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## REVIEW ARTICLE

# Perspectives and control of hepatitis B virus infection in Taiwan

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Hepatitis B virus (HBV) infection is endemic in Taiwan. After the implementation of universal hepatitis B vaccination, there was a significant reduction of hepatitis B surface antigen (HBsAg) seropositivity and HBV-related hepatocellular carcinoma (HCC) incidence in children, teenagers, and young adults. However, the incidence of HBV-related HCC in adults remains high. Through several community- and hospital-based cohort studies, the viral factors affecting the prognosis of HBV carriers have been illustrated. Serum HBV DNA level > 2000 IU/mL at study entry starts to increase the risks of cirrhosis and HCC in adult patients with chronic HBV infection. In addition, serum HBsAg level > 1000 IU/mL is associated with a higher risk of HCC in HBeAg-negative patients with low viral load. Virologically, HBV genotype C/D and core promote/pre-S mutations correlate with an increased HCC risk. Recently, a risk calculator has been developed to predict HCC in noncirrhotic patients with external validation. Therapeutically, hospital-based cohort and population-based nationwide studies indicated that interferon and nucleos(t)ide analogue treatments could reduce the incidence of HCC over time. Towards the ultimate goal of HBV eradication, several novel agents aiming at viral and host targets are under development. In addition, the immune therapy may play a key role in HBV cure in the foreseeable future.

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## Introduction

Hepatitis B virus (HBV) is one of the most common viral infections in humans. Global prevalence of chronic HBV infection is heterogeneous, with 2–20% of a given population being infected with HBV.<sup>1</sup> Persistent HBV infection has a wide disease spectrum, including inactive carrier state, chronic hepatitis, liver cirrhosis, and hepatocellular carcinoma (HCC).<sup>1</sup> The lifetime risk of patients with chronic HBV infection to develop cirrhosis, liver failure, or HCC is estimated to be as high as 15–40%.<sup>2</sup> Through previous investigations on the molecular epidemiology of HBV, several hepatitis B viral biomarkers, including HBV viral load, quantitative hepatitis B surface antigen (HBsAg), and HBV genotype are known to affect the long term outcomes of patients with chronic HBV infection.<sup>3,4</sup> The impact of hepatitis B viral biomarkers on the natural course of patients with chronic HBV infection has been explored in both community- and hospital-based cohort studies.<sup>5</sup> In addition, recent advances in anti-HBV treatment have documented the prevention of disease progression and reduction of HCC risk in patients with chronic HBV infection. In this review, the predictive roles of hepatitis B viral biomarkers on the clinical outcomes and challenges of anti-HBV therapy in Taiwan will be summarized and discussed.

## Changing burden of HBV infection and HBV-related HCC after HBV vaccination in Taiwan

HBV infection is endemic in Taiwan and most patients with chronic HBV infection acquire the virus early in life.<sup>6</sup> Before the launch of the universal hepatitis B vaccination in 1984, the prevalence of HBsAg in the general population of Taiwan was approximately 11–20%, which was the highest in the world.<sup>7,8</sup> After the implementation of universal hepatitis B vaccination, the HBsAg seropositive rate in children decreased dramatically from 11% to 0.9% in 2012.<sup>9</sup> Of particular note, the incidence of HCC in children also decreased from 0.52 per 100,000 children in those born before universal vaccination program to 0.13 per 100,000 in those born afterwards.<sup>10</sup> However, a recent analysis to determine the total and changing burden of chronic HBV infection and to evaluate the serological status between vaccinated and unvaccinated individuals in Taiwan was performed with participants of “The Taiwanese Survey on Prevalence of Hyperglycemia, Hyperlipidemia and Hypertension” in 2002 ( $n = 6602$ ), and 4088 participants had follow-up surveys in 2007. The results showed that the overall prevalence of chronic HBV infection was 13.7% and two thirds had past exposure (antibody to hepatitis B core antigen; anti-HBc, 68.46%) in 2002. The vaccinated cohort tended to have lower prevalence of HBsAg and anti-HBc, and higher proportion of anti-HBs and HBeAg positivity, genotype C and high viral load. In the vaccinated cohort, none acquired new exposure and became HBsAg positive, demonstrating the durability of vaccination through teenage and young adulthood. This comprehensive, population-representative survey indicated that 20 years after universal vaccination, the backlog still composed a substantial burden of chronic HBV infections in Taiwan.<sup>11</sup>

The etiological association between persistent HBV infection and HCC has been well documented, including the geographical correlation between the prevalence of HBsAg and the incidence of HCC, the high prevalence of HBsAg in HCC patients, the increased relative risk of HCC in HBsAg carriers, the presence of integrated HBV DNA in HCC tissue, and the association of chronic hepadnavirus infection with HCC in animal models.<sup>1,12</sup> The reduction of childhood HCC incidence after universal HBV vaccination in Taiwan and other countries further lends support to the fact that HBV-related HCC can be primarily prevented by HBV vaccination and it is thus the first cancer preventive vaccine.<sup>13</sup>

Despite effective immunoprophylaxis to interrupt mother-to-infant HBV transmission in our clinical practice, mothers positive for HBeAg with a high viral load are the major causes of immunoprophylactic failure. The estimated predictive rates of mother-to-infant transmission at maternal viral load levels of 5–9 log<sub>10</sub> copies/ml were 0.9%, 2.6%, 6.6%, 14.6%, and 27.7%, respectively.<sup>14</sup> To further reduce mother-to-infant HBV transmission, a prospective, multicenter trial enrolling 118 HBeAg-positive pregnant women with HBV DNA  $\geq 7.5$  log<sub>10</sub> IU/mL was conducted in Taiwan. The mothers received no medication or 300 mg tenofovir disoproxil fumarate (TDF) daily from weeks 30–32 of gestation until 1 month postpartum. The encouraging data showed that maternal TDF treatment could decrease infant HBV DNA level at birth, infant HBsAg positivity at 6 months, and ameliorate maternal alanine transaminase (ALT) elevations postpartum.<sup>15</sup>

Nevertheless, the annual incidence of HCC in the unvaccinated adult Taiwanese population remains high, ranging from 15/100,000 in the 1980s to approximately 30/100,000 in the 2000s.<sup>16</sup> Among these unvaccinated people, a large proportion of patients with chronic HBV infection are still at risk of developing cirrhosis and HCC if left untreated. Therefore, administration of effective antiviral agents to those in need, to slow disease progression is of paramount importance in clinical practice.

## Factors associated with long term outcomes from HBV cohort studies in Taiwan

### Hepatitis B viral load and quantitative HBsAg in predicting HCC risk

With results from several large cohort studies in Taiwan, the hepatitis B viral factors affecting the prognosis of patients with chronic HBV infection have been unveiled (Table 1).<sup>17–19</sup> The impact of baseline serum HBV DNA level on cirrhosis and HCC development in adult patients with chronic HBV infection was first assessed in the Risk Evaluation of Viral Load Elevation and Associated Liver Disease/Cancer-Hepatitis B Virus (REVEAL-HBV) study.<sup>17,20</sup> Among 3653 adult Taiwanese patients with chronic HBV infection over a mean follow-up period of 11.4 years, the cumulative incidence of HCC increased with serum HBV DNA levels in a dose dependent manner. The incidence increased from 1.3% to 14.9% in patients with an HBV DNA level of  $< 300$  copies/mL ( $\sim 60$  IU/mL) and  $\geq 10^6$  copies/mL ( $\sim 200,000$  IU/mL), respectively ( $p < 0.001$ ). The relative risk began to increase at an entry HBV DNA level of 2000 IU/

**Table 1** Summary of landmark hepatitis B virus cohort studies from Taiwan.

| Cohort          | REVEAL-HBV  | ERADICATE-B   | C-TEAM   | REACH-B   |
|-----------------|---|---|--|---|
| Study design    | Community-based cohort  | Hospital-based cohort   | Hospital-based cohort  | International collaboration   |
| Disease stage   | Including HBeAg-positive & -negative phases   | Including HBeAg-positive & -negative phases   | Compensated cirrhosis  | Including HBeAg-positive & -negative noncirrhotic patients  |
| No. of patients | 3653  | 2688  | Control group: 503<br>Entecavir treatment: 1123  | 4634  |
| Follow-up (y)   | 11.4  | 14.7  | Control group: 6.8<br>Entecavir treatment: 3.6   | -   |
| Main results    | Baseline serum HBV DNA level > 2000 IU/mL start to increase risk of cirrhosis & HCC | In HBeAg-negative patients with an HBV DNA level < 2000 IU/mL, an HBsAg level > 1000 IU/mL is associated with higher risks of hepatocellular carcinoma, cirrhosis, & HBeAg-negative hepatitis | Entecavir treatment was associated with 60% reduction of HCC risk (adjusted hazard ratio: 0.40, 95% CI: 0.27–0.60) in cirrhosis patients | A 17-point risk score of HCC, including 5 predictors (sex, age, serum ALT level, HBeAg status, & serum HBV DNA levels) was developed. The risk score precisely estimated the risk of developing HCC at 3 y, 5 y, & 10 y of follow-up. |

mL (hazard ratio; HR: 2.3; 95% confidence interval; CI: 1.1–4.9;  $p = 0.02$ ). Patients with HBV DNA levels of  $\geq 200,000$  IU/mL had the greatest risk (HR: 6.1; 95% CI: 2.9–12.1;  $p < 0.001$ ). In particular, the dose-dependent relationship was most marked in HBeAg seronegative patients with normal serum ALT levels and no cirrhosis at study entry.<sup>17,21</sup> In summary, the REVEAL-HBV cohort study revealed that HBV viral load is a strong risk predictor for HCC development in patients with chronic HBV infection, regardless of HBeAg status, ALT level, and other known risk factors.

As quantification of serum HBsAg becomes widely available, the association between HBsAg level and HCC was first addressed by the Elucidation of Risk fActors for Disease Control or Advancement in Taiwanese Hepatitis B Carriers (ERADICATE-B) hospital-based cohort study.<sup>19</sup> Similar to the findings derived from the REVEAL-HBV community-based cohort study, both elevated HBV DNA and HBsAg levels were positively associated with HCC development in a dose–response manner in patients with HBV DNA levels > 2000 IU/mL. Particularly, among 1068 HBeAg-negative patients with HBV DNA levels < 2000 IU/mL, the HCC risk significantly increased in those with HBsAg level  $\geq 1000$  IU/mL compared with those with HBsAg level < 1000 IU/mL (HR:5.4; 95% CI:2.1–14.2). Multivariate analysis revealed that a HBsAg level  $\geq 1000$  IU/mL was an independent risk factor for HCC development (HR:13.7; 95% CI:4.8–39.3). Furthermore, the relationship of HCC risk with dynamic changes of serum HBV DNA, HBsAg, and ALT levels was evaluated in the ERADICATE-B cohort. Compared to patients with persistently low HBV DNA, HBsAg, or ALT levels, those with persistently high levels of these three factors were at a higher risk of HCC over time. The recent update of the REVEAL-HBV cohort also confirmed that the HBsAg level was complementary to the HBV DNA level in stratifying HCC risks, especially in patients with HBV DNA

level < 200,000 IU/mL.<sup>22</sup> Taken together, both cohorts firmly suggest the promising predictive role of HBsAg in HBV patients with low viral load.

### HBV genotype/variants and risk of cirrhosis/HCC

At least 10 HBV genotypes (A–J) have been defined virologically, and HBV genotype has been documented to have specific geographic distribution and impact on the long term outcomes of HBV infection.<sup>23</sup> Two community based prospective cohort studies of Taiwanese patients with chronic HBV infection demonstrated that HBV genotype C was associated with an increased risk of cirrhosis/HCC compared to genotype B. For example, genotype C-infected patients who also had a very high viral load had a 26-fold higher risk of HCC than those with other genotypes and low or undetectable viral loads.<sup>24,25</sup> The ERADICATE-B cohort study also showed genotype C patients had a higher annual incidence rate of HCC than genotype B patients by univariate analysis.<sup>19</sup> These findings confirmed that genotype C correlates with a higher risk of HCC development than genotype B.

Of naturally occurring HBV mutants, several mutations in the X gene of the HBV genome are frequently found in patients with HBV-related HCC.<sup>26–29</sup> The 3'-end of the X gene is frequently deleted in HCC cells, leading to a C-terminal truncated HBx protein, and may contribute to hepatocarcinogenesis via the activation of cell proliferation and loss of proapoptotic ability.<sup>29–31</sup> Several cross-sectional studies revealed that dual mutations in basal core promoter (BCP) A1762T/G1764A were strongly associated with the risk of HCC development.<sup>32–36</sup> The REVEAL-HBV cohort study further confirmed BCP A1762T/G1764A mutations as an independent predictor for HCC development (HR:1.73; 95% CI:1.13–2.67;  $p = 0.013$ ).<sup>24</sup> A subgroup analysis from the ERADICATE-B cohort showed that a higher proportion of

BCP mutations increased the risk of cirrhosis development in HBV patients with genotype B or C infection.<sup>37</sup> For example, the risk of cirrhosis was higher in patients with BCP mutations  $\geq 45\%$  compared to  $< 45\%$  in the longitudinal cohort.

Previous reports also showed that the deletion mutations within the pre-S gene were significantly associated with the development of cirrhosis and HCC.<sup>38–40</sup> HBV pre-S deletion mutations may lead to defective immunity against HBV and contribute directly to progressive liver cell damage and ultimately hepatocarcinogenesis.<sup>41</sup>

### Risk calculator for HBV-related HCC in treatment-naïve chronic hepatitis B patients

Because HCC is the most common cause of mortality in patients with chronic HBV infection, a simple formula with different weights for different clinical and virological variables may help clinicians predict the risk of HCC development in patients with chronic HBV infection. Based on the REVEAL-HBV cohort along with international collaboration, the Risk Estimation for Hepatocellular Carcinoma in Chronic Hepatitis B study (REACH-B) developed and validated a predictive score for the risk of development of HBV-related HCC.<sup>42,43</sup> This study included a risk score development cohort with 3584 noncirrhotic, chronic hepatitis B (CHB) patients without antiviral treatment (REVEAL-HBV cohort) and a validation cohort with 1050 patients from three independent hospitals in Hong Kong and South Korea. The 17-point risk score included five predictors of HCC, including sex, age, serum ALT level, HBeAg status, and serum HBV DNA levels (SALED). The risk score precisely estimated the risk of developing HCC at 3 years, 5 years, and 10 years of follow-up in the validation cohort. Additional receiver operating characteristic (ROC) curves and calibration charts also confirmed the predictive value of this risk score in noncirrhotic patients. The areas under an ROC curve (AUROCs) for predicting 3 year, 5 year, and 10 year HCC risk were 0.811 (95% CI: 0.790–0.831), 0.796 (95% CI: 0.775–0.816), and 0.769 (95% CI: 0.747–0.790), respectively.<sup>43</sup>

Although the REACH-B risk calculator for HCC has been externally validated in noncirrhotic CHB patients, this scoring system may underestimate the risk in patients with low viral load at baseline. Based on the ERADICATE-B cohort, HBsAg level is known to be a complementary marker for HCC risk in the low viral load group.<sup>19</sup> In the ERADICATE-B subcohort study, a total of 2165 Taiwanese HBeAg-negative non-cirrhotic patients were followed for 14.9 years. There was no association between HBsAg level and HCC in patients with higher viral loads ( $\geq 20,000$  IU/mL). HBsAg level was then included to stratify the risk of HCC in patients with low ( $< 2000$  IU/mL) and intermediate viral loads (2000–19,999 IU/mL). ROC curve analysis showed that combining HBV DNA and HBsAg level improved the prediction of the development of HCC at 10 years compared to the HBV DNA level alone in patients with low ( $< 2000$  IU/mL) and intermediate viral loads (2000–19,999 IU/mL) as well as the overall cohort ( $p = 0.004$  and  $p = 0.028$ , respectively).<sup>44</sup> In the recent update of the REVEAL-HBV study, HBsAg level was

incorporated into the HCC risk prediction model.<sup>22</sup> Predictive factors of risk included age, sex, family history of HCC, HBeAg status, serum HBV DNA, ALT, HBsAg levels, and HBV genotype. The sum risk scores ranged from 0 to 19 in the HCC prediction model. Patients were categorized by their sum risk scores into low risk (risk score  $< 9$ ), medium risk (risk score 9–12), and high risk (risk score  $\geq 13$ ) groups. The observed cumulative risk of HCC for the high risk group was significantly higher than in the low risk and medium risk groups ( $p < 0.001$ ).<sup>22</sup> Although the predictive accuracy of the upgraded REVEAL-HBV risk calculator for HCC was excellent in noncirrhotic CHB patients, its cost-effectiveness needs to be surveyed.<sup>45</sup>

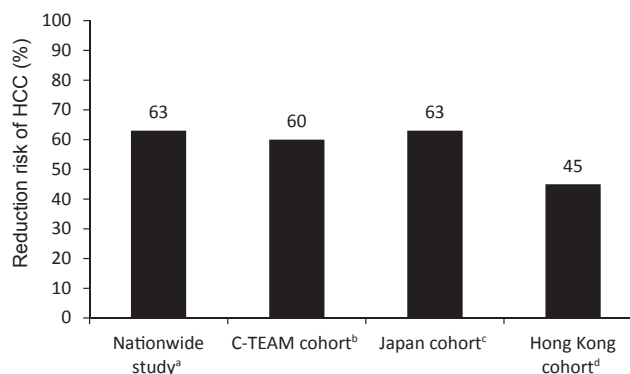
### Impact of antiviral therapy on long-term outcomes of CHB

Cirrhosis with necroinflammation, deposition of extracellular matrix and shortening of telomeres, leads to cellular senescence and regeneration nodule. HCC may subsequently emerge in the setting of cirrhosis.<sup>46</sup> The most effective tool to prevent the occurrence of HCC in patients with chronic HBV infection is to receive effective antiviral agents and halt the disease progression from chronic hepatitis to cirrhosis and eventually HCC.<sup>47,48</sup> At the time of writing, seven agents are globally approved for the treatment of CHB, they are standard interferon- $\alpha$  (IFN- $\alpha$ ) or pegylated interferon- $\alpha$  (PEG-IFN- $\alpha$ ) and five nucleos(t)ide analogues (NAs), including lamivudine (LAM), telbivudine (LdT), entecavir (ETV), adefovir dipivoxil (ADV), and tenofovir disoproxil fumarate (TDF).<sup>49–52</sup> Previous studies revealed that long term NA therapy resulted in the improvement of histologic necroinflammation as well as reversal of advanced fibrosis or even cirrhosis.<sup>53–56</sup> Earlier randomized control studies showed that IFN and LAM therapy were protective against HCC development in patients with chronic HBV infection.<sup>57,58</sup> In a meta-analysis including six eligible studies (3644 patients in total), the incidence of long term complications (including decompensated cirrhosis, CHB-related death, and HBV-related HCC) in patients receiving NAs (mainly of LAM and ADV) as treatment was reduced by 74% (relative risk; RR:0.26, 95% CI: 0.15–0.47) compared to patients without NA treatment.<sup>59</sup> Similarly, another meta-analysis including 11 studies (2122 patients in total) reported that IFN therapy reduced the risk of cirrhotic complications by 54% (RR:0.46, 95% CI:0.32–0.67,  $p < 0.001$ ).<sup>60</sup> Another meta-analysis also confirmed that the risk of HCC after IFN and NA (mainly of LAM and ADV) treatment was reduced by 34% (RR: 0.66, 95% CI: 0.48–0.89) and 78% (RR: 0.22, 95% CI: 0.10–0.50), respectively.<sup>61</sup> However, there is no large randomized clinical trial to show a more beneficial effect of NAs with high potency as well as a high barrier to drug resistance (ETV and TDF) than earlier generation NAs (LAM and ADV) in the prevention of HBV-related HCC. A small scale study for propensity score matching CHB patients with and without ETV treatment revealed that the cumulative HCC incidence rates at 5 years were 3.7% and 13.7% for the ETV and control groups, respectively ( $p < 0.001$ ). The ETV group were less likely to develop HCC than those in the control group (HR: 0.37; 95% CI: 0.15–0.91;  $p = 0.030$ ).<sup>62</sup> Similarly, Wong

et al<sup>63</sup> reported that cirrhotic patients with ETV treatment had reduced risks of HCC when compared with historical untreated patients with cirrhosis (HR:0.55; 95% CI, 0.31–0.99;  $p = 0.049$ ). The effect of long term ETV therapy on the reduction of HCC risk in hepatitis B-related cirrhosis patients was also investigated in the Cirrhosis Taiwanese Entecavir Multicenter (C-TEAM) study in Taiwan. In this large hospital-based cohort study, 503 patients in the control group and 1123 in the ETV group from 24 academic centers were enrolled. ETV treatment was associated with a 60% reduction of HCC risk (adjusted HR: 0.40, 95% CI: 0.27–0.60) in cirrhosis patients.<sup>64</sup> The protective effect of NAs in the development of HCC was further confirmed in a retrospective, population-based nationwide study in Taiwan. CHB patients treated with NAs had a significantly lower 7 year incidence of HCC (7.32%; 95% CI: 6.77%–7.87%) than patients without NA treatment (22.7%; 95% CI: 22.1%–23.3%;  $p < 0.001$ ). NA treatment was associated with a reduced risk of HCC, with an adjusted hazard ratio of 0.37 (95% CI, 0.34–0.39;  $p < 0.001$ ).<sup>65</sup> In addition, the nationwide study in Taiwan also revealed that NA treated HCC patients had a significantly lower HCC recurrence rate after liver resection (45.6%; 95% CI: 36.5%–54.6% vs. untreated, 54.6%; 95% CI: 52.5%–56.6%;  $p < 0.001$ ). NA treatment was independently associated with a reduced risk of HCC recurrence (HR:0.67; 95% CI: 0.55–0.81;  $p < 0.001$ ).<sup>66</sup> Through long term viral suppression, NA treatment reduced the risk of HCC development in cirrhotic patients (Figure 1).<sup>62,63,64,65</sup> However, HCC still occurred in some patients during virological remission.<sup>47</sup> A recent systematic review indicated that current NA therapy consistently resulted in a significantly lower HCC incidence in Asian patients with cirrhosis, an overall HCC risk reduction of 30%; in noncirrhotic patients, HCC risk reduction was 80% overall in some studies. For Caucasian patients, no appropriate comparative studies are available to evaluate the impact of NA treatment on HCC. In addition, achievement of viral suppression under current NA therapy was associated with a lower HCC risk in Asians, but not Caucasians. Studies comparing ETV or TDF with older NAs generally found no difference in HCC risk reduction between agents.<sup>67</sup> Overall, these data imply that current potent NAs can reduce HCC but not eliminate it. Thus HCC surveillance remains mandatory in treated patients, and new treatment strategies to eradicate intrahepatic HBV cccDNA are urgently required for patients who are at risk of developing cirrhosis and HCC.

### Hidden menace: HBV reactivation from immunosuppression

Acute flares in the natural course of chronic HBV infection are not unusual. Pathogenesis of acute flares is related to the interactions between virus and host immune responses to HBV-encoded antigens.<sup>68</sup> Therefore, patients who have been infected with HBV, including HBsAg-positive carriers and those with resolved HBV infections with or without anti-HBs, are vulnerable to increased viral replication and widespread infection of hepatocytes during immunosuppressive therapy or chemotherapy. After discontinuation of immunosuppressive therapy or chemotherapy, restored



**Figure 1** Nucleos(t)ide analogues reduce risk of hepatocellular carcinoma in chronic hepatitis B patients in population-based and hospital-based cohort studies in Asia. <sup>a</sup> Nationwide study: Population-based nationwide cohort study in Taiwan.<sup>65</sup> <sup>b</sup> C-TEAM cohort: Cirrhosis Taiwanese Entecavir Multicenter study in Taiwan, a hospital-based cohort study.<sup>64</sup> <sup>c</sup> Japan cohort: A hospital-based cohort study in Japan.<sup>62</sup> <sup>d</sup> Hong Kong cohort: A hospital-based cohort study in Hong Kong.<sup>63</sup> HCC = hepatocellular carcinoma.

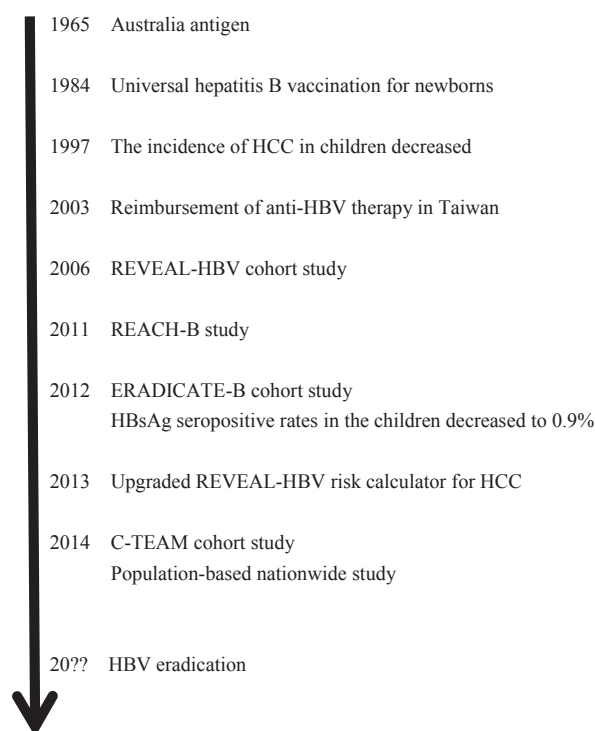
immune competence will respond to reactive HBV and lead to hepatitis.<sup>69–71</sup> HBV reactivation is well recognized in patients with malignancies following chemotherapy,<sup>72</sup> as well as in patients with nonmalignant diseases after treatment of immunosuppressive therapy or biologic agents.<sup>73,74</sup> The clinical manifestations of HBV reactivation in patients receiving immunosuppressive therapy or chemotherapy varies from asymptomatic hepatitis flares to fatal liver failure. Furthermore, anticancer therapy may be interrupted due to hepatitis flares and affect the prognosis of cancer. There is ample evidence to support the use of prophylactic antiviral therapy to avoid HBV reactivation from immunosuppression.<sup>75–79</sup> In the international guidelines for the management of HBV infection, prophylaxis with an antiviral agent before the start of chemotherapy or immunosuppressive agent is recommended.<sup>49–51</sup> However, patients who have a resolved HBV infection as reflected by HBsAg negativity with or without anti-HBs and hepatitis B core antibody positivity also have a risk of HBV reactivation when host immunity is severely compromised by chemotherapy or immunosuppressive agents. In a prospective study that included 150 lymphoma patients with resolved HBV infection who received rituximab-based chemotherapy, the incidence of HBV reactivation and HBV-related hepatitis flares was 10.4 per 100 people/y and 6.4 per 100 people/y, respectively. Patients with hepatitis flare exhibited a significantly higher incidence of reappearance of HBsAg after HBV reactivation (100% vs. 28.5%;  $p = 0.003$ ).<sup>80</sup> The role of antiviral prophylaxis in preventing HBV reactivation before rituximab-based chemotherapy in patients with lymphoma with resolved hepatitis B was elucidated in a randomized controlled trial in Taiwan.<sup>81</sup> In this study, 80 patients with lymphoma and resolved hepatitis B undergoing chemotherapy were randomly assigned to prophylactic ETV therapy or therapeutic ETV at the time of HBV reactivation. During a mean of 18 months follow-up period, the ETV prophylactic group had a significantly

lower rate of HBV reactivation than the control group (2.4% vs. 17.9%,  $p = 0.027$ ). The cumulative HBV reactivation rates at 6 months, 12 months, and 18 months after chemotherapy were 8%, 11.2%, and 25.9%, respectively, in the control group, and 0%, 0%, and 4.3% in the ETV prophylactic group ( $p = 0.019$ ).<sup>81</sup> Taken together, accumulating evidence strongly recommends that all patients should be screened for evidence of HBV infection prior to chemotherapy or immunosuppressive treatment. The prophylactic use of NAs can effectively prevent HBV reactivation and diminish the risk of severe or fatal HBV reactivation.<sup>79,82</sup>

## Perspectives in Taiwan: Towards HBV eradication

Although the efficacy of antiviral therapy has dramatically improved the long term outcomes of CHB, clearance of HBsAg is only observed in a small portion of Asian patients with chronic HBV infection. For example, 1 year PEG-IFN monotherapy led to HBsAg seroconversion in 3–5% of patients at 6 months of therapy.<sup>83–85</sup> Similarly, the HBsAg loss rates for HBeAg-positive patients treated with ETV or LAM for 2 years were 5% and 3%, respectively.<sup>86</sup> Although HBsAg loss could be achieved in 11.8% of HBeAg-positive patients after 7 years of TDF therapy, it only occurred in Caucasian patients with genotype A or D infection.<sup>87</sup> Therefore, eradication of HBV infection is still a daunting challenge ahead, especially in Asian patients who acquire the virus early in life. Recently, several potential strategies for HBV eradication aimed at viral and host targets responsible for HBV persistence have been developed.<sup>88</sup> Several novel therapeutic approaches will be briefly introduced. The first one, Myrcludex-B, a lipopeptide derived from the pre-S1 domain of the HBV envelope protein, which targets the HBV receptor, the sodium taurocholate co-transporting polypeptide (NTCP),<sup>89</sup> has been shown to efficiently inhibit HBV entry.<sup>90</sup> The second one is zinc-finger nuclease (ZFN). Through blocking transcription of cccDNA, ZFN can be used to inhibit viral transcription and replication of duck HBV.<sup>91</sup> *In vitro* studies also revealed effective cleavage of viral DNA targets by HBV-specific ZFN within cultured cells.<sup>92</sup> Lucifora et al<sup>93</sup> recently reported that IFN- $\alpha$  and lymphotoxin- $\beta$  receptor activation upregulated apolipoprotein B mRNA editing enzyme, catalytic polypeptide 3A (APOBEC3A) and APOBEC3B cytidine deaminases in HBV infected cells, resulting in noncytolytic clearance of cccDNA and preventing HBV reactivation.

Agents targeting host factors to enhance innate and adaptive immune responses may play an important role in elimination of HBV infected cells. The toll-like receptor (TLR) family is an important regulator of innate and adaptive immune responses to various pathogens.<sup>94</sup> Exogenous interferon stimulation by TLR agonists may reinstate endogenous IFN- $\alpha$  responses and result in innate and adaptive immune reconstitution.<sup>95</sup> GS-9620, a selective oral TLR7 agonist, could induce prolonged suppression of HBV DNA in the serum and liver in chronically infected chimpanzees. Furthermore, serum HBsAg and HBeAg levels, and numbers of HBV antigen-positive hepatocytes, were reduced while hepatocyte apoptosis were increased.<sup>96</sup>



**Figure 2** Strategies and achievements for the control of hepatitis B disease in Taiwan. HBsAg = hepatitis B surface antigen; HBV = hepatitis B virus; HCC = hepatocellular carcinoma.

Antiviral immunity plays an important role in the control of HBV infection, and therapeutic vaccines have been considered as a promising strategy. Martin et al<sup>97</sup> developed a novel adenovirus-based therapeutic vaccine, TG1050, encoding three HBV antigens or domains, including core, polymerase, and envelope proteins. They demonstrated that long-lasting HBV-specific memory CD8 T cells could be induced by TG1050 in mouse models.<sup>97</sup> Although reduction of HBsAg and HBV DNA levels could be observed in mouse models, the magnitude of reduction might not be enough to eradicate persistent HBV or control viral replication in humans.<sup>98</sup> In the future, the new agents in conjunction with long term NA therapy may lead to the development of protective immunity against HBV with subsequent eradication of HBV.<sup>88,95</sup>

## Conclusion

Effective vaccines against HBV infection have been available for more than three decades, and the effectiveness of universal hepatitis B vaccination to decrease hepatitis B carriage and HCC in children and young adults is confirmed by the long term follow-up survey.<sup>12</sup> In addition, HBV factors affecting liver disease remission or progression have been elucidated and risk calculator for HBV-related HCC has been developed and validated from HBV natural history cohorts from Taiwan. Ample evidence also indicates current anti-HBV therapies significantly reduce the risk of HBV-related cirrhosis and HCC (Figure 2). However, CHB still remains a major global health threat because of the

existence of 240 million patients with chronic HBV infection across the world. The recent publication of "Guidelines for the prevention, care and treatment of persons with chronic hepatitis B infection" by the World Health Organization further highlights this important health issue.<sup>99</sup> With the development of more effective measures, the ultimate goal of a HBV cure can be achieved in the foreseeable future.

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