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Sero-Prevalence of Hepatitis and HIV Co-Infection among Women of Reproductive Age in Port Harcourt, Nigeria

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Abstract

Background: The study is aimed at determining the prevalence rate of HIV/ Hepatitis co-infection amongst women of reproductive age.

Materials and Methods: A total of 204 blood samples were collected from pregnant women, HIV-positive patients, outpatients, and healthy donors.

Results: This study investigated the prevalence of hepatitis and HIV among 204 patients, revealing a significant prevalence of 26.1% for hepatitis B (HBV), with no cases of hepatitis A or C reported. Socio-demographic analysis indicated a higher HIV prevalence among married women (59.3%) compared to single women (40.7%), with the majority of cases found in individuals with secondary education (75.9%). The co-infection ratio for HIV and HBV was 26.1/4.8%. Occupational analysis showed a lower prevalence among public servants (5.5%) compared to self-employed (6.5%) and unemployed individuals (6.3%). A notable 92.6% of subjects reported having 1-5 lifetime sex partners. Previous testing rates were high for HIV (87.7%) and moderate for HBV (21.6%), with significant p-values indicating prior awareness of these diseases, contrasting with other sub-Saharan regions' awareness levels.

Conclusion: The study's findings underscore a substantial prevalence of HBV and HIV, particularly among married women and those with secondary education, highlighting the need for targeted public health interventions. The high rate of prior testing suggests a baseline awareness in the population, which can be leveraged to enhance prevention and control measures. These results contribute valuable insights into the epidemiology of HBV and HIV co-infection, paving the way for further research and policy development in sub-Saharan Africa.

INTRODUCTION

Human immunodeficiency virus (HIV) and hepatitis virus cause significant morbidity and mortality across the World especially hepatitis B virus (HBV) [1]. Human immunodeficiency virus and hepatitis B virus are blood-borne viruses transmitted mainly through sexual contact and use of unsterilized needles. Their similar means of transmission increases the risk of contracting both infections concurrently [2].

Viral hepatitis is an acute or chronic inflammation of the liver caused by viral infection [3]. HBV is globally the leading cause of death due to liver disease in people living with HIV/AIDS [4]. In 2016, the World Health Organization (WHO) estimated that 36.9 million people are living with HIV [5]. It was also reported that 248 million people have chronic HBV infection [5]. It is estimated that chronic HBV infection affects an estimated 5–20% of people living with HIV [6].

Human Immunodeficiency Virus (HIV) positive persons who become infected with HBV or hepatitis C virus (HCV) are at increased risk for developing chronic hepatitis. In addition, persons who are co-infected with HIV and hepatitis can have serious medical complications, including an increased risk for liver-related morbidity and mortality, chronic hepatitis, cirrhosis, and hepatocellular carcinoma, all of which are of serious public health concern. [4]

Human Immunodeficiency Virus (HIV) and Hepatitis B Virus (HBV) are both endemic in Nigeria. There is a heavy burden of HIV – HBV and HIV – HCV co-infections in many regions of the developing world [7], including Nigeria [8]. The burden of these co-infections is greatest in the African and South-East Asian regions [7]. HIV is associated with a higher prevalence of both HBV and HCV in Sub-Saharan Africa. In this region, many people, especially women living with HIV, are co-infected with HBV or HCV [9].

In Nigeria, hepatitis co-infection with HIV is associated with increased morbidity and mortality [10]. Nigeria has the second-largest HIV prevalence in the world and one of the highest rates of new infection in Sub-Saharan Africa [3]. Unprotected heterosexual sex accounts for 80% of new HIV infections in Nigeria, with the majority of remaining HIV infections occurring in key affected populations such as sex workers [3]. Rivers State is among the tops in the prevalence rate chart, with many of its residents living with the virus, especially women of reproductive age [11].

The notable relationship between HIV and HBV is that they can be transmitted in the same way. Several studies strongly suggest that the influence of HIV on HBV is characterized by a chronic infection, an increased viral replication rate, higher viral levels, accelerated liver damage and an increased risk of liver cancer [12].

On the other hand, the HBV infection aggravates the progression of HIV towards AIDS and an increased in vitro replication of HIV [13]. Mortality among HIV/HBV co-infected persons is substantially higher than among HIV mono-infected persons [14]. A study in the USA revealed that HIV/HBV co-infection increased mortality by 8-fold or 19-fold when compared with single infection with HIV or HBV alone, respectively [14].

The exact impact of HBV on HIV disease progression is less clear [15], although it is postulated that HBV infection may potentially lead to a blunted immune response in patients receiving antiretroviral therapy (ART) and increase patient susceptibility to ART-related liver toxicity [8]. Unfortunately, access to anti-viral medication is limited in resource-poor countries and only 41% of people living with HIV have access to ART in sub-Saharan Africa [16]. The objective of this study was to determine the prevalence of hepatitis B virus (HBV) and HIV Co-infection among women of Reproductive age in Rivers State, Nigeria.

MATERIALS AND METHODS

Study Design

This project was an experimental work at the Medical Microbiology Laboratory Department of Rivers State University Teaching Hospital.

Study Area

Rivers State University Teaching Hospital, formerly known as Braithwaite Memorial Specialist Hospital (abbreviated as BMSH), is a government-owned hospital named after Eldred Curwen Braithwaite, a British doctor and a surgery pioneer. It is located at 6-8 Harley Street, Old GRA, Rivers State, a neighborhood of Port Harcourt, and is operated by Rivers State Hospital Management Board. It lies between latitude and longitude 4.7802° N, 7.0142° E. It was established in March 1925 as Braithwaite Memorial Hospital and was originally a medical facility for senior civil servants. It later became a General Hospital and has since become a "Specialist Health Institution". In 2018, it was renamed to serve as a Teaching Hospital for the state-owned university following the establishment of the College of Medical Sciences. Officially recognized by the Federal Ministry of Health, Braithwaite Memorial Specialist Hospital is ranked among the largest hospitals in the Niger Delta. The facility has 375 licensed beds and 731 medical staff members. Its departments include Medicine, Paediatrics,

Laboratories, Radiology, Family Medicine, Obstetrics & Gynaecology, Anaesthesia, Surgery, Pathology, Ophthalmology, Accident Centre and the Surgical/Medical Emergency. Some other departments are Pharmacy, Finance, Maintenance, General Administration.

Study Population

The study population will comprise of women of reproductive age from four different groups

I. Pregnant women
II. HIV patients
III. Outpatients
IV. Healthy donors

Specimen Collection and Processing

Five millilitres of blood will be aseptically collected from each female by venipuncture of the cubital vein using sterile disposable vacutainer blood collection needles and bottles. Samples will be placed in EDTA bottles, and the sera will be separated into sample vials and stored at -20°C until analyzed. Serum samples will be analyzed for HIV and Hepatitis Virus infection using a rapid lateral flow chromatographic immunoassay kit by Abbot and Tell, respectively. The test will be carried out and interpreted according to the manufacturer's instructions.

To obtain information, a pretested structured questionnaire will be given to all participating females of reproductive age. The questionnaire includes close-ended and open-ended questions and will be categorized into socio-demographic characteristics, risk factors, and vaccination history. The laboratory test results for participants will be anonymously linked to their questionnaire information through unique identifiers.

Serological analysis
Rapid Assay for Hepatitis B Surface Antigen (HBsAg)

The assay was carried out using DiaSpotHBsAg rapid test strip (DiaSpot Diagnostics, USA) and interpreted according to the manufacturer's specifications. The test strip is a rapid, one-step test for qualitatively detecting HBsAg in serum or plasma. The test strip uses the immuno-chromatographic method to detect the presence or absence of HBsAg in serum or plasma. All test strips, serum or plasma specimens, and controls were allowed to equilibrate to room temperature (15-30°C) before testing. The assay was performed within 1 hour to obtain the best results according to the manufacturer's specifications. The result was read at 15 minutes, and no result was interpreted after 30 minutes because a low HBsAg concentration might result in a weak line appearing in the test region (T) after an extended period of time. The interpretation of test results was performed according to the manufacturer's specifications.

Procedures:

Specimens and test kits were brought to room temperature before testing. The foil wrap pouch was opened, and the cassette was removed. The cassette was kept on a flat, clean surface, and the test kit was immediately after opening. Using a dropper provided, 3 drops of the serum or plasma were transferred to the cassette, making sure no air bubble was trapped. The results were read within 15 minutes.

ELISA for Detection of Hepatitis B Surface Antigen (HBsAg)

Serum samples were analyzed for hepatitis B surface antigen (HBsAg) using the ELISA kit (DIA.PRO Diagnostic Bioprobes, Italy). The tests were performed according to the manufacturer's instructions. The test results were calculated using a cut-off value determined on the mean OD450nm value of the negative control (NC) with the following formula: NC + 0.050 = Cut-Off (Co). Test results are interpreted as the ratio of the sample OD450nm (S) and the Cut-Off value (Co), mathematically S/Co, according to the following: < 0.9 = negative, 0.9 – 1.1 = equivocal, and > 1.1 = positive. A negative result indicated that the patient was not infected by HBV. A positive result was indicative of HBV infection, and therefore, the patients should be treated accordingly.

Procedures:

The sample was added to a well containing an anti-HBsperoxidase solution. It was then Incubated at 37°C. Thereafter, the plate was washed. Diluted TMB was added as a substrate. It was then Incubated at room temperature. A stop solution (sulphuric acid) was then added, and the results were read using an ELISA reader or spectrophotometer, as shown below.

CD4 T cell count enumeration

EDTA-treated blood samples were used for CD4 T cell count using PartecCyFlow® Counter (Partec GmbH, Germany), following the manufacturer's instructions. The specimens were analyzed on a flow cytometer to detect cell surface markers for CD4 cells. Results were classified based on the CDC (1997) guidelines.

Procedures:

The test was performed within 48 hours. The samples were placed on a gentle blood rocker for 5 minutes to ensure that the samples were uniformly distributed. The blood was pipetted in a uniform volume accurately and reproducibly. All tubes were incubated in the dark during the staining procedure. A lyse/no-wash method was carefully followed as directed by the manufacturer, which is required for SPT. Immediately after the specimens were processed, the tubes were capped, and all stained samples were stored in the dark, under refrigeration (39°--50°F [4°--10°C]) until flow cytometric analysis for 24 hours

HIV-1 Viral Load Testing (Abbott Real-Time Assay)

To assess the viral load (VL) of HIV-1-positive individuals used in this study, the Abbott Real Time HIV-1 (m2000sp) assay was used to determine the viral load according to the manufacturer's instructions. The results were presented in copies/ mL of plasma.

Data Analysis

The data generated from this study will be represented in percentages and mean±SD where necessary. Where appropriate, Statistical Package for Social Sciences (SPSS) Version 23 statistical analyses. Statistical significance will be defined as a p-value of less than 0.05 at a 95% confidence interval.

RESULTS

The prevalence of HIV infection in women of reproduction is 26%; 3(1.47) of pregnant women were positive, 30 (24.5%) of HIV subjects were positive, 1(0.4%) of outpatient subjects were positive, 0(0%) of healthy donors were positive. For Hepatitis, 3(1.47%) pregnant women were positive, 4(1.9%), HIV patient were positive, 3(1.47%) of outpatient were positive.

The prevalence of HIV and Hepatitis Co-infection among the various study populations in RSUTH, Port Harcourt, Rivers State, was studied to ascertain the Sero-prevalence of HIV and Hepatitis Virus in the hospital. The results obtained showed that out of 204 sample population screened, 54(26.5%) subjects indicated were positive for HIV, 150 (73.5%) were negative for HIV, 10 (4.9%) were positive for Hepatitis B Virus, 204 (100%) for both Hepatitis B and C virus were negative as seen in Table 1.

Table 3 shows the socio-demographic specific prevalence of HIV infection: in terms of spread according to marital status, 32(59.3%) were for married women while single women were 22 (40.7%); educational status shows those in secondary education as 41 (75.9%) while those in Tertiary were 13 (24.10%). In occupation, those in public service were 3 (5.5%), self-employed 32 (6.5%) while unemployed were 17 (6.3%)

Socio-Demographic Specific Prevalence of HBV Infection

Table 4 shows the socio-demographic prevalence of HBV infection. Those married had a prevalence of 5(5%), and those singles had a prevalence rate of 5(5%). In terms of education, those in secondary school were 7(7%), while those in tertiary institutions recorded 3(3%). Regarding occupation, those in public service (public servant) had none

while the self-employed recorded 5(5%).

Socio-Demographic Specific Prevalence of HBV Infection

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Table 5 shows the percentage distribution of the prevalence in ratio to their P-values and Chi-square values. From the result, about 179 (87.7%) of the subjects had a previous HIV test, 25 (12.3%) has not been tested given a P-value of <0.001 and Chi square value of 8.412. For Hepatitis, 44 (21.6%) has a test before on the infection and 160 (78.4%) has nor, given a P-value of 0.002 and Chi-square of 10.222. From the study, 2(1%) have a previous transfusion history, and 202 (99%) have not been transfused before the Chi-square value of 4.381. 27 (13.2%) has a previous surgery, and 177 (86.8%) have no surgical history, P-value <0.001, and Chi-square 23.123. about 28 (13.7%) have multiple sexual parties compared to 176 (86.8%) who do not. Persons who have had sex with non-marital partners, 87 (42.6%) compared to 117 (57.4%) have P-values 0.009 and 0.002 respectively and Chi-square 2.314 and 30.124 respectively. Only 9 (4.4%) have been infected with sexually transmitted disease, 195(95.6%) have not been, this gave a P-value of 0.001 and Chi-square of 12.415.

Table 1 Overall Prevalence Rate of HIV/Hepatitis Co-Infection Sample Collection

	Total No.	HIV Positive	HBV Positive	
				Negative
Pregnant Women	51	3(1.47%)	3(1.47%)	45(22%)
HIV patients	50	50(24.5%)	4(1.9%)	0(0%)
Out Patients	50	1 (0.4%)	3(1.47%)	46(22.5%)
Healthy Donors	53	0(0%)	0(0%)	53(26%)
	204(100%)	54(26%)	10(4.8%)	144

No. of Co-Infection = 10
Overall Prevalence is (26.1%)
Hepatitis (4.8%)

Table 2: Demographic Data showing positive and negative results in percentages

	Total No.	No. of Positive %	No. of Negative %
HIV Status	204	54 (26.5)	150 (73.5)
HAV	204	0 (0)	204 (100)
HBV	204	10 (4.9)	194 (95.1)
HCV	204	64 (31.3)	204 (100)

Table 3: Socio-Demographic Specific Prevalence of HIV Infection

Variables	No. of Positive %	No. of Negative %
Marital status		
Married	32 (59.3)	68 (45.3)
Single	22 (40.7)	82 (54.7)
Educational status		
Secondary	41 (75.9)	30 (20)
Tertiary	13 (24.1)	120 (80)
Occupation		
Public servant	3 (5.5)	13 (8.7)
Self Employed	32 (6.3)	52 (34.7)
Unemployed	17 (31.5)	85 (56.6)
No. Of life time Sex partners		
1-5	50 (92.6)	134 (89.3)
6-10	4 (7.4)	13 (8.7)
11-20	0 (0)	1 (0.7)

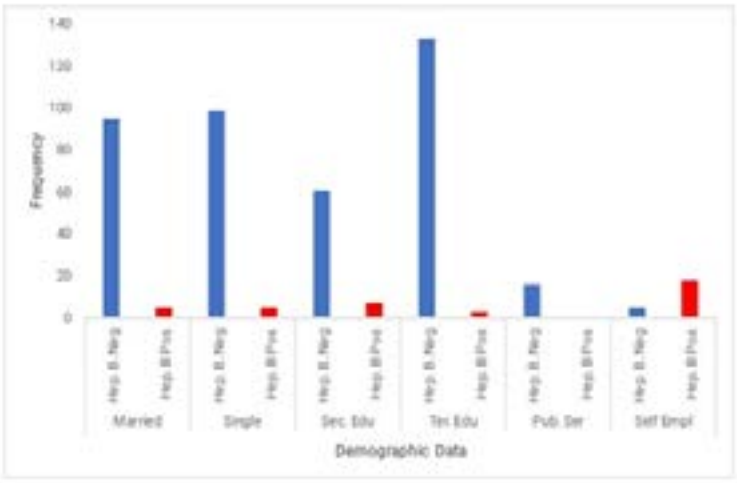

Figure 1: Demographic Data of Subjects

Table 4.5: Demographic data of responses from the Questionnaire

	Yes	No	P-Value	Chi Square X^2 Value
Have you done HIV Test before?	179(87.7)	25(12.3)	<0.001	8.412
Have you done HBV Test?	44(21.6)	160(78.4)	0.002	10.222
Have you done Blood Transfusion?	2(1)	202(99)	<0.001	4.381
History of Surgery?	27(13.2)	177(86.8)	<0.001	23.123
Share Sharp Objects with others?	51(25)	153(75)	<0.001	9.113
Multiple Sexual Partners?	28(13.7)	176(86.8)	0.009	2.314
Sex with non-marital partners?	87(42.6)	117(57.4)	0.002	30.124
History of Sexual Transmitted Infection?	9(4.4)	195(95.6)	0.001	12.415

DISCUSSION

The results obtained showed overall prevalence of HIV and Hepatitis Co-infection in women of reproductive age to be 26%. Positive cases for pregnant women 3(1.47%), 30(24.5%) of HIV subjects were positive, 1(0.4%) of Out-patients subject and no positive result for healthy donors to HIV. Pregnant women showed 3(1.47%) positive result to Hepatitis, in HIV patients 4(1.9%) were positive and 3(1.47%) of Out-patient were positive. From the result, pregnant women has less prevalence value than the average woman of reproductive age because it is believed that those pregnant women might be married and as this sticks to the husbands (one life sex partner)

Socio-Demographic specific prevalence results of HIV infection were also captured having variables indicating prevalence. In terms of spread according to marital status, 32(59.3%) were for married while single women were 22(40.7%). Educational status shows those in secondary education as 41(75.9%) while those in Tertiary were 13(24.1%). In occupation, those in public service were 3(5.5%), self-employed 32(6.5%) while those unemployed were 17(6.3%). In terms of the number of life time sex partners, sex partners between 1-5 were 50(92.6%), sex partners between 6-10 were 4(7.4%).

Whereas, HBV infection prevalence from the socio-demographic analysis showed married women to have a prevalence of 5(50%) and those single, prevalence of 5(50%). In terms of education those in secondary school to be 7(70%) while those in Tertiary institution recorded 3(30%). In terms of occupation, the self-employed recorded 5(5%), while those in public service no positive result.

The Socio-demographic results also shows that occupation as well as educational level is critical in these examinations. This is true because according to the study, those in public service had no positive results may be due to the fact, only very few of those in public have time for sexual activities. So we see 5% value for the self-employed as those self-employed can have much leisure for sexual activities compared to those employed or in public service.

From the result, 179 (87.7%) of the subjects had at one time or the other been tested for HIV and 44 (21.6%) for Hepatitis B Virus with p-values of < 0.001 and < 0.002 respectively with chi-square of 8.412 and 10.222 respectively. This is an indication that there has been previous knowledge about the disease which is different from other sub-Saharan countries with little awareness.

Only about 2 (1%) has been transfused, 27 (13.2%) having previous surgery, 28 (13.7%) has had sex with multiple partners, 87 (42.6%) having sex with non-marital partners and 9 (4.4%) having history of sexually transmitted disease, having p-values/chi-square values of < 0.001/ 4.381, <0.001/ 23.123, < 0.009/ 2.314, < 0.002/ 30.124, and < 0.001/ 12.415 respectively.

Prevalence of the co-infection from transfusion is almost insignificant while history of surgery has some significant value of prevalence. It is imperative to say that majority of the test subjects who tested positive contracted the through sexual interactions with multiple and non-marital partners thus, giving the very significant values.

The results showed a prevalence of HBV of almost 5% (4.9%) which is comparable to previous studies among different populations in the Sub-African region. In Togo, this prevalence was 9.7% [4] and 10.6% among women of childbearing age in 2017. Across the region, prevalence of HBV was approximately similar in Côte d'Ivoire, Burkina Faso and Mali with a prevalence of 8.5% among HIV [17].

A study in South Africa, found a prevalence

of 4% in 2017 and another one in Kenya a prevalence of 10.1% in 2020 [18]. Those results demonstrate that HBV remains endemic in sub-Saharan Africa, despite the framework for action for viral hepatitis in the African region aiming for the elimination of viral hepatitis as a major public health threat by 2030.

The American College of Physicians also advises as a primary best practice for HBV prevention the vaccination against HBV in all unvaccinated adults at risk of infection (due to sexual, percutaneous, or mucosal exposure; health care and public safety workers at risk of blood exposure; adults with chronic liver diseases, end-stage renal infection, HIV infection). In this sense, additional efforts and strategies should be geared towards adults' screening and vaccinations, including high risk individuals such as Female Sex Workers.

CONCLUSION

Our study showed that more than a quarter of female sex workers never used condoms and were approximately three times more likely to be infected with HBV. A similar result was found in a study in Thailand among migrant sex workers. This subgroup of female sex workers who do not systematically use condoms with multiple sex partners is where immunization strategies should be focused on. Outreach programs and awareness campaigns specifically designed for female sex workers on viral hepatitis could potentially have an impact both on condom use and HBV screening and immunization uptake.

To our knowledge, no studies have explored the reasons for this correlation. However, hypotheses include genetic or cultural factors. Further studies should specifically explore this aspect of the viral hepatitis pandemic in other regions to identify specific ways to prevent transmission.

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