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Diagnosis and Treatment

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Dedication

To my daughters, Ophira and Feridey

Preface

You are not obliged to complete the task,
nor are you free to desist from trying.
—Talmud, Avot

Hepatocellular carcinoma (HCC) used to be regarded as a rare disease. The increasing numbers of chronic hepatitis C virus carriers in the United States and subsequent increased incidence of HCC seen in most large medical centers means that it is no longer an uncommon disease for most gastroenterologists or oncologists to encounter.

During the times when liver resection or systemic chemotherapy were the only real therapeutic modalities available, the outcomes were generally dismal, especially because most patients presented with advanced-stage tumors. Several recent factors seem to have changed this. They include the more frequent use of aggressive surveillance by ultrasound and computed tomography (CT) scanning in patients who have chronic hepatitis or cirrhosis from any cause (and thus are known to be at risk for subsequent development of HCC) to detect tumors at an earlier and therefore more treatable stage. Advances in CT scanning, particularly the introduction of multihead fast helical scans, mean that this vascular tumor can often be detected at an earlier stage, or multiple lesions can be diagnosed when only large single lesions were formerly seen, so that unnecessary resections are not performed.

Liver transplantation has had a profound effect on the therapeutic landscape. There have always been two hopes for this modality: namely, to eliminate cirrhosis as a limiting factor for surgical resection and also to extend the ability of the surgeon to remove ever-larger tumors confined to the liver. Regional chemotherapy and hepatic artery chemoembolization have been around for a long time and have been practiced mainly in the Far East and Europe.

There has not been a consensus for which drug or drug combination is best or whether embolization is important and, if so, what type and size of particle are optimal. Although there is still no consensus on these matters, it has recently become clear from two randomized controlled clinical trials that hepatic artery chemoembolization for unresectable nonmetastatic HCC seems to bestow a survival advantage compared to no treatment. The high recurrence rates after resection have led numerous investigators to evaluate preresection and postresection chemotherapy in the hope of decreasing recurrence rates. Only recently have clinical trials begun to provide evidence of enhanced survival for multimodality therapy involving resection and either chemotherapy or ^{131}I-lipiodol. The introduction of ^{90}Yttrium microspheres, which appear to offer the promise of relatively nontoxic tumoricidal therapy to the liver, appears to be a major therapeutic addition to our treatment choices, and its role alone or in combination with other therapies is just beginning to be explored.

In addition, we are beginning to enter the phase in which proteomics is applied to many tumor types, including HCC. This raises the possibility of being able to categorize patients into prognostic subsets, prior to any therapy. We are also just at the beginning of the age of cell cycle modulating factors including hormones, growth factors, and growth factor receptor antagonists and agents that specifically alter defined aspects of the cell cycle.

For these reasons, it seemed reasonable to produce a book that represents much of the current therapy and thinking on HCC. Admittedly, there is a bias toward expressing the experience of one center, the Liver Cancer Center at the University of Pittsburgh Starzl Transplant Institute, in which over 250 new cases of HCC have been seen each year for the last 15 years. This is an exciting time to be in the field of HCC basic science as well as clinical management because so many changes are simultaneously occurring at multiple levels of our understanding and management of the disease.

Brian I. Carr, *MD, FRCP, PhD*

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Color Plates

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1 Etiology and Epidemiology of Hepatocellular Carcinoma

Jawad Ahmad, MD
and Mordechai Rabinovitz, MD

CONTENTS

1. INTRODUCTION

Hepatocellular carcinoma (HCC) is one of the leading causes of worldwide cancer mortality, with an estimated 1 million deaths annually and a 5-year survival rate of less than 5% *(1)*. Cirrhosis of the liver is the main risk factor for the development of HCC, but the incidence of HCC varies considerably depending on, among other factors, geographical location and the cause of liver disease. HCC incidence of 4 to 15 per 100,000 has been reported in Western countries, compared with 120 per 100,000 in Asia and Africa *(2)*. Similarly, higher incidences of HCC have been reported in chronic liver disease related to viral hepatitis B and C compared with alcoholic liver disease. Tables 1 and 2 list some of the causes and possible mechanisms of HCC development.

2. RISK FACTORS FOR HCC

2.1. Hepatitis B Virus

Several case-control studies in the 1970s demonstrated that hepatitis B surface antigen (HBsAg) was present in the serum of patients with HCC substantially more frequently than in controls *(3–5)*. This was contrary to earlier reports that doubted such an association *(6,7)*. The issue was resolved by the landmark cohort

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Table 1
Risk Factors for the Development of HCC

Hepatitis B
Hepatitis C
Cirrhosis
Aflatoxin β1
Hereditary hemochromatosis
α-1-antitrypsin deficiency
Hereditary tyrosinemia
Alcohol
Male gender
Age
Tobacco smoking
Radiation exposure
Inorganic arsenic
Radioactive thorium dioxide
Oral contraceptive use
Glycogen storage disease
Membranous obstruction of the vena cava
Safrole oil

Table 2
Possible Pathogenic Mechanisms in the Development of HCC

Risk factor	*Possible mechanism*
Hepatitis B	Integration into host genome
	HBx protein and p53 suppression
Hepatitis C	Persistent inflammation and cirrhosis
	HCV capsid induced genetic modulation
Aflatoxin	Genetic polymorphism, p53 tumor suppression
Male gender	Androgenic stimulation of transforming growth factor α

study by Beasley et al. *(8)*, who followed up almost 23,000 Chinese subjects in Taiwan for an average of 3.3 years, 15% of whom were HBsAg positive. Of the 41 people who died of HCC, all but one was HBsAg positive, a relative risk of 223 compared with HBsAg-negative controls. A similar increased risk of HCC was seen in a prospective study of 1400 Alaskan natives with chronic hepatitis B *(9)*.

The precise mechanism for HCC development in chronic hepatitis B virus (HBV) is unclear, but integration of the viral genome into cellular DNA has been found in almost all HBV-induced HCC cases *(10)*. The X gene of the viral genome codes for functionally active transactivator proteins, including the X protein (HBx), which is oncogenic in transgenic mice *(11)*. The HBx protein interacts directly with promoter-bound transcription factors, or proto-oncogene promoters such as NFκB and also stimulates cellular second messenger systems, including the *ras* mitogen-activated protein-kinase signaling cascade and the protein kinase C pathway *(12)*. The pivotal role of HBx protein in initiating carcinogenesis is given further credence by the demonstration that it binds to the p53 tumor-suppressor protein and may inhibit p53-mediated apoptosis, leading to a clonally selected population of hepatocytes that expresses the incorporated viral genome *(13,14)*.

The strong association between hepatitis B infection and HCC is demonstrated further by examining the effect of interferon (IFN)-α therapy. IFN-α leads to loss of serological markers, including hepatitis B DNA and hepatitis B e antigen, appearance of hepatitis B e antibody (seroconversion), and can even result in the loss of HBsAg. This, in turn, would be expected to reduce the risk of HCC. Indeed, a recent meta-analysis of nine randomized controlled trials suggested that IFN-α therapy may reduce the absolute cumulative lifetime incidence of HCC by 4% *(15)*. It is unclear whether this is related to the direct anticarcinogenic effect of IFN-α or its antiviral effect. This argument is supported further by the striking decline in HCC incidence after the introduction of hepatitis B vaccine. Chang et al. *(16)* examined the effect on the incidence of HCC of a nationwide hepatitis B vaccination program begun in Taiwan in 1984. There was a stepwise decline in the annual incidence of HCC in children age 6 to 14 years from 0.7 per 100,000 between 1981 and 1986, to 0.57 per 100,000 between 1986 and 1990, to 0.36 per 100,000 between 1990 and 1994. Similarly, the incidence in children age 6 to 9 years declined from 0.52 per 100,000 in those born between 1974 and 1984 to 0.13 per 100,000 for those born between 1984 and 1986. The same group also suggested that the reduction in HCC attributed to hepatitis B vaccination is significant only in male children, with no decline witnessed in female children vaccinated over the same time period *(17)*. However, a recent study in the same population demonstrated that HBV vaccination decreased childhood HCC by up to 70% in boys and by up to 62% in girls *(18)*.

Previous infection with hepatitis B also may confer a risk for carcinogenesis. Matsuzaki et al. *(19)* recently demonstrated an increased incidence of hepatitis B DNA integration into the host genome in some Japanese patients with HCC who had no serological evidence of hepatitis B or hepatitis C infections. This suggests that prior hepatitis B infection may play a role in neoplastic progression.

2.2. Hepatitis C Virus

Hepatitis C virus (HCV) is an RNA virus and does not integrate into the host genome as seen with HBV *(20)*. Despite this, numerous series from throughout the world have demonstrated the high incidence of serum antibodies to hepatitis C (anti-HCV) in patients with HCC *(21–33)* and also have detected HCV RNA in liver and tumor tissue *(34–36)*. Most of the HCC cases develop in patients with underlying cirrhosis several decades after the initial infection, at a rate of 1–7% per year *(37,38)*. Tong et al. *(39)* reported a cohort of 131 patients with chronic posttransfusion hepatitis seen over a 14-year period and followed up for 4 years. Of these, 5% had evidence of HCC at presentation, and a further 5% developed HCC during the follow-up period.

Although cirrhosis greatly increases the risk of HCC, it can also develop in patients without cirrhosis. In patients with chronic hepatitis without cirrhosis, the incidence of HCC over a 3-year cumulative period was 4%, compared with almost 13% in those with established cirrhosis *(40)*. Indeed, HCV RNA can be found in patients with HCC in the absence of cirrhosis or fibrosis *(41)*.

The rate of liver cell proliferation also may play a role in the development of HCC. Persistent regeneration and proliferation of liver cells is a key feature of the hepatitis process, and products of the HCV genome may be involved in regulating liver cell proliferation. The HCV capsid can modulate the effect of several cellular genes, such as the human c-*myc* promoter gene *(42)*, and can transform rat embryo fibroblasts into a malignant phenotype when transfected with the *ras* oncogene *(43)*. Both in vitro and in vivo studies have underscored the capacity of viral proteins to induce tumorigenesis *(44)*. Some investigators have demonstrated clinical correlation of these models with higher rates of HCC in patients with chronic HCV or cirrhosis who expressed markers of liver cell dysplasia or periportal hepatocyte regeneration *(45–47)*. Recently, a retrospective study of 115 patients with chronic viral hepatitis identified large liver cell dysplasia as an independent risk factor for the development of HCC *(48)*. This may be related to the association of HCV replication with overexpression of transforming growth factor (TGF)-α and insulin-like growth factor II, both of which are related to hepatocyte transformation *(49)*.

The effect of hepatitis C treatment further emphasizes the role of HCV in the development of HCC. IFN-α with or without ribavirin is effective in reducing hepatic inflammation, in clearing HCV RNA in some patients with chronic HCV infection, or both *(50,51)*. Nishiguchi et al. conducted a randomized trial of IFN-α in patients with chronic HCV infection and cirrhosis *(52)*. Ninety patients were randomized to IFN-α or placebo and were followed up for 2–7 years. HCC developed in only 4% of the IFN-α groupcompared with 38% of the placebo group, although most patients in this study had HCV genotype 2. Another study analyzed the effect of IFN-α on more than 1500 patients with chronic HCV, most of whom had genotype 1. The risk of HCC was decreased in treated patients, but this effect was seen mostly in those patients who cleared HCV RNA during treatment or normalized their serum transaminases *(53)*. Other studies also demonstrated the effect of treatment on HCC. In patients with a sustained response, relapse, or nonresponse to IFN therapy, the relative risk of developing HCC compared with historical controls was 0.06, 0.51, and 0.95, respectively *(54)*. Further analysis of these findings suggested that age of 50 years or older, male gender, and advanced fibrotic stage were associated with an increased risk of HCC.

Liver fibrosis seems to be a crucial factor in determining carcinogenesis. A recent retrospective study of 2890 patients in Japan demonstrated a strong correlation between the annual incidence of HCC and the degree of liver fibrosis. IFN therapy was associated with a reduced risk of HCC, again most significantly in those who had a virological or biochemical response *(55)*.

In patients with chronic hepatitis C, several other factors seem to affect the progression to HCC. Coinfection with the human immunodeficiency virus (HIV), which is a frequent problem because of its common mode of acquisition, has been made increasingly relevant by the improved life expectancy of HIV-infected persons with the introduction of highly effective antiretroviral therapies. Although data are limited, there is some evidence that HCC may occur earlier and after a shorter period of HCV infection in patients coinfected with HIV *(56)*.

The role of alcohol in persons with hepatitis C also has received some attention. Yamauchi et al. *(57)* retrospectively studied 133 patients with either alcohol- or HCV-induced cirrhosis and determined that the 10-year cumulative occurrence rate of HCC was 18.5% in anti-HCV-negative alcoholic cirrhosis, 56.5% in nonalcoholic HCV-induced cirrhosis, and 80.7% in patients with alcoholic cirrhosis who were positive for anti-HCV. Similarly, diabetes mellitus may have a synergistic effect on progression to HCC in chronic hepatitis C *(58)*. A recent study from Japan indicated that genetic polymorphisms in proinflammatory cytokines such as the interleukin-1 family also may increase the risk of HCC in HCV patients *(59)*.

2.3. Hepatitis B and Hepatitis C Coinfection

The common modes of acquisition of hepatitis B and C infection may result in coinfection. Several investigators have hypothesized a possible synergistic effect on the development of HCC. Donato et al. *(60)* carried out a meta-analysis of 32 case-control studies looking at the impact of HBV and HCV infection on HCC development. Although the odds ratio for developing HCC in anti-HCV-positive persons was 17.3 and was 22.5 for HBsAg-positive subjects, the odds ratio for those with combined infection was 165. Similarly, a recent Italian study of 259 cirrhotic patients followed over 5 years suggested that patients positive for both anti-HCV and HbsAg were twice as likely to experience HCC compared with patients with hepatitis C infection alone and four times as likely compared with those with just hepatitis B infection *(61)*. However, data from a prospective Taiwanese cohort of more than 12,000 men contradict these earlier reports and suggest that HCV and HBV act independently in the pathogenesis of HCC, with a similar relative risk of developing HCC in patients who were HBsAg positive, anti-HCV positive, or positive for both *(62)*.

The mechanisms for HCC development in coinfection are unclear but probably occur through common and different pathways, because the data suggest a less than multiplicative effect of the two viruses. One simple model would involve HBV acting as an initiator by integrating into the host genome, and both HBV and HCV may then promote carcinogenesis by stimulating repeated cycles of inflammation, necrosis, and regeneration.

2.4. Hereditary Hemochromatosis

Hereditary hemochromatosis (HH) is an autosomal recessive disorder with homozygous frequency estimates of 1 in 220 to 1 in 400 in some populations *(63)*. Niederau et al. *(64)* followed 163 patients with HH for more than 10 years and found that in patients with cirrhosis, the frequency of HCC was more than 200 times that of an age- and gender-matched population. The same study demonstrated that venesection before the development of cirrhosis is associated with a normal life expectancy. The importance of cirrhosis as a risk factor in HH has been shown in a similar cohort of homozygous Italian patients who were followed prospectively for up to 229 months. Of the 97 patients who had cirrhosis, HCC developed in 28 during follow-up, whereas there were no cases of HCC observed in the 55 noncirrhotic patients *(65)*. However, the presence of cirrhosis is not an absolute prerequisite for the development of HCC, as some investigators have shown *(66,67)*. Interestingly, the development of HCC in HH, although associated with cirrhosis in most cases, is not usually associated with markers of hepatitis B or C infection or with the degree of iron deposition *(68)*.

2.5. α-1 Antitrypsin Deficiency

α-1 antitrypsin (A1AT) deficiency is an autosomal recessive disorder that is a common cause of liver disease and liver transplantation in children and may cause cirrhosis and HCC in adults *(69)*. The homozygous PiZZ phenotype is associated with liver injury, presumably because of the accumulation of the abnormally folded A1AT deficiency mutant in the endoplasmic reticulum of liver cells. Transgenic mice carrying the mutant Z allele develop severe liver disease and HCC with intrahepatocytic globules of A1AT *(70)*.

As seen with HCV infection, liver cell dysplasia often is found in patients with A1AT deficiency *(71)*. In a retrospective Swedish study *(72)*, homozygous PiZZ type was found more frequently in patients with cirrhosis and HCC compared with age- and gender-matched controls. Also in that study, HCC developed even in patients without evidence of cirrhosis and in those who had the heterozygous PiZ state *(73)*. However, other investigators found no correlation with an increased HCC risk *(74,75)*. The exact mechanism of carcinogenesis is unclear but may follow a stepwise model of preneoplastic nodules, adenomas, and HCC over several years *(76)*.

2.6. Aflatoxin Exposure

Aspergillus flavus is an ubiquitous fungus that produces hepatotoxins called aflatoxins, which contaminate staple foodstuffs in several tropical and subtropical regions. Exposure to aflatoxins, particularly aflatoxin β1, is associated with HCC as proved by several ecological and molecular epidemiological studies *(77–79)*. The magnitude of risk of HCC may be related to genetic polymorphism of hepatic metabolizing enzymes, particularly microsomal epoxide hydrolase *(80)*. There is a close association between aflatoxin exposure and mutation in the p53 tumor suppressor gene, one of the most frequently mutated genes in human cancers. Aflatoxin β1 induces a G to T transversion at the third position of codon 249 of p53 in human HCC cells *(81)*, and the same mutation has been observed in patients with HCC in areas of the world with high levels of aflatoxin exposure *(82,83)*. Results of a recent cohort study of some 6500 subjects in the Penghu Islets of Taiwan suggested that in patients with HCC, there was an adjusted odds ratio of 5.5 for heavy exposure to aflatoxins *(84)*. Although the vast majority of these HCC patients were HBsAg positive, the earlier onset of HCC in these patients compared with the rest of Taiwan suggests a synergistic effect of aflatoxin exposure and hepatitis B infection.

2.7. Other Risk Factors

Porphyria cutanea tarda is the most common and readily treated of the porphyrias and is characterized by chronic, blistering lesions of sun-exposed skin. Reports suggest that the prevalence of HCC is up to 16% in these patients, although this may be related to the presence of underlying hepatic fibrosis and the 80–90% incidence of HCV infection in these patients *(85,86)*. Similarly, acute intermittent porphyria is associated with HCC (87), particularly in women *(88)*.

Primary sclerosing cholangitis is well known to increase the risk of cholangiocarcinoma, but also may increase the risk of HCC *(89)*. Primary biliary cirrhosis also recently has been shown to result in a similar incidence of HCC as other causes of cirrhosis *(90,91)*.

Older age is another possible risk factor, but likely reflects the several decades required for cirrhosis to develop in patients with viral hepatitis. A recent European study examined this more closely by using birth order as a surrogate marker for age at infection with hepatitis B *(92)*. First- or second-born children tend to acquire common infections at school, whereas later-born children are exposed earlier through their older siblings. Compared with HBsAg-positive controls, those with HCC who were HBsAg-positive were twice as likely to have been later-born children, reflecting a longer period of infection. This was true even allowing for the confounding effect of an increased carrier state in patients infected at a younger age. The same relationship was not seen with hepatitis C-related HCC, reflecting the later onset of infection.

Tobacco smoke and alcohol have been implicated in the cause of many cancers and may play a role in the causation of HCC in patients with underlying liver disease. A recent European study of 333 patients with HCC indicated a dose–response association between smoking and HCC risk and a supermultiplicative effect of heavy smoking and heavy drinking. Interestingly, the effect was more noticeable in patients without evidence of HBV or HCV infection *(93)*. However, in a Chinese cohort of almost 90,000 people followed for 8 years, HCC did not seem to be associated with alcohol consumption. In males, there was no association with smoking, although a positive association between HCC and smoking was seen in females *(94)*. Alcohol itself has some carcinogenic potential through its metabolism to acetaldehyde, which inhibits nuclear repair enzymes, and also because of the generation of oxygen free radicals induced by cytochrome P450 2E1. This enzyme is involved in activating several compounds, including nitrosamines, to carcinogens. In addition, chronic alcohol use depletes liver retinoic acid, which is associated with increased expression of *AP1* genes (c-*fos* and c-*jun*), leading to cellular hyperproliferation. This effect can be reversed in experimental animals by adding retinoic acid and subsequent normalization of *AP1* gene expression and cellular regeneration *(95)*.

There are case reports of plant products associated with HCC. Safrole is an oil found in high concentration in *Piper betle*, a leafy plant used in betel quid that is chewed in many countries. Safrole is a rodent hepatocarcinogen and has been found in the livers of patients with HCC in the form of safrole DNA adducts, in the absence of other causes of tumor *(96)*.

Membranous obstruction of the inferior vena cava (MOVC) is an uncommon disorder of unclear origin leading to blockage of hepatic outflow, similar to Budd–Chiari syndrome (BCS). However, unlike BCS, MOVC is associated with the development of HCC in up to 40% of cases, in the absence of hepatitis viral markers *(97)*.

Male gender is associated with a two- to fourfold increase in HCC, even when the higher rates of confounding factors, such as viral hepatitis and alcoholic liver disease in men, are adjusted for *(98,99)*. The reasons for this discrepancy are unclear but may be related to androgenic stimulation. Studies in a transgenic mouse model in which overexpression of TGF-α leads to spontaneous development of liver tumors in 75% of male mice in 12 months, found that castration of male mice led to a decrease in liver tumors, which could be reversed by the addition of dihydrotestosterone *(100)*. Similarly, oophorectomy can increase the incidence of tumor formation to levels seen in male mice *(101)*.

Cirrhosis of any cause predisposes to HCC, although the risk varies according to the cause of cirrhosis. For example, hemochromatosis and viral liver disease are associated with higher rates of HCC than alcoholic liver disease. Nonetheless, the duration of cirrhosis seems to be the most important factor, regardless of cause *(102)*. Cirrhosis resulting from multiple risk factors present at one time may be associated with an increased incidence of HCC, and indeed, a recent study suggested that multinodular HCC is more prevalent in this group of patients *(103)*.

Cryptogenic cirrhosis or cirrhosis thought to be associated with nonalcoholic fatty liver disease (NAFLD) also may increase the risk of HCC. A recent study indicated that obesity was an independent predictor for HCC in patients with alcoholic cirrhosis or cryptogenic cirrhosis *(104)*. There also seems to be an increased incidence of NAFLD in patients with HCC, with one report from the United States suggesting that, although hepatitis C was still the predominant cause, NAFLD contributed to at least 13% of cases *(105)*.

Greater hepatocyte turnover in patients with cirrhosis may lead to an increased incidence of HCC as seen with HCV infection. One method of assessing cell turnover is liver cell proliferative activity measured by immunostaining of liver tissue for proliferating cell nuclear antigen (PCNA). In a cohort of 208 well-compensated cirrhotic patients, Donato et al. *(106)* were able to demonstrate a relative risk of almost 5 for the development of HCC in patients with a high level of PCNA compared with those with lower levels. This relative risk extended to patient survival. Cirrhotic patients with spontaneous bacterial peritonitis are also at increased risk of developing HCC *(107)*.

The presence of cirrhosis is not a prerequisite for HCC formation. However, even in cases of HCC in noncirrhotic livers, abnormal histological results, including fibrosis and iron overload, often are seen in the nontumorous liver, underscoring the importance of hepatocyte dysplasia or regeneration in the cause of HCC *(108,109)*.

Oral contraceptive pills (OCPs) are associated with the development of hepatic adenomas *(110)*. Several studies and case reports have demonstrated that prolonged use (particularly longer than 8 years) of OCPs and higher synthetic estrogen content, increases the relative risk of hepatic adenoma and HCC *(111–116)*. The occurrence of neoplastic change may be related to the propensity of estrogen to promote hepatocyte proliferation *(117)*. The effect on adenoma formation may be reversible with discontinuation of the OCPs *(118)*, but HCC still can occur after resolution of the adenoma *(119)*. The absolute risk of HCC is small, but the relative risk of 2.6 reported in case-control studies could have implications in societies where OCP use is prevalent and other risk factors are uncommon, especially because the risk of HCC does not decrease for at least 10 years after prolonged OCP use *(120)*.

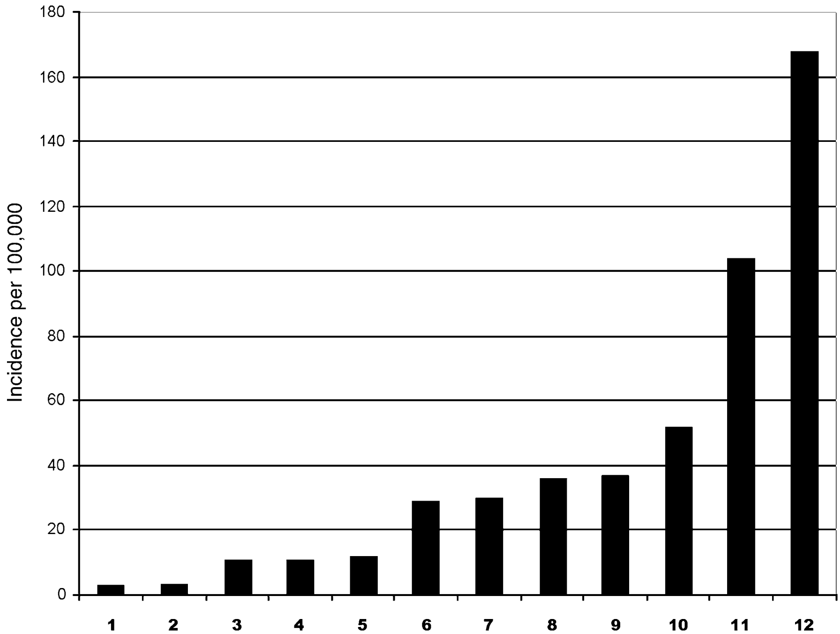

Fig. 1. Age-adjusted incidence of HCC in men according to geographical location. Data from refs. *1*,*132*,*133*,and *144* and from *Cancer Incidence in Five Continents*, Vol VII. IARC Scientific Publication No. 143. Cancer Base no. 2, 1997. Key: 1, United States; 2, U.K.; 3, Italy; 4, France; 5, Nigeria; 6, South Africa; 7, Zimbabwe; 8, Hong Kong; 9, Japan; 10, Thailand; 11, Mozambique; 12, Haimen City, China.

Hereditary tyrosinemia is an autosomal recessive disorder of tyrosine metabolism that results in liver disease and HCC in childhood. There is evidence of hepatocellular dysplasia before the development of HCC and DNA ploidy *(121)*. Therefore, it has been suggested that liver transplantation should be considered at younger than 2 years, because the risks of dysplasia and HCC increase substantially after this age *(122)*. Also, radiation exposure, inorganic arsenic ingestion, and radioactive thorium dioxide all may increase HCC risk *(123,124)*.

3. WORLDWIDE DISTRIBUTION OF HCC

The global distribution of HCC varies widely (Fig. 1), with two-thirds of the estimated 350,000 new cases per year occurring in the Far East, only several thousand occurring annually in the United States, and some 30,000 cases being diagnosed per year in Europe *(125)*. Even within these locations, HCC incidence differs according to age, gender, and cause. The situation becomes complex by the effect of immigration, particularly in the United States, where Chinese Americans have rates of HCC much higher than the indigenous population, and

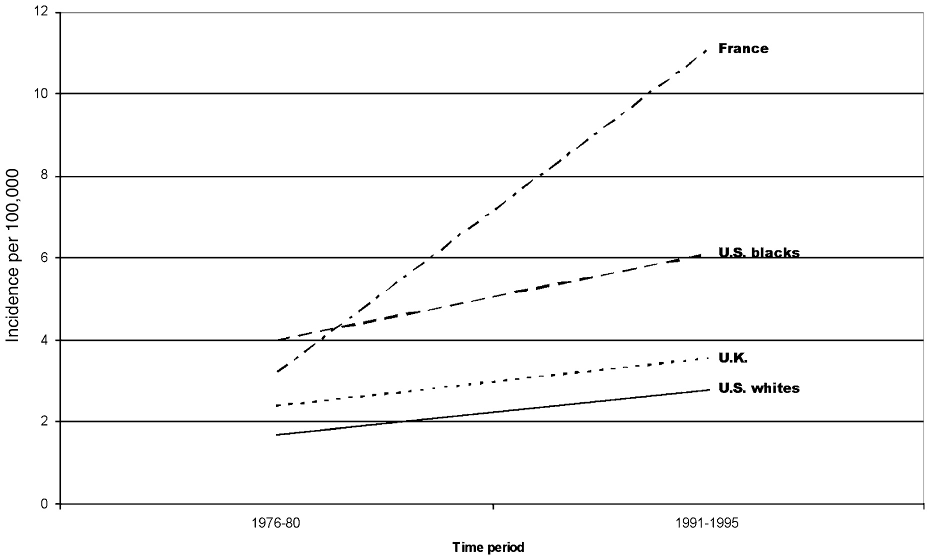

Fig. 2. Rising age-adjusted incidence of HCC among males in the West from 1976 through 1995. The data for 1976 through 1980 and for 1991 through 1995 from France and the UK refer to data from 1979 and 1994, respectively. Data from refs. *1*, *132*, and *133*.

rates may take several generations to fall *(126)*. In the Western world overall, HCC incidence is rising (Fig. 2).

3.1. HCC in the United States

The lower incidence of HCC in the United States (approx 2.5 per 100,000) is likely a result of lower rates of chronic HBV infection. A recent report demonstrated that in patients with HBV, rates of HCC are as high as 1000 per 100,000, very similar to those seen in high-incidence countries *(127)*. The emergence of chronic HCV infection over the last 30 years is likely to impact on the HCC incidence in the United States as the population ages.

Viral hepatitis accounts for most cases of HCC worldwide, as in the United States. Liang et al. *(128)* studied 91 American patients with HBsAg-negative HCC. Using sensitive polymerase chain reaction and three sets of primers for both HBV DNA and HCV RNA to optimize the detection of the viral genomes, they were able to demonstrate that 71% of these patients had evidence of either or both viruses in serum or liver tissue. Almost half the cases had evidence of HCV infection, which seems to be the most common cause of HCC in the United States.

The incidence of HCC cases in the United States has risen over the last two decades *(1)*. El-Serag and Mason *(129)* examined three national databases and found a 41% increase in the mortality rate from HCC and a 46% increase in hospitalizations resulting from HCC from the late 1970s to the early 1990s. It has been attributed to a rise in the overall age-adjusted incidence of HCC from 1.4 per 100,000 during the period from 1976 through 1980 to 2.4 per 100,000 from 1991 through 1995. The incidence of HCC increased particularly in younger persons age 40–60 years, and there were a disproportionate number of males and nonwhites affected. The authors hypothesized that HBV and HCV infections acquired during the 1960s and 1970s were responsible, at least in part, for the increase in the incidence in males and blacks, who are at greater risk of HBV and HCV infection. Further investigation revealed that HCV infection accounted for most of the increase in HCC cases, whereas the rates for HCC associated with HBV, alcohol, and other risk factors were unchanged. These findings were confirmed by the same group controlling for differences in age, gender, race, and geographic region. The increase in incidence of HCC rose the fastest in white males age 45–54 years, perhaps explained by the consequences of HCV infection acquired during the 1960s and 1970s *(130)*. Even within the United States, significant geographic variation exists in the incidence of HCC that cannot be explained by differences in race, age, or gender, with age-adjusted incidence rates as high as 4.6 per 100,000 in Hawaii, compared with 1.1 per 100,000 in Iowa and 1.0 per 100,000 in Utah *(131)*.

3.2. HCC in Europe

As in the United States, there has been a rise in the incidence of HCC in the United Kingdom *(132)*. Age-adjusted mortality rates increased from 2.39 to 3.56 per 100,000 in the male population between 1979 and 1994. An even greater rise was seen in women. The authors hypothesized that HCV infection was responsible for this trend and warned that HCC incident rates would continue to increase as those infected in the 1960s and 1970s from intravenous drugs experienced cirrhosis, and subsequently HCC, at an annual rate of 5%. Similar trends have been seen in France *(133)*. A recent case-control study in southern Italy, an area where HCV infection is hyperendemic, demonstrated a high incidence of HCC *(134)*. National statistics in Italy have shown that the age-standardized mortality rate from HCC rose from 4.8 to 10.9 per 100,000 over a 25-year period (1969–1994). Hepatitis C infection again would seem to be chiefly responsible, because more than 70% of cases of HCC diagnosed from 1996 through 1997 were anti-HCV-positive and only 11.5% were HBsAg-positive *(135)*. Closer analysis of these data revealed that HCC in older patients was more likely to be HCV-related, and the discrepancy in the incidence of HCC between males and females was more marked in HBsAg-positive patients. More than 90% of all patients had underlying cirrhosis. These results are in contrast to the one-third of cirrhotic patients who were HBsAg-positive in the early 1970s *(136)*. The use of reusable glass syringes in the 1950s and 1960s may have led to an increase in HCV infection rates.

In Greece, where the prevalence of chronic HBV infection is 10 times higher than that of chronic HCV infection, almost 60% of HCC cases are attributed to HBV and only 12% to HCV *(137)*. Even in Germany, which has a lower incidence of HCC, most cases are related to viral infection *(2)*.

The impact of viral hepatitis on HCC actually may be underestimated. Most studies have used the presence of HBsAg and anti-HCV to document the presence of HBV or HCV infection. A European cooperative group reported that HBsAg was present in 19% of HCC cases and anti-HCV was present in 40%. However, HBV DNA was detected in 33% of HBsAg-negative patients, and HCV RNA was detected in 7% of anti-HCV-negative cases of HCC. These findings emphasize the importance of HBV and HCV in the etiology of HCC and also stress the importance of using viral genome detection methods in epidemiological studies of HCC *(138)*.

3.3. HCC in Asia

In areas of the world with the highest rates of HCC, HBV infection is the most common etiological agent. In parts of China and Taiwan, almost one-fifth of the population are carriers of HBV, and the vast majority of persons with HCC are HBsAg-positive *(139)*. Although HCV infection is the main risk factor in HBsAg-negative HCC, HCV prevalence is low, with only 0.9% of healthy blood donors positive for anti-HCV *(140)*. The positive impact of HBV vaccination programs as detailed above may alter the cause of HCC in the Far East in the future.

A similar situation exists in India *(141)* , where a recent study showed that 53 of 74 HCC cases had evidence of HBV infection, compared with only 3 patients with HCV infection alone and 6 patients with dual infection with both HBV and HCV.

In contrast to the rest of Asia, cases of HCC in Japan are mainly related to HCV infection, and its incidence, as in the West, is rising but on a larger scale *(99)*. The reasons underlying this difference are likely related to the wide transmission of HCV to young people in Japan from contaminated blood and needles after the Second World War. It also seems that in Japanese patients with chronic viral hepatitis, the progression to HCC occurs at an accelerated rate in HCV infection compared with HBV infection. Takano et al. followed a cohort of 251 patients for a mean of 6 years and demonstrated that HCC was 2.7 times more common in patients with HCV than in patients with HBV *(142)*. Similarly, in a prospective study of 2215 patients followed for 10 years, HCC developed in 13.6% of the patients who were anti-HCV-positive. In contrast, HCC developed in only 4.9% of HBsAg-positive patients during that period *(143)*.

4. SUMMARY

The cause of HCC varies according to geographical location, but viral hepatitis is involved in most cases. Hepatitis B remains the most common cause in the developing world, and integration of the viral genome into the host is an important step in carcinogenesis. Mass vaccination programs are likely to dramatically alter the incidence of HCC in Asia. Hepatocellular carcinoma related to HCV is becoming an increasing problem in the developed world, but the factors leading to tumor formation are not yet well-understood. More effective treatments of HCV infection will be required to impact on the predicted increase in HCC in the coming decades.

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