e-ISSN: 0976-822X, p-ISSN:2961-6042

Available online on http://www.ijcpr.com/

International Journal of Current Pharmaceutical Review and Research 2023; 15(10); 692-695

Original Research Article

# Study to Investigate the Seroprevalence of Hepatitis B Surface Antigen (HBsAg) and Possible Risk Factors for Perinatal Hepatitis B Virus (HBV) Transmission in Pregnant Women

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**Received: 04-07-2023 Revised: 12-08-2023 / Accepted: 28-09-2023**
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**Conflict of interest: Nil**

**Abstract**

**Aim:** The aim of the present study was to investigate the seroprevalence of hepatitis B surface antigen (HBsAg) in pregnant women and possible risk factors for perinatal hepatitis B virus (HBV) transmission.

**Methods:** The study was carried out in the Department of Gastroenterology for 12 months. Women in any trimester of pregnancy with or without jaundice attending the antenatal clinic were included. After a complete general, systemic and obstetrical examination, 4 mL blood was collected after consent. Sera were tested for HBsAg using ELISA (Span Diagnostic Ltd, India). Of 2,000 women studied, 18 (0.9%) tested positive for HBsAg.

**Results:** Seroprevalence of HBsAg was found to be 0.9% (18/2,000). The highest prevalence rate was observed in the age group of 21–25 years followed by the 26–30 year age group. The difference in HBsAg prevalence rates in different age groups was not significant. Of 2 women with acute hepatitis B, 1 transmitted infection to their babies.

**Conclusion:** In conclusion, seroprevalence of HBsAg in antenatal women was found to be 0.9%. HBeAg and HBV DNA positivity was associated with a significantly higher chance of vertical transmission.

**Keywords:** Diagnosis, HBeAg, HBsAg, HBV DNA, Hepatitis B, Perinatal transmission, Prevalence, Virus



## Introduction

Hepatitis B virus infection occurs globally and constitutes a major public health problem. [1] Over 20 million people are infected globally with this virus every year and there are around 350-400 million chronic carriers of hepatitis B virus. [2] More than 1.2 million deaths occur annually from HBV related disease making HBV infection the 10th leading cause of death globally. [3,4] It has been estimated that up to 10% of the 350 million hepatitis B chronic carriers are in India. The carrier rate of hepatitis B in India may vary in the different regions and is being quoted as being 4.7%. [5,6] Hepatitis B infection leads to a wide spectrum of clinical presentations ranging from asymptomatic carrier state to acute self-limiting infections or fulminating hepatic failure, chronic hepatitis with progression to cirrhosis and hepatocellular carcinoma. [7]

The hepatitis B surface antigen (HBsAg) in serum is the first seromarker to indicate active HBV infection either acute or chronic. [8] Hepatitis B virus (HBV) infection usually goes undetected. Unawareness of an on-going infection delays the diagnosis of HBV related liver disease and favours the spread of the virus. [9] Pregnant women are a vulnerable group and there is risk of transmitting infection to the new-born if mother is infectious. Vertical transmission of infection from mother to infants is a very important route of transmission of HBV. [10] Fetal and neonatal hepatitis acquired from mother during pregnancy leads to impaired cognitive and physical development in later life of the children. [11] In the absence of immunoprophylaxis 10-20% of women seropositive for HBsAg transmit the virus to their neonates. [12] Effective strategies for reducing incidence of chronic infections include maternal screening combined with post exposure prophylaxis consisting of HBV vaccination immediately after delivery in all children born to HBsAg positive mothers ideally with immunoglobulin prophyaxis. [13] Several studies worldwide have recommended that pregnant women should be screened for hepatitis B before delivery as this offers an opportunity to prevent another generation from being chronically infected by the virus. [14]

The aim of the present study was to investigate the seroprevalence of hepatitis B surface antigen (HBsAg) in pregnant women and possible risk factors for perinatal hepatitis B virus (HBV) transmission.

## Materials and Methods

The study was carried out in the Department of Gastroenterology, PARAS, HMRI Hospital, Patna, Bihar, India for 12 months Women in any trimester of pregnancy with or without jaundice attending the antenatal clinic were included. After a complete general, systemic and obstetrical examination, 4 mL blood was collected after consent. Sera were tested for HBsAg using ELISA (Span Diagnostic Ltd, India). Of 2,000 women studied, 18 (0.9%) tested positive for HBsAg.

Women who tested HBsAg positive were enrolled in the follow up study after an informed consent. Personal history, history of risk factors, chronic hepatitis and obstetric history was obtained. At the time of admission for delivery, again a detailed history was taken, and general, systemic and obstetrical examination was done. Data on ultrasound findings, mode of delivery, indication for cesarean section if done, weight and maturity of babies at delivery, and results of neonatal physical and neurological examination were recorded. Cord blood was collected at the time of delivery and tested for HBsAg, HBeAg and HBV DNA; presence of HBV DNA in cord blood was taken as evidence of vertical transmission of HBV.

Biochemical and serological tests

One aliquot of serum was used for liver biochemical tests including bilirubin, alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (Autospan) and enzyme immunoassays for HBsAg and HBeAg (EIAgen kit; Adaltis Italia SPA, Milano, Italy). All HBsAg positive specimens were tested further for HBV DNA, anti-HBe antibody and anti-HBc IgM using Autospan Diagnostic kit as manufacturer instruction (Span Diagnostic Ltd, Surat, India). Infants born to HBsAg positive mothers received HBV vaccine (10 μg) and hepatitis B immunoglobulin (HBIG; 0.5 mL) were administered, one in each thigh, within 12 h of birth.

### HBV DNA Analysis

From blood specimens, serum was separated and stored immediately at −20°C until use. From one aliquot, total DNA was extracted using QIAamp DNA Blood Mini Kits (Qiagen, GmbH, Germany). HBV DNA was amplified using primers corresponding to S gene (5′-TCACCA TATTCTTGGGAACAAGA-3′ and 5′-TTCCTGAACTG GAGCCACCA-3′); cycling conditions were as have been reported previously.[15] Amplified PCR products were analyzed using 2% agarose gel electrophoresis and visualized under ultraviolet light.

### Statistical Analysis

Inter-group comparisons were done using Student's t test and $\chi 2$-test with and without Yates' correction as applicable; p values below 0.05 were taken as significant. All analyses were done using Statistical Program for Social Sciences (SPSS 10.0 for Windows).

## Results

**Table 1: Prevalence of HBsAg in pregnant women in different age groups**

| Age groups in years | Number of women studied | HBsAg positive |
|---|---|---|
| 20 | 122 | – |
| 21–25 | 924 | 10 |
| 26–30 | 877 | 7 |
| 31–35 | 77 | 1 |
| Total | 2,000 | 18 |

Seroprevalence of HBsAg was found to be 0.9% (18/2,000). The highest prevalence rate was observed in the age group of 21–25 years followed by the 26–30 year age group. The difference in HBsAg prevalence rates in different age groups was not significant.

**Table 2: Maternal HBV infection status and vertical transmission**

| Maternal serological status for HBV infection | Number of mothers | Number with vertical transmission |
|---|---|---|
| Acute hepatitis B (HBsAg +ve, HBeAg +ve, IgM anti-HBc +ve) | 2 | 1 |
| HBsAg +ve, HBeAg +ve chronic HBV infection | 8 | 4 |
| HBsAg +ve, HBeAg −ve, anti-HBe +ve, HBV DNA +ve | 2 | 2 |
| HBsAg +ve, HBeAg −ve, anti-HBe +ve, HBV DNA −ve | 4 | 0 |
| HBsAg alone | 2 | 1 |
| Total | 18 | 8 |

Of 2 women with acute hepatitis B, 1 transmitted infection to their babies.

## Discussion

Infection with hepatitis B virus (HBV) is a serious public health problem worldwide and a major cause of chronic hepatitis, cirrhosis, and hepatocellular carcinoma (HCC). Transmission of HBV from carrier mothers to their babies can occur during the perinatal period, and appears to be the most important factor in determining the prevalence of infection in high endemicity areas, particularly in China and Southeast Asia. Before HBV vaccine was integrated into the routine immunization program, the proportion of babies that became HBV carriers was about 10% to 30% for mothers who were HBsAg positive but HBeAg negative. However, the frequency of perinatal infection was higher, i.e. 70% to 90%, when the mother was also HBeAg positive. [16,17]

Seroprevalence of HBsAg was found to be 0.9% (18/2,000). The highest prevalence rate was observed in the age group of 21–25 years followed by the 26–30 year age group. This finding is similar to the data from a previous study (3.8% in 20–24 years, 3.4% in 25–29 years, 2.7% in 30–34 years and 1.8% above 35 years) of antenatal women. [18] However, other studies have reported an increase in seropositivity with increasing age of antenatal women. [19,20] We also found a higher frequency of HBsAg positivity in multigravida women. The difference in HBsAg prevalence rates in different age groups was not significant. Of 2 women with acute hepatitis B, 1 transmitted infection to their babies. Rate of HBV infection to baby was higher if mother had acute hepatitis in third trimester as compared to those that suffered in 2nd trimester. Similar finding was also shown by Reinus et al [21] who showed transmission rate of 80% to 90% in infants, if mother suffered from acute hepatitis B during 3rd trimester of pregnancy.

The USPSTF found convincing evidence that universal prenatal screening for HBV infection substantially reduces perinatal transmission of HBV and the subsequent development of chronic HBV infection. They also found no published studies that describe harms of screening for HBV infection in pregnant women. [22] In order to prevent perinatal transmission and spread of infection within the larger community, pregnant mothers should be screened for hepatitis B. Neonates who are infected by hepatitis B will have an almost 90% risk of developing chronic hepatitis B surface antigen carriage and chronic liver disease. [23]

## Conclusion

In conclusion, seroprevalence of HBsAg in antenatal women was found to be 0.9%. HBeAg and HBV DNA positivity was associated with a significantly higher chance of vertical transmission.

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