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Prevalence and Viral Load Determination of Hepatitis B Virus among Hiv Seropositive Patients Attending Kogi State Specialist Hospital Lokoja, Kogi State

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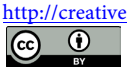
Abstract

Human Immunodeficiency Virus (HIV) and Hepatitis B Virus (HBV) share common risk factors and HBV occurs in people with HIV resulting in an increased risk for HIV/HBV co-infection. Globally, hepatitis B virus infection is of serious public health causing morbidity and mortality. The increasing incidence of liver diseases caused by HBV is emerging as a significant cause of morbidity and mortality among HIV-infected individuals. A clearer knowledge of HBV prevalence in Kogi State is important in order to educate, inform the population and control epidemics through extensive vaccination and treatment programme. The aim of this study was to determine the seroprevalence of Hepatitis B infection and to evaluate molecularly HBV infection among HIV seropositive individuals. Sera samples were obtained from 218 consented HIV participants and screened for HBsAg using the commercial membrane based rapid qualitative test kit and real-time PCR was performed using Tianlong to assay the virus quantitatively. A structured questionnaire was used to collect information on patient's demographic variables and risk factors for



HBV transmission. Overall, 17 of the participants were seropositive to HBsAg. There was a significant difference between the age distribution with (*P*-value = 0.006) and marital status with (*P*-value = 0.044). Type of marriage, occupation, place of residence and risk factors associated with HIV and HBV co-infection do not show significant differences. A total of 17 HBsAg positive samples were subjected to viral load analysis, out of which 7 were highly unsuppressed, 5 were suppressed while the remaining 5 were undetectable. This study confirmed a moderately high HIV/HBV co-infection rate (7.8%). The highly unsuppressed viral load obtained from the study is a potential risk for Hepatocellular carcinoma among the study population. Enlightenment programme on routes of virus acquisition with a view to reducing the morbidity and mortality of HIV/HBV co-infection should be intensified.

Keywords

Real-Time PCR, Co-Infection, Morbidity, Mortality, CD4 Receptor, Monocytes, Macrophages, Dendritic, Viral Hepatitis

1. Introduction

Despite major advances in treatment over recent decades, the human immunodeficiency virus (HIV) remains a significant and complex global health issue [1]. Use of the term AIDS (acquired immune deficiency syndrome) is now rare, but it was previously used to describe the advanced stage of HIV infection, in which the immune system has declined to such a level that infections and cancers can emerge and proliferate. This phase of the infection is now more commonly called advanced or late-stage HIV infection [1]. HIV infects immune system cells which have a CD4 receptor on the surface. These cells include T-lymphocytes (also known as T cells), monocytes, macrophages and dendritic cells [2]. HIV recognizes host cells by binding to the CD4 cell membrane receptor. Infection may be latent or chronic low level. Activation leads to the development of immune dysfunction as a result of direct and indirect killing of T4 cells and functional impairment of viable T4 cells. The infection is found primarily in homosexual and bisexual men, intravenous drug users, haemophiliacs, transfusion recipients, sexual partners of infected persons, and infants born to infected mothers. Transmission occurs through sexual contact, exposure to contaminated blood or blood products, and perinatally. Acquired immune deficiency syndrome (AIDS) is a late manifestation of infection with HIV. AIDS is characterized by a marked depletion of T4 cells. It has been established that following reduction in the CD4 positive cells count to lower than 200 cells/mL, the immune system of HIV positive individuals fails to develop an adequate response against microbial agents, as a result, re-activation of HBV infection and its related complications will occur [3]. The progressive immune deficiency is accompanied by a wide range of life-threatening opportunistic infections and neoplasms.

One important opportunistic infection associated with immune deficiency is

hepatitis B infection [4]. It has been documented that one of the frequent complications of HIV is hepatitis B co-infection and due to the common methods of transmission of these two viruses, the incidence rate of co-infection is increasing [5] [6]. Hepatitis B infection is a disease of the liver caused by the viral pathogen Hepatitis B. Viral hepatitis has emerged as a major public health problem throughout the world affecting several hundreds of millions of people. Viral hepatitis is a cause of considerable morbidity and mortality in the human population, both from acute and chronic infections. Hepatocellular carcinoma which is one of the ten most common cancers worldwide is closely associated with hepatitis B, and at least in some regions of the world with hepatitis C virus [7] [8].

Hepatitis B virus is endemic in the human population and hyper-endemic in many parts of the world. Although various body fluids (blood, saliva, menstrual and vaginal discharges, serous exudates, seminal fluid, and breast milk) have been implicated in the spread of infection, infectivity appears to be especially related to blood [9] [10]. The epidemiologic propensities of this infection are therefore wide. They include infection by inadequately sterilized syringes and instruments, transmission by unscreened blood transfusion and blood products, by close contact, and by sexual contact. Antenatal (rarely) and perinatal (frequently) transmission of hepatitis B infection from mother to child may take place. HIV positive individuals constantly stand the chance of opportunistic infections if prompt and effective treatment is not administered. More so, individuals who are not faithful to medications may gradually have their immune system depleted [11]. Hepatitis B infection which affects the liver also poses serious health challenge to individuals. This is because the target site of the viral pathogen is an indispensable organ for humans. The liver serves too many functions that anyone would dare to compromise [12]. This study was carried out to determine the seroprevalence of Hepatitis B infection and to evaluate molecularly HBV infection among HIV seropositive individuals. It also assessed the risk factors likely associated with Hepatitis B virus infection among HIV positive individuals.

2. Methods

Study Location: This study was carried out among HIV seropositive individuals aged 15 years and above attending Kogi State Specialist Hospital (KSSH) in Lokoja, Nigeria who consented to the research. Lokoja is a city in Nigeria. It is situated at the confluence of the Niger and Benue Rivers. Lokoja, the capital of Kogi State is located on latitude 7˚45'N - 7˚51'N and longitude 6˚45'E and lies at an attitude of 45 to 125 meters above sea level. Lokoja lies about 7.8023˚ North of the equator and 6.7333˚E east of the meridian. It is about 165 km Southwest of Abuja as the crow flies and 390 km of Lagos by same measure. Lokoja has a total area of 1230 m^2 (3180 km^2). The hospital provides diagnostic and treatment services for substantial number of individuals within and beyond the State. The ethical clearance was obtained from the ethical review board of Kogi State Specialist Hospital.

Two hundred and eighteen HIV seropositive individuals who attended Kogi

State Specialist Hospital (KSSH) between August and November, 2022 were included as subjects. Each individual's consent was granted before being enrolled into the study. After explaining the objective of the study, consent was obtained from each participant to collect blood samples and obtain data on socio-demographics.

Data Collection: All seropositive individuals re-diagnosed/confirmed for HIV infection participated in the questionnaire form and underwent diagnosis for HBsAg using standard strip method.

Sample Collection and Storage: Blood samples were collected aseptically from each of the individuals in the study using sterile needle and syringe. The blood sample was transferred into a sterile anti-coagulant bottle (EDTA) to avoid haemolysis. After some minutes, samples were centrifuged at 200 revolutions per minute (rpm) for 10 minutes to separate the serum from the whole blood. The serum and the test strips were stored at room temperature (15˚C - 30˚C) before being screened for HBsAg.

2.1. Procedure for HIV Testing

Determine (initial test) and Uni-gold test kits were used to test the HIV status of the participants using the procedure described by Arora and Arora (2011). Where positive result was obtained with determined test kit, a confirmatory test was carried out with Uni-gold test kit using the same procedure. A positive test with Uni-gold confirms the participant's HIV positive status.

2.2. Procedure for Viral Load Test

The hepatitis B viral load was determined using Tianlong Real-time PCR System (model: Gentier 48E. SN TL23EL20061990).

2.3. Statistical Analysis

The statistical analysis was performed using SPSS software (Version 21.0). The age is expressed in range. All other data are expressed in percentages. The Chi-square test was applied wherever necessary. Statistical significance was set at $P \leq 0.05$.

3. Results

A total of 218 seropositive individuals attending HIV Clinic at Kogi State Specialist Hospital, Lokoja participated in this research. The majority (68.3%) of the participants were females and 31.7% were males. While all participants tested positive to HIV rescreening, only 17 were co-infected for hepatitis B Surface Antigen (HBsAg) giving rise to co-infection rate of 7.8%.

3.1. Social Demographic Characteristics of Hepatitis B Co-Infection in HIV Seropositive Individuals

Table 1 shows the age distribution of the HIV/HBV co-infection among individuals attending Kogi State Specialist Hospital, Lokoja, Kogi State. The age range 16 - 25 (33.3%) had the highest co-infection rate followed by 46 - 55 (11.1%)

Table 1. Age distribution of HIV/HBV co-infected individuals attending Kogi State Specialist Hospital Lokoja, Kogi State.

Age	Number of Respondents (n = 218) (%)	Number of Positive (%)	*P*-Value	Chi-square
16 - 25	24 (11.0)	8 (33.3)	0.0006	21.595
26 - 35	63 (28.9)	0 (0)		
36 - 45	89 (40.8)	6 (6.7)		
46 - 55	27 (12.4)	3 (11.1)		
56 - 65	9 (4.1)	0 (0)		
Above 65	6 (2.8)	0 (0)		

and 36 - 45 (6.7%), respectively. There is no co-infection in the age brackets 26 - 35, 56 - 65, and above 65. Statistical analysis shows that there is significant difference between the age distribution ($P = 0.0006$).

Table 2 shows other social demographic characteristics as regards to marital status, type of marriage, occupation and place of residence of the individuals attending Kogi State Specialist Hospital Lokoja, Kogi State.

HIV/HBV co-infection rate is higher among the single (26.67%) followed by the divorced (9.52%) and married (7.38%) while none of the widows had HBV infection. The statistics show that there was significant difference in the HIV/HBV co-infection in the marital class ($P = 0.044$).

There was no significant difference in the statistical analysis in the kind of marriage of the patients ($P = 0.385$).

The highest incidence rate of HIV/HBV co-infection is found among the civil servants and traders compared to farmers, students and others (Table 2). There was no significant difference in the occupational status of the respondents ($P = 0.165$).

Table 2 also shows the place of residence of the respondents (urban versus rural). The result indicates a higher HIV/HBV co-infection among urban dwellers. There was no significant difference as regards the place of residence ($P = 0.321$).

3.2. Risk Factors Associated with HIV/HBV Co-Infection

Table 3 shows the risk factors associated with HIV/HBV co-infected participants attending Kogi State Specialist Hospital, Lokoja, Kogi State.

History of surgery is shown in Table 3. Lower population of the sample (42) had been exposed to surgery while 176 have never had surgery. Among those exposed to surgery, HIV/HBV co-infection was found among 21.43%. There was no significant difference in the HIV/HBV positive rates among those exposed to surgery or not ($P = 0.377$). History of tribal marks and tattoos is also shown in Table 3. A greater percentage of the respondents do not have tribal marks or tattoos on their bodies. Co-infection rate among the respondents with tribal

Table 2. Type of marriage, occupation specific, residence specific distribution of HIV/HBV co-infected participants attending Kogi State Specialist Hospital Lokoja, Kogi State.

Marriage	Number of respondents (n = 218) (%)	Number of co-infected Respondents (n = 17) (%)	*P*-value	Chi-Square	T-test	Degree of Freedom
Marital Status						
1Single	15 (6.88)	4 (26.67)	0.0443	8.084		3
Married	149 (68.35)	11 (7.38)				
Divorced	21 (9.63)	2 (9.52)				
Widow	33 (15.14)	0 (0)				
Type of Marriage						
Monogamy	183 (83.94)	13 (7.10)	0.3852		1.446	1
Polygamy	35 (16.06	4 (11.43)				
Occupation						
Civil Servant	59 (27.06)	8 (13.56)	0.1650	6.497		4
Trader	105 (48.17)	9 (8.57)				
Farmer	6 (2.75)	0 (0)				
Student	15 (6.88)	0 (0)				
Others	33 (15.14)	0 (0)				
Place of Residence						
Urban	171 (78.44)	15 (8.77)	0.3212	1.811		1
Rural	47 (21.56)	2 (4.26)				

Table 3. Risk factors associated with HBV co-infection in HIV seropositive participants attending Kogi State Specialist Hospital, Lokoja, Kogi State.

	Number of respondents (n = 218) (%)	Number of co-infection (n = 17) (%)	*P*-Value	Chi-Square	T-test	Degree of Freedom
History of Surgery						
Yes	42 (19.27)	9 (21.43)	0.3765		1.489	1
No	176 (80.73)	8 (4.55)				
Tribal Marks/Tattoos						
Yes	77 (35.32)	5 (6.49)	0.1759	3.526		1
No	141 (64.68)	12 (8.51)				
History of Blood Transfusion						
Yes	36 (16.51)	6 (16.67)	0.3894	1.426		1
No	182 (83.49)	11 (6.04)				
History of Dental Treatment						
Yes	49 (22.48)	7 (14.29)	0.3356	1.718		1
No	169 (77.52)	10 (5.92)				

Continued

Intravenous Substance Abuse					
Yes	63 (28.90)	15 (23.81)	0.3065	1.914	1
No	155 (71.10)	2 (1.29)			
History of Previous Sexually Transmitted Diseases					
No	18 (8.26)	0 (0)	0.4376	1.218	1
Yes	200 (91.74)	17 (8.5)			
Multiple Sexual Partners					
None	49 (22.48)	4 (8.16)	0.3803	3.075	3
1	136 (62.39)	13 (9.56)			
More than 1	9 (4.13)	0 (0)			
Rather not say	24 (11.00)	0 (0)			
Awareness of Sexual Partner Being Exposed to HBV					
No	187 (85.78)	13 (6.95)	0.4020	1.367	1
I don't Know	31 (14.22)	4 (12.90)			

marks or tattoos is 6.49%. There was no significant difference ($P = 0.176$).

Table 3 also shows the history of blood transfusion. 36 (16.51%) of the respondents have had history of blood transfusion while 182 (83.49%) have not been exposed. Co-infection rate among the respondents was 16.67%. There was no significant difference of those who had had blood transfusion or not ($P = 0.3894$, t = 1.43).

A trend in blood transfusion was observed among those who had undergone dental treatment or intravenous substance abuse. Co-infection rate of HIV/HBV among intravenous substance users was 23.81%. There was no significant difference among those exposed to dental treatment as well as intravenous substance abuse at 0.05 probability limit ($P = 0.3356$ and 0.307, respectively).

The history of sexually transmitted diseases (STD) and number of sexual partners are shown in Table 3. A lower number of the respondents, 18 (8.26%) had no history of STD and there was no co-infection in this population. This is reflected in the statistically non-significant difference of this aspect of the study ($P = 0.438$). 136 (62.39%) of the population have only one sexual partner as shown in Table 3. This accounts for a higher population of respondents. Co-infection rate was the highest (9.56%) among this population (respondents with only one sexual partner). There was also no significant statistical difference among the participants having multiple sexual partners.

The knowledge of sexual partner having hepatitis B virus is also shown in Table 3. A higher number of respondents were not aware of their partner's status while very few (31) were not bothered by the HBV status of their sexual partner. The co-infection rate was highest among respondents who were not bothered by

HBV status of their partner. There was no significant difference in the HIV/HBV co-infection at 0.05 probability limit.

3.3. Viral Load of HBsAg in HIV/HBV Co-Infected Participants Attending Kogi State Specialist Hospital, Lokoja, Kogi State

Viral load of hepatitis B in HIV/HBV co-infected patients is shown in Table 4. All six males positive for co-infection were distributed in the ratio 1:1:1 at 33.3% into the viral load reading of >20%, 15% - 19%, and 0% - 14% while the females were distributed at 45.4% into the reading > 20% and 27.3% into 15% - 19% and 0% - 14% viral load reading frame each. Females (45.4%) were more highly suppressed than the males (33.3%) while the males (33.3%) were more moderately suppressed and unsuppressed than the females. The statistical analysis for the set of data showed that they were not significantly different.

4. Discussion

Globally, it is estimated that 5% - 10% of people living with HIV are co-infected with hepatitis (HBV), while HIV/HBV frequency in Sub-Saharan Africa vary from 0.0% to 28.4% [13]. HIV and HBV share common risk factors and many cases of HBV occur in people with HIV resulting in an increased risk for HIV/HBV co-infection [14].

Nigeria is also known to be highly endemic for hepatitis B viral infection [15]. In HBV endemic countries, of which Nigeria is one, the severity of HBV infection has been classified into low (<2%), moderate (2% - 8%) and high (>8%) prevalence by WHO (2018).

In this study, the overall seroprevalence of HBV among the 218 HIV positive participants was 7.8% suggesting a moderate prevalence of HIV/HBV co-infection among the respondents. The co-infection rate of 7.8% in this study is a clear indication that HBV is a threat to HIV individuals in Nigeria.

The prevalence of HIV/HBV co-infection from this study is slightly higher than the 6.3% obtained in a similar study by Okonko *et al.* (2021) [16] in Uyo, Akwa Ibom, Nigeria. However, it is higher than the 2.2% prevalence documented in South-Eastern part of Nigeria [17] but lower than 15.0% reported in North Central region of Nigeria [18].

This is comparable to previous rates in studies conducted in other parts of the

Table 4. Viral load of hepatitis B in HIV/HBV co-infected participants attending Kogi State Specialist Hospital, Lokoja, Kogi State.

Variable	No of Samples	>20%	15% - 19%	0% - 14%	*P*-value	Chi-square
			Gender (%)			
Male	6 (35)	2 (33.3%)	2 (33.3%)	2 (33.3%)	0.2355	0.8889
Female	11 (65)	5 (45.4%)	3 (27.3%)	3 (27.3%)		
Total	17	7(41.2%)	5(29.4%)	5(29.4%)		

Degree of freedom: 2. Key: >20%; highly suppressed, 15% - 19%; moderately suppressed, and 0% - 14%; unsuppressed.

world. Other researchers identified HBsAg positivity in individuals as 7.8% in France [19], 7.3% in India [20], 7.8 in Iran [21], 6.0% in South Africa and Greece [21] [22], and 9.4% in Germany [23]. Much lower prevalence rates were reported in Enugu as 0.24% [24], Anambra State as 0.7% [25], 0.78% in Cameroon [26] and 1.4% in the United States [27]. Very high prevalence rates have been documented in different parts of Nigeria and other parts of the world; 28.4% in Lagos [28], 28.7% in Jos [29], 30.4% in Ilorin [30], 70.5% in Kano [31], 45.0% in France [32], and 33.8% in India [33].

The differences in the prevalence rates can be attributed to differences in the number and behaviour of study group and knowledge of HBV infection and prevention.

The prevalence of HBV was highest (33.3%) among participants aged 16 - 25 and this is supported by previous studies in Uyo, Akwa Ibom Nigeria [15], Cameroon [26] [34], and Abuja, Nigeria [35]. This may be associated with higher social activities within this age group. This finding is in agreement with the finding of Olokaba *et al.*, (2011) [36] who reported that age group between 20 - 29 years has a greater prevalence rate. Katamba *et al.*, 2020 also reported that one of the correlates of HIV and Hepatitis B co-infection was age between 20 - 29 years [37] [38].

Younger age groups have always proved to be the most important factor, the age at which many infections occur, calls for concern and concerted effort aimed at implementing preventive measure that will reduce lifestyle practice among the most susceptible age bracket. Statistical analysis shows that there was significant difference between the age distribution ($P = 0.0006$).

Marital status specific prevalence of HBV was highest among HIV-infected individual who were single (26.6%) as compared to the divorced (8.5%), married (7.4%) and widow (0%). The statistical analysis shows there is significant difference in the HIV/HBV co-infection in the marital class ($P = 0.0443$). This may imply that the marital status is really a risk factor for HBV infection but an indicator to consider the sexual partner as a risk for infection since unmarried (single) people tend to have many sexual partner or unprotected sex. Also, positivity of HBsAg among the married and divorced subjects in this study could be an indication that the infection might be through unprotected heterosexual intercourse or close contact with their intended spouse as the virus can be spread through body fluids.

In relation to occupation, the civil servants had the highest prevalence (13.6%) followed by traders (8.6%) and those others with zero prevalence. This might be due to the high risk of social behaviour. However, the statistics showed that co-infection of HBV with HIV among the respondents is independent of occupation.

With respect to residence, co-infection rate was higher (8.77%) among urban residents as compared to 4.26% among rural dwellers. Though statistically not significant, the sampling method, selection criteria, study location and number of participants may have influenced this outcome.

The higher hepatitis B surface HIV/HBV co-infection rate in participants with history of surgical application is similar to previous reports in Anyigba (Omatola *et al.*, 2019), Lokoja (Omatola *et al.*, 2020) and Benue State (Mbaawuaga *et al.*, 2014).

A trend is observed in the predisposal of respondents to blood transfusion, dental treatment, and intravenous substance abuse. Prevalence of HIV/HBV co-infection was highest in participants predisposed to the measured parameters. Although none of the parameters defined showed statistical significance, it is a pointer to the fact that these parameters are important risk factors among the population studied. In contrast, the presence of tribal marks and tattoos as well as previous sexually transmitted diseases did not count for higher significance as compared to the population who do not have them.

A higher rate was found among those who have one sexual partner (9.56%) followed by those without sexual partner (8.16%). Individuals with more than more sexual partner and those who would not say recorded zero prevalence (0%) each. Poor knowledge of HBV infection and prevention among the study group could be the reason for prevalence distribution among the study groups.

It will be more appropriate to relate the findings of this study to the general population concerned. Considering that the patients attending the Antiretroviral Therapy (ART) clinic come from various places with different background and practices. The HIV/HBV co-infected individuals studied serve as representation partially of different age group and occupation. The results, therefore, emphasize the need for all recently diagnosed HIV cases to be screened for HBV as this will improve management of HIV/HBV co-infection and all ART individuals confirmed for HBV must be routinely accessed for viral load as this will help to avoid unnecessary therapies, commence treatment as appropriate and save cost.

5. Conclusion

The key highlights of this study are: HIV/HBV co-infection had a prevalence of 7.8% among HIV positive individuals, the prevalence of HBV was highest (33.3%) among participants aged 16 - 25, Marital status specific prevalence of HBV was highest among HIV-infected individual who were single (26.6%) as compared to the divorced (8.5%), married (7.4%) and widow (0%), In relation to occupation, the civil servants had the highest prevalence (13.6%) followed by the traders (8.6%) and those with the others with zero prevalence, With respect to residence, co-infection rate was higher (8.77%) among urban residents as compared to the 4.26% among rural dwellers, A trend is observed in the predisposal of respondents to blood transfusion, dental treatment, and intravenous substance abuse A higher rate was found among those who have one sexual partner (9.56%) followed by those without sexual partner (8.16%) attending Kogi State Specialist Hospital, Lokoja, Kogi State. The high prevalence of HIV/HBV viral load in individuals with no knowledge underscores the need for continuous public health education on ways of avoiding risky behaviours and unprotected sexual intercourse. There is a need to screen all HIV-positive individuals for HBV infection.

Public enlightenment programmes on routes of virus acquisition with a view to reducing the morbidity and mortality of HIV/HBV co-infection should be intensified.

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Authors' Contributions

ENO, OMO, OOE, CIA designed the studies. ENO, EMO, OOE, EMO, NTS, OZP, COA, EOE performed viral testing, and PCR. ENO, OMO, OOE, CIA wrote the manuscript with contributions from all authors. NTS, OZP, EOE and ENO revised the manuscript. All authors contributed to data analysis, drafting and critically revising the paper and agree to be accountable for all aspects of the work. All authors read and approved the final manuscript.

Availability of Data and Materials

All data generated or analyzed during this study are included here and are available from the corresponding author on reasonable request.

Consent to Participate

All authors declare that "written" informed consent was obtained from the participants with assurance of anonymity and confidentiality before the commencement of the study.

Ethical Approval

Ethical approval for the study was obtained from the Kogi State Specialist Hospital Research Ethics Committee with ethical approval registration number: KSSH/RECA/966/VOL.1/55.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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Abbreviations

HIV: Human Immunodeficiency; HBV: Virus Hepatitis B Virus; HBsAg: Hepatitis B Surface Antigen; CD4: Cluster of Differentiation; PCR: Polymerase Chain Reaction; AIDS: Acquired Immune Deficiency Syndrome; STD: Sexually Transmitted Diseases; ART: Anti-Retroviral Therapy; WHO: World Health Organization.