

Project Title: Studies on chronic type B, C and D viral hepatitis : molecular epidemiology, pathogenesis, natural history and treatment

Project Number: DOH96-DC-1018

Executing Institute: Hepatitis Research Center, National Taiwan University College of Medicine and National Taiwan University Hospital

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Abstract:

1. Study on the necessity of hepatitis B booster vaccination in adolescents

Universal hepatitis B vaccination in infancy has proved to be highly efficacious in preventing mother-infant transmission of hepatitis B. However, the duration of protection induced by hepatitis B vaccine remains unknown. Our earlier works have raised the concern that certain percentage of plasma-derived hepatitis B vaccinees would lose their vaccine-conferred protection 15-18 years after the neonatal vaccination. The purpose of this study was to test the reliability of the 2nd generation (recombinant) hepatitis B vaccine 13-14 years after the neonatal vaccination.

Up to the present, we have recruited 933 high school students aged 13-14 years. We have done several things for these students as follows. (1) We have checked their vaccination record from the NIIS. (2) We have tested their hepatitis B seromarkers including HBsAg, anti-HBs, and anti-HBc. (3) For those who were negative for HBsAg, anti-HBs, and anti-HBc, we have given them one dose of hepatitis B vaccine booster.

The current results showed vaccination records were available for 874 (93.7%) out of 933 study subjects. In subjects whose vaccination records were available, the hepatitis B vaccination rate (≥ 3 doses) was 95.8% (835/872). Within subjects with complete neonatal hepatitis B vaccination (at least 3 doses), HBsAg was positive in 0.2% (0-0.8%) subjects. Anti-HBc was positive in 1.7%. Anti-HBs was positive in 27.9%. Totally 72% of all subjects were negative for HBsAg, anti-HBs, and anti-HBc.

We have given a booster dose of hepatitis B vaccine to 543 subjects whose HBsAg, anti-HBs, and anti-HBc were all negative. 72.9% (396/543) of them developed protective anti-HBs (≥ 10 mIU/mL) after the booster. Still, 27.1% of them remained negative for anti-HBs. Considering some of these subjects were actually vaccine non-responders, we calculated 14.5% of the adolescents aged 13-14 years have lost hepatitis B vaccine-conferred protection. For those who remain seronegative, a second booster dose was given. 94% of these developed anti-HBs ≥ 10 mIU/mL.

We concluded here that 14.5% of adolescents who have received recombinant hepatitis B vaccine at infancy would have lost the vaccine-induced protection at aged 13-14 years old. However, as new

hepatitis B infection is rare at present, no booster vaccination is needed at present. In the following year, we will continue to give additional doses to those who failed to develop protective anti-HBs according to the standard vaccination schedule (0-1-6 months). We expect to understand how many doses of booster is optimal once booster vaccination is to be given.

2. The effect of HBIG on the prevention of HBsAg carriage and antibody responses in children born to HBeAg negative/HBsAg carrier mothers

A nationwide HBV vaccination program in Taiwan since 1984 has resulted in a significant decrease in the incidence of HBV carrier, hepatocellular carcinoma, and fulminant hepatitis in children. Chronic carriage rate of HBsAg in children has decreased to below 1% in the general population. In our current vaccination program, HBIG is not given to HBeAg negative/HBsAg carrier mother, which is different from some other countries where HBIG is given to all HBsAg carrier mother in addition to 3 doses of HBV vaccines. We have shown that fulminant hepatitis B has developed in a small number of infants born to HBsAg carrier mothers who were most likely to be HBeAg negative but not HBeAg positive. The benefits and risks of the two different vaccination policies have not been well-established. Recently many hospitals in Taiwan had provided choices of self-paid HBIG for infants born to HBsAg positive/HBeAg negative mothers and the impact on the vaccination efficacy has not been evaluated.

The current study aims to investigate the incidence and risk factors of breakthrough infection in HBeAg negative/HBsAg carrier mothers, focusing on the effect of HBIG. The antibody titer responses in infants with or without HBIG will also be evaluated. We will also analyze the current status of self-paid HBIG in hospitals and clinics in Taiwan.

The first part is a cohort study of children aged 0-10 years of HBsAg carrier mother, including children of HBeAg negative mother with or without HBIG, and children of HBeAg positive mothers who had received routine HBV immunization of three doses of HBV vaccine were collected for comparisons. We have collected 976 children aged 0-10 years of HBsAg carrier mothers, including 765 children of HBsAg(+)/HBeAg (-) mothers. Among them, 4 (0.5%) were tested positive for HBsAg, 9 (1.1%) positive for Anti-HBc, and 460 (60.1%) positive for Anti-HBs. Among the 765 children, 207 received self-paid HBIG within 24 hours after birth. The carrier rate were 0.6% and 0.5% in children receiving and not receiving HBIG, respectively. In 211 children of HBsAg (+)/HBeAg (+) children, 19 (9.0%) were tested positive for HBsAg, 27 (12.7%) for Anti-HBc, and 107 (50.7%) for Anti-HBs.

The second part is a prospective study. We have collected 45 mother-infant pairs since birth. Serum samples from mother and infant at age 6 and 12 months will be collected prospectively, including mother of HBsAg(+)/HBeAg (+) or HBeAg (-), receiving or not receiving HBIG. The carrier rate, frequency of liver dysfunction, and viral loads will be tested.

The present study will give important information especially in the outcome of infants born to HBeAg negative/HBsAg carrier mothers, the risk and clinical significant for breakthrough infection in these

children. The results will be useful in the revision of further vaccination programs.

3. Studies on chronic type B, C and D viral hepatitis : molecular epidemiology, pathogenesis, natural history and treatment

I. HBV:

1. Lower serum viral loads in young patients with hepatitis-B-virus-related hepatocellular carcinoma (J Viral Hepat. 2007 Mar;14(3):153-60.)

A total of 183 HBV-related HCC patients and 202 HBV carriers were enrolled to compare serum viral loads in young (≤ 40 years of age) and old (> 40 years of age) age groups. High serum HBV DNA level was associated with the development of HCC in old patients rather than in young patients. Thus, viral factors in association with the development of HBV-related HCC in young patients may be different from their old counterparts.

2. Two decades of universal hepatitis B vaccination in Taiwan: impact and implication for future strategies (Gastroenterology. 2007 Apr;132(4):1287-93.)

HBV seromarkers were studied in 18,779 subjects from neonates to adults below 30 years of age in 2004. The absence of an increase in HBsAg seropositive subjects at different ages in the same birth cohorts born after the vaccination program implied no increased risk of persistent HBV infection with aging. Thus, universal HBV vaccination provides long-term protection up to 20 years, and a universal booster is not indicated for the primary HBV vaccinees before adulthood.

3. Hepatitis B virus-related hepatocellular carcinoma: epidemiology and pathogenic role of viral factors (J Chin Med Assoc. 2007 Apr;70(4):141-5.)

Since the carcinogenic process involves the interplay between the hepatitis virus and the host hepatocytes, both genomes contribute to the final pathogenic outcome, either individually or synergistically. Studying the genetic factors predisposing hepatocarcinogenesis in both host and viral genomes will help illuminate the critical carcinogenic mechanisms, and create molecular targets for future therapy.

4. Hepatitis B viral factors in HBeAg-negative carriers with persistently normal serum alanine aminotransferase levels (Hepatology. 2007 May;45(5):1193-8.)

Baseline clinical and virological features of 414 HBeAg-negative carriers, including 176 (42.5%) with low-normal ALT (levels of less than 0.5x upper limit of normal) and 238 (57.5%) with high-normal ALT, were compared. Factors associated with a high-normal serum ALT level included male sex, increasing age and serum HBV DNA level $> 10^4$ copies/ml. Thus, HBeAg-negative patients with persistently normal ALT are not a homogenous group, and those with high-normal ALT share some of the characteristics that have been associated with adverse long-term outcomes.

5. To Genotype or Not To Genotype: Toward an Optimal Tailoring of Treatment of Chronic Hepatitis B (Clin Infect Dis. 2007 Jun 15;44(12):1665-6.)

6. Association of pre-S deletion mutant of hepatitis B virus with risk of hepatocellular carcinoma (J Gastroenterol Hepatol. 2007 Jul;22(7):1098-103.)

Pre-S deletion mutant of HBV were determined in 266 patients with chronic HBV genotype B or C infection. Hepatocellular carcinoma and genotype C were independently associated with the presence of pre-S deletion mutant. Thus, pre-S deletion mutant is more frequent in HBV carriers with genotype C infection, and those with pre-S deletion mutant may be associated with the development of HCC.

7. Appropriate use of interferon for treatment of chronic hepatitis B (Hepatol Res. 2007 Jul;37(s1):S47-S54.)

Individualized chronic hepatitis B treatment algorithms should be tailored to host (immune status, ALT level and genomic polymorphisms), virus (HBeAg status, HBV DNA level, genotype, precore/basal core promoter mutants and pre-S deletion mutant) as well as liver disease status (hepatitis activity and fibrosis stage).

8. Subgenotypes of hepatitis B virus genotype C do not correlate with disease progression of chronic hepatitis B in Taiwan (Liver Int. 2007 Sep;27(7):983-8.)

Subgenotypes of HBV/C were determined in 242 Taiwanese HBV carriers with various stages of liver disease. HBV/Ce was the predominant subgenotype in Taiwan. There was no significant difference in the distribution of the HBV/C subgenotypes among patients with different stages of liver disease. Thus, subgenotypes of HBV/C may not have a clinical impact on the disease progression of chronic hepatitis B in Taiwan.

9. Hepatocellular carcinoma in Taiwan. (Hepatol Res. 2007 Sep;37 Suppl 2:S101-5.)

Hepatocellular carcinoma (HCC) is common in Taiwan. The main causes are chronic hepatitis B and C infections, with >90% of patients positive for hepatitis B surface antigen (HBsAg) or antibody to hepatitis C virus (anti-HCV). Through efforts to control HBV and HCV infection, virally-induced HCC will be controlled in 20-30 years, and a decrease of approximately 85% is anticipated by 2040. Then, HCC will not be commonly seen in Taiwanese people.

10. High prevalence of occult hepatitis B virus infection in Taiwanese intravenous drug users (J Med Virol. 2007 Nov;79(11):1674-8.)

Molecular assays were used to determine the level of serum HBV DNA and the genotype in 304 intravenous drug users negative for both HBsAg and anti-HCV. Of 304 intravenous drug users, 125 (41.1%) were positive for serum HBV DNA. The amino acid sequence determination of HBV surface gene in 20 intravenous drug users with occult HBV infection selected at random showed no mutation of amino acid at codon 145. Thus, the prevalence of occult HBV infection and mixed HBV genotype infections are not uncommon in intravenous drug users residing in an HBV endemic areas.

II. HCV:

1. Peginterferon alfa-2a and ribavirin for 16 or 24 weeks in HCV (N Engl J Med. 2007 Oct 18;357(16):1661)

2. Metabolic Profiles in Patients with Chronic Hepatitis C: A Case-Control Study (in submission)

We enrolled 500 patients with chronic hepatitis C and 536 sex and age-matched controls. Stratifying ALT level according to its upper limit of normal, HCV infection was associated with younger age, female gender and higher TC levels in chronic hepatitis C patients with normal ALT levels, but with lower TC and lower TG levels in those with abnormal ALT levels. Presence of HCV infection was independently associated with higher serum adiponectin. Metabolic profiles of chronic hepatitis C patients are affected by age, gender, serum adiponectin and ALT levels.

Key words: Hepatitis B 、 long-term protection 、 newborn 、 hepatitis B immunoglobulin 、 molecular epidemiology 、 pathogenesis