

Interrupting the transmission of hepatitis B virus from mothers with both positive HBsAg and HBeAg to infants

Qi-Rong Zhu, Hui Yu, Hui Chen, Zuo-Quan Dong, Lin-E Fei, Xin-Huan Gu and Xin-Zhen Zhang

Shanghai, China

Background: Although hepatitis B vaccine has been highly effective and passive-active immunoprophylaxis has been used, 20%-30% babies whose mothers are hepatitis B virus (HBV) carriers may fail with vaccination against HBV. Since intrauterine HBV infection is responsible for most failures of immunoprophylaxis, this study was focused on combined antepartum and postpartum immunoprophylaxis for interruption of HBV transmission from mothers with both positive HBsAg and HBeAg to their infants.

Methods: One hundred and four pregnant women were HBsAg carriers with HBeAg positive. They were randomly divided into HBV specific immunoglobulin (HBIG) group and control group after informed consent was obtained and the study design was approved by the institutional ethics committee. The HBIG group received 400 IU HBIG at months 3, 2, 1 before delivery, whereas the control group did not. A total of 105 neonates (including twins) in the two groups were given a dose of 200 IU HBIG at birth and 2 weeks after birth, followed by 3 doses of Hepatitis B vaccine at 1, 2 and 7 months of age. A series of blood specimens obtained from the neonates at birth and 1, 2, and 7 months of age were tested for HBsAg, HBeAg, HBV-DNA, and anti-HBs.

Results: In the HBIG group, 3 of 51 neonates were infected by HBV at birth, which was found to be persistent for one year. The average titers of anti-HBs in 47 neonates at 1 month and 48 neonates at 12

months were 46 ± 9.7 and 36 ± 15.1 , respectively. In the control group, 12 of 54 neonates were infected by HBV at birth. Ten of the 12 HBV infected neonates were found to be persistent for 4 months and 9 for 12 months. The average titers of anti-HBs in 42 neonates at 1 month and 45 neonates at 12 months were 41 ± 8.2 and 35 ± 12.9 , respectively.

Conclusions: The rates of intrauterine HBV infection in the HBIG group and control group were 5.9% and 18.5% respectively ($\chi^2 = 3.86$, $P < 0.05$). The average values of anti-HBs at one month of age in the 2 groups were 46 ± 9.7 and 41 ± 8.2 ($t = 2.609$, $P < 0.05$). More than 94% high risk infants at one year of age can be protected by the combined antepartum and postpartum immunoprophylaxis by significantly interrupting the transmission of HBV from mothers with both HBsAg and HBeAg positive to their infants.

World J Pediatr 2005;1:30-33

Key words: hepatitis B virus; antepartum and postpartum; interruption; transmission;

Introduction

Since 1992, the immunization of hepatitis B (HB) vaccine has been brought into the Expanded Program on Immunization (EPI) Service in China. HB vaccine is highly effective in decreasing HBsAg carrier rate to less than 1% in children below 5 years of age in large cities.^[1] Despite this as well as the practice of passive-active immunoprophylaxis, 20%-30% of babies, whose mothers who are hepatitis B virus (HBV) carriers may fail with vaccination against HBV,^[2,3] to which intrauterine HBV infection is attributive.^[4] To assess the interruptive effect of HBV transmission from mother to infant, we used combined antepartum and postpartum immunoprophylaxis to interrupt the transmission of HBV from mothers both HBsAg and HBeAg positive from January 1995 to December 2001.

Author Affiliations: Department of Infectious Diseases, Children's Hospital, Fudan University, Shanghai 200032, China (Zhu QR, Yu H, Chen H, Dong ZQ, Fei LE and Gu XH); Department of Obstetrics, Zhongshan Hospital, Fudan University, Shanghai 200032, China (Zhang XZ)

Corresponding Author: Qi-Rong Zhu, MD, Department of Infectious Diseases, Children's Hospital, Fudan University, Shanghai 200032, China (Tel: 86-21-54524666; Fax: 86-21-64038992; Email: qrzhu@shmu.edu.cn)

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Methods

Study participants

One hundred and four healthy pregnant women were HBsAg and HBeAg positive carriers without symptoms aged 19-33 years (average 23 years) from the Obstetric Department of Zhongshan Hospital, Fudan University, Shanghai, and the Shanghai Ninth People's Hospital of Shanghai Second University, Shanghai, China. A total of 105 neonates (49 were male and 56 female, including twins) were periodically followed up until one year of age.

Methods

The pregnant women were randomly divided into 2 groups: HBV specific immunoglobulin (HBIG) and control, after informed consent was obtained and the study proposal was approved by the institutional ethics committee. The HBIG group was given an intramuscular dose of 400 IU HBIG at months 3, 2 and 1 before delivery whereas the control group received no HBIG at all. The neonates in the two groups received a intramuscular dose of 200 IU HBIG after inguinal venipuncture of blood sample at birth and 2 weeks after birth, followed by 30 µg plasma-derived hepatitis B vaccine or 5 µg recombinant yeast-derived HB vaccine at 1, 2 and 7 months of age. A series of specimens were obtained from the peripheral venous blood of the neonates at birth and 1, 4, 7, and 12 months of age.

Serological assay

The blood specimens were tested for HBsAg and HBeAg by monoclonal enzyme immunoassay (Abbott kits). Antibodies to HBsAg (anti-HBs) were determined by EIA with the commercially available kits of Kehua Biotech, Shanghai, China. The concentration of anti-HBs was indicated by optical density (OD). The cut off value of OD was determined by comparison of positive control mean (S) and negative control mean (N). Specimens with values greater than or equal to 2.1 were considered positive. The value greater than or equal to 10 was protected persistently by immunoprophylaxis. HBV DNA was determined by the dot-blot hybridization of digoxin-sign (Laboratory of Kehong-Bioengineering, Shanghai Medical University, Shanghai, China). The abnormal level of ALT for liv-

er function was less than 40 IU/L. The persistence of HBsAg and/or HBeAg, HBV DNA for more than 1 to 4 months in the sera of a neonate may be the evidence of HBV intrauterine infection and the persistence for more than 6 months was diagnosed as chronic HBV infection.^[5]

A sample equation for randomized control trial was suggested:

$$N_1 = N_2 = \left[t_{\alpha} \sqrt{2p(1-p)} + t_{\beta} \sqrt{p_t(1+p_t) - p_c(1-p_c)} \right]^2 / (p_t - p_c)^2 \quad [6, 7]$$

According to a preliminary study:^[6] $p_t = 95\%$, $p_c = 75\%$; $p = (p_t - p_c) / 2$, $\alpha = 0.05$, $t_{\alpha} = 1.96$; $\beta = 0.10$, $t_{\beta} = 1.28$; $N_1 = N_2 = 43$. As usual, sample size plus 5% of an estimate ($N_1 = N_2$) was 46. A total of 46 cases were included in each group.

Statistical analysis

The chi-square or *t* test was used to compare the results in each group.

Results

In the HBIG group, 5 of 51 neonates had seroconversion of HBV infectious marker positive at birth. Of the 5 HBV positive neonates, 3 were found to be persistent for 4 months, and then still persistent for one year. In the 3 neonates, 2 were persistent for one year with abnormal level of ALT in venous blood. In the control group, 12 of 54 neonates were detected HBV infection marker positive at birth. Ten of the 12 HBV positive neonates were detected persistent for 4 months and 9 for one year, including 5 neonates with abnormal level of ALT in venous blood. All the 7 neonates with abnormal ALT on physical examination showed nothing abnormal except for the liver palpable 1-2 cm below the right costal margin in the period of follow-up. The status of both intrauterine HBV infected (IHI) neonates and chronic HBV infected (CHI) neonates in the two groups is shown in Table 1. The proportions of neonates with anti-HBs positive and average values (S/N) during the period of follow-up in the two groups are shown in Table 2. These recipients of HBIG and HBV vaccine injection had no side-effects in the study.

Table 1. Intrauterine HBV infected neonates and chronic HBV infected neonates in the two groups

Group	Mothers	Neonates	No of HBV infected neonates					IHI		CHI	
			Birth	1	4	7	12 (mon)	No	%	No	%
HBIG	51	51	5	4	3	3	3	5.9	3	5.9	
Control	53	54	12	12	10	9	9	18.5	9	16.7	
Total	104	105	17	16	13	12	12	13	12		

Comparison of IHI; $\chi^2 = 3.86$, $P < 0.05$.

Table 2. Conditions of neonates with anti-HBs positive and average values of S/N (mean \pm SD) during the period of follow-up in the two groups

Group	Neonates	1		4		7		12 (mon)	
		No	average *	No	average	No	average	No	average **
HBIG	51	47	46 \pm 9.7	48	32 \pm 11.6	48	12 \pm 4.6	48	36 \pm 15.1
Control	54	42	41 \pm 8.2	44	31 \pm 9.8	45	13 \pm 5.4	45	35 \pm 12.9

* : $t=2.609$, $P<0.05$; * * : $t=0.3423$, $P>0.05$.

Discussion

Hepatitis B virus infection acquired perinatally and in early childhood is usually asymptomatic, becoming chronic in 90% and 30% of cases, respectively.^[7-9] Mother to infant transmission of HBV is an important cause for a high chronic infection rate in infants born to HBV carrier mothers. When the mother is alone HBsAg positive in her serum, the transmission rate is about 40% to 50% after 6 months of age in her neonates.^[10,11] If the mother with both HBsAg and HBeAg positive or viral DNA in serum, the transmission rate is estimated to be greater than 90%. Hence the high risk of HBV transmission consists of neonates born to HBV carrier mother.^[12] The efficacy of hepatitis B vaccine against HBV infection in clinical practice is well documented. Combined passive and active immunizations have shown an effective rate of 70% to 90%, but intrauterine infection of HBV is the major cause of failure of the vaccinations designed to combat hepatitis B infection in neonates.^[13] HBV infection in the uterus may be interrupted by injecting multiple intramuscular HBIG before delivery.^[14-16]

In this study, combined antepartum and postpartum immunoprophylaxis was given to interrupt the transmission of HBV from mothers with both HBsAg and HBeAg positive to their infants. The positive rates of HBV infection markers (HBsAg and/or HBeAg, HBV DNA) at birth in the HBIG group and control group were 9.8% and 22.2% respectively ($\chi^2 = 2.980$, $P = 0.0857$). The persistent positive rates at more than 4 months of age in the 2 groups, which indicate intrauterine HBV infection, were 5.9% and 18.5% ($\chi^2 = 3.86$, $P = 0.0494$). Although the chronic HBV infection rates at one year of age in the HBIG group and control group were 5.9% and 16.7% ($\chi^2 = 3.312$, $P = 0.0825$), the rate of chronic HBV infection (CHI) reduced by 10% in the 2 groups. The increased doses of HBIG before delivery decreased effectively both intrauterine infection and chronic HBV infection in high risk infants without any side-effects.

Passive immunization followed by active immunization after birth showed average values (S/N) of anti-HBs at one month of age in the 2 groups were 46 \pm 9.7 and 41 \pm 8.2 ($t=2.609$, $P=0.011$). Obviously, the changes were statistically significant in the HBIG group. The increased average values of anti-HBs in the HBIG group was induced by using multiple intramuscu-

lar injections of HBIG to HBV carrier mothers before delivery.^[14-16] Increasing evidence suggests that HBIG at the antepartum can cross the human placenta and produce passive immunoprophylaxis to the fetus.^[17,18] In this study, the preventive rate for neonates at one year of age in the HBIG group and control group were 83.3% (45/54) and 94.1% (48/51). The results indicate that more than 94% of the high risk infants who are born to HBsAg positive and HBeAg or HBV DNA positive carrier mothers are protected by combined antepartum and postpartum immunoprophylaxis, i. e., interrupting the transmission of HBV from the mothers to their infants. Injecting HBIG during the pregnancy can decrease the intrauterine HBV infection effectively and one of its mechanisms may be that the fetuses acquire the transplacental anti-HBs.^[19,20]

Funding: This study was supported by a grant from Clinical Key Projects of Chinese Health Ministry (No. 97030223).

Ethical approval: This study was approved by the regional committee on medical research ethics.

Competing interest: No benefits in any form have been received or will be received from a commercial party related directly or indirectly to the subject of this article.

Contributors: ZQR designed the study and wrote the draft. YH analyzed the data. All authors contributed to the clinical trial.

References

- Zhu ZH. Challenges to and strategies for the new century's child health care. *Chin J Pediatr* 2000;38:267-270.
- Lazizi Y, Badur S, Perk Y, Ilter O, Pillot J. Selective unresponsiveness to HBsAg vaccine in newborns related with an utero passage of hepatitis B virus DNA. *Vaccine* 1997;15:1095-1100.
- Deng XQ, Xa ZY, Liu Y, Zhuang YL, Duan SC, Lu PH, et al. Failure with hepatitis B vaccine in neonates. *Chin Med Sci J* 1999;14 (Suppl): 26-31.
- Li XM, Yang YB, Hou HY, Shi ZJ, Shen HM, Teng BQ, et al. Interruption of HBV intrauterine transmission: a clinical study. *World J Gastroenterol* 2003;9:1501-1503.
- Wang JS, Zhu QR. Correlation risk factors of chronic hepatitis B virus infection after hepatitis B vaccination in children whose mothers were HBsAg carriers. *J Clin Pediatr* 2001;19:134-136.
- Zhu QR, Yu GJ, Lv Q, Yu H, Gu XH, Zhang XZ, et al. A randomized control trial on interruption of HBV transmission in uterus. *Chin J Pediatr* 2002;40:478-480.
- Theodore Colton. *Clinical trials. In: Statistics in medicine.* Boston: Little, Brown and Company;1974:257-264.

- 8 Roberto DF, Antoine H, George KKL, Daieil L, Anna SL, Neil MI, et al. Easl international consensus conference on hepatitis B. *Hepatology* 2003;38:533-540.
- 9 Shiraki K. Perinatal transmission of hepatitis B virus and its prevention. *J Gastroenterol Hepatol* 2000;15(Suppl):E11-15.
- 10 Lok AS, Lai CL. A longitudinal follow-up of asymptomatic hepatitis B surface antigen positive Chinese children. *Hepatology* 1988;8:1130-1133.
- 11 Chang MH, Hsu HY, Hsu HC, Ni YH, Chen JS, Chen DS. The significance of spontaneous hepatitis E antigen seroconversion in children; with special emphasis on the clearance of hepatitis E antigen before 3 years ago. *Hepatology* 1995;22:1389-1392.
- 12 Ranger-Rogez S, Alais S, Denis F. Hepatitis viruses: mother to child transmission. *Pathol Boil* 2002;50:568-576.
- 13 Wang JS, Zhu QR. Interruption of the transmission of hepatitis B virus from mother to babies. *Chin J Hepatol* 2002;10:308-310.
- 14 Yue YF, Yang XJ, Zhang SL, Han XB. Prevention of intrauterine infection by hepatitis B virus with hepatitis B immune globulin: efficacy and mechanism. *Chin Med J* 1999;112:37-39.
- 15 Hu YW, Liu HM, Yuan J, Wen B. Effects of blocking intrauterine and perinatal hepatitis B virus infection with HBIG made at home or abroad. *Chin J Pediatr* 2002;40:733-735.
- 16 Li H, Wang ZM, Huang Y, Zhang BM. Study of the mechanism of maternal-infant transmission of HBV by interruption injected hepatitis B virus-specific immunoglobulin. *Chin J Birth Health Heredity* 2002;10:65-69.
- 17 Yue YF, Li YJ, Zhang SL. Clinical study of preventing HBV intrauterine infection by active and passive immunization of pregnant women. *Chin J Perinatal* 2000;3:3-5.
- 18 Ayoola EA, Johnson AO. Hepatitis B vaccine in pregnancy: immunogenicity, safety and transfer of antibodies to infants. *Int J Gynecol Obstet* 1987;25:297-301.
- 19 Chen N, Zhu QR, Wang JS. Transplacental serum markers of hepatitis B virus from mothers to infants and its existing time. *Chin J Infect Dis* 2004;22:335-336.
- 20 Li XM, Yang YB, Hou HY, Shi ZJ, Chen HM, Teng BQ, et al. Interruption of HBV intrauterine transmission: a clinical study. *World J Gastroenterol* 2003;9:1501-1503.

Received January 11, 2004

Accepted after revision March 18, 2005