Small Hepatocellular Carcinoma Associated with Wilson’s Disease

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Abstract

We report a 66-year-old male patient with hepatocellular carcinoma (HCC) associated with Wilson’s disease. The patient presented with an unresolving abnormal liver function test, decreased serum ceruloplasmin levels and increased 24-hour urine copper excretion. Liver biopsy specimen revealed the presence of increased levels of copper and features suggestive of Wilson’s disease. Abdominal imaging showed the existence of a small HCC. Three years after chemoembolization and microwave coagulation therapy for HCC, he died of hepatic failure, which apparently resulted from chemoembolization. Patients with Wilson’s disease should be screened for HCC. We should elude therapies such as chemoembolization in these patients.

Case Report

A 60-year-old Japanese man visited an outpatient department in 1988 due to ulceration in the stomach. He was treated for ulceration with histamine 2-receptor blocker and it was improved. He gave the history of an abnormal liver function test, which was first pointed out at a local hospital in 1968 during mass screening. He received blood transfusion in 1963 during an operation for intestinal obstruction. Ultrasonography at our hospital revealed that the surface of the liver was irregular with a rounded edge. The echogenic texture of the liver was irregular and he also had a mild degree of splenomegaly. Multiple esophageal varices were detected at the lower end of the esophagus by upper gastrointestinal endoscopy. The levels of asparate aminotransferase (AST, 18 IU/l), alanine aminotransferase (ALT, 26 IU/l), bilirubin (0.9 mg/dl) and albumin (4.3 g/dl) in the sera were within normal limits at this point. Platelet count in the peripheral blood was low (100,000/mm³). He was diagnosed as liver cirrhosis clinically. He was not given any specific therapy, but followed-up routinely once every 6 months.

In August 1994, a space-occupying lesion was detected in the liver and he was admitted for further diagnosis and therapy. On admission, he did not show any abnormality in physical examination except for the presence of a mildly enlarged spleen. Laboratory tests at this time showed very low levels of copper (23 μg/dl, normal range 78–131 μg/dl) and ceruloplasmin (2.5 mg/dl, normal range 21–37 mg/dl) in his sera. Daily urinary excretion of copper was 65 μg/day (normal <50 μg/day). Penicillamine therapy induced increased daily excretion of copper in the urine (898 μg/day). However, he did not show the presence of Kayser-Fleischer
rings with slit lamp. He did not complain of any neurological symptoms. HBV DNA, HCV RNA or hepatitis G virus (HGV) RNA were not detected in his sera by polymerase chain reaction. Autoantibodies, such as anti-nuclear antibody, anti-mitochondria antibody and anti-smooth muscle antibody were all negative in his sera. The levels of tumor markers, such as α-fetoprotein and des-gamma-carboxy prothrombin (PIVKA II) were within normal range. Both carcinoembryonic antigen and carbohydrate antigen 19-9 were higher than normal, 6.1 ng/ml and 77 U/ml, respectively. Ferritin was normal, 212 ng/dl. His family members had not been suffering from liver diseases and metabolic disorders. A hypoechoic mass with mosaic pattern was detected in the liver by ultrasonography (Fig. 1A). The size of the lesion was 1.5 cm in maximum diameter. Helical-dynamic computed tomography (CT) scanning of the abdomen showed one round well-defined mass in the S4 region of the liver, which was enhanced in the arterial phase. The liver volume was 1,126 cm$^3$ as estimated by CT (Fig. 2A). He was diagnosed as HCC associated with Wilson’s disease.

In September 1994, he was treated for HCC with chemoembolization: injection of 20 mg of epirubicin with 1 ml of lipiodol from the medial branch of the left hepatic artery and right hepatic artery followed by embolization with gelform from the medial branch of left hepatic artery. After chemoembolization, CT revealed strong accumulation of lipiodol into the tumor (Fig. 1B). Subsequently, surgical microwave coagulation therapy was done. A liver biopsy was performed at this point. Hematoxylin-eosin staining showed large regenerative nodules with strong fatty degeneration at the non-cancerous portion of the resected liver (Fig. 3A). Rhodamine and Orcein staining detected copper granules in the hepatocytes (Fig. 3B, C). However, deposits of iron were seen in only a few hepatocytes. Hepatic copper content was 198 μg/100 g wet weight (normal <10 μg/100 g wet weight) in the non-cancerous portion as detected by the quantitative method. These findings led to a definite diagnosis of Wilson’s disease with liver cirrhosis. Investigation of the responsible mutation of ATP7B for Wilson’s disease in this patient was homozygote of Ala874Val, which was done by single strand conformational polymorphism analysis followed by non-radioactive sequencing method (12).

Just before chemoembolization, bilirubin, albumin, AST and ALT in the sera were 1.0 mg/dl, 3.7 g/dl, 42 IU/l and 29 IU/l, respectively. However, seven days after chemoembolization, abdominal CT revealed a reduced liver volume (973 cm$^3$, Fig. 2B) without deterioration of liver function (bilirubin 1.0 mg/dl, albumin 3.2 g/dl) nor increase of serum transaminase levels (AST 27 IU/l, ALT 35 IU/l). He was administered penicillamine from December 1994 at the dose of 1,000 mg per day and hematuria was reported 10 days after the start of therapy. Next, he was given trientine, which was also discontinued due to interstitial pneumonia. Finally, he was maintained on a low copper diet.
In 1997, 3 years after the first chemoembolization, he developed another HCC at S6 region (2 cm in diameter) and was treated with percutaneous ethanol injection therapy (PEIT). By this time, he developed features of liver failure such as increased bilirubin (5 mg/dl) and decreased prothrombin time (50%) and the liver volume was highly reduced (537 cm$^3$, Fig. 2C). He was managed by conservative therapy, but he died in December 1998. The cause of his death was liver failure, but not HCC.

At autopsy, the color of the liver was yellowish-brown and the surface showed many regenerative nodules of intermediate size up to 8 mm in diameter (Fig. 4A). The weight of the liver was 640 g and the cut section of the liver showed multiple HCC nodules measuring up to 2 cm in diameter; one in S4, two in S6 and several in S8. There was no evidence of metastatic tumors in any other organs. Histopathological examination showed the presence of moderately differentiated HCC (trabecular pattern), existence of bile plug and necrotic tissues (Fig. 4B). The non-cancerous portion was characterized by the presence of mixed nodular cirrhosis with established thick fibrous septum containing regenerative small bile ducts and infiltration of mononuclear cells. Hepatocytes with positive granules for Orcein and Rhodanine staining were seen, especially in the peripheral area of the regenerative nodules. Liver copper level in the non-cancerous portion and cancerous portion were 128 μg/100 g and 42 μg/100 g, respectively (wet weight).

**Discussion**

Only eleven cases of HCC associated with Wilson’s disease have been reported to date (1–11). Of these 11 cases, 10 cases were reported before 1989 and only one case was reported in 1992. HCV is a major etiological agent related to the development of HCC and a serological diagnosis of HCV was established in 1989. Although we are not sure about the
virological markers about the 10 cases of HCC-associated Wilson’s diseases reported before 1989, a role of HCV in those cases cannot be discarded completely. Again, there is no description about the HCV marker in HCC patient associated with Wilson’s disease reported in 1992. The case reported here was the first case, which was completely free from all hepatitis viral markers.

On the other hand, there are several unique features in the reported case. First, this is the oldest male patient of HCC associated with Wilson’s disease. Second, the size of the HCC nodule was 1.5 cm during first detection. This is the smallest HCC detected so far in patients with Wilson’s disease. In other cases the size of HCC was too large for therapy. Only one patient has to date received surgical operation for small HCC. Third, we did chemoembolization, microwave therapy and PEIT in this patient. In addition, the liver volume was dramatically reduced after chemoembolization.

There are many reports on the high appearance rates of HCC in patients chronically infected with HBV or and HCV. Due to this, many patients with HBV or and HCV are screened well regarding for HCC complication. In some of them small HCC is detected and several types of therapies are undertaken. Although rare in incidence, patients with Wilson’s disease should be carefully considered the possible complication with HCC, similar to the patients with liver cirrhosis and multiple HCC nodules were seen at autopsy (A). Histopathological examination of the tumor by hematoxylin-eosin staining showed moderately differentiated HCC (trabecular pattern), existence of a bile plug and necrotic tissues (B, ×200).
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cirrhosis due to HBV or/and HCV infections. Such a procedure may afford patients the chance for therapy for HCC in an early stage.

Many groups have reported the role of copper in hepatocarcinogenesis. Some support for its protective role (13–15). In contrast however, the mutant strain Long-Evans Cinnamon (LEC) rat, a model of Wilson’s disease, is known to develop hepatitis and subsequently HCC in spite of having a high copper accumulation in the liver (16, 17). Therefore, it is difficult to explain the occurrence of HCC in Wilson’s disease solely based on the role of copper. However, the role of liver cirrhosis seems to be important in this regard. The common feature shared by all cases with HCC associated with Wilson’s diseases was liver cirrhosis (18).

To prevent the prevalence of HCC in Wilson’s disease, it is important to make an early diagnosis of Wilson’s disease to arrest further progression of the pathological processes (19). All patients with unresolving abnormal liver function tests should be screened for the possibility of Wilson’s disease.

Although we treated the present patient with chemoembolization and PEIT for HCC, the role of the therapy in this patient remains to be reevaluated. His liver volume was highly diminished after chemoembolization (from 1,126 cm³ to 973 cm³). The mechanisms of liver atrophy in this case remain to be clarified, especially the relationship with apoptosis. However, Ishikawa et al (20) and Kobayashi et al (21) have reported a role of chemoembolization in the induction of apoptosis and anoxic stress. Moreover, Strand et al reported the induction of apoptosis both in vivo and in vitro due to copper overload in hepatocytes (22). Since epirubicin or other anticancer agents may also induce apoptosis (23–26), chemoembolization, especially the injection of anticancer agents may have induced or accelerated liver failure in this case. Therefore, we should be aware of chemoembolization in liver diseases, which are at the pre-apoptotic state, such as Wilson’s disease. Further study is necessary in the future to design the appropriate therapeutic intervention for such cases.

References