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# ASSESSMENT OF SEROLOGICAL MARKERS FOR HEPATITIS B VIRUS INFECTION AMONG BLOOD DONORS IN BAUCHI METROPOLIS, BAUCHI STATE, NIGERIA

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***ABSTRACT***
***Hepatitis B virus infection is a public health problem that compromised the safety of blood donated for transfusion and therefore remains one of the transfusion transmissible infections of great concern globally. The infection is usually defined by the presence of hepatitis B surface antigen in serum or plasma. The aim of the study was to assess serological markers for hepatitis B virus infection among blood donors in Bauchi metropolis. A cross-sectional study was designed and total of 216 blood donors were enrolled for this study. ELISA was used for qualitative detection of Hepatitis B surface antigen (HBsAg) and Combo test panel was used to assess various markers for HBV infection. The overall prevalence for HBsAg and Hepatitis B core antibody (HBcAb) was found to be 10.7% and 4.6%. Other prevalence observed for various markers for HBV included; Hepatitis B surface antibody (HBsAb) 6 (2.8%), Hepatitis B envelop antigen (HBeAg) 2 (0.9%) and Hepatitis B envelop antibody (HBeAb) 3 (1.3%). The age group 26-30 years had the highest prevalence for HBsAg (4.63%) followed by the 21-25 years age group (2.78%) while in the 41-45years age group no sero-conversion was observed. The presence of antibodies against HBc is an indication of active infection and possibility of infectiousness. In conclusion, the outcomes of this study revealed a high prevalence of both hepatitis B surface antigen (HBsAg) and hepatitis B core antibody (HBcAb) in the study area. The study recommends increase in awareness campaign among the general public and improving the standard of screening and testing for Hepatitis B virus infection in the facilities***
***Keywords: Hepatitis B Virus Serological Markers, Blood Donors***

## INTRODUCTION

Hepatitis B virus (HBV) is said to be a hepatotropic virus with a double stranded DNA molecule and one of the family members of Hepadnaviridae (WHO, 2020). Hepatitis B virus is a small, circular, 3200-base pair size DNA virus which is enclosed in an icosahedral capsid compose of HBV core (HBc) and outer lipid envelop, the outer lipid envelop measures about 30-42nm in diameter (Daniel and Syria, 2018).

Hepatitis B virus can cause acute and chronic liver disease, the liver injury as a result of hepatitis B infection is believed to be caused primarily by the immune response of the host to the viral infection of the hepatocyte, the clinical presentation ranges from asymptomatic to symptomatic hepatitis and in rare instances, fulminant hepatitis which occurs in 0.1 to 0.5 percent of patient with viral hepatitis and it is believed to be caused by massive immune-mediated lysis of infected hepatocytes (WHO, 2020). The long term complications of HBV are liver cirrhosis and hepatocellular carcinoma (WHO, 2020).

Hepatitis B virus has a very compact structure that encode proteins from four overlapping genes (*S*, *C*, *P* and *X*), the *S* gene codes for Hepatitis B surface antigen (HBsAg), the *C* gene codes for nucleocapsid proteins, Hepatitis B core antigen (HBcAg) and Hepatitis B envelop antigen (HBeAg). The *P* gene codes the Deoxyribonucleic acid (DNA) polymerase, the *X* gene codes for a small, non-particulate protein that is capable of transactivating the transcription of both viral and cellular genes (Kramvis, 2014; Daniel and Syria, 2018).

There are five serologic markers that can be detected in serum following HBV infection; HBsAg which is the hallmark of HBV infection, Anti-HBs (HBsAb) which is a neutralising antibody that confers long-term immunity, Anti-HBc (HBcAb), that indicates active infection which can be acute or chronic HBeAg, and Anti-HBe (HBeAb) that indicates both infectivity and viral replication. Though, HBV infection is usually defined by the presence of HBsAg in serum or plasma, the serologic markers are used to elucidate chronic HBV infection, assessing clinical phases of the infection and monitor antiviral theraphy (Gerlich *et al.,* 2010; Song and Kim, 2016).

Sero-prevalence of HBsAg varies according to geographical regions with reported high endemicity in Sub-Saharan Africa, South East Asia, China and Amazon Basin (Schweitzer *et al*., 2015). Regions with low endemicity include Mediterranean area, Europe,

South America, North America and Australia (Schweitzer *et al.*, 2015). Nigeria falls among the African countries that are highly endemic for HBV infection (Olotu *et al.,* 2016).

Besides, unprotected sexual contact, reuse of contaminated sharp object and vertical transmission from mother to foetus, human blood transfusion is also one of the ways HBV infection can be transmitted (El-Ishaq and Liman, 2015; WHO, 2018). Hepatitis B virus infection is a public health problem that compromised the safety of blood donated for transfusion and therefore remains one of the transfusion transmissible infections of great concern (Shittu *et al.,* 2014). The aim of this study was to assess serological markers for Hepatitis B virus (HBV) infection among blood donors in Bauchi metropolis.

## MATERIALS AND METHODS

Bauchi state metropolis was selected to be the study area, where two secondary health facilities comprising of Bauchi State Specialist Hospital (BSSH) and General Hospital Tashan-Babiye (GHTB) were domicile. The study participants were mainly male blood donors between the ages of 21-45 years who attended the two Hospitals during the period of the sample collection. Ethical clearance (MOH/GEN/5/1409/1) was obtained from Bauchi State Ministry of Health Research and Ethics committee and consent was sort from the donors before sample collection.

Systematic random sampling technique was used to select consented blood donors while female blood donors and those participants who unconsent or tested positive for HCV, HIV and Syphilis were excluded. The sample size (216) was calculated based on the previous prevalence (16.6%) of Yakubu *et al.* (2016) obtained in Kebbi State.

Five milliliters of venous blood sample was collected aseptically into plain containers, allowed to clot at room temperature for 20mins and were centrifuged at 3000rpm for 20 minutes. The serum was collected in a separate plain container and stored at -20$^0$C for further analysis. Qualitative ELISA was used for detection of HBsAg in the serum according to manufacturer's instructions (Bio-Rad Laboratories). All reagents and samples were brought to room temperature (30°C) for the assay, while the incubator was brought to 37°C. 50 µL of each positive and negative controls and samples were added to appropriate wells of the microtiter plate. Another 50 µL of Anti HBs Peroxidase solution was added to each well and the plate was tapped gently. The plate was sealed using the adhesive slip and incubated at 37°C for 80 minutes. After incubation, the adhesive slip was removed and discarded. The plate was washed using the auto washer. Equal volumes of TMB substrate solutions A and B were prepared and 100 µL of the mixture was added to each well for colour development. The plate was covered with a black cover and incubated at room temperature for 30 minutes. The reaction was stopped by adding 100 µL of stop solution to each well and visually read. Yellow wells indicate a positive reaction while colorless wells indicate negative reactions. These were compared with the controls. Hepatitis B Virus Markers **(**HBsAg, HBsAb, HBeAg, HBeAb and HBcAb) were detected using HBV profile test kit (Micropoint) according to manufacturer's instructions (Micropoint). The test is base on immunochromatography, where the samples added will migrate and the antibodies or antigens bind to the corresponding molecules on the band area, thereby giving reactions as band lines.

## RESULTS

A total of 157 participants were recruited from BSSH while only 59 were enrolled from GHTB. The age of the participants ranges from 21 to 45 years with a mean of 32.6 ±7.6 years (SD). A prevalence of 10.7% (23/216) for HBsAg was observed in the participants, where 16 were from BSSH while GHTB accounted for only 7 of the subjects as presented in Table 1. The age group 26-30 years had the highest prevalence of 4.6% (10/216) for the HBsAg seropositivity (Table 1). Plate I shows the HBsAg microtitre plate with some positive (yellow wells) and negative (colourless) reactions.

Out of the 23 participants that were observed to be sero-positive to HBsAg, 10 (4.63%) were positive for the HBcAb biomarker while 6 (2.8%), 3 (1.3%) and 2 (0.9%) were positive for HBsAb, HBeAb and HBeAg biomarkers respectively as presented in Figure 1.

**Table 1: Prevalence of HBsAg among Blood Donors based on Age groups and Hospitals**

| Age groups (Years) | BSSH | | | GHTB | | | Total | | |
|---|---|---|---|---|---|---|---|---|---|
| | Number Screened | No. Positive | % | Number Screened | No. Positive | % | Number Screened | No Positive | % |
| 21-25 | 40 | 5 | 3.19 | 13 | 1 | 1.70 | 53 | 6 | 2.78 |
| 26-30 | 61 | 7 | 4.46 | 20 | 3 | 5.09 | 81 | 10 | 4.63 |
| 31-35 | 28 | 2 | 1.27 | 12 | 2 | 3.39 | 40 | 4 | 1.85 |
| 36-40 | 17 | 2 | 1.27 | 11 | 1 | 1.70 | 28 | 3 | 1.39 |
| 41-45 | 11 | 0 | 0.0 | 3 | 0 | 0.0 | 14 | 0 | 0.0 |
| **Total** | **157** | **16** | **10.2** | **59** | **7** | **11.9** | **216** | **23** | **10.7** |

Key: BSSH- Bauchi State Specialist Hospital, GHTB- General Hospital Tashan-Babiye.

**Plate I: HBsAg ELISA Microtitre Plate**

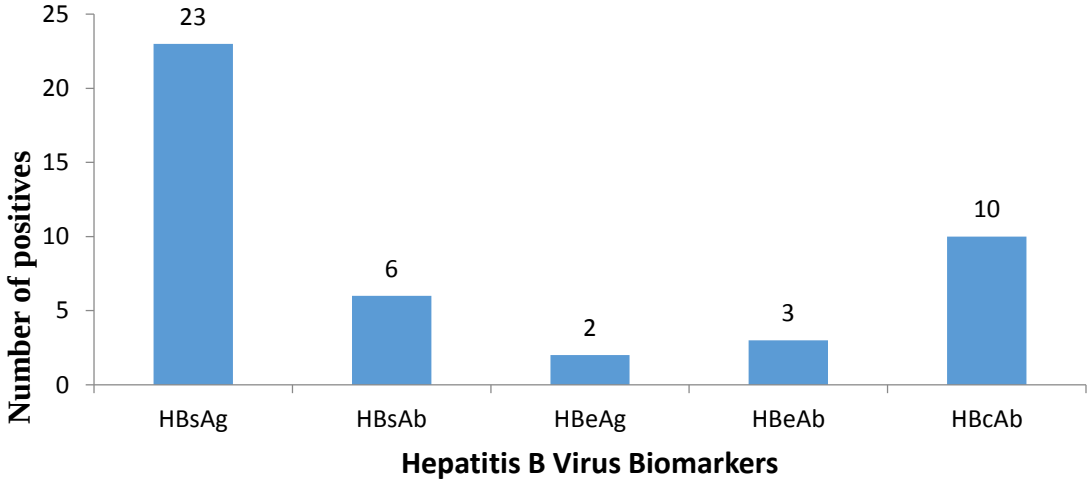


**Figure 1:** Distribution of Hepatitis B Virus biomarkers among HBV positive blood donors

**DISCUSSION**

The results for the prevalence indicated a high prevalence in the blood donors who are apparently healthy and are expected to be free of the infection. Our prevalence of 10.7% for HBsAg is closely similar to the prevalence obtained in other studies conducted in Nigeria by Aminu *et al.* (2013) in Kaduna and Mbamara and Obiechina (2010) in Onitsha who reported a prevalence of 12.5% and 12% respectively. The similarity may be due to similar geographic location and socioeconomic status of the populations involved in the study because most of the participants are from low income communities. People from such communities tend to share a lot of sharp objects such as nail cutter, scissors and hair clippers as a common practice in such locations. However, Umar *et al.* (2020) and Mustapha *et al.* (2020) reported a prevalence of 6.0% and 6.7% for HBsAg among pregnant women in Azare and Gamawa LGAs of Bauchi State. Some studies conducted on blood donors indicated prevalences of 7.5%, 8.3% and 8.8% in Kano, Makurdi and Ilorin respectively (Emmanuel *et al.,* 2012; Arun *et al.*, 2014; Olawumi *et al.,* 2016). These reports are lower than the prevalence obtained in this study and might be as a result of increased health awareness campaigns in the regions and adherence to the HBV infection preventive measures because most of the blood donors in the region are educated. On the other hand a higher prevalence of 16.6% was reported in Kebbi State during a similar study on blood donors (Yakubu *et al.,* 2016). An even higher prevalence was observed in Benue State (Alao *et al.,* 2008) and North Central region (Ndako *et al.,* 2016) with prevalence of 20.0% and 22.5% respectively among blood donors. The high prevalence might be associated with some multicultural differences that exist in the north central part of Nigeria which allow them to have cultural marks that might involve the use of unsterilized equipment such as razor blades. In the African region generally, HBV infection has been classified as highly endemic according to Schweizer *et al.* (2015) and there is variation in the outcome of most the studies conducted within the African region, some studies reported a high prevalence, while some lower prevalence.

Transfusion of blood unit collected from a donor who is in the HBV infection window period can result in post transfusion Hepatitis B virus infection in the recipient (Muhlbacher *et al.,* 2001; Hennig *et al.,* 2002). This study reported a prevalence of 5.7% for HBcAb, which indicates active infection hence the possibility of being infectious. Prevalence of 48.8% was recorded by Fasola *et al.* (2022) in Ibadan in a study to determine prevalence of hepatitis B virus infection among blood donors with total antibodies (IgG and IgM) to hepatitis B core antigen which is higher than the result recorded in this study which used IgM based Anti-HBc and concentrated on those tested positive for HBsAg only.

It is understood that only 3/23 of the HBsAg positive donors have persistence of HBeAg in blood which is always associated with progress towards a liver disease as well as an increase probability of transmitting the virus (Akazong *et al.,* 2021).

## CONCLUSION

The outcome of this study revealed a high prevalence (10.7%) of Hepatitis B surface antigen and Hepatitis B core antibody (4.63%) among the blood donors**.** While the prevalence for HBcAg, HBsAb and HBeAg were low. The study recommends increase in awareness campaign among the general public and improving the standard of screening and testing for Hepatitis B virus infection in the blood transfusion services.

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