Original Article

Hepatocellular Carcinoma in Explanted Livers of Patients with Genotype D HBV Cirrhosis: Report of the First Experience from Iran

Bita Geramizadeh MD•[1,2], Saman Nikeghbalian MD[2,3], Kurosh Kazemi MD[3], Alireza Shamsaifar MD[3], Ali Bahador MD[3], Heshmatollah Salahi MD[3], Seyed Ali Malekhosseini MD[3], Marzieh Kashtkar Jahromi MD[1], Ali Bakhshai Dehkordi MD[1]

Abstract

Background: This study was conducted to determine the impact of hepatitis B virus (HBV) as a cause of hepatocellular carcinoma (HCC) in a single liver transplant center in Iran.

Methods: We included all hepatectomy specimens from patients with HBV-related cirrhosis who underwent transplants from May 1993 until January 2012 in this study. From these, we determined the number that had HBV-induced HCC. Nested PCR results were used to determine the HBV genotype from sections of the hepatectomy pathology specimens.

Results: During this time period there were 1361 cirrhotic livers transplanted in our center. Of these, 249 were attributed to HBV cirrhosis. Overall, HCC was detected in 40 (2.9%) subjects, of which 29 (1.2%) had HBV-related HCC. Genotype D was only genotype observed in all HBV subjects.

Conclusion: The results revealed that although HBV-related cirrhosis was the most frequent single cause for liver transplant, the frequency of HBV-induced HCC was very low among transplant recipients. Out of 1361 transplant recipients, only 29 (2.1%) were diagnosed with HBV-related HCC. All HBV subjects had genotype D.

Keywords: Explanted liver, genotype D, hepatocellular carcinoma

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Introduction

Globally, infection with hepatitis B virus (HBV) is the major risk factor for development of hepatocellular carcinoma (HCC).[1] Iran is located in an intermediate geographic region for HBV infection; however, for HCC, Iran is in a low incidence geographic region.[2,3] In Middle Eastern countries, HCC prevalence is lower in comparison to sub-Saharan Africa and some Far East countries.[4]

Factors increasing the risk of HCC in a patient infected with HBV include cirrhosis, presence of HBeAg, viral load, genotype, alcohol and tobacco use,[5] exposure to aflatoxin, male gender and older age.[1] There are some reports of the independent effects of HBV genotype on the prevalence of HCC. Estimated incidence rates of HCC in subjects with chronic HBV infection in East Asian countries is 3.7 person per 100000 person-years for those with compensated cirrhosis.[6] According to reports, the most common HBV genotype in Iran is genotype D.[7] Controversial reports exist about the effect of HBV genotype on the risk of chronic liver disease and development of HCC.[8]

In studies from Asia, there is a greater association between genotype C infection and severe liver disease, cirrhosis, and HCC when compared with genotype B. In Western countries, individuals with genotype D have a greater incidence of severe liver disease or HCC than those with genotype A.[9]

We conducted this study to evaluate hepatectomy specimens of patients who underwent liver transplants for HBV cirrhosis. This study determined the percentage of HCC in those patients and compared the results with other countries according to HBV genotype.

Patients and Methods

From May 1993 to January 2012, we reviewed clinical charts of all HBV cirrhotic liver transplanted patients in Nemazee Hospital, Shiraz, Iran. Cirrhosis was diagnosed by the combination of clinical and paraclinical findings. Demographic information that included age and gender was obtained from the patients' clinical charts. Clinical and paraclinical data were extracted from the patients' files.

The diagnosis of HCC was based upon the pathology result. HBV DNA was extracted from the formalin fixed paraffin embedded tissue sections of the hepatectomy specimens. We evaluated HBV genotype by nested PCR[10] according to a previously described method.[9]

Results

From May 1993 to January 2012, there were 1361 liver transplants performed in Nemazee Hospital, affiliated with Shiraz University of Medical Sciences. Of these, 249 (18.3%) subjects had HBV-related cirrhosis. The other causes for cirrhosis in this hos-

Authors' affiliations: [1]Department of Pathology, Shiraz University of Medical Sciences, Shiraz, Iran. [2]Transplant Research Center, Shiraz University of Medical Sciences, Shiraz, Iran. [3]Transplant ward, Shiraz University of Medical Sciences, Shiraz, Iran.

•Corresponding author and reprints: Bita Geramizadeh MD, Pathology Department, Transplant Research Center, Shiraz University of Medical Sciences, P. O. Box: 71345-1864, Shiraz, Iran. Telefax: +98-711-6474331,
E-mail: geramib@sums.ac.ir.
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Table 1. Clinical and paraclinical characteristics of hepatitis B virus (HBV)-infected patients with and without hepatocellular carcinoma (HCC).

Variables	HBV cirrhosis with HCC *n* = 29	HBV cirrhosis without HCC *n* = 220	*P*-value
Female	5 (17.2%)	25 (11.3%)	0.57
Male	24 (82.8%)	195 (88.7%)	0.45
Age (years)	51.5 ± 9.7	46.5 ± 10.36	0.43
MELD score	12.5 ± 5.6	16 ± 9	0.1
AFP*(ng/mL)	229.8 ± 510	16.9 ± 30	< 0.001
ALT*(IU/L)	152 ± 129	129 ± 148	0.1
AST*(IU/L)	148 ± 420	128 ± 387	0.2
Alk-P*(IU/mL)	140 ± 39	160 ± 27	0.1

*AFP = alpha-fetoprotein; ALT = alanine aminotransferase, AST = aspartate aminotransferase, Alk-P = alkaline phosphatase.

Table 2. Underlying causes of cirrhosis and hepatocellular carcinoma (HCC) in transplant patients.

Cause for transplant	Total transplants *n* = 1361 (%)	Causes of HCC *n* = 40 (%)
HBV	249 (18.3)	29 (1.2)
HCV	39 (2.8)	8 (20)
Other	1073 (78.9)	13 (1.2)

pital were primary sclerosing cholangitis, autoimmune hepatitis, and Hepatitis C.

Patients with HBV cirrhosis ranged in age from 10 to 83 years, with a mean age of 46.5 ± 10.8 years. There were 219 male subjects with a male to female ratio of 7.3:1. Demographic characteristics of the subjects are presented in Table 1. HCC was detected in 29 (1.2%) HBV-related cirrhosis subjects.

There were significant differences observed in MELD score, AFP, ALT, AST, and alkaline phosphatase (Alk-P) between HCC subjects compared to those without HCC (Table 1; $P < 0.05$).

Out of the 1361 liver transplants, there were 21cases of HCC secondary to other causes (Table 2). HBV was the most common underlying single cause of HCC followed by HCV (8), tyrosinemia (2) and cryptogenic (2). There were 9 cases of HCC without cirrhosis.

Nested PCR of the liver specimens from the 249 HBV cases were all genotype D, regardless of HCC status.

Discussion

HCC is one of the major causes of cancer-related deaths worldwide.[1] Epidemiologic studies have shown marked variations in the incidence of HCC in different geographic areas of the world.[2] HCC is common in the Asia-Pacific region because of the high prevalence of main etiological factors such as HBV and HCV.[11]

Although HBV is the most common etiological agent of cirrhosis in Iran, HCC is the 16th most prevalent cancer. This contrasts other regions that have a similar prevalence of HBV, however in those regions HCC is among the top seven cancers in both incidence and prevalence.[12]

Case control studies have reported the relative risk of HCC in HBV with and without cirrhosis as 5 to 49.[13] Other reports from high incidence areas of the world have shown that the 5–10 year prevalence of HCC following HBV cirrhosis is 4% and for chronic hepatitis, it is 7%.[14] However this prevalence is from areas that have a high incidence of genotypes B and C.[15]

HBV genotype C has a higher frequency of basal core promoter mutation which suggests important pathogenic differences between HBV genotypes.[16] In addition genotype C tends to have a higher level of HBV DNA and HBeAg positivity as well as delayed HBe seroconversion in the immune clearance phase of chronic hepatitis.[17] Genotype C is closely associated with a high risk of HCC among cirrhotic patients, whereas genotype B is more commonly associated with HCC in non-cirrhotic young patients.[17]

In Iran, previous reports as well as the current investigation have documented only genotype D in patients with HBV infection.[18,19] Genotype D is widely distributed throughout the Middle East, Eastern Europe, Russia, Northern Asia and the Mediterranean region. Patients with genotype D often convert from HBeAg to anti-HBe in early adulthood and adolescence.[20]

Iran is placed in the world's low incidence area of HCC, with a reported rate of <4/100,000 persons.[21]

In the current study we have observed an HCC incidence rate of 1.2% observed among the 249 liver transplant patients with HBV cirrhosis, which is low in comparison with other countries that have the same incidence of HBV infection.[22]

Few studies have been performed regarding the risk of HCC in genotype D[23]; most previous reports are from high incidence areas of the world with genotypes B and C.[24]

This is the first report regarding the low incidence of HCC in a geographic region of genotype D HBV cirrhosis. It should be emphasized that although the study has been performed in Southern Iran, this transplant center receives patients from throughout Iran as it is the most active liver transplant center in Iran.

Further studies should be performed to follow and evaluate patients with all clinical syndromes of HBV infection to determine the presence of different mutations and other interfering factors (genetic and environmental) in order to find the true incidence of HCC in patients with genotype D.

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