

Sero prevalence of Hepatitis B surface antigen in patients attending a tertiary care hospital

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Abstract

Introduction: Hepatitis B virus (HBV) infection is one of the global health problems which can lead to complications like chronic hepatitis and carcinoma of liver. HBV is an emerging threat to the treating clinicians due to the risk of transmission of the virus, increased mortality rate and disease progression to fatal complications.

Aim: The aim of the present study is to determine the baseline prevalence of Hepatitis B surface antigen in patients attending a tertiary care hospital.

Materials and Method: A total of 8190 patients were included in the present study. Screening of HBsAg was done by rapid card test. Positive cases were further confirmed by both Enzyme linked Immunosorbant Assay (ELISA) and Enzyme linked fluorescent assay (ELFA) methods.

Results: Out of 8190 cases, 0.86% of the cases were diagnosed to be positive for HBsAg. Among the positive cases, 0.92% were males and 0.76% were female patients. The seropositivity was higher in the age group of 40-60 years.

Conclusion: The Seroprevalence of hepatitis B surface antigen is more in age group of 40 – 60 years with a male preponderance and is comparatively similar to the previous studies. This indicates the importance of need for strengthening the hepatitis B vaccination in the population, with a high coverage.

Keywords: Hepatitis B virus, Hepatitis B surface antigen, Seroprevalence

Introduction

Hepatitis B virus (HBV) infection is one of the serious global threats. World Health Organisation (WHO) estimates more than 2 billion people to be infected with HBV worldwide, with 240 million to develop the risk of chronic liver diseases like cirrhosis liver and hepatocellular carcinoma.⁽¹⁾ Hepatitis B is an acute inflammation of liver cells (i.e. hepatocytes). It can be caused by both infectious and non-infectious agents. The infectious agents include viruses such as hepatitis A, B, C, D and E. The non-infectious causes include drug-usage, alcoholism, chemicals and environmental toxins.⁽²⁾ There are various routes of transmission of HBV infections namely perinatal transmission during child birth, contact with infected person's blood and body fluids, tattoos, needle stick injuries, unprotected sexual contact, etc. HBV is found to be 50 to 100 time more infectious than Human Immunodeficiency Virus (HIV).⁽¹⁾

India has one-fifth of the world's population and accounts for a large proportion of the worldwide HBV burden.⁽³⁾ HBV infection stands tenth in the leading causes of deaths due to communicable diseases throughout the world.⁽⁴⁾ Around 780000 people globally and 100000 people from India die of complications of hepatitis every year.⁽¹⁾

Prevalence of Hepatitis B surface Antigen (HBsAg) among different areas of the world are classified as high (>8%), intermediate (2–7%) and low (<2%) HBV endemicity. Highly endemic areas include South-East Asia, China, most of Africa, most of Pacific Islands, the Amazon basin and parts of the Middle East.

Intermediately endemic areas are South Asia, Eastern and Southern Europe, Russia and Central and South America. Low endemic areas include United States, Western Europe and Australia.⁽³⁾ The HBV prevalence of India ranges between 3 – 4.2%.⁽¹⁾ India has around 40 million HBV carriers which contributes to 10–15% of the global HBV carriers.⁽⁵⁾ The estimated carrier rate in India is 4.7% and carrier population is 56.5 million.⁽³⁾

Every year in India, 2.6 crore infants are born and approximately 10 Lakhs have the chance of developing chronic HBV infection.⁽³⁾ The early identification of HBV-related liver disease in primary-care setting is essential for its appropriate management and identifying its basic epidemiological correlates helps in prevention and control.⁽⁴⁾ The HBsAg positivity rate has been estimated to range between 2% and 8%. Most of the studies are based on blood bank screening which may be biased and may not truly account for the national prevalence.⁽³⁾ Details about region and age-specific prevalence of hepatitis B infection play a key role for evaluating vaccination programs and national disease prevention and control efforts.⁽⁶⁾ There is some paucity in the large scale population studies to determine the prevalence of HBV burden in India. Hence this study was taken up to evaluate the seroprevalence of Hepatitis B surface antigen in the patients of different age groups attending our tertiary care hospital in Chennai.

Materials and Method

After obtaining Institutional Research and ethical committee clearance, the study was carried out at Sree Balaji Medical College and Hospital, Chennai over a

period of 4 months from November 2016 to February 2017. After obtaining informed consent, the patients were screened for the diagnosis of Hepatitis B surface antigen. Serum samples were collected from the patients using standard procedure aseptically. The samples were initially screened by commercial HBsAg rapid card test (SD BIOLINE HBsAg test) according to manufacturer's instructions. It is a rapid in vitro immunochromatographic test for qualitative detection of HBsAg. The test cassette contains a membrane strip precoated with mouse monoclonal anti- HBs capture antibody on test band region. Antibody – antigen – antibody gold particle complex forms a visible line (Test line). The control line is used for procedural control. Appearance of two colour bands (Test line and control line) is considered as positive. The sensitivity of the test is $\geq 99\%$ with specificity $\geq 98\%$. The positive cases were further confirmed by Enzyme linked Immunosorbent Assay (ELISA, HBSAg ULTRA, Bio-Rad France) and Enzyme linked fluorescent assay (ELFA, VIDAS® HBsAg, bioMérieux, France) methods.

HBsAg Ultra ELISA test is a one-step sandwich enzyme immunoassay using a solid phase coated with monoclonal antibodies. The solid phase is coated with monoclonal antibodies. The assay procedure includes distribution of red coloured conjugate into the wells followed by incubation and washing. The subsequent steps are distribution of coloured Substrate solution followed by incubation and washing, distribution of stopping solution, reading of the optical densities at 450/620-700 nm and finally the results were interpreted.

ELFA, VIDAS® HBsAg is a compact automated immunoassay utilizing Enzyme linked Fluorescent Assay phage and immuno concentration technology. The VIDAS HBsAg test is performed in the automated Mini Vidas (BioMerieux) system and the results were calculated by the computer software. The solid phase receptacle (SPR) serves as the solid phase as well as the pipetting device for the assay. The assay works with the principle that after the preliminary washing step, the antigen present in the sample will bind to the monoclonal antibody coating the interior SPR and to the antibody conjugated with biotin. The unbound sample components are washed away. The antigen bound to solid phase and to the biotinylated antibody is in contact with streptavidine conjugated with alkaline phosphatase which will bind with biotin. The conjugate enzyme catalyses the hydrolysis of the substrate (4-methylumbelliferyl phosphate) into a fluorescent product (4 – methyl umbelliferone), the intensity of which is proportional to the concentration of antigen present in the sample.

Both the ELISA and ELFA tests were carried out based on manufacturer's protocol. Standard positive and negative controls from manufacturers were used in testing each batch of samples.

Result

Out of the 8190 samples screened for HBsAg, 71 (0.86%) were diagnosed to be positive for Hepatitis B surface antigen. The prevalence of HBsAg was found to be high among the male patients (0.92%) compared with the female patients (0.76%). This has been shown in the Table 1. Also, it has been found that the prevalence of HBsAg is higher in the age group 40 – 60 years as depicted in the Table 2.

Table 1: Sex wise distribution of total cases

Patients tested	Number	Percentage
Male	5432	66.32
Female	2758	33.67

Table 2: Sex wise prevalence of positive cases HBsAg

Gender	Total number of cases tested	Number of positive HBsAg cases	% of positive cases
Male	5432	50	0.92
Female	2758	21	0.76

Table 2: Age wise distribution of positive cases of HBsAg

Age in years	Number of positive cases
0-20	4
20-40	15
40-60	46
>60	6

Discussion

The prevalence of Hepatitis B infection differs among various countries. The prevalence is found to be low in countries like North America, North Europe with high standards of living such as better sanitation, effective vaccination, safe transfusion practices and the prevalence is reported to be high in countries like South East Asia, South America with relatively low standards of living. Most people with chronic Hepatitis B or C are oblivious of their infection, putting them at the risk of developing cirrhosis or carcinoma of liver.^(3,7,8) The prevalence of HBsAg varies in diverse regions of India. This may be due to the type of study population, different geographical area, socioeconomic conditions, genetic and health factors.

WHO has estimated that more than 2 billion population have been infected with HBV. India is one of the countries which account for a high HBV burden. India stands second to China in the prevalence of Hepatitis B. The awareness of the complications of hepatitis is still lower in the general population. A meta-analysis has found that the HBV burden is high in tribal population than in nontribal people⁹. In the report on

prevention of HBV in India, WHO reported that the prevalence of HBsAg among general population ranges from 0.1% to 11.7%, but most of the studies reported between 2% and 8%.^(10,11,12)

In the present study, the seroprevalence was found to be 0.86%. It was similar to the studies from Rajasthan (0.87%) however slightly higher prevalence was documented from Vellore (1.7%) and Chennai (1.9%).^(13,14) In a study conducted by Chaudhary A, the prevalence of Hepatitis B infection in Indian population was found to be 3-4%.⁽¹⁵⁾ The highest prevalence was among the aborigines of Andaman and Arunachal Pradesh.⁽¹⁵⁾ The prevalence rate was found to be 2.5% in a study conducted at Kathmandu, Nepal.⁽¹⁶⁾

The prevalence of HBsAg was found to be high among the male patients (0.92%) as compared to the female patients (0.76%). This was similar to the study by Dutta *et al*⁽¹⁷⁾ and Sood S *et al*.⁽¹⁴⁾ The possible reason for the higher prevalence in males could be because of higher exposure to occupational HBV risk factors.^(18,19) The prevalence of HBV infection in a study conducted in Puducherry was 2.4%.⁽²⁰⁾ The same study showed a male preponderance in all age groups.⁽²⁰⁾ Most of the previous studies also show a male preponderance for HBsAg.^(21,22,23) Men are found to be more prone to remain infected with HBV and are more susceptible to chronic hepatitis. Women are said to develop anti-HBs rapidly and are found to show an enhanced immune response to HBsAg.^(24,25)

It has been found that the prevalence of HBsAg is higher in the age group 40 – 60 years and lowest among 0-20 years. This data indicates the adequate prevention of perinatally transmitted HBV infection in this locality by improved standards of living and effective immunization. This was similar to the study by Vazhavandal *et al*⁽²⁶⁾ but in contrast to the study by Singh J *et al* who reported highest incidence among 0-5 years and thereafter a progressive decline.⁽²⁷⁾

The limitations of the study are failure to observe the association of HBV infection with history of prolonged hospital stay, invasive procedures such as ear piercing, tooth extraction, and multiple sexual partners.^(28,29)

Conclusion

Large scale multi-centric epidemiological studies are needed to accurately estimate HBV burden in India, to identify the high risk areas of endemicity and to analyze the risk factors of transmission of the virus. In spite of campaigns and awareness programs on HBV vaccinations, there is higher prevalence of HBV infections in India. The routine hepatitis B vaccination programmes need to be strengthened with a high coverage. Timely initiation and strict adherence to the immunization schedule for Hepatitis B helps in prevention of the disease. It has been found that the prevalence of HBsAg is higher among the males in the age group of 40 to 60 years. It is recommended that all

the patients have to be screened for HBsAg. Further tests need to be done to screen the liver function. Since India is one of the major contributors of the global HBV carrier pool, it is very much essential to identify the high prevalence areas and focus on the public health measures to decrease the disease burden in our country.

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