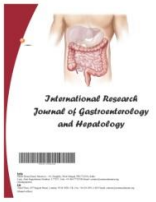

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Patterns of Hepatitis B Virus Infection Serologic Markers among Blood Donors at a Tertiary Healthcare Facility in Central Nigeria

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Authors' contributions

This work was carried out in collaboration among all authors. Author HCE designed the study, collected samples, performed laboratory and statistical analyses and wrote the first draft of the manuscript. Authors HIM and GRP designed and supervised the study, manage literature searches, wrote the protocols and managed the analyses of the study. All authors read and approved the final manuscript.

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ABSTRACT

Aims: This study was conducted to evaluate the patterns of hepatitis B virus infection serologic markers among blood donors at a tertiary healthcare Facility in Central Nigeria
Study Design: The study was a cross sectional study.
Place and Duration of Study: Keffi, Nasarawa State, between January and October, 2018.
Methodology: Blood sample (3 ml) was collected from each of the 400 consenting blood donors at Federal Medical Centre, Keffi, Nasarawa State and their socio-demographic information obtained using structured questionnaires. The sera were screened for HBV infection serologic markers (HBsAg, HBsAb, HBeAg, HBeAb and HBcAb) using HBV-5 rapid panel test kit (CTK Biotech. Inc. San Diego, USA). Data collected were analysed using Smith's Statistical Package (version 2.8, California, USA) and *P* value of ≤ 0.05 was considered statistically significant.

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Results: Majority of the 400 blood donors screened were males (391/400) and aged 25-34 years (203/400). Of these, 31(7.8%) were positive for HBsAg, 113(28.3%) for HBsAb, 11(2.8%) for HBeAg, 18(4.5%) for HBeAb and 78(19.5%) for HBcAb. Analysi;s of these sero-markers indicated that 1.5% of the donors had chronic infection with high viral replication, 1.2% had acute infection with high viral replication, 4.5% were carriers with low viral replication, 0.7% had occult infection, 0.5% were recently vaccinated, 15.5% were immuned due to successful vaccination, 12.8% were immuned as a result of natural previous exposure to the virus while 63.3% were not exposed to the virus. Age and gender were not associated with HBV infection in this study ($P > 0.05$). However there was statistically significant difference between age and gender with rate of HBcAb ($P < 0.05$).
Conclusion: We confirmed the presence occult HBV infection among prospective blood donors in the study area. Hence, HBV profiling for routine screening of blood donors should be made mandatory to avoid transfusion-associated hepatitis B virus infection.

Keywords: Hepatitis B virus; infection; seromarkers; blood donors; Central Nigeria.

1. INTRODUCTION

Viral hepatitis B, caused by hepatitis B virus (HBV) has emerged as a major disease of mankind and is a serious global public health problem, accounting for over 360 million cases of chronic hepatitis and 620,000 deaths per year [1]. HBV is a double-stranded DNA virus of a complex structure that causes infection of the liver [2]. The virus belongs to the *Hepadnaviridae* family and is the most common cause of chronic liver disease; hepatocellular carcinoma and necrotizing vasculitis [3]. HBV can cause both acute and chronic infections; and during the acute phase of infection, symptoms are not experienced by most people. Nevertheless, certain individuals develop acute illness with symptoms that last several weeks, including yellowing of the skin and eyes (jaundice), nausea, dark urine, extreme fatigue, abdominal pain and vomiting [1]. Additionally, in individuals with acute hepatitis, a small subset can develop life-threatening acute liver failure whereas in certain individuals, HBV establishes a chronic liver infection that progresses to cirrhosis or cancer of the liver [1,3]. It is reported that HBV is 50 to 100 times more infectious than HIV [4].

The most common outcome after HBV infection is the expression of diverse serological markers of varying epidemiological and clinical significance namely; hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (HBsAb), hepatitis B envelope antigen (HBeAg), hepatitis B envelope antibody (HBeAb), hepatitis B core antigen (HBcAg) and hepatitis B core antibody (HBcAb) [5]. Symptomatic and asymptomatic forms of both acute and chronic infections may be discovered incidentally only through laboratory assay of these viral markers [6]. These markers may occur singly or in various combinations depending on the natural history of the infection [5,7].

Early in the course of HBV infection, HBsAg is present in the serum and disappears with the production of specific antibodies against it (HBsAb) and recovery from the disease. HBeAg appears early in acute infection and is associated with high rate of infectivity and higher chances of chronicity, but may also recede on seroconversion to HBeAb. HBcAg is not detectable in the blood stream; however, detection of HBcAb suggests acute or chronic infection [6-8]. Hence, detection of these markers and the serological patterns they present in individuals help to determine stages of HBV infection and plan better management strategies [5].

Transfusion-associated hepatitis B virus (TAHBV) infection continues to be a major public health problem particularly in developing countries [9]. This is due to the fact that the presence of HBsAg is the commonly used method for detecting HBV infection in blood banks [10] because most hospitals cannot afford DNA testing of all collected units of blood which serves as the only possibility of achieving zero risk of post-transfusion HBV infection [11]. However, in the absence of HBsAg, Occult hepatitis B infection (OBI) may occur which basically refers to the presence of HBV DNA in the serum of individuals without HBsAg but with other serological evidence of the infection such as HBcAb and HBeAg [9]. Additionally, there is also a potential risk of transmission of HBV during the window period. This is the period during the host's serological response between the removal of HBsAg and the appearance of HBsAb and in this case, HBcAb may be the only serological evidence of disease [11].

Although, there are data on HBV infection serologic markers in different population groups from different part of Nigeria [5-7,12-13], however, this is the first research work that has successfully reported the prevalence and patterns of HBV infection serologic markers among blood donors at Federal Medical Centre, Keffi, Nasarawa State, Nigeria. Furthermore, the results from this study can be used for intervention strategies for prevention and control of HBV infection especially post-transfusion associated HBV infection.

2. MATERIALS AND METHODS

2.1 Study Area

This study was conducted at Federal Medical Centre, Keffi, Nasarawa State, Nigeria. It is one of the 22 Federal Medical Centres established by the Federal Government of Nigeria to provide basic and advanced health needs of people living in the State and other neighboring States. Keffi city, where the centre is located is approximately 68 km from Abuja, the Federal Capital Territory and 128 km from Lafia, the capital of Nasarawa State. It is located geographically between latitude 8°3'N of the equator and longitude 7°50'E and situated on an altitude of 850 m above sea level [14].

2.2 Study Population

The study population comprises of male and female commercial donors and adult relatives of patients requiring blood transfusion aged18- 65 years who came to donate blood at the Blood Group and Serology unit of the Federal Medical Centre, Keffi.

2.3 Sample Size Determination

The sample size for this study was determined using the formula by Naing et al. [15] for sample size calculation at 0.05 level of precision;

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

Where:
n = required sample size
Z = standard normal deviation at the required confidence interval (1.96) which corresponds to 95% confidence interval.
P = prevalence of HBV infection from previous study (9.7%) (0.1) [6].
Q = 1 – p = 0.9
d = degree of precision expected (0.05)

$$n = \frac{(1.96)^2(0.1)(0.9)}{(0.05)^2} = \frac{3.8416\times0.09}{0.0025} = \frac{0.3457}{0.0025}$$
$$= 138.3$$
$$n = 138$$

To minimize error, this was however rounded up to 400 samples.

2.4 Sample Collection, Processing and Storage

The Blood Group and Serology unit of the Medical laboratory was used as the sample collection point. Three mls of blood sample was obtained from each consenting participant by venepuncture and placed in an appropriately labelled plain tube. This was allowed to clot at room temperature and spun for 5 min at 3000 rpm to separate out the serum [16]. The resultant sera were harvested into well-labelled cryovials and stored at −20°C until use.

2.5 Laboratory Analysis

2.5.1 Detection of HBV infection serologic markers

All sera were screened for HBV infection serologic markers (HBsAg, HBsAb, HBeAg, HBeAb and HBcAb) using HBV-5 rapid panel test kit (CTK Biotech. Inc. San Diego, USA). The test was conducted and results interpreted according to manufacturer's instructions.

2.6 Data Analysis

The data obtained were analyzed using Smith's Statistical Package (version 2.8, California, USA). Chi-square test was conducted at 95% confidence interval and *P* values ≤ 0.05 were considered statistically significant.

3. RESULTS AND DISCUSSION

Blood transfusion is a very important therapeutic procedure. Therefore, providing safe blood devoid of transfusion transmissible agent is needed [11]. This current study was conducted to evaluate patterns of HBV infection serologic markers among prospective blood donors in tertiary healthcare facility in Central Nigeria. A total of 400 eligible blood donors majority of whom were males (391/400) and aged 25-34 years (203/400) were screened for HBV infection serologic markers. Of these, 31(7.8%) were positive for HBsAg, 113(28.3%) for HBsAb,

11(2.8%) for HBeAg, 18(4.5%) for HBeAb and 78(19.5%) for HBcAb (Fig. 1).

The recorded 7.8% rate of HBsAg in this study which is the most frequently used seromarker for screening of HBV infection [10] is regarded as moderate according to World Health Organization's classification [17]. However, this rate (7.8%) was higher than the 6.7% reported by Okonkwo et al. [18] among blood donors at University of Port-harcourt Teaching Hospital, 7.5% by Alaku et al. [19] among HIV patients at a tertiary health care facility in Central Nigeria and 4.4% by Argaw et al. [20] among children in Southern Ethiopia. Notwithstanding, it was lower than the 17.5% reported by Agbesor et al. [21] among blood donors in Asokoro General Hospital Abuja, 9.7% by Mohammed et al. [6] in a subset of young people in central Nigeria and 9.8% by Guimaraes et al. [22] among people living in poverty in Brazil. It is likely that the reported varying rates from the different studies were impacted by location and study population type with different peculiar risk factors.

We found that 113(28.3%) of the blood donors in this study had HBsAb which is usually a neutralizing antibody produced by the immune system against HBsAg as a results of previous natural exposure or due to successful vaccination against the virus [7-8]. This result compares well with 22.5% prevalence of natural HBsAb among healthy individuals in Benue [5], 22.2% among Surgeons in Lagos [23] and 27.5% among hospital personnel in Cairo, Egypt [24].

In this study, HBeAg which is an indicative of the replicating phase of the virus was prevalent in 2.5% of the participants (Fig. 1). This rate is lower than the 6.5% reported by Abah and Aminu [13] among pregnant Nigerian women and 4.7% among individuals with HBsAg seropositivity in Benue State [7]. However, there is a cause for alarm because these donors (2.5%) have 70-90% chance of transmitting the virus [13, 25]. They are also expected to have high chances of developing persistent liver disease leading to cirrhosis and even primary liver cancer if not treated [26].

HBeAb is the antibody produced by the body against HBeAg and its presence indicates lowered infectivity and transmission of the virus [8]. Just like the HBsAb, it may also an indication of recovery from HBV infection [27]. There was a prevalence of 4.5% of this antibody (HBeAb) in this study (Fig. 1). This is however lower than the 4.7% reported in a subset of young people in Central Nigeria [6], 8.0% among HBsAg seropositive individuals in Makurdi [7] and 51.6% among pregnant Nigerian Women [13]. Differences in study populations may account for the observed differences in findings

The first antibody to appear in HBV infection is HbcAb and its presence in an individual symbolizes earlier contact with virus [8]. This seromaker was found in 19.3% of the donors and these were those that have had contact with the virus at one time or the other in their lives [8]. Although the recorded rate of this antibody in this study is lower than the 32.5% reported among blood donors in Ilorin [28], 38.2% in Benue State [5] and 28.0% in a subset of young people in Central Nigeria [6]. It was however lower than the 10.5% and 16.6% reported among blood donors in India and Egypt respectively [29,30]. Notwithstanding, the detection of HBcAb among prospective apparently healthy blood donors in this study calls for concerns because some of these donors may have occult HBV infection or may probably be in the window period of the infection.

Furthermore, in this current study, the rate HBcAb was significantly associated with age of the participants ($P<0.05$). However, rates of other seromarkers (HBsAg, HBsAb, HBeAg and HBeAb) were not associated with age ($P>0.05$). Nevertheless, HBsAg was detected with the highest prevalence (14.0%) among donors aged 15-24 years and lowest (6.3%) among those aged ≥45 years (Table 1). This observation agrees with previous reports of Buseri et al. [31] in Niger Delta, Kuta et al. [32] in Kwara State and Bagiyalakshim et al. [33] in India who reported higher rate of the seromarker among younger subjects aged 18-30 years. On the other hand, it contradicts that of Alaku et al. [19] and Agbesor et al. [21] who reported higher rate among participants aged 40-60 years in Central Nigeria. The age of peak infection in this study falls within the age range of greatest sexual activity, hence, suggesting the role of sexual intercourse in the viral transmission.

Similarly, gender was significantly associated with rate of HBcAb in this study ($P<0.05$) and other markers of HBV infection (HBsAg, HBsAb, HBeAg and HBeAb) were statistically insignificant ($P>0.05$). However, the rates of HBsAg (7.9%), HBsAb (28.4%), HBeAg (2.8%) and HBcAb (19.7%) were higher among males than females indicating that males were likely for infected with HBV infection than their female counterparts (Table 1). This finding agrees with

that of Isa et al. [12] in a tertiary institution in North Western Nigeria and Pennap et al. [34] among students of a Nigerian tertiary institution. In contrast, Mustapha et al. [35] reported higher prevalence of HBV infection in females than males among HIV patients in Gombe State. The high prevalence of HBsAg among males in this study may be connected to the higher rate of promiscuity among males than females in Nigeria [36]. It may also be as a result of restriction of females to their houses which reduced the chances of being exposed to HBV risk factors.

Detection of HBV infection serologic markers and the pattern they present in an individual help in determining the status and stages of the infection [8]. In this study, based on these patterns, 1.5% of the donors had chronic HBV infection with high viral replication, 1.2% had acute infection with high viral replication, 4.5% were carriers with low viral replication, 0.7% had occult infection, 0.5% were recently vaccinated, 15.5% were immuned due to successful vaccination, 12.8% were immuned as a result of natural previous exposure to the virus and 63.3% were not yet exposed to the virus (Table 2).

The recorded prevalence rates of 1.5% and 1.2% for chronic and acute infection respectively in this study were lower than the 3.8% and 8.7% reported by Mbaawuaga *et al*. [5] in Benue State. This observed difference may be because while Mbaawuaga *et al*. [5] conducted their study among pregnant women, who naturally have suppressed immunity [37], we recruited apparently healthy blood donors in this current study.

It is worthy of note that, the observed 0.7% prevalence of occult HBV infection among blood donors in this current study is a reason to panic. This is because these individuals (0.7%) were HBsAg sero-negative and their blood must have been declared free of HBV infection and safe for transfusion since in Federal Medical Centre Keffi, Blood Group and Serology unit where blood is screened before transfusion, only HBsAg seromarker is used to detect the presence of HBV infection. Hence, patients transfused with such blood are at high risk of post-transfusion HBV infection from blood that was not properly screened before transfusion.

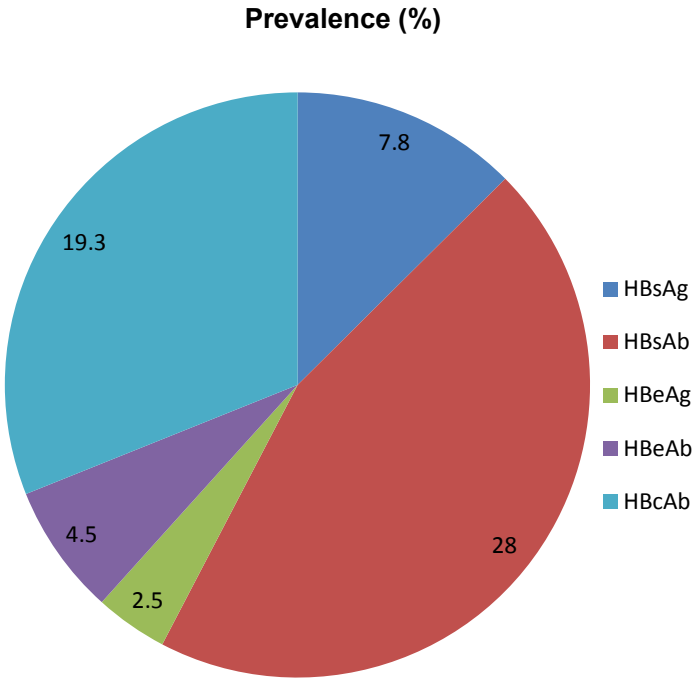

Fig. 1. Prevalence of HBV infection serologic markers among prospective blood donors at Federal Medical Centre, Keffi, Nigeria

Table 1. Prevalence and distribution of HBV infection serologic markers in relation to age and gender among prospective blood donors at Federal Medical Centre Keffi, Nigeria

Parameter	No. Examined	No. Positive (%)				
		HBsAg	HBsAb	HBeAg	HBeAb	HBcAb
Age (Years)						
15-24	50	7(14.0)	13(26.0)	3(6.0)	2(4.0)	13(26.0)
25-34	203	13(6.4)	52(25.6)	3(1.5)	8(3.9)	35(17.2)
35-44	131	10(7.6)	41(31.3)	4(3.0)	8(6.1)	24(18.3)
≥45	16	1(6.3)	7(43.8)	1(6.3)	0(0.0)	6(37.5)
Total	400	31(7.8)	113(28.3)	11(2.8)	18(4.5)	78(19.5)
p-value		0.4621	0.1001	0.9999	0.1777	0.0005
Gender						
Male	391	31(7.9)	111(28.4)	11(2.8)	17(4.3)	77(19.7)
Female	9	0(0.0)	2(22.2)	0(0.0)	1(11.1)	1(11.1)
Total	400	31(7.8)	113(28.3)	11(2.8)	18(4.5)	78(19.5)
p-value		0.1435	0.0735	0.2646	0.9920	0.0222*

**Statistically significant*

Table 2. Patterns of HBV infection serologic markers among prospective blood donors at Federal Medical Centre Keffi, Nigeria

Pattern of HBV serologic markers	Interpretation	Prevalence (%)
HBsAg+, HBsAb-, HBcAb+, HBeAg+, HBeAb	Chronic infection with high viral replication	6(1.5)
HBsAg+, HBsAb-, HBcAb-, HBeAg+, HBeAb	Acute infection with high viral replication	5(1.2)
HBsAg+, HBsAb-, HBcAb+, HBeAg-, HBeAb	Carrier with low viral replication	18(4.5)
HBsAg-, HBsAb-, HBcAb+, HBeAg-, HBeAb	Occult infection	3(0.7)
HBsAg+, HBsAb-, HBcAb-, HBeAg-, HBeAb	Recently vaccinated	2(0.5)
HBsAg-, HBsAb+, HBcAb-, HBeAg-, HBeAb	Immune due to vaccination	62(15.5)
HBsAg-, HBsAb+, HBcAb+, HBeAg-, HBeAb	Immune due to natural previous exposure	51(12.8)
HBsAg-, HBsAb-, HBcAb-, HBeAg-, HBeAb	Unexposed (Susceptible)	253(63.3)
Total		**400(100)**

4. CONCLUSION

This study reveals the presence HBV infection among prospective blood donors in the study area. This is frightening since the infected donors; particularly those with occult HBV infection may serve as source of infection to blood recipients. Hence, HBV profiling for routine screening of blood donors should be made mandatory to avoid transfusion-associated hepatitis B virus infection.

CONSENT

All authors declare that written informed consent was obtained from each participant (or other approved parties) for publication of this research work.

ETHICAL APPROVAL

All authors hereby declare that all experiments have been examined and approved by the appropriate ethics committee and have been conducted in accordance with the ethical standards laid down in the 1975 Declaration of Helsinki.

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DISCLAIMER

The Products used for this research are commonly and predominantly use products in our area of research and country. There is absolute no conflict of interest between the authors and producer of the products because we do now intend to use these product as an avenue for any litigation but for the advancement of knowledge. Also, the research was not founded by the producing company rather it was funded by personal efforts of the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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