Viral hepatitis B and hepatocellular carcinoma

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Abstract

Hepatocellular carcinoma (HCC) is the sixth most common cancer in the world, some 630,000 new cases being diagnosed each year. 82% of cases are related to viral hepatitis, 55% to hepatitis B virus (HBV), 89% of those in regions where HBV is endemic. There is a striking parallel between the geographical distribution of the rates of chronic HBV infection and that of HCC. In the majority of HCC cases (70-90%) there is underlying liver cirrhosis. However, because HBV is an oncogenic virus, it can cause HCC in the absence of cirrhosis. The annual risk of HBV-induced HCC varies according to the presence or absence of concomitant cirrhosis. In HBV carriers without cirrhosis, the risk is 0.02-0.3% in Caucasians and 0.4-0.6% per year in Asians. In those with cirrhosis, the risk is 2.2% and 3.7% respectively in Caucasians and Asians.

HBV likely causes HCC via both indirect (necro-inflammation and regeneration injury) and direct (by integration of its DNA in the host genome) pathways.

During recent years it has become evident that HBV viral load > 2000 IU/mL is associated with a high risk of malignant transformation.

The most effective measure of prevention of HBV-related HCC is prevention of HBV infection by vaccination. A universal vaccination program in Taiwan was shown to be effective in reducing the rate of childhood and early adulthood HCC. In patients already infected with HBV, antiviral therapy remains the best strategy. Interferon-alfa therapy appears to be effective in preventing HCC in cirrhosis in Asia but not in Europe. Medium-term nucleos/tide-analogue therapy significantly reduces but does not eliminate the risk of HCC, especially in patients with pre-existing cirrhosis. Maintenance of virological remission is important for the reduction of HCC risk. With more potent antiviral drugs currently available (entecavir, tenofovir), long-term HBV DNA suppression is now possible with very low risk of drug resistance. (Acta gastroenterol. belg., **2011**, 74, **4-8**).

Key words : hepatitis B, natural history, hepatocellular carcinoma, interferon, nucleos(t)ide analogues.

Introduction

Hepatocellular carcinoma (HCC) is the sixth most common cancer in the world (the fifth in males, the eighth in females), and the third most common cause of cancer death. Because of its poor prognosis, the number of deaths per year (in 2002 : 598,000) almost equals the number of cases diagnosed each year (in 2002 : 625,000) (1). HCC does not have a uniform geographical distribution. Eighty per cent of all cases occur in developing countries. Regions with the highest incidences are eastern and southeastern Asia, Melanesia, sub-Saharan Africa and the Amazon basin. Age-adjusted incidences of HCC per 100,000 persons per year in developing countries (17.4 and 6.7 resp. in males and females) are double than those in the developed world (8.7 and 2.8 respectively in males and females) (2). The geographical distribution of HCC is determined by the distribution of its major causes. The regions with the highest incidences of the tumour are those that are endemic for hepatitis B virus (HBV) infection. The close association between HBV infection and the development of HCC was first reported in 1975 (3). HBV is now recognized as the predominant cause of HCC : 55% of all HCC worldwide, 89% of HCC in regions where the virus is endemic or hyperendemic. Of the 360 million global carriers of HBV as many as one quarter will develop HCC (4). Countries with low incidences of HCC have low rates of HBV carriage but other risk factors associated with HCC e.g. hepatitis C, alcohol and metabolic syndrome (4).

Risk factors for HBV-related HCC

Both host and viral factors have been associated with the risk of HCC among people with chronic hepatitis B infection.

Host factors

- Gender : Chronically infected males are at a greater risk than are females. In Taiwan the adjusted relative risk for developing HCC in males in comparison with females was 3.6 (5).
- Age : In most populations older age (≥ 40 y) is strongly associated with HCC risk (6). One exception is black Africans : 73.5% of HCC patients < 30 y have HBV-induced tumours compared with 28.6% of those > 60 y (7).
- Concomitant cirrhosis : In the majority of HBV-related HCC cases (80-90%) there is underlying liver cirrhosis (8). However, because HBV is an oncogenic virus, it can cause HCC in the absence of cirrhosis. In studies conducted in East Asian countries, the summary HCC incidence rate ranged from 0.2 per 100 person years among inactive carriers to 0.6 in persons with chronic hepatitis B but without cirrhosis and 3.7 in subjects with compensated cirrhosis ; the

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corresponding 5-year HCC cumulative incidences were 1%, 3% and 17%. In studies performed in Europe and in the United States, the summary HCC incidence rate was 0.02 per 100 person years in inactive carriers, 0.3 in subjects with chronic hepatitis B without cirrhosis and 2.2 in subjects with compensated cirrhosis ; the corresponding 5-year HCC cumulative incidences were 0.1%, 1% and 10% (9).

- Co-morbidity such as excessive alcohol consumption and metabolic factors such as obesity or diabetes (10,11).
- Aflatoxin : A synergistic interaction between HBV and the fungal toxin aflatoxin B1 has been proven in sub-Saharan Africa, China, Taiwan, Thailand, but aflatoxin B1-induced HCC has not been reported in regions where chronic hepatitis B is rare (12).

Viral factors

- HBV replication status, particularly HBV viral load, is a very strong predictor of HCC. Viral loads $> 10^4$ copies/mL (approx. 2000 IU/mL) are associated with a high risk of malignant transformation. In the REVEAL study from Taiwan, Chen *et al*. showed that the cumulative number of HCC cases increased significantly if the HBV DNA was equal to or exceeded 10^5 copies/mL (ca 20,000 IU/mL) compared to carriers whose viral load was less than 10^4 copies/mL. The number of cases was intermediate for intermediate viral loads (13). A case-control study in The Gambia also showed that HBV DNA levels are strongly associated with HCC risk, and that the risk is notably increased at HBV DNA levels $\geq 10^4$ copies/mL (14).
- HBV genotype is also associated with HCC. The risk of HCC is higher in subjects with genotype C compared with genotype B in the Asia-Pacific region (15).
- HBV and HCV coinfection may synergistically increase the risk of HCC (16,17). The data on HDV and HIV coinfection are not clear concerning possible increased risk for HCC.

Irrespective of the severity of the underlying liver disease, the risk of HCC in persons with HBV infection is higher in East Asian countries than in Western countries, possibly because of earlier acquisition of the infection, longer duration of disease and/or exposure to environmental factors (9).

HBV carcinogenesis

Carcinogenesis of HCC is considered to be a multistep process. It is postulated that HBV infection causes HCC via direct and indirect pathways.

- Indirect : Continual necrosis of hepatocytes accompanied by regeneration may lead to accumulation of mutations and selection of cells with malignant phenotype (18).
- Direct : HBV belongs to the oncogenic hepadnaviruses.
  - HBV is able to integrate its DNA into the genome of the infected cell. This integration may give a selective growth advantage to the target cells and lead to tumour progression or lead to chromosomal instability (19). However, in many cases the HBV DNA integration site does not appear to be in a critical location and the process appears to be at random (20), and the integration near known oncogenes appears to be a rare event.
  - In a large proportion of tumor cells, sequences encoding for the HBx protein and/or truncated preS2/S proteins are found. HBx is known to be a transcription activator. It can interfere with regulatory elements and so interfere with the hepatocyte DNA repair system and controlling elements of cellular proliferation (21). It can also bind with p53 with subsequent inhibition of p53-mediated apoptosis (22). Truncated preS2/S encodes a group of regulatory proteins called transcription activators which after phosphorylation activate pathways causing increase in hepatocyte proliferation (23).

Surveillance for HCC

Because individuals with chronic hepatitis B infection can be readily identified and because they are known to be at risk for HCC, surveillance programs have been instituted in order to identify HCC at an early stage when more treatment options are available. The AASLD recommends surveillance with liver ultrasound at six month intervals in following groups of HBsAg carriers (24) :

- Asian males > 40 y
- Asian females > 50 y
- Those born in Africa > 20 y
- All patients with cirrhosis
- Those with a family history of HCC
- Patients with high serum HBV DNA and ongoing liver injury

Prevention of HCC related to HBV

Three levels of prevention can be considered :

- Primary prevention represents treating or preventing the diseases leading to cirrhosis. This can be established through vaccination and medical antiviral treatment.
- Secondary prevention is the prevention of a first occurrence of HCC once cirrhosis is established.
- Tertiary prevention refers to treatments that aim at preventing a second occurrence of HCC after curative treatment of a first tumour.

Vaccination

A vaccine protecting against HBV infection was invented in 1969 and is now one of the most commonly used vaccines in the world. National vaccination programs have dramatically reduced the prevalence of HBV infection and carriers with a concomitant decrease in the incidence of HCC in the vaccinated populations. HBV vaccine is therefore the first widely used cancer prevention vaccine. Many countries have introduced universal infant vaccination, particularly in regions in which there is a high incidence of HCC.

One of the best examples is Taiwan where universal infant vaccination was introduced in 1984. This national program has been shown to be associated with a decrease in the rate of HBsAg positivity from 9.8% in 1984 to 4.8% in 1989, 1.3% in 1994 and 0.7% in 1999, remaining around that level in 2004 (25). In conjunction with this decrease, a significant reduction in the incidence of childhood HCC occurred : the rate of childhood HCC (between 6-14 y) was 0.7/100,000 in the early 1980s, whereas between 1996 and 1999 it fell to 0.19/100,000 (26,27). It was recently shown that the incidence rate of HCC in children and adolescents aged 6-19 y was significantly reduced for those born after the initiation of the vaccination program. These data suggest that the effectiveness of the universal HBV immunization program to prevent HCC has extended beyond childhood into young adulthood over the past two decades (28).

A similar study in Korea including adults showed results in the same direction. The risk ratio for HCC among those who were not vaccinated and did not have natural protection was 18.1 compared to unvaccinated and initially uninfected individuals. It was 0.58 in the vaccinated group and 0.34 in those with naturally acquired immunity (29).

Antiviral treatment

Current therapeutic options for patients with chronic HBV infection may be summarized into treatment with standard or pegylated interferon alpha (IFN-α), a drug with antiviral, immunomodulatory and perhaps antitumoural activities, and treatment with nucleoside or nucleotide analogues (30).

IFN-α

– Noncirrhotics : 3 studies of HBeAg positive chronic hepatitis B treated with a 4-6 month course of IFN-α therapy showed a reduced risk of HCC in IFN-α-treated patients (31-33). By contrast a study from Hong Kong did not show a long-term benefit of IFN-α therapy in the prevention of HCC (34). The long-term benefit of IFN-α therapy in HBeAg negative patients is less clear. A six-year follow-up study from Greece showed a significant reduction in incidence of HCC in patients with a sustained response over non-sustained responders or untreated patients (35). By contrast, a similar benefit in sustained responders was not observed in an Italian cohort study (36).
– Cirrhotics : Seven non-randomized controlled studies investigated the possible effect of IFN-α treatment on the development of HCC in patients with already established cirrhosis (37-43). A meta-analysis on these studies (44) suggested that IFN-α therapy decreased the incidence of HCC. The difference was significant in only 3 trials. The pooled estimate was significantly in favour of the preventive effect of IFN-α therapy. The inconsistency among the trials was a major problem in reaching a firm conclusion. Subgroup analysis showed no preventive effect in the European patients, perhaps because of the lower incidence of HCC development in Europe vs. Asia. In two of the studies reduced risk of HCC was strongly associated with virological response suggesting that arrest of the viral replication is a critical factor.

Oral antiviral therapy

Long-term data on the effect of direct antiviral agents are limited.

In what has become recognized as a landmark study, the impact of lamivudine was tested in a randomized controlled study in patients with advanced fibrosis or cirrhosis due to hepatitis B, 436 patients receiving lamivudine, 215 placebo. The trial was terminated after 36 months based on significant differences in clinical outcomes between the two groups. Lamivudine treatment was associated with fewer cases of HCC vs placebo (7.4% vs 3.9%, $p = 0.047$) (45).

Another study of lamivudine treatment in 353 patients with chronic hepatitis B and 303 patients with cirrhosis, the risk of HCC was threefold higher among patients who had virological breakthrough (46).

A systematic review (47) shows that the medium-term risk of HCC is significantly lower in chronic hepatitis B patients receiving effective oral antiviral therapy. Among the treated patients, HCC developed significantly more frequent in patients with cirrhosis compared to patients without cirrhosis. In the patients with lamivudine resistance, HCC was reported to develop exclusively in cirrhotics. In patients under rescue therapy, HCC was detected in 20.2% of patients with viremia vs 5.9% in patients in virological remission ($p < 0.001$).

It should be concluded that medium-term nucleos(t)ide analogue therapy significantly reduces but not completely eliminates the risk of HCC, especially in patients with preexisting cirrhosis. HCC surveillance (*vide supra*) should be continued. Maintenance of virological remission is important for the reduction of HCC risk. He HCC risk seems increased in patients who experience virological breakthrough.

It is anticipated that the newer nucleos(t)ide analogues (entecavir, tenofovir) will further reduce HCC incidence given their greater potency and better resistance profiles.

Tertiary prophylaxis

Hepatic resection or transplantation can provide a complete cure of HCC. Postoperative recurrence is common and is the main cause of death in HCC patients after resection. Most recurrences occur within 2 years of surgery (early recurrence). Factors linked to early recurrence are more closely related to those predisposing to tumour progression or metastases. These tumours were more likely to show the same clonal origins as the original tumour, suggesting that they had their metastatic origin shortly after the operation. Late recurrence (> 2 years after surgery) often shows clonal origins different from the original tumours, suggesting *de novo* second primary HCC. Factors linked to late recurrence are more related to factors predisposing to hepatocarcinogenesis such as high viral load and hepatic inflammatory activity, providing a basis for adjuvant antiviral treatment (48).

Wu *et al*. showed that HCC late recurrence after resection in patients with hepatitis B-related HCC was significantly higher in patients with HBV DNA > 10^6 copies/mL at time of surgery (49). Qu *et al*. confirmed that persistent HBV DNA level ≥ 10^4 copies/mL at surgery and follow-up had the highest late recurrence rate, and that postoperative IFN-α treatment significantly decreased the risk (50). Also Hung *et al*. showed that HBV viral load > 2000 IU/mL (10^4 copies) at the time of surgery was an independent factor predicting recurrence of HCC after curative resection. Antiviral treatment with lamivudine after the resection was associated with significantly lower cumulative risk of recurrence, though this was not proven as an independent factor by multivariate analysis, probably due to the relatively small number of patients treated (51). A meta-analysis of postoperative adjuvant antiviral therapy for HBV-related HCC (1 study on IFN-α, 1 study on lamivudine) showed a trend of fewer recurrences in the treatment group than in the controls (52). Limited number of studies, limited number of patients and emergence of lamivudine-resistant mutations hamper the interpretation of the potential benefit of antiviral treatment. Also in this context it is hypothesized that new potent oral nucleos(t)ide analogues can overcome these issues, but further studies are required to confirm this.

Finally, it has been shown that HCC is associated with an increased risk of HCC recurrence after liver transplantation in spite of optimal post-transplant prophylaxis, most likely due to HBV replication in residual metastatic tumour cells, which could act as potential reservoir for HBV recurrence (53,54).

Conclusions

The strong, multifactorial significance of the association of chronic hepatitis B infection and hepatocellular carcinoma is met in importance by the role of HBV DNA being the prime predictive tool in the management of chronic HBV. Although further study is required, profound and long-term suppression of HBV DNA using potent nucleos(t)ide analogues seems to reduce the risk of HCC. Treating patients with pre-existing severe liver disease (cirrhosis) reduces the risk for HCC, but continued and careful monitoring of virological and hepatological parameters, is required. The use of HBV DNA goes beyond secondary prophylaxis : in tertiary prophylaxis HBV DNA predicts HCC recurrence after surgery. Finally, in primary prophylaxis of HCC, an effective and universal vaccination programme reduces the risk and occurrence of this tumour.

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