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BRIEF ARTICLE

Hepatocellular carcinoma in patients with chronic hepatitis C virus infection without cirrhosis

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Abstract

AIM: To investigate and characterise patients with chronic hepatitis C virus (HCV) infection presenting with hepatocellular carcinoma (HCC) in the absence of cirrhosis.

METHODS: Patients with chronic hepatitis C infection without cirrhosis presenting with HCC over a 2-year period were identified. The clinical case notes, blood test results and histological specimens were reviewed to identify whether additional risk factors for the development of HCC were present.

RESULTS: Six patients (five male, one female) with chronic hepatitis C infection without cirrhosis presented to a single centre with HCC over a 2-year period. Five patients were treated by surgical resection and one patient underwent liver transplantation. Evaluation of generous histological specimens confirmed the presence of HCC and the absence of cirrhosis in all cases. The degree of fibrosis of the background liver was staged as mild (n = 1), moderate (n = 4) or bridging fibrosis (n = 1). Review of the clinical case notes revealed that all cases had an additional risk factor for the development of HCC (four had evidence of past hepatitis B virus infection; two had a history of excessive alcohol consumption; a further patient had prolonged exposure to immune suppression).

CONCLUSION: HCC does occur in patients with non-cirrhotic HCV infection who have other risk factors for hepatocarcinogenesis.

Key words: Hepatitis C virus; Hepatocellular carcinoma; Non-cirrhotic; Screening

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INTRODUCTION

Hepatocellular carcinoma (HCC) is a well-recognised complication of cirrhosis regardless of aetiology; the risk of malignancy differs according to the underlying cause of liver damage. As a consequence, patients with cirrhosis undergo routine interval screening in most liver centres using a combination of serum α-foetoprotein (AFP) and liver ultrasound, although solid evidence to support this

approach post-dates adoption of the strategy[1,2]. Surveillance is restricted to those at higher risk in some centres. Patients with cirrhosis secondary to chronic infection with hepatitis B virus (HBV) or hepatitis C virus (HCV) are at particular risk of HCC[3]. Furthermore, there are numerous reports indicating that chronic infection with HBV is associated with HCC in the absence of cirrhosis.

In contrast, HCC in patients with chronic HCV infection without cirrhosis appears to be very rare[4-7]. We describe six such cases that presented to one tertiary referral centre in the past 2 years (Table 1).

MATERIALS AND METHODS

Patients with chronic HCV infection without cirrhosis presenting with HCC over a 2-year period were identified. The clinical cases were reviewed to identify any additional risk factors for hepatocarcinogenesis. Details obtained included ethnic origin, alcohol consumption, past or present infection with HBV, medical co-morbidity, medication history and family history of liver disease or HCC. Histological specimens were reviewed by an experienced liver pathologist to confirm the presence of HCC and to assess the degree of fibrosis of the background liver tissue. Furthermore, histological specimens were evaluated for the presence of additional hepatic pathology including the presence of iron, steatosis and α-1 antitrypsin globules.

RESULTS

Six cases with non-cirrhotic chronic HCV infection and HCC presented to one centre over a 2-year period. Table 1 summarises the clinical and histological characteristics of the cases including additional risk factors for hepatocarcinogenesis. The first case, a 53-year-old Caucasian male, was referred to the service having been found to be HCV-RNA positive (genotype 1). He had a history of previous injecting drug use and a high alcohol intake exceeding 30 U/wk. Evaluation of hepatitis B serology demonstrated that he was surface antigen (HBsAg) negative, and positive for core antibody (anti-HBc) in keeping with past infection. Screening liver ultrasound demonstrated a focal lesion. Computerised tomography (CT) confirmed a mass with arterial enhancement. Serum AFP was 239 IU/L (normal range < 10 IU/L). Histology of the resected mass revealed a 3.5-cm multi-focal HCC, with moderate to poor differentiation and vascular invasion. Histology of the background liver revealed minimal inflammation but moderate fibrosis. He is recurrence-free 24-mo post resection. HCV-RNA was undetectable at completion of 48-wk therapy with pegylated interferon-α and ribavirin.

The second case, a 57-year-old Caucasian male, presented with fever and abdominal pain. CT of the abdomen revealed a 5 cm × 3 cm, inflammatory mass in the right side of the abdomen in close proximity to bowel and in addition a 2 cm hypodense lesion within the liver. A right hemi-colectomy was performed which revealed extra-colonic fibrosis and abscess formation secondary to a caecal diverticulum. Biopsy of the liver mass demonstrated HCC; the background liver comprised moderate inflammation and mild fibrosis consistent with chronic HCV infection. He was HCV-RNA positive (genotype-3), HBsAg negative, anti-HBc positive with a history of previous injecting drug use. Liver function tests were normal. Serum AFP was 3 IU/L. After hepatic resection, histology revealed a 3.1 cm HCC with moderate differentiation, without vascular invasion; the background liver confirmed mild inflammation and mild fibrosis. He has completed therapy with pegylated interferon-α and ribavirin with sustained virological response. He is recurrence-free 22 mo following surgery.

Case three, a 67-year-old Nigerian lady with chronic HCV infection, had a 4 cm focal lesion detected on screening liver ultrasound. CT imaging confirmed the mass with arterial enhancement. Biopsy of the lesion demonstrated HCC and the background liver revealed moderate inflammation and mild fibrosis consistent with chronic HCV infection. She was HCV-RNA positive (genotype 1), HBsAg negative, anti-HBc positive. Liver function tests were unremarkable. Serum AFP was 206 IU/L. After hepatic resection, histology revealed a 4.5 cm HCC with moderate to poor differentiation and lymphovascular invasion on a background liver with moderate fibrosis. She was unable to tolerate antiviral therapy due to profound anaemia and remains HCV-RNA positive. She is recurrence-free 20 mo after resection.

The fourth case was a 46-year-old Caucasian man known to have chronic HCV (genotype 1) with a history of previous injecting drug use; he was HBsAg negative, anti-HBc positive. Liver biopsy demonstrated moderate activity and moderate fibrosis consistent with chronic HCV infection. He had failed to respond to pegylated interferon and ribavirin. Six years later investigation of a raised AFP (77 IU/L) revealed a 1.5 cm arterially enhancing lesion in the liver. Surgical resection revealed a 1.7 cm moderately - well differentiated HCC without vascular invasion on a background liver with bridging fibrosis but not cirrhosis. He is recurrence-free 16 mo after resection.

Case 5 was a 46-year-old Indian man with chronic HCV infection with a history of renal transplantation 15-year previously, treated with ciclosporin and prednisolone. Liver function tests were unremarkable. Screening ultrasound detected a 3 cm focal lesion in the liver; biopsy of the mass revealed HCC. He was HCV-RNA positive (genotype 1) without evidence of exposure to HBV. Serum AFP was 5 IU/L. Further imaging with CT, magnetic resonance imaging and angiography confirmed the mass and revealed two further smaller lesions. He was deemed to meet Mazzaferro criteria[8] and listed for liver transplantation. The explant revealed a 3 cm HCC with poor differentiation and vascular invasion and several small satellite lesions. The background liver demonstrated moderate fibrosis and moderate inflammation. He developed HCC within the grafted liver after 6 mo leading to death 8 mo following transplantation.

The final case, a 63-year-old Russian doctor, acquired HCV (genotype 1) 9 years previously following a needle stick injury from a patient. He had a long history of excess alcohol consumption (30 U/wk) but was immune

Table 1 Patient characteristics

Age (yr)	Sex	HCC	Background liver histology at time of HCC treatment	AFP (IU/L)	HCV status	HBV status	Alcohol (U/wk)
53	M	3.5 cm Moderate/poor differentiation Vascular invasion	Minimal inflammation Moderate fibrosis	239	RNA positive Genotype 1	HBsAg negative Anti-HBc positive	10-30
57	M	3.1 cm Moderate differentiation No vascular invasion	Mild inflammation Mild fibrosis	3	RNA positive Genotype 3	HBsAg negative Anti-HBc positive	0
67	F	4.5 cm Moderate/poor differentiation Lymphovascular invasion	Moderate inflammation Moderate fibrosis	206	RNA positive Genotype 1	HBsAg negative Anti-HBc positive	0
52	M	1.7 cm Moderate differentiation No vascular invasion	Moderate inflammation Bridging fibrosis	77	RNA positive Genotype 3	HBsAg negative Anti-HBc positive	20
46	M	3 cm Poorly differentiated with satellite lesions	Moderate inflammation Moderate fibrosis	5	RNA positive Genotype 1	Negative	0
62	M	6.5 cm Moderate differentiation Vascular invasion	Moderate inflammation Moderate fibrosis	10	RNA positive Genotype 1	Negative	30

HCC: Hepatocellular carcinoma; AFP: α-foetoprotein; HCV: Hepatitis C virus; HBV: Hepatitis B virus; HBsAg: Hepatitis B surface antigen.

to HBV and was anti-HBc negative. At presentation, liver biopsy revealed mild fibrosis with moderate inflammation. Nine years later he was found to have a 6.5 cm HCC. Serum AFP was 10 IU/L. He underwent surgical resection; histology revealed a moderately differentiated HCC on a background liver with moderate fibrosis. He is well 5 mo after.

DISCUSSION

Six cases with non-cirrhotic chronic HCV infection and HCC presented to one centre over a 2-year period. All were considered at low risk of HCC and none was in a surveillance programme. One had symptoms but the identification of a liver mass in the remaining five patients was fortuitous. Cirrhosis was excluded confidently in all cases by careful histological review of generous tissue specimens revealing at worst bridging fibrosis. No patient had histological features to suggest any diagnosis other than HCV related injury. All patients had a normal body mass index and none were diabetic. However, all patients had an additional risk factor for liver injury or HCC: four had evidence of past HBV infection; two had a history of excessive alcohol consumption; a further patient had prolonged exposure to immune suppression.

The incidence of HCC is increasing across the developed world[9]. Cirrhosis of any cause is an important precursor for HCC, although liver disorders including chronic HBV or HCV infection, haemochromatosis, non-alcohol-related fatty liver disease (NAFLD) and alcohol-related liver disease carry a particular risk[3]. The risk is also much higher in men and older patients[10,11]. The current increase in the prevalence of chronic liver disease secondary to chronic HCV infection and NAFLD (a consequence of the increasing prevalence of obesity and an ageing population) are the main reasons for the increasing incidence of HCC in the developed world[12-15], although improved screening and diagnosis may also play a part. The underlying mechanisms that lead to malignant transformation of HCV-infected hepatocytes, however, remain uncertain, but as most HCV-related HCC occurs on a background of severe fibrosis or cirrhosis it is thought that the mechanism of carcinogenesis is more likely to be indirect, such that the process of tissue damage, regeneration and repair are important, rather than a direct oncogenic effect of HCV infection or the inflammatory response to the virus.

It is well recognised that chronic HBV infection can lead to HCC in the absence of cirrhosis[16]. HBV is a DNA virus that can integrate into the host cell genome; integration may be mutagenic directly by causing genomic instability, loss of tumour suppressor activity or over-expression of genes involved in regulation of cell cycle proliferation. In addition, HBV encodes HBx protein, which functions as a transcriptional trans-activator of cellular genes that are involved in cell proliferation control such as c-jun, c-fos and c-myc. This may lead to dysregulation of the cell cycle and interference with cellular DNA repair and apoptosis[17].

This series raises the possibility that HCV may be oncogenic. HCV is an RNA virus, which replicates in the cytoplasm and does not integrate into host cellular DNA. However, some HCV proteins, such as HCV core and non-structural proteins NS3, NS4B and NS5A have a regulatory effect on cellular promoters and interact with a number of cellular proteins involved in carcinogenesis under certain conditions[18-21]. In addition, hepatocytes from patients with chronic HCV infection are arrested in G1 and may undergo replicative senescence, which may predispose to malignancy[22]. Direct evidence for a carcinogenic role for HCV *in vivo* is lacking.

HCC has been described very rarely in chronic HCV infection in the absence of cirrhosis[4-7], which suggests that other aetiological factors may be more important. Analyses of case series in Japan have suggested that ageing increases the risk of developing HCC in patients with HCV who do not have cirrhosis, particularly in wom-

en[10,11]. In the series presented here, however, five of six cases were men and the median age at diagnosis of HCC was 55, suggesting that alternative factors are important. None of the cases described had histological evidence of additional injury such as steatohepatitis or iron accumulation that are recognised co-factors in the development of liver injury and HCC[23,24]. However, four of six patients had serological evidence of previous exposure to HBV. It is possible that integration of HBV genes had occurred, increasing the risk of HCC as in HBV infection without cirrhosis. Long-term immune suppression in another may have increased the risk of HCC[25]. Furthermore, it has been reported that renal transplant patients might have an increased susceptibility to HCC even without viral infection purely as a result of immune suppression[26]. In the final patient there was a long history of excess alcohol use but no evidence of alcohol related liver damage on biopsy.

A third of patients with HCV have been exposed to HBV because of common risk factors[27]. In the Cambridge series 35% of 1500 patients with chronic HCV infection were anti-HBc positive/HBsAg negative while 2% were co-infected with HBV. Past HBV infection is associated with an increased risk of progressive liver injury in some series of patients with chronic HCV infection and other liver disorders[28,29] although the presence of anti-HBc was not associated with progressive fibrosis in our own series[30]. HBV genomic material has been identified in liver from patients with HCC who were HBsAg negative but HCV RNA positive[31,32] and the presence of HBV genes in this setting has been linked to HCC[33]. In a prospective observational study, serum anti-HBc was a marker of high risk for HCC among patients with HCV related cirrhosis, but was not a significant risk factor in those without cirrhosis[34]. The presence of HBV genes in HCC tissue of HBsAg negative, HCV negative patients has also been described[35]. Thus, long-term persistence of HBV genes in liver tissue may cause HCC without inflammation, necrosis or regeneration. While abnormal alanine aminotransferase (ALT) fluctuation is associated with carcinogenesis in HCV positive patients[36], the presence of integrated HBV DNA in the liver may promote carcinogenesis independently. Patients with low levels of ALT and minimal histological change may still be at risk of HCC development if they have had previous exposure to HBV.

Surveillance for HCC has been conducted for many years but a survival benefit for screening with 6-monthly ultrasound and AFP monitoring has only been demonstrated recently[1]. However, surveillance is practiced widely and recommended in high-risk groups such as those with cirrhosis due to HBV, HCV, alcohol, or haemochromatosis[2]. In addition, because of the high risk of HCC in non-cirrhotic HBV infection, current guidelines recommend screening in high-risk groups (family history of HCC, Asian males > 40 years, Asian females > 50 years and Africans > 20 years and could be extended to those with high serum HBV DNA levels). In HCV infection with cirrhosis there is a high risk of HCC development (2%-8% per year); surveillance is recommended and cost effective[37]. At the current time it is unclear whether patients with bridging fibrosis should be offered screening and it is not recommended for patients with mild or moderate disease regardless of patient age or length of time of infection.

All HCV-infected patients with cirrhosis in our centre are offered 6-monthly screening with liver ultrasound by a small group of experienced liver radiologists or ultrasonographers; serum AFP is not sought in this particular cohort because of a lack of specificity and sensitivity, as demonstrated in this series. This experience has prompted review of our policy; for example should older men with chronic HCV infection without cirrhosis but with an additional risk factor for HCC, such as anti-HBc or excess alcohol consumption, undergo ultrasound screening? The increased workload for our service would be enormous and it is probable that the number of patients identified with HCC that could be cured by intervention would be too low to justify such an approach. Further data on the incidence of HCC in HCV-infected patients without cirrhosis are required before a change in policy can be recommended as routine practice; a national register of such cases could be helpful.

COMMENTS

Background

Cirrhosis of any cause is associated with a significant risk of developing hepatocellular carcinoma (HCC). Chronic hepatitis B virus (HBV) infection in the absence of cirrhosis is also a recognised risk factor for HCC and screening is recommended in some high risk groups. HCC occurs rarely in non-cirrhotic hepatitis C virus (HCV) infection and there are no recommendations for screening in these patients.

Research frontiers

Due to the observation of an increase in HCC in non-cirrhotic HCV patients, a detailed evaluation of these patients was undertaken.

Innovations and breakthroughs

In each patient with HCC and non-cirrhotic HCV infection, an additional risk factor for hepatocarcinogenesis was identified. These included previous infection with HBV, high alcohol intake and immunosuppression.

Applications

Patients with chronic HCV without cirrhosis may be at risk of developing HCC if there are other risk factors for liver injury and carcinogenesis present. It is possible that these patients should be considered for surveillance programmes although this would result in a dramatic increase in workload for radiological departments and may not be cost effective.

Peer review

This is a well written article.

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