P.O.Box 2345, Beijing 100023,China
Fax: +86-10-85381893
E-mail: wjg@wjgnet.com www.wjgnet.com

World J Gastroenterol 2003;9(7):1501-1503
World Journal of Gastroenterology
 ISSN 1007-9327

• VIRAL HEPATITIS •

Interruption of HBV intrauterine transmission: A clinical study

Xiao-Mao Li, Yue-Bo Yang, Hong-Ying Hou, Zhong-Jie Shi, Hui-Min Shen, Ben-Qi Teng, Ai-Min Li, Min-Feng Shi, Ling Zou

Xiao-Mao Li, Yue-Bo Yang, Hong-Ying Hou, Zhong-Jie Shi, Hui-Min Shen, Ben-Qi Teng, Ai-Min Li, Ling Zou, Department of Obstetrics and Gynecology, The Third Affiliated Hospital, Sun Yat-Sen University, Guangzhou 510630, Guangdong Province, China
Min-Feng Shi, Maternity Hospital, School of Medicine, Zhejiang University, Hangzhou 310006, Zhejiang Province, China
Supported by the Science and Research Foundations of Sun Yat-Sen University and Guangzhou Science Committee, No.1999-J-005-01
Correspondence to: Xiao-Mao Li, Department of Obstetrics and Gynecology, The Third Affiliated Hospital, Sun Yat-Sen University, Guangzhou 510630, Guangdong Province, China. tigerli777@163.com
Telephone: +86-20-85515609 **Fax**: +86-20-87565575
Received: 2003-03-13 **Accepted:** 2003-04-11

Abstract

AIM: To investigate the effect of hepatitis B virus (HBV) specific immunoglobin (HBIG) and lamivudine on HBV intrauterine transmission in HBsAg positive pregnant women.

METHODS: Each subject in the HBIG group (56 cases) was given 200 IU HBIG intramuscularly (im.) every 4 weeks from 28-week (wk) of gestation, while each subject in the lamivudine group (43 cases) received 100 mg lamivudine orally (po.) every day from 28-wk of gestation until the 30th day after labor. Subjects in the control group (52 cases) received no specific treatment. Blood specimens were tested for HBsAg, HBeAg, and HBV-DNA in all maternities at 28-wk of gestation, before delivery, and in their newborns 24 hours before the administration of immune prophylaxis.

RESULTS: Reductions of HBV DNA in both treatments were significant ($P<0.05$). The rate of neonatal intrauterine HBV infection was significantly lower in HBIG group (16.1 %) and lamivudine group (16.3 %) compared with control group (32.7 %) ($P<0.05$), but there was no significant difference between HBIG group and lamivudine group ($P>0.05$). No side effects were found in all the pregnant women or their newborns.

CONCLUSION: The risk of HBV intrauterine infection can be effectively reduced by administration of HBIG or Lamivudine in the 3rd trimester of HBsAg positive pregnant women.

Li XM, Yang YB, Hou HY, Shi ZJ, Shen HM, Teng BQ, Li AM, Shi MF, Zou L. Interruption of HBV intrauterine transmission: A clinical study. *World J Gastroenterol* 2003; 9(7): 1501-1503
http://www.wjgnet.com/1007-9327/9/1501.asp

INTRODUCTION

It is of vital importance to interrupt the transmission of viral hepatitis B from mother to fetus in control of its prevalence[1-3], including HBV intrauterine infection[4-7]. This study investigated the effect of administration of HBIG (im.) and lamivudine (po.) on the interruption of HBV intrauterine infection from the 3rd trimester of gestation.

MATERIALS AND METHODS

Subjects

One hundred and fifty one pairs of women and their newborns who followed the antepartum care were selected and admitted for labor in our hospital from January of 1999 to December of 2001. These pregnant women were HBsAg positive, with normal liver and kidney function. Serial tests were negative for HAV, HCV, HDV and HEV in these women and no other severe complications were found and no other drugs, including the ones that were studied, anti-virus, cytotoxic, steroid hormones, or immune regulating drugs were administrated. The patients were randomly allocated into 3 groups. There were 56 patients in the HBIG group (22 were both HBsAg and HBeAg positive) and 43 in the lamivudine group (33 were both HBsAg and HBeAg positive). There were 52 patients in the control group (17 were both HBsAg and HBeAg positive). No significant differences were found in age, race, time of gestation and parturition, gestational age, way of delivery, and incidence of threatened abortion, threatened labor or pregnancy-induced hypertension syndrome (PIH). The 151 pregnant women delivered 151 newborns.

Methods

Patients in the HBIG group were administered HBIG 200IU intramuscularly (im.) from 28-wk of gestation, once every 4 weeks till labor. Patients in the lamivudine group were administered 100 mg (po.) lamivudine orally daily till the 30th day after labor. Patients in the control group were given no specific treatment. Blood specimens were tested for HBsAg, HBeAg, and HBV-DNA in all the subjects at 28-wk and before delivery, and their newborns (blood from the femoral vein) 24 hours before administration of immune prophylaxis.

HBsAg and HBeAg were assessed by ELISA, the assay kits were produced by Zhongshan Biological and Engineering Co. Ltd. HBV-DNA was assessed by fluorogenic quantitative polymerase chain reaction (FQ-PCR), and the assay kits were produced by Da'an Gene Diagnosis Center, Sun Yat-Sen University.

Before the administration of positive and/or active prophylaxis at 24 hours after delivery, intrauterine HBV infection would be considered if HBsAg and/or HBeAg were tested positive in neonatal peripheral blood.

Statistics

The *t*-test and χ^2 test were used to analyze our data using Excel software. Statistical significance was set at $P<0.05$. HBV DNA values were expressed as $\bar{x}\pm s$, and neonatal intrauterine HBV infection rates were expressed as percentage of total cases in each group.

RESULTS

Changes of HBsAg, HBeAg and HBV DNA

HBsAg turned negative in 1 case of the HBIG group, but HBeAg turned negative in no case. HBsAg and HBeAg turned negative in 1 case of the lamivudine group. No cases turned negative of HBsAg or HBeAg in the control group.

Before administration of agents, there was no significant difference in the values of HBV DNA among 3 groups

(P>0.05). But there was significant difference between the values of HBV DNA in HBIG group and lamivudine group after administration of either reagent respectively (both values reduced, P<0.05). The reduction of value before and after administration of the reagents was significantly different between the administered groups and control group (P<0.05). (Table 1).

Table 1 Comparison of HBV DNA values before and after administration of the reagents

Group	*n*	Log10 HBV DNA before administration of drugs (copies / ml)	Log10 HBV DNA before labor (copies / ml)	Minus value of log10 HBV DNA before and after administration of agents (copies / ml)
HBIG	56	7.38±1.17[a]	5.28±2.77[bd]	2.09±2.28[b]
Lamivudine	43	7.49±0.54[a]	5.33±1.34[bd]	2.16±1.27[b]
Conrol	52	7.05±1.29[a]	6.23±3.66[c]	0.82±2.73[c]

[a]P>0.05 *vs* other groups; [b]P>0.05 between HBIG group and lamivudine group; [c]P<0.05 *vs* HBIG group or lamivudine group; [d]P<0.05 (before *vs* after administration).

Incidence of HBV intrauterine infection

Three newborns were HBsAg positive, and 7 cases were HBeAg positive, one of them was doubly positive for HBsAg and HBeAg in HBIG group. Corresponding cases in lamivudine group and control group were 1, 7, and 1, or 8, 11, and 2 respectively. The infection rates of HBIG, lamivudine, and control groups were 16.1 %, 16.3 %, and 32.7 %, respectively. There were significant differences between the incidence of HBV intrauterine infection in either reagent administrated group and control group (P<0.05), while there was no significant difference between HBIG group and lamivudine group (P>0.05). (Table 2).

Table 2 Incidence of neonatal intrauterine infection in 3 groups

Group	*n*	HBsAg(+) *n*	HBeAg(+) *n*	Intrauterine infection	
				n	%
HBIG	56	3	7	9	16.1[a]
Lamivudine	43	1	7	7	16.3[a]
Control	52	8	11	17	32.7[b]

[a]P>0.05 between HBIG group and lamivudine group; [b]P<0.05 *vs* HBIG group or lamivudine group.

Safety

There were no incidences of fever, rigor, rash, or other complaints and dysfunction of the liver and kidney in subjects throughout administration and follow-ups. There were no significant differences in gestational age, severity of postpartum hemorrhage, rate of cesarean section, neonatal weight, neonatal height, circumference of neonatal head and Apgar score (P>0.05).

DISCUSSION

There are several thoughts about the mechanisms of HBV intrauterine transmission, including placental infection[8], placental exudation and transudation[9-11], peripheral blood monocyte (PBMC)[12] infection, fraternal transmission, etc. Infection through placenta is the most active pathway in maternity-fetus transmission. It is suggested that infection mainly occurs in the 3rd trimester. This might be resulted from the fact that the layer of trophoblastic cells becomes thinner and turns into chorion-vessel membrane, which makes it easier for HBV to pass the placental barrier[13]. The organs of fetus during this period have already developed, therefore, it is safe for the administration of reagents. So we chose this period to begin the interruption of infection. Lamivudine (po.) or HBIG (im.) was administered from 28-wk of gestation.

Barrier-destroying factors, such as threatened abortion, threatened premature labor and TORCH (toxoplasmosis, others, rubella, cytomegalovirus, herpes) infection, were the highly risk factors for HBV intrauterine infection[14]. It is generally considered that intrauterine infection might be the general effect of maternity and virus. In this study, there were no significant differences among the 3 groups in the highly risk factors (threatened abortion, threatened premature labor) or age, time of gestation and delivery, pregnant complication, medical or surgical complication, gestational age at labor or way of delivery.

It has been clinically accepted to administer joint immune reagents (HBIG together with HBV vaccine) to neonates with high risks, but the immune failure rate is still about 10-20 %[15,16], the main reason is intrauterine infection. So, it is important to study the mechanism of HBV intrauterine infection, and we further investigated the intrauterine prevention and interruption of HBV infection.

HBIG is a highly effective immune globulin[17], which is purified from highly effective plasma or serum taken from healthy individuals after the use of HBV vaccine. HBs antibody can bind HBsAg, activate the complimentary system at the same time and strengthen humoral immune, clear HBV, and reduce the number of virus in the maternal blood. It can prevent and decrease the incidence of normal cell infection and might reduce HBV copy in the body. Placenta has the function of transmitting antibody in the form of IgG to the fetus. It is suggested that after maternal administration of HBIG (im.), HBsAb can be transmitted to fetus, which makes it possible for the fetus to obtain the protection of intrauterine passive immunization and to prevent intrauterine infection[18]. The results of this study suggest that regular administration of HBIG (im.) to HBV positive pregnant women might reduce the amount of HBV DNA in blood, and neonatal intrauterine infection rate also reduced significantly when compared with control group. DNA polymerase of HBV has many functions in the process of virus replication. After infection of liver cells by HBV, the incomplete double strand DNA integrates into a complete one, enters the nuclei, forming super helix covalent closed circular DNA (cccDNA). cccDNA is extremely stable, and is the resource of viral DNA and directs the formation of viral protein. The whole mRNA replicated from cccDNA model can form a single strand minus-DNA by reverse-transcription of the HBV DNA polymerase. This DNA can form incomplete double strand DNA through DNA polymerase. The latter can also integrate with antigen proteins in the endoplast, forming new, contagious mature viral particles and be released into blood, or migrate into the nuclei to supply cccDNA there. The multiple functions of HBV polymerase enable it to become one of the most prosperous anti-virus targets.

Lamivudine is a potent anti-virus nucleotide analogue to HBV and HIV. Through competitive inhibition of HBV DNA polymerase and formation of new HBV DNA strand, it can terminate the synthesis of new strand[19,20]. After several days of the administration of lamivudine, the level of HBV DNA drops dramatically, and throughout the treatment, HBV DNA will be suppressed continuously. It can reduce the necrosis and inflammation of the liver and bring ALT level to normal without significant side effects or malformation-causing effects[21-31]. We found the amount of HBV DNA in blood and the rate of neonatal intrauterine infection after administration of lamivudine in the 3rd trimester were significantly lower than that in control group. This suggests that administration of lamivudine of HBV positive pregnant women in the 3rd trimester can effectively decrease the rate of intrauterine HBV infection.

As a passive antibody, the main effect of HBIG is to neutralize HBV in the body, prevent and decrease infection of normal cells[32]; while lamivudine is a potent anti-virus agent, which can suppress the replication of HBV actively, decrease HBV level during pregnancy. Our data showed that the neonatal infection rates, after these two reagents were used in the 3rd trimester to interrupt intrauterine HBV infection, were 16.1 % and 16.3 %, respectively, with no significant difference between these 2 groups (P>0.05). But compared with control group, the infection rates of both groups were significantly lower. These data indicate that both of them are safe and effective in the interruption of intrauterine HBV infection.

We found in our previous studies that HBV DNA level in maternal serum was an important factor for intrauterine infection[1]. Especially when HBV DNA is $\geqslant 10^8$ copies/ml, it has significant correlation with neonatal HBV infection[33]. Administration of HBIG in combination with lamivudine in these patients might decrease the neonatal HBV infection rate more effectively. Further studies are required to improve our understanding about this problem.

REFERENCES

1 **Li XM**, Liu SL, Li X, Huang HJ, Lu JX, Gao ZL. The level of HBV DNA in peripheral, umbilical, and milk of maternal and its correlation. *Zhongshan Yike Daxue Xuebao* 2000; **21**: 233-235

2 **Merle P**, Trepo C, Zoulim F. Current management strategies for hepatitis B in the elderly. *Drugs Aging* 2001; **18**: 725-735

3 **Hamdani-Belghiti S**, Bouazzaou NL. Mother-child transmission of hepatitis B virus. State of the problem and prevention. *Arch Pediatr* 2000; **7**: 879-882

4 **Zhang SL**, Han XB, Yue YF. Relationship between HBV viremia level of pregnant women and intrauterine infection: neated PCR for detection of HBV DNA. *World J Gastroenterol* 1998; **4**: 61-63

5 **Shiraki K**. Perinatal transmission of hepatitis B virus and its prevention. *J Gastroenterol Hepatol* 2000; **15** (Suppl): E11-15

6 **Michielsen PP**, Van Damme P. Viral hepatitis and pregnancy. *Acta Gastroenterol Belg* 1999; **62**: 21-29

7 **Tang JR**, Hsu HY, Lin HH, Ni YH, Chang MH. Hepatitis B surface antigenemia at birth: a long-term follow-up study. *J Pediatr* 1998; **133**: 374-377

8 **Xu DZ**, Yan YP, Zou S, Choi BC, Wang S, Liu P, Bai G, Wang X, Shi M, Wang X. Role of placental tissues in the intrauterine transmission of hepatitis B virus. *Am J Obstet Gynecol* 2001; **185**: 981-987

9 **Kroes AC**, Quint WG, Heijtink RA. Significance of isolated hepatitis B core antibodies detected by enzyme immunoassay in a high risk population. *J Med Virol* 1991; **35**: 96-100

10 **Suga M**, Shibata K, Kodama T, Arima K, Yamada S, Yachi A. A case of HBs antigen negative fulminant hepatitis with IgM antibody to hepatitis B core antigen persisting more than seven years. *Gastroenterol Jpn* 1991; **26**: 661-665

11 **Wang JS**, Zhu QR. Infection of the fetus with hepatitis B e antigen via the placenta. *Lancet* 2000; **355**: 989

12 **Leung NW**, Tam JS, Lau GT, Leung TW, Lau WY, Li AK. Hepatitis B virus DNA in peripheral blood leukocytes. A comparison between hepatal cellulo carcinoma and other hepatitis B virus-related chronic liver diseases. *Cancer* 1994; **73**: 1143-1148

13 **Yan YP**, Xu DZ, Wang WL, Liu B, Liu ZH, Men K, Zhang JX, Xu JQ. The relation between HBV placenta infection and intrauterine transmission. *Zhonghua Fuchanke Zazhi* 1999; **34**: 392-395

14 **Del Canho R**, Grosheide PM, Schalm SW, de Vries RR, Heijtink RA. Failure of neonatal hepatitis B vaccination: the role of HBV-DNA levels in hepatitis B carrier mothers and HLA antigens in neonates. *J Hepatol* 1994; **20**: 483-486

15 **Zhu Q**, Lu Q, Gu X, Xu H, Duan S. A preliminary study on interruption of HBV transmission in uterus. *Chin Medl J (Engl)* 1997; **110**: 145-147

16 **Xu DZ**, Yan YP, Choi BC, Xu JQ, Men K, Zhang JX, Liu ZH, Wang FS. Risk factors and mechanism of transplacental transmission of hepatitis B virus:A case-control study. *J Med Virol* 2002; **67**: 20-26

17 **Ghendon Y**. Perinatal transmission of hepatitis B virus in high-incidence countries. *J Virol Methods* 1987; **17**: 69-79

18 **Yue YF**, Yang XJ, Zhang SL, Han XB. Clinical research of the effect of intramuscular administration of HBIG on HbsAg positive pregnant women to prevent vertical transmission. *Zhongguo Shiyong Fuke Yu Chanke Zazhi* 1999; **15**: 547-548

19 **Yao GB**, Wang SB, Cui ZY, Yao JL, Zeng MD. A multi-center random double-blind case-control study of the treatment of chronic hepatitis B by Lamivudine. *Zhongguo Xinyao Yu Linchuang Zazhi* 1999; **18**: 131-135

20 **Johnson MA**, Moore KH, Yuen GJ, Bye A, Pakes GE. Clinical pharmacokinetics of lamivudine. *Clin Pharmacokinet* 1999; **36**: 41-66

21 **Rizzetto M**. Efficacy of lamivudine in HBeAg-negative chronic hepatitis B. *J Med Virol* 2002; **66**: 435-451

22 **Ricceri L**, Venerosi A, Valanzano A, Sorace A, Alleva E. Prenatal AZT or 3TC and mouse development of locomotor activity and hot-plate responding upon administration of the GABA(A) receptor agonist muscimol. *Psychopharmacology (Berl)* 2001; **153**: 434-442

23 **Calamandrei G**, Venerosi A, Branchi I, Valanzano A, Alleva E. Prenatal exposure to anti-HIV drugs. long-term neurobehavioral effects of lamivudine (3TC) in CD-1 mice. *Neurotoxicol Teratol* 2000; **22**: 369-379

24 **Culnane M**, Fowler M, Lee SS, McSherry G, Brady M, O'Donnell K, Mofenson L, Gortmaker SL, Shapiro DE, Scott G, Jimenez E, Moore EC, Diaz C, Flynn PM, Cunningham B, Oleske J. Lack of long-term effects of in utero exposure to zidovudine among uninfected children born to HIV-infected women. *JAMA* 1999; **281**: 151-157

25 **Conner EM**, Sperling RS, Gelber R Kiselev P, Scott G, O'Sullivan MJ, VanDyke R, Bey M, Shearer W, Jacobson RL. Reduction of maternal-infant transmission of human immunodeficiency virus type-1 with zidovudine. *N Engl J Med* 1994; **331**: 1173-1180

26 **Vuthipongse R**, Bhadrakom C, Roongpisuthipong P. Administration of Zidovudine during late pregnancy and delivery to prevent perinatal HIV transmission-Thailand 1996-1998. *MMWR* 1998; **47**: 151-154 / / *JAMA* 1998; **279**: 1061-1062

27 **Moodley J**, Moodley D, Pillay K, Coovadia H, Saba J, van Leeuwen R, Goodwin C, Harrigan PR, Moore KH, Stone C, Plumb R, Johnson MA. Pharmacokinetics and antiretroviral activity of lamivudine alone or when coadministered with zidovudine in human immunodeficiency virus type 1-Infected pregnant women and their offspring. *J Infect Dis* 1998; **178**: 1327-1333

28 **van Leeuwen R**, Lange JM, Nijhuis M, Schuurman R, Reiss P, Danner SA, Boucher CA. Results of long-term follow-up of HIV-infected patients treated with lamivudine monotherapy, followed by a combination of lamivudine and zidovudine. *Antivir Ther* 1997; **2**: 79-90

29 **Dienstag JL**, Perillo RP, Schiff ER, Bartholomew M, Vicary C, Rubin M. A preliminary trial of lamivudine for chronic hepatitis B infection. *N Engl J Med* 1995; **333**: 1657-1661

30 **Liaw YF.** Current therapeutic trends in therapy for chronic viral hepatitis. *J Gastroenterol Hepatol* 1997; **12**: S346-353

31 **Lai CL**, Ching CK, Tung AK, Li E, Young J, Hill A, Wong BC, Dent J, Wu PC. Lamivudine is effective in suppressing hepatitis B virus DNA in Chinese hepatitis B surface antigen carriers: a placebo-control trial. *Hepatology* 1997; **25**: 241-244

32 **Yue Y**, Yang X, Zhang S. Prevention of intrauterine infection by hepatitis B virus with hepatitis B immuneglobulin: efficacy and mechanism. *Chin Med J (Engl)* 1999; **112**: 37-39

33 **Ngui SL**, Andrews NJ, Underhill GS, Heptonstall J, Teo CG. Failed postnatal immunoprophylaxis for hepatitis B: characteristics of maternal hepatitis B virus as risk factors. *Clinical Infectious Diseases* 1998; **27**: 100-106

Edited by Yuan HT and Wang XL