MMWR
Weekly
June 10, 1988 / 37(22);341-6,351



# Recommendations of the Immunization Practices Advisory Committee Prevention of Perinatal Transmission of Hepatitis B Virus: Prenatal Screening of all Pregnant Women for Hepatitis B Surface Antigen

Transmission of hepatitis B virus (HBV) from mother to infant during the perinatal period represents one of the most efficient modes of HBV infection and often leads to severe long-term sequelae. Infants born to mothers positive for hepatitis B surface antigen (HBsAg) and hepatitis B "e" antigen (HBeAg) have a 70%-90% chance of acquiring perinatal HBV infection, and 85%-90% of infected infants will become chronic HBV carriers (1,2). It has been estimated that more than 25% of these carriers will die from primary hepatocellular carcinoma or cirrhosis of the liver (3). These deaths usually occur during adulthood, when familial and financial responsibilities make them particularly devastating. In the United States, an estimated 16,500 births occur to HBsAg-positive women each year (about 4,300 of whom are also HBeAg- positive), and approximately 3,500 of these infants become chronic HBV carriers. Prenatal screening of all pregnant women would identify those who are HBsAg- positive and thus would allow treatment of their newborns with hepatitis B immune globulin (HBIG) and hepatitis B (HB) vaccine, a regimen that is 85%-95% effective in preventing the development of the HBV chronic carrier state (2,4-6).

In 1984, the Immunization Practices Advisory Committee (ACIP) recommended that pregnant women in certain groups at high risk for HBV infection be screened for HBsAg during a prenatal visit and, if found to be HBsAg-positive, that their newborns receive HBIG and HB vaccine at birth (7). No data are available regarding the proportion of high-risk women currently being screened in clinical practice, but several studies and the experience of public health workers indicate that major problems have been encountered

MMWR

Recommendations and Reports

December 23, 2005 / 54(RR16);1-23

# A Comprehensive Immunization Strategy to Eliminate Transmission of Hepatitis B Virus Infection in the United States

## Recommendations of the Advisory Committee on Immunization Practices (ACIP) Part 1: Immunization of Infants, Children, and Adolescents

*Please note: Errata have been published for this article. To view the errata, please click here and here.*

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## *Summary*

*This report is the first of a two-part statement from the Advisory Committee on Immunization Practices (ACIP) that updates the strategy to eliminate hepatitis B virus (HBV) transmission in the United States. The report provides updated recommendations to improve prevention of perinatal and early childhood HBV transmission, including implementation of universal infant vaccination beginning at birth, and to increase vaccine coverage among previously unvaccinated children and adolescents. Strategies to enhance implementation of the recommendations include 1) establishing standing orders for administration of hepatitis B vaccination beginning at birth; 2) instituting delivery hospital policies and procedures and case management programs to improve identification of and administration of immunoprophylaxis to infants born to mothers who are hepatitis B surface antigen (HBsAg)*

# Perinatal transmission of hepatitis B virus: an Australian experience

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## ABSTRACT

**Objective:** To determine the rate of perinatal hepatitis B virus (HBV) transmission in an Australian setting and to identify maternal virological factors associated with highest risk of transmission.

**Design, participants and setting:** A prospective, observational study of perinatal transmission of HBV. Participants were pregnant women attending Sydney South West Area Health Service antenatal clinics who tested positive for hepatitis B surface antigen (HBsAg), and their babies. All babies were routinely offered hepatitis B immunoglobulin (HBIG) and HBV vaccination. Babies positive for HBsAg at 9-month follow-up underwent further virological testing, including HBV DNA sequencing. The study was conducted between August 2002 and May 2008.

**Main outcome measures:** HBV DNA levels and demographic characteristics of HBsAg-positive pregnant women; proportion of their infants with active HBV infection at 9-month follow-up; maternal characteristics affecting transmission rate; HBV DNA sequencing of infected infants and their mothers.

**Results:** Of 313 HBsAg-positive pregnant women, 213 (68%) were HBV DNA-positive and 92 (29%) were positive for hepatitis B "e" antigen (HBeAg); 138 babies born to HBV DNA-positive mothers were tested for HBV infection (HBsAg positivity) at about 9 months of age. Four cases of transmission were identified. All four mothers had very high HBV DNA levels ($> 10^8$ copies/mL) and were HBeAg-positive. Three of the four infants were infected with wild-type HBV strains, with identical maternal/infant isolates. The fourth mother–infant pair had an S gene variant, HBV D144E, which has been previously reported in association with vaccine/HBIG escape. (Unfortunately, HBIG was inadvertently omitted from the immunisation schedule of this infant.) Transmission rates were 4/138 (3%) from HBV DNA-positive mothers overall, 4/61 (7%) from HBeAg-positive mothers, and 4/47 (9%) from mothers with very high HBV DNA levels. No transmission was seen in 91 babies of mothers with HBV DNA levels $< 10^8$ copies/mL.

**Conclusion:** In this cohort, HBV perinatal transmission was restricted to HBeAg-positive mothers with very high viral loads.

MJA 2009; 190: 489–492

Perinatal transmission is the predominant mode of hepatitis B virus (HBV) transmission in areas of high disease prevalence, and still occurs despite immunoprophylaxis with hepatitis B immunoglobulin (HBIG) (passive immunisation) and infant HBV vaccination (active immunisation). Reported rates of transmission from mothers who are positive for hepatitis B "e" antigen (HBeAg) vary from 7%[1] to 28%.[2,3] Several studies have implicated high maternal viraemia as the most important factor associated with failure of neonatal vaccination.[4]

In Australia there is a low prevalence of HBV infection (0.49%–0.87%),[5] the majority of cases being among immigrants from North and South-East Asia. Perinatal transmission in Australia has not been reported and is not routinely monitored. The *Australian immunisation handbook*[6] has recently recommended that infants born to mothers positive for hepatitis B surface antigen (HBsAg) be screened for HBsAg and antibodies to HBsAg (anti-HBs) after completion of vaccination.

The aim of our study was to determine the rate of perinatal HBV infection from a cohort of HBsAg-positive, HBV DNA-positive women in Australia.

## METHODS

### Study population

From August 2002 to May 2008, 313 asymptomatic, HBsAg-positive pregnant women from Sydney South West Area Health Service antenatal clinics were assessed in the Liverpool Hospital hepatitis clinic. Their initial clinical and biochemical assessment included tests for liver enzymes, HBV DNA and HBeAg, and hepatitis C and HIV serology. The study was approved by the Sydney South West Area Health Service ethics committee, and all participants gave informed consent for participation.

Passive and active immunoprophylaxis was given to babies according to the Australian HBV vaccination schedule.[6] Within 12 hours of delivery, infants were given 100 IU HBIG by intramuscular injection (human hepatitis B immunoglobulin-VF; CSL Bioplasma) and a dose of hepatitis B vaccine (either H-B-VAX II [thiomersal-free, 5 µg recombinant HBsAg protein; CSL Biotherapies/Merck Sharp & Dohme] or ENGERIX-B [10 µg recombinant HBsAg protein; GlaxoSmithKline]). Vaccination was completed with doses at 2, 4 and 6 months of age. Completion of the vaccination schedule was assessed for each infant using child health records.

Infants born to women with detectable HBV DNA were tested at about 9 months of age for HBsAg, anti-HBs and antibodies to hepatitis B core antigen (anti-HBc) (total anti-HBc and, if positive, anti-HBc IgM, if adequate serum was available) to determine rates of perinatal transmission. HBsAg-positive infants were further assessed for HBV DNA and viral sequencing.

### Laboratory methods

Before November 2006, HBV serology was performed using the AxSYM microparticle enzyme immunoassay (Abbott Laboratories). Subsequently, serology testing was

**Abbreviations**

| | |
|---|---|
| Anti-HBc | Antibodies to hepatitis B core antigen |
| Anti-HBs | Antibodies to hepatitis B surface antigen |
| HBeAg | Hepatitis B "e" antigen |
| HBsAg | Hepatitis B surface antigen |
| HBIG | Hepatitis B immunoglobulin |
| HBV | Hepatitis B virus |

# Outcome of Perinatal Hepatitis B Virus Exposure Is Dependent on Maternal Virus Load

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To evaluate the role of maternal hepatitis B virus (HBV) DNA levels in perinatal infection, two nested case-control studies were done within a cohort of 773 hepatitis B surface antigen (HBsAg)–positive Taiwanese women and their infants. As serum HBV DNA levels increased from <0.005 to ≥1.4 ng/mL among the hepatitis B e antigen (HBeAg)–positive mothers, the odds ratio (OR) for having a persistently infected infant increased from 1.0 to 147.0 ($P$ for trend < .001). Among HBeAg-negative mothers, the OR for having a persistently infected infant was 19.2 (95% confidence interval, 2.3–176.6) in mothers with high versus low levels of serum HBV DNA. A logistic regression analysis identified maternal HBV DNA to be a stronger independent predictor of persistent infection than HBeAg status. Thus, perinatal exposure to high levels of maternal HBV DNA is the most important determinant of infection outcome in the infant.

Perinatal transmission of hepatitis B virus (HBV) by asymptomatic carrier mothers remains a major medical problem in many areas of the world (e.g., Asia, sub-Saharan Africa) [1]. In fact, the majority of infants infected with HBV in the perinatal period become chronic HBV carriers [2, 3]. The sequelae of chronic HBV infection include chronic hepatitis, cirrhosis, and hepatocellular carcinoma. To eliminate HBV infection, the World Health Organization, the Advisory Committee on Immunization Practices, and the American Academy of Pediatrics have recently recommended universal childhood immunization against HBV (for review see [4]).

The presence of hepatitis B e antigen (HBeAg) in maternal serum is associated with a high risk of HBV transmission [2, 3, 5]. In fact, >70% of infants born to HBeAg-positive mothers become chronically infected with the virus unless they are treated prophylactically [2, 6]. However, a number of studies have found a discordance between maternal HBeAg status and perinatal HBV transmission, indicating that HBeAg is not always a reliable marker of potential infectivity [7–9]. Recently, two studies suggested that HBV DNA may be a more precise determinant for predicting persistent infection in infants, although the numbers of subjects in those studies were small [10, 11]. In the present investigation, we did two nested case-control studies in a large group of patients in which the HBeAg status of the mother incorrectly predicted the outcome of infection in her infant. We sought to determine whether HBV DNA in the maternal circulation correlated with perinatal transmission and subsequent development of chronic infection.

## Subjects and Methods

*Background.* To evaluate the role of maternal HBV DNA to predict postnatal HBV infection in infants, stored prenatal sera were quantitatively tested for HBV DNA. We did a nested case-control evaluation of mother-infant pairs who had been participants in several large cohort studies of HBV vertical transmission in Taipei, Taiwan, between 1972 and 1980, before the widespread use of immunoprophylaxis with hepatitis B immune globulin or with vaccine [5, 12, 13]. In those cohorts, women attending routine prenatal clinics at several large public hospitals were tested for hepatitis B surface antigen (HBsAg). Testing typically occurred during the first visit, which usually took place within the first or second trimester of pregnancy. Those found positive were asked to join the study and were tested for HBeAg and antibody to HBeAg (anti-HBe). The infants of mothers who agreed to participate in the study were tested for HBsAg at birth, at 3-month intervals until the child reached 24 months of age, and annually thereafter [13]. All participating mothers found to be HBsAg-positive during the prenatal period were retested after the birth of the child to confirm their carrier status. Mothers whose infants received immunoprophylaxis against HBV were not included in this study.

The population of this cohort consisted of 773 chronic HBsAg-positive mothers and their infants. As shown in table 1,

Received 11 April 1994; revised 28 July 1994.
Informed consent was obtained from all subjects.
Grant support: National Institutes of Health; American Cancer Society (Faculty Award to R.D.B.).
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**The Journal of Infectious Diseases 1994;170:1418–23**

0022-1899/94/7006-0009$01.00

Scand J Infect Dis 35: 814–819, 2003

Taylor & Francis
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# Hepatitis B Virus DNA during Pregnancy and Post Partum: Aspects on Vertical Transmission

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**Little is known about how pregnancy influences viremia levels in women with chronic hepatitis B virus infection. In this study, we first retrospectively analysed changes in HBV DNA levels during and after 55 pregnancies in HBsAg-positive women, of whom 9 were HBeAg-positive. Secondly, HBV DNA levels in 3 HBeAg-positive mothers whose babies became chronic HBV carriers, were compared with levels in 18 mothers whose babies were not infected by HBV. We found that HBV DNA ranged from $10^{8.1}$ to $10^{9.5}$ copies/mL in HBeAg-positive, and from undetectable ( $< 100$) to$10^{6.8}$ copies/mL in HBeAg-negative mothers. HBV DNA increased by a mean of 0.4 log late in pregnancy or early post partum; in 4 out of 16 HBeAg negative mothers by $> 1$ log during pregnancy. Post partum ALT increased in both HBeAg-positive and negative women. HBV DNA was $10^{9.4}$–$10^{10.4}$ copies/mL in 3 HBeAg-positive mothers whose babies were, as compared to $< 100$–$10^{10.4}$ copies/mL in 18 whose babies were not, vertically infected. Although the majority of HBeAg-negative women had low and relatively stable HBV DNA during pregnancy, viremia was also relatively high in some HBeAg-negative mothers, and both viremia and ALT increased significantly late in pregnancy or shortly after delivery. Vertical transmission was only seen in HBeAg-positive mothers with very high levels of viremia. The value of measuring HBV DNA in the pregnant woman to modify immunoprophylaxis to her infant needs further study.**

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## INTRODUCTION

The impact on HBV DNA levels of immunosuppressive hormones produced during pregnancy is largely unknown. Post partum subsidence of hepatitis B viral replication has been described in HBeAg-positive carrier mothers as an effect of immune restoration (1). It has, however, been shown that HCV RNA levels increase during pregnancy in chronic hepatitis C (2). Similar effects in chronic hepatitis B might result in increased HBV DNA levels and a higher risk of transmission during delivery.

The incidence of perinatal transmission of hepatitis B virus (HBV) from mother to child is related to the HBeAg/anti-HBe status of the mother. If immunoprophylaxis is not given, 90% of babies born to HBeAg-positive mothers will become infected, in contrast to only 10–20% of infants born to HBeAg-negative/anti-HBe-positive mothers (3). HBV infection in early life is particularly important because it most often results in a chronic carrier state. Passive-active immunisation with hepatitis B immune globulin (HBIG) and hepatitis B vaccine has proved to be highly effective in preventing perinatal transmission of HBV (4, 5). Nevertheless, 1–2% of properly vaccinated infants born to HBsAg-positive mothers become HBsAg-positive and another 1–2% of the infants do not develop long-lasting immunity against HBV (6). Among children born to HBeAg-positive mothers, even higher rates of transmission in spite of immunoprophylaxis are seen (7).

In Sweden, universal hepatitis B vaccination has not been implemented and pregnant women born outside Scandinavia or with special risk factors are screened for HBV infection during pregnancy. The immunoprophylaxis regimens for infants born to HBsAg-positive women in our region are based on the HBeAg status of the mother. Infants born to HBeAg-positive mothers are given both HBIG and HBsAg vaccine; those with HBeAg-negative mothers receive vaccine only. However, some HBeAg-negative carriers have HBV DNA at levels similar to those in HBeAg-positive carriers (8). Furthermore, some authors have found that HBeAg-positive mothers with particularly high HBV DNA levels run an even higher risk of transmission and failure of neonatal hepatitis B vaccination (5, 9–11).

The aim of this study was to investigate the HBV DNA levels during pregnancy in HBsAg-positive women using a highly sensitive quantitative PCR. We also compared viremia levels in these pregnancies with those seen in 3 cases of vertical transmission of HBV in spite of immunoprophylaxis.

## PATIENTS AND METHODS

### *Patients and samples*

Serum samples from HBsAg-positive women who had postnatal care at the Department of Infectious Diseases during 1998, 1999 and 2000 were retrospectively included and analysed (group I), if at least 2 stored samples were available from pregnancy and a follow-up period of 1 y after delivery. Samples from previous pregnancies ($n = 17$) of these women were also studied. The times of sampling are shown in Fig. 1. The blood samples drawn during pregnancy were divided into those drawn early ( $> 120$ d before partus) and late ( $<$ 90 d before partus) in pregnancy. If more than one sample was obtained during a period, the mean value was used in the analysis. Altogether 55 pregnancies in 33 mothers were analysed; 4 women had had 3 pregnancies, 14 women had had 2 pregnancies and 15 women had had 1 pregnancy. The children of HBeAg-positive mothers received immunoglobulin (250 IU, Aunativ, Biovitrum,

This study was supported by grants from the Swedish Medical Society, the Swedish Medical Research Council and Magn. Bergwall foundation.

 ISSN 0036-5548

DOI: 10.1080/00365540310016547

Journal of Medical Virology 29:1–6 (1989)

# Postpartum Subsidence of Hepatitis B Viral Replication in HBeAg-Positive Carrier Mothers

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To elucidate the effects of pregnancy and delivery on hepatitis B e antigen (HBeAg)-positive carrier mothers, 31 HBeAg-positive carrier mothers were followed-up postpartum 1 year, with 30 HBeAg-positive nonpregnant female carriers as controls. Serum hepatitis B surface antigen (HBsAg), HBeAg titer, and hepatitis B virus (HBV)-DNA concentration were studied at defined intervals. The results revealed that in the control group HBeAg titers and HBV-DNA concentrations fluctuated, whereas the HBsAg titers showed little change, but HBeAg clearance or seroconversion to anti-HBe were not noted on follow-up. In contrast, one carrier mother seroconverted to anti-HBe during pregnancy and the antibody persisted thereafter. Five of the remaining 30 carrier mothers cleared HBeAg postpartum, and among these five cases, one also seroconverted to anti-HBe. In addition, in another five of the 30 cases, the HBV-DNA fell to undetectable level (< 0.04 ng/ml). All these ten cases had a common tendency of showing a decrease in HBeAg titers and/or HBV-DNA concentrations 1–2 months after delivery. The HBeAg titers and HBV-DNA concentrations in the other 11 cases remained unchanged, whereas the remaining nine cases had increased levels. It is concluded that subsidence of HBV replication is precipitated by delivery in one-third of HBeAg-positive carrier mothers in Taiwan, and this occurs most frequently 1–2 months postpartum.

**KEY WORDS:** hepatitis B virus, HBV-DNA, asymptomatic

## INTRODUCTION

The control of hepatitis B virus (HBV) infection in the world is an urgent problem [Szmuness, 1978; Sampliner, 1985; Chen and Sung, 1987; Zuckerman 1988], and this can be achieved in two ways: first, by prevention of perinatal transmission by immunoprophylaxis in the newborns of hepatitis B surface antigen (HBsAg) carrier mothers [Beasley et al., 1983; Wong et al., 1984; Chen and Sung, 1987; Chen et al., 1987]; and second, by reducing horizontal infection of HBV, such as reduction of infectivity of hepatitis B e antigen (HBeAg)-positive carriers, screening of blood donors, and education.

During pregnancy, there is increased production of certain hormones, including adrenal corticosteroids, estrogen, and progesterone, which have immunosuppressive properties. After delivery, these hormones decrease rapidly and the immunosuppresion is then removed. Whether these effects will affect HBV carrier women is uncertain. There are only a few reports that some HBeAg-positive carrier mothers cleared HBeAg or seroconvered to anti-HBe postpartum [Tagawa et al., 1987; Tashima et al., 1987]. However, the relationship between HBeAg/anti-HBe and HBV is indirect and the dynamic changes of HBV status exerted by pregnancy and delivery were not elucidated. We therefore carried out a prospective study, investigating serial HBsAg titers, HBeAg titers, and HBV-DNA concentrations to monitor HBV activity in HBeAg-positive carrier mothers during pregnancy and postpartum.

## MATERIALS AND METHODS

Between August 1986 and July 1988, 30 asymptomatic HBsAg carriers from volunteer blood donors of Taipei Blood Donation Center were recruited. All were nonpregnant, nullipara women of reproductive age and were HBeAg-positive (designated as group A); also recruited were 31 cases of HBeAg-positive carrier mothers who received prenatal care at the Departments of Obstetrics and Gynecology of National Taiwan Univer-

Accepted for publication April 27, 1989.

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Journal of Medical Virology 44:13–21 (1994)

# Novel Application of a Point Mutation Assay: Evidence for Transmission of Hepatitis B Viruses With Precore Mutations and Their Detection in Infants With Fulminant Hepatitis B

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Mutations of the precore region of hepatitis B virus (HBV) genome have been associated with fulminant and severe chronic hepatitis. However uncertainty remains about the clinical significance and transmissibility of these mutant strains. A point mutation assay (PMA) was developed to identify qualitatively and quantitatively mutations affecting precore amino acids 1 and 28. We have analysed serum samples from six mother-infant pairs where perinatal transmission of HBV has occurred and where the mothers were HBV carriers without detectable serum HBeAg. In three cases fulminant hepatitis developed in the infant, in two cases acute hepatitis resolved, and in one case the infant was immunised and did not become infected. We also examined serum from a healthcare worker, an anti-HBe-seropositive HBV carrier, believed to have transmitted HBV infection to a patient.

The PMA results were confirmed in all cases by direct sequencing of polymerase chain reaction (PCR) products using nested and double-nested PCR with primers to the precore and X region. Precore aa28 mutant-type virus was detected in the serum of one mother at the time of delivery of three of her children, two of whom developed fulminant hepatitis. Another mother of an infant with fulminant hepatitis had no precore mutations. In one mother-infant pair a mixed viral population was found; the acute hepatitis B in the infant resolved. The HBV sequence from the healthcare worker was also of aa28 mutant type. No mutations of aa1 were detected in any of the specimens.

The study supports the association of precore mutations with some cases of transmission of HBV infection from HBeAg-negative mothers to their infants. Precore mutations may also be associated with fulminant hepatitis B in infants. Transmission of HBV infection from an HBeAg-negative healthcare worker may be associated with HBV precore mutation.



**KEY WORDS:** polymerase chain reaction, sequencing, immunisation, healthcare worker

## INTRODUCTION

The precore and core genes of the hepatitis B virus (HBV) genome encode the viral nucleocapsid or core protein. Posttranslational modification of the precore-core protein produces the soluble e antigen (HBeAg) [Ou et al., 1986; Uy et al., 1986] while the presence of this antigen in serum is associated with active viral replication [Hoofnagle et al., 1987]. Loss of serum HBeAg and seroconversion to anti-HBe is associated with reduced viral replication and a consequent decrease in infectivity.

In some carriers a high level of viral replication, indicated by the concentration of serum HBV-DNA, is present despite anti-HBe seropositivity [Lok et al., 1984; Bonino et al., 1986]. Carman et al. [1989] have studied such patients and found a sequence variation in the precore region at amino acid 28 with substitution of a stop codon for a tryptophan residue. This prevents transcription of the precore region and subsequent translation of the HBeAg. By site-directed mutagenesis, Ulrich et al. [1990] have shown that HBeAg translation is not necessary for HBV replication, a finding

Accepted for publication November 19, 1993.

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