Clinical Significance of Hepatitis B Surface Antigen in Cord Blood of Hepatitis B e-Antigen-Negative Chronic Hepatitis B Virus-Infected Mothers

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Key Words

Hepatitis B surface antigen · Cord blood · Hepatitis B virus · Vertical transmission

Abstract

Vertical transmission of hepatitis B virus (HBV) infection during the perinatal period is the major cause of HBV transmission. The aim of our study was to evaluate the serological and virological profiles of HBV infection in cord blood samples obtained from HBeAg-negative chronic HBV-infected women, at delivery, and to investigate their relationship with the clinical outcome (possible transmission of HBV) in neonates receiving the currently approved passive-active immunoprophylaxis schedule. Sixteen women (32%) exhibited HBsAg positivity in the cord blood but HBV-DNA has not been detected in any of the 50 cord blood samples evaluated. We conclude that HBsAg can be transferred through the placental barrier, as with other proteins, in about one third of HBeAg-negative chronic HBV-infected pregnant women, irrespective of the maternal viral load, the mode of delivery or the placenta HBV pathology. The clinical impact of this phenomenon on the intrauterine-transplacental or perinatal transmission of HBV infection and/or passive-active immunoprophylaxis failure does not seem to be important.

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Vertical transmission of hepatitis B virus (HBV) infection during the perinatal period is the major cause of HBV transmission in endemic countries [1]. In countries with high HBV prevalence, intrauterine/transplacental transmission is observed in a significant proportion of pregnancies, resulting in passive-active immunoprophylaxis failure and dissemination of the HBV infection [2, 3].

In Greece, the overall seroprevalence of HBsAg positivity among women of reproductive age is relatively low, but is higher among immigrants who live and work in our country [4]. The vast majority of HBsAg-positive women in our country (>95%) are HBeAg negative [5]. Despite that, about a third of them exhibit high or even extremely high viral load during the perinatal period [5], mainly due to a precore mutation (G1896A) of the HBV genome. Data concerning the serological and virological profiles of cord blood, as well as of the placental pathology for HBV antigens, from HBeAg-negative chronic HBV-infected women in Europe are limited.

The aim of our study was to evaluate the serological and virological profiles of HBV infection in cord blood samples obtained from HBeAg-negative chronic HBV-infected women at delivery, and to investigate their relationship with the clinical outcome (possible transmission of HBV) in neonates receiving the currently approved passive-active immunoprophylaxis schedule.

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Fig. 1. HBeAg-positive stained placental tissue (**a**, **b**, the arrows show brown-colored positive staining), HBeAg-negative stained placental tissue (**c**) and HBeAg-negative stained placental tissue (**d**). In 3- μ m formalin-fixed and paraffin-embedded sections, the presence of HB surface and HB core antigens were studied using the Envision HRP immunohistochemical method. The anti-HBs

Ab was mouse anti-human monoclonal (clone 3E7) and was used at a dilution of 1:50. The anti-HB core antigen was rabbit-polyclonal and was used at a dilution of 1:500. A piece of liver biopsy tissue from a patient with chronic hepatitis B was used as control.

All pregnant women with chronic hepatitis B who had delivered between July 2005 and December 2006 at the First Department of Obstetrics and Gynecology of Athens University were evaluated for HBV serologically and virologically. Cord blood was obtained at the time of delivery and was evaluated serologically and virologically; serum samples were collected from all the neonates (at the first day of life, before the initiation of the immunoprophylaxis schedule, and at the third day of life) for virological evaluation. Moreover, immunohistochemical staining of the placental tissues, obtained after the delivery, for HBsAg and HBeAg was performed using the Envision HRP immunohistochemical method. HBeAg-positive chronic HBV-infected pregnant women as well as women coinfecting with HIV/HBV, HCV/HBV or HBV/

HDV were excluded from the study. Serological re-evaluation of the infants was performed at the age of 12 months. Transmission of HBV infection to neonates was confirmed by detectable HBV-DNA in their serum and/or serum HBsAg positivity at the age of 12 months.

During the study period, 72 chronically HBV-infected women had delivered at our department and 50 of them were eligible for the final analysis. Twenty-two (44%) had caesarean sections and 28 (56%) had natural deliveries. Twenty-two women (44%) exhibited detectable serum HBV-DNA, while in 28 (56%), serum HBV-DNA was undetectable (<400 copies/ml, Amplicor/Roche) at the perinatal period. Four of 22 (18.2%) HBV-DNA-positive women had high viral load, defined as HBV-DNA >10,000 copies/ml.

Table 1. Virological and serological data obtained from serum and cord blood samples of pregnant women and data obtained from placenta staining for HBV antigens

Women's serum (n = 50)	
HBV-DNA positive	22/50 (44)
HBV-DNA negative	28/50 (56)
HBV-DNA >10,000 c/ml	4/22 (18.2)
Cord blood (n = 50)	
HBsAg positive	16/50 (32)
antiHBc positive	50/50 (100)
antiHBe positive	50/50 (100)
HBV-DNA positive	0/50
Placenta immunopathology (n = 25)	
HBsAg positive	0/25
HBcAg positive	8/25 (32)

Numbers in parentheses denote percent values.

Sixteen women (32%) exhibited HBsAg positivity in the cord blood, whereas all the cord blood samples evaluated were HBeAg negative, anti-HBe positive, anti-HBc positive and anti-HBs negative. The statistical analysis did not reveal any correlation between the maternal HBV viremia and the presence of HBsAg in the cord blood ($p = 0.761$). It is important to note that HBV-DNA have not been detected in any of the 50 cord blood samples evaluated. Virological and serological data obtained from pregnant women and cord blood samples are summarized in table 1.

Twenty-five specimens of placental tissue were evaluated for the presence of HBsAg and/or HBcAg. In all of them, staining for HBsAg was negative, whereas 8 (32%) were weakly positive for HBcAg (table 1, fig. 1), irrespective of the presence or absence of viremia or the maternal viral load.

The presence of HBsAg in the cord blood of chronic HBV-infected women was not linked to the mode of delivery in the whole group ($p = 0.559$) or in the subgroups of viremic women with high or low serum HBV-DNA levels ($p = 0.675$). Moreover, the mode of delivery did not seem to have an impact on the placenta staining for HBV antigens.

All 50 neonates evaluated at days 1 and 3 after birth were HBV-DNA negative, and HBV transmission was not observed in any of them. It is important to note that 2/4 (50%) infants born from HBeAg-positive mothers (who were excluded from the final analysis) exhibited passive-active immunoprophylaxis failure and finally acquired HBV infection (HBsAg and HBV-DNA positivity at the age of 12 months).

In conclusion, HBsAg can be transferred through the placental barrier, as with other proteins, in about one third of HBeAg-negative chronic HBV-infected pregnant women, irrespective of the maternal viral load, the mode of delivery or the placenta HBV pathology. The clinical impact of this phenomenon on the intrauterine-transplacental or perinatal transmission of HBV infection and/or passive-active immunoprophylaxis failure does not seem to be important.

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