

PREVALENCE OF OCCULT HEPATITIS B VIRUS INFECTION AMONG HEALTHY HBsAg NEGATIVE BLOOD DONORS IN OWERRI, SOUTH-EAST NIGERIA.

ABSTRACT

Background: Hepatitis B virus (HBV) infection with its attendant complications is a disease of major public health importance being the 10th leading cause of death globally. Transfusion transmitted HBV continues to be a major problem despite the routine screening of donor blood products using the ELISA method. This study is aimed to determine the prevalence of Occult Hepatitis B virus infection among healthy HBsAg negative blood donors in Owerri, South-East Nigeria.

Methods: This was a cross-sectional descriptive study conducted in consecutive prospective blood donors in Owerri, South-East Nigeria. A total of 180 participants were enrolled for the study.

Results: The age of the subjects ranged from 21 to 45 years with a mean [SD] of 26.85±5.21 years. Male to female ratio was 4.6:1. Of the 180 participants, 33 (18.3%) had anti HBc IgG positive antibody. Out of the 33 anti HBcIgG positive participants, 5(15.2%) of them were anti-HBs positive while 13 (15.2%) of them had detectable HBV DNA. The prevalence of occult HBV infection was 7.2% (13/180).

Conclusions: Occult HBV infection is not uncommon among Nigerian blood donors. This study signifies the high prevalence of OBI and proposes that the blood samples in Nigeria should be pre-tested for OBI by Nucleic Acid Testing and or anti-HBV prior to transfusion to

minimize the HBV transmission risk. More awareness should be created on the need for immunization against HBV to further reduce the risk of transmission.

Keywords: Occult Hepatitis B virus infection, blood transfusion, HBV DNA load, Nigeria

INTRODUCTION

Hepatitis B virus (HBV) infection with its attendant complications is a disease of major public health importance being the 10th leading cause of death globally.¹ Hepatitis B and C viruses (HBV and HCV) are significant global health burdens, with millions of individuals affected worldwide. They are communicable diseases for which deaths are increasing. About 1.3 million people died of viral hepatitis in 2022.² Nigeria is among the countries with a high burden of viral hepatitis with prevalence of Hepatitis B Virus (HBV) and Hepatitis C Virus (HCV) as 11% and 2.2%, respectively (FMOH 2013) and Nigeria also accounts for 8.3% and 4.5% of the global burden of chronic HBV and HCV respectively.³ and approximately 15-40% of infected persons will develop cirrhosis, liver failure or hepatocellular carcinoma.^{4,5} About 350 million of the 2 billion people infected worldwide are chronic carriers of hepatitis B virus (HBV).⁶

An international workshop on occult hepatitis B virus infection (OBI) endorsed by the European Association for the Study of the Liver (EASL)⁷, and the Toarmina consensus conference defined OBI as the presence of HBV DNA in the liver of individuals testing HBsAg negative with currently available assays; and introduced a cut off value for serum HBV DNA less than 200 IU/ml.⁸

The serological markers of hepatitis B virus are HBsAg, anti HBs, anti HBc (IgM and IgG), HBeAg, anti-HBe, and HBVDNA. These are important as they can be used in the diagnosis, and determination of the severity of the infection.⁹

The first marker to appear in the blood following Hepatitis B Virus Infection is HBV DNA, followed by HBsAg, the DNA polymerase and HBeAg. Thereafter the antibodies to the antigens can be detected.¹⁰

Routine screening of blood for prospective donation is usually by detection of HBsAg using the Enzyme Linked Immunosorbent Assay [ELISA] method¹⁰. Other screening assays include the HBVDNA load. However, this is not routinely done due to the cost implication. Also, most centers do not have the necessary facilities for detection of HBVDNA load. Thus, this may expose blood recipients to HBV infection if the donor blood is HBsAg negative but has HBV DNA present. In the absence of other hepatitis B viral makers, anti HBe may be the only indication of an existing hepatitis B virus infection.¹¹

Patients may have HBV infection, and HBsAg will be absent in their blood but may have antibodies to hepatitis B core antigen¹². Such has been demonstrated in healthy blood donors in which their liver harbors the HBV genome.^{13, 14, 15}

Therefore, in order to achieve the WHO goal of eliminating viral hepatitis by 2030, there is need to comply with the who global report that advises taking action in viral hepatitis interventions that will enable countries to regain the trajectory to achieve the Sustainable Development Goals – saving lives; preventing a future generation of new infections, cancers and deaths; and reducing costs. One of these interventions is to ensure safe blood transfusion.

LITERATURE REVIEW

OBI are mainly found in older donors, nearly 100% carry anti HBe and approximately 50% also carry anti HBs.¹⁶ This suggests that OBIs occur largely in individuals having recovered from the infection but unable to develop a totally effective immune control.¹⁷ Co-infection

with Human immunodeficiency virus(HIV),drug abuse or immunosuppression can trigger an enhancement of HBV DNA levels without the presence of HBsAg^{18, 19, 20}. Transmission of HBV from individuals with occult HBV infection may occur via organ transplant and blood transfusion.²¹

It is presently unclear to what extent occult HBV infection represents a risk factor for the community other than for the infected individual.²²

The prevalence of anti HBc and HBV DNA alone is most reported within this definition of OBI, but various serological patterns including the presence of anti HBs can exist.²³

The prevalence of OBI is unclear and depends in part on the sensitivity of the HBsAg and DNA assays used as well as the prevalence of HBV infection in the study population.²⁴ OBI varies significantly between different geographical regions.²⁵ Studies have shown that the prevalence of OBI is closely related to the endemicity of HBV infection.^{26, 27} Patients from countries highly endemic for HBV are more likely to develop OBI.²⁸ Nna and his colleagues observed a prevalence of 8% among 100 repeat blood donors in Enugu South-East Nigeria with a median HBVDNA of 51IU/ml.²⁹ Another study by Oluyinka and his colleagues showed a prevalence of 17% among healthy blood donors in South-West Nigeria.³⁰ Japhet and his colleagues in Ile-Ife, Osun State recorded a prevalence of 13.0% of anti HBc positivity with 5.4% of the blood donors having anti HBc IgM alone.³¹ Salawu and his colleagues observed a prevalence of 4.37% of anti-HBc positivity in 457 blood donors in South-West Nigeria.³² Olotu and his colleagues observed a prevalence of 5.4% among 325 blood donors in South-West Nigeria.³³ Eightannan and his colleagues in Egypt noted a 9% prevalence of antiHBc among multiply transfused haemodialysis patients, 18.9% of them had both anti HBc and anti HBs. Hassan and her colleagues in Egypt observed that 22.5% of Hepatoma patients had serum occult HBV and 10% of anti HBc positive had OBI.³⁴ Also, Shambesh and his

colleagues observed a prevalence of 10.5% among blood donors in Western Libya.³⁵ Medawi and his colleagues and Yuman and his colleagues observed prevalence of 15% and 10% among HIV infected adults in Sudan and Cote d'Ivoire respectively.^{36, 37}

Manuchehi and his colleagues observed an OBI prevalence of 10% among blood donors in Iran. Fang and his colleagues observed a prevalence of 10.6% among 659 healthy individuals in China.³⁸

Strata and his colleagues in Italy had a 3.3% prevalence of occult HBV infection among patients waiting for kidney transplant being more frequent among Asians and Africans.³⁹

Toyoda and his colleagues showed an OBI prevalence of 51.2% among 44 patients with Hemophilia who have received multiple transfusions. Said and her colleagues in Egypt observed that 17.2% of the anti HBc [total] positive patients had detectable HBVDNA.⁴⁰

Tsui and her colleagues in USA noted that 2% of 400 HIV infected women with anti HBc positivity alone had occult HBV infection [ie detectable HBV DNA] with no correlation to Aminotransferase levels.⁴¹ Kew and his colleagues in South Africa observed a 48.4% prevalence in patients with PLCC with positive anti HBc.⁴² Selin and his colleagues noticed that OBI has also been associated with liver enzyme flares in HCV infection without changes in HCVRNA levels.⁴³ It was observed by Win and his colleagues that persistently infected patients with no clinical disease were found to have anti HBc with greater affinity compared with chronic liver disease group.³⁶ There are also strong suggestions of viral replication and gene expression in OBI patients.³⁷ Much evidence also suggests that it can favour progression to hepatic fibrosis and above all development of PLCC.²⁹ Several studies have approached the issue of HBV persistence differently by screening for HBV DNA in different populations of individuals especially blood donors positive for both anti HBc and anti HBs or anti HBc alone. In blood donors carrying anti HBc, only 0.3 -15% have detectable HBV DNA. The

variation in prevalence depends on the sensitivity of the Nucleic Acid Amplification Testing [NAT] assay, the genotype[s] prevalent in the population and the specificity of the anti HBc assay.³⁰

As in highly endemic countries, the majority of HBV infections are contracted perinatally or in early childhood; a higher proportion of the infected adults have late chronic HBV with undetectable HBsAg. This may account for the higher rate of OBI in anti-HBc-positive populations in these areas.³⁴ Prevalence may also vary depending on the nature of biological material tested, with a higher proportion for liver compared to serum specimens.²⁸

Occult HBV infection has been reported in 0.1%-2.4% of HBsAg-negative, anti-HBc-positive (\pm anti-HBs) blood donors in Western countries such as the United States, where only 5% of the population has prior exposure to HBV, and in up to 6% of a similar cohort of donors who reside in endemic areas of Africa where 70%-90% of the population has been exposed to HBV.^{25, 44} When anti-HBc only data is evaluated, the rates range from 0% to 15% (median of 1.1%).²⁵ The low prevalence seen in some African studies may be due to the sensitivity of the assays used in both HBsAg and HBVDNA detection.

The Federal Medical Centre Owerri is a major center for blood donation in Imo State and South-East Nigeria. Blood donors are screened for HBsAg using the Rapid test method, but anti HBc and HBVDNA are not done. This will most likely result in post transfusion HBV infection in future. On the average, eight hundred blood donors are screened for HBV infection annually with a 3.4% prevalence of HBsAg positivity [unpublished data obtained from the hematology laboratory and NBTS records]. Knowledge of the prevalence of Occult hepatitis B virus infection among blood donors in the center will go a long way in preventing the spread of transfusion transmitted HBV and subsequent development of chronic liver disease [Cirrhosis and Hepatoma] in the area.

Transfusion transmitted Hepatitis B virus infection continues to be a major problem despite the routine screening of blood for prospective donation for HBV with HBsAg using the ELISA method. This may be due to Occult Hepatitis B virus infection.

This study is expected to identify the prevalence of chronic HBV infection using serologic markers other than HBsAg. This will enable a possible recommendation of using these other serologic markers as part of the routine screening of donor blood products for HBV to further reduce transfusion transmitted HBV. Also, subjects with Occult Hepatitis B Virus infection [anti HBc positive and detectable DNA] can then be followed up and monitored for overt HBV and early detection of Hepatocellular carcinoma. Furthermore, those who require treatment can be detected. The data obtained will also serve as a background for further related studies.

GENERAL OBJECTIVE OF THE STUDY

To determine the prevalence of Occult Hepatitis B virus infection among healthy HBsAg negative blood donors in Owerri, South-East Nigeria

SPECIFIC OBJECTIVES

1. To determine the seroprevalence of positive anti HBc antibody among healthy HBsAg negative blood donors in Owerri
2. To determine the prevalence of detectable serum HBV DNA among the anti HBc positive blood donors

MATERIALS AND METHODS

STUDY SITE

The study was carried out at the Federal Medical Centre Owerri, which is a major blood donation center in the region with an average of fifty blood donations monthly between 2009 and 2014. It is a 417 bedded tertiary health institution serving as a major referral center in the state. Also, the National Blood Transfusion Service [NBTS] center for South-East Nigeria established in March 2006 is located inside the institution and this serves as a major source of blood products for transfusion for several hospitals in this region. The blood bank and NBTS center were used for the study.

STUDY POPULATION AND SAMPLE SIZE

This was a cross-sectional descriptive study conducted in consecutive prospective blood donors who were HBsAg negative seen in the hospital that satisfied the inclusion criteria.

SAMPLE SIZE

The sample size was calculated using the formula

$$n = \frac{Z^2 pq}{d^2}$$

where n=estimated sample size

z= standard deviation set at 1.96

p= estimated prevalence of anti HBc

q= 1-p

d= degree of accuracy desired [desired precision limit]=0.05

Therefore using 13% prevalence⁸⁶

$$n = \frac{1,96^2 \times 0.13 [1-0.13]}{0.05^2}$$

$$0.05^2$$

$$n = 173.6$$

so, the estimated sample size was 174

For the study, Occult Hepatitis B Virus Infection [OBI] was defined as HBsAg negative subjects who are anti HBc positive with detectable HBV DNA in the serum.³²

SELECTION/RECRUITMENT OF SUBJECTS

INCLUSION CRITERIA

1. Healthy prospective HBsAg negative blood donors.
2. Adults of both sexes aged 18years and above.
3. Adults who give consent to participate in the study.
4. Adults who are Human Immunodeficiency Virus [HIV] negative.

EXCLUSION CRITERIA

1. Prospective blood donors who are HBsAg positive.
2. Adults with clinical evidence of liver disease.

ETHICAL APPROVAL

Ethical approval for the study was sought for and obtained from the Ethical Committee of Federal Medical Centre Owerri.

CLINICAL EVALUATION

After Ethical approval, permission to carry out the study was obtained from the Head of Haematology department and the Zonal Coordinator of the National Blood Transfusion Service. The study was explained to the subjects in English or in their local languages for full comprehension and written informed consent or oral consent with thumb print [for those who cannot read and write] was obtained from them. The individuals who met the study criteria were enrolled into the study

The biodata and other relevant history were obtained from all subjects by a structured questionnaire which was administered by the researcher. A thorough physical examination was performed and documented by the researcher to confirm the absence of stigmata and other clinical signs of chronic liver disease. Blood pressure of less than 140/90mmHg was taken as normal while blood pressure of 140/90mmHg and above were taken as hypertension. Liver span of between 8 and 12cm were taken as normal, less than 8cm were taken as shrunken and above 12cm were taken as hepatomegaly

LABORATORY TESTS

The following lab tests were conducted on the subjects.

1. Anti HBc antibody assay [IgG]
2. Anti HBs antibody assay
3. HBV DNA

All recruited subjects were potential blood donors who had HBsAg serology done in the blood bank and NBTS by the laboratory Scientists working in these laboratories using ELISA. Also, HIV serology was done using Polymerase Chain Reaction by the laboratory

Scientists and those who were seronegative were recruited in the study as indicated by the inclusion criteria.

SAMPLE COLLECTION AND ANALYSIS

Using a pair of latex gloves for each subject, 10 milliliters [mls] of blood was obtained after cleaning the area of needle insertion with Chlorhexidine [savlon] and then spirit soaked in cotton wool.

Anti HBc and Anti HBs

4mls of the sample was collected and using an automated microparticle Enzyme Linked Immunosorbent Assay [ELISA] screened for Anti HBc [AcSYM Core, Abbott, Wiesbaden] and AntiHBs [AcSYM AUSAB Abbott]. These samples were frozen until they were analysed. Anti HBc [IgG] values < 1 IU/ml were taken as positive while values ≥ 1 IU/ml were taken as negative. Anti HBs values ≥ 10 IU/ml were taken as positive while values < 10 IU/ml were taken as negative.

HBV DNA

3mls each of the Anti HBc positive blood samples were stored in a potassium Ethylene Diamine Tetracetic Acid [EDTA] bottle at 4°C to 8°C and transported via ice packs to Safety Molecular Pathology Laboratory located at Independence layout Enugu where HBV DNA assay was done by the laboratory scientist using Real time nested Polymerase Chain Reaction [PCR] after preparation using the NucliSens Extractor [Organon Teknika Boxtel] and aysis buffer to increase the nucleic acid yield. Detected DNA was estimated in International Units per Milliliter [IU/ml]. The limit of detection by this method is 20 IU/ml.

DATA ANALYSIS

Data obtained were subsequently analyzed using SPSS Version 20 statistical software. Variables were compared using relevant statistical methods

- Means were expressed as means \pm [SD]
- Categorical variables were compared using chi-square while continuous variables were compared using paired t-test. For test of significance, P values <0.05 was taken as statistically significant.

RESULTS

A total of one hundred and eighty (180) participants were enrolled for the study, The age of the subjects range from 21 to 45 years with a mean [SD] of 26.85 ± 5.21 years

The sociodemographic and other clinical characteristics of the study participants are shown in Tables 1 and 2.

TABLE 1: Sociodemographic characteristics of the 180 study participants

Clinical characteristic	Frequency	Percentage
Age		
20-29 years	142	78.8
30-39 years	33	18.4
40-49 years	5	2.8

Occupation		
Unemployed	9	5
Artisans	3	1.7
Civil servants	39	21.7
Students	119	66.1
Business/Trading	10	5.6
Sex		
Male	148	82.2
Female	32	17.8
Tribe		
Igbo	161	89.4
Yoruba	14	7.8
Hausa	5	2.8

TABLE 2: CLINICAL CHARACTERISTICS OF THE 180 STUDY PARTICIPANTS

Clinical characteristic	Frequency	Percentage
Blood transfusion history		
Yes	20	11.1
No	160	88.9
Sharing of sharp objects		
Yes	77	42.8

No	103	57.2
Leg swelling		
Yes	0	0.0
No	180	100.0
Multiple sexual partners		
Yes	51	28.3
No	129	71.7
Contact with anybody with yellowness of the eyes		
Yes	42	23.3
No	138	76.7
Liver span (cm)		
8	46	25.6
9	27	15.0
10	45	25.0
11	11	6.1
12	51	28.3
Blood pressure (mmhg)		
Normotensive (<140/90)	158	87.8
Hypertensive (>140/90)	22	12.2

Figure 1 shows the distribution of anti HBc (IgG) among the 180 study participants. Of the 180 participants, 33 (18.3%) had anti HBc IgG +ve antibody.

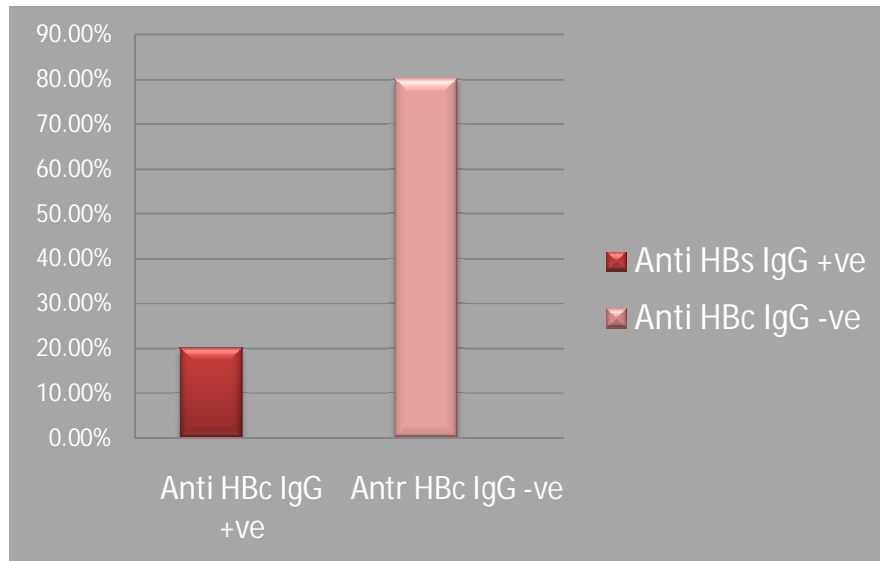


Figure 1: Anti HBc (IgG) distribution of the 180 study participants.

Table 3 shows Sociodemographic and other characteristics of AntiHBc(IgG) Positive/Negative study participants.

The age, occupation or sex were not statistically associated with anti HBc IgG Antibody.

Table 3: Sociodemographic and other characteristics of AntiHBc(IgG) Positive/Negative study participants.

	Anti HBc		Total	X(P value)
	Positive (%)	Negative (%)		
Age				
20-29 years	29(16.1)	113(62.7)	142(78.8)	10.97(0.03)
30-39 years	4(2.2)	29(16.2)	33[18.4]	
40-49 years	0[0]	5(2.8)	5[2.8]	
Total	33(18.3)	147(81.7)	180(100)	
Occupation				
Unemployed	2(1.1)	7(3.9)	9(5)	4.71(0.32)
Artisans	0(0)	3(1.7)	3(1.7)	
Civil servants	3(1.7)	36(20)	39(21.7)	
Students	26(14.4)	93(51.7)	119(66.1)	
Business/Trading	2(1.1)	8(4.5)	10(5.6)	
Total	33(18.3)	147(81.7)	180(100)	
Sex				
Male	27(15)	121(67.3)	148(82.2)	0.29[0.81]
Female	6(3.3)	26(14.4)	32(17.8)	
Total	33(18.3)	147(81.7)	180(100)	

Out of the 33 anti HBc[IgG] positive participants,5(15.2%) of them were anti HBs positive while 28(84.8%) of them were anti HBs negative. Of the 5 antiHBs positive participants who were antiHBc[IgG] positive, 3[60%] were males and 2[40%] were females. This is shown in table 2

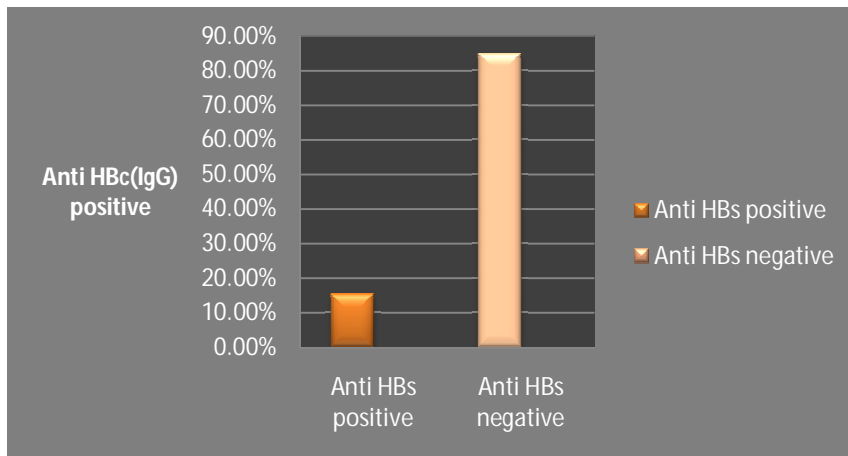


Figure 2: Anti HBs distribution of the 33 Anti HBc (IgG) positive participants.

Figure 3 shows the distribution of detectable HBV DNA among the 33 anti-HBc [IgG] positive participants.

Of the 33 anti HBc [IgG] positive study participants,13[39.4%] had detectable HBV DNA \geq 20IU/ml] while 20 [60.6%] had undetectable HBV DNA [$<$ 20IU/ml]

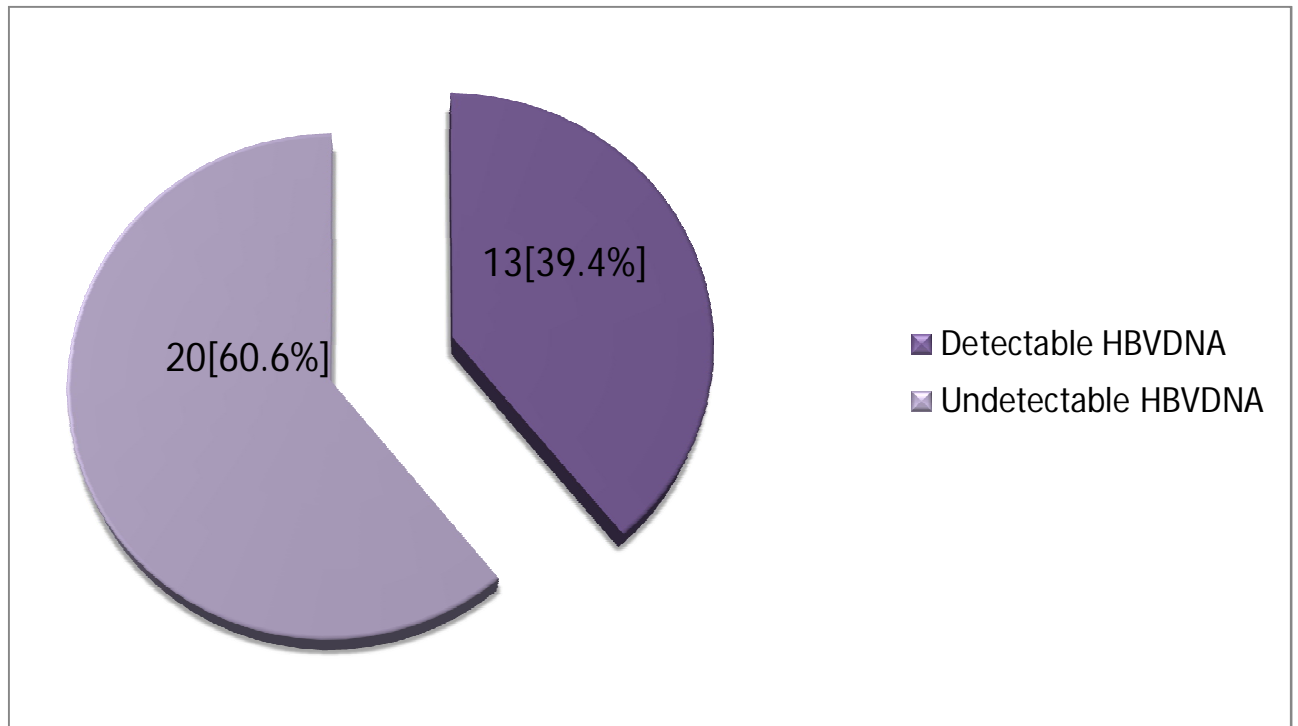


Figure 3: Distribution of detectable HBVDNA among the 33 Anti HBc(IgG) positive participants.

Figure 4- Out of the 180 study participants, 13(7.2%) were OBI positive while 167 [92.8%] of them were OBI negative

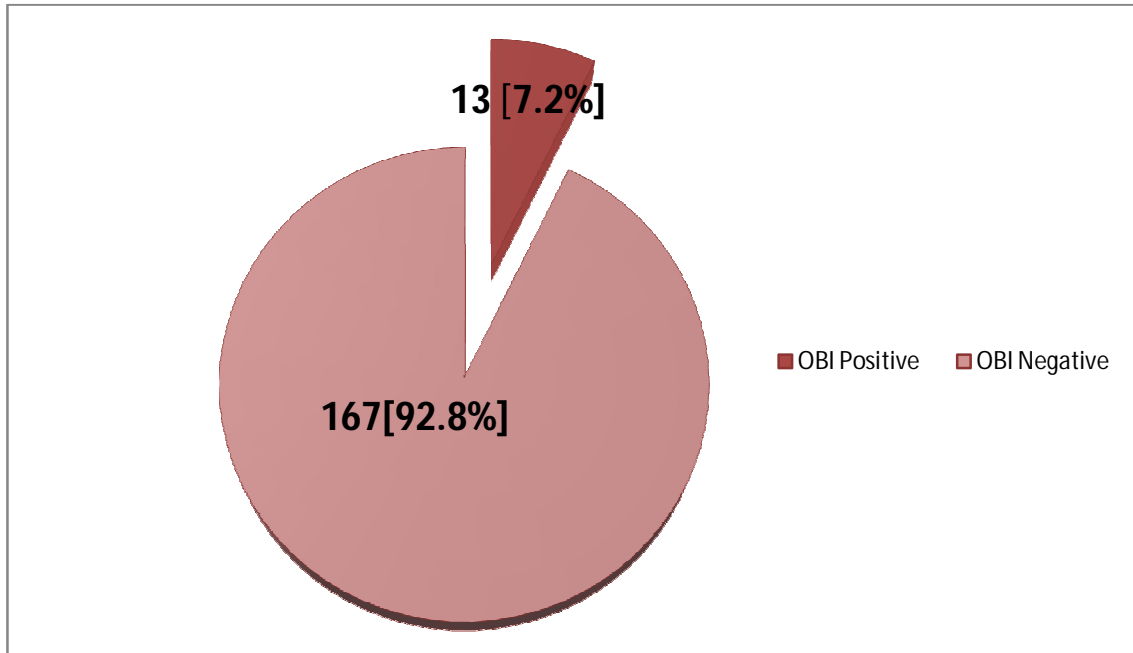


Figure 4: Distribution of OBI among the study participants

Table 4 gives a summary of the laboratory findings among the 180 study participants

TABLE 4 :Laboratory investigation distribution of the study participants

	Frequency	Percentage
Anti HBc[IgG]		
Positive	33	18.3
Negative	147	81.7
Anti HBs		
Positive	30	16.7
Negative	150	83.3
HBV DNA		
Positive	13	39.4
Negative	20	60.6

DISCUSSION

The ages of the participants were between 21 and 45 years with a mean of 26.85 ± 5.21 years.

Out of the 180 participants, 142[78.8%] were in the age group of 20-29 years. This high prevalence in this age group is not surprising as individuals within this age bracket are more likely to undertake voluntary blood donation which is the major source of blood for NBTS.²⁹

This was also the highest age group of participants who had OBI. The age group of 30-39 years was the highest age group prevalence of OBI observed by Oluyinka and his colleagues.³⁰ The participants were mostly students [66.1%] and Civil servants [21.7%] which form the predominant population in the area of study.

The relatively higher haemoglobin concentration in males, make them more likely to meet the eligibility criteria for blood donation than females. This was reflected in the higher male prevalence of 82.2% observed in this study. This was similar to the study done by Olotu and his colleagues who observed a high male prevalence of 97%.³³

All the study participants had normal liver span of between 8 and 12cm. This is not surprising because abnormal liver span is one of the clinical evidence of liver disease which was part of the exclusion criteria. 11.1% of the study participants and 7.7% of the participants with OBI had previous history of blood transfusion while 42.8% and 30.8% of the study participants and those with OBI shared sharp objects respectively. These constitute major risk factors of HBV infection.

Of the 180 participants, 158[87.8%] of them were normotensive while 22[12.2%] were hypertensive. This high prevalence of normotensive individuals is related to the age distribution of study participants as individuals in the predominant age groups are less likely to be hypertensive.

In this study 18.3% were antiHBc[IgG] positive. This is higher than the prevalence of 13.6% observed by Japhet and his colleagues at Ile-Ife.³¹ Also, a low prevalence of 4.37% was observed by Salawu and his colleagues in Soth-West Nigeria.³² The prevalence of hepatitis B in various states of the country could help explain this variation in prevalence rates. The nature of kits used could also explain this. Said and her colleagues in Egypt observed a prevalence of 16.6%.⁴⁰ The slightly lower prevalence in this study done in Egypt is due to the fact that two assays were used to confirm anti HBc: ABBOT and ACHITECT anti HBc assays and only those samples reactive to the two assays were regarded as positive while in this study only one assay [ABBOT] was used. A lower prevalence of 9% was also noted by Eightamman and his colleagues among blood donors in Egypt, 18.9% of them had both antiHBc and antiHBs.⁴⁵ Higher prevalence of antiHBc positivity were also observed in areas with high HBV endemicity.³⁴

Of the 33 antiHBc[IgG] positive participants, 13 [39.4%] of them had detectable DNA in this study. This is higher than the prevalence of 5.4% observed by Olotu and his colleagues in Ile-Ife³³ and 10.5% observed by Shambesh and his colleagues in Libya among 123 antiHBc positive blood donors.³⁵ This study in Libya used Real time Polymerase Chain Reaction [COBAS, Ampliprep COBAS, TaqMan] with a sensitivity of 20IU/ml which was the same assay used in this study. The higher prevalence of HBV DNA among antiHBc positive blood donors seen in this study may be due to the fact that the Enzyme Linked Immunosorbent Assay [ELISA] test kit used for HBsAg determination in this study has a sensitivity of 0.45IU/ml which is low compared to the VITROS 3600 immunodiagnostic system with a sensitivity of 0.085IU/ml used in the study by Shambesh and his colleagues. This means that some of the blood donors in this study may actually have been HBsAg positive if a more sensitive ELISA test kit was used. The high prevalence seen in this study may also be due to the high endemicity of HBV infection in the area.³⁴

Of the 180 study participants, 30[16.7%] of them were anti HBs positive. This shows that less than 20% of the adult population in the study area is immune to HBV. This relatively low prevalence could be due to poor awareness of access to immunization against HBV. Of the 33 anti HBcIgG positive participants, 5[15.2%] of them were also anti HBs positive. This is similar to the prevalence of 18.9% observed by Eightaman and his colleagues in Egypt. Only one of participants with OBI was anti HBs positive. This low prevalence of anti HBs in individuals with OBI shows that the presence of anti HBs in the serum of individuals may make them less likely to have detectable HBV DNA.

The prevalence of 7.2% of OBI in this study agrees with the documented evidence that there is high prevalence of OBI in Nigeria and Sub-Saharan Africa. In Nigeria, recent studies have shown a prevalence of 8% among blood donors in South East Nigeria by Nna and his colleagues and 17% and 5.4% in studies in South-West Nigeria by Oluyinka and his colleagues and Olotu and his colleagues respectively.^{29, 30, 33} This is not surprising as this could be due to the high prevalence of HBV infection in the area. This is evident in the systematic review and meta-analysis from 46 studies in Nigeria from 2000 to 2013 by Musa and his colleagues which showed a prevalence of 13.6% among 34,376 persons with a prevalence of 14% among blood donors.⁴⁶ Emechebe and his colleagues also showed a HBV prevalence of 9 to 39% in South-East Nigeria.⁴⁷ WHO in their weekly epidemiological report in 2009 stated that areas with a HBV prevalence of up to 8% were classified as endemic for HBV infection.⁴⁸

In the study by Oluyinka and his colleagues, Nested Polymerase Chain Reaction assay which covers the HBV genome Core, Surface and X genes was used to test for HBV DNA which was the same assay used in this study. Therefore, the higher prevalence in the study by Oluyinka and his colleagues is due to the fact that, HBV DNA was tested for both anti HBc positive and negative participants, whereas in this study, only anti HBc positive participants

were tested for HBV DNA. In the USA, Hollinger and his colleagues observed an OBI prevalence of 0.1 to 2.4% in an area where only 5% of the population has prior exposure to HBV.²⁵

In Asia an OBI prevalence of 10.6% was observed among 659 healthy individual in China.

⁹¹In Sub-Saharan Africa, OBI prevalence of 15% & 10% respectively were shown in Sudan and Ivory Coast.^{36, 37}

The median viral load determined by Nested PCR-Taqman biochemistry in the participants who had detectable viral load was 43IU/ml which was similar to 51IU/ml observed by Nna. E. and his colleagues who also observed a viral load which ranged from unquantifiable to 58IU/ml.²⁹ The low level of viral load seen in these studies agree with another report which showed almost all OBI cases are infected with replication competent HBV, revealing a strong suppression of replication activity and gene expression thereby resulting in a reduced viral load. A higher median viral load of 200IU/ml was observed by Said and her colleagues in Egypt.⁴⁰ This higher median viral load may be due to the higher sensitivity of the assay used in the HBV DNA detection, the real time PCR[QAGEN Germany] with 3.8IU/ ml detection limit as compared to the assay detection limit of 20IU/ml used in this study.

One of the participants with OBI in this study had a viral load of 240 IU/ml which is above the cutoff point of less than 200 IU/ml used to define OBI at the Toarmina conference.³⁴

However, the participant was included as OBI by virtue of its definition for the purpose of this study which is the presence of anti HBcIgG and detectable HBV DNA. This relatively high viral load observed in this participant may be due to the low sensitivity of the HBsAg detection assay used as the individual might have been HBsAg positive if a more sensitive assay was used.

CONCLUSION

Occult HBV infection is not uncommon among Nigerian blood donors. High prevalence is seen in high endemic areas worldwide and this is a general burden on the safety of blood transfusion. The potential infectivity of OBI in blood transfusion cannot be excluded. Most OBI patients are asymptomatic and would only be detectable by systematic screening of large population. This study signifies the high prevalence of OBI and proposes that the blood samples in Nigeria should be pre-tested for OBI by Nucleic Acid Testing and or anti HBc prior to transfusion to minimize the HBV transmission risk.

RECOMMENDATIONS

The findings of this study warrant the following recommendations:

1. Anti HBc screening should be made mandatory for all blood donors as this would possibly eliminate the risk of unsafe blood donation.
2. More awareness should be created on the need for immunization against HBV to further reduce the risk of transmission of OBI.
3. More research should be undertaken to derive more cost effective methods for HBV DNA determination to maximize its use in screening blood donors.
4. Specific management strategy for OBI should be implemented.

LIMITATIONS OF THE STUDY

1. Some individuals who were anti HBc negative may still have detectable HBV DNA
2. Some HBsAg negative donors may truly be positive but missed because of the sensitivity of the assay used.

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