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Immuno-prophylaxis of babies borne to hepatitis B carrier mothers

針對乙型肝炎母親誕下嬰兒的免疫預防治療

Objectives. To examine the efficacy of current hepatitis B immuno-prophylaxis and estimate the prevalence of *S*-mutant infections among local newborn babies.
Design. Prospective study.
Setting. Regional hospital, Hong Kong.
Patients. A total of 137 newborn babies delivered between the period of November 2000 and 30 June 2001 inclusive, whose mothers were chronic hepatitis B surface antigen carriers.
Results. Of the 121 infants who were followed up for 12 months, three were found to be chronic hepatitis B virus carriers, giving a vertical transmission rate of 2.5%. One (0.8%) was suspected to be infected by the *S*-mutant. All the three hepatitis B virus carrier babies were born to mothers with hepatitis B e antigen, but none to the eight mothers suspected to have *S*-mutants. Of 119 (98.3%) infants who developed hepatitis B surface antibody upon follow-up at 12 months, 35 were found to have hepatitis B e antigen at birth. All were born to hepatitis B e antigen–positive mothers. Only three of the 35 babies were found to be hepatitis B virus carriers. Most babies lost the hepatitis B e antigen by 6 months of age; only the infected babies had the antigen persisting at 1 year of age. The non-infected infants' hepatitis B e antigen is likely transplacental.
Conclusions. Our hepatitis B virus prophylaxis programme was effective at preventing perinatal infection and the non-infected infants' hepatitis B e antigen was likely transplacental.

目的：檢查現時香港新生嬰兒的乙型肝炎免疫預防治療的效用，並估計發生表面抗原突變而受感染的情況。
設計：前瞻性研究。
安排：地區醫院，香港。
患者：在2000年11月至2001年6月30日出生，而母親長期帶有乙型肝炎表面抗原的新生嬰兒；共137人。
結果：121名跟進了12個月的嬰兒中，3名為長期乙型肝炎病毒帶菌者，即垂直感染率為2.5%，其中1人（0.8%）疑似受表面抗原突變感染。3人的母親都帶有乙型肝炎e抗原，卻無一人來自疑似受表面抗原突變感染的8名母親。在隨後12個月跟進期，119人（98.3%）出現乙型肝炎表面抗體，當中35人於出生時已帶有乙型肝炎e抗原，他們的母親亦全部對乙型肝炎e抗原呈陽性反應。35名嬰兒中只有3人是乙型肝炎帶菌者。大部分嬰兒在6個月大時乙型肝炎e抗原會消失，只有受感染嬰兒的乙型肝炎e抗原至1歲仍然存在。未受感染嬰兒的乙型肝炎e抗原可能通過胎盤得到。
結論：香港的乙型肝炎免疫預防計劃，有效防止嬰兒於懷孕期間受感染，而未受感染嬰兒的乙型肝炎e抗原可能通過胎盤得到。

Key words:
Hepatitis B e antigens;
Hepatitis B surface antigens;
Hepatitis B vaccines;
Infant, newborn

關鍵詞：
乙型肝炎e抗原；
乙型肝炎表面抗原；
乙型肝炎防疫注射；
嬰兒，新生的

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Introduction

Hepatitis B virus (HBV) infection is a global health problem with about 350 million chronic carriers, of whom it is estimated that one quarter will die of hepatitis B (HB)–related chronic liver diseases.[1] Ninety percent of the infants of hepatitis B e antigen (HBeAg)–positive mothers become HBV carriers.[2] Ten percent of the Hong Kong population are chronic HBV carriers.[3,4] Vertical (mother-to-infant) transmission accounts for half of the carriers, and the rest occurs from horizontal transmission in childhood.[3] In Hong Kong, active immunisation for newborn babies of HBV-carrier mothers was introduced in

1983 and has been offered for all newborn babies since 1988.[4] In 1984, Wong et al[5] reported that combined immuno-prophylaxis was very effective in preventing the vertical transmission; 96 to 100% of the infants born to hepatitis B surface antigen (HBsAg) and HBeAg carrier mothers had hepatitis B surface antibody (that is they were anti-HBs positive) at the age of 1 year.[5] The more effective combined use of active HBV vaccine plus passive immunisation with hepatitis B immunoglobulin (HBIG) was incorporated into the local childhood vaccination programme in 1989.[3]

Hepatitis B virus produces enormous viral loads during active replication, without killing the infected cell directly and its uniquely evolved life cycle appears to have interesting consequences.[6] Thus, because HBV uses reverse transcription, which lacks a proofreading function when copying its deoxyribonucleic acid (DNA) genome, mutant viral genomes emerge frequently. Moreover, both endogenous (host immune clearance) and exogenous (vaccines and antiviral drugs) selection pressures readily select out these escape mutants.

The viral envelope encoded by the *S* gene contains three distinct configurations synthesised in all people.[1] Several specific antigenic determinants, including the labelled 'a' (amino acids 124-147),[7] are common to all HBsAg.[1] Most HB vaccines contain the major HBsAg protein and induce an immune response to the major hydrophilic region, located from residue 99 to 170.[6] This anti-HBs response produces protective immunity.[6] Mutations within this epitope have been selected during vaccination.[8] Most mutant isolates contain a mutation from glycine to arginine at residue 145 of HBsAg (sG145R) and this mutation has been associated with vaccine failure.[8] Commercially available assay kits detect HBV by measuring the binding affinity of anti-HBs antibodies.[7] Thus, mutations in the *S* gene can limit or abrogate the diagnostic value of commercially available assays.[7]

In 1988, the first HB variant was reported in Europe.[9] A group of Italian investigators found the presence of HBsAg in the presence of specific antibody (anti-HBs) after active immunisation with and without HBIG being given.[9] Analysis of the HBsAg with monoclonal antibodies revealed that the circulating antigen did not carry the 'a' determinant or that this determinant was masked.[9] These findings suggest that the circulating viral antigen has unusual epitopic conformation and is not neutralised by anti-a surface antibody.[9] Their observation suggests the emergence of a variant of HBV, possibly due to epidemiological pressure associated with immunisation in areas of endemic infection.[9] Since then, a variety of *S* antigen–mutants have been reported from different parts of the world.

Recent reports showed that about 5 to 10% of infants of HBeAg-positive mothers became chronic HBV carriers, despite immuno-prophylaxis.[10,11] In Hong Kong, a recent population-based study reported that the overall prevalence of HBsAg in children between 11 and 19 years old was 5.8% (7.9% in boys and 4.1% in girls).[12] It is therefore important to ensure that the current immuno-prophylaxis programme is effective. The primary objective of this study was to examine the efficacy of the current immuno-prophylaxis programme and to estimate the *S*-mutant infection rate in infants whose mothers are chronic HBV carriers.

Methods

The recruitment period was from November 2000 to June 2001 inclusive. Study details were explained to the mothers by paediatricians, during antenatal classes about HB infection. Pregnant mothers being admitted to the Tuen Mun Hospital (TMH) for delivery were invited to join the study and provide written consent for themselves and signed another written consent for their babies to be tested. The Ethics Committee of the New Territories West Hospital Cluster approved the study protocol.

Serological tests for HBsAg, HBeAg, and anti-HBs were performed with electrochemiluminescent immunoassays using the Elecsys 2010 System[13] (Roche Diagnostics GmbH, Mannheim, Germany). Serum samples, which showed a positive result with the Elecsys HBsAg Assay, were retested with the Elecsys HBsAg Confirmatory Assay to confirm the presence of HBsAg. The Elecsys anti-HBs Assay and Elecsys HBeAg Assay were used to detect anti-HBs and HBeAg, respectively.

Data entry sheets for mother and infant (Appendices 1 and 2) were used to collect the relevant data. While the obstetrician filled out the mother's data sheet, the paediatrician filled out that of the baby.

All pregnant mothers attending the antenatal clinics of TMH were screened for HBsAg at the first antenatal visit. For mothers with positive HBsAg, their HBeAg and anti-HBs status and liver function were determined. The mother would be regarded as infected by an *S*-mutant if she simultaneously had (i) anti-HBs and (ii) HBsAg and/or HBeAg, which conforms to the definition of having an *S*-mutant used in the first HBV variant report.[9]

In this study, chronic HBV carrier was defined as having HBsAg present in serum for at least 6 months.[14] The newborn babies of chronic HBsAg carrier mothers were screened for HBsAg, anti-HBs, and HBeAg at birth (using cord blood), before the immuno-prophylaxis and subsequently at 1, 3, 6, and 12 months. Liver function tests were also checked at 1, 3, 6, and 12 months. If HBsAg or HBeAg was detected with the anti-HBs, the baby was regarded as infected by an *S*-mutant.

Statistics

All results were analysed with the Statistical Package for the Social Sciences (Windows version 10.0; SPSS Inc,

Chicago [IL], US). Categorical variables were compared using two-tailed Chi squared tests and Fisher's exact test. Continuous and ordinal variables were compared using two-tailed *t* tests and Mann-Whitney *U* tests, respectively. For small data sets, Poison approximation or exact formulas were used. A P value of less than 0.05 (two-tailed) was considered statistically significant.

Results

Maternal status of hepatitis B surface antigen, hepatitis B e antigen, and anti-HBs status

The flowchart of maternal HBsAg and HBeAg status and their relationships to HBsAg and HBeAg status in their babies is shown in the Figure. Of 1256 mothers recruited, 149 (11.9%) were found to be chronic HBV carriers, 47 (31.5%) of the latter were HBeAg positive. Five of these HBeAg-positive mothers refused permission to have their babies tested. Of the 42 remaining HBeAg-positive carrier mothers, one delivered twins, as did two non-HBeAg carrier mothers. Of 134 mothers who agreed to have their babies tested, eight (6.0%) conformed to our definition of *S*-mutant, of whom three (2.2%) were both HBeAg and anti-HBs positive.

Epidemiological data of the babies

A total of 137 neonates (72 girls and 65 boys) were followed up. Maternal gestation periods ranged from 33.86 to 42.14 weeks (mean, 39.21; standard deviation [SD], 1.62 weeks). Of these, 92 were born by spontaneous vaginal delivery, 11 by vacuum extraction, and the others by caesarean section. Respective minimal and maximal Apgar's score at 1 and 5 minutes ranged from 5 to 9 (mean, 7.9; SD, 0.5), and 8 to 10 (mean, 8.9; SD, 0.3). The body weight of the babies at birth ranged from 1.73 to 4.64 kg (mean, 3.26; SD, 0.44 kg). Seven infants defaulted 1-month follow-up, two more at the 3-month follow-up, an additional six at the 6-month follow-up, and one more at the 12-month follow-up. Hence, the calculated drop-out rate at 12 months was 11.7% (16/137).

Hepatitis B surface antigen status of the babies

Six were HBsAg positive at birth; two did not test positive for HBsAg on re-check at 1, 3, 6, and 12 months (one of whom was HBeAg positive at birth and at 1 and 3 months old, and the other was HBeAg negative throughout the whole study period). One baby had HBsAg and HBeAg present in the cord blood at birth and at 1 month but turned negative for both antigens at 3 months. In this baby, the disappearance of the HBsAg and HBeAg at 3 months was associated with the appearance of anti-HBs and a concomitant surge in serum alanine transferase (ALT) and bilirubin (SB). The ALT rose from 128 U/L at 1 month to 593 U/L at 3 months and the SB from 27 μmol/L at 1 month to 322 μmol/L at 3 months. However, that infant returned to Mainland China and defaulted subsequent follow-up. The remaining three infants remained HBsAg positive throughout the first year and fulfilled the diagnostic criteria for being chronic HBV carriers. It was found that HBeAg was present in the blood of these three infants throughout the first year, one of whom also had anti-HBs present in her blood at birth, and at 1, 3, 6, and 12 months. Thus, she was an *S*-mutant carrier.

Vertical transmission and S-mutant rates

Of the 121 babies who attended for follow-up from birth till 12 months old, three were HBsAg positive, of whom one had *S*-mutant infection. The vertical transmission rate was therefore 2.5% (3/121) and for the *S*-mutant carrier it was 0.8% (1/121), whereas 98.3% (119/121) of the infants had developed anti-HBs at 12-month follow-up. None of the eight mothers that were *S*-mutant carriers gave birth to an infected baby.

From the 47 chronic HBV-carrier mothers tested HBeAg positive, consent for testing was obtained for 42 of their babies. At 1 year of age, 32 of these babies were HBeAg negative with only three being chronic HBV carriers. None of the 92 HBeAg-negative mothers gave birth to an HBeAg-positive baby. The vertical transmission rate among the HBeAg-positive mothers was 7.1% (3/42).

Maternal variables

The three HBsAg carrier babies were born to HBeAg- and HBsAg-positive mothers. No increases in ALT or clinical evidence of liver disease was encountered in the mothers. The three infected babies were delivered by three different means; two had antenatal complications, including foetal distress and maternal group B streptococcus carrier status.

Neonatal variables

Of the 121 gastric lavage (GL) specimens saved, one from a carrier baby and 32 from non-infected babies were unfit for the testing. Of the three carrier babies, HBsAg was present in the GL of two. Of the 118 non-carrier babies, HBsAg was present in the GL of 46 babies but absent in the GL of 40 babies (that is among HBsAg carriers, 2/2 GL specimens were positive versus 46/86 non-carriers, P=0.5).

Neonatal hepatitis B e antigen status

Thirty-five newborns (33 singleton and one pair of dizygotic twins) were HBeAg positive at birth. The positive HBeAg status among newborns decreases with time. The numbers of HBeAg-positive babies were 35 at birth, 30 at 1 month, 25 at 3 months, 7 at 6 months, and 3 at 12 months (chronic HBV carriers). For the 32 babies born HBeAg positive but becoming HBeAg negative on subsequent follow-up, they acquired anti-HBs from the age of 1 month and were all HBsAg negative at 1 year old. The positive HBeAg status was not related to the infants' serum ALT and SB levels at 1, 3, 6, or 12 month.

Discussion

Hepatitis B virus vaccine

Since the introduction of HB vaccine in the early 1980s,

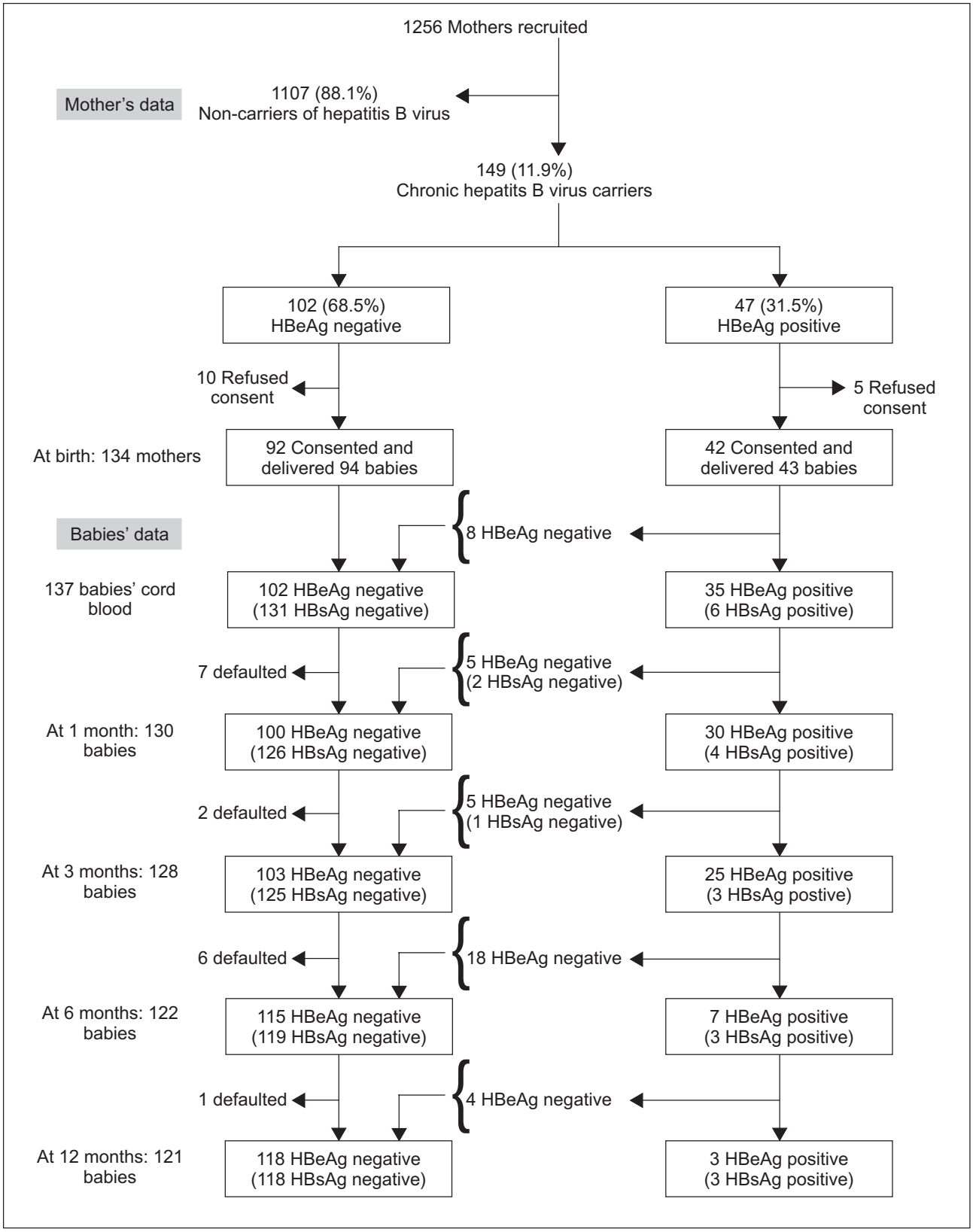

Fig. Flowchart of maternal hepatitis B e antigen (HBeAg) and hepatitis B surface antigen (HBsAg) status and their relationships to neonatal HBeAg and HBsAg status

passive-active immuno-prophylaxis with HBIG and HB vaccine has proved to be highly effective in preventing perinatal transmission of HBV infection.[5,15] In the yeast-recombinant HB vaccine clinical trial, the vaccine was administered in combination with HBIG to high-risk newborns.[15] If mothers positive for both HBsAg and HBeAg

receive no immuno-prophylaxis, 70 to 90% of infants will be infected with the virus and become chronic carriers. Among infants who received HBIG at birth and three 5-mg doses of yeast-recombinant HB vaccine, only 4.8% became chronic carriers, a better than 90% level of protection being obtained.[15] In our study, the vertical transmission rate among the chronic HBV carrier mothers with HBeAg was 7.1%, which is in line with previous findings.[15]

Perinatal factors

As early as 1976, before the introduction of active immunisation for the newborns of HBV carrier mothers, local pioneers had already carried out a prospective study to determine the relative importance of various perinatal factors in the vertical transmission of HBV from symptom-free carrier mothers to their infants.[16] In 1980, Wong et al[16] reported that the presence of HBeAg in maternal serum correlated strongly with the subsequent presence of HBsAg in their infants. Our findings are similar; 32% of the chronic HBV carrier mothers were HBeAg positive. The vertical transmission rate among these HBeAg-positive mothers was 7.1%, whereas there was 0% transmission in HBeAg-negative carrier mothers.

Hepatitis B surface antigen in neonatal gastric lavage

Whilst HBsAg in the GL of newborns has been associated with HBsAg in the blood of babies aged 3 months,[16] use of such testing to predict perinatal HBV infections was addressed in only two studies. In a prospective study from Taiwan involving 147 HBV carrier mothers,[17] neonatal gastric aspirate positive for HBsAg was a predictor of perinatal HBV infection. However, in a study from Mexico involving 113 HBV carrier mothers,[18] HBV-DNA was detected in the serum of seven women, but in the GL of only one newborn. In our study, HBsAg in GL at birth did not correlate with the eventual carrier status of the infants, but the high number of unsuitable gastric specimens was a cause of concern with respect to this form of testing.

Hepatitis B surface antigen in the cord blood and probable intrauterine infection

Hepatitis B surface antigen in the cord blood may be the result of intrauterine infection, materno-foetal transfusion during labour, or contamination by maternal blood during sample collection.[16] Materno-foetal transfusion during labour is a distinct possibility because the presence of cord blood HBsAg has been positively correlated with the duration of labour.[16]

Wong et al[5] performed a prospective double-blind randomised placebo-controlled study to compare the efficacy of an active HB vaccination programme with a combined active-passive immunisation programme (HB vaccination and HBIG injections) in preventing the HBsAg carrier state. In their study, they inferred probable intrauterine infection based on the HBsAg level in the third-day sample. If the third-day HBsAg level was the same or more than the level in the cord blood, the baby was likely to have acquired intrauterine infection (assuming that newborns not infected in utero should have no viral replication). Thus, if the level of HBsAg persisted or even increased in the third-day sample, it was assumed that there was HBV replication in the infant. Using this method, the reported figure for intrauterine infection was about 3%.[19] In the present study, the vertical transmission rate was 2.5% and hence similar to the previous figure.

Hepatitis B e antigen clearance

In our study, one infant was found to have HBsAg/anti-HBs seroconversion and HBeAg clearance at 3 months. However, she defaulted subsequent follow-up. We believed that she had entered the HBeAg clearance phase, as a surge in ALT and SB levels coincided with the seroconversion. Gradual clearance of serum HBeAg with reduced viral replication has been observed previously and is usually preceded by an elevation of aminotransferases; in most such individuals, whilst this insidious HBe seroconversion process takes place over a period of 2 to 7 years.[2] After the elevation of aminotransferases, around 40% of the children clear the HBeAg within 1 year. Rarely, HBeAg/anti-HBe seroconversion may occur as early as infancy with an unclear or brief HBeAg seropositive phase.[2] However, in most instances HBeAg clearance rate is very low before 3 years of age and particularly during infancy, which might be due to immune tolerance to hepatitis B core antigen (HBcAg) and HBeAg.[2]

Hepatitis B surface antigen mutant

Mutations in surface HBsAg, such as those in the 'a' determinant, have been identified as possible causes for vaccine failure.[11] Although these variants were more frequently found in vaccinated than in unvaccinated infected children,[20] 'a' determinant vaccine escape mutants have only a marginal role (if any) to account for unsatisfactory protection.[11,20,21]

A Taiwan serosurvey published in 2004 (conducted in 1999[20]) analysed nucleotide sequences encoding of the 'a' determinant region of HBsAg in all HBV-DNA positive sera from 1357 children and 219 adolescents and compared their findings with the results of prior surveys in the same area. These investigators reported the 1999 prevalence and changes in mutations in relation to data obtained in 1984 (just before vaccination), 1989, and 1994. Their study showed that in 1999 (with universal vaccination in childhood), the prevalence of wild-type HBV continued to decrease. Whereas the absolute numbers of children with infections of 'a' determinant mutants was stable over a 5- to 10-year period and eventually decreased 15 years post-vaccination. The authors concluded that the 'a' determinant variants have an advantage in infecting immunised children, but do not threaten current HBV vaccination strategies in Taiwan. In the present study, only 0.8% (1/121) of the immunised infants was an *S*-mutant carrier and all the eight mothers with *S*-mutant infection

did not pass their infection to their babies. We suggest that the infection by HBV with escape mutations in the *S* open reading frame does not account for most cases of failed immuno-prophylaxis.

Hepatitis B e antigen

Hepatitis B e antigen is a circulating peptide derived from the core gene. It serves as a marker of active viral replication, and its presence in the circulation has been associated with a diminished immune response.[1] Whether HBeAg can pass the placenta from mother directly to baby and induce T-cell tolerance in the uterus, is still being debated. It has been suggested that HBeAg can be more easily transmitted via the placental route than HBsAg, as it is smaller and does not agglutinate.[22]

It was suggested that HBeAg could cross human placenta based on the detection of antigen in the cord blood of the neonate—a procedure that does not exclude maternal blood contamination. Wang and Zhu[23] collected neonatal serum by femoral puncture at birth and reported that 47% (7/15) of neonates from HBeAg-positive mothers were also HBeAg positive compared to 0% (0/18) of neonates from HBeAg-negative mothers. One baby was simultaneously positive for HBsAg and HBeAg. No babies born to HBeAg-negative mothers tested positive for HBeAg or HBsAg. All babies were re-examined at the age of 1 month for serum HBsAg, but no new HBsAg neonates were found. The baby positive for HBsAg at birth was still positive at the age of 1 month, suggesting intrauterine HBV infection.[23]

The serum samples of the seven HBeAg-positive neonates and their mothers were further analysed for HBsAg and HBeAg titre.[23] For the single infected baby, the HBsAg titre was significantly higher than the HBeAg titre, in a ratio similar to the corresponding infected mother.[23] For the other six non-infected babies, their HBeAg titres were low and they were all HBsAg negative.[23]

In another study, the HBeAg titres and HBV DNA levels were measured in 54 mother-infant pairs.[24] Peripheral blood samples were collected from both the mothers before delivery and the neonates (femoral venous) at birth and at 6 and 12 months after birth.[24] Seventy percent (23/33) of neonates from HBeAg-positive mothers were HBeAg positive at birth compared with 0% (0/21) of neonates from HBeAg-negative mothers.[24] Four infants became HBV DNA positive during the 12-month follow-up period, but this did not occur in any of the 10 HBeAg-negative infants. The HBeAg levels of these four HBV-infected babies were high at birth, declined to very low levels at 6 months, and rebounded to a high level at 12 months. Hepatitis B e antigen was cleared from the serum of all the 19 babies who did not develop HBV infection. The changes in serial HBeAg titres suggest that HBeAg can cross the placenta in humans and persists for less than 6 months in most babies with or without HBV infection.[24] The increased HBeAg titres after 6 months in the four infected babies indicates that 'new' HBeAg is generated (possibly from the liver of the newly infected babies). The higher concentrations of HBeAg in the infants with HBV infection suggests that the extent of HBeAg present in post-natal blood may have a role in establishing a chronic HBV infection in neonates.[24] We have similar findings; 81% (35/43) of neonates from HBeAg-positive mothers were HBeAg positive at birth compared with 0% (0/94) from HBeAg-negative mothers. Moreover, HBeAg was cleared from the sera in 32 of these babies by 1 year, of whom 88% (28/32) cleared their passively acquired HBeAg before they were 6 months old.

Potential ways to interrupt hepatitis B virus intrauterine infection

Intrauterine transmission is one of the main sources of HBV vertical infection, but till now there is no definite prophylaxis for this eventality.[25,26] In Guangzhou, a randomised controlled trial investigated the effect of HBIG and lamivudine on intrauterine HBV transmission in HBsAg-positive pregnant women.[27] Either treatment administered in the third trimester effectively reduced neonatal HBV infection rates to a similar level; 16.1% and 16.3%, respectively, versus 32.7% in the control group ($P<0.05$). No side-effects were reported in either the pregnant women or their neonates. The same group also performed a randomised controlled trial to evaluate the efficacy of HBIG as a means of interrupting HBV intrauterine infection during late pregnancy.[28] The neonatal infection rates in the HBIG group and controls were 10.5% and 27.3%, respectively ($P<0.05$). Another large randomised controlled trial involving 980 HBsAg carrier pregnant women performed in Shanghai also reported a significantly lower chronic HB infection in the babies of mothers treated with HBIG as opposed to controls[29]; the respective rates of chronic HB were 5.7% versus 14.3 % ($P<0.001$).[30]

Limitations of the study

Active immunisation for the newborns of HBV carrier mothers in Hong Kong was introduced about 2 decades ago and since then there have been rapid advances in the study of HBV. Serum HBV DNA quantitative assays, extraction and subsequent amplification and sequencing analysis has greatly increased understanding about the real impact of immuno-prophylaxis on overt/occult HBV infection. Due to financial restraints, such molecular evaluation was not undertaken in our study, which limited our estimation of the *S* mutant rate.

Mesenas et al[30] showed that *S*-mutant might be present in HBsAg carriers regardless of anti-HBs positivity or negativity. In their study, the serum HBV DNA test performed on 13 chronic HBV carriers positive for both HBsAg and anti-HBs, eight were infected with 'a' epitope variant (three with variant mixed with wild type and five carrying the epitope variant only). In the group that was HBsAg positive and anti-HBs negative, three of five patients had both wild type and *S*-mutant. Thus, our definition of

S-mutant by the simultaneous presence of HBsAg and HBeAg is not accurate, without molecular testing.

Another limitation of our study was the 11.7% default rate, which is high compared with previous studies.[5,23,24] It may also have been worth collaborating with other major hospitals in different clusters so as to assess a more representative Hong Kong population.

Conclusion

The current HBV prophylaxis programme is effective in preventing vertical transmission of HBV infection, as reflected by the low transmission rate (2.5%) and a high percentage of infants (98.3%) who developed anti-HBs upon follow-up at age 12 months. The estimated *S*-mutant infection rate was 0.8% and maternal HBeAg carrier status was predictive of neonatal carrier status. Moreover, HBeAg can cross the placenta and may have a role in establishing chronic HBV infection in neonates.

Appendix

Additional material related to this articie can be found on the HKMJ website. Please go to <http://www.hkmj.org.hk>, search for the appropriate article, and click on Full Article in PDF following the title.

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Appendix 1. Data sheet of the survey on the efficacy of the immuno-prophylaxis of babies borne to hepatitis B carrier mothers

Mother's data

Mother lives in:
1) Hong Kong, 2) China

Antenatal visit:
1) at Tuen Mun Hospital, 2) elsewhere in Hong Kong, 3) No antenatal visit

Mother's knowledge on her hepatitis B status: she
1) knows, she has; 2) knows, she has not; 3) does not know

Age of infection _____
Diagnosed by:
1) self, 2) herbal doctor, 3) western doctor, 4) clinical, 5) blood test

Diagnosis:
1) hepatitis, unspecified, 2) hepatitis A, 3) hepatitis B, 4) hepatitis C, 5) hepatitis D, 6) hepatitis E, 7) others _____

Symptoms of infection during pregnancy:
1) asymptomatic, 2) recurrent right upper quadrant pain, 3) jaundice, 4) malaise, 5) deranged liver function tests

Complications:
1) chronic active hepatitis, 2) compensated cirrhosis, 3) uncompensated cirrhosis (ascites/gastrointestinal bleeding), 4) hepatocellular carcinoma

Treatment received:
1) no, 2) self, 3) herbs, 4) interferon, 5) nucleoside analogue, 6) others _____

Mother's blood result

Hepatitis B surface antigen:
1) positive in Department of Health, 2) negative in Department of Health, 3) weakly positive

Hepatitis B surface antigen:
1) positive in Tuen Mun Hospital, 2) negative in Tuen Mun Hospital, 3) weakly positive

Hepatitis B surface antibody:
1) positive, 2) negative, 3) weakly positive

Hepatitis B e antigen:
1) positive, 2) negative, 3) weakly positive

Alanine transferase level: _____
Mother has received transfusion within 3 months before blood taking for this study:
1) yes, in Hong Kong, 2) yes, outside Hong Kong, 3) no, 4) do not know

Please be informed that if the mother has received transfusion, she has to be called back 6 months later to have the blood rechecked

Appendix 2. Baby's data

P _ G _ Gestation ____ weeks ____ days

Antenatal visit at Tuen Mun Hospital:
1) yes, 2) no

Mode of delivery:
1) normal spontaneous delivery, 2) vacuum extraction, 3) forcep, 4) elective caesarean section, 5) emergent caesarean section

Complication of pregnancy:
1) toxaemia of pregnancy, 2) antepartum haemorrhage, 3) placenta previa, 4) prolonged rupture of membrane, 5) others _____

Apgar score: 1 min_____, 5 min_____
Birth weight: _____ kg
Time of delivery: 200_ yr, ___ mth, ___day, ____hr (am/pm), ____ min
Time of vaccine: ____ mth, ___ day, hr (am/pm), ____ min; dose _____
Time of hepatitis B immunoglobulin:_____mth, ____day, hr (am/pm), ____ min; dose _____

Laboratory result

	G/L	0 mth	1 mth	3 mth	6 mth	12 mth
HBsAg						
Anti-HBs	---					
HBeAg						
ALT level	---	---				
Bilirubin	---	---				

History, symptoms and signs

	0 mth	1 mth	3 mth	6 mth	12 mth
Jaundice					
Liver size					
Liver failure					

Transfusion, age, 1) _____ 2) _____ 3) _____ 4) _____ 5) _____

Drug with potential liver toxicity

Drug							
Age							

Other congenital liver diseases _____
Default, age _____
Death, age _____, cause _____