

Farming and Non-Farming Individuals Attending Grimard Catholic Hospital, Anyigba, Kogi State, Nigeria Were Comparable in Hepatitis B Surface Antigen Seroprevalence

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Abstract: A previous study had shown that women farmers or those having farmer husbands attending Grimard Catholic Hospital, Anyigba were seronegative for hepatitis B surface antigen (HBsAg) (0.0% prevalence for HBsAg). We therefore designed this study to test the hypothesis that HBsAg seroprevalence among farming and non-farming individuals attending the hospital is the same. Two hundred apparently healthy adults attending the hospital were consecutively selected and screened for HBsAg including 106 (53.0%) females and 94 (47.0%) males aged 20 to 51 years (mean age = 35 yrs). The sera of the participants were tested for the presence of HBsAg using Diaspot® HBsAg test kit. The patients were categorized into farming and non-farming groups based on the demographic data obtained with questionnaire forms. This study showed an overall HBsAg seroprevalence of 11.0%; with seroprevalence of 17.4% (n = 46) and 9.1% (n = 154) respectively for the farming and non-farming groups. Variables (gender, age and occupation) examined were not statistically associated with prevalence rates of HBsAg. Broad categorization of the participants into farming and non-farming with their respective spouses also revealed no association (p = 0.19) with HBsAg seropositivity. Moreover, some of the women farmers and those having farmer spouses were HBsAg seropositive. We therefore concluded that farmers and non-farmers enrolled in this study were not different in HBsAg seroprevalence; they appeared equally susceptible to HBV infection. Our findings represent the endemic HBV situation in many hospitals in Nigeria. This study could serve to direct any national effort aimed toward reducing the HBV burdens of our local hospitals. The study will be of immense value as a public health tool for planning, delivery, monitoring and evaluation of HBV interventions.

Key words: Farmers, HBsAg seroprevalence, HBV infection, Nigeria, non-farmers

INTRODUCTION

Hepatitis B virus (HBV) is an ancient disease that was first discovered in the 5th century BC. Epidemics of jaundice have been identified since that time with the

earliest recognized blood-borne outbreak of hepatitis occurring in Bremen, Germany in 1983 among shipyard workers who received the smallpox vaccine. HBV was however, identified and characterized following the discovery of Australian antigen in human serum by

Blumberg (1971); the Australian antigen is designated hepatitis B surface antigen which is present in blood of both acutely and chronically infected hepatitis B patients (Patrick *et al.*, 2003).

Hepatitis literally means inflammation of the liver. It is caused by many agents, such as viruses, alcohol and drugs among others, however, the focus here is on viral hepatitis, that is, infection caused by a group of viruses that primarily infect liver cells, with accompanying damage to the liver. Hepatitis may be acute (that is, short term inflammation of the liver), or chronic (that is, long term inflammation of the liver). Different viruses cause hepatitis, but five of the viruses have been identified and are well characterized, they include; hepatitis A, B, C, D, E (Prescott *et al.*, 2005), TT (Saket *et al.*, 2005) and SEN viruses.

Hepatitis B infection is caused by HBV, a partially double-stranded circular DNA virus in the *Hepadnaviridae* family (CDC, 2008). HBV is found in highest concentrations in the blood, and lower concentrations in saliva, semen, vaginal secretions, and wound exudates. HBV can remain viable for >7 days on environmental surfaces at room temperature (CDC, 2008). Electron microscopy of serum containing HBV showed three distinct antigenic particles: a spherical 22nm particle that is hepatitis B surface antigen (HBsAg), 42 nm spherical particle containing DNA with the associated DNA polymerase called the Dane particle and the tubular or filamentous particle that vary in length. The virus consists of an outer lipid envelope containing an icosahedral nucleocapsid core composed of the viral proteins. The nucleocapsid encloses the viral DNA and a DNA polymerase that has reverse transcriptase activity. HBV is one of a few known non-retroviruses, which employ reverse transcription in its replication process. The outer envelope contains embedded proteins which are responsible for binding of the viral molecule to its cognate cellular receptor (i.e., HBV-binding factor) thereby making liver cells i.e. hepatocytes susceptible to infection by HBV (Schneider-Schaulies, 2000). The virus is generally spherical in morphology but pleomorphic forms also exist (Redd *et al.*, 2007). The DNA genome is not segmented but it contains long and short segments, which overlap approximately 240 nucleotides to form an open circle.

Following its attachment to and penetration into hepatocytes, HBV core particles migrates to the nucleus where the partially double stranded, relaxed circular genome is converted to covalently closed circular DNA (cccDNA), which forms the template for transcription of the viral DNA genome into mRNA and pregenomic RNA by cellular RNA polymerase II. Once the pregenome is transferred to the cytoplasm, the reverse-transcriptase component of HBV polymerase converts the RNA pregenome into new circular genomic DNA; the latter is

selectively package into viral capsid proteins. This can either bud via the cellular endoplasmic reticulum in order to be enveloped and subsequently released from the infected liver cell or recycled back into the nucleus for conversion to cccDNA (Alter, 2003). The viral mRNA is also transported to the cytoplasm where it is translated by cellular ribosome into various HBV polypeptides, abundant between, which is HBsAg. Infected cells into blood circulation release the latter. Hence the detection of HBsAg in human's blood is used as a veritable indicator of HBV infection (Redd *et al.*, 2007). Detection of HBsAg is therefore used for seroepidemiology of HBV infection.

The average incubation period is 90 days from time of exposure to onset of symptoms, but may vary from 6 weeks to 6 months (Mast *et al.*, 2005; CDC, 2006, 2008; ACOG, 2007). After exposure of susceptible human to infectious dose of HBV, they develop HBV infection, which may either be acute (self-limited) or chronic (long-standing). Persons with self-limited infection clear the infection spontaneously within weeks to months. Children are less likely than adult to clear the infection. More than 95% of people who become infected as adults or older children exhibit full recovery and develop protective immunity to the virus. When the infection is not cleared, the infected becomes a chronic carrier of the virus. Acutely infected individuals develop clinically apparent hepatitis with loss of appetite, nausea, vomiting, fever, abdominal pain body ache, and development of jaundice (CDC, 2006). Some may have dark urine and gray stool (ACOG, 2007). About one half of acute HBV infections in adults are symptomatic. About 1% of cases result in acute liver failure and death (CDC, 2008). The illness lasts for a few weeks and gradually improves in most people. A few patients may have more severe liver disease and may die as result of it. The infection may be entirely asymptomatic and may go undiagnosed.

Chronic infection with HBV on the other hand may be either symptomatic or associated with a chronic inflammation of liver. A sequela of long-term damage to infected liver is cirrhosis, the massive fibrosis of the liver resulting from the organ's effort to repair the damaged liver. This sequela increases the incidence of liver cancer. Co-infection with hepatitis D virus increases the risk of liver cirrhosis and subsequently liver cancer. Polyarteritis nodosa is also common in people with HBV infection (Lok, 2005). Chronic infection occurs in about 90% of infected infants, 30% of infected children aged <5 years, and 2-6% of adults (CDC, 2008). Among persons with chronic HBV infection, the risk of death from cirrhosis or hepatocellular carcinoma is 15-25% (Workowski and Berman, 2006; CDC, 2008).

In infected individuals, HBV is present in blood, sweat, tears, saliva, semen and vaginal secretions. The transmission of the virus is therefore through parenteral

routes such as needle stick injury, sharing of sharp objects, blood transfusion and vertically from infected mother to her child; it can also spread by sexual contact with infected person. Abstinence from unprotected sexual intercourse is a guaranteed way of preventing sexual transmission of HBV. Familial spread among members of a household is also possible through contact with secretions or saliva containing HBV (Alter, 2003). Sexual transmission accounts for most adult HBV infections in the United States (CDC, 2006). Approximately 25% of the regular sexual contacts of infected individuals will themselves become seropositive (ACOG, 2007). About 10-20% of women seropositive for HBsAg transmit the virus to their neonates in the absence of immunoprophylaxis (CDC, 2008). In women who are seropositive for both HBsAg and HBeAg vertical transmission is approximately 90% (ACOG, 2007). In patients with acute hepatitis B vertical transmission occurs in up to 10% of neonates when infection occurs in the first trimester and in 80-90% of neonates when acute infection occurs in the third trimester (ACOG, 2007; CDC, 2008).

The assay for detection of HBV infection involves serum or blood test that detects either viral antigens or virus-specific antibodies produced by infected host. However, humans suspected of HBV infection are frequently screened for the presence of HBsAg. This is because this antigen is the first detectable viral antigen to appear during infection with the virus. Early in infection, this antigen may not be present and it may also be undetectable later in the infection as it is cleared from blood circulation by the host immune system. The infectious virion contains an inner core particle enclosing viral genome; this particle is known as hepatitis B core antigen (HBcAg). Shortly after the appearance of HBsAg, another antigen named as hepatitis B e antigen (HBeAg) appears. The presence of HBeAg in a host serum indicates active viral replication and infectiousness of infected persons. However, some variants of HBV do not produce this 'e' antigen. During the window period when the infected successfully clears the HBsAg thereby making it undetectable, IgM antibodies to the HBcAg (i.e., anti-HBcAg IgM) may be the only serologic evidence of the infection/disease. During the natural course of HBV infection, the HBeAg may be cleared while concurrently producing antibody to the 'e' antigen (i.e., anti-HBeAg). If the host is able to clear the infection, the HBsAg becomes undetectable and antibodies to HBsAg (anti-HBs) develops (Ray and Ray, 2004). A person negative for HBsAg but positive for anti-HBs has either cleared the infection or has been previously vaccinated against HBV. A number of persons who are positive for HBsAg have very little viral multiplication and hence may be at little risk of long-term complication or of transmitting the infection to others.

In Nigeria, there is a dearth of literature on seroprevalence of HBsAg for farming and non-farming humans. A previous study done by Sule *et al.* (2007) observed zero HBsAg seroprevalence for pregnant women that were farmers and those that had farmers as their spouses. This present study therefore aimed at determining the seroprevalence of HBsAg among farming and non-farming persons attending Grimard Catholic Hospital, Anyigba, Kogi State, Nigeria with the view to determining statistical association or lack thereof between occupational farming and prevalence rates of HBsAg.

MATERIALS AND METHODS

Study area/population: Anyigba town is located in Dekina local government area of Kogi state. It has the latitude of 8°43' and 9°45' and longitude of 6°65' and 7°45'. Egume, Abejukolo and Ofakaga villages surround it. It enjoys both wet and dry climatic conditions. The annual rainfall period is between April and October; the rainfall ranges from 120-150 mm, while the dry season lasts between November and March (Ocholi, 2006). Anyigba has the total population of 18,907 according to the United National Population Commission survey of 2002 with the growing rate of 3.61%. About forty five percent of its population was farmers, while about 30% were civil servants and businessmen, with about 25% being vocational workers/students.

Study design: The study was carried out between June and November 2007. The location of the present study was Grimard Catholic Hospital, Anyigba; and the participants were also those attending the hospital. We obtained permission to carry out the study from the Management of the hospital. Subsequently, the objective and procedures of the study were explained to all consecutive adult patients visiting the hospital and consenting individuals recruited into the study. With well-structured questionnaire forms, pertinent demographic data were obtained from the participants. The data collected included age, gender, patient and their spouses' occupation. About 5 mL blood sample was aseptically collected by venipuncture from each participant into sterile plane bottle. The blood samples were left to form clots at room temperature, after which they were centrifuged for 10 min at 200 revolutions per minute (rpm) to separate serum from clot. Each blood sample was screened using immunoassay based DiaSpot® test strip for qualitative detection of HBsAg in serum (relative sensitivity and specificity of > 99% and 97.0% respectively with accuracy of 98.5%); the test and result interpretations were done according to the test kit manufacturer's instruction. In order to exclude participants' and or researchers' bias, it was after the

laboratory HBsAg screening we categorized them into the two groups - the farming and non-farming.

Data analysis: We presented the results of this study with descriptive statistics. In addition, we used CHI² statistical test to establish difference or absence thereof of HBsAg seroprevalence between farming and non-farming groups and between other categories; $p \leq 0.05$ was used as indicator of statistical significance. We used SPSS 13.0 for the analysis.

RESULTS AND DISCUSSION

Sera were obtained from blood samples of 200 adults and screened for HBsAg, the participants comprised 106 (53.0%) females and 94 (47.0%) males (Table 1) aged 20 to 51 years (mean age = 35 years). No invalid test result was observed in this study. Of the 200 subjects tested for HBsAg, 22 (11.0%) were HBsAg-positive as shown in Table 1 and 2. The Tables also reveal HBsAg seroprevalence rates for different categories. Since the study encompassed male and female adults having 11.0% seroprevalence rate, Anyigba was hence further confirmed endemic for hepatitis B virus infection (Sule *et al.*, 2007).

The overall HBsAg seroprevalence of 11.0% observed in this study is comparable to previous studies (Saillour *et al.*, 1996; Olubuyide *et al.*, 1997; Ola *et al.*, 2004; Agbaji *et al.*, 2005; Sule *et al.*, 2007). Similar prevalence was also reported by Agbaji *et al.* (2005) from Jos University Teaching Hospital (JUTH) in which they reported a prevalence of 14.8% for HBV amongst HIV positive patients. The infection rates of HBsAg in the present study among farmers and non-farmers showed that this infection is still high among Nigerians. These observations are similar to the findings in other African countries and the world in general (Saillour *et al.*, 1996).

The seroprevalence of HBsAg among the farming group was 17.4% (n = 46) while it was 9.1% for the non-farmers (n = 154); the two groups were however, comparable (p = 0.11). When the participants were re-grouped as shown in Table 2 for the purpose of comparison; the group with and without farmers had 15.5 and 9.2% HBsAg seropositivity. However, these were statistically comparable (p = 0.19). This suggested that both categories were not different in HBsAg seroprevalence. This implied that HBV infection was apparently not associated with the occupational categories.

In this study, without reference to pregnancy status, there were 18 women farmers of which only 3 (16.7%) were seropositive for HBsAg (data not shown). Fifteen women had farmer husbands of which the same 3 (20.0%) women were seropositive. In addition, in all, 4 couples were farmers, three (75.0%) wives of which were HBsAg

Table 1: HBsAg seropositivity of different groups of participants in Grimard Catholic Hospital, Anyigba, Kogi State, Nigeria, 2007

Subjects' variables	No. tested (%)	No. positive (%)	p-value
Gender			
Female	106 (53.0)	9 (8.5)	0.23
Male	94 (47.0)	13 (13.8)	
Age (years)			
20-39	132 (66.0)	14 (10.6)	0.80
40-51	68 (34.0)	8 (11.8)	
Occupation			
Farmers			0.11
Non-farmers	46 (23.0)	8 (17.4)	
	154(77.0)	14 (9.1)	

Table 2: Comparison of HBsAg seroprevalence among farming and non-farming groups attending Grimard Catholic Hospital, Anyigba, Kogi State, Nigeria, 2007

Category	No. tested (%)	No. positive (%)	p-value
Farmers and non-farmers having farmer spouses	58 (29.0)	9 (15.5)	0.19
Non-farmers having non-farmer spouses	42(71.0)	13 (9.2)	

positive. All these contrast the previous observations in the same health facility where we reported zero HBsAg prevalence for pregnant women that were farmers, as well as, for those having farmers as spouses (Sule *et al.*, 2007).

This study showed that though more male subjects than the females were seropositive, however, they were comparable (p = 0.23) in HBsAg seropositivity, Table 1. This suggested that both the male and female subjects were apparently equal in exposure to HBV. This contrasts the report that globally, male gender appeared to be higher risk of contracting HBV than female gender (Groshiede and Damme, 1996).

We also observed that there was no age difference (p = 0.80) in seropositivity for HBsAg though higher seroprevalence occurred in 20-39 years category, Table 1. While we might attribute this finding to the smaller samples size used in this study; the observation is consistent with the report of Motta-Castro *et al.* (2003) who reported no association of age with HBsAg seropositivity. In conclusion, farmers and non-farmers enrolled in this study were not different in HBsAg seroprevalence; they appeared equally susceptible to HBV infection.

Our findings represent the endemic HBV situation in many hospitals in Nigeria. This study could serve to direct any national effort aimed toward reducing the HBV burdens of our local hospitals. The study will be of immense value as a public health tool for planning, delivery, monitoring and evaluation of HBV interventions. It is recommended, that people attending Grimard Catholic Hospital, Anyigba be enlightened on the need for HBsAg screening; confirmation of the test should also be done so that appropriate drug or vaccination of true positive patients can be instituted.

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