost effective.*

Keywords

HMTLV-1, Pregnancy, Blood Donors, Transfusion, Transmission.

Introduction

Human T-cell lymphoma/Leukemia virus type 1(HTLV-1) is one of the re-emerging blood-borne infections that is transmitted through blood transfusion and vertically from mother to the child. The first identified retrovirus, a member of the delta-retroviruses like HTLV-II, Bovine Leukemia Virus (BLV) and Simian T-cell Leukemia virus (STLV) [1-3]. An oncogenic human RNA virus with heterogenous and restricted distribution with high endemicity associated with certain geographic areas and ethnicity [1,4,5].

Global affectation is over 20 million and the highly endemic areas include the Caribbean basin, South America, tropical Africa, Iran and Melanesia [1,3,5-7]. Studies, have reported high prevalence of HTLV-1 infection Sub-Saharan Africa especially in Guinea, Nigeria, Cameroon, Democratic republic of Congo and Ivory Coast with seroprevalence of the antibodies ranging from 0.5 to 33% in the general population [1,5,6].

Transmission is via infected lymphocytes [8,9] sexually and vertically (transplacental and breast feeding), parenterally (via blood transfusion, needle sharing and accidental pricks). Some countries have incorporated routine screening of donors for HTLV-1 antibodies in their blood transfusion safety protocols as a result of the mode of transmission [3,10,11]. In Nigeria and most parts of Africa, routine screening and diagnosis of HTLV-1 infection

among blood donors is not practiced [5]. Studies have shown 13 to 75% risk of transmission in recipients of HTLV-1 infected cellular blood products [12,13].

In Nigeria, 7%, 3.7%, and 0.7% and 0 prevalence were reported among blood donors in Ibadan, Zaria, Lagos and Enugu respectively [14-17]. In Ghana, a seroprevalence of 2.1% HTLV-1 antibodies among pregnant women was reported [18]. Studies have shown that the seroprevalence rate in pregnant women can be 5 to 10 folds higher than in blood donors, however HTLV-1 infection does not appear to be associated with congenital malformations but mother-to-child transmission constitutes over 90% of the cause of HTLV-1 seropositivity in children with breast feeding accounting for most cases of vertical transmission of HTLV-1 infection [6,19,20].

HTLV-1 has been implicated in the pathogenesis of lymphoproliferative malignancy of CD4+ cells known as Adult-T-Cell Lymphoma (ATL) and a chronic neurological disease, Tropical Spastic Paralysis/ HTLV-1 Associated Myelopathy (TSP/ HAM, HTLV-1 infection has also been associated with a number of inflammatory diseases such as pediatric infectious dermatitis, uveitis and some cases of myositis [20-23]. Infection with HTLV-1 confers a lifelong risk of 2–4% for ATL and 0.2 to 5% for HAM. The incubation period for HAM, ATL and other HTLV-1 associated diseases may be several decades [8,24-26]. The majority of infected persons are asymptomatic. Considering the importance of blood transfusion in clinical practice, the need for exclusive breast feeding in our environment and the rising incidence/awareness of malignant diseases, this study is undertaken to shed more light on the magnitude of this re-emerging transmissible viral infection by determining the seroprevalence of anti- HTLV-1 antibodies in blood donors and pregnant women in Ebonyi State, Southeast, Nigeria. The study also determined whether the prevalence is high enough for it to be included in routine screening for blood donors and screening tests for pregnant women in our setting as it is not done presently.

Materials and Methods

This is a cross-sectional prospective study that was carried out at the Alex Ekwueme Federal University Teaching Hospital, Abakaliki, Ebonyi State, South-eastern Nigeria. The study lasted for a period of 3 months from September 14th to December 13th 2023. The qualified blood donors as well as pregnant women at various stages of gestation were consecutively recruited from the blood bank unit and the antenatal clinics respectively till the sample size is obtained.

The minimum sample size for this study was obtained using the formula for cross-sectional study proposed by Lwanga et al. [27];
$N = z^2p(1-p)/d^2$ where
N = Sample size
Z = A number relating to the confidence interval = 1.96
P = prevalence from a similar study in the same geographic region Donors [16] (7%) and pregnant women [18] (2.1%)
d = standard error margin tolerable = 0.05
For blood donors $N = \frac{1.96^2 (0.07) (1 - 0.07)}{0.05^2} = 100$
For pregnant women $N = \frac{1.96^2 (0.021) (1- 0.02)}{0.05^2} = 32$
Anticipated response rate due to attrition = 90%
Therefore, total N = 132/0.9 = 147
N for Donors will be 111participants
While pregnant women will be 36 participants

Participants who signed informed consent form were consecutively recruited into the study. Demographic data and history of blood donation/transfusion were obtained from each participant using a questionnaire. Inclusion criteria were those that signed the informed consent form, blood donors that satisfy all the requirements for blood donation, screened negative for HBsAg, HCV, VDRL, RVST and other infections that may contradict blood donation. The woman must be pregnant, booked and attending antenatal clinic at hospital and also screened negative for the above infections. The exclusion Criteria were those who fail to sign consent, unbooked pregnant women, or pregnancy complicated by other problems and diseases, or those who fail to meet the criteria for blood donation.

Samples Collection

Five milliliters (5mls) of blood were collected from the peripheral vein of each participant ensuring sterile procedure. The blood was transferred into a sterile plain bottle and allowed to clot. The serum was then separated into another sterile plain bottle and stored immediately at − 20°C. Sample collection lasted about 3 months. The entire specimen were screened for anti-HTLV-1 antibody using a commercially available ELISA kit. based on one-step incubation, double antigen sandwich principle. The sample that tested positive to HTLV-1antibodies was then subjected to Western blotting to confirm that indeed it is HTLV-1infection.

HTLV- 1 Antibody Screening

The enzyme-linked immunosorbent assay (ELISA) was used to screen all the sera from the participants. The ELISA kit uses strips pre- coated with recombinant HTLV antigen polystyrene microwell. Participant's serum sample was incubated in the microwells together with a second recombinant HTLV-1 antigen conjugated to horseradish peroxidase (HRP-Conjugated). If anti- HTLV antibody is present in the serum, the pre-coated and HRP-conjugated antigens will be bound to the two variable domains of the antibody during incubation. The specific antigen-anti-body immunocomplex formed, is captured on the solid phase of the microwell. After the serum and the unbound HRP-Conjugate were washed away and chromogen solutions containing tetramethylbenzidine (TMB) and urea peroxide was added to each of the wells. In the presence of antigen-antibody-antigen (HRP) sandwich complex, the colorless chromogens will be hydrolyzed to a blue colored product. The blue color turns to yellow after stopping the reaction with sulfuric acid. Wells containing samples negative for anti-HTLV antibody will remain colorless. For each test quality control was ensured by using the criteria for reactive and non-reactive results as clearly stated in the manufacturers' quality control ranges for the validation of results before the sample is interpreted. Reactive samples was then subjected to qualitative real time polymerase chain reaction (PCR) targeting the HTLV-1/2 polygene for confirmation [28].

Data Management/Analysis

The data collected was fed into the computer using the Statistical Package for Social Science (SPSS) version 22 (SPSS Inc. Chicago, IL, USA) and analysis done. Descriptive statistics was used for quantitative variables while the results were presented in simple percentages and tables, means and standard deviation.

Ethical Issue

Ethical approval was gotten from the research and ethics committee of the hospital. Informed consent was obtained from the clients before recruiting them into the study and was given the option to opt out at any time if they do not want to continue. Confidentiality was also ensured as the data collected was used only for the purpose of the study. The cost was borne by the researcher.

Results

A total of 250 participants who signed consent were recruited into the study. One hundred and fifty blood donors and 100 pregnant women. Among the 150 blood donors only 10 were females. The mean age was 28.2 ± 9.1 and ranged between 18 years – 55 years. The mean parity for the female participants was 2.8 ± 1.2 and ranged between 1 – 6. Majority of the participants were Christians (98%), married (90%) civil servants (60%) with tertiary education (80%). Table 1.

Table 1: Characteristics of respondents.

Variables	Participants (n=250)	Percentage (%)
Age (years)		
<20	20	8
20-29	100	40
30-39	110	44
40-49	15	6
≥50	5	2
Mean ±SD (Range)	28.2 ± 9.1 (18–55)	
Gender		
Male	140	56
Female	110	44
Parity (n=110)		
1	25	23
2-4	75	68
≥5	10	9
Median ±SD (Range)	2.0 ± 2 (0–7)	
Marital status		
Unmarried	25	10
Married	225	90
Occupation		
Civil servants	150	60
Students	60	24
Traders	25	10
Farmer	10	4
House wives	5	2
Religion		
Christian	245	98
Others	5	2
Level of Education		
Primary education	12	4.8
Secondary education	38	15.2
Tertiary education	200	80
Blood donors (n=150)		
Male	140	93.3
Female	10	6.7
Pregnant women	100	40

Table 2: History of Blood Donation and Transfusion.

Characteristics	Number	%
Donated Blood in the Past (Donors 150)		
Yes	140	93.3
No	10	6.7
(Pregnant women 100)		
Yes	6	6
No	94	94
Total	**250**	**100**
Received blood in the Past (Donors 150)		
Yes	6	4.0
No	144	96.0
(Pregnant women 100)		
Yes	10	10
No	90	90
Total	**250**	**100**

Among the blood donors, 93.3% had donated blood in the past and 96% of them had not received blood transfusion in the past. On the other hand, only 6% of the pregnant women had donated blood in the past but 10% of them had received blood transfusion in the past. Table 2.

Seroprevalence of HTLV-1

The seroprevalence of HTLV-1 in this study was zero percent (0) for both the blood donors and the pregnant women.

Discussion

Human T-cell lymphoma/Leukemia virus type 1(HTLV-1) is one of the blood-borne re-emerging infections that is transmitted through blood transfusion and vertically from mother to the child or sexually. It has been implicated in the pathogenesis of lymphoproliferative malignancy of CD4+ cells known as Adult-T-Cell Lymphoma (ATL) and a chronic neurological disease, Tropical Spastic Paralysis/ HTLV-1 Associated Myelopathy (TSP/HAM). HTLV-1 infection has also been associated with a number of inflammatory diseases such as pediatric infectious dermatitis, uveitis and some cases of myositis [20-23]. The difficulty encountered in its diagnosis and lack of definitive treatment has made prevention the best approach to its management.

In this study, there is zero prevalence of the antibodies in the 150 and 100 blood donors and pregnant women respectively. This is similar to the study in Enugu, Southeast Nigeria that reported zero prevalence in blood donors but 0.5% in pregnant women. This may be as a result of the application of similar methodology in the studies as well as similar geographical location. This is however different from the studies in Ibadan, Zaria and Lagos respectively that reported 7%, 3.7%, and 0.7% prevalence among blood donors in [14-17]. This may be as a result of different methodologies

and non-application of PCR for specificity. One study in Nigeria reported high seroprevalence of 16.7% among pregnant women [5]. This also may be due to the reason above. Other African countries like Ghana, Zimbabwe, Mozambique, Congo, Namibia and Senegal reported seroprevalence of 0% to 2.0% HTLV-1 antibodies among pregnant women [18,29-34]. These reports are in contrast to the studies that reported high endemicity in Africa [6,20]. Thus, this infection may not be endemic in Africa, especially Nigeria.

Conclusion

The zero-prevalence reported in this study makes the inclusion of HTMLV-1 in routine antenatal and blood screening tests non-cost effective.

Limitations

This is a hospital-based study with small sample size, hence may not be a true representation of the population. A multi-center or general population study may be required to have a true picture the endemicity of the infection.

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