Abstract

**Purpose.** The aim of this study was to establish the relation between observed ultrasonographic (US) images produced with a galactose-based contrast agent and histologic characteristics of small hepatocellular carcinomas (HCCs).

**Materials and methods.** A total of 64 nodules in 64 patients, 22 well differentiated and 42 moderately differentiated with a histologically proven HCC, smaller than 3.0cm in diameter and who had undergone hepatectomy were consecutively examined by contrast-enhanced US using a galactose-based contrast agent. Perfusion images were acquired by intermittent high-intensity, harmonic power Doppler sonography using a high pulse-repetition frequency and high-pass filter setting. Perfusion images of the arterial and late phases were classified into several patterns and compared with the histologic findings obtained from resected specimens.

**Results.** Most of the well- and moderately differentiated resected HCCs showed hyperechoic change during the arterial phase. However, 13 (59%) of the well-differentiated HCCs showed isoechoic change and 27 (64%) of the moderately differentiated HCCs showed hypoechoic change during the late phase. The difference is statistically significant ($P < 0.0001$). In a comparison of microscopic portal invasion (vp) of HCCs using enhanced US patterns, both vp(−) and vp(+) groups showed a high incidence of the hypervascular pattern during the arterial phase; in contrast, during the late phase 11 (73%) of 15 vp(+) nodules showed hypoechoic change with spotty signals. This difference is statistically significant ($P < 0.0001$) when compared with a high incidence (52%) of signal defect in the vp(−) group. The existence of well-differentiated components associated with the periphery of moderately differentiated HCCs also correlated closely with patterns during the late phase ($P < 0.01$).

**Conclusions.** Late-phase contrast-enhanced US images of small HCCs with a galactose-based contrast agent are useful for predicting specific histologic characteristics.

**Keywords** contrast enhancement · galactose-based contrast agent · hepatocellular carcinoma · ultrasound

Introduction

Advances in contrast-enhanced ultrasonography (US) with intravenous application of commercially available contrast agents and such new ultrasonic imaging technologies as harmonic mode and flash echo imaging are facilitating accurate studies of the hemodynamics of hepatocellular carcinoma (HCC).

Portal invasion, a well-known cause of intrahepatic spreading of HCC, is a primary factor limiting the long-term outlook for patients after treatment of an HCC. Furthermore, microscopic venous invasion is an important determinant of outcome after surgically removing these nodules.

Macroscopic portal or hepatic venous tumor thrombi can be detected with US, hepatic arteriography, computed tomography (CT), magnetic resonance imaging (MRI), and fine-needle aspiration biopsy, among other methods. Microscopic venous invasion of a small HCC is difficult or impossible to depict with imaging methods. Rather, its detection requires precise exploration of resected specimens.

Multicentric carcinogenesis, an HCC with well-differentiated components, is also a recognized cause of frequent intrahepatic recurrence after curative treatment or simultaneous occurrence of HCC. Identifying multicentric carcinogenesis is important to the treatment and outcome of patients with recurrent or simultaneous multi-
Detection of microscopic portal invasions of small HCCs or well-differentiated components associated with moderately differentiated HCCs in which images were later compared with resected specimens has not yet been reported.

In this study, intravenous application of a galactose-based contrast agent (Leovist; Schering AG, Berlin, Germany) was used to enhance ultrasonographic images that were classified into several patterns. Contrast-enhanced US patterns were then compared with the histologic findings from the resected specimens to determine the relation between the patterns and the findings, which in turn helped determine the value of the images for predicting the existence of microscopic portal invasion of small HCCs. We further tried to detect well-differentiated components associated with small, moderately differentiated HCCs. The clinical meanings of contrast-enhanced US findings during the late phase are discussed.

**Materials and methods**

**Subjects**

A total of 64 nodules from 64 patients with small HCCs (<3.0 cm in diameter) without macroscopic tumor thrombi included 22 predominantly well-differentiated and 42 moderately differentiated HCCs proven by histologic examination of surgically resected specimens. They were consecutively examined by contrast-enhanced US with intravenous injection of a galactose-based contrast agent at this institution between November 1999 and September 2002. Among these patients were 44 men and 20 women, with a mean age of 67 years.

**Tumors**

The tumors ranged from 1.2 to 3.0 cm (mean 2.1 cm) in diameter and were located 2.0–10.0 cm from the body surface (mean 6.1 cm). The sizes of predominantly well-differentiated tumors, moderately differentiated tumors with well-differentiated components in the periphery, and moderately differentiated tumors without well-differentiated components were 2.0 ± 0.5 cm (n = 22), 2.1 ± 0.5 cm (n = 21), and 2.2 ± 0.6 cm (n = 21), respectively. These differences are not statistically significant.

Differentiated HCCs were defined as follows. Predominantly well-differentiated HCCs were nodules with well-differentiated components accounting for more than 50% of the nodule in the maximum section of surgically resected specimens; predominantly moderately differentiated HCCs were those with well-differentiated components comprising less than 50% of the maximum section of surgically resected specimens. No poorly differentiated components were included in this series. Histologic analysis of resected specimens proved portal invasion in one well-differentiated HCC nodule and 15 moderately differentiated HCC nodules. Peliotic changes in HCCs were classified as grade 0, indicating no peliotic change in the entire nodule; grade 1, partial existence of peliotic changes in the nodule; and grade 2, many peliotic changes scattered throughout the nodule.

**Laboratory examinations**

Leovist-enhanced US images of well-differentiated HCCs were compared with those of moderately differentiated HCCs to evaluate whether contrast-enhanced US increases the diagnostic confidence regarding small HCCs. All examinations, including contrast-enhanced US, CT, MRI, and arteriography, were undertaken with each patient’s informed consent and in compliance with rules formulated by the ethics committee of this institution.

**Ultrasonographic examination**

We used a commercially available Model SSA 390A ultrasound system (Toshiba, Tokyo, Japan) with a convex-array scanner (PVN-375AT) for the contrast-enhanced US examination and designed the scanning mode for contrast studies as follows. Contrast-enhanced images were acquired by intermittent scanning using high-intensity, harmonic power Doppler sonography with a high pulse-repetition rate and high-pass filter setting. High-intensity intermittent scanning was performed at a high frame rate and low-intensity (mechanical index 0.2) monitoring in dual-display mode. The power Doppler mode transmitted at 2.1 MHz and received at 4.2 MHz in the harmonic mode. For deeply located lesions (≥7 cm from the body surface), the 2.0- to 3.0-MHz fundamental mode was used. The mechanical index (MI) for scanning mode was 0.8–1.6 with a pulse-repetition frequency (PRF) as high as 4.5 KHz and the high-pass filter set as high as 1.34 KHz to eliminate motion artifacts and velocity-dependent Doppler signals. This left only signals with a wide distribution of the frequencies caused by disruption of the microbubbles (flash echo) produced by high-intensity scanning for contrast imaging.

We set intermittent scanning triggers by electrocardiographic R waves at every heartbeat during the arterial phase and every two or three heartbeats during the portal phase, after taking into consideration that the cardiac output would modulate the inflow of contrast agent into the liver. Scanning during the initial vascular phase, including the portal phase, was limited to no more than 1 min for acquiring necessary images and then stopped until the late phase. During the late phase, test scanning of intrahepatic portal veins of the opposite lobe of the liver was performed every minute for 5 min or longer after injecting the contrast agent but before the tumor was scanned. After signals in the intrahepatic vessels of the opposite lobe disappeared, a single manual scan of the tumor was performed in the same plane as that scanned during the arterial phase. The concentration of the contrast agent was 300 mg/ml, and the volume of the bolus injection was 1 ml/10 kg body weight.